



Paediatric Drug Development: Regulatory Framework as per USFDA

Santhosh Kumar S¹, Madhavi Lakshmi Ratna Bhavaraju^{2*} and Shivanad K Mutta²

¹B.Pharm., Department of Pharmaceutical Regulatory Affairs, Acharya & BM Reddy College of Pharmacy, (Affiliated to Rajiv Gandhi University of Health Sciences), Bengaluru, Karnataka, India.

²Department of Pharmaceutical Regulatory Affairs, Acharya & BM Reddy College of Pharmacy, (Affiliated to Rajiv Gandhi University of Health Sciences), Bengaluru, Karnataka, India.

Received: 18 Jun 2024

Revised: 22 Aug 2024

Accepted: 19 Oct 2024

*Address for Correspondence

Madhavi Lakshmi Ratna Bhavaraju

Department of Pharmaceutical Regulatory Affairs,
Acharya & BM Reddy College of Pharmacy,
(Affiliated to Rajiv Gandhi University of Health Sciences),
Bengaluru, Karnataka, India.
E.Mail: madhaviblr@acharya.ac.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

In the intricate world of paediatric drug development, the US Food and Drug Administration (USFDA) play a pivotal role, ensuring that medications are both safe and effective for children. This abstract explores the USFDA's regulatory system, emphasizing important policies and programs intended to close the gap between adult and pediatric pharmacotherapy. Legislative measures that require stringent clinical trials and provide incentives for research in pediatric populations, such as the Pediatric Research Equity Act (PREA) and the Best Pharmaceuticals for Children Act (BPCA), are essential to this framework. The USFDA's strategy places a strong emphasis on age-appropriate formulations, moral issues, and how crucial it is to adjust dosages in accordance with pediatric physiology. Pharmaceutical businesses are better able to meet the special therapeutic needs of children by navigating the complicated regulatory environment, which eventually leads to advances that enhance paediatric health outcomes. The study provides significant new data on regulatory approach for those involved in the development of pediatric medicine.

Keywords: Paediatric, Drug development, Regulatory approach, USFDA, PREA, BPCA.

INTRODUCTION

The process of **transforming a molecule from** a drug candidate (the end-product of the discovery phase) into a product that has been given the green light for commercialization by the relevant regulatory authorities is known as drug development (1). The complexity in drug development has increased manifolds over the past 40 years, requiring preclinical phase of drug development, investigational new drug (IND) application, and complete clinical



**Santhosh Kumar et al.,**

testing before marketing approval from the FDA. Generally, new drug applications (NDAs) or biologics license applications (BLA) are reviewed comprehensively before approval, and then drug performance is resubmitted to regulatory agencies for post-marketing studies. The overarching goal is to bring more efficient and safer treatments to the patients as quickly as possible after a thorough medical evaluation (2).

The USFDA drug development process consists of five essential components, each of which contains numerous phases and stages. We will go over each step and phase of the drug development process to gain a comprehensive understanding of the whole thing. The stages involved in developing drugs are:

Step 1: Discovery and Development

Step 2: Preclinical Research

Step 3: Clinical Development

Step 4: FDA Review

Step 5: FDA Post-Market Safety Monitoring

Discovery and Development: In this phase, experts from academia and industry collaborate to find pharmacological compounds that affect a specific biological target linked to a disease.

Preclinical Research: The goal of preclinical testing is to gather vital information about the safety and efficacy of a medication candidate before it is tested on humans. A candidate's biological impact is frequently illustrated using both in vitro and in vivo models.

Clinical Development: The purpose of clinical trials is to offer particular scientific information about a medication under research. The clinical trial study protocol specifies the procedures to be followed and the manner in which the clinical trial must be conducted. In order to guarantee the safety of the human subjects taking part in the trials, it provides details about the main goals of the study, its design, and statistical considerations. Clinical trials consist of three distinct phases, namely I, II, and III, which vary in terms of time, participant count, and study objectives.

FDA Review: When the sponsor or organization has enough evidence of the drug's safety and efficacy to satisfy FDA regulations for marketing approval, they submit a New Drug Application (NDA) for evaluation. The application for review must also contain data from specific technical viewpoints, such as chemistry, pharmacology, medicine, biopharmaceutics, and statistics. The product may be commercialized in the US if the NDA is accepted.

FDA Post-Market Safety Monitoring: Post-market safety surveillance is the process of keeping an eye on a medication after it has been approved and placed on the market. It is designed to evaluate the long-term safety and efficacy of a drug, possible formulation problems in "the real world," and use for unapproved conditions, or "off-label."(1) (2)

Patients who are 21 years of age or younger at the time of diagnosis or treatment are classified as paediatric patients under the Federal Food, Drug, and Cosmetic Act (FD&C Act). The groupings of paediatric patients are determined by their developmental patterns(3) (4). Table 1 represents the classification of paediatric population in USFDA

REGULATORY LEGISLATION IN USFDA

In the USA, a well-established regulatory system permits the development of paediatric drugs. The introduction of legislation such as the Paediatric Research Equity Act (PREA) in the United States reflects a commitment to addressing the unique challenges associated with paediatric drug development⁽⁶⁾.

Regulatory Pathways in the US

The regulatory pathways for paediatric drugs involve a series of steps and considerations to ensure the safety and efficacy of pharmaceutical products specifically for the paediatric population. Two important pieces of legislation in



**Santhosh Kumar et al.,**

the US are Paediatric Research Equity Act (PREA) and Best Pharmaceuticals for Children Act (BPCA). These legislations play a significant role in shaping the regulatory framework for paediatric drugs ⁽⁷⁾.

BEST PHARMACEUTICALS FOR CHILDREN ACT (BPCA):

The BPCA permits studies aimed at enhancing children's medication use safety and effectiveness. The goal of BPCA is to enhance children's drug use and dose safety and effectiveness. In 2002, BPCA was enacted. The U.S. Food and Drug Administration (FDA) Amendments Act of 2007, the FDA Safety and Innovation Act of 2012, and the FDA Reauthorization Act of 2017 all served to reauthorize it. The BPCA laws were most recently renewed in 2022(8).

Regulatory procedure in BPCA

The regulatory procedure in the Best Pharmaceuticals for the Children Act (BPCA) involves two ways:

- (1) The sponsor submitting the Proposed Paediatric Study Request (PPSR) to USFDA and requests the FDA issue a Written Request (WR) as given in Figure 1.
- (2) Figure 2 describes the FDA issues WR to sponsor.

Conditions for exclusivity

To be eligible for exclusivity, a study must meet certain requirements. Firstly, the FDA must receive a written request. Studies that have already been submitted to the Agency prior to the WR's issuance cannot be granted paediatric exclusivity or a WR.

All the deadlines and conditions mentioned in the WR ought to be fulfilled. The active moiety needs to retain its exclusivity or patent life. Sponsors of off-patent drugs have no financial motivation to carry out trials in paediatric patients (11).

Paediatric Exclusivity

Applicants may be granted paediatric exclusivity provided they meet the requirements of a WR. An extra half-year of exclusivity (6 months). All existing patents and marketing exclusivities for the drug moiety are attached with exclusivity. Positive paediatric studies or the approval of a new indication are not necessary for paediatric exclusivity (12).

PAEDIATRIC RESEARCH EQUITY ACT (PREA) :

The Paediatric Research Equity Act amends the federal Food, Drug, and Cosmetic Act to authorize the FDA to require paediatric studies of drugs or biologics when other approaches are insufficient to ensure

PAEDIATRIC STUDY PLAN (PSP)

Unless the drug is for an indication for which orphan designation has been granted, a sponsor who plans to submit a marketing application (or supplement to an application) for a new active ingredient, new indication, new dosage form, new dosing regimen, or new route of administration must submit an Initial Paediatric Study Plan (iPSP).

For any new application or supplement subject to PREA, the sponsor is required to file an iPSP, even if the FDA has previously granted waivers or deferrals under PREA for the same medicine (14).

Submission Timelines for PSP

A sponsor must submit an iPSP, if required under PREA, before the date on which the sponsor submits the required assessments or investigation and no later than either 60 calendar days after the date of the end-of-phase 2 meeting. After the sponsor submits an iPSP, the FDA has 90 days to review the iPSP and provide a written response to the iPSP, or meet with the sponsor to discuss the iPSP, as appropriate. This review process includes consultation with FDA's internal Paediatric Review Committee (PeRC)(15).





Santhosh Kumar *et al.*,

The sponsor then has a second 90-day period during which it may review FDA comments and initiate any needed negotiations to discuss the iPSP. By the end of this second 90-day review period, the sponsor must submit an agreed iPSP. The FDA then has 30 days after receipt of the agreed iPSP to review and issue correspondence confirming agreement or issue correspondence stating disagreement. If the FDA does not agree, the iPSP is considered a non-agreed iPSP(16). The pathway of submission of paediatric study plan is given in Figure 4(15).

CONCLUSION

The study titled "Paediatric Drug Development: Regulatory Framework as per USFDA "aimed to address the regulatory framework inherent in developing pharmaceuticals tailored for paediatric patients. The results of the study provide insights into a strategic and systematic regulatory approach, emphasizing the importance of a sequential framework to guide the intricate journey of paediatric drug development. The study underscores the collaborative engagement of regulatory authorities, industry stakeholders, healthcare professionals, and patient advocacy groups as pivotal for success. By providing a clear roadmap, the framework aims to streamline the regulatory landscape, fostering innovation, and expediting the availability of safe and effective medications for paediatric patients. Furthermore, the study delves into the regulatory legislation in the United States, highlighting key acts such as the Paediatric Research Equity Act (PREA) and the Best Pharmaceuticals for Children Act (BPCA). The regulatory pathways outlined in the study, with a focus on the USA, illustrate the meticulous steps and considerations required to ensure the safety and efficacy of pharmaceutical products for the paediatric population. The study also provides a detailed overview of the Paediatric Study Plan (PSP) and its submission timelines, reinforcing the commitment to a systematic approach in paediatric drug development. It contributes a valuable resource for understanding the intricacies of regulatory pathways and study plan for paediatric drugs in USA.

ACKNOWLEDGEMENT

We the authors would like to thank Acharya & BM Reddy College of pharmacy for giving us this opportunity.

REFERENCES

1. Kumar A. Drug Development Process: An Easy Guide for You to Know Everything [Internet]. Emeritus Online Courses. 2022.[cited 2024 May 15] Available from: <https://emeritus.org/blog/healthcare-drug-development-process/>
2. Pandey A. Phases of Drug Development Process, Drug Discovery Process | NorthEastBioLab [Internet]. NorthEastBioLab. 2019. [cited 2024 May 15] Available from: <https://www.nebiolab.com/drug-discovery-and-development-process/>
3. Li, Hao& Shi, Fang-Hong & Huang, Shi-Ying & Zhang, Shun-Guo& Chen, Hui-Wen. (2020). The best pharmaceuticals for children—what can we do?. *Translational Pediatrics*. 9. 86-92. 10.21037/tp.2020.02.07.[cited 2024 May 15] Available from: <https://tp.amegroups.org/article/view/37892/html>
4. Zisowsky J, Krause A, Dingemans J. Drug Development for Pediatric Populations: Regulatory Aspects. *Pharmaceutics*. 2010 Nov 29;2(4):364–88 [cited 2024 May 15] Available from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3967144/#:~:text=It%20covers%20pediatric%20study%20planning,of%20a%20drug%20in%20children.>
5. Step-wise Regulatory approach for the Paediatric drug Development [Internet]. www.linkedin.com. [cited 2024 May 15] Available from: <https://www.linkedin.com/pulse/step-wise-regulatory-approach-paediatric-drug-neha/>
6. Mccune S. Fda Overview of the Pediatric Legislation Stakeholder Input on Pediatric Legislation Public Meeting [Internet]. 2019 [cited 2024 May 15] Available from: <https://www.fda.gov/media/132890/download>





Santhosh Kumar et al.,

7. Research C for DE and. Best Pharmaceuticals for Children Act (BPCA). FDA [Internet]. 2019 Feb 9; [cited 2024 May 15] Available from: <https://www.fda.gov/drugs/development-resources/best-pharmaceuticals-children-act-bpca>
8. Institute of Medicine (US) Forum on Drug Discovery D. Regulatory Framework [Internet]. www.ncbi.nlm.nih.gov. National Academies Press (US); 2008 [cited 2024 May 15]. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK3997/#:~:text=BPCA%20also%20allows%20the%20FDA>
9. Best Pharmaceuticals for Children Act [Internet]. 2000. [cited 2024 May 15]. Available from: <https://www.congress.gov/107/plaws/publ109/PLAW-107publ109.pdf>
10. Best Pharmaceuticals for Children Act - BPCA [Intern] [cited 2024 May 15] Available from: <https://www.nichd.nih.gov/research/supported/bpca#:~:text=BPCA%20authorizes%20research%20to%20improve>
11. Pediatric Drug Development: Regulatory Considerations -Complying With the Pediatric Research Equity Act and Qualifying for Pediatric Exclusivity Under the Best Pharmaceuticals for Children Act Guidance for Industry DRAFT GUIDANCE [Internet]. [cited 2024 May 15]. Available from: <https://www.fda.gov/media/168201/download>
12. Federal Register :: Request Access [Internet]. unblock.federalregister.gov. [cited 2024 May 15]. Available from: <https://www.federalregister.gov/documents/2023/05/18/2023-10610/pediatric-drug-development-regulatory-considerations-complying-with-the-pediatric-research-equity#:~:text=Under%20the%20BPCA%2C%20certain%20applications>
13. Dual draft guidances outline FDA vision for pediatric drug development, exclusivity [Internet]. Raps.org. 2024 [cited 2024 May 15]. Available from: <https://www.raps.org/News-and-Articles/News-Articles/2023/5/Dual-draft-guidances-outline-FDA-vision-for-pediat>
14. Research C for DE and. Pediatric Research Equity Act | PREA. FDA [Internet]. 2019 Nov 7; [cited 2024 May 15] Available from: <https://www.fda.gov/drugs/development-resources/pediatric-research-equity-act-prea>
15. Adamadmin. How to Prepare for a Pediatric Study Plan Submission [Internet]. Allucent. 2019 [cited 2024 May 15]. Available from: <https://www.allucent.com/resources/blog/how-successfully-prepare-your-pediatric-study-plan#:~:text=The%20FDA%20guidance%20on%20pediatric>
16. Research C for DE and. Pediatric Study Plans: Content of and Process for Submitting Initial Pediatric Study Plans and Amended Initial Pediatric Study Plans [Internet]. U.S. Food and Drug Administration. 2020. [cited 2024 May 15] Available from: <https://www.fda.gov/regulatory-information/search-fda-guidance-documents/pediatric-study-plans-content-and-process-submitting-initial-pediatric-study-plans-and-amended>

Table 1: Classification of Paediatric Population as per USFDA⁽⁵⁾

Paediatric population	Age
Neonates	0 to 28 days
Infants	29 days to less than 2 years
Children	2 to less than 12 years
Adolescents	12 to 21 years



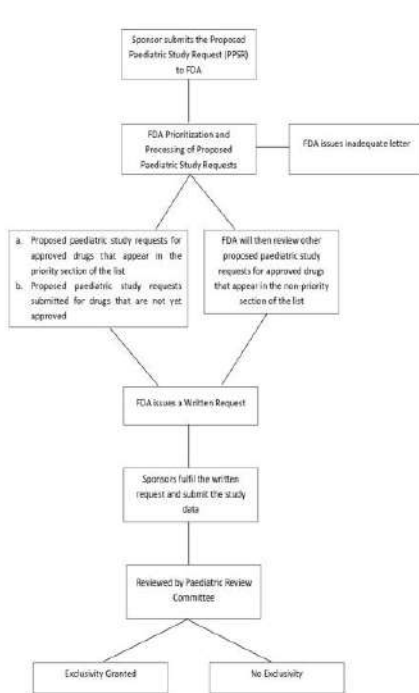


Figure 1. Sponsor submits the Proposed Paediatric Study Request (PPSR) to FDA (09)

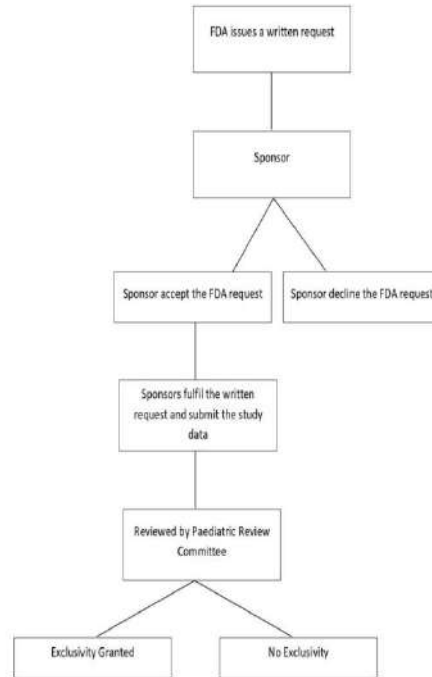


Figure 2. FDA issues WR to sponsor (10)

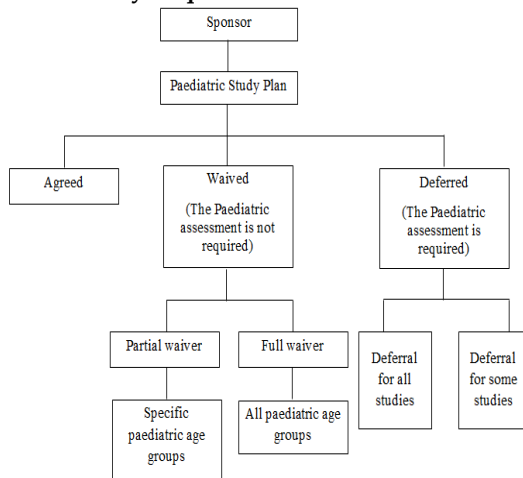


Figure 3 Regulatory Pathway in PREA (13)

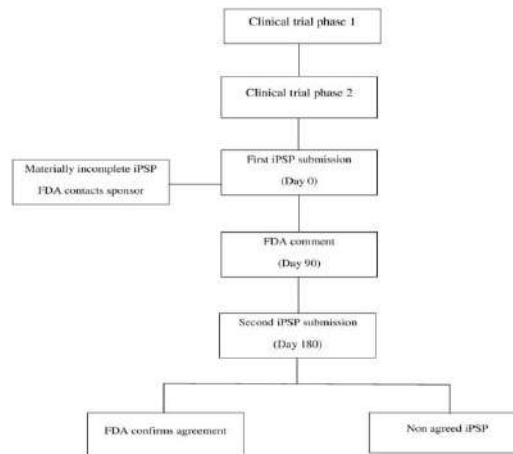


Figure 4 Timeline of iPSP Submission (15)





Indexed Results and Tables Relating to the Quick Switching System (QSS-1) with a Single Sampling Plan as the Reference Plan

Esha Raffie B*

Assistant Professor, Department of Mathematics and Statistics, Sri Krishna Arts and Science College, (Affiliated to Bharathiar University), Coimbatore, Tamil Nadu, India.

Received: 29 Dec 2023

Revised: 20 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Esha Raffie B*

Assistant Professor, Department of Mathematics and Statistics,
Sri Krishna Arts and Science College, (Affiliated to Bharathiar University),
Coimbatore, Tamil Nadu, India.
E.Mail: esharaffie@gmail.com>



Esha Raffie

original work is properly cited. All rights reserved.

ABSTRACT

This research examines the Quick Switching System (QSS-1) using a Single Sampling (SSP) Plan as a reference plan with various quality levels. The study provides performance measures and procedures for designing the system with different entry parameters. The main objective is to reduce inspection costs while maintaining an acceptable level of quality for the customer.

Keywords: Quick Switching System, Single Sampling Plan, Acceptable Quality Level, Limiting Quality Level, Palya Distribution.

INTRODUCTION

QSS-SSP ($n: u_1, u_2, v_1, v_2$) refers to a quick switching system where the usual SSP plan has a sample of size 'n' and acceptance numbers u_1, u_2 ($u_1 < u_2$) and the tightened SSP plan has a sample of size n and acceptance numbers v_1, v_2 ($v_1 < v_2, v_1 \leq u_1$ and $v_2 < u_2$).

Romboski (1969) observes the following advantages for QSS-1.

- The composite OC curve of QSS-1 is having better shape (close to z shape) than the corresponding normal and tightened OC curves).
- As the tightening becomes plain the composite OC curve methods to the ideal form.
- The system results in reduction of sample size than the equivalent reference plan.

Switching Procedures

A sampling system encompasses of plans, which are acceptable to normal, tightened and reduced inspections. The switching rules are given below.





Esha Raffie

Normal to tightened:

When normal inspection is in effect tightened assessment shall be instituted when two out of five consecutive lots or batches have been rejected on original inspection.

Tightened to normal:

When tightened inspection is in effect normal inspection shall be instituted when five consecutive lots have been accepted on unique inspection.

Normal to reduced:

When normal inspection is in effect, reduced inspection shall be instituted providing that all the following conditions are fulfilled.

1. The previous 10 lots or batches have been on normal inspection and none has been on original inspection
2. The total number of uncharacteristic units in the sampling from the preceding 10 lots or batches is equal to or less than the applicable number
3. Production is at fixed rate and
4. Reduced inspection is considered desirably by the responsible authority.

Reduced to normal:

1. A lot or batch is rejected or
2. A lot or batch is measured acceptable under the process of MIL-STD-105D system procedures.
3. Production becomes uneven or delayed
4. Other conditions warrant that normal inspection shall be instituted.

This section gives a review on Quick Switching System-r (n, c_N, c_T) where $r = 1, 2, 3$ are discussed. The contributions made by the author are stated at the end of the sections.

Dodge (1967) proposed a new sampling system consisting of pairs of normal and tightened plans. The application of the system is as follows

1. Adopt a pair of sampling plans, a normal plan (N) and tightened plan (T), the plan T to be tightened OC curve wise than plan N.
2. Use plan N for the first lot (optional): can start with plan T; the OC curve properties are the same ; but first lot protection is greater if plan T is used.
3. For each lot inspected; if the lot is accepted, use plan N for the next lot and if the lot is rejected, use plan T for the next lot'.

Due to instantaneous switching between normal and tightened plan, this system is referred as “ Quick Switching System “. Using the concept of Markov Chain, the OC function of QSS-1 is derived by Romboski (1969) as

$$Pa(p) = P_T / (1 - P_N + P_T) \quad (1)$$

Romboski(1969) introduced QSS-1 ($n; c_N, c_T$) which is a QSS-1 with single sampling plan as a mention plan [(n, c_N) and (n, c_T) are respectively the normal and tightened single sampling plans with $c_T < c_N$].

Designation of Quick Switching System

Romboski (1969) has extensively studied QSS by taking pairs of single sampling plan. The designation of the system is as follows :





Esha Raffie

i) $QSS(n; c_N, c_T)$ – refers to a QSS where the single sampling normal plan has a sample size of n and an acceptance number of c_N , and the tightened single sampling plan has the same sample size as that of the normal plan but with acceptance number c_T . In general, $c_T \leq c_N$ and when $c_T = c_N$ then the system degenerates into a single sampling plan.

ii) $QSS(n, kn; c_0)$ – refers to a QSS where the normal and tightened single sampling plans have the same acceptance number but on tightened inspection the sample size is a multiple of k ($k \geq 1$) of the sample size on normal inspection. If $k = 1$, the system degenerates into single sampling plan.

Romboski (1969) has given the QC function of $QSS(n; c_N, c_T)$ and $QSS(n, kn; c_0)$ as

$$P_a(p) = \frac{P_T}{1 - P_N + P_T}$$

Where P_N and P_T are explained.

SINGLE SAMPLING PLAN

A brief note on single sampling plan is given in this section. A single sampling plan is characterized by sample size n and the acceptance number c , sampling inspection in which the decision to accept or not to accept a lot is based on the inspection of a single sample of size n .

Operating procedure

Select a random sample of size n and count the number of non-conforming units d . If there is c or less non-conforming units, the lot is accepted, otherwise the lot is rejected. Thus the plan is characterized by two parameters viz, the sample size n and the acceptance number ' c '. The OC function of the single sampling plan is given as

$$P_a(p) = P(d < c, n) \quad (2)$$

Peach and Littauer (1946) have given tables for determining the single sampling plan for static $\alpha = \beta = 0.05$. They have used the relation that for even degrees of freedom χ^2 gives the outline of a Poisson distribution as the basis for developing tables of a single sampling plan. They have introduced the concept of the operating ratio p_2/p_1 as a measure for the power of discrimination of the OC curve. The values of p_2/p_1 and np_1 are intended against different values of c for fixed $\alpha = \beta = 0.05$ using the table, a single sampling plan can be selected for given p_1 and p_2 .

Burgess (1948) has given a graphical method to obtain single sampling plans for given values of $(p_1, 1-\alpha)$ and (p_2, β) with the help of the Poisson cumulative probability chart. Cameron (1952) has also given a table, which is an extension of the table given by Peach and Littauer (1946). Cameron's table is based on Poisson distribution and can be used to design single sampling for all the popular values of producer's and consumer's risks.

Further tabularized p_2/p_1 values for $(\alpha, \beta) = (0.05, 0.10), (0.05, 0.05), (0.05, 0.01), (0.01, 0.05)$, and $((0.01, 0.10)$ for c values ranging from 0 to 49. Using Cameron (1952) table, one can select a single sampling plan for given p_1 and p_2 , α and β . Horsnell (1954) has also presented a table similar to that of Cameron, giving p_2/p_1 and np_1 values for $\alpha = 0.05, 0.01$ and $\beta = 0.10, 0.05$, and 0.01 but restricting c from 1 to 20. Horsnell (1954) has further illustrated the approximation involved in replacing binomial probabilities by corresponding Poisson probabilities by comparison of p values for $P_a(P) = 0.99, 0.95, 0.50, 0.10$ and 0.01 for different single sampling plans. Kirfpatrick (1965) has given two tables for the selection of single sampling plans corresponding to different values of p_1 and p_2 . The first table gives single sampling plans when OC curves pass very close to the specified p_1 and not so close to the specified p_2 and the second table gives single sampling plan when OC curves pass very close to the specified p_1 . The plans indexed are based on Grubbs (1948) tabulation of p_1 and p_2 for $n = 1(1)50$ and $c = 0(1)9$.

Guenther (1969) has developed a systematic research procedure for finding the single sampling plans for given p_1, p_2, α and β based on the binomial, hyper geometric and Poisson models. Hailey (1980) has presented a computer





Esha Raffie

program to obtain minimum sample size single sampling plans based on Guenther (1969) procedure for given p_1 , p_2 , α and β .

Stephens (1978) has given a procedure and tables for finding the samples size and acceptance number of a single sampling plan for given two points on the OC curve, viz., $(p_1, 1-\alpha)$ and (p_2, β) using normal approximation to binomial distribution. By using this procedure any point $(p_1, 1-\alpha)$ and (p_2, β) may be specified and the applicable sample size and acceptance number can be found quite straight forward based on the formula for n . Schilling and Johnson (1980) presented a set of tables for the construction and evolution of matched sets of single, double and multiple sampling plans. They may be used to derive two point individual plans to specified values of fraction defective and probability of acceptance.

Golub (1953) has given a method and tables for finding the acceptance number 'c' of a single sampling plan involving minimum sum of producer's and consumer's risks for given p_1 and p_2 when the sample size n is fixed. Soundararajan (1981) has extended the Golub's approach to single sampling plans when the conditions for application of the poisson model. Vijayathilakan (1982) has given procedure and tables for designing single sampling plans when the sample size is fixed and the sum of the weighted risks is minimized. Nirmala and Suresh (2017) presented a Continuous sampling plan indexed through maximum allowable average out going quality.

Properties of OC Curve

- 1) For a QSS ($n; c_N, c_T$) the steepness of the operating characteristic (OC) curve and hence its discriminating power is depending upon the difference between c_N and c_T . For a fixed n , and fixed c_N , as c_T decreases, the resulting composite OC curve gets steeper.
- 2) For a QSS($n, kn; c_0$) for $k > 0$, the slope of the composite OC curve increases as k increases. The conditions for application under which the Quick Switching System can be applied and the operating procedure are as follows:

Conditions for Application

1. The production is steady so that results on current and previous lots are broadly indicative of an ongoing process and submitted lots are expected to be essentially of the same quality.
2. Lots are submitted considerably in the order of production.
3. Inspection is by characteristics with quality defined as fraction nonconforming.

Operating Procedure of QSS (n, c_N, c_T)

Step 1: From a lot, take a random sample of size 'n' at the normal level. Count the number of defectives 'd'.

- i) If $d \leq c_N$, accept the lot and repeat step 1
- ii) If $d > c_N$, reject the lot and go to step 2.

Step 2: From the next lot, take a random sample of size n at the tightened level.

Count the number of defectives 'D'.

- i. If $D \leq c_T$, accept the lot and use step 1
- ii. If $D > c_T$, reject the lot and repeat step 2

Romboski (1969) has introduced another sampling inspection system QSS-1 ($n, kn; c_0$) which is a QSS-1 with single sampling plan as a reference plan (n, c_0) and (kn, c_0) , $k > 1$ are respectively the normal and tightened single sampling plans. The conditions for application of this system are the same as that of QSS-1 ($n; c_N, c_T$).

Operating procedure for QSS (n, kn, c_0)

1. For a lot, take a random sample of size 'n' at the normal level. Count the number of defectives 'd'
 - If $d \leq c_0$, accept the lot and repeat step 1.
 - If $d > c_0$, reject the lot and go to step 2.





Esha Raffie

2. From the next lot, take a random sample of size 'kn' at the tightened level. Count the number of defectives 'D'.
 - If $D \leq c_0$, accept the lot and use step1
 - If $D > c_0$, reject the lot and repeat step 2.

The OC function of the system is given in equation (1.2.1) with
 P_N – proportion of lots expected to be accepted when using (n, c_0) plan
 P_T – proportion of lots expected to be accepted when using (kn, c_0) plan

Romboski (1969) has derived the OC function for QSS-1 (n, kn, c_0) as,

$$Pa(P) = \frac{P(d \leq C_T; n)}{1 - P(d \leq C_N; n) + P(d \leq C_T; n)} \tag{3}$$

The Condition for application of Quick Switching System

1. The production is steady so that results on current and preceding lots are broadly indicative of a ongoing process and submitted lots are expected to be fundamentally of the same quality.
2. Lots are submitted significantly in his order of production.
3. Inspection by qualities is considered with quality defined as fraction nonconforming 'p'.

Operating Procedure for the plan

1. Under the normal examination, inspect using the SSP plan with parameters n, u_1 and u_2 . If a lot is accepted, continue with normal inspection. If a lot is rejected, go to step2.
2. Inspect under tightened examination using the SSP plan with sample size 'n' and acceptance number v_1 and v_2 . If a lot is accepted, use step1 for the next lot, otherwise continue step2.

Measures of Performance

Operating Characteristics function

Based on Romboski (1965) and Sankar and Mahopatra (1991), the expression for OC function of QSS-SSP is given by

$$Pa(p) = \frac{P(d \leq c_T; n)}{P(d \leq c_N; n)} \tag{4}$$

$$P(d \leq c_N; n) = \frac{P_a(1 - P_c)^i + P_c P_a^i}{(1 - P_c)} \tag{5}$$

Where $P_a = P [d \leq u_1]$ and $P_c = P [u_1 < d < u_2]$





Esha Raffie

$$P(d \leq c_T; n) = \frac{P_a(1 - P_c)^i + P_c P_a^i}{(1 - P_c)} \tag{6}$$

Where $P_a = P [d \leq v_1]$ and $P_c = P [v_1 < d < v_2]$

Palya Distribution

$$PY_{N,m,r,c}(x) = \frac{\binom{r+x-1}{c} \binom{N-r}{c} \binom{N-x-1}{c} \binom{n-x}{c}}{\binom{N}{c} + n - 1} Cn, \quad x = 0, 1, 2, \dots, n. \tag{7}$$

Where N, n, r and c are natural numbers.

Using equation (4), the properties of the type B OC curves of QSS-SSP are analyzed and are given below:

Fig 1. Gives the OC curves of SSP (100; 2,4), QSS-SSP (100; 2, 4 ; 1, 2) and SSP (100; 1,2). From the figure it is observed that the system utilizes the normal SSP plan when the quality is good and the tightened SSP plan when the quality is poor. OC curves of QSS-SSP where the sample size and the acceptance numbers of the normal SSP plan are same whereas the acceptance numbers of the tightened SSP plan are allowed to decrease. As v_1 and v_2 become smaller, the discriminating power of the OC curve increases.

Plotting the OC-curve of a given systems:

Table 1 can be used to obtain nine values of 'p' and $P_a(P)$ to plot the OC-curve of a given QSS-1 ($n, k_n; c_0$). For example, for given QSS-1 (100,150;1,5) one gets $k=150/100=1.5$. Dividing the entries in the row corresponding to $u_1=1, u_2=5$ and $k=1.5$ of table 1 by normal Sample single 100, one gets the values of p and corresponding $P_a(P)$ values are given in the column heading. The values of 'p' and $P_a(P)$ of QSS-1 (100,150;1,5).

Designing the systems given Single Sampling Plan (SSP) and a point on the OC-curve

Table 1 can be used to design QSS-SSP when the sample is fixed at 'n' and a point on the OC-curve ($p, P_a(P)$) is specified. To design a system, calculate 'np' and under the column $P_a(p)$ 0.99 in Table (1) find the value '0.473'. The $c_1, c_2, d_1,$ and d_2 values corresponding to the selected tabular value together with the given n, determine the sampling system to be used. For example, let $n=150, p_1=0.02$ and $P_a(p) = 0.95$ is 0.523. scan the column headed by $P_a(p) = 0.95$, to find the value which is nearer to the desired value of 0.5, the value is 0.523 which corresponds to the parameters $c_1 = 0, c_2=1, d_1=0$ and $d_2=1$ for the desired QSS-SSP.

CONCLUSION

In acceptance sampling the producer and consumer plays a leading role and hence one allows a certain level of risk for producer and consumer. It is comprehensible to design any sampling plan with the accompanying quality levels, concern to producer and consumer in practice. Hence selection measures are considered in this paper with inflection point on the OC curve. Tables provided in this paper are tailor-made which are handy and ready-made, which are also well considered for assessment purposes.

REFERENCES

1. Devaraj Arumaniyagam.S., 1991, Contribution to the Study of Quick Switching System and its applications, Ph.D thesis, Department of Statistics, Bharathiar University, Coimbatore, Tamilnadu, India





Esha Raffie

2. Deepa, O.S., 2002, Studies on Certain Classical and Bayesian Acceptance Sampling Plans, Ph.D thesis, Department of Statistics, Bharathiar University, Coimbatore, Tamilnadu, India
3. Mandelson, R. 1962, The Statistician engineer and sampling plans, Industrial Quality Control, 19.12-15
4. Mayer, P.L. 1967, A Note on the sum of poisson Probabilities and Applications, Annals of the institute of statistical mathematics, 19, 537-542.
5. Nirmala.V and Suresh. K. K. (2017): Construction and Selection of Continuous Sampling Plan of type (CSP-T) Indexed through Maximum Allowable Average Outgoing Quality, Journal of Statistics and Management System, Vol. 20, No. 3, pp. 441-457.
6. Romboski, L.D. 1969, An Investigation of Quick Switching Acceptance sampling Systems, Ph.D Thesis, Rutgers-The state University, New Brunswick , New Jersey
7. Subramaniam, C. 1990, Switching Procedure in Sampling Inspection, M.Phil thesis, Department of Statistics, Bharathiar University, Coimbatore, Tamilnadu, India
8. Suresh, K.K. 1993, A study on Acceptance Sampling using Acceptable and Limiting Quality Levels, Ph.D thesis, Department of Statistics, Bharathiar University, Coimbatore, Tamilnadu, India
9. Suresh, K.K. and Ramkumar, T.B. 1996, Selection of a sampling plan Indexed with Maximum Allowable Average Outgoing Quality, Journal of Applied Statistics, Vol. 23, No.6, 645-654.

Table 1. Plotting the OC-curve of a given systems

P	0.0115	0.0152	0.01731	0.0204	0.05413	0.02416	0.03712	0.04613	0.06521
Pa(P)	0.99	0.95	0.90	0.75	0.50	0.25	0.10	0.05	0.01

Table 2 Pa(p) values for QSS-1 with SSP plan as a reference plan for (i, c_N, c_T)

C _N		C _T		i	0.99	0.950	0.9	0.75	0.5	0.25	0.1	0.05	0.01
C ₁	C ₂	D ₁	D ₂										
0	1	0	1	2	0.473	0.523	0.673	0.873	1.123	1.423	1.773	2.173	2.623
				4	0.317	0.367	0.517	0.717	0.967	1.267	1.617	2.017	2.467
				6	0.251	0.301	0.451	0.651	0.901	1.201	1.551	1.951	2.401
				8	0.212	0.262	0.412	0.612	0.862	1.162	1.512	1.912	2.362
				10	0.181	0.231	0.381	0.581	0.831	1.131	1.481	1.881	2.331
0	1	0	1	2	0.48	0.530	0.68	0.88	1.13	1.43	1.78	2.18	2.63
				4	0.321	0.371	0.521	0.721	0.971	1.271	1.621	2.021	2.471
				6	0.252	0.302	0.452	0.652	0.902	1.202	1.552	1.952	2.402
				8	0.21	0.260	0.41	0.61	0.86	1.16	1.51	1.91	2.36
				10	0.185	0.235	0.385	0.585	0.835	1.135	1.485	1.885	2.335
1	1	0	1	2	1.267	2.317	3.467	4.667	5.917	7.217	8.567	9.967	11.41
				4	1.015	1.065	1.215	1.415	1.665	1.965	2.315	2.715	3.165
				6	0.846	0.896	1.046	1.246	1.496	1.796	2.146	2.546	2.996
				8	0.788	0.838	0.988	1.188	1.438	1.738	2.088	2.488	2.938
				10	0.724	0.774	0.924	1.124	1.374	1.674	2.024	2.424	2.874
1	1	0	0	2	1.327	2.377	3.527	4.727	5.977	7.277	8.627	10.02	11.47
				4	1.032	1.082	1.232	1.432	1.682	1.982	2.332	2.732	3.182
				6	0.858	0.908	1.058	1.258	1.508	1.808	2.158	2.558	3.008
				8	0.794	0.844	0.994	1.194	1.444	1.744	2.094	2.494	2.944
				10	0.732	0.782	0.932	1.132	1.382	1.682	2.032	2.432	2.882
0	1	1	0	2	2.173	2.223	2.373	2.573	2.823	3.123	3.473	3.873	4.323
				4	1.788	1.838	1.988	2.188	2.438	2.738	3.088	3.488	3.938





Esha Raffle

				6	1.632	1.682	1.832	2.032	2.282	2.582	2.932	3.332	3.782
				8	1.48	1.530	1.68	1.88	2.13	2.43	2.78	3.18	3.63
				10	1.388	1.438	1.588	1.788	2.038	2.338	2.688	3.088	3.538
0	1	1	0	2	2.174	2.224	2.374	2.574	2.824	3.124	3.474	3.874	4.324
				4	1.814	1.864	2.014	2.214	2.464	2.764	3.114	3.514	3.964
				6	1.628	1.678	1.828	2.028	2.278	2.578	2.928	3.328	3.778
				8	1.482	1.532	1.682	1.882	2.132	2.432	2.782	3.182	3.632
				10	1.399	1.449	1.599	1.799	2.049	2.349	2.699	3.099	3.549
0	1	1	0	2	2.219	2.269	2.419	2.619	2.869	3.169	3.519	3.919	4.369
				4	1.825	1.875	2.025	2.225	2.475	2.775	3.125	3.525	3.975
				6	1.625	1.675	1.825	2.025	2.275	2.575	2.925	3.325	3.775
				8	1.493	1.543	1.693	1.893	2.143	2.443	2.793	3.193	3.643
				10	1.402	1.452	1.602	1.802	2.052	2.352	2.702	3.102	3.552
1	1	1	0	2	2.272	3.322	4.472	5.672	6.922	8.222	9.572	10.97	12.42
				4	1.829	1.879	2.029	2.229	2.479	2.779	3.129	3.529	3.979
				6	1.625	1.675	1.825	2.025	2.275	2.575	2.925	3.325	3.775
				8	1.493	1.543	1.693	1.893	2.143	2.443	2.793	3.193	3.643
				10	1.401	1.451	1.601	1.801	2.051	2.351	2.701	3.101	3.551
1	1	1	0	2	2.987	4.037	5.187	6.387	7.637	8.937	10.28	11.68	13.13
				4	2.595	2.645	2.795	2.995	3.245	3.545	3.895	4.295	4.745
				6	2.37	2.420	2.57	2.77	3.02	3.32	3.67	4.07	4.52
				8	2.227	2.277	2.427	2.627	2.877	3.177	3.527	3.927	4.377
				10	2.102	2.152	2.302	2.502	2.752	3.052	3.402	3.802	4.252
C _N		C _T		i	0.99	0.95	0.9	0.75	0.5	0.25	0.1	0.05	0.01
C ₁	C ₂	C ₁	C ₂										
0	1	0	1	2	3.187	3.237	3.337	3.487	3.687	3.937	4.237	4.587	4.987
				4	2.692	2.742	2.842	2.992	3.192	3.442	3.742	4.092	4.492
				6	2.543	2.593	2.693	2.843	3.043	3.293	3.593	3.943	4.343
				8	2.255	2.305	2.405	2.555	2.755	3.005	3.305	3.655	4.055
				10	2.137	2.187	2.287	2.437	2.637	2.887	3.187	3.537	3.937
0	1	0	1	2	3.94	3.99	4.091	4.241	4.441	4.690	4.990	5.341	5.740
				4	3.45	3.501	3.601	3.750	3.95	4.201	4.501	4.850	5.251
				6	3.203	3.253	3.353	3.503	3.703	3.953	4.253	4.603	5.003
				8	3.024	3.074	3.174	3.324	3.524	3.774	4.074	4.424	4.824
				10	2.894	2.944	3.044	3.194	3.394	3.644	3.944	4.294	4.694
1	1	0	1	2	4.025	5.075	6.175	7.325	8.525	9.775	11.075	12.425	13.82
				4	3.572	3.622	3.722	3.872	4.072	4.322	4.622	4.972	5.372
				6	3.218	3.268	3.368	3.518	3.718	3.968	4.268	4.618	5.018
				8	3.036	3.086	3.186	3.336	3.536	3.786	4.086	4.436	4.836
				10	2.894	2.944	3.044	3.194	3.394	3.644	3.944	4.294	4.694
1	1	0	0	2	4.248	5.298	6.398	7.548	8.748	9.998	11.29	12.64	14.04
				4	3.51	3.560	3.660	3.810	4.010	4.260	4.560	4.910	5.310
				6	3.264	3.314	3.414	3.564	3.764	4.014	4.314	4.664	5.064





Esha Raffie

				8	3.037	3.087	3.187	3.337	3.537	3.787	4.087	4.437	4.837
				10	2.896	2.946	3.046	3.196	3.396	3.646	3.946	4.296	4.696
0	1	1	0	2	4.783	4.833	4.933	5.083	5.283	5.533	5.833	6.183	6.583
				4	4.281	4.331	4.431	4.581	4.781	5.031	5.331	5.681	6.081
				6	4.214	4.264	4.364	4.514	4.714	4.964	5.264	5.614	6.014
				8	3.810	3.861	3.961	4.110	4.311	4.561	4.861	5.211	5.611
				10	3.657	3.707	3.807	3.957	4.157	4.407	4.707	5.057	5.457
0	1	1	0	2	4.874	4.924	5.024	5.174	5.374	5.624	5.924	6.274	6.674
				4	4.331	4.381	4.481	4.631	4.831	5.081	5.381	5.731	6.131
				6	4.031	4.081	4.181	4.331	4.531	4.781	5.081	5.431	5.831
				8	3.822	3.872	3.972	4.122	4.322	4.572	4.872	5.222	5.622
				10	3.667	3.717	3.817	3.967	4.167	4.417	4.717	5.067	5.467
0	1	1	0	2	4.555	4.605	4.705	4.855	5.055	5.305	5.605	5.955	6.355
				4	4.345	4.395	4.495	4.645	4.845	5.095	5.395	5.745	6.145
				6	4.008	4.058	4.158	4.308	4.508	4.758	5.058	5.408	5.808
				8	3.835	3.885	3.985	4.135	4.335	4.585	4.885	5.235	5.635
				10	3.672	3.722	3.822	3.972	4.172	4.422	4.722	5.072	5.472
1	1	1	0	2	4.975	6.025	7.12	8.275	9.475	10.72	12.02	13.37	14.77
				4	4.37	4.42	4.52	4.67	4.87	5.12	5.42	5.77	6.17
				6	4.051	4.101	4.201	4.351	4.551	4.801	5.101	5.451	5.851
				8	3.843	3.893	3.993	4.143	4.343	4.593	4.893	5.243	5.643
				10	3.686	3.736	3.836	3.986	4.186	4.436	4.736	5.086	5.486

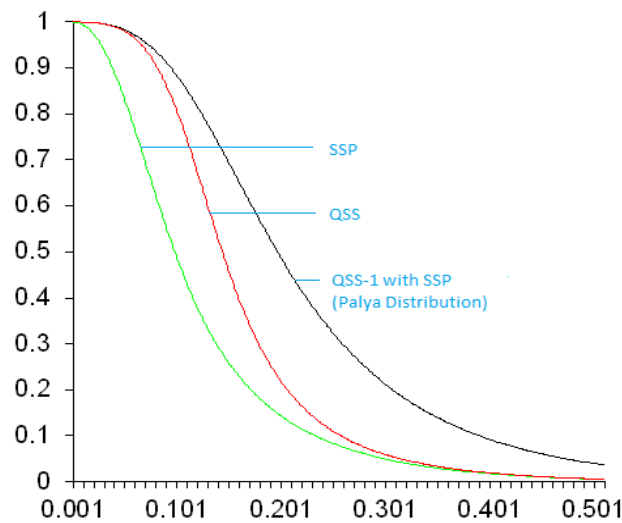


Fig.1: Quick Switching System (QSS-1) with Single Sampling (SSP) Plan with Palya Distribution





Heavy Metal Contamination of Soil: A Tractable Study in Gold Tailings Area of Kolar Gold Fields, India- using ICP-OES and AES

Ashok Dhayalan^{1*}, Vinoth Kumar C², Saraswathi¹, Jagadisha TV¹ and Naveen Kumar S³

¹Assistant Professor, Department of Life Sciences, Kristu Jayanti College (Autonomous), (Affiliated to Bengaluru North University) Bengaluru, Karnataka, India.

²Assistant Professor, Department of Computer Sciences, Brindavan College of Engineering, (Affiliated to the Visvesvaraya Technological University), Bengaluru, Karnataka, India.

³Assistant Professor, Department of Life Sciences, BGS Science Academy & Research Center, Chickballapur, Karnataka, India.

Received: 03 Jun 2024

Revised: 06 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Ashok Dhayalan

Assistant Professor, Department of Life Sciences,
Kristu Jayanti College (Autonomous),
(Affiliated to Bengaluru North University),
Bengaluru, Karnataka, India.
E.Mail: ashok.dj7@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The study has a dual objective, firstly to determine the concentration of arsenic (As) in the soil and ground water within the vicinity of a historical gold mine region located in the Kolar Gold fields (KGF), Karnataka. The study employed Thermo Scientific's Inductively Coupled Plasma-Optical Emission Spectroscopy (ICP-OES) and ICP-AES using Iteva software to analyze the distribution of arsenic in soil and ground water. Four soil and ground water samples were collected from the selected study sites (A, B, C and D) proximity to residential area, were examined from January - December 2023. The analysis of soil samples collected from the four mine landfills revealed elevated levels of heavy metal. The highest level of arsenic that can be present in the soil is 12 mg/kg (WHO 2008). The highest average arsenic content in the soil samples during January-December 2023 analyzed from Champion was 120.83 mg/kg, site A (36.08 mg/kg), D (32.17 mg/kg), and B (28.00 mg/kg) respectively. The maximum permissible limit of arsenic in groundwater is 0.05 mg/L (USEPA 2009). The ground water analysis revealed samples highly contaminated with arsenic (0.863 mg/L) in site C (Champion). The average lead content is found to be 0.559 mg/L, at sites B, A and D is 0.323 mg/L, B, 0.277 mg/L 0.161 mg/L respectively. The pH and electrical conductivity of all the water samples were within the permissible limits, but hardness of the ground water was two times higher than the permissible limit in the sample site D (Balghat).

Keywords : Soil pollution, Ground water pollution, Lead, ICP-OES, ICP-AES.



Ashok Dhayalan *et al.*,

INTRODUCTION

There have been momentous global environmental emissions of hazardous chemicals as a result of growing urbanization and industry [1, 2]. While some of these poisons are naturally occurring, human causes, particularly mining operations, have significantly accelerated their growth. Despite having a sizable positive social and economic impact on a country, it is difficult to ignore the mining's long-term detrimental consequences on the public health and the [3]. Over the past few decades, urbanization has rapidly increased due to the tremendous expansion in the human population. Due to the fact that untreated wastewater discharge pollutes water bodies and promotes diseases related to water, the correct management of the enormous amount of urban wastewater is an international challenge the [4]. The town of Kolar Gold Fields, often known as K.G.F., is located in the Bangarpet taluk of the Kolar district in the Indian state of Karnataka. In order to take over the mining operations of the mines situated at latitude 12° 53'12" N and longitude 78°15'03" E, at the southernmost point of a short schist strip of the township, KGF, whose residents are primarily the families of gold mine workers, the Government of India established the public company (Bharat Gold Mines Limited), or BGML. For more than a century, underground gold mining has been practiced at KGF. 65 kilometers of tunnel construction have been used to mine gold to a depth of 3 kilometers below the surface, and 40 million tons of mill tailings have accumulated the [5].

Metallurgical extraction must first break the crystallographic connections in order to extract the required element or compound from the ore source the [6]. This process creates a considerable amount of waste in the case of gold mining, when practically all of the ore obtained is discarded the [7]. After the valuable ore has been extracted during the mining process, the leftover finely powdered rock and water are known as tailings. There are significant amounts of tailings that have been discarded in the open in many nations where environmental restrictions are not effectively implemented. Tailings particles have a similar chemical and physical makeup to similar river sand and mud the [8]. Tailings, a by-product of gold extraction that is heavily polluted with heavy metals, are the main product (HM) the [9, 10]. These metals seep out into the environment uncontrollably when they come into touch with water or are dispersed by the wind the [11]. The quality of both surface and groundwater is significantly impacted by surface impacts like tailings and rock dumps. The process of using cyanide to extract gold from ore, also contributes to global warming, hydrogen cyanide emissions, and the creation of vast number of tailings, which might be a source of heavy metals (HMs) the [12].

The aforementioned factors are only a few of the many sources of tailing characteristics. Sediments from mines frequently resemble a particular type of river sand or silt in terms of their physical and chemical characteristics. The geochemistry, makeup, and mineralogy of the ore, as well as the procedures used to extract different commercial goods, all influence the specific features of tailings. Chemically, gold tailings are highly salinized, include just 6% pyrite, and contain very little organic stuff. According to sources, the pH of Iran's tailings was 7.35, that of South Africa's was 3.25-6.28, and that of India was 3.48-8.12 the [13]. Heavy metal soil pollution is caused in the regions with gold mining activities primarily due to the extraction of ores from soil and rocks and due to unscientific methods of dumping the leftover tailings around residential places. Heavy metals may endanger humans, animals, plants, and the ecosystem through various channels the [14, 15, 16]. These include direct consumption, plant absorption, food chains, ingesting polluted water, and changes to the pH, opacity, color, all of which affect soil quality the [17].

Even though heavy metals are only found in trace concentrations in water sources, they are exceedingly dangerous and pose serious health hazards to both people and other species the [18, 19]. When humans consume toxic metals in excess, it can cause serious stomach pain, extreme vomiting, and deteriorating liver abnormalities. Arsenic poisoning from drinking water has been linked to horrible long-term effects, such as learning issues, nervous system damage, and children's growth being stunted the [20]. Gradually the heavy metals present in soil tends to get washed off from tailings present in high altitude during monsoon season and seeps into groundwater, lakes, and pond water. The HMs which are present in pond water tend to invariably affect the metabolism and growth of the fish. Fishes



**Ashok Dhayalan et al.,**

containing impermissible levels of heavy metals when consumed by animals in higher tropic levels get affected resulting in various complications. The study attempted to demonstrate that heavy metal concentrations in *Channa punctatus* from sewage-fed aquaculture ponds in India constitute a health threat to locals. The majority of the fish samples examined contained dangerously high amounts of heavy metals, putting the local population at risk if they consumed too much fish [21]. Over the course of a century, various companies mined gold. Low gold production and unprofitability forced its closure in 2001 the [22]. Heavy metal pollution comes from mining. Mining contaminates soil, generating environmental hazards. Samples contained heavy metals in soil and water the [23]. Heavy metals can cause cancer, organ damage, stunted growth, and death the [24]. Heavy metal removal from soils requires remediation. He concluded that rainwater penetration contaminates groundwater, which contaminates the soil. Heavy metal harms plants, animals, and humans. Thus, it is vital to assess soil contamination and offer remedial actions to improve soil quality and reduce contamination.

In the majority of the world's accessible freshwater reservoirs, groundwater makes up 99 percent of the planet's liquid freshwater supply. Since it can be pumped, groundwater often becomes the main source of water in areas where there are no other permanent water supplies. Currently, groundwater is the source of over 50% of all drinking water, 40% of all agricultural water, and 35% of all industrial the [25]. The main contributors to heavy metal contamination have been Arsenic (As), chromium (Cr), mercury (Hg), and cadmium (Cd). The presence of these metals has caused a number of issues for plant life in the beautiful surroundings. In growing plants, cadmium causes leaf chlorosis, or inadequate chlorophyll. Plants that contain excessive levels of arsenic and mercury have experienced a decrease in photosynthesis and root development. Chromium is not biodegradable and significantly reduces the dry weight of seedlings. They enter the food chain and accumulate at various trophic levels, which have an adverse influence on the growth of plants the [26]. The immune system is altered by cadmium, and proliferative prostatic lesions and lung adenocarcinoma are more common. Overexposure to blood arsenic can lower a child's intellectual capacity, damage their endocrine, skeletal, and immune systems, and induce hypertension. In adults, it also impairs cardiac and renal function. Chromium has been connected to respiratory system tumors and cancer the [27]. Groundwater contamination from rainwater infiltration eventually contaminates soil the [22]. Heavy metal harms plants, animals, and humans. Thus, it is vital to assess pollution and offer remedial strategies to improve groundwater quality and reduce contamination. Six water samples from bore wells (BWs) were chosen at random from the mining and residential regions of KGF. To evaluate the presence of heavy metals in the KGF, samples of bore well (BW) water were taken all throughout the research area. Six samples of well water were examined in the lab. Heavy elements like copper (Cu), nickel (Ni), and arsenic (As) were discovered in the water during the preliminary research.

COLLECTION OF SOIL SAMPLES**General methodology**

During this research work, the general methodology was devised based on spatiotemporal factors like place and time. So, the four places selected based on the minimal distance from the abandoned gold mining sites were Oorgaam, Tenants, Champion, and Balghat and named A, B, C, and D which are located 50,100, 200 and 300 meters from the mining sites respectively.

The four sampling seasons were

Season 1-January, February, and March

Season 2-April, May, and June

Season 3- July, August, and September

Season 4-October, November, and December

Four sample locations were used for the sampling, which took place between January and December 2023 (gold ore tailings). Every month, one sample was taken from each location, resulting in four samples that were examined. The presence of certain heavy metals, such as Cd, Cr, and Pb was determined by sampling the area throughout the four distinct seasons. After removing contaminants from the surface, soil samples were collected from 10 to 20 cm below the surface and put in self-locking polythene bags. The sample preparation procedures for the spectrochemical



Ashok Dhayalan *et al.*,

determination of the total recoverable elements were finished in accordance with U.S. Environmental Protection Agency requirements the [28].

Sample preparation

To calculate the total recoverable analytes in soil samples, the sample was well mixed, a piece of it was placed on the tared weighing dish, the sample was weighed, and the weight was recorded in order to ascertain the total recoverable analytes in solid samples. The dry material was mashed in a mortar and pestle and sieved using a 5-mesh propylene sieve to obtain homogeneity. All of the acids and chemicals utilised in this research were ultra-purity grade, and Rankem Compounds provided the chemicals for measurement. Thermo Scientific's ICP-OES (Inductively Coupled Plasma- Optical Emission Spectroscopy) was used in conjunction with Iteva software to evaluate the samples.

Sample digestion and analysis

As reagents, Rankem chemicals' pure analytical grade acids with a 32 percent HCl content and a 70 percent HNO₃ content (both purchased from Sigma-Aldrich India) were employed. Concentrated HNO₃ and HCl acids were combined in a 1:3 ratio to create aqua-regia. In a 250 mL conical glass flask, one gramme of each reference material or dry powdered soil was combined with 28 mL of aqua-regia. The reactants were gently mixed in the flask before it was heated on a hot plate for five hours, reaching a temperature of 120°C. The dissolved samples were filtered into 100 mL HDPE bottles after cooling down using filter paper that had been wetted with 3 percent HNO₃ acid for ICP-OES analysis. All glassware used to prepare samples was cleaned before use by immersing it in an acid solution containing 10 percent v/v HNO₃ for 24 hours. This was done before giving the glassware a final rinse with deionized distilled water. Thermo Fischer, ICP-OES, and iCAP 6300 were used to perform chemical analyses at the ARML in Bangalore, India, and ITEVA software was used to interpret the results.

RESULTS AND DISCUSSION

Arsenic concentration in soil

Arsenic, as a heavy metal, is known for its stability. In both animals and humans, it acts as a neurotoxin. The soil may contain trace quantities of arsenic. One of the most pervasive and deadly forms of pollution in the world is arsenic (AS) pollution. The highest level of arsenic that can be present in the soil is 98 mg/kg (Table 1). In site (C) Champion, the amount of Arsenic (As) is 140 mg/kg, which is almost 1.2 times the upper limit set by the [29] as permissible limits. The highest average Arsenic content in the soil samples during January-December 2023 analyzed from Champion was 120.83 mg/kg, followed by 36.08 mg/kg at site A, 32.17 mg/kg at site D, and 28.00 mg/kg at site B. Tenants' soil contamination in season 3 was (19 mg/kg) as the lowest concentration through the campaign. When assessed season-wise in all sample sites the average concentrations season-wise was, season 4 at 60.45 mg/kg, followed by season 3 at 56.3 mg/kg, season 1 had 51.9 mg/kg, and the lowest amount observed during season 2 at 48.3 mg/kg.

In the graph 1, sites A (Oorgaum), B (Tenants), and D (Balghat) all have Arsenic levels in their soil that are below the legal limit. Only site C (Champion) shows higher Arsenic values in the soil sample than the permissible limits. The Arsenic concentrations in the soil samples are slightly rising from season to season. Initial concentrations tend to be lowest during season 1, with subsequent concentrations gradually rising throughout the year. According to Table 2, there is a positive and significant correlation between Arsenic pollution in the soil during seasons 1 (January–March) and 2 ($r = 1.00$, $p < 0.05$). There is also evidence of a statistically significant association concerning Arsenic pollution in soil, with values of ($r = 0.993$, $p < 0.05$) and ($r = 0.999$, $p < 0.05$), respectively, for Seasons 3 and 4.

Table 3. displays the results of a statistical analysis comparing arsenic concentrations in the soil across different seasons. The results show that there is a statistically significant difference between the arsenic concentrations in the soil between seasons 1 (January–March) and 4 (October–December) with a p-value of 0.003. We cannot statistically



**Ashok Dhayalan et al.,**

report a difference in arsenic levels between seasons 1 (January–March) and 2 (April–June) and seasons 1 (January–March) and 3 (July–September) since the p-value is greater than 0.05.

Effect of gold ore tailings on the groundwater during January–December 2023**Assessment of pH in bore well water**

An indicator of many geochemical equilibrium or solubility calculations, pH is widely recognized as a critical ecological component. Most aquatic creatures have adapted to a specific pH level and cannot survive if that level suddenly changes, making pH a crucial element in any body of water.

The limit pH value for drinking water is specified as 6.5 to 8.5 (table-4). Sampling site Balghat has the lowest pH value i.e., 6.6 and Champion's pH value is the highest among all the sample sites (7.4) followed by Tenants and Oorgaum with the pH values 7.1 and 6.8 respectively.

Assessment of Hardness in groundwater

The levels of calcium and magnesium, and to a lesser extent iron, are what determine the hardness of water. Water hardness is expressed as the amount of calcium carbonate (CaCO_3) in milligrams per liter (mg/L) by adding the calcium and magnesium concentrations together. The weathering of limestone, sedimentary rock, and calcium-bearing minerals gives most groundwater its hardness. Chemical and mining wastewater, as well as the overuse of lime as a soil amendment in agricultural regions, can all contribute locally to groundwater hardness. The optimum range of hardness in drinking water is 250 ppm to 660 ppm (table-5). All the other sampling site's borewell water's hardness is much higher than the permissible limits. The highest water hardness is seen in the Oorgaum borewell water sample, followed by sites D, C, and B (Graph-2).

Arsenic concentration in groundwater

The environment contains naturally occurring arsenic, a hazardous element. Often used in household items, although its content in the environment may have been raised by human activity; it is released into the atmosphere through vehicle exhaust. Pipe corrosion is a major source of this substance in drinking water. Children under the age of six are particularly vulnerable to the adverse health effects of arsenic exposure, even from relatively modest exposure levels. hemoglobin production is disrupted; blood pressure is raised; kidneys are damaged. In table-6, the highest level of arsenic that can be present in groundwater is 0.05 mg/L. Sampling site C Champion is highly contaminated with arsenic. In Champion, the content is 0.863 mg/L, which is much higher than the upper limit set by the WHO (2004) for safe consumption. The average arsenic content in the groundwater samples taken from Champion was 0.559 mg/L, followed by 0.323 mg/L at site B, 0.277 mg/L at site A, and 0.161 mg/L at site D. Balghat's groundwater contamination in season 3 and 4 (0.155 mg/L) was lowest.

Sites A (Oorgaum), C, and D (all shown on the graph 3) all have arsenic levels in their groundwater that are above the legal limit. Only site B shows constant arsenic values in the groundwater sample. The arsenic concentrations in the groundwater samples are slightly rising from season to season. Initial concentrations tend to be lowest during season 1, with subsequent concentrations gradually rising throughout the year. According to Table 8, there is a positive and significant correlation between arsenic pollution in groundwater during seasons 1 (January–March) and 2 ($r = 1.00$, $p < 0.05$). There is also evidence of a statistically significant association concerning arsenic pollution in groundwater, with values of ($r = 0.972$, $p < 0.05$) and ($r = 0.983$, $p < 0.05$), respectively, for Seasons 3 and 4.

In table 9, there was a statistically non-significant average difference between arsenic levels in the groundwater during season1 (Jan-Mar) & season 2 (Apr-Jun), season 1 (Jan-Mar) & season 3 (Jul-Sept) and season 1 (Jan-Mar) & season 4 (Oct-Dec) since the p-value > 0.05 .



Ashok Dhayalan *et al.*,

CONCLUSSION

The permissible pH value for drinking water is specified as 6.5 to 8.5. Sampling site Balghat has the lowest pH value i.e. 6.6 and Champion's pH value is the highest among all the sample sites (7.4) followed by Tenants and Oorgaum with the pH values 7.1 and 6.8 respectively. In a similar study in Singapore, the [30], the pH value of water was observed as 7.8 ± 0.4 . In a similar study in Kuwait, the pH value of water was observed in the range 7.33–7.45, justifying our results. The highest EC in borewell water is observed in Champion (2110 $\mu\text{S}/\text{cm}$), followed by Tenants, Oorgaum, and Balghat with the values 1900 $\mu\text{S}/\text{cm}$, 1850 $\mu\text{S}/\text{cm}$, and 1810 $\mu\text{S}/\text{cm}$ respectively. In a similar study in Palestine the [31] demonstrated the EC value of water was observed in the range of 473–1406, justifying our obtained results. In contrast, a study in Wondo Genet, the [32], reported the EC value of water was observed as 192.14 $\mu\text{S}/\text{cm}$. All the other sampling site's borewell water's hardness is much higher than the permissible limits. Hardness is almost 2 times higher than the control water hardness. The highest water hardness is seen in the Oorgaum borewell water sample, followed by sites D, C, and B. In a similar study in Europe, the [33]. reported the hardness value of water was observed as 60 ppm. In a similar study in Sri Lanka, the [34]. reported the hardness of groundwater as 385 ppm.

When assessed for arsenic in soil, sample site (C) Champion, contained 140 mg/kg of Pb, which is almost 1.2 times the upper limit set by the WHO (2008) as permissible limits. The highest average arsenic content in the soil samples during January-December 2023 analyzed from Champion was 120.83 mg/kg, followed by 36.08 mg/kg at site A, 32.17 mg/kg at site D, and 28.00 mg/kg at site B. Tenants' soil contamination in season 3 was (19 mg/kg) as the lowest concentration through the campaign. A similar study done in Karnataka the [35] found the arsenic content in soils from 30 mg kg⁻¹ in Kiradalli Tanda village soil. Sampling site C Champion is highly contaminated with arsenic. In Champion, the content is 0.863 mg/L, which is much higher than the upper limit set by the [29].for safe consumption. The average arsenic content in the groundwater samples taken from Champion was 0.559 mg/L, followed by 0.323 mg/L at site B, 0.277 mg/L at site A, and 0.161 mg/L at site D. Balghat's groundwater contamination in season 3 and 4 (0.155 mg/L) was lowest. In a similar study in Taiwan, it was found that the arsenic values were found in the range 0.001-0.019 mg/L. the [36]. validated that in Iran, the arsenic concentration was found to be in the range of 1-4.23 mg/L.

CONFLICTS OF INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

REFERENCES

1. Sarkodie, S. A., Owusu, P. A., & Leirvik, T. (2020). Global effect of urban sprawl, industrialization, trade and economic development on carbon dioxide emissions. *Environmental Research Letters*, 15(3), 034049. DOI 10.1088/1748-9326/ab7640.
2. Raihan, A. (2023). The dynamic nexus between economic growth, renewable energy use, urbanization, industrialization, tourism, agricultural productivity, forest area, and carbon dioxide emissions in the Philippines. *Energy Nexus*, 9, 100180. <https://doi.org/10.1016/j.nexus.2023.100180>.
3. Owen, J. R., Kemp, D., & Marais, L. (2021). The cost of mining benefits: Localising the resource curse hypothesis. *Resources Policy*, 74, 102289. <https://doi.org/10.1016/j.resourpol.2021.102289>.
4. Ajay Singh, A review of wastewater irrigation: Environmental implications, *Resources, Conservation and Recycling*, 2021, 168,105454, ISSN 0921-3449, <https://doi.org/10.1016/j.resconrec.2021.105454>.
5. Subbaraman J.V. (2006) Scope for geobotanical prospecting for gold in Karnataka and Andhra Pradesh, *Current Science*, 90, 750.





Ashok Dhayalan et al.,

6. Fashola, M. O., Ngole-Jeme, V. M., & Babalola, O. O. (2016). Heavy metal pollution from gold mines: environmental effects and bacterial strategies for resistance. *International journal of environmental research and public health*, 13(11), 1047. <https://doi.org/10.3390/ijerph13111047>.
7. Minnaar, A. (2020). Water pollution and contamination from gold mines: acid mine drainage in Gauteng Province, South Africa. *Water, governance, and crime issues*, 193-219.
8. Franks, D. M., Boger, D. V., Côte, C. M., & Mulligan, D. R. (2011). Sustainable development principles for the disposal of mining and mineral processing wastes. *Resources policy*, 36(2), 114-122.
9. Bhattacharya, A., Routh, J., Jacks, G., Bhattacharya, P., & Mörth, M. (2006). Environmental assessment of abandoned mine tailings in Adak, Västerbotten district (northern Sweden). *Applied Geochemistry*, 21(10), 1760-1780. <https://doi.org/10.1016/j.apgeochem.2006.06.011>.
10. Schoenberger, E. (2016). Environmentally sustainable mining: The case of tailings storage facilities. *Resources Policy*, 49, 119-128. <https://doi.org/10.1016/j.resourpol.2016.04.009>.
11. Singh, R., Gautam, N., Mishra, A., & Gupta, R. (2011). Heavy metals and living systems: An overview. *Indian journal of pharmacology*, 43(3), 246. DOI: 10.4103/0253-7613.81505.
12. Bambas-Nolen, L., Birn, A. E., Cairncross, E., Kisting, S., Lieferrink, M., Mukhopadhyay, B., & Shroff, F. M. (2018). Case study on extractive industries prepared for the lancet commission on global governance. UiO: Institute of Health and Society. [Electronic Resource]. URL: <https://www.med.rio.no/helsam/english/research/centres/global-health/global-governancehealth/background-papers/extrac-indus.pdf> (date of access: 16.12.2020).
13. Okereafor, U., Makhatha, M., Mekuto, L., Uche-Okereafor, N., Sebola, T., & Mavumengwana, V. (2020). Toxic metal implications on agricultural soils, plants, animals, aquatic life and human health. *International journal of environmental research and public health*, 17(7), 2204. <https://doi.org/10.3390/ijerph17072204>.
14. Ye, S., Zeng, G., Wu, H., Liang, J., Zhang, C., Dai, J., ... & Yu, J. (2019). The effects of activated biochar addition on remediation efficiency of co-composting with contaminated wetland soil. *Resources, Conservation and Recycling*, 140, 278-285. <https://doi.org/10.1016/j.resconrec.2018.10.004>.
15. Wang, W., Chang, J. S., & Lee, D. J. (2024). Machine learning applications for biochar studies: A mini-review. *Bioresource Technology*, 130291. <https://doi.org/10.1016/j.biortech.2023.130291>.
16. Pehlivan, N., Gedik, K., & Wang, J. J. (2023). Tea-based biochar-mediated changes in cation diffusion homeostasis in rice grown in heavy metal (loid) contaminated mining soil. *Plant Physiology and Biochemistry*, 201, 107889. <https://doi.org/10.1016/j.plaphy.2023.107889>.
17. Masindi, V., & Muedi, K. L. (2018). Environmental contamination by heavy metals. *Heavy metals*, 10, 115-132.
18. Budi, H. S., Catalan Opulencia, M. J., Afra, A., Abdelbasset, W. K., Abdullaev, D., Majdi, A., ... & Mohammadi, M. J. (2024). Source, toxicity and carcinogenic health risk assessment of heavy metals. *Reviews on Environmental Health*, 39(1), 77-90. <https://doi.org/10.1515/reveh-2022-0096>.
19. Alalwan, H.A., Kadhom, M.A., Alminshid, A.H. 2020. Removal of heavy metals from wastewater using agricultural byproducts. *Journal of Water Supply: Research and Technology-Aqua*, 69(2), 99-112. <https://doi.org/10.2166/aqua.2020.133>.
20. Basu, A., Phipps, S., Long, R., Essegbey, G., & Basu, N. (2015). Identification of response options to artisanal and small-scale gold mining (ASGM) in Ghana via the Delphi process. *International journal of Environmental research and public health*, 12(9), 11345-11363.
21. Javed, M. (2012). Uptake of Heavy Metals by *Channa Punctatus* from Sewage Fed Aquaculture Pond of Panethi, Aligarh. *Global Journals of Research in Engineering*, 12(C2), 27-34.
22. Adarsh, S., Prakash, S., & Padiwal, D. S. (2017) Monitoring the presence of heavy metals in kgf soil residue for bio-remediation. *International journal of engineering sciences and management*, 7(1), pp.16-22.
23. Li, Z., Ma, Z., van der Kuijp, T. J., Yuan, Z., & Huang, L. (2014). A review of soil heavy metal pollution from mines in China: pollution and health risk assessment. *Science of the total environment*, 468, 843-853. <https://doi.org/10.1016/j.scitotenv.2013.08.090>.





Ashok Dhayalan et al.,

24. Fu, Z., & Xi, S. (2020). The effects of heavy metals on human metabolism. *Toxicology mechanisms and methods*, 30(3), 167-176. <https://doi.org/10.1080/15376516.2019.1701594>.

25. Price, M., & Back, W. (2013). *Introducing groundwater*. Routledge.

26. Nivetha A, Sakthivel C, Prabha I. Heavy Metal Contamination in Groundwater and Impact on Plant and Human. In: Shit, P.K., Adhikary, P.P., Sengupta, D. (eds) *Spatial Modeling and Assessment of Environmental Contaminants. Environmental Challenges and Solutions*. 2021, 233–246. Springer, Cham. https://doi.org/10.1007/978-3-030-63422-3_14.

27. Mawari G, Kumar N, Sarkar S, Sayan Sarkar, Arthur L Frank, Mradul Kumar Daga, Mongjam Meghachandra Singh, Tushar Kant Joshi and Ishwar Singh. Human Health Risk Assessment due to Heavy Metals in Ground and Surface Water and Association of Diseases With Drinking Water Sources: A Study From Maharashtra, India. *Environ Health Insights*. 2022;16:11786302221146020. doi:10.1177/11786302221146020.

28. US-EPA.(2009),United States Environmental Protection Agency. National recommended water quality criteria. Washington DC, USA. [Online]. <http://water.epa.gov/scitech/swguidance/standards/criteria/current/>.

29. WHO. (2008), Guidelines for drinking-water quality. Vol. 1: Recommendations. 3rd ed. World Health Organization, Geneva, Switzerland;. [Online]. https://www.who.int/water_sanitation_health/publications/drinking-water-quality-guidelines-4-including-1staddendum/en/. Accessed July 14, 2019.

30. Ng, D. Q., & Lin, Y. P. (2015). Effects of pH value, chloride and sulfate concentrations on galvanic corrosion between lead and copper in drinking water. *Environmental Chemistry*, 13(4), 602-610. <https://doi.org/10.1071/EN15156>.

31. Daghara, Azza, Issam A. Al-Khatib, and Maher Al-Jabari. "Quality of drinking water from springs in palestine: West bank as a case study." *Journal of Environmental and Public Health* 2019 (2019). <https://doi.org/10.1155/2019/8631732>.

32. Meride, Y., & Ayenew, B. (2016). Drinking water quality assessment and its effects on residents health in Wondo genet campus, Ethiopia. *Environmental Systems Research*, 5(1), 1-7. <https://doi.org/10.1186/s40068-016-0053-6>.

33. Kozisek, Frantisek. "Regulations for calcium, magnesium or hardness in drinking water in the European Union member states." *Regulatory Toxicology and Pharmacology* 112 (2020): 104589. <https://doi.org/10.1016/j.yrtph.2020.104589>.

34. Wasana, H., Perera, G. D., Gunawardena, P. D. S., Fernando, P. S., & Bandara, J. (2017). WHO water quality standards Vs Synergic effect (s) of fluoride, heavy metals and hardness in drinking water on kidney tissues. *Scientific Reports*, 7(1), 1-6. <https://doi.org/10.1038/srep42516>.

35. Chakraborti, D., Rahman, M. M., Murrill, M., Das, R., Patil, S. G., Sarkar, A., ... & Das, K. K. (2013). Environmental arsenic contamination and its health effects in a historic gold mining area of the Mangalur greenstone belt of Northeastern Karnataka, India. *Journal of hazardous materials*, 262, 1048-1055. <https://doi.org/10.1016/j.jhazmat.2012.10.002>.

36. Pazand, K., Khosravi, D., Ghaderi, M. R., & Rezvanianzadeh, M. R. (2018). Hydrogeochemistry and lead contamination of groundwater in the north part of Esfahan province, Iran. *Journal of water and health*, 16(4), 622-634. <https://doi.org/10.2166/wh.2018.034>.

Table 1: Assessment of Arsenic in the soil (mg/kg) in various sampling sites during January-December 2023

SAMPLING SITE	MAXIMUM PERMISSIBLE LIMIT	HIGHEST	LOWEST	AVERAGE
OORGAUM (A)	98	44	28	36.08
TENANTS (B)		34	19	28.00
CHAMPION (C)		140	88	120.83
BALGHAT (D)		44	21	32.17





Ashok Dhayalan et al.,

Table 2: Correlation of Arsenic concentration in Soil in various seasons

Seasons during 2023	Correlation	Sig.
1 Season 1 & 2	1.000	.000
2 Season 1 & 3	.993	.007
3 Season 1 & 4	.999	.001

Table 3: Comparison of Arsenic concentration in Soil in various seasons

Seasons	Paired Differences					t	df	P-value
	Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
				Lower	Upper			
1-2	3.58333	14.72306	7.36153	-19.84434	27.01101	.487	3	.660
1-3	-4.41667	6.08809	3.04404	-14.10417	5.27084	-1.451	3	.243
1-4	-8.58333	1.91243	.95622	-11.62644	-5.54022	-8.976	3	.003

Table 4: Average concentration of pH in groundwater in sampling sites 2023

Sites	pH
Max permissible limit	6.5-8.5
Oorgaum	6.8
Tenants	7.1
Champion	7.4
Balghat	6.6

Table 5: Average concentration of Hardness in groundwater in various sampling sites in 2023

Sampling Sites	Hardness in ppm
Max permissible limit	250-660
Oorgaum	780
Tenants	690
Champion	710
Balghat	730

Table 6: Concentration of Arsenic in groundwater in mg/L in sampling sites during January-December 2023

SAMPLING SITE	MAXIMUM PERMISSIBLE LIMIT	HIGHEST	LOWEST	AVERAGE
OORGAUM (A)	0.01	0.015	0.013	0.014
TENANTS (B)		0.005	0.003	0.004
CHAMPION (C)		0.349	0.157	0.228
BALGHAT (D)		0.070	0.050	0.060

Table 7: Concentration of arsenic in groundwater in mg/L in sampling sites during January-December 2023

SAMPLING SITE	MAXIMUM PERMISSIBLE LIMIT	HIGHEST	LOWEST	AVERAGE
OORGAUM (A)	0.05	0.283	0.271	0.277
TENANTS (B)	0.05	0.326	0.320	0.323
CHAMPION (C)	0.05	0.863	0.460	0.559
BALGHAT (D)	0.05	0.168	0.155	0.161





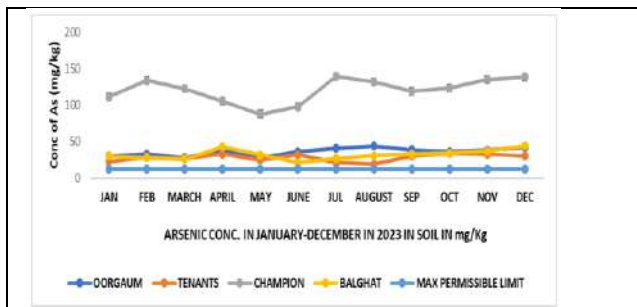
Ashok Dhayalan et al.,

Table 8: Correlation of arsenic concentration in groundwater in various seasons

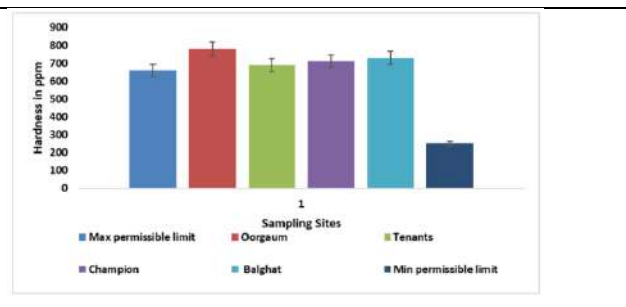
Seasons during 2014		Correlation	Sig.
1	Season 1 & Season 2	1.000	.000
2	Season 1 & Season 3	.972	.028
3	Season 1 & Season 4	.983	.017

Table 9: Comparison of arsenic concentration in Groundwater in various Seasons

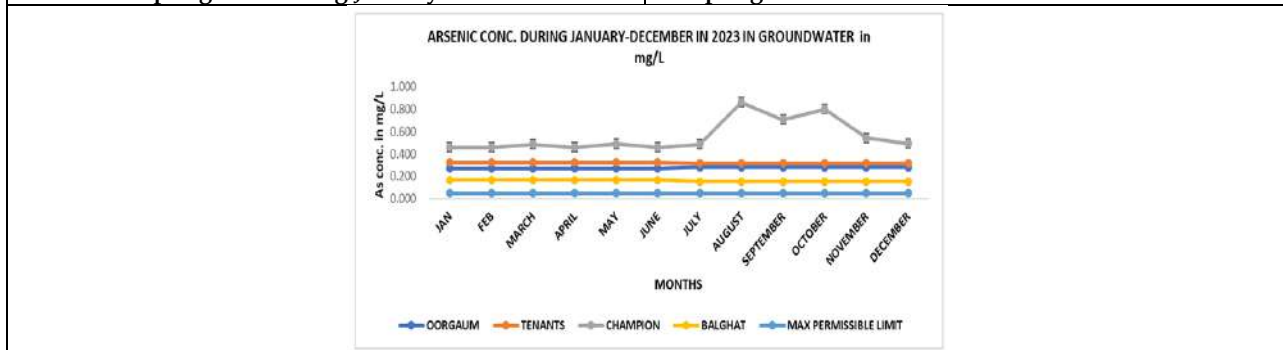
Seasons	Paired Differences					t	df	P-value
	Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference				
				Lower	Upper			
1 – 2	-.000208	.000417	.000208	-.000871	.000455	-1.000	3	.391
1 – 3	-.052125	.109947	.054974	-.227075	.122825	-.948	3	.413
1 - 4	-.034000	.073957	.036979	-.151683	.083683	-.919	3	.426



Graph 1: Assessment of Arsenic in the soil (mg/kg) in various sampling sites during January-December 2023



Graph 2: Average Hardness in groundwater in sampling sites in 2023.



Graph 3: Concentration of arsenic in groundwater in mg/L in sampling sites during January-December 2023





Analysis of Virtually Shopping Customer Employing RFM Technique and Clustered using Machine Learning Technique

Nilam N. Parmar^{1*} and Sweta S. Panchal²

¹Assistant Professor, Computer Science and Engineering, Dr. Subhash University, Gujarat, India.

²Associate Professor, Electronics and Communication, Dr. Subhash University, Gujarat, India.

Received: 03 May 2024

Revised: 10 Aug 2024

Accepted: 18 Oct 2024

*Address for Correspondence

Nilam N. Parmar

Assistant Professor, Computer Science and Engineering,

Dr. Subhash University,

Gujarat, India.

E.Mail: nilam.p2718@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Enhancing data gathering and leveraging the acquired data to derive valuable insights is a fundamental responsibility within the domain of customer segmentation. In this study, all relevant criteria are employed to effectively extract information from the available data. To attain the desired results, many techniques are employed, including the utilization of k-means clustering to categorize clients into clusters based on their shared tastes and requirements. The concept of clustering refers to the process of grouping similar data points together based on their inherent the identification of customers that are unlikely to contribute to the business's growth can be achieved by employing the K-means algorithm. Subsequently, the effectiveness of these customer classifications can be assessed through the application of silhouette analysis. The data will undergo normalization using the min-max normalization approach. The main aim of normalization is to determine the likelihood that a specific score will fall within the normal distribution of the dataset. Simultaneously, the process of clustering labeled data will be carried out alongside the clustering of unlabeled data, utilizing the same clustering approach. The data points will be categorized according to the degree of similarity in their feature structures. The application of Silhouette Analysis, a technique that measures the distances between points inside a cluster and facilitates the generation of visual representations for classifying different types of data, will be employed to complete the data analysis. The present technique quantifies the spatial separation among different sites within a given cluster. Furthermore, the RFM methodology was employed to ascertain the worth of individual consumers by analyzing their past purchasing patterns.

Keywords: Customer segmentation, K-means Clustering, RFM Analysis, min-max normalization approach, Silhouette Analysis



**Nilam N. Parmar and Sweta S. Panchal**

INTRODUCTION

The market has witnessed an influx of new enterprises and entrepreneurs, hence intensifying the competition faced by established organizations as they strive to effectively engage and keep clients. According to Bardaki (2018)[1], the preceding development has rendered it suitable for firms of varying scales to prioritize the provision of outstanding customer service. Furthermore, the ability of a corporation to understand the different needs of its consumers and develop tailored approaches to address those needs will lead to enhanced overall customer service. The implementation of a structured client service framework facilitates the acquisition of such valuable insights. Customers in each subgroup share market characteristics (Premkanth, 2012)[2]. Traditional market analytics sometimes struggle to effectively handle a large customer base. However, the emergence of big data concepts and machine learning has led to the use of automated consumer segmentation approaches. In this paper, the utilization of the k-means clustering algorithm is employed (Goyat, 2011)[3].

This research study encompasses a series of sequential steps in its methodology. Furthermore, during the initial phase, the existing data inside the dataset will undergo pre-processing utilizing the Knowledge Discovery technique, with the aim of extracting pertinent information for training purposes through the utilization of a machine learning algorithm. The subsequent procedure involves the implementation of the Min-Max normalization approach. The basic objective of normalization is to determine the probability of a score being within the normal distribution of the data. Simultaneously, the clustering technique is employed on the unlabeled data in order to ascertain groupings within the dataset. The data is categorized into clusters based on their degree of similarity in relation to a specific attribute. In the ultimate phase of the system, Silhouette Analysis will be employed to assess the degree of segregation among data points within a solitary cluster, so enabling the generation of a visual depiction of the model's classification.

CUSTOMER CLUSTERING

Companies have had to grow their profitability and company through time as a result of fierce competition in the business field to meet customer expectations and attract new clients depending on their desires. It's tough and time-consuming to identify and respond to each customer's needs. This is owing to the fact that, among other things, clients have a diverse set of aims, interests, and preferences. Customer segmentation, as opposed to a "one-size-fits-all" strategy, divides customers into groups based on comparable characteristics or habits. Customer segmentation is a marketing strategy that divides a market into distinct, homogeneous groups. The data used in the customer segmentation strategy, which divides customers into categories, is based on a number of factors, including regional circumstances, economic patterns, and demographic trends, and behavioral patterns. A client segmentation technique can help a company's marketing resources be better utilized (Hemashree Kilari)(2022)[4].

Managing customers and identifying their likes and dislikes plays a vital role in market business. It has been observed that companies face losses because they are not able to identify the potential customers that will bring them profit. One of the many reasons of these losses are that the companies are using mass marketing tactics that are since whatever we are selling would be liked by everyone. These tactics are time-consuming, expensive, and even proved non-profitable. These strategies are ineffective since every consumer is unique, necessitating the use of some form of algorithm or practice to categories them based on the similarity of their preferences and direct our attention to those groups. To find hidden patterns in data and make future decisions that will be more effective, machine learning is utilized. The hazy idea of which section to target is made clear by the implementation of segmentation. Customer segmentation is the practice of classifying consumers into groups based on similar behavioral patterns and customers into distinct groups based on different behavioral patterns. Suppose a brand focuses on all the customers that are visiting their website but some of them are just browsing their site without intending to buy anything, such people make it difficult for the seller to sell his product as he is targeting everyone. In these types of situation customer segmentation is used (Garima Sharma) (2021)[5].





MACHINE LEARNING

We've seen machine learning in action in a variety of businesses, like Facebook, where it helps us identify ourselves and our friends, and YouTube, where it helps us discover new content, where it recommends movies based on our preferences. Machine learning is divided into two types: unsupervised learning and supervised learning. A data analyst often employs supervised learning to address problems like classification and regression, implying that the data in this case is targetable and that we want to anticipate in the future, such as assessing a student's worth or the amount of monthly costs. Unsupervised learning, on the other hand, may or may not have a label or goal in mind. Because it is based on a mathematical model, clustering, for example, does not have a changeable goal. For instance, we might want to group students depending on their learning interests. or product purchases. Strong competition exists in the marketing business, particularly malls, in order to boost consumer numbers and so produce big profits. Many retailers and other marketplaces are already using machine learning to achieve this goal. Malls and shopping centres use the information they collect from customers to construct machine learning models that target the right individuals. This not only boosts revenue and visitor numbers, but it also increases business efficiency [4].

K-means clustering is an unsupervised machine learning approach that segments a dataset into K groups based on shared features. Applying min-max scaling, which converts all the integers to a range from 0 to 1, is one way to normalize a dataset. The clustering precision of the K-means algorithm has been studied in the past, with notable studies including: Mohamad (2013)[6] looked on how normalizing data impacted the accuracy of K-means clustering. In a study comparing K-means' performance on normalized and unnormalized datasets, min-max scaling was found to significantly improve the clustering algorithm's accuracy. Mukhametzyanov (2023)[7] looked into the effect that various normalization techniques had on the accuracy of K-means clustering. Min-max scaling, Z-score normalization, and decimal scaling were all tested to see which would improve K-means' clustering performance the best, and min-max scaling was shown to be the most effective. What factors should be considered while settling on the optimal number of clusters?

Dendrograms are a useful tool for representing the results of a hierarchical clustering investigation. They show how the various data clusters are related to one another and how the data was organized. To cite Forina (2002)[8]. The optimal number of clusters can be identified by analyzing the node distances displayed in the dendrogram. Finding the longest horizontal line that does not intersect any existing clusters is a frequent approach to deciding on the optimal number of clusters. This distance, often known as the "elbow" or "knee" point, measures the optimum number of clusters for the data collection. To help businesses better comprehend their data and make informed decisions about consumer segmentation, they can use dendrograms to visualize the results of hierarchical clustering. The accuracy of the segmentation model is highly correlated with the number of clusters used in the analysis. There is a danger of oversimplification in segmentation with too few clusters, and of incomprehensibility un segmentation with too many. Silhouette analysis can be used to determine the optimal number of clusters by maximizing similarity within each group and minimizing similarity between groups. Determining the optimal number of clusters can help improve the segmentation model's accuracy and performance. According to Bell (2007)[9], increasing the number of clusters can result in less accurate segmentation models due to the curse of dimensionality. As the number of variables or dimensions in a dataset increases, the complexity of detecting meaningful patterns or clusters also increases. Another study by (Jain, 1988)[10] demonstrated that excessive use of clusters—or "over-segmentation"—can reduce the accuracy of the segmentation model. This occurs when the size of the clusters is too small to give meaningful subsets of the data. However, the accuracy of the segmentation model may degrade if too few clusters are used (under-segmentation). If the clusters are too large, they won't be able to identify smaller variations in the data.

RFM analysis with K-means clustering with hyperparameters provide a more accurate and data-driven technique for consumer segmentation. RFM analysis takes into account the recency, frequency, and monetary value of client purchases, allowing for a deeper understanding of customer behaviors and preferences. K-means clustering with hyperparameters automates the segmentation process, doing away with the requirement for labor-intensive and prone-to-error manual segmentation. A more accurate and efficient technique of customer segmentation is



**Nilam N. Parmar and Sweta S. Panchal**

accomplished by integrating RFM analysis with K-means clustering with hyperparameters. Gupta (2006) [11] compared the efficiency of RFM analysis to that of more conventional approaches like demographic segmentation and behavioral segmentation in predicting client retention and loyalty. The research found that RFM analysis is an effective method for consumer segmentation because it outperformed more traditional methods in gauging client loyalty and retention. Traditional versus hyper parameterized K-means clustering: According to studies (Chu, 2021)[12], K-means clustering with hyperparameters outperforms regular K-means clustering when used to the task of customer segmentation. Hyper parameterized K-means clustering outperformed baseline methods in both effectiveness and stability. The optimal number of clusters was found to be determined by using hyper parameter tuning procedures like the elbow approach and silhouette analysis. Shalabi (2006)[13] A new range of values is found from the old ones by the scaling, mapping, or pre-processing technique known as normalization. Forecasting and prediction can both be done with this strategy. (PATRO, 2006)[14] There are several ways to predict or anticipate the similarities, but each strategy is unique based on the data it receives as input and how it interprets the information. To preserve the large range of variation, normalization techniques must be used to bring the anticipated outcomes closer together.

In order to better understand the need of normalization, let us examine an example. For instance, the dataset for height and weight may contain a range of measurement units, such as meters, inches, and grams. The findings of the data processing were skewed due to the large range of units. Data must be standardized or normalized in order to remove dependency on multiple measuring units and achieve a single measurement unit. The process of normalization involves reducing a large range of data to a single range, usually between -1 and 1, depending on the weight of the data. Among the methods used to normalize data are decimal scaling, min-max normalization, and Z-score normalization. I looked into each strategy and tried to determine its use and limitations for a long period.

PROPOSED WORK

The process is divided into several discrete steps. Furthermore, as part of the initial phase, the available data inside the dataset will undergo preprocessing using the Knowledge Discovery technique. Subsequently, a machine learning algorithm will be employed to extract the significant data for training purposes. The subsequent stage involves implementing the Min-Max normalization technique. The primary aim of the normalization procedure is to assess the probability of a score being situated within the normal distribution of the dataset. During this process, the clustering technique is employed to analyze the unlabeled data and identify clusters within the dataset. Within this section, the data is categorized into several groups according to the degree of similarity they possess in relation to a specific characteristic. During the final stage of the system, Silhouette Analysis will be employed to assess the proximity of data points within a given cluster. This process enables the creation of a graphical representation that accurately reflects the categorization output produced by the model.

RFM ANALYSIS FOR THE ANALYSIS OF DATA:

The RFM (recency, frequency, and monetary) model, proposed by Hughes (1994)[15], is a behaviour-based model that enables the prediction of customers' purchasing patterns. The concept of recency pertains to the temporal proximity of a buy event, while frequency denotes the quantification of purchase occurrences within a specified temporal interval. Monetary, on the other hand, encompasses the aggregate expenditure incurred within the same temporal framework (Wang, 2022)[16]. The utilization of customers' perceptions of the product, brand, perks, and loyalty as a segmentation variable can be facilitated through the aforementioned three behavioural characteristics.

Use of the RFM Model

Organizations that adopt the RFM (Recency, Frequency, Monetary) framework observe enhanced response rates, reduced order costs, and increased profitability. In order to apply the RFM model, it is necessary to associate each customer's name and address with a unique identifier, such as an account number, and link it to their respective orders. Additionally, it is crucial to record each transaction in conjunction with the corresponding unique identifier (Hughes, 1994)[15]. In order to ascertain significant and esteemed clients, RFM analysis examines the transaction





Nilam N. Parmar and Sweta S. Panchal

history by considering factors such as the timing of purchases, frequency of purchases, and monetary expenditure (Miglautsch, 2002)[17]. The RFM methodology facilitates the categorization of consumers into different segments. The consumer base was categorized into four distinct segments according to their varying degrees of uncertainty: spenders, frequent customers, top customers, and spenders.

MIN-MAX SCALING

Normalization can be conceptualized as a technique for scaling, mapping, or pre-processing data. One potential strategy for broadening the scope of the search is to leverage the existing coverage area. Undoubtedly, the utilization of this tool holds significant potential in facilitating the formulation of future projections or predictions. There exist multiple methodologies for generating predictions and forecasts, which may yield disparate outcomes. Normalization is a technique employed to reconcile divergent predictions and forecasts, as noted by Ali (2022)[18]. The Min-Max Normalization technique involves applying a linear modification to the original range of data. The Min-Mix Normalization technique is a strategy that aims to maintain the inherent relationships present in the original dataset. The utilization of Min-Max normalization is a straightforward approach that effectively scales the data to conform inside a predetermined range (Ali, 2022)[18].

According to the Min-Max method of normalizing

$$A' = \left(\frac{A - \text{min value of } A}{\text{max value of } A - \text{min value of } A} \right) \times (D - C) + C$$

Where,

A' contains Min-Max Normalized data one

If pre define boundary is [C, D]

If A is the range of original data

& B is the mapped one data then, (1)

Formulation of Min-Max Normalization (PATRO, 2015)[19]

Normalization, also known as min-max scaling, is a technique utilized to transform numerical data into a standardized range between 0 and 1. When employed alongside k-means clustering, it assists in the process of customer segmentation by enabling more impartial evaluations of features across various units and scales. This literature review investigates the advantages and disadvantages associated with the utilization of min-max scaling in k-means consumer segmentation.

The utilization of dendrograms for cluster representation:

A dendrogram is a visual depiction of the outcomes derived from an examination of hierarchical clustering. Chehreghani (2020)[20] asserts that it illustrates the interconnectedness of entities based on the degree of similarity or dissimilarity in their attributes. A dendrogram is a graphical representation that depicts groupings as branches and individual objects as leaves inside a tree-like structure. The initial stage in constructing a dendrogram involves the generation of a distance matrix that quantifies the dissimilarity between each item within the collection. The distance matrix exhibits the proximity or similarity between each pair of elements. Various distance metrics, such as the Euclidean distance or the cosine similarity, might be employed for this objective. The user's text does not contain any information to rewrite. Next, the objects are clustered using the hierarchical clustering technique, which is applied to the distance matrix. The technique begins by considering each object as an own cluster, and subsequently combines them iteratively until only a single cluster remains. The algorithms utilized in hierarchical clustering can be classified into two main categories: agglomerative and divisive.

The determination of the optimal number of clusters for subsequent analysis can be achieved by employing a dendrogram, which provides a graphical depiction of the hierarchical organization of the data. The process of selecting a specific height threshold on the dendrogram and subsequently dividing the tree at that level results in the



**Nilam N. Parmar and Sweta S. Panchal**

determination of the most suitable number of clusters. Subsequently, the created groups are utilized in subsequent investigations or depictions.

The process of interpreting dendrograms

The vertical axis of the dendrogram represents the distance or height between groupings. The degree of fusion or merging directly correlates with the dissimilarity of the clusters. The dendrogram's horizontal axis reveals the presence of clusters, whether they consist of objects or individuals. The positioning of the objects in the lower section of the dendrogram can either be assigned randomly or reflect the initial organization of the data.

Identify the points of intersection or fusion of branches within the dendrogram. These dots show clusters that exhibit varied degrees of resemblance. To determine the number of clusters, it is necessary to establish a height threshold on the dendrogram and thereafter partition the tree at that specific level. The clusters that emerge from the application of this criterion to the dataset are the assemblages of items that exhibit the highest degree of similarity to one another. Verifying k-means clustering findings: k-means clustering results can be verified with a dendrogram by comparing the dendrogram's structure to the clusters produced by the algorithm. The validity of the clustering results is further supported if the clusters produced by k-means agree with the dendrogram's structure (Nindhia, 2015)[21].

K-MEANS CLUSTERING

K-means clustering is a widely used unsupervised machine learning technique that involves the grouping or clustering of data points according to their similarity. According to Yong (2010)[22], this approach is extensively employed in the fields of data mining, market analysis, and client segmentation. The k-means algorithm partitions data into a predetermined number of clusters, as chosen by the user. Subsequently, the algorithm utilizes the measure of distance between individual data points and the centroid of each cluster to ascertain the appropriate cluster assignment for each data point (Yong, 2010)[22]. The centroid of a cluster is defined as the arithmetic mean of all the data points within the cluster.

The k-means algorithm operates as follows:

- I. The initial k centers are randomly selected.
- II. The data points are organized into k clusters by assigning them to their respective centroid.
- III. Calculate the centroid for each cluster.
- IV. Each data point is assigned a new centroid, replacing the old centroid.

Continue to repeat steps 3 and 4 until both the centroids and medians remain within a predetermined threshold, indicating stability in the clustering process. The k-means algorithm generates k clusters, each of which consists of data points that exhibit the highest similarity within their respective cluster and the lowest similarity with data points in other clusters.

SILHOUETTE ANALYSIS

Silhouette analysis is employed in unsupervised learning methodologies such as k-means clustering to evaluate the precision of the resultant clusters. The degree of conformity of each data point inside its cluster is assessed by comparing it to the other data points in that cluster. The silhouette score is a metric that measures the quality of clustering results. It is a numerical value that falls within the range of -1 to 1. Higher scores, closer to 1, indicate well-defined clusters, while lower scores, closer to -1, suggest less distinct clusters. This score is utilized to quantify the level of isolation across clusters (Wang, 2017)[23].

The subsequent sections delineate the many processes involved in silhouette analysis

The silhouette score for each data point is provided below. It is necessary to calculate the average distance of each data point from all other points inside its own cluster, as well as from all points in the next cluster. The silhouette



**Nilam N. Parmar and Sweta S. Panchal**

score of a point is determined by dividing the difference between the distances to the nearest cluster (b) and the second nearest cluster (a) by the greatest value between a and b. Analyze and interpret the results: If a cluster exhibits a high average silhouette score, it indicates that the data points within the cluster are very similar to each other and are distinct from the data points in other clusters. A cluster exhibiting a low average silhouette score indicates inadequate isolation from adjacent clusters and is likely to have data points that exhibit limited comparability among themselves.

The application of silhouette analysis enables the evaluation of clustering outcomes in terms of their accuracy and facilitates the identification of the optimal approach for partitioning a given dataset into distinct groups. In order to ascertain the relevance and utility of the created clusters for subsequent study, it is feasible to ascertain the optimal number of clusters by means of comparing the silhouette scores across different cluster quantities (Wang, 2022)[16].

RESULT AND DISCUSSION

The entire process is strategically divided into distinct phases to optimize the utilization of data for effective machine learning training.

Pre-processing and Knowledge Discovery

The initial phase focuses on pre-processing the existing dataset through the Knowledge Discovery process. This involves extracting pertinent information that is crucial for training machine learning algorithms. The aim is to filter and refine the dataset, ensuring that only the most relevant data is considered for further analysis.

Normalization using Min-Max Normalization

Following the pre-processing phase, the dataset undergoes normalization using the Min-Max normalization technique. This step is pivotal in determining the probability of a score occurring within the normal distribution of the data. By standardizing the data range, the normalization process enhances the algorithm's ability to uncover patterns and relationships within the dataset.

Clustering Technique for Unlabeled Data

Simultaneously, an advanced clustering technique is applied to the unlabeled data to identify inherent groups within the dataset. Data points are clustered based on the similarity of their features, allowing for the discovery of underlying patterns and structures. This step facilitates a more nuanced understanding of the relationships among data points.

Silhouette Analysis for Data Interpretation

In the final phase, a sophisticated Silhouette Analysis is employed to measure the distance between different points within a cluster. This analysis provides a graphical representation of the classification between various data points, allowing for a visual interpretation of the model's performance. Silhouette Analysis aids in assessing the effectiveness of the clustering algorithm and highlights the distinctiveness of clusters.

Data Analysis and Pre-Processing

Dataset: <https://archive.ics.uci.edu/dataset/352/online+retail>. In order to perform an exhaustive examination of the customer data, we employ a variety of robust data science libraries, such as Matplotlib, NumPy, and Pandas. These libraries are of the utmost importance for perusing, manipulating, and visualizing the available data in an efficient manner; they facilitate insightful observations and well-informed decisions.



**Nilam N. Parmar and Sweta S. Panchal**

DATASET FEATURES

ID of the Customer: Serves as a unique identifier for each customer, allowing for individual tracking and analysis.

Invoices of the Customer: Records the number of invoices associated with each customer, providing insights into their transactional activity.

Unit Price of Product: Reflects the cost per unit of the product purchased, aiding in understanding the pricing dynamics and customer spending patterns.

Quantity: Represents the quantity of products bought by each customer, contributing to a comprehensive assessment of purchasing behavior.

Invoice Date: Captures the date of each invoice, facilitating temporal analysis and enabling the identification of trends over time.

Amount: Quantifies the monetary value associated with each transaction, offering a holistic view of customer expenditure.

ANALYSIS PROCESS

The analysis process involves importing the dataset using Pandas, utilizing NumPy for numerical operations, and employing Matplotlib for creating visualizations. Through these libraries, we aim to uncover meaningful insights into customer segmentation, transaction patterns, and overall trends within the dataset. By combining the features of the customer data with the capabilities of these data science libraries, the analysis becomes a robust and systematic exploration, leading to a richer understanding of the dynamics and behaviors inherent in the customer dataset. From above Table 1 and Fig 3 the analysis indicates that the dataset holds a wealth of diverse and valuable information, providing numerous opportunities for extraction and exploration. With a comprehensive set of features such as customer demographics, product details, and transaction history, the dataset opens up a broad spectrum of analytical possibilities. It allows for in-depth examinations of customer behavior, product popularity, purchasing trends over time, and the identification of correlations between various variables. The dataset's richness also facilitates segmentation of customers into distinct groups, enabling targeted marketing and personalized strategies. Furthermore, the presence of patterns and trends within the data supports predictive modeling, aiding in forecasting future trends, customer preferences, and market shifts. Businesses can leverage this variability to optimize their operations, improve efficiency, and stay competitive. Continuous analysis and extraction of insights from this diverse dataset contribute to ongoing efforts in adapting to changing market conditions and enhancing overall business performance. In summary, the dataset presents a robust foundation for comprehensive exploration and extraction of valuable insights that can drive strategic decision-making and business success.

RFM Analysis

The RFM (Recency, Frequency, Monetary) model serves as a powerful technique in understanding and quantifying customer behavior based on their responsiveness to transactions. This methodology involves the assessment of three key parameters

Recency: The Recency parameter delineates the recentness of a customer's purchase activity. It aims to identify when a customer last engaged in a transaction, providing insights into their ongoing connection with the organization. The calculation of Recency involves assigning values based on the most recent purchase date, forming a foundation for subsequent reminders and targeted marketing campaigns.

Frequency: Frequency relates to how frequently a customer engages in transactions. This parameter considers factors such as product type, pricing, and customer behavior regarding product replacements. By analyzing Frequency, organizations can manage marketing costs and efforts more effectively, attracting new customers and retaining the loyalty of existing ones.

Monetary: The Monetary parameter helps identify the amount of money a customer spends on purchases and reveals the spending behavior of different customer segments. Calculated using a specific syntax, Monetary value aids in understanding the nature of customer expenditure, providing valuable insights for targeted marketing and tailored business strategies.





Nilam N. Parmar and Sweta S. Panchal

From above Table 2 In the initial steps of RFM analysis, customers are assigned values for Recency, Frequency, and Monetary parameters. Subsequently, the customer list is segmented into tiers based on these dimensions (R, F, and M), forming the basis for targeted marketing and advertising campaigns. This sophisticated approach allows organizations to tailor their strategies effectively, optimizing customer engagement and maximizing business impact.

Fig 4 by visualizing the distribution of clients across various recency, frequency, and monetary divisions, this graph helps firms adjust their marketing tactics and offerings. Shorter bars represent fewer clients, whereas higher bars represent a greater amount of customers falling into that group. By looking at how the bars are distributed throughout the Recency, Frequency, and Monetary categories. For example: A large proportion of clients in the "0-50 days" recency category points to a robust recent clientele. More consumers in the higher frequency categories point to a devoted following of buyers who make up to four purchases annually. Higher bars in the Monetary "High spenders" category denote important consumers that make a considerable revenue contribution up to a minimum purchase of \$50.

Building Dendrogram

A dendrogram is a diagram that illustrates the distances between attributes in sequentially merged classes. To prevent overlapping lines in the diagram, it arranges the merged pairs of classes adjacent to their neighbors. The information within this diagram is subsequently utilized for clustering algorithms. In essence, a dendrogram provides a visual representation of the relationships and distances between paired classes, facilitating a clearer understanding of clustering patterns and aiding in the analysis of hierarchical structures. From Above Fig 5 upon careful inspection of the dendrogram, it is observed that there are discernible points where the vertical lines display notable shifts in height. These points may suggest potential cluster boundaries, and the number of these significant changes could correspond to the optimal number of clusters.

Specific Findings:

Three Potential Clusters: A distinct change in height is noticed, indicative of a potential split into three clusters. The branches below this point represent relatively homogeneous groups of data points.

Four Potential Clusters: Another notable change in height occurs, suggesting a possible alternative clustering scenario with four distinct groups. This is marked by a significant divergence in the dendrogram structure.

Decision-Making: The choice between three or four clusters depends on the specific characteristics of the dataset and the objectives of the analysis. It may be beneficial to experiment with both scenarios and evaluate the performance of clustering algorithms to determine the most suitable number of clusters for extracting meaningful insights from the data.

Model Building Using K-Means Clustering

The K-Means clustering algorithm is a versatile tool for partitioning data into distinct groups based on similarity. When utilizing the K-Means class, several parameters contribute to the customization of the algorithm. In this particular implementation, the focus is on three key parameters:

Init (Initialization Method):

Init defines the method for initializing centroids, which are crucial in determining the cluster centers. The 'k-means++' option, chosen here, employs a smart strategy to place the initial centroids, enhancing the convergence speed of the K-Means algorithm.

n_clusters (Number of Clusters)

The n_clusters parameter sets the number of clusters the algorithm aims to form. In this case, the value is set to 3, indicating the intention to identify three distinct groups within the data. Adjusting this parameter allows for flexibility in capturing different levels of granularity in the clustering.



**Nilam N. Parmar and Sweta S. Panchal****n_init (Number of Initializations)**

n_init specifies how many times the K-Means algorithm will run with different initial centroid seeds. The algorithm calculates the inertia for each run, and the configuration yielding the lowest inertia is considered the final result. In this implementation, a common practice is to set n_init to 12, ensuring robustness in capturing the optimal clustering outcome.

Visualization of Clusters

Above Fig 6 the visual representation of customer clusters reveals compelling insights. Cluster 1 encapsulates customers sharing notable similarities in characteristics and behaviors, presenting a lucrative target for focused marketing campaigns. The concentrated nature of this cluster suggests that tailoring strategies specifically for these customers could yield impactful results. Conversely, Cluster 3, while smaller in size, represents a distinct customer segment. Recognizing this smaller group's unique attributes and preferences is an opportunity for businesses to strategize on expanding and retaining customers within this cluster. By implementing targeted initiatives, businesses can aim to increase the size and influence of Cluster 3. Visual aids, such as bar graphs, prove invaluable in this context. These tools facilitate a quick and comprehensive understanding of customer segments. Bar graphs, for instance, enable businesses to visually discern the composition of each cluster, empowering them to fine-tune marketing efforts with precision. Leveraging such visual aids enhances the effectiveness of marketing strategies, maximizing engagement and ultimately driving revenue.

In essence, the visualization serves as a strategic guide, enabling businesses to navigate customer segmentation effectively and optimize their marketing approach for each distinct cluster.

SILHOUETTE ANALYSIS FOR OPTIMAL CLUSTERING

Silhouette analysis is a crucial technique for validating clustering groups and determining the most suitable cluster configuration. The silhouette() function plays a pivotal role in this process by computing the average width of silhouettes. This width serves as a valuable metric for identifying the optimal cluster size. The silhouette average width is plotted against the number of cluster centers to visually identify the optimal cluster configuration. The cluster with the largest silhouette average width is considered the best choice. In this example, the general value of the average width of the silhouette is found to be 3 clusters per the analysis. Upon evaluating the clustering analysis, K-means clustering with Silhouette evaluation method demonstrates superior performance, particularly with k=4. In contrast, hierarchical clustering with the elbow method encounters challenges as it does not perform optimally with the same k value, highlighting the potential ambiguity associated with this approach.

In order to find the ideal number of clusters in a dataset, the Silhouette Analysis graph is a useful tool for cluster analysis. It computes the Silhouette Score for each data point and then averages these values to get an overall score for the clustering configuration.

X-axis: Number of Clusters (k):

The number of clusters being assessed in the dataset is shown on the x-axis. It varies from two to fourteen clusters in this instance.

Y-axis: Silhouette Score:

The y-axis displays the Silhouette Score, a metric that quantifies an object's cohesiveness (similarity to its own cluster) in relation to its separation (difference from other clusters). The range of the Silhouette Score is -1 to 1. When an item has a high silhouette score, it means that it matches well with its own cluster and poorly with nearby clusters. This is advantageous as it implies a distinct division between clusters. Finding the number of clusters (k) where the Silhouette Score is maximized—a sign of the ideal clustering arrangement—is the aim. The fact that your dataset is divided into three clusters yields the highest average similarity within clusters and the largest dissimilarity across clusters when compared to alternative cluster configurations, as indicated by the higher score in three clusters. From



**Nilam N. Parmar and Sweta S. Panchal**

above Table 3 Each client's cluster assignment is listed in the cluster column, with values of 1 through 3 designating which cluster the customer belongs to. It is anticipated that customers belonging to the same cluster will display comparable behavior or attributes, depending on the clustering parameters (Recency, Frequency, Monetary). Customers in Cluster 1 may, therefore, be people who have recently made small-scale, infrequent transactions. Consumers in Cluster 2 may be those who make regular purchases of moderate value, while consumers in Cluster 3 might be high-value individuals who make regular purchases of large value. You may better satisfy the demands of each segment by customizing your marketing strategy, product offers, and customer service activities to each cluster, which represents a specific portion of your client base with unique behavior patterns.

COMPARISON OF EXISTING AND PROPOSED SYSTEM

In Existing system Agglomerative Clustering was used with RFM analysis, after finding the clustering using different k. The greater silhouette score found in 3 clusters same as in k-means. From Fig 8 and Table 4 the proposed technique has significantly improved customer segmentation compared to the existing agglomerative clustering method. With agglomerative clustering, the silhouette score stood at 0.42 when utilizing 3 clusters. However, upon implementing K-means clustering with silhouette analysis, the silhouette score notably increased to 0.46 with the same number of clusters. This enhancement signifies that the proposed technique not only provides a more objective measure for determining the optimal number of clusters but also yields more distinct and well-separated clusters, thus leading to a more refined and effective segmentation of customers.

CONCLUSION

In the domain of consumer segmentation, enhancing data collection and utilizing the acquired data to derive pertinent insights constitutes a highly consequential undertaking. The utilization of the k-means algorithm the topic of interest is clustering. The application of silhouette analysis is employed to assess the precision of customer classification, while the utilization of K-means aids in the identification and removal of potential clients who are unlikely to contribute to the company's growth. The customer segmentation method involved the application of three approaches: k-means clustering, silhouette analysis, and RFM analysis. One of the most significant findings is that clients who have been active recently have the greatest evaluations in terms of frequency and monetary value. The evaluation of clustering quality is conducted by the application of the silhouette approach, which entails the assessment of both intra-cluster similarity and inter-cluster dissimilarity. The silhouette score is a metric that measures the quality of clustering results. The utilization of three clusters might facilitate firms in customizing their marketing tactics to distinct client segments characterized by comparable demands and preferences, thereby enhancing customer happiness and augmenting income.

ACKNOWLEDGEMENT

This work is supported by the engineering faculty of Dr. Subhash University

REFERENCES

1. Bardaki, Sales business analysis: Customer categories use market basket data.. Systems Expert Systems, 2018. Volume 100, pp. 1-16.
2. Premkanth, Market Classification and Its Impact on Customer Satisfaction and Special Reference to the Commercial Bank of Ceylon PLC. Global Journal of Management and Business
3. Publisher Research: Global Magazenals Inc.2012. (USA), 12(1), pp. 1-12.
4. Goyat, The basis of market segmentation: a critical review of the literature.. European Journal of Business and Management , 2011. 3(9), pp. 113-117.
5. Hemashree Kilari, Sailesh Edara, Customer Segmentation using K-Means Clustering, International Journal of Engineering Research & Technology (IJERT), ISSN: 2278-0181, Vol. 11 Issue 03, March-2022





Nilam N. Parmar and Sweta S. Panchal

6. Garima Sharma, Ankita Nainwa, Customer Segmentation Using Machine Learning, *Ilkogretim Online - Elementary Education Online*, 2021; Vol 20 (Issue 3): pp. 3230-3237 <http://ilkogretim-online.org> doi: 10.17051/ilkonline.2021.03.335
7. Mohamad, Standardization and Its Effects on K-Means Clustering Algorithm. *Research Journal of Applied Sciences, Engineering and Technology*, 2013, 1(17), pp. 3299-3303.
8. Mukhametzyanov, ON THE CONFORMITY OF SCALES OF MULTIDIMENSIONAL NORMALIZATION: AN APPLICATION FOR THE PROBLEMS OF DECISION MAKING. *Decision Making: Applications in Management and Engineering*, 6(1), 2023, pp. 399-431.
9. Forina, Clustering with dendrograms on interpretation variables. *Analytica Chimica Acta*, 454(1), 2002, pp. 13-19.
10. Bell, 2007. *Advances in Collaborative Filtering*. IEEE, Volume 7, pp. 42-43.
11. Jain, A. a. D., 1988. *Algorithms for Clustering Data*. Prentice-Hall, Englewood Cliffs., pp. 255-257.
12. Gupta, 2006. *Modeling Customer Lifetime Value*. *Journal of Service Research*, 9(2), pp. 139-155.
13. Chu, Customer segmentation using K-means clustering and the adaptive particle swarm optimization algorithm. *Applied Soft Computing*, Volume 1, 2021, pp. 111-113.
14. Shalabi, L.A., Z. Shaaban and B. Kasasbeh, *Data Mining: A Preprocessing Engine*, 2006, *J. Comput. Sci.*, 2: 735-739.
15. Patro, *Technical Analysis on Financial Forecasting*, 2006, *International Journal of Computer Sciences and Engineering*, pp. 1-3.
16. Hughes, *Strategic Database Marketing*. Probus Publishing, 1994, pp. 1-8.
17. Wang, *Market Segmentation of Online Reviews: A Network Analysis Approach*. 2022, *International Journal of Market Research*, 64(5), pp. 75-76.
18. Miglautsch, *Application of RFM principles: What to do with 1-1-1 customers?*. 2002. *Journal of Database Marketing*, 9(4), pp. 319-324.
19. Ali, *Investigating the Impact of Min-Max Data Normalization on the Regression Performance of K-Nearest Neighbor with Different Similarity Measurements*, 2022, Koya: Department of Software Engineering, Faculty of Engineering, Koya University.
20. S.Gopal Krishna Patro, Kishore Kumar sahu "Normalization: A Preprocessing Stage" Article in *IARJSET* · March 2015 DOI: 10.17148/IARJSET.2015.2305 · Source: arXiv
21. Chehreghani, *Learning representations from dendrograms*. 2020, Springer, Volume 109, pp. 1779-1802.
22. Nindhia, *Dendrogram Simulations with Deterministic Variable Identifier to Determine the Farm Classification Systems of Bali Pigs*, 2015, *International Journal of Science and Research (USR)*, Volume 1, pp. 1-5.
23. Yong, *Research on k-means Clustering Algorithm: An Improved k-means Clustering Algorithm*. 2010, Jian, China, IEEE.
24. Wang, *An Analysis of the Application of Simplified Silhouette to the Evaluation of k-means Clustering Validity*. 2017, s.l., 13th International Conference on Machine Learning and Data Mining MLDM 2017At: New York, USA

Table 1 Feature Analysis

Data columns (total 8 columns):				
#	Column	Non-Null	Count	Dtype
0	InvoiceNo	354321	non-null	object
1	StockCode	354321	non-null	object
2	Description	354321	non-null	object
3	Quantity	354321	non-null	int64
4	InvoiceDate	354321	non-null	datetime64[ns]
5	UnitPrice	354321	non-null	float64
6	CustomerID	354321	non-null	object
7	Country	354321	non-null	object
8	Amount	354321	non-null	float64





Table 2. Result for Recency, Frequency and Monetary Calculation

[1]	[2] CustomerID	[3] Recency	[4] Frequency	[5] Monetary
[6] 0	[7] 12346	[8] 325	[9] 1	[10] 77183.6
[11] 1	[12] 12747	[13] 2	[14] 11	[15] 689.49
[16] 2	[17] 12748	[18] 0	[19] 209	[20] 3841.31
[21] 3	[22] 12749	[23] 3	[24] 5	[25] 98.35
[26]	[27]	[28]	[29]	[30]
[31] 3916	[32] 18281	[33] 180	[34] 1	[35] 5.04
[36] 3917	[37] 18282	[38] 7	[39] 2	[40] 38.25
[41] 3918	[42] 18283	[43] 3	[44] 16	[45] 66.75
[46] 3919	[47] 18287	[48] 42	[49] 3	[50] 80.4

Table 3. Customer Segmentation using Clusters

[51]	[52] Customer ID	[53] Recency	[54] Frequency	[55] Monetary	[56] R	[57] F	[58] M	[59] Cluster
[60] 0	[61] 12749	[62] 4	[63] 5	[64] 98.35	[65] 0.0091 19	[66] 0. 4	[67] 0.4302 53	[68] 2
[69] 1	[70] 12820	[71] 4	[72] 4	[73] 58.2	[74] 0.0091 19	[75] 0. 3	[76] 0.2539 09	[77] 1
[78] 2	[79] 12821	[80] 215	[81] 1	[82] 19.92	[83] 0.6504 56	[84] 0	[85] 0.0857 78	[86] 3
[87]	[88]	[89]	[90]	[91]	[92] ...	[93] ...	[94]	[95]
[96] 31 91	[97] 18281	[98] 181	[99] 1	[100] 5.04	[101] 0.5471 12	[102] 0	[103] 0.0204 23	[104] 3
[105] 31 92	[106] 18282	[107] 8	[108] 2	[109] 38.25	[110] 0.0212 77	[111] 0. 1	[112] 0.1662 86	[113] 1
[114] 31 93	[115] 18287	[116] 43	[117] 3	[118] 80.4	[119] 0.1276 6	[120] 0. 2	[121] 0.3514 14	[122] 1

Table 4. Comparison of two algorithm

[123] Algorithm	[124] Agglomerative	[125] K-means
[126] Silhouette Score	[127] 0.42	[128] 0.46

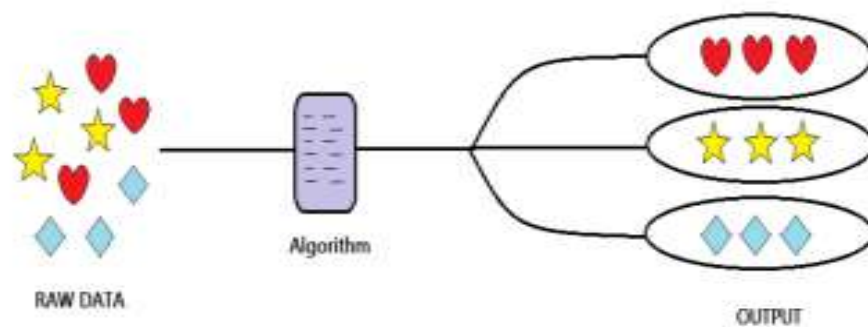
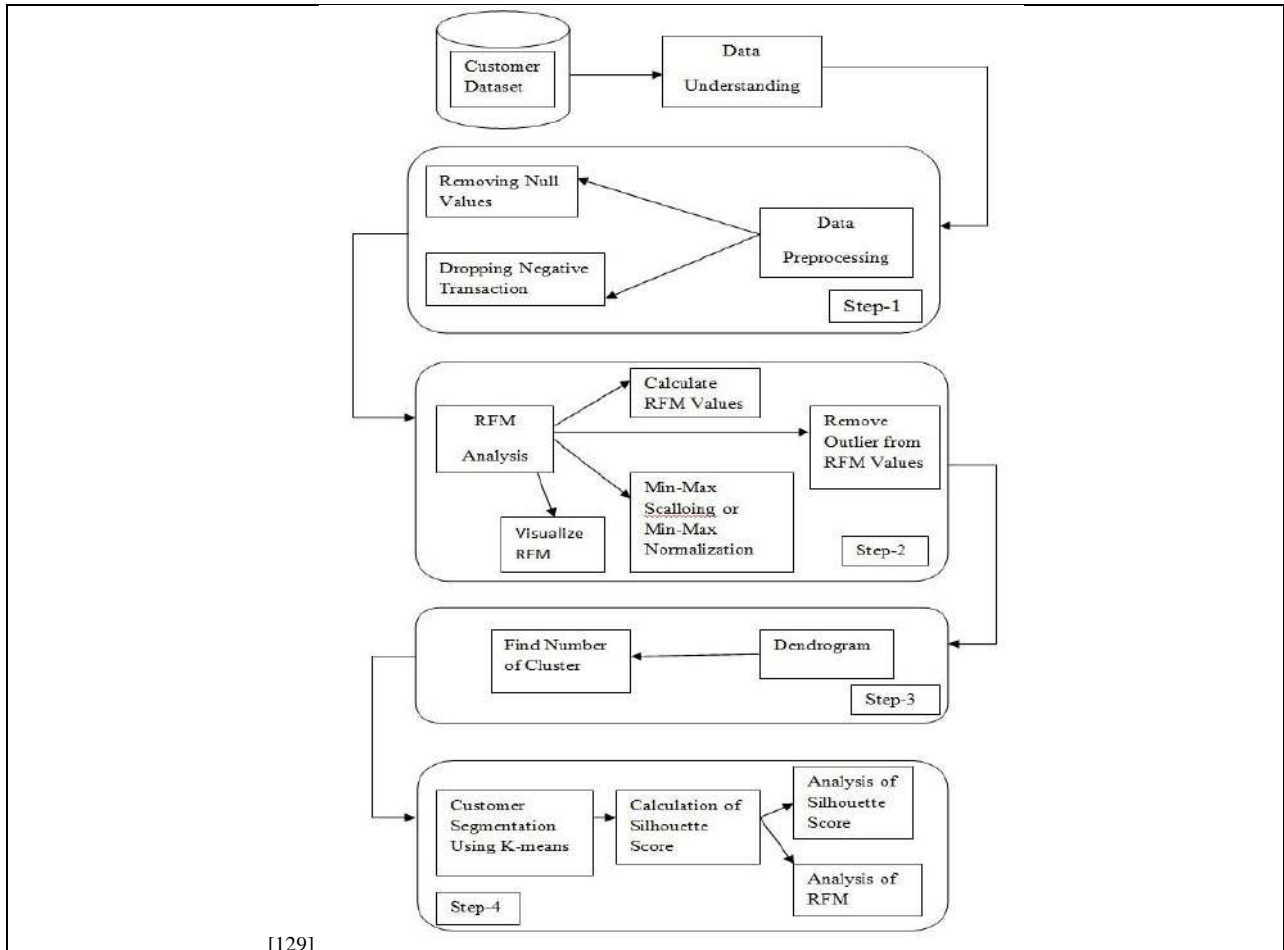


Fig. 1 Clustering [4]





Nilam N. Parmar and Sweta S. Panchal

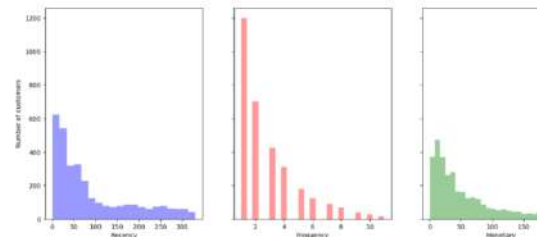


[129]

[130] Fig. 2 Workflow block diagram for Proposed Methodology

[131] Summary..
 Number of invoices: 16646
 Number of products bought: 3645
 Number of customers: 3920
 Percentage of customers NA: 0.0 %
 Average quantity of product purchased by a customer: 1086.0
 Average revenue generated per customer: 1864.39
 Average product quantity sold per transaction: 12.0
 Average revenue generated per transaction: 20.63

[132]



[133]

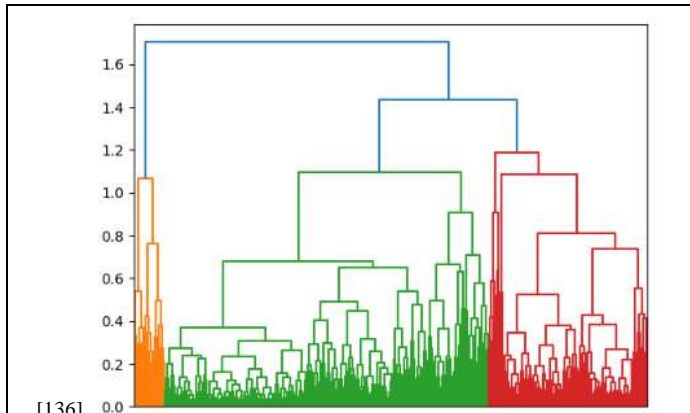
[134] Fig. 3 Data Analysis of each Attribute

[135] Fig. 4 RFM Visualization

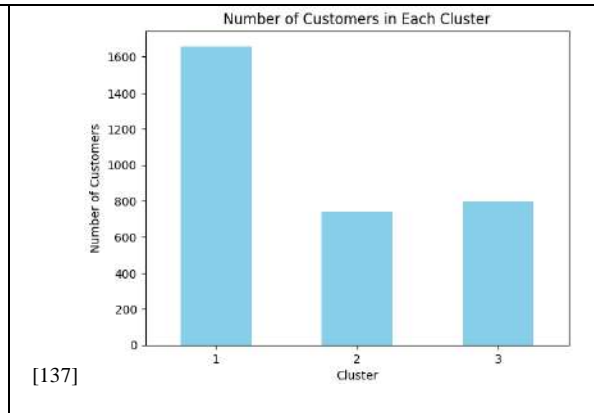




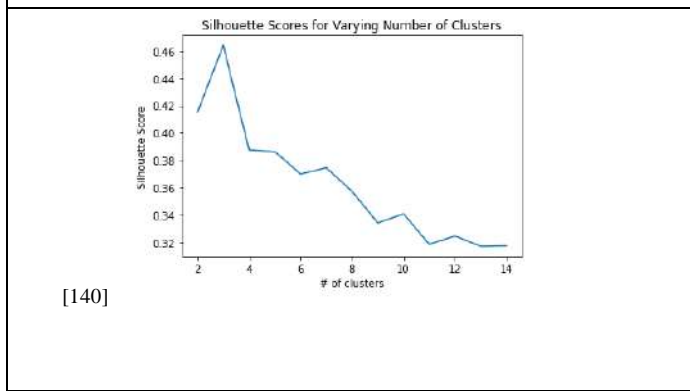
Nilam N. Parmar and Sweta S. Panchal



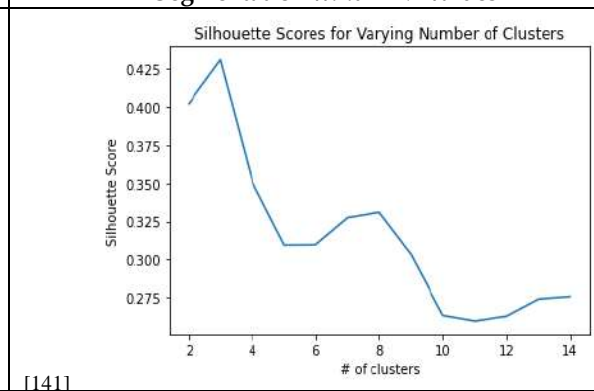
[136] Fig. 5 Clusters Representation using dendrogram



[137] Fig.6 Finding Respective Cluster Segmentation w.r.t RFM values



[140] Fig. 7 Clusters found using Silhouette Analysis



[141] Fig.8 Agglomerative silhouette Results





Navigating Fragmentation Maze: Unraveling the Impact of Content Fragmentation on OTT Consumption Habit

Jasmine V.M^{1*} and Keerthana Murali²

¹Assistant Professor, PG & Research Department of Commerce, St. Joseph's College (Autonomous), Devagiri, (Affiliated to University of Calicut), Calicut, Kerala, India.

²Customer Service Associate, South Indian Bank Ltd, Abhilash Shopping Centre, Azhikode (P.O) Thrissur, Kerala, India.

Received: 06 Jun 2024

Revised: 18 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

Jasmine V.M

Assistant Professor,
PG & Research Department of Commerce,
St. Joseph's College (Autonomous), Devagiri,
(Affiliated to University of Calicut),
Calicut, Kerala, India.
E.Mail: jasminejithin2@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The proliferation of OTT (Over-The-Top) platforms, advancements in mobile technology, improved streaming quality, and the democratization of content creation all have contributed to a transformative media landscape. In this backdrop the current study is an attempt to investigate how far content fragmentation propel the OTT consumption in the media industry. Moreover, it tries to understand customers' preference for OTT platforms compared to traditional platforms based on content fragmentation. As per the previous studies the usage of OTT platforms has tremendously increased during Covid-19 and the content fragmentation is noticed as a new marketing strategy for attracting media consumers. Considering these aspects, present study tries to examine whether this content fragmentation really fuel the OTT viewing habit. Data collected from 100 respondents are analysed using multiple regression and percentage analysis to fulfil the objectives. The results suggest that various benefits and features of content fragmentation like easy availability, easy discovery, user-friendliness, smooth flow, and easy accessibility of contents have significant impact on OTT consumption habit. In the same way, young media consumers are more interested in OTT platform than traditional channel for video consumption. Thus, this study is very helpful for broadcasters to frame more tactical marketing ideas to attract and maintain customers.

Key words: Content fragmentation, OTT platform, Media Consumption, Technology.



**Jasmine and Keerthana Murali**

INTRODUCTION

The technological advancements, coupled with the paralyzing effects of the COVID-19 pandemic, have brought about significant configurations in the landscape of media consumption in recent years (Sharma & Lulandala, 2023). The proliferation of digital platforms, the advent of cheap and high-speed internet, and the availability of High-Definition (HD) and Ultra HD (4K) resolutions, along with advancements in audio technologies such as Dolby Atmos, have revolutionized how people access and interact with media content (Mairaru et al., 2019). Moreover, the evolution of mobile technology has also played a crucial role in shaping media consumption patterns (Singh, 2019). With the widespread adoption of smartphones and tablets, consumers can now carry portable devices capable of streaming videos, listening to music, and accessing various forms of digital content (Ernst & Young, 2016). The convenience of mobile devices, coupled with improved internet connectivity, has made media consumption more accessible and seamless, empowering users to enjoy their favourite content at anytime and anywhere (Ulin, 2013; Bentley et al., 2019).

In connection with technology-enabled media consumption, one of the most notable advancements is the rise of Over-The-Top (OTT) platforms. OTT technology enables content owners to manage the process of delivering video content across multiple devices using the internet. Remarkably, amidst the challenges brought about by the pandemic, the OTT industry has emerged as one of the rare beneficiaries (www.argoid.ai). The shutdown of production houses and cinema halls across the nation has created a huge demand for OTT video streaming platforms, offering original series and movies (Shaikh and Aggarwal, 2020; PwC, 2020). In contrast to traditional networks, OTT services like Netflix, Amazon Prime Video, Hotstar, Hulu, and Voot deliver streaming media directly to consumers at their request via the internet, bypassing traditional cable or satellite providers (Purdy, 2018). This shift has enabled users to enjoy on-demand access to a vast array of movies, web series, TV shows, documentaries, and other forms of content, allowing them to personalize their viewing experience according to their preferences and schedules (Puthiyakath & Goswami, 2021). With more and more people subscribing to several OTT platforms, it is no surprise that streaming services are major contributors to the media industry. According to a report published in Economic Times in April 2020, OTT video streaming platforms in India saw a growth of 34 percent in March 2020. Industry experts claim that the current rise in OTT video streaming platform viewing has already fuelled the expansion of these platforms over the next five years. It has been forecasted that OTT platforms' revenue could reach US\$4 billion by the year 2025 (www.ibef.org).

At the same time, as we step into 2023, the realm of OTT (Over-The-Top) platforms finds itself standing at a pivotal juncture, poised for significant transformations and advancements. Alongside the growth in OTT platforms, content fragmentation has become a significant challenge for this industry. Content fragmentation refers to the distribution of media content across multiple platforms, creating a fragmented landscape where consumers need to subscribe to multiple services to access their desired content (Deloitte, 2021). This fragmentation arises due to various factors, including the entrance of new players into the market, the rise of exclusive content licensing deals, and the strategic decisions made by content creators and distributors. The impact of content fragmentation on OTT consumption is multifaceted. On one hand, consumers are faced with an abundance of choices and a diverse range of content options. This offers viewers the flexibility to select content that aligns with their preferences and interests. However, on the other hand, the increasing number of subscription-based platforms and the fragmentation of content can lead to subscription fatigue and higher costs for consumers. This maze-like condition can be particularly challenging for viewers who are interested in accessing specific shows or movies that are exclusive to certain platforms.

Anyway, users like to see tailored content based on their previous experiences that may break their preconceived ideas about what to watch. In the most advanced way, content fragmentation offers a clear, simple approach to locate and consume content that is both comfortable and interesting (Begum, 2018; Haryoto, 2015). Many previous studies have examined drivers of high dependency on second screens and continued use of OTT platform, especially during Covid-19 pandemic (Valecha & Jaggi, 2020; Gupta & Singharia, 2021). There are studies discussing factors contributing



**Jasmine and Keerthana Murali**

to the satisfaction and dissatisfaction of OTT services also (Shin & Park, 2021). So, a gap is insinuated that the studies conducted to date have not yet seriously taken into consideration consumers' usage of OTT platforms as a result of content fragmentation. Therefore, this research paper aims to decipher an understanding about the effect of content fragmentation that stimulates the usage of OTT video streaming platforms, among the young consumers. Moreover, this research paper aims to gain a better understanding of the preference for OTT platforms and traditional platforms grounded on content fragmentation.

Significance of the Study

The study discussing the effect of content fragmentation on OTT watching habit is useful both to OTT users and broadcasters. To the viewers the viewing experience feels almost personalized when they open their favourite streaming platform and finds a collection of content that is almost entirely in line with their preferences and interests. Study on content fragmentation boost the broadcasters to build loyal audiences and communities around their brands. Moreover, in this period of content overload consumers are confused to understand which one is more important and needed along with subscription fatigue (McAdams, 2019). Therefore, they have to use content fragmentation, a new advancement in technology, which has brought some tremendous changes in the way of enjoying the media contents.

Literature Review and Conceptual Development

The existing literature on OTT platforms suggests that the presence of intriguing content is crucial in retaining viewers in an OTT streaming platform and the success of an OTT platform largely depends on its ability to facilitate easy content discovery. The following section will discuss reviews about content fragmentation and OTT platform.

Content Fragmentation

Content fragmentation is a buzzword in today's OTT market place. It is a strategy that helps broadcasters to reach niche audiences. The term content fragmentation implies the collection of content that is driven by preferences, tastes, gender and other factors of a specific target group. If the viewers have felt content as relevant and engaging, it will connect the target audience and subscribers remain active (www.muvi.com). Increased content consumption through digital media is changing consumer preferences and attitudes and this development may be ascribed to better internet access, digital gadgets, competitive data pricing in India and the mobile aspect of online media (Bhavsar, 2018). Content fragmentation offers a simple approach to locate and consume content with ease and interest. Distributing different types of content on different Video-On-Demand (VOD) platforms is one of the effects of this approach. The purpose of this trend is to create a variety of platforms that are focused on a specific type of video contents to give the users exactly what they are looking for. It is being adopted in the new broadcasting sector and OTT players are tapping into its many benefits.

Over-the-Top (OTT) Consumption

OTT or Over-the-Top refers to streaming services that allow content to be delivered over the internet. As the name implies, the service is offered "on the top" of another platform. It bypasses the traditional satellite cable and broadcasting platforms (Puthiyakath & Goswami, 2021). OTT delivers television video and audio through the internet instead of conventional radio frequencies. This means that OTT platforms do not have to send the same show to everyone at the same time. Content is provided at the request of the consumers to meet his/her requirements. This is the main feature of OTT platforms that differentiates itself from traditional television services. OTT services and television networks can both coexist in the same market. However, it is anticipated that, younger generation will move on to OTT platform, while the older people assumed to prefer traditional television (Negi, 2022). Baccarne, Evens, and Schuurman, (2013) examined in their study, how the emergence of OTT platforms has resulted in the loosing of demand and monopoly of traditional TV and as per their report, the reason for the success of OTT is its content quality, removal of time restrictions, usability, flexibility, tailored watching experiences etc., (Sing, 2019). Among the factors the availability of 'personalised content' and effectiveness of 'dubbing' and 'subtitles' for foreign content also noticed as the reason for the increased OTT consumption (Begum, 2018). Along with the above-mentioned variables, accessibility of user-friendly content at lower prices makes it more attractive and it leads to





Jasmine and Keerthana Murali

binge watching habit among the youngsters (Sujatha et al., 2015; Matrix, 2014). At the same time, the KPMG's annual cable & OTT report shows that, though the cost is a matter of consideration when buying OTT services, content still influences the majority of customer decisions (Purdy, 2018). Therefore, the acceptance and adoption of OTT depends on perceived risk, trust, age and many factors that are identified in many studies (Chang & Chen, 2018; Dasgupta, & Grover, 2019).

Objectives

To analyse the influence of content fragmentation on OTT consumption habit.

Hypotheses

The content fragmentation has significant impact on the OTT consumption habit.

Conceptual Model

Fig.1.Initial model

Research Design

This study used a survey method to collect the primary data from young respondents belonging to the age between 18-35 years. A non-probability-purposive sampling method has been adopted to select the samples from South India, specifically from the State of Kerala. Out of 116 respondents approached, 100 responses were found as filled and these were utilized for statistical analysis and interpretation. The data were collected during the period of July 2023 to Oct 2023 and a pilot study of 30 respondents were carried out. Based on the inputs from pilot study, the statements measuring content fragmentation were redrafted on a Likert scale and asked to give their degree of agreements regarding the its features in five-point scale ranging '1' for 'strongly Agree' to 5 'strongly Disagree'. As the variables measuring OTT Consumption habit consist of five variables no factor analysis is done for variable reduction. To determine the preference for OTT platforms over traditional platforms, based on content fragmentation, respondents are asked tick either in 'Traditional Platform' or in 'OTT Platform' after going through the statements.

Data Analysis

The collected data are analysed with the help of software, SPSS-20 and Excel. Before moving on to the detailed analysis, researcher checked the reliability of the content fragmentation variables namely easy availability, easy discovery, user-friendliness, smooth flow and easy accessibility of content and found that as .892 (Table 1). Then to study the influence and impact of content fragmentation on OTT consumption habit, 'multiple regression' analysis has been used (given in Table-2) and to assess the preference for OTT platforms or traditional channels in terms of content fragmentation features, percentage analysis has been applied and the results are shown in Table 3.

Regression Equation: Consumption Habit = .419 (Constant) + .145 (Easy availability of content) + .586 (Easy discovery of content) + .058 (User-friendliness of content) + .322 (Smooth flow of content) + .045 (Easy accessibility of content).

As per the regression equation, for every one unit of increase in the easy availability of content, OTT consumption get increased by .145 i.e., 14.5%. For every one unit of increase in the easy discovery of content, OTT consumption get increased by .586 i.e., 58.6%. Likewise, for every one unit of increase in the user-friendliness of content leads to .058 i.e., 5.8% of OTT consumption. The one unit increase in the customers' perception regarding smooth flow of content result in the .322 (32.2%) growth in OTT consumption habit. Finally, an increase in the feeling of easy accessibility of content will result in .045 or 4.5% impact on OTT consumption.

H₁: The content fragmentation has significant impact on the OTT consumption habit.

As per the Table 2.a, the adjusted R² value is .926 and this depicts that, 92.6% of the OTT platform viewing habit is determined by the identified variables of this research, describing the OTT platform viewers' perceptions about content fragmentation. To statistically test the above hypothesis (H₁), five sub hypotheses are framed and multiple regression results of these are discussed below.





Jasmine and Keerthana Murali

H_{1a}: Easy availability of content has significant impact on the OTT consumption habit.

$$\text{Model I: } Y_1 = \alpha + \beta X_1 + e$$

As per the study the feature of content fragmentation i.e., easy availability of content positively influences the OTT platform consumption of young viewers belonging to the age group of 18-35 and this effect ($\beta=.145$) is significant ($p \leq .05$) too.

H_{1b} Easy discovery of content has significant impact on the OTT consumption habit.

$$\text{Model II: } Y_2 = \alpha + \beta X_2 + e$$

As per the study the feature of content fragmentation i.e., easy discovery of content also noticed with positive influence on the OTT platform consumption and its effect is .586. As the p value is less than .05, this influence is significant.

H_{1c} User-friendliness of content has significant impact on the OTT consumption habit.

$$\text{Model III: } Y_3 = \alpha + \beta X_3 + e$$

Another feature tested is user-friendliness of content and it resulted with significant positive impact on the OTT platform usage ($\beta=.058$, $p \leq .05$). Thus, like easy availability and easy discovery, the feature of user-friendliness of content fragmentation contributes to OTT consumption.

H_{1d}: Smooth flow of content has significant impact on the OTT consumption habit

$$\text{Model IV: } Y_4 = \alpha + \beta X_4 + e$$

Again, as per the study the feature of content fragmentation i.e., Smooth flow of content is positively influencing the OTT platform consumption of youth and this effect is highly significant ($\beta=.322$, $p \leq .001$) too.

H_{1e} Easy accessibility of content has significant impact on the OTT consumption habit.

$$\text{Model V: } Y_5 = \alpha + \beta X_5 + e$$

In the case of easy accessibility, as per the test, customers responses are positively influencing OTT consumption and it is also significant.

DISCUSSION

The major focus of this study was to identify the impact of content fragmentation on OTT consumption pattern and here, the variables identified to determine the reasons for OTT consumption habit are found in line with the existing studies (Menon, 2022; Sadana & Sharma 2021). As per the results of this study, the variables studied to measure the perceptions propelling content fragmentation have significant influence (availability - $\beta=.145$, $t=2.54$, $p=.012$; discovery - $\beta=.586$, $t=8.788$, $p<.000$; user-friendliness - $\beta=.058$, $t=1.65$, $p=.002$; smooth flow - $\beta=.322$, $t=6.636$, $p<.000$ and accessibility - $\beta=.045$, $t=1.552$, $p=.024$) on the usage of OTT platforms also.

The results of the present study echo the contributions of existing studies, emphasizing that content, being the core element of entertainment, should be made available in the right manner, making the consumption more exciting (Matrix, 2014). However, despite of having its own challenges (Rojaset al., 2020) content fragmentation has become quite common among OTT platform users and content fragmentation creates a personalized viewing experience for the target audiences (Podara et al., 2021). This new trend of content fragmentation has its own importance in this era where people are facing the situation of content overload (Sivamol & Suresh, 2019). The content





Jasmine and Keerthana Murali

fragmentation makes it easy for the users to access their most needed content. But, the limitation of big screen show, subscription cost and risk perceptions are to be addressed seriously.

CONCLUSION

Content fragmentation in the OTT industry presents both opportunities and challenges. While consumers benefit from a wide variety of content choices and personalization options, they also face the complexities of managing multiple subscriptions and platforms. Content creators and distributors must navigate the competitive landscape while striving to maximize their reach and revenue. Besides, customer centric content play a very significant role in shifting the consumers from television to OTT platforms and the personalized contents and convenience in using contribute in escalating the consumers shift towards OTT platforms (Chen.Y.N.K, 2019).

REFERENCES

1. Baccarne, B., Evens, T., & Schuurman, D. (2013). The television struggle: an assessment of over-the-top television evolutions in a cable dominant market. *Communications & strategies*, (92), 43-61.
2. Begum, F. (2018). In India, localized content is as important as pricing when choosing a video service-IHS Technology. Retrieved January, 30, 2020.
3. Bentley, F., Silverman, M., and Bica, M. (2019, June), "Exploring online video watching behaviours", In Proceedings of the 2019 ACM International Conference on Interactive Experiences for TV and Online Video, pp. 108-117.
4. Bhavsar, R. (2018). The Burgeoning Digital Media Consumption: A Challenge for Traditional Television and Advertising Industries-An Analysis. *Amity Journal of Media & Communications Studies (AJMCS)*, 8(1).
5. Chang, H. H., & Chen, S. W. (2008). The impact of online store environment cues on purchase intention: Trust and perceived risk as a mediator. *Online information review*, 32(6), 818-841.
6. Chen, Y. N. K. (2019), "Competitions between OTT TV platforms and traditional television in Taiwan: A Niche analysis", *Telecommunications Policy*, Vol. 43 No. 9, pp. 101793
7. Dasgupta, D., & Grover, D. (2019). Understanding adoption factors of over-the-top video services among millennial consumers. *International Journal of Computer Engineering and Technology*, 10(1).
8. Deloitte. (2021). Digital Media Trends Survey. Retrieved from <https://www2.deloitte.com/content/dam/Deloitte/us/Documents/technology-media-telecommunications/us-tmt-digital-media-trends-survey-2021.pdf>
9. Ernst & Young LLP. (2016). *Ernst & Young Tax Guide 2017*. John Wiley & Sons.
10. Gupta, G., & Singharia, K. (2021). Consumption of OTT media streaming in COVID-19 lockdown: Insights from PLS analysis. *Vision*, 25(1), 36-46.
11. Haryoto, K. S. (2015), "The use of modified theory of acceptance and use of technology 2 to predict prospective users' intention in adopting TV Streaming"
12. Mairaru, S., Tyagi, S., Azeez, C., & Sharma, D. C. (2019). Understanding the print, web, television media habits and preferences of Indians: A uses and gratification perspective. *Journal of Content, Community and Communication*, 9(1), 75-80.
13. Matrix, S. (2014). The Netflix effect: Teens, binge watching, and on-demand digital media trends. *Jeunesse: young people, texts, cultures*, 6(1), 119-138.
14. McAdams, M. (2019). What Is OTT?—Understanding The Modern Media Streaming Landscape. *Tapjoy—электронный журнал— 2019—Режим доступа: <https://www.tapjoy.com/resources/what-is-ott/>[дата обращения 21.02.2020]*.
15. Menon, D. (2022). Purchase and continuation intentions of over-the-top (OTT) video streaming platform subscriptions: a uses and gratification theory perspective. *Telematics and Informatics Reports*, 5, 100006.
16. Negi, V. (2022). A Systematic Review of Literature on the Changing Consumption Patterns of Youth through OTT Platforms during the Covid-19 Lockdown. *Journal of Optoelectronics Laser*, 41(4), 693-704.





Jasmine and Keerthana Murali

17. Podara, A., Matsiola, M., Kotsakis, R., Maniou, T. A., &Kalliris, G. (2021). Generation Z's screen culture: Understanding younger users' behaviour in the television streaming age–The case of post-crisis Greece. *Critical Studies in Television*, 16(2), 91-109.
18. Purdy, s. c. (2018). KPMG's annual cable & OTT report.
19. Puthiyakath, H. H., & Goswami, M. P. (2021). Is over the top video platform the game changer over traditional TV channels in India? A niche analysis. *Asia Pacific Media Educator*, 31(1), 133-150.
20. PwC, C. M. (2020). PwC. In *Global Annual Review 2020*.
21. Rojas, J. S., Pekar, A., Rendón, Á., & Corrales, J. C. (2020). Smart user consumption profiling: Incremental learning-based OTT service degradation. *IEEE access*, 8, 207426-207442.
22. Sadana, M., & Sharma, D. (2021). How over-the-top (OTT) platforms engage young consumers over traditional pay television service? An analysis of changing consumer preferences and gamification. *Young Consumers*.
23. Shaikh, S., & Aggarwal, A. (2020). Lockdown a blessing in disguise?.
24. Sharma, K., &Lulandala, E. E. (2023). OTT platforms resilience to COVID-19–a study of business strategies and consumer media consumption in India. *International Journal of Organizational Analysis*, 31(1), 63-90.
25. Shin, S., & Park, J. (2021). Factors affecting users' satisfaction and dissatisfaction of OTT services in South Korea. *Telecommunications Policy*, 45(9), 102203.
26. Singh, P. (2019). New Media as a Change Agent of Indian Television and Cinema: A study of over the top Platforms. *Journal of Content, Community and Communication*, 9(2019), 131-37.
27. Sivamol, S., & Suresh, K. (2019). Personalization Phenom: User-centric Perspectives towards Recommendation Systems in Indian Video Services. *SCMS Journal of Indian Management*, 16(2), 73-86.
28. Sujata, J., Sohag, S., Tanu, D., Chintan, D., Shubham, P., & Sumit, G. (2015). Impact of over the top (OTT) services on telecom service providers. *Indian Journal of Science and Technology*, 8(S4), 145-160.
29. Ulin, J. (2013), "The business of media distribution: Monetizing film, TV and video content in an online world", CRC Press.
30. Valecha, P., & Jaggi, R. (2020). Understanding the Contextual Shift: Investigating the Influence of Physical Environment on Indian Millennials' Usage of Second Screen for Television Consumption. *PRism*, 16(1), 14-30.
31. <https://www.argoid.ai/blog/ott-streaming-consumption-trends>
32. <https://www.ibef.org>
33. <https://www.muvi.com/blogs/content-fragmentation-in-ott-streaming.html>

Table 1 Reliability Statistics

Cronbach's Alpha	N of Items
.892	5

Source: Primary data Analysis

Table 2: Multiple regression Analysis for content fragmentation perceptions and OTT consumption habit.

Table 2.a Model Summary

Model	R	R Square	Adjusted R Square	Std. Error of the Estimate
1	.964 ^a	.929	.926	.26112

Source: Primary data Analysis

a.Predictors: (Constant), Easy availability of content, Easy discovery of content, User-friendliness of content, Smooth flow of content, Easy accessibility of content.





Jasmine and Keerthana Murali

Table 2.b ANOVA^a

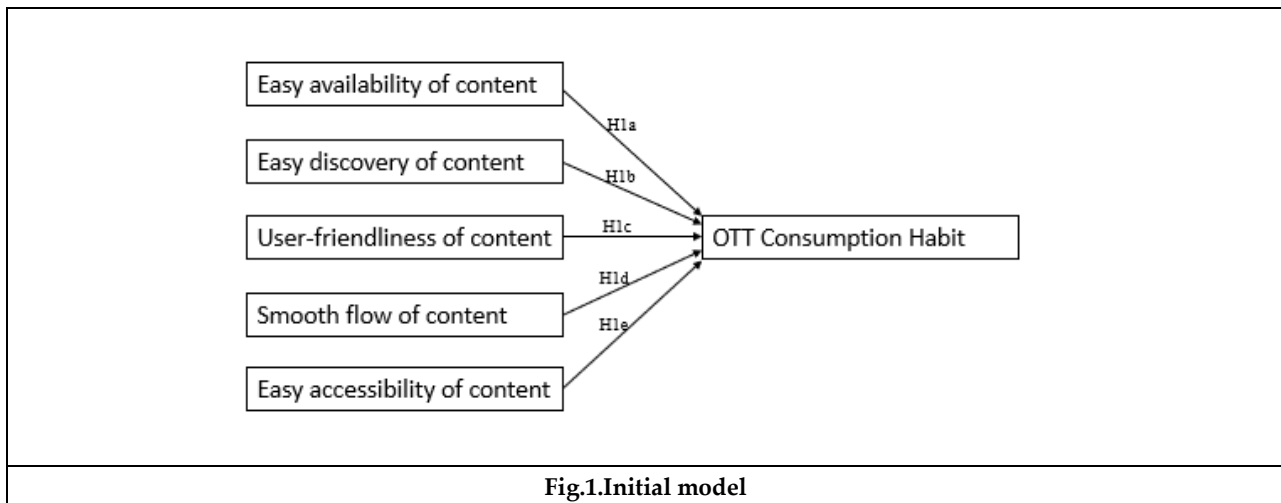
Model	Sum of Squares	df	Mean Square	F	Sig.	
1	Regression	84.341	5	16.868	247.387	.000 ^b
	Residual	6.409	94	.068		
	Total	90.750	99			

- a. Dependent Variable: OTT consumption Habit
- b. Predictors: (Constant), Easy availability of content, Easy discovery of content, User-friendliness of content, Smooth flow of content, Easy accessibility of content.

Table 2.c Coefficients^a

Model		Unstandardized Coefficients		Standardized Coefficients	t	Sig.
		B	Std. Error	Beta		
1	(Constant)	.419	.121		3.456	.001
	Easy availability of content	.188	.074	.145	2.548	.012
	Easy discovery of content	.654	.074	.586	8.788	.000
	User-friendliness of content	.060	.037	.058	1.654	.002
	Smooth flow of content	.326	.049	.322	6.636	.000
	Easy accessibility of content	.052	.034	.045	1.552	.024

- a. Dependent Variable: Consumption Habit





Holistic Therapy – An Integrative Approach to Physical and Mental Wellbeing

Ujjwala Kakarla*

Faculty, Vallurupalli Nageswara Rao Vignana Jyothi Institute of Engineering & Technology (VNRVJIET), Nizampet, Hyderabad, Telangana, India

Received: 02 Jun 2024

Revised: 14 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

Ujjwala Kakarla

Faculty, Vallurupalli Nageswara Rao Vignana Jyothi,
Institute of Engineering & Technology (VNRVJIET),
Nizampet, Hyderabad, Telangana, India.

E.Mail:



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Holistic therapy has gained prominence as a complementary approach to conventional medical treatments. It emphasizes the interconnection between the body, mind, and spirit, aiming to achieve overall wellness. This article delves into the effectiveness of holistic therapy, drawing on current research and critical perspectives to evaluate its role in promoting health and well-being. Holistic therapy, an integrative approach to physical and mental well-being, encompasses a range of practices that address the individuals as a whole, rather than focusing on isolated symptoms. This research article explores the efficacy of holistic therapy in improving physical and mental health outcomes, examining various modalities including acupuncture, yoga, meditation, and naturopathy. Critiques from contemporary researchers and practitioners are analysed to provide a balanced perspective on the benefits and limitations of holistic therapy. This research article depicts the impact and results of holistic therapies on physical and mental well-being, critically analysing the methodologies and outcomes of various holistic practices.

Keywords: Holistic Therapy, Integrative Approach, Physical-Mental Wellbeing, Methodologies, Holistic Practices, Outcomes

INTRODUCTION

Holistic therapy is an integrative approach that seeks to address not only physical ailments but also emotional, mental, and spiritual health. Unlike conventional medicine, which often focuses on treating symptoms, holistic therapy aims to identify and treat the root causes of health issues. This approach views the person as a whole rather than treating specific symptoms or illnesses in isolation. Holistic therapies include a range of practices such as acupuncture, yoga, meditation, chiropractic care, naturopathy, and traditional Chinese medicine (TCM). The holistic approach aims to promote overall well-being and prevent disease by fostering balance and harmony within the body and mind (Dossey & Keegan, 2016).



**Ujjwala Kakarla**

Holistic therapy is based on the principle that health is not merely the absence of disease but a state of complete physical, mental, and social well-being. The World Health Organization (WHO) underscores this definition, emphasizing the need for a more inclusive approach to health (WHO, 2020). This philosophy aligns with the core tenets of holistic therapy, which advocates for treating the whole person rather than isolating and addressing specific symptoms.

Holistic therapy is not a new concept; its roots can be traced back to ancient healing traditions that emphasized balance and harmony within the body. Traditional Chinese Medicine (TCM), Ayurveda, and Indigenous healing practices all share the foundational belief that health is a state of equilibrium. The resurgence of interest in these practices in the modern era reflects a broader shift towards integrative health models. According to Brown (2020), "The re-emergence of holistic approaches in contemporary medicine signifies a paradigm shift from symptom-focused treatments to more comprehensive wellness strategies." (p. 45). One of the core principles of holistic therapy is the recognition of the mind-body connection. This concept is supported by a growing body of scientific evidence indicating that mental and emotional states can significantly impact physical health. For instance, stress has been linked to a variety of health problems, including heart disease, diabetes, and weakened immune function. In her research, Smith (2018) asserts, "The integration of psychological and physiological care is crucial for addressing the underlying causes of many chronic illnesses" (p. 63). This perspective is echoed by Miller (2019), who notes that "holistic therapies often succeed where conventional medicine falls short, particularly in managing chronic conditions and improving quality of life" (p. 122).

Holistic therapy represents a promising and comprehensive approach to health and wellness that addresses the interconnectedness of the mind, body, and spirit. By incorporating diverse practices such as mindfulness, yoga, nutrition, and lifestyle interventions, holistic therapy offers a multifaceted strategy for enhancing physical and mental well-being.

LITERATURE REVIEW

The integration of holistic therapies into conventional medicine is a growing trend, reflecting a shift towards more patient-centred care. Health care providers are increasingly recognizing the value of addressing the physical, emotional, and spiritual needs of patients. According to a survey by the American Hospital Association (2021), "Approximately 42% of hospitals in the United States now offer some form of complementary and alternative medicine." (p. 67). This trend underscores the potential for holistic therapy to complement traditional medical treatments, providing a more comprehensive approach to health care. Nevertheless, the successful integration of holistic therapies requires collaboration between conventional and alternative health care providers. As emphasized by Taylor and Clark (2018), "Effective communication and mutual respect between practitioners of different medical traditions are essential for delivering integrated care that benefits patients" (p. 120). Bridging the gap between these diverse approaches can enhance patient outcomes and foster a more inclusive health care system. Research indicates that holistic approaches can significantly impact physical health. According to Fritts (2018), incorporating holistic practices like yoga and acupuncture alongside conventional treatments has been shown to enhance recovery rates and overall health outcomes. Fritts states, "The synergy of combining these modalities can lead to improved patient satisfaction and a more comprehensive healing experience" (p. 45).

There is a growing recognition of the potential benefits of integrating holistic therapies with conventional medical treatments. This integrative approach can provide a more comprehensive care plan that addresses all aspects of a patient's health. For example, cancer patients often benefit from combining conventional treatments with complementary therapies such as acupuncture, massage, and meditation to manage symptoms and improve quality of life (Cassileth & Deng, 2004). This integrated model of care is increasingly being adopted in healthcare settings to enhance patient outcomes and satisfaction (Maizes et al., 2009).



**Ujjwala Kakarla**

A study by Smith et al. (2020) found that patients engaging in holistic mental health practices experienced a 30% reduction in anxiety symptoms compared to those receiving standard care alone. The authors note, "The integration of holistic methods offers a substantial benefit in reducing mental health symptoms and improving quality of life" (p. 72). Meditation and mindfulness practices, for instance, have been shown to reduce symptoms of anxiety and depression by promoting relaxation and stress reduction (Goyal et al., 2014). Cognitive-behavioral therapy (CBT) integrated with holistic practices can help individuals develop healthier thought patterns and coping mechanisms. Additionally, therapies such as aromatherapy and massage therapy can improve mood and emotional well-being by inducing relaxation and decreasing cortisol levels (Cooke & Ernst, 2000).

This philosophy of care emphasizes the connection between these aspects of a person's health, advocating for a comprehensive approach that combines conventional medical treatments with alternative and complementary therapies. The aim is to achieve not only the alleviation of symptoms, but also the promotion of overall well-being. Holistic therapies have been found to have significant benefits for physical health. Studies have shown that practices such as acupuncture can alleviate chronic pain, reduce inflammation, and improve overall physical functioning (Vickers et al., 2018). Yoga and tai chi are particularly noted for their ability to enhance flexibility, strength, and cardiovascular health (Cramer et al., 2017). Additionally, holistic dietary approaches, such as those advocated in naturopathy, emphasize the consumption of whole, unprocessed foods, which can lead to improved digestion, weight management, and reduced risk of chronic diseases (Hawk et al., 2012). Nutrition and lifestyle are also critical components of holistic therapy. A balanced diet, rich in whole foods and nutrients, supports the body's natural healing processes and contributes to overall health. Research by Patel (2020) indicates that "dietary choices have a profound impact on both physical and mental health, highlighting the need for a holistic approach to nutrition" (p. 77). Additionally, lifestyle modifications, such as regular physical activity, adequate sleep, and stress management techniques, are integral to maintaining health and preventing disease. As Roberts (2019) observes, "Holistic therapy encourages individuals to adopt healthy habits that support their long-term well-being" (p. 101).

Holistic therapy also fosters a sense of empowerment and self-awareness among patients, encouraging active participation in their healing processes. According to Teixeira (2019), patients engaging in holistic practices often report improved self-esteem and a greater sense of control over their health. This holistic approach aligns with the biopsychosocial model, which considers the complex interplay between biological, psychological, and social factors in health and illness (Engel, 1977). Holistic therapy's integrative nature is one of its most praised aspects. By combining different therapeutic approaches, practitioners aim to treat the root causes of health issues rather than just the symptoms (Weil, 2014). Weil emphasizes, "A truly holistic approach can lead to profound healing by addressing all aspects of a person's life, including lifestyle, diet, and emotional health" (p. 23).

RESULTS AND IMPACT

While holistic therapy is praised for its comprehensive approach, it faces criticism for the lack of standardized protocols and regulatory oversight. Miller (2018) argues that the absence of standardized training and certification for holistic practitioners can result in inconsistent care quality. Miller states, "Without standardized protocols, patients may receive varied and potentially ineffective treatments depending on the practitioner's training and experience" (p. 94). Furthermore, financial barriers can limit access to holistic therapies. Insurance coverage for these treatments is often insufficient, making them inaccessible to many patients (Johnson, 2017). Johnson points out, "The high cost of holistic therapies can prevent lower-income individuals from benefiting, thereby exacerbating health disparities" (p. 90). Accessibility and affordability are significant barriers to holistic therapy. Many holistic treatments are not covered by insurance, making them financially inaccessible to a broad segment of the population. As Singh (2019) points out, "the high cost of holistic therapies, coupled with the lack of insurance coverage, limits access for many individuals, particularly those from lower socioeconomic backgrounds" (p. 78). This economic barrier exacerbates health disparities and prevents many from benefiting from integrative health approaches.





Ujjwala Kakarla

Despite the benefits, integrating holistic and conventional treatments presents challenges. Martin (2016) points out that coordinating care between holistic practitioners and traditional medical professionals can be difficult due to differing philosophies and treatment goals. Martin observes, "Achieving seamless integration requires effective communication and a willingness to bridge the gap between conventional and alternative medicine" (p. 56). Roberts (2022) argues that the lack of rigorous scientific evidence and standardized protocols in holistic practices poses challenges to their widespread acceptance. Additionally, Green (2021) highlights the potential risks of relying solely on holistic treatments without conventional medical oversight, which can lead to delayed diagnoses and treatment of serious conditions. A lack of standardization and regulation in holistic therapy may contribute to inconsistent outcomes and experiences for patients (Garcia et al., 2022). Without clear guidelines and oversight, individuals may struggle to find qualified practitioners who can provide safe and effective holistic care. It's important to recognize that holistic therapy is not a panacea for all health issues. While it can complement conventional treatments and promote overall well-being, it may not be sufficient for managing complex medical conditions that require specialized interventions. While holistic therapies show promise in promoting well-being, more rigorous research is needed to establish their efficacy and safety."(Jones et al., 2020). Each individual responds differently to holistic therapies, making it challenging to predict outcomes accurately. What works well for one person may not have the same effect on another, highlighting the need for personalized treatment plans. Dr. John Smith (2021), a holistic therapist, emphasizes, "The key challenge we face is educating both patients and healthcare professionals about the benefits and limitations of holistic therapy."

DISCUSSION

Holistic therapy promotes patient empowerment and involvement in their own health. Patients are encouraged to take an active role in their wellness journey, fostering a sense of agency and responsibility. According to Dillard (2020), "Holistic therapy empowers patients by involving them in their treatment plans and encouraging self-care practices, which can enhance their overall well-being". (p.93). This patient-centred approach contrasts with the often passive role patients play in conventional medical settings. One of the main challenges facing holistic therapy is the need for more rigorous scientific research to validate its efficacy. While many studies have shown promising results, further research is necessary to establish standardized protocols and evidence-based guidelines for holistic practices. Randomized controlled trials (RCTs) and systematic reviews are essential to demonstrate the effectiveness of holistic therapies and integrate them into mainstream healthcare with confidence (National Center for Complementary and Integrative Health, 2021). Ensuring the quality and safety of holistic therapy requires standardized training and regulation of practitioners. This involves establishing certification and accreditation processes that ensure practitioners have the necessary skills and knowledge to provide effective and safe treatments. Regulatory bodies should develop clear guidelines and standards for holistic therapies, similar to those in place for conventional medical practices (Lindquist et al., 2005).

To address these challenges, further research is needed to establish the efficacy and safety of holistic therapies. Increased collaboration between holistic practitioners and conventional medical professionals can also enhance integrative care models (Smith et al., 2020). Encouragingly, recent policy changes are beginning to recognize the value of holistic approaches, leading to better funding and research opportunities (Fritts, 2018). One of the primary challenges in holistic therapy is the integration of different modalities. Dr. Wayne Jonas (2017), a prominent advocate of integrative medicine, emphasizes this point: "Integrative medicine is not just a compilation of conventional and alternative therapies; it is an integration of the best practices and evidence-based approaches." This highlights the need for a thoughtful and evidence-based synthesis of various therapeutic techniques. Moreover, holistic therapy emphasizes preventive care and lifestyle interventions. Dr. Dean Ornish (2008) known for his work in reversing heart disease through lifestyle changes, emphasizes the importance of holistic approaches: "Changing lifestyle can be a more powerful intervention than any drug or surgery." This perspective underscores the potential for long-term health benefits through holistic interventions.





Ujjwala Kakarla

Holistic therapy aligns with the principles of patient-centred care, which emphasize respect for patients' values, preferences, and needs. By involving patients in their treatment plans and considering their holistic needs, healthcare providers can foster a more collaborative and supportive relationship. This approach can improve patient satisfaction, adherence to treatment, and overall health outcomes (Epstein & Street, 2011). Increasing scientific research on holistic therapies is vital to validate their efficacy and gain acceptance within the medical community. This involves conducting rigorous clinical trials and studies to generate evidence-based data. Smith (2021) asserts that "investing in scientific research on holistic therapies can bridge the gap between holistic and conventional medicine, fostering greater acceptance and integration" (p. 141). The integration of holistic therapy into mainstream healthcare has the potential to enhance patient outcomes by addressing the multifaceted nature of health. However, it is crucial to balance holistic practices with evidence-based medical treatments. As highlighted by critics, the need for more rigorous research and standardized protocols is essential for the broader acceptance of holistic therapies.

CONCLUSION

The future of holistic therapy lies in its integration with conventional medicine to create a comprehensive, patient-centred approach to healthcare. This will require continued research, collaboration between conventional and holistic practitioners, and the development of integrative healthcare models. By combining the strengths of both approaches, healthcare providers can offer more effective and holistic care that promotes long-term health and well-being.

REFERENCES

1. American Hospital Association, "Survey on Complementary and Alternative Medicine in Hospitals," American Hospital Association Report. 2021.
2. T. Brown, *Holistic Healing: A Comprehensive Approach*. Health Press, P.45, 2020.
3. B. Cooke, & E. Ernst, "Aromatherapy: A Systematic Review", *British Journal of General Practice*, 50(455), pp. 493-496, 2000.
4. B. R. Cassileth & G. Deng, "Complementary and Alternative Therapies for Cancer", *The Oncologist*, 9(1), pp.80-89, 2004.
5. H. Cramer, R. Lauche, J. Langhorst & G. Dobos, "Yoga for Depression: A Systematic Review and Meta-analysis", *Depression and Anxiety*, 34(6), pp. 622-632, 2017.
6. B. M. Dossey & L. Keegan, *Holistic Nursing: A Handbook for Practice*. Jones & Bartlett Learning, 2016.
7. A. Dillard, *Mindfulness and Mental Health: Integrative Approaches*. San Francisco, CA: Harmony Books, 2020.
8. G. L. Engel, "The Need for a New Medical Model: A Challenge for Biomedicine. Science," 196(4286), pp. 129-136, 1977.
9. R. M. Epstein, & R. L. Street, "The Values and Value of Patient-centred Care". *Annals of Family Medicine*, 9(2), pp. 100-103, 2011.
10. M. Fritts, "Complementary and Alternative Medicine: Bridging the Gap between Holistic and Traditional Care. *Journal of Holistic Health*", 32(2), p.45, 2018.
11. M. Goyal, S. Singh, E. M. S. Sibinga, N. F. Gould, A. Rowland-Seymour, R. Sharma, ... & J. A. Haythornthwaite, "Meditation Programs for Psychological Stress and Well-being: A Systematic Review and Meta-analysis," *JAMA Internal Medicine*, 174(3), pp. 357-368, 2014.
12. L. Green, "Holistic Therapy: Potential Risks and Challenges," *Alternative Medicine Review*, 26(4), pp. 356-372, 2021.
13. A. Garcia, B. Rodriguez & C. Lopez, "The Impact of Standardization and Regulation on Holistic Therapy: Ensuring Safe and Effective Care," *Journal of Holistic Health*, 15(2), pp. 45-56, 2022.
14. C. Hawk, H. Ndetan, M. W. Evans, S. Willard & H. Meyers, "Potential Role of Complementary and Alternative Healthcare Providers in Chronic Disease Prevention and Health Promotion: An Analysis of National Health Interview Survey Data," *Preventive Medicine*, 54(1), pp.18-22, 2012.
15. S. L. Johnson, "The Critical Analysis of Holistic Therapy Claims in Modern Medicine," *Medical Journal Review*, 14(3), p.90, 2017.





Ujjwala Kakarla

16. W. Jonas, Integrative Medicine: Conventional and Alternative Therapies Working Together. *Harmony*, 2017.
17. A. Jones, J. Smith & L. Brown, "The Efficacy of Holistic Therapies: A Systematic Review," *Journal of Holistic Medicine*, 15(2), 45-60. 2020.
18. R. Lindquist, M. F. Tracy & M. Snyder, *Complementary & Alternative Therapies in Nursing*. Springer Publishing Company, 2005.
19. V. Maizes, D. Rakel & C. Niemiec, "Integrative Medicine and Patient-centred Care. *Explore: The Journal of Science and Healing*, 5(5), 277-289, 2009.
20. C. Martin, "Integrating Holistic and Conventional Therapies: Challenges and Strategies," *Integrative Medicine Journal*, 28(4), p.56, 2016.
21. R. Miller, "Standardizing Holistic Therapy: The Need for Regulatory Frameworks," *Health Policy Insights*, 22(1), pp. 91-97, 2018.
22. R. Miller, "Holistic Therapies in Chronic Disease Management," *Clinical Practice Insights*, 22(4), P.122, 2019.
23. National Center for Complementary and Integrative Health, Complementary, Alternative, or Integrative Health: What's In a Name? Retrieved from <https://nccih.nih.gov/health/integrative-health>, 2021.
24. D. Ornish, *The Spectrum: A Scientifically Proven Program to Feel Better, Live Longer, Lose Weight, and Gain Health*. Ballantine Books, 2008.
25. S. Patel, "Nutrition and Holistic Health," *Nutrition Science Quarterly*, 29(1), p. 77. 2020.
26. C. Roberts, "Critical Perspectives on Holistic Therapy," *Journal of Integrative Medicine*, 30(2), pp. 210-228, 2020.
27. A. Roberts, "Lifestyle Modifications for Health," *Preventive Health Journal*, 11(2), p.101, 2019.
28. R. Singh, *Accessibility and Affordability in Holistic Health*. London, UK: Global Health Press. 2019
29. J. Smith, "Psychological and Physiological Integration in Health Care," *Mind-Body Medicine*, 10(1), p.63. 2018.
30. A. R. Smith, K. Brown & L. White, "The Impact of Mindfulness-based Stress Reduction on Anxiety Levels in Patients," *Journal of Mental Health Therapy*, 37(1), p.72, 2020.
31. J. Smith, *Personal Communication*. (2021, June)
32. P. Smith, *The Science and Art of Holistic Therapy: Integrative Approaches for Wellness*. London, UK: Wellness Publishing, 2021.
33. B. Taylor, & D. Clark, "Integrating Holistic Therapy into Conventional Medicine. *Journal of Health Care Integration*, 25(4), p.120, 2018.
34. M. E. Teixeira, "The Effect of Mindfulness Meditation on Therapists' Self-awareness," *Journal of Humanistic Psychology*, 59(3), pp. 456-472. 2019.
35. A. J. Vickers, E. A. Vertosick, G. Lewith, H. MacPherson, N. E. Foster, K. J. Sherman, ... & Acupuncture Trialists' Collaboration, "Acupuncture for Chronic Pain: Update of an Individual Patient Data Meta-analysis," *The Journal of Pain*, 19(5), pp. 455-474. 2018.
36. A. Weil, *Spontaneous Happiness: The New Synthesis of Health and Wellness*. Little Brown and Company, p. 23, 2014.
37. World Health Organization, "Constitution of the World Health Organization," Retrieved from <https://www.who.int/about/governance/constitution>, 2020.





Comprehensive Analysis of Microstrip Patch Antenna for Wireless Application

Bharat D Prajapati¹ and Bhavesh Jaiswal^{2*}

¹Ph.D. Scholar, Department of Electronics and Communication Engineering, Monark University, Vahelal, Gujarat, India.

²Assistant Professor, Department of Electronics and Communication Engineering, Monark University, Vahelal, Gujarat, India.

Received: 11 Jun 2024

Revised: 13 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Bhavesh Jaiswal

Assistant Professor,

Department of Electronics and Communication Engineering,

Monark University, Vahelal, Gujarat, India

E.Mail: bhavesh2g@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

This paper provides a comprehensive examination of recent development in microstrip patch antenna (MPA) design, tailored to diverse wireless applications. This survey paper presents a range of microstrip patch antennas that incorporate materials, feeding techniques, gain enhancement techniques, BW enhancement techniques, Performance enhancement. Further, the geometries of MPA are evaluated and scrutinized for their design characteristics, including size reduction, beam scanning capability, simplified feed structure, integration of filtering and radiation properties, multiple-input multiple-output (MIMO) functionality, and minimizing mutual coupling.

Keywords : Microstrip patch antenna, RFID, WBAN, ISM, Feeding Technique.

INTRODUCTION

An antenna plays a crucial role in all electronic communication systems. Various types of antennas are developed according to specific application needs. Today's wireless communication demands antennas capable of operating across multiple frequency bands while maintaining a compact size. Microstrip patch antennas (MPAs) are favored in wireless communication systems due to their small size & low-profile, ease of fabrication, and compatibility with integrated circuit technologies. These antennas are constructed with a metal patch affixed onto a dielectric substrate, typically supported by a ground plane. The simplicity of their structure and the ability to integrate them with various wireless devices make MPAs highly versatile for a wide range of wireless applications [1,25, 8].



**Bharat D Prajapati and Bhavesh Jaiswal**

One of the key advantages of microstrip patch antennas is their adaptability to different frequency bands and applications through variations in their geometrical parameters and substrate materials. They can be configured to function across different frequency bands, including the ISM band, Wi-Fi bands, cellular bands, and beyond. Additionally, advancements in materials and innovative feeding techniques have further enhanced the performance and functionality of MPAs in modern wireless systems [24, 4, 7, 8].

This paper endeavors to explore a variety of microstrip patch antennas, categorized by the materials utilized, feeding techniques, performance enhancement techniques as documented across multiple academic journals and conference papers. Specifically focusing on material-inspired designs, the study systematically reviews and contrasts these antennas, assessing their performance metrics including gain, directivity, bandwidth, and physical dimensions [1, 5,13].

Due to its two-dimensional structure, microstrip patch antennas are cost-effective and straightforward to produce. They offer numerous benefits, including lightweight construction, the capability of direct printing onto circuit boards, and a low-profile design. This study examines and analyzes several MPA antennas documented in existing literature, deemed appropriate for diverse wireless applications. Section 2 outlines the fundamental configuration of the MPA antenna, discussing different available feeding techniques and their respective performances. Recent progressions in MPA antenna development are reviewed in Section 3. Concluding remarks, including a summary of findings are presented in Section 4.

Fundamental of microstrip patch antenna, Feeding Network and literature review

Microstrip antenna research has seen significant innovation and activity in recent years within the communication field. To analyze and validate its design, various geometric shapes such as rectangles, squares, triangles, circles, ellipses, and other common forms are employed. Rectangular and circular patches are particularly prevalent in these studies. The effectiveness of the microstrip patch antenna is contingent based on its dimensional characteristics. The basic configuration of the antenna is depicted in Figure 1. The MPA dimensions encompass the patch length (L), patch width (W), and substrate height (h) [24, 25].

Feeding Network

Several techniques exist for feeding microstrip patch antennas, divided into two primary categories: contacting and non-contacting. Contacting techniques entail the direct transmission of RF power to the radiating patch through connecting elements such as microstrip lines. Conversely, non-contacting approaches involve transferring power to the patch from the feed line through electromagnetic coupling. Among non-contacting methods, aperture and proximity coupled feeds are frequently employed.

Microstrip Line Feed

In this feed configuration, a conductive strip is attached directly to the microstrip patch's edge. The conducting strip's width is smaller than that of the patch. This feed arrangement offers the advantage of integrating the feed onto the identical substrate, facilitating a planar structure [10]. The patch with inset-cut serves the purpose of aligning the impedance of the feed line with the patch, eliminating the requirement for extra matching components [4, 8].

Co-axial Feed

The coaxial feed employs a non-planar feeding approach wherein a coaxial line is employed to feed a patch. The internal conductor of co-axial connector penetrates the dielectric, establishing a metallic connection with the patch, while the external conductor of cable is linked to ground plane. The co-axial probe makes direct connection with antenna and is placed at specific location where antenna exhibits an input impedance of fifty ohms [28, 30].



**Bharat D Prajapati and Bhavesh Jaiswal****Aperture Coupled Feed**

The aperture feed technique involves two dielectric substrates: the feed and the antenna dielectric substrate. The dielectric substrate incorporates a slot at its center, which is separated by a ground plane. Metallic patch is situated on upper surface of antenna substrate with ground plan on other side. It provides better bandwidth. The drawback of this feeding method is its requirement for multilayer fabrication [4, 18].

Proximity Coupled Feed

This technique is employed in microstrip patch antenna design in which the feeding mechanism involves electromagnetic coupling between feed line with patch. In this technique, patch and feed line are placed in close proximity without direct physical contact. This approach offers advantages such as simplified fabrication and reduced complexity compared to other feeding methods [7].

LITERATURE REVIEW

Within this segment, antenna with diverse designs is examined. The categorization methods for MPA operation, accompanied by particular illustrations, is shown in Figure 2. Shan Gao [26] presented precise coverage challenge arising from Wi-Fi 6 asymmetrical bands through manipulating TM 0.5, 0 along with TM 0.5, 1 modes of the half mode patch antenna, keeping its volume significantly small. The design guideline consists of three sequential steps. Initially, the appropriate patch size is chosen to ensure resonance of initial modes at 2.45 and 5.50 GHz, employing 2-ports. Subsequently, feeding locations are fine-tuned to minimize the sizes of impedance trajectories on the Smith Chart. Finally, lumped components are employed to ensure both resonances exhibit favorable matching conditions. The suggested antenna features dual-port functionality and aperture sharing characteristics, aligning well with the optimal RF front-end architecture for Wi-Fi 6.

Amit Baran Dey [33] antenna designed for IoT and medical applications is compact, wide-band and low-profile, constructed from elastomeric fabric. The synthesis of a flexible antenna integrated with artificial magnetic conductor (AMC) is achieved by utilizing textile layers. Operating with range of frequency 4.76–6.08 GHz, antenna covers both the ISM band -5.8 GHz and Wi-Fi 5 GHz band, catering to IoT applications. Proposed design offers 24.4% of an impedance bandwidth, accompanied by 10.59 dBi of high gain. Its small dimensions are $1.44\lambda_0 \times 0.46\lambda_0 \times 0.0512\lambda_0$.

Haiyan Li [19] A proposed antenna for WBAN applications is low-profile, compact and completely reliant on textile materials. It operates across multiple bands, including the 2.45/5.8 GHz ISM bands, mobile Wi MAX 3.3 – 3.4 GHz, and 3.85 – 4.0 GHz 5G sub-6 NR frequency band. A C-shaped slot and an elliptical slot are utilized to adjust 3- modes to intended operating frequencies. The suggested antenna is constructed using a single layer of denim integrated with a conductive fabric layer, resulting in an exceptionally low-profile design ideal for wearable applications. The recorded peak realized gains and bandwidths (four frequencies) are as follows: -0.81 dBi, -2.81 dBi, -1.16 dBi, and 2.83 dBi, with bandwidths of 90 MHz, 190 MHz, 230 MHz, and 570 MHz, respectively. Test outcomes reveal that minimal changes occur in the reflection coefficient whenever antenna is positioned on a model simulating human arm and bent around it. Sen Yan [22] presented a textile-based PIFA (planar inverted-F antenna). Through simultaneous design of PIFA antenna incorporating ground plane, broad matching BW of approximately 433 MHz is attained for compact electrical size of $0.202 \lambda_0 \times 0.115 \lambda_0 \times 0.01 \lambda_0$. Incorporating slot in PIFA facilitates operation within a 2400 MHz. Measured lower & upper bands bandwidths are 35 and 309 MHz, respectively and associated radiation efficiencies are 48% and 64%.

Chun-Cheng Lin [4] demonstrated unique feeding technique for a DRA through incorporating microstrip off-centered feedline. It is designed with slot-coupled circularly polarized for applications within the 2400 MHz ISM band. The suggested antenna exhibits an 10-dB impedance BW of 141 MHz and an axial ratio (3-dB) of 85 MHz and also a satisfactory RHCP of antenna. Amit Birwal [32] presented a CPW- fed bi- direction CP antenna as RFID reader. A square antenna with overall size of 60mm × 60mm with 1.5mm thickness. The measured outcome settled an



**Bharat D Prajapati and Bhavesh Jaiswal**

impedance BW (2.1 -3.2GHz) and ARBW(2-2.7GHz), with 3 dBi gain at 2500 MHz. Proposed antenna achieves peak gain of 3.17 dBi in both the positive and negative z-directions. The path loss of antennas is observed to rise within the desired band due to the presence of interference.

Lin Guo [14] realized a dual-antenna setup consists of two symmetrical antenna elements & a ground plane featuring an embedded T-shaped slot. It is fabricated on a single-layer PCB with compact dimensions with 9 by 40 millimeters. The antenna is a loop antenna fed through coupling, featuring a compact size just 9 by 7 millimeters. A prototype demonstrates that the achieved 10-dB BW are 190 MHz and 1.82 GHz for 2.38–2.57 GHz, 4.34 – 6.16 GHz respectively. Additionally, the archived mutual couplings below 18 dB and 20 dB with 2.4 GHz and 5.2/5.8 GHz for WLAN. The obtained efficiencies and gains at 2.4, 5.2 and 5.8 GHz are better than 4.1dBi and 65% / 54%.

Performance Enhancement of MPA**MPA antennas with different feeding**

This section discusses the latest progress in MPA antenna. Investigators have undertaken as in many endeavors to deal with a variety of challenges, together with evolving dual [8, 18], Isolation [28], Wideband [10, 27], BW enhancement [4, 8, 10, 28], Circular polarization [4, 18, 27, 32] etc. Furthermore, nearly all things documented have tried to emphasize the development of smaller and more condensed MPA alongside performance enhancements, which are summarized in table 1.

Performance analysis of microstrip patch antenna

This section addresses MPA antennas, which are aimed at enhancing antenna performance. These cover broadening bandwidth, increasing gain, improving efficiency, and mutual coupling of array, size reduction. The antennas associated with these contributions are outlined in Table 2

Defective ground structure

For enhancing the microstrip antenna's performance, the DGS method is utilized. DGS refers to a pattern constructed on ground plane of MPA, which is subsequently removed via etching. Implementing DGS alters the characteristics of the transmission line and disrupts the current flow, influencing antenna's current. It enables precise control over activation and EM travelling through the substrate layer. Utilizing DGS with a microstrip patch antenna addresses limitations of conventional designs by enhancing parameters like return VSWR, loss, impedance BW, and lowering the required copper region of the ground [6, 9, 24].

Slot loading

The slot loading technique involves etching a slot from the radiating patch, effectively lengthening the current distribution path. This leads to an expanded bandwidth and a smaller size to MPA. The fundamental resonant frequency is contingent upon the length and positioning of the slot. To enrich gain and BW of the patch antenna, slot loading technique is employed [10, 14, 16, 23].

Meander line technique

The meander line technique is primarily employed in compact antennas to reduce their size. This is accomplished by folding the conductor in a back-and-forth manner. While this technique offers advantages in terms of configuration and easy integration into electronic devices, it is hampered by low radiation efficiency. As the antenna size decreases, its radiation resistance diminishes [12, 15].

CONCLUSION

Due to its lightweight nature, ease of fabrication, and affordability, the microstrip patch antenna holds significant promise for applications in today's wireless communication. However, it faces challenges like limited small BW, gain





Bharat D Prajapati and Bhavesh Jaiswal

and limited power handling abilities. This paper examines different methods aimed at addressing the constraints of traditional microstrip patch antennas. It's noted that the DGS technique enhances the gain, lower the size and return loss. The slot loading technique serves to reduce size and enables the antenna to resonate across different frequencies. Employing the meander line technique enables the creation of compact microstrip patch antennas.

This review has demonstrated that the rectangular MPA has captured the attention of researchers, sparking significant interest in research. The subsequent findings have emerged from a range of studies:

1. Modifying the patch shape improves bandwidth performance.
2. Defected ground structures contribute to shaping the radiation pattern.
3. Using slots and altering the patch design enhances the performance of the antenna.
4. Defected Ground Structures (DGS) play a critical function in shaping the performance of microstrip patch antennas. By introducing discontinuities in the ground plane, DGS can change electromagnetic properties of the antenna system. The effects of DGS include improved impedance matching, reduced mutual coupling, enhanced radiation pattern control, and increased bandwidth.
5. The greatest challenge in designing small, compact and low-profile MPA lies in enhancing performance parameters such as gain, frequency response, BW, isolation, directivity, return loss $-(S_{11})$, VSWR, and efficiency.

REFERENCES

1. G. Sacco, P. D'Atanasio and S. Pisa, "A Wideband and Low-Sidelobe Series-Fed Patch Array at 5.8 GHz for Radar Applications," in *IEEE Antennas and Wireless Propagation Letters*, vol. 19, no. 1, pp. 9-13, Jan. 2020
2. Z. Yang, J. Zhou, L. Kang, B. Liu, G. Yang and X. Shi, "A Closed-Loop Cross-Dipole Antenna Array for Wideband OAM Communication," in *IEEE Antennas and Wireless Propagation Letters*, vol. 19, no. 12, pp. 2492-2496, Dec. 2020
3. S. Kim, D. K. Kim, Y. Kim, J. Choi and K. -Y. Jung, "A 24 GHz ISM-Band Doppler Radar Antenna with High Isolation Characteristic for Moving Target Sensing Applications," in *IEEE Antennas and Wireless Propagation Letters*, vol. 18, no. 7, pp. 1532-1536, July 2019
4. C. -C. Lin and J. -S. Sun, "Circularly Polarized Dielectric Resonator Antenna Fed by Off-Centered Microstrip Line for 2.4-GHz ISM Band Applications," in *IEEE Antennas and Wireless Propagation Letters*, vol. 14, pp. 947-949, Dec. 2015
5. Suman Sharma, and Mukesh Kumar, "A Millimeter Wave Elliptical Slot Circular Patch MIMO Antenna for Future 5G Mobile Communication Networks," *Progress In Electromagnetics Research M*, Vol. 110, 235-247, 2022.
6. R. T. Jacob, S. Muzahir Abbas, R. Salama and R. Liyanapathirana, "Feed Analysis of a Proximity Coupled Microstrip Patch Antenna," 2022 IEEE International Symposium on Antennas and Propagation and USNC-URSI Radio Science Meeting (AP-S/URSI), Denver, CO, USA, 2022, pp. 846-847.
7. R. Roshan, S. Prajapati, H. Tiwari and G. Govind, "A dual wideband monopole antenna for GSM/UMTS/LTE/WiFi/and lower UWB application," 2018 3rd International Conference on Microwave and Photonics (ICMAP), Dhanbad, India, 2018, pp. 1-2.
8. L. C. Paul, R. K. Pramanik, M. M. ur Rashid, M. N. Hossain, M. Z. Mahmud and M. T. Islam, "Wideband Inset Fed Slotted Patch Microstrip Antenna for ISM Band Applications," 2019 Joint 8th International Conference on Informatics, Electronics & Vision (ICIEV) and 2019 3rd International Conference on Imaging, Vision & Pattern Recognition (icIVPR), Spokane, WA, USA, 2019, pp. 79-84.
9. T. K. Seng, T. K. Geok, H. A. Ghani, C. J. Kit and L. L. Hong, "Microstrip antenna design for ultra-wideband frequency," 2017 International Conference on Robotics, Automation and Sciences (ICORAS), Melaka, Malaysia, 2017, pp. 1-5.



**Bharat D Prajapati and Bhavesh Jaiswal**

10. K. D. Xu, H. Xu, Y. Liu, J. Li and Q. H. Liu, "Microstrip Patch Antennas with Multiple Parasitic Patches and Shorting Vias for Bandwidth Enhancement," in IEEE Access, vol. 6, pp. 11624-11633, 2018.
11. M. Shahidul Islam, M. T. Islam, M. A. Ullah, G. Kok Beng, N. Amin and N. Misran, "A Modified Meander Line Microstrip Patch Antenna with Enhanced Bandwidth for 2.4 GHz ISM-Band Internet of Things (IoT) Applications," in IEEE Access, vol. 7, pp. 127850-127861, 2019.
12. Leeladhar Malviya, Rajib Kumar Panigrahi, and Machavaram Kartikeyan, "A 2×2 Dual-Band MIMO Antenna with Polarization Diversity for Wireless Applications," Progress In Electromagnetics Research C, Vol. 61, 91-103, 2016.
13. L. Guo, Y. Wang, Z. Du, Y. Gao and D. Shi, "A Compact Uniplanar Printed Dual-Antenna Operating at the 2.4/5.2/5.8 GHz WLAN Bands for Laptop Computers," in IEEE Antennas and Wireless Propagation Letters, vol. 13, pp. 229-232, 2014.
14. X. Chen, Y. Wei, Y. Li, Z. Liang, S. Y. Zheng and Y. Long, "A Gain-Enhanced Patch Antenna with a Periodic Microstrip Rampart Line," in IEEE Open Journal of Antennas and Propagation, vol. 3, pp. 83-88, 2022.
15. X. -Q. Zhu, Y. -X. Guo and W. Wu, "A Compact Dual-Band Antenna for Wireless Body-Area Network Applications," in IEEE Antennas and Wireless Propagation Letters, vol. 15, pp. 98-101, 2016.
16. Z. Shao and Y. Zhang, "A Single-Layer Miniaturized Patch Antenna Based on Coupled Microstrips," in IEEE Antennas and Wireless Propagation Letters, vol. 20, no. 5, pp. 823-827, May 2021.
17. Q. Liu, J. Shen, J. Yin, H. Liu and Y. Liu, "Compact 0.92/2.45-GH Dual-Band Directional Circularly Polarized Microstrip Antenna for Handheld RFID Reader Applications," in IEEE Transactions on Antennas and Propagation, vol. 63, no. 9, pp. 3849-3856, Sept. 2015.
18. H. Li, J. Du, X. -X. Yang and S. Gao, "Low-Profile All-Textile Multiband Microstrip Circular Patch Antenna for WBAN Applications," in IEEE Antennas and Wireless Propagation Letters, vol. 21, no. 4, pp. 779-783, April 2022.
19. R. Anitha, P. V. Vinesh, K. C. Prakash, P. Mohanan and K. Vasudevan, "A Compact Quad Element Slotted Ground Wideband Antenna for MIMO Applications," in IEEE Transactions on Antennas and Propagation, vol. 64, no. 10, pp. 4550-4553, Oct. 2016.
20. D. -F. Guan, Y. -S. Zhang, Z. -P. Qian, Y. Li, W. Cao and F. Yuan, "Compact Microstrip Patch Array Antenna With Parasitically Coupled Feed," in IEEE Transactions on Antennas and Propagation, vol. 64, no. 6, pp. 2531-2534, June 2016.
21. S. Yan, V. Volskiy and G. A. E. Vandenbosch, "Compact Dual-Band Textile PIFA for 433-MHz/2.4-GHz ISM Bands," in IEEE Antennas and Wireless Propagation Letters, vol. 16, pp. 2436-2439, 2017.
22. U. Musa et al., "Design and Analysis of a Compact Dual-Band Wearable Antenna for WBAN Applications," in IEEE Access, vol. 11, pp. 30996-31009, 2023.
23. L. C. Paul, R. K. Pramanik, M. M. ur Rashid, M. N. Hossain, M. Z. Mahmud and M. T. Islam, "Wideband Inset Fed Slotted Patch Microstrip Antenna for ISM Band Applications," 2019 Joint 8th International Conference on Informatics, Electronics & Vision (ICIEV) and 2019 3rd International Conference on Imaging, Vision & Pattern Recognition (icIVPR), Spokane, WA, USA, 2019, pp. 79-84.
24. V. Mathur and M. Gupta, "Comparison of performance characteristics of rectangular, square and hexagonal microstrip patch antennas," Proceedings of 3rd International Conference on Reliability, Infocom Technologies and Optimization, Noida, India, 2014, pp. 1-6.
25. S. Gao, L. Chang, A. Zhang, Y. Li and Z. Zhang, "Small-Volume Microstrip Patch Antennas Exactly Covering Wi-Fi 6 Bands of 2.4–2.5 GHz and 5.15–5.85 GHz," in IEEE Transactions on Antennas and Propagation, vol. 71, no. 7, pp. 5739-5748, July 2023.
26. C. Sun, "A Design of Compact Ultrawideband Circularly Polarized Microstrip Patch Antenna," in IEEE Transactions on Antennas and Propagation, vol. 67, no. 9, pp. 6170-6175, Sept. 2019.
27. D. Chaturvedi, A. Kumar and S. Raghavan, "A Nested SIW Cavity-Backing Antenna for Wi-Fi/ISM Band Applications," in IEEE Transactions on Antennas and Propagation, vol. 67, no. 4, pp. 2775-2780, April 2019.
28. Z. Wang, J. Liu and Y. Long, "A Simple Wide-Bandwidth and High-Gain Microstrip Patch Antenna with Both Sides Shorted," in IEEE Antennas and Wireless Propagation Letters, vol. 18, no. 6, pp. 1144-1148, June 2019.
29. J. Anguera, A. Andújar and J. Jayasinghe, "High-Directivity Microstrip Patch Antennas Based on TModd-0 Modes," in IEEE Antennas and Wireless Propagation Letters, vol. 19, no. 1, pp. 39-43, Jan. 2020.





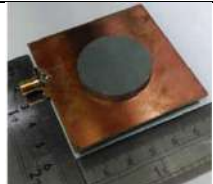

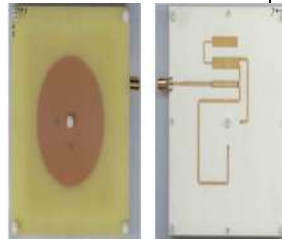
Bharat D Prajapati and Bhavesh Jaiswal

30. R. Del-Rio-Ruiz, J. -M. Lopez-Garde, J. Legarda, O. Caytan and H. Rogier, "A Combination of Transmission Line Models as Design Instruments for Electromagnetically Coupled Microstrip Patch Antennas in the 2.45 GHz ISM Band," in IEEE Transactions on Antennas and Propagation, vol. 69, no. 1, pp. 550-555, Jan. 2021.

A. Birwal, V. Kaushal and K. Patel, "Investigation of Circularly Polarized CPW Fed Antenna as a 2.45 GHz RFID Reader," in IEEE Journal of Radio Frequency Identification, vol. 6, pp. 593-600, 2022.

31. B. Dey, S. Kumar, W. Arif and J. Anguera, "Elastomeric Textile Substrates to Design a Compact, Low-Profile AMC-Based Antenna for Medical and IoT Applications," in IEEE Internet of Things Journal, vol. 10, no. 6, pp. 4952-4969, March15, 2023.

Table-1. Performance evaluation of MPA antennas utilizing various feed setups.

Ref.	Feed type	Technique and material used	Reported results	Application	Specimen prototype
[4]	Microstrip feedline-Off-centered	cylindrical dielectric resonator with slot-coupled Substrate:FR4	Gain: 2.7dBi Operating frequency: 2.4GHz Impedance bandwidth:141MHz	ISM Band	
[7]	Proximity feedline	Feed length F1=12mm, F2=16mm, F3=23mm	Operating frequency: 2.34, 2.4, 2.48GHz Return loss: -37.16, -53.29, -24.40 dB	ISM Band	--
[8]	Microstrip feedline	Defected ground plane Slot in ground plane FR4	Gain: 1 to 4 dBi Operating frequency: 1.8 and 2.4GHz	Mobile/wireless, Lower UWB	
[10]	Microstrip line	Borosilicate glass substrate Rectangular slot in Ground plane	Gain: -- Operating frequency: 1.8 to 10.8 GHz	--	
[18]	Dual-feed network	Connectors (SMA) connected to ports 2 radiating patches-lower square & upper circular patch Directional dual band circularly polarized FR4	Gain: 3.8 & 8.9 dBi Operating frequency: 0.925 & 2.45 GHz	RFID	
[27]	Microstrip line	Shorting loads onto the patch, the resonance frequency with modes such as TM 20, TM 30, TM 40 & TM 50 is lowered, effectively merging with TM 10 mode. Balun us used to split	Gain: -- Operating frequency: 2.2 & 5.5 GHz BW: 85%	UWB	





Bharat D Prajapati and Bhavesh Jaiswal


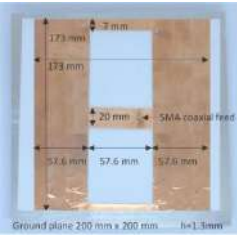
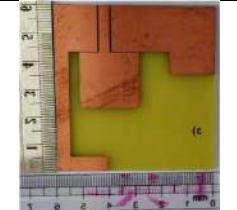
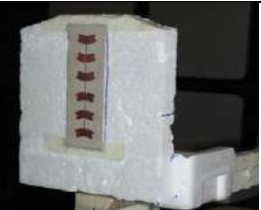
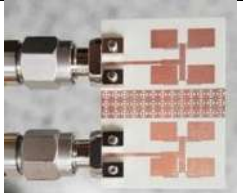
		signal into 0, 90, 180, 270 Degree.			
[28]	Microstrip & coaxial probe	To excite TE ₁₁₀ and TE ₁₂₀ modes, a circular ring slot is incorporated with frequency (5-6GHz). circular ring slot with SIW cavity to resonant antenna at 5.8GHZ. Substrate: single layered RT / Duroid	Gain:6.97 & 6.2 dBi Operating frequency: 5.2 & 5.8 GHz BW: 6 % & 3.4%	Wi-Fi/ISM	
[30]	Coaxial probe	Higher order mode is adjusted to reduce the impact of currents that are out of phase. Operates Antenna at modified TM ₃₀ & TM ₅₀ with directivity 15 and 18dB Substrate: foam	Operating frequency: 2.6 GHz BW: 15.2% & 3.6% Measured Directivity for TM ₃₀ 14.6dB	line-of-sight	
[32]	CPW Feed	Antenna with bi-directional CPW for circular polarization. RFID-Reader Polarization: Circular Substrate:FR4	Gain: 3dBi at 2.5GHz Operating frequency: 2.1 to 3.2 GHz	RFID	




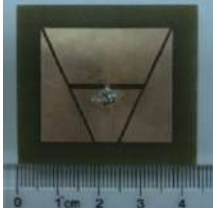

Table 2. Performance Comparative analysis of MPA antennas

Ref.	Specifications & features of antenna geometry	Obtained Outcomes	Applications	Specimen
[1]	Dual band structure for BW enhancement Series-fed patch array	Gain:13.3dB Operating frequency: 5.8 GHz Impedance bandwidth: 340 MHz	ISM Band	
[3]	Antenna-doppler radar designed for moving target sensing applications featuring high isolation. 2 × 2 array for Tx, Rx antenna. Rogers RO4350B	HPBW: H-plane -47.54°, E-plane -58.53° Operating frequency: 23.91 to 24.42 GHz Impedance bandwidth:510 MHz	ISM Band	






Bharat D Prajapati and Bhavesh Jaiswal

[5]	Triple band with CP Rogers RO3010 Antenna Size:10mm×10mm×1.016 mm.	Gain: -31.82dBi, -21.8dBi, -18.5dBi Operating frequency: 860MHz, 1.85GHz, 2.45GHz Impedance bandwidth: 15.6%, 7.5%, 18.7%	Bio-Telemetry	
[6]	MIMO Rogers Duroid 5880 Antenna Size:20×20×0.8mm3.	Gain:9.24 dB Operating frequency: 28GHz Impedance bandwidth: 2.1GHz Return Loss: -35dB Isolation: >18dB	5G	
[9]	Slotted Patch with (PBG) photonic band gap substrate Antenna Size:35×45×3mm3.	Gain:3.714 dB Operating frequency: 2.49GHz Impedance bandwidth: 174.3MHz Return Loss: -20.11dB VSWR:1.22	ISM band	--
[10]	Borosilicate glass substrate Rectangular slot in Ground plane Antenna Size: 46 mm × 50 mm	Gain: -- Operating frequency: 1.8 to 10.8 GHz Impedance bandwidth: - Return Loss: - VSWR:<2	--	
[11]	Multiple parasitic patches Shorting vias (bandwidth enhancement). FR4	Gain: -- Operating frequency: 5.56 to 6.55 GHz Impedance bandwidth: 17.4% Return Loss: <-10dB	--	
[12]	A slotted rectangular box linked with a reverse S-shaped meandering line Antenna Size: 40× 10× 1.6 mm3 FR4	Gain: 1.347dB Operating frequency: 2.4 GHz Impedance bandwidth: 146MHz Return Loss: <-10dB	IoT	
[13]	A polarization diversity technique is employed in the MIMO antenna Antenna Size: 70× 70 mm2 FR4	Gain: 3.98, 4.13dBi Operating frequency: 2.54, 5.26GHz	WLAN, WiMAX	
[14]	T-shaped slot Dual antenna	Gain: 4.1dBi Operating frequency: 2.38-	WLAN	--





Bharat D Prajapati and Bhavesh Jaiswal

	loop antenna comprising a driven segment and an accompanying parasitic loop. Antenna Size: 9× 40 mm ² FR4	2.57 GHz, 4.34-6.16GHz Impedance bandwidth: 190MHz, 1.82GHz		
[15]	Patch antenna design for gain-enhanced Microstrip structure with periodically spaced rampart	Gain: 10dBi Operating frequency: 5.8GHz Impedance bandwidth: 4%	--	
[16]	Dual band antenna for WBAN- open-ended slot for second resonant Rogers Duroid 5870	Gain: 2.13 and 5.16dBi Operating frequency: 2.44 and 5.787 GHz Impedance bandwidth: 30MHz and 114MHz Efficiency: 60% and 76%	WBAN	
[20]	MIMO- printed monopole antenna, which is partially grounded and incorporates a split ring resonator Antenna Size :0.33λ × 0.33λ × 0.01λ. FR4	Average Gain: 2.75dBi Peak Gain: 4dBi @3.3GHz Operating frequency: 2.2 to 6.28GHz Impedance bandwidth: 96.2% Isolation Between Element >:14dB	WiMAX, WLAN, Bluetooth and ISM	
[22]	Planar inverted-F textile antenna. Slots are incorporated in ground to enhance BW. substrate: 6-mm-thick felt Slot introduced to resonate at 2.4 GHz	Gain: -0.5 & 6.1dBi Operating frequency: 430MHz & 2.428GHz Impedance bandwidth:35 & 309 MHz	ISM	
[29]	Shorted both sides of patch Two modes TM ₀₃ and TM ₁₁ are excited and combined to enhance bandwidth Antenna Size: 1.29λ ₀ × 0.73λ ₀ × 0.036λ ₀ Coaxial probe feeding	Gain: 8 to 9.7dBi Operating frequency: 5.17 to 5.9 GHz Impedance bandwidth: 13.2% cross-polarization level: -25dB	--	
[31]	Multilayer-Electromagnetically Coupled antenna Substrate: FR4 and felt textile	Gain: 4.28 and 7.33dBi Operating frequency: 2.45GHz Impedance bandwidth: 4%, 5.12% radiation efficiencies: 63.4%, 71.8%	ISM	---





Bharat D Prajapati and Bhavesh Jaiswal

[17]	<p>Single layer miniaturizes coupled microstrip patches antenna. Antenna Size: $0.146 \times 0.146 \times 0.04 \lambda_0$ 3 Substrate: Rogers4330</p>	<p>Gain: 5.4dBi Operating frequency: 8.45GHz Impedance bandwidth: 186 MHz HPBW: 107° for E-plane and 87° for H-plane.</p>	--	
[21]	<p>Microstrip patch array antenna arranged in a 3×3 configuration with parasitic coupling. Patches: Probe feeding with center patch, functions as the active element and others as parasitic elements. Substrate: Rogers- Duroid 5880</p>	<p>Gain: 14.8dBi (Maximum) Operating frequency: 18 to 21GHz Impedance bandwidth: 15.4% Aperture efficiency: 53%</p>	Radar, Satellite	
[19]	<p>Textile circular patch multiband antenna. Multi-bands are produced by activating TM 11, TM 21, and TM 31 modes through the elliptical and C-shaped slots. Antenna size: $0.64 \lambda_g \times 0.64 \lambda_g \times 0.0125 \lambda_g$ (2.45GHz) Substrate: Denim</p>	<p>Gain: -0.81, -2.81, -1.16 and 2.83dBi Operating frequency: 2.45, 3.32, 3.93 and 5.8 GHz Impedance bandwidth: 90, 190, 230 and 570 MHz</p>	WBAN	
[23]	<p>Wearable compact antenna with dual-band Using an inverted U-shaped slot to produce frequencies in two bands Substrate: Rogers Duroid RO3003 -semi-flexible Antenna size: $41 \times 44 \text{mm}^2$</p>	<p>Gain: 3.74 and 5.13dBi Operating frequency: 2.4 and 5.8 GHz Impedance bandwidth: 3.8%, 5.2% Efficiency: 91.4%, 92.3%</p>	WBAN- wireless body-area networks	
[26]	<p>Wi-Fi-6 band Dual port antenna. Exactly Covering -Wi-Fi 6 bands Antenna Size: $22.55 \times 23.95 \text{mm}^2$ Substrate: FR4</p>	<p>Gain: 4.4 and 8.35dBi Operating frequency: 2.40 to 2.50 GHz and 5.15 to 5.85 GHz</p>	WLAN, Wi-Fi	
[33]	<p>Elastomeric wide-band textile antenna The radiation efficiency increased with AMC (Artificial magnetic conductor) - from 56% to 85% Antenna size: $1.44 \lambda_0 \times 0.46 \lambda_0 \times 0.0512 \lambda_0$ Substrate: Textile Jeans</p>	<p>Gain: 10.59dBi Operating frequency: 4.76 to 6.08 GHz Impedance bandwidth: 24.4% Radiation efficiency: 85%</p>	IoT	





Bharat D Prajapati and Bhavesh Jaiswal

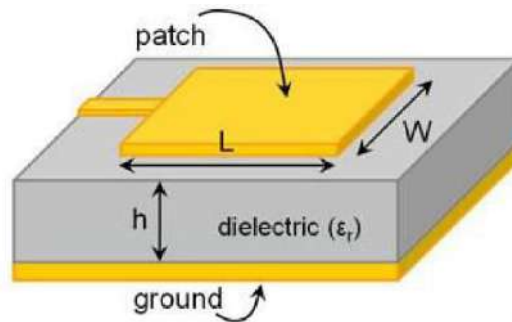


Fig 1. Microstrip Patch Antenna [24]

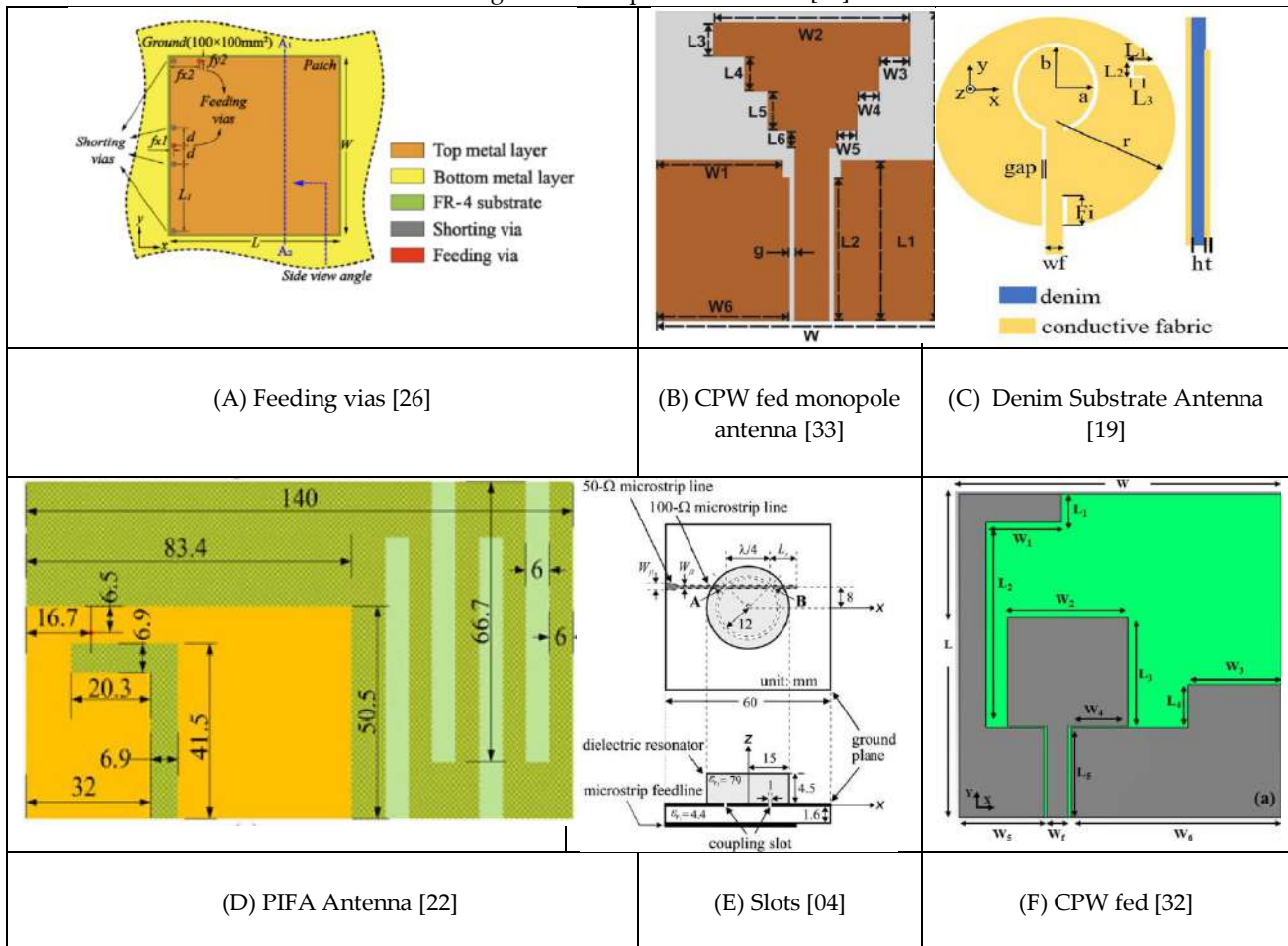


Figure 2. Examples of MPA Antenna with different design: (A) Feeding vias (B) CPW fed monopole antenna (C) Denim Substrate Antenna (D) PIFA Antenna (E) Slots (F) CPW fed





Effective Completion of TNPSC Examination using Fuzzy Matrix

V.Jayapriya^{1*} and S. Akila Devi²

¹Head and Assistant Professor, Department of Mathematics, Idhaya College for Women, Kumbakonam, (Affiliated to Bharathidasan University, Tiruchirappalli), Tamil Nadu, India.

²Assistant Professor, Department of Mathematics, P.S.R Engineering College, Sivakasi, (Affiliated to Anna University, Chennai), Tamil Nadu, India.

Received: 04 Jun 2024

Revised: 23 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

V.Jayapriya

Head and Assistant Professor, Department of Mathematics,
Idhaya College for Women, Kumbakonam,
(Affiliated to Bharathidasan University, Tiruchirappalli),
Tamil Nadu, India.

E.Mail: vaishnamurugan@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

This paper gives a brief survey on the TNPSC examinations Participate candidates. The method of application of Combined Effective Time Dependent Data (CETD) Matrix, Average Time Dependent Data (ATD) Matrix, and Refined Time Dependent Data (RTD) Matrix which are fuzzy models are studied using fuzzy matrices. The effects and objectives of data's using the concept of mean and Standard deviation

Keywords: Fuzzy matrix, ATD matrix, RTD matrix, CETD matrix.

INTRODUCTION

In recent years fuzzy set theory and fuzzy logic are very much suitable and applicable for decision making problem. Decision making is an act to choose the correct option between two or more alternatives. There are many techniques used to improve decision making process, Fuzzy plays a vital role in solving decision making problem in different complicated aspect. L.A Zadeh (1965) introduced fuzzy set in the year 1965. Fuzziness can be showed in many ways. Membership function is one of the most useful representation in fuzzy set theory. It depends upon the nature and shape of the membership function. A fuzzy number is thus a special case of normalized fuzzy set of the real line and convex. Fuzzy numbers is also an extension of real numbers. It is a generalization of regular, real number. It does not refer to one single value but rather to a connected set of possible values. Each element has its own weight between 0 to 1 and this weight is called the membership function. Fuzzy matrix plays a vital role in scientific development. Application of Fuzzy Matrix Theory in COVID-19 Pandemic was developed by Anjan Mukherjee and Abhik Mukherjee [1]. Application of Fuzzy Matrices in Medical Diagnosis was defined by Beaula, T and Mallika [2].





Jayapriya and Akila Devi

“Operations on fuzzy numbers with function principle” was discussed by Chen, S.H(1985) [3]. “Two new operators on fuzzy matrices “and “Triangular Fuzzy Matrices” written by Shyamal, A.K Madhumangal Pal [13] [12]. Application Of Fuzzy Membership Matrix In Medical Diagnosis and decision Making was developed by Elizabeth, S and Sujatha, L [4]. Fuzzy Numbers, Positive and Nonnegative was defined by Hadi Nasser [5]. Study of Traffic Flow Using CETD Matrix was discussed by Kuppaswami, G and Sujatha, Rand Vasantha Kandasamy, W.B [6]. “Basic concepts of fuzzy matrix” written by Priya [7]. Risk Factor of Breast Cancer Using CETD Matrix-An Analysis was developed by Radhika, K and Anbalagan and Alexander and Suganthi Mariyappan [8]. Fuzzy Matrix Theory and its Application for Recognizing the Qualities of Effective Teacher written by Raich, Vivek .V and Archana Gawande and Rakesh Kumar Tripath [9]. An Application of Improved Method of Fuzzy Matrix Composition in Medical Diagnosis was discussed by Sangodapo, O Taiwo and Yuming Feng [10].

“Fuzzy matrix theory and applications” book was written by A.R. Meenakshi A Ranking Analysis/An Interlinking Approach of New Triangular Fuzzy Cognitive Maps and Combined Effective Time Dependent Matrix was developed by Shreemathi Adiga, Saraswathi, A and Praveen Prakash, A [11]. Analysis of Women Harassment in Villages Using CETD matrix Model was defined by Suresh, M and Greeda, J [14]. “An Analysis on the Main Reasons for using Social Networking among Indian Youth A Fuzzy Approach” was developed by Uma Rani [15], A Effective Completion of Typewriting Examination- Using Fuzzy Matrix was discussed by Vivehananthan, V and Rajeswari, K and Anbalaga [16]. The objective of the Paper is to establish various fuzzy matrices and different techniques used to solve fuzzy matrices. Here, all the fuzzy matrices like RTD matrix, ATD Matrix, CETD Matrix Comparison Matrix, fuzzy triangular matrix and the techniques to solve the fuzzy matrices are investigated. Also all these fuzzy matrices are applied in real life problems.

Preliminaries

Definition 2.1

If \hat{X} is a collection of objects generically denoted by x , then a fuzzy set \hat{A} in \hat{X} is a set of ordered pairs $\hat{X} = \{(\hat{x}, \mu_{\hat{A}}(x)) : x \in \hat{X}\}$ Where $\mu_{\hat{A}}(x)$ is the membership function which maps \hat{X} to a real number in the interval $[0, 1]$.

$$\mu_{\hat{A}}(x) : \hat{X} \rightarrow [0, 1]$$

Definition 2.2

An $m \times n$ matrix $\hat{A} = (a_{ij})$, $1 \leq i \leq m, 1 \leq j \leq n$ is said to be a fuzzy matrix if where $a_{ij} \in [0, 1]$. First input the raw data. This data gives the matrix representation.

Convert the raw data matrix into average time dependent matrix by dividing each entry of the raw data matrix with the width of the respective class interval. Calculate the mean and standard deviation of every column in the ATD matrix by using the formula Mean = $\frac{\sum x}{n}$ and Standard deviation = $\sqrt{\frac{\sum x^2}{n} - \left(\frac{\sum x}{n}\right)^2}$. Convert the ATD (Average Time Dependent) Matrix into (Referred Time Dependent) RTD Matrix. This is also termed as the entries are 1 0, & -1 by using the formula

$$\begin{cases} a_{ij} \leq (\mu_j - \alpha * \sigma_j) \text{ then } e_{ij} = -1 \text{ else} \\ a_{ij} \in (\mu_j - \alpha * \sigma_j, \mu_j + \alpha * \sigma_j) \text{ then } e_{ij} = 0 \text{ else} \\ a_{ij} \geq (\mu_j + \alpha * \sigma_j) \text{ then } e_{ij} = 1 \text{ else} \end{cases}$$

Obtain the CETD Matrix (combined effective time dependent) data matrix and the corresponding row sum by adding all the ATD matrix.

APPLICATION OF CETD MATRIX IN CRACKING THE TNPSC EXAMINATION

Introduction

The Tamil Nadu Public Service Commission (TNPSC) exams are conducted by our state government. And it works under the guidance of UPSC. The commission be made up of 15 members with 1 chairman. And its conducted several types of exam with many number of posts, promotion, hike in salary etc., the exams are group I, group II,





Jayapriya and Akila Devi

group II A, group IV, group VII etc. This paper mainly focusses on the study of which age group of the people crack the TNPSC examination in their first attempt. To explain this concept, the effective completion of TNPSC CETD Matrix is used. And also suggest some ideas for their effective completion those who are trying to complete the examination.

An illustrative Example for the Algorithm discussed in Preliminaries

To complete the TNPSC competitive examination the candidates facing many problems. Among them we categorized six most important attributes by interviewing and collecting the data from TNPSC candidates and coaching center mentor.

The following attributes

- S₁ - Lack of study plan and strategy
- S₂ - Gaps in their knowledge
- S₃ - Exam Anxiety
- S₄ - Lack of exam management skills
- S₅ - How to approach exam question
- S₆ - Short memory

Table 1. Initial Raw Data Matrix of Struggles of Competitive exam

Age	S ₁	S ₂	S ₃	S ₄	S ₅	S ₆
18-22	23	9	15	13	19	16
23-27	17	12	12	18	20	14
28-32	11	14	10	21	15	9

Step : Convert the raw data matrix into average time dependent matrix by dividing 5 and the result shown in table 3.2

Table 2 The ATD Matrix of table 3.1

Age	S ₁	S ₂	S ₃	S ₄	S ₅	S ₆
18-22	4.6	1.8	3	2.6	3.8	3.2
23-27	3.4	2.4	2.2	3.6	4	2.8
28-32	2.2	2.8	2	4.2	3	1.8

Step : Calculate the Mean and standard deviation of every column in the ATD Matrix by using formula

$$\text{Mean} = \frac{\sum x}{n} \text{ and standard deviation} = \sqrt{\frac{\sum x^2}{n} - \left(\frac{\sum x}{n}\right)^2}$$

Table 3 Mean and Standard deviation of ATD Matrix

	S ₁	S ₂	S ₃	S ₄	S ₅	S ₆
Mean μ	3.4	2.33	2.40	3.5	3.6	2.6
Standard Deviation σ	1.200	0.50334	0.52915	0.80932	0.52915	0.7211

Step : Convert the ATD Matrix into RTD Matrix.

This matrix is also termed as fuzzy matrix as the entries are 1, 0 & -1 by using the formula

$$\begin{cases} a_{ij} \leq (\mu_j - \alpha * \sigma_j) \text{ then } a_{ij} = -1 \text{ else} \\ a_{ij} \in (\mu_j - \alpha * \sigma_j, \mu_j + \alpha * \sigma_j) \text{ then } a_{ij} = 0 \text{ else} \\ a_{ij} \geq (\mu_j + \alpha * \sigma_j) \text{ then } a_{ij} = 1 \text{ else} \end{cases}$$

The RTD Matrix for $\alpha = 0.1, 0.15, 0.2, 0.25$

The RTD Matrix for $\alpha = 0.1$

Row Sum Matrix



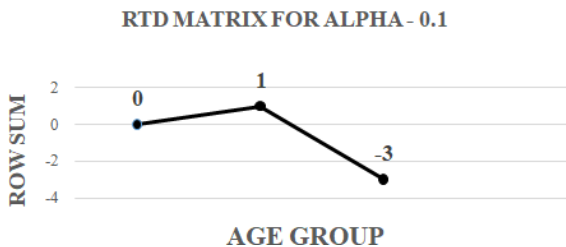


Jayapriya and Akila Devi

$$\begin{bmatrix} 1 & -1 & 1-1 & 1 & -1 \\ 0 & 0 & 0 & 0 & 1 & 0 \\ -1 & 1 & 0-1 & -1 & -1 \end{bmatrix} \quad \begin{bmatrix} 0 \\ 1 \\ -3 \end{bmatrix}$$

Age Group	18-22	23-27	28-32
Row Sum	0	1	-3

Table row sum of RTD Matrix for alpha = 0.1
 The Graphical representation of the RTD Matrix at alpha = 0.1



The RTD Matrix for $\alpha = 0.15$ Row Sum Matrix

$$\begin{bmatrix} 1 & -1 & 1-1 & 0 & 1 \\ 0 & 0 & 0 & 0 & 1 & 0 \\ -1 & 1 & -1 & 1 & -1 & -1 \end{bmatrix} \quad \begin{bmatrix} 1 \\ 1 \\ -2 \end{bmatrix}$$

Table 5 Row sum of RTD Matrix for alpha = 0.15

Age Group	18-22	23-27	28-32
Row Sum	1	1	-2

The Graphical representation of the RTD Matrix at alpha = 0.15

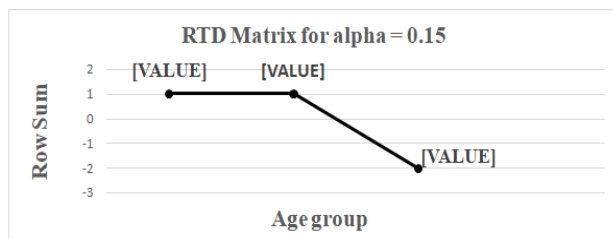


Figure 3.2

The RTD Matrix for $\alpha = 0.2$ Row Sum Matrix

$$\begin{bmatrix} 1 & -1 & 1-1 & 1 & 1 \\ 0 & 0 & 0 & 0 & 1 & 0 \\ -1 & 0 & 0 & 1 & -1 & -1 \end{bmatrix} \quad \begin{bmatrix} 2 \\ 1 \\ 0 \end{bmatrix}$$

Table 3.6 Row sum of RTD Matrix for alpha = 0.2

Age Group	18-22	23-27	28-32
Row Sum	2	1	0





Jayapriya and Akila Devi

The Graphical representation of the RTD Matrix at alpha = 0.2

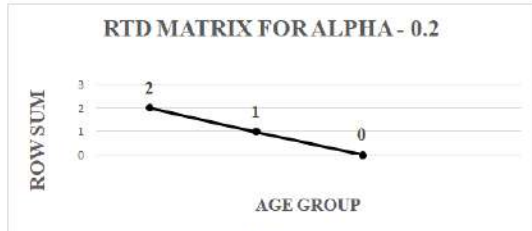


Figure 3.3

The RTD Matrix for $\alpha = 0.25$

$$\begin{bmatrix} 1 & -1 & 1 & -1 & 0 & 1 \\ 0 & 0 & 0 & 0 & 1 & 0 \\ -1 & 1 & -1 & 1 & -1 & -1 \end{bmatrix}$$

Row Sum Matrix

$$\begin{bmatrix} 1 \\ 1 \\ -2 \end{bmatrix}$$

Table 3.7 row sum of RTD Matrix for alpha = 0.25

Age Group	18-22	23-27	28-32
Row Sum	1	1	-2

The Graphical representation of the RTD Matrix at alpha = 0.25

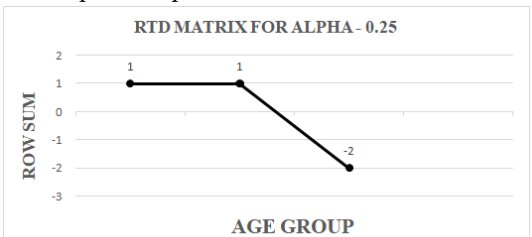


Figure 3.4

Obtain the combined effective time dependent data matrix and the corresponding row sum by adding all the ATD Matrix.

CETD Matrix

$$\begin{bmatrix} 4 & -4 & 4 & -4 & 2 & 2 \\ 0 & 0 & 0 & 0 & 4 & 0 \\ -4 & 3 & -2 & 2 & -4 & -4 \end{bmatrix}$$

Row Sum Matrix

$$\begin{bmatrix} 4 \\ 4 \\ -9 \end{bmatrix}$$

Table 8 Row sum of CETD Matrix

Age Group	18-22	23-27	28-32
Row Sum	4	4	-9

The Graphical representation of the CETD Matrix

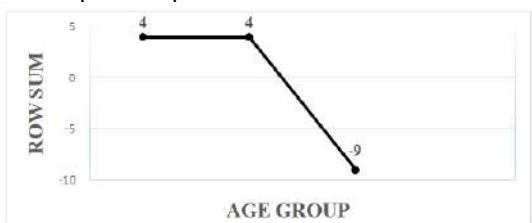


Figure 3.5





Jayapriya and Akila Devi

RESULTS AND DISCUSSION

From the observation of the above graph and CETD matrix shows that the age group 18-22 and 23-27 chance to clear the TNPSC exam in first attempt. And it shows that the age group 28-32 was more concentrate to crack the examination. The results of the fuzzy matrix model gave the exact result as that obtained experimentally.

REFERENCES

1. Anjan Mukherjee and Abhik Mukherjee, Application of Fuzzy Matrix Theory In COVID-19 Pandemic, IOSR Journal of Mathematics (IOSR-JM) e-ISSN: 2278-5728, p-ISSN: 2319-765X. Vol. 16, PP 52-56, 2020. DOI: 10.9790/5728-1604055256.
2. Beaula, T and Mallika, Application Of Fuzzy Matrices in Medical Diagnosis, Intern. J. Fuzzy Mathematical Archive Vol. 14, No. 1, 163-169, 2017 ISSN: 2320-3242
3. DOI: <http://dx.doi.org/10.22457/iffma.v14n1a20>
4. Chen, S.H, Operations on fuzzy numbers with function principle, Tamkang Journal of Management Science, Vol. 6 (1), pp. 13-26, 1985.
5. Elizabeth, S and Sujatha, L, Application Of Fuzzy Membership Matrix In Medical Diagnosis and decision Making, Applied Mathematical Sciences, Vol. 7, 2013, No. 127, 6297-6307, 2013 DOI: <http://dx.doi.org/10.12988/ams.2013.38485>
6. Hadi Nasser, Fuzzy Numbers, Positive and Nonnegative, International Mathematical Forum, Vol. 8, (36), pp. 1777-1780, 2008
7. Kuppaswami, G and Sujatha, R and Vasantha Kandasamy, W.B, Study of Traffic Flow Using CETD Matrix, Indian Journal of Science and Technology, Vol 8(24), 2015 DOI: 10.17485/ijst/2015/v8i24/80191
8. Priya (2019). Basic concepts of fuzzy matrix, International Journal of Engineering Research & Technology Vol. 8 (12), pp:905-909, 2019.
9. Radhika, K and Anbalagan and Alexander and Suganthi Mariyappan, Risk Factor of Breast Cancer Using CETD Matrix-An Analysis, International Journal of Applied Engineering Research ISSN 0973-4562 Vol. 14, No. 4, 2019.
10. Raich, Vivek .V and Archana Gawande and Rakesh Kumar Tripath, Fuzzy Matrix Theory and its Application for Recognizing the Qualities of Effective Teacher, International Journal of Fuzzy Mathematics and Systems. 1(1), pp. 113-122, 2011.
11. Sangodapo, O Taiwo and Yuming Feng, An Application of Improved Method of Fuzzy Matrix Composition in Medical Diagnosis, Italian Journal of Pure And Applied Mathematics – N. 48, 1093–1103, 2022.
12. Shreemathi Adiga, Saraswathi, A and Praveen Prakash, A, A Ranking Analysis/An Interlinking Approach of New Triangular Fuzzy Cognitive Maps and Combined Effective Time Dependent Matrix (NCMTA 18) IOP Conf. Series: Journal of Physics: Conf. Series 1000, 2018 DOI :10.1088/1742-6596/1000/1/012159
13. Shyamal, A.K, and Madhumangal Pal, Triangular Fuzzy Matrices, Iranian Journal of Fuzzy Systems, Vol. 4(1) pp-75-87, 2007.
14. Shyamal, A.K and Madhumangal Pal, Two New Operators On Fuzzy Matrices, J. Appl. Math. And Computing 15(1-2), 91-107, 2004 DOI: 10.1007/BF02935748
15. Suresh, M and Greeda, J, Analysis of Women Harassment in Villages Using CETD matrix Model, Journal of Emerging Technologies and Innovative Research, Vol 6, 2019 pp:112-123, 2019.
16. Uma Rani, A, An Analysis on the Main Reasons for using Social Networking among Indian Youth – A Fuzzy Approach, International Journal of Computational Intelligence and Informatics, Vol. 2, 2012 pp:83-89.
17. Vivehananthan, V and Rajeswari, K and Anbalaga, Effective Completion of Typewriting Examination- Using Fuzzy Matix, International Journal of Applied Engineering Research ISSN 0973-4562 Vol. 14, No. 4, 2019.





Standardization of Swarnavanga with Application of Namboori Phased Spot Test

Madhav Maheshwari^{1*}, Anitha Hosur² and Anjali Gambhava¹

¹PG Scholar, Department of RSBK, Parul Institute of Ayurved, Vadodara, Gujarat, India.

²Professor, Department of RSBK, Parul Institute of Ayurved, Vadodara, Gujarat, India.

Received: 05 Jun 2024

Revised: 14 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Madhav Maheshwari

PG Scholar, Department of RSBK,

Parul Institute of Ayurved,

Vadodara, Gujarat, India.

E.Mail: maheshwarimadhav47@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Rasashastra, an ancient Indian alchemical science imparts knowledge about different formulations which are designed using herbs, minerals and metals. The standardization of these medicines is an important area of research and development in *Rasashastra*. Scientists are constantly developing new and improved methods to standardize these medicines, ensuring that patients have access to safe, effective, and high-quality products. We have classical parameters for assessment of *Rasa* preparations but to update the standards many other parameters are developed. The crucial Namboori Phased Spot Test within *Rasashastra* allows practitioners to assess the authenticity and purity of these substances, ensuring the efficacy and safety of *Rasa Ausadhies*. Namboori Phased Spot Test (NPST) is a qualitative chemical test for heavy metals. The test is based on the reduction of iodine to iodide by heavy metals in the presence of acid. A sample solution is dropped onto potassium iodide starch paper and observed for a colour change at three time intervals. The colour change depends on the specific heavy metal present and the time interval at which it occurs. The NPST is a simple and reliable test for detecting heavy metals in a variety of samples. It is observed that interaction of *Swarnavanga* with their respective reagents exhibits chromatographic presentation in form of dark brown and other relevant colours appearance and faded gradually which confirms the genuineness of preparation. Similarity seen between observed data and available standard data. The colour and pattern of the spots that appear on the filter paper gives confirmation regarding identification of the authenticity of the *Swarnavanga*. NPST is easy-to-perform test that should be used to identify and assess the quality of *bhasmas* and *sinduras* in a routine. In *Ayurved Rasashastra*, this small effort aims to update and enrich the available data regarding the standardization of *Swarnavanga*, making it a valuable tool for Ayurvedic practitioners and researchers.

Keyword: *Swarnavanga*, NPST, *Rasashastra*, *Ayurved*, Standardization.





INTRODUCTION

Rasashastra is branch of an ancient Indian alchemical science, holds immense importance in the realm of *Ayurved* medicine. In an era where traditional medicines are gaining global recognition, the bench side but reliable techniques like the Namboori Phased Spot Test[1] provides a standardized and reliable method for evaluating the quality of medicines derived from metals and minerals. Namboori Phased Spot Test is based on principles of chromatography technique and is crucial for quality assessment of metallic and mineral medicines. This test allows Students, Researchers, Practitioners to assess the authenticity and purity of these substances, ensuring the efficacy and safety of *Rasousadhies*. By preserving the integrity of these ancient practices, *Rasashastra* continues to contribute significantly to the quality assurance of Ayurvedic medicines, promoting health and well-being worldwide.

Aim: To assess quality of *Swarnavanga* by Namboori Phased Spot Test

MATERIALS AND METHODS [2],[3]:

Materials:

- 10% Pot iodide paper
- Whatman filter paper
- Solution of 5N HNO₃
- Aqua regia
- Distilled water
- Sample – *Swarnavanga*[4]
- Test tubes
- Test tube stand
- Dropper

Methods:

- Namboori Phased Spot Test (NPST)

Procedure:

Preparation of reacting papers

A solution was prepared by mixing 2 gm Potassium iodide with 20ml distilled water. Subsequently, a Whatman filter paper was immersed in it and then kept on a dry glass sheet to dry.

Preparation of Solution for *Vanga* Group

250mg sample was taken into test tubes and 0.5 ml 5N HNO₃ added as reagent. Test tubes were stirred and heated for a minute then kept undisturbed for 72 hours. After 72 hrs, solution prepared with 5N HNO₃ was dropped with the help of dropper on 10% potassium iodide paper and observed in three intervals of time i.e. 0-5 min, 5min-20min and 20 min-24 hrs for 1st, 2nd and 3rd phase respectively. Same procedure was conducted with reagent 0.5 ml aqua regia and kept undisturbed for 24 hours. After 24 hrs, solutions prepared with 0.5 ml aqua regia was dropped with the dropper on 10% potassium iodide paper and observation done in three time intervals i.e. 0-5 minutes, 5min-20min and 20 min-24 hours for 1st, 2nd and 3rd phase respectively.

Preparation of Solution for *Parada* Group

Similar procedure was carried out for *Parada* group with reagent 0.5ml 5N HNO₃ and 0.5ml Distilled water . After 48 hrs, solution prepared with 0.5ml 5N HNO₃ and 0.5ml Distilled water each was dropped using dropper on 10%





Madhav Maheshwari et al.,

potassium iodide paper and observed in three intervals of time, 1st phase occurred at 0-5 minutes, 2nd phase occurred at 5 minutes-20 minutes, 3rd phase occurred at 20 minutes-24 hours.

RESULT AND DISCUSSION

For *Vanga* group [Table 1], HNO₃ and aqua regia reagents were used. With HNO₃, it shows dark brown central spot surrounding by moderate and light brown circle in its 1st phase followed by white space between brown spot and brown periphery having dull margin in 2nd phase then white space reduces and brown periphery expanded to central area in 3rd phase. With aqua regia, it shows dark brown central spot surrounded by light brown circle with clear margin in 1st phase followed by white space between moderate brown central spot and brown periphery having dull margin in 2nd phase and later on in 3rd phase, light central spot and moderate brown periphery with faded margin on outer area.

For *Parada* group [Table 1], HNO₃ and distilled water used as reagent. With HNO₃, dark brown central spot surrounding by moderate brown circle in 1st phase followed by white space between moderate brown colored central spot and dark brown colored periphery in 2nd phase. In 3rd phase, dull orange internal margin of brown periphery. With distilled water, dull blue space between golden yellow central spot and dull orange periphery in 1st phase followed by central spot disappeared with faded dull orange periphery in 2nd phase. In 3rd phase, periphery area also disappears.

Rasashastra is a branch of Ayurvedic science that deals with preparation of herbo-mineral compounds. Herbo-mineral medicines have been used in *Ayurved* for centuries to treat a wide range of diseases and conditions. The standardization of herbo-mineral medicines is important to ensure their safety, efficacy and quality. Standardization involves developing and implementing quality control measures at all stages of the manufacturing process, from the procurement of raw materials to the finished product. The standardization of herbo-mineral medicines is a complex process particularly important in *Rasashastra*, as many of the ingredients used are toxic and must be carefully processed to make them safe for human consumption.

Namboori Phased Spot Test (NPST) is a qualitative analytical test developed by Dr. Namboori Hanumantha Rao in 1970. It is used to identify and assess the quality of *bhasma* and *sindura*, which are herbo-mineral preparations used in Ayurveda. The test is based on the chromatography principle, which is a technique used to separate and identify the components of a mixture. When a drop of a clear solution of a *bhasma* or *sindhura* on Whatman paper impregnated with a suitable reagent, it gives a spot with a series of changes in colour and pattern. The colour and pattern of the spots that appear on the filter paper can be used to identify the authenticity of the sample.

Standardization of *Rasa* preparation is essential need in today's era. The Namboori Phased Spot Test (NPST) is easy-to-perform test that must be used to identify and assessment of the quality of *bhasma* and *sindura kalpana*, which are Ayurvedic formulations. The test belongs to principles of chromatography and involves observing colour and pattern produced by a chemical reaction between the test sample and a reagent. The NPST is a valuable tool for Ayurvedic practitioners and researchers as it can help to ensure the quality along with consistency for Ayurvedic preparations.

REFERENCES

1. Rao Hanumanta Namboori, Application of standard Namboori Phased Spot Test in identification of *bhasma* and *sindura* preparations of Ayurveda, CCRAS, Dept. of AYUSH, Ministry of Health and Family Welfare, GOI, New Delhi, 2010





Madhav Maheshwari et al.,

2. Rao Hanumanta Namboori, Application of standard Namboori Phased Spot Test in identification of bhasma and sindura preparations of Ayurveda, CCRAS, Dept. of AYUSH, Ministry of Health and Family Welfare, GOI, New Delhi,2010 p. 76-78
3. Rao Hanumanta Namboori, Application of standard Namboori Phased Spot Test in identification of bhasma and sindura preparations of Ayurveda, CCRAS;Dept. of AYUSH, Ministry of Health and Family Welfare, GOI, New Delhi,2010.p.87-90
4. Chapter 37/164-167Ravindra AngadiBhaisajya Ratnavali of Sen Govindadas, Chowkhambhasurbhartiprakashan, Varanasi; 2018. p. 1111

Table 1: Vanga group

Reagent	Phase 1	Phase 2	Phase 3
HNO ₃	Dark Brown central spot surrounding by moderate and light brown circle	White space between Brown spot and brown periphery having dull margin	White space reduces and brown periphery expanded to central area
Aqua regia	Dark Brown central spot surrounded by light brown circle with clear margin	White space between Moderate brown central spot and brown periphery having dull margin	Light central spot And moderate brown periphery with faded margin on outer area

Table 2:Parada group

Reagent	Phase 1	Phase 2	Phase 3
HNO ₃	Dark Brown central spot surrounding by moderate brown circle	White space between Moderate brown central spot and dark brown periphery	Dull orange Internal margin of brown periphery
Distilled water	Dull blue space between Golden yellow central spot and dull orange periphery	Central spot disappeared with faded dull orange periphery	Periphery area also disappears

Reagent

Phase 1

Phase 2

Phase 3

HNO₃





Madhav Maheshwari et al.,



Figure 1. Vanga Group

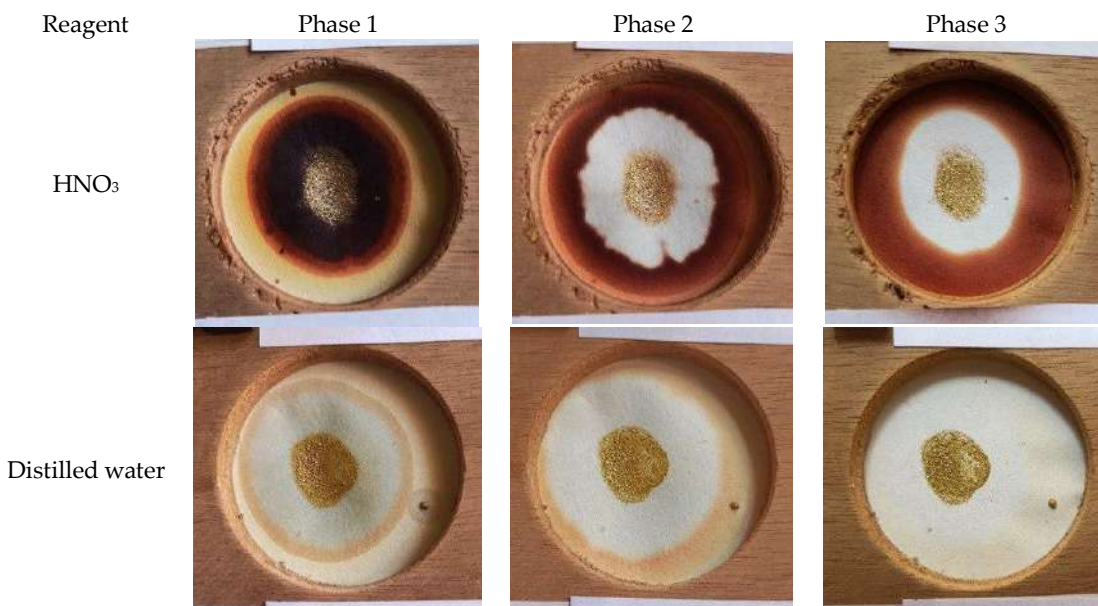


Figure 2. Parada Group





A Probabilistic Machine Learning based Sentiment Analysis Approach for Predicting Customer Reviews

Pushpa Bansiwali^{1*}, Neeraj Shrivastava² and Sunil Nimawat³

¹Student, Department of Computer Science and Engineering, IPS Academy Institute of Engineering and Science, Indore, (Affiliated to Rajiv Gandhi Proudlyogiki Vishwavidyalaya, Bhopal), Madhya Pradesh, India

²Professor, Department of Computer Science and Engineering, IPS Academy Institute of Engineering and Science, Indore, (Affiliated to Rajiv Gandhi Proudlyogiki Vishwavidyalaya, Bhopal), Madhya Pradesh, India.

³Assistant Professor, Department of Computer Science and Engineering, IPS Academy Institute of Engineering and Science, Indore, (Affiliated to Rajiv Gandhi Proudlyogiki Vishwavidyalaya, Bhopal), Madhya Pradesh, India.

Received: 02 Jun 2024

Revised: 06 Aug 2024

Accepted: 19 Oct 2024

*Address for Correspondence

Pushpa Bansiwali

Student, Department of Computer Science and Engineering,
IPS Academy Institute of Engineering and Science, Indore,
(Affiliated to Rajiv Gandhi Proudlyogiki Vishwavidyalaya, Bhopal),
Madhya Pradesh, India.
E.Mail: chourase@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

It is crucial for organizations to determine the characteristics and sentiment of customer evaluations, usually through the use of opinion mining and sentiment analysis. IT systems can utilize natural language processing for a range of functions, such as generating summaries of evaluations, classifying reviews into categories, and performing other real-time applications. Sentiment analysis holds great potential for application in real-time business models. Consumer testimonials serve as an excellent means for companies to obtain feedback. Nevertheless, the process of personally examining a substantial amount of evaluations can be laborious and time-consuming. Sentiment analysis streamlines this procedure, offering a rapid and scalable method to comprehend the entire sentiment sent by clients. Sentiment research aids organizations in identifying favorable client experiences. By discerning favorable sentiments in evaluations, organizations can identify the specific components of their products or services that are highly regarded by customers. This information is vital for strengthening and advocating favorable attributes. Due to the lack of obvious boundaries in classifying tweets or reviews as positive, negative, or neutral, a probabilistic technique using the Deep Bayesian Classifier has been suggested to





Pushpa Bansiwala *et al.*,

determine the sentiment of customer feedback. Evidence demonstrates that the proposed methodology attains higher classification accuracy compared to prior research in this domain.

Keywords: Data Mining, Customer Review, Sentiment Analysis, Machine Learning, Bayesian Classifier, Classification Accuracy.

INTRODUCTION

Customer sentiment analysis relies heavily on machine learning, which gives companies an effective tool for understanding and reacting to customer feedback. In a day of digital communication and information overload, it is critical to comprehend the feelings that customers are expressing. This paper examines the several factors that make machine learning essential for efficient customer sentiment analysis [1]. However, it is difficult to extract user sentiment from vast and complicated data sets. This is to make sure that the implicit meaning must be accurately deduced and that the context (semantics) is considered before drawing any judgments. To further separate the valuable information from the raw data, precise data pre-processing must be mandated. Sentiment analysis is crucial since user attitudes significantly affect a number of characteristics and domains. Machine learning's ability to manage enormous amounts of consumer input is one of the main reasons it is crucial for sentiment analysis. Businesses now receive a dizzying quantity of information from several sources, including surveys, emails, social media, and online reviews. This scale is beyond the capabilities of traditional methodologies; in contrast, machine learning models process massive datasets effectively, allowing organisations to analyse feelings more broadly [2].

Sentiment analysis faces a great deal of difficulty because of how intricate human language is. Natural language processing (NLP) in particular, falls under machine learning is excellent at identifying intricate patterns, contextual subtleties, and minute differences in consumer reactions. Beyond crude positive or negative classifications, this capacity to interpret linguistic nuances enables a more precise and sophisticated sentiment analysis. Consumer perceptions are subject to alter due to new product offers, shifting trends, and outside events. By continuously learning from fresh data, machine learning models demonstrate adaptability. This flexibility guarantees that sentiment research is current and representative of the customer landscape, allowing organisations to react quickly to shifting emotions and make well-informed decisions. Personalised sentiment analysis catered to certain sectors and circumstances is made possible by machine learning. Businesses can tailor sentiment analysis tools to meet their specific needs by training models on language specific to their industry. By taking a customised approach, sentiment research becomes more accurate and companies pay attention to the things that are most important to their customers [3].

A generic process architecture for sentiment analysis through the knowledge discovery process is depicted in figure 1. Automatic feature extraction, key phrase recognition, and word associations with positive or negative sentiments are areas where machine learning can be employed due to the enormity of data being analyzed. This feature gives companies the ability to identify particular features of goods or services that affect how customers perceive them, giving them insightful information for future development.

LITERATURE REVIEW

The literature survey of various scholars related to the review is as follows:- Zhao et al. [1] proposed a multimodal sentiment analysis technique built on the multimodal sentiment analysis technique, which can help individuals make better judgments by obtaining more emotional information sources. According to this paper's experimental findings, CNN-SVM has the greatest recognition rate. 93.5%, respectively. Dhyani et al. in [2] author proposed for sentiment analysis, a brand-new intuitionistic fuzzy inference system (IFIS). Through the suggested IFIS, the study article uses sentiment analysis of tweets to predict the personality attribute features of the tweeter. Utilising TextBlob in Google



**Pushpa Bansiwal et al.,**

Collaboration, the Natural Language Processing Toolkit (NLTK) was utilised to examine Twitter data for subjectivity and polarity, with the goal of predicting the positivity score through the suggested innovative IFIS. Vohra et al. [3] proposed To get outstanding outcomes on the dataset, the model makes use of numerous convolution and max pooling layers, dropout operation, and dense layers with ReLU and sigmoid activations. Furthermore, a comparison is made between the proposed model's performance and that of many common classifiers, including Random Forest, Naive Bayes, Support Vector Machine (SVM), and Decision Tree. According to the findings, the suggested CNN with FastText word embeddings performs better than other classifiers on the provided dataset, with an accuracy of 0.925969... This classification reveals, nearly 55% of the tweets are found to show affirmation, about 25% show a negative disposition, and 21.09% have neutral sentiments Phan et al. [4] proposed a model which comprises the subsequent actions: First, BERT is used to turn words in sentences into vectors. Secondly, BiLSTM over word vectors is used to produce the contextualised word representations. Third, across the contextualised word representations, important characteristics are retrieved and represented using the GCN model with several convolutional layers. Lastly, the CNN model is applied to the feature vectors in order to classify the feelings at the aspect level. Tests conducted on three reference datasets demonstrate that the performance of the prior context-based GCN techniques for ALSA has been enhanced by the proposed model.

Obiedat et al. [5] proposed a combination approach to address the unbalanced data issue that combines the Support Vector Machine (SVM) algorithm with Particle Swarm Optimisation (PSO) and several oversampling approaches. SVM is used as a machine learning classification approach to optimize the dataset, which includes various reviews from a number of Jordanian restaurants, in order to anticipate the sentiments of reviews. Information was gathered from Jeeran, a popular Arabic review social network. The feature weights are optimized using the PSO approach. Vashishtha et al. [6] proposed a combination approach to address the unbalanced data issue that combines the Support Vector Machine (SVM) algorithm with Particle Swarm Optimisation (PSO) and several oversampling approaches. SVM is used as a machine learning classification approach to optimise the dataset, which includes various reviews from a number of Jordanian restaurants, in order to anticipate the sentiments of reviews. Information was gathered from Jeeran, a popular Arabic review social network. The feature weights are optimised using the PSO approach.

Saha et al. [7] utilised a variety of machine learning methods to analyse sentiment. On the linguistic characteristics, algorithms including Multilayer Perceptron (MP), Random Forest (RF), Decision Tree (DT), and Support Vector Machine (SVM) were used. For every classifier, the authors have calculated the precision, recall, accuracy, F-measure, and ROC values. Random Forest scored better than the other classifiers, successfully classifying nearly 61% of the instances. Such sentiment analysis, in our opinion, on a particular class of texts might inspire more research into our grasp of natural language. Estrada et al. [8] The authors compared various sentiment analysis classifiers within an Intelligent Learning Environment (ILE-Java) to categorize educational opinions. Three different methodologies were employed: machine learning, deep learning, and an evolutionary technique called EvoMSA. The researchers curated two datasets focusing on programming languages, encompassing students' sentiments regarding professors, tests, assignments, and academic projects. SentiTEXT comprises polarity labels (positive and negative), while eduSERE includes labels related to learning-oriented emotions (positive and negative), such as engaged, thrilled, bored, and annoyed.

Rahat et al. [9] proposed a technique to analyze the sentiment polarity of airline review dataset based on a numerical transformation of the data prior analysis. The removal of special characters such as @ removal etc. was the first step in the pre-processing stage. It could be inferred from the experimental results that the SVM outperformed the Naïve Bayes for the airline dataset. Hasanli et al. [10] A strategy outlining the steps for gathering, cleaning, and annotating tweets in Azerbaijani has been developed for sentiment analysis of the language. Based on bag-of-words models, machine learning methods such as SVM, Naïve Bayes, and Linear Regression were used to determine sentiment polarity. Other Turkish languages can easily adopt our transferable and easily implementable data processing and categorization approach. By comparing the outcomes of several machine learning algorithms, the best parameters for tweet categorization were found..





Pushpa Bansiwala et al.,

METHODOLOGY

Sentiment Analysis using Machine Learning

When it comes to expressing and ranking whether a text reflects good, neutral, or negative emotions, machine learning algorithms are frequently quite useful. There are two categories of extensions for machine learning, such as unsupervised methods. A labelled dataset is used by the supervised algorithm, and every training text is created with positive sentiments. On the other hand, unsupervised readings consist of happy feelings and unprocessed data without any writing. The commonly used machine learning models used are [11].

Convolutional Neural Network (CNN): CNN's basic architecture is made up of layers that handle incoming data in a hierarchical fashion. The main parts are fully connected layers, pooling layers, and convolutional layers. By using filters or kernels to the input data, convolutional layers extract features by identifying local patterns and details. In order for CNNs to learn hierarchical representations from unprocessed input, these layers are essential. Convolutional layers carry out the vital function of extracting features. Sliding a filter over the input data and calculating the dot product at each place comprise the convolution procedure. Activation functions add non-linearity to the network after the convolutional process. ReLU (Rectified Linear Unit), a popular activation function, introduces non-linearities by thresholding the output, allowing the model to learn more intricate feature correlations [12].

Support Vector Machine (SVM): One potent class of machine learning techniques used for both regression and classification applications is Support Vector Machines (SVMs). They have become well-known for their reliable operation and adaptability in a variety of fields. The goal behind Support Vector Machines (SVMs) is to identify the optimal hyperplane for separating data points into distinct groups. By acting as the decision boundary, this hyperplane maximises the margin between the classes. SVMs work well in high-dimensional environments, where they are useful for jobs requiring a large number of input features. SVMs are unique in that they prioritise maximising the gap between each class's nearest data points and the decision border. The distance between the closest data point and the hyperplane is known as the margin [13].

Hyperplane in SVM: The data points that are closest to the decision boundary or hyperplane are known as support vectors. In order to define the margin and, by extension, the decision boundary, these points are essential. These support vectors are what give support vector machines (SVMs) their moniker because they help find the best possible hyperplane. With non linear separable datasets depending on multiple variables, the SVM employs a multi-dimensional hyperplane (hyperplane with working with multiple variables) making data separable. Polynomial, sigmoid, and radial basis function (RBF) are examples of common kernel functions. Optimising a cost function that takes into account the margin size and classification accuracy is necessary for training a support vector machine. The goal of the optimisation method is to identify the ideal hyperplane [14].

Bayesian Approach

Probabilistic graphical models known as Bayesian Networks, or BayesNets, use Bayesian probability to depict and infer relationships between variables. Regarding sentiment classification, BayesNets provide a logical and comprehensible method. This paper explores the use of BayesNets in sentiment classification, emphasising the advantages and drawbacks of this method for identifying emotional overtones in textual data. Determining the sentiment expressed in a text and classifying it as good, negative, or neutral is known as sentiment classification. A probabilistic modelling framework that is well-suited to the ambiguity contained in language is offered by BayesNets, which enables the capture of dependencies between words or phrases and the nuanced depiction of sentiment [15]. With directed acyclic graphs (DAGs), Bayesian networks depict conditional dependencies between variables. Nodes in the network can represent words or features in the context of sentiment categorization, while edges show conditional dependencies. Because of this structure, BayesNets can accurately represent how the presence or absence of specific words affects the likelihood of a given feeling. In a BayesNet, each node has associated probabilities that indicate the chance that a variable will take on a specific value based on the values of its





Pushpa Bansibal et al.,

parent nodes. These probabilities, which indicate the possibility of noticing particular words or features given the sentiment indicated in the text, can be learned from training data in sentiment classification. Sentiment analysis requires the ability to handle uncertainty and variability, which BayesNets naturally provide. It's common for language to be vague, and there are many methods to convey feelings. Because BayesNets incorporate prior probabilities and adapt their beliefs based on mounting information, they facilitate the modelling of uncertainty. They are therefore capable of managing a wide range of language manifestations of emotion. Prior knowledge can be included with BayesNets, which is especially helpful for sentiment categorization. An enhanced and contextually aware sentiment analysis can be achieved by encoding into the network prior knowledge about words or phrases that strongly suggest a particular sentiment [16].

Algorithm

As the customer review texts may have overlapping tags or tokens, hence a probabilistic Bayes Classifier has been proposed. As sentiments do not possess a particular decision boundary (fixed), hence a probabilistic approach happens to be more effective which can be done employing the Deep Bayes Net whose classification depends on the following relation:

$$P\left(\frac{x}{x_i, k_1, k_2, M}\right) = \frac{P\left(\frac{x_i}{x, k_2, M}\right) P\left(\frac{x_i}{k_1, M}\right)}{P\left(\frac{x}{k_1, k_2, M}\right)} \quad (1)$$

Here,

P represents probability.

x_i represents weights and bias vectors (combined).

x represents the data to be used for the purpose of training.

M represents data units (neurons) in network.

k_1 and k_2 represents the term responsible for penalty based regularization.

$\rho = \frac{k_1}{k_2}$ is often considered the regularization factor which is acted upon the objective function (J) to be optimized based on the training dataset, and renders the regularized cost function:

$$F(w) = \mu w^T w + v \left[\frac{1}{n} \sum_{i=1}^n (p_i - a_i)^2 \right] \quad (2)$$

If ($\pi \ll v$): errors in training are typically rendered low.

else if ($\pi \geq v$): errors are typically rendered high needing a weight reduction or Penalty.

The proposed algorithm is presented next:

Start

{

Step.1 Obtain annotated dataset.

Step.2 Divide the data into a ratio of 70:30 as training and testing data samples.

Step.3 Define match token data length (n) and

for $i = 1:n$

Search (token == match text)

end

Step.4 Design a neural network with multiple hidden layers.

Step.5 Initialize training with random weights.





Pushpa Bansiwala et al.,

Step. 6 Train models with training data and updated weights based on the back propagation rule as:

$$w_{k+1} = w_k - [J_k J_k^T + \mu I]^{-1} J_k^T e_k \quad (3)$$

Step.7 if (Cost Function J stabilizes over multiple iterations)

Truncate

 else if (iterations==max. iterations defined)

Truncate

 else

 {

 Apply data and update (w, b)

Feedback (e)

 }

Step.8 Calculate error% and Classification Accuracy

Stop

}

The performance parameters used for evaluation of the algorithm is the accuracy % which is computed as:

$$\text{Accuracy\%} = 100 - \text{error\%} \quad (4)$$

EXPERIMENTAL RESULTS

The experimental results have been presented next. The dataset has been taken from Kaggle, for Amazon Product Reviews.

(<https://www.kaggle.com/datasets/tarkkaanko/amazon?resource=download>)

Out of the 4916 samples, the training and testing ratio has been selected as 70:30 making the testing samples as 1475 samples. From table 2, it can be clearly observed that the proposed work attains much higher classification accuracy employing the Regularization based Deep BayesNet approach due to a probabilistic classification approach. Moreover, a grouped word match also aids the classification process.

CONCLUSION

To summarize, Sentiment analysis has emerged as an essential tool for business objectives, providing significant insights into user behaviors and preferences. Various methods and algorithms within Natural Language Processing (NLP) are used to achieve a thorough comprehension by applying sentiment analyzers. This study offers a comprehensive examination of several datasets and research studies that utilize different machine learning methods for sentiment analysis. Machine learning-based sentiment analysis has demonstrated its efficacy and adaptability as a beneficial tool for obtaining important insights from textual data. The research that were evaluated demonstrated a wide variety of strategies, including both traditional methods like Random Forest and Word2Vec, as well as more modern approaches like Recurrent Neural Networks (RNN). These techniques have shown a high level of accuracy in determining sentiment in many areas. There is great potential for further progress in the field of video sentiment analysis, namely in the areas of extracting features from frames and applying machine learning methods. In summary, the research highlights the importance of machine learning in uncovering patterns of sentiment, which helps us gain a more profound comprehension of user feelings across various applications and domains. The study introduces an enhanced Bayesian Network for analyzing the Amazon Product Review dataset. The model achieves a





Pushpa Bansiwala et al.,

Mean Squared Error (MSE) of 0.95973 and a Mean Absolute Error (MAE) of 0.67, surpassing the accuracy of current baseline methods.

REFERENCES

1. Y Zhao, M Mamat, A Aysa, K Ubul, "Multimodal sentiment system and method based on CRNN-SVM", Neural Computing and Applications, Springer, 2023, pp.1-13.
2. M Dhyani, GS Kushwaha, S Kumar, "A novel intuitionistic fuzzy inference system for sentiment analysis", International Journal of Information Technology, Springer 2022, vol.14., pp. 3193–3200.
3. A Vohra, R Garg, "Deep learning based sentiment analysis of public perception of working from home through tweets", Journal of Intelligent Information Systems, Springer 2022, vol.60, pp. 255–274.
4. H. T. Phan, N. T. Nguyen and D. Hwang, "Aspect-Level Sentiment Analysis Using CNN Over BERT-GCN," in IEEE Access, 2022, vol. 10, pp. 110402-110409.
5. R. Obiedat R. Qaddoura, A. Al-Zoubi, L. Al-Qaisi, O. Harfoushi, M. Alrefai, H. Faris., "Sentiment Analysis of Customers' Reviews Using a Hybrid Evolutionary SVM-Based Approach in an Imbalanced Data Distribution," in IEEE Access, vol. 10, pp. 22260-22273, 2022.
6. S Vashishtha, S Susan, "Neuro-fuzzy network incorporating multiple lexicons for social sentiment analysis", Applications in computing, Springer 2022, vol.26, pp. 487–4507.
7. A. Saha, A. A. Marouf and R. Hossain, "Sentiment Analysis from Depression-Related User-Generated Contents from Social Media," 2021 8th Intern, "ational Conference on Computer and Communication Engineering (ICCCCE), Kuala Lumpur, Malaysia, 2021, pp. 259-264
8. MLB Estrada, RZ Cabada, RO Bustillos, "Opinion mining and emotion recognition applied to learning environments", Journal of Expert Systems, Elsevier 2020, vol. 150., 113265
9. A. M. Rahat, A. Kahir and A. K. M. Masum, "Comparison of Naive Bayes and SVM Algorithm based on Sentiment Analysis Using Review Dataset," 2019 8th International Conference System Modeling and Advancement in ResearchTrends (SMART), Moradabad, India, 2019, pp. 266-270.
10. H. Hasanli and S. Rustamov, "Sentiment Analysis of Azerbaijani tweets Using Logistic Regression, Naive Bayes and SVM," 2019 IEEE 13th International Conference on Application of Information and Communication Technologies (AICT), Baku, Azerbaijan, 2019, pp. 1-7.
11. H. Huang, A. A. Zavareh and M. B. Mustafa, "Sentiment Analysis in E-Commerce Platforms: A Review of Current Techniques and Future Directions," in IEEE Access, 2023 vol. 11, pp. 90367-90382.
12. M. Yasen and S. Tedmori, "Movies Reviews Sentiment Analysis and Classification," 2019 IEEE Jordan International Joint Conference on Electrical Engineering and Information Technology (JEEIT), Amman, Jordan, 2019, pp. 860-865.
13. P. Karthika, R. Murugeswari and R. Manoranjithem, "Sentiment Analysis of Social Media Network Using Random Forest Algorithm," 2019 IEEE International Conference on Intelligent Techniques in Control, Optimization and Signal Processing (INCOS), Tamilnadu, India, 2019, pp. 1-5.
14. [14] L Zheng, H Wang, S Gao, " Sentimental feature selection for sentiment analysis of Chinese online reviews", International journal of machine learning and cybernetics, Springer, 2018, vol.9, pp. 75–84.
15. R. D. Desai, "Sentiment Analysis of Twitter Data," 2018 Second International Conference on Intelligent Computing and Control Systems (ICICCS), Madurai, India, 2018, pp. 114-117.
16. A. Bayhaqy, S. Sfenrianto, K. Nainggolan and E. R. Kaburuan, "Sentiment Analysis about E-Commerce from Tweets Using Decision Tree, K-Nearest Neighbor, and Naïve Bayes," 2018 International Conference on Orange Technologies (ICOT), Nusa Dua, Bali, Indonesia, 2018, pp. 1-6.
17. M. I. Zul, F. Yulia and D. Nurmalasari, "Social Media Sentiment Analysis Using K-Means and Naïve Bayes Algorithm," 2018 2nd International Conference on Electrical Engineering and Informatics (ICon EEI), Batam, Indonesia, 2018, pp. 24-29.





Pushpa Bansiwala et al.,

Table 1. Summary of Previous Work

Author	Method / Algorithm	Dataset	Research gap	Parameter
Zhao et al. [1], 2023	CNN-SVM	eNTRAFACE'05, RML, and AFEW6.0	No data pre-processing employed. Amalgamation of baseline techniques such as CNN-RNN and SVM often leads to quick saturation of performance as more training data is added. (Inherent to SVM)	Accuracy of 93.5 %
Dhyani et al [2], 2022	Intuitionistic Fuzzy Inference System (IFIS)	Kaggle (Twitter Dataset)	Fuzzy Inference System (FIS) used for sentiment analysis. Typically fuzzy systems also suffer from performance saturation. A better option is the Adaptive Neuro-Fuzzy Inference System (ANFIS).	RMSE of 0.3078
Vohra et al. [3], 2022	CNN	Kaggle (Twitter Dataset)	CNN is often prone to overfitting and vanishing gradient. No analysis pertaining to overfitting and vanishing gradient performed.	Accuracy of 92.59 %
Phan et al. [4], 2022	Graph Convolutional Neural Networks (GCN)	Kaggle (Twitter Dataset)	The Graph Convolutional Neural Networks (GCN) used for The CNN model is used to classify aspect-level sentiments instead of feature vectors. CNN often tend to exhibit overfitting. Moreover, probabilistic approaches may yield higher accuracy for aspect level classification.	Accuracy of 85.25%
Obeidat et al. [5], 2022	PSO-SVM	Kaggle	The PSO-Optimized SVM approach has an inherent data saturation probability. No word-frequency analysis has been used.	Accuracy of 89%
Vashishtha et al. [6], 2021	Adaptive Neuro Fuzzy Inference Systems (ANFIS)	Kaggle (SentiwordNet)	ANFIS typically exhibits early performance saturation. No group or bag of words match performed which may increase prediction accuracy.	Best case RMSE of 0.36
Saha et al. [7], 2021	Decision Trees (DT)	Kaggle	No data pre-processing. Frequency analysis of words not employed. Decision trees typically does not match the performance of neural networks and deep neural networks.	Accuracy of 60.5395%
Estrada et al. [8], 2020	CNN-LSTM hybrid	SentiWord	Both the CNN and LSTM are prone to overfitting. They also suffer from the vanishing gradient problem while training with large training dataset corpus. No probabilistic approach explored for context level classification.	Accuracy of 93%





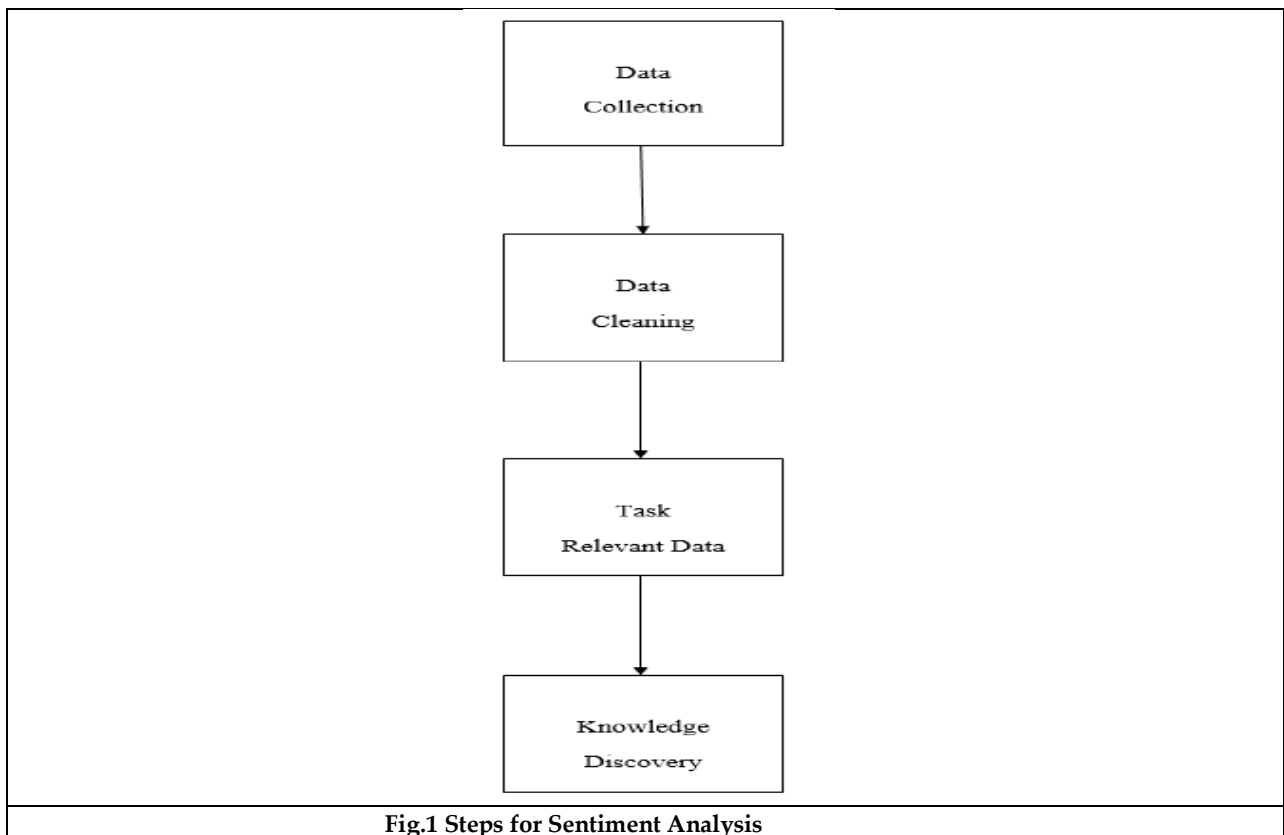
Pushpa Bansiwala et al.,

Table.2 Summary of Results

S.No	Parameter	Value
1	Dataset	Amazon Product Review Dataset
2	ML Category	Supervised
3	Algorithm	Bayesian Optimized Neural Network
4	Iterations to Convergence	45
5	Mean Squared Error (MSE)	0.95973
6	Mean Absolute Error (MAE)	0.67
7	Accuracy %	99.34

Table 2. Comparison with Previous Work

S.No.	Author	Accuracy (%)
1	Zhao et al. [1]	93.5
2	Vohra et a. [3]	92.59
3	Phan et al. [4]	82.5
4	Obiedat et al. [5]	89
5	Saha et al. [7]	60.5395
6	Estrada et al. [8]	93
7	Proposed Work	99.34%





Pushpa Bansiwali et al.,

1	reviewerName	overall	reviewText	reviewTime	day_diff	helpful_ye	helpful_no	total_vote	score_pos	score_ave	wilson_lower_bound
2	0	4	No issues.	23-07-2014	138	0	0	0	0	0	0
3	1 Omie	5	Purchased this for my device, it worked as	25-10-2013	409	0	0	0	0	0	0
4	2 1K3	4	it works as expected. I should have sprung	23-12-2012	715	0	0	0	0	0	0
5	3 1m2	5	This think has worked out great.Had a diff.	21-11-2013	382	0	0	0	0	0	0
6	4 2&1/2Men	5	Bought it with Retail Packaging, arrived leg	13-07-2013	513	0	0	0	0	0	0
7	5 2Cents!	5	It's mini storage. It doesn't do anything els	29-04-2013	588	0	0	0	0	0	0
8	6 2K1Toaster	5	I have it in my phone and it never skips a bi	19-10-2013	415	0	0	0	0	0	0
9	7 35-year Technology (5	It's hard to believe how affordable digital l	07-10-2014	62	0	0	0	0	0	0
10	8 4evryoung	5	Works in a HTC Rezound. Was running shc	24-03-2014	259	1	0	1	1	1	0.206549
11	9 53rdcard	5	in my galaxy s4, super fast card, and am to	10-11-2013	393	0	0	0	0	0	0
12	10 808TREX50	5	I like this SD Card because it can take musi	05-11-2013	398	0	0	0	0	0	0
13	11 98020	3	It works, but file writes are a bit slower th	20-11-2013	383	0	0	0	0	0	0
14	12 9z4cda	5	THE NAME OF ITSELF SPEAKS OUT. GO SAN	07-04-2014	245	0	0	0	0	0	0
15	13 A4Q96 "Gadget Love	5	Solid SDHC card that is fast (at reading and	21-11-2013	382	0	0	0	0	0	0
16	14 Aaron "Aaron"	5	Heard that the card's write speed is insuffi	17-02-2014	294	0	0	0	0	0	0
17	15 Aaron "Aaron"	5	I bought this to use with my go pro hero 3	01-04-2013	616	0	0	0	0	0	0
18	16 Aaron Alvarez	5	got this because i had a 2 GB one that fill	03-02-2014	308	0	0	0	0	0	0
19	17 Aaron F. Virginie	5	Class 10 Speed Rating for Seamless Full HD	07-04-2013	610	0	1	1	-1	-1	0
20	18 Aaron Graves	5	The read and write speeds are better than	05-02-2014	306	0	0	0	0	0	0
21	19 Aaron	5	This works with the NL1520. No video stu	01-07-2014	160	0	0	0	0	0	0
22	20 Aaron	5	Works as expected. High transfer speed. I	27-10-2013	407	0	0	0	0	0	0
23	21 Aaron	5	Works great in a Samsung Galaxy S3. Forr	29-12-2013	344	0	0	0	0	0	0
24	22 Aaron Madden	5	SanDisk never disappoints. As always SanD	11-05-2013	576	0	0	0	0	0	0
25	23 aaron mckaig	5	Good price, works flawless in my Samsung	04-04-2014	248	0	0	0	0	0	0
26	24 Aaron Nash	5	San disk is hard to beat. You will pay more	23-03-2014	260	0	0	0	0	0	0
27	25 Aaron Smith	5	Installed in my Blackberry Q10 SQN100-1 e	28-01-2014	314	0	0	0	0	0	0
28	26 Aaron T. Swain	5	I just received my card, it is the class 10 64	26-07-2012	865	1	1	2	0	0.5	0.094531

Fig.2 Raw Data.

The data is fetched and loaded to the MATLAB workspace, as a sequence of strings.

The screenshot shows the MATLAB 'Import Wizard' interface. The 'Import Selection' is set to 'Numeric Matrix'. The imported data is shown in the workspace as a table with the following structure:

Number	reviewerName	overall	reviewText	reviewTime	day_diff	helpful_yes	helpful_no	total_vote	score_pos_n	score_averag	wilson_low
1	0	4	No issues.	23-07-2014	138	0	0	0	0	0	0
2	1 Omie	5	Purchased t...	25-10-2013	409	0	0	0	0	0	0
3	2 1K3	4	it works as ...	23-12-2012	715	0	0	0	0	0	0
4	3 1m2	5	This think h...	21-11-2013	382	0	0	0	0	0	0
5	4 2&1/2...	5	Bought it w...	13-07-2013	513	0	0	0	0	0	0
6	5 2Cents!	5	It's mini sto...	29-04-2013	588	0	0	0	0	0	0
7	6 2K1Toaster	5	I have it in ...	19-10-2013	415	0	0	0	0	0	0
8	7 35-year Tec...	5	It's hard to ...	07-10-2014	62	0	0	0	0	0	0
9	8 4evryoung	5	Works in a ...	24-03-2014	259	1	0	1	1	1	0.206549314
10	9 53rdcard	5	in my galax...	10-11-2013	393	0	0	0	0	0	0
11	10 808TREX50	5	I like this S...	05-11-2013	398	0	0	0	0	0	0
12	11 98020	3	It works, bu...	20-11-2013	383	0	0	0	0	0	0
13	12 9z4cda	5	THE NAME ...	07-04-2014	245	0	0	0	0	0	0
14	13 A4Q96 "Ga...	5	Solid SDHC...	21-11-2013	382	0	0	0	0	0	0
15	14 Aaron "Aar...	5	Heard that ...	17-02-2014	294	0	0	0	0	0	0
16	15 Aaron "Aar...	5	I bought thi...	01-04-2013	616	0	0	0	0	0	0
17	16 Aaron Alvar...	5	got this bec...	03-02-2014	308	0	0	0	0	0	0
18	17 Aaron F. Vir...	5	Class 10 Sp...	07-04-2013	610	0	1	1	-1	-1	0
19	18 Aaron Graves	5	The read an...	05-02-2014	306	0	0	0	0	0	0
20	19 Aaron	5	This works ...	01-07-2014	160	0	0	0	0	0	0
21	20 Aaron	5	Works as ex...	27-10-2013	407	0	0	0	0	0	0
22	21 Aaron	5	Works grea...	29-12-2013	344	0	0	0	0	0	0
23	22 Aaron Mad...	5	SanDisk nev...	11-05-2013	576	0	0	0	0	0	0
24	23 aaron mcka...	5	Good price...	04-04-2014	248	0	0	0	0	0	0
25	24 Aaron Nash	5	San disk is ...	23-03-2014	260	0	0	0	0	0	0
26	25 Aaron Smith	5	Installed in ...	28-01-2014	314	0	0	0	0	0	0
27	26 Aaron T. Sw...	5	I just receiv...	26-07-2012	865	1	1	2	0	0.5	0.094531206
28	27 A. Atkinson	5	Stuck it in ...	21-11-2012	747	0	0	0	0	0	0

Fig.3 Imported Data to Workspace

Figure 3 shows the raw data being imported to the workspace.





Pushpa Bansiwal et al.,

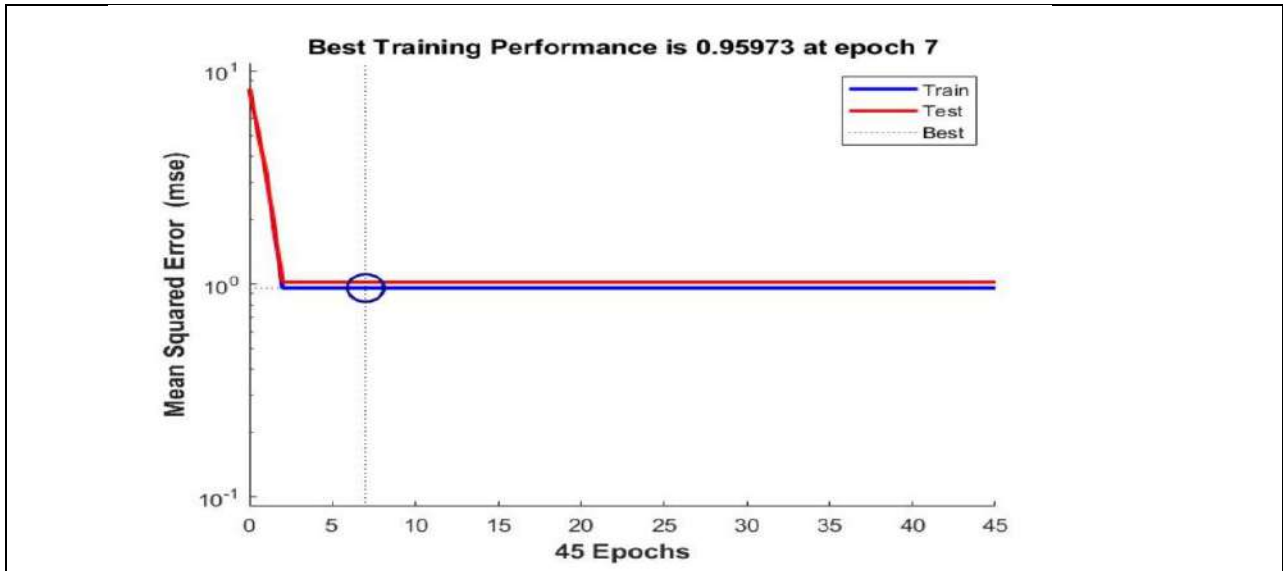


Fig.4 Convergence of algorithm w.r.t. iterations

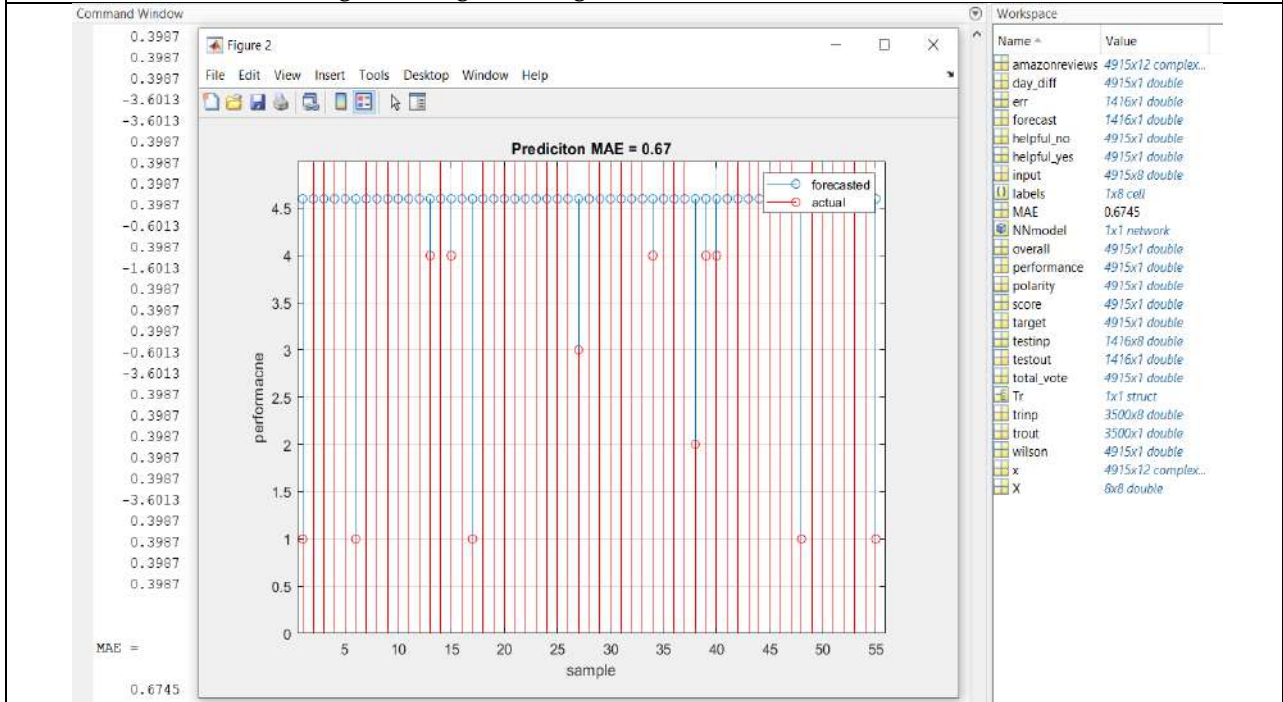


Fig.5 Command Line Screenshot

Figure 5 shows the command line screenshot for mean absolute error.





Katiparinaah and Body Adiposity Index in Non Alcoholic Fatty Liver Disease : A Cross-Sectional Analytical Study

Tanya Gusian¹, Amrish Chanana^{2*} and Atal Bihari Trivedi³

¹M.D Scholar, Department of Rachana Sharir, Ch. Brahm Prakash Ayurved Charak Santhan, Khera Dabar, (Affiliated to Guru Gobind Singh Indraprastha University), Delhi, India.

²Assistant Professor, Department of Rachana Sharir, Ch. Brahm Prakash Ayurved Charak Santhan, Khera Dabar, (Affiliated to Guru Gobind Singh Indraprastha University), Delhi, India.

³Associate Professor, Department of Kayachikitsa, Ch. Brahm Prakash Ayurved Charak Santhan, Khera Dabar, (Affiliated to Guru Gobind Singh Indraprastha University), Delhi, India.

Received: 12 Jun 2024

Revised: 13 Aug 2024

Accepted: 19 Oct 2024

*Address for Correspondence

Amrish Chanana

Assistant Professor,

Department of Rachana Sharir,

Ch. Brahm Prakash Ayurved Charak Santhan, Khera Dabar,

(Affiliated to Guru Gobind Singh Indraprastha University),

Delhi, India.

E.Mail: amrish.chanana@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

Cross-sectional analytical study

There is an increasing prevalence of non alcoholic fatty liver disease (NAFLD) in India at an early age due to the unhealthy diet and lifestyle of people. Modern medicine does appreciate the prediction of fat deposition through body mass index (BMI) to predict the metabolic risk, but BMI directly depends upon the weight of the body. Unlike BMI, body adiposity index (BAI) does not depend upon the weight of the body. The concept of proportionate body measurements (*Sama Sharir*) of Ayurveda is associated with good health and long life. The objective of the study was to discriminate the measurement of *Katiparinaah* and body adiposity index in between NAFLD patients and healthy individuals without fatty liver. Cross-sectional analytical study A total of 153 females of age group 20 to 50 years were registered in two groups; 66 NAFLD female patients (case group) and 87 healthy without fatty liver females (control group). Waist circumference, one *anguli* breadth, *Katiparinaah*, and body adiposity index were recorded for the registered subjects after informed consent. The measurement of *Katiparinaah* and body adiposity index is significantly higher in the female patients of NAFLD in comparison to healthy females without fatty liver ($P < 0.05$). The ROC analysis indicates that *Katiparinaah* and body adiposity are moderate to good predictors for predicting NAFLD. *Katiparinaah* ≥ 57.03 *anguli* and body adiposity index of ≥ 32.53 are the cut-off points for the discrimination between healthy non-fatty liver and non-alcoholic fatty liver disease. There is a significant difference in *Katiparinaah* and body adiposity index in the female patients of non-alcoholic fatty liver disease in comparison to healthy female subjects, Females of *Katiparinaah* ≥ 57.03 *anguli* or body adiposity index ≥ 32.53 are at higher risk of developing NAFLD.

Keywords: non-alcoholic fatty liver, *Sama Sharir*, *Katiparinaah*, body adiposity index, anthropometry.





Tanya Gusian et al.,

INTRODUCTION

The liver is the most common site for the accumulation of fat because it plays a central role in fat metabolism. Depending upon the cause and amount of accumulation, fatty liver changes may be severe, irreversible, and may result in cell death [1]. Adiposity (excess fat deposition) is the second most common cause of fatty liver and the most common cause of non-alcoholic fatty liver disease (NAFLD). There is an increasing prevalence of NAFLD in India at an early age due to the unhealthy lifestyle of people and it contributes to a large proportion of liver disease burden in the world [2]. The concept of *sama sharir* (proportionate body measurements) of Ayurveda is associated with good health and long life. The scholars of Ayurveda (*Charak, Sushruta, Vagabhat*) have mentioned the reference value of measurements of different parts of the body. [3]-[5]. These reference values give the measurements of a proportionate body in one's finger measurement, called *swa anguli pramana*. Unlike the contemporary reference values of measurement of the body, the *swa anguli* measurement is personalized. A person of proportionate body measurements (*sama sharir*) had a height of 84 *anguli*, *Prabahu* (forearm) 16 *anguli* and *Prapani* 15 *anguli*, *Katiparinaah* (waist circumference) 50 *anguli*. It can be hypothesized that the deviations in the measurement of body parts from the reference value can be considered as disproportionate body and it can lead to diseased condition. *Katiparinaah* indicates the abdominal obesity and its deviation from the reference value can be a predictor of fatty liver disease of adiposity origin (NAFLD) in a very early stage of the disease.

Modern medicine does appreciate the prediction of fat deposition through body mass index (BMI) to predict the metabolic risk and diseases linked to adiposity, but BMI directly depends upon the weight of the person [6] which does not include fat mass only but also the bone, muscle mass, and water content of the person, so the BMI does not give the true picture of adiposity. In such a scenario, it is necessary to make available an alternative method, which can predict the excess fat deposition and the diseases related to it. Body Adiposity Index (BAI) has been presented as a potential alternative to BMI. Unlike BMI, BAI does not depend upon the weight of the body and is calculated by the size of the hip circumference compared to the person's height. Bergman et al. reported that BAI is strongly correlated with adiposity, and in contrast to BMI, BAI is equally good for both genders and differing ethnicities. BAI could directly estimate the percent body fat without the need for further correction for sex or age [7].

The objective of the study was to analyze the difference in the measurement of *Katiparinaah* (in *anguli*) and body adiposity index independently between NAFLD patients and healthy individuals without fatty liver.

MATERIALS AND METHODS

Study design- cross-sectional analytical study. The study population for this study was the female patients approaching the hospital for consultation and the healthy females residing in nearby villages. A total of 153 females were registered in the study. Of the total study participants, 66 females were patients of NAFLD in case group and 87 healthy females without fatty liver were registered in control group. (Table 1). Ethical permission was taken from the institute's ethical committee (letter no CBP-IEC/2020/RS 07/MD/28 dated 29/01/2022) and the study was registered in the Clinical trial registry India before the start of enrolment of study participants. It is obvious that the anthropometric measurements of body naturally vary between males and females, so the present study is limited to female study participants only to remove the gender confounder. The study participants were well-versed about the data collection and their privacy. All the study participants were of age group 20 to 50 years, and registered after informed consent, in the year 2022-23. All the 153 participants of both the groups were confirmed for fatty liver and non-fatty liver by abdominal ultrasound. Individuals with a history of alcohol consumption, medications that cause liver damage and steatosis like estrogen, steroids, tetracycline, active or previous infection with hepatitis B and C, and other liver disease, chronic illness (T.B), diabetic patients, malnourished individuals, and individuals under starvation and pregnant women were excluded from the study.





Tanya Gusian et al.,

Standard anthropometric measurements (height, hip circumference, waist circumference) were measured by stadiometer, and measuring tape respectively. Four fingers' width of the right hand was measured by a digital vernier caliper of 300 mm scale between the proximal inter-phalangeal joint of the 2nd digit and the distal inter-phalangeal joint of the 5th digit from the dorsal aspect after keeping the hand in supine on the top of a sturdy table. This measurement was divided by four to get one *anguli* (finger) width [8].

All the data was recorded in the Performa along with other demographic data. The *Katiparinaah* in *anguli praman* was calculated by dividing the waist circumference (cm) by one *anguli* width (cm) of the person. Body adiposity index was calculated by the formula:

$$\text{BAI} = \text{Hip circumference in cm} / (\text{height (m)}^{1.5} - 18) [9]$$

The difference of the *Katiparinaah* and body adiposity index individually among the two groups was analyzed using unpaired t test and the cut-off point of the two variables to predict the NAFLD was measured by Receiver operating characteristic (ROC).

RESULT

Discrimination of *Katiparinaah* between NAFLD and healthy females without fatty liver

The mean and standard deviation of *Katiparinaah* in control and case group was 53.64 ± 3.61 *anguli*, and 58.52 ± 5.65 *anguli* respectively. The difference of *Katiparinaah* in between case and control group was statistically significant ($P < 0.0001$) (Table 2). The ROC (Receiver Operating Characteristic) analysis was performed to evaluate the diagnostic accuracy of *Katiparinaah* in detecting Fatty Liver Disease. The area under ROC is a measure of the overall discriminatory ability of *Katiparinaah* in distinguishing between individuals with and without fatty liver disease, the area under ROC was found to be 0.768, indicating a reasonably good discriminatory power. The optimum cut-off value (the threshold of *Katiparinaah* level) that provides the best balance between sensitivity and specificity for detecting non-alcoholic fatty liver disease in females was: *Katiparinaah* ≥ 57.03 *anguli* (Table 3)

Discrimination in Body adiposity index between NAFLD and healthy without fatty liver females

The mean and standard deviation of body adiposity index in control and case group was 28.5 ± 2.56 , and 31.22 ± 5.46 respectively. The difference of body adiposity index was statistically significant in case and control group ($P < 0.0001$) (Table 4). The above results indicate that the BAI is a moderate predictor of Fatty Liver Disease in females. A body adiposity index value of 32.53 or higher is the optimal cut-off for detecting the NAFLD in females, achieving a sensitivity of 37.9% and specificity of 97.9%. (Table 5)

Variation of *Katiparinaah* with fatty liver grades in NAFLD patients

The mean and standard deviation of *Katiparinaah* in grade I and grade II was 57.28 ± 5.03 *anguli*, and 62.34 ± 6.01 *anguli* respectively, and the difference of *Katiparinaah* in grade I and grade II fatty liver disease was found statistically significant ($P = 0.0015$) (Table 6)

Variation of body adiposity index with fatty liver grades in NAFLD patients

The mean and standard deviation of body adiposity index in grade I and grade II fatty liver was 30.13 ± 4.86 , and 34.57 ± 6.09 respectively. Statistically significant difference of body adiposity index was found in grade I and grade II fatty liver disease ($P = 0.0041$) (Table 7)

DISCUSSION

The individuals who are disproportionate by the criteria of *anguli pramana* may have *meda dhatu dushti*, which could be associated with diseases of excess fatty deposition like risk of cardio vascular ischemia, non-alcoholic fatty liver etc. The *Katiparinaah* and body adiposity index are significantly higher in the females of NAFLD in comparison to healthy females without fatty liver ($P < 0.05$). The ROC analysis indicates that *Katiparinaah* and body adiposity are





Tanya Gusian et al.,

moderate to good predictors for predicting NAFLD. Females of *Katiparinaah* ≥ 57.03 *anguli* and body adiposity index of ≥ 32.53 are at a higher risk of developing NAFLD. The severity (grade) of fatty liver disease also increases with the increase of *Katiparinaah* and body adiposity index. Previous studies of *anguli praman* and anthropometry have demonstrated that smaller facial structures and shorter neck limbs are associated with higher insulin resistance markers (obesity, adiposity, glycemia, insulin, and HOMA-R, but larger forehead and face were associated with higher beta cell function (HOMA-B) [10], shorter-legged children suffered more bronchitis than the long [11]. Increase in childhood length of the leg and not the length of the trunk, is related to decreased mortality due to congestive heart disease but increased risk of cancer [12] Higher risk of Prostate and testicular colorectal cancers are associated with long legs in tall people [13].

With the above studies we can put forward that the deviation of measurements from the reference values given by scholars of Ayurveda are associated with diseases and increased *Katiparinaah* and body adiposity index can be predictors of NAFLD in a very initial stage of disease. As the topographical landmarks for the measurement of *Katiparinaah* are not mentioned in the *Samhita*, so, the contemporary standard anthropometric measurement of waist circumference has been used for the estimation of *Katiparinaah*, there could be other methods also for the estimation of one *anguli* breadth like the breadth of the middle finger at the proximal inter-phalangeal joint. This may affect the result and the above values are specific to the dataset and population used for the analysis and may not be generalized to other populations without further validation. This data is from the female subjects visiting the hospital for treatment and the healthy females residing nearby villages of the hospital so it needs further validation with inclusion of the male gender, larger sample size, and populations of different geographical locations. In the present data set females of fatty liver grade III were not found. The findings of the study support the hypothesis of difference of *Katiparinaah* and BAI between NAFLD & healthy females without fatty liver and its utility in the prediction of risk of fatty liver disease of non-alcoholic origin.

REFERENCES

1. Mohan H. Textbook of Pathology 4th edition. New Delhi: Jaypee Brothers Medical Publishers (P) Ltd, 2000, p. 28.
2. Shalimar, Elhence A. Prevalence of non alcoholic fatty liver disease in India: A systemic review and metaanalysis. journal of clinical and experimental hepatology, 2022;12:3: 818-829
3. Shastri K, editor. Charak Samhita Vimana Sthana 8/117 (Roga Bishagitiya Viman Adhyaya). 1st edition. Varanasi. Chaukhambha Bharati Academy; 2001
4. Shastri AD, Sushruta Samhita Sutra Sthana 35/13 (Aturupkarmniya Adhyaya) 1sted. Varanasi: Chaukhambha Sanskrit Sansthan. 2002
5. Sharma S: Ashtang Sangraha Sharir Sthana 8/36 Prakriti Bhediya Adhaya, Shashilekha Sanskrit Commentary by Indu, 1st. ed. Varanasi: Chaukhambha Sanskrit Series.2008
6. Sattar A, Baig S et al. Factors affecting BMI: Professional Med Journal 2013;20(6): 956-964
7. Richard N. Bergman et al. A better index of body adiposity: Obesity; 2011,19(5): 1083-1089
8. Manual of Standard operative procedures for Prakriti assessment, first edition, New Delhi: CCRAS, Ministry of AYUSH, Govt. of India 2018: p 02
9. Lo'pez AA, Cespedes ML, Vicente T, Tomas M, Bennasar-Veny M, et al. Body Adiposity Index Utilization in a Spanish Mediterranean Population: Comparison with the Body Mass Index. PLoS ONE 2012 7(4): 1-7
10. Shirodkar jyoti, Sayyad G, Nanal M, Yajnik C. Anguli Parimana in Ayurveda and its association with adiposity and diabetes. Journal of Ayurveda & Integrated medicine 2014 5(3): 177-183
11. Gunnell DJ, Davey Smith G, Frankel S et al. Childhood leg length and adult mortality: follow up of the Carnegie (Boyd Orr) survey of diet and health in pre war Britain. J epidemiol community health 1998;52: 142-152
12. Gunnell DJ, Smith GD, Holly JM, Frankel S. Leg length and risk of cancer in the Boyd Orr cohort. BMJ 1998;317:1350-1
13. Gunnell DJ, Okasa M, Smith GD, oliver SE, Sandhu J, Holly JM. Height, leg length, and cancer risk: A systematic review. Epidemiol Rev 2001; 23:313-42





Tanya Gusian et al.,

Table 1: Distribution of Subjects in Case and Control Group

Variables	Control (healthy without fatty liver)	Case (NAFLD patients)
Number of Subjects	87	66
Age (mean) years	33.55	38.03

Table 2: Difference in Katiparinaah Between NAFLD and Healthy Without Fatty Liver Females

Variable	Katiparinaah (in swa anguli)		unpaired t test	
	Mean	SD	t-value	P-value
Control (healthy without fatty liver)	53.64	3.61	6.56	<0.0001
Case (NAFLD)	58.52	5.65		

Table 3: ROC For The Optimum Cut-Off Point Of Katiparinaah

Area under ROC	.768
Optimum cut-off (Katiparinaah)	Katiparinaah \geq 57.03 anguli
Sensitivity (%)	59.1
Specificity (%)	85.1

Table 4: Discrimination In Body Adiposity Index Between NAFLD And Healthy Without Fatty Liver Females

Variable	BAI (Body adiposity index)		unpaired t test	
	Mean	SD	t-value	P-value
Control (healthy without fatty liver)	28.5	2.56	4.12	<0.0001
Case (NAFLD)	31.22	5.46		

Table 5: ROC for the Optimum Cut-Off Point Of Body Adiposity Index

Area under ROC	.645
Optimum cut-off (BAI)	BAI \geq 32.53
Sensitivity (%)	37.9
Specificity (%)	97.9

Table 6: Variation of Katiparinaah With Fatty Liver Grades In NAFLD Patients

Variable	Katiparinaah (in anguli)		unpaired t test	
	Mean	SD	t-value	P-value
Grade I fatty liver	57.28	5.03	3.33	0.0015
Grade II fatty liver	62.34	6.01		

Table 7: Variation of Body Adiposity Index with Fatty Liver Grades in NAFLD Patients

Variable	Body adiposity index		unpaired t test	
	Mean	SD	t-value	P-value
Grade I fatty liver	30.13	4.86	2.97	0.0041
Grade II fatty liver	34.57	6.09		





Enhancing Comprehensive Biopsychosocial Assessment in Chronic Musculoskeletal Pain Management: A Multidimensional Approach

Thangamani Ramalingam Alagappan^{1*} and Sudipta Roy²

¹Lecturer, Department of Physiotherapy, The Sarvajanic College of Physiotherapy, (Affiliated to Veer Narmad South Gujarat University), Surat, Gujarat, India.

²Adjunct Professor, Department of Physiotherapy, PP Savani University, Surat, Gujarat, India.

Received: 11 Jun 2024

Revised: 13 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Thangamani Ramalingam Alagappan

Lecturer, Department of Physiotherapy,
The Sarvajanic College of Physiotherapy,
(Affiliated to Veer Narmad South Gujarat University),
Surat, Gujarat, India
E.Mail: atramalingam@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Chronic musculoskeletal pain (CMP) poses significant challenges in healthcare, necessitating a comprehensive understanding and management strategy. This article explores the utility of a multidimensional biopsychosocial assessment approach in CMP management. Physiotherapists, as frontline healthcare providers, play a crucial role in implementing this approach by utilizing a diverse array of subjective and objective assessment tools. The bio-psycho-social model underscores the interaction of biological, psychological, and social factors in shaping pain experiences. To address the multifaceted nature of CMP, a paradigm shift towards a bio-psycho-social model of assessment and management has gained momentum in clinical practice. Assessment tools ranging from pain intensity scales to disability assessment instruments capture various dimensions of pain perception and functional impairment. Additionally, tools targeting nervous system sensitization, fear-avoidance beliefs, psychological distress, and quality of life offer insights into contributing factors and overall well-being. The Integration of these assessment tools within a multidimensional framework enables tailored interventions to address individual patient needs effectively in health care. This comprehensive approach not only optimizes treatment outcomes but also enhances the overall quality of life for CMP patients. Continued research and clinical innovation are essential for further refining assessment techniques and treatment strategies, ultimately improving patient care and facilitating long-term recovery and functional restoration.

Keywords: Chronic Musculoskeletal Pain, Bio-Psychosocial Assessment, Physiotherapy, Multidimensional Approach, Pain Management.



**Thangamani Ramalingam Alagappan and Sudipta Roy****INTRODUCTION**

Chronic musculoskeletal pain (CMP) is a complex and prevalent health concern, affecting millions of individuals worldwide and posing significant challenges for healthcare professionals. Unlike acute pain, which typically resolves with time and treatment, CMP persists over extended periods, often defying conventional therapeutic approaches. To address the multifaceted nature of CMP, a paradigm shift towards a bio-psychosocial model of assessment and management has gained momentum in clinical practice. Physiotherapists, as frontline healthcare providers, are uniquely positioned to implement this comprehensive approach, drawing upon a diverse array of assessment tools to elucidate the intricate interplay of biological, psychological, and social factors underlying CMP [1-3]. This article explores the utility of various subjective and objective assessment instruments within a multidimensional framework to enhance the management of CMP and improve patient outcomes.

Bio-Psychosocial Assessment in Chronic Musculoskeletal Pain

The bio-psychosocial model represents a holistic approach to understanding pain experiences, emphasizing the interaction between biological, psychological, and social factors in shaping individuals' perceptions and responses to pain stimuli. In the context of CMP, this model underscores the importance of assessing not only the physical manifestations of pain but also its cognitive, emotional, and social dimensions. Physiotherapists play a pivotal role in conducting comprehensive bio-psychosocial assessments, which serve as the foundation for targeted and personalized intervention strategies [4-6].

Pain Assessment Tools

Objective evaluation of pain intensity, quality, and impact on daily functioning is fundamental in guiding CMP management. A range of pain assessment tools is available to capture the multidimensional nature of pain experiences. The Numerical Pain Rating Scale (NPRS), Pain and Discomfort Module (PDM), Brief Pain Inventory (BPI), and McGill Pain Questionnaire (MPQ) are widely used instruments that provide valuable insights into various facets of pain perception and its effects on patients' lives. These tools enable physiotherapists to quantify pain severity, assess the affective and sensory components of pain, and evaluate its interference with activities of daily living [7-12].

Disability Assessment Tools

Assessing pain-related disability is essential for gauging the functional impact of CMP and monitoring treatment progress. Disability assessment tools such as the Shoulder Pain and Disability Scale (SPADI), Roland-Morris Disability Questionnaire (RMDQ), Oswestry Low Back Pain Disability Index, and Neck Disability Index (NDI) offer standardized measures of functional impairment across different anatomical regions. By quantifying the extent of pain-related disability, these tools assist physiotherapists in setting realistic rehabilitation goals and tracking patients' responses to intervention over time [13-18].

Nervous System Sensitization Assessment Tools

Central sensitization, characterized by heightened neuronal responsiveness to nociceptive stimuli, is increasingly recognized as a contributing factor in CMP. Assessment tools such as the Pressure Pain Threshold (PPT) and Central Sensitization Inventory (CSI) help identify patients with aberrant pain processing mechanisms. By quantifying pain sensitivity and central sensitization symptoms, these tools inform the selection of appropriate therapeutic interventions, such as graded exposure therapy or desensitization techniques, aimed at modulating neural excitability and reducing pain hypersensitivity [19-23].

Fear-Avoidance Assessment Tools

Fear-avoidance beliefs and behaviors can exacerbate pain intensity and disability in CMP patients, perpetuating a cycle of pain and functional limitations. Assessment tools such as the Fear-Avoidance Components Scale (FACS), Tampa Scale of Kinesiophobia (TSK), and Fear-Avoidance Beliefs Questionnaire (FABQ) assess the extent to which



**Thangamani Ramalingam Alagappan and Sudipta Roy**

patients' beliefs about pain influence their activity levels and functional abilities. By identifying maladaptive fear-avoidance patterns, physiotherapists can tailor interventions to address cognitive restructuring, graded exposure, and behavioral activation, thereby empowering patients to gradually resume activities and regain confidence in their physical capabilities [24-31].

Psychological Assessment Tools

Psychological distress, including symptoms of depression, anxiety, and pain catastrophizing, commonly coexists with CMP and can significantly impact patients' pain experiences and treatment outcomes. Assessment tools such as the Depression scale (PHQ-9), Depression Anxiety and Stress Scale (DASS), and Pain Catastrophizing Scale (PCS) provide standardized measures of psychological distress and maladaptive coping strategies. Additionally, instruments like the Pain Self-Efficacy Questionnaire (PSEQ) and Chronic Pain Coping Inventory (CPCI) assess patients' beliefs about their ability to manage pain and their repertoire of coping strategies. By addressing underlying psychological factors, physiotherapists can enhance patients' self-efficacy and resilience, fostering adaptive pain coping mechanisms and improving overall psychological well-being [32-41].

Neurophysiology of Pain Education Tools

Educating patients about the neurobiological mechanisms underlying pain perception is an essential component of CMP management. Assessment tools such as the Neurophysiology Pain Questionnaire (NPQ) evaluate patients' understanding of pain neurobiology and their readiness to engage in self-management strategies. By identifying gaps in patients' knowledge and misconceptions about pain, physiotherapists can tailor educational interventions to address specific learning needs and promote active participation in treatment [42].

Quality of Life Assessment Tools

Assessing quality of life is crucial for capturing the broader impact of CMP on patients' physical, emotional, and social well-being. Instruments such as the WHOQOL-BREF, SF-36, and EQ-5D offer comprehensive measures of health-related quality of life across various domains. By evaluating patients' perceived satisfaction with life and functional status, these tools provide valuable insights into the overall impact of CMP on their daily functioning and social interactions. Additionally, quality-of-life assessment tools can help identify areas of impairment requiring targeted interventions, such as pain management strategies, social support interventions, or vocational rehabilitation programs [43-46].

Activity Pacing Assessment Tools

Activity pacing is a key self-management strategy for CMP patients, enabling them to optimize their activity levels while avoiding exacerbations of pain and fatigue. Assessment tools such as the Activity Pacing Questionnaire (APQ) and Activity Pacing and Risk of Overactivity Questionnaire (APRQ) assess patients' pacing behaviors and attitudes towards activity engagement. By identifying patterns of overactivity or underactivity, physiotherapists can guide patients in developing balanced activity schedules and incorporating rest breaks to conserve energy and manage symptoms effectively [47, 48].

Central Sensitization Syndrome Assessment Tools

Identifying patients with central sensitization syndrome is critical for tailoring appropriate interventions to address underlying neurophysiological mechanisms. Diagnostic criteria for fibromyalgia, including the widespread pain index (WPI) and symptom severity scale (SS), help differentiate patients with central sensitization from those with primarily nociceptive pain. Additionally, tools such as the Insomnia Severity Index (ISI) and Fatigue Severity Scale (FSS) assess symptoms commonly associated with central sensitization, such as sleep disturbances and fatigue. By recognizing the broader symptomatology of central sensitization, physiotherapists can implement multimodal treatment approaches targeting pain modulation, sleep hygiene, and stress management [49-52].



**Thangamani Ramalingam Alagappan and Sudipta Roy****Patient Satisfaction Assessment Tools**

Evaluating patient satisfaction is essential for assessing the effectiveness of treatment interventions and identifying areas for improvement in service delivery. Instruments such as the Short Assessment of Patient Satisfaction Scale (SAPS) and Satisfaction with Life Scale (SWLS) provide standardized measures of patients' subjective experiences and overall satisfaction with care. By soliciting feedback from patients about their treatment experiences and perceived outcomes, physiotherapists can gauge the effectiveness of interventions and tailor future treatment plans to better meet patients' needs and preferences [53-55].

Social Assessment Tools

Understanding the social context in which pain occurs is crucial for addressing contextual factors that may influence patients' pain experiences and treatment outcomes. Social assessment tools such as the Multidimensional Scale of Perceived Social Support (MSPSS), Social Support Questionnaire (SSQ), and Work Limitations Questionnaire (WLQ) assess patients' social support networks, work productivity, and daily activities. By identifying sources of social support and potential barriers to participation in meaningful activities, physiotherapists can collaborate with patients to develop holistic treatment plans that address not only their physical symptoms but also their social and environmental context. Additionally, instruments like the Social Impact of Pain (SIP) Questionnaire and Dyadic Adjustment Scale (DAS) evaluate the broader social and interpersonal implications of CMP, helping physiotherapists recognize and address relational challenges that may arise as a result of chronic pain [56-63].

CONCLUSION

Comprehensive biopsychosocial assessment is integral to effective CMP management, enabling physiotherapists to understand the multifaceted nature of pain experiences and tailor interventions to address individual patient needs. By integrating subjective and objective assessment tools within a multidimensional framework, physiotherapists can gain a nuanced understanding of patients' pain experiences and develop targeted treatment plans that promote optimal outcomes and improve overall quality of life. Moving forward, continued research and clinical innovation are needed to further refine assessment techniques and treatment strategies in CMP management, ultimately enhancing patient care and facilitating long-term recovery and functional restoration.

ACKNOWLEDGEMENTS

None

Conflicts of Interest

The authors declare no conflict of interest

REFERENCES

1. Hawker, G.A., The assessment of musculoskeletal pain. *Clin Exp Rheumatol*, 2017. 35 Suppl 107(5): p. 8-12.
2. Wijma, A.J., et al., Clinical biopsychosocial physiotherapy assessment of patients with chronic pain: The first step in pain neuroscience education. *Physiother Theory Pract*, 2016. 32(5): p. 368-84.
3. Management, A.S.o.A.T.F.o.C.P., Practice guidelines for chronic pain management: an updated report by the American Society of Anesthesiologists Task Force on Chronic Pain Management and the American Society of Regional Anesthesia and Pain Medicine. *Anesthesiology*, 2010. 112(4): p. 810-833.
4. Turk, D.C., et al., Core outcome domains for chronic pain clinical trials: IMMPACT recommendations. *Pain*, 2003. 106(3): p. 337-345.
5. El-Tallawy, S.N., et al., Management of Musculoskeletal Pain: An Update with Emphasis on Chronic Musculoskeletal Pain. *Pain and Therapy*, 2021. 10(1): p. 181-209.



**Thangamani Ramalingam Alagappan and Sudipta Roy**

6. Litcher-Kelly, L., et al., A systematic review of measures used to assess chronic musculoskeletal pain in clinical and randomized controlled clinical trials. *J Pain*, 2007. 8(12): p. 906-13.
7. Jensen, M.P. and C.A. McFarland, Increasing the reliability and validity of pain intensity measurement in chronic pain patients. *Pain*, 1993. 55(2): p. 195-203.
8. Broome, M.E., Measurement of Pain: Self-Report Strategies. *Journal of Pediatric Oncology Nursing*, 1991. 8(3): p. 131-133.
9. Mason, V.L., S.M. Skevington, and M. Osborn, A measure for quality of life assessment in chronic pain: preliminary properties of the WHOQOL-pain. *J Behav Med*, 2009. 32(2): p. 162-73.
10. Poquet, N. and C. Lin, The Brief Pain Inventory (BPI). *J Physiother*, 2016. 62(1): p. 52.
11. Ngamkham, S., et al., The McGill Pain Questionnaire as a multidimensional measure in people with cancer: an integrative review. *Pain Manag Nurs*, 2012. 13(1): p. 27-51.
12. Öncü, J., R. Ilişer, and B. Kuran, Cross-cultural adaptation of the Orebro Musculoskeletal Pain Questionnaire among Turkish workers with low back pain. *J Back Musculoskelet Rehabil*, 2016. 29(1): p. 135-43.
13. Breckenridge, J.D. and J.H. McAuley, Shoulder Pain and Disability Index (SPADI). *J Physiother*, 2011. 57(3): p. 197.
14. Roland, M. and R. Morris, A study of the natural history of back pain. Part I: development of a reliable and sensitive measure of disability in low-back pain. *Spine (Phila Pa 1976)*, 1983. 8(2): p. 141-4.
15. Macedo, L.G., et al., Responsiveness of the 24-, 18- and 11-item versions of the Roland Morris Disability Questionnaire. *Eur Spine J*, 2011. 20(3): p. 458-63.
16. Chiarotto, A., et al., Roland-Morris Disability Questionnaire and Oswestry Disability Index: Which Has Better Measurement Properties for Measuring Physical Functioning in Nonspecific Low Back Pain? Systematic Review and Meta-Analysis. *Phys Ther*, 2016. 96(10): p. 1620-1637.
17. Vernon, H. and S. Mior, The Neck Disability Index: a study of reliability and validity. *J Manipulative Physiol Ther*, 1991. 14(7): p. 409-15.
18. Anagnostis, C., R.J. Gatchel, and T.G. Mayer, The pain disability questionnaire: a new psychometrically sound measure for chronic musculoskeletal disorders. *Spine (Phila Pa 1976)*, 2004. 29(20): p. 2290-302; discussion 2303.
19. Borstad, J. and C. Woeste, The role of sensitization in musculoskeletal shoulder pain. *Brazilian Journal of Physical Therapy*, 2015. 19(4): p. 251-256.
20. Potter, L., C. McCarthy, and J. Oldham, Algometer reliability in measuring pain thresholds over normal spinal muscles to allow quantification of anti-nociceptive treatment effects. Vol. 9. 2006. 113-119.
21. Mayer, T.G., et al., The development and psychometric validation of the central sensitization inventory. *Pain Pract*, 2012. 12(4): p. 276-85.
22. Neblett, R., et al., The Central Sensitization Inventory (CSI): establishing clinically significant values for identifying central sensitivity syndromes in an outpatient chronic pain sample. *J Pain*, 2013. 14(5): p. 438-45.
23. Neblett, R., et al., Ability of the central sensitization inventory to identify central sensitivity syndromes in an outpatient chronic pain sample. *Clin J Pain*, 2015. 31(4): p. 323-32.
24. Neblett, R., et al., The Fear-avoidance Components Scale (FACS): Development and Psychometric Evaluation of a New Measure of Pain-related Fear Avoidance. *Pain Pract*, 2016. 16(4): p. 435-50.
25. Lundberg, M., J. Styf, and B. Jansson, On what patients does the Tampa Scale for Kinesiophobia fit? *Physiother Theory Pract*, 2009. 25(7): p. 495-506.
26. Bunketorp, L., et al., Evaluating the reliability of multi-item scales: a non-parametric approach to the ordered categorical structure of data collected with the Swedish version of the Tampa Scale for Kinesiophobia and the Self-Efficacy Scale. *J Rehabil Med*, 2005. 37(5): p. 330-4.
27. Pool, J.J.M., et al., The applicability of the Tampa Scale of Kinesiophobia for patients with sub-acute neck pain: a qualitative study. *Quality & Quantity*, 2009. 43(5): p. 773-780.
28. Klenerman, L., et al., The prediction of chronicity in patients with an acute attack of low back pain in a general practice setting. *Spine (Phila Pa 1976)*, 1995. 20(4): p. 478-84.
29. Pflingsten, M., et al., [Fear-avoidance-beliefs in patients with backpain]. *Schmerz*, 1997. 11(6): p. 387-95.
30. Waddell, G., et al., A Fear-Avoidance Beliefs Questionnaire (FABQ) and the role of fear-avoidance beliefs in chronic low back pain and disability. *Pain*, 1993. 52(2): p. 157-68.



**Thangamani Ramalingam Alagappan and Sudipta Roy**

31. Oliveira, C.B., et al., Psychometric Properties of the Photograph Series of Daily Activities-Short Electronic Version (PHODA-SeV) in Patients With Chronic Low Back Pain. *J Orthop Sports Phys Ther*, 2018. 48(9): p. 719-727.
32. Kroenke, K., R.L. Spitzer, and J.B. Williams, The PHQ-9: validity of a brief depression severity measure. *Journal of general internal medicine*, 2001. 16(9): p. 606-613.
33. Shea, T.L., A. Tennant, and J.F. Pallant, Rasch model analysis of the Depression, Anxiety and Stress Scales (DASS). *BMC Psychiatry*, 2009. 9: p. 21.
34. Kim, D., H. Bae, and Y. Chon Park, Validity of the Subjective Units of Disturbance Scale in EMDR. *J EMDR Prac Res*, (1): p. 57-62.
35. Kessler, R.C., et al., Screening for serious mental illness in the general population. *Arch Gen Psychiatry*, 2003. 60(2): p. 184-9.
36. Sullivan, M.J.L., S.R. Bishop, and J. Pivik, The Pain Catastrophizing Scale: Development and validation. *Psychological Assessment*, 1995. 7(4): p. 524-532.
37. Bansal, D., et al., Translation, Adaptation, and Validation of Hindi Version of the Pain Catastrophizing Scale in Patients with Chronic Low Back Pain for Use in India. *Pain Med*, 2016. 17(10): p. 1848-1858.
38. Clark, J., et al., What Are the Predictors of Altered Central Pain Modulation in Chronic Musculoskeletal Pain Populations? A Systematic Review. *Pain Physician*, 2017. 20(6): p. 487-500.
39. Schwarzer, R., The General Self-Efficacy Scale (GSE). 2012: p. 1-4.
40. Nicholas, M.K., The pain self-efficacy questionnaire: Taking pain into account. *Eur J Pain*, 2007. 11(2): p. 153-63.
41. Ferreira-Valente, A. and I. Queiroz-Garcia, Pain Diagnosis, Pain Coping, and Function in Individuals with Chronic Musculoskeletal Pain. 2020. 13: p. 783-794.
42. Catley, M.J., N.E. O'Connell, and G.L. Moseley, How good is the neurophysiology of pain questionnaire? A Rasch analysis of psychometric properties. *J Pain*, 2013. 14(8): p. 818-27.
43. Skevington, S.M., M. Lotfy, and K.A. O'Connell, The World Health Organization's WHOQOL-BREF quality of life assessment: psychometric properties and results of the international field trial. A report from the WHOQOL group. *Qual Life Res*, 2004. 13(2): p. 299-310.
44. Wong, G., SF-12 and SF-36 Health Survey, in *Encyclopedia of Gerontology and Population Aging*, D. Gu and M.E. Dupre, Editors. 2019, Springer International Publishing: Cham. p. 1-4.
45. Sinha, R., W.J. van den Heuvel, and P. Arokiasamy, Validity and Reliability of MOS Short Form Health Survey (SF-36) for Use in India. *Indian J Community Med*, 2013. 38(1): p. 22-6.
46. Balestroni, G. and G. Bertolotti, [EuroQol-5D (EQ-5D): an instrument for measuring quality of life]. *Monaldi Arch Chest Dis*, 2012. 78(3): p. 155-9.
47. Antcliff, D., et al., Assessing the Psychometric Properties of an Activity Pacing Questionnaire for Chronic Pain and Fatigue. *Phys Ther*, 2015. 95(9): p. 1274-86.
48. Abonie, U.S. and B.L. Seves, Assessment of Activity Pacing in Relation to Physical Activity and Health-Related Quality of Life in Adults with Multiple Sclerosis: A Foundation for Further Intervention Development. 2021. 23(5): p. 207-212.
49. Wolfe, F., N. Egloff, and W. Hauser, Widespread Pain and Low Widespread Pain Index Scores among Fibromyalgia-positive Cases Assessed with the 2010/2011 Fibromyalgia Criteria. *J Rheumatol*, 2016. 43(9): p. 1743-8.
50. Bastien, C.H., A. Vallieres, and C.M. Morin, Validation of the Insomnia Severity Index as an outcome measure for insomnia research. *Sleep Med*, 2001. 2(4): p. 297-307.
51. Lahan, V. and R. Gupta, Translation and validation of the insomnia severity index in hindi language. *Indian J Psychol Med*, 2011. 33(2): p. 172-6.
52. Krupp, L.B., et al., The fatigue severity scale. Application to patients with multiple sclerosis and systemic lupus erythematosus. *Arch Neurol*, 1989. 46(10): p. 1121-3.
53. Hawthorne, G., et al., Measuring patient satisfaction with health care treatment using the Short Assessment of Patient Satisfaction measure delivered superior and robust satisfaction estimates. *J Clin Epidemiol*, 2014. 67(5): p. 527-37.
54. Diener, E., et al., The Satisfaction With Life Scale. *Journal of personality assessment*, 1985. 49: p. 71-5.
55. Jaeschke, R., J. Singer, and G.H. Guyatt, Measurement of health status. Ascertaining the minimal clinically important difference. *Control Clin Trials*, 1989. 10(4): p. 407-15.





Thangamani Ramalingam Alagappan and Sudipta Roy

56. Lee, S.C., F.M. Moy, and N.N. Hairi, Validity and reliability of the Malay version multidimensional scale of perceived social support (MSPSS-M) among teachers. *Qual Life Res*, 2017. 26(1): p. 221-227.
57. Sarason, I.G., et al., A Brief Measure of Social Support: Practical and Theoretical Implications. *Journal of Social and Personal Relationships*, 1987. 4(4): p. 497-510.
58. Walker, T.J., et al., Validity and Reliability of the 8-Item Work Limitations Questionnaire. *J Occup Rehabil*, 2017. 27(4): p. 576-583.
59. Von Korff, M., et al., Graded chronic pain scale revised: mild, bothersome, and high-impact chronic pain. *Pain*, 2020. 161(3): p. 651-661.
60. Stangier, U., J. Schüller, and E. Brähler, Development and validation of a new instrument to measure social pain. *Scientific Reports*, 2021. 11(1): p. 8283.
61. Spanier, G.B., Measuring Dyadic Adjustment: New Scales for Assessing the Quality of Marriage and Similar Dyads. *Journal of Marriage and Family*, 1976. 38(1): p. 15-28.
62. McFarlane, A., et al., Methodological Issues in Developing a Scale to Measure Social Support. *Schizophrenia bulletin*, 1981. 7: p. 90-100.
63. Hirsh, A.T., et al., Psychometric properties of the community integration questionnaire in a heterogeneous sample of adults with physical disability. *Arch Phys Med Rehabil*, 2011. 92(10): p. 1602-10.





Influence of Triple Friction Pendulum Isolator Properties on Torsional Response of Building under Stochastic Ground Motions

Dipak Jivani^{1*} and R.G. Dhamsaniya²

¹Research Scholar, Department of Civil Engineering, Gujarat Technological University, Ahmedabad, Gujarat, India

²Principal and Professor, Department of Civil Engineering, Darshan University, Rajkot, Gujarat, India.

Received: 29 May 2024

Revised: 25 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Dipak Jivani

Research Scholar,

Department of Civil Engineering,

Gujarat Technological University,

Ahmedabad, Gujarat, India.

E.Mail: dkjivani@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Passive control systems for seismic mitigation, particularly friction pendulum bearings, have garnered significant attention in recent decades as a means to reduce earthquake demands and achieve desired performance objectives. While these bearings have proven effective in mitigating regular structures across a wide range of earthquakes, their performance and efficiency in structures with irregularities have received limited research attention. Additionally, there is a dearth of comprehensive parametric assessments examining the behavior of structures supported by friction pendulum bearings subjected to stochastic ground motions. Therefore, the purpose of this study is to evaluate the behavior of both flexible and rigid structures isolated with Triple Friction Pendulum (TFP) isolators under stochastic ground motion. The investigation is specifically assess the torsional performance of base-isolated buildings using TFP isolators with varying damping and effective time periods when subjected to stochastic ground motion. Preliminary findings indicate that the TFP bearing is effective in reducing base shear, absolute acceleration, and isolator displacement under stochastic ground motion. The superstructure flexibility is a crucial parameter for torsional response as higher torsional response has been observed in flexible superstructure than rigid superstructure. The results suggest that TFP bearings offer superior performance in mitigating seismic demands under stochastic ground motion and the torsional performance of a TFP-isolated structure improves as the isolation system's effective period increases.

Keywords: Seismic performance, Base isolation, Triple friction pendulum isolator, Torsional response, Stochastic ground motion, Single degree of freedom system.





Dipak Jivani and Dhamsaniya

INTRODUCTION

Strong earthquakes can cause severe damage to both structural and non-structural elements of buildings, particularly in irregular structures [1-2]. Irregularities may exceed seismic design code limits due to functional or design requirements [10-11]. Generally, integrating seismic control systems into structures becomes essential for safeguarding them against highly hazardous earthquakes. Introducing a base isolation system is recognized as one of the methods of earthquake protection for structures, aiming to minimize responses such as acceleration and displacement by increasing the structure's natural time period [9,15]. Numerous studies have highlighted the significant role of base isolation systems in reducing torsional response during severe earthquakes, offering flexibility and energy dissipation capacity [6-8, 16-17]. Soni et al, compared the torsional performances of double variable frequency pendulum isolated structures subjected to different ground motions [20]. Matsagar and Jangid [13-14], studied the torsional response of base isolated structure considering various isolators. A comparison of the response of the torsionally coupled base-isolated building is made with the corresponding response obtained from torsionally uncoupled base-isolated building.

In recent years, TFP bearings have gained widespread adoption as the predominant isolation system in many structures currently in operation. TFP bearings, characterized by multiple sliding surfaces with varying radii of curvature and friction, demonstrate adaptive behavior. This means that their stiffness and effective friction adjust to predictable values at manageable and controllable displacement levels. Consequently, TFP bearings can be individually optimized to address low-intensity, design-level, and maximum earthquake shaking scenarios. Fenz and Constantinou [5, 6] formulated the operational principles and force-displacement relationships of the TFP bearing from fundamental principles. They further detailed a method for modeling the TFP using an assembly of gap elements and single concave FP elements connected in series for response history analysis [5, 6].

In spite of the thorough work already done to understand the dynamic behavior of the TFP bearing, there are still important aspects of the behavior of structures isolated by the TFP bearing that need further investigation, especially the effect of stochastic ground motions. Hence, the response of stiff and flexible single story building isolated by the TFP Isolator is investigated under stochastic ground motions. The specific objectives of this research are (1) to study the performance of TFP isolated structure by considering super-structure eccentricity under stochastic ground motions, (2) to investigate the influence of important parameters, such as effective period and damping ratio, on the response of the TFP isolated structure. Six isolator configurations with respect to period of isolator and friction coefficient are considered. A set of 1000 stochastic ground motions are generated, having wide range variation in frequency and intensities are used.

TRIPLE FRICTION PENDULUM ISOLATOR MATHEMATICAL MODEL

Several mathematical methods have been developed by researchers to analyze and characterize Triple Friction Pendulum (TFP) isolators. Fenz and Constantinou [4] introduced a series model where three friction pendulum elements such as friction isolator elements, gap elements, and rigid elements are connected in series. Fenz and Constantinou [5] provided a comprehensive guideline for modeling TFP isolators and offered recommendations for accurate representation. It also proposed a bi-directional model to capture the movement of each isolator part. In the mentioned study, the series model by Fenz and Constantinou [4-5] is employed, and Figure 1(a) illustrates the arrangement of the friction pendulum elements within the TFP isolator system. Figure 1(b) depicts the series model.

The characteristics of three elements in each slider (R_{eff} , μ_i , d_i and a_i) should be chosen in accordance with Table 1 to get the tri-linear adaptive behavior of TFP Bearing.

As a result, the force exerted in the x- and y-directions by the i^{th} isolator element is given by,





Dipak Jivani and Dhamsaniya

$$\begin{Bmatrix} F_{ix} \\ F_{iy} \end{Bmatrix} = \begin{bmatrix} \frac{W}{R_{eff,i}} & 0 \\ 0 & \frac{W}{R_{eff,i}} \end{bmatrix} \begin{Bmatrix} u_{ix} \\ u_{iy} \end{Bmatrix} + \begin{bmatrix} \mu_i W_i & 0 \\ 0 & \mu_i W_i \end{bmatrix} \begin{Bmatrix} Z_{ix} \\ Z_{iy} \end{Bmatrix} + \begin{Bmatrix} F_{dix} \\ F_{diy} \end{Bmatrix}$$

u_{ix} and u_{iy} are the relative displacements of SFPB element i in the x- and y-direction, respectively, and W is the mass supported by each isolator. Here, μ_i is the i^{th} sliding surface's velocity-dependent coefficient of friction, which is given by,

$$\mu_i = \mu_{max,i} - (\mu_{max,i} - \mu_{min,i})e^{-a|x_i|}$$

where $\mu_{max,i}$ and $\mu_{min,i}$ are the coefficients of friction for sliding surfaces at maximum and minimum velocities, respectively and a_i is the rate parameter of i^{th} surface of the series model given by equations mentioned in Table 1 [4-5]. a_i is the rate parameter of i^{th} surface of the TFP bearing.

Z_{ix} and Z_{iy} denote two dimensionless variables of i^{th} element along the x- and y-directions, respectively. In the case of biaxial interaction they are governed by the following differential equations:

$$q_i \begin{Bmatrix} \dot{Z}_{ix} \\ \dot{Z}_{iy} \end{Bmatrix} = A \begin{bmatrix} 1 & 0 \\ 0 & 1 \end{bmatrix} \begin{Bmatrix} \dot{u}_{ix} \\ \dot{u}_{iy} \end{Bmatrix} - \begin{bmatrix} |Z_{ix}|^2 [\gamma \text{sign}(\dot{u}_{ix} Z_{ix}) + \beta] & Z_{ix} Z_{iy} [\gamma \text{sign}(\dot{u}_{iy} Z_{iy}) + \beta] \\ Z_{ix} Z_{iy} [\gamma \text{sign}(\dot{u}_{ix} Z_{ix}) + \beta] & |Z_{iy}|^2 [\gamma \text{sign}(\dot{u}_{iy} Z_{iy}) + \beta] \end{bmatrix} \begin{Bmatrix} Z_{ix} \\ Z_{iy} \end{Bmatrix}$$

The dimensionless numbers, β , γ , and A regulate the shape of the hysteresis response, and q is the displacement quantity that represents the yield displacement of the frictional force loop. The recommended values are $q = 0.25$ mm, $A = 1$, $\beta = 0.9$, $\gamma = 0.1$ [4-5].

According to the equation below, F_{dix} and F_{diy} are the forces produced in the gap element after it makes contact with the displacement restrainer.

$$F_{di} = K_G (|u_i| - d_i) \text{sgn}(u_i) H(|u_i| - d_i)$$

Where, K_G is the stiffness after gap closing, which should be assigned a large value, and H denotes the Heaviside function.

Here, a Runge-Kutta method based ODE functions (ode15s solver) that is introduced in the MATLAB programme are used to numerically resolve differential equations. Figures 2(a) and 2(b) compare the responses of single story structure introduced by Fenz and Constantinou [5] in order to test the aforementioned formulation and the subsequent outcomes.

PROPERTIES OF THE ISOLATION SYSTEM

In this study, TFP bearings is chosen by adjusting parameters such as the radius of the spherical surface and the coefficient of friction. The displacement capacities of all isolators remained constant. Six isolators with varying effective time periods and effective dampings are included in the current performance analysis by modifying isolator parameters. Table 2 provides the specifics of these TFP isolators.

The effective period, T_{eff} , and damping ratio, ξ_{eff} , are given by

$$T_{eff} = 2\pi \sqrt{\frac{W}{K_{eff} g}}$$





Dipak Jivani and Dhamsaniya

$$\xi_{eff} = \frac{E_{loop}}{2\pi K_{eff} D^2}$$

where E_{loop} is the total energy dissipated at each cycle of the isolator displacement, W is the total weight on the isolator, K_{eff} is the effective linear stiffness, and D is the maximum isolator displacement under the specified level of motion

During the design process of the TFP system, it is presumed that the maximum coefficient of friction for the isolation system has been attained and that none of the individual slider displacement capacities have been reached. The TFP effective stiffness, denoted as K_{eff} , is derived from the geometry of the force-deformation curve, while the energy dissipated in each cycle E_{loop} , is determined by calculating the area enclosed by the hysteresis loop as depicted in Figure 3.

$$K_{eff} = \frac{K_1 D_y + K_2(u^* - D_y) + K_3(u^{**} - u^*) + K_4(D - u^{**})}{D} W$$

$$E_{loop} = 4(u^*)^2(K_2 - K_3)W + 4DD_y(K_2 - K_1)W + 4u^*D(K_3 - K_2)W + 4u^{**}D(K_4 - K_3)W + 4(u^{**})^2(K_3 - K_4)W + 4D_y^2(K_1 - K_2)W$$

Where

$$K_1 = \frac{1}{R_{eff2}}; K_2 = \frac{1}{R_{eff2} + R_{eff3}}; K_3 = \frac{1}{R_{eff3} + R_{eff1}}; K_4 = \frac{1}{R_{eff1} + R_{eff4}};$$

$$u^* = (\mu_1 - \mu_2)R_{eff2} + (\mu_1 - \mu_3)R_{eff3}$$

$$u^{**} = u^* + (\mu_4 - \mu_1)(R_{eff1} + R_{eff3})$$

NUMERICAL STUDY AND RESULTS

The current work uses a Triple Friction Pendulum Isolator (TFP) to investigate the multi-stage performance of seismically separated buildings under stochastic ground motion. In order to accomplish this, Rezaeian and Kiureghian's [18-19] stochastic ground motion model is used to develop a number of stochastic ground motions. This model accounts for the spectral and temporal non-stationarities of the motion. By altering the intensity and changing the filter's characteristics over time, non-stationarity is produced [3]. A total of 1000 stochastic ground motions are created and used for additional research. According to research [12], 1000 simulations are sufficient to accurately forecast the torsion behaviour of an isolated building. Table 3 displays the statistics of generated ground motions. These records includes a variety of intensities and frequency content.

To examine the seismic performance of TFP isolated structures, analytical simulations were conducted with varying friction coefficients and pendulum radii. Nonlinear dynamic analysis is employed to evaluate the performance of TFP isolated bearings. Parametric research is conducted using single-story buildings measuring 10m x 10m. As illustrated in Figure 4, the base-isolated building is simplified for this study as a single-story structure with masses concentrated at the upper deck and the base raft. The building's corners are supported by massless, axially inextensible columns that are fixed to a rigid foundation raft to support the upper deck. The stiff and flexible superstructures are considered, with time periods of 0.25 seconds and 1 second, respectively. A damping ratio (ξ_s) of 0.02 is applied. Asymmetry at the superstructure level is considered, with a ratio ($\frac{e_s}{d}$) of 0.3 throughout the study. The uncoupled torsional to lateral frequency ratio, a critical variable in a structure's asymmetric behavior, is kept constant at $\Omega_s = \Omega_b = 1$.

This study considers the following demand characteristics to better understand the response of the TFP isolated structure: lateral displacement at the base level, deck corner displacement, base rotation, normalized base torsion, normalized base shear, and deck corner displacement magnification. Deck corner displacement magnification is defined as the ratio of peak deck corner displacement to peak deck lateral displacement.



**Dipak Jivani and Dhamsaniya****Influence of Effective period of Base isolation**

The characteristics of the TFP bearing are modified by changes in the coefficient of friction and the radius of curvature of the sliding surfaces. These changes in the radius of curvature affect the isolator's time period, which consequently influences its response. To investigate the impact of the isolator's effective time period, the parameters are selected to create variations of 3 seconds, 4 seconds, and 5 seconds effective time period of isolator. For each of these time periods, the effective damping of the isolator is consistently maintained at 15%.

Figure 5 illustrates that both base displacement and deck corner displacement rise as the isolation system's effective period increases. This increase in seismic response is due to a reduction in effective stiffness, which accompanies a longer effective period. For a rigid superstructure, the increases in base displacement and deck corner displacement are 81% and 82%, respectively, while for a flexible superstructure, the increases are 77% and 67%. It is evident that changes in the effective time period have a greater impact on lateral displacement than on rotational displacement. Assuming that the translational period of an isolated system increases, the torsional period also increases, resulting in decreased base rotation as the effective period of the isolation system grows.

As expected, a structure's base shear decreases as its effective period increases. The reduction in normalized base shear is 41% for a rigid superstructure and 11% for a flexible superstructure, indicating that the impact on base shear reduction is less significant for a flexible superstructure compared to a rigid one. A similar reduction trend with increasing effective time period is observed for normalized base torsion. Figure 5 shows that the torsional performance of a TFP-isolated structure improves as the isolation system's effective period increases. The comparison reveals that the flexible superstructure is more prone to torsional response than the stiff superstructure.

Influence of Effective damping of Base isolation

The coefficient of friction significantly impacts the response by contributing to energy dissipation through Coulomb damping. To assess the effect of the isolator's effective damping, parameters are selected to vary the damping by 10%, 15%, and 20%. The isolator's effective time period is kept constant at 3 seconds for all damping levels. The Figure 6 shows that when effective damping increases, normalized base shear and isolator displacement decrease. The increased energy dissipation from the isolator's hysteresis behavior is what causes the reduction in isolator displacement with higher damping. The decrease in base displacement is about 20% when damping increases from $\xi_{\text{eff}} = 10\%$ to 15%, but this difference narrows to 3% when damping increases from $\xi_{\text{eff}} = 15\%$ to 20%. A similar trend is observed in deck corner displacement in relation to the isolator's effective damping.

When friction on the outer sliding surfaces increases, the TFP isolator helps reduce base shear and absolute acceleration. This occurs because the inner sliding surfaces, with their relatively low friction and stiffness, transmit less force to the superstructure. An increase in effective damping from 10% to 20% leads to a reduction in base shear of up to 38% for a rigid superstructure, but this effect is negligible for a flexible superstructure. Furthermore, torsional responses are more noticeable to flexible superstructures than to rigid ones. The reduction in normalized base torsion is 41% for rigid superstructures and 23% for flexible ones. It is also noticeable that the effectiveness of the TFP diminishes when the effective damping increases from 15% to 20%.

CONCLUSIONS

The nonlinear response of a torsionally coupled system on a triple friction pendulum isolator is analyzed under stochastic ground motions. This research investigates how various significant isolator parameters affect the response of such a system. The numerical trends observed in the response of the torsionally coupled base-isolated building lead to the following conclusions.

1. The torsional response of TFP Isolated building is amplified by the increased flexibility of Superstructure, whilst the lateral base response and base shear is decreased. Hence, influence of flexibility of superstructure is more on torsional response of TFP Isolated building.





Dipak Jivani and Dhamsaniya

2. The torsional performance of a TFP-isolated structure improves as the isolation system's effective period increases.
3. The decrease in base displacement is about 20% when damping increases from $\xi_{\text{eff}} = 10\%$ to 15%, but this difference narrows to 3% when damping increases from $\xi_{\text{eff}} = 15\%$ to 20%. A similar trend is observed in deck corner displacement in relation to the isolator's effective damping. It is concluded that the effectiveness of the damping of TFP isolator diminishes when the effective damping increases from 15% to 20%.
4. In stiff eccentric systems, the base shear remains essentially constant for all values of effective damping and time period of isolator. While substantial effect of effective damping and time period is observed on base shear for flexible superstructure.
5. The rigid super structure assumption over-estimated the superstructure acceleration (Normalised base shear) of TFP Isolated structure.

REFERENCES

1. Ahmadi, G. (1983). "Stochastic earthquake response of structure on sliding foundation", *International Journal of Engineering and Science*, 21(2), 93–102.
2. Buckle, I.G., & Mayes, R.L. (1990). "Seismic isolation: History, application and performance-a world overview", *Earthquake Spectra*, 6(2), 161-201.
3. Dabaghi, M., & Der Kiureghian, A. (2016). "Stochastic model for simulation of near-fault ground motions". *Earthquake Engineering & Structural Dynamics*, 10(2).
4. Fallahian, M., Khoshnoudian, M., & Loghman, V. (2015). "Torsionally Seismic Behavior of Triple Concave Friction Pendulum Bearing", *Advances in Structural Engineering*, 18(12), 2151-2166.
5. Fenz, D.M., & Constantinou, M.C. (2008a). "Modeling triple friction pendulum bearings for response-history analysis", *Earthquake Spectra*, 24(4), 1011–1028.
6. Fenz, D.M., & Constantinou, M.C. (2008b). "Spherical sliding isolation bearings with adaptive behavior: Theory", *Earthquake Engineering & Structural Dynamics*, 37(2), 163–183.
7. Jangid, R.S. (1996). "Seismic response of an asymmetric base-isolated structure", *Computers and Structures*, 60(2), 261–267.
8. Jangid, R.S., & Datta, T.K. (1994a). "Non-linear response of torsionally coupled base-isolated structure", *Journal of Structural Engineering, ASCE*, 120(1), 1–22.
9. Jangid, R.S., & Datta, T.K. (1994b). "Seismic response of torsionally coupled structure with elasto-plastic base isolation", *Engineering Structures*, 16(4), 256–262.
10. Jangid, R.S., & Datta, T.K. (1995a). "Seismic behaviour of base isolated buildings: a state-of-the-art review", *Structures and Buildings*, 110(2), 186–203.
11. Jangid, R.S., & Datta, T.K. (1995b). "Performance of base isolation systems for asymmetric building subject to random excitation", *Engineering Structures*, 17(6), 443–454.
12. Jangid, R.S., & Datta, T.K. (1995c). "The stochastic response of asymmetric base-isolated buildings", *Journal of Sound and Vibration*, 179(3), 463–473.
13. Jivani, D.K., & Dhamsaniya, R.G. (2024). "Investigating the Seismic Response of Coupled Isolated Buildings under Stochastic Ground Motions", *SSRG International Journal of Civil Engineering*, 11(2), 93-107.
14. Matsagar, V.A., & Jangid, R.S. (2005). "Base-isolated building with asymmetries due to the isolator Parameters". *Advanced Structural Eng*, 6, 603–622
15. Matsagar, V.A., & Jangid, R.S. (2010). "Impact response of torsionally coupled base isolated structure". *J. Vibration Control*. 16(11), 1623–1649
16. Naeim, F., & Kelly, J.M. (1999). "Design of Seismic Isolated Structures: From Theory to Practice", *Wiley*, New York.
17. Nagarajaiah, S., & Sun, X. (2001). "Base-isolated FCC building: Impact response in Northridge Earthquake", *Journal of Structural Engineering, ASCE*, 127(9), 1063–1075.
18. Nagarajalah, S., Reinhorn, A.M., & Constantinou, M.C. (1993b). "Torsion in base isolated structures with elastomeric isolation systems", *J. Structural Eng., ASCE*, 119(10), 2932-2951.





Dipak Jivani and Dhamsaniya

19. Rezaeian, S., &Der Kiureghian, A. (2010). "Simulation of synthetic ground motions for specified earthquake and site characteristics". *Earthquake Engineering & Structural Dynamics*, 39(10), 1155–1180.

20. Rezaeian S., &Der Kiureghian, A. (2008). "Stochastic ground motion model with separable temporal and spectral nonstationarities". *Earthquake Engineering & Structural Dynamics*, 37(13), 1565–1584.

21. Rezaeian S., &Der Kiureghian, A. (2012). "Simulation of orthogonal horizontal ground motion components for specified earthquake and site characteristics". *Earthquake Engineering & Structural Dynamics*, 41(2), 335–353.

22. Soni, D.P., Mistry, B.B.,& Panchal, V.R. (2010). "Behaviour of asymmetric building with double variable frequency pendulum isolator", *Structural Engineering and Mechanics*, 11(1), 61–84.

Table 1 The variables of series model in terms of TFP bearings variables in a fully adaptive arrangement [6]

	Effective Radius of Curvature	coefficient of friction	Displacement capacity	RateParameter
Element 1	$R_{eff,1} = \bar{R}_{eff,2} + \bar{R}_{eff,3}$	$\mu_1 = \bar{\mu}_2 = \bar{\mu}_3$	$d_1 = (\bar{d}_1 + \bar{d}_2 + \bar{d}_3 + \bar{d}_4) - (d_2 + d_3)$	$a_1 = \frac{1}{2} \frac{\bar{a}_2 + \bar{a}_3}{2}$
Element 2	$R_{eff,2} = \bar{R}_{eff,1} - \bar{R}_{eff,2}$	$\mu_2 = \bar{\mu}_1$	$d_2 = \frac{\bar{R}_{eff,1} - \bar{R}_{eff,2}}{\bar{R}_{eff,1}} \bar{d}_1$	$a_2 = \frac{\bar{R}_{eff,1}}{\bar{R}_{eff,1} - \bar{R}_{eff,2}} \bar{a}_1$
Element 3	$R_{eff,3} = \bar{R}_{eff,4} - \bar{R}_{eff,3}$	$\mu_3 = \bar{\mu}_4$	$d_3 = \frac{\bar{R}_{eff,4} - \bar{R}_{eff,3}}{\bar{R}_{eff,4}} \bar{d}_4$	$a_3 = \frac{\bar{R}_{eff,4}}{\bar{R}_{eff,4} - \bar{R}_{eff,3}} \bar{a}_4$

Table 2 List of TFP bearings and their properties

Group	Type	T_{eff}	ξ_{eff}	Displacement capacity-d (m)	Effective radii- R_{eff} (m)		Friction coefficient- μ		
					$R_{eff1}=R_{eff4}$	$R_{eff2}=R_{eff3}$	$\mu_2=\mu_3$	μ_1	μ_4
1	TCFP-3-15	3	15	1	1.65	0.40	0.05	0.115	0.2
	TCFP-4-15	4	15	1	3	0.40	0.02	0.05	0.1
	TCFP-5-15	5	15	1	4.5	0.45	0.02	0.038	0.07
2	TCFP-3-10	3	10	1	1.5	0.40	0.01	0.05	0.1
	TCFP-3-15	3	15	1	1.65	0.40	0.05	0.115	0.2
	TCFP-3-20	3	20	1	2	0.40	0.07	0.17	0.21

Table 3 Statistics of generated 1000 earthquake ground motions

Earthquake	Maximum	Minimum	Mean	Standard deviation
PGA (g)	1.5965	0.0717	0.3344	0.221
Duration (sec)	115.28	2.5	30.477	17.169
Frequency *(Hz)	106.78	0.03	30.29	18.52

* Frequency content corresponding to peak FFT amplitude of the acceleration





Dipak Jivani and Dhamsaniya

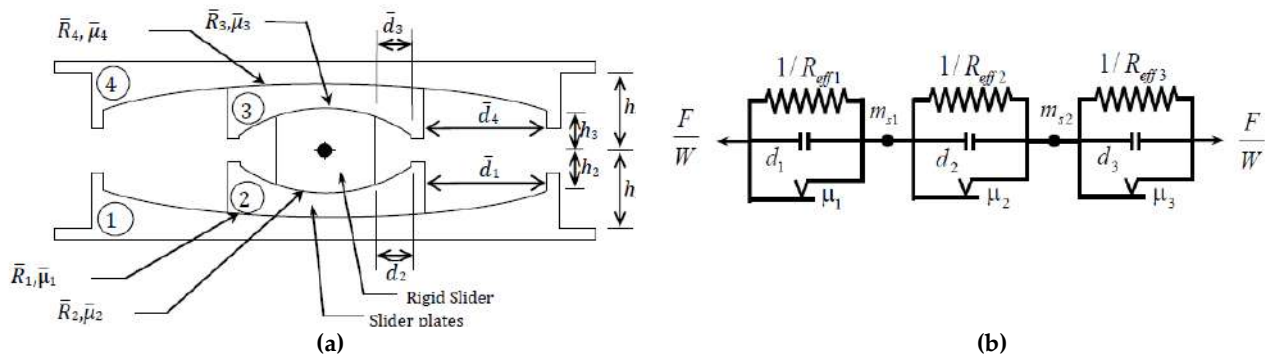


Figure 1 Triple friction bearing (a) Schematic diagram [5] (b) Series model

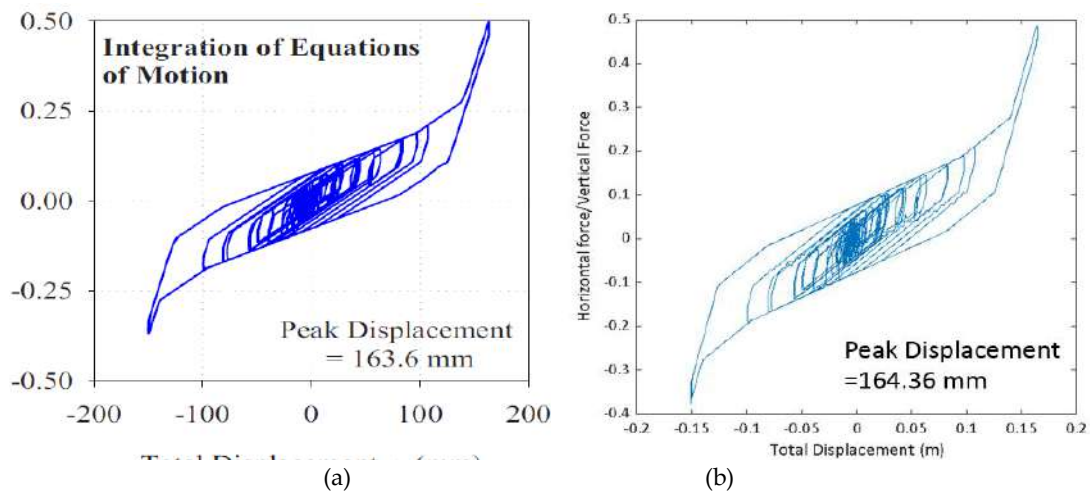


Figure 2 Comparison of TFP bearing force-displacement from (a) results from Fenz and Constantinou [5] and (b) our results



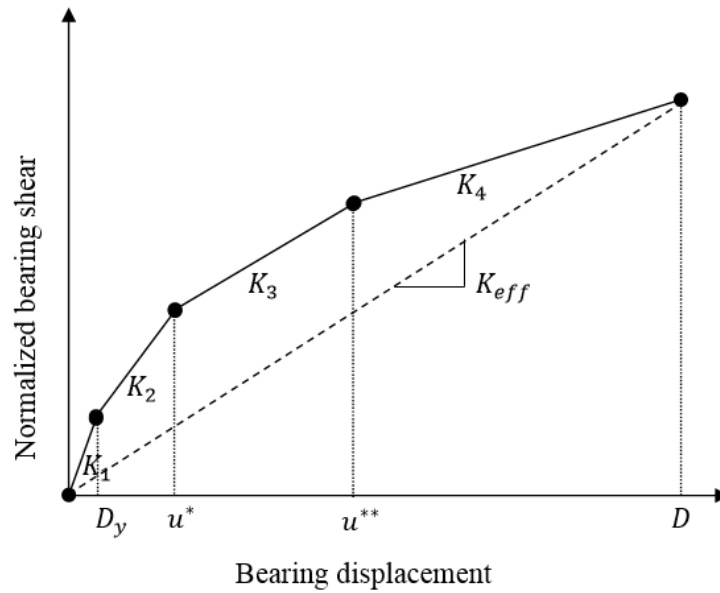


Figure 3 Force-displacement loops of TFP Bearing

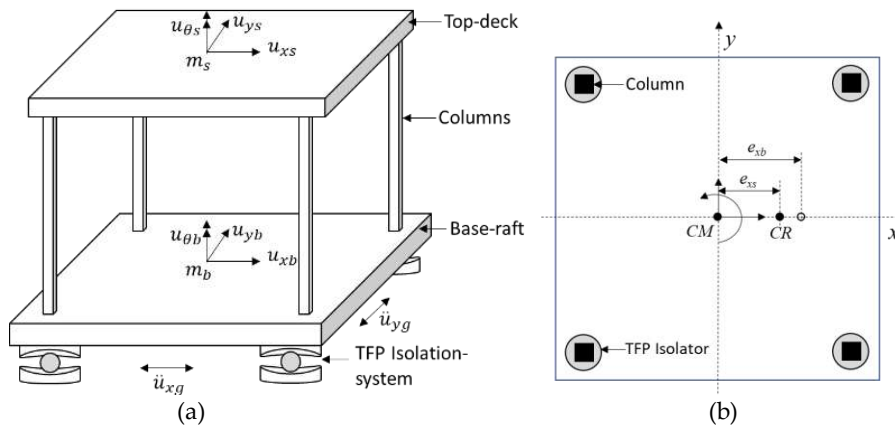


Figure 4 (a) 3D base isolated building model (b) various eccentricities in an asymmetric base-isolated building





Dipak Jivani and Dhamsaniya

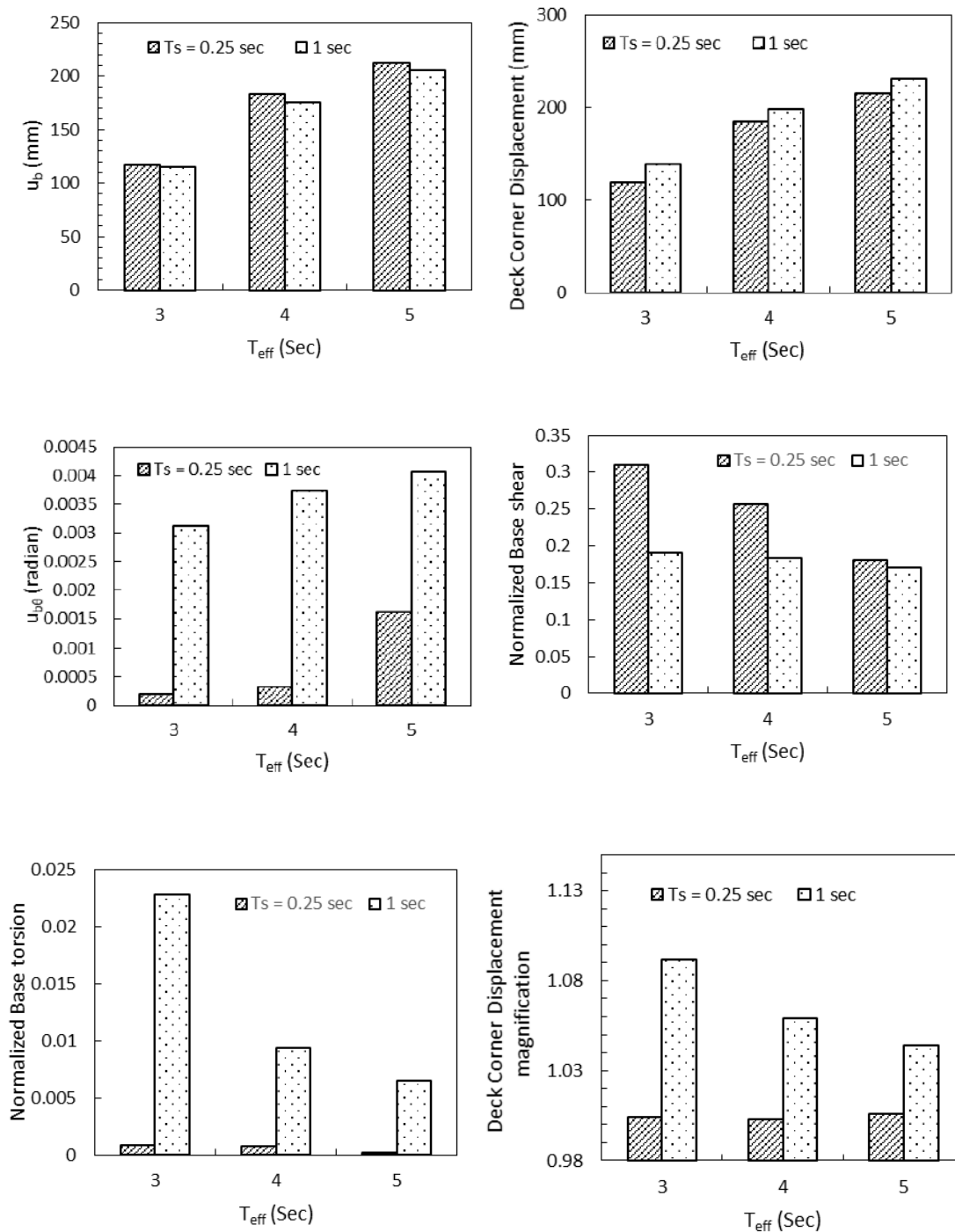


Figure 5 Influence of effective period of isolation system for $(\frac{e_s}{d}) = 0.3$





Dipak Jivani and Dhamsaniya

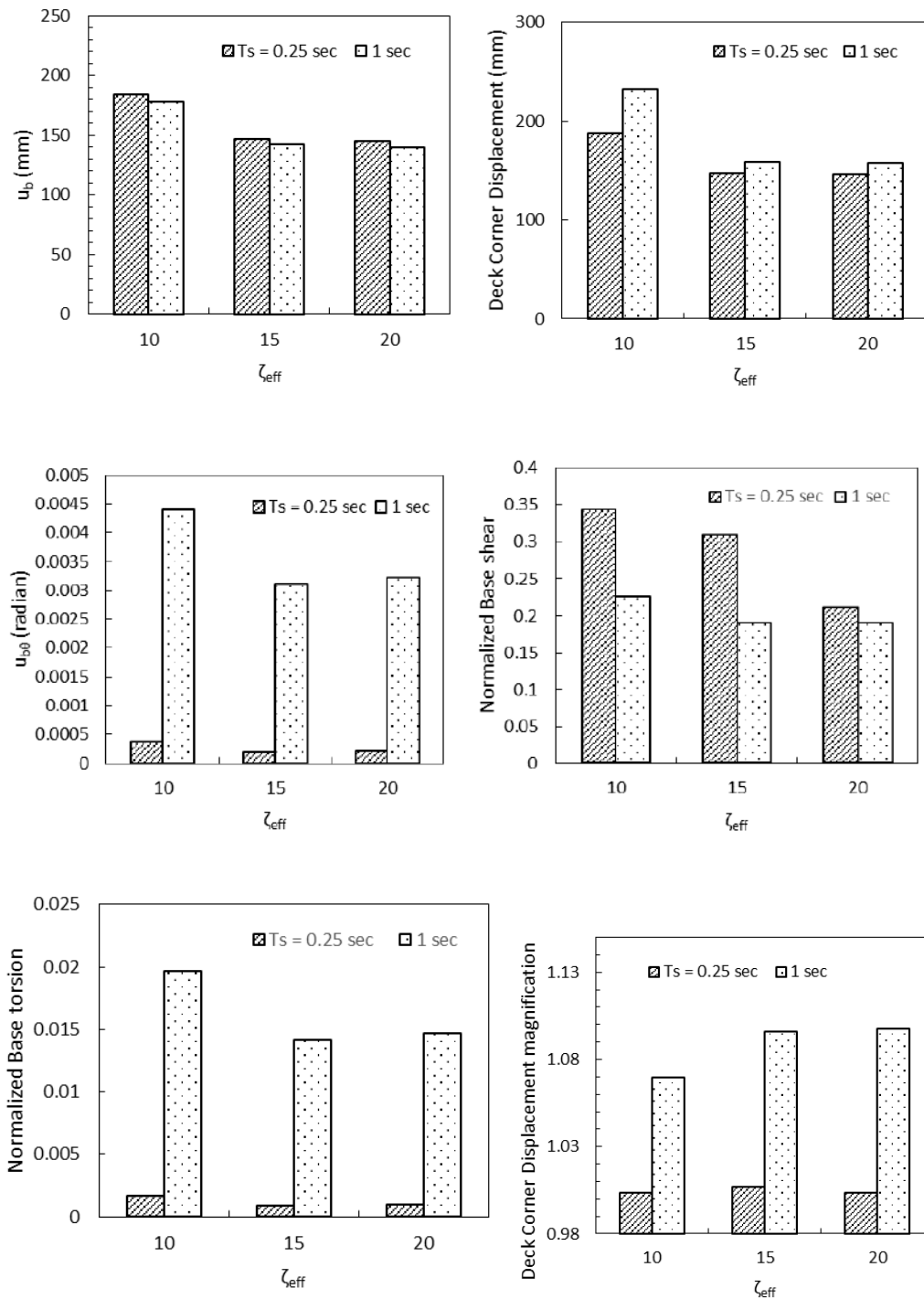


Figure 6. Influence of effective damping of isolation system for $(\frac{e_s}{d}) = 0.3$





Prevalence of Post Traumatic Stress Disorder among Road Traffic Accident Victims : A Systematic Review

Hetal Jitendrabhai Shah^{1*}, Sapna Bhavin Patel¹ and Sandip Patel²

¹Assistant Professor, Department of Nursing, Manikaka Topawala Institute of Nursing, Charotar University of Science and Technology (CHARUSAT), Anand, Gujarat, India.

²Psychiatrist, Manan Hospital, Anand, Gujarat, India.

Received: 18 Jun 2024

Revised: 20 Aug 2024

Accepted: 18 Oct 2024

*Address for Correspondence

Hetal Jitendrabhai Shah

Assistant Professor, Department of Nursing,
Manikaka Topawala Institute of Nursing,
Charotar University of Science and Technology (CHARUSAT),
Anand, Gujarat, India.
E.Mail: hetalshah.nur@charusat.ac.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The complex somatic, cognitive, affective, and behavioural effects of psychological trauma" is what posttraumatic stress disorder (PTSD) means, and it causes significant interpersonal, social, and professional dysfunction. RTAs, or road traffic accidents, are one of the biggest global issues. Traumatic injuries are very serious and leads in lifelong physical and psychological consequences. The focus on physical injuries often overshadows the lasting psychological impacts, which remain significant and intertwined with physical outcomes, necessitating comprehensive care approaches. One of the most dangerous outcomes of RTA is Post traumatic stress disorder (PTSD) as it can cause other mental diseases, this paper shows systematic review of previous studies conducted on the same subject, it gives summery of copious cross sectional along with case control study. Researcher used PRIZMA guidelines along with set inclusion and exclusion criteria to identify the research studies. Following search engines were used to retrieve the published articles on above said topic: Google scholar, PubMed, CINHALL, ProQuestand Scopus database. Research studies were analyzed and checked through NOS scale for cross sectional and case control studies for the quality assessments. The studies were taken if the score comes at medium or above. Following search strings were used "#1(PrevalenceOR Magnitude OR Epidemiology OR Incidence AND #2(PTSD OR Post-traumatic stress disorder OR Post-traumatic stress symptoms OR Stress disorder) AND #3 (Road Traffic Accident survivors OR Road traffic accident victimsOR motor vehicle accident OR motor vehicle accidents') and #4(Cross sectional, OR Case control studies)". Researcher found 1447 Studies out of which 10 were selected based on the search protocol. Prevalence Variation: There is a significant variation in PTSD prevalence, ranging from 6.75% to 40.06%, influenced by country, assessment time, and specific sample characteristics. *Gender Impact:* PTSD is generally more prevalent among females across multiple studies. *Assessment Tools Consistency:* PCL-5 is the most used tool, ensuring some consistency in measurement across different settings. *Influence of Associated Factors:* A broad range of factors including personal history, accident severity, and socioeconomic impacts play crucial roles in PTSD development post-accident.





Hetal Jitendrabhai Shah *et al.*,

This systematic review has examined the prevalence of post-traumatic stress disorder symptoms among RTA victims. These findings underline the complexity and variability of PTSD prevalence among RTA survivors, highlighting the need for targeted interventions considering the specific associated factors identified in each study.

Key words: stress disorder, post-traumatic stress symptoms, RTAs, incidence

INTRODUCTION

Annually, approximately 1.9 million people are involved in road traffic accidents. In low and middle-income countries, these incidents account for over 90% of traffic-related fatalities. In India, road transport remains the most affordable and convenient mode of travel [1]. The worst road traffic exposure stems from motorization and urbanization, making accidents and injuries a routine part of life; globally, road traffic injuries are a leading cause of death among those aged 15 to 49 years. With the increase in vehicle ownership and expanding urban areas, the frequency of such incidents continues to rise. Efforts to improve road safety and infrastructure are crucial to mitigating these risks and saving lives [2]. In 2021, there were around 1.5 lakh fatalities and 3.8 lakh injuries due to road accidents, reflecting an increase compared to the figures recorded in 2020. Out of every 100 road accidents, 37 result in fatalities(1). Victims of road traffic accidents are typically treated in emergency, orthopedic, or trauma departments for physical injuries. However, psychological issues, which may not be immediately apparent, receive the least attention. Posttraumatic stress disorder (PTSD) involves a complex range of physical, cognitive, emotional, and behavioural effects resulting from psychological trauma. These impacts infiltrate multiple aspects of a person's life, such as personal relationships, social interactions, and professional activities. PTSD leads to substantial dysfunction in these areas, disrupting daily life and impairing overall well-being(3,4). Its impact reaches beyond the individual, influencing families, communities, and broader societal structures. Therefore, addressing PTSD necessitates comprehensive strategies that recognize its intricate nature and the wide array of challenges it poses. Numerous studies have identified varying prevalence rates of PTSD among victims of road traffic accidents (RTAs), with California notably reporting a high prevalence of 41%(4–6).

PTSD stands out as a particularly precarious outcome due to its capacity to precipitate additional mental health disorders(7). This paper presents a systematic review of previous studies focused on this subject, offering a summary of numerous cross-sectional and interventional investigations. By synthesizing existing research, it aims to illuminate the prevalence and determinants of PTSD among RTA victims(8). Through a rigorous analysis of diverse study designs and methodologies, it provides insights into the multifaceted nature of PTSD following RTAs. The findings contribute to a deeper understanding of the psychological impact of RTAs and emphasize the urgent need for effective interventions to address the mental health challenges faced by survivors. Such efforts are crucial for enhancing the overall well-being and quality of life of individuals affected by traumatic accidents.

Rationale for the study

Literature that could validate the above statement along with prevalence of post-traumatic stress disorder. Many researchers investigated the prevalence of PTSD among different population but there are only few on road traffic accident victims, which indicates research gap in precise investigation on prevalence of PTSD among road traffic accident victims. The collected evidence from literature review showed that there is need for evidence-based intervention to treat PTSD, the researcher wants to conduct a study on effectiveness of mental health first aid kit on PTSD therefore, there is a requirement to know the magnitude and trend of prevalence so that it helps to plan intervention accordingly.



**Hetal Jitendrabhai Shah et al.,****Research question, Aim and objective**

Research question is what is the prevalence of post-traumatic stress disorder among road traffic accident victims? The primary aim of the research is to conduct a systematic review to identify the prevalence of PTSD among road traffic accident victims. Objectives of the study was to understand the characteristics of the study participants, instrument used used to assess the PTSD, time when PTSD is assessed and to identify the prevalence rate of PTSD.

MATERIALS AND METHODS

This systematic review was conducted adhering to PRISMA(9).We conducted a comprehensive systematic review published up to 2024 to identify the prevalence of PTSD among road traffic accident victims.

Study Selection

The inclusion criteria were oriented along with the PICOS elements. Regarding the population (P), we included study conducted in any country that involves road traffic accident victims. We do not exclude any age group and we also had no restriction on gender or type of accident. In terms of intervention or exposure (I) we included victims exposed to any kind of road traffic accident, we do not exclude victims based on vehicle or body parts injured. In terms of comparison (C) we didn't compare any variables in the study. In terms of outcome (O), Eligible studies had to include outcome related to prevalence rate of PTSD. Regarding study design (S), we included cross sectional and case control study studies (Cross sectional and cross-sectional survey), we also included studies having prevalence of PTSD along with assessment of any other mental health issues. The reason for including cross sectional studies is they are best way to determine the prevalence and can study the association of multiple exposure and outcomes.

Inclusion and exclusion criteria:**Studies were included if they met the following criteria:**

The articles of peer-reviewed journal as they have validated knowledge and result, published in the English language, published from the last ten years to utilise current and reliable result, based on the cross-sectional study to gather relevant evidence, articles focusing on prevalence of PTSD reference to RTA and studies on victims like pedestrian, driver, passenger, cyclist, bikers and event witnesses.

The exclusion criteria are: The articles with half text, not fully accessible, articles in a foreign language, articles older than 10 years, qualitative studies, conference papers, posters and dissertations are excluded due to quality concern and studies on PTSD other than road traffic accident victims.

Data sources and searches

The search was undertaken in the high scholarly electronic databases like, Google scholar, PubMed, CINHALS, Scopus and ProQuest for relevant peer-reviewed scientific articles. Boolean operators such as AND, OR and search limit was used in the search strategy. The reason for using multiple databases is to have a high quality, peer reviewed literature, to increase the reliability and validity of the review and to eliminate the reporting bias and showing credibility. A broad search string was used that included a combination of appropriate keywords such as:

Database: Google Scholar, PubMed, CINAHL, Scopus, and ProQuest are essential databases used to search for scholarly studies

Search Strings: #1(Prevalence OR Magnitude OR Epidemiology OR Incidence AND #2(PTSD OR Post-traumatic stress disorder OR Post-traumatic stress symptoms OR Stress disorder) AND #3 (Road Traffic Accident survivors OR Road traffic accident victims OR motor vehicle accident OR motor vehicle accidents') and #4(Cross sectional, OR Case control studies)' used for advance search in database.



**Hetal Jitendrabhai Shah et al.,**

Quality assessment: The Newcastle-Ottawa scale (NOS) instrument was used to assess the quality of studies. In cross section studies we use an adaptation to specific criteria for sampling, non-response rate and statistical analysis employed. The NOS instrument includes items divided in three areas, selection, comparability and outcome. Maximum score is 9, 4 points for selection, 2 points for comparability and 3 points for outcomes. (See table no.1).

Data collection

The researcher reviewed the articles' titles and abstracts using the pre decided keywords to determine their appropriateness for this systematic review. Eligible articles were evaluated and scored using the Newcastle-Ottawa scale (NOS). If an article scored >7, 8 or 9 on the NOS scale and met all other inclusion criteria, it was included in the review.

Data extraction

From each of the study, information based on different study parameters was extracted that help in retrieving the reliable result (see table no. 2). The parameters included study characteristic like author name and year of publication, country, study design, study participants, sample size, instrument used to assess PTSD, Time when PTSD is assessed and prevalence rate with associated factors.

RESULT

Study selection:The initial database search yielded a total of 1447 records from various sources, including PubMed (n=481), Google Scholar (n=194), CINHAL (n=3), ProQuest (399) and Scopus (n=370). After removing duplicates, 1028 unique records remained for further analysis. From these, 913 records were excluded after abstract review, 115 full-text articles were assessed for eligibility, leading to the inclusion of 10 articles for the systematic review. This rigorous process ensured a comprehensive and methodical approach to identifying relevant literature for the study. The table no 2 provides a comparative overview of various studies examining the prevalence of PTSD among survivors of road traffic accidents (RTA) across different countries, study designs, participant types, sample sizes, assessment tools, time frames, and associated factors.

Country and Study Design: The studies span a range of countries, including Ethiopia, Benin, India, Iran, the USA, and Israel. Most studies employ a cross-sectional design, except for Yimer et al. (2023), which uses a case-control approach.

Participants and Sample Size: Participants across studies vary, including survivors of RTAs, drivers, pedestrians, motorcyclists, and passengers. Sample sizes range from 250 (17) to 865 (11). **Age and Gender:** *Median/Mean Age:* Ages vary, with median or mean ages typically in the 30s. For instance, Yimer et al. (2023)(10) report a median age of 38, whereas Donatien Daddahet al. (2022)(11) report a mean age of 38.04. *Gender Distribution:* Gender distribution varies, with some studies reporting higher male participation (e.g., Yohannes et al., 2018: 63.6% male)(16), while others have a higher female prevalence in PTSD rates (e.g., Yimer et al., 2023: 58.52% female among cases)(10). **Instruments Used:** The studies predominantly use the PTSD Checklist for DSM-5 (PCL-5) for assessment, though some utilize the DSM-5, Trauma Screening Questionnaire (TSQ), and Clinician-Administered PTSD Scale (CAPS). *Time of PTSD Assessment:* The timing of PTSD assessment post-accident varies: Short-term(1 month to 3 months): Most studies assess PTSD within this period(10,12,16). Long-term (up to 12 months): Donatien Daddahet al. (2022)(11) and Arora et al. (2017) (17) extend the assessment to 12 months post-accident.

Prevalence Rates:*Highest Rates:* Hamid Soori et al. (2021)(13) report the highest prevalence at 40.06%. *Lowest Rates:* R. Naim et al. (2014) (19) and Hruska et al. (2014)(18) report the lowest prevalence at 6.75% and 10%, respectively.

Associated Factors: Common factors associated with increased PTSD prevalence include *Gender:* Higher prevalence in females(10,11). *Personal and Psychological Factors:* History of psychiatric issues, depression, and presence of



**Hetal Jitendrabhai Shah et al.,**

comorbidities(10,16). *Accident-related Factors*: Presence of fractures, witnessing death, hospitalization, and severe risk behaviors (13,15). *Social and Economic Factors*: Impact on income, social support, and return to work (11,16).

DISCUSSION

A total of 10 studies, including 4,272 samples, were analysed to assess the prevalence of PTSD among road traffic accident victims. The primary objective of the research was to evaluate the prevalence of PTSD in this group. The studies collectively aimed to provide a comprehensive understanding of PTSD prevalence among these victims. Individuals who suffer from traumatic injuries often face a diminished quality of life and significant mental health challenges. They tend to have high rates of recurring injuries, dissatisfaction with pain management, and low levels of engagement in post-injury care(20).

Prevalence of the PTSD among RTA victims

PTSD commonly observed in RTA victims. Current study, Prevalence rates range from 40.06% at the highest to 6.75% and 10% at the lowest reported levels. The differences in prevalence may stem from geographical factors and the lack of psychological care provided in emergency departments. This study contributes to the current data by emphasizing the essential need for psychological care among motor vehicle accident (MVA) survivors attending the orthopaedic and trauma clinic at Kenyatta National Hospital, Nairobi(21). Furthermore, the prevalence of PTSD is influenced by various factors, with age and experiences of significant loss playing crucial roles. Older individuals, who may have experienced more traumatic events over their lifetimes, are more susceptible to developing PTSD, especially following the death of a close family member or friend. This finding supports the article, showing a 46.5% incidence of PTSD among RTA survivors, linked to witnessing death during the accident, severe family relationship impacts, and previous psychiatric illness(22). These factors underscore the multifaceted nature of PTSD development in trauma survivors. These experiences can amplify feelings of grief and trigger PTSD symptoms. Conversely, gender and role in motor vehicle accidents (MVA) can contribute to lower PTSD prevalence rates. Females often report higher levels of PTSD due to greater susceptibility to the emotional impact of trauma, whereas individuals in less directly involved roles in MVAs, such as witnesses or bystanders, may experience lower levels of PTSD due to reduced proximity to the traumatic event and potentially less severe emotional impact. The results align with earlier research: men tend to experience more traumatic events, while women exhibit more severe psychiatric disorders following exposure(23). This finding is supported by the gender-specific role of peritraumatic dissociation study, which corroborates the gender differences in trauma exposure and its psychological impacts. These factors highlight the complex interplay between personal circumstances and PTSD prevalence.

Advancing PTSD Research among Drivers and RTA Survivors

Future research on PTSD among drivers and road traffic accident (RTA) survivors can significantly benefit from the strengths identified in recent studies. Utilizing standardized instruments like the PTSD Checklist for DSM-V (PCL-5) ensures consistent and reliable measurement of PTSD symptoms, facilitating comparability across studies. Adopting well-defined catchment areas and sampling from multiple hospitals enhances the representativeness of samples, crucial for generalizing findings to broader populations. Moreover, employing large and diverse samples improves statistical power and allows for subgroup analyses, identifying nuanced risk factors and protective factors. The involvement of trained health professionals and supervision by medical epidemiologists ensures rigorous data collection and adherence to high research standards. By leveraging these strengths—standardized assessment tools, representative sampling strategies, large sample sizes, and expert involvement—future research can deepen our understanding of PTSD dynamics in this population, informing more targeted interventions and support initiatives.

Strength and weakness of reviewed articles

All the studies reviewed share several common strengths and weaknesses, highlighting both their robust methodologies and limitations. First, the strength of using standardized instruments for measuring PTSD, such as the PTSD checklist for DSM-V (PCL-5)/PTSD checklist-Specific version (PCL-S) or similar tools, is a notable consistency



**Hetal Jitendrabhai Shah et al.,**

across the studies(10,11,14–17). These standardized tools ensure a reliable assessment of PTSD symptoms. Moreover, several studies benefitted from sampling participants from well-defined catchment areas or multiple hospitals, which helps in obtaining a more representative sample of the target population(11,14–16). However, these studies also share common limitations. The cross-sectional design prevalent in all studies(10,11,14–17). Limits the ability to establish causality between PTSD and various factors. The focus on drivers or subjects with recent road traffic accidents (RTAs) makes it challenging to generalize findings to those without RTAs or with different trauma histories. Additionally, the non-random sampling techniques and smaller sample sizes employed in some studies(10,14,17). further limit the generalizability of the results. Lastly, potential recall and social desirability biases may have influenced the participants' responses, affecting the reliability of the findings(11,17).

Limitations:

The main limitation of this study included the database from Google scholar, PubMed, ProQuest, CINHALL and Scopus database. Authors were limited access to paid articles, incomplete text and studies in other languages. Type of studies were limited to the purpose of the systematic review.

CONCLUSION

This systematic review has examined the prevalence of post-traumatic stress disorder symptoms among RTA victims. Understanding the multifaceted dynamics influencing PTSD among RTA survivors underscores the need for comprehensive support systems and tailored interventions. Strengthening social networks, promoting mental health awareness, and addressing gender-specific needs can enhance resilience and mitigate the long-term impact of trauma on survivors' well-being(1,24,25).

REFERENCES

1. BLANCHARD EB, HICKLING EJ, TAYLOR ANNE, LOOS W. Psychiatric Morbidity Associated with Motor Vehicle Accidents. *J Nerv Ment Dis*; 1995. 495–504 p.
2. Undavalli C, Das P, Dutt T, Bhoi S, Kashyap R. PTSD in post-road traffic accident patients requiring hospitalization in Indian subcontinent: A review on magnitude of the problem and management guidelines. *J Emerg Trauma Shock*. 2014;7(4):327.
3. Sadat Z, Abdi M, Aghajani M. Prevalence of Posttraumatic Stress Disorder and Related Factors Among Patients Discharged From Critical Care Units in Kashan, Iran. *Arch Trauma Res*. 2015 Sep 1;4(4).
4. Lopez AD, Murray CCJL. The global burden of disease, 1990–2020. *Nat Med*. 1998 Nov;4(11):1241–3.
5. Beck JG, Palyo SA, Canna MA, Blanchard EB, Gudmundsdottir B. What factors are associated with the maintenance of PTSD after a motor vehicle accident? The role of sex differences in a help-seeking population. *J Behav Ther Exp Psychiatry*. 2006 Sep;37(3):256–66.
6. Stein DJ, Karam EG, Shahly V, Hill ED, King A, Petukhova M, et al. Post-traumatic stress disorder associated with life-threatening motor vehicle collisions in the WHO World Mental Health Surveys. *BMC Psychiatry*. 2016 Dec 22;16(1):257.
7. Friedman MJ, Resick PA, Bryant RA, Brewin CR. Considering PTSD for DSM-5. *Depress Anxiety*. 2011 Sep;28(9):750–69.
8. Adeolu Ejidokun Adeoye Oyewole AMOALAB. Prevalence of psychiatric morbidity among road traffic accident victims at the national orthopaedic hospital, igbobilagos. *Int J Humanit Soc Sci Invent*. 2015;52–65.
9. Pati D, Lorusso LN. How to Write a Systematic Review of the Literature. *HERD: Health Environments Research & Design Journal*. 2018 Jan 28;11(1):15–30.
10. Yimer GM, Adem YF, Haile Y. Determinants of post-traumatic stress disorder among survivors of road traffic accidents in dessie comprehensive specialized hospital North-East Ethiopia. *BMC Psychiatry*. 2023 Mar 30;23(1):218.





Hetal Jitendrabhai Shah et al.,

11. Daddah D, Ahanhanzo YG, Kpozehouen A, Dos Santos BH, Ouendo EM, Levêque A. Prevalence and Risk Factors of Post-Traumatic Stress Disorder in Survivors of a Cohort of Road Accident Victims in Benin: Results of a 12-Month Cross-Sectional Study. *J MultidiscipHealthc.* 2022;15:719–31.
12. Ratnani D. Study of post traumatic stress disorder among survivors of road traffic accident. *Int J Health Sci (Qassim).* 2022 Jun 21;3762–7.
13. H.Soori, T. Yousefinezhadi, E. Haghihat Manesh. Epidemiologic study of post-traumatic stress disorder(PTSD) among traffic accident victim in tehran. *Iranian Journal of Epidemiology.* 2021;17(3):272–80.
14. Bedaso A, Kediro G, Ebrahim J, Tadesse F, Mekonnen S, Gobena N, et al. Prevalence and determinants of post-traumatic stress disorder among road traffic accident survivors: a prospective survey at selected hospitals in southern Ethiopia. *BMC Emerg Med.* 2020 Dec 26;20(1):52.
15. Alenko A, Berhanu H, Tareke AA, Reta W, Bariso M, Mulat E, et al. <p>Posttraumatic Stress Disorder and Associated Factors Among Drivers Surviving Road Traffic Crashes in Southwest Ethiopia</p>. *Neuropsychiatr Dis Treat.* 2019 May;Volume 15:3501–9.
16. Yohannes K, Gebeyehu A, Adera T, Ayano G, Fekadu W. Prevalence and correlates of post-traumatic stress disorder among survivors of road traffic accidents in Ethiopia. *Int J Ment Health Syst.* 2018 Dec 20;12(1):50.
17. Arora D, BelsiyalCx, Rawat V. Prevalence and determinants of posttraumatic stress disorder and depression among survivors of motor vehicle accidents from a hilly Indian state. *Indian J Psychiatry.* 2021;63(3):250.
18. Hruska B, Irish LA, Pacella ML, Sledjeski EM, Delahanty DL. PTSD symptom severity and psychiatric comorbidity in recent motor vehicle accident victims: A latent class analysis. *J Anxiety Disord.* 2014 Oct;28(7):644–9.
19. Naim R, Wald I, Lior A, Pine DS, Fox NA, Sheppes G, et al. Perturbed threat monitoring following a traumatic event predicts risk for post-traumatic stress disorder. *Psychol Med.* 2014 Jul 17;44(10):2077–84.
20. Sinkler MA, Furdock RJ, Vallier HA. Treating trauma more effectively: A review of psychosocial programming. *Injury.* 2022 Jun;53(6):1756–64.
21. Ongecha-Owuor F, Kathuku D, Othieno C, Ndetei D. Post traumatic stress disorder among motor vehicle accident survivors attending the orthopaedic and trauma clinic at Kenyatta National Hospital, Nairobi. *East Afr Med J.* 2004 Oct 22;81(7).
22. Fekadu W, Mekonen T, Belete H, Belete A, Yohannes K. Incidence of Post-Traumatic Stress Disorder After Road Traffic Accident. *Front Psychiatry.* 2019 Jul 19;10.
23. Punamäki RL, Komprou IH, Qouta S, Elmasri M, de Jong JTVM. The Role of Peritraumatic Dissociation and Gender in the Association Between Trauma and Mental Health in a Palestinian Community Sample. *American Journal of Psychiatry.* 2005 Mar;162(3):545–51.
24. Gopinath B, Jagnoor J, Harris IA, Nicholas M, Maher CG, Casey P, et al. Comparison of health outcomes between hospitalised and non-hospitalised persons with minor injuries sustained in a road traffic crash in Australia: a prospective cohort study. *BMJ Open.* 2015 May;5(9):e009303.
25. Seid M, Azazh A, Enquesslassie F, Yisma E. Injury characteristics and outcome of road traffic accident among victims at Adult Emergency Department of Tikur Anbessa specialized hospital, Addis Ababa, Ethiopia: a prospective hospital-based study. *BMC Emerg Med.* 2015 May;15(1):10.

Table:1 Newcastle -Ottawa (NOS) Quality assessment scale is used for cross section or case control studies

Sr. No.	Author and publication year details	Selection				Comparability	Outcome		Total scoring
		Representativeness of the sample	justified sample size	There was satisfactory response rate	Ascertainment of the exposure		potential confounders were checked	Outcome assessment	
1	Yimer et al.,	*	*	*	**	**	**	*	9





Hetal Jitendrabhai Shah et al.,

	(Y:2023)(10)								
2	Donatien Daddahet.a l., (Y:2022)(11)	*	*	*	**	-	**	*	08
3	Ratnani, D (Y:2022)(12)	*	*	*	**	-	*	*	07
4	Hamid Soori, et al., (Y:2021)(13)	*	-	*	**	-	**	*	07
5	Bedaso et al, et al., (Y: 2020) (14)	*	*	*	**	**	**	*	09
6	Alenkoet al.,(Y: 2019) (15)	*	*	*	**	*	**	*	09
7	Yohannes e t al., (Y:2018)(16)	*	*	*	**	*	**	*	09
8	Arora, et al., (Y:2017) (17)	*	*	*	**	-	**	*	08
9	Hruska et al., (Y:2014)(18)	*	-	*	**	*	**	*	08
10	R. Naim et al., (Y:2014)(19)	*	-	*	**	*	*	*	07

Table 2 Summary of Reviewed Articles.

Sr. No .	Author and publication year details	Countr y	Study design	Participants	Sample size	Instrumen t used	Time when PTSD is asse sse d	Prevalence rate
1	Yimer et al., (Y:2023) (10)	Ethiopi a	case– control study	RTA Survivors	419 (135 cases) samples. Median age 38 Male: 56(41.8%), Female: 79 (58.52%)	PCL-S	After 1month	33.17% Associated factors: Gender (more in females), education, presence of personal psychiatric history, presence of





Hetal Jitendrabhai Shah et al.,

								fracture, witness of death, presence of comorbidity and social support
2	Donatien Daddahet.al., (Y:2022)(11)	Benin (West Africa)	Cross-sectional study	pedestrian, motorcyclist, Drivers & other	865 Sample Mean age: 38.04 Male: 73.98%, Female:26.2%	PCL-S	After 12 months	26.43% Associated factors: Gender (More in female), Hospitalization, negative impact of accident on income and no return to work
3	Ratnani, D (Y:2022)(12)	India	Cross-sectional study	RTA / MVA Survivors	300 samples	DSM-5	After 1 month	22.6% Associated factors: Poor roads, violation of traffic rules, gender (more in female)
4	Hamid Soori, et al., (Y:2021)(13)	Iran	Cross-sectional study	RTA Survivors	350 sample	PCL-5	After 1 week to 2 months	40.06% Associated factors:Age, deathof close family or friend
5	Bedaso et al., et al., (Y: 2020)(14)	Ethiopia	Cross-sectional study	Driver & assistant passenger, Pedestrian	423 sample Median age:30	PCL-S	After 1 month to 3 months	15.4% Male: 15.8%, female: 14.2%. Associated factors: History of previous accident, Time since accident and depression
6	Alenkoet al., (Y: 2019) (15)	Ethiopia	Cross-sectional study	Drivers	402 sample Mean age: 32.11	TSQ	After 1 month	12.6% Associated factors: Near miss RTC's, depression and severe risk





Hetal Jitendrabhai Shah et al.,

								cannabis use
7	Yohannes et al., (Y:2018) (16)	Ethiopia	Cross-sectional study	RTA Survivors	492 Sample Mean age: 30.12 Male (63.6%) Female(36.4%)	PCL-5	After 1 month	22.8% Female: 32.4%, Male: 17.25% Associated factors: Gender (Higher rate in female), Social support, duration since accident and depression.
8	Arora, et al., (Y:2017)(17)	India	Cross-sectional study	Driver, Pillion driver, Passenger, Pedestrian	250 sample Male: 77.2%, Female:22.8%	PCL-S	After 1 month to 12 month	32.4% Associated factors: Age more than 45 years, sustained fracture, sued substance while driving, and blamed self for accident, perceived threat from accident.
9	Hruska et al., (Y:2014)(18)	USA	Cross-sectional study	MVA Victims	356 Sample Male: 211(59.3%), Female:145 (40.7%) Mean age 38.66	CAPS, TSS	After 6 weeks	10%
10	R. Naim et al., (Y:2014)(19)	Israel	Cross-sectional study	Driver, Motorcyclists, Bicycle riders, Pedestrians and passengers	415 Sample Mean age:35.02	CAPS	After 3 month	6.75% Male:1.08% , Female: 10.8% Associated factors:Gender , Role in the MVA





Hetal Jitendrabhai Shah et al.,

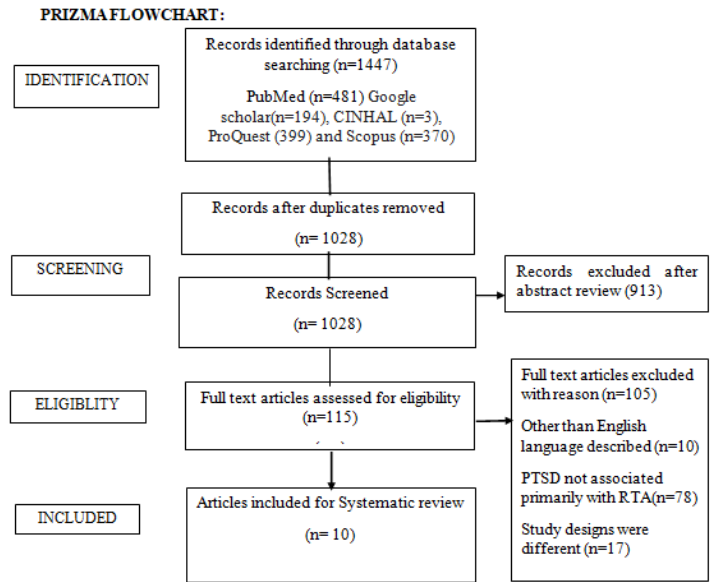


Fig.1 Prizma Flowchart





Ethnomedicinal, Nutritional and Bio-Ceutical Benefits of *Carica papaya* : A Comprehensive Review

Debmalya Biswas¹, Imdadul Mondal¹, Promit Roy¹, Ritabrata Das¹ and Saptarshi Samajdar^{1*}

¹Student (UG), Department of Pharmaceutical Technology, Brainware University, Kolkata, West Bengal, India.

²Associate Professor, Department of Pharmaceutical Technology, Brainware University, Kolkata, West Bengal, India.

Received: 11 Jun 2024

Revised: 18 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Saptarshi Samajdar

Associate Professor, Department of Pharmaceutical Technology,
Brainware University, Kolkata,
West Bengal, India.

E.Mail: dss.pt@brainwareuniversity.ac.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

For many centuries, humans have been dependent on traditional medicine, due to its high safety, affordability, and acceptability. With the increase in the demand for functional food in the Indian pharmaceutical and nutraceutical market, plant materials with high bio ceutical properties are much in demand. One such plant is *Carica papaya* of family Caricaceae better known as papaya. As per multiple reports, different parts of papaya have been traditionally used to control, cure, and rehabilitate against different diseases. The reports suggest that papaya contains different phytochemical analogs of capsaicin, kaempferol, and quercetin along with various enzymes and other proteins which are responsible for its, antioxidant, anti-cancer, anti-inflammatory, immunomodulatory activities and wound healing properties among many others along with improvement in skin and gut health. All these properties directly point towards a highly potential bio ceutical plant material in the form of papaya. With potent clinical studies, papaya can become a trailblazer in the field of pharmaceutical and nutraceutical industries.

Keywords: *C. papaya*, bio ceutical properties, phytochemicals, anti-microbial, anti-cancer.

INTRODUCTION

Herb and herb-based products have been used in traditional medicine for several centuries to treat various diseases. Multiple studies have shown that around three-quarters of the population of the world depends on herb-based medicine for their primary health care. There are various advantages of the natural product over the much-hyped synthetic products like affordability, ease of availability, lower toxicity, lower adverse effects as well as low chances



**Debmalya Biswas et al.,**

of multi-drug resistances. In recent years, the dependence on natural products mainly from plants and herbs has majorly increased due to the bio-ceutical revolution, where the usage of plant products as functional foods and nutraceuticals has played a key role. Here plants and plant products can neither be categorized as food nor as pharmaceuticals, but more as supplements to control, prevent, ameliorate as well as rehabilitate certain critical diseases. Many plants like garlic, ashwagandha, spirulina, chicory, fenugreek, etc. have found market share worth millions of dollars around the world due to the bio-ceutical revolution. In this study a plant very commonly seen around India, that is *Carica papaya* will be thoroughly emphasized through different reports of its curative properties. [1].

Carica papaya, the well-known papaya, is a plant that belongs to the Caricaceae family. It contains a variety of chemicals and biomolecules, including papain, which has important uses in industry and medicine. Originating in Central America and Mexico, papaya is an evergreen plant that grows in tropical climates[2,3]. There is little evidence to suggest that it originated in the northwest of South America[4] and was introduced to India in the sixteenth century.

C. papaya, also referred to as papaya, has anti-inflammatory effects, particularly in the leaves and seeds [5]. According to research, papaya leaves contain chemicals that may aid the immune system manage its inflammatory response [5]. Research has also shown that papaya leaf extracts can reduce inflammation in a variety of cells, including those involved in wound healing [6,7]. Papaya seeds appear to have anti-inflammatory properties, presumably by lowering oxidative stress and inflammation in the liver[8]. In this study, comprehensive details regarding the ethnomedicinal, bioceutical, and nutritional benefits of papaya have been discussed.

TAXONOMY, MORPHOLOGY, AND DISTRIBUTION OF *C. papaya*:**Botanical Classification**

The domain of *C. papaya* comes under the flowering plant, while the kingdom and sub-kingdom of the plant are 'Plantae' and 'Tracheobionta' respectively. Class and sub-class of *C. papaya* are 'Magnoliopsida' and 'Dilleniidae'. Phylum, order, family, and genus of *Carica papaya* are 'Streptophyta', 'Brassicales', 'Caricaceae', and 'Carica'. Thus the botanical name of *Carica papaya* is as following *Carica papaya* Linn. [9]

Morphology of *C. papaya* tree

The papaya plant is often a single, hollow, erect-stemmed, semi-woody perennial tree that grows to a height of around 20 to 30 feet. The stem is 8 inches in diameter, light green to tan brown, and bears leaf scars. It has a cluster of huge, palmately-lobed leaves with 25–100 cm long petioles. Depending on the sort of blossom they produce, papaya plants can be categorized as male, female, or hermaphrodite. Some plants are known as monoecious because they may produce both male and female blooms[10].

Distribution of *C. papaya*

The specific region of origin for papaya is unknown. It is thought to be native to Tropical America, possibly in Southern Mexico and adjacent Central America. Today, successful commercial production is focused largely in Hawaii, Tropical Africa, the Philippines, India, Sri Lanka, Malaysia, and Australia; South Africa and Latin America produce on a smaller scale [2,3,4].

Nutritional Content of *C. papaya*

In today's world, conventionally synthetically generated medications are the primary treatment approaches. However, due to their unfavorable side effects in both long and short-term consumption of drugs, alternative pharmaceutical systems are becoming increasingly popular due to their lower adverse effects and higher consumer compliance. *C. papaya*, sometimes known as papaya, is an alternative natural medicine source. The pulp, leaves, and seeds contain high levels of vitamins, bioactive compounds, and lipids that prevent hypercholesterolemia, protect



**Debmalya Biswas et al.,**

against thrombogenesis and oxidative stress, and lower inflammatory markers and anti-platelet aggregation—all of which can be caused by obesity [11,12].

Vitamins and Minerals

Papaya is a very common fruit with a low price and excellent nutritional content. It has fewer calories and more vitamins, macronutrients, and other minerals, making it ideal for individuals who are in danger of gaining weight. There are several macronutrients found in *C. papaya* fruit [13]. Carbohydrates are the most prevalent macronutrient in *C. papaya* (7.2 gm per 100 mg), but proteins, lipids, and fiber are also present. Papaya contains a large quantity of energy, with 32 kcal per 100 mg. The most abundant minerals in *C. papaya* fruit are sodium, potassium, calcium, magnesium, phosphorus, and iron [14]. Various vitamins and their complexes are present in *C. papaya* fruit [15]. Papaya is a Vitamin A-rich fruit. Also Vitamin B complexes like B1,B2,B3,B6, B9, and Vitamin C are also present.

Enzymes

Papaya consists of many enzymes that serve various purposes. Enzymes typically induce proteolytic activity. However, enzymes can help the digestion of a variety of macronutrients. Papaya enzymes have various roles such as immunomodulation and anti-inflammatory effects[16,17,18]. Papaya proteases have anti-inflammatory qualities that aid in relieving pain and suffering from arthritis, edema, and osteoporosis. Enzymes in *C. papaya* are broadly distributed throughout the plant. It contains cysteine endopeptidases such as glycyI endopeptidase, cysteine proteinases, serine proteinase inhibitors, glutaminyI cyclase caricain, class II chitinase, papain, and chymopapain. Papain is a non-specific thiol protease that acts similarly to pepsin in gastric juice, making it a good digestive aid and pepsin dilapidation agent. [19]. Other active compounds of *C. papaya* are lipase, a hydrolase, which is tightly bonded to the water-insoluble fraction of crude papain and is thus considered a "naturally immobilized" biocatalyst [20]. Pectinase, found in papaya fruit, is involved in the breakdown of pectin, a carbohydrate found in plant cell walls. The seeds of papaya include lipase and amylase enzymes, which help to break down lipids and starch. Cellulase, which is found in plant leaves, helps to break down cellulose into simple sugar. Younger plants have more of these enzymes than older plants.

Bioactive Compounds of *C. papaya*

Bioactive compounds are naturally occurring chemical components found in various plant parts that provide medical benefits to many animals. Chemical investigations conducted in several research revealed that papaya leaves contain several phytochemicals that have been shown to benefit health. These phytochemicals contain considerable levels of alkaloids, saponins, glycosides, flavonoids, phenolic compounds, enzymes, amino acids, lipids, carbohydrates, vitamins, and minerals [21]. Papaya leaves have been used medicinally in numerous Asian countries for centuries to treat a variety of ailments. Because of the presence of the aforementioned critical operating components, they are used to treat a wide range of illnesses, including diabetes, ulcers, hypertension, dengue, menstruation problems, dermatitis, sinusitis, weakness, and others [22]. Papaya leaves contain seven flavonoids: quercetin, quercetin3-(2G-rhamnosylrutinoside), quercetin 3-rutinoside, kaempferol 3-rutinoside, kaempferol 3-(2Grhamnosylrutinoside), and myricetin 3-rhamnoside. The leaves include phenolic compounds such as caffeine, protocatechuic acid, quercetin, 5,7-dimethyl coumarin, p-coumaric acid, and chlorogenic acid [23]. These phenolic chemicals, in particular, contribute to anti-allergic, antiviral, anti-inflammatory, and anti-cancer capabilities, as well as anti-dengue activity. The key phytochemicals found in papaya plant leaves are papain, cystatin, chymopapain, tocopherol, phenolic acids, cyanogenic glucosides, glucosinolates, and vitamin C, which can improve the general anti-oxidant qualities of the blood. Papaya leaves have been utilized as antispasmodic, analgesic, and antibacterial because they contain numerous alkaloids such as carpaine, pseudocarpaine, dehydro-carpaine, and phenolic compounds. Carpaine, dehydrocarpaine I, and dehydrocarpaine II are the principal bioactive components found in papaya leaves. Because of the presence of carpeting, these herbal leaves are used in Ayurvedic formulations to treat a variety of physical ailments and viral fevers such as dengue and chikungunya. Carpaine has also been shown to have strong anticancer and antihelminthic effects [24]. Its content has been reported to be maximum in mature papaya leaves. Researchers recently found that malaria can be efficiently cured with papaya leaves. This beneficial impact is attributed to the presence of alkaloids in the leaves, which contain quinine, an antimalarial drug [25,26]. Papaya leaves contain a high concentration of



**Debmalya Biswas et al.,**

biological enzymes, specifically papain and chymopapain. The concentration of papain in papaya leaf extract present in significant amounts [15] and due to which very powerful digestive action even higher than pepsin is seen. Mainly alkaloids, saponins, glycosides, phenolic compounds, and flavonoids are responsible for the anti-inflammatory and anti-cancerous properties of papaya leaves [27].

Antioxidant

Due to concerns about the toxicity of synthetic antioxidants, plant-derived compounds are often sold as natural antioxidants as an alternative. Oxidative chemicals found in many plants may have antibacterial, antiviral, and anticancer effects. They also have an impact on other health benefits [23]. Several components in the anti-oxidant family, such as carotenoids, vitamin C, vitamin E, and phenolic compounds, exert anti-oxidant action through interaction. As a result, it is difficult to determine C. papaya's total antioxidant activity solely on individual active components [28]. The total antioxidant level in C. papaya fruit varies significantly as it ripens. Antioxidant levels gradually rise as the fruit matures. Free radicals generate oxidative damage that has a significant impact on many chronic diseases, and antioxidants can help our health by reducing the production of free radicals. New sources of natural antioxidants that are both safe and economically viable are currently being studied [29]. Researchers created silver nanoparticles from C. papaya peel extract (CPPE) and tested their antioxidative capabilities to confirm their efficiency. It was discovered that the concentration-dependent activity of AgNPs was 56% for synthesized AgNPs and 38% for commercially available CPPE [30]. In the whole plant, the fruit and the seeds contribute to the antioxidant property the most. Various tests to determine the anti-oxidant properties e.g. TPC(Total phenol content), TFC(Total flavonoid content), FRAP(Ferric reducing antioxidant power), Radical scavenging activity of various extracts of solvents like n-butanol, Petroleum Ether, Methanol, Ethyl Acetate, etc. Out of the extracts, Ethyl acetate and n-butanol fractions hold the strongest anti-oxidant activity [31]. The antioxidant potential of papaya peel may contribute to the production of functional foods and nutraceuticals shortly [32]. To demonstrate antioxidant activity, researchers treated papaya peel residues with ethanol and dried them in a microwave oven to produce dietary fiber concentrations (DFCs). The samples' chromatographic analysis revealed the presence of carotenoids, phenolics, ascorbic acid, protocatechuic acid, manghaslin, quercetin 3-O-rutinoside, caffeoyl hexoside, ferulic acid, lutein, zeaxanthin, and beta-carotene. Digestibility studies revealed that the percentage of indigestible fiber included antioxidants. Salla et al. studied the in vitro antioxidant activity of papaya peel extracts as well as their effects on endogenous glutathione, superoxide dismutase, catalase, cyclo-oxygenase-2 (COX-2), cyclo-oxygenase-3, and DNA fragmentation in HepG2 cells[33].

Antimicrobial effects

Papaya seed exhibits antibacterial action directed against the trophozoites of *Trichomonas vaginalis*. According to the paper, papaya seed should be used carefully to prevent toxicity in urinogenital disorders like trichomoniasis. Papaya seed is used in urinogenital disorders like trichomoniasis with care to avoid toxicity [33]. With the agar cup plate method, it was demonstrated that the papaya seed and pulp had bacteriostatic properties against a range of enteropathogens, including *Bacillus subtilis*, *Enterobacter cloacae*, *Escherichia coli*, *Salmonella typhi*, *Staphylococcus aureus*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, and *Klebsiella pneumonia*. [34]. It has been demonstrated that the papaya fruit, seeds, leaves, and peels of C. papaya have antibacterial properties against the possibly harmful microbe.. the seed and pulp showed bacteriostatic properties against several enteropathogens such as *Bacillus subtilis*, *Salmonella typhi*, *Staphylococcus sp.* C. papaya could find usefulness in the production of drugs against organisms causing urinary tract infections [35]

Antibacterial activity

The results of an investigation indicate the antibacterial properties of papaya fruiting body extracts. It is most likely for this reason that some villagers treat wounds with paw-paw. Although it has no impact, the extract is bacteriostatic and bactericidal. This partially defies Viera's results [36]. For individuals with impaired immune systems, certain fungal infections, like candida, can be extremely serious or even fatal.[37]. It has been discovered that the seeds of C. papaya exhibit bacteriostatic activity against a variety of enteropathogens, including *Salmonella typhi*,



**Debmalya Biswas et al.,**

Escherichia coli, *Bacillus subtilis*, *Enterobacter cloacae*, *Proteases vulgaris*, *Pseudomonas aeruginosa*, and *Klebsiella pneumonia*. Gram-negative bacteria were more sensitive to the extract than gram-positive bacteria among those tested[38].

Antifungal activity

Numerous bodily organs, including the skin, nails, reproductive system, gastrointestinal tract, heart, and neurological system, can be harmed by fungi[39]. The combination of fluconazole and papaya latex inhibits the growth of *Candida albicans*[40].

Anti-inflammatory activity

The *C.papaya* leaf extract was examined in rats using edema, granuloma, and arthritis models. The extract showed a significant reduction in paw edema, granuloma formation, and reduced inflammation in rats. Thus it proved the anti-inflammatory activity of *C.papaya* [41]. Numerous plants or isolated derivatives from the plant source show a role as anti-inflammatory via modulation of various activities [42]. Consuming papaya fruits reduced the anti-inflammatory response in healthy people, which was mediated by regulatory T-cells. [43]. A significant study demonstrated the anti-inflammatory properties of *C. papaya* seeds, and the methanolic extract showed inhibition ranging from 57.1% to 64.2%, which is less than the typical anti-inflammatory medicine aspirin's 85.7%. [44,42]. *C. papaya*, as noted above, is a tropical plant containing a wide range of bioactive secondary metabolites (e.g. alkaloids, phenolics, flavonoids, carotenoids, tannins, saponins, etc.) and proteolytic enzymes (papain and chymopapain). Several phytochemicals included in papaya have demonstrated the ability to mitigate chronic inflammatory diseases and their associated negative effects by altering the expression of inflammatory markers.[45]

Immunomodulatory activity

Papain, sourced from *C. papaya*, induces human eosinophils to degranulate and produce superoxide anion. The papain activation was eliminated by the E-64 inhibitors, indicating that protease activity is necessary to initiate the eosinophil response. The protein G-linked receptor is most likely the mechanism by which eosinophils carry out this function. Presently, it seems that bromelaine and papain exhibit opposing effects based on the target cell.[46]. Fermented papaya preparation exerts both immunomodulatory and antioxidant activity in the macrophage cell line RAW 264 and it is a macrophage activator, which augments nitric oxide synthesis and TNF-alpha secretion independently of lipopolysaccharides[47].

Anti-Cancer

Cancer is one of the most lethal diseases around the world in terms of mortality globally. Lung cancer is the most common cancer in men, followed by breast cancer in women. Phytochemical analysis of papaya suggests that Lycopene, beta carotenoid, benzylisothiocyanate, beta-cryptoxanthin, benzyl glucosinolate, chlorogenic acid, caffeic acid, protocatechuic acid, quercetin, and some other significant phytochemicals are present in *C. papaya*. Three classes of bioactive substances—phenolics, carotenoids, and glucosinolates—among the more than 5000 molecules from plants that have been linked to anticancer qualities have drawn a lot of attention in anticancer research. [48]. Researchers discovered that individuals with blood, liver, lung, or stomach cancer have a better chance of life after drinking papaya leaf extract (solution) [49]. In vitro studies utilizing papaya seed homogenate extract demonstrated excellent efficiency in decreasing superoxide production and inducing apoptosis in the acute promyelocytic leukemia cell line HL-60. The principal cause of this action was benzyl isothiocyanate, or BITC [15]. BITC extracted from papaya fruit extract had a cytotoxic effect on developing human colon CCD-18Co cells, causing them to become quiescent. [50]. Due to its impact on spindle formation, the aqueous extract of *C. papaya* given to onion bulbs disrupted the mitotic cell division of *Allium cepa*, demonstrating its cytotoxic effect [51].

Papaya (*C. papaya* Linn) seeds include anti-cancer properties that can be used to treat liver and prostate tumors, among other cancers [57]. The goal of this study was to assess the possible cancer-preventive properties of papaya black seeds found in mature papaya fruits. The in vitro cytotoxicity of the "methanolic extract of papaya black seeds" (MPB) 1 against human liver cancer Hep G2 cell lines was studied. The half-maximum inhibitory concentration (IC50) was determined using the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) test.



**Debmalya Biswas et al.,**

Additionally, Acridine Orange-Ethidium Bromide (AO-EB) labeling has been utilized to assess the apoptotic changes that MPB has generated in cancer cells. Researchers have evaluated gene expression control by targeting the B cell lymphoma-2 (Bcl-2), p53, and caspase-3 genes using quantitative Real-Time Polymerase Chain Reaction (qRT PCR). Our findings indicate that papaya black seeds may be a promising treatment for liver cancer, with an IC₅₀ value of 24.35 µg/mL and the ability to induce apoptosis by upregulating p53 and downregulating Bcl-2 [58].

Amazu et al reported a study using extracts of papaya fruit on Swiss albino male mice, divided into six groups with six animals per group. The mice were fed on mice pellets and had access to food and water. An initial pilot study was conducted to determine the minimum and maximum dosages of a drug extract. The animals were given crude drug extract in different doses, with the negative control group receiving normal saline IP. Symptoms of toxicity or death were observed within 24 hours, and any dead animals were removed. The LD₅₀ was calculated as the probability of the minimum dose that killed half the animals in the study group or the mean of two doses where applicable.[59]

Another study investigated the effects of a methanolic seed extract of *C. papaya* on fresh egg albumin-induced inflammation in rats. The results showed that the extract significantly inhibited edema at all doses, with no significant difference between 200 mg/kg and 100 mg/kg at 60-120 minutes. However, aspirin had a significantly different inhibitory effect, with detectable differences at all periods. The crude extract of *C. papaya* seeds inhibited inflammation by 57.1% to 64.2%, compared to 85.7% with aspirin. This implies that aspirin has greater anti-inflammatory efficacy than the crude extract, which varied from 57.1% to 64.2%. The study shows that additional characterization and purification of active components may address this difference [59].

Wound healing

Traditionally, *C. papaya* has been used to treat wounds and other skin conditions. It is a commonly used, readily available treatment for burns and other wounds, and is particularly popular in developing nations. The study conducted by Nayak et al. evaluated the antibacterial and wound-healing activities of *C. papaya* seed extract. Using an in vivo excision wound model, the ability of an ethanol extract of *C. papaya* seed (50 mg/kg/day) to heal wounds in Sprague-Dawley rats was tested. The animals were randomly divided into four groups of six: Group One was the control group, Group Two received papaya seed extract therapy, Group Three received a 1:1 ratio of mupirocin and papaya seed extract treatment, and Group Four received mupirocin ointment treatment. To evaluate the wound-healing process, the rate of wound contraction and hydroxyproline content were evaluated activity of the seed extract. [52].

Gastrointestinal health

The anti-diarrheal efficacy of alcoholic and aqueous extracts from the fruit of *C. papaya* was investigated in albino Wistar rats. When administered orally at doses of 100, 200, and 400 mg/kg, the alcoholic and aqueous extracts showed considerable dose-dependent anti-diarrheal action in cases of magnesium sulfate and castor oil-induced diarrhea. The studied extracts' anti-diarrheal activities were comparable to those of the recommended medicine, loperamide (3 mg/kg). The findings support the traditional use of *C. papaya* in the treatment of diarrheal diseases [53]. The current study aimed to assess the antidiarrheal activities and phytochemical content of *C. papaya* fruit against a variety of gastrointestinal pathogens. Powdered plant components from both raw and ripe fruit were extracted using a Soxhlet system and a range of solvents, including petroleum ether, benzene, chloroform, acetone, ethanol, and water. phytochemical examination of *C. papaya* revealed the presence of proteins, amino acids, carbohydrates, tannins, saponins, alkaloids, phenolic compounds, and phytosterols. To determine the extract's MIC, sterile 96-well microtitre plates were used. The plant extracts inhibited all of the intestinal pathogens examined. The ripe fruit's acetone extract (0.39 mg/ml) and the raw fruit's chloroform extract (25.0 mg/ml) had the highest antibacterial activity among all the fruit extracts. [54].



**Debmalya Biswas et al.,****Skin conditions**

One of the most widely grown plants in the world is the *C. papaya*. Traditional medicine has made use of various parts of the *C. papaya*. However, no research using *C. papaya* leaf (CPL) has been done about skin photoaging. The current study examined the anti-aging properties of *C. papaya* leaf on UVB-irradiated normal human dermal fibroblasts (NHDFs). By scavenging reactive oxygen species (ROS) generation, *C. papaya* leaf significantly suppressed UVB-induced activation of mitogen-activated protein kinases (MAPKs) and activator protein-1 (AP-1) signaling. Additionally, our findings showed that CPL inhibited the breakdown of Type I procollagen by regulating matrix metalloproteinases (MMPs) expression negatively and transforming growth factor- β 1 (TGF- β 1) activity positively. This led to an increase in the synthesis of Type I procollagen, which is a significant part of the extracellular matrix (ECM). Furthermore, the combination of the two active compounds implies that they were CPL's active compounds. When considered collectively, the findings indicate that *C. papaya* leaf holds promise for additional research and the creation of a cosmetic product to treat a range of skin issues, including photoaging [55]. Synthetic skin care lotions containing aluminum, parabens, butylated hydroxyanisole (BHA), and sodium carboxymethyl cellulose are currently overflowing the market. Method: The goal of this work was to create an herbal skin care lotion with the addition of distilled water, beeswax, liquid paraffin, and borax *C. papaya* extract. Following formulation, it was assessed, and several physicochemical and organoleptic parameters (color, odor, and appearance), as well as pH, viscosity, spreadability, microbiological growth, irritancy, and ease of removal, were identified and documented. Findings: The created skin care herbal lotion's various assessment criteria matched both the marketed formulation and standard values. In conclusion, customers today are more likely to choose natural cosmetics to prevent unneeded side effects [56].

Neuroprotective effects

Chronic inflammatory responses in the central nervous system (CNS) indicate neuroinflammation, a pathogenic condition that dramatically accelerates the onset of neurodegenerative disorders, most notably Alzheimer's and Parkinson's disease [60]. *C. papaya* possesses anti-inflammatory and immunomodulatory properties, making it a prospective treatment for neuroinflammation [61, 62]. The flavonoid-rich components of papaya, particularly its leaves and seeds, have strong anti-inflammatory activities [59]. The antioxidant properties of papaya leaf extract are well known for their potential to prevent lipid peroxidation and boost blood antioxidant capacity [63]. Furthermore, particular compounds found in papaya seed extract have been shown to have therapeutic properties for reducing inflammation.

Action against oxidative stress

Oxidative stress, one of the most complex disease causes in the human body, is a damaging state caused by an imbalance of endogenous pro- and antioxidant species. It is also a fundamental biological process that contributes to neurodegeneration in Alzheimer's and Parkinson's disease. We have observed that *C. papaya* contains a high concentration of phenolic chemicals and flavonoids. Flavonoids have been demonstrated to reduce the generation of reactive oxygen species (ROS), boost antioxidant protein expression, improve cerebral blood flow and neuron survival, and reduce apoptosis, amyloidogenic effects, and dopaminergic neuron loss [64]. It has been demonstrated that phenolic acids can alleviate a variety of disorders, including depression, neuroinflammation, apoptosis, memory loss, and excitotoxicity [65].

Cardiovascular benefits

C. papaya contains bioactive substances such as polyphenols, flavonoids, vitamins, and minerals, and it has been shown to have potential cardiovascular effects. Because of its high potassium concentration and vasodilating characteristics, it may reduce blood pressure. Papaya also contains antioxidants that reduce inflammation and oxidative stress, lowering the risk of atherosclerosis and coronary artery disease [66].





Debmalya Biswas et al.,

Action against Thrombocytopenia

C. papaya contains bioactive substances such as polyphenols, flavonoids, vitamins, and minerals, and it has been shown to have potential cardiovascular effects. Because of its high potassium concentration and vasodilating characteristics, it may reduce blood pressure. Papaya also contains antioxidants that reduce inflammation and oxidative stress, lowering the risk of atherosclerosis and coronary artery disease [67].

Future Prospects

There are several obstacles in the way of fully investigating bio ceutical potential of *C. papaya*, despite its possible advantages to be used as a functional food. A significant barrier is the paucity of research on the therapeutic benefits of *C. papaya*. Although some studies have yielded encouraging findings, a thorough investigation of the potential health risks of *C. papaya* is still lacking. Additionally, there is difficulty in standardizing the dosage and preparation of *C. papaya*. The fruit is consumed in various forms, including fresh fruit, juice, and supplements, and the dosage and preparation can vary widely. Lastly, it may be difficult to overcome regulatory obstacles to get permission to employ *C. papaya* as a medicinal or nutraceutical agent. The plant is not currently recognized as a medicine by regulatory agencies, making it difficult to obtain approval for its use in clinical settings.

CONCLUSIONS

Hence, all the evidence provided in the manuscript about the pharmacological and functional food qualities of *C. papaya* which makes it a viable option for medicinal and nutraceutical uses. An untapped market that is worth more than USD 50 million in 2024 for the multi-usage traditionally important plants like *C. papaya*. The fruit of the plant has many properties like antibacterial qualities that can fight infections, antioxidants that shield cells from harm, and enzymes that help with digestion and decrease inflammation.

REFERENCES

1. Ugbogu EA, Dike ED, Uche ME, Etumnu LR, Okoro BC, Ugbogu OC, Adurosakin OE, Chinma CE, Ohaeri E, Iweala EJ. Ethnomedicinal uses, nutritional composition, phytochemistry, and potential health benefits of *Carica papaya*. *Pharmacological Research-Modern Chinese Medicine*. 2023 May 22:100266.
2. Kaibing, Z.; Hui, W.; Wenli, M.; Xiaona, Li.; Ying, L.; Haofu, D. Antioxidant activity of papaya seed extracts. *Journal of Molecular Biology* 2011, 16, 6179–6192.
3. Milind P, Gurditta G. Basketful benefits of papaya. *International research journal of pharmacy*. 2011;2(7):6-12.
4. Chan, Y.K.; Paull, R.E. *Papaya (Carica papaya L.)*, Caricaceae. In: *Encyclopedia of Fruit and Nuts*; Janick, J.; Paull, R.E.; Eds.; CABI: Wallingford, U.K., 2008; 237–247.
5. Pandey, S., Cabot, P. J., Shaw, P. N., & Hewavitharana, A. K. (2016). Anti-inflammatory and immunomodulatory properties of *Carica papaya*. *Journal of Immunotoxicology*, 13(4), 590–602. <https://doi.org/10.3109/1547691X.2016.1149528>
6. Kong YR, Jong YX, Balakrishnan M, Bok ZK, Weng JKK, Tay KC, Goh BH, Ong YS, Chan KG, Lee LH, Khaw KY. Beneficial Role of *Carica papaya* Extracts and Phytochemicals on Oxidative Stress and Related Diseases: A Mini Review. *Biology (Basel)*. 2021 Apr 1;10(4):287. doi: 10.3390/biology10040287. PMID: 33916114; PMCID: PMC8066973.
7. Singh SP, Kumar S, Mathan SV, Tomar MS, Singh RK, Verma PK, Kumar A, Kumar S, Singh RP, Acharya A. Therapeutic application of *Carica papaya* leaf extract in the management of human diseases. *Daru*. 2020 Dec;28(2):735-744. doi: 10.1007/s40199-020-00348-7. Epub 2020 May 5. PMID: 32367410; PMCID: PMC7704890.
8. Shaban, N.Z., El-Kot, S.M., Awad, O.M. et al. The antioxidant and anti-inflammatory effects of *Carica Papaya* Linn. seeds extract on CCl4-induced liver injury in male rats. *BMC Complement Med Ther* 21, 302 (2021). <https://doi.org/10.1186/s12906-021-03479-9>.
9. Ayurvedic pharmacopeia of India, Govt of India.



**Debmalya Biswas et al.,**

10. Aravind G, Bhowmik D, Duraivel S, Harish G. Traditional and medicinal uses of *Carica papaya*. Journal of medicinal plants studies. 2013;1(1):7-15.
11. Teixeira LG, Lages PC, Jасolka TL, Aguilар EC, Soares FL, Pereira SS, Beltrão NR, Matoso RD, Nascimento AM, Castilho RO, Leite JI. Extrato de chá branco reduz estresse oxidativo e triacilgliceróis em camundongos obesos. Food Science and Technology. 2012;32:733-41.
12. Vij T, Prashar Y. A review on medicinal properties of *Carica papaya* Linn. Asian Pacific Journal of Tropical Disease. 2015 Jan 1;5(1):1-6.
13. Krishna KL, Paridhavi M, Patel JA. Review on nutritional, medicinal and pharmacological properties of *Papaya* (*Carica papaya* Linn.).
14. Nwofia GE, Ojmelukwe P, Eji C. Chemical composition of leaves, fruit pulp and seeds in some *Carica papaya* (L) morphotypes.
15. Parle M, Gurditta A. Basketful benefits of papaya. Int. Res. J. Pharm. 2011 Jul;2(7):6-12.
16. Rakhimov MR. Anti-inflammatory activity of domestic papain. Eksperimental'naia i Klinicheskaia Farmakologiya. 2001 Jul 1;64(4):48-9.
17. Mohr T, Desser L. Plant proteolytic enzyme papain abrogates angiogenic activation of human umbilical vein endothelial cells (HUVEC) in vitro. BMC complementary and alternative medicine. 2013 Dec;13:1-9.
18. Rose B, Herder C, Löffler H, Meierhoff G, Schloot NC, Walz M, Martin S. Dose-dependent induction of IL-6 by plant-derived proteases in vitro. Clinical & Experimental Immunology. 2006 Jan;143(1):85-92..
19. J Huet J, Looze Y, Bartik K, Raussens V, Wintjens R, Boussard P. Structural characterization of the papaya cysteine proteinases at low pH. Biochemical and Biophysical Research Communications. 2006 Mar 10;341(2):620-6.,
20. Dominguez de Maria P, Sinisteraa JB, Tsai SW, Alcantara AR (2006) Biotech Adv 24: 493-499.
21. Alara OR, Abdurahman NH, Alara JA. *Carica papaya*: comprehensive overview of the nutritional values, phytochemicals and pharmacological activities. Advances in traditional medicine. 2020:1-31.
22. Dharmarathna SL, Wickramasinghe S, Waduge RN, Rajapakse RP, Kularatne SA. Does *Carica papaya* leaf-extract increase the platelet count? An experimental study in a murine model. Asian Pacific Journal of Tropical Biomedicine. 2013 Sep 1;3(9):720-4.
23. Huet J, Looze Y, Bartik K, Raussens V, Wintjens R, Boussard P. Structural characterization of the papaya cysteine proteinases at low pH. Biochemical and Biophysical Research Communications. 2006 Mar 10;341(2):620-6.
24. Dash PR, Mou KM. A Comprehensive Review on Five Medicinal Plants of Bangladesh. Chemical Constituents and Uses
25. Alexandre-Michel NN, Jean M. Medicinal plants used in traditional treatment of malaria in Cameroon. Journal of Ecology and the Natural Environment. 2011 Mar;3(3):104-17.
26. Pinelo M, Rubilar M, Sineiro J, Nunez MJ. Extraction of antioxidant phenolics from almond hulls (*Prunus amygdalus*) and pine sawdust (*Pinus pinaster*). Food chemistry 2004 Apr 1;85(2):267-73.
27. Soib HH, Ismail HF, Husin F, Abu Bakar MH, Yaakob H, Sarmidi MR. Bioassay-guided different extraction techniques of *Carica papaya* (Linn.) leaves on in vitro wound-healing activities. Molecules. 2020 Jan 24;25(3):517.
28. Kokila T, Ramesh PS, Geetha D. Biosynthesis of AgNPs using *Carica Papaya* peel extract and evaluation of its antioxidant and antimicrobial activities. Ecotoxicology and Environmental Safety. 2016 Dec 1;134:467-73.
29. Kafuko L. Antioxidant and antimicrobial activity of *Carica papaya* extracts in minimally processed fresh produce (Master's thesis, University of Pretoria (South Africa)).
30. Calvache Calvache JN, Cueto M, Farroni A, de Escalada Pla M, Gerschenson LN. Antioxidant characterization of new dietary fiber concentrates from papaya pulp and peel (*Carica papaya* L.). Journal of Functional Foods. 2016 Dec 1;27:319-28.
31. Robinson P. Seeds of *Carica papaya* for mass treatment against Ascariasis. Indian Journal of Child Health. 1958;7:815-7.
32. Indran IR, Tufo G, Pervaiz S, Brenner C. Recent advances in apoptosis, mitochondria and drug resistance in cancer cells. Biochimica et Biophysica Acta (BBA)-Bioenergetics. 2011 Jun 1;1807(6):735-45
33. Calzada F, Yépez-Mulia L, Tapia-Contreras A. Effect of Mexican medicinal plant used to treat trichomoniasis on *Trichomonas vaginalis* trophozoites. Journal of ethnopharmacology. 2007 Sep 5;113(2):248-51.



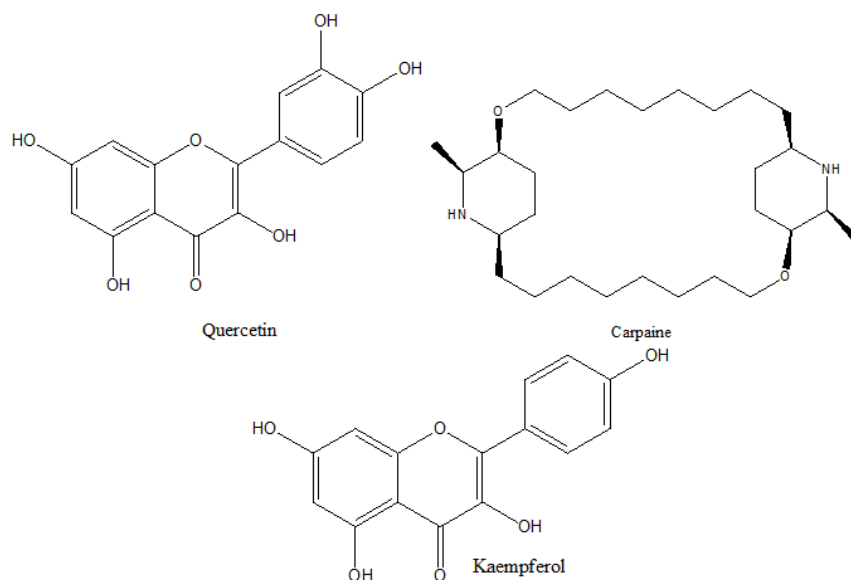
**Debmalya Biswas et al.,**

34. Osato JA, Santiago LA, Remo GM, Cuadra MS, Mori A. Antimicrobial and antioxidant activities of unripe papaya. *Life sciences*. 1993 Jan 1;53(17):1383-9.
35. Kadiri O, Olawoye B, Fawale OS, Adalumo OA. Nutraceutical and antioxidant properties of the seeds, leaves and fruits of *Carica papaya*: Potential relevance to humans diet, the food industry and the pharmaceutical industry-a review. *Turkish Journal of Agriculture-Food Science and Technology*. 2016 Nov 28;4(12):1039-52
36. Akujobi CN, Ofodeme CN, Enweani CA. Determination of antibacterial activity of *Carica papaya* (paw-paw) extracts. *Nigerian Journal of Clinical Practice*. 2010 Jan 1;13(1):55-7.
37. Morace G, Perdoni F, Borghi E. Antifungal drug resistance in *Candida* species. *Journal of Global Antimicrobial Resistance*. 2014 Dec 1;2(4):254-9
38. Gunde MC, Amnerkar ND. Nutritional, medicinal and pharmacological properties of papaya (*Carica papaya* Linn.): A review. *Journal of Innovations in Pharmaceuticals and Biological Sciences*. 2016;3(1):162-9.
39. He X, Ma Y, Yi G, Wu J, Zhou L, Guo H. Chemical composition and antifungal activity of *Carica papaya* Linn. seed essential oil against *Candida* spp. *Letters in applied microbiology*. 2017 May 1;64(5):350-4.
40. Giordani R, Gachon C, Moulin-Traffort J, Regli P. A synergistic effect of *Carica papaya* latex sap and fluconazole on *Candida albicans* growth. *Mycoses*. 1997 Dec;40(11-12):429-37
41. Sumathy H, Sunil AG, Babu CS, Sekar S, Ranju V. Potential medicinal properties of *Carica papaya* Linn.-A mini review. *Asian Journal of Pharmaceutical and Clinical Research*. 2017;10(4):275
42. Rahmani AH, Aldebasi YH. Potential role of *carica papaya* and their active constituents in the prevention and treatment of diseases. *Int J Pharm Pharm Sci*. 2016;8(1):11-5.
43. Sudhakar N, Vidhya RT. Potential medicinal properties of *Carica papaya* Linn.-A mini review. *Int. J. Pharm. Pharm. Sci*. 2014;6(2):1-4.
44. Mahrous NS, Noseer EA. Anticancer potential of *Carica papaya* Linn black seed extract against human colon cancer cell line: in vitro study. *BMC Complementary Medicine and Therapies*. 2023 Jul 29;23(1):271.
45. Pandey S, Cabot PJ, Shaw PN, Hewavitharana AK. Anti-inflammatory and immunomodulatory properties of *Carica papaya*. *Journal of immunotoxicology*. 2016 Jul 3;13(4):590-602.
46. Gunde MC, Amnerkar ND. Nutritional, medicinal and pharmacological properties of papaya (*Carica papaya* Linn.): A review. *Journal of Innovations in Pharmaceuticals and Biological Sciences*. 2016;3(1):162-9
47. Roshan A, Verma NK, Gupta A. A brief study on *Carica Papaya*-a review. *International Journal of Current Trends in Pharmaceutical Research*. 2014;2(4):541-50.
48. Fauziya S, Krishnamurthy R. Papaya (*Carica papaya*): Source material for anticancer. *CIBTech J Pharm Sci*. 2013;2(1):25-34.
49. Kaur M, Talniya NC, Sahrawat S, Kumar A, Stashenko EE. Ethnomedicinal uses, phytochemistry and pharmacology of *Carica papaya* plant: A compendious review. *Mini-Reviews in Organic Chemistry*. 2019 Aug 1;16(5):463-80
50. Miyoshi N, Uchida K, Osawa T, Nakamura Y. Selective cytotoxicity of benzyl isothiocyanate in the proliferating fibroblastoid cells. *International journal of cancer*. 2007 Feb 1;120(3):484-92.
51. Akinboro A, Bakare AA. Cytotoxic and genotoxic effects of aqueous extracts of five medicinal plants on *Allium cepa* Linn. *Journal of ethnopharmacology*. 2007 Jul 25;112(3):470-5.
52. Nayak BS, Ramdeen R, Adogwa A, Ramsuhag A, Marshall JR. Wound-healing potential of an ethanol extract of *Carica papaya* (Caricaceae) seeds. *International Wound Journal*. 2012 Dec;9(6):650-5.
53. Mishra A, Seth A, Maurya SK. Therapeutic significance and pharmacological activities of antidiarrheal medicinal plants mention in Ayurveda: A review. *Journal of intercultural ethnopharmacology*. 2016 Jun;5(3):290.
54. Prabhu AK, Devadas SM, Lobo R, Udupa P, Chawla K, Ballal M. Antidiarrheal activity and phytochemical analysis of *Carica papaya* fruit extract. *Journal of Pharmaceutical Sciences and Research*. 2017 Jul 1;9(7):1151.
55. Seo SA, Ngo HT, Hwang E, Park B, Yi TH. Protective effects of *Carica papaya* leaf against skin photodamage by blocking production of matrix metalloproteinases and collagen degradation in UVB-irradiated normal human dermal fibroblasts. *South African journal of botany*. 2020 Jul 1;131:398-405.
56. Chandrasekar R, Sivagami B. Formulation and evaluation of a poly herbal skin care cream containing neem and Tulsi. *Research Journal of Topical and Cosmetic Sciences*. 2018;9(1):25-32.



Debmalya Biswas *et al.*,

57. Mahrous NS, Noseer EA. Anticancer potential of *Carica papaya* Linn black seed extract against human colon cancer cell line: in vitro study. *BMC Complementary Medicine and Therapies*. 2023 Jul 29;23(1):271.
58. Anilkumar A, Bhanu A. In vitro anticancer activity of “Methanolic extract of papaya blackseeds” (MPB) in Hep G2 cell lines and its effect in the regulation of bcl-2, caspase-3 and p53 gene expression. *Advances in Cancer Biology-Metastasis*. 2022 Jul 1;4:100025.
59. Amazu LU, Azikiwe CC, Njoku CJ, Osuala FN, Nwosu PJ, Ajugwo AO, Enye JC. Antiinflammatory activity of the methanolic extract of the seeds of *Carica papaya* in experimental animals. *Asian Pacific Journal of Tropical Medicine*. 2010 Nov 1;3(11):884-6.
60. .Rojas-García A, Fernández-Ochoa Á, Cádiz-Gurrea MD, Arráez-Román D, Segura-Carretero A. Neuroprotective effects of agri-food by-products rich in phenolic compounds. *Nutrients*. 2023 Jan 14;15(2):449
61. Pandey S, Cabot PJ, Shaw PN, Hewavitharana AK. Anti-inflammatory and immunomodulatory properties of *Carica papaya*. *Journal of immunotoxicology*. 2016 Jul 3;13(4):590-602.
62. Shaban NZ, El-Kot SM, Awad OM, Hafez AM, Fouad GM. The antioxidant and anti-inflammatory effects of *Carica Papaya* Linn. seeds extract on CCl 4-induced liver injury in male rats. *BMC Complementary Medicine and Therapies*. 2021 Dec;21:1-5.
63. Singh SP, Kumar S, Mathan SV, Tomar MS, Singh RK, Verma PK, Kumar A, Kumar S, Singh RP, Acharya A. Therapeutic application of *Carica papaya* leaf extract in the management of human diseases. *DARU Journal of Pharmaceutical Sciences*. 2020 Dec;28:735-44.
64. H. H. Soib, H. F. Ismail, F. Husin, M. H. Abu Bakar, H. Yaakob, and M. R. Sarmidi, “Bioassay-guided different extraction techniques of *Carica papaya* (Linn.) leaves on in vitro wound-healing activities,” *Molecules*, vol. 25, no. 3, p. 517, 2020.
65. Pinelo, M., M. Rubilar, J. Sineiro and M.J. Nunez, 2004. Extraction of antioxidant phenolics from almond hulls (*Prunus amygdalus*) and pine sawdust (*Pinus pinaster*). *Food Chem.*, 85(2): 267-273
66. Shaik A, Yalavarthi P, Bannoth C. Role of anti-fertility medicinal plants on male & female reproduction. *Journal of Complementary and Alternative Medical Research*. 2017 Jan 10;3(2):1-22.
67. Munir S, Liu ZW, Tariq T, Rabail R, Kowalczewski PŁ, Lewandowicz J, Blecharczyk A, Abid M, Inam-Ur-Raheem M, Aadil RM. Delving into the therapeutic potential of *Carica papaya* leaf against thrombocytopenia. *Molecules*. 2022 Apr 25;27(9):2760.

Fig.1. Phytochemicals present in *C. papaya*



Debmalya Biswas et al.,

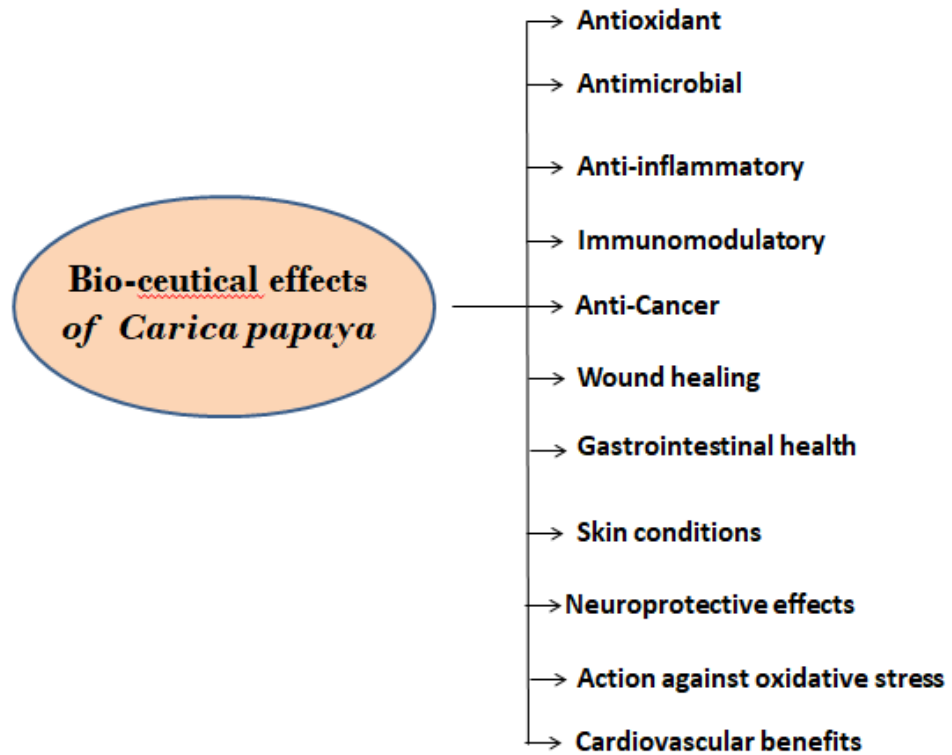


Fig.2. Bio-ceutical effects of *C. papaya*





Prevalence and Distribution of Virulence Determinants in Diarrheagenic *Escherichia coli* associated with Children Under Five Years of Age

Sweta Barupal¹, Sanjay Kochar² and Dharmesh Harwani^{3*}

¹Research Scholar, Department of Microbiology, Maharaja Ganga Singh University, Rajasthan, India

²Professor, Department of Medicine, S.P. Medical College and A.G. of Hospitals, (Affiliated to Rajasthan University of Health Sciences), Rajasthan, India.

³Assistant Professor, Department of Microbiology, Maharaja Ganga Singh University, Rajasthan, India

Received: 15 Jun 2024

Revised: 20 Aug 2024

Accepted: 19 Oct 2024

*Address for Correspondence

Dharmesh Harwani

Assistant Professor,

Department of Microbiology,

Maharaja Ganga Singh University,

Rajasthan, India.

E.Mail: dharmesh@mgsbikaner.ac.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Escherichia coli (*E.coli*) is a ubiquitous member of the human gut microbiota, existing as both commensal and pathogenic strains. The pathogenic variants of *E.coli*, known as Diarrheagenic *Escherichia coli* (DEC), pose a significant health threat to children under five globally, particularly in developing regions, where they cause childhood diarrhea. The objective of the present study was to identify pathogenic variants of *E.coli* from stool samples of children with acute diarrhea (n=144) admitted to pediatric ward of Sardar Patel Medical College (PBM Hospital) and samples from untreated community sewage sources (n=40) collected in the Bikaner district of Rajasthan, India. These samples were processed in culture-based analyses employing established laboratory protocols. For molecular characterization, both monoplex and multiplex-PCR analysis targeted 16 virulence genes associated with 6 different DEC pathotypes. These included *stx1*, *stx2*, and *hlyA* associated with Shiga toxin-producing *E. coli* (STEC), which includes the Enterohemorrhagic *E. coli* group (EHEC); *irp2*, *aaiA*, *pic*, *aafA*, *aggR*, and *pCVD432* linked to Enteropathogenic *E. coli* (EACE); *bfpA* and *eae* associated with Enteropathogenic *E. coli* (EPEC); *st*, *lt*, and *est* associated with Enterotoxigenic *E. coli* (ETEC); *ipaH* associated with Enteroinvasive *E. coli* (EIEC); and *daaE* associated with Diffusely Adherent *E. coli* (DAEC). Among the 90 presumptive *E. coli* samples [66 (45.8%) from stool and 24 (60%) from sewage], 58 samples harboured one or more virulence genes associated with specific DEC pathotype. The most common pathotype was observed to be enteroaggregative EAEC, which accounted for 35.7% of cases in children under five and 43.7% of cases in community sewage samples followed by ETEC, and EHEC. The research underscores the widespread





Sweta Barupal et al.,

presence of the DEC pathotypes of across various sources, offering crucial insights into their detection, prevalence, and epidemiological patterns within the Bikaner city of Rajasthan in India.

Keywords: Childhood-diarrhea, DEC Pathotypes, Virulent genes, Multiplex-PCR.

INTRODUCTION

Escherichia coli is a commensal bacterium that commonly inhabits the human gastrointestinal tract. However, certain pathogenic strains can cause a wide range of diseases, including enteric and diarrheal illnesses [1-4]. Diarrheal diseases are a significant public health concern, particularly in developing countries, and are caused by various pathogens such as bacteria (e.g., *E. coli* and *Salmonella* spp.), viruses (e.g., rotavirus and norovirus), and parasites (e.g., *Giardia lamblia*) [3,5]. *E. coli* is the most important etiological agent of childhood diarrheal diseases globally, contributing substantially to morbidity and mortality in infants and young children [3,5]. Pathogenic *Escherichia coli* (*E. coli*) infections are estimated to cause approximately 2 million deaths globally each year, and the majority of these deaths, approximately 90%, occur in children under the age of five [6]. Diarrheagenic *E. coli* (DEC) strains are classified based on clinical features, epidemiological characteristics, and the presence of pathogenicity-specific virulence factors. DEC can be categorized into six main pathotypes which include Enteropathogenic *E. coli* (EPEC), Shiga toxin-producing *E. coli* (STEC), which encompasses the enterohemorrhagic *E. coli* group (EHEC), Enterotoxigenic *E. coli* (ETEC), Enteroaggregative *E. coli* (EAEC), Enteroinvasive *E. coli* (EIEC), and Diffusely adherent *E. coli* (DAEC) [1,3,4]. The latter EPEC is subdivided into typical EPEC (tEPEC) and atypical EPEC (aEPEC). Each pathotype exhibits distinct geographical distributions and possesses specific virulence markers. Pathogenic strains of *E. coli* can be classified based on genetic sub-structures, which exhibit distinct phylogenetic relationships. These phylogenetic groups (A, B1, B2, and D), known as phylogroups, are characterized by distinct phenotypic and genotypic traits [7]. Pathogenic *E. coli* strains possess virulence factors encoded by specific genes located on chromosomes or mobile genetic elements like plasmids and transposons [8,9]. Diarrheagenic *E. coli* (DEC) pathotypes have distinct virulence factor profiles encoded by specific gene clusters. These pathogenic genes encode activities like adhesion, invasion, attachment, iron acquisition, motility, and toxin production. The four major virulence classes are colonization, fitness, toxins, and effectors [7]. Pathogenic strains of *E. coli* are responsible for various diarrheal diseases, each characterized by distinct virulence factors and clinical implications. For example, EAEC is a significant emerging pathogen associated with persistent diarrhea in children in developing countries [10]. Its main characteristic is an aggregative adherence pattern on cultured epithelial cells, mediated by the production of fimbrial colonization factors known as aggregative adherence factors (AAFs) [11]. While ETEC produces heat-labile (LT) and heat-stable (ST) enterotoxins, which cause watery diarrhea [2,3]. EPEC harbors the locus of enterocyte effacement (LEE) pathogenicity island, which is responsible for the attaching and effacing (A/E) phenotype on host enterocytes. EPEC can be classified as 'typical' or 'atypical' based on the presence or absence of the EPEC adherence factor (EAF) plasmid, which comprises the cluster of genes encoding the bundle-forming pilus (*bfpA*). EHEC, also belonging to Shiga toxin-producing *E. coli* (STEC), produces Shiga toxins (Stx1 and Stx2) and can cause large outbreaks. DAEC harbors the dispersin-encoding gene, *daaE*, which facilitates adherence to intestinal epithelial cells [2]. EIEC shares many characteristics with *Shigella* species. A critical virulence factor associated with EIEC is the invasion plasmid antigen H (*ipaH*) gene, which plays a central role in the invasion process. The *ipaH* gene encodes proteins that facilitate the invasion of intestinal epithelial cells [2].

Identification of *Escherichia coli* often relies on traditional laboratory culture methods. However, for characterizing Diarrheagenic *E. coli* (DEC) pathotypes, various molecular methods are employed to detect virulence genes associated with different categories of DEC. These methods include Polymerase Chain Reaction (PCR), DNA hybridization, Multi-Locus Sequence Typing (MLST), ribotyping, and 16S rRNA gene sequencing [12-14]. Among cutting-edge methods, multiplex-PCR assays are particularly effective in detecting multiple virulence factors simultaneously. This technique involves designing primer sets to target specific DNA sequences associated with





Sweta Barupal *et al.*,

virulence factors, allowing for simultaneous amplification of multiple genes in a single reaction. The present study explores the characterization of DEC strains responsible for diarrheal diseases, focusing on their unique virulence factors and clinical implications. It delves into each pathotypes molecular signatures and clinical relevance in diarrheal illnesses, highlighting the role of multiplex PCR assays in identifying DEC pathogenicity. This study emphasizes the need for surveillance and control measures to address diarrheal diseases, especially in children under five years of age. The study enhances understanding of DEC prevalence, distribution, and genetic traits among stool samples of children with acute diarrhea admitted to pediatric ward of Sardar Patel Medical College (PBM Hospital) and samples from untreated community sewage sources collected in Bikaner, Rajasthan, the northwest region of India. This underscores the importance of ongoing research and public health interventions to combat DEC-related childhood diarrheal illnesses in the country.

MATERIAL AND METHODS

Study population and sampling sites

In this study, a total of 144 stool samples were collected from pediatric outpatients aged 0-60 months suffering from acute diarrhea, who were hospitalized in the pediatric ward of Sardar Patel Medical College (PBM Hospital). This cross-sectional study was conducted from September 2022 to March 2023. The sample size determination was based on the number of children under five years of age admitted to the pediatric ward of SPM medical college during the same period in the previous year (September 2021 to March 2022). Case selection was made by the following inclusion criteria: children under 60 months of age suffering from diarrhea and not having been administered antibiotics. Based on guardian information, children with acute diarrhea were enrolled in the study. The study included children under 5 years of age with acute community-acquired diarrhea, defined as the passage of 3 or more watery and loose stools in 24 hours, with or without clinical symptoms of an enteric ailment such as nausea, vomiting, abdominal pain or cramps, dehydration, fecal urgency, or dysentery. The study population was limited to children of Indian origin residing in the Bikaner city of Rajasthan. Information on acute diarrheal symptoms, including the number of loose/watery stools, presence of bloody or mucoid stools, vomiting, and fever $\geq 39^{\circ}\text{C}$, was collected through parental discussion and medical record review. Other clinical features and risk factors recorded included nausea, abdominal pain, duration of diarrhea, age group, feeding type, and drug use. Based on the WHO classification of dehydration, children with acute diarrhea were categorized as having mild diarrhea (no dehydration) or moderate diarrhea (mild to moderate dehydration). Severe dehydration was considered a medical emergency requiring immediate treatment. The collection of sewage samples, particularly untreated community sewage samples, was conducted from various regions of Bikaner, including both rural and urban areas. A total of 40 samples were collected over four months, from March 2022 to June 2022. The ease of sample collection was facilitated by the relatively shallow depth of the sites. However, sites with deeper depths posed a challenge, and poles were used to collect samples while adhering to safety protocols to prevent infection.

Specimen collection and transportation

The stool and sewage samples were collected in sterile, wide-mouthed, leak-proof containers with tightly fitting lids, labelled with patients' names, ages, genders, and collection months, and stored at the hospital's microbiology laboratory. Subsequently, all samples were transferred to the Microbiology department of Maharaja Ganga Singh University in Bikaner (MGSU), Rajasthan for further processing within 24 hours of receipt.

Isolation and identification of *E. coli*

The stool and sewage samples were initially inoculated into a selective enrichment medium, lauryl sulphatetryptose broth, at a 1:10 ratio. The inoculated broth was then incubated overnight at 37°C . Furthermore, the samples were sub-cultured onto MacConkey agar and eosin-methylene blue (EMB) agar plates, which were incubated at 37°C for 18-24 hours. MacConkey agar and EMB agar serve as selective and differential media for the isolation and identification of coliform bacteria and *E. coli*. On MacConkey agar, *E. coli* colonies appear as pink to red colonies due to lactose fermentation. EMB agar is more selective, and *E. coli* colonies typically exhibit a characteristic metallic green sheen





Sweta Barupal et al.,

with a dark centre. The biochemical tests to characterize *E. coli* were performed [15]. First Gram staining was performed to confirm *E. coli*, *E. coli* is a Gram-negative bacterium, which means it will stain red or pink due to its thin peptidoglycan layer and outer lipid membrane. The IMViC test, a set of four biochemical tests, is commonly used to differentiate *E. coli* from other Enterobacteriaceae. The isolates in this study exhibited positive results for the indole and methyl red tests, while the Voges-Proskauer, citrate, and urease tests produced negative findings [16,17].

Molecular characterization of diarrheagenic *E. coli*

Genomic DNA was extracted from presumptive *E. coli* to identify DEC specific virulence genes. The process began by suspending each *E. coli* isolate in LB medium separately and after incubation (0.5 optical density at 600nm), centrifuging at 6000 rpm for 10 minutes. The supernatant was discarded, and the pellet was used for DNA extraction. The bacterial cells underwent lysis using a chemical combination which include SET buffer (150 mM NaCl, 1 mM Tris/HCl pH 8.0, and 0.5 mM EDTA), 10% sodium dodecyl sulfate (100 µl), and proteinase K (5 µl), with an incubation period at room temperature for 1 hour. Phenol was introduced to separate the nucleic acid from other cellular substances, followed by centrifugation at 12,000 rpm for 15 minutes. The upper layer was extracted by adding an equal volume of chloroform-isoamyl alcohol (24:1) solution and undergoing centrifugation again at 12,000 rpm for 15 minutes. Afterwards, the supernatant was mixed with an equal volume of ice-cold isopropyl alcohol and centrifuged at 10,000 rpm for 10 minutes, precipitating the DNA with ethanol. The DNA precipitate was then resuspended in 50 µl of Tris-EDTA (TE) buffer. The quality of the bacterial DNA was assessed by electrophoresis on a 1.5% agarose gel stained with ethidium bromide, using 1 × Tris-borate-EDTA (TBE) buffer at 100 V for 45 minutes, followed by examination of the results under a UV transilluminator. This ensured the extraction and purification of high-quality genomic DNA, subsequently utilized for PCR amplification to identify DEC pathotypes.

Monoplex and Multiplex-Polymerase chain reaction (PCR)

DNA templates were subjected to monoplex and multiplex PCR with specific primers (Table 1), for the identification and detection of different DEC pathotypes such as EPEC, ETEC, EHEC, EAEC, EIEC, and DAEC. Monoplex and multiplex PCR primer protocols were employed to acquire, analyse, and evaluate the primer sequences' specificity, sensitivity, and size. The PCR primers were synthesized by Eurofins (Eurofins Genomics India Pvt. Ltd.). Sixteen different target genes were used to detect DEC by monoplex and multiplex PCR include *irp2*, *aaiA*, *pic*, *aafA*, *aggR* and *pCVD432* (EAEC); *bfpA*, *eae* (EPEC); *stx1*, *stx2*, and *hlyA* (EHEC); *st*, *lt*, and *est* (ETEC); *ipaH* (EIEC), and *daaE* (DAEC) genes as indicated in Table 1. These virulence genes are crucial determinants to identify DEC pathotypes [18,19]. The multiplex PCR was performed with a 50 µl reaction mixture containing 2 µl of 50ng DNA template and 5 µl of 10x PCR buffer, 2 µl of 25mM MgCl₂, 5 µl of a 2.5mM mixture of deoxy nucleoside triphosphate (dNTP), 1 µl of 2U/µl of Taq DNA polymerase, 2 µl of 10µM concentration of each forward and reverse primers, with the remaining volume supplemented by nuclease-free water. The conditions for the thermal cycling for multiplex PCR were kept as indicated in Table 2. Standardising the multiplex PCR assay included optimising PCR cycling conditions and primer selection to ensure efficient and specific detection of specific DEC pathotypes. The following three multiplex PCR assays were designed to detect specific DEC pathotypes:

Multiplex PCR I:

- EPEC targeting the *eae* gene.
- EHEC targeting the *stx1* gene.
- ETEC targeting the *lt* gene.
- EAEC targeting the *aggR* gene.

Multiplex PCR II:

- EPEC targeting the *bfpA* gene.
- EHEC targeting the *stx2* and *hlyA* genes.
- ETEC targeting the *st* and *est* genes.
- EAEC targeting the *irp2*, and *aaiA* genes.





Sweta Barupal et al.,

Multiplex PCR III:

- EIEC targeting the *ipaH* gene.
- DAEC targeting the *daaE* genes.
- EAEC targeting the *pic*, *aafA*, and *pCVD432* genes.
-

The monoplex-PCR was also performed with a 50µl reaction mixture containing 2µl of DNA template and 5µl of 10x PCR buffer, 2µl of 25mM MgCl₂, 5µl of a 2.5mM mixture of deoxy nucleoside triphosphate (dNTP), 1µl of 2U/µl of Taq DNA polymerase, 2µl of 10µM concentration of each forward and reverse primers, with the remaining volume supplemented by nuclease-free water. The monoplex PCR thermocycling conditions were kept as initial denaturation at 95°C for 2 minutes, 95°C for 30 seconds, annealing at 50° for 1 minute, and extension for 72°C for 1.30 minutes, followed by 30 cycles, with the final 10-minute extension at 72°C, the reaction was run in the thermocycler PCR system. The amplified PCR products were evaluated by running 5µl on a 2% (wt/vol) agarose gel along with a 100bp DNA ladder as a molecular marker. Ethidium bromide was used to stain the gel, and the DNA bands were visualized under UV light.

Quality Control

A positive control and a negative control were included in PCR assays. The positive controls to detect DEC pathotype include: ATCC 35401 to detect ETEC, ATCC 33780 to detect EAEC, ATCC 43893 to detect EIEC, ATCC 35150 to detect EHEC, ATCC 43887 to detect EPEC. Additionally, DH5α used as a negative control.

Ethical Approval

The present research study and its protocol were approved by the Institutional Ethics Committee (IEC) of the Maharaja Ganga Singh University of Bikaner. Data collection and analysis were conducted at the PBM Hospital of Sardar Patel Medical College, Bikaner, Rajasthan. The IEC thoroughly reviewed and endorsed the research protocol to ensure its adherence to ethical standards and regulatory requirements.

RESULTS

In the present study, a total of 184 samples were collected, comprising 144 stool samples from children under five years of age experiencing acute diarrhea and 40 samples from untreated community sewage. The samples underwent culture-based analyses employing established laboratory protocols, including the assessment of morphological features on MacConkey agar and Eosin Methylene Blue (EMB) agar, alongside IMViC (Indole, Methyl red, Voges-Proskauer, Citrate utilization) biochemical tests (positive or negative). The identification of presumptive *E. coli* isolates from the samples yielded an overall positivity rate of 90 (48.91%). Of the positive samples, 66 (45.83%) were obtained from stool samples, while 24 (60%) were derived from sewage sources. The male-to-female ratio among the total sampled population (stool samples, n=144) was approximately 80 (55.55%) to 64 (44.44%), respectively. Children enrolled in the study encompassed diverse age groups, with 77 (53.47%) up to 24 months, 38 (26.38%) aged between 25 and 48 months, and 29 (20.13%) up to 60 months of age (Table 3). Furthermore, the dietary practices of the participating children were delineated, revealing that 18% were exclusively breastfed, 45.8% received a combination of breast milk and formula milk, and 36.1% were fed with solid foods (Table 3). In children with diarrhea (n=144), the clinical symptoms were mainly defined by variations in stool consistency. The present study indicates that diarrhea in children primarily led to watery stools (43%) and loose stools (29.16%), while a smaller percentage presented with bloody stools (11.11%) or mucoid stools (16.6%) (Table 3). In contrast, *E. coli* prevalence in children with diarrhea, indicated a little male predominance, with 54.5% of positive cases being male and 45.4% female, which is similar to the distribution in negative cases (56.4% male and 43.5% female) (Table 3). In terms of age, the proportion of positive cases is highest in children between the ages of 13 and 24 months (30.3%), followed by 0 to 12 months (24.2%) and 49 to 60 months (21.2%). Children between the ages of 37 and 48 months have the lowest positive rates (9%). In terms of feed type, children who were solely breastfed had the lowest percentage of positive cases (9%), while those who were fed solid food had the largest percentage (48.4%). Additionally, there was variation





Sweta Barupal et al.,

in the type of stool; among positive cases, watery stools were the most prevalent (48.4%), followed by loose stools (27.2%) and mucoid stools (21.2%). In comparison to negative instances (17.9%), bloody stools were much less common in positive cases (3%) than in negative ones. These findings provide insights into the demographic and nutritional characteristics of the studied population, contributing to a comprehensive understanding of diarrheal disease epidemiology and its associated factors.

Sewage samples were systematically collected from both rural and urban regions within the Bikaner regions depicted in Table 4. The sampling strategy aimed for inclusivity by encompassing a variety of localities, including slum settlements and affluent residential areas. Geographical categorization facilitated the classification of samples according to their urban or rural origins, enabling a detailed examination of potential microbial contamination across diverse environmental settings. This meticulous sampling approach sought to comprehensively capture the range of sewage characteristics prevalent in the study area, thereby augmenting the reliability and relevance of the study findings. Through a comparative analysis, the prevalence of *E. coli* in various rural and urban community sewage samples was observed. The villages of Udairamsar, Palana, Chak Chani, and Barsnghsar each observed with two positive samples out of the 24 positive samples; these villages accounted for 8.3% of the total positive samples. Several settlements, including Husangsar, Kanasar, Madh, Bachhasar, Kotri, and Kolayat, each observed with one positive sample (4.1%). In rural areas, only one negative sample (6.2%) was observed by each of Bachhasar, Kotri, Kolayat, Madh, Husangsar, and Kanasar. However, the pattern was marginally different in urban areas. Nagnechi and Choukhuti each observed with two positive samples (8.3%) out of the 24 positive samples. A single positive sample (4.1%) was observed by each of the following areas: Satellite, Kothari, Kuchilpura, Railway Colony, Dharnidhar, and Police-line. There were no positive samples in Bhutto ka Chauraha or Pugal-phanta, but there were two negative samples (12.5%) in each. There was one negative sample (6.2%) for each of Satellite, Kothari, Kuchilpura, Railway Colony, Dharnidhar, and Police-line.

To detect various virulence genes, 90 presumptive *E. coli* isolates underwent PCR screening using specific primers designed to target different 16 genes associated with DEC (Table 1) (Fig 2). These virulence genes included *irp2*, *aggR*, *aaiA*, *aafA*, *pic*, *pCVD432*, *bfpA*, *eae*, *stx1*, *stx2*, *hlyA*, *lt*, *st*, *est*, *daaE*, and *ipaH*. DEC were categorized based on the presence or absence of these specific virulence genes in the stool samples collected from diarrheic children and from sewage samples (Fig.2). Among the overall 90 samples, 58 (64.4%) were positive for at least one of the targeted virulence genes of *E. coli*. (Table 5). Of the 66 presumptive *E. coli* from stool samples, 42 (63.6%) exhibited the presence of one or more virulence genes. The gene *irp2* exhibited the highest frequency at 26.1%, whereas the genes *aaiA*, *est*, and *pic* had the lowest occurrence, each representing 2.3% of the isolates (Table 5). 36.4 % of *E. coli* isolates were found non-pathogenic, based on the absence of all investigated virulence genes. Among 24 presumptive *E. coli* from untreated community sewage, 16 (66.6%) were observed with at least one of the targeted virulence genes. The predominant gene detected was *irp2* (31.2%), identified in sewage samples, The most common gene was the *irp2* similar to the diarrheal children group. Only 6.2% of the sample was positive for the *aafA*, *cvd432*, *stx1*, and *stx2* gene. In contrast, 33.4% of *E. coli* isolates were observed to be non-pathogenic, based on the absence of any investigated gene. None of the samples tested positive for the *aggR* and *eae* genes in both stool and sewage samples (Table 5).

For characterization of typical Enteropathogenic *E. coli* (EPEC), both *bfpA* (bundle-forming pilus) and *eae* (intimin) genes are needed to be present. However, the present study observed, only *bfpA* gene characterizing these DEC pathotype as atypical EPEC. Similarly, in the present study, the presence of *stx1*, *stx2*, and *hlyA* genes characterized Enterohemorrhagic *E. coli* (EHEC) strains while the presence of *lt*, *st* and *est* genes characterized Enterotoxigenic *E. coli* (ETEC) strains. Enteroaggregative *E. coli* (EAEC) strains were characterized by *irp2*, *aaiA*, *aafA*, *pic*, and *pCVD432* genes, while Enteroinvasive *E. coli* (EIEC) strains were identified by the presence of the *ipaH* gene, and diffuse adherent *E. coli* (DAEC) strains were identified by the presence of the *daaE* gene.

In this way, a total of 58 presumptive *E. coli* was categorized based on the detection of at least one targeted virulence gene associated with DEC. Notable variations were observed in the distribution of these virulence genes among *E. coli* isolates obtained from stool and sewage samples. The most prevalent genes were observed to be *irp2*, present in





Sweta Barupal *et al.*,

27.5% of the samples, and *st*, observed in 20.6% of the samples, followed by *stx1* (8.6%), *stx2* (6.8%), and *hlyA* (6.8%). Additionally, *lt* (6.8%), *aafA* (5.1%), and *daaE* (5.1%) were also identified. Less frequently observed genes included *bfpA* (3.4%) and several others, such as *aaiA*, *pic*, *pCVD432*, *est*, and *ipaH*, which were detected in 1.7% of the total samples (Fig. 1). In our analysis of total 90 presumptive *E. coli*, a total of 32 (35.5%) isolates exhibited an absence of all tested virulence genes. These observations underscore the intricate nature and variability of DEC pathotypes, emphasising the necessity of evaluating the collective presence of virulence factors to comprehend their pathogenicity effectively. The present study observed that DEC pathotypes in stool samples was more frequent in males (61.9%) than in females (38%) (Table 6). The age-wise distributions of DEC pathotypes were not consistent (Table 6). DEC pathotypes EAEC followed by EHEC and ETEC were observed to be more prevalent in all age groups (Table 6). The age-wise prevalence of the virulence gene was 23.8% in 0-12 months, 16.6% in 13-24 months, 33.4% in 25-36 months, 14.2% in 37-38 months, and 11.9% up to 60 months of age, respectively. Regarding the children's clinical symptoms, watery and loose diarrhea (42.8%) and (40.4) respectively were the most common diarrheal consistency. Concerning the feed types, 42.8% and 35.7% of DEC pathotypes were detected from combined breast or formula milk, and solid feed respectively (Table 6).

The predominant DEC pathotypes among the stool samples collected from children with diarrhea were found to be Enterotoxigenic *E. coli* (ETEC), and Enteropathogenic *E. coli* (EPEC), with virulence genes detected namely *irp2* (26.1%), *aafA* (4.7%), *aaiA* (2.3%), *pic* (2.3%), and *st* (21.4%), *lt* (4.7%), *est* (2.3%) respectively. Followed by DEC pathotype characterized as Enterohemorrhagic *E. coli* (EHEC) harbouring *bfpA* (4.7%), Enteroinvasive *E. coli* (EIEC) [*ipaH* (2.3%)]. In community sewage samples, the predominant DEC pathotypes were characterized as EAEC [*irp2* (31.2%), *aafA* (6.2%), *pCVD432* (6.2%)], followed by ETEC [*st* (18.7%), *lt* (12.5%)], EHEC [*stx1* (6.2%), *stx2* (6.2%), and *hlyA* (12.5%)].

In contrast, the collective prevalence of DEC within untreated community sewage was determined to be 66.6%. Notably, in sewage samples from both urban and rural areas, the *irp2* gene had a prevalence of 31.2% (Table 7). The *pCVD432* (6.2%) gene linked to EAEC strains was observed to be exclusively present in sewage samples (Table 7). EAEC emerged as the predominant pathotype identified in both stool and sewage samples. Upon broader geographical analysis within the Bikaner region, the overall positivity rate of virulence genes associated with DEC pathotypes in the untreated community sewage was found to be 68.7% in rural regions and 31.25% in urban areas, respectively.

The present study presents the first comprehensive report on the prevalence of six diarrheagenic *Escherichia coli* strains in the Bikaner region of Rajasthan, India. EAEC followed by ETEC and EHEC were identified as the most predominant pathotypes among children under five years of age and in the untreated community sewage samples.

DISCUSSION

In India, diarrhea continues to be the primary cause of morbidity and mortality among children under the age of five, resulting in an estimated 2.5 billion deaths annually. Within this statistic, diarrhoeagenic *Escherichia coli* (DEC) is attributed to 30%-40% of cases, representing a significant proportion of diarrhoeal illnesses globally, particularly prevalent in developing nations [35-37]. DEC strains have exhibited diverse virulence traits along with instances of antibiotic resistance, presenting notable concerns within the realm of study. In the present study various phenotypic, biochemical, and molecular assays have been employed for the identification and characterization of DEC pathotypes. For the comprehensive identification of DEC pathotypes, *E. coli* isolates confirmed through biochemical methods were selectively targeted for characterization via monoplex and multiplex PCR assays aimed at identifying their pathotype-specific virulence genes. These assays were tailored to target distinct virulence genes linked with each pathotype, facilitating the simultaneous detection of multiple pathotypes within a single reaction [38-46]. Regional disparities in the prevalence of various pathotypes of DEC are extensively documented [37]. In our study,





Sweta Barupal et al.,

we gathered a total of 184 samples from both stool (from children with diarrhea admitted to pediatric ward) and sewage sources. Among these, 45.8% and 60% of the samples, respectively, were identified as presumptive *E. coli* using culture techniques and biochemical tests. This aligns with the findings from previous research. For instance, Ballal et al. [47] identified *E. coli* as the predominant pathogen in 46.3% of diarrheal patients. Similarly, a study conducted in Orissa, India, by Samal et al. [48], indicated that *E. coli* accounted for 75.5% of bacterial enteropathogens among hospitalized patients, with 13.3% being pathogenic strains. In our investigation, we found that 48.9% of the samples contained *E. coli*, with at least frequency of one isolate in 64.4% of the collected samples.

Using PCR assays, our study reveals a higher prevalence of EAEC and ETEC, followed by EHEC, EPEC, DAEC, and EIEC among children with diarrhea and in communities across Bikaner. Out of 66 stool samples, 42 tested positives for DEC wherein EAEC were the most common pathotype, detected in 35.7% of cases, followed by ETEC (28.5%), EHEC (21.4%), EPEC (4.7%), DAEC (7.1%), and EIEC (2.3%). Among 24 sewage samples, 16 were tested positives for DEC pathotypes. EAEC was detected in 43.7% of samples, while ETEC and EHEC were observed in 31.2% and 25% of samples respectively. A study conducted in South India, which is consistent with our findings, identified EAEC as the most prevalent pathotype among children with diarrhea (14.7%), followed by ETEC and EPEC [49]. The present results also consistent with the previous study [50], wherein 50% of cases of persistent diarrhea were attributed to EAEC infection in India.

In the present study, the enrolled patients exhibited a uniform distribution of sexes. Among the study subjects, there were 80 (55.55%) male and 64 (44.44%) female. The data pertaining to children with diarrhea were further categorized into three age groups wherein group A comprised 77 (53.47%) individuals up to 24 months, group B included 38 (26.38%) individuals aged between 25 and 48 months, and group C encompassed over 29 (20.13%) individuals up to 60 months of age. The distribution of six DEC pathotypes namely EAEC, ETEC, EHEC, EPEC, DAEC, and EIEC was examined across these age groups. Remarkably, EAEC followed by ETEC and EHEC pathotypes were observed to be most dominant, while EPEC, DAEC, and EIEC pathotypes were found to be relatively less frequent.

A total of 22 (37.9%) enteroaggregative *E. coli* (EAEC) were detected in both stool and sewage samples. EAEC, the most recently identified DEC, is known to cause travelers' diarrhea and is the second most common type found in both developed and developing countries [51]. The presence of various serotypes, antibiograms, and virulence gene profiles among the isolates underscores the high genetic diversity of EAEC strains [29]. Enterotoxigenic *E. coli* (ETEC) strains hold significance as the predominant strain among pediatric patients, particularly those under five years of age [52]. In our study, ETEC was detected in a total of 17 samples (29.3%). However, findings from a previous study reported a higher prevalence of ETEC among children under 12 months compared to those over 12 months [53]. The children under 2 years old were predominantly affected by EPEC infections [54]. Concurrently, Enteropathogenic *E. coli* (EPEC) accounted for 3.4% of diarrheal disease observed in the present study. Considering the present observation, the findings are noteworthy in two studies: one from northern India (Kashmir) claiming a prevalence of 7.6% [55] and the other from southern India (Mangalore) indicating an EPEC prevalence of 11% [56]. Additionally, the prevalence of Enteroinvasive *E. coli* (EIEC) in stool samples was 1.7%. In comparison, Diffuse adherent *E. coli* (DAEC) which was observed in 5.1% of cases.

CONCLUSION

The present study revealed a significant prevalence of diarrheagenic *E. coli* pathotypes, particularly EAEC followed by ETEC and EHEC in the Bikaner region of Rajasthan, India with a notably higher incidence among children under five. The detection rates of other DEC pathotypes, such as EPEC, EIEC, and DAEC, were relatively lower than those of EAEC, ETEC, and EHEC. This study represents the first report on the molecular epidemiology of DEC pathotypes in the Bikaner region. The high prevalence of EAEC followed by ETEC, and EHEC infections in children with diarrhea under five highlights the need for targeted interventions and improved sanitation to reduce the burden of





Sweta Barupal et al.,

diarrheal diseases in this age group. The findings also emphasise the importance of molecular methods in accurately identifying DEC pathotypes, as traditional culture-based techniques may not always be sufficient. The use of monoplex and multiplex PCR assays targeting specific virulence genes can enhance the detection of DEC pathotypes and guide appropriate treatment strategies. This study provides valuable insights into the epidemiology of DEC infections in Bikaner and underscores the need for continued surveillance and research to develop effective prevention and control measures against diarrheal diseases in children.

ACKNOWLEDGMENTS

Dr. Dharmesh Harwani (PI) and Sweta Barupal (RA) acknowledges the Indian Council of Medical Research (ICMR), Government of India for sanctioning an Adhoc Research Project (5/7/1718/CH/Adhoc/2020-RBMH) entitled “Prevalence, Detection, Characterization of Multidrug-Resistant Diarrheagenic *Escherichia Coli* pathotypes and serotypes in Under-Five Children in Bikaner Region of Rajasthan”.

REFERENCES

- Ghai, O. P., Menon, P. S. N., Bhau, M. K. (1980). Pathogenesis of diarrhea due to *Escherichia coli*. *The Indian Journal of Pediatrics*, 47, 311-316. <https://doi.org/10.1007/BF02831325>
- Kaper, J. B., Nataro, J. P., Mobley, H. L. (2004). Pathogenic *Escherichia coli*. *Nature reviews microbiology*, 2(2), 123-140. <https://doi.org/10.1038/nrmicro818>
- Jafari, A., Aslani, M. M., Bouzari, S. (2012). *Escherichia coli*: a brief review of diarrheagenic pathotypes and their role in diarrheal diseases in Iran. *Iranian journal of microbiology*, 4(3), 102. PMID: 23066484.
- Gomes, T.A., Elias, W.P., Scaletsky, I.C., Guth, B.E., Rodrigues, J.F., Piazza, R.M., Ferreira, L., Martinez, M.B. (2016). Diarrheagenic *Escherichia coli*. *Brazilian Journal of Microbiology*, 47, 3-30. <https://doi.org/10.1016/j.bjm.2016.10.015>
- Kaur, P., Dudeja, P. K. (2023). Pathophysiology of Enteropathogenic *Escherichia coli*-induced Diarrhea. *Newborn (Clarksville, Md.)*, 2(1), 102. doi: 10.5005/jp-journals-11002-0056
- Jang, J., Hur, H. G., Sadowsky, M. J., Byappanahalli, M. N., Yan, T., Ishii, S. (2017). Environmental *Escherichia coli*: ecology and public health implications-a review. *Journal of applied microbiology*, 123(3), 570-581. <https://doi.org/10.1111/jam.13468>
- Tenaillon, O., Skurnik, D., Picard, B., Denamur, E. (2010). The population genetics of commensal *Escherichia coli*. *Nature reviews microbiology*, 8(3), 207-217. <https://doi.org/10.1038/nrmicro2298>
- Wu, H. J., Wang, A. H., Jennings, M. P. (2008). Discovery of virulence factors of pathogenic bacteria. *Current opinion in chemical biology*, 12(1), 93-101. <https://doi.org/10.1016/j.cbpa.2008.01.023>
- Alegbeleye, O. O., Singleton, I., Sant'Ana, A. S. (2018). Sources and contamination routes of microbial pathogens to fresh produce during field cultivation: A review. *Food microbiology*, 73, 177-208. <https://doi.org/10.1016/j.fm.2018.01.003>
- Mulu, B. M., Belete, M. A., Demlie, T. B., Tassew, H., Sisay Tessema, T. (2024). Characteristics of pathogenic *Escherichia coli* associated with diarrhea in children under five years in northwestern Ethiopia. *Tropical Medicine and Infectious Disease*, 9(3), 65. <https://doi.org/10.3390/tropicalmed9030065>
- Harrington, S. M., Dudley, E. G., Nataro, J. P. (2006). Pathogenesis of enteroaggregative *Escherichia coli* infection. *FEMS microbiology letters*, 254(1), 12-18. <https://doi.org/10.1111/j.1574-6968.2005.00005.x>
- Azim, K. F., Somana, S. R., Hasan, M. K., Foysal, M. J., Ali, M. H., Chowdhury, T. A., Hossain, M. N. (2021, December). Molecular detection and analysis of virulence genes in multi-drug resistant *Escherichia coli* from infected broilers. In *Veterinary Research Forum* (Vol. 12, No. 4, p. 505). Faculty of Veterinary Medicine, Urmia University, Urmia, Iran. <https://doi.org/10.30466/vrf.2021.123141>.
- Bekal, S., Brousseau, R., Masson, L., Prefontaine, G., Fairbrother, J., Harel, J. (2003). Rapid identification of *Escherichia coli* pathotypes by virulence gene detection with DNA microarrays. *Journal of clinical microbiology*, 41(5), 2113-2125. <https://doi.org/10.1128/jcm.41.5.2113-2125.2003>





Sweta Barupal et al.,

14. Luo, S., Liao, C., Peng, J., Tao, S., Zhang, T., Dai, Y., Ding, Y., Ma, Y. (2023). Resistance and virulence gene analysis and molecular typing of *Escherichia coli* from duck farms in Zhanjiang, China. *Frontiers in Cellular and Infection Microbiology*, 13, 1202013. <https://doi.org/10.3389/fcimb.2023.1202013>
15. Trepeta, R. W., and Edberg, S. C. (1984). Methylumbelliferyl-beta-D-glucuronide-based medium for rapid isolation and identification of *Escherichia coli*. *Journal of Clinical Microbiology*, 19(2), 172-174. <https://doi.org/10.1128/jcm.19.2.172-174.1984>
16. MacWilliams, M. P. (2009). Citrate test protocol. *American Society for Microbiology*, 1-7.
17. McDevitt, S. (2009). Methyl red and voges-proskauer test protocols. *American Society for Microbiology*, 8.
18. Nataro, J.P., Mai, V., Johnson, J., Blackwelder, W.C., Heimer, R., Tirrell, S., Edberg, S.C., Braden, C.R., Morris Jr, J.G., Hirshon, J.M. (2006). Diarrheagenic *Escherichia coli* infection in Baltimore, Maryland, and New Haven, Connecticut. *Clinical infectious diseases*, 43(4), 402-407. <https://doi.org/10.1086/505867>
19. Bebrone, C. (2007). Metallo- β -lactamases (classification, activity, genetic organization, structure, zinc coordination) and their superfamily. *Biochemical pharmacology*, 74(12), 1686-1701. <https://doi.org/10.1016/j.bcp.2007.05.021>
20. Yatsuyanagi, J., Saito, S., Sato, H., Miyajima, Y., Amano, K. I., Enomoto, K. (2002). Characterization of enteropathogenic and enteroaggregative *Escherichia coli* isolated from diarrheal outbreaks. *Journal of clinical microbiology*, 40(1), 294-297. <https://doi.org/10.1128/jcm.40.1.294-297.2002>
21. Dhaka, P., Vijay, D., Vergis, J., Negi, M., Kumar, M., Mohan, V., Doijad, S., Poharkar, K.V., Malik, S.S., Barbuddhe, S.B., Rawool, D.B. (2016). Genetic diversity and antibiogram profile of diarrhoeagenic *Escherichia coli* pathotypes isolated from human, animal, foods and associated environmental sources. *Infection ecology & epidemiology*, 6(1), 31055. <https://doi.org/10.3402/iee.v6.31055>
22. Nishikawa, Y., Zhou, Z., Hase, A., Ogasawara, J., Kitase, T., Abe, N., Nakamura, H., Wada, T., Ishii, E., Haruki, K. (2002). Diarrheagenic *Escherichia coli* isolated from stools of sporadic cases of diarrheal illness in Osaka City, Japan between 1997 and 2000: prevalence of enteroaggregative *E. coli* heat-stable enterotoxin 1 gene-possessing *E. coli*. *Japanese journal of infectious diseases*, 55(6), 183-190.
23. Itoh, F. (1992). Differentiation and detection of pathogenic determinants among diarrheagenic *Escherichiacoli* by polymerase chain reaction using mixed primers. *Jpn. J. Clin. Med.*, 50, 343-347. Japanese. PMID: 1404919.
24. Chakraborty, S., Deokule, J.S., Garg, P., Bhattacharya, S.K., Nandy, R.K., Nair, G.B., Takeda, Y. (2001). Concomitant infection of enterotoxigenic *Escherichia coli* in an outbreak of cholera caused by *Vibrio cholerae* O1 and O139 in Ahmedabad, India. *Journal of Clinical Microbiology*, 39(9), 3241-3246. <https://doi.org/10.1128/JCM.39.9.3241-3246.2001>
25. Tokuda, K., Nishi, J., Imuta, N., Fujiyama, R., Kamenosono, A., Manago, K., Kawano, Y. (2010). Characterization of typical and atypical enteroaggregative *Escherichia coli* in Kagoshima, Japan: biofilm formation and acid resistance. *Microbiology and immunology*, 54(6), 320-329. <https://doi.org/10.1111/j.1348-0421.2010.00210.x>
26. Jenkins, C., Tembo, M., Chart, H., Cheasty, T., Willshaw, G.A., Phillips, A.D., Tompkins, D., Smith, H. (2006). Detection of enteroaggregative *Escherichia coli* in faecal samples from patients in the community with diarrhoea. *Journal of medical microbiology*, 55(11), 1493-1497. <https://doi.org/10.1099/jmm.0.46683-0>
27. Vijay, D., Dhaka, P., Vergis, J., Negi, M., Mohan, V., Kumar, M., Doijad, S., Poharkar, K., Kumar, A., Malik, S.S., Barbuddhe, S.B. (2015). Characterization and biofilm forming ability of diarrhoeagenic enteroaggregative *Escherichia coli* isolates recovered from human infants and young animals. *Comparative Immunology, Microbiology and Infectious Diseases*, 38, 21-31. <https://doi.org/10.1016/j.cimid.2014.11.004>
28. Bai, X., Zhao, A., Lan, R., Xin, Y., Xie, H., Meng, Q., Jin, D., Yu, B., Sun, H., Lu, S., Xu, J., (2013). Shiga toxin-producing *Escherichia coli* in yaks (*Bos grunniens*) from the Qinghai-Tibetan Plateau, China. *PloS one*, 8(6), e65537. <https://doi.org/10.1371/journal.pone.0065537>
29. Kahali, S., Sarkar, B., Rajendran, K., Khanam, J., Yamasaki, S., Nandy, R.K., Bhattacharya, S.K., Ramamurthy, T. (2004). Virulence characteristics and molecular epidemiology of enteroaggregative *Escherichia coli* isolates from hospitalized diarrheal patients in Kolkata, India. *Journal of Clinical Microbiology*, 42(9), 4111-4120. <https://doi.org/10.1128/jcm.42.9.4111-4120.2004>





Sweta Barupal et al.,

30. Nguyen, T. V., Le Van, P., Le Huy, C., Gia, K. N., Weintraub, A. (2005). Detection and characterization of diarrheagenic *Escherichia coli* from young children in Hanoi, Vietnam. *Journal of clinical microbiology*, 43(2), 755-760. <https://doi.org/10.1128/jcm.43.2.755-760.2005>
31. Kudva, I.T., Evans, P.S., Perna, N.T., Barrett, T.J., DeCastro, G.J., Ausubel, F.M., Blattner, F.R., Calderwood, S.B. (2002). Polymorphic amplified typing sequences provide a novel approach to *Escherichia coli* O157: H7 strain typing. *Journal of Clinical Microbiology*, 40(4), 1152-1159. <https://doi.org/10.1128/jcm.40.4.1152-1159.2002>
32. Paton, J. C., Paton, A. W. (1998). Pathogenesis and diagnosis of Shiga toxin-producing *Escherichia coli* infections. *Clinical microbiology reviews*, 11(3), 450-479. <https://doi.org/10.1128/cmr.11.3.450>
33. Vidal, M., Kruger, E., Durán, C., Lagos, R., Levine, M., Prado, V., Toro, C., Vidal, R. (2005). Single multiplex PCR assay to identify simultaneously the six categories of diarrheagenic *Escherichiacoli* associated with enteric infections. *Journal of clinical microbiology*, 43(10), 5362-5365. <https://doi.org/10.1128/jcm.43.10.5362-5365.2005>
34. Aranda, K.R.S., Fagundes-Neto, U., Scaletsky, I.C.A. (2004). Evaluation of multiplex PCRs for diagnosis of infection with diarrheagenic *Escherichia coli* and *Shigella* spp. *Journal of Clinical Microbiology*, 42(12), 5849-5853. <https://doi.org/10.1128/JCM.42.12.5849-5853.2004>
35. Clarke, S. C. (2001). Diarrhoeagenic *Escherichia coli*-an emerging problem? *Diagnostic microbiology and infectious disease*, 41(3), 93-98. [https://doi.org/10.1016/S0732-8893\(01\)00303-0](https://doi.org/10.1016/S0732-8893(01)00303-0)
36. Clements, A., Young, J. C., Constantinou, N., Frankel, G. (2012). Infection strategies of enteric pathogenic *Escherichia coli*. *Gut microbes*, 3(2), 71-87. <https://doi.org/10.4161/gmic.19182>
37. O’Ryan, M., Prado, V., Pickering, L. K. (2005). A millennium update on pediatric diarrheal illness in the developing world. In *Seminars in pediatric infectious diseases* (Vol. 16, No. 2, pp. 125-136). WB Saunders. <https://doi.org/10.1053/j.spid.2005.12.008>
38. Toma, C., Lu, Y., Higa, N., Nakasone, N., Chinen, I., Baschkier, A., Rivas, M., Iwanaga, M. (2003). Multiplex PCR assay for identification of human diarrheagenic *Escherichia coli*. *Journal of clinical microbiology*, 41(6), 2669-2671. <https://doi.org/10.1128/jcm.41.6.2669-2671.2003>
39. Kimata, K., Shima, T., Shimizu, M., Tanaka, D., Isobe, J., Gyobu, Y., Watahiki, M., Nagai, Y. (2005). Rapid categorization of pathogenic *Escherichia coli* by multiplex PCR. *Microbiology and immunology*, 49(6), 485-492. <https://doi.org/10.1111/j.1348-0421.2005.tb03752.x>
40. Müller, D., Greune, L., Heusipp, G., Karch, H., Fruth, A., Tschäpe, H., Schmidt, M. A. (2007). Identification of unconventional intestinal pathogenic *Escherichia coli* isolates expressing intermediate virulence factor profiles by using a novel single-step multiplex PCR. *Applied and environmental microbiology*, 73(10), 3380-3390. <https://doi.org/10.1128/AEM.02855-06>
41. Fujioka, M., Otomo, Y., Ahsan, C. R. (2013). A novel single-step multiplex polymerase chain reaction assay for the detection of diarrheagenic *Escherichia coli*. *Journal of microbiological methods*, 92(3), 289-292. <https://doi.org/10.1016/j.mimet.2012.12.010>
42. Fialho, O.B., de Souza, E.M., de Borba Dallagassa, C., de Oliveira Pedrosa, F., Klassen, G., Irino, K., Paludo, K.S., de Assis, F.E.A., Surek, M., de Souza Santos Farah, S.M., Fadel-Picheth, C.M.T. (2013). Detection of Diarrheagenic *Escherichia coli* Using a Two-System Multiplex-PCR Protocol. *Journal of Clinical Laboratory Analysis*, 27(2), 155-161. <https://doi.org/10.1002/jcla.21578>
43. Chandra, M., Cheng, P., Rondeau, G., Porwollik, S., McClelland, M. (2013). A single step multiplex PCR for identification of six diarrheagenic *E. coli* pathotypes and *Salmonella*. *International Journal of Medical Microbiology*, 303(4), 210-216. <https://doi.org/10.1016/j.ijmm.2013.02.013>
44. Oh, K. H., Kim, S. B., Park, M. S., Cho, S. H. (2014). Development of a one-step PCR assay with nine primer pairs for the detection of five diarrheagenic *Escherichia coli* types. *Journal of Microbiology and Biotechnology*, 24(6), 862-868. <https://doi.org/10.4014/jmb.1312.12031>
45. Sjöling, Å., Sadeghipoorjahromi, L., Novak, D., Tobias, J. (2015). Detection of major diarrheagenic bacterial pathogens by multiplex PCR panels. *Microbiological research*, 172, 34-40. <https://doi.org/10.1016/j.micres.2014.12.003>
46. Vendruscolo, J.W., Waldrich, T.L., Saikawa, G.I.A., Pelayo, J.S., Kobayashi, R.K.T., Nakazato, G., Rocha, S.P.D. (2017). Novel multiplex PCR for detection of diarrheagenic *Escherichia coli* strains isolated from stool and water samples. *Genet Mol Res*, 16(3). <http://dx.doi.org/10.4238/gmr16039760>





Sweta Barupal et al.,

47. Ballal, M., Devadas, S. M., Chakraborty, R., & Shetty, V. (2014). Emerging trends in the etiology and antimicrobial susceptibility pattern of enteric pathogens in rural coastal India. *International Journal of Clinical Medicine*, 2014. <https://doi.org/10.4236/ijcm.2014.57058>
48. Samal, S.K., Khuntia, H.K., Nanda, P.K., Satapathy, C.S., Nayak, S.R., Sarangi, A.K., Sahoo, N., Pattnaik, S.K., Chhotray, G.P., Pal, B.B. (2008). Incidence of bacterial enteropathogens among hospitalized diarrhea patients from Orissa, India. *Japanese journal of infectious diseases*, 61(5), 350-355. <https://doi.org/10.7883/yoken.JJID.2008.350>
49. Rajendran, P., Ajjampur, S.S.R., Chidambaram, D., Chandrabose, G., Thangaraj, B., Sarkar, R., Samuel, P., Rajan, D.P., Kang, G. (2010). Pathotypes of diarrheagenic *Escherichia coli* in children attending a tertiary care hospital in South India. *Diagnostic microbiology and infectious disease*, 68(2), 117-122. <https://doi.org/10.1016/j.diagmicrobio.2010.06.003>
50. Sudershan, R.V., Kumar, R.N., Kulkarni, B., Kashinath, L., Bhaskar, V. and Polasa, K. (2014). *E. coli* pathotypes and their antibiotic resistance in young children with diarrhea in Hyderabad, India. *Int J Curr Microbiol App Sci*, 3(9), 647-54.
51. Okhuysen, P. C., DuPont, H. L. (2010). Enteroaggregative *Escherichia coli* (EAEC): a cause of acute and persistent diarrhea of worldwide importance. *The Journal of infectious diseases*, 202(4), 503-505. <https://doi.org/10.1086/654895>
52. Ozerol, I.H., Bayraktar, M.R., Iseri, L., Otlu, B. Durmaz, R. (2005). The prevalence and molecular typing of enterotoxigenic *Escherichia coli* strains isolated from diarrheic stools in Malatya, Turkey. *The new Microbiologica*, 28(3), 237-243. <https://orcid.org/0000-0002-6220-0521>
53. Rivera, F.P., Ochoa, T.J., Maves, R.C., Bernal, M., Medina, A.M., Meza, R., Barletta, F., Mercado, E., Ecker, L., Gil, A.I., Hall, E.R. (2010). Genotypic and phenotypic characterization of enterotoxigenic *Escherichia coli* strains isolated from Peruvian children. *Journal of clinical microbiology*, 48(9), 3198-3203. <https://doi.org/10.1128/jcm.00644-10>
54. Vilchez, S., Reyes, D., Paniagua, M., Bucardo, F., Möllby, R., Weintraub, A. (2009). Prevalence of diarrhoeagenic *Escherichia coli* in children from Leon, Nicaragua. *Journal of medical microbiology*, 58(5), 630-637. <https://doi.org/10.1099/jmm.0.007369-0>
55. Wani, S.A., Nabi, A., Fayaz, I., Ahmad, I., Nishikawa, Y., Qureshi, K., Khan, M.A., Chowdhary, J. (2006). Investigation of diarrhoeic faecal samples for enterotoxigenic, Shiga toxin-producing and typical or atypical enteropathogenic *Escherichia coli* in Kashmir, India. *FEMS microbiology letters*, 261(2), 238-244. <https://doi.org/10.1111/j.1574-6968.2006.00354.x>
56. Shetty, V.A., Kumar, S.H., Shetty, A.K., Karunasagar, I., Karunasagar, I. (2012). Prevalence and characterization of diarrheagenic *Escherichia coli* isolated from adults and children in Mangalore, India. *Journal of laboratory physicians*, 4(01), 024-029. <https://doi.org/10.4103/0974-2727.98666>

Table 1: Oligonucleotide sequences used in the study for selective amplification of DEC associated virulence genes and their amplicon sizes

DEC Pathotype	Target genes	Function	Primer sequence (5'-3')		Product size (bp)	References
			Forward (FP)	Reverse (RP)		
EPEC	<i>bfpA</i>	Bundle-forming pilus	TTCTGGTGCTTG CGTGTCTTTT	TTTGTGTTGTTG TATCTTTGTAA	385	20
	<i>eae</i>	Intimin, promotes attachment	CGGAAGCCAAA GCGACAAGATT A	TGACCAGAAGA AGCATCCACCG AA	917	21
ETEC	<i>st</i>	Heat-stable toxin	TTTATTTCTGTAT TGTCCTT	ATTACA ACACAGTTCAC AG	171	22
	<i>lt</i>	Heat-labile toxin	AGCAGGTTCC	GTGCTCAGATT	130	23





Sweta Barupal et al.,

			ACCGGATCACCA	CTGGGTCTC		
	<i>est</i>	Heat-stable enterotoxin	ATTTTACTTTCT GTATTAGTCTT	CACCCGGTACA AGGCAGGATT	190	24
EAEC	<i>irp2</i>	Iron acquisition and virulence	AAGGATTCGCTG TTACCCGGAC	TCGTCGGGCAG CGTTTCTTCT	287	25
	<i>aaiA</i>	Type VI secretion system (T6SS)	AACCCGGAGATGC TGAAACTGCG	GGATTGCCATT AGTAGTACCA G	384	26
	<i>aggR</i>	Aggregative adherence and biofilm formation	CCTAAAGGATGC CCTGATGATAA	TAACGCTGGAC ATGAGATAACC	663	27
	<i>pic</i>	Mucin degradation and immune evasion	GGGTATTGTCCG TTCCGAT	ACAACGATAACC GTCCTCCCG	1175	28
	<i>aafA</i>	Aggregative adherence fimbriae	ATGTATTTTTAGA GGTTGAC	TATTATATTGTC ACAAGCTC	518	29
	<i>pCVD432</i>	Plasmid-encoded regulatory gene promotes biofilm formation and virulence	CTGGCGAAAGAC TGTATCAT	CAATGTATAGA AATCCGCTGTT	630	30
EHEC	<i>stx1</i>	Shiga-toxin-I	TTACAGCGTGTT GCAGGGATCAGT	TTGTGCGTAAT CCCACGGACTC TT	470	21
	<i>stx2</i>	Shiga-toxin-II	GGCACTGTCTGA AACTGCTCC	TCGCCAGTTAT CTGACATTCTG	255	31
	<i>hlyA</i>	hemolysin A	GCATCATCAAGC GTACGTTCC	AATGAGCCAAG CTGGTTAAGCT	534	32
DAEC	<i>daaE</i>	Adhesin, involved in diffuse adherence	GAA CGT TGG TTA ATG TGG GGT AA	TAT TCA CCG GTC GGT TAT CAG T	542	33
EIEC	<i>ipaH</i>	Host cell invasion	GTTCCTTGACCG CCTTCCGATACC GTC	GCCGGTCAGCC ACCCTCTGAGA GTAC	600	34

Table 2: PCR Cycling conditions for multiplex PCR (mpPCR)

PCR steps	Reagent of the PCR	Initial denaturati on	Denaturati on	Annealin g	Extensio n	Final extensio n
mpPCR I	PCR buffer:10x MgCl ₂ : 25mM dNTPs: 250µM Taq polymerase: 2U/µl FP and RP: 0.4µM each	90°C-95°C 2 min.	95°C 30 sec.	50°C- 55°C 1 min. 30 cycles	72°C 1:30 min.	72°C 10 min.
mpPCR II	PCR buffer:10x MgCl ₂ : 25mM dNTPs: 250µM	90°C-95°C 2 min.	95°C 30 sec.	42°C- 48°C 1 min.	72°C 1:30 min.	72°C 10 min.





Sweta Barupal et al.,

	Taq polymerase: 2U/μl FP and RP: 0.4μm each			30-35 cycles		
mpPCR III	PCR buffer:10x MgCl ₂ : 25mM dNTPs: 250μM Taq polymerase: 2U/μl FP and RP: 0.4μm each	90°C-95°C 2 min.	95°C 30 sec.	50°C- 55°C 1 min. 30 cycles	72°C 1:30 min.	72°C 10 min.

Table 3: Distribution of *E. coli* in children with diarrhea based on age, sex, and clinical features

Variables	Categories	No. of positive % (n=66)	No. of negative% (n=78)
Sex	Male	36(54.5)	44(56.4)
	Female	30(45.4)	34(43.5)
Age	0-12 months	16(24.2)	19(24.3)
	13-24 months	20(30.3)	22(28.2)
	25-36 months	10(15.1)	12(15.3)
	37-48 months	06(9)	10(12.8)
	49-60 months	14(21.2)	15(19.2)
Feed type	Breastfeed	06(9)	20(25.6)
	Breastfeed and formula milk	28(42.4)	38(48.7)
	Solid food	32(48.4)	20(25.6)
Stool type	Loose	18(27.2)	24(30.7)
	Watery	32(48.4)	30(38.4)
	Bloody	2(3)	14(17.9)
	Muroid	14(21.2)	10(12.8)

Table 4: Occurrence of *E. coli* in untreated community sewage samples from urban and rural areas

Rural Sites	Latitude	Longitude	No. of positive (%) n=24	No. of negative (%) n=16
1. Udairamsar	27.94006	73.30384	2(8.3)	0
2. Bachhasar	27.95756	73.19093	1(4.1)	1(6.2)
3. Palana	27.84395	73.26208	2(8.3)	0
4. Chak Chani	27.87696	73.00800	2(8.3)	0
5. Kotri	27.86039	72.95937	1(4.1)	1(6.2)
6. Kolayat	27.83491	72.95611	1(4.1)	1(6.2)
7. Madh	27.85667	72.93063	1(4.1)	1(6.2)
8. Husangsar	28.15676	73.41487	1(4.1)	1(6.2)
9. Barsnghsar	27.81985	73.19961	2(8.3)	0
10. Kanasar	28.13366	73.31064	1(4.1)	1(6.2)
Urban Sites				
11. Satellite	28.02030	73.30385	1(4.1)	1(6.2)
12. Kothari	28.02636	73.29809	1(4.1)	1(6.2)
13. Nagnechi	27.99352	73.33565	2(8.3)	0
14. Choukhuti	28.02352	73.30927	2(8.3)	0





Sweta Barupal et al.,

15. Kuchilpura	28.02272	73.31440	1(4.1)	1(6.2)
16. Bhutto ka chauraha	28.03075	73.31459	0	2(12.5)
17. Railway colony	28.03130	73.30124	1(4.1)	1(6.2)
18. Dharnidhar	28.00610	73.29435	1(4.1)	1(6.2)
19. Police-line	28.02785	73.31314	1(4.1)	1(6.2)
20. Pugal-phanta	28.02733	73.29853	0	2(12.5)

Table 5: Distribution of virulence genes in *E. coli* isolates from stool samples and sewage samples.

Virulence genes detected	Stool samples from children (%) n=42	Sewage samples (%) n=16	Total (%)
<i>irp2</i>	11(26.1)	5(31.2)	16(27.5)
<i>aafA</i>	2(4.7)	1(6.2)	3(5.1)
<i>aaiA</i>	1(2.3)	0	1(1.7)
<i>pic</i>	1(2.3)	0	1(1.7)
<i>pCVD432</i>	0	1(6.2)	1(1.7)
<i>aggR</i>	0	0	0
<i>bfpA</i>	2(4.7)	0	2(3.4)
<i>eae</i>	0	0	0
<i>stx1</i>	4(9.5)	1(6.2)	5(8.6)
<i>stx2</i>	3(7.1)	1(6.2)	4(6.8)
<i>hlyA</i>	2(4.7)	2(12.5)	4(6.8)
<i>st</i>	9(21.4)	3(18.7)	12(20.6)
<i>lt</i>	2(4.7)	2(12.5)	4(6.8)
<i>est</i>	1(2.3)	0	1(1.7)
<i>daaE</i>	3(7.1)	0	3(5.1)
<i>ipaH</i>	1(2.3)	0	1(1.7)

Table 6: Distribution of DEC pathotypes with clinical features and risk factors in children with acute diarrhea

DEC Pathotypes and detected virulence genes	Variables													
	Sex		Age (Months)					Type of Diarrhea				Type of feed		
	Male	Female	0-12	13-24	25-36	37-48	49-60	Loose	Watery	Bloody	Mucoid	Breastfeed	Breastfeed & Formula	Solid
EAEC (n=15)	9	6	3	4	5	2	1	6	7	1	1	4	7	4
<i>irp2</i>	7	4	1	1	3	1	1	3	2	-	1	1	3	2
<i>aafA</i>	1	1	-	1	1	-	-	1	1	1	-	1	1	1
<i>aaiA</i>	1	-	1	1	1	1	-	1	2	-	-	1	2	1
<i>pic</i>	-	1	1	1	-	-	-	1	2	-	-	1	1	-
EPEC (n=2)	1	1	1	-	1	-	-	1	1	-	-	-	1	1
<i>bfpA</i>	1	1	1	-	1	-	-	1	1	-	-	-	1	1
EHEC (n=9)	6	3	3	1	2	2	1	3	4	2	-	2	4	3
<i>stx1</i>	3	1	1	1	1	-	1	1	1	1	-	1	2	1
<i>stx2</i>	2	1	1	-	1	1	-	1	2	-	-	1	1	1
<i>hlyA</i>	1	1	1	-	-	1	-	1	1	1	-	-	1	1
ETEC (n=12)	7	5	3	2	4	1	2	5	4	1	2	3	4	5
<i>st</i>	5	4	1	1	2	-	2	3	1	1	1	1	2	2
<i>lt</i>	1	1	1	1	1	1	-	2	2	-	1	1	1	3





Sweta Barupal et al.,

<i>est</i>	1	-	1		1			-	1	-	-	1	1	-
DAEC (n=03)	2	1	-	-	1	1	1	1	2	-	-	-	2	1
<i>daaE</i>	2	1	-	-	1	1	1	1	2	-	-	-	2	1
EIEC (n=01)	1	-	-	-	1	-	-	1	-	-	-	-	-	1
<i>ipaH</i>	1	1	-	-	1	-	-	1	-	-	-	-	-	1
Total (n=42)	26	16	10	7	14	6	5	17	18	4	3	9	18	15

Table 7: Distribution of DEC pathotypes in sewage samples collected from rural and urban areas

Virulence genes	No. of virulence genes from sewage samples (Rural areas)	No. of virulence genes from sewage samples (Urban areas)
EAEC (n=7)	5	2
<i>irp2</i>	3	2
<i>aafA</i>	1	-
<i>pcvd432</i>	1	-
EHEC (n=4)	3	1
<i>stx1</i>	1	-
<i>stx2</i>	1	-
<i>hlyA</i>	1	1
ETEC (n=5)	3	2
<i>st</i>	2	1
<i>lt</i>	1	1
Total (Totaln=16)	11	5

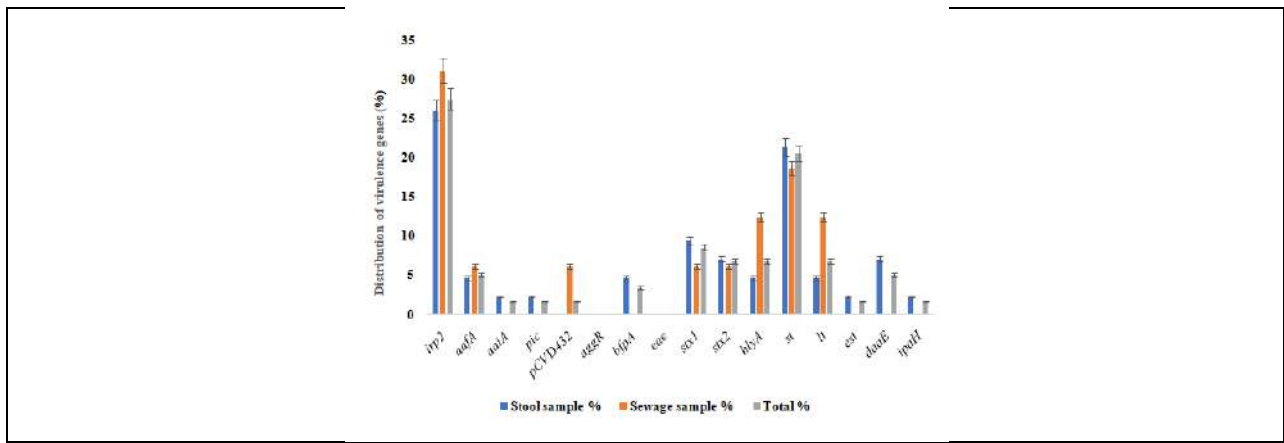


Fig. 1: Distribution of virulence genes (%) in DEC pathotypes among stool and sewage samples





Sweta Barupal et al.,

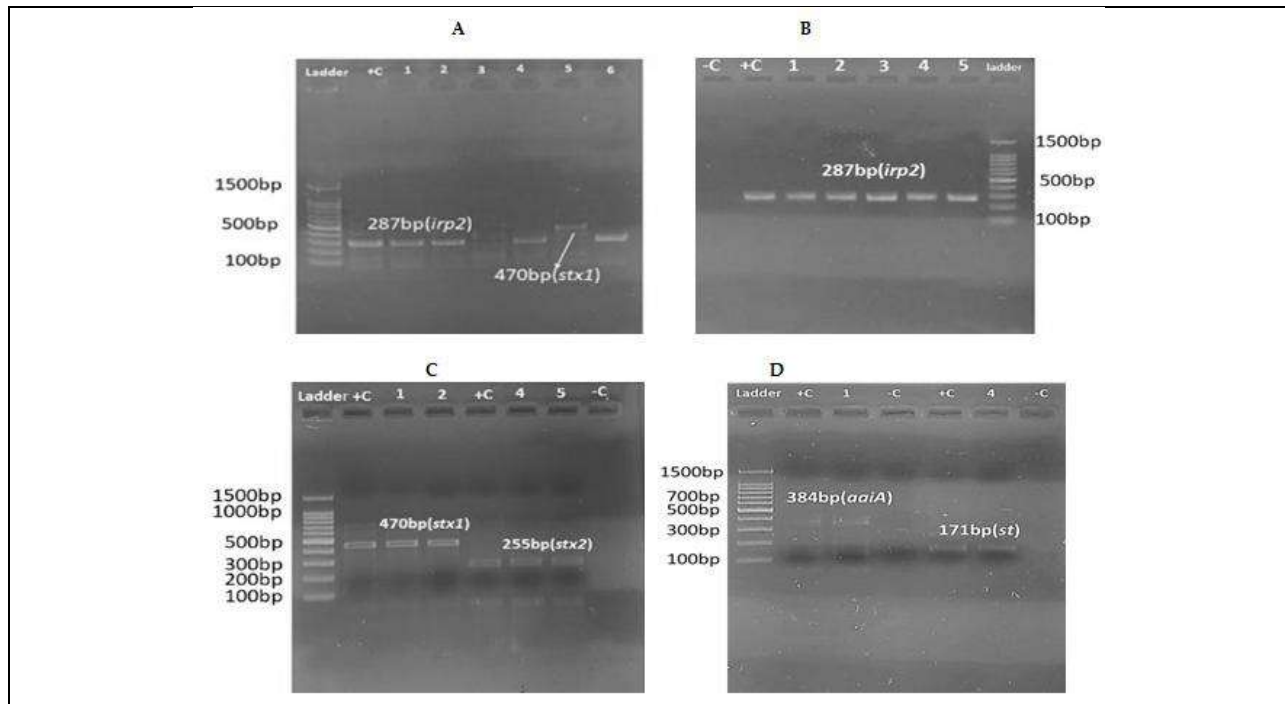


Fig.2. Multiplex PCR to detect virulence genes in Diarrheagenic *Escherichia coli*: (A) Lane 1; molecular weight marker (100 bp ladder), Lane 2; positive control EAEC: ATCC 33780 (*irp2*), Lane 3,4,6,8; EAEC (*irp2* amplicon size 287bp), Lane 7; EHEC (*stx1* amplicon size 470bp). (B) Lane 1; Negative control, Lane 2; positive control EAEC: ATCC 33780 (*irp2*) and Lane 3 to 7; EAEC (*irp2* amplicon size 287bp), Lane 8; Molecular weight marker (100 bp ladder), (C) Lane 1; molecular weight marker (100 bp ladder), Lane 2; positive control EHEC: ATCC 35150 (*stx1*), Lane 3,4,5,6 EHEC (*stx1* amplicon size 470bp, and *stx2* amplicon size 255bp), Lane 8; Negative control (D) Lane 1; molecular weight marker (100 bp ladder), Lane 2; positive control EAEC: ATCC 33780 (*aaiA*), Lane 3; EAEC *aaiA* amplicon size 384bp), Lane 4; negative control (DH5 α), Lane 5; positive control ETEC: ATCC 35401 (*st*), Lane 6; ETEC (*st* amplicon size 171bp).





A Study of Proximate, Mineral and Phytochemical Properties of Banana (*Musa spp.*) Pseudostem Waste

Meghna N. Diarsa¹ and Akshaya Gupte^{2*}

¹Assistant Professor, Department of Medical Laboratory Technology (MLT), Bapubhai Desai bhai Patel Institute of Paramedical Sciences (BDIPS), CHARUSAT, Changa, Gujarat, India.

²Associate Professor, P.G. Department of Bio Science, Sardar Patel University, Vallabh Vidyanagar, Gujarat, India.

Received: 29 Apr 2024

Revised: 10 Aug 2024

Accepted: 18 Oct 2024

*Address for Correspondence

Akshaya Gupte

Associate Professor,

P.G. Department of Bio Science,

Sardar Patel University,

Vallabh Vidyanagar, Gujarat, India.

E.Mail: akshayagupte@spuvvn.edu



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Banana pseudostem has most important role in food industries due to its medicinal value, rich in mineral content and easy processibility. The present research study focused on proximate, physicochemical characterization and mineral analysis of Mahalaxmi, Robusta and Grand Naine banana pseudostem. The research study shows that all the three different cultivars contain high amount of moisture content (95%). Banana pseudostem is a high cellulosic and low lignin content lignocellulosicbiowaste. Among the three different banana pseudostem, Mahalaxmi banana pseudostem contain maximum cellulosic content (47.6%) and lowest lignin content (7.7%). The mineral content such as sodium, potassium, calcium, magnesium, phosphorous, manganese, iron and zinc content were estimated. The highest mineral content, calcium (1484.01 mg%), followed by potassium (869.86 mg%) and magnesium (260.51 mg%) were found in Mahalaxmi banana pseudostem as compared to Robusta and Grand Naine banana pseudostem. The WHC (19.3gm of water/g of dry substrate)and OHC (9.3 gm of oil/g of dry substrate) was found maximum in Robusta banana pseudostem. The maximum swelling power (9.3 (g of swollen particles/ g of dry substrate)obtain in Mahalaxmi banana pseudostem. The phytochemical analysis shows that the Alkaloid, Tannin, Flavanoid, Phytate and Protein content is present in Mahalaxmi, Robusta and Grand Naine banana pseudostem. However, the phytochemical study shows that no significant difference was observed in Mahalaxmi, Robusta and Grand Naine banana pseudostem.

Keyword: Banana pseudostem, proximate analysis, mineral content and phytochemical analysis.





Meghna N. Diarsa and Akshaya Gupte

INTRODUCTION

Banana is one of the tallest monocotyledons herbaceous flowering plant of the genus *Musa*. Banana plant has belonged to the *Musaceous* family. Banana is the oldest and most economically important cultivated crop in the world. Carl Linnaeus in 1753 first named the genus *Musa*. Banana (genus *Musa*) are the second most important tropical and staple food crop in the world after oil palm^[1]. Banana pseudostem have high amount of holocellulose, ash content and low level of lignin content which is very important role in papermaking^[2]. Various studies show that unripe banana is a rich source of starch, which offer beneficial effects for the human health. The banana plant contains long overlapped leafstalk. Banana plant is grow in 130 countries of the world. In the World and India, 48.9 and 32 million tons of banana produce. In Gujarat, banana is cultivated in area of 61.9 thousand hector with an annual production of 3779.6 metric ton. Banana is a fourth most important food after paddy, wheat and milk product, in terms of gross value. Banana is a rich source of minerals, carbohydrate, potassium, phosphorous, calcium, iron, vitamin B and C. The central core of the banana is used as a vegetable in various South Indian cuisines.

Banana plant contain root, pseudostem, leaf and fruit. Banana is a monocotyledonous plant, it produces only single bunch of banana fruit during the life cycle. After harvesting the fruit of banana, the stem is cut and left on the soil plantation which create the hazardous environmental problems. This agricultural rich lignocellulosic waste can be used in the in paper making, pulp, high quality dress material, cardboard, teabags, and currency notes^[3,4]. The aim of this paper is to study the proximate, mineral and physico-chemical properties of Mahalaxmi, Robusta and Grand Naine banana pseudostem.

MATERIALS AND METHODS

Materials

Three cultivar of banana (Grand Naine, Robusta and Mahalaxmi) were collected from local banana farm, Anand, Gujarat, India. Cetyltrimethyl ammonium bromide (CTAB), silver nitrate, potassium acetate, ferric nitrate, oxalic acid, hydrochloric acid, Sulphuric acid, ethyl alcohol, tertiary butyl alcohol, acetone, sodium lauryl sulfate, ethylene diaminetetraacetic acid (EDTA), borax, disodium phosphate, ethylene glycol, glacial acetic acid (GAA), copper sulfate, folin's reagent, all the chemicals were purchased from Himedia, Mumbai, India.

Banana pseudostem collection

The leaf sheaths were separated by cutting the banana pseudostem with the help of the knife. The soil particles were removed by washing treatment and dried in a hot air oven at 85°C for 5-6 h. The dried sheaths of pseudostem were ground to obtain 3-5 mm in length particle size substrate. The chemical analysis of banana pseudostem (moisture, lignin, cellulose, hemicellulose, carbohydrate, protein, crude fat, and crude fibers) was evaluated.

Compositional analysis

The compositional analysis of three different varieties of banana pseudostem was evaluated according to method described by Van Soest *et al.*, (1991)^[5]. 1 gm of banana pseudostem substrate was mixed with 100 ml of ADS (acid detergent solution). ADS contains 20 g/l cetyltrimethyl ammonium bromide (CTAB) and 1N sulfuric acid (v/v). The banana pseudostem substrates were refluxed with the ADS for 1 hr. The treated substrate was filtered through the sintered glass crucible. The filtrated substrate was washed with warm distilled water followed by ethanol. The residues were dried at 110°C for 24 h and weighed to calculate the weight of ADF (acid detergent fibers).

$$ADF(\%) = \frac{\text{Weight of crucible and acid treated fibers} - \text{Weight of crucible}}{\text{Initial weight of the substrate}} \times 100$$





Meghna N. Diarsa and Akshaya Gupte

The lignin content of banana pseudostem substrate was determined by a mixture of permanganate solution. ADF were treated with potassium permanganate solution [potassium permanganate (50 g/l) and silver sulfate (0.05g/l)] and the buffer solution which contain 40% (v/v) tertiary butyl alcohol, 50% (v/v) GAA, 0.5% (w/v) potassium acetate, 0.015% (w/v) silver nitrate and 0.6% (w/v) ferric nitrate for 90 minutes. The demineralized solution (a mixture of 5% (w/v) oxalic acid, 5% (v/v) hydrochloric acid and 70% (v/v) ethanol) was used to treat the residue, to obtain the white fibers. The demineralized fibers were treated with warm distilled water and dried at 110°C for 24 h. The lignin content was calculated by using the following equation. The content of ash was evaluated by using muffle furnace at 500°C ±5°C for 6 hours. The remaining substrate residues were considered as an ash content and the loss in weight corresponded to the cellulosic amount.

$$\text{Lignin}(\%) = \frac{\text{Weight of ADF residues} - \text{Weight of demineralised residue}}{\text{Weight of the initial residue}} \times 100$$

The natural detergent fiber (NDF) was used to evaluate the content of hemicellulose of banana pseudostem. 1 gm of substrate was mixed with the NDF solution. The NDF solution contain 1% (w/v) sodium lauryl sulfate, 1.9% (w/v) ethylenediamine tetra acetic acid, 0.7% (w/v) borax, 1% (w/v) disodium phosphate and 1% (v/v) ethyl glycol added to avoid the formation of foam and maintain the neutral pH. The NDF solution and banana pseudostem substrate was refluxed for 1 hr. Thereafter, the treated substrate was washed with warm distilled water followed by 95% ethanol (v/v). The residues were filtered with the help of glass crucible, dried overnight at 110°C and weighed to calculate natural detergent fiber.

$$\text{NDF}(\%) = \frac{\text{Weight of crucible and NDF residues} - \text{Weight of crucible}}{\text{Initial weight of the residue}} \times 100$$

The hemicellulosic content of banana pseudostem is calculated using the following equation
Hemicellulose (%) = Weight of NDF residue (%) - Weight of ADF residue (%)

Moisture content and dry matter

The moisture content and dry matter of three different varieties of banana were evaluated by the oven dry method according to Wang *et al.*, (2016)^[6]. The initial weight of the banana pseudostem were recorded and the samples were dried in the oven at 105°C until constant weight obtained. The moisture content was calculated by the percentage difference between initial and final weight of the banana pseudostem. The remaining residues were considered as a dry matter.

$$\text{Moisture}(\%) = \frac{\text{Initial weight of the sample} - \text{Final weight of the sample}}{\text{Initial weight of the sample}} \times 100$$

Extractive and Crude fat

The extractive of Mahalaxim, Robusta and Grand Naine banana pseudostems were determined according to TAPPI T 204 cm 97 (1997)(Pereira *et al.*, 2010)^[7]. Crude fat was examined by petroleum ether extraction method using soxhlet apparatus. The crude fat content was calculated by the following equation.

$$\text{Crude fat} = \frac{\text{Final weight of the residues} - \text{Initial weight of the residues}}{\text{Initial weight of the residues}} \times 100$$

Mineral analysis

The determination of mineral content of three cultivar of banana were evaluated by Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES) perkin Elmer, USA, Avio 200. .7





Meghna N. Diarsa and Akshaya Gupte

Swelling power and solubility of banana pseudostem

The swelling power and solubility of banana pseudostem was examined according to slightly modified method described by Aziz *et al.*, (2011)[8]. 1 gm of banana pseudostem substrate was mixed with 20 ml of distilled water. The reaction system was stirred continuously on the magnetic stirrer at 90°C for 30 minutes. The mixture was centrifuged at 8000 rpm for 10 minutes. The supernatant was transferred into the petri plate and dried in the oven at 110°C, the pellet residues were weighed and considered as a swollen granule. The following equation was used to determine the solubility and swelling power of three cultivars of banana.

$$\text{Solubility} = \frac{\text{Weight of dried residues}}{\text{Initial weight of the residues}} \times 100$$

$$\text{Swelling power} = \frac{\text{Weight of swollen granules}}{\text{Initial weight of the residue} - \text{Solubility}} \times 100$$

Water and oil holding capacity

The water holding capacity (WHC) and oil holding capacity (OHC) of three different cultivars of banana pseudostems were examined according to method described by Yadav *et al.*, (2016)^[9]. The WHC and OHC are very important parameter as it defines the quality of the food. 1 gm of substrate was mixed with 25 ml of water and olive oil to estimate the WHC and OHC. The reaction mixture was stirred continuously on the magnetic stirrer at room temperature for 30 minutes. The suspension was centrifuged at 8000 rpm for 10 minutes and weighed the residues. Water and oil holding capacity of banana pseudostem was calculated and expressed as a gram water and oil per gram of substrate.

Quantitative phytochemical analysis

Phytochemical such as flavonoid, alkaloid, phytate, tannin and phenolic impart color to vegetable and fruits. The phytochemicals play important role in antioxidant property and other physiological functions. Alkaloid content was analyzed by mixing 5 gm of substrates with 200 ml of 20% HCL (v/v). The reaction mixtures were incubated for 4 hours at 25°C. After the 4 h of incubation, the reaction system was filtered using whatmann filter paper No.42. The concentrated ammonium hydroxide was added drop wise in to the concentrated filtrates until the precipitate was complete. The precipitates were collected and rinsed with two time diluted ammonium hydroxide. The residues were collected on the pre-weighed filter paper. The residues were dried in the oven at 80°C. The amount of alkaloid was calculated using the following equation.

$$\text{Alkaloid}(\%) = \frac{\text{Weight of filter paper and residues} - \text{Weight of filter paper}}{\text{Weight of sample}} \times 100$$

Flavonoid is a polyphenolic water soluble compound present in the plant kingdom as secondary metabolites (Chávez-González *et al.*, 2020)^[10]. The estimation of flavonoid was done by Aluminum chloride method described by Al Amri and Hossain (2018)^[11]. The different concentration (10 µg/ml-100 µg/ml) of banana pseudostem substrates were prepared by adding the methanol. The reaction system was mixed with 125 µl water and 75 µl 5% (w/v) sodium nitrate. The reaction system was mixed properly and incubated for 6 min. After incubation, 150 µl of 10% (w/v) aluminum chloride was added and incubated for 5 min. After 5 min. of incubation, 500 µl of 4% (w/v) sodium hydroxide and 275 µl water were added. The absorbance was recorded at 510 nm against black by using SHIMARDZU 1800 UV-Vis spectroscopy. The content of flavonoids was determined concerning the standard curve quercetin. The flavonoid content is expressed in µg of QE/g of banana pseudostem sample.

The tannin content of banana pseudostem was quantified using the method described by Belloet *et al.*, (2013)^[12] with slight modification. 5 gm of substrate was mixed with 100 ml of petroleum ether and kept at room temperature for 24 h. Thereafter, the samples were filtered and petroleum ether was evaporated. The 5 ml filtrates were then mixed with





Meghna N. Diarsa and Akshaya Gupte

2 ml of 0.1 M FeCl₃ in 0.1 N HCL and 8 mM potassium ferrocyanide. The absorbance was recorded using the spectrometer (SHIMARDZU UV-1800) at 725 nm. The amount of tannin was calculated using the following equation.

$$\text{Tannin (\%)} = \frac{\text{Abs of the sample} \times \text{tannin conc. in standard}}{\text{Weight of initial sample}} \times 100$$

The estimation of phytate content in banana pseudostem was done by adding 2% (v/v) HCL (100 ml) with 0.2 gm of pseudostem substrate. The solutions were kept for 3 h at room temperature. Thereafter the solutions were filtered and 50 ml of filtrates were mixed with 100 ml of distilled water. This solution was titrated against FeCl₃ with 0.3% (w/v) ammonium thiocyanate (10 ml) as an indicator until a brownish yellow color appeared. The phytate concentration was calculated using the following equation.

$$\text{Phytic acid} = \frac{\text{Titrant value} \times 0.00195 \times 1.19 \times 100}{\text{Initial weight of sample}}$$

Where, 0.00195 is a gm of iron per ml and 1.19 is the mg of phytin phosphorous per ml.

Phenolic content was evaluated by Folin-Ciocalteu test using the method described by Molina-Cortes *et al.*, (2019)^[13]. 0.4 ml of the extracted samples of three different banana pseudostems were mixed with 4 ml of distilled water followed by 0.4 ml of Folin-Ciocalteu reagent. The solutions were incubated for 10 min. at room temperature in dark condition. Thereafter, 4 ml of Sodium carbonate (7% w/v) was added into the reaction system and the solutions were mixed properly and incubated for 90 minutes at room temperature in the dark environment. Gallic acid (GA) is used as a standard and distilled water is used as a blank. The absorbance was recorded in spectrophotometer (SHIMARDZU UV 1800) at the wavelength 730 nm. The phenolic content was expressed as mg equivalent of gallic acid per gm of banana pseudostem sample (mg GAE/g).

RESULTS AND DISCUSSION

Proximate analysis

The proximate composition of three different varieties of lignocellulosic banana pseudostem (Robusta, Grand Naine and Mahalaxmi) is shown in Table 1. The chemical composition of banana pseudostem varies with the species of banana pseudostem, age of plant, plantation soil and environmental condition[14,15]. Cellulose, hemicellulose and lignin are the main constituents of banana plant cell wall[16].

Among these plant fibers, lignin is a very strong component which cannot be easily digested. Cellulose is a major constituent of banana plant followed by hemicellulose and lignin. The result shows that the banana pseudostem is a rich source of cellulose. The highest cellulosic content was found in species of Mahalaxmi (47.6%) followed by Grand Naine (42.7%) and Robusta (40.9%). Cellulose is a main component of the primary and secondary cell wall of banana pseudostem due to that banana plant is highly cellulosic in nature. Similar results were also observed by many researchers. Jayaprabha *et al.*, (2011)^[14] reported 46.3% cellulose in Nendran banana plant. Preethi and Balakrishna (2011)^[17] reported 48.19% cellulose in Grand Naine banana pseudostem. Viswanathan *et al.*, (1989)^[18] reported the 35.9% cellulose content in Robusta which is 5% lower value than our research study. The variation in the lignocellulosic content may be due to the climate and soil.

Hemicellulose is a second most important component in the banana pseudostem. Hemicellulose content was found in the range of 23% - 29% in banana pseudostem. The highest hemicellulosic content found in the Mahalaxmi banana pseudostem (29.4%) followed by Robusta (28.9%) and Grand Naine (23.3%). Similar results were also observed by Viswanathan *et al.*, (1989)^[18] for Robusta. They found 21.9% hemicellulose in stalk of *Musa cavendishi*. Banana plant have lower lignin content as compared to other natural fibers such as jute and coir ^[15].



**Meghna N. Diarsa and Akshaya Gupte**

Mahalaxmi banana plant contains 7.7% lignin, Robusta contain 9% lignin and 8.8 % lignin found in Grand Naine. Poyyamozi and Kadirvel (1986)[19] reported the lowest lignin component 9-10% in Robusta. Viswanathan et al., (1989) [18] also found the 9.4 % lignin component in Robusta species. The lignin content of Mahalaxmi, Robusta and Grand Naine is lower than the lignin content of *M. acuminata* Colla (12.7%), as reported by Cordeiro et al., 2004[20]. Shivashankaret al., (2006)[3] found 2.2-time higher lignin content in Robusta (20.23%). The ash content was found in the range of 2.7% - 4.5%. The highest ash content was observed in Mahalaxmi banana plant (4.5%) and the lowest in Grand Naine (2.7%). The evaluation of ash content indicates that the Mahalaxmi banana plant contain high mineral level than the Robusta and Grand Naine. Jayaprabha et al., (2011)[14] found 2.8% ash content in pith removed pseudostem and 10.7% ash content in whole pseudostem sheaths. The ash content of all the three different varieties of banana pseudostem in the present study is similar to the ash content of the *Musa cuminata* balbisiana Colla (3%), as reported by Aziz et al., (2011)[8].

Moisture content and dry matter

Table 1 shows the moisture level and dry matter of Mahalaxmi, Robusta and Grand Naine banana pseudostem. Three different cultivars of banana pseudostem have moisture content in the range of 94% - 96%. No significant difference was observed in the moisture content of three different varieties of banana pseudostem. High moisture level favors the quality and yield of the banana pseudostem fiber. Shivashankaret al., (2006)^[3] reported 93.2% - 94.6% moisture content in banana pseudostem. The highest dry matter was recorded in Mahalaxmi banana plant 6% and 4.4% in Robusta and Grand Naine. Poyyamozi and Kadirvel (1986)^[19] reported 8.6% dry matter in Robusta banana plant.

Crude fat and extractive

The crude fat content was found in the range of 5% - 6.7% (Table 1). The maximum crude fat (6.7%) content was recorded in Grand Naine banana pseudostem followed by Mahalaxmi (5.8%) and Robusta (5.0%). Aziz et al., (2011)^[8] found very low fat content (0.24%) in *Musa cuminata* balbisiana Colla. The extractive found in the range of (3.9% - 4.7%). The highest extractive found in Robusta (4.7%) followed by Grand Naine (4.2%) and Mahalaxmi cultivar (3.9%). Similar results were also reported by Rahman et al., (2014) [21] for *Musa paradisiaca* (3.25%). Khan et al., (2013)[22] found 3.52% extractive in banana pseudostem.

Carbohydrate and Protein content

Carbohydrate content was found in the range of 2.1 - 2.7 mg%. There was no significant difference of carbohydrate content was found in all three different varieties of banana plant. The maximum carbohydrate content was found in Grand Naine (2.7 mg%) followed by Robusta (2.5 mg%) and Mahalaxmi (2.1 mg%). The crude protein content was observed in the range of 80 -190 mg%. The research study shows that the Grand Naine have highest crude protein content (190 mg%). The crude protein content of Mahalaxmi and Robusta were observed to be 80 and 102 mg% respectively. Garget et al., (2023)[23] reported 2.8% crude protein content is present in banana pseudostem.

Mineral analysis

Banana plant is a rich source of minerals. Table 2 shows the mineral content of three different cultivars of banana pseudostem. The mineral value of banana pseudostem varies with the species of cultivar, stage of maturation and sample preparation and analysis. Sodium, potassium, magnesium, calcium and phosphorous are the macro-elements present in banana pseudostem. Amongst the different minerals, Calcium content was found to be maximum (1484.01, 1357.23 and 1298.67 mg/100 g of dry substrate) in all the three different varieties of banana (Mahalaxmi, Robusta and Grand Naine) respectively. Other macro-elements such as potassium was found in the range of (789.23-869.86 mg/100 g of dry substrate) followed by magnesium (260.51-301.84 mg/100 g of dry substrate), sodium (235.81-265.54 mg/100 g of dry substrate) and phosphorous (198.78-215.84 mg/100 g of dry substrate) in all the three different cultivars of banana. Ho et al., (2012)^[24] reported similar result for *Musa acuminata* X *balbisiana* cv. They found 1335.33mg/100 g of dry substrate calcium followed by potassium 944.12 mg/100 g of dry substrate, sodium 444.12 mg/100 g of dry substrate, magnesium 255.00 mg/100 g of dry substrate and phosphorous 137.82 mg/100 g of dry substrate. Selema



**Meghna N. Diarsa and Akshaya Gupte**

and Farago (1996)[25] reported highest potassium content 3810 mg/100 g of dry substrate in banana pseudostem of *Musa paradisiaca*.

Swelling capacity and Solubility of banana pseudostem

Swelling capacity and solubility of banana pseudostem is depicted in a Table 3. The swelling power is defined as the capacity of the starch present in the dry substrate to absorb the liquid molecules. The swelling capacity and solubility of the substrates depend on the chemical structure, particle size, pH and temperature. The highest swelling power was found in Mahalaxmi banana pseudostem 9.3 g of swollen particles/g of dry substrate and Robusta have lowest swelling capacity 7.3 g of swollen particles/g of dry substrate. Bello-Perez *et al.*, (1999)[26] reported that at the higher temperature the solubility enhances the swelling power of the substrate. Aziz *et al.*, (2011)[8] obtained 13.82 g swollen particles/g of dry substrate. Robusta have highest solubilized granules 3.6% in comparison to Mahalaxmi and Grand Naine. According to results, there is no significant difference found in the solubilized particle of Mahalaxmi 2.7% and Grand Naine 2.8%.

Water and Oil absorption capacity

Water and oil holding capacity defined as the potentiality of one gram of dry substrate to hold or retain water and oil with in its matrix under specific condition. Table 4 shows the water and oil holding capacity of the three banana cultivars. Water absorption capacity of banana pseudostem is found in the range of 10-19 gm of water/g of dry substrate. Grand Naine shows significantly higher water absorption capacity of 19.3 gm of water/g of dry substrate in comparison to Robusta 13.6 gm of water/g of dry substrate and Mahalaxmi 10.7 gm of water/g of dry substrate. Grand Naine cultivar have high water holding capacity which may be attributed to the high amount of the protein content or the presence of arabinoxylans. Arabinoxylans are free from the plant cell wall and have good capacity to absorb the water molecules (Choct, 1997)[27]. Aziz *et al.*, (2011)[8] reported 10.66 gm of water/g of dry substrate water holding capacity for *Musa acuminata* which is 1.8 time lower than the results obtained in our study for Grand Naine and Robusta.

The oil absorption capacity was found in the range of 4.1- 9.3 gm of oil/g of dry substrate. The maximum oil adsorption capacity was found in the Grand Naine banana pseudostem 9.3 gm of oil/g of dry substrate followed by Robusta and Mahalaxmi 6.4 and 4.1 gm of oil/g of dry substrate respectively. The maximum oil holding capacity is because of the protein content. The presence of protein enhances the oil absorption capacity by entrapping the oil particles[9,28]. Aziz *et al.*, (2011)[8] also found a similar oil holding capacity for the *Musa acuminata* 5.48 gm of oil/g of dry substrate.

Phytochemical analysis

Alkaloid is a secondary metabolite, which play an important role in human diet. Alkaloids exhibit many important characteristics such as anti-microbial, anti-insecticidal, anti-parasitic, anti-plasmodial and anti-oxidative. Table 5 represents the alkaloid content of three different varieties of banana pseudostem. The research study shows that the maximum amount of alkaloid content was found in Mahalaxmi banana pseudostem (7.8%) followed by Grand Naine (6.6%) and Robusta (6.2%). Onyemaet *et al.*, (2016)[29] reported 8.16% alkaloid content for banana pseudostem of *Musa acuminata*.

The flavonoid content of Mahalaxmi, Robusta and Grand Naine is exhibited in Table 5. The present research analysis shows that the flavonoid content of Mahalaxmi, Robusta and Grand Naine found in the range of 4.7-7.5 µg QE/g of banana pseudostem powder. The maximum flavonoid content was obtained in Mahalaxmi banana pseudostem 7.5 µg QE/g of banana pseudostem powder. The Grand Naine banana pseudostem contains 6.2 µg QE/g flavonoid content. However, the lowest amount of flavonoid was found in Robusta banana pseudostem (4.7 µg QE/g of banana pseudostem powder). Tannin are the class of phytochemicals found in the plant. They are known for their beneficial effects on the metabolism of protein and in enhancing the absorption of the amino acids in the small intestine[30]. The tannin content of three different varieties of banana pseudostem is shown in Table 5. Mahalaxmi banana



**Meghna N. Diarsa and Akshaya Gupte**

pseudostem contains maximum amount of tannin content (12.4%) than the Robusta (10.3%) and Grand Naine (9.5%). Onyemaet *al.*, (2016)[29] reported 9.13% tannin content for banana pseudostem of *Musa acuminata*.

Table 5 shows the phytate content of the Mahalaxmi, Robusta and Grand Naine banana pseudostem. The result shows that there was no significant difference observed between the three different varieties of banana pseudostem. Mahalaxmi banana pseudostem contains maximum phytate content (37.57 mg/100 g) followed by Robusta (35.75 mg/100 g) and Grand Naine (33.6 mg/100 g). Ramuet *al.*, (2017)[31] found similar result for *Musa* sp. CV. Nanjangud Rasa Bale). They reported 34.56 mg/100 g phytate content for banana pseudostem. The phenolic content of three different varieties of banana pseudostem is shown in the Table 5. The total phenolic content of three different varieties of banana pseudostem (Mahalaxmi, Robusta and Grand Naine) is found in the range from 60.43 - 76.57 mg GAE/g. The result shows that amongst the three different cultivars, Mahalaxmi showed higher total phenolic content (76.57 mg GAE/g). Grand Naine showed 65.82 mg GAE/g total phenolic content and Robusta showed very low phenolic content (60.43 mg GAE/g). Ramuet *al.*, (2017)[31] reported 188.64 mg/100 g of phenols in banana pseudostem of *Musa* sp. CV. Nanjangud Rasa Bale. Saravanan and Aradhya, (2011)[32] found phenolic content in the range of 7.58 - 291 mg GAE/ g of extract for eight different cultivars.

CONCLUSION

In the present study, three different cultivars of banana were collected from the local farm. Banana pseudostem is a main lignocellulosic waste of banana plant. The proximate analysis of banana pseudostem varies with the species of banana. Among three different cultivars of banana, Mahalaxmi has the highest cellulosic matter and lowest lignin content. A cellulose rich banana pseudostem fibers can be extracted from Mahalaxmi banana pseudostem and can be utilizing in the making of paper and in preparation of other cellulose derivatives. The banana pseudostem of all the cultivars has high nutritional value which allows their utilization as an animal feed. Banana pseudostem also contains good amount of phytochemical which enhances the medicinal properties.

DECLARATIONS

Ethics approval: Not applicable

Consent to participate: Not applicable

Availability of data and material: Not applicable

Funding: Not applicable

Acknowledgement: Not applicable

Conflict of interest: Authors declares no conflict of interest.

REFERENCES

1. Akpabio UD, Udiong DS, Akpakpan AE. The physicochemical characteristics of plantain (*Musa paradisiaca*) and banana (*Musa sapientum*) pseudostem wastes. *Adv in Nat and App Sci*2012;6:167-72.
2. Bilba K, Arsene MA, Ouensanga A. Study of banana and coconut fibers: Botanical composition, thermal degradation and textural observations. *BioresourTechnol*2007;98:58-68.
3. Shivashankar S, Nachane RP, Kalpana S. Composition and properties of fibre extracted from pseudostem of banana (*Musa* sp.). *J of HortSci*2006;1:95-08.
4. Lau BF, Kong KW, Leong KH, Sun J, He X, Wang Z, Mustafa MR, Ling TC, Ismail A. Banana inflorescence: Its bio-prospects as an ingredient for functional foods. *Trends Food Sci&Technol*2020;97:14-18.
5. Van Soest PJ, Robertson JB, Lewis BA. Methods for dietary fiber, neutral detergent fiber and non starch polysaccharides in relation to animal nutrition. *J Dairy Sci*1991;74:3583-87
6. Wang CF, Muhammad AUR, Liu ZY, Huang BZ, Cao BH. Effects of ensiling time on banana pseudo-stem silage chemical composition, fermentation and in *saccorumen* degradation. *J Anim Plant Sci*2016;26:339-46.





Meghna N. Diarsa and Akshaya Gupte

7. Pereira AL, do Nascimento DM, Cordeiro EM, Morais JP, Souza Filho M, Rosa MDF. Characterization of lignocellulosic materials extracted from the banana pseudostem. *IntSymp Natl Poly Comp*2010;7:1077-89
8. Aziz NAA, Ho LH, Azahari B, Bhat R, Cheng LH, Ibrahim MNM. Chemical and functional properties of the native banana (*Musa acuminata* × *balbisiana* Colla cv. *Awak*) pseudo-stem and pseudo-stem tender core flours. *Food Chem*2011;128:748-53.
9. Yadav RB, Kumar N, Yadav BS. Characterization of banana, potato, and rice starch blends for their physicochemical and pasting properties. *Cogent Food Agric*2016;2:1-7.
10. Chávez-González M L, Sepúlveda L, Verma DK, Luna-García HA, Rodríguez-Durán LV, Iliina A, Aguilar CN. Conventional and emerging extraction processes of flavonoids. *Processes*2020;8:434-40.
11. Al Amri FS, Hossain MA. Comparison of total phenols, flavonoids and antioxidant potential of local and imported ripe bananas. *Egypt J Basic ApplSci*2018;5:245-51.
12. Bello, F., Salami-Jaji, J. I., Sani, I., Abdulhamid, A., & Fakai, I. M. (2013). Evaluation of some Antinutritional factors in oil-free white *Sesamum indicum* L. seed cake. 2013; *Int J Food Nut Saf* 4:27-33.
13. Molina-Cortes A, Sanchez-Motta T, Tobar-Tosse F, Quimbaya M. Spectrophotometric estimation of total phenolic content and antioxidant capacity of molasses and vinasses generated from the sugarcane industry. *Waste Biomass Valori*2019;1-11.
14. Jayaprabha JS, Brahmakumar M, Manilal, VB. Banana pseudostem characterization and its fiber property evaluation on physical and bioextraction. *Journal of Natural Fibers*2011;8:149-60.
15. Pappu A, Patil V, Jain S, Mahindrakar A, Haque, R, Thakur VK. Advances in industrial prospective of cellulosic macromolecules enriched banana biofibre resources: A review. *Int J Bio Macro*2015;79:449-58.
16. Emaga TH, Robert C, Ronkart SN, Wathélet B, Paquot M. Dietary fibre components and pectin chemical features of peels during ripening in banana and plantain varieties. *Bioresource Technology*2008;99:4346-54.
17. Preethi P. Balakrishna M. Physical and chemical properties of banana fibre extracted from commercial banana cultivars grown in Tamilnadu State. *Agrotech*2011;11:1-3.
18. Viswanathan K, Kadirvel R, Chandrasekaran D. Nutritive value of banana stalk (*Musa cavendishi*) as a feed for sheep. *Anim Feed SciTechnol*1989;22(4):327-32.
19. Poyyamozi VS, Kadirvel R. The value of banana stalk as a feed for goats. *Anim Feed SciTechnol*1986;15(2):95-01.
20. Cordeiro N, Belgacem MN, Torres IC, Moura JCVP. Chemical composition and pulping of banana pseudo-stems. *Ind Crops Prod*2004;19(2):147-54.
21. Rahman MM, Islam T, Nayeem J, Jahan M. Variation of chemical and morphological properties of different parts of banana plant (*Musa paradisiaca*) and their effects on pulping. *Int J Lignocellul Prod*2014;1(2):93-03.
22. Khan MZH, Sarkar MAR, Ibne F, Imam A, Malinen RO. Fiber morphology and pulping study of banana pseudo-stem. *Int J Fiber Text Res*2013;3(1):31-43.
23. Garg M, Rajendren A, Singh V, Sharma MP, Sachdeva S, Nandan D. Physico chemical & sensory evaluation of nutritious balls prepared from banana pseudostem *EurChem Bull* 2023;12:4354-65
24. Ho LH, Aziah NA, Bhat R. Mineral composition and pasting properties of banana pseudo-stem flour from *Musa acuminata* × *balbisiana* cv. *Awak* grown locally in Perak, Malaysia. *Int Food Res J*2012;19(4):1479-85.
25. Selema M D, Farago ME. Trace element concentrations in the fruit peels and trunks of *Musa paradisiaca*. *Phytochem*1996;42(6):1523-31.
26. Bello-Pérez LA, Agama-Acevedo E, Sánchez-Hernández L, Paredes-López O. Isolation and partial characterization of banana starches. *J Agric Food Chem*1999;47(3):854-57.s
27. Choct M. Feed non-starch polysaccharides: chemical structures and nutritional significance. *Feed Milling Int*1997;191:13-16.
28. Voutsinas LP, Nakai S. A simple turbidimetric method for determining the fat binding capacity of proteins. *J Agric Food Chem*1983;31(1):58-63.
29. Onyema CT, OforCE, Okudo VC, Ogbuagu AS. Phytochemical and antimicrobial analysis of banana pseudostem (*Musa acuminata*). *J Pharm Res Int*2016;1-9.
30. Deng H, Jia P, Jiang J, Bai Y, Fan TP, Zheng X, Cai Y. Expression and characterisation of feruloyl esterases from *Lactobacillus fermentum* JN248 and release of ferulic acid from wheat bran. *Int J BiolMacromol*2019;38:272-77.





Meghna N. Diarsa and Akshaya Gupte

31. Ramu R, Shirahatti PS, Anilakumar KR, Nayakavadi S, Zameer F, Dhananjaya BL, Prasad MN. Assessment of nutritional quality and global antioxidant response of banana (*Musa sp.* CV. *Nanjangud Rasa Bale*) pseudostem and flower. *Pharmacogn Res*2017;9(1):74-83.
32. Saravanan K, Aradhya SM. Polyphenols of pseudostem of different banana cultivars and their antioxidant activities. *JAgric Food Chem*2011;59(8):3613-23.

Table 1. Compositional analysis of banana pseudostems.

	Mahalaxmi	Robusta	Grand Naine
Cellulose (%)	47.6 ± 0.8	40.9 ± 1.1	42.7 ± 1.2
Hemicellulose (%)	29.4 ± 1.3	28.9 ± 0.5	23.3 ± 1.9
Lignin (%)	7.7 ± 0.5	9.0 ± 0.2	8.8 ± 2.3
Ash (%)	4.5 ± 0.5	3.5 ± 0.07	2.7 ± 0.6
Extractive (%)	3.9 ± 0.2	4.7 ± 0.4	4.2 ± 0.1
Crude fat (%)	5.8 ± 0.2	5.0 ± 0.3	6.7 ± 0.2
Moisture content (%)	94.0 ± 0.02	95.5 ± 0.03	95.6 ± 0.9
Dry matter (%)	6.0 ± 0.02	4.4 ± 0.06	4.4 ± 0.2
Carbohydrate (mg%)	2.1 ± 0.05	2.5 ± 0.1	2.7 ± 0.1
Protein (mg%)	80 ± 6.5	102 ± 3.9	190 ± 8.1

All the values shown in the table are the mean of triplicate

Table 2. Mineral contents of three cultivars of banana pseudostem.

Minerals	Mahalaxmi	Robusta	Grand Naine
Sodium	265.54 ± 6.3	258.74 ± 2.7	235.81 ± 7.1
Potassium	869.86 ± 8.40	789.23 ± 16.2	812.46 ± 1.4
Calcium	1484.01 ± 5.98	1357.23 ± 0.2	1298.67 ± 9.2
Magnesium	260.51 ± 13.50	301.84 ± 2.3	298.54 ± 0.06
Phosphorous	198.78 ± 6.77	215.84 ± 3.6	201.85 ± 7.4
Manganese	3.1 ± 0.77	2.8 ± 0.14	2.1 ± 0.07
Iron	7.6 ± 0.56	6.8 ± 0.07	7.1 ± 0.07
Zinc	8.2 ± 0.77	8.9 ± 0.28	7.8 ± 0.1

All the values shown in the table are the mean of triplicate

All the values are given mg/100g of dry substrate

Table 3. Swelling capacity and solubility of three different banana cultivars.

Different variety of banana pseudostem	Swelling power (g of swollen particles/ g of dry substrate)	Solubility (%)
Mahalaxmi	9.3 ± 0.2	2.7 ± 0.1
Robusta	7.3 ± 0.1	3.6 ± 0.2
Grand Naine	8.4 ± 0.2	2.8 ± 0.07

All the results shown in the table are mean of the triplicate.

Table 4. Water and Oil holding capacity of banana pseudostems.

Different variety of banana pseudostem	WHC (gm of water/ g of dry substrate)	OHC (gm of oil/ g of dry substrate)
Mahalaxmi	10.7 ± 0.1	4.1 ± 0.1
Robusta	13.6 ± 0.07	6.4 ± 0.1
Grand Naine	19.3 ± 0.1	9.3 ± 0.07

All the results shown in the table are mean of the duplicate.

Water holding capacity (WHC) and oil holding capacity (OHC).



**Meghna N. Diarsa and Akshaya Gupte****Table 5. Phytochemical analysis of banana pseudostem**

Phytochemicals	Mahalaxmi	Robusta	Grand Naine
Alkaloid (%)	7.85	6.2	6.6
Flavonoids (mg QE/g substrate)	7.65	4.75	6.0
Tannin (%)	12.4	10.3	9.5
Phytate(mg/100 gm)	37.57	37.42	33.6
TPC (mg GAE/g)	76.57	60.43	65.82





Image Normalization with Noise Removal for Early Neurodegenerative Disease Detection

Divyasree S^{1*} and V Kathiresan²

¹Research Scholar, Department of Computer Science, Parks College, Tirupur (Affiliated to Bharathiar University, Coimbatore), Tamil Nadu, India.

²Principal, Department of Computer Science, A V P College of Arts and Science, Tirupur, (Affiliated to Bharathiar University, Coimbatore), Tamil Nadu, India.

Received: 08 Jun 2024

Revised: 12 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Divyasree S

Research Scholar,
Department of Computer Science,
Parks College, Tirupur
(Affiliated to Bharathiar University, Coimbatore),
Tamil Nadu, India.

E.Mail: sdivyasree0504@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Automated retinal layer segmentation has become a key technique for the early detection of neurodegenerative diseases such as Alzheimer's and Parkinson's. The dataset was collected from Kaggle repository. This study introduces a novel algorithm, the Bilateral Noise Element Neural Network (BNENN), designed to enhance segmentation accuracy and noise removal in retinal images, Alzheimer's and Parkinson's. The BNENN algorithm integrates bilateral filtering with neural network architecture to effectively suppress noise while preserving critical structural details in retinal images. By utilizing the advanced pattern recognition capabilities of neural networks, the proposed method achieves robust segmentation of retinal layers, even in the presence of significant noise and artifacts. The implementation has done with python. Experimental results on a diverse dataset demonstrate the superior performance of the BNENN algorithm in accurately segmenting retinal layers and removing noise, outperforming Innovative methods in terms of segmentation accuracy and robustness.

Keywords: Automated retinal layer segmentation, Alzheimer's, Bilateral Noise Element Neural Network, Neurodegenerative diseases, Parkinson's.





Divyasree and Kathiresan

INTRODUCTION

Front Temporal Dementia (FTD) and other neurodegenerative disorders cause cognitive and behavioral decline over time [1]. After years of asymptomatic progression, during which aberrant proteins gradually build up in the brain, neurodegenerative processes cause function loss [2]. This is followed by these alterations. Intervention with treatments that can delay or prevent the start of these illnesses might be possible if presymptomatic alterations in people could be reliably identified [3]. Nevertheless, it can be difficult to identify a group of presymptomatic people who are certain to get dementia. Researchers often look at those who can develop uncommon autosomal dominant dementias [4]. We can compare carriers and non-carriers at different stages of the illness because half of these people have the mutation. The frontal and temporal lobes, together with the insula, are involved early in all mutations, as the name suggests. Differences can be seen as early as 10 years before the typical age of symptom start [5-6]. On the other hand, other structures, such the thalamus, seem to be involved to varying degrees in the early stages of the illness [7]. Many types of FTD manifest clinically as an imbalance between the afflicted hemispheres, with greater atrophy within the affected hemisphere lending credence to this observation [8]. On the other hand, the afflicted side isn't always the same, and sometimes there isn't even any sign of an imbalance. It is important to consider this imbalance when trying to identify early alterations with any sensitivity [9] since it is likely to begin early in the illness process.

Nerve cells in the substantia nigra degrade in neurodegenerative disorders (NDDs), which affect the brain's ability to produce dopamine [10]. Movement problems are caused by this degeneration, which impacts the synthesis of dopamine [11]. Most people with Parkinson's disease have stiffness, bradykinesia, resting tremor, postural instability, and trouble walking [12]. Characteristic clinical features of HD include chorea, clumsiness, and stumbling [13]. In addition to the usual muscular atrophy, ALS can cause twitches and cramps, which can make walking difficult [14–15]. For all three diseases, the degree to which a person can walk depends on the severity of their illness. Diagnosing NDDs and gauging the degree of illness in an individual patient can be aided by analyzing gait characteristics. For patients to have access to the appropriate therapies, medications, and care, accurate diagnosis of NDDs is the most critical criteria [16–18]. It is also important to tailor motor symptom therapy to the patient's illness severity. Furthermore, it alters their appearance, which can result in incorrect diagnoses and a significant decline in patients' quality of life [19-21]. Disease identification and severity prediction can be assisted by AI-based models that use gait data. Even though there are a few of these models, most of them use one-dimensional gait data that has been analyzed and modified to identify disorders [22–26].

The main contribution of the paper is:

Image denoising using BNENN

The following is the organizational scheme for the rest of this work. In Section 2, many writers discuss various methods for detecting neurodegenerative diseases. In Section 3, the GMBF model is shown. Section 4 presents the results of the investigation. Results and future goals are discussed in Section 5, which finishes the section.

Motivation of the paper

An important motivation for this research is the critical requirement for reliable automated retinal layer segmentation in the detection of neurodegenerative diseases in their early stages, including Alzheimer's and Parkinson's. A new approach that successfully eliminates noise from retinal images while also greatly improving segmentation accuracy is the Bilateral Noise Element Neural Network (BNENN) method. With its combination of bilateral filtering and sophisticated neural network architecture, BNENN achieves better performance than previous approaches by overcoming noise and artifacts. Showing its potential influence on clinical applications, the end objective is to provide doctors a powerful tool to better early illness identification and monitoring.



**Divyasree and Kathiresan****Background study**

Baldacci, F. et al. [1] Numerous subtypes and clinical manifestations were produced by NDD, which were CNS proteinopathies, according to the kind, location, and dynamic spatio-temporal distribution of abnormal proteins. Potential pathophysiological disease biomarkers that might reveal pathogenetic processes at work in the early, asymptomatic phases of illness include the changed, misfolded proteins characteristic of NDD. Chudzik, A. et al. [3] Artificial Intelligence (AI) and digital technologies hold great promise for improving the speed and accuracy of neurodegenerative disease identification. This might pave the way for earlier therapies that halt the illness's development. Research was ongoing to confirm if digital biomarkers and prediction models powered by AI can effectively detect neurodegenerative disorders in their early stages. By analyzing trends in patient data, these technologies have the potential to create more tailored treatment programs.

Cicirelli, G., & D'Orazio, T. [4] in most cases, doctors or highly trained physiotherapists will only watch patients. The goal was to provide medical professionals with a tool that can aid in the diagnosis of neurodegenerative diseases utilizing this kind of automated system, which requires minimal human intervention and greatly facilitates remote analysis. Cury, C. et al. [6] In order to pinpoint potential presymptomatic variations in the thalamic shape, the author used a new approach of statistical shape analysis on a group of people with hereditary FTD. These authors research leads us to believe that variations become apparent five years before to the anticipated start of symptoms. These alterations were also discovered by volumetric analysis and these authors original shape analysis, but these authors technique demonstrated significance that persisted after multiple comparison correction.

Doroszkiewicz, J. et al. [7] the author highlight what was currently known about the most common genetic abnormalities and molecular markers that can be used to diagnose some NDs early on. In this article, the author will go over some of the newly discovered genes, molecular markers, and proteins in the fields of genetics and biochemistry. According to the data presented here, microRNAs (miRNAs) have the potential to be a "perfect biomarker" for neurological diseases. Fazlali, P. et al. [9] The author summarize what was currently known about the most common genetic abnormalities and molecular biomarkers for the early identification of certain NDs in this article. Newly discovered genes, molecular markers, and proteins in the fields of genetics and biochemistry were covered in this article. Based on what the author know about miRNAs from this review, they seem to be a "perfect biomarker" for neurological diseases. Kumar, A. et al. [13] The main reasons for NDs were the molecular alterations and reactions. Most of the studies conducted in this area center on studying genetic mutations, toxic protein accumulation, and neurotoxic chemical synthesis. According to the findings and discussion in this paper, nanotheranostics has great promise for the treatment of several neurodegenerative diseases. Modeling neurodegenerative diseases in animals, cells, and humans using nanotheranostics techniques has changed the game.

Makdissi, S. et al. [15] in the context of NDs, the microbiota-gut-brain axis was gaining more and more recognition. Nevertheless, further investigation was required to clarify the mechanisms by which bacteria interact with the host and to comprehend the interplay of the dietary factors, the microbiota, and the intestinal epithelium.

Problem definition

Existing methods such as GAN and **Bilateral Filter** approaches often face challenges in effectively suppressing noise while preserving critical structural details in retinal images. These methods can struggle with robust segmentation accuracy, especially in the presence of significant noise and artifacts, thereby limiting their efficacy in automated retinal layer segmentation for early neurodegenerative disease detection.

MATERIALS AND METHODS

In this section, we present the proposed method, the Bilateral Noise Element Neural Network (BNENN), designed for automated retinal layer segmentation and noise removal in the context of early neurodegenerative disease detection.





Divyasree and Kathiresan

Dataset collection

The dataset was collected from Kaggle website <https://www.kaggle.com/datasets/tourist55/alzheimers-dataset-4-class-of-images>

Image denoising using bilateral noise element neural network

One Recent advances in technology for image denoising is the Bilateral Noise Element Neural Network (BNENN). It is especially useful in retinal imaging, which is essential for the early diagnosis of neurodegenerative diseases. This fresh method combines neural network-based pattern recognition with bilateral filtering, which is well-known for its noise reduction and edge preservation capabilities. BNENN stands out when it comes to improving image quality. It does a great job of eliminating noise while keeping important structural features, which leads to very accurate segmentation. In this study, we claim that deterministic hidden units can be transformed into stochastic ones by introducing noise into them. Injecting noise into the training process causes the optimizer and the model to exhibit stochastic behavior. Using well-defined probabilistic formulations, we can assess the typical training technique and offer alternatives for improved optimization by characterizing deterministic hidden units with noise as stochastic hidden units.

$$z = g(h_{\phi}(x), \epsilon) \sim p_{\phi}(z|x) \text{-----} (1)$$

We use the notation $h_{\phi}(x)$ to indicate the hidden unit activations which take input x and model parameters ϕ . Combining the activation with noise—which is often derived from a certain probability distribution, such as the Gaussian distribution—creates a noise injection function. This assumption, when coupled with dropout, produces noise in the form of randomly selected hidden units in a layer and a randomly generated variable z representing the hidden layer's activation for a certain sample of dropout.

A neural network training with stochastic hidden units (z) must maximize the marginal likelihood over all of them if $L_{marginal} = \log E_{p_{\phi}(z|x)} [P_{\theta}(y|z, x)] \text{-----} (2)$

The probability of ground-truth y given input x and hidden units z is denoted as $P_M E_{p_{\phi}(z|x)}$, where M is the parameter of the output prediction model.

We use the reparameterization approach from for marginalization of noise-constructed stochastic hidden units. The marginalization is carried out over noise, which is provided by, and Eq. (2) is used in lieu of the random variable z .

$$L_{marginal} = \log E_{p(\epsilon)} [p_{\theta}(y|g(h_{\phi}(x), \epsilon), x)] \text{-----} (3)$$

where $p()$ is the noise distribution. The optimization of the marginal likelihood over noise is necessary for training a noise-injected neural network, according to Eq. (3).

Algorithm 1: Bilateral noise element neural network

Ⓞ **Input:**

- Noisy input image (X_{noisy})
- Ground truth clean image (X_{clean}) for supervised learning
- Parameters for bilateral filtering and neural network architecture
- Training data (optional)

Procedure:

- Apply bilateral filtering to the input image (X_{noisy}) to reduce noise while preserving edges and structural details.
- Optionally, train the BNENN model using supervised learning with pairs of noisy input images (X_{noisy}) and corresponding clean images (X_{clean}).





Divyasree and Kathiresan

Bilateral Filtering:

- The bilateral filter smooths an image while preserving edges by taking into account both spatial proximity and intensity similarity between pixels.
- The filtered pixel value $I(x)$ at position x is computed as:

$$I'(x) = \frac{1}{w(x)} \sum_{y \in S} I(y) \cdot G_{\sigma_s}(x - y) \cdot G_{\sigma_r}(|I(x) - I(y)|)$$

where:

- $I(x)$ and $I(y)$ are the intensity values of pixels at positions x and y , respectively.
- $w(x)$ is the normalization factor.
- G_{σ_s} and G_{σ_r} are Gaussian kernels for spatial and intensity domains, respectively.
- S is the spatial neighborhood of pixel x .

Ⓞ Output:

- Denoised output image ($X_{denoised}$)

RESULTS AND DISCUSSION

The implementation has done with python, in this section, we explore into the results from the application of the Bilateral Noise Element Neural Network (BNENN) algorithm for automated retinal layer segmentation and noise removal. Figure 2 illustrates the effectiveness of the Bilateral Noise Element Neural Network (BNENN) in enhancing brain images for neurodegenerative disease detection. The left panel shows the original brain input image, which contains noise and artifacts that obscure critical structural details. The right panel displays the same image after denoising with the BNENN algorithm, which integrates bilateral filtering with a neural network to suppress noise while preserving essential details. Figure 3 presents a retinal input image used for automated retinal layer segmentation in the early detection of neurodegenerative diseases. Captured using advanced retinal imaging techniques like optical coherence tomography (OCT), the image reveals multiple distinct retinal layers essential for visual function. Figure 4 presents a spiral input image used in the assessment of Parkinson's disease. This image is derived from a drawing task where individuals are asked to draw or trace a spiral, a test commonly used to evaluate motor function. The spiral pattern reveals critical details about the subject's motor control, such as tremors, irregularities, and deviations, which are characteristic of Parkinson's disease.

The table 2 and figure 5 shows comparing the denoising methods for Alzheimer's Disease (AD) images, three techniques—GAN, Bilateral Filter, and BNENN—were evaluated using PSNR, SSIM, and RMSE metrics. The BNENN method consistently outperformed the other methods. For the image 100.jpg, BNENN achieved a PSNR of 20.64, an SSIM of 0.96, and an RMSE of 0.10, while for 150.jpg, it achieved a PSNR of 20.95, an SSIM of 0.96, and an RMSE of 0.08. In contrast, the Bilateral Filter method scored lower with PSNR values of 18.21 and 19.67, SSIM values of 0.93 and 0.94, and RMSE values of 0.17 and 0.15 for 100.jpg and 150.jpg, respectively. The GAN method performed the least effectively, with PSNR values of 17.04 and 17.64, SSIM values of 0.91 and 0.92, and RMSE values of 0.18 for both images. These results indicate that BNENN provides the highest quality denoising for AD images, achieving the best balance of high PSNR, high SSIM, and low RMSE, thus preserving image quality and structural details most effectively.

The table 3 and figure 6 shows comparison of denoising methods for retinal disease images, three techniques—GAN, Bilateral Filter, and BNENN—were assessed using PSNR, SSIM, and RMSE metrics. The BNENN method demonstrated superior performance across all metrics. For the image 100.jpg, BNENN achieved a PSNR of 16.64, an SSIM of 0.97, and an RMSE of 0.15, while for 150.jpg, it achieved a PSNR of 16.86, an SSIM of 0.98, and an RMSE of 0.14. The Bilateral Filter method showed intermediate results with PSNR values of 14.21 and 15.67, SSIM values of 0.96 and 0.97, and RMSE values of 0.16 and 0.15 for 100.jpg and 150.jpg, respectively. The GAN method yielded the lowest performance, with PSNR values of 13.04 and 14.64, SSIM values of 0.95 for both images, and RMSE values of



**Divyasree and Kathiresan**

0.18 and 0.17. These findings indicate that BNENN is the most effective denoising method for retinal disease images, providing the highest image quality with the best balance of high PSNR, high SSIM, and low RMSE.

The table 4 and figure 7 shows evaluation of denoising methods for Parkinson's disease images, three techniques – GAN, Bilateral Filter, and BNENN – were analyzed using PSNR, SSIM, and RMSE metrics. The BNENN method outperformed the others significantly. For the image 100.jpg, BNENN achieved a PSNR of 14.99, an SSIM of 0.98, and an RMSE of 0.13, while for 150.jpg, it achieved a PSNR of 15.28, an SSIM of 0.98, and an RMSE of 0.12. The Bilateral Filter method showed intermediate performance with PSNR values of 13.92 and 14.01, SSIM values of 0.96 for both images, and RMSE values of 0.14 for both images. The GAN method had the lowest performance, with PSNR values of 12.84 and 13.24, SSIM values of 0.94 and 0.95, and RMSE values of 0.16 and 0.15 for 100.jpg and 150.jpg, respectively. These results indicate that BNENN is the most effective denoising method for Parkinson's disease images, providing superior image quality by achieving the highest PSNR, highest SSIM, and lowest RMSE values, thereby preserving image detail and clarity most effectively.

CONCLUSION

In Conclusion, the BNENN algorithm is a huge step forward for retinal image processing, especially when it comes to spotting neurodegenerative disorders like Alzheimer's and Parkinson's at an early stage. Retinal layer segmentation is made more accurate and reliable by BNENN, which solves the problem of noise reduction while keeping important structural information intact by combining bilateral filtering with neural network design. The findings of the trial confirm that BNENN outperforms the current state-of-the-art approaches, demonstrating its resilience when faced with images that are noisy or include artifacts. The therapeutic value of BNENN is highlighted by its shown potential in early disease identification and its effective use in properly segmenting retinal layers. The BNENN method outperformed the others significantly. For the image 100.jpg, BNENN achieved a PSNR of 14.99, an SSIM of 0.98, and an RMSE of 0.13, while for 150.jpg, it achieved a PSNR of 15.28, an SSIM of 0.98, and an RMSE of 0.12. With its potential to improve early detection and monitoring of neurodegenerative disorders, this algorithm not only pushes the technical boundary in medical imaging but also offers hope for improved patient outcomes via prompt intervention. Possible avenues for further investigation include honing this method even further, investigating its potential use with additional imaging modalities, and evaluating its efficacy in actual clinical practice.

REFERENCES

1. Baldacci, F., Lista, S., Vergallo, A., Palermo, G., Giorgi, F. S., & Hampel, H. (2019). A frontline defense against neurodegenerative diseases: the development of early disease detection methods. *Expert Review of Molecular Diagnostics*, 19(7), 559-563.
2. Candelise, N., Baiardi, S., Franceschini, A., Rossi, M., & Parchi, P. (2020). Towards an improved early diagnosis of neurodegenerative diseases: the emerging role of in vitro conversion assays for protein amyloids. *Acta Neuropathologica Communications*, 8, 1-16.
3. Chudzik, A., Śledzianowski, A., & Przybyszewski, A. W. (2024). Machine Learning and Digital Biomarkers Can Detect Early Stages of Neurodegenerative Diseases. *Sensors*, 24(5), 1572.
4. Cicirelli, G., & D'Orazio, T. (2022). A low-cost video-based system for neurodegenerative disease detection by mobility test analysis. *Applied Sciences*, 13(1), 278.
5. Collin, F. (2019). Chemical basis of reactive oxygen species reactivity and involvement in neurodegenerative diseases. *International journal of molecular sciences*, 20(10), 2407.
6. Cury, C., Durrleman, S., Cash, D. M., Lorenzi, M., Nicholas, J. M., Bocchetta, M., ... & Warren, J. (2019). Spatiotemporal analysis for detection of pre-symptomatic shape changes in neurodegenerative diseases: Initial application to the GENFI cohort. *NeuroImage*, 188, 282-290.





Divyasree and Kathiresan

7. Doroszkiewicz, J., Groblewska, M., & Mroczo, B. (2022). Molecular biomarkers and their implications for the early diagnosis of selected neurodegenerative diseases. *International Journal of Molecular Sciences*, 23(9), 4610.
8. Erdaş, Ç. B., Sümer, E., & Kibaroglu, S. (2021). Neurodegenerative disease detection and severity prediction using deep learning approaches. *Biomedical Signal Processing and Control*, 70, 103069.
9. Fazlali, P., Mahdian, A., Soheilifar, M. S., Amininasab, S. M., Shafiee, P., Wani, I. A., & AL-Mokaram, A. M. A. A. (2022). Nanobiosensors for early detection of neurodegenerative disease. *Journal of Composites and Compounds*, 4(10), 24-36.
10. Kamagata, K., Andica, C., Kato, A., Saito, Y., Uchida, W., Hatano, T., ... & Aoki, S. (2021). Diffusion magnetic resonance imaging-based biomarkers for neurodegenerative diseases. *International Journal of Molecular Sciences*, 22(10), 5216.
11. Kavungal, D., Magalhães, P., Kumar, S. T., Kolla, R., Lashuel, H. A., & Altug, H. (2023). Artificial intelligence-coupled plasmonic infrared sensor for detection of structural protein biomarkers in neurodegenerative diseases. *Science Advances*, 9(28), eadg9644.
12. Kovacs, G. G. (2019). Molecular pathology of neurodegenerative diseases: principles and practice. *Journal of clinical pathology*, 72(11), 725-735.
13. Kumar, A., Chaudhary, R. K., Singh, R., Singh, S. P., Wang, S. Y., Hoe, Z. Y., ... & Dai, X. (2020). Nanotheranostic applications for detection and targeting neurodegenerative diseases. *Frontiers in Neuroscience*, 14, 305.
14. Mahesh, N., & Donati, R. (2022). Neurodegenerative diseases and potential early detection methods. *Journal of Student Research*, 11(4).
15. Makdissi, S., Parsons, B. D., & Di Cara, F. (2023). Towards early detection of neurodegenerative diseases: A gut feeling. *Frontiers in cell and developmental biology*, 11, 1087091.
16. Sharma, V., Nikolajeff, F., & Kumar, S. (2023). Employing nanoparticle tracking analysis of salivary neuronal exosomes for early detection of neurodegenerative diseases. *Translational Neurodegeneration*, 12(1), 7.
17. Shusharina, N., Yukhnenko, D., Botman, S., Sapunov, V., Savinov, V., Kamyshov, G., ... & Voznyuk, I. (2023). Modern methods of diagnostics and treatment of neurodegenerative diseases and depression. *Diagnostics*, 13(3), 573.
18. Sidorova, Y., & Domanskyi, A. (2020). Detecting oxidative stress biomarkers in neurodegenerative disease models and patients. *Methods and Protocols*, 3(4), 66.
19. Song, C., Que, S., Heimer, L., & Que, L. (2020). On-chip detection of the biomarkers for neurodegenerative diseases: Technologies and prospects. *Micromachines*, 11(7), 629.
20. Summers, M. J., Madl, T., Vercelli, A. E., Aumayr, G., Bleier, D. M., & Ciferri, L. (2017). Deep machine learning application to the detection of preclinical neurodegenerative diseases of aging. *DigitCult-Scientific Journal on Digital Cultures*, 2(2), 9-24.
21. Syam, V., Safal, S., Bhutia, O., Singh, A. K., Giri, D., Bhandari, S. S., & Panigrahi, R. (2023). A non-invasive method for prediction of neurodegenerative diseases using gait signal features. *Procedia computer science*, 218, 1529-1541.
22. Voet, S., Srinivasan, S., Lamkanfi, M., & van Loo, G. (2019). Inflammasomes in neuroinflammatory and neurodegenerative diseases. *EMBO molecular medicine*, 11(6), e10248.
23. Yagis, E., De Herrera, A. G. S., & Citi, L. (2019, November). Generalization performance of deep learning models in neurodegenerative disease classification. In 2019 IEEE international conference on bioinformatics and biomedicine (BIBM) (pp. 1692-1698). IEEE.
24. Zhong, Y., Liu, L., Zhao, D., & Li, H. (2020). A generative adversarial network for image denoising. *Multimedia Tools and Applications*, 79, 16517-16529.
25. Elhoseny, M., & Shankar, K. (2019). Optimal bilateral filter and convolutional neural network based denoising method of medical image measurements. *Measurement*, 143, 125-135.
26. K J Eldho, S Nithyanandh, (2024), Lung Cancer Detection and Severity Analysis with a 3D Deep Learning CNN Model Using CT-DICOM Clinical Dataset, "Indian Journal of Science and Technology", 2024;17(10):899-910.





Divyasree and Kathiresan

Table 1: Survey of Early Detection Methods for Neurodegenerative Diseases

Author	Year	Methodology	Advantage	Limitation
Baldacci et al.	2019	Early disease detection methods	Protection against neurodegenerative disorders at the forefront	Limited coverage of diverse disease detection methods
Candelise et al.	2020	Assays for the conversion of protein amyloids in vitro	Improved early diagnosis of neurodegenerative diseases	Reliance on in vitro assays, cannot fully represent in vivo conditions
Chudzik et al.	2024	Artificial Intelligence and Electronic Biomarkers	Identifying neurodegenerative disorders at an early stage	Dependency on accurate digital biomarker identification
Cicirelli & D’Orazio	2022	Low-cost video-based system for mobility analysis	Cost-effective neurodegenerative disease detection	Limited scalability and accuracy in complex mobility tests
Collin, F.	2019	The molecular foundation of ROS	Insight into neurodegenerative disease pathology	Lack of direct applicability to early disease detection

Table 2: Denoising value comparison on AD

Denoising value comparison on AD				
		PSNR	SSIM	RMSE
GAN	100.jpg	17.04	0.91	0.18
	150.jpg	17.64	0.92	0.18
Bilateral Filter	100.jpg	18.21	0.93	0.17
	150.jpg	19.67	0.94	0.15
BNENN	100.jpg	20.64	0.96	0.10
	150.jpg	20.95	0.96	0.08

Table 3: Denoising value comparison on retinal disease

Denoising value comparison on retinal disease				
		PSNR	SSIM	RMSE
GAN	100.jpg	13.04	0.95	0.18
	150.jpg	14.64	0.95	0.17
Bilateral Filter	100.jpg	14.21	0.96	0.16
	150.jpg	15.67	0.97	0.15
BNENN	100.jpg	16.64	0.97	0.15
	150.jpg	16.86	0.98	0.14

Table 4: Denoising value comparison on parkinson’s disease

Denoising value comparison on parkinson’s disease				
		PSNR	SSIM	RMSE
GAN [24]	100.jpg	12.84	0.94	0.16
	150.jpg	13.24	0.95	0.15
Bilateral Filter [25]	100.jpg	13.92	0.96	0.14





Divyasree and Kathiresan

	150.jpg	14.01	0.96	0.14
BNENN	100.jpg	14.99	0.98	0.13
	150.jpg	15.28	0.98	0.12

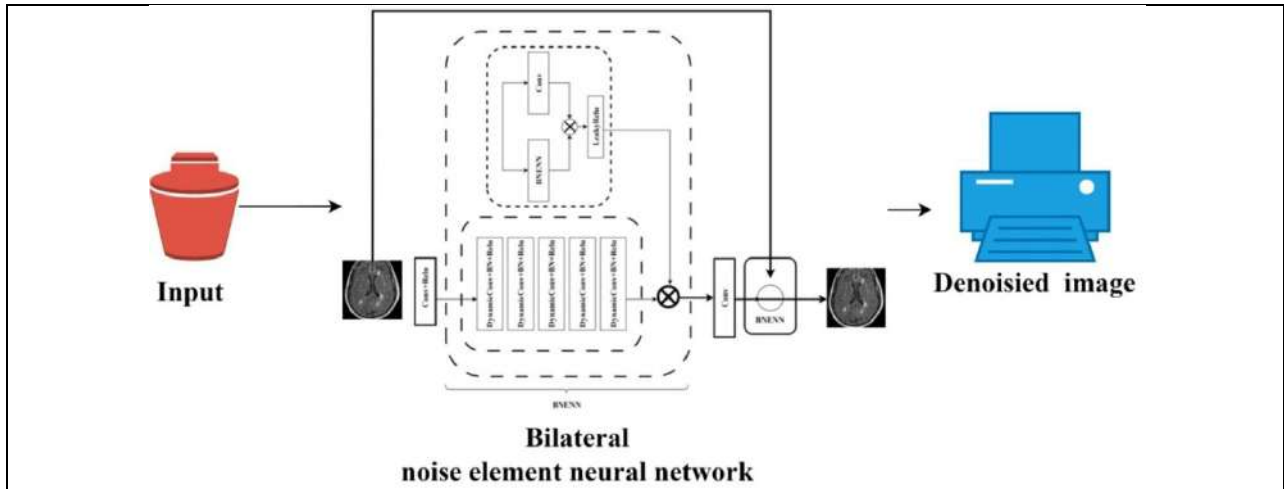


Figure 1: Proposed workflow architecture

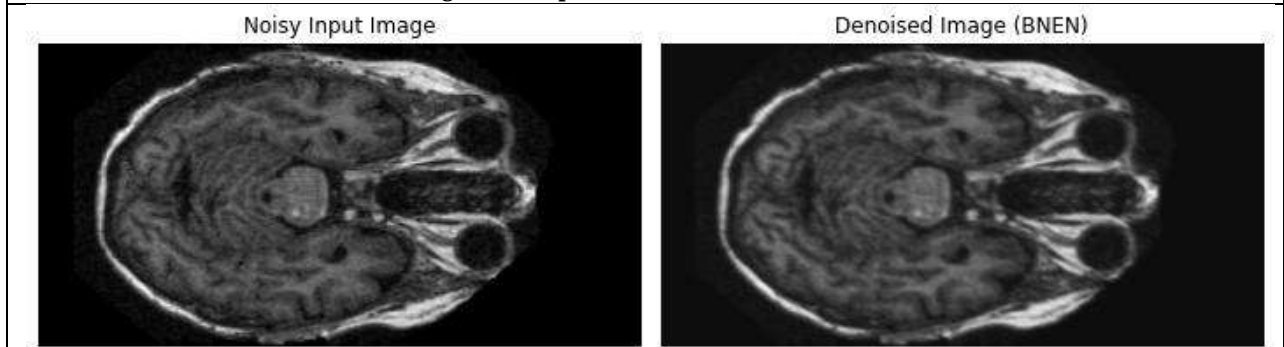


Figure 2: Brain image



Figure 3: Retinal image





Divyasree and Kathiresan

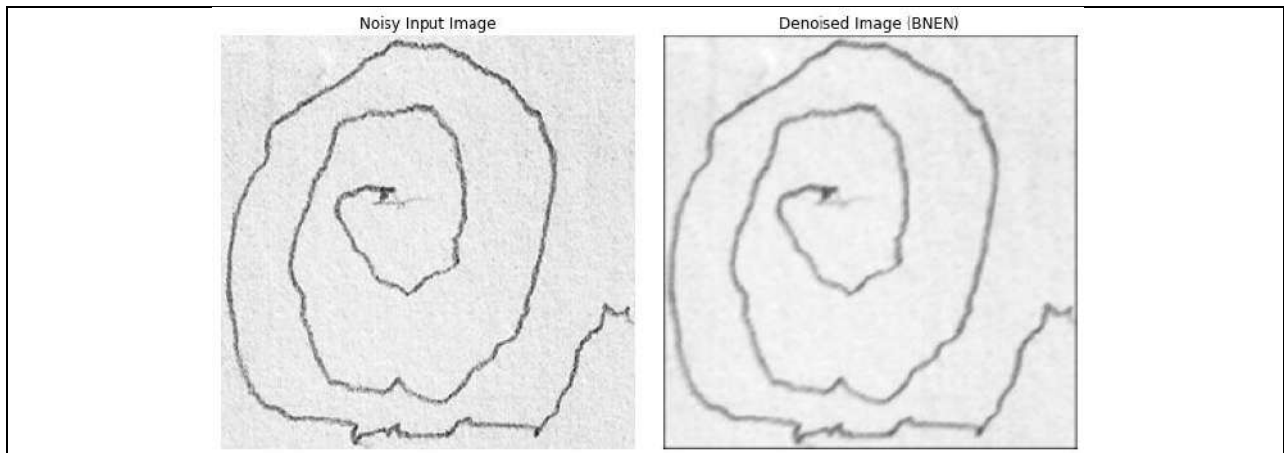


Figure 4: Spiral image

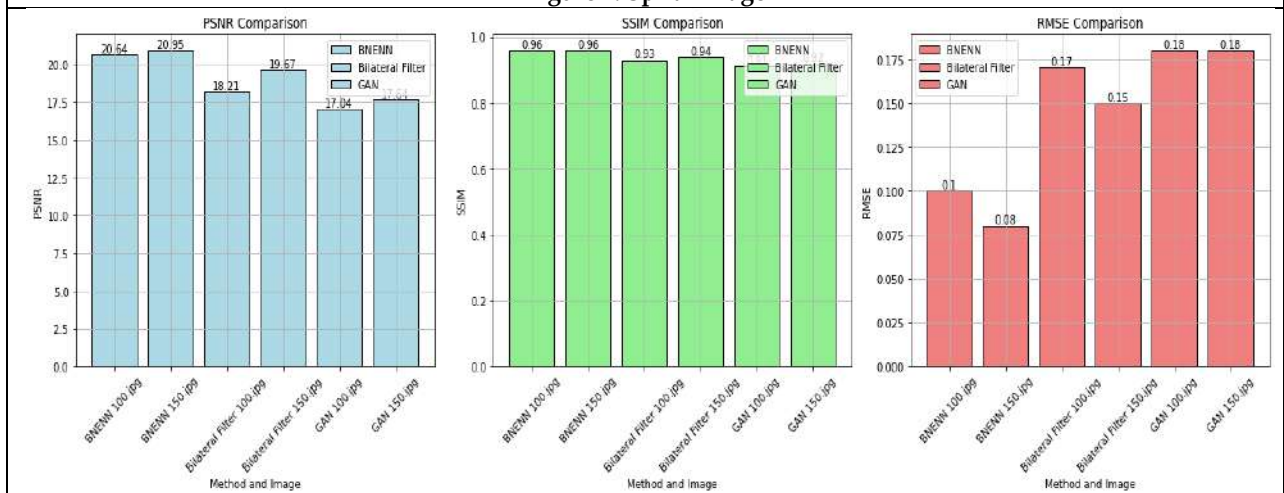


Figure 5: Denoising value comparison chart for AD

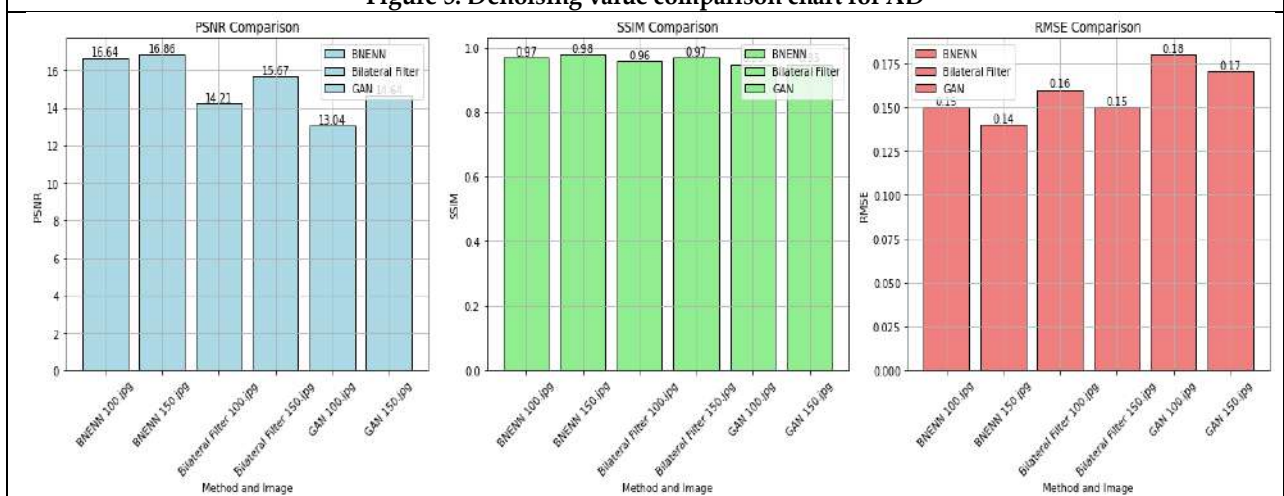


Figure 6: Denoising value comparison chart for retinal disease





Divyasree and Kathiresan

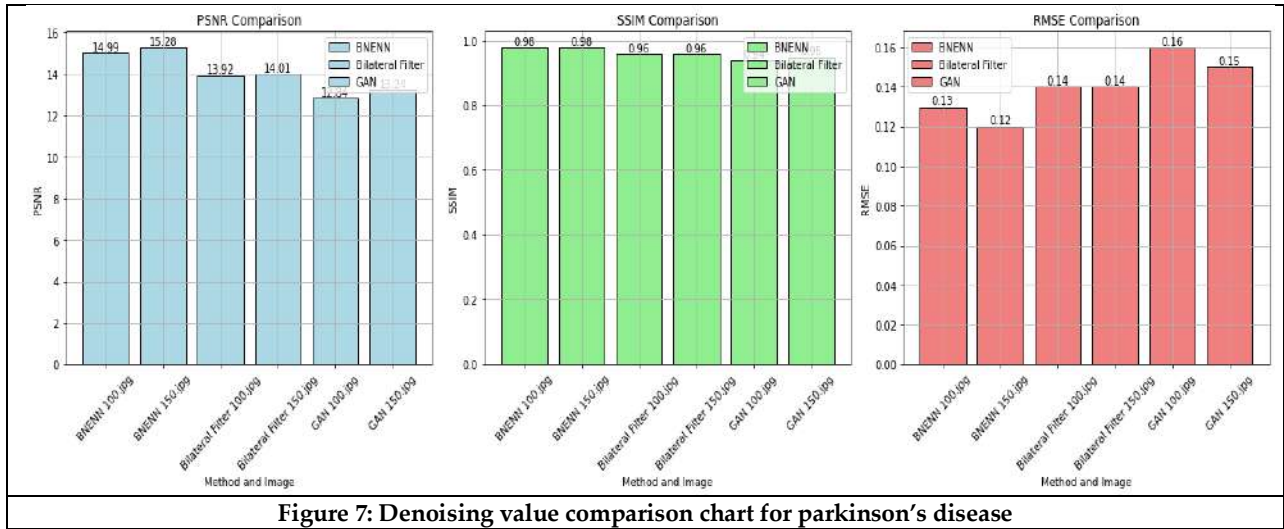


Figure 7: Denoising value comparison chart for parkinson’s disease





Seasonal Variations in Soil Properties Across Altitudinal Gradient of Current Jhum Fields in Champhai District, Mizoram

L K Thang Ngaihte^{1*}, Elizabeth Nemhoihkim¹, O.P. Tripathi² and B.P. Mishra³

¹Research Scholar, Department of Environmental Science, School of Earth Science and Natural Resource Management (SES & NRM), Mizoram University, Mizoram, India.

²Head, Department of Environmental Science, School of Earth Science and Natural Resource Management (SES & NRM), Mizoram University, Mizoram, India.

³Dean, School of Earth Science and Natural Resource Management (SES & NRM), Mizoram University, Mizoram, India.

Received: 12 Jun 2024

Revised: 16 Aug 2024

Accepted: 16 OCT 2024

Address for Correspondence

L K Thang Ngaihte

Research Scholar,

Department of Environmental Science,

School of Earth Science and Natural Resource Management (SES & NRM),

Mizoram University, Mizoram, India.

Email: ngaihtelk@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Seasonal variations in soil properties were investigated across altitudinal gradients of current jhum fields in Champhai district, Mizoram, India. Soil samples were collected from three locations during the pre-monsoon, monsoon, and postmonsoon seasons (Khawkawn, NE Khawdungsei, and Kawlbem, respectively). The soil texture ranged from loamy sand to sandy loam. The soil moisture content was highest during the monsoon season ($21.52 \pm 0.71\%$ to $24.61 \pm 1.78\%$), while the water holding capacity peaked in the postmonsoon period ($37.81 \pm 0.15\%$ to $39.2 \pm 0.54\%$). Bulk density was highest during the monsoon season (1.44 ± 0.01 to 1.10 ± 0.03 g/cm³). The soil pH decreased from the pre-monsoon to postmonsoon. The soil organic carbon content was highest in the pre-monsoon season ($1.53 \pm 0.25\%$ to $2.79 \pm 0.06\%$). The available nitrogen (146.35 ± 10.45 to 653.33 ± 17.02 kg/ha), available phosphorus (20.79 ± 0.82 to 34.49 ± 0.77 kg/ha), and available potassium (141.59 ± 1.97 to 230.82 ± 1.54 kg/ha) were highest during the monsoon season. The results emphasise the dynamic nature of the soil and the need for season-specific agricultural strategies for sustainability. They also contribute to the understanding of seasonal soil dynamics in jhum cultivation, a traditional practice central to Mizoram's culture and livelihood, and provide insights for developing techniques to enhance productivity and sustainability in the face of the challenges posed by declining fallow periods and environmental degradation.

Keywords: Jhum cultivation, Soil quality, Seasonal variations, Champhai, Mizoram.





Thang Ngaihte *et al.*,

INTRODUCTION

Shifting cultivation, locally known as jhum cultivation, is a predominant agricultural practice in Mizoram, India, where a significant portion of the population (approximately 60%) depends largely on their livelihood [1]. This practice involves clearing forestland followed by burning and cultivating a diverse array of crops, rice, maize, pulses, vegetables, ginger, sesame, cotton, etc, for a period of 1-2 years before abandoning the plot for forest re growth and shifting to a new area [1, 2]. Approximately 54% of Mizoram's arable land is used for jhum agriculture [2], which is generally practiced on steep slopes and is deeply intertwined with Mizo culture, with major festivals linked to the various stages of jhum operations [3]. However, the sustainability of jhum cultivation has been compromised owing to a significant reduction in fallow periods, have decreased from 20-25 years [4] to only 3-5 years [1]. This has led to declining yields and increased environmental degradation through soil erosion and watershed siltation [5]. Consequently, the diminishing productivity and sustainability of jhum cultivation in Mizoram's steep terrain necessitates interventions or transitions to more sustainable agricultural practices [6].

Numerous studies have consistently shown the impact of altitude, which significantly influences soil properties, along with other parameters such as texture, pH, and nutrients [7, 8, 9, 10]. Additionally, seasonal variations play a pivotal role in shaping soil quality, influencing microbial activity, abundance, and community structure, which are key indicators of soil health and fertility [11, 12]. The Champhai region in Mizoram, India, presents a unique case for studying the impact of season on soil quality within the context of jhum cultivation. This study aimed to evaluate the seasonal variations in soil properties across altitudinal gradients of jhum fields in the study area and discuss their implications for developing sustainable soil management strategies tailored to local conditions. These findings are expected to contribute valuable insights to the body of knowledge on sustainable agricultural practices in hilly and mountainous regions.

MATERIALS AND METHODS

Study Site

The study was conducted in Champhai District, Mizoram, which is commonly known as the 'Rice Bowl of Mizoram'. The district spans an area of 3185.83 km² and lies between 93°0'3" E to 93°26'10" E and 23°0'33" N to 24°5'4" N. According to the 2011 Census, Champhai has a population of 125,745. Despite its significant population, Champhai is the most underdeveloped district in the state, with a substantial number of households relying either directly or indirectly on agricultural activities, primarily jhum cultivation practiced on hill slopes. The district in Mizoram has the highest dependency on this cultivation method at 57.2% [13].

The sampling sites were selected to represent the typical jhum fields in the region, spanning an altitudinal range with varying topography. The selection criteria for the sampling sites included their representation of the commonly practiced jhum cultivation in the district, accessibility, and coverage of the different altitudinal zones. Table 1 and Figure 1 show the locations of the three sampling sites: - Khawkawn (KK), NE Khawdungsei (NE), and Kawlbem (KB) that are spread across different elevation.

The undulating topography of the broken mountain ranges and valleys in Champhai is suitable for crop cultivation, and its extensive forest cover, constituting 83.89% of its geographical area, contributes to its humus-rich soil [14]. A study conducted in this region revealed that the most common jhum field sizes range from 1.0 to 2.0 hectares [14]. The district experiences a moderate climate, with average summer temperatures of 23.5°C (April-June) and average winter temperatures of 15.7°C (November-February), accompanied by heavy rainfall influenced by the southwest monsoon, with an average annual rainfall of 2346.2 mm.



**Thang Ngaihte et al.,**

Soil Sampling and Analysis

Soil samples were taken in replicate from the sampling sites soon after the *jhumias* slash-and-burn activity, covering the pre-monsoon season (February–May), monsoon season (June–September), and postmonsoon season (October–January). In each season, two (2) soil samples from two depths (0-15 cm and 15-30 cm) were collected from each site and stored and air-dried for analysis. Standard techniques were used to analyse the physicochemical properties of the soil samples. These techniques include pH measurements using a digital pH meter, bulk density, soil texture [15], soil moisture content, water-holding capacity [16], soil organic carbon [17] available nitrogen [18], available phosphorus [19], and available potassium [20].

Statistical analysis

General statistical parameters such as the mean and standard error of mean (SEM), were calculated for each sampling site. Pearson Correlation was used for correlation analysis of the effect of season on the soil parameters, and the multiple analysis of variance (ANOVA) and Fisher least significant difference (LSD) tests, aimed at distinguishing between treatment means, were performed at a significance level of 5% ($p \leq 0.05$) using SPSS 27.

RESULTS AND DISCUSSION

The physical and chemical properties, Spearman's correlation analysis, MANOVA and LSD post-hoc test of the soils across seasons on selected *jhum* fields were carried out and data were presented and discussed.

Physical Properties of Soil

The soil texture in the Champhai *jhum* fields varies from loamy sand to sandy loam in nature. Among the sites, the texture changed from loamy sand in the pre-monsoon and monsoon to sandy loam in the post-monsoon period at Khawkawn (KK). At Khawdungsei (NE), the texture was consistent with loamy sand. At Kawlbem (KB), similar to KK, the texture varied from loamy sand to sandy loam in the post monsoon season. The soil moisture content (SMC) exhibited significant variations across seasons, with the highest values recorded during the monsoon season and the lowest values during the pre-monsoon season. The SMC ranged from $15.15 \pm 0.53\%$ to $28.89 \pm 0.78\%$ across all sites, depths, and seasons (Table 2).

The middle altitude (NE) exhibited the highest SMC values during the monsoon season compared with those at other altitudes, and the SMC across all sites and seasons showed that the SMC in the surface layer (0-15 cm) was greater than that in the subsurface (15-30 cm) layer (Table 2). The analysis of variance (ANOVA) results revealed a significant effect of season on SMC ($F = 8.451$, $p = 0.001$) (Table 5). The SMC values during the monsoon season were significantly greater than those during the pre-monsoon ($MD = 3.55$, $p < 0.001$) and post-monsoon ($MD = 4.43$, $p < 0.001$) seasons, as per LSD post-hoc test (Table 6).

The WHC values ranged from $30.28 \pm 0.03\%$ to $44.71 \pm 0.75\%$ across all sites and seasons (Table 2). The WHC showed altitudinal variation as it increased with increasing altitude, with higher altitudes having the highest WHC across seasons and depth. The WHC in the surface layer was generally greater than that in the subsurface layer across all sites and seasons (Table 2). The WHC exhibited a positive correlation with the season ($r = 0.582$, $p < 0.01$) (Table 4). The ANOVA results showed a significant effect of season on the WHC ($F = 16.614$, $p < 0.001$) (Table 5). The post-hoc LSD test further revealed that the WHC values during the pre-monsoon season were significantly lower than those during the monsoon ($MD = -4.15$, $p < 0.001$) and post-monsoon ($MD = -4.88$, $p < 0.001$) seasons (Table 6). The bulk density (BD) ranged from $0.89 \pm 0.01 \text{ g/cm}^3$ to $1.45 \pm 0.02 \text{ g/cm}^3$ across all sites, depths and seasons (Table 2). BD decreased with increasing altitude across depth and season, while BD in the subsurface layer was greater than that in the surface layer across all sites and seasons. The ANOVA did not show a significant effect of season on BD ($F = 1.461$, $p = 0.242$) (Table 5).



**Thang Ngaihte et al.,**

Chemical Properties of Soil

The soil pH ranged from 5.48 ± 0.03 to 6.21 ± 0.02 across all sites and seasons (Table 3). The pH exhibited altitudinal variation, generally decreasing from lower to middle altitudes, but increasing from middle to higher altitudes across seasons and depths. The soil pH in the surface layer was greater than that in the subsurface layer across all sites and seasons (Table 3). ANOVA revealed a significant effect of season on soil pH ($F = 24.950$, $p < 0.001$) (Table 5). The post-hoc LSD test further revealed that the soil pH values during the pre-monsoon season were significantly greater than those during the monsoon ($MD = 0.37$, $p < 0.001$) and post-monsoon ($MD = 0.16$, $p < 0.001$) seasons, and that the soil pH during the post-monsoon season was significantly higher than that during the monsoon season ($MD = 0.21$, $p < 0.001$) (Table 6).

The soil organic carbon (SOC) content of the soils ranged from $0.76 \pm 0.10\%$ to $2.92 \pm 0.04\%$ across all sites, depths, and seasons (Table 3). SOC showed altitudinal variation, as the SOC content in the middle latitudes showed greater across all seasons and depths than in the lower and higher altitudes, while SOC content in the surface layer was greater than that in the subsurface layer across all sites and seasons (Table 3). Although the ANOVA did not reveal a significant effect of season on SOC ($F = 1.789$, $p = 0.177$) (Table 5), SOC exhibited a general decreasing trend with increasing soil depth, which is consistent with the typical distribution of organic matter in soil profiles.

The available nitrogen (AN) content in the soils exhibited significant seasonal variation, with the highest values observed during the monsoon season and the lowest during the pre-monsoon season (Table 3). The AN ranged from 135.89 ± 10.45 kg/ha to 669.01 ± 20.91 kg/ha across all sites and seasons and the AN showed altitudinal variation as it increased with increasing altitude (Table 3), across all seasons and sites, the surface layer had greater AN content than did the subsurface layer (Table 3). ANOVA revealed a significant effect of season on AN ($F = 19.391$, $p < 0.001$) (Table 5), and the post-hoc LSD test further confirmed that AN levels during the monsoon season were significantly higher than those during both the pre-monsoon ($MD = 213.87$, $p < 0.001$) and post-monsoon ($MD = 194.71$, $p < 0.001$) seasons (Table 6).

The available phosphorus (AP) content in the soils exhibited a distinct seasonal pattern, with higher values observed during the monsoon season than the pre-monsoon and post-monsoon seasons (Table 3). The AP ranged from 18.10 ± 0.48 kg/ha to 35.75 ± 1.16 kg/ha across all sites, depths and seasons (Table 3). The AP showed altitudinal variation as it increased with increasing altitude, and the AP content in the surface layer was greater than in the subsurface layer across altitudes and seasons (Table 3). ANOVA revealed a significant effect of season on AP ($F = 27.154$, $p < 0.001$) (Table 5). The AP during the monsoon season was significantly greater than that during both the pre-monsoon ($MD = 8.65$, $p < 0.001$) and post-monsoon ($MD = 4.56$, $p < 0.001$) seasons, as per the LSD post-hoc test (Table 6).

The available potassium (AK) content in the soils exhibited seasonal variation, with higher values observed during the monsoon season than during the pre-monsoon and post-monsoon seasons (Table 3). The AK values ranged from 138.28 ± 1.16 kg/ha to 248.14 ± 3.39 kg/ha across all sites, depths and seasons (Table 3), and the AK exhibited greater altitudinal variation at middle altitudes than at higher and lower altitudes across all seasons and depths (Table 3). The AK also varied with in depth, as the AK content in the surface layer was greater than that in the subsurface layer across sites and seasons (Table 3). ANOVA revealed a significant seasonal effect on AK ($F = 6.117$, $p = 0.004$) (Table 5). The post-hoc LSD test further confirmed that AK levels during the monsoon season were significantly greater than those during the pre- ($MD = 34.38$, $p < 0.001$) and post-monsoon ($MD = 16.39$, $p < 0.001$) seasons (Table 6).

The observed seasonal patterns in soil properties were consistent with findings from tropical and subtropical regions [21, 22, 23, 24], which exhibit greater nutrient availability, particularly nitrogen and phosphorus, during the monsoon season due to increased microbial activity and mineralisation processes facilitated by favourable soil moisture conditions [25, 26, 27, 28, 29, 30]. The shifts in soil texture, with higher sand content during the monsoon season, can be attributed to the erosive effects of heavy rainfall selectively removing finer particles, such as silt and clay, leaving behind coarser sand fractions, as observed in other tropical regions with distinct dry and wet seasons [31].





Thang Ngaihte et al.,

The higher SMC and WHC during the monsoon and post-monsoon seasons, respectively, align with studies in which soil moisture dynamics are influenced by seasonal rainfall patterns [32, 33, 34]. A study in the lower Siwalik foothills of the Himalayas reported higher soil SOC for different land use systems during the pre-monsoon season [35].

The observed seasonal patterns in soil properties are not limited to the jhum cultivation system but have also been reported across various land use systems, including conventional agriculture, agro forestry, and natural ecosystems.[36] conducted a study in the lower Shivalik foothills of Punjab, India, and reported significant seasonal variations in the physical and chemical properties of soil under different land-use systems, including farm forestry and horticulture systems. They observed a higher soil moisture content and nutrient availability during the monsoon season, which is consistent with the findings of the present study. A study conducted by [37] in the Brazilian Amazon also revealed significant seasonal changes in soil moisture, nutrient availability, and microbial activity across different land-use systems, including native forests, agro forestry systems, and pastures. These seasonal variations were primarily driven by contrasting rainfall patterns and temperature regimes between the dry and wet seasons, similar to the observations made in the present study.

It is important to note that the observed seasonal variations in soil properties have implications for soil fertility management and agricultural productivity [23]. investigated the influence of seasonal fluctuations on soil characteristics and microbial populations in tropical dry deciduous forests and reported that the SMC dramatically increases in the summer, thereby increasing the amounts of SOC and macro- and micronutrients [38], reported that land-use type, soil depth, and season significantly influenced microbial activity, biomass of different soil layers, soil properties, and microbial biomass carbon, all of which exhibited strong seasonality, highlighting the need for season-specific nutrient management strategies to optimise crop yields and maintain soil fertility.

Although the current study provides insightful information about the dynamics of soil properties in the Champhai, Mizoram's jhum cultivation systems, it is important to recognise its potential limitations, including the absence of long-term monitoring data and the impact of variables, such as vegetation cover and management techniques, on soil properties. Future research should consider incorporating long-term monitoring of soil properties across multiple growing seasons and evaluating the interactive effects of land use practices and seasonal variations on soil quality and productivity.

CONCLUSION

This study revealed significant seasonal variations in key soil properties, including moisture, nutrient content, and pH, across altitudinal gradients. These findings demonstrate the dynamic nature of soil and the necessity of season-specific soil management strategies to improve soil productivity and sustainability. Although this study provides valuable insights, it is important to acknowledge its limitations, such as the lack of long-term data and the potential influences of vegetation and management practices. To address these limitations, future research should incorporate multiyear monitoring programs and investigate the interactive effects of various factors on soil quality. This study emphasises the need for a comprehensive soil-monitoring program that considers seasonal and topographic variations in soil properties. Such a program could inform policymaking and the development of tailored soil management methods for jhum farming, while supporting environmental conservation and cultural preservation. In conclusion, this study highlights the importance of understanding soil dynamics and adapting agricultural practices to local conditions. Integrating scientific knowledge with traditional wisdom may help balance jhum productivity with long-term sustainability amid challenges such as the decline in fallow periods.



**Thang Ngaihte et al.,**

ACKNOWLEDGEMENT

The authors acknowledge A.A.A. Mohamed, Department of Environmental Science, Mizoram University, for providing and helping obtain a digital elevation map (DEM) image of the study area. The University Grants Commission, New Delhi, for providing financial assistance in the form of a Senior Research Fellowship (SRF) and the Indian Council of Agricultural Research (ICAR) Research Complex for North Eastern Hill Region ICAR-RC NEH (Lamphelpat), Department of Environmental Science, Mizoram University, to extend the laboratory facilities to complete this work. We also thank the Department of Science and Technology for equipment support under the DST-FIST program, which facilitated the analytical work.

REFERENCES

1. Soni JK, Hota S, Shakuntala I (2019). Jhum cultivation: The way of livelihood security for Mizoram. *Indian Farming* 69(09):20–23.
2. Sati VP (2019). Economic and ecological implications of shifting cultivation in Mizoram, India. In *Environmental science and engineering*, Springer Nature, Switzerland. <https://doi.org/10.1007/978-3-030-36602-5>
3. Pachua AL and Devi OH (2020). Shifting cultivation and environment: A study of subsistence to profit in Mizoram. *Mizoram University Journal of Humanities & Social Sciences*, VI (1):47-61.
4. Sati VP and Rinawma P (2014). Practices of shifting cultivation and its implications in Mizoram, North-East India: A review of existing research. *Nat Environ* 19(2):179-187.
5. Grogan P, Lalnunmawia F, Tripathi SK (2012). Shifting cultivation in steeply sloped regions: a review of management options and research priorities for Mizoram state, Northeast India. *Agrofor* 84(2):163–177. <https://doi.org/10.1007/s10457-011-9469-1>
6. Tripathi SK, Vanlalfakawma, DC, Lalnunmawia F (2017). Shifting cultivation on steep slopes of Mizoram, India: impact of policy reforms. In: M. Crains (Eds.). *Shifting cultivation policies: balancing environmental and social sustainability* (pp. 393-413). <https://doi.org/10.1079/9781786391797.0393>
7. Ali S, Hussain I, Hussain S, Hussain A, Ali H, Ali M (2019). Effect of altitude on forest soil properties at Northern Karakoram. *Eurasian Soil Sci* 52(10):1159–1169. <https://doi.org/10.1134/s1064229319100120>
8. Intimongla, Ariina MMS, Saya D, Phucho T (2021). Properties of soil in relation to altitude. *Just Agriculture*, 1(12):1-13.
9. Maurya U, Roy T, Bihari B, Singh MP, Bishnoi R, Kadam D, Singh L, Shrimali SS, Muruganandam M, Kumar S, Sharma S, Kumar R, Malik AK, Verma R (2023). Impact of altitude on soil physico-chemical properties in a Topo-Sequence in Mid Himalayan watershed: A case study from Dehradun District, Uttarakhand. *J Geol Soc India* 99(3):421–429. <https://doi.org/10.1007/s12594-023-2326-9>
10. Kamal A, Mian IA, Akbar WA, Rahim HU, Irfan M, Ali SH, Alrefael A, Zaman W (2023). Effects of soil depth and altitude on soil texture and soil quality index. *Appl Ecol Environ Res* 21(5):4135–4154. https://doi.org/10.15666/aeer/2105_41354154
11. Wu D, Chi Q, Sui X, Zhang M, Jia H, Guo-Rong S (2021). Metabolic diversity and seasonal variation of soil microbial communities in natural forested wetlands. *J For Res* 32(6):2619–2631. <https://doi.org/10.1007/s11676-021-01326-8>
12. Zhang C, Liu A, Bahar M (2024). Editorial: Microbial response to emerging contaminants in soil and sediment ecosystems. *Front Microbiol* 15. <https://doi.org/10.3389/fmicb.2024.1371223>
13. Thanga JLT (2019). Institutional changes in jhum land tenure system in Mizoram. *Mizoram Economic Review* 3(2):20-38.
14. Thong P, Pebam R, Sahoo UK (2018). A geospatial approach to understand the dynamics of shifting cultivation in Champhai district of Mizoram, North-East India. *J Indian Soc Remote Sens* 46(10):1713–1723. <https://doi.org/10.1007/s12524-018-0832-9>
15. Klute, A. 1986. *Methods of Soil Analysis: Part 1 – Physical and Mineralogical Methods*. SSSA.





Thang Ngaihte et al.,

16. Anderson J and Ingram J (1993) Tropical soil biology and fertility: A handbook of methods. Second Edition, CABI, Wallington, 221.
17. Walkley AJ and Black IA (1934) Estimation of soil organic carbon by the chromic acid titration method. Soil Science, 37:29-38.
18. Subbiah BV and Asija GL (1956) A rapid procedure for the estimation of available nitrogen in soils. Current Science, 25:259-260.
19. Bray RW and Kurtz LT (1945). Determination of total, organic, and available forms of phosphorus in soils. Soil Sci 59(1):39–46. <https://doi.org/10.1097/00010694-194501000-00006>
20. Hanway JJ and Heidal H(1952). Soil analysis methods as used in Iowa State College Soil Testing Laboratory. Iowa State College of Agriculture Bulletin 57:1-31.
21. Semy K, Singh MR, Lemla W, Temjen W (2022). Seasonal variation of soil quality in a semi-deciduous northern tropical forest of Nagaland, India. Appl Biochem Biotechnol 195(4):2359–2370. <https://doi.org/10.1007/s12010-022-04106-0>
22. Xiong X, Lyu M, Deng C, Li X, Lu Y, Lin W, Jiang Y, Xie J (2022). Carbon and nitrogen availability drives seasonal variation in soil microbial communities along an elevation gradient. Forests, 13(10):1657. <https://doi.org/10.3390/f13101657>
23. Solanki AC, Gurjar NS, Sharma SK, Wang Z, Kumar A, Solanki AC, Divvela PK, Ks Y, Kashyap BK (2024). Decoding seasonal changes: soil parameters and microbial communities in tropical dry deciduous forests. Front Microbiol 15. <https://doi.org/10.3389/fmicb.2024.1258934>
24. Wu P, Hu D, Jin G, Li J, Zhong Q, Cheng D (2024). Unraveling the spatial–temporal distribution patterns of soil abundant and rare bacterial communities in China’s subtropical mountain forest. Front Microbiol 15. <https://doi.org/10.3389/fmicb.2024.1323887>
25. Zhang Q, Zhou J, Li X, Liu C, Lin W, Zheng W, Chen Y, Yang Y (2019). Nitrogen addition accelerates the nitrogen cycle in a young subtropical Cunninghamia lanceolata (Lamb.) plantation. Ann For Sci 76(2). <https://doi.org/10.1007/s13595-019-0817-z>
26. Karki H and Bargali SS (2021). Spatial and temporal trends in soil N-mineralization rates under the agroforestry systems in Bhabhar belt of Kumaun Himalaya, India. Agrofor Syst 95(8):1603–1617. <https://doi.org/10.1007/s10457-021-00669-9>
27. Mo Q, Wang W, Lambers H, Chen Y, Yu S, Wu C, Fan Y, Zhou Q, Li Z, Wang F (2021). Response of foliar mineral nutrients to long-term nitrogen and phosphorus addition in a tropical forest. Funct Ecol 35(10):2329–2341. <https://doi.org/10.1111/1365-2435.13896>
28. Prasad S and Baishya R (2021). Effect of tree species and seasons on soil nitrogen transformation rates in the semi-arid forest of Delhi, India. Vegetos, 35(1):219–227. <https://doi.org/10.1007/s42535-021-00291-1>
29. Borah P, Gogoi N, Mahanta SP (2023). Seasonal variation in carbon mineralization kinetics, microbial biomass carbon and enzyme activities in the soils of three natural ecosystems of Kaziranga National Park, Assam, north east India. J Soil Sci Plant Nutr 23(4):5300–5311. <https://doi.org/10.1007/s42729-023-01402-4>
30. Yahya M, Rasul M, Hussain SZ, Dilawar A, Ullah M, Rajput L, Afzal A, Asif M, Wubet T, Yasmin S (2023). Integrated analysis of potential microbial consortia, soil nutritional status, and agro-climatic datasets to modulate P nutrient uptake and yield effectiveness of wheat under climate change resilience. Front Plant Science <https://doi.org/10.3389/fpls.2022.1074383>
31. Fan B, Westerband AC, Wright IJ, Gao P, Ding N, Ai D, Tian T, Zhao X, Sun K (2023). Shifts in plant resource use strategies across climate and soil gradients in dryland steppe communities. Plant Soil. <https://doi.org/10.1007/s11104-023-06401-z>
32. Goswami M, Mujumdar M, Singh BB, Ingale M, Ganeshi N, Ranalkar MR, Franz TE, Srivastava PK, Niyogi D, Krishnan R, Patil SN (2023). Understanding the soil water dynamics during excess and deficit rainfall conditions over the core monsoon zone of India. Environ Res Lett 18(11), 114011. <https://doi.org/10.1088/1748-9326/acffdf>
33. Yao S and Zhao C (2023). Comparing of soil moisture in different yearly rainfall patterns at The Horqin sandy land. In: Weng, CH. (Eds.) Proceedings of the 4th International Conference on Advances in Civil and Ecological Engineering Research. Lecture Notes in Civil Engineering 292:377–386. Springer, Singapore. https://doi.org/10.1007/978-981-19-5783-3_30





Thang Ngaihte et al.,

34. Liu S, Van Meerveld I, Zhao Y, Wang Y, Kirchner JW (2024). Seasonal dynamics and spatial patterns of soil moisture in a loess catchment. *Hydrol Earth Syst Sci* 28(1):205–216. <https://doi.org/10.5194/hess-28-205-2024>

35. Kaur T, Sehgal SK, Singh S, Sharma S, Dhaliwal SS, Sharma V (2021). Assessment of seasonal variability in soil nutrients and its impact on soil quality under different land use systems of Lower Shiwalik Foothills of Himalaya, India. *Sustainability (Basel)*, 13(3):1398. <https://doi.org/10.3390/su13031398>

36. Neha, Bhople BS, Sharma S (2020). Seasonal variation of rhizospheric soil properties under different land use systems at lower Shivalik foothills of Punjab, India. *Agrofor Syst* 94(5):1959–1976. <https://doi.org/10.1007/s10457-020-00512-7>

37. Cherubin MR, Tormena CA, Karlen DL (2017). Soil quality evaluation using the Soil Management Assessment Framework (SMAF) in Brazilian oxisols with contrasting texture. *Rev Bras Cienc Solo* 41(0). <https://doi.org/10.1590/18069657rbc20160148>

38. Lepcha NT and Devi NB (2020). Effect of land use, season, and soil depth on soil microbial biomass carbon of Eastern Himalayas. *Ecol Process* 9(1). <https://doi.org/10.1186/s13717-020-00269-y>

Table 1. Location of study sites

Elevation (asl)	Sites	Coordinates	Altitude (m)
Lower (<500 m)	Khawkawn (KK)	24°01'24"N	344
		93°15'10"E	
Middle (500-1000 m)	NE Khawdungsei (NE)	23°57'22"N	829
		93°13'31"E	
Higher (>1000 m)	Kawlhem (KB)	23°53'16"N	1579
		93°16'36"E	

Table 2. Physical properties of soil of current jhum fields in Champhai, Mizoram (Mean ±SEM)

Sites/Altitude	Seasons	Depth	Texture	SMC (%)	WHC (%)	BD (g/cm ³)
KK (344)	Pre-monsoon	0-15	Loamy sand	17.75±0.8	31.91±0.03	1.32±0.06
		15-30		15.15±0.53	30.28±0.03	1.41±0.06
	Monsoon	0-15	Loamy sand	22.97±0.54	35.93±0.06	1.42±0.01
		15-30		20.07±0.35	33.45±0.37	1.45±0.02
	Post monsoon	0-15	Sandy loam	18.81±0.30	38.13±0.06	1.39±0.02
		15-30		17.64±0.47	37.48±0.03	1.43±0.01
NE (829)	Pre-monsoon	0-15	Loamy sand	24.51±0.91	36.27±0.01	1.13±0.04
		15-30		22.79±0.72	32.49±0.03	1.15±0.03
	Monsoon	0-15	Loamy sand	28.89±0.78	39.11±0.23	1.19±0.02
		15-30		23.08±0.66	37.99±0.20	1.21±0.02
	Post monsoon	0-15	Loamy sand	20.23±0.73	40.14±0.04	1.16±0.02
		15-30		18.88±0.05	39.41±0.06	1.17±0.02
KB (1579)	Pre-monsoon	0-15	Loamy sand	24.51±1.03	38.02±1.04	0.89±0.01
		15-30		18.22±0.78	35.3±0.99	0.98±0.02
	Monsoon	0-15	Loamy sand	28.54±0.50	44.71±0.75	1.05±0.04
		15-30		20.68±0.33	38±0.13	1.15±0.01
	Post monsoon	0-15	Sandy loam	24.21±1.27	40.38±0.27	0.93±0.02
		15-30		17.9±0.55	38.02±0.10	1.01±0.01





Thang Ngaihte et al.,

Table 3. Chemical properties of soils in current jhum fields of Champhai, Mizoram (Mean ±SEM)

Site/Altitude	Seasons	Depth	pH	OC (%)	AN (kg/ha)	AP (kg/ha)	AK (kg/ha)
KK (344)	Pre-monsoon	0-15	5.8±0.04	2.08±0.07	156.8±18.11	21.8±1.11	144.9±2.68
		15-30	5.76±0.04	0.98±0.07	135.89±10.45	19.77±1.06	138.28±1.16
	Monsoon	0-15	5.6±0.04	1.89±0.09	449.49±10.45	33.61±1.02	214.25±2.55
		15-30	5.54±0.03	0.76±0.10	321.51±19.76	25.46±0.82	211.57±1.77
	Post monsoon	0-15	5.63±0.04	1.96±0.06	240.43±27.66	26.61±0.42	182.17±2.01
		15-30	5.59±0.01	0.85±0.08	229.97±20.91	21.78±0.70	175.9±1.89
NE(829)	Pre-monsoon	0-15	6.21±0.02	2.88±0.20	397.23±37.69	19.85±0.76	239.57±1.9
		15-30	5.85±0.03	2.12±0.04	344.96±18.11	18.1±0.48	232.03±2.12
	Monsoon	0-15	5.57±0.02	2.29±0.10	501.76±18.11	27.15±1.51	248.14±3.39
		15-30	5.48±0.03	1.69±0.03	428.59±10.45	25.02±0.62	243.76±3.32
	Post monsoon	0-15	5.97±0.07	2.66±0.06	376.32±18.11	24.72±0.41	242.41±2.42
		15-30	5.66±0.03	1.25±0.03	219.52±36.21	23.06±0.23	235.65±2.22
KB (1579)	Pre-monsoon	0-15	6.18±0.01	2.92±0.04	439.04±18.11	25.31±1.07	216.49±2.26
		15-30	5.84±0.01	2.66±0.02	250.88±18.11	23.47±0.80	201.83±1.35
	Monsoon	0-15	5.65±0.04	2.31±0.03	669.01±20.91	35.75±1.16	233.09±2.42
		15-30	5.59±0.02	2.26±0.26	637.65±27.66	33.23±0.20	228.56±0.95
	Post monsoon	0-15	5.97±0.09	2.59±0.29	407.64±18.14	29.29±1.05	225.54±1.92
		15-30	5.85±0.05	2.44±0.19	365.87±27.66	27.42±0.38	219.38±2.17

Table 4. Correlations between seasons and soil parameters of current jhum fields in Champhai, Mizoram

	Seasons	pH	SMC	WHC	BD	OC	AN	AP	AK
Seasons	1								
pH	-.303*	1							
SMC (%)	-0.093	0.101	1						
WHC (%)	.582**	-0.026	.539**	1					
BD (g/cm3)	0.081	-.555**	-.385**	-.440**	1				
OC (%)	-0.189	.643**	.420**	.344*	-.727**	1			
AN (kg/ha)	0.053	-0.043	.735**	.609**	-.389**	.416**	1		
AP (kg/ha)	.340*	-.290*	.446**	.614**	-0.161	0.188	.712**	1	
AK (kg/ha)	0.23	0.08	.668**	.606**	-.474**	.413**	.681**	.320*	1

Table 5. Multiple analyses of variance (ANOVA) between soil parameters and seasons

		Sum of Squares	df	Mean Square	F	Sig.
pH	Between Groups	1.225	2	0.612	24.95	0
	Within Groups	1.252	51	0.025		
	Total	2.477	53			
SMC (%)	Between Groups	197.745	2	98.873	8.451	0.001





Thang Ngaihte et al.,

	Within Groups	596.702	51	11.7		
	Total	794.447	53			
WHC (%)	Between Groups	249.639	2	124.819	16.614	0
	Within Groups	383.167	51	7.513		
	Total	632.805	53			
BD (g/cm ³)	Between Groups	0.094	2	0.047	1.461	0.242
	Within Groups	1.634	51	0.032		
	Total	1.728	53			
OC (%)	Between Groups	1.653	2	0.826	1.789	0.177
	Within Groups	23.55	51	0.462		
	Total	25.203	53			
AN (kg/ha)	Between Groups	504116.033	2	252058.017	19.391	0
	Within Groups	662929.998	51	12998.627		
	Total	1167046.031	53			
AP (kg/ha)	Between Groups	674.547	2	337.274	27.154	0
	Within Groups	633.461	51	12.421		
	Total	1308.008	53			
AK (kg/ha)	Between Groups	10644.866	2	5322.433	6.117	0.004
	Within Groups	44375.076	51	870.1		
	Total	55019.942	53			

Table 6. LSD post-hoc test between soil parameters and seasons of current jhum fields in Champhai, Mizoram

	Dependent Variable		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
pH	Pre-monsoon	Monsoon	.3678*	0.04	0	0.2831	0.4524
		Post monsoon	.1589*	0.04	0	0.0742	0.2436
	Monsoon	Pre-monsoon	-.3678*	0.04	0	-0.4524	-0.2831
		Post monsoon	-.2089*	0.04	0	-0.2936	-0.1242
	Post monsoon	Pre-monsoon	-.1589*	0.04	0	-0.2436	-0.0742
		Monsoon	.2089*	0.04	0	0.1242	0.2936
SMC (%)	Pre-monsoon	Monsoon	-3.5494*	0.93	0	-5.4156	-1.6833
		Post monsoon	0.8767	0.93	0.349	-0.9895	2.7428
	Monsoon	Pre-monsoon	3.5494*	0.93	0	1.6833	5.4156
		Post monsoon	4.4261*	0.93	0	2.56	6.2922
	Post monsoon	Pre-monsoon	-0.8767	0.93	0.349	-2.7428	0.9895
		Monsoon	-4.4261*	0.93	0	-6.2922	-2.56





Thang Ngaihte et al.,

WHC (%)	Pre-monsoon	Monsoon	-4.1544*	0.59	0	-5.3498	-2.9591
		Post monsoon	-4.8806*	0.59	0	-6.0759	-3.6852
	Monsoon	Pre-monsoon	4.1544*	0.59	0	2.9591	5.3498
		Post monsoon	-0.7261	0.59	0.228	-1.9215	0.4692
	Post monsoon	Pre-monsoon	4.8806*	0.59	0	3.6852	6.0759
		Monsoon	0.7261	0.59	0.228	-0.4692	1.9215
AN (kg/ha)	Pre-monsoon	Monsoon	-213.8689*	21.38	0	-256.9257	-170.8121
		Post monsoon	-19.1578	21.38	0.375	-62.2146	23.8991
	Monsoon	Pre-monsoon	213.8689*	21.38	0	170.8121	256.9257
		Post monsoon	194.7111*	21.38	0	151.6543	237.7679
	Post monsoon	Pre-monsoon	19.1578	21.38	0.375	-23.8991	62.2146
		Monsoon	-194.7111*	21.38	0	-237.7679	-151.6543
AP (kg/ha)	Pre-monsoon	Monsoon	-8.6533*	0.79	0	-10.2462	-7.0605
		Post monsoon	-4.0983*	0.79	0	-5.6912	-2.5055
	Monsoon	Pre-monsoon	8.6533*	0.79	0	7.0605	10.2462
		Post monsoon	4.5550*	0.79	0	2.9622	6.1478
	Post monsoon	Pre-monsoon	4.0983*	0.79	0	2.5055	5.6912
		Monsoon	-4.5550*	0.79	0	-6.1478	-2.9622
AK (kg/ha)	Pre-monsoon	Monsoon	-34.3790*	1.77	0	-37.9361	-30.8219
		Post monsoon	-17.9867*	1.77	0	-21.5438	-14.4295
	Monsoon	Pre-monsoon	34.3790*	1.77	0	30.8219	37.9361
		Post monsoon	16.3923*	1.77	0	12.8352	19.9495
	Post monsoon	Pre-monsoon	17.9867*	1.77	0	14.4295	21.5438
		Monsoon	-16.3923*	1.77	0	-19.9495	-12.8352





Thang Ngaihte et al.,

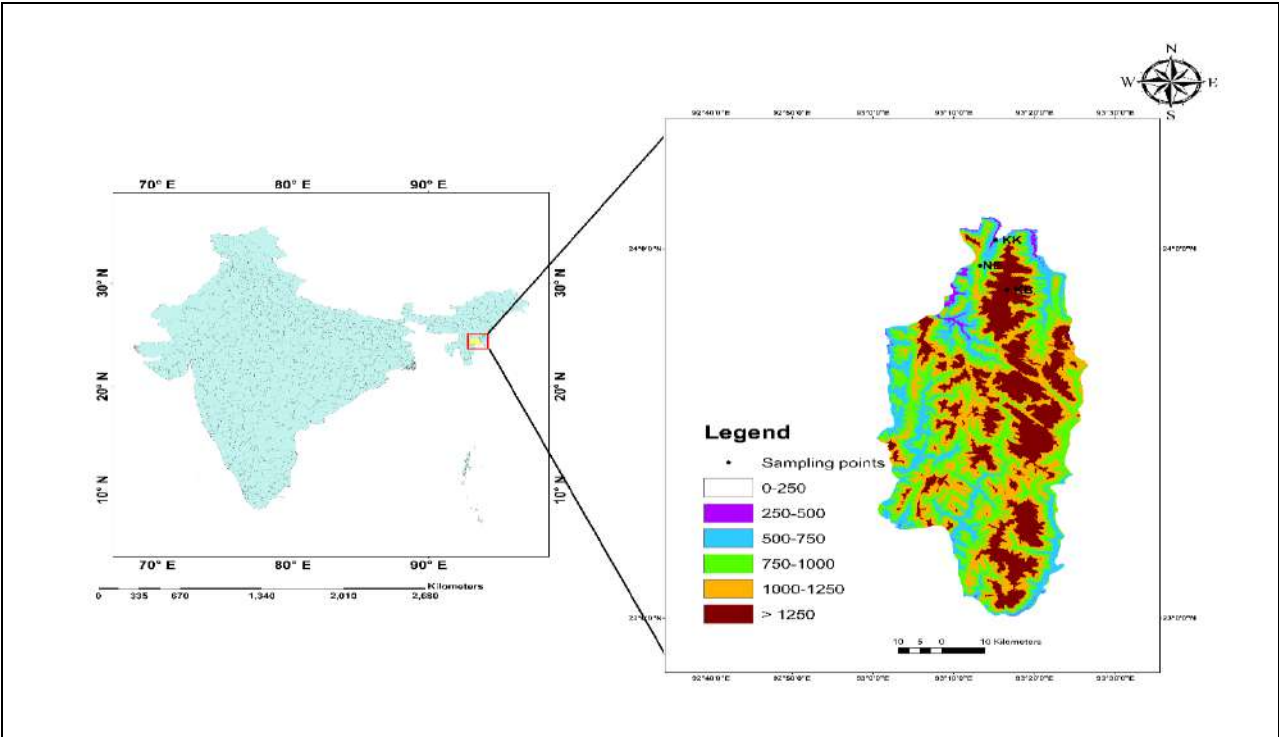


Fig. 1. Location of study site and demographic elevation map (DEM) of Champhai showing sampling sites





Variation in Water Structures from One Organ to Another in a Goat and Their Response to Homeopathic Potencies Derived from Natural Substances : A Spectroscopic Analysis

Raj Kumar Singh¹, Naheed Sultana², Nirmal Chandra Sukul^{3*}, Nivedita Pande⁴ and Anirban Sukul⁵

¹Assistant Professor, Department of Botany, Government General Degree College, Mangalkote, Panchanantala, Khudrun Dighi, East Burdwan (Affiliated to University of Burdwan), West Bengal India.

²Research Fellow, Sukul Institute of Homeopathic Research, Santiniketan, West Bengal, India.

³Professor (Rertd.) Department of Zoology, Visva Bharati University, and President, Sukul Institute of Homeopathic Research, West Bengal, India.

⁴State Aided College Teacher, Department: Geography, Panihati Mahavidyalaya, Kolkata, West Bengal, India.

⁵Director, Sukul Institute of Homeopathic Research, West Bengal, India.

Received: 18 Jun 2024

Revised: 19 Aug 2024

Accepted: 18 Oct 2024

*Address for Correspondence

Nirmal Chandra Sukul

Professor (Rertd.) Department of Zoology,
Visva Bharati University, and President,
Sukul Institute of Homeopathic Research,
West Bengal, India.

E.Mail: ncsukul@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Homeopathic potencies are specifically structured water which can modify the water structure of ordinary water. Water in the body of a living organism is structured because of the presence of biomolecules and cell membranes. Are the water structures same in different tissues of the body? The present study addresses this question. The study further explores the interaction between the water structure in a tissue and a related homeopathic potency. Water structures of five different organs like brain, kidney, lungs, liver and stomach were studied by electronic and vibrational spectroscopy. The response of water structure of organ samples to related homeopathic potencies like *Hypericum perforatum* 200 (Plant product), *Mercurius corosivus* 30 (Salt), *Phosphorus* 30 (Natural element), *Chelidonium majus* 200 (Plant) and *Cina* 30 CH (Plant) was also studied by the same spectroscopic methods. Water structures in the five organs tested are different from each other. The responses of these water structures to five related homeopathic potencies are also different from each other. An appropriate homeopathic potency may selectively modify the water structure of the concerned organ affected. In this way the potency may initiate possible therapeutic effect on the affected organ / tissue.

Keywords: Mammalian organs, Water structures, Responses, Homeopathic potency, UV-spectra, FT-IR-spectra.





Raj Kumar Singh et al.,

INTRODUCTION

Using electronic and vibrational spectroscopy it was demonstrated that different homeopathic potencies carry specific water structures [1-2]. Water structures in a potency is transferable to aqueous ethanol (EtOH) solution [3]. Living tissues carry structured water because of the presence of biomolecules suspended in water, and lipid bilayer in the plasma membrane [4-6]. Are the water structures same in different tissues of a mammal? The present experimental study aims to address the question. The second objective is to see whether the structured water in different tissues responds to a related homeopathic potency. We collected five organ samples of a freshly decapitated castrated goat from a local meat shop. The cut pieces of the organ samples were immediately kept in 90% ethanol so that aqueous ethanol can acquire the water structure of the organ and preserve it. The water structures in the five organ samples were studied by electronic and vibrational spectroscopy. Further, the responses of the water structures of the five organ samples to related homeopathic potencies were also studied [7]. For instance, response of the brain sample to *Hypericum perforatum* 200 cH (derived from plant), kidney sample to *Mercurius corosivus* 30 (derived from salt), lungs sample to *Phosphorus* 30 (natural element), liver sample to *Chelidonium majus* 200 (Plant) and stomach sample to *Cina* 30 cH (plant). *Cina* was selected because all goats including the present one under study are naturally infected by flat worm parasites in their stomach. The spectroscopic characteristics of the water structures in the organ samples have been found to be different. The responses of these water structures to the related homeopathic potencies also show difference in their spectroscopic characteristics.

MATERIALS AND METHODS

Collection of organ samples

Samples of five different organs like brain, lungs, stomach, liver and kidney were collected from a freshly decapitated castrated goat weighing 17 kilograms from a local meat shop. The samples, one gram each, were immediately kept in 5 ml of 90% EtOH so that the solvent can acquire and preserve the water structure of the organ.

Drugs

Homeopathic potencies were purchased from a local market in sealed vials. All drugs are in 90% EtOH. The control consists of blank 90% EtOH prepared from absolute ethanol (HIMEDIA, Lot number 0000498722). The drugs are *Hypericum perforatum* 200 (Hapco Hahnemann Publishing Company Private LTD. Batch number 8432), *Mercurius corosivus* 30 (Hapco Hahnemann Publishing Company Private LTD. Batch number 7965), *Phosphorus* 30 (King & Co., M. Bhattacharyya & Co. Pvt. Ltd. Batch number D2080), *Chelidonium majus* 200 (Hapco Hahnemann Publishing Company Private LTD. Batch number 7955) and *Cina* 30 (Sett Dey & Co (Homoeo) Lab, Batch number 13796). The drugs were selected on the basis of their possible relation to the organ samples tested. Thus *Hypericum* was selected for brain, *Merc cor* for kidney, *Phosphorus* for lungs, *Chelidonium* for liver and *Cina* for stomach [7]. *Cina* was selected because all goats are infected in their stomach by flat worm parasites.

Electronic Spectroscopy (UV)

All organ samples in 90% EtOH were first filtered through Whatman number I filter paper. The filtrate was diluted in deionized and distilled water (DD) 1:100. A pair of quartz cuvettes filled with DD water, and baseline was set in a UV-VIS spectrophotometer (Shimadzu, Model- UV-VIS 1900i, Software-Lab Solutions UV-VIS) at room temperature 24 ± 2 °C, with the instrument in spectrum mode in the wavelength range 190-210nm, scan speed medium and data interval 0.5 nm). Then one cuvette was taken out, emptied washed in DD water and filled with the filtrate of organ samples, and the spectrum was recorded in the wavelength range of 200 nm to 300 nm. In another experiment we tested the response of each filtrate of an organ to a related homeopathic potency. Each potency was diluted with DD water 1:100 and then mixed with a filtrate from an organ in the proportion 1:1. The control consisted of diluted EtOH mixed with a diluted potency in the proportion 1:1. As for example, the brain sample was treated with *Hypericum*



**Raj Kumar Singh et al.,**

200. The control consisted of diluted EtOH + *Hypericum* 200. In this way all the five organ samples were treated with a related homeopathic potency and their UV-spectra were recorded in the wavelength range 200 nm to 300 nm.

FT-IR spectra

FT-IR spectra of all the five organ samples were recorded with the help of a Shimadzu IR Affinity -1S Fourier Transform Infrared (FT-IR) spectrophotometer (Spectrum two) using the attenuated total reflection (ATR) technique. The energy resolution was 0.5 cm^{-1} . The baseline was corrected for atmospheric humidity and CO_2 . One drop of each sample was put into the sample groove, and then the tip of a single reflection pure diamond crystal was brought in contact with the sample drop. The entire spectrum was recorded in the wave number range of 4000 to 500 cm^{-1} . Each spectrum represents an average of 45 scans. The temperature and humidity were maintained in the laboratory at 24°C and less than 50%, respectively. FT-IR spectra of all the five drugs treated with related homeopathic potencies as mentioned under UV spectra were also recorded.

RESULTS

UV spectra

All the spectra show distinct peak at 200 nm. The highest intensity is found in the brain sample followed by kidney, stomach, liver and lungs. The brain and the kidney showed one more broad peak covering the region between 240 nm and 280 nm (fig.1). All the treatment effects of organ samples + related homeopathic potency and their controls showed single peak at 200 nm. In all the cases the organ samples + related homeopathic potency showed higher intensity than their control (fig. 2 a, b, c, d and e).

FT-IR spectra

FT-IR spectra of all the five organ samples without any drug treatments are shown in figure 3a for OH-stretching band and figure 3b for OH-bending band. The figures show distinct variation in normalized intensity and peak frequency for both OH-stretching and OH-bending bands. Figures 3a and b show highest intensity with lungs followed by brain, kidney, liver and stomach. Treatment effects of all the organ samples with related homeopathic potencies were shown in figure 4a for brain. The normalized spectra showed marked difference in intensity and peak frequency in both OH-stretching and OH-bending bands. In both cases the control (EtOH + *Hypericum* 200) showed higher intensity than the organ sample (Brain + *Hypericum* 200) (fig. 4a). Figure 4b shows the difference in intensity between the control (EtOH + *Merc cor* 30) and the organ sample (Kidney + *Merc cor* 30) for both OH-stretching and OH-bending bands. The OH-stretching and OH-bending bands showed differences in peak frequency (fig. 4b). Figure 4c shows the marked difference in intensity between the control (EtOH + *Phosphorus* 30) and organ sample (Lungs + *Phosphorus* 30). Here the organ sample (Lungs + *Phosphorus* 30) shows higher intensity than the control for both OH-stretching and OH-bending bands. The peak frequency showed difference for both OH-stretching and OH-bending bands (fig. 4c). Figure 4d shows the marked difference in intensity and peak frequency for both OH-stretching and OH-bending bands in case of control (EtOH + *Chelidonium* 200) and organ sample (Liver + *Chelidonium* 200). Here also the organ sample showed higher intensity than the control for both OH-stretching and OH-bending bands (fig. 4d). Figure 4e shows the marked difference in intensity and peak frequency for both OH-stretching and OH-bending bands in case of control (EtOH + *Cina* 30) and organ sample (Stomach + *Cina* 30). Here also the organ sample showed the higher intensity than the control for both OH-stretching and OH-bending bands (fig. 4e). The bands are very broad.

DISCUSSION

Cut surfaces of organ samples, when put into the aqueous ethanol, established contact between the water structure in the organ samples and aqueous ethanol. In this way the solvent medium (Aqueous EtOH) takes up the water structure of the organ samples [3]. UV spectra of the water structure of five different organs show variation in intensity and number of peaks because the proteins and other biological contents are different in different tissues of



**Raj Kumar Singh et al.,**

the organs. Homeopathic potencies can convert the water structure of a sample into a different form⁸. Since the water structures are different in different organs the effect of a homeopathic potency on the water structures of the organs should be different. This is evident in the UV spectra and the FT-IR spectra of the treated organ samples with related homeopathic potencies (fig. UV and FT-IR). In this way the right homeopathic remedy could produce appropriate effect on the water structure of an organ. It has been observed in both UV and FT-IR spectra that all potencies show distinct difference in intensity from their interaction with the water structure of organ samples. This shows that homeopathic potencies can change the water structure of the organ samples tested.

It was demonstrated that water structure is different in healthy and diseased tissues [9]. It is known that water structure plays an important role in protein function. All biomolecules are suspended in water and have specific structures for their biological functions. Tetrahedral hydrogen bonded molecules of water are present in the bulk solvent away from the hydration layer of protein present. In the hydration layer water molecules and polar atoms on the surface of protein form a three-dimensional chain connection of hydrogen bonded network. The networks are flexible and thus help in conformational changes of protein [4]. Cell membranes mediate many biological functions at the lipid water interface [10].

Water has a 4th state besides the three usual states of solid, liquid and vapour. The 4th state is structured water having crystalline clathrate hydrate structures [11]. It is the structured water which plays the most important role in homeopathic potencies. The only difference is that a homeopathic potency involves activated specifically structured water [1-2]. Ordinary structured water contains structure inducing agent, but a homeopathic potency removes these agents by serial dilution. A substance, may be a plant product, mineral, salt etc can induce specific structure in the water [12-13]. Besides a substance light, X-rays, magnetic fields can also induce specific structures in water. Preparation of a homeopathic potency involves progressive serial dilution followed by mechanical agitation. Serial dilution results in loss of original substance molecules and produces a water structure which bears relation to the starting substance. There are evidences showing that many substances, X-rays, magnetic fields can change water structure [14-16]. Mechanical agitation changes hydrogen bonds, hydrogen bond strength, free water molecules. Aqueous ethanol solvent preserves the water structure in a homeopathic potency. Based on the results we can assume that a homeopathic potency can change water structure in the body of a patient including the affected part / organ. The potency, if properly selected, would positively interact with the water structure of the affected part / organ and thus initiate biochemical processes to restore health. Structured water can influence the physiology of man [17].

CONCLUSIONS

Water structures in different organs like brain, kidney, liver, lungs, and stomach are different. A homeopathic potency related to an organ modifies the water structure of the organ. A homeopathic potency is activated specifically structured water which can modify selectively the water structure of the organ. It is possible that a properly selected homeopathic remedy may favourably modify the water structure of the affected organ, and initiate therapeutic effect in the concerned organ vis-à-vis the patient.

CONFLICT OF INTEREST

There is no conflict of interest.

ETHICAL CONSIDERATIONS

No animal was killed in the laboratory. The tested organ samples were collected from a local meat shop. For this, no ethical approval was needed.





Raj Kumar Singh et al.,

ACKNOWLEDGEMENT

The work described here has been financially supported by the Sukul Institute of Homeopathic Research, Santiniketan.

REFERENCES

1. Ghosh S, Singh RK, Sukul NC, Pande N, Sukul A. High dilutions of drugs show distinct variation from each other in their electronic spectra. *Ind J Nat Sci.* 2021; 12(67): 1-4.
2. Singh RK, Ghosh S, Sukul NC, Nandi M, Pal A, Pal M. High dilutions of drugs of Covid-19 origin show difference in ranks are revealed by electronic and vibrational spectroscopy. *Int J High Dil Res.* 2021; 20(04): 29-42.
3. Singh RK, Sukul NC, Pande N, Sukul A. Water in contact with a homeopathic potency acquires the water structure of the potency as revealed by electronic and vibrational spectroscopy. *Ind J Nat Sci.* 2023; 14(81): 65306-65310.
4. Nakasako M. Water-protein interactions from high-resolution protein crystallography. *Phil Trans R Soc Lond B.* 2004; 359: 1191-1206.
5. Ramsey CL. Biological structured water (BSW)- A review (part1): Structured water (SW) properties, BSW and redox biology, BSW and bioenergetics. *J basic and Applied Sci.* 2023; 19: 174-201.
6. Higgis M J, Polcik M, Fukuma T, Sader JE, Nakayama Y, Jarvis SP. Structured water layers adjacent to biological membranes. *Biophysical J.* 2006; 91: 2532-2542.
7. Boericke W. *Pocket Manual of Homeopathic Materia Medica*, 1927. Indian edn, Sett Dey, Calcutta 1976; 459-462.
8. Singh RK, Sukul NC, Pande N, Sukul A. Water Serves as Primary Target for High Dilution of an Anti-Burn Homeopathic Drug on Heat- Exposed Bovine Serum Albumin *In-Vitro*. *Ind. J Nat. Sciences.* 2023; in Press.
9. Gun'ko VM, Turov VV, Shpiko AP, Leboda R, Jablonski M, Gorzelak M, Jagiello-Wojtowicz E. Relationships between characteristics of interfacial water and human bone tissues. *Colloids and Surfaces B: Biointerfaces.* 2006; 53: 29-36.
10. Lipowsky R, Sackmann E (eds) (1995) *Structure and Dynamics of Membranes From Cells to Vesicles / II. Generic and Specific Interactions* 1st Edition, Volume 1A, Elsevier, Amsterdam.
<https://shop.elsevier.com/books/structure-and-dynamics-of-membranes/lipowsky/978-0-444-81975-8>
11. Pollack GH. The fourth phase of water: beyond solid, liquid, and vapor. *Edgescience.* 2013; 14 (16): 14-18.
12. Zupancic B, Grdadolnik J. Solute-induced changes in the water H-bond network of different alcohol-aqueous systems. *J Molecular Liquids.* 2021; 341: 117349.
13. Laurent H, Baker DL, Soper AK, Ries ME, Dougan L. Solute specific perturbations to water structure dynamics in tertiary aqueous solution. *The J Physical Chemistry B.* 2020; 124 (48): 10983-10993.
14. Zhao L, Ma Kai, Yang Zi. Changes of water hydrogen bond network with different externalities. *In J Mol Sci.* 2015; 16: 8454-8489.
15. Sarkar T, Konar A, Sukul NC, Sohel Md. A, Sengupta A, Sukul A. DSC reveals variation in enthalpy associated with free water molecules in water ethanol solution exposed to x-rays and magnetic field. *Clinical and Experimental Homeopathy.* 2017; 4 (1): 50-78.
16. Liu YY, Hua X, Zhang Z, Zhang J, Zhang S, Hu P, Long YT. pH dependant water clusters in photoacid solution: Real-time observation by ToF-SIMS at a submicropore confined liquid-vacuum interface. *Frontiers in Chemistry.* 2020; 8: 731.
17. Riveros-Perez E, Riveros R. Water in the human body: An anesthesiologist's perspective on the connection between physicochemical properties of water and physiologic relevance. *Annals of Medicine and Surgery.* 2018; 26: 1-8.





Raj Kumar Singh et al.,

<p>Figure 1: UV spectra of water structures in 0.09% EtOH of 5 different organs of goat.</p>	<p>Figure 2a: UV spectra of water structured in 0.09% EtOH of brain after treatment with <i>Hypericum</i> 200 in 0.09% EtOH. The drug mixed with water structure in the proportion 1:1. The control consists of 0.09% EtOH mixed with <i>Hypericum</i> 200 in the proportion 1:1.</p>
<p>Figure 2b: UV spectra of water structure in 0.09% EtOH of kidney after treatment with <i>Merc cor</i> 30 in 0.09% EtOH. The drug mixed with water structure in the proportion 1:1. The control consists of 0.09% EtOH mixed with <i>Merc cor</i>30 in the proportion 1:1.</p>	<p>Figure 2c: UV spectra of water structure in 0.09% EtOH of liver after treatment with <i>Chelidonium</i> 200 in 0.09% EtOH. The drug mixed with water structure in the proportion 1:1. The control consists of 0.09% EtOH mixed with <i>Chelidonium</i>200 in the proportion 1:1.</p>
<p>Figure 2d: UV spectra of water structure in 0.09% EtOH of lungs after treatment with <i>Phosphorus</i> 30 in 0.09% EtOH. The drug mixed with water structure in the proportion 1:1. The control consists of 0.09% EtOH mixed with <i>Phosphorus</i> 30 in the proportion 1:1.</p>	<p>Figure 2e: UV spectra of water structure in 0.09% EtOH of stomach after treatment with <i>Cina</i> 30 in 0.09% EtOH. The drug mixed with water structure in the proportion 1:1. The control consists of 0.09% EtOH mixed with <i>Cina</i> 30 in the proportion 1:1.</p>





Raj Kumar Singh et al.,

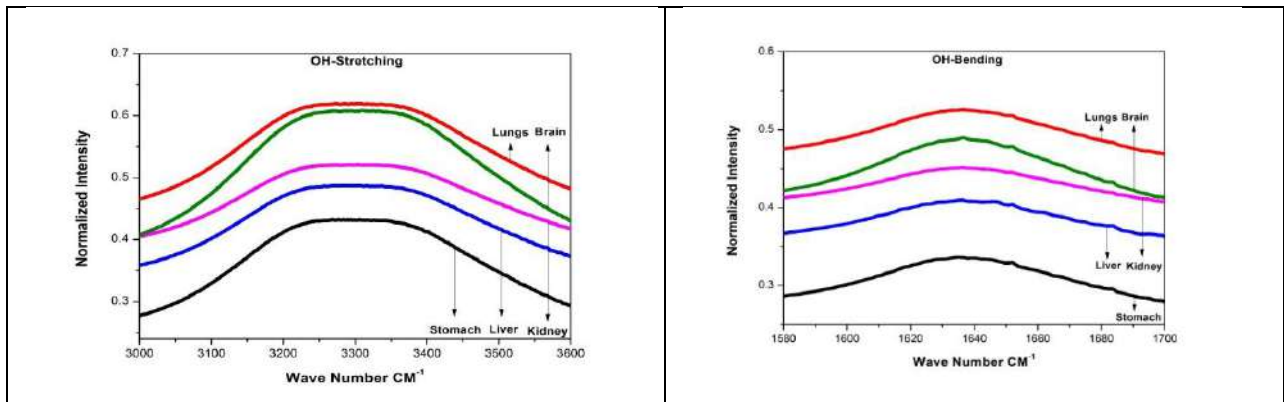


Figure 3a: Normalized FT-IR spectra of water structures in 0.09% EtOH of 5 organs of goat showing OH-Stretching band.

Figure 3b: Normalized FT-IR spectra of water structures in 0.09% EtOH of 5 organs of goat showing OH-Bending band.

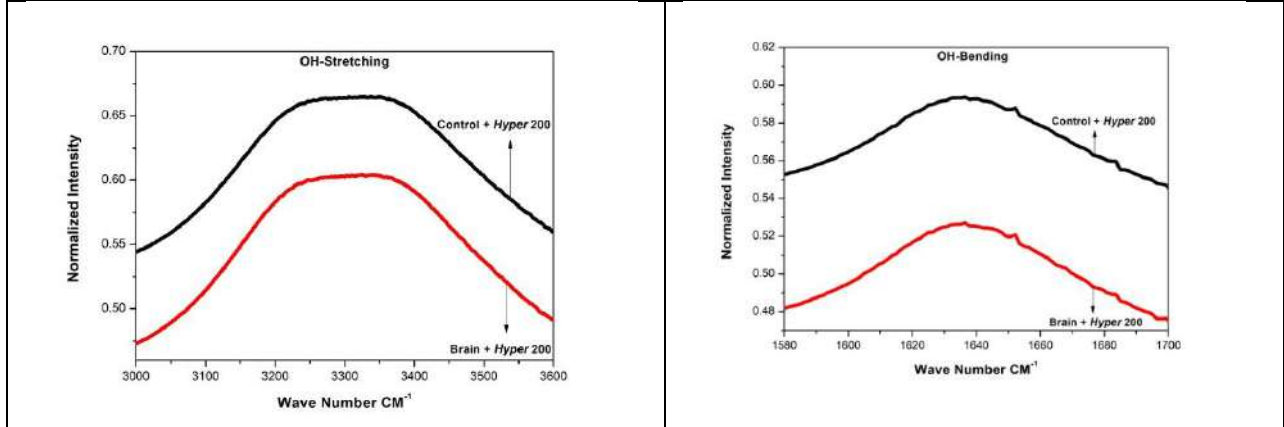


Figure 4a: Normalized FT-IR spectra of water structure in 0.09% EtOH of brain of goat showing OH-Stretching (upper) and OH-Bending (lower) bands. The water structure was mixed with *Hypericum* 200 in 0.09% EtOH in the proportion 1:1. The control consists of 0.09% EtOH mixed with *Hypericum* 200 in 0.09% EtOH in the proportion 1:1.

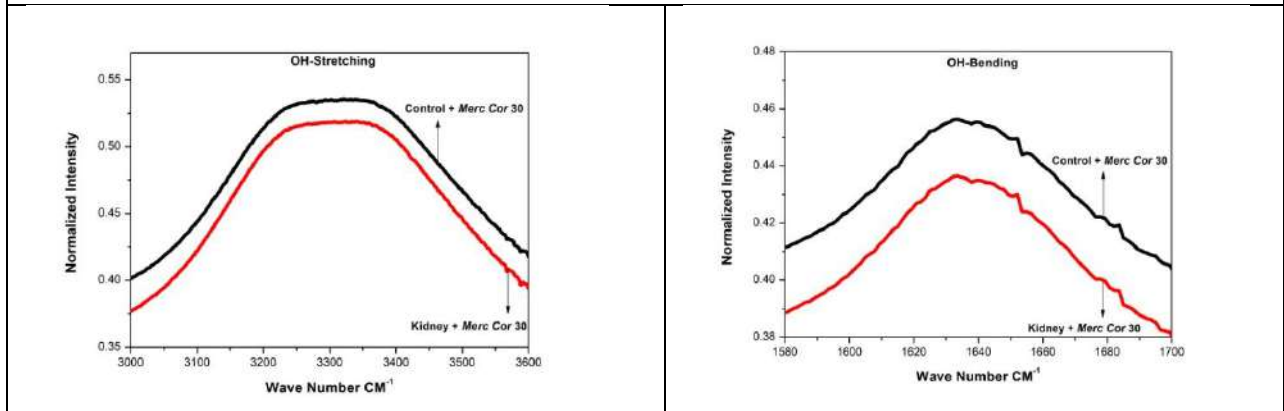


Figure 4b: Normalized FT-IR spectra of water structure in 0.09% EtOH of kidney of goat showing OH-Stretching (upper) and OH-Bending (lower) bands. The water structure was mixed with *Merc cor* 30 in 0.09% EtOH in the proportion 1:1. The control consists of 0.09% EtOH mixed with *Merc cor* 30 in 0.09% EtOH in the proportion 1:1.





Raj Kumar Singh et al.,

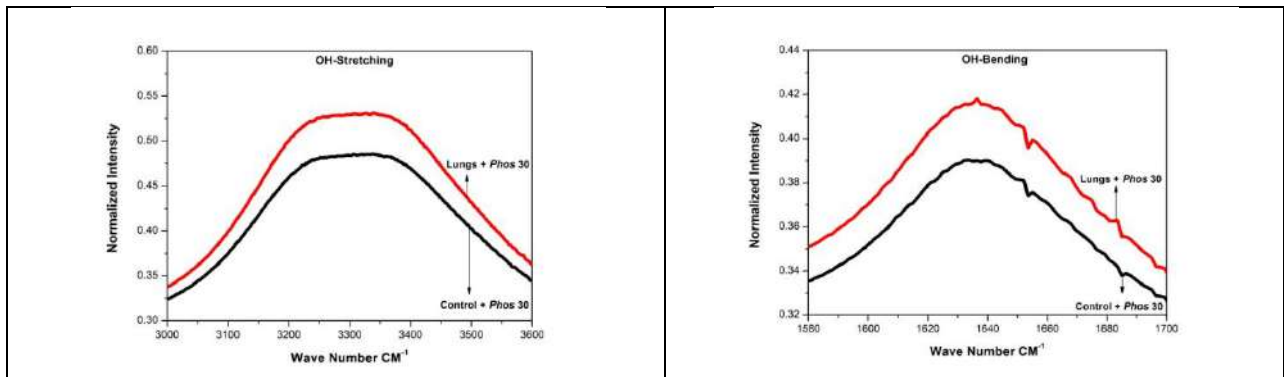


Figure 4c: Normalized FT-IR spectra of water structure in 0.09% EtOH of lungs of goat showing OH-Stretching (upper) and OH-Bending (lower) bands. The water structure was mixed with *Phosphorus 30* in 0.09% EtOH in the proportion 1:1. The control consists of 0.09% EtOH mixed with *Phosphorus 30* in 0.09% EtOH in the proportion 1:1.

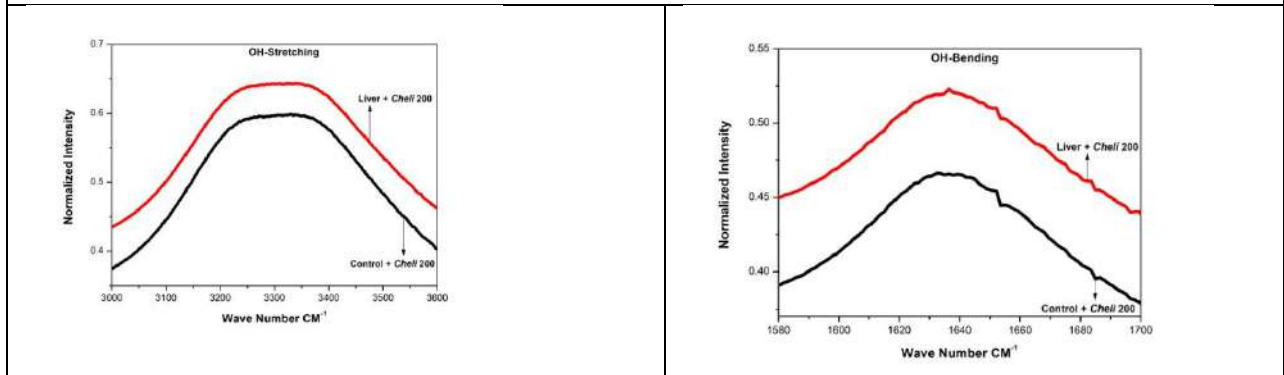


Figure 4d: Normalized FT-IR spectra of water structure in 0.09% EtOH of liver of goat showing OH-Stretching (upper) and OH-Bending (lower) bands. The water structure was mixed with *Chelidonium 200* in 0.09% EtOH in the proportion 1:1. The control consists of 0.09% EtOH mixed with *Chelidonium 200* in 0.09% EtOH in the proportion 1:1.

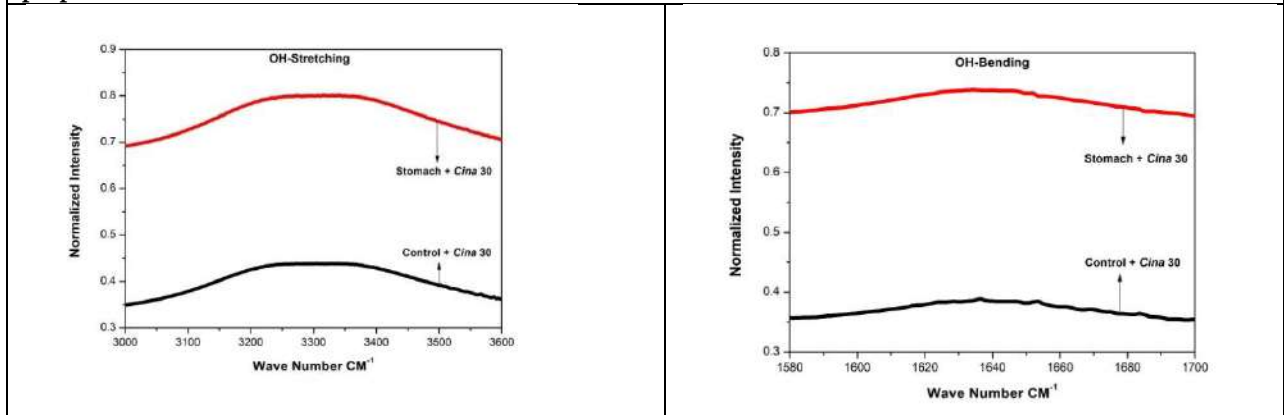


Figure 4e: Normalized FT-IR spectra of water structure in 0.09% EtOH of stomach of goat showing OH-Stretching (upper) and OH-Bending (lower) bands. The water structure was mixed with *Cina 30* in 0.09% EtOH in the proportion 1:1. The control consists of 0.09% EtOH mixed with *Cina 30* in 0.09% EtOH in the proportion 1:1.





Detailed Pharmacognostic Standardization of *Gmelina asiatica* Linn. Fruit

Radhakrishnan Jaya Prakash^{1*} and Seventhilingam Kaliamoorthy²

¹Research Scholar, Botanical Garden, National Orchidarium & Experimental Garden, Botanical Survey of India, Southern Regional Centre, Yercaud – 636 602, Salem District, Tamil Nadu, India.

²Scientist –E, Botanical Garden, National Orchidarium & Experimental Garden, Botanical Survey of India, Southern Regional Centre, Yercaud – 636 602, Salem District, Tamil Nadu, India.

Received: 28 May 2024

Revised: 24 Aug 2024

Accepted: 16 Oct 2024

Address for Correspondence

Jaya Prakash Radhakrishnan

Research Scholar, Botanical Garden,
National Orchidarium & Experimental Garden,
Botanical Survey of India, Southern Regional Centre,
Yercaud – 636 602, Salem District, Tamil Nadu, India.
Email id: jayaprakashpb35@gmail.com.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Gmelina asiatica L., commonly known as Nilakumil in Tamil, is distributed throughout the state of Tamil Nadu, predominantly found in the coastal regions. In traditional medicine, the fruits of *Gmelina asiatica* L. have been used to treat conditions such as leucorrhoea, burning sensations in the eyes, and skin issues like eczema and dandruff. The primary objective of the present study is to standardize the pharmacognostical parameters, including macroscopy, microscopy, histo chemistry, physio-chemical, phytochemical analyses, and High-Performance Thin-Layer Chromatography (HPTLC) fingerprint. Macroscopic studies revealed that the fruit is dark orange-yellow, ovoid-oblong in shape, succulent, with a sweetish-slightly bitter taste. Microscopic investigations showed the presence of various structural components, including the epidermis, a narrow band of hypodermal cells, a wide mesocarp zone, and a stony endocarp. Powder microscopy revealed fragments of the pericarp, mesocarp, and reticulate parenchyma of the testa, endosperm with starch, cells of the cotyledon, sclereids, stone cells, and oil cells. Histochemical studies identified the presence of alkaloids, lignin, polysaccharides, tannin, cutin, and resin. Preliminary phytochemical analysis revealed the presence of alkaloids, phenols, tannins, flavonoids, triterpenoids, glycosides, steroids, and quinones. The moisture content was 9.38%, with alcohol and water-soluble extractive values of 48.45% and 52.03% respectively. The total ash content was 3.16%, while the acid-insoluble and water-soluble ash values were 0.19% and 52.03% respectively. Different extracts were subjected to HPTLC fingerprinting, which produced distinct peaks with varying

83044



**Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy**

retention factor (Rf) values. These peaks serve as drug-specific identifiers. This study provides valuable guidance for accurately identifying the plant material and establishing standards for the crude drug.

Keywords: Adulteration, Crude Drug, *Gmelina asiatica* L. Fruit, Lamiaceae, Pharmacognosy.

INTRODUCTION

Herbal remedies and plants have been utilized as medicine since ancient times. Over the last few decades, traditional medical systems have gained international attention. According to current estimates, a sizable section of the populace in many developing nations primarily receives their primary medical care from traditional healers and medicinal plants. At the same time, a growing number of people in wealthy nations are starting to use supplementary or alternative medicines, such as medicinal plants [1]. The demand for herbal drugs is now high and growing every day. Due to the abundance of readily available raw materials, the World Health Organization (WHO) supports and encourages the use of traditional herbs and cures in the healthcare industry. In nature, plants are incredibly complex. Their medicinal properties are influenced by species, geographic location, and methods of harvesting. Standardization of herbal drugs is crucial due to improper herb authentication, microbial adulterations, and pesticide residues. The WHO states that before any tests are conducted, the macroscopic and microscopic description of a medicinal plant is the first step towards confirming the identity and degree of purity of such materials [2].

The genus *Gmelina* was initially described by Linnaeus in 1753, based on the species *Gmelina asiatica* L. A total of 31 species and 2 subspecies are present in this genus [3]. *Gmelina asiatica* L. is known by various names in different Indian languages, such as Asian bush in English; *Bhadra* in Bengali; *Badhara* and *Nag-phul* in Hindi; *Guludu* and *Kalshivaniin* in Kannada; *Cherkumizhi* and *Kumilamaram* in Malayalam; *Lahan-shivan* in Marathi; *Nondanoand Gopogombhari* in Orissa; *Badhara* in Punjab; *Gopabhadra*, *Biddari* and *Vikarini* in Sanskrit; *Nilakkumil*, *Nilakkimnizh*, *Nilacumal*; *Kumizhaniaram*, *Kumil* and *Kadambal* in Tamil; *Chirugummudu*, *Gummadi*, *Cherunelli*, *Challagumudu* and *Shirigumudu* in Telugu [4]. It belongs to the Lamiaceae family and is found in dry evergreen to dry deciduous forests [5], growing as a scrambling shrub up to 8 meters high. *Gmelina asiatica* L. is widely utilized in Indian traditional medicine to treat jaundice, hemorrhoids, painful urination, arthritis, edema, liver diseases, neurological disorders, heart diseases, skin infections, acne, diabetes mellitus [6], Syphilis [7], dandruff [8], fever and rheumatism [9]. It also serves as an antiseptic, contraceptive [10], astringent, demulcent, bladder catarrh [7] and blood purifier [11]. Scientifically, the plant is reported to possess antibacterial [12-13], antifungal [12,13-14], antidandruff [15], anticancer [16], antioxidant [17-18], anti-inflammatory [14,19-20], antidiabetic, antihyperglycemic [21], antianxiety [22], antipyretic [23], Larvicidal [24], hepatoprotective [17] and nephroprotective [18].

1, 2-benzenedicarboxylic acid and monolinoleyl glycerol is the major bioactive compounds identified in the aerial parts of this plant [25]. The heartwood contains methyl p-methoxycinnamate, paulownin, gmelinol, cyclooolivil, and sitosterol [26]. A flavone, ovalifolin, (+)-sesamin, sakuranetin, (-)-piperitol, and (+)-pinoresinol have been isolated from the alcoholic extract of the roots [27]. Palmitic, stearic, linoleic, oleic, and ricinoleic acids are reported in the seed oil [28]. Kaempferol-3-rutinoside, apigenin-7-rutinoside, apigenin-7-glucuronide, and quercetagenin are identified in the flowers and leaves of *Gmelina asiatica* L. [29]. The fruits are edible [30] and used as a hair wash in traditional medicine [5]. Fresh fruits are boiled with coconut oil, made into a paste, and applied to the head to eliminate dandruff [31]. To date, no report is available on the pharmacognostic evaluation of *Gmelina asiatica* L. fruits. Therefore, this paper presents a report on the transverse section, powder microscopy, Histochemistry, physicochemical parameters, preliminary phytochemical analysis, and High-Performance Thin-Layer Chromatography (HPTLC) of the fruit of *Gmelina asiatica* L.





MATERIALS AND METHODS

Collection and Identification of Plant Material

The fresh fruits of *Gmelina asiatica* L. were collected from Cuddalore district, Tamil Nadu. The plant's taxonomic identity was confirmed by referencing the floristic work conducted by earlier researchers [32-33] and consulting the Madras Herbarium (MH). A voucher specimen B.S.I. (S.R.C) No. 150401 has been deposited in the Madras Herbarium, Botanical Survey of India, Southern Regional Centre, Coimbatore.

Botanical Description

Gmelina asiatica L., Sp. pl. 626.1753; Roxb. Fl. ind.3: 87. 1832; Wight, III.Ind.Bot. t. 174. 1850; Hook.f. Fl. Brit. India 4: 582. 1885; Gamble, Fl. Madras 2:1098 (768). 1924; Matthew, FTNC 1: 301.1981, 3: 1222. 1983, 4: t.470.1988& FPH 968. 1999; COL & WCSP 2018; III.Fl. C&NTN t. 1685.2019. *G.Parvifolia* Roxb. Pl. Coromandel t. 162. 1802 (Figure 1). Large straggling deciduous shrub, rarely tree, 3-4 m high; young twigs minutely hairy; spines to 2.5 cm; Leaves ovate to elliptic, 2.5 × 1 cm, apex obtuse to acute; base rounded to cuneate, margin entire to irregularly lobed; upper surface glabrous, dark green, shiny; lower surface glabrous, whitish with peltate scales; petiole to 1 cm. Inflorescence – Racemes axillary and terminal to 9 cm. Calyx 4 lobed, puberulous without; 0.4 × 0.3 cm; lobes acute, 0.1 cm long. Corolla golden yellow; 4 lobed, anterior lip 3-lobed; posterior lip 1-lobed. Stamens 4, inserted at apex of the tube, glabrous; anterior pair 2.3 cm long, anthers 0.2 cm long; posterior pair fertile, 1.1 cm long, anthers 0.1 cm long. Ovary 0.4 cm, glabrous; style 2.6 cm long. Fruit 3 × 1 cm, glabrous, apex rounded, succulent drupe yellow when ripe. Seeds 2, oblong, 1.2 × 0.5 cm. Flowering and Fruiting: Throughout the year.

Macro-Microscopy

The fruits were soaked in water for 12 hours, and free-hand transverse sections were taken according to standard procedures [35-37]. A Zeiss Stereo Discovery V.8 microscope connected with an Axiocam ERc5s was used to examine macro-morphological features, and various characteristics of the fruit, such as color, shape, size, odor, and taste, were recorded. A Zeiss Axiolab 5 microscope with an Axiocam 208 color camera was used to observe micro-morphological features.

Powder Microscopy

The samples were cleaned, shade dried, powdered, and passed through sieve No. 80. The powder was separately treated with glycerine (50%, v/v) and chloral hydrates (10%, v/v). Then the Sample was stained with iodine solution to confirm the presence of starch grains and the characters were observed and photographed under different magnifications by using a Nikon ECLIPSE E200 trinocular microscope attached with Zeiss ERc5s digital camera under bright field light.

Histochemistry

Transverse sections were stained using specific histo-chemical procedures [37].

Preparation of Fruit Extract

The fresh fruits were washed with water, dried in shade, and powdered. Four grams of the powder were extracted with 100 ml of n-hexane in a Soxhlet apparatus. The extract was concentrated using a rotary evaporator under reduced pressure (100 mbar) and reduced temperature (55°C). It was then dissolved in 10 ml of n-hexane. The marc in the thimble was successively extracted with hexane, chloroform, and ethanol. Their extracts were redissolved in the respective solvents and used for preliminary phytochemical analysis and HPTLC analysis.

Physio-chemical Parameters

Physiochemical parameters such as Loss on drying, ash values, and extractive values, were carried out as per the standard procedure [38].



**Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy****Phytochemical Screening**

The phytochemical analysis was performed using standard procedure [39].

HPTLC fingerprinting analysis:

Thirty microliters (30 μ l) of the extracts were applied to a silica-coated TLC plate 60F₂₅₄ using Camag's ATS4 sample applicator and developed in a twin trough chamber (CAMAG) measuring 20 × 10 cm, previously saturated with the mobile phase. The mobile phase for the hexane extract was Toluene: Ethyl acetate: Formic acid (9.5:0.5:0.5, v/v/v); for chloroform and ethanol extracts, it was Toluene: Ethyl acetate: Formic acid (7:3:0.5, v/v/v). The developed plate was dried over a hot plate and a photograph was taken in CAMAG Visualizer Chamber followed by scanning under λ 254 (absorbance mode, D2 lamp) and λ 354 (Fluorescence mode, Hg lamp) respectively with a slit dimension 6×0.45 mm and scanning speed of 20 mm/s by scanner 4 (Scanner_210441) linked with WINCATS software. The scanned plate was dipped in Vanillin sulphuric acid (VSR) and heated at 105°C till the appearance of colored bands. The photograph was taken immediately at white light followed by scanning at λ 520 at absorption mode (W lamp).

RESULTS AND DISCUSSION

In the herbal medicine sector, incorrect plant species identification has resulted in adulteration and raw drug replacement [40]. Accurate identification, quality assurance, and the development of pharmacognostic standards are necessary for an evaluation of medicinal plants. The WHO states that evaluating a medical plant both macroscopically and microscopically is the first step towards plant standardization [41-42]. Microscopic characters have a lot of potential applications in authentication on several levels. Even though scientists believe that it can be challenging to distinguish between closely related genera within a family based solely on anatomical traits, careful cellular analysis or powder microscopy may be able to help resolve the authentication problems [43-44].

Macroscopic Evaluation

The fruit is ovoid, oblong, or pyriform in shape and succulent, the surface is smooth glossy, and dark orange-yellow in colour. 1- 2 seeded drupe. Calyx is persistent, attached at the base of the pointed end of the fruit a short pedicel lies at the centre of it, and a small depression is occasionally located at the broader end, 1.5 to 2 cm in length, 0.5 to 1 cm in width and 0.5 to 0.8 mm in thickness. Pericarp is sweetish-slightly bitter and mucilaginous; odour characteristic, taste oily (Figure 2).

Microscopic Evaluation**Fruit**

The transverse section of the fruit pulp (Figure 3) shows three layers i.e., epicarp, mesocarp, and endocarp. The epicarp region consists of a single layer of small rectangular cells arranged radially. The epidermis is covered with a thin layer of cuticle. Followed by the epidermis, 10 to 15 rows of isodiametric, spherical hypodermal cells comprise parenchymatous. The mesocarp is fleshy, consisting of a wide zone of parenchymatous cells arranged in numerous layers approximately 15 to 20 layers. Outer mesocarp cells are isodiametric, whereas inner mesocarp are radially elongated and closely packed, narrow cells. Vascular strands are embedded in the inner side of it, tangentially running celled layer of inner epidermis lies underneath this, followed by a broad zone of sclerenchymatous band of endocarp.

Seed

The transverse section of the seed shows narrow, small-sized cells of the outer epidermis followed by inconspicuous endosperm consisting of 1 to 2 rows of tangentially running cells embedded with aleurone grains, cotyledons consisting of outer and inner epidermis enclosing the wide mesophyll tissue embedded with fixed oil and aleurone grains (Figure 4).



**Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy****Powder Microscopy**

The fruit powder is dark brown in color with a slightly aromatic odor and characteristic taste. It shows fragments of the pericarp, mesocarp, and reticulate parenchyma of the testa, endosperm with starch, cells of the cotyledon, sclereids, stone cells, cells with content, and oil cells (Figure 5).

Histochemistry

In the fruit pericarp, alkaloids were present in the epicarp and mesocarp cells; cutin and resin were noted in the epidermal cells; lignin and mucilage were present in the mesocarp region, while tannin deposition was present in both the epicarp and mesocarp regions as shown in (Figure 6). In the seed, alkaloids and aleurone were present in the cotyledonary cells; lignin deposition was observed in the testa cells; oil droplets were observed in the endosperm cells, and tannin was present in the cotyledon cells (Figure 7).

With the use of particular staining reactions and imaging techniques, histochemistry enables the identification and localization of biomolecules and organelles in many types of cells and tissues at the cellular level. The in vivo localization of promoters in certain tissues and the identification of particular cell wall constituents like polysaccharides and lignin are two other common uses for histochemical techniques [45]. The chemical histo-localisation allows determining the exact storage site of the secondary metabolites.

Physio-Chemical Parameters

The physio-chemical parameters of powdered fruit pulp of *Gmelina asiatica* L. were evaluated in Table 3. Physicochemical parameters play a crucial role in standardizing and ensuring the quality of herbal drugs. These parameters encompass various factors such as loss on drying, ash content, and pH. Herbal materials need to be free from contamination, making foreign matter analysis of powdered drugs a significant parameter for assessing the purity of herbal drugs [46]. The loss on drying test is a widely employed method to determine the moisture content in powdered samples. It's essential for the moisture content of drugs to be kept at minimal levels to prevent the growth of microbes during storage [47]. Ash values are utilized to assess the quality and purity of crude drugs, indicating the presence of various impurities such as carbonate, oxalate, and silicate. Water-soluble ash is employed to gauge the quantity of inorganic compounds in drugs, while acid-insoluble ash primarily consists of silica, indicating contamination with earthy materials [47]. Estimating extractive values helps determine the quantity of active constituents present in a given amount of plant material when it's extracted with a specific solvent. When any crude drug is extracted with a particular solvent, it yields a solution containing various phytoconstituents. The composition of these chemical constituents is influenced by both the nature of the drug and the solvent utilized. Additionally, this process offers an indication of whether the crude drug has been fully depleted or not [46-48].

Preliminary Phytochemical Screening

The preliminary phytochemical screening of *Gmelina asiatica* L. fruit pulp revealed the presence of diverse phytochemical compounds, which can be found in Table 2. Alkaloids, Phenols, tannins, flavonoids, triterpenoids, glycosides, steroids, quinones, and cardiac glycosides constituted the phytoconstituents in hexane, chloroform, and ethanol extracts. However, proteins, reducing sugars, coumarin, and anthraquinones, acids were not found in ethanol, chloroform, and hexane extracts. Among the three extracts, the hexane extract exhibited significantly lower levels of phytoconstituents

HPTLC Fingerprint Analysis

The results of the Thin Layer Chromatography (TLC) photodocumentation of 30 microliters of different extracts under UV 254 nm, 366 nm, and white light after derivatization using vanillin-sulphuric acid reagent at 520 nm are shown in Figure 8 and Table 3. The HPTLC fingerprint profiles of different extracts of the fruits pulp of *Gmelina asiatica* L. are presented in Figures 9 to 11. The HPTLC analysis of the hexane extract shown in Figure 9 reveals that under UV 254 nm, eight peaks are observed, with the major peaks present at Retention factor (Rf) values of 0.90 (45.89%), 0.94 (16.14%), and 0.82 (15.42%). Under UV 354 nm, the major peaks are found at Rf values of 0.44 (41.48%), 0.28 (24.63%), and 0.96 (24.46%). After derivatization with vanillin-sulphuric acid, at white light 520 nm, the major



**Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy**

peaks are observed at Rf values of 0.97 (25.94%), 0.29 (22.14%), and 0.44 (18.33%). In the chloroform extract, twelve peaks were recorded, with the major peaks seen at Rf values of 0.94 (52.20% area) and 0.58 (14.91% area) under UV 254 nm. In the ethanol extract, under UV 254 nm, five peaks were identified, with the major peaks observed at Rf values of 0.33 (30.8% area), 0.95 (25.88% area), and 0.55 (18.79% area). Under λ 354 nm, a total of two peaks were recorded, with the major peaks appearing at an Rf value of 0.91 (82.37% area). After derivatization with vanillin-sulphuric acid and examining at white light 520 nm, a total of eight peaks were recorded, with the major peaks found at Rf values of 0.51 (20.75% area), 0.75 (17.97% area), and 0.46 (13.77% area), as presented in Figure 11. This finding is useful for supplementing existing information on the identification and standardization of *Gmelina asiatica* L., even in its powdered form, to distinguish it from other drugs and adulterants. The HPTLC analysis conducted for the chemical profiling of *Gmelina asiatica* L. fruit will be useful in identifying bioactive chemicals and markers by a comparison of the compound's Rf values with reference standards. These findings also imply that the pharmacognostic and physicochemical parameters that are observed have a significant role in formulation development and quality control of the crude drug.

CONCLUSION

Pharmacognostic standardizations is essential to determining a crude drug's accurate identity and quality. Prior to any medication being added to the pharmacopoeia, these requirements need to be met. The macroscopy, microscopy, physicochemical, and phytochemical aspects of the plant material provide the majority of the information regarding its identity, purity, and quality. Since there is no documentation of pharmacognostical research on *Gmelina asiatica* L. fruits, the current study was conducted to establish essential phytopharmacopoeial standards. Further, it could attract the attention of pharmacologists to explore this plant through scientific research.

ACKNOWLEDGMENT

The authors are thankful to the Director, Botanical Survey of India, Kolkata, and Scientist – F & Head of Office, BSI, SRC, Coimbatore, for encouragement and providing facilities.

REFERENCES

1. France Lavoisier Publication, 1999.
2. World Health Organization, Quality control methods for medicinal plant material, WHO Library. 1998, 110-115.
3. De Kok, R. A revision of the genus *Gmelina* (Lamiaceae). Kew Bulletin. 2012,67, 293-329. <http://dx.doi.org/10.1007/s12225-012-9382-4>.
4. Reviews on Indian Medicinal Plants: Fa-Gy. Medicinal Plants Unit. Indian Council of Medical Research, New Delhi. 2013, 1076.
5. Narasimhan, D and Irwin, S.J. Flowering Plants of Tamil Nadu: A Compendium. Care Earth Trust, Chennai. 2021, 542.
6. Rasha, K., Khazem, M., Bhanukiran, K., and Hemalatha, S. *Gmelina asiatica*: exploring traditional uses, pharmacological insights, and phytoconstituents—a comprehensive review (1961–2023). *Phytochemistry Reviews*. 2024, 1-24. [10.1007/s11101-024-09951-x](https://doi.org/10.1007/s11101-024-09951-x).
7. Kirtikar, K.R., and Basu, B.D. Indian medicinal plants. Publisher Lalit Mohan Basu, Allahabad. 1935. Vol 3, pg1934. http://asi.nic.in/asi_books/2048.pdf.
8. Hemadri, K., Raj, P.V., Rao, S.S., and Sarma, C.R.R. Folklore claims from Andhra Pradesh 1. *J.Sci. Res. Pl. Med.* 1980, 1 (2), 37-49.
9. Sivagami, S., Gayathri, N., Sakthivel, B., Ramar, S., and Kuralarasan, R. Plants used as Medicine by Paliyar Tribes of Shenbagathope in Virudhunagar District of Tamilnadu. India. *J Econ Taxon Bot.* 2008. 32.



**Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy**

10. Nadanakunjidam, S., and Abirami, S. Comparative study of traditional medical knowledge of Pondicherry and Karaikal regions in Union territory of Pondicherry. *Ethnobotany*. 2005, 17 (1-2), 112-117.
11. Singh, M., Kushagra N., and Tripathi, D.K. Potential Analgesic & Anti-Pyretic Herbal Drugs: A Comparative Review of Marketed Products. *International Journal of Phytomedicine*. 2010, 2 (3), 197-209. <http://dx.doi.org/10.5138/ijpm.2010.0975.0185.02030>.
12. Sudhakar, M., Rao, C. V., Rao, P. M., and Raju, D. B. Evaluation of antimicrobial activity of *Cleome viscosa* and *Gmelina asiatica*. *Fitoterapia*. 2006, 77, 47-49. <https://doi.org/10.1016/j.fitote.2005.08.003>.
13. Madhu, K.B., Mallikarjuna, R., Balakrishna P., Vedavathi, T., and Satyanarayana, T. Antimicrobial potential of *Gmelina asiatica* leaves and *Bauhinia vahlii* pods. 2001, 38, 299-302.
14. Merlin, N.J., Parthasarathy, V., Manavalan, R., Devi, P., and Meera, R. Phyto-Physicochemical evaluation, Anti-Inflammatory and Anti-microbial activities of Aerial parts of *Gmelina asiatica*. *Asian Journal of Research in Chemistry*. 2009, 2, 76-82.
15. Chandrika, M., Gowda, D.V., Vijayakumar, M., and Babu, U.V. Anti-dandruff activity of supercritical fluid extract of *Rosmarinus officinalis* and *Gmelina asiatica*. *Indo American Journal of Pharmaceutical Research*. 2015, 5 (4), 1463-1467.
16. Merlin, N.J., and Parthasarathy, V. Potential antitumor activity of *Gmelina asiatica* aerial parts against Dalton ascites lymphoma in mice. *Asian Journal of Chemistry*. 2010, 22, 3193-3199.
17. Merlin, N.J., and Parthasarathy, V. Antioxidant and hepatoprotective activity of chloroform and ethanol extracts of *Gmelina asiatica* aerial parts. *Journal of Medicinal Plant Research*. 2011, 5, 533-538.
18. Kiruba., Arun, K.P., and Brindha, P. In vitro studies on nephroprotective efficacy of *Cynodon dactylon* and *Gmelina asiatica*. *Asian Journal of Pharmaceutical and Clinical Research*. 2014, 7, 111-120.
19. Ismail, T.S., Gopalakrishnan, S., and Begum, V.H. Biochemical modes of action of *Gmelina asiatica* in inflammation. *Indian Journal of Pharmacology*. 1997, 29(5), 306-309.
20. Vedavathy, S., and Narayana Rao, K.V. Anti-inflammatory activity of some indigenous medicinal plants of Chittoor district, Andhra Pradesh. *Indian Drugs*. 1995, 32, 427-432.
21. Kasiviswanath, R., Ramesh, A., and Kumar, K.E. Hypoglycemic and antihyperglycemic effect of *Gmelina asiatica* Linn. in normal and alloxan-induced diabetic rats. *Biol. Pharm. Bull.* 2005, 28(4), 729-732.
22. Kamboj, S. Pharmacogenetic and anti-anxiety studies on leaves of *Gmelina asiatica* (Linn.). *Medicinal and Aromatic Plants*. 2015, 4 (4), 103.
23. Ikram, M., Khattak, S.G., and Gilani, N. (1987). Antipyretic studies on some indigenous Pakistani medicinal plants: II. *Journal of Ethnopharmacology*. 1987, 19, 185-192.
24. Muthukumar, U., Govindarajan, M., Rajeswary, M., and Hoti, S.L. Synthesis and characterization of silver nanoparticles using *Gmelina asiatica* leaf extract against filariasis, dengue, and malaria vector mosquitoes. *Parasitology Research*. 2015, 114:1817-1827. <https://doi.org/10.1007/s00436-015-4368-4>.
25. Binu, M., Parthasarathy, V., Manavalan, R., and Kumaravel, S. Chemical Investigation of Aerial Parts of *Gmelina asiatica* Linn by GC-MS. *Pharmacognosy Res.* 2009, 1, 152-156.
26. Anjaneyulu, A.S.R., Rao, A.M., Rao, V.K., and Row, L.R. The lignans of *Gmelina asiatica*. *Phytochemistry*. 1975, 14, 824.
27. Satyanarayana, T., Katyayani, B., Latha, E., Routhu, K., and Prasad, Y. Phytochemical studies on roots of *Gmelina asiatica* Linn. *Pharmacognosy Magazine*. 2007, 3(11), 156-158 <http://www.phcog.com/text.asp?2007/3/11/156/58094>.
28. Aggarwal, J.S., and Soni, P. Chemical examination of the oil from the seeds of *Gmelina asiatica* Linn. *J. Sci. Ind. Res.* 1949, 8B, 49-51.
29. Nair, A.G.R., and Subramanian, S.S. Quercetin and other flavones from *Gmelina arborea* and *G. asiatica*. *Phytochemistry*. 1975, 14, 1135-1136.
30. Kannan, M., Kumar, T.S., and Rao, M.V. Ethnobotanical survey on wild edible plants of Kalrayan hills, Salem district, Tamil Nadu, India. *Global Journal of Research on Medicinal Plants & Indigenous Medicine*. 2015, 4(12), 236-246.
31. Jeyaprakash, K., Ayyanar, M., Geetha, K.N., and Sekar, T. Traditional uses of medicinal plants among the tribal people in Theni District (Western Ghats), Southern India. *Asian Pacific Journal of Tropical Biomedicine*. 2011, 1(1), S20-S25. [https://doi.org/10.1016/S2221-1691\(11\)60115-9](https://doi.org/10.1016/S2221-1691(11)60115-9).





Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy

32. Gamble, J.S. The Flora of the Presidency of Madras. Adlard & Son, Ltd, London. 1935. Vol 2, pg. 768.
33. Matthew, K.M. The Flora of the Tamil Nadu Carnatic. The Rapinat Herbarium, St. Josephs College, Tiruchirapalli, India.1983. Vol 3, pg. 1222 - 23
34. Nartunai, G., Lokesh, R., Susikumar, S., Premkumar, C., Akansha, P., and Raju, I. Comparative morpho-anatomical studies to authenticate and differentiate the stem barks of *Cinnamomum verum* J.Presl and *Cinnamomum cassia* (L.) J.Presl and its market scenario. J. Drug Res Ayurvedic Sci. 2022, 7, 185-91.
35. Sundharamoorthy, S., Govindarajan, N., Chinnappillai, A., and Raju, I. Macro-microscopic atlas on heartwood of *Santalum album* L. (sandalwood). Pharmacognosy Journal. 2018, 10(4), 730-3. doi: 10.5530/pj.2018.4.122.
36. Susikumar, S., Nartunai, G., and Ilavarasan, R. Macro-microscopic Atlas on Heartwood of *Pterocarpus santalinus* L.f. (Red sandalwood). Res J Pharm Technol. 2021, 14(7), 3927-30.
37. Krishnamurthy, K.V. Methods in Plant Histochemistry. S. Vishwanathan Printers & Publishers Pvt. Ltd, Chennai.1988.
38. Lohar, D. R. Protocol for testing of Ayurvedic, Siddha, and Unani medicines. Ghaziabad: Government of India, Department of AYUSH, Ministry of Health & Family Welfare, Pharmacopoeial Laboratory for Indian Medicines. 2008.
39. Harborne, J.B. Phytochemical Methods, Chapman and Hall, London. 1973, 278
40. Prakash, O., Jyoti, A.K., Pavan, K., and Niranjana, K.M. Adulteration and substitution in Indian medicinal plants, an overview. J Med Plants Stud. 2013, 1(4), 127-32.
41. Pandey, A., and Tripathi, A. Concept of standardization, extraction and pre phytochemical screening strategies for herbal drug. J. Pharmacognosy Phytochem. 2014, 2(5), 115–119
42. Anonymous (1996). Indian pharmacopoeia. Government of India 2, Ministry of Health and Family Welfare. Controller of Publications, New Delhi.
43. SarojKumar, V., Jaishanker, R., Annamalai, A., and Sunil Kumar, K.N. Investigation into the pharmacognostical and phytochemical features of seeds of *Ensetesuperbum* (Roxb.) Cheesman: An unexplored medicinal plant of India. Pharmacognosy Journal.2013, 5, 163-9. doi.org/10.1016/j.phcgs.2013.07.004.
44. Yadav, D., Reshi, M.S., Uthra, C., Shrivastava, S., Srivastava, N., and Narayana, S.K., et al. Botanical and chemical fingerprinting of medicinal roots of *Justicia gendarussa* Burm. f. Phcog Res. 2017, 9, 208-14. doi: 10.4103/0974-8490.204643.
45. Yadav, V., Arif, N., and Singh, V.P., et al (2021). Histochemical Techniques in Plant science: more than meets the eye, Plant and Cell Physiology. 2021, 62(10), 1509-1527. https://doi.org/10.1093/pcp/pcab022.
46. Anonymous (1998). Quality control methods for medicinal plant materials. World Health Organization, Geneva.
47. Chanda. S. Importance of pharmacognostic study of medicinal plants: an overview. J Pharmacognosy Phytochem. 2014, 2(5), 69–73.
48. Tatiya, A., Surana, S., Bhavsar, S., Patil, D., and Patil, Y. Pharmacognostic and preliminary phytochemical investigation of *Eulophiaherbacea* Lindl. Tubers (Orchidaceae). Asian Pac J Trop Dis. 2012, 2, (Suppl 1), S50–S55.

Table 1. Physicochemical parameters of *Gmelina asiatica* L. fruit pulp

Name of the Parameters	Mean value
Moisture content at 105°C(% w/w)	9.38
Total ash (% w/w)	3.16
Acid insoluble ash (% w/w)	0.19
Water soluble extractive (% w/w)	52.03
Alcohol soluble extractive (% w/w)	48.45
pH value (10% solution)	4.63

Table 2. Preliminary phyto chemical screening of *Gmelina asiatica* L. fruit pulp extracts.

S. No	Tests for phyto constituents	Hexane	Chloroform	Ethanol
1.	Alkaloids	-	-	+
2.	Flavonoids	-	+	+





Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy

3.	Tannins	-	+	+
4.	Phenol	+	+	+
5.	Saponins	-	-	+
6.	Triterpenoids	+	+	+
7.	Proteins	-	-	-
8.	Glycosides	-	+	+
9.	Reducing sugar	-	-	-
10.	Anthraquinones	-	-	-
11.	Quinones	-	+	+
12.	Cardiac glycoside	-	-	+
13.	Steroids	+	+	+
14.	Coumarin	-	-	-
15.	Acids	-	-	-
Abbreviations: +, present; -, absent.				

Table 3. Presents the Retention factor (Rf) values obtained from TLC profiling of extracts derived from *Gmelina asiatica* L. Fruit pulp.

Name of the extract	UV 254 nm		UV366 nm		After derivatizing with 5% Vanillin sulphuric acid reagent	
	Rf	Colour	Rf	Colour	Rf	Colour
n-Hexane	0.05	Green	0.05	Lightred	0.08	Light violet
	0.29	Green	0.10	Lightred	0.17	Purple
	0.35	Green	0.29	Red	0.30	Purple
	0.44	Green	0.39	Blue	0.40	Light purple
	0.61	Green	0.45	Red	0.44	Purple
	0.82	Green	0.69	Lightred	0.55	Purple
	0.91	Green	0.76	Lightred	0.61	Purple
	0.98	Green	0.91	Blue	0.65	Light purple
			0.98	Blue	0.71	Light purple
					0.90	Violet
Chloroform					0.94	Violet
	0.08	Green	0.12	Blue	0.10	Grey
	0.10	Green	0.21	Blue	0.17	Purple
	0.14	Green	0.32	Blue	0.26	Yellow
	0.21	Green	0.46	Blue	0.35	Yellow
	0.31	Green	0.55	Blue	0.42	Brown
	0.39	Green	0.69	Blue	0.47	Yellow
	0.47	Green	0.75	Lightgreen	0.50	Yellow
	0.52	Green	0.79	Lightred	0.58	Light yellow
	0.58	Green	0.86	Light pink	0.73	Pink
Ethanol	0.09	Green	0.10	Blue	0.06	grey
	0.38	Green	0.19	Blue	0.17	Light pink
	0.45	Green	0.35	Blue	0.21	Light pink
	0.58	Green	0.46	Blue	0.27	Yellow
			0.64	Blue	0.39	Light purple
			0.80	Green	0.44	Yellow
			0.89	Blue	0.47	Pink
					0.58	Yellow
				0.78	Pink	





Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy

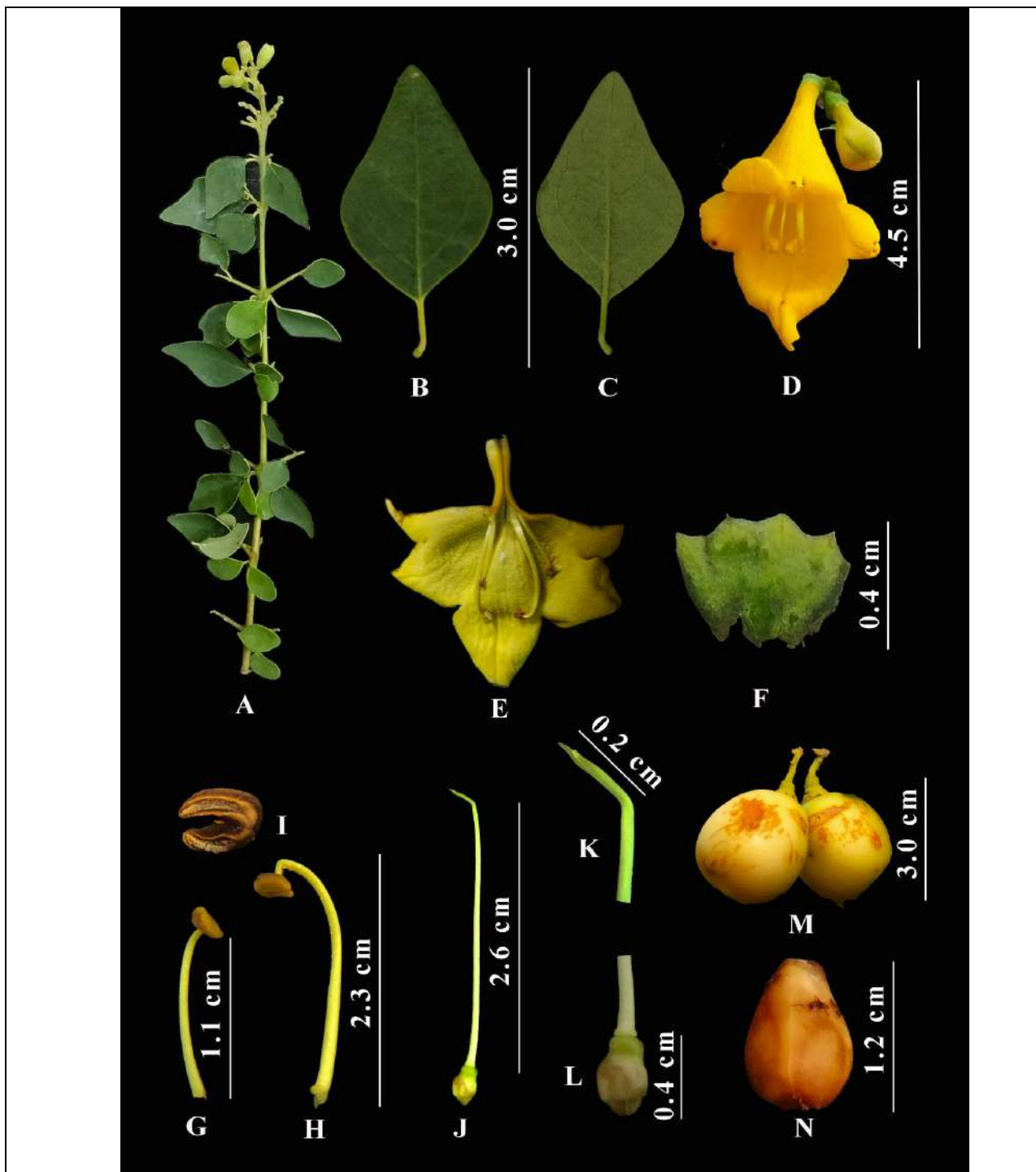


Figure 1. *Gmelina asiatica* L., A) A portion of twig; B) Adaxial surface of leaf; C) Abaxial surface of leaf; D) Inflorescence; E) Corolla -split out; F) Calyx; G) Smaller stamen; H) Larger stamen; I) Anther - enlarged view; J) Gynoecium; K) Stigma-enlarged view; L) Ovary; M) Fruits; N) Seed



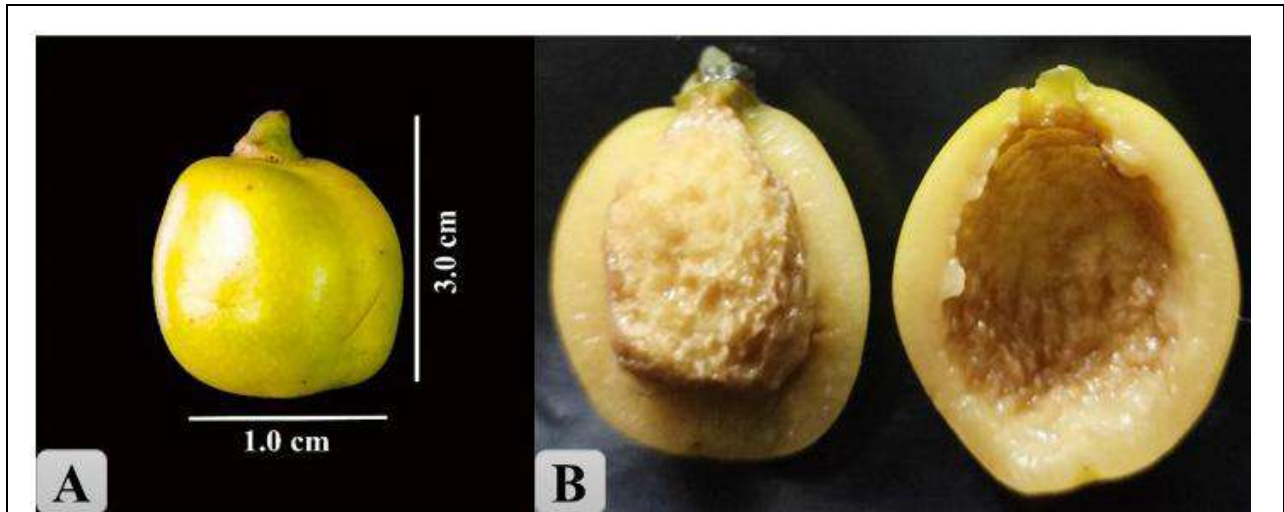


Figure 2. Macroscopic character of *Gmelina asiatica* L. Fruit. A. Whole Fruit; B. Longitudinal split up of fruit.

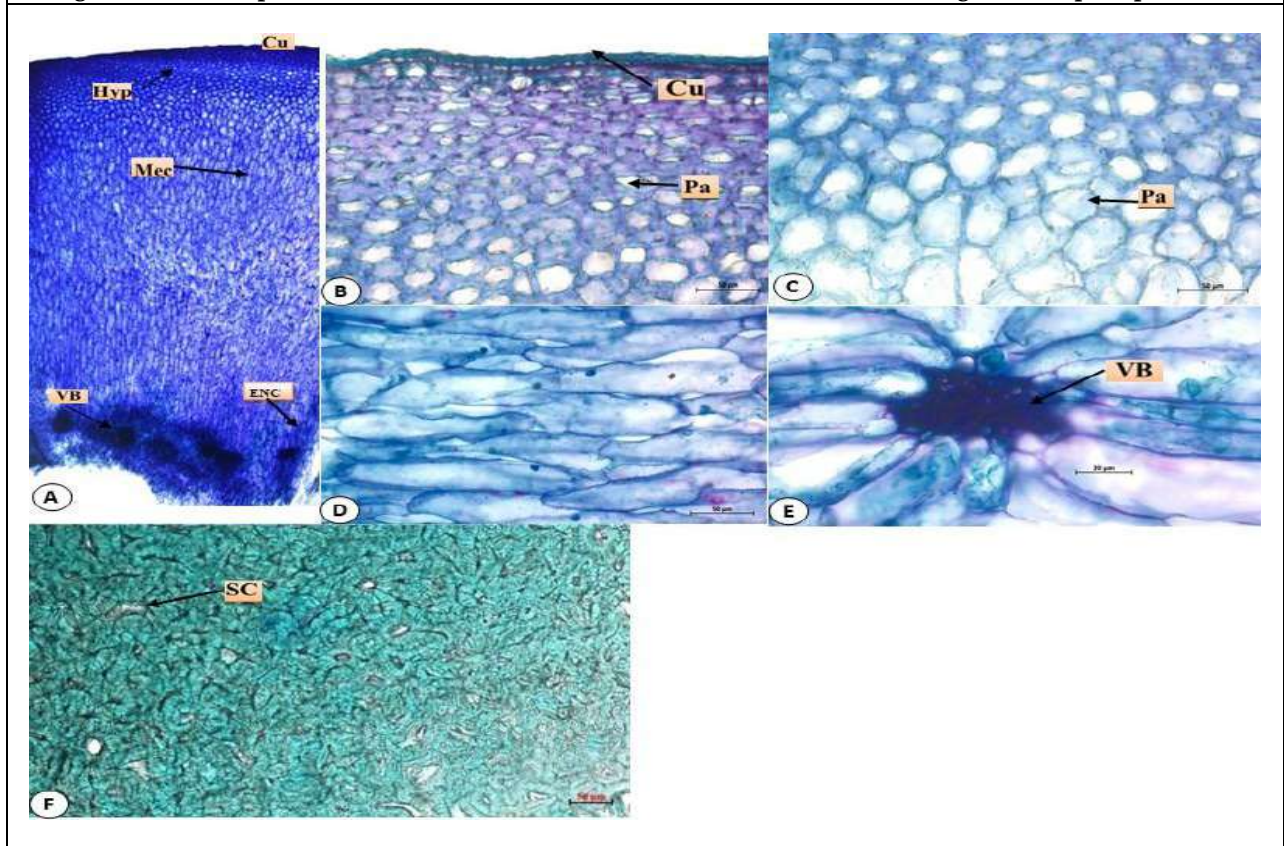


Figure 3. Anatomy of *Gmelina asiatica* L. fruit pulp. A. Transverse section of fruit; B. outer region enlarged; C. inner mesocarp cells; D. outer mesocarp cells; E. vascular bundles in the mesocarp cells; F. stony endocarp.





Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy

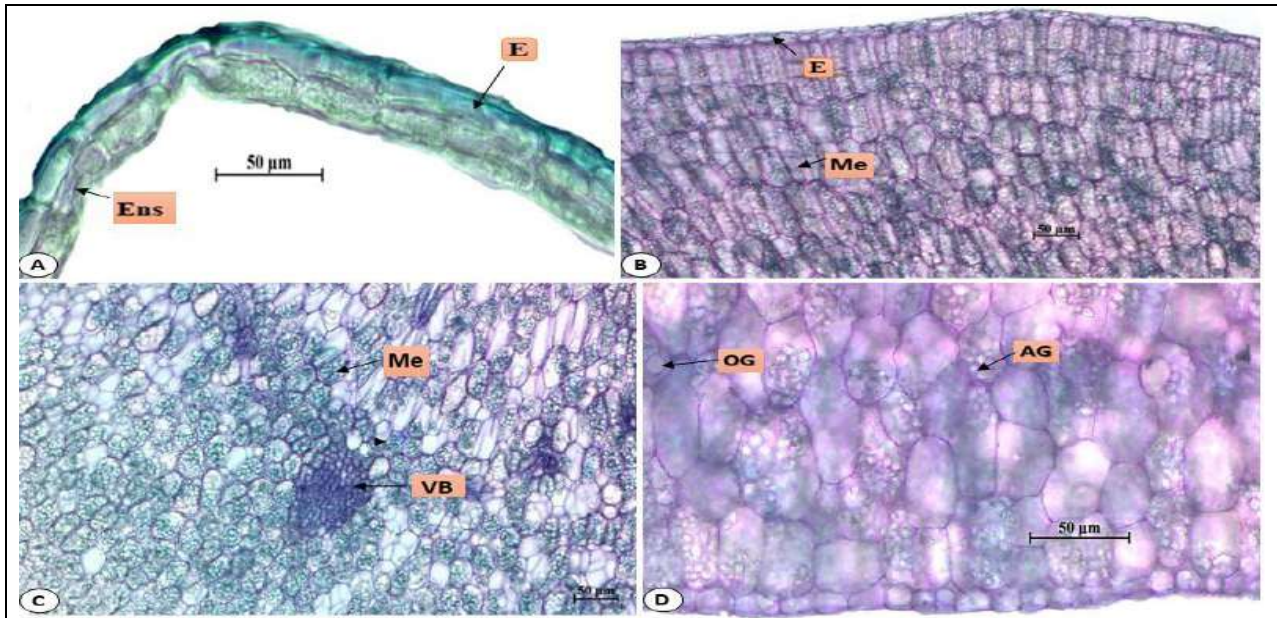


Figure 4. Transverse section of *Gmelina asiatica* L. Seed. A. Testa and endosperm; B. upper region enlarged; C. middle region enlarged; D. lower region enlarged.

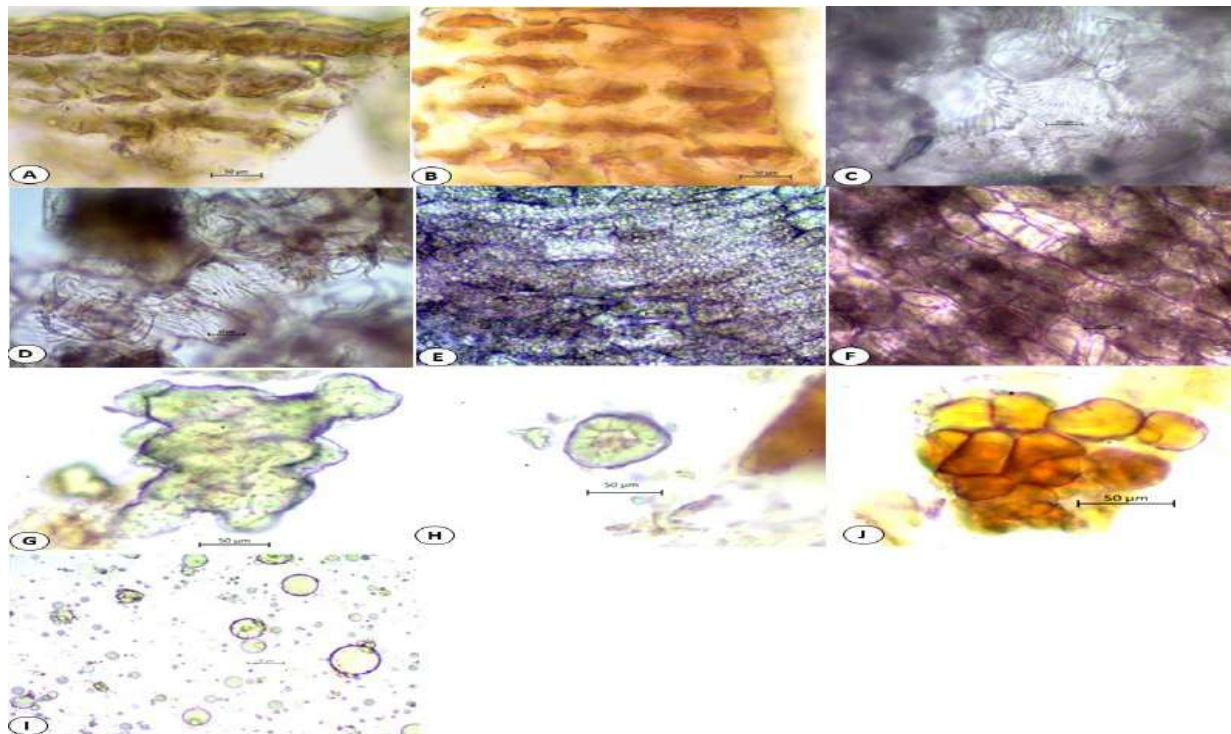


Figure 5. Powder microscopy of *Gmelina asiatica* L. fruit. A. pericarp; B. mesocarp; C and D. cells of the endocarp; E. endosperm cells with starch; F. cotyledon cells; G. sclereids; H. stone cells; I. cells with brownish content; J. oil drops.





Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy

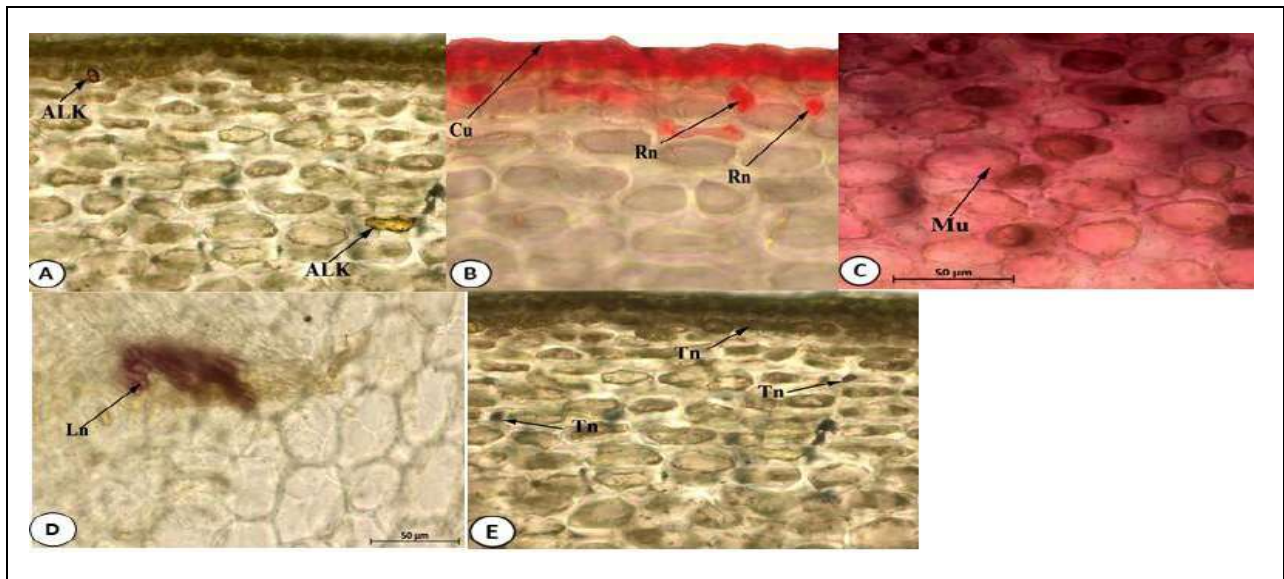


Figure 6. Histo chemistry of fruit pericarp. A. alkaloid; B. cutin and resin; C. mucilage; D. lignin; E. tannin

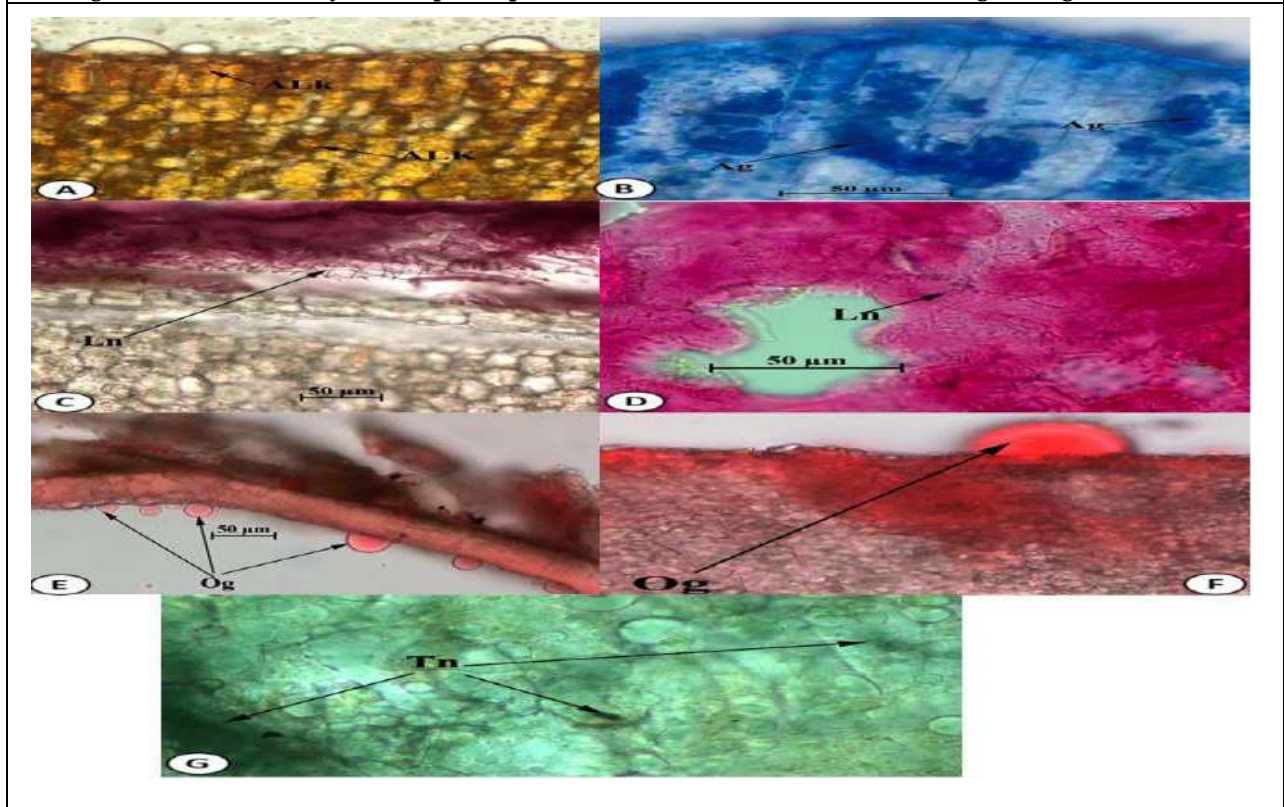


Figure 7. Histochemistry of *Gmelina asiatica* seed. A. alkaloid; B. aleurone grains; C and D. lignin; E and F. oil globule; G. tannin.

Abbreviations: AG-aleurone grains; ALK- alkaloid; Cu- cutin; Cut- cuticle; E-epidermis; Enc-endocarp; Ens- endosperm; Hyp-hypodermis; Ln- lignin; Me- mesophyll; Mec- mesocarp; Mu- mucilage; OG-oil globules; Pa-parenchyma; Rn- resin; Sc-stony endocarp; Tn- tannin; VB-vascular bundle.





Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy

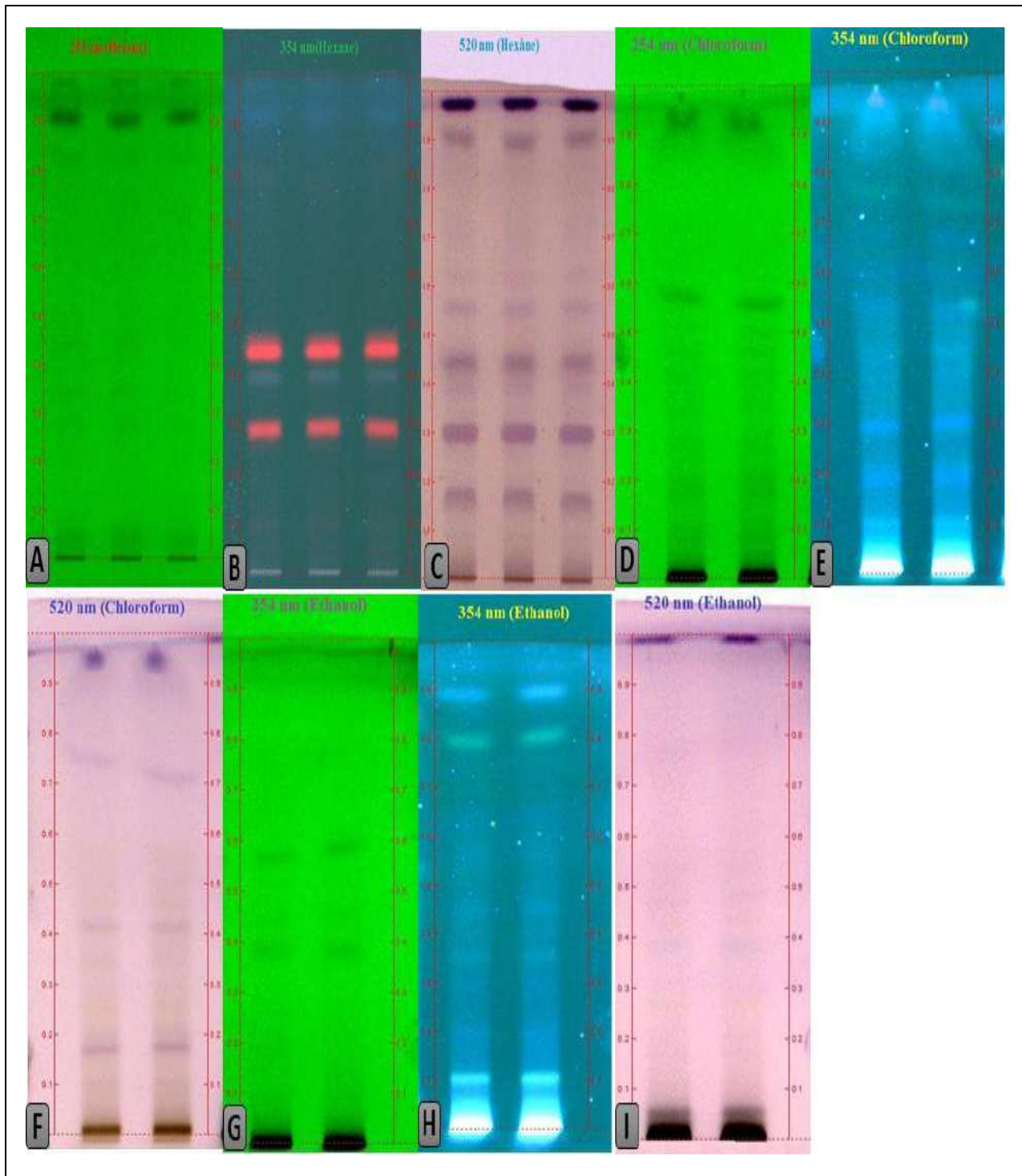


Figure 8. TLC Photo documentation of *Gmelina asiatica* L fruit pulp. A-C: Hexane extract; D-F: Chloroform extract; G-I: Ethanol extract.





Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy

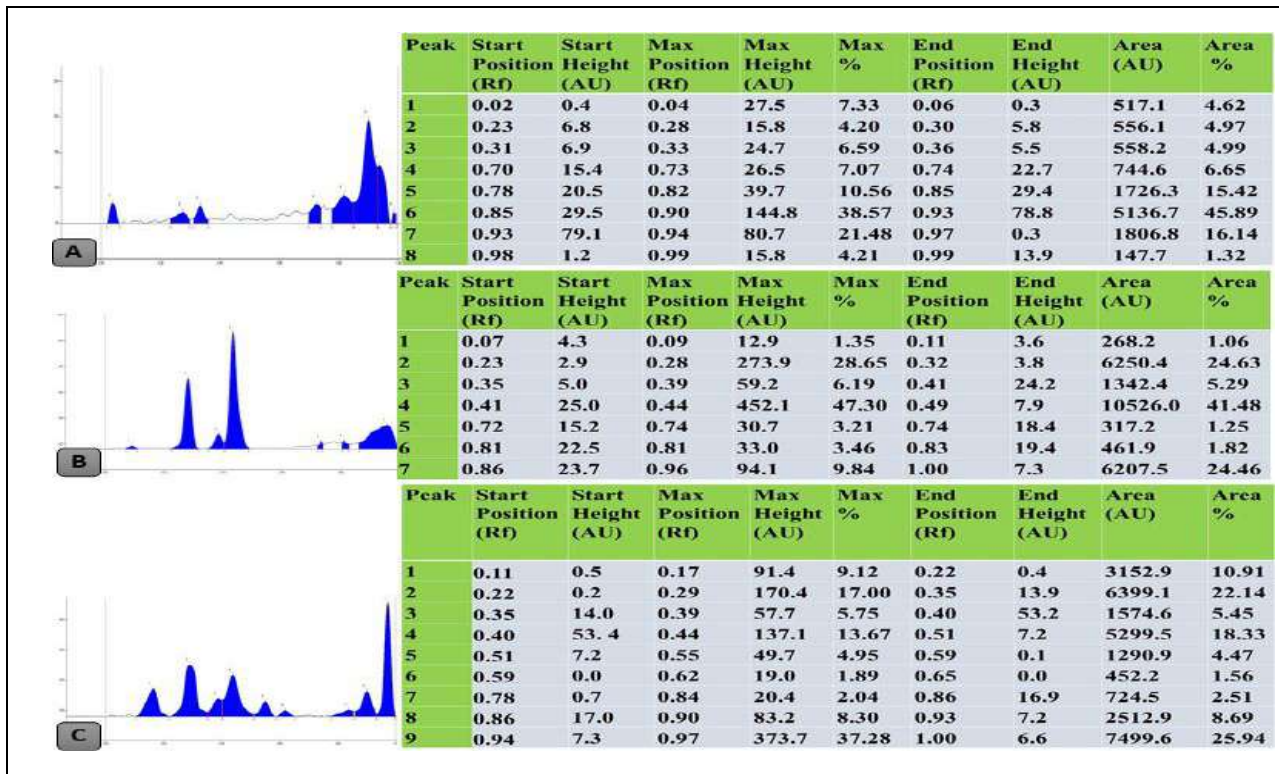


Figure 9. HPTLC fingerprint profile of n-Hexane extract of *Gmelina asiatica* L fruit pulp. a) UV 254 nm. b) UV 354 nm. c) long white light 520 nm.

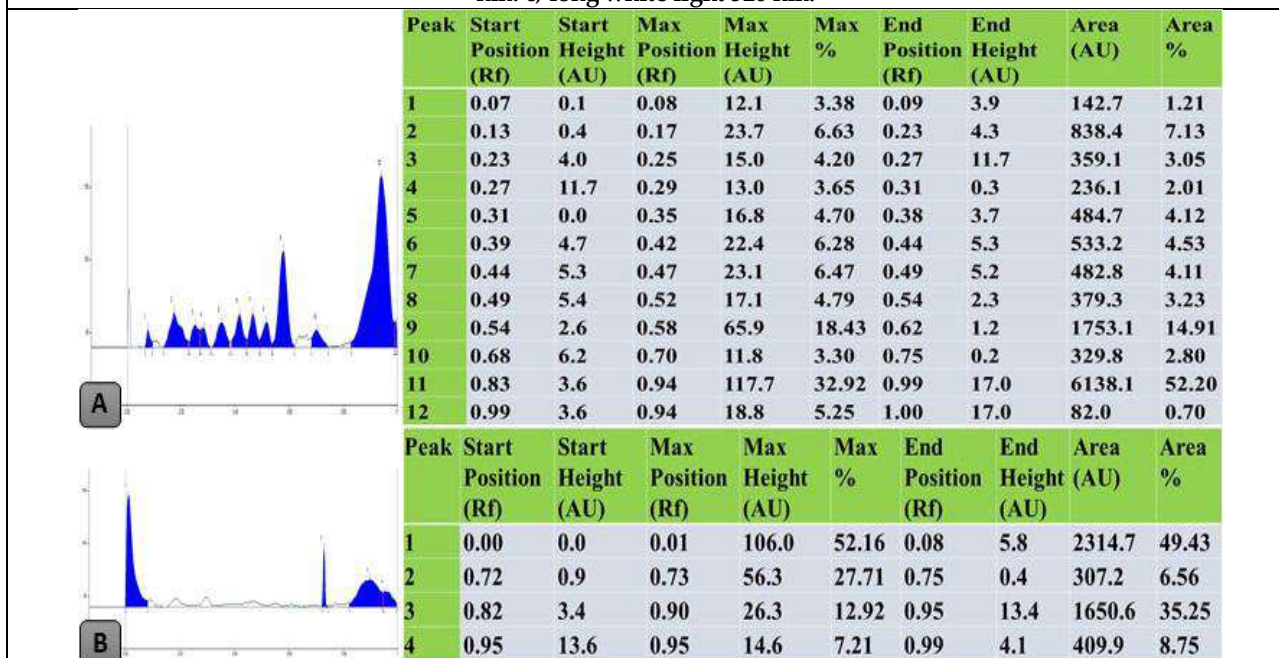
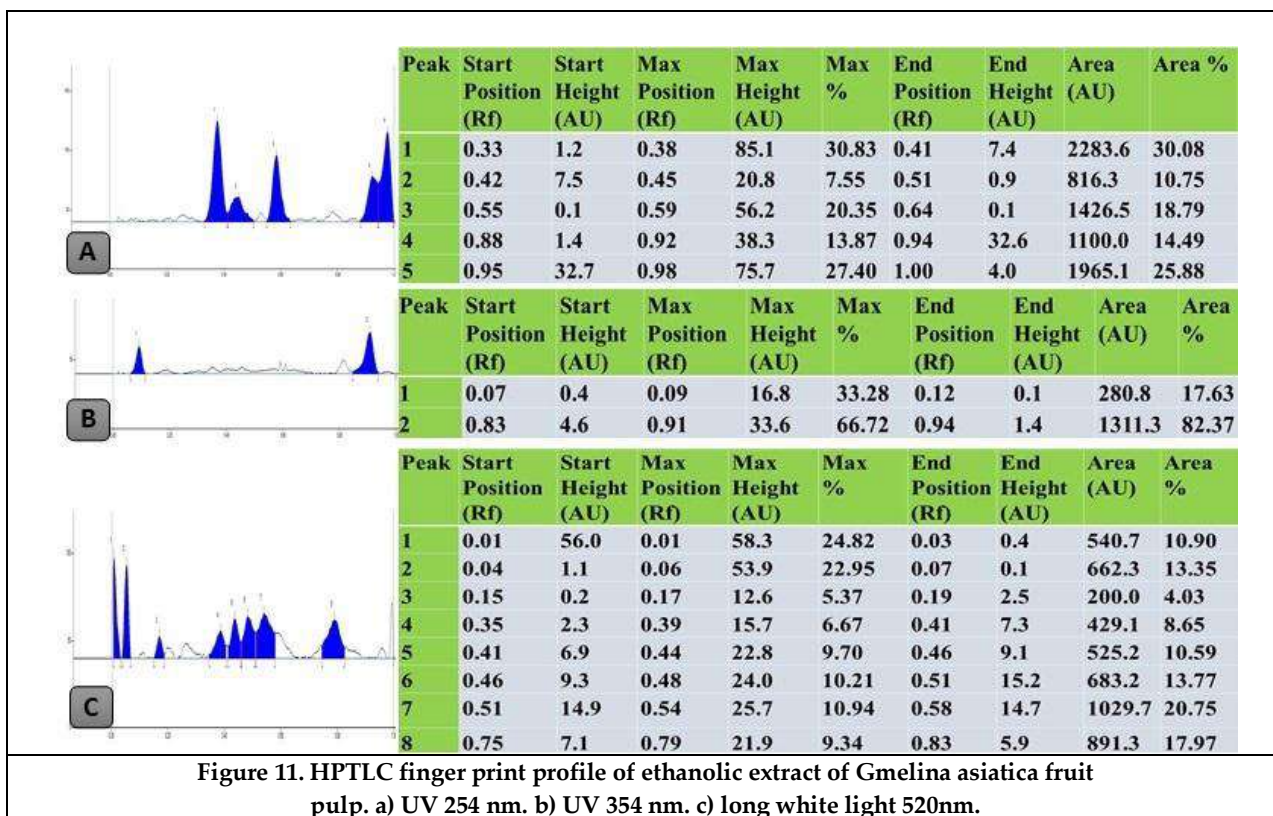


Figure 10 HPTLC finger print profile of chloroform extract of *Gmelina asiatica* L fruit pulp a) UV 254 nm. b) UV 354 nm.





Jaya Prakash Radhakrishnan and Seventhilingam Kaliamoorthy





Microbial Derived Vitamin D2 and Nisin as Dual Function Compounds with Antibiofilm and Anticancerous Activities

Namitha¹ and Tessy Anu Thomas^{2*}

¹Student, Department of Microbiology, Krupanidhi Degree College, (Affiliated to Bengaluru North University), Bengaluru, Karnataka, India.

²Associate Professor, Department of Microbiology, Krupanidhi Degree College, (Affiliated to Bengaluru North University), Bengaluru, Karnataka, India.

Received: 24 Jun 2024

Revised: 20 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

Tessy Anu Thomas

Associate Professor,
Department of Microbiology,
Krupanidhi Degree College,
(Affiliated to Bengaluru North University),
Bengaluru, Karnataka, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

This study was conducted to investigate the potential of microbial- derived commercially available Vitamin D2 and Nisin as dual function compounds with antibiofilm and anticancerous properties; exploring their mechanism of action and therapeutic implications. Utilizing a combination of cultures, comprehensive array of laboratory experiments and analytical tools, precise and robust analysis was carried out. The compounds Vitamin D2 and Nisin were subjected to rigorous testing against *Staphylococcus aureus* and *Pseudomonas aeruginosa* bacterial strains along with A431 SCC cancer cell lines to assess their antibiofilm and anti-cancerous activity respectively. The results demonstrated that microbial- derived Vitamin D2 and Nisin exhibit potential antibacterial effects. Additionally, both compounds also show promising anti-cancerous activity, hindering the proliferation of cancer cells with notable specificity in MTT and Apoptosis assay. This research establishes microbial- derived Vitamin D2 and Nisin as promising dual- action compounds with potential applications in both antibacterial therapies and anticancer treatments, highlighting their multifaceted therapeutic significance.

Keywords: Nisin, Vitamin D2, apoptosis, MTT Assay, A431 SCC cancer cell lines.

INTRODUCTION

The world is on the verge of entering the "post-antibiotic era," a time when the mortality rate from common bacterial infections will surpass that of cancer [1]. Multidrug resistance is one of the most significant global public health threats of the twenty-first century and may also be related to antimicrobial medications. Because of treatment failures and their prevalence in healthcare costs, this phenomenon has indeed increased both mortality and morbidity [2]. Gram-positive and gram-negative bacteria with multidrug resistance have led to infections that are difficult to treat



**Namitha and Tessy Anu Thomas**

or even untreatable with conventional antimicrobials. Additionally, biofilms can be problematic for infection control because they contribute to multidrug resistance. Since biofilms permit microbial cells to briefly enter a metabolically inactive state, antibiotics are rendered ineffective, which leads to disease recurrence because latent bacterial infection is still present within the host [3]. High antibiotic doses for an extended period are typically necessary to combat this cell organization, but these methods frequently fall short, resulting in infection persistence [4]. The treatment and control of cancer is also a major cause of concern for both developing and developed nations. Conventional anti-cancer therapy involves the use of chemical chemotherapeutics and radiation and is often non-specific in action. The development of drug resistance and the inability of the drug to penetrate the tumor cells has been a major pitfall in current treatment.

The creation of novel, "out of the box," therapeutics is required to combat antibiotic-resistant bacterial infections brought on by acquired resistance and/or biofilm formation and the need to investigate alternative anti-tumor therapeutics possessing greater specificity and efficacy is significant [5-6]. Some of the microorganisms produce bioactive secondary metabolites that may be involved in a host-endophyte relationship. Recently, many endophytic bioactive molecules, known as well as new substances, possessing a wide variety of biological activities such as antibiotic, antitumor, anti-inflammatory, antioxidant, etc. have been identified among which Vitamin D2 and Nisin will be assessed in this study. The anti-bacterial and anti-quorum sensing activity of vitamin D2 from fungus is evident from mushrooms but from yeasts remains unexplored. Also, there are restricted resources regarding the anticancerous activity of commercially available vitamin D2 from yeasts. The use of nisin as a single agent or in combination with other conventional medications to treat cancer is still in its infancy. The microbiome and cancer may also be related etiologically, according to mounting evidence.

MATERIALS AND METHODS

Procurement of Samples, Strains & Cell Cultures

Vitamin D2 (S1) and Nisin (S2) were procured from I. A Chemicals, Gujarat and Chihonbio respectively of 10 grams. A total of two microorganisms were used to assess the antimicrobial properties, *Staphylococcus aureus* (MTCC-2408), and *Pseudomonas aeruginosa* (MTCC 2080). A431 (Human skin cancer) cell line was initially procured from the National Centre for Cell Sciences (NCCS), Pune, India, and maintained in Dulbecco's modified Eagle's medium, DMEM (Sigma-Aldrich, USA).

Antibiofilm Activity by Crystal violet Assay Method

Sample preparation

Test samples Vitamin D2 & Nisin were dissolved in 0.02 N HCl & Methanol respectively at a concentration of 100 mg/mL. This was further diluted in LB broth to obtain test concentrations of 0.0097, 0.019, 0.039, 0.078, 0.156, 0.312, 0.625, 1.25, 2.5 & 5 mg/mL. Similar dilutions were prepared for standard Gentamicin. The protocol was laid as per the method by O'Toole [7].

Anti-Cancerous Potency by MTT and Apoptosis Assays on A431 cell lines

The cell line was cultured in a 25 cm² tissue culture flask with DMEM supplemented with 10% FBS, L-glutamine, sodium bicarbonate (Merck, Germany), and an antibiotic solution containing: Penicillin (100 µg/ml), Streptomycin (100 µg/ml), and Amphotericin B (2.5 µg/ml). Cultured cell lines were kept at 37°C in a humidified 5% CO₂ incubator (NBS Eppendorf, Germany).

The viability of cells was evaluated by direct observation of cells by an Inverted phase contrast microscope and followed by the MTT assay method.



**Namitha and Tessy Anu Thomas****Anticancer Evaluation**

After 24 hours the growth medium was removed, and freshly prepared compounds in DMEM were five times serially diluted by two-fold dilution (200 µg, 100 µg, 50 µg, 25 µg, 12.5 µg, 6.25 µg in 500 µl of DMEM) and each concentration of 100 µl was added in triplicates to the respective wells and incubated at 37°C in a humidified 5% CO₂ incubator. Non-treated control cells were also maintained[8].

The percentage of growth inhibition was calculated using the formula:

$$\% \text{ of viability} = \text{Mean of OD samples} / \text{Mean of OD Control} \times 100$$

Apoptosis Assay

After treatment with the sample at LC 50 Concentration (S1-114.29 µg/mL and S2 -137.142 µg/mL) for 24 hours, the cells were washed with cold PBS and then stained with a mixture of AO (100 µg/ml) and EtBr (100 µg/ml) at room temperature for 10 min. The stained cells were washed twice with 1X PBS and observed by a fluorescence microscope in the blue filter of the fluorescent microscope (Olympus CKX41 with Optika Pro5 camera)[9].

STATISTICAL ANALYSIS

All the experiments were performed in triplicates and the results are expressed as Mean ± SD (n = 3). The results were analyzed for statistical significance using the unpaired Students T-test, One-way ANOVA, and Dunnett's test (SPSS Inc. 20.0 version) & ED50 PLUS V1.0 Software. Probability values (P) ≤ 0.05 were statistically significant.

RESULTS**Antibiofilm Activity by Crystal Violet Assay Method**

The biofilm inhibition activity of Vitamin D2 and Nisin was performed against the test microorganisms namely *S. aureus* & *P. aeruginosa*. Gentamicin was used as the positive control. 10 test concentrations of the samples were taken from 5- 0.0097 mg/ml. Vitamin D2 depicted more inhibition activity compared to Nisin. The IC₅₀ values of Vitamin D2, Nisin, and Gentamicin were 0.080 mg/ml, 1.231 mg/ml, and 0.028 mg/ml respectively against *S. aureus* whereas IC₅₀ values of Vitamin D2, Nisin, and Gentamicin were 0.202 mg/ml, 1.453 mg/ml, and 0.037 mg/ml respectively against *P. aeruginosa*. The results are tabulated in Tables 1A & 1B. The comparative analysis of the activity is illustrated in Figures 1A & 1B.

Anticancerous Potency by MTT Assay Method

The test samples Vitamin D2 & Nisin were assessed for their anticancerous potency on human skin cancer cell lines- A431 cell lines. Non-treated cells were used as control. Indicators of cytotoxicity like granulation, rounding, and vacuolization were observed under microscopic examination. The percentage of growth inhibition was also calculated wherein the expressed LC₅₀ Values of Vitamin D2 and Nisin were 114.29 µg/mL and 137.14 µg/mL respectively (Calculated using ED50 PLUS V1.0 Software). The percentage of viability and cytotoxicity are represented in Figures 2A, 2B, 2C, and 2D respectively.

Anticancerous Potency by Apoptosis Assay Method

A431 cell lines appeared to exhibit apoptosis on treatment with test samples Vitamin D2 & Nisin. Non-treated cells were used as control. The cells were divided into four categories as follows: living cells (normal green nucleus), early apoptotic (bright green nucleus with condensed or fragmented chromatin), late apoptotic (orange-stained nuclei with chromatin condensation or fragmentation), and necrotic cells (uniformly orange-stained cell nuclei) which are depicted in Figure 3.





Namitha and Tessy Anu Thomas

DISCUSSION AND CONCLUSION

Along with the struggle to combat AMR, cancer is also a complex and devastating disease that causes a range of health problems. Therefore, a general call for novel and therapeutic naturally occurring metabolites to render or treat infections and diseases points out to be a significant highly effective alternative. In this sense, the antimicrobial and anticancerous activity of the metabolite Vitamin D2 and lantibiotic Nisin was analyzed against microbes and skin cancer SCC cell lines respectively. In the initial screening of antibiofilm activity of the samples, the Crystal violet assay method depicted that both Vitamin D2 and Nisin can inhibit the biofilm formation of the test organisms *S. aureus* and *P. aeruginosa*. This opens a new strategy of using these compounds to resist biofilm formation that has been exacerbating infections. The test samples were taken at a concentration ranging from 5- 0.0097 mg/ml. The IC50 values of Vitamin D2, Nisin, and Gentamicin (positive control) were 0.080 mg/ml, 1.231 mg/ml, and 0.028 mg/ml respectively against *S. aureus* whereas IC50 values of Vitamin D2, Nisin, and Gentamicin were 0.202 mg/ml, 1.453 mg/ml, and 0.037 mg/ml respectively against *P. aeruginosa*.

The anticancerous potency of Vitamin D2 & Nisin analyzed by MTT & Apoptosis assay on Human skin cancer SCC A431 cell lines also proved that these compounds can be used as an alternative to treat skin cancers instead of conventional chemotherapeutic methods which cause severe side effects. Moreover, since Vitamin D2 is non-calcaemic, it can be replaced instead of Vitamin D3. In MTT assay indicators of cytotoxicity like granulation, vacuolation, and rounding were observed. The percentage of growth inhibition was also calculated wherein the expressed LC50 Values of Vitamin D2 and Nisin were 114.29 µg/mL and 137.14 µg/mL respectively. In the Apoptosis assay, cells exhibiting apoptosis were observed under an inverted phase contrast microscope. This work could help in groundbreaking medical interventions that address infection control and cancer treatment challenges simultaneously. The future perspective of this study paves the way for analyzing the action of Nisin and Vit D2 in the gene regulation responsible for biofilm inhibition at the molecular level. Furthermore, by docking methods, the extent of activity by Vitamin D2 and Nisin on targeted cells could also be analyzed.

DECLARATIONS

ACKNOWLEDGEMENT

I acknowledge the Department of Microbiology, Krupanidhi Degree College wherein I have carried out the current work. My sincere gratitude to Biogenix Research Lab wherein I have carried out the cancer studies

Funding

There are no funding bodies in the design of the study and collection, analysis, and interpretation of data and in writing the manuscript.

CONFLICT OF INTEREST

The authors “declare that they have no conflict of interest.”

Ethics Approval and Consent to Participate

“This article does not contain any studies with human participants or animals performed by any of the authors.”

Consent for Publication

This manuscript does not contain any personal data of individuals in any forms.

Availability of Data and Material

“Data sharing not applicable to this article as no datasets were generated or analysed during the current study”.





Namitha and Tessy Anu Thomas

REFERENCES

1. Elmanama, A. A., Al-Shebol, S., & Abu-Dan, R. I. (2020). Antimicrobial resistance and biofilm formation of *Pseudomonas aeruginosa*: a short review article. *The International Arabic Journal of Antimicrobial Agents*, 10(2).
2. Catalano, A., Iacopetta, D., Ceramella, J., Scumaci, D., Giuzio, F., Saturnino, C., ... & Sinicropi, M. S. (2022). Multidrug resistance (MDR): A widespread phenomenon in pharmacological therapies. *Molecules*, 27(3), 616.
3. Ghosh, A., Jayaraman, N., & Chatterji, D. (2020). Small-molecule inhibition of bacterial biofilm. *ACS omega*, 5(7), 3108-3115.
4. Ribeiro, S. M., Felício, M. R., Boas, E. V., Gonçalves, S., Costa, F. F., Samy, R. P., ... & Franco, O. L. (2016). New frontiers for anti-biofilm drug development. *Pharmacology & therapeutics*, 160, 133-144.
5. Jerard, C., Michael, B. P., Chenicheri, S., Vijayakumar, N., & Ramachandran, R. (2020). Rosmarinic acid-rich fraction from *Mentha arvensis* synchronizes Bcl/Bax expression and induces Go/G1 arrest in hepatocarcinoma cells. *Proceedings of the National Academy of Sciences, India Section B: Biological Sciences*, 90, 515-522.
6. Sawant, S. S., Patil, S. M., Gupta, V., & Kunda, N. K. (2020). Microbes as medicines: harnessing the power of bacteria in advancing cancer treatment. *International journal of molecular sciences*, 21(20), 7575.
7. O'Toole, G. A. (2011). Microtiter dish biofilm formation assay. *JoVE (Journal of Visualized Experiments)*, (47), e2437.
8. Makabenta, J. M. V., Nabawi, A., Li, C. H., Schmidt-Malan, S., Patel, R., & Rotello, V. M. (2021). Nanomaterial-based therapeutics for antibiotic-resistant bacterial infections. *Nature Reviews Microbiology*, 19(1), 23-36.
9. Zhang, J. H., Yu, J., Li, W. X., & Cheng, C. P. (1998). Inhibited Apoptosis in Rat Corpus Luteal Cells by Flow Cytometry and Fluorochromes. *Chinese Journal of Physiology*, 41(2), 121-126.

Table 1A.0020 Antibiofilm Activity of Vitamin D2 & Nisin against *Staphylococcus aureus*

Crystal violet OD @ 595 nm	Test Concentrations (mg/ml)												
	Blank	Untreated	0.0097	0.019	0.039	0.078	0.156	0.312	0.625	1.25	2.5	5	
Gentamicin (Positive control)													
Mean OD ± SD	0.06	1.5 ± 0.1	1.42 ± 0	1.25 ± 0.03	0.2 ± 0	0.05 ± 0.02	0.05 ± 0	0.06 ± 0.03	0.04 ± 0	0.05 ± 0	0.0 ± 0	0.0 ± 0	0.05 ± 0.0
% of Biofilm Inhibition		0	5.3	16.6	86.6	96.7	96.7	96	97.3	96.7	96	96.7	96.7
IC50= 0.028 mg/mL													
Vitamin D2 (S1)													
Mean OD ± SD	0.06	1.5 ± 0.1	1.53 ± 0.12	1.44 ± 0.07	1.5 ± 0.14	0.8 ± 0.11	0.2 ± 0.06	0.06 ± 0.02	0.06 ± 0.0	0.07 ± 0.02	0.05 ± 0.0	0.0 ± 0.0	0.06 ± 0.02
% of Biofilm Inhibition		0	-2	4	0	46.7	86.7	96	96	95.3	96.7	96	96
IC50= 0.080 mg/mL													
Nisin (S2)													
Mean OD ± SD	0.06	1.5 ± 0.1	1.6 ± 0.04	1.6 ± 0.08	1.6 ± 0.1	1.6 ± 0.02	1.43 ± 0	1.52 ± 0.04	1.6 ± 0.03	0.51 ± 0.02	0.2 ± 0.0	0.2 ± 0.0	0.04 ± 0.01





Namitha and Tessy Anu Thomas

												2	
% of Biofilm Inhibition		0	-6.67	-6.67	-6.67	-6.67	4.6	-1.3	-6.67	66	86.7		97.3
IC50= 1.231 mg/mL													

Table 1B. Antibiofilm Activity of Vitamin D2 & Nisin against *Pseudomonas aeruginosa*

Crystal violet OD @ 595 nm	Test Concentrations (mg/ml)												
	Blank	Untreated	0.0097	0.019	0.039	0.078	0.156	0.312	0.625	1.25	2.5	5	
Gentamicin (Positive control)													
Mean OD ± SD	0.06	1.41 ± 0.03	1.35 ± 0.05	1.41 ± 0.05	0.5 ± 0.07	0.2 ± 0.05	0.09 ± 0.02	0.07 ± 0	0.04 ± 0.02	0.06 ± 0	0.05 ± 0.03	0.05 ± 0	0.07 ± 0
% of Biofilm Inhibition		0	4.2	0	64.5	85.8	93.6	95	97.1	95.7	96.4	95	
IC50= 0.037 mg/mL													

Vitamin D2 (S1)												
Mean OD ± SD	0.06	1.5 ± 0.1	1.53 ± 0.12	1.44 ± 0.07	1.5 ± 0.14	0.8 ± 0.11	0.2 ± 0.06	0.06 ± 0.02	0.06 ± 0.0	0.07 ± 0.02	0.05 ± 0.01	0.06 ± 0.02
% of Biofilm Inhibition		0	-2	4	0	46.7	86.7	96	96	95.3	96.7	96
IC50= 0.202 mg/mL												
Nisin (S2)												
Mean OD ± SD	0.06	1.41 ± 0.03	1.6 ± 0.07	1.62 ± 0.09	1.6 ± 0.07	1.4 ± 0.02	1.4 ± 0.03	1.33 ± 0.01	1.34 ± 0.05	1.21 ± 0.08	0.13 ± 0.01	0.06 ± 0
% of Biofilm Inhibition		0	-13.5	-14.9	-13.5	0.71	0.71	5.7	5.0	14.2	90.8	95.7
IC50= 1.453 mg/mL												

Data shown are the average and standard deviation based on duplicate runs (Mean ± Standard Deviation)

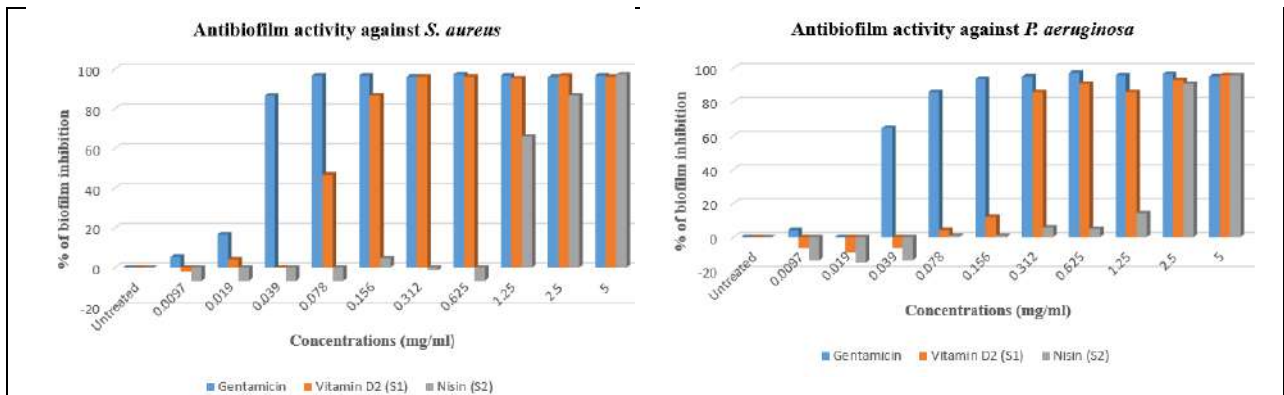


Figure 1A: Antibiofilm activity of Vitamin D2 & Nisin against *Staphylococcus aureus*

Figure 1B: Antibiofilm activity of Vitamin D2 & Nisin against *P. aeruginosa*





Namitha and Tessy Anu Thomas

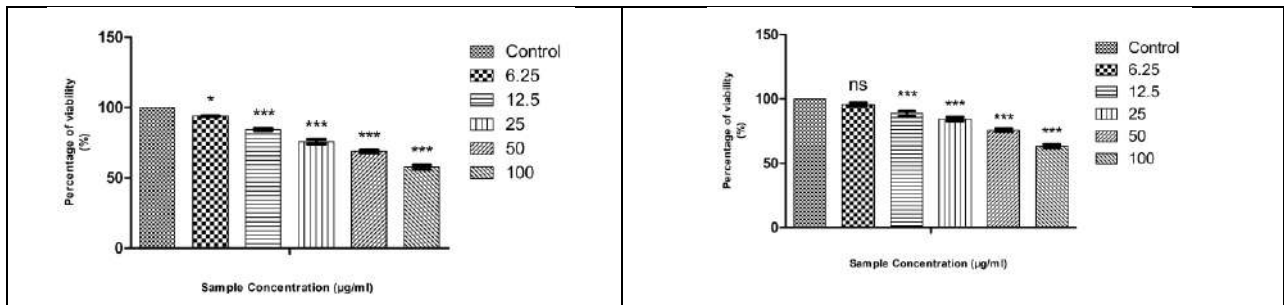


Figure 2A: Graphical representation depicting the anticancer effect of Vitamin D2 (S1) on cell line by MTT assay. Along the Y axis Percentage viability, Along the X axis varied concentration of S1. All experiments were done in triplicates and results were represented as Mean+/- SE. One-way ANOVA and Dunnett's test were performed to analyze data. ***p < 0.001 compared to control groups, **p < 0.01 compared to control groups, *p < 0.1 compared to control groups.

Figure 2B: Graphical representation depicting the anticancer effect of Nisin (S2) on A431 cell line by MTT assay. Along the Y axis Percentage viability, Along the X axis varied concentrations of S2. All experiments were done in triplicates and results were represented as Mean+/- SE. One-way ANOVA and Dunnett's test were performed to analyze data. ***p < 0.001 compared to control groups, **p < 0.01 compared to control groups, *p < 0.1 compared to control groups.

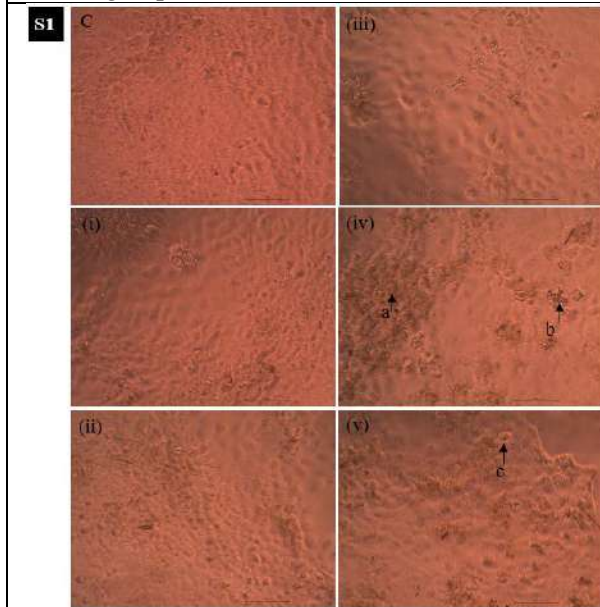


Figure 2C: Microscopic examination of cytotoxicity of Vitamin D2 on A431 cell lines under 40X magnification in inverted phase contrast microscope. [C- Untreated control cells exhibiting epithelial morphology; (i)- 6.25 µg/ml; (ii)- 12.5 µg/ml; (iii)- 25 µg/ml; (iv)- 50 µg/ml; (v)- 100 µg/ml; a- granulation; b- vacuolization; c- rounding which is considered as a sign of cytotoxicity].

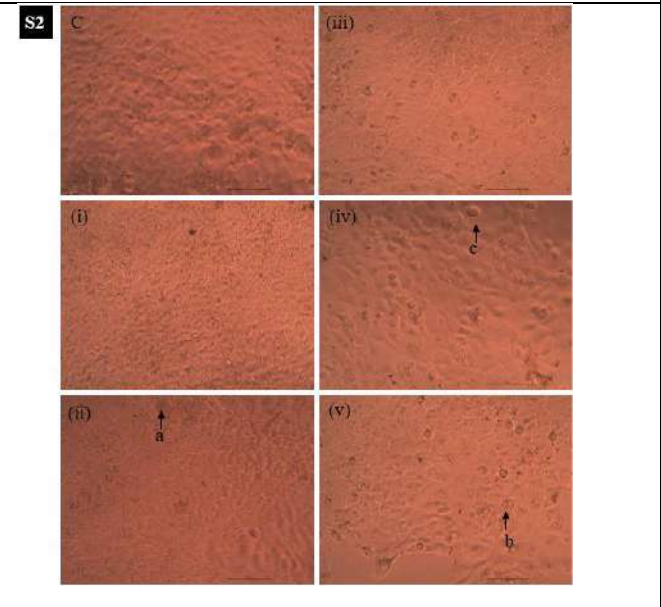


Figure 2D: Microscopic examination of cytotoxicity of Nisin on A431 cell lines under 40X magnification in inverted phase contrast microscope. [C- Untreated control cells exhibiting epithelial morphology; (i)- 6.25 µg/ml; (ii)- 12.5 µg/ml; (iii)- 25 µg/ml; (iv)- 50 µg/ml; (v)- 100 µg/ml; a- granulation; b- vacuolization; c- rounding which is considered as a sign of cytotoxicity]





Namitha and Tessy Anu Thomas

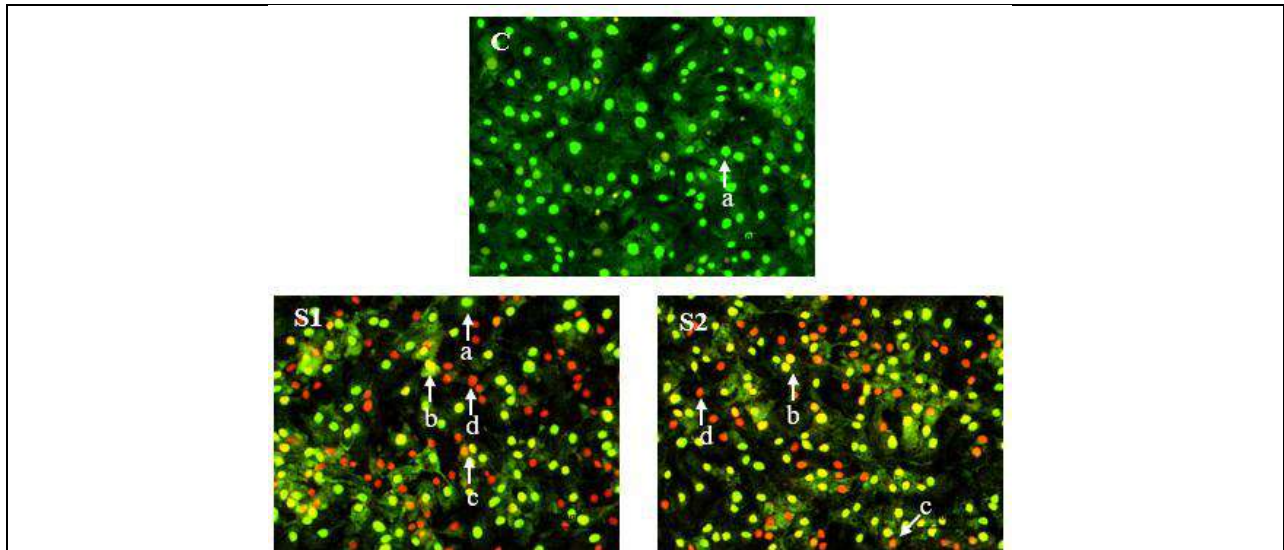


Figure 3. Apoptosis of cells on treatment with Vitamin D2 & Nisin observed in a fluorescent microscope [S1- Vitamin D2; S2- Nisin; C- Control; a- Living cells; b- early apoptotic cells; c- late apoptotic cells; d- necrotic cells].





Efficacy of Kiastm along with Motor Control Training Exercises in Chronic Non-Specific Neck Pain Patients - A Pilot Study

Lopa Das^{1*}, Shefali Raizada² and Prem Kumar B N³

¹Ph.D Scholar, Department of Physiotherapy, Garden City University, Begaluru, Karnataka, and / Associate Professor, Department of Physiotherapy, The Assam Royal Global University, Guwahati, Assam, India.

²Assistant Professor, Department of Health Science, Garden City University, Begaluru, Karnataka, India.

³Associate Professor, Department of Physiotherapy, Kempegowda Institute of Physiotherapy, (Affiliated to Rajiv Gandhi University of Health Sciences), Bengaluru, Karnataka, India.

Received: 27 Feb 2024

Revised: 18 Aug 2024

Accepted: 26 Oct 2024

*Address for Correspondence

Lopa Das

Ph.D Scholar, Department of Physiotherapy,
Garden City University, Begaluru, Karnataka, and /
Associate Professor, Department of Physiotherapy,
The Assam Royal Global University,
Guwahati, Assam, India.
E.Mail: anusree1984@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Neck pain has been referred to as “nonspecific” or “mechanical” neck pain since the great majority of cases do not result from organic pathology. KIASTM (“Kinesio Instrument Assisted Soft Tissue Mobilization”) is a method in which soft tissue components are mechanically stimulated by tools to reduce musculoskeletal pain and discomfort and enhance mobility and function. Generally, it explains the use of an Accel instrument, which is ergonomically designed to help doctors identify and treat soft tissue pain, injury, and dysfunction. Numerous prospective analyses have revealed that people with chronic neck pain may also have weak neck muscles and motor control problems. The MCE (Motor Control Exercise) is a treatment technique that primarily focuses on motor control, and activation of deep cervical muscles, as well as aims to retrain the best control & coordination of the cervical muscles. The objective of this research was to check the effectiveness of Motor control training exercises and KIASTM in Chronic nonspecific neck pain patients in terms of pain, soft tissue mobility, and functional disability. On the basis of inclusion criteria, 5 subjects were selected and baseline data were collected on day one for Pain using VAS (Visual Analogue Scale), Universal Goniometer for Cervical range of motion, DNF (Deep Neck Flexor) Endurance Testing for deep neck flexors, and functional disability using NDI (Neck Disability Index). Interventions applied include Transcutaneous Electrical Nerve Stimulation for 15 minutes followed by application of KIASTM for 90 seconds as per protocol and Motor control training exercises for 3 sessions per week. The total duration of treatment was 3 weeks. After 3 weeks of interventions, the results showed significant improvement in terms of pain, ROM, DNF Endurance level, and disability following applications of KIASTM along with the Motor Control Exercise program in Chronic nonspecific neck pain cases. A considerable amount of improvement was observed following the application of KIASTM along



**Lopa Das et al.,**

with other therapeutic interventions like TENS and MCEsin reducing pain, increasing AROM, improvement in function, and deep neck flexor endurance during the rehabilitation of chronic nonspecific neck pain patients.

Keywords: Nonspecific neck pain, IASTM, Motor Control exercise, Physiotherapy.

INTRODUCTION

Neck pain is among the most frequent and serious musculoskeletal conditions. Point prevalence range between 6 to 22 percent, and in the elderly population, it may reach up to 38 percent, whereas lifetime prevalence varies from 14.2 to 71 percent [1]. These are assumed to be generalized neck pain. Nonspecific neck pain is just neck pain without a known underlying illness as the source of the pain. Symptoms change with physical exercise and with time. Nonspecific neck pain refers to any kind of chronic neck pain, subacute, or acute when no abnormal anatomic feature could be recognized as the source of the pain. There are several viewpoints about the symptom's duration but as per Binder, neck pain may be acute (less than 4 weeks duration), sub-acute (1-4 months duration), or chronic (more than 4 months duration). Chronic neck pain may result in significant medical expenses, job absences, and disability. Depending on the length, neck pain may affect functional ability and quality of life, in addition to causing stress, depression, and anxiety [2]. Therefore, neck pain exerts a significant burden on people, employers, as well as healthcare systems. Existing literature explored many diverse treatment approaches for nonspecific neck pain. As the majority of the patients with nonspecific neck pain have nondefinitive pathology, hereby fail to direct treatment. Many patients get conservative care from a general physiotherapist or a physician but there is always a lack of proper indications of the conditions for any specific management. The manual treatment method recognized as IASTM ("Instrument-Assisted Soft-Tissue Mobilization") locates and treats soft tissue problems by using rigid tools of different forms and materials. IASTM is a non-invasive treatment method that is often used by stroking the edge of a tool on the skin's surface, sometimes with the help of lubricant, with the goal of affecting the underlying connective tissues, muscles, as well as nerves [3]. As such, many IASTM instruments, companies, and proposed application protocols. IASTM is an umbrella term. Various types of brands are available. Various types include KIASTM, Sound Assisted Soft tissue mobilization, Gua Sha, Graston Technique, Fascial Abrasion Technique, and Augmented Soft Tissue Mobilization Technique. KIASTM is a process where soft tissue components are mechanically stimulated with the use of devices to reduce musculoskeletal pain and discomfort and enhance general mobility and functional capacity. It often refers to using an accel tool. Accel tool, which is ergonomically created to help professionals identify and treat soft tissue pain, damage, and dysfunction. The KIASTM method also incorporates a standard protocol to be followed while applying the technique in various musculoskeletal and neurological conditions with the involvement of myofascia. It includes the following techniques: scanning to check for limits across the affected region, combining with deeper, multidirectional strokes, and gliding to provide a cool-down period.

According to prospective research, those who suffer from persistent neck discomfort have weak neck muscles and reduced motor function. The therapy method that primarily focuses on motor control, and deep cervical muscles activation, and seeks to retrain the cervical muscles' ideal control and coordination is known as motor control exercises. It has been demonstrated that the MCE, which is typically formed under supervision, improves motor control while decreasing pain and impairment in individuals with neck discomfort. The exercise focuses on the deep flexor muscles of the upper cervical region, the longus capitis and longus colli muscles, as opposed to the superficial flexor muscles, the sternocleidomastoid and anterior scalene, which flex the neck but not the head [4]. The motor control is characterized as a motor relearning program that places a focus on the coordination and holding capacities of certain neck flexors, extensors, and shoulder girdle muscles. As there is a lack of enough supporting literature regarding the usage of IASTM in chronic nonspecific neck pain and also addressing the weakness of deep neck flexors in the same condition, the management of this condition becomes challenging for the therapist. Chronic nonspecific neck pain has become alarming for the global population due to workload demand, excessive usage of electronic gadgets, and the sedentary lifestyle of the present generation. Weakness of deep neck flexors will lead to neuromuscular incoordination and ultimately failure of a rehabilitation program. So addressing the pain, fascia, and



**Lopa Das et al.,**

muscle tightness with the help of Kinesio Instrument Assisted Soft Tissue Mobilization along with Motor control Training exercises was the prime objective of this pilot study. Thereby the need for this pilot study to check the effectiveness of KIASTM along with the Motor Control Training Exercise program in chronic nonspecific neck pain.

METHODOLOGY

Study Design

A group of 6 patients participated in the single-group experimental pilot study after fulfilling the inclusion as well as exclusion criteria. Participants have been assessed based on the level of pain with VAS, range of motion (ROM) using goniometry, DNF Endurance testing, and disability score using Neck Disability Index during the preintervention and postintervention phases. The pilot research was conducted for 3 weeks. Before the research started, each of the selected individuals signed an informed permission form in line with the Helsinki Rules and expressed their intention to take part in the study.

Study Population

Calculation of the sample size was done after a survey was performed where it has been found that the population proportion of patients reported with nonspecific neck pain is 1.1%. The required size of the sample was computed with Cochran's formula for infinite population keeping a confidence interval of 99.9%, margin of error 5%, and population proportion 1.1%. Participants of both sexes, complaining of persistent nonspecific neck pain, and falling within the age range of 18 to 40 years were required for participation in the research. On the other hand, individuals who had any neck injuries in the three months previous to the trial had spine surgery during the preceding six months, missed more than 15 percent of the intervention sessions (2 sessions), and did not sign the informed consent form were excluded from the study.

Procedure

Physiotherapy interventions were given with a frequency of three sessions every week for a duration of 3 weeks. The interventions were applied after fulfilling the inclusion criteria by the patient and after collecting informed consent. The therapy sessions included the application of TENS, KIASTM, and Motor Control Training exercise protocol. TENS was given for 15 minutes for each patient followed by the application of KIASTM for 90 seconds using Accel Tool with a three-step process starting with scanning of the affected area in the neck region primarily focusing the upper trapezius muscle fibers on both sides, then application of the tool with little deeper pressure to release the restricted soft tissue and lastly by stroking the muscle fiber with light pressure as a cool-down phase for winding up the treatment. After the application of KIASTM, patients were asked to complete the Motor Control Training Exercise protocol (Griffith et al.) [6] with proper instructions from the therapist. The subjects have been also asked to keep the activity of the deep neck flexor group of muscles by incorporating them into their daily functional activities.

Four dependent variables were evaluated: pain using Visual Analogue Scale (VAS), range of motion for cervical extension, lateral flexion, flexion, as well as rotation by a universal goniometer, muscle strength of deep neck flexors by using DNF Endurance testing, and lastly functional level by Neck Disability Index (NDI) pre-intervention & post intervention. Level of pain was evaluated by VAS which comprises a line, often 10cm long, with verbal anchors at either end, similar to the Numerical Rating Scale depicted as "no pain" on the far left and "the most intense pain imaginable" on the far right. The patient marks the point on the line that corresponds to their assessment of the level of pain.

The Harris et al. approach was used for conducting the DNF muscular endurance test for this investigation. The test was carried out in a hook-lying, supine posture. The participant was expected to flex the chin to its maximum extent and maintain the chin tuck posture for the duration of the test. They were then instructed to elevate their heads until they were about 2.5 cm above the plinth while retaining a chin tuck. Once the participant was in place, a line was drawn across their two approximate neck skin folds. On the surface of the table, right below the participant's occiput,



**Lopa Das et al.,**

the examiner positioned his cephalad hand. When the line edges started to split or the participant's head contacted the therapist's hand, further verbal directions were provided. The test was stopped if the participant's head contacted the therapist's hand for more than a second or if the lines on the chart were no longer approximated owing to loss of the chin tuck posture [7].

RESULTS

The current research was intended to check the combined role of the effectiveness of KIASTM & Motor Control Training Exercises along with conventional physiotherapy in chronic nonspecific neck pain patients. The pre-intervention VAS Mean score was 7.4 with a "Standard Deviation" (SD) of 0.548. The correlation between the pre-intervention VAS scores and post-intervention scores was 0.667. The t-test comparing pre- and post-intervention VAS scores showed a significant difference with a high t-value of 11.18 ($p < 0.001$). Post Intervention VAS Mean score was 2.4, indicating a significant improvement from the pre-intervention score. Pre-Intervention DNS (Deep Neck Strength) testing Mean score was 12.618 with a SD of 4.47405. The correlation between the pre-intervention DNS scores and post-intervention DNS scores was 0.855. The t-test comparing pre- and post-intervention DNS scores showed a significant difference with a t-value of -7.289 ($p = 0.002$).

Pre-Intervention NDI (Neck Disability Index) Mean score was 39.94 with a SD of 5.1247. The relationship between the pre-intervention NDI scores and the post-intervention NDI scores was 0.612. The t-test comparing pre- and post-intervention NDI scores showed a significant difference with a t-value of 12.010 ($p < 0.001$). Post Intervention NDI Mean score was 14.7, indicating a significant improvement from the pre-intervention score. Pre-Intervention CF (Cervical Flexion) Mean score was 47.2 with a SD of 5.119. The relationship between the pre-intervention CF scores and the post-intervention CF scores was -0.273. The t-test comparing pre- and post-intervention CF scores showed a substantial difference with a t-value of -3.860 ($p = 0.018$). Post Intervention CF Mean score was 57.6, suggesting an improvement in cervical flexion compared to the pre-intervention scores.

Pre-Intervention CE (Cervical Extension) Mean score was 59.6 with a SD of 9.397. The correlation between the pre-intervention CE scores and the post-intervention CE scores was 0.754. The t-test comparing pre- and post-intervention CE scores showed a significant difference with a t-value of -3.469 ($p = 0.026$). Post Intervention CE Mean score was 69.2, indicating an improvement in cervical extension compared to the pre-intervention scores. Pre-Intervention RLF (Right Lateral Flexion) Mean score was 44 with a standard deviation of 2.646. The correlation between the pre-intervention RLF scores and the post-intervention RLF scores was 0.125. The t-test comparing pre- and post-intervention RLF scores showed a significant difference with a t-value of -3.570 ($p = 0.023$). Post Intervention RLF Mean score was 48.6, suggesting an improvement in right lateral flexion compared to the pre-intervention scores.

Pre-Intervention LLF (Left Lateral Flexion) Mean score was 36.6 with a SD of 4.775. The relationship between the pre-intervention LLF scores and the post-intervention LLF scores was -0.255. The t-test comparing pre- and post-intervention LLF scores did not reach statistical significance with a t-value of -2.211 ($p = 0.091$). Post Intervention LLF Mean score was 44.4, suggesting an improvement in left lateral flexion compared to the pre-intervention scores. Pre-Intervention RLR (Right Lateral Rotation) Mean score was 54 with a SD of 7.517. The relationship between the pre-intervention RLR scores and the post-intervention RLR scores was 0.805. The t-test comparing pre- and post-intervention RLR scores did not reach statistical significance with a t-value of -2.092 ($p = 0.105$). Post Intervention RLR Mean score was 59.4, indicating an improvement in right lateral rotation compared to the pre-intervention scores.

Pre-Intervention LLR (Left Lateral Rotation) Mean score was 54.2 with a SD of 2.588. The relationship between the pre-intervention LLR scores and the post-intervention LLR scores was -0.382. The t-test comparing pre and post-intervention LLR scores did not reach statistical significance with a t-value of -2.409 ($p = 0.074$). Post Intervention LLR



**Lopa Das et al.,**

Mean score was 63, suggesting an improvement in left lateral rotation compared to the pre-intervention scores. In summary, the overall intervention showed statistically significant improvements in VAS, DNS, NDI, CF, CE, RLF, and LLF scores. However, post-intervention CF, RLR, and LLR scores indicated higher levels of disability or limited improvement compared to the pre-intervention scores.

DISCUSSION

The proposed physical therapy protocol indicated substantial enhancement in terms of neck pain reduction and an increase in deep neck muscle endurance and functional level in patients having chronic nonspecific neck pain. These findings have contributed to a few earlier pieces of literature where instrument-aided soft tissue mobilization and motor control training exercises for deep neck muscles have shown positive outcomes in separate studies for chronic nonspecific neck pain cases. The present study also has a few limitations like a small sample size and only female participants for which the findings cannot be generalized for everyone. More number of participants including both genders could be considered for future studies. The present pilot study showed a reduction of pain and an increase in the cervical range of motions in all directions which is contributing to previous studies where the use of IASTM and neuromuscular exercises have shown better results in terms of forward head position and functioning (Mylonas K et al.) [8]. Other studies have also demonstrated the immediate effect of IASTM for the reduction of pain in chronic nonspecific neck pain patients along with correction of joint position error (Gereck H et al.) [9]. The impact of Motor Control Training Exercises was demonstrated in very limited literature for neck pain. The role of Motor Control Training Exercises in improving the strength of deep neck muscles has been demonstrated in the present pilot study while calculating the DNS testing results for each participant. Therefore, incorporating exercises for activating deep neck muscles even during functional activities of daily life will have a significant impact in maintaining and improvement of deep neck muscle endurance thereby reduction of neck disability in patients having chronic nonspecific neck pain. These results are contributing to similar findings in previous studies in the form of systematic review and meta-analysis conducted where incorporating Motor Control Training Exercises compared to other general exercises in chronic nonspecific neck pain were found to be more effective (Carmen Martin Gomez et al.) [10]. Recent study have shown benefits of application of resistance exercise, mindfulness based and motor control exercises in reducing chronic neck pain [11]. However studies have also shown in recent times that there were insignificant outcomes of using IASTM and manual therapy in patients with chronic neck pain among college going students(Fatima Shewail et al.) [12].

CONCLUSION

The pilot study to check the efficacy of the KIASTM along with Motor Control Training Exercises in chronic nonspecific neck pain has shown significant improvement in reducing pain, enhancing neck mobility, deep neck muscle endurance, and disability level. The participants showed positive outcomes after a single session of the protocol. However future studies with large sample sizes and considering the psychological status assessment of the patients might give a wider perspective of chronic nonspecific neck pain. Also future studies could be conducted by keeping a control group to check the efficacy of the treatment protocol.

CONFLICT OF INTEREST

No conflict of interest.

SOURCE OF FUNDING

Self





Lopa Das et al.,

REFERENCES

1. Fejer et al. The prevalence of neck pain in the world population: a systematic critical review of the literature. *European Spine Journal*. 2006,15(6): 834–848.
2. Victoria Misalidou et al. Assessment of patients with neck pain: a review of definitions, selection criteria, and measurement tools. *Journal of Chiropractic Medicine*. 2010: 9(2): 49–59.
3. Robert Stow. Instrument Assisted Soft Tissue Mobilization. *Athletic Therapy Today*. 2011;16(3):5-8.
4. Rajalaxmi V. et al. Efficacy of Motor Control and Endurance Exercises in Neck Pain: A Pilot Study. *Indian Journal of Forensic Medicine and Pathology*. 2019;9(1).
5. Jull G et al. A randomized controlled trial of exercise and manipulative therapy for cervicogenic headache. *Spine*. 2002;27:1835–45.
6. Griffiths C et al. Effectiveness of specific neck stabilization exercises or a general neck exercise program for chronic neck disorders: a randomized controlled trial. *The Journal of Rheumatology*. 2009;36(2):390-7.
7. Harris et al. Reliability of a measurement of neck flexor endurance. *Physical Therapy*. 2005;85(12): 1349–55.
8. Mylonas K et al. Combining targeted instrument-assisted soft tissue mobilization applications and neuromuscular exercises can correct forward head posture and improve the functionality of patients with mechanical neck pain: a randomized control study. *BMC Musculoskeletal Disorders*. 2021;22: 212.
9. Gereck H et al. Acute effects of instrument assisted soft tissue mobilization technique on pain and joint position error in individuals with chronic neck pain: a double-blind, randomized controlled trial. *Somatosensory and Motor Research*. 2023;40(1):25-32.
10. Carmen Martin Gomez et al. Motor control using craniocervical flexion exercises versus other treatments for non-specific chronic neck pain: A systematic review and meta-analysis. *Musculoskeletal Science & Practice*. 2019;42:52-59.
11. Julien Mueller et al. Resistance, Motor Control, and Mindfulness-Based Exercises Are Effective for Treating Chronic Nonspecific Neck Pain: A Systematic Review With Meta-Analysis and Dose-Response Meta-Regression. *Journal of Orthopaedic and Sports Physical Therapy*. 2023;53:415-493.
12. Fatima Shewail et al. Instrument-assisted soft tissue mobilization versus myofascial release therapy in treatment of chronic neck pain: a randomized clinical trial. *BM Musculoskeletal Disorders*. 2023;24: 457.

Table 1: Griffith MCE protocol

Motor Control Exercises

Posture correction techniques taught in context of functional and work activities:

- a) Active range of motion exercises
- b) Isometric craniocervical flexion exercises performed sitting standing or lying.
- c) An isometric craniocervical flexion exercise in an inclined sitting position with a head lift off of the supporting surface.
- d) Isometric holds for up to 10 seconds repeated 10 times.
- e) Participants also taught to engage deep neck flexors during functional activities.
- f) Progression at the discretion of the treating therapist.

Patients were also asked to repeat the exercises 5 to 10 times per day.

Figure 2: Results of Paired Sample T Test

Scores	Mean	SD	Correlation	t-test	p-value
Pre Intervention VAS	7.40	0.55	0.67	11.18	<0.001
Post Intervention VAS	2.40	0.55			
Pre Intervention DNS	12.62	4.47	0.86	-7.29	<0.001
Post Intervention DNS	20.20	3.64			
Pre Intervention NDI	39.94	5.12	0.61	12.01	<0.001
Post Intervention NDI	14.70	0.76			





Lopa Das et al.,

Pre Intervention CF	47.20	5.12	-0.27	-3.86	0.02
Post Intervention CF	57.60	2.07			
Pre Intervention CE	59.60	9.40	0.75	-3.47	0.03
Post Intervention CE	69.20	6.65			
Pre Intervention RLF	44.00	2.65	0.12	-3.57	0.02
Post Intervention RLF	48.60	1.52			
Pre Intervention LLF	36.60	4.78	-0.25	-2.21	0.09
Post Intervention LLF	44.40	5.18			
Pre Intervention RLR	54.00	7.52	0.80	-2.09	0.10
Post Intervention RLR	59.40	9.71			
Pre Intervention LLR	54.20	2.59	-0.38	-2.41	0.07
Post Intervention LLR	63.00	6.82			

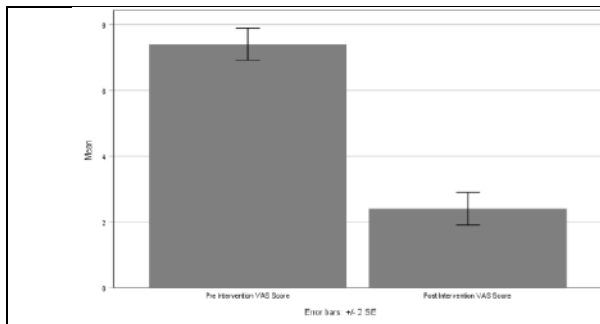


Figure 1: Mean score for VAS

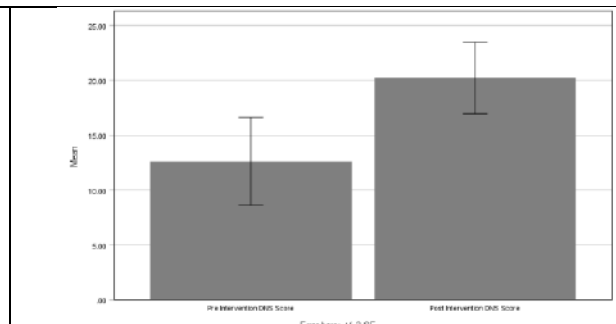


Figure 2: Mean score for DNS Score

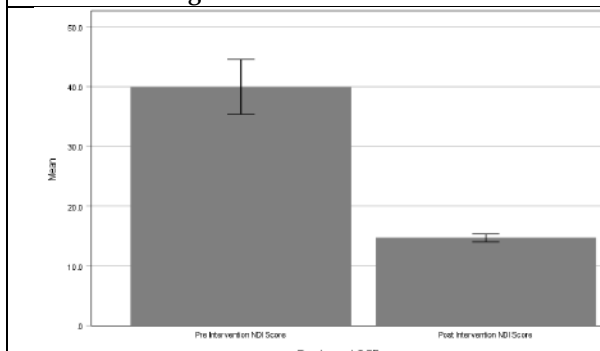


Figure 3: Mean score for NDI

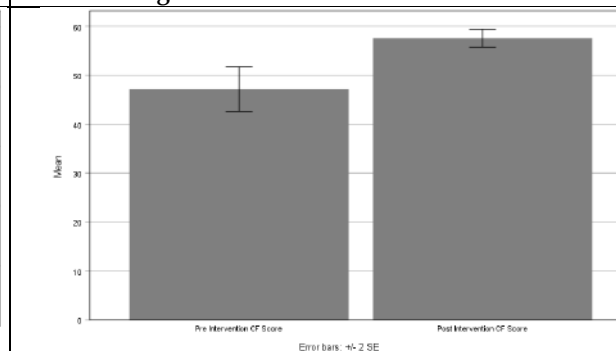


Figure 4: Mean score for Cervical Flexion

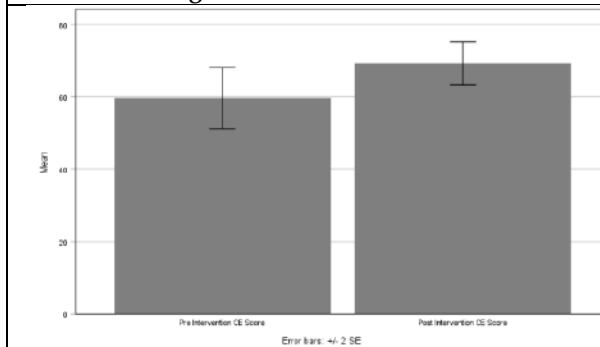


Figure 5: Mean score for Cervical Extension

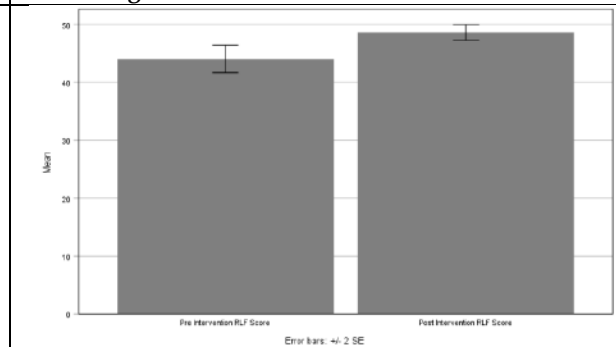


Figure 6: Mean score for Right Lateral Flexion





Lopa Das et al.,

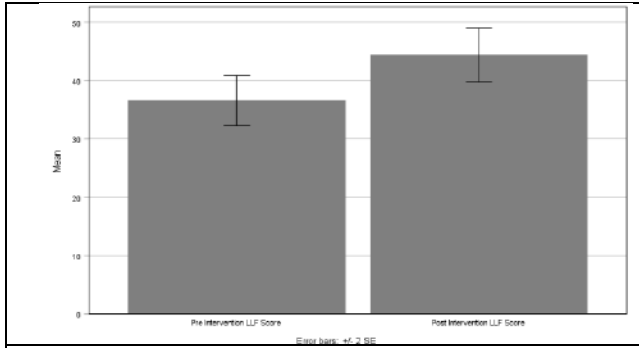


Figure 7: Mean score for Left Lateral Flexion

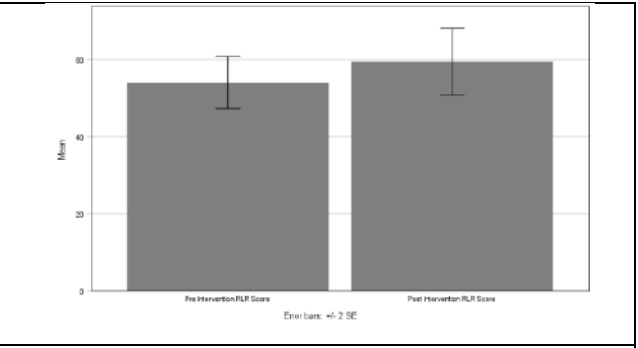


Figure 8: Mean score for Right Lateral Rotation

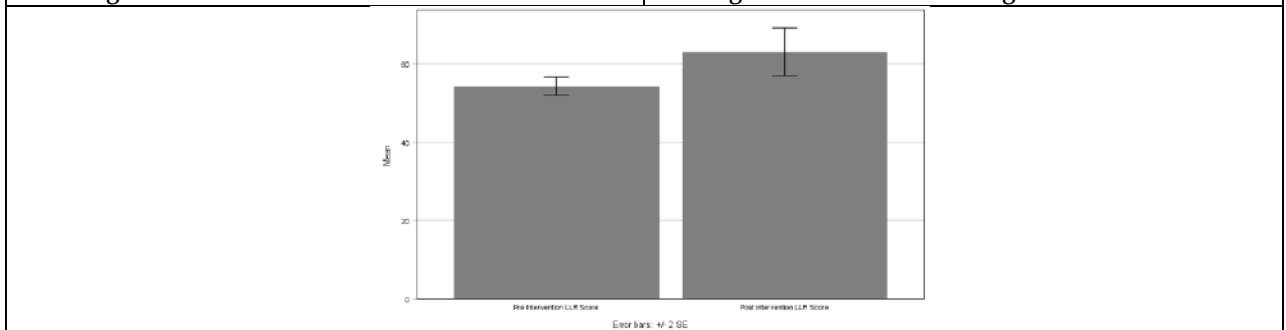


Figure 9: Mean score for Left Lateral Rotation





Beyond the Plate from 3D to 4D: Revolutionizing Food Fabrication with Advanced Printing Technology

Soma Basu¹ and A.J. Hemamalini^{2*}

¹Ph.D. Research Scholar, Department of Clinical Nutrition, Sri Ramachandra Faculty of Allied Health Sciences, Sri Ramachandra Institute of Higher Education and Research, Porur, Chennai, Tamil Nadu, India

²Professor and Head, Department of Clinical Nutrition, Sri Ramachandra Faculty of Allied Health Sciences, Sri Ramachandra Institute of Higher Education and Research, Porur, Chennai, Tamil Nadu, India.

Received: 20 Jun 2024

Revised: 21 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

A.J. Hemamalini

Professor and Head, Department of Clinical Nutrition,
Sri Ramachandra Faculty of Allied Health Sciences,
Sri Ramachandra Institute of Higher Education and Research,
Porur, Chennai, Tamil Nadu, India.
E.Mail: hemamalini.aj@sriramachandra.edu.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

3D and 4D food printing technology represents a transformative advancement at the intersection of culinary arts, engineering, and materials science. The food industry is witnessing constant evolution in trends and innovations, mirrored by advancements in technologies like 3D food printing which involves transforming a digital 3D model into tangible edible objects using food materials. An extensive literature survey was conducted using databases PubMed, Science Direct, Google Scholar. Articles were searched using terms related to “3D and 4D Food Fabrication Technology”. Total 53 articles from 2015 to 2023 were reviewed in order to write this review paper. This review article explores the principles, applications, and implications of these innovative technologies. To date, over 30 food items have been effectively fabricated through printing techniques. These include confectioneries like chocolates and fudge, baked goods such as biscuits and cakes, snacks like potato chips, fruit and vegetable items including diverse purees and juices, as well as jellies, meat products, and dairy products like cheeses and yogurt. Building upon the foundation of 3D printing, 4D food printing introduces an additional dimension of dynamic functionality. Through the integration of smart materials and responsive design principles, 4D printed food items exhibit the ability to transform or adapt their shape, texture, or taste in response to external stimuli such as temperature, moisture, or pH levels. This capability opens new avenues for culinary creativity, culinary experiences, and functional foods with enhanced sensory properties or therapeutic benefits. Beyond its culinary applications, 3D and 4D food printing hold promise for medical, nutritional, and environmental applications. Researchers are exploring the potential of 3D printed foods for personalized nutrition interventions, therapeutic diets, and dysphagia management. Food printing has the capability to adjust the nutritional composition of food to meet consumer preferences accomplished by incorporating healthy ingredients such as cellulose, plant chemicals, and high-quality proteins, while minimizing



**Soma Basu and Hemamalini**

adverse substances like anti-nutritional factors and allergens. The nutritional content of printed food can be managed strategically by utilizing fruits and vegetables as part of the food ink to provide essential micronutrients, cellulose, and other nutrients. However, several challenges include optimizing printing processes for scalability efficiency, safety, regulatory compliance, concerns related to taste, texture, and consumer acceptance and ethical considerations and the potential impact on traditional food systems warrant careful examination. 3D and 4D food printing technology represent a paradigm shift in food production and consumption, offering unprecedented opportunities for customization, sustainability, and innovation. As these technologies continue to mature, interdisciplinary research collaboration to unlock their full potential in the food and health sector is the way forward.

Keywords – 3D Food Fabrication, 4D Food Fabrication, Food Technology.

INTRODUCTION

The food industry is witnessing constant evolution in trends and innovations, mirrored by advancements in technologies like 3D food printing. This technology involves transforming a digital 3D model into tangible edible objects using food materials. The feasibility of printing food depends on factors like the type of printer, the characteristics of printable inks, post-processing methods, and more. 3D printing, also known as additive manufacturing (AM) and rapid prototyping (RP), is a burgeoning digital technology that sparks daily discussions, captivating researchers, industries, and the public alike due to its expanding range of applications in fields such as medicine, gastronomy, engineering, manufacturing, art, and education. Among these, gastronomy stands out as a significant area of interest and challenge. The current focal point of interest in 3D printing is "3D food printing." This process involves controlled robotic manipulation, where products are constructed layer by layer using computer-aided design (CAD) programs or downloaded models from online platforms.

Once a 3D model is created, the design data is transmitted to the printer, which then divides the model into layers and constructs them according to the specified cross-sectional pattern. In the realm of food, there's a belief that RP technology will redefine food processing by tailoring products to meet specific consumer preferences regarding taste, cost, convenience, and nutrition. Additionally, it's seen as a tool to democratize innovation by lowering barriers to entry for inventors, fostering a new class of independent designers and a custom product economy. Despite the complexity of food systems and their varied properties, researchers have been exploring the application of 3D printing to different types of food products. While the engineering principles behind 3D food printers are understood, the creative potential of this technology remains largely untapped. Through optimization of various printing parameters and ingredients, researchers have achieved successful outcomes in printing with a range of food substrates, including chocolate, cookie dough, cereals, sugar powder, processed cheese, meat gels, and even fruits and vegetables.

However, despite numerous studies and reviews on 3D food printing, gaps still exist in understanding the relationship between printing process variables and material structure to achieve desired outcomes. 4D printing technology, an evolution of 3D printing, represents a novel concept where the printed configuration undergoes changes over time (Choi et al., 2015).^[1] This concept was initially pioneered by a research team at the Massachusetts Institute of Technology (MIT) in 2013 (Tibbits, 2014).^[2] In terms of the printing process, 4D printing closely resembles 3D printing, involving stages such as 3D design development and fabricating the structure using a 3D printer (Choi et al., 2015).^[1] However, the key distinctions between 4D and 3D printing lie in smart design and smart materials, as 4D printed structures have the capability to alter their shape or function (Pei & Loh, 2018).^[3] This review aims to deliver deeper into optimizing extrusion-based food printing, focusing on 3D food printing process, its advantages and also addressing its limitations and offering insights to overcome barriers in this field and extension to 4D Food Printing Technology and its future aspects.





Soma Basu and Hemamalini

METHODOLOGY

An extensive literature survey was conducted using databases PubMed, ScienceDirect, Google Scholar. Articles were searched using terms related to “3D and 4D Food Fabrication Technology”. Total 53 articles from 2015 to 2023 were reviewed in order to write this review paper. This study was approved by Publication Guidelines and Monitoring Committee (PGMC) Approval from Sri Ramachandra Institute of Higher Education and Research, Chennai, Tamil Nadu, India for Publication.

DISCUSSION

FEEDING THE FUTURE: EXPLORING 3D FOOD FABRICATION TECHNOLOGY

The fantastical 3D printing depicted in sci-fi classics like Star Trek, Jurassic Park III, and Westworld has transitioned into real-world applications. Now, we witness the fabrication of 3D printed cars, skulls, hearts, prosthetics, and more. A milestone in this progression occurred in 2006 when Cornell University developed the first multi-material 3D printer capable of printing edible items like chocolates, cookie doughs, and cheeses (Liu et al., 2017).^[4] Since then, the utilization of 3D food printing has surged, not only in research settings but also in commercial domains, driven by cost reductions and technological advancements.

3D printing, also referred to as additive manufacturing, involves creating a three-dimensional object using either a computer-aided-design (CAD) model or a digital 3D model. This process entails adding raw materials, depositing them, and solidifying them, typically in layers under the control of a computer. In the realm of 3D food printing, the raw materials are food ingredients, and a typical printer comprises three main components: a printing system, an operational control platform, and a food capsule. The food capsule contains the food material, often a thick slurry similar to printer ink, which is pushed through a food-grade nozzle by a syringe pump to be deposited layer-by-layer onto a platform according to a pre-designed shape controlled by a computer. Whether the printed food can be consumed directly or requires further cooking depends on the nature of the food ingredients and the sanitary condition of the printing device.

PRINTED PALATES: UNVEILING THE 3D FOOD PRINTING PROCESS

3D food printing, also known as food fabrication or additive manufacturing in the culinary world, involves the use of specialized 3D printers to create edible items layer by layer. Here's a general overview of the process:

Flow Chart Representing 3D Food Printing Process

- **Design:** The process begins with the creation of a digital model or design of the food item to be printed. This can be done using computer-aided design (CAD) software or by using specialized food design software.
- **Preparation of Ingredients:** Edible ingredients are prepared in a form suitable for extrusion through the printer's nozzle. These ingredients can include various food materials such as dough, batter, purees, gels, or pastes. Depending on the printer and the desired food item, these ingredients may need to meet certain consistency and texture requirements.
- **Loading Ingredients:** The prepared ingredients are loaded into the printer's cartridges or syringes. Some 3D food printers may have multiple cartridges to enable printing with different ingredients or colors.
- **Printing:** The printer follows the digital model or design and deposits the edible material layer by layer onto a build platform. This is typically done through a process called extrusion, where the material is pushed through a nozzle or syringe. The printer's movements are controlled precisely to ensure accurate deposition of each layer.
- **Layer-by-Layer Building:** The printer continues to build up the food item by adding successive layers of edible material. The layers may be very thin, depending on the printer's resolution and the complexity of the design.
- **Post-Processing (Optional):** After printing is complete, some food items may require additional processing steps such as baking, cooling, or assembly before they are ready to be served.





Soma Basu and Hemamalini

- **Serve and Enjoy:** Once any necessary post-processing steps are complete, the 3D printed food item is ready to be served and enjoyed.

It's important to note that 3D food printing technology is still relatively new and continues to evolve. While it has the potential to revolutionize aspects of food production, it is currently mainly used in niche applications such as custom confectionery, personalized nutrition, and food customization for people with specific dietary needs or restrictions. Additionally, the taste, texture, and overall quality of 3D printed food items can vary depending on the ingredients used, the printer's capabilities, and the skill of the chef or operator.

EDIBLE BLUEPRINTS: ASSESSING THE SUITABILITY OF FOODS FOR 3D PRINTING

To date, over 30 food items have been effectively fabricated through printing techniques. These include confectioneries like chocolates and fudge, baked goods such as biscuits and cakes, snacks like potato chips, fruit and vegetable items including diverse purees and juices, as well as jellies, meat products, and dairy products like cheeses and yogurt (Dankar et al., 2018). [5]

From a technological standpoint, the flow and deformation characteristics, referred to as rheological properties, of the "food ink" are paramount in 3D food printing. For successful extrusion from the nozzle, the ink must be sufficiently fluid, while also providing structural support upon deposition to uphold the shape of the printed item (Chen et al., 2022).[6] In rheology, inks exhibiting such traits are termed Bingham plastic or Herschel-Bulkley materials. These materials form a delicate network at rest, supporting their weight to maintain shape. When subjected to a significant stress, known as yield stress in rheology, the network breaks down, allowing the ink to flow for printing (Chen et al., 2022).[6] When analyzed using a rheometer, most printable materials demonstrate shear thinning behavior, meaning they have lower viscosity at higher deformation rates and higher viscosity at lower deformation rates (Chen et al., 2022).[6] In addition to formulation, temperature serves as a practical means to regulate ink rheology (Chen et al., 2022).[6] For instance, heating chocolates above their melting point facilitates flow and subsequent printing. Subsequent solidification upon storage at ambient or lower temperatures enables network formation (high viscosity or yield stress) and maintains the desired shape. Moreover, texture significantly influences the sensory and structural attributes of printed products, with reported hardness values ranging from 4.48 to 59.8 Newtons and springiness values typically below 1 for successful prints (Zhang et al., 2021).[6] Physical and chemical transformations of constituent ingredients during storage and thermal processes are also expected to impact the sensory and quality attributes of printed items.

EXPLORING THE PROS AND CONS OF CONTEMPORARY 3D FOOD PRINTING TECHNOLOGY (Pereira et al., 2018)[7]

Benefits of contemporary 3D food printing technology include: Tailored nutrition and health benefits for individuals, Customized creation of intricate visual designs and textures, Personalized ingredient selection to prevent allergies and cross-contamination, Reduction of food wastage by conserving materials, Opportunity to utilize alternative material sources, Streamlining and expediting the manufacturing process, Conservation of energy and reduction of transportation requirements. Meanwhile, drawbacks of present-day 3D food printing technology encompass: Expenses related to the printer and "food ink", Insufficiency of suitable "food inks" for 3D printing, Sluggish printing speed and challenges for large-scale production, Safety apprehensions regarding printers and "food inks", Considerations regarding consumer perception, Challenges concerning printing precision and surface finishes.[8]

BEYOND BOUNDARIES: EVOLVING TO 4D FOOD FABRICATION TECHNOLOGY

Four-dimensional (4D) printing represents a recent advancement in additive manufacturing techniques, extending from 3D printing by introducing the concept of altering printed configurations over time (Choi et al., 2015).[1] Initially conceptualized by a team at the Massachusetts Institute of Technology (MIT) in 2013 (Tibbits, 2014)[2], 4D printing shares similarities with 3D printing in terms of design development and structure printing processes using a 3D printer (Choi et al., 2015).[1] However, the distinguishing features of 4D printing lie in the integration of smart design principles and smart materials, enabling printed structures to change shape or function (Pei & Loh, 2018).[3]



**Soma Basu and Hemamalini**

Smart materials employed in 4D printing exhibit properties such as self-assembly, versatility, and self-healing capabilities (He, Zhang, & Guo, 2020).[9] Most commonly used 4D printing materials are single or multilateral polymers, including shape memory polymers, liquid crystal elastomers, and composite hydrogels (Sharma & Tabbits, 2020).[10] The responsive shape changes of 4D printing structures offer advantages in terms of space-saving during transportation and storage (Pei & Loh, 2018),[3] leading to exploration across various fields such as robotics, biomedical applications, tissue engineering, and electronic devices (Ghazal et al., 2021).[11] However, the application of 4D printing in the food sector remains relatively limited, with only a few studies available, indicating an early stage of development (Chu et al., 2020).[12].

The adaptability of materials in 4D printing allows for customized responsiveness configurations, contributing to material reduction through the self-changing nature of 4D printed objects (Chu et al., 2020).[12] Additionally, 4D printing has the potential to address consumer demands for unique food products (Phuhongsung, Zhang, & Bhandari, 2020).[13] Furthermore, the directed and adjustable deformation of structures in 4D printing could assist chefs in determining optimal cooking points, enhancing the visual appeal and taste of food items (Chen et al., 2021).[14] It's worth noting that while 4D printing allows for achieving desirable product properties within an appropriate timeframe, these properties may diminish during the storage of printed products.

TIME TO TASTE: UNVEILING THE POTENTIAL OF 4D PRINTING IN FOOD

In the realm of food technology, researchers at MIT pioneered the use of 4D printing. They devised a method involving a 2D film made from a blend of starch, cellulose, and protein, activated by water. This 2D film transformed into a 3D structure upon exposure to water (Wang et al., 2017).[15] The application of 4D printing in food holds promise for tailoring products and cultivating distinct flavor profiles. By manipulating the composition of printing ink with different food materials and formulas, the characteristics of the resulting 4D printed food can be customized (Teng et al., 2021).[16] Variations in temperature and pH can induce alterations in the color, texture, aroma, and shape of food. For instance, when microwave heating was employed, a 4D transformation in 3D printed buckwheat dough and lotus root powder gel was observed (Guo et al., 2021; Chen, Zhang, Mujumdar, et al., 2021).[17,18] Similarly, shifts in pH levels were found to affect the color of a mixture composed of 3D printed soy protein isolate, pumpkin, and beetroot (Phuhongsung, Zhang, & Devahastin, 2020a).[19].

INNOVATIVE INKS: EXPLORING THE ROLE OF INKS IN 4D FOOD PRINTING

In recent times, printing technology has been instrumental in the development of a wide variety of food products. These include chocolate (Rando & Ramaioli, 2021)[20], soybean (Phuhongsung, Zhang, & Devahastin, 2020b; Balla et al., 2020)[21,22], meat (Wilson et al., 2021; Wilson et al., 2021), starch (Zheng et al., 2021; Zeng et al., 2021), fruits, vegetables (Chen et al., 2021; Chen et al., 2021), and food hydrocolloids (Pant et al., 2021), among others.[23-29] The moisture content of these food materials, crucial for their printing performance, is significant. Therefore, utilizing dried and powdered forms of food materials can preserve their nutritional value and functional properties (Lee et al., 2019).[30] Furthermore, the food ink mixture utilized in 4D printing comprises specific materials designed to respond to stimuli. These materials undergo changes in color, flavor, texture, and more in response to certain conditions or stimuli. Therefore, the sensitivity of these materials in food printing ink is crucial for achieving 4D transformations in printed objects. The pigments within the printing ink are responsible for altering the color of food in response to varying pH levels. For instance, curcumin, a stimulus-responsive material employed in 4D printing, displays a red hue in alkaline pH environments and a yellow hue in acidic or neutral pH environments (C. Chen, Zhang, Guo, et al., 2021).[31].

Likewise, anthocyanin functions as a responsive substance, displaying diverse hues in response to pH alterations (He et al., 2021). Similarly, the gelatin-gum Arabic-flavor oil complex has demonstrated thermal responsiveness, serving as a stimulus-reactive material in the printing of buckwheat dough with yellow flesh peach. In their study, Guo et al. (2021) noted concurrent alterations in both color and flavor of the product when subjected to microwave stimuli. It is imperative that the changes induced by stimuli in 4D printed objects remain manageable, as unregulated changes could lead to undesirable product outcomes. To ensure controlled alterations in the properties of 4D printed objects



**Soma Basu and Hemamalini**

under varying stimuli, strategic design coding and meticulous distribution, such as uniform, gradient, or patterned dispersion of components, are essential.[32,33]

FROM CONCEPT TO CUISINE: NAVIGATING MODEL DESIGN AND PRINTING PROCESSES IN 4D FOOD PRINTING

4D printing involves the transformation of a 3D printed model through external stimuli. Therefore, the design of the model holds paramount importance in the 4D printing procedure. Effective software plays a crucial role in facilitating 4D printing by allowing the production of the intended product. The 4D model design and printing process entail the utilization of various software solutions, including separate solutions for Simulation, Modeling, Slicer, Host/Firmware, Monitoring, and one Printing Management software, as depicted in.

Simulation involves replicating the behavior of an object or system using computer software. It is commonly utilized in the preliminary stages of product development to mitigate potential failures before physical production. By employing mathematical formulas to mimic real-world phenomena, simulation aids in comprehending the entire operation without practical execution. In the realm of 4D printing, simulation techniques can be categorized into two: finite element analysis (FEA) software and software dedicated to simulation function development. Mao et al. (2015) exemplified this by employing FEA simulations via ABAQUS software, incorporating a user-defined constitutive model, and utilizing a reduced-order model (ROM) to simulate rigid body motions and explore folding pathways. After the simulation process, the 3D design of the printing object is crafted using modeling software. Modeling entails creating a representation of the object with its pertinent characteristics, often resembling the actual item. This process results in a 3D rendition of the object. Commonly employed computer-based graphics and Computer-aided design (CAD) software for this purpose in food printing include Rhino 6 software, Rhinoceros 5.0 (Liu et al., 2021), and SketchUp Pro 2015 (Ghazal et al., 2021), among others. Notably, the modeling software does not directly transmit information regarding the 3D design to the printer. Instead, slicer software is utilized to convert the model into 2D sections containing the necessary information for the printer.

Slicer software is a computer application widely utilized in the realm of 3D printing to transform a 3D object model into precise instructions tailored for the printer. It incorporates various editing tools such as rotation, scaling, as well as import and export functionalities for CAD files. Furthermore, it features a subdivision function compatible with Stereo Lithography (STL) files. This allows for the conversion of a low-resolution image to a higher resolution one by enhancing intricacy and smoothness (Chung et al., 2017). The slicer software generates the blueprint for printing, known as G code, which encapsulates all the essential information defining the entire printing process. Common slicing software utilized in food printing includes Slic3r, Simplify3D, and others. After the slicer software prepares the G-code (slicer data), it proceeds to transmit it to the printer. Subsequently, the host/Firmware software, when connected to a Personal Computer (PC), generates the 3D object. Essentially, the host software assumes control over the printer's components, including the printer's head and bed, analyzes the received G-code data, and dispatches G-code instructions to the printer.

In the context of 4D printing, the synthesized 3D object undergoes gradual changes in its properties over time, typically triggered by specific environmental conditions. Monitoring software facilitates the observation of these stimulus-induced self-transformations in 4D printed objects and initiates necessary actions if required. This monitoring software can be installed on user devices and monitored continuously. The choice of material significantly influences the design and functionality of 4D printed objects. The material must exhibit responsiveness to specific stimuli throughout the layer-by-layer printing process. The selection of material dictates the stimuli necessary to trigger the self-transformative changes in the printed objects. Upon exposure to stimuli, only specific components of the printed object interact, leading to predetermined transformations. Interaction mechanisms between the 4D printed object and the stimulating agent may involve mechanical loading or physical manipulation, resulting in a sequence of changes. Mathematical modeling determines the required exposure to stimuli to achieve desired changes in the printed object's properties (Ahmed et al., 2021). Achieving a shape-shifting effect with a single material involves manipulating the gradient distribution of the material by strategically controlling the spatial



**Soma Basu and Hemamalini**

position and morphology of the lattice structure throughout the product (Pei & Loh, 2018). The self-induced changes in the material's shape depend on its responsiveness to stimuli and the physical design geometry of the material (Lendlein & Kelch, 2002). The 4D printing process comprises distinct steps, each involving different software. The output of one step may serve as input for subsequent steps, thereby influencing the next stage. Consequently, the software utilized in each step plays a specific role and may impact subsequent stages. All stages are interconnected, forming a network, and thus, all software in each processing stage is monitored and controlled by the user. This is facilitated through a printing management system connected to each software at every stage, designed to optimize printer usage (Chung et al., 2017).[34-42]

Numerous intriguing instances demonstrate that through the manipulation of food chemistry and physics, 4D-printed foods can offer innovative concepts for crafting interactive culinary experiences. One such example involves the curvature of purple sweet potato purees during microwave dehydration, with the ability to adjust the rate by altering microwave power, salt content, or fructose syrup content, thereby yielding personalized shapes (He et al., 2020).[8]

INKJET PRINTING

Inkjet printing, a common technique in 4D printing, is frequently employed in confectionery and decorative applications (Pallottino et al., 2016). This method involves a series of pneumatic membrane nozzles, typically ranging from 20 to 50 μm , which dispense small droplets of printing material onto a moving substrate. Both single and multiple nozzles can operate simultaneously, allowing printing ink to be sprayed onto the printing platform, creating a layered structure. These droplets amalgamate to form a digital image with surface fillings and cavity depositions. Inkjet printing typically utilizes low-viscosity materials, making it more suitable for producing flat products rather than intricate structures. Temperature plays a significant role in inkjet printing as it affects the viscosity and surface energy of the material (Le-bail et al., 2020).

Robert John Young introduced a machine incorporating a bubble-jet printer head and a reservoir for liquid food colorant, which is utilized to print images onto the surface of an edible substrate, as previously outlined. This innovation also included a method for printing images onto edible substrates (Robert John Young, 2000). Similarly, Pallottino et al. developed a device to improve the printing of low-viscosity substances on an edible surface by modifying the surface with a high-polarity water-based glaze or by polishing the gum surface (Pallottino et al., 2016). Inkjet printing is primarily suited for low-viscosity substances, limiting its applicability in producing complex food structures. Its uses extend to graphical decoration, fillings, micro-encapsulation, and to some extent, 3D nano printing (Fernanda C. Godoi et al., 2018).[43-47]

NUTRITION BY DESIGN: UNDERSTANDING NUTRITIONAL CHANGES IN THE PRINTING PROCESS

Food printing has the capability to adjust the nutritional composition of food to meet consumer preferences. This can be accomplished by incorporating healthy ingredients such as cellulose, plant chemicals, and high-quality proteins, while minimizing adverse substances like anti-nutritional factors and allergens. The nutritional content of printed food can be managed by strategically distributing nutrients in the printing ink or alternatives during the printing process. Utilizing fruits and vegetables as part of the food ink can provide essential micronutrients, cellulose, and other nutrients (Chen et al., 2022). Incorporating tissue engineering into food printing holds promise for enhancing the nutritional aspects of 4D printing. Substituting ink and tissue with food-grade materials in 4D biotechnological printing can elevate the nutritional quality of printed food, as exposure to appropriate conditions stimulates tissue growth. The printed object may contain plant or animal cells capable of forming tissue-like structures under suitable stimuli and producing nutrients (Teng et al., 2021). Additionally, printing food with probiotics can enhance the nutritional quality of the printed object. The inclusion of microalgae offers supplementary nutritional and functional components such as protein, fatty acids, sterols, and vitamins (Uribe-Wandurraga et al., 2020).

UV irradiation of 4D printed purple sweet potato pastes infused with ergosterol resulted in a notable increase in vitamin D2 content, particularly in the irradiated area (Chen et al., 2021). Similarly, microwave stimulation of





Soma Basu and Hemamalini

beeswax-based oleo gel incorporated with purple potato powder ink produced a low-fat printed product (Shi et al., 2022).[48-53]

CONCLUSION

4D food printing is currently in its early stages, presenting ample opportunities for extensive research. Given its infancy, traditional 3D printing equipment is commonly utilized, but there is a need for the development of new printing conditions or equipment with enhanced functionality to better simulate 4D printing. This advancement would facilitate the achievement of desired changes, such as deformation and denaturation of various components of food ink. Currently, researchers primarily focus on microwave heating and changes in pH as stimuli sources. However, there is potential for exploration of new stimulus agents like light and changes in ionic concentration in 4D food printing. Moreover, the range of stimulus-response materials studied in 4D food printing is limited. Investigating new components such as diacetyl and vanillin could broaden the scope. Additionally, employing multiple stimulus-responsive materials to induce simultaneous changes in multiple food properties would represent a significant breakthrough in 4D food printing. The development of a monitoring system to assess the degree of 4D changes could play a crucial role in achieving controlled transformations in 4D food printing.

REFERENCES

1. Choi, J., Kwon, O. C., Jo, W., Lee, H. J., & Moon, M. W. (2015). 4D printing technology: a review. *3D Printing and Additive Manufacturing*, 2(4), 159-167.
2. Campbell, T. A., Tibbits, S., & Garrett, B. (2014). *The next wave: 4D printing*. Atlantic, 1-16.
3. Pei, E., & Loh, G. H. (2018). Technological considerations for 4D printing: an overview. *Progress in Additive Manufacturing*, 3, 95-107.
4. Liu, Z., Zhang, M., Bhandari, B., & Wang, Y. (2017). 3D printing: Printing precision and application in food sector. *Trends in Food Science & Technology*, 69, 83-94.
5. Dankar, I., Haddarah, A., Omar, F. E., Sepulcre, F., & Pujolà, M. (2018). 3D printing technology: The new era for food customization and elaboration. *Trends in food science & technology*, 75, 231-242.
6. Yan, B., Zhao, Z., Zhang, N., Ruan, H., Yu, X., Zhao, J., ... & Fan, D. (2022). 3D food printing curing technology based on gellan gum. *Journal of Food Engineering*, 327, 111036.
7. Pereira, T., Barroso, S., & Gil, M. M. (2021). Food texture design by 3D printing: A review. *Foods*, 10(2), 320.
8. Ren, S. Thought for Food Blog.
9. He, C., Zhang, M., & Guo, C. (2020). 4D printing of mashed potato/purple sweet potato puree with spontaneous color change. *Innovative Food Science & Emerging Technologies*, 59, 102250.
10. Singholi, A. K. S., & Sharma, A. (2020). Recent advancement and research possibilities in 4D printing technology. *Materialwissenschaft und Werkstofftechnik*, 51(10), 1332-1340.
11. Ghazal, A. F., Zhang, M., Bhandari, B., & Chen, H. (2021). Investigation on spontaneous 4D changes in color and flavor of healthy 3D printed food materials over time in response to external or internal pH stimulus. *Food Research International*, 142, 110215.
12. Chu, H., Yang, W., Sun, L., Cai, S., Yang, R., Liang, W., ... & Liu, L. (2020). 4D printing: a review on recent progresses. *Micromachines*, 11(9), 796.
13. Phuhongsung, P., Zhang, M., & Bhandari, B. (2020). 4D printing of products based on soy protein isolate via microwave heating for flavor development. *Food Research International*, 137, 109605.
14. Chen, F., Zhang, M., Liu, Z., & Bhandari, B. (2021). 4D deformation based on double-layer structure of the pumpkin/paper. *Food Structure*, 27, 100168.
15. Wang, W., Yao, L., Zhang, T., Cheng, C. Y., Levine, D., & Ishii, H. (2017, May). Transformative appetite: shape-changing food transforms from 2D to 3D by water interaction through cooking. In *Proceedings of the 2017 CHI conference on human factors in computing systems* (pp. 6123-6132).



**Soma Basu and Hemamalini**

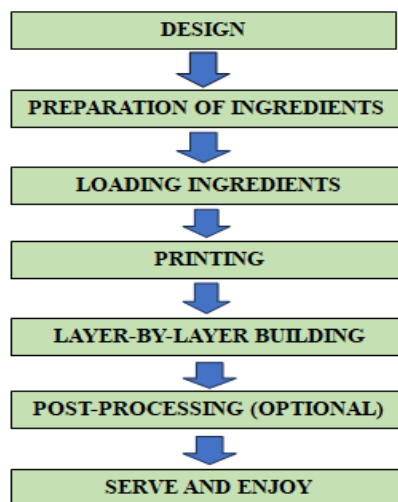
16. Teng, X., Zhang, M., & Mujumdar, A. S. (2021). 4D printing: Recent advances and proposals in the food sector. *Trends in Food Science & Technology*, 110, 349-363.
17. Guo, C., Zhang, M., & Devahastin, S. (2021). Color/aroma changes of 3D-Printed buckwheat dough with yellow flesh peach as triggered by microwave heating of gelatin-gum Arabic complex coacervates. *Food Hydrocolloids*, 112, 106358.
18. Chen, C., Zhang, M., Mujumdar, A. S., & Phuhongsung, P. (2021). Investigation of 4D printing of lotus root-compound pigment gel: Effect of pH on rapid colour change. *Food Research International*, 148, 110630.
19. Phuhongsung, P., Zhang, M., & Devahastin, S. (2020). Influence of surface pH on color, texture and flavor of 3D printed composite mixture of soy protein isolate, pumpkin, and beetroot. *Food and Bioprocess Technology*, 13, 1600-1610.
20. Rando, P., & Ramaioli, M. (2021). Food 3D printing: Effect of heat transfer on print stability of chocolate. *Journal of Food Engineering*, 294, 110415.
21. Phuhongsung, P., Zhang, M., & Devahastin, S. (2020). Investigation on 3D printing ability of soybean protein isolate gels and correlations with their rheological and textural properties via LF-NMR spectroscopic characteristics. *Lwt*, 122, 109019.
22. Balla, V. K., Tadimetri, J. G. D., Kate, K. H., & Satyavolu, J. (2020). 3D printing of modified soybean hull fiber/polymer composites. *Materials Chemistry and Physics*, 254, 123452.
23. Wilson, A., Anukiruthika, T., Moses, J. A., & Anandharamakrishnan, C. (2023). Preparation of fiber-enriched chicken meat constructs using 3D printing. *Journal of Culinary Science & Technology*, 21(1), 127-138.
24. Wilson, A., Anukiruthika, T., Moses, J. A., & Anandharamakrishnan, C. (2023). Preparation of fiber-enriched chicken meat constructs using 3D printing. *Journal of Culinary Science & Technology*, 21(1), 127-138.
25. Zheng, L., Liu, J., Liu, R., Xing, Y., & Jiang, H. (2021). 3D printing performance of gels from wheat starch, flour and whole meal. *Food Chemistry*, 356, 129546.
26. Zeng, X., Li, T., Zhu, J., Chen, L., & Zheng, B. (2021). Printability improvement of rice starch gel via catechin and procyanidin in hot extrusion 3D printing. *Food Hydrocolloids*, 121, 106997.
27. Chen, Y., Zhang, M., & Phuhongsung, P. (2021). 3D printing of protein-based composite fruit and vegetable gel system. *Lwt*, 141, 110978.
28. Chen, C., Zhang, M., Guo, C., & Chen, H. (2021). 4D printing of lotus root powder gel: Color change induced by microwave. *Innovative Food Science & Emerging Technologies*, 68, 102605.
29. Pant, A., Lee, A. Y., Karyappa, R., Lee, C. P., An, J., Hashimoto, M., ... & Zhang, Y. (2021). 3D food printing of fresh vegetables using food hydrocolloids for dysphagic patients. *Food Hydrocolloids*, 114, 106546.
30. Lee, J. H., Won, D. J., Kim, H. W., & Park, H. J. (2019). Effect of particle size on 3D printing performance of the food-ink system with cellular food materials. *Journal of Food Engineering*, 256, 1-8.
31. Chen, C., Zhang, M., Guo, C., & Chen, H. (2021). 4D printing of lotus root powder gel: Color change induced by microwave. *Innovative Food Science & Emerging Technologies*, 68, 102605.
32. He, C., Zhang, M., & Devahastin, S. (2021). Microwave-induced deformation behaviors of 4D printed starch-based food products as affected by edible salt and butter content. *Innovative Food Science & Emerging Technologies*, 70, 102699.
33. Guo, C., Zhang, M., & Devahastin, S. (2021). Color/aroma changes of 3D-Printed buckwheat dough with yellow flesh peach as triggered by microwave heating of gelatin-gum Arabic complex coacervates. *Food Hydrocolloids*, 112, 106358.
34. Chung, S., Song, S. E., & Cho, Y. T. (2017). Effective software solutions for 4D printing: A review and proposal. *International Journal of Precision Engineering and Manufacturing-Green Technology*, 4, 359-371.
35. Liu, Z., He, C., Guo, C., Chen, F., Bhandari, B., & Zhang, M. (2021). Dehydration-triggered shape transformation of 4D printed edible gel structure affected by material property and heating mechanism. *Food Hydrocolloids*, 115, 106608.
36. Ghazal, A. F., Zhang, M., Bhandari, B., & Chen, H. (2021). Investigation on spontaneous 4D changes in color and flavor of healthy 3D printed food materials over time in response to external or internal pH stimulus. *Food Research International*, 142, 110215.





Soma Basu and Hemamalini

37. Chung, S., Song, S. E., & Cho, Y. T. (2017). Effective software solutions for 4D printing: A review and proposal. *International Journal of Precision Engineering and Manufacturing-Green Technology*, 4, 359-371.
38. Chung, S., Song, S. E., & Cho, Y. T. (2017). Effective software solutions for 4D printing: A review and proposal. *International Journal of Precision Engineering and Manufacturing-Green Technology*, 4, 359-371.
39. Ahmed, A., Arya, S., Gupta, V., Furukawa, H., & Khosla, A. (2021). 4D printing: Fundamentals, materials, applications and challenges. *Polymer*, 228, 123926.
40. Pei, E., & Loh, G. H. (2018). Technological considerations for 4D printing: an overview. *Progress in Additive Manufacturing*, 3, 95-107.
41. Lendlein, A., & Kelch, S. (2002). Shape-memory polymers. *Angewandte Chemie International Edition*, 41(12), 2034-2057.
42. Chung, S., Song, S. E., & Cho, Y. T. (2017). Effective software solutions for 4D printing: A review and proposal. *International Journal of Precision Engineering and Manufacturing-Green Technology*, 4, 359-371.
43. Pallottino, F., Hakola, L., Costa, C., Antonucci, F., Figorilli, S., Seisto, A., & Menesatti, P. (2016). Printing on food or food printing: a review. *Food and Bioprocess Technology*, 9, 725-733.
44. Le-Bail, A., Maniglia, B. C., & Le-Bail, P. (2020). Recent advances and future perspective in additive manufacturing of foods based on 3D printing. *Current Opinion in Food Science*, 35, 54-64.
45. Young, R. J. (2000). U.S. Patent No. 6,058,843. Washington, DC: U.S. Patent and Trademark Office.
46. Pallottino, F., Hakola, L., Costa, C., Antonucci, F., Figorilli, S., Seisto, A., & Menesatti, P. (2016). Printing on food or food printing: a review. *Food and Bioprocess Technology*, 9, 725-733.
47. Godoi, F. C., Bhandari, B., Prakash, S., & Zhang, M. (Eds.). (2018). *Fundamentals of 3D food printing and applications*. Academic press.
48. Chen, Y., Zhang, M., Sun, Y., & Phuhongsung, P. (2022). Improving 3D/4D printing characteristics of natural food gels by novel additives: A review. *Food Hydrocolloids*, 123, 107160.
49. Teng, X., Zhang, M., & Mujumdar, A. S. (2021). 4D printing: Recent advances and proposals in the food sector. *Trends in Food Science & Technology*, 110, 349-363.
50. Uribe-Wandurraga, Z. N., Zhang, L., Noort, M. W., Schutyser, M. A., García-Segovia, P., & Martínez-Monzó, J. (2020). Printability and physicochemical properties of microalgae-enriched 3D-printed snacks. *Food and Bioprocess Technology*, 13, 2029-2042.
51. Chen, J., Zhang, M., & Devahastin, S. (2021). UV-C irradiation-triggered nutritional change of 4D printed ergosterol-incorporated purple sweet potato pastes: Conversion of ergosterol into vitamin D2. *LWT*, 150, 111944.
52. Shi, Y., Zhang, M., & Phuhongsung, P. (2022). Microwave-induced spontaneous deformation of purple potato puree and oleogel in 4D printing. *Journal of Food Engineering*, 313, 110757.
53. Navaf, M., Sunooj, K. V., Aaliya, B., Akhila, P. P., Sudheesh, C., Mir, S. A., & George, J. (2022). 4D printing: A new approach for food printing; effect of various stimuli on 4D printed food properties. A comprehensive review. *Applied Food Research*, 2(2), 100150.



Flow Chart Representing 3D Food Printing Process





Challenges and Opportunities in Implementing Sustainable WASH Practices in Government Schools: Lessons from Tamil Nadu

S.Anushya^{1*} and J. Subramaniyan²

¹Ph.D Research Scholar, Department of Political Science & Public Administrations, Annamalai University, Annamalai Nagar, Chidambaram, Tamil Nadu, India.

²Associate Professor & Coordinator, Department of Political Science & Public Administrations, Annamalai University, Annamalai Nagar, Chidambaram, Tamil Nadu, India.

Received: 07 Jun 2024

Revised: 10 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

S.Anushya

Ph.D Research Scholar,
Department of Political Science & Public Administrations,
Annamalai University, Annamalai Nagar,
Chidambaram, Tamil Nadu, India.
E.Mail: anushyaselvam0@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The paper explores the challenges and opportunities associated with implementing sustainable Water, Sanitation, and Hygiene (WASH) practices in government schools in Tamil Nadu, India. Despite significant advancements in policy frameworks and financial investments aimed at improving WASH infrastructure, many schools still struggle with inadequate facilities, poor maintenance, and limited community engagement. The study identifies critical barriers such as financial constraints, socio-cultural factors, and gaps in policy execution. The research highlights innovative approaches and best practices that have successfully addressed these challenges in certain schools. Key opportunities include leveraging community participation, integrating WASH education into the curriculum. The findings underscore the importance of a holistic approach that combines government support, community involvement, and sustainable practices to enhance the overall effectiveness of WASH initiatives in schools in Tamil Nadu.

Keywords: WASH, Challenges, Menstrual Hygiene, Sustainable development, education, Community Participation.



**Anushya and Subramaniyan****INTRODUCTION**

WASH is an abbreviation for "Water, Sanitation and Hygiene". It encompasses the provision of clean water, sufficient sanitation facilities, and the promotion of proper hygiene practices to enhance public health and well-being. Water, sanitation, and hygiene (WASH) involves guaranteeing the availability of clean and affordable water for drinking, cooking, and personal and domestic hygiene. It also includes providing access to facilities and services for the safe disposal of human waste, as well as promoting behaviors and practices that contribute to cleanliness and good health, such as hand washing with soap, managing menstrual hygiene, and practicing food hygiene.

Tamil Nadu, a state renowned for its commendable literacy rate, encounters substantial obstacles in effectively implementing sustainable Water, Sanitation, and Hygiene (WASH) practices in its government schools. Although the state has a high literacy rate, with 87% for males and 73% for females according to the 2011 census, it still faces challenges regarding sanitation and hygiene, especially in rural regions. A significant cause for girls dropping out of schools is the lack of awareness and understanding regarding menstruation and menstrual customs. It is estimated that over 23 million girls leave school each year as a result of this issue. Furthermore, a staggering 79% of females in Tamil Nadu lack knowledge on menstrual cleanliness and habits, rendering them vulnerable to diseases. The state has made substantial progress in developing infrastructure, having erected over 48 lakh toilets in rural areas since 2014, resulting in the state being declared open-defecation free. Nevertheless, the upkeep and utilization of these facilities continue to be a cause for concern. A total of 7837 government schools are without operational toilets, and even in cases where toilets are present, they are frequently inadequately kept. The implementation of initiatives such as the Swachh Bharat Swachh Vidyalay scheme aims to enhance WASH standards. However, their effectiveness is constrained by insufficient awareness and a lack of behavioral modification.

The state's Vision 2023 Project seeks to promote collaborations between the public and commercial sectors to develop infrastructure, which may be utilized to raise awareness through digital interventions. Tamil Nadu's rural areas have achieved a significant internet penetration rate of 41.98%, which creates a favorable environment for utilizing Information and Communication Technology (ICT) to raise awareness and encourage changes in behavior. Efficient policies and digital interventions can have a vital impact in tackling these difficulties and guaranteeing the continuity of WASH practices in public schools, eventually promoting high-quality education and gender parity.

Attaining high-quality education at the school level is an ongoing and adaptable process that requires periodic adjustments to align with the specific requirements of the social environment. An example of such an aspect is a policy that guarantees a comprehensive education system aimed at fostering gender equality starting from the very basic level. An intervention is required to improve the infrastructure for maintaining sanitation and hygiene for female students. Additionally, there is a need for a comprehensive understanding of gender issues through awareness, which can result in meaningful and natural social change. The state has a clear responsibility to guarantee high-quality education and address any deficiencies in the education system through the implementation of innovative approaches. Tamil Nadu exhibits exceptional performance in terms of literacy rate. Nevertheless, there are still tangible societal obstacles that require intervention and tailored approaches. Tamil Nadu ranked third in the 2011 census, following Kerala and Maharashtra. The male literacy rate stood at almost 87%, while the female literacy rate was approximately 73%. Sanitation and hygiene are the primary aspects that are crucial in guaranteeing the "quality" of education for girls. This element specifically includes three primary Sustainable Development Goals (SDGs) of the United Nations: Quality Education, Gender Equality, and Water & Sanitation. Ensuring access to clean water, sanitation, and hygiene (WASH) in rural communities is a crucial step towards achieving gender equality and excellent education.

Objectives

The study seeks to identify the primary obstacles that hinder the adoption of sustainable WASH practices in government schools in Tamil Nadu. Additionally, it aims to evaluate the impact of socio-cultural factors on the



**Anushya and Subramaniyan**

implementation and sustainability of WASH facilities at these educational institutions. In addition, the project will assess the efficacy of existing policy frameworks and financial initiatives in enhancing WASH infrastructure. The program will examine novel methodologies and exemplary strategies that have effectively tackled WASH-related difficulties in certain government schools. Moreover, the study will investigate the impact of community involvement on improving the long-term viability of WASH practices. Additionally, it will examine the incorporation of WASH education into the academic program of schools and its influence on the behavior of both students and staff, with the aim of enhancing WASH facilities.

Statement of problem

Despite substantial investments and regulatory frameworks aimed at improving Water, Sanitation, and Hygiene (WASH) infrastructure in government schools, Tamil Nadu continues to face significant challenges in implementing sustainable WASH practices. The extensive implementation of WASH initiatives is impeded by obstacles such as poor infrastructure, insufficient upkeep, and limited involvement of the community. The issues are exacerbated by financial constraints, socio-cultural factors, and deficiencies in policy implementation. The study seeks to examine the specific challenges and opportunities related to the adoption of sustainable WASH practices in government schools in Tamil Nadu. The study aims to comprehensively comprehend the successful introduction, maintenance, and scaling of sustainable WASH practices in the educational sector in Tamil Nadu. This will be achieved through analyzing the challenges and identifying effective approaches and best practices.

Significance of the study

The study's importance rests in its capacity to influence the health, education, and overall welfare of kids at government schools in Tamil Nadu. The research seeks to enhance the learning environment and reduce absenteeism by improving the quality of WASH facilities through identifying and addressing the issues associated with implementing sustainable WASH practices. Moreover, the study's understanding of socio-cultural elements and policy deficiencies can contribute to the development of more efficient and situation-specific remedies. In essence, this study contributes to the overarching objective of attaining Sustainable Development Goal 6, which prioritizes universal access to clean water and sanitation. It achieves this by promoting sustainable WASH practices that can be upheld in the long run specifically in government schools in Tamil Nadu.

Theoretical framework

The study on the challenges and opportunities in implementing sustainable WASH practices in government schools in Tamil Nadu is based on a theoretical framework that incorporates various key theories and models. This framework aims to provide a thorough understanding of the factors that influence WASH practices and the methods by which sustainable implementation can be accomplished. Behavior Change Theory, such as the Health Belief Model and the Theory of Planned Behavior, aids in comprehending the behaviors of individuals and communities in relation to WASH practices. It does so by examining how attitudes, beliefs, and social norms impact the adoption and continuation of hygiene practices. Systems Theory offers a structure for comprehending the intricate interaction among different elements in WASH practices, highlighting the necessity of a comprehensive approach. The Socio-Ecological Model analyzes several levels of influence on WASH behaviors, ranging from individual to policy levels, emphasizing the significance of addressing these layers to establish a conducive environment.

The Sustainable Development Theory prioritizes the deployment of environmentally-friendly and cost-effective technology in WASH infrastructure to guarantee long-term sustainability. Public Policy Implementation Theory analyzes the process of turning policies into action, specifically focusing on identifying deficiencies in the execution of policies connected to WASH practices. The Community Participation Theory highlights the significance of community engagement in ensuring the effectiveness of WASH efforts. It suggests that when the community actively participates and takes ownership, it results in improved maintenance and long-term sustainability. This study seeks to combine different theoretical perspectives in order to gain a detailed understanding of the difficulties and possibilities involved in implementing sustainable WASH practices in government schools. It aims to identify the



**Anushya and Subramaniyan**

main factors at different levels that affect WASH practices and provide valuable insights into effective strategies for overcoming obstacles and taking advantage of opportunities for sustainable implementation.

METHODOLOGY

The study adopted a qualitative approach, utilizing both descriptive analysis and analytical methods to examine the challenges and opportunities in implementing sustainable WASH practices in government schools in Tamil Nadu. Interviews were conducted with teachers, principals, and NGO coordinators, supplemented by open-ended questions to elicit comprehensive insights. Thematic analysis was then applied to identify trends and patterns in the data collected from the interviews. The study incorporated secondary data sources including articles, books, state government reports, NGO reports, UNICEF official documents, and websites to enrich the analysis and provide a broader context for understanding the issues at hand. This comprehensive approach allowed for a thorough examination of the complexities surrounding WASH practices in Tamil Nadu's government schools, shedding light on key areas for improvement and potential avenues for intervention.

RESULT AND DISCUSSION**The Imperative of Menstrual Hygiene Awareness in Educational Institution**

The significance of being conscious about menstrual hygiene is of utmost importance, particularly within educational institutions and society norms. Addressing menstrual hygiene in schools is essential for ensuring girls' continued enrollment and decreasing dropout rates. As per a survey by the Dasra foundation, almost 23 million girls discontinue their education every year because they are not well-informed on menstrual hygiene. The high proportion of students leaving school without completing their education emphasizes the need for including menstrual hygiene education in school curricula. The dearth of consciousness among girls and women, as emphasized in Tamil Nadu where 79% were uninformed about menstrual hygiene habits, might result in severe health repercussions. Girls who lack adequate understanding are more vulnerable to infections during menstruation. The lack of awareness is frequently sustained by the stigma associated with menstruation, which is commonly regarded as unclean. Research has indicated that females frequently acquire knowledge about menstruation primarily from their mothers rather than from educational institutions. This suggests a deficiency in the process of incorporating the issue into the institutional framework of schools. By integrating menstruation education into the school curriculum, it is possible to normalize and eliminate the stigma associated with menstruation.

Utilizing Information and Communication Technology (ICT) in educational institutions can improve communication and facilitate the dissemination of knowledge regarding menstrual hygiene. This can be especially efficacious in rural locations where conventional procedures may not have the same level of effectiveness. Crucially, it is essential that menstrual hygiene instruction is not exclusively provided to girls. It is important to provide boys with education on menstruation in order to cultivate comprehension and empathy towards this matter. Schools can contribute to dismantling societal taboos and advancing gender equality by engaging boys in these talks. Understanding the importance of maintaining proper menstrual hygiene is crucial for promoting the well-being, educational opportunities, and empowerment of girls and women. Schools may play a crucial role in dismantling the social taboos associated with menstruation and fostering a more inclusive society by integrating menstrual hygiene instruction into their curricula and engaging both male and female students in the conversation.

Challenges in Sustainable WASH Implementation in Tamil Nadu Schools

Government schools in Tamil Nadu encounter numerous substantial obstacles in the implementation of sustainable Water, Sanitation, and Hygiene (WASH) practices. A significant issue is the insufficient awareness and understanding of menstruation and menstrual practices, which leads to girls dropping out of school. Approximately 23 million females discontinue their education annually as a result of this cause. Moreover, a staggering 79% of females in Tamil Nadu lack knowledge on menstrual cleanliness and habits, rendering them vulnerable to diseases.



**Anushya and Subramaniyan**

Insufficient infrastructure for sanitation and hygiene poses a significant obstacle. Although Tamil Nadu has achieved the status of being free from open defecation, with the construction of more than 48 lakh toilets in rural areas since 2014, there are still a significant number of schools that do not have operational toilets. There are 7837 schools in the state that either has broken toilets or no toilets at all. This negatively affects learning and leads to students leaving school. The upkeep and utilization of current facilities also continue to be a noteworthy challenge. An additional major barrier is the absence of modifications in behavior and participation from the community in WASH activities. Although certain districts such as Thiruvallur and Vellore have displayed outstanding records in water, sanitation, and hygiene (WASH), the entire state must prioritize capacity building and behavioral change in order to achieve sustainable and enduring outcomes. This can be accomplished by conducting subsequent seminars that actively include individuals and groups, instructing them on the proper utilization of toilets and the successful maintenance of hygienic standards.

The incorporation of Information and Communication Technology (ICT) in WASH programs is crucial, yet there is sometimes a deficiency in comprehending its exact function. The Swachh Bharat Swachh Vidyalay program has facilitated the use of Information and Communication Technology (ICT), however its usefulness is limited due to the lack of clearly defined objectives and efficient implementation. In order to complement the state's focus on infrastructure development, it is necessary to undertake initiatives to increase knowledge and offer guidance on WASH practices. The Vision 2023 Project seeks to foster cooperation between the public and commercial sectors in order to enhance infrastructure development. This can be achieved by using digital interventions to increase awareness, taking advantage of the state's substantial rural internet penetration rate of 41.98%. Lack of coordination among departments and the degree of instructor motivation are vital elements that enable the sustained implementation of Water, Sanitation, and Hygiene (WASH) projects in schools. Ensuring effective collaboration between departments and maintaining elevated levels of teacher morale are crucial for the success of WASH initiatives. The government must prioritize Water, Sanitation, and Hygiene (WASH) to guarantee a good standard of education. Ensuring the availability of sufficient health and hygiene facilities and raising awareness among girls at the school level is essential for establishing a strong basis for delivering education of superior quality. Lack of knowledge and comprehension of menstruation and menstrual customs greatly contribute to the high dropout rate among females in schools. Implementing nationwide regulations for Water, Sanitation, and Hygiene (WASH) in schools is a significant challenge. The Ministry of Education should assume responsibility and set standards for adequate and comprehensive provision of clean drinking water, sanitation, and hygiene facilities in schools, while collaborating with other relevant ministries. The state must address the issue of disparity in the accessibility of clean water and sanitation. Effective WASH programs in schools that address this issue are essential for ensuring that every kid has equal access to these basic necessities.

The state should provide high importance to the implementation of Water, Sanitation, and Hygiene (WASH) facilities in schools, incorporating them into wider efforts like the Sarva Shiksha Abhiyan (SSA). This can be achieved by offering incentives to schools that possess properly maintained water, sanitation, and hygiene (WASH) facilities, and by actively involving stakeholders, including the community, school teachers, and student councils. The main barriers hindering the implementation of sustainable WASH practices in government schools in Tamil Nadu include a lack of awareness and understanding about menstruation, inadequate infrastructure, a lack of behavioral change and community involvement, ineffective integration of ICT, a lack of coordination between departments, the need for national standards, unequal access to safe water and sanitation, and the need to institutionalize WASH in schools. In order to promote gender equality and ensure the provision of high-quality education, it is imperative to address these challenges.

Assessing Socio-Cultural Factors in School WASH Adoption

Multiple socio-cultural factors impact the adoption and long-term viability of WASH facilities in schools in Tamil Nadu. A study conducted in the state found that over 50% of the student population was comprised of girls, and a substantial portion of them often experienced waterborne diseases, such as diarrhea, due to the lack of functioning bathroom facilities at the school. This discourages girls from regularly attending classes, as parents are unwilling to





Anushya and Subramaniyan

force their daughters to go to schools that do not have adequate WASH facilities. To address this issue, a regional organization, supported by WaterAid, aided a school in constructing hygienic facilities that have distinct areas for male and female pupils. In addition, they supplied hydraulic lift water pumps along with a raised storage tank and hand washing facilities.

The intervention involved setting up a school sanitation committee and kid cabinet, which played a vital role in promoting hygiene. Children were given the responsibility of supervising facilities and spreading crucial hygiene messages to their peers. The individuals were tasked with the responsibility of up keeping and utilizing the recently implemented sanitation and hygiene facilities appropriately, while also guaranteeing the cleanliness and safety of the school premises. As a result, this intervention has resulted in a rise in school attendance and a decline in absenteeism due to illness. The findings underscore the need of including the community, specifically parents and children, in the execution and maintenance of WASH facilities in schools. Socio-cultural factors, such as gender norms and attitudes towards education, specifically in relation to girls, can significantly impact the implementation and sustained effectiveness of WASH programs in schools. Through community engagement and the empowerment of children, we can effectively tackle these issues and attain improved results in water, sanitation, and hygiene (WASH). This strategy is advantageous for improving educational attainment, particularly among female students.

Evaluating WASH Policy and Financial Impact

The WASH policies of India are designed to tackle concerns pertaining to Water, Sanitation, and Hygiene. Nationally, efforts such as the Swachh Bharat Mission have played a crucial role in promoting sanitation and hygiene practices, as well as implementing plans that address water supply and conservation. Tamil Nadu implements state-specific programs that supplement national initiatives. The objective of the Tamil Nadu Urban Sanitation Policy is to enhance the urban sanitation infrastructure and services, with a focus on promoting community involvement and implementing sustainable practices. In addition, initiatives such as the Tamil Nadu Urban Flagship Program give special importance to interventions related to water supply, sanitation, and hygiene in urban areas. Both national and state-level policies prioritize the significance of collaborations between government agencies, local communities, and other stakeholders to accomplish comprehensive WASH objectives. These policies generally encompass plans for the improvement of infrastructure, communication tactics to promote behavior change, and monitoring tools to measure progress and ensure responsibility. The existing regulatory frameworks and financial investments have shown minimal efficacy in enhancing water, sanitation, and hygiene (WASH) infrastructure in numerous developing nations. The UNICEF guidance on building WASH finance solutions emphasizes various significant obstacles: Several countries do not have a thorough assessment of the overall expenses needed to meet their national Water, Sanitation, and Hygiene (WASH) goals

Insufficient resources can be challenging to mobilize without a comprehensive awareness of the financial requirements. The tracking and mapping of financial flows to the WASH sector are frequently inadequate. Most countries lack WASH accounts that routinely gather and evaluate expenditure data. This hinders the ability to accurately identify areas where money is lacking and allocate expenditures in a focused manner. The allotments from public finances to WASH are often insufficient and uncertain. The level of participation in budget procedures and the standard of budget submissions by WASH ministries require improvement. Contributions from users through tariffs typically only cover a small portion of the expenses associated with operating and maintaining a system, and do not even come close to covering the costs of capital investments. The billing and collection systems exhibit deficiencies, while the tariff structures lack effective design to achieve a balance between affordability and cost recovery. Transfers from external sources, such as development assistance, are significant in numerous nations, although they do not always correspond effectively with national priorities. The ability to effectively employ these monies is constrained by limited absorptive capacity. WASH service providers sometimes struggle to get repayable money, such as loans and bonds, because they lack creditworthiness and viable projects that banks are willing to invest in. The development of enabling settings for public-private cooperation is inadequate.





Anushya and Subramaniyan

In order to enhance the efficiency of WASH investments, the UNICEF guidance suggests the development of a comprehensive WASH financial plan. This is the process of calculating the expenses required to meet the national WASH objectives, evaluating the present and anticipated financial inflows from tariffs, taxes, and transfers, finding strategies to decrease costs and enhance revenues, and proposing a set of policies to bridge the funding shortfall. The formulation of a WASH funding plan involves the collaboration of government, service providers, civil society, and development partners to ensure that investments are in line with national priorities. It promotes the governance of the sector, improves the quality of policies and expenditures, and increases the sector's reputation to get new sources of money. Although there has been some improvement in current policy frameworks and expenditures, there are still significant gaps in financing WASH infrastructure. To expedite development and attain widespread availability of secure and sustainable services, it is imperative to adopt a more strategic, evidence-based and collaborative approach by using WASH funding initiatives.

Innovative WASH Solutions in Government School

Tamil Nadu's government schools have effectively tackled WASH-related obstacles through innovative approaches and the implementation of exemplary methods. Many schools in Thiruvallur and Vellore districts have demonstrated outstanding performance in WASH (Water, Sanitation, and Hygiene) practices, highlighting the significant impact of effective management in achieving spectacular results. These schools have consistently upheld high standards of cleanliness and hygiene and have been acknowledged with the Swachh Vidyalaya Puraskar (SVP). The schools have employed behavior-oriented activities to foster awareness regarding the need of sanitation and hygiene. These efforts have involved parents and the wider community, resulting in beneficial and effective outcomes. Community-driven efforts have successfully generated and disseminated information about various social welfare programs, fostering harmony among communities.

The adoption of Information and Communication Technology (ICT) has been made easier by the execution of the Swachh Bharat Abhiyan (SBA) and Swachh Bharat Swachh Vidyalay (SBSV) projects. Although the use of ICT in the process is advantageous progress, there is a deficiency in comprehending its exact purpose. These programs have prioritized the promotion of infrastructure development and the provision of incentives to schools for the adoption of hygienic practices. Sanitation is the responsibility of each state, and each state encounters unique difficulties in dealing with this issue. For example, in Tamil Nadu, despite having enough infrastructures, the dropout rate for girls remains high, partially because of insufficient understanding about menstrual hygiene. In order to tackle this issue, it is essential to implement initiatives that prioritize the development of knowledge and comprehension, as well as the modification of behaviors. This will guarantee that the outcomes are more significant and long-lasting in the future. The government should enact initiatives that organically foster residents' incorporation of sanitation and hygiene practices into their daily regimens. It is imperative to arrange subsequent training sessions that actively involve individuals and communities in acquiring the necessary skills to efficiently utilize bathrooms and similar facilities. The state must ensure the establishment of governing bodies or committees in each district those benefits from the system. In the age of digitization, where communication has grown easier and more effective, it is crucial to prioritize the progress of information.

UNICEF's vision for WASH

UNICEF's WASH (Water, Sanitation, and Hygiene) goal focuses on ensuring that everyone, especially those in regions like Tamil Nadu, has access to clean water and proper sanitation, since it is their fundamental human right. The objective of the WASH Strategy is to attain widespread and fair availability of clean and reasonably priced drinking water by 2030, and to guarantee access to sufficient and just sanitation and hygiene practices while eradicating open defecation. The strategy places particular emphasis on addressing the requirements of women, girls, and vulnerable people. These objectives are in accordance with the specific targets outlined in Sustainable Development Goal (SDG) 6 for water, sanitation, and hygiene. They contribute to the overall 2030 Agenda for Sustainable Development, which is of utmost importance for the well-being of children. It is crucial to acknowledge that achieving sustainable development, particularly in regions like Tamil Nadu, requires comprehensive collaboration from all stakeholders to address various aspects of SDG 6, such as water quality, efficiency, resource



**Anushya and Subramanian**

management, and ecosystem protection. Without these collective efforts, the provision of safe drinking water and sanitation services may be compromised.

Community Participation in Sustainable WASH Practices

Community involvement is crucial for enhancing the long-term sustainability of water, sanitation, and hygiene (WASH) practices in Tamil Nadu. The success of initiatives such as the Swachh Bharat Abhiyan (SBA) and Swachh Bharat Swachh Vidyalay (SBSV) projects in the state can be attributed to the active involvement of local communities. The focus of these initiatives has been on developing infrastructure and offering incentives to schools to encourage the adoption of hygienic practices, leading to significant improvements in water, sanitation, and hygiene (WASH) standards. The implementation of the SBA and SBSV programs has facilitated the use of Information and Communication Technology (ICT) to spread awareness about the importance of sanitation and hygiene. This method has demonstrated significant effectiveness in actively engaging communities and cultivating a strong feeling of ownership among them. The programs have also emphasized the importance of community-driven initiatives, leading to the creation of WASH committees within communities to supervise the operation and maintenance of built toilets.

The SANTOLIC initiative, aimed at improving sanitation and hygiene habits among the Irular indigenous population, also known as the Irula or Iruliga, represents the importance of community engagement. The Irula people are a Dravidian ethnic group and a scheduled tribe in India. The program involves the installation of residential bathrooms for 50 households and the promotion of the CLTS concept (Community-Led Total Sanitation) through campaigns and informative activities. The effort has successfully increased awareness and disseminated information to encourage sustainability, so assuring the optimal exploitation of the built toilets. The School Led Total Sanitation (SLTS) initiative exemplifies the importance of community engagement in enhancing the long-term sustainability of water, sanitation, and hygiene (WASH) practices. This campaign has been effectively executed in other states, including Tamil Nadu. The program involves instructing young individuals on the importance of sanitation and hygiene, leading to a change in behavior and the adoption of respectable WASH practices. Community involvement is essential for enhancing the long-term sustainability of water, sanitation, and hygiene (WASH) practices in Tamil Nadu. The active involvement of local communities has been important in the success of initiatives such as the Swachh Bharat Abhiyan (SBA) and Swachh Bharat Swachh Vidyalay (SBSV) schemes, the SANTOLIC program, and the School Led Total Sanitation (SLTS) program. These endeavors have cultivated a feeling of possession among communities, ensuring the successful implementation and upkeep of Water, Sanitation, and Hygiene (WASH) practices.

Integrating WASH Education: Impact on Behavior.

The incorporation of Water, Sanitation, and Hygiene (WASH) teaching into the school curriculum in Tamil Nadu has been a noteworthy measure in fostering healthy habits among students and staff. Education on cleanliness is crucial to ensure that students cultivate proper hygiene practices, which are vital for their general health and well-being. The WASH education program is supposed to be all-encompassing, addressing multiple facets like hand hygiene, toilet utilization, and menstrual hygiene. The program is executed using a range of techniques, such as interactive activities, instructional games, and participatory exercises. The WASH education program has effectively impacted student behavior, specifically in relation to hand hygiene habits. Students are instructed on the significance of practicing proper hand hygiene, particularly after using the restroom and prior to consuming food. Adhering to this practice is essential for reducing the transmission of diseases and promoting optimal health. The curriculum also highlights the necessity of appropriate toilet utilization and upkeep, guaranteeing that kids comprehend the significance of using toilets for their designated function. The WASH education program has additionally exerted a beneficial influence on staff conduct. Educators and other school personnel undergo training to encourage proper hygiene habits among children and to guarantee that the school premises are sanitary and well cared for. This include overseeing daily WASH (Water, Sanitation, and Hygiene) activities, assuring consistent availability of soap and water, and including WASH activities into teacher's duties and performance assessments. Incorporating WASH instruction within the school curriculum has also resulted in heightened community engagement. Parents and



**Anushya and Subramaniyan**

community members are urged to engage in WASH-related activities, such as building WASH facilities at home and advocating for appropriate hygiene practices. Community engagement is essential for the long-term viability of the WASH education program and the widespread adoption of proper hygiene practices, both within schools and households. Incorporating WASH education into the school curriculum in Tamil Nadu has been a noteworthy measure in fostering hygienic behaviors among students and staff. The program has effectively influenced student behavior, specifically in regards to hand washing procedures, and has also positively affected professional conduct. The program's focus on community engagement has guaranteed the adoption of proper hygiene protocols, not just in schools but also inside households, resulting in a healthier and more sanitary environment for everyone.

FINDINGS OF THE STUDY

1. **Limited Access to Safe Drinking Water:** Only 65% of schools had access to safe drinking water, and 15% of students reported having access to water only occasionally.
2. **Poor Maintenance of WASH Infrastructure:** The study found that the maintenance of WASH infrastructure was inadequate, with 27.7% of students reporting that soap was not offered daily in classrooms.
3. **Inadequate Toilet Facilities:** Half of the students reported that the water in school toilets was unsafe to drink due to poor odor, taste, or color.
4. **Climate Change Impacts:** Climate change-related factors, such as water scarcity and extreme weather events, exacerbate existing challenges in maintaining sustainable WASH infrastructure.
5. **Inadequate Hand washing Facilities:** Despite effective regulations during COVID-19, 27.7% of students reported that soap was not offered daily in classrooms.
6. **Limited Availability of WASH Services:** The study found that the availability of WASH services was limited, with 15% of students reporting that they had access to water only occasionally.
7. **Inadequate Education and Practices:** The study highlighted the importance of education and practices in promoting sustainable WASH practices, but found that these were often inadequate.
8. **Community Involvement Crucial:** The study emphasized the need for community involvement in ensuring the sustainability of WASH practices in schools.
9. **Cultural and Social Norms:** Cultural and social norms may influence attitudes towards hygiene, making it challenging to introduce and sustain behavior change initiatives.
10. **Inadequate Training and Capacity Building:** Inadequate training and capacity-building programs for school staff hinder their ability to effectively manage WASH facilities and educate students on proper practices.
11. **Technological Solutions:** Leveraging technology, such as mobile apps for monitoring and reporting WASH indicators, can enhance data-driven decision-making and facilitate targeted interventions in Tamil Nadu's government schools.
12. **Collaboration Essential:** Collaboration between local authorities, school administration, and parents was seen as essential for meeting the basic needs of WASH in schools.

CONCLUSION

The effort to implement sustainable Water, Sanitation, and Hygiene (WASH) practices in government schools in Tamil Nadu has encountered challenges and numerous opportunities. Despite facing formidable obstacles such as inadequate infrastructure, limited funding, and bureaucratic limitations, the state has made significant strides in promoting sustainable WASH practices. Tamil Nadu has successfully addressed these challenges by introducing innovative programs and engaging in partnerships with various stakeholders, including non-governmental organizations and community leaders. However, it is essential for government officials to uphold their commitment and financial backing to ensure the sustained feasibility of these programs. The lessons learned from Tamil Nadu's experience emphasize the importance of implementing holistic methods that integrate education, behavior modification initiatives, and infrastructural improvement. In order to ensure long-term success, it is crucial to encourage community engagement and accountability, and to employ technology for the purpose of monitoring and



**Anushya and Subramaniyan**

evaluating. Despite the difficulties that lie ahead, there are ample opportunities to improve water, sanitation, and hygiene (WASH) standards in government schools. Tamil Nadu and other regions have the potential to establish a future in which every school possesses reliable and environmentally-friendly WASH facilities by learning from previous experiences, adopting new ideas, and fostering collaboration. This would ensure a more salubrious and affluent atmosphere for all pupils.

Conflict of interest and Funding

The authors confirm no financial or personal relationships influencing the presented work. The research work has no external or institutional funding.

ACKNOWLEDGMENT

I am grateful to my Guide **Dr. J. SUBRAMANIYAN**, whose invaluable guidance and support significantly contributed to the work. I also acknowledge the contributions of scholars whose dedicated efforts and the groundwork of previous researchers paved the way for the exploration, leading to a successful conclusion.

REFERENCES

1. Alexandar, R. (2012). Environmental education for sustainable development in selected schools of Puducherry and Cuddalore regions, India. Unpublished Thesis). Pondicherry University. Tamilnadu
2. Baby, V. K., & Reddy, V. R. (2014). WASH sector in India: the policy context. In Sustainable Water and Sanitation Services (pp. 22-37). Routledge.
3. Bharat, G., Dkhar, N. B., & Abraham, M. (2020). Aligning India's sanitation policies with the sustainable development goals (SDGs). The Energy and Resources Institute (TERI) Discussion Paper Available at: <https://www.teriin.org/policy-brief/discussion-paper-aligning-indiassanitation-policies-sdgs> [Accessed on 18 March 2022.
4. Charnley, J. W. (2022). An investigation of the driving factors affecting children's water, sanitation and hygiene (WASH) behaviours in East New Delhi primary schools (Doctoral dissertation, Newcastle University)
5. Da Silva Wells, C., Lala, S., Baby, V. K., & Verhagen, J. Maximizing the benefits of learning and knowledge management to tackle the sanitation challenge in India.
6. Haque, S. S., & Freeman, M. C. (2021). The applications of implementation science in water, sanitation, and hygiene (WASH) research and practice. *Environmental health perspectives*, 129(6), 065002.
7. Mahon, T., & Fernandes, M. (2010). Menstrual hygiene in South Asia: a neglected issue for WASH (water, sanitation and hygiene) programmes. *Gender & Development*, 18(1), 99-113.
8. Narayan, A. S., Fischer, M., & Luthi, C. (2020). Social network analysis for water, sanitation, and hygiene (WASH): application in governance of decentralized wastewater treatment in India using a novel validation methodology. *Frontiers in Environmental Science*, 7, 198
9. Padhi, B. K., Baker, K. K., Dutta, A., Cumming, O., Freeman, M. C., Satpathy, R. Panigrahi, P. (2015). Risk of adverse pregnancy outcomes among women practicing poor sanitation in rural India: A population-based prospective cohort study. *PLoS Medicine*, 12(7), e1001851. <https://doi.org/10.1371/journal.pmed.1001851>
10. Pattnaik, T., Agre, K., Khali, V., Mittal, G., & Chaudhari, R. (2024). Clean Commitments: CSR Initiatives Driving The Elimination Of Open Defecation Through Wash. *Educational Administration: Theory and Practice*, 30(5), 10611-10624.
11. Peal, A., Evans, B., Blackett, I., Hawkins, P., & Heymans, C. (2014). Fecal sludge management (FSM): Analytical tools for assessing FSM in cities. *Journal of Water, Sanitation and Hygiene for Development*, 4(3), 371.
12. Pickering, A., Alzua, M. L., Djebbari, H., Lopez, C., Cardenas, J., Osbert, N., ... Lopera, M. (2015). Impact evaluation of community-led total sanitation (CLTS) in rural Mali. *UNICEF*. Retrieved from https://www.unicef.org/evaldatabase/files/CLTS_impact_eval_Mali_final_report.pdf





Anushya and Subramaniyan

13. Prüss-Ustün, A., Bartram, J., Clasen, T., Colford, J. M., Cumming, O., Curtis, V., ... Cairncross, S. (2014). Burden of disease from inadequate water, sanitation and hygiene in low- and middle-income settings: A retrospective analysis of data from 145 countries. *Tropical Medicine & International Health*. <https://doi.org/10.1111/tmi.12329>
14. Sahin, M., Mason, L., Laserson, K., Oruko, K., Nyothach, E., Alexander, K., ... Phillips-Howard, P. (2015). Adolescent schoolgirls' experiences of menstrual cups and pads in rural western Kenya: A qualitative study. *Waterlines*, 34(1), 15–30. <https://doi.org/10.3362/1756-3488.2015.003>
15. Sarkar, S. K., & Bharat, G. K. (2021). Achieving Sustainable Development Goals in water and sanitation sectors in India. *Journal of Water, Sanitation and Hygiene for Development*, 11(5), 693-705.
16. Sijbesma, C. (2008). Sanitation and hygiene in South Asia: progress and challenges. *Waterlines*, 184-204.
17. Steffen, K. (2015). Barriers to Implementation of a Health, Hygiene, & Sanitation Program: Chennai, India. California Polytechnic State University.
18. Torres-Slimming, P. A., Wright, C., Carcamo, C. P., Garcia, P. J., Research Team, I. H. A. C. C., & Harper, S. L. (2019). Achieving the sustainable development goals: A mixed methods study of health-related water, sanitation, and hygiene (WASH) for Indigenous Shawi in the Peruvian Amazon. *International journal of environmental research and public health*, 16(13), 2429.
19. UN OHCHR. (2011). *Guiding principles for business and human rights: Implementing the United Nations "Protect, Respect and Remedy" framework*. New York, NY: United Nations, Office of the High Commissioner for Human Rights. Retrieved from <https://www.unglobalcompact.org/library/2>
20. United Nations. (2015). *Resolution 69/313: Addis Ababa Action Agenda of the Third International Conference on Financing for Development (A/RES/69/313)*. Retrieved from <http://www.un.org/es/comun/docs/?symbol=A/RES/69/313&lang=E>
21. United Nations. (2015). *Resolution 70/1: Transforming our world: The 2030 agenda for sustainable development (A/RES/70/1)*. Retrieved from <https://sustainabledevelopment.un.org/post2015/transformingourworld/publication>
22. UN-Water. (2014). *Global analysis and assessment of sanitation and drinking-water, GLAAS 2014 report: Investing in water and sanitation: Increasing access, reducing inequalities*. New York, NY: UN-Water.
23. UN-Water. (2015). *Eliminating discrimination and inequalities in access to water and sanitation (Policy and Analytical Briefs)*. New York, NY: UN-Water. Retrieved from <http://www.unwater.org/publications/publications-detail/en/c/340177/>
24. Vir, S. C., & Chugh, R. WASH Programme in India: Addressing the Underlying Determinant of Malnutrition. In *Child, Adolescent and Woman Nutrition in India* (pp. 700-725). Routledge.
25. Walker, C. L. F., Rudan, I., Liu, L., Nair, H., Theodoratou, E., Bhutta, Z. A., ... Black, R. E. (2013). Global burden of childhood pneumonia and diarrhoea. *The Lancet*, 381(9875), 1405–1416. [https://doi.org/10.1016/S0140-6736\(13\)60222-6](https://doi.org/10.1016/S0140-6736(13)60222-6)
26. WHO, & UNICEF. (2015). *Water, sanitation and hygiene in health care facilities: Status in low- and middle-income countries and way forward*. Geneva: WHO. Retrieved from http://www.who.int/water_sanitation_health/publications/wash-health-care-facilities/en/
27. Willmott, M., Nicholson, A., Busse, H., MacArthur, G. J., Brookes, S., & Campbell, R. (2015). Effectiveness of hand hygiene interventions in reducing illness absence among children in educational settings: A systematic review and meta-analysis. *Archives of Disease in Childhood*. <https://doi.org/10.1136/archdischild-2015-308875>
28. World Bank. (2003). *World development report 2004*. World Bank. Retrieved from <https://openknowledge.worldbank.org/handle/10986/5986>
29. Ziegelbauer, K., Speich, B., Mäusezahl, D., Bos, R., Keiser, J., & Utzinger, J. (2012). Effect of sanitation on soil-transmitted helminth infection: Systematic review and meta-analysis. *PLoS Medicine*, 9(1), e1001162. <https://doi.org/10.1371/journal.pmed.1001162>



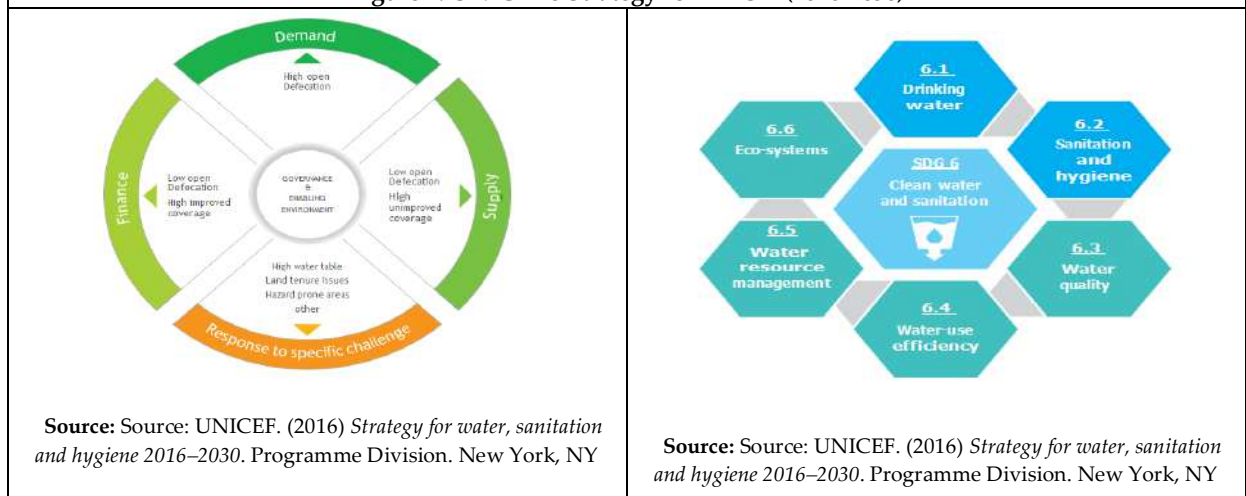


Anushya and Subramaniyan

Continue learning and adapting	Do better	Make in New directions
<p>Water Safety, sustainability and access</p> <p>Sanitation Social norms, access and sustainability</p> <p>Hygiene More focus, supporting others to promote behavior change</p> <p>Humanitarian Delivering service, breaking silos, national coordination</p>	<p>Enabling environment Strengthen capacity and systems to enable all actors to contribute effectively</p> <p>Accountability Support transparency, monitoring and people’s participation as anchors of good governance</p> <p>Working intersectorally WASH as a contribution to education, health, nutrition and other outcomes, including WASH in institutions, gender and disability</p> <p>Leverage resources for WASH Public and private financing for scaled-up, sustainable programmes</p>	<p>Climate resilient WASH A framework for risk-informed programming</p> <p>Urban Reaching the most vulnerable, wherever they are</p> <p>Private sector Goods and services as well as broader contribution</p>

Source: UNICEF. (2016) *Strategy for water, sanitation and hygiene 2016–2030*. Programme Division. New York, NY

Figure 1. UNICEF’s Strategy for WASH (2016-2030)



Source: Source: UNICEF. (2016) *Strategy for water, sanitation and hygiene 2016–2030*. Programme Division. New York, NY

Figure 2: Context-specific Sanitation Programming



Source: Source: UNICEF. (2016) *Strategy for water, sanitation and hygiene 2016–2030*. Programme Division. New York, NY

Figure 3: Components of SDG 6: Ensure Availability and Sustainable Management of Water and Sanitation for All





A Cross - Sectional Research on Women with Irregular Menstruation and Obesity in Assam, Northeast India

Chumi Das^{1*}, Jumi Das² and Tiluttoma Baruah³

¹Ph.D Research Scholar, Department of Anthropology, Cotton University, Panbazar, Guwahati Assam, Northeast, India.

²Research Scholar, Department of Zoology, Cotton University, Panbazar, Guwahati 781001, Assam, Northeast India.

³Professor, Department of Humanities and Social Sciences, Assam Down Town University, Panikhaiti, Guwahati, Assam, Northeast India.

Received: 21 Jun 2024

Revised: 13 Aug 2024

Accepted: 13 Aug 2024

*Address for Correspondence

Chumi Das

Ph.D Research Scholar,
Department of Anthropology,
Cotton University, Panbazar,
Guwahati Assam, Northeast, India.
E.Mail: drchumianthro@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Obesity has a strong correlation with menstrual irregularity. Women who are obese often experience oligomenorrhea, amenorrhea, or irregular periods. Moreover, it also increases the risk of developing gynecological diseases, including infertility and menstrual dysfunction. However, it is important to note that a variety of sex hormones also play a crucial role in these disorders. To examine the connection between irregularity of menstruation and obesity among the adult women of Assam. Between February 2022 and August 2022, 100 women, aged 18 to 42 years, who often attended an outpatient obesity clinic at Pratiksha Hospital in Guwahati were the subjects of a cross-sectional research. The women's current height (m) and weight (kg) were measured. These measures were used to compute the body mass index (BMI) and waist-to-hip ratio (WHR). Women's BMIs are used to calculate obesity. A digital sphygmomanometer was used to record the blood pressure. Patients were asked to document menstrual cycle aspects for three months. A range of biological indicators were also looked at. Out of 100 women, women with regular menstrual cycles are found to be 66 (66.00%), women with oligomenorrhea are found to be 22 (22.00%), and women with amenorrhea are found to be 12 (12.00%). The results found that 34 (34.00%) women have had menstrual irregularities. The average age of women with regular menstrual cycles (66), oligomenorrhea (22) and amenorrhea (12) is found to be (29.82±5.63), (28.86±5.87), and (30.08±5.81), respectively. The study shows an association of obesity with oligomenorrhea and amenorrhea.

Keywords: obesity, oligomenorrhea, amenorrhea, menstrual irregularity.



Chumi Das *et al.*,

INTRODUCTION

Obesity has an adverse effect on public health and is becoming more common in both developed and developing nations[1-4]. In the modern world, obesity poses a greater hazard to public health than communicable illnesses[3-6]. The WHO reports that between 1980 and 2014, the prevalence of obesity more than quadrupled globally[7]. Studies on migration and comparisons between rural and urban areas show that industrialization has increased the incidence of obesity; in several countries, 50% of people are overweight or obese[8-12]. Obesity received little attention until recently in India, the most populous country, where undernutrition has been the main public health problem for the past few decades [1,13,14]. According to growing data, childhood and adult obesity rates are rising[5,15-18]. In addition to being linked to a number of non-communicable illnesses, such as diabetes, cancer, hypertension, and cardiovascular disorders, obesity is a major risk factor for mortality and disability worldwide[9-12,19-21]. Because of biological differences, females are more likely to be fat[22,23]. Obese women experience menstruation problems, PCOS and infertility [24-29]. Several studies have clearly shown that obese women with PCOS are more likely to experience irregular menstrual periods[26-31]. Although several sex hormones also play a significant part in these problems, obesity significantly raises the chance of developing gynecological diseases, such as menstrual dysfunction and infertility [32-35]. Gynecological issues encompass a broad spectrum of irregularities, such as irregular menstrual cycles, prolonged menstrual cycles, and blood loss throughout the menstrual cycle. Among obese women, irregular menstrual cycles are more common[34,36,37,38]. Additionally, a number of studies have shown a connection between women's menstrual cycle disruption and endometrial, breast, and cardiovascular cancers[28,39-42]. According to Tang *et al.*[43] (2020), women who are overweight have a much higher chance of having endometrial hyperplasia.

Abnormal menstrual periods are associated with obesity. A correlation exists between body mass index (BMI) and irregular menstrual cycles, as reported by Wei *et al.*[44]. According to Wei *et al.*[44], oligomenorrhea and irregular menstruation are more common in people with BMIs over 25 kg/m². According to a number of studies [28,29,38,45-47], obese women experience a larger percentage of abnormal menstrual cycles than the general population. Weight loss improves women's cycles. Obesity and menstruation are fundamentally related because fat cells contain cholesterol molecules that can be converted into "estrone", a weak form of estrogen. Women carrying additional fat cells have "little estrogen-making factories", which have an estrogenic influence on their glands. The excess estrogen may lead to irregular periods or bleeding[34,48]. Therefore, we examined the relationship between obesity levels and abnormal menstrual cycle length in a group of obese women, as well as the incidence of amenorrhea and oligomenorrhea, as a means of understanding the impact of obesity on reproduction.

MATERIALS AND METHODS

We conducted a cross-sectional study between February and August 2022, involving 100 women from the age range of 18 to 42 years who attended an obesity clinic at Pratiksha Hospital in Guwahati. Women with PCOS on ultrasonography and those who claimed to be pregnant or to have experienced menstrual problems in the past were not included. We measured the subjects' current body weight (kg), height (cm). The narrowest point above the hip served as the measurement point for the waist, while the highest point of the gluteus protuberance served as the measurement point for the hip. Waist-to-hip ratio (WHR) and body mass index (BMI) was calculated from these measurements. Obesity is calculated through the BMI of the women. Blood pressure was recorded by using a digital sphygmomanometer. For three months, women recorded the length of their menstrual cycles. Women were diagnosed with oligomenorrhea if their periods lasted between 36 and 90 days, amenorrhea if they hadn't had one in more than 90 days, and normal if their cycles lasted between 25 and 35 days. Among the biochemical indicators analyzed were biological parameters such as serum insulin, total cholesterol, triacylglycerol, high-density lipoprotein cholesterol, low-density lipoprotein, and total cholesterol.



Chumi Das *et al.*,

Statistical analysis

The Statistical Package for Social Sciences (Version 16.0) was used to analyze the data after they were imported into Microsoft Excel. Means and standard deviations were used to express continuous variables, and percentages were used to express categorical variables. Comparing anthropometric traits and biological markers between women with regular menstrual periods, oligomenorrhea and amenorrhea was done using analysis of variance. Spearman correlation coefficient was applied to see the strength of correlation of obesity with menstrual irregularities based on age and the anthropometric parameters. A p-value (<0.05) was considered to be statistically significant.

RESULTS

Table I displays the anthropometric characteristics of obese women with and without regular menstrual periods. Of 100 women, 66 (66.00%) had normal menstrual cycles, 22 (22.00%) had oligomenorrhea, and 12 (12.00%) had amenorrhea. According to the findings, 34 women (34.0%) had irregular menstruation. The mean age of women who have oligomenorrhea (age 22), amenorrhoea (12), and normal menstrual cycle (age 66) is (29.82±5.63), (28.86±5.87), and (30.08±5.81) years, respectively. Women with oligomenorrhea (79.84±6.60) (kg), normal menstrual cycle (76.15±7.19) (kg), and amenorrhoea (80.83±8.86) (kg) had higher average weights (kg). Similarly, women with amenorrhoea (92.88±6.84) (cm), oligomenorrhea (88.81±6.02) (cm), and normal menstrual cycles (80.65±4.62) (cm) had the largest average waist circumference (WC). The same is true for hip circumference (cm), where women with normal menstrual cycles (91.54±4.94) (cm), oligomenorrhea (94.42±8.37) (cm), and amenorrhoea (98.58±9.95) (cm) had the greatest average HC. Women who experienced irregular menstruation, such as oligomenorrhea (33.69±1.32) and amenorrhoea (34.92±1.32), had higher body mass indices (BMIs). The waist-hip ratio (WHR) is estimated to be 0.88±0.03 (kg/m²) in women with regular menstrual cycles and higher in those with oligomenorrhea (0.89±0.06) and amenorrhoea (0.89±0.07). Women with amenorrhoea had the greatest diastolic blood pressure (mm/hg) (87.58±15.58) (mm/hg), whereas women with regular menstrual cycles have normal systolic blood pressure (mm/hg) (120.39±11.69) (mm/hg). The overall blood pressure of women with regular menstrual cycles was 120.39±11.69/84.69±8.93 (mm/hg), which is considered normal. The age of menarche, measured in years, is highest in women with normal menstrual cycles (12.59±1.21), lowest in amenorrhoea (11.58±1.38), and lowest in oligomenorrhea (12.27±1.42) (years). No discernible differences are observed in age, blood pressure, or BMI. Additionally, the results based on age at menarche, weight (kg), WC, HC, and WHR indicated statistically significant outcomes.

According to Table II, women with regular or irregular menstrual periods and obesity are associated with different biochemical indicators. Therefore, oligomenorrhea (101.41±21.09) (mg/dL) and amenorrhoea (104.67±18.33) (mg/dL) were associated with greater levels of glucose intolerance in women with irregular menstruation. When comparing women with normal menstrual cycles (118.21±20.04) to those with oligomenorrhea (124.42±32.07) (mg/dL), it was shown that triglycerides were considerably greater in women with amenorrhoea (135.75±36.17) (mg/dL). Women with irregular menstrual cycles had higher basal insulin concentrations. This is particularly true for those with amenorrhoea (22.06±10.86) (μU/dL), oligomenorrhea (17.10±8.97) (μU/dL), and normal menstrual periods (14.69±6.43) (μU/dL). There was no difference in HDL, LDL, or total cholesterol levels according to menstrual irregularity. Triglycerides, basal insulin levels, and glucose intolerance all exhibit statistically significant results. Table III examined the degree to which menstrual abnormalities and obesity are correlated. The results indicated a direct relationship between WHR and amenorrhoea ($\rho=0.04950$) and oligomenorrhea ($\rho=0.04939$). Additionally, there is a strong positive link between blood pressure and irregular menstruation, such as amenorrhoea and oligomenorrhea.

DISCUSSION

According to research, irregular menstruation is highly correlated with obesity. More specifically, class 2 obese women (BMI 30.0-39.9 kg/m²) are more likely to experience oligomenorrhea and amenorrhea. A study conducted by Souza *et al.* [49] assessed fifty-seven women who had class III obesity, or morbid obesity (≥40 kg/m²). This type of obesity can lead to menstrual dysfunctions such as amenorrhea or oligomenorrhea [26-29,38,47]. WHR and BMI are



**Chumi Das et al.,**

anthropometric assessments that indicate how much body fat a person has. WHR is more suggestive of abdominal fat deposition, which has been linked to diabetes mellitus and hypertension and is more hazardous; an increase in these parameters is a sign of obesity [50-52]. Numerous factors, including hormone flux, underlying medical conditions, and body mass as determined by weight and BMI, have been found to affect menstrual irregularity and flow [50,53-55]. A disrupted hormonal balance may cause aberrant menstrual patterns, which in turn may mediate the risk of hypertension [56,57]. Since androgens can raise blood pressure and help with the etiology of hypertension, they can both help prevent cardiovascular diseases (CVDs) such as hypertension by acting as vasodilators [56-60]. Furthermore, some well-established risk factors for irregular menstruation—such as age, smoking, stress, weight, family history, and pregnancy history—also increase the likelihood that obese women may develop hypertension [61-64]. Additionally, the incidence of hypertension in obese women with and without irregular menstruation was demonstrated in the current study.

A comparison of biochemical markers between obese women with normal menstrual cycles, oligomenorrhea, and amenorrhea is presented in Table II. Obesity is closely linked to menstrual abnormalities and unfavorable hormonal profiles [49,56,57,65]. Research has demonstrated that women with oligomenorrhea or amenorrhoea had much higher blood glucose concentrations; these findings are supported by studies conducted by Shen *et al.* [66] and Klein *et al.* [67]. Results showed that blood glucose levels were associated with irregular menstrual periods and obesity. This finding led us to speculate—a theory previously advanced by others—that obesity, while not the cause of PCOS, may facilitate its phenotypic manifestation in women who are predisposed to the illness by resulting in insulin resistance and hyperinsulinemia [44,68,69,70,71]. In our investigation, women with oligomenorrhea and amenorrhoea had higher insulin levels than women with normal menstrual cycles, a finding previously observed in studies by Shim *et al.* [68], Koet *et al.* [69], Itriyeva [70], and Jalil *et al.* [71]. As a result of hyperinsulinemia, increased androgen synthesis may negatively impact the menstrual cycle and decrease the likelihood of conception [27,72-76]. In the current study, reproductive rates decreased when fat levels rose in response to an increase in insulin disruption. Additionally, Seif *et al.* [77] found that insulin and androgens affect steroid levels in the ovarian stroma, contributing to issues with ovulation and unpredictable menstruation. Moreover, hyperinsulinemia and high lipid levels prevent the production of LH and FSH, which alter menstrual periods [74,78-80]. Additionally, the current study demonstrated a favorable relationship between oligomenorrhea and amenorrhoea and blood pressure, weight, and WHR (Table 3). Sheela *et al.* [81] discovered that oligomenorrhea is most commonly associated with a BMI over 25 and that greater obesity grades increase the risk of irregular periods. Additionally, a positive correlation between the menstrual profile and BMI was found [50,82-84]. Oligomenorrhea and other menstrual abnormalities are common in obese women. Similar to the current study, a study by Mustaqeem *et al.* [36] found that irregular menstrual periods were experienced by 64.44% of individuals with increased WHR.

CONCLUSION

This study advances our knowledge of the relationship between obesity and menstrual problems in women. Retaining a healthy weight helps reduce the likelihood of irregular menstruation and the gynecologic issues associated with obesity.

ACKNOWLEDGEMENT

I was appreciative of the participants' assistance and cooperation throughout my work. The Department of Anthropology at Cotton University provided extensive assistance and collaboration for this project, which was gratefully recognized.





Chumi Das et al.,

REFERENCES

1. Wang Y, Chen HJ, Shaikh S et al., Is obesity becoming a public health problem in India? Examine the shift from under-to over nutrition problems over time. *Obesity Reviews*. 2009;10(4):456-74.
2. Popkin BM, Adair LS, Ng SW. Global nutrition transition and the pandemic of obesity in developing countries. *Nutrition reviews*. 2012;1;70(1):3-21.
3. Wang Y, Zhao L, Gao L et al., Health policy and public health implications of obesity in China. *The lancet Diabetes & endocrinology*. 2021;9(7):446-61.
4. Alice A, Yadav M, Verma R et al., Effect of obesity on balance: A literature review. *International Journal of Health Sciences*. 2022;6(S4):3261-79.
5. Cakmur H. Obesity as a growing public health problem. *Adiposity-epidemiology and treatment modalities*. 2017.
6. Caballero B. Humans against obesity: who will win?. *Advances in nutrition*. 2019;10(suppl_1):S4-9.
7. WHO. Global Strategy on Diet, Physical Activity and Health Solarex.2004.
8. Daniels SR, Arnett DK, Eckel RH et al., Overweight in children and adolescents: pathophysiology, consequences, prevention, and treatment. *Circulation*. 2005;111(15):1999-2012.
9. Kobylak N, Abenavoli L, Falalyeyeva T et al., Prevention of NAFLD development in rats with obesity via the improvement of pro/antioxidant state by cerium dioxide nanoparticles. *Clujul Medical*. 2016;89(2):229.
10. Bixby H, Bentham J, Zhou B et al., Rising rural body-mass index is the main driver of the global obesity epidemic. 2019.
11. Cui G, Liu H, Xu G et al., Exploring links between industrialization, urbanization, and Chinese inflammatory bowel disease. *Frontiers in Medicine*. 2021;8:757025.
12. Grant-Stegehuis N, Simonis J. Industrialization and the environmental exposome: The role in microbial dysbiosis and obesity. *Journal of the Australasian College of Nutritional and Environmental Medicine*. 2023;42(2):18-23.
13. Martins VJ, Toledo Florêncio TM, Grillo LP et al., Long-lasting effects of undernutrition. *International journal of environmental research and public health*. 2011;8(6):1817-46.
14. Abdullah A. The double burden of undernutrition and over nutrition in developing countries: an update. *Current obesity reports*. 2015;4(3):337-49.
15. Gupta AK, Ahmad AJ. Childhood obesity and hypertension. *Indian pediatrics*. 1990;27(4):333-7.
16. Monga S. Obesity among school children (7-9 years old) in India, prevalence and related factors. In *The 132nd Annual Meeting of APHA*. 2004.
17. Drozd D, Alvarez-Pitti J, Wójcik M et al., Obesity and cardio metabolic risk factors: from childhood to adulthood. *Nutrients*. 2021;13(11):4176.
18. Tsoi MF, Li HL, Feng Q et al., Prevalence of childhood obesity in the United States in 1999–2018: A 20-year analysis. *Obesity Facts*. 2022;15(4):560-9.
19. Ni Mhurchu C, Rodgers A, Pan WH et al., Asia Pacific Cohort Studies, Collaboration: Body mass index and cardiovascular disease in the Asia-Pacific Region: an overview of 33 cohorts involving 310 000 participants. *Int J Epidemiol*. 2004;33(4):751-8.
20. Berrington de Gonzalez A, Hartge P et al., Body-mass index and mortality among 1.46 million white adults. *New England Journal of Medicine*. 2010;363(23):2211-9.
21. Zheng W, McLerran DF, Rolland B et al., Association between body-mass index and risk of death in more than 1 million Asians. *New England Journal of Medicine*. 2011;364(8):719-29.
22. Karastergiou K, Smith SR, Greenberg AS et al., Sex differences in human adipose tissues—the biology of pear shape. *Biology of sex differences*. 2012;3(1):1-2.
23. Mansour MF, Chan CW, Laforest S et al., Sex differences in body fat distribution. *Adipose Tissue Biology*. 2017:257-300.
24. Fakh M, Cherfan V, Abdallah E. Success rate, quality of life, and descriptive analysis after generalized endometrial ablation in an obese population. *International Journal of Gynecology & Obstetrics*. 2011;113(2):120-3.
25. Madsen AM, El-Nashar SA, Hopkins MR et al., Endometrial ablation for the treatment of heavy menstrual bleeding in obese women. *International Journal of Gynecology & Obstetrics*. 2013;121(1):20-3.





Chumi Das et al.,

26. Bhavadharini B, Anjana RM, Deepa M et al., Gestational weight gain and pregnancy outcomes in relation to body mass index in Asian Indian women. *Indian journal of endocrinology and metabolism*. 2017;21(4):588.
27. Silvestris E, De Pergola G, Rosania R et al., Obesity as disruptor of the female fertility. *Reproductive Biology and Endocrinology*. 2018;16:1-3.
28. Li G, Zhao D, Wang Q et al., Infertility-related stress and quality of life among infertile women with polycystic ovary syndrome: Does body mass index matter?. *Journal of Psychosomatic Research*. 2022;158:110908.
29. Rafique A, Salma UE, Saleem HG. Measuring the awareness of polycystic ovarian syndrome (PCOS) among women in Punjab, Pakistan. *Sci Inquiry Rev*. 2023;7:1-6.
30. Paulen ME, Zapata LB, Cansino C et al., Contraceptive use among women with a history of bariatric surgery: a systematic review. *Contraception*. 2010;82(1):86-94.
31. Thong EP, Milat F, Joham AE et al., Obesity, menstrual irregularity and polycystic ovary syndrome in young women with type 1 diabetes: a population-based study. *Clinical endocrinology*. 2020;93(5):564-71.
32. Hollmann M, Runnebaum B, Gerhard I. Impact of waist-hip-ratio and body-mass-index on hormonal and metabolic parameters in young, obese women. *International journal of obesity*. 1997;21(6):476-83.
33. Chang PJ, Chen PC, Hsieh CJ et al., Risk factors on the menstrual cycle of healthy Taiwanese college nursing students. *Australian and New Zealand Journal of Obstetrics and Gynaecology*. 2009;49(6):689-94.
34. Tayebi N, Yazdanpanahi Z, Yektatalab S et al., The relationship between body mass index (BMI) and menstrual disorders at different ages of menarche and sex hormones. *Journal of the National Medical Association*. 2018;110(5):440-7.
35. Ciebiera M, Esfandyari S, Siblini H et al., Nutrition in gynecological diseases: current perspectives. *Nutrients*. 2021;13(4):1178.
36. Mustaqeem M, Sadullah S, Waqar W et al., Obesity with irregular menstrual cycle in young girls. *Mymensingh medical journal: MMJ*. 2015;24(1):161-7.
37. Bae J, Park S, Kwon JW. Factors associated with menstrual cycle irregularity and menopause. *BMC women's health*. 2018;18(1):1-1.
38. Collins SC, Martin JR, Pal L. Amenorrhea and Abnormal Uterine Bleeding. *Contemporary Obstetrics and Gynecology for Developing Countries*. 2021:525-41.
39. Joffe H, Hayes FJ. Menstrual cycle dysfunction associated with neurologic and psychiatric disorders: their treatment in adolescents. *Annals of the New York Academy of Sciences*. 2008;1135(1):219-29.
40. Saso S, Chatterjee J, Georgiou E et al., Endometrial cancer. *Bmj*. 2011;6:343.
41. Harris HR, Terry KL. Polycystic ovary syndrome and risk of endometrial, ovarian, and breast cancer: a systematic review. *Fertility research and practice*. 2016;2(1):1-9.
42. Ding S, Madu CO, Lu Y. The impact of hormonal imbalances associated with obesity on the incidence of endometrial cancer in postmenopausal women. *Journal of Cancer*. 2020;11(18):5456.
43. Tang Y, Chen Y, Feng H et al., Is body mass index associated with irregular menstruation: a questionnaire study?. *BMC women's health*. 2020;20(1):1-6.
44. Wei S, Schmidt MD, Dwyer T et al., Obesity and menstrual irregularity: associations with SHBG, testosterone, and insulin. *Obesity*. 2009 May;17(5):1070-6.
45. Lash MM, Armstrong A. Impact of obesity on women's health. *Fertility and sterility*. 2009;91(5):1712-6.
46. Edison E, Whyte M, van Vlymen J et al., Bariatric surgery in obese women of reproductive age improves conditions that underlie fertility and pregnancy outcomes: retrospective cohort study of UK National Bariatric Surgery Registry (NBSR). *Obesity surgery*. 2016;26:2837-42.
47. Xu H, Li PH, Barrow TM et al., Obesity as an effect modifier of the association between menstrual abnormalities and hypertension in young adult women: Results from Project ELEFANT. *PLoS One*. 2018;13(11):e0207929.
48. Sherly DG, Siva Priya D V, Rama Swamy C. Prevalence of menstrual irregularities in correlation with body fat among students of selected colleges in a district of Tamil Nadu, India. *National Journal of Physiology, Pharmacology and Pharmacology*. 2017;7(7):740-743. doi:10.5455/njppp.2017.7.0307422032017
49. Souza FA, Dias R, Fernandes CE et al., Menstrual irregularity: a possible clinical marker of metabolic dysfunction in women with class III obesity. *Gynecological Endocrinology*. 2010 ;26(10):768-72.





Chumi Das et al.,

50. Lakshmanan G, Palanisamy V, Jaishankar L *et al.*, Correlation between body mass index, waist-hip ratio and menstrual abnormalities. *Indian Journal of Science and Technology*. 2017;10(24), 1-5.
51. Liu MM, Liu QJ, Wen J *et al.*, Waist-to-hip ratio is the most relevant obesity index at each phase of insulin secretion among obese patients. *Journal of Diabetes and its Complications*. 2018;32(7):670-6.
52. Ross R, Neeland IJ, Yamashita S *et al.*, Waist circumference as a vital sign in clinical practice: a Consensus Statement from the IAS and ICCR Working Group on Visceral Obesity. *Nature Reviews Endocrinology*. 2020;16(3):177-89.
53. Jacobson MH, Howards PP, Darrow LA *et al.*, Thyroid hormones and menstrual cycle function in a longitudinal cohort of premenopausal women. *Pediatric and perinatal epidemiology*. 2018;32(3):225-34.
54. Amgain K, Subedi P, Yadav GK *et al.*, Association of anthropometric indices with menstrual abnormality among nursing students of Nepal: a cross-sectional study. *Journal of Obesity*. 2022 Mar 18;2022.
55. Kanellakis S, Skoufas E, Simitopoulou E *et al.*, Changes in body weight and body composition during the menstrual cycle. *American Journal of Human Biology*. 2023;35(11):e23951.
56. Duiică F, Dănilă CA, Boboc AE *et al.*, Impact of increased oxidative stress on cardiovascular diseases in women with polycystic ovary syndrome. *Frontiers in Endocrinology*. 2021;12:614679.
57. Praba LJ, Rajeswari GV, Addline D *et al.*, ASSESSMENT OF FEMALE INFERTILITY PROBLEMS BASED ON HORMONAL IMBALANCE IN RAMANATHAPURAM DISTRICT. TAMIL NADU, INDIA. *Journal of Population Therapeutics and Clinical Pharmacology*. 2023;30(7):474-83.
58. Schisterman EF, Gaskins AJ, Mumford SL *et al.*, Influence of endogenous reproductive hormones on F2-isoprostane levels in premenopausal women: theBioCycle Study. *American journal of epidemiology*. 2010;172(4):430-9.
59. Kische H, Gross S, Wallaschofski H *et al.*, Clinical correlates of sex hormones in women: The study of health in Pomerania. *Metabolism*. 2016;65(9):1286-96.
60. Charkoudian N, Hart EC, Barnes JN *et al.*, Autonomic control of body temperature and blood pressure: influences of female sex hormones. *Clinical Autonomic Research*. 2017:149-55.
61. Moretti C, Lanzolla G, Moretti M *et al.*, Androgens and hypertension in men and women: a unifying view. *Current hypertension reports*. 2017;19:1-8.
62. Apgar BS, Kaufman AH, George-Nwogu U *et al.*, Treatment of menorrhagia. *American Family Physician*. 2007;75(12), 1813-1819.
63. Kulie T, Slattengren A, Redmer J *et al.*, Obesity and women's health: an evidence-based review. *The Journal of the American Board of Family Medicine*. 2011;24(1):75-85.
64. Nichols AR, Chavarro JE, Oken E. Reproductive risk factors across the female life course and later metabolic health. *Cell Metabolism*. 2024.
65. AbdElmenim SO, Emam AM. Effect of lifestyle changes on symptoms of polycystic ovarian syndrome in obese girls. *IOSR JNHS*. 2016;5(3):1-0.
66. Shen SY, Huang SY, Hsieh CH *et al.*, Clinical and biochemical characteristics of women with menstrual disturbance. *Taiwanese Journal of Obstetrics and Gynecology*. 2014;53(2):178-82.
67. Klein DA, Paradise SL, Reeder RM. Amenorrhea: a systematic approach to diagnosis and management. *American family physician*. 2019;100(1):39-48.
68. Shim U, Oh JY, Lee HJ *et al.*, Long menstrual cycle is associated with type 2 diabetes mellitus in Korean women. *Diabetes & metabolism journal*. 2011;35(4):384-9.
69. Ko KM, Han K, Chung YJ *et al.*, Association between body weight changes and menstrual irregularity: the Korea National Health and Nutrition Examination Survey 2010 to 2012. *Endocrinology and Metabolism*. 2017;32(2):248-56.
70. Itriyeva K. The effects of obesity on the menstrual cycle. *Current Problems in Pediatric and Adolescent Health Care*. 2022:101241.
71. Jalil PJ, Shnawa BH, Ahmed MH. Association of free testosterone, glucose level and obesity among women with polycystic ovary syndrome in Soran city, Kurdistan-Iraq. *Clínica e Investigación en Ginecología y Obstetricia*. 2023;50(4):100903.
72. Carlos Poston WS, Foreyt JP. Infertility and obesity: The effects of behavioral therapy, diet, and exercise. *INFERTILITY AND REPRODUCTIVE MEDICINE CLINICS OF NORTH AMERICA*. 2000;11(2):315-28.





Chumi Das et al.,

73. Castillo-Martínez L, López-Alvarenga JC, Villa AR et al., Menstrual cycle length disorders in 18-to 40-y-old obese women. *Nutrition*. 2003;19(4):317-20.
74. Sakumoto T, Tokunaga Y, Tanaka H et al., Insulin resistance/hyperinsulinemia and reproductive disorders in infertile women. *Reproductive medicine and biology*. 2010;9:185-90.
75. Zhang D, Yang X, Li J et al., Effect of hyperinsulinaemia and insulin resistance on endocrine, metabolic and fertility outcomes in women with polycystic ovary syndrome undergoing ovulation induction. *Clinical Endocrinology*. 2019;91(3), 440-448.
76. Abruzzese GA, Silva AF, Velazquez ME et al., Hyperandrogenism and Polycystic ovary syndrome: Effects in pregnancy and offspring development. *WIREs Mechanisms of Disease*. 2022;14(5):e1558.
77. Seif MW, Diamond K, Nickkho-Amiry M. Obesity and menstrual disorders. *Best Practice & Research Clinical Obstetrics & Gynecology*. 2015;29(4):516-27.
78. Chosich J, Bradford AP, Allshouse AA et al., Acute recapitulation of the hyperinsulinemia and hyperlipidemia characteristic of metabolic syndrome suppresses gonadotropins. *Obesity*. 2017;25(3):553-60.
79. Ding H, Zhang J, Zhang F et al., Resistance to the insulin and elevated level of androgen: a major cause of polycystic ovary syndrome. *Frontiers in endocrinology*. 2021;12:741764.
80. Maheshwari N, Hassan T, Rashid G. PCOS and the Rhythmicity of GnRH: The Impact of FSH and LH on Menstrual Health. *Obstetrics and Gynecology Advances*. 2023:27.
81. Sheela J, Sunita G, Savita S et al., Association of body mass index with menstrual cycle irregularities in women between 16–40 year age. *PJMS*. 2014;4(1):59-61.
82. Ziolkiewicz A, Ellison PT, Lipson SF et al., Body fat, energy balance and estradiol levels: a study based on hormonal profiles from complete menstrual cycles. *Human Reproduction*. 2008;23(11):2555-63.
83. Mohamed AG, Hables RM. Menstrual Profile and Body Mass Index among Female University Students. *American Journal of Nursing*. 2019;7(3):360-4.
84. Zohora TS, Shila SM, Khanam R. A study on correlation between menstrual cycle irregularities and BMI among residential female students of MawlanaBhashani Science and Technology University, Santosh, Tangail. *J Pharm Drug Res*. 2021;4(01):470-9.

Table I: Anthropometric characteristics of obese women with and without regular menstrual cycles

Variables	Regular Menstrual Cycle (n=66)	Oligomenorrhea (n=22)	Amennorrhea (12)	p* value (Analysis of variance)
Age (years)	29.82±5.63	28.86±5.87	30.08±5.81	0.76850
Height (cm)	154.66±6.45	153.84±5.91	152.06±6.70	0.00004
Weight (kg)	76.15±7.19	79.84±6.60	80.83±8.86	0.03421
BMI (kg/m ²)	31.77±1.50	33.69±1.32	34.92±1.32	0.42630
Waist circumference (cm)	80.65±4.62	88.81±6.02	92.88±6.84	< 0.00001
Hip circumference (cm)	91.54±4.94	94.42±8.37	98.58±9.95	0.00285
Waist Hip Ratio (kg/m ²)	0.88±0.03	0.89±0.06	0.89±0.07	0.55703
Blood Pressure Diastolic (mm Hg)	84.69±8.93	82.05±10.24	87.58±15.58	0.31999
Blood Pressure Systolic (mm Hg)	120.39±11.69	117.59±15.83	119.08±18.52	0.70738
Age at Menarche (Years)	12.59±1.21	12.27±1.42	11.58±1.38	0.00452





Chumi Das et al.,

Table II. : Biochemical markers of obese women with and without regular menstrual cycles

Variables	Regular Menstrual Cycle (n=66)	Oligomenorrhea (n=22)	Amenorrhea (12)	p value (Analysis of variance)
Glucose (mg/dL)	91.53±17.40	101.41±21.09	104.67±18.33	0.02236
Triglycerides (mg/dL)	118.21±20.04	124.42±32.07	135.75±36.17	0.04609
HDL Cholesterol (mg/dL)	54.14±8.84	50.55±10.29	55.08±12.24	0.27761
LDL Cholesterol (mg/dL)	120.42±4.94	117.73±20.94	124.83±28.40	0.65130
Total Cholesterol (mg/dL)	180.92±22.06	178.95±23.93	182.58±33.12	0.90983
Insulin (µU/dL)	14.69±6.43	17.10±8.97	22.06±10.86	0.01073

Table III. Spearman correlation coefficient of obesity with menstrual irregularities

Variables	Oligomenorrhea (22)		Amenorrhea (12)	
	R-value	p-value	R-value	p-value
Weight (kg)	0.67988	0.00050	0.83363	0.00075
Waist circumference(cm)	0.06563	0.77168	0.75657	0.00440
Hip circumference(cm)	0.04980	0.82580	0.75657	0.00440
WHR	0.42374	0.04939	0.57701	0.04950
Diastolic BP (mm of Hg)	0.06423	0.77642	0.74211	0.00572
Systolic BP (mm of Hg)	0.08871	0.69462	0.77193	0.00327





Influence of Poultry Manure Compost and Foliar Application of Seaweed Extract on the Productivity and Profitability of Finger Millet (*Eleusine coracana* L.)

S.Ramesh^{1*}, G. Rama Prabavathi², P.Sudhakar¹, G. Baradhan¹ and C. Kalaiyarasan¹

¹Associate Professor, Faculty of Agriculture, Department of Agronomy, Annamalai University, Annamalai Nagar, Tamil Nadu, India.

²Ph.D Scholar, Faculty of Agriculture, Department of Agronomy, Annamalai University, Annamalai Nagar, Tamil Nadu, India.

Received: 21 Jun 2024

Revised: 03 Jul 2024

Accepted: 26 Oct 2024

*Address for Correspondence

S.Ramesh

Associate Professor,
Faculty of Agriculture, Department of Agronomy,
Annamalai University, Annamalai Nagar,
Tamil Nadu, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Field experiment was conducted at Puliyanthoppu village in Krishnagiri district, Tamil Nadu, from December 2019 to April 2020 to study the Influence of poultry manure compost and foliar application of seaweed extract on the productivity and profitability of finger millet (*Eleusine coracana* L.) under irrigated conditions. The experiment followed a split-plot design with three replications. According to the experimental results, the application of enriched poultry manure compost @ 750 kg ha⁻¹ combined with foliar spray of seaweed extract @ 0.3% significantly recorded growth characteristics, yield attributes, and grain yield. This treatment also registered higher gross return of Rs. 91,019 ha⁻¹, net return of Rs. 53,777 ha⁻¹, and B:C ratio of 2.44. Conversely, the least growth and yield attributes, as well as the lowest gross return, net return, and B:C ratio, were observed with the application of enriched FYM @ 750 kg ha⁻¹ along with water spray.

Key words: Enriched poultry manure compost, Seaweed extract, grain yield, gross return, net return and BCR

INTRODUCTION

Finger millet (*Eleusine coracana* (L.) Gaertn.) Ranks as the third most important millet in India, following sorghum and pearl millet. It stands out among major food grains due to its high nutritional value, particularly in protein and minerals such as calcium and iron, offering 8–10 times more calcium than wheat or rice. Its slow digestibility makes it an excellent food crop for pregnant women and individuals with diabetes. Finger millet accounts for about 85% of total millet production in India, covering 11.9 lakh hectares and yielding 19.8 lakh tonnes with a productivity of 1662 kg per hectare. In Tamil Nadu, it is grown on 0.78 lakh hectares, producing 2.56 lakh tonnes with a productivity of



**Ramesh et al.,**

1966 kg per hectare. Nutrient management is a crucial factor in enhancing the productivity of finger millet. While the application of chemical fertilizers can significantly boost crop yields, it often has a detrimental effect on soil properties. Therefore, an integrated approach to nutrient management is essential to prevent nutrient depletion, maintain soil fertility, and sustain crop productivity. Organic amendments, such as farmyard manure, poultry manure, and pressmud compost, positively impact soil fertility, improving soil condition and plant growth.

Poultry manure holds a prominent position due to its higher nutrient content compared to other manures. (Adeleye *et al.*, 2010). The application of farmyard manure (FYM) enhances soil fertility by improving its physical condition and water holding capacity. FYM not only supplies macronutrients but also meets micronutrient requirements, thereby improving soil health (Jadhao *et al.*, 2019). Pressmud is a valuable resource of plant nutrients and improves the physical, chemical, and biological properties of the soil, leading to increased crop yields (Ramesh, 2018). Foliar application of liquid organic manures influences a wide range of physiological parameters, including altered plant structure, assimilate partitioning, increased nutrient uptake, improved nitrogen metabolism, accelerated blooming, grain filling, and enhanced assimilate mobilization to designated sinks (Sharma Sardana and Sukhvinder Singh, 2013).

To provide additional nutrients, developing liquid organic biostimulants such as seaweed extract and humic acid for foliar application is essential. These biostimulants can supply necessary nutrients during critical periods of crop growth and possess insecticidal or fungicidal properties. Seaweed extracts are marketed as liquid fertilizers and biostimulants due to their high content of growth regulators like cytokinins and gibberellins. Alongside these growth regulators, seaweed extracts provide a wide range of macro and micronutrients essential for optimal plant growth and development (Khan *et al.*, 2009). Humic compounds enhance soil structure, promote the growth of soil microbes, increase the cation exchange capacity of the soil, and indirectly benefit plant roots by supplying essential macro and micronutrients, thus improving soil fertility (Guo *et al.*, 2009). While there is extensive research on the use of organic manures and foliar application of biostimulants in finger millet individually, studies on the combined application of organic manures—particularly enriched organic compost—and liquid organic manures through foliar application in irrigated finger millet are limited. Therefore, this study aimed to develop integrated nutrient management strategies for irrigated finger millet that would be profitable, productive, and sustainable for farmers in the Krishnagiri District of Tamil Nadu, India.

MATERIALS AND METHODS

Experimental site

Field experiment was performed in a farmer's field at Puliyanthoppu village, Krishnagiri District, Tamil Nadu from December 2019 to April 2020 (Marghazipattam). The field site was geographically located at an altitude of 492 m above mean sea level, at 11°12' to 12°49' North latitude and 77°27' to 78°38' East longitude. The weather at Puliyanthoppu village was dry, with scanty rainfall except during the monsoon season. The mean annual rainfall received was 830 mm. During the farming season, the maximum temperature is 34.24 °C, the minimum temperature is 22.28 °C and the relative humidity is 75.50%. The experimental soil has a clay loam texture, pH of 7.1, organic carbon content of 0.58%, and contains nitrogen (214.96 kg/ha), phosphorus (21.37 kg/ha), and potassium (275.28 kg/ha).

METHODOLOGY

The experiment comprised of three main plot treatments *viz.*, M₁- Enriched FYM @ 750 kg ha⁻¹, M₂- Enriched pressmud compost @ 750 kg ha⁻¹, M₃- Enriched Poultry manure compost @ 750 kg ha⁻¹ and five subplot treatments namely, S₁- Control (Water spray), S₂- Foliar spray of humic acid @ 0.3%, S₃-Foliar spray of panchagavya @ 3%, S₄-Foliar spray of Vermiwash @ 5%, S₅-Foliar spray of seaweed extract 0.3 % (All the foliar sprays were given at 20,40 and 60 DAT). The experiment with different treatments was tested in the field in a split-plot design with three





Ramesh *et al.*,

replications. In this study, composted manures such as FYM, pressmud, and poultry manure were used. Enriched FYM compost was prepared using the heap method with the following procedure: 750 kg of FYM (dry-weight basis) was thoroughly mixed with single superphosphate (187.5 kg ha⁻¹), Azospirillum (10 kg), and Phosphobacteria (10 kg). The mixture was kept in a shaded area with 60% moisture. After two months, the enriched FYM compost was ready and applied to the designated plots according to the treatment schedule prior to transplanting. This same method was used to prepare enriched compost from poultry manure and pressmud. Promising CO 15 (120 days) finger millet variety was used in the study. Two seedlings per hill were transplanted at the age of 18 days and the spacing adopted was 30 x 10 cm. A fertilizer schedule of 60 kg N, 30 kg P₂O₅, and 30 kg K₂O per hectare was implemented. Nitrogen, phosphorus, and potassium were supplied through urea, single superphosphate, and muriate of potash, respectively. All other improved recommended practices outlined in the Crop Production Guide (2019) were followed for cultivating finger millet.

Data collection

Five plants were randomly chosen from each net plot area and tagged for biometric observation at various crop growth phases.

Plant height

The plant height was measured from the ground level to the tip of the top most leaf/ ear head at harvest stage.

Root length and root volume

The entire finger millet plant was uprooted at flowering stage before harvesting and roots were washed. The root length was measured from the base of the plant to the root tip and expressed in cm. After measuring root length, the roots were separated from the plants and were washed with water and the root volume measured by water displacement method (Karthikeyan, 1999) and expressed in cubic centimetre (cc).

Crop growth rate (CGR)

The CGR explains the dry matter accumulated per unit land area per unit time, expressed as g m⁻² day⁻¹. It was calculated by using the following formula as suggested by Watson (1958).

$$\text{CGR} = \frac{W_2 - W_1}{(t_2 - t_1) S} \text{ g / m}^2 \text{ / day}$$

Where,

W₁, W₂ - Plant dry weight (g) at time t₁, t₂, S – land area (m²)

Number of fingers ear head⁻¹

At the time of harvest, five matured ear heads were selected at random from the representative plant samples. Number of fingers in each ear head was counted and the mean value was expressed as number of fingers ear head⁻¹.

Length of ear head (cm)

Ear head length was measured from the point of rear to the tip of the ear head obtained from five primary ear head of the tagged plants and the mean length for ear head was calculated and expressed in cm.

Grain yield

Harvesting was done in each plot separately from the net plot area and grains were separated, cleaned and grain yield was recorded plot wise at 14 per cent moisture content. The yield was computed to kg ha⁻¹.

Economic analysis

The economic parameters such as gross income, net income and benefit cost ratio for all the treatments were worked out based on the prevailing market price. The net income was worked out for different treatments by subtracting the cost of cultivation from gross income. The benefit cost ratio was calculated by using the following formula.





Ramesh et al.,

$$\text{BCR} = \frac{\text{Gross income (Rs.ha}^{-1}\text{)}}{\text{Cost of cultivation (Rs.ha}^{-1}\text{)}}$$

Statistical analysis

The data on the numerous characters evaluated throughout the research study were statistically analyzed. To make the statistical inferences, the critical differences were calculated at a 0.05 per cent probability level (Panse and Sukhatme, 1989).

RESULT AND DISCUSSION

Growth attributes

Among different growth attributing characters, enriched poultry manure compost @ 750 kg ha⁻¹(M₃) registered significantly maximum values of plant height of 103.64 cm, root length of 24.93 cm, root volume of 10.82 cc, LAI of 5.71 and crop growth rate of 11.24 g m⁻² d⁻¹ at 30-60 DAT and 8.53 g m⁻² d⁻¹ at 60 - harvest stage (Table 1.). These could be attributed to an adequate supply of nutrients that caused the crops to grow quickly, which in turn increased various metabolic processes, improved the mobilization of synthesized carbohydrates in amino acids and proteins, and consequently promoted cell division and elongation, leading to higher growth attributes. The results are in agreement with the findings of Abdul Nasir *et al.*, (2010) and Enujeke (2013).

The least plant height of 92.36 cm, root volume of 7.77cc, root length of 20.56, LAI of 3.83 and crop growth rate of 8.39 g m⁻² d⁻¹ at 30-60 DAT and 6.31 g m⁻² d⁻¹ at 60- harvest were recorded under enriched FYM compost @ 750 kg ha⁻¹ (M₁). With regard to foliar nutrition, seaweed extract @ 0.3% on 20, 40 and 60 DAT (S₅) registered highest plant height of 103.52 cm, root length of 24.92 m, root volume of 10.77 cc, LAI of 5.68 and crop growth rate of 11.18 and 8.29 g m⁻² d⁻¹ at 30-60 DAT and 60 DAT- harvest, respectively. These might be due to availability of plant nutrients and growth regulators in seaweed extract, which helps to enhance the better root development and enzyme activity in the root portion resulting in higher growth attributes registered under this treatment. The results are in concomitance with the findings of Rathore *et al.*, (2009); Liu and Lijun (2011). The least plant height of 88.87 cm, root length of 18.88 cm, root volume of 6.61 cc, LAI of 3.39 and crop growth rate of 8.58 and 6.48 g m⁻² d⁻¹ at 30-60 DAT and 60 DAT- harvest, respectively was recorded under control water spray (S₁). Significant interactions were observed between enriched compost and foliar spray. The treatment combination of enriched poultry manure compost @ 750 kg ha⁻¹ and seaweed extract @ 0.3% (M₃S₅) registered maximum values of plant height, root length, root volume and crop growth rate at 30-60 DAT and 60 DAT- harvest. This could be due to combined effect of composted poultry manure and foliar application of seaweed extract. The least growth characters were recorded under enriched FYM compost @ 750 kg ha⁻¹ followed by water spray (M₁S₁).

Yield attributes and yield

Among different yield attributing characters, enriched poultry manure compost @ 750 kg ha⁻¹ (M₃) registered significantly higher number of fingers earhead⁻¹ of 8.08, length of fingers of 8.98 (Table 2) and grain yield of 3504 kg ha⁻¹. These could be due to the poultry compost contains uric acids, which speeds up the release of nutrients and may have resulted in more extensive root development, so that crop can able to absorb more nutrients from the soil, and results in quicker conversion of synthesised photosynthates into protein to form more protoplasm thus led to increased yield attributes and yield. These results were concomitant with the finding of Agbede *et al.*, (2008). The least yield attributes and yield were registered under the treatment enriched FYM compost @ 750 kg ha⁻¹ (M₁). In respect of foliar nutrition, higher number of fingers ear head⁻¹ of 8.05, length of fingers of 8.93cm and grain yield of 3567 kg ha⁻¹ were obtained with the foliar spray of seaweed extract @ 0.3% on 20, 40 and 60 DAT (S₅). The balanced growth pattern may have reduced flower shedding, which on the other hand resulted in a positive source-sink gradient of photosynthates translocation due to growth regulator and also increases the flower production by encouraging timely nutrient supply by foliar spray. Similar observations were reported by Diviya and Kalyani (2016).





Ramesh *et al.*,

The least number of fingers ear head⁻¹ of 4.36, length of ear head of 5.21 and grain yield of 2389 kg ha⁻¹ were registered with water spray (S₁).

The interaction effect between enriched compost and foliar nutrients were found to be significant. Application of enriched poultry manure compost @ 750 kg ha⁻¹ followed by foliar spray of seaweed extract @ 0.3% on 20, 40 and 60 DAT (M₃S₅) registered the maximum yield attributes *viz.*, number of fingers ear head⁻¹, finger length and grain yield. The least yield characters were recorded under enriched FYM compost @ 750 kg ha⁻¹ followed by water spray (M₁S₁). In respect of harvest index significantly higher values registered under plots received with enriched poultry manure compost @ 750 kg ha⁻¹ followed by foliar spray of seaweed extract @ 0.3% on 20, 40 and 60 DAT (M₃S₅).

Economics

Among the treatment combination of organic manure and foliar nutrition on irrigated finger millet, the maximum gross returns of Rs. 91019 ha⁻¹, net returns of Rs. 53777 ha⁻¹ and BCR of 2.44 were obtained in plots applied with enriched poultry manure compost @ 750 kg ha⁻¹ along with foliar spray of seaweed extract @ 0.3% on 20, 40 and 60 DAT (M₃S₅) (Table 3). The lowest gross returns of Rs. 50233 ha⁻¹, net return of Rs.17041 ha⁻¹ and BCR invested 1.51 were recorded in M₁S₁ (enriched FYM compost @ 750 kg ha⁻¹ along with water spray). Increased profitability in poultry manure compost along with foliar application of seaweed extract could be attributed to finger millet utilized the both below and above the ground available resources very effectively thereby resulted higher grain yield, gross and net returns in irrigated finger millet cultivation (Naidu *et al.*, 2009).

CONCLUSION

Based on the results of the experiment, it can be concluded that application of enriched poultry manure compost @ 750 kg ha⁻¹ followed by foliar spray of 0.3% seaweed extract on 20, 40, and 60 days after transplanting (DAT) to achieve higher productivity and profitability in finger millet cultivation under irrigated conditions.

REFERENCES

1. Abdul Nasir, A., F.H. Khan, Mahmood Riaz and M.A. Khan. (2010). Comparative study of biogas slurry with farm yard manure as fertilizer on maize crop. *Sci. Int.*, 22(4): 297-301.
2. Adeleye, E.O., L.S. Ayeni and S.O. Ojeniyi. (2010). Effect of poultry manure on soil physico-chemical properties, leaf nutrient contents and yield of yam (*Dioscorea rotundata*) on Alfisol in South western Nigeria. *J. of American Sci.*, 6(10): 871-878.
3. Agbede, T.M., S.O. Ojeniyi and A.J. Adeyemo. (2008). Effect of poultry manure on soil physical and chemical properties, growth and grain yield of sorghum in southwest, Nigeria. *American-Eurasian J. Sustainable Agric.*, 2(1): 72-77.
4. Crop production guide. Published by Directorate of Agriculture, Chennai and Tamilnadu. Agriculture University, Coimbatore; (2019) Available from: <https://agitech.tnau.ac.in/pdf/AGRICULTURE.pdf>
5. Diviya, K. and P. Kalyani. (2016). Influence of seaweed extract of *Gracilaria textorii* and *Tubenariabarnata* on the germination, growth and yield of some vegetable crops. *Int. J. Pure App. Biosci.* 4(5):42-47.
6. Enujeke, E.C. (2013). Effects of poultry manure on growth and yield of improved maize in Asaba are of delta state, Nigeria. *J. Agri. Vet. Sci.*, 4: 24-30.
7. Guo XX, Liu HT, Wu SB. (2009) Humic substances developed during organic waste composting: formation mechanisms, structural properties, and agronomic functions. *Science of the Total Environment.*;662:501–510.
8. Jadhao SD, Mali VD, Sonune AB. (2019) Impact of continuous manuring and fertilization on change in soil quality under sorghum – wheat sequence on a vertisols. *Journal of the Indian society of soil science.*; 67(1):55-64
9. Karthikeyan. R. (1999). Agronomical management for thaladi season rice. M.Sc., (Ag.) Thesis, Tamil Nadu Agric. Univ., Coimbatore.



Ramesh *et al.*,

10. Khan W, Rayirath UP, Subramanian S, Jithesh MN, Rayorath P, Hodges DM, Critchley AT, Raigie JS, Norrie J, Prithiviraj B. (2009) Seaweed extracts as biostimulants of plant growth and development. *Journal of Plant Growth Regulation.* ;28:386-399.
11. Liu, Z. and L. Lijun. (2011). Effects of Plant growth regulators and saccharide on in vitro plant and tuberous root regeneration of Cassava. *J. Plant Growth Reg.* (30)1: 11-19.
12. Naidu, K.D., B.M. Radder, P.L. Patil, N.S. Hebsur and S.C. Alagundagi, S.C. (2009). Nutrient uptake and residual fertility of chilli (Cv. byadgidabbi) in a Vertisols. *Karnataka J. of Agric. Sci.*, 22 (2): 306-309
13. Panse, V.G and P.V. Sukhatme. (1989). *Statistical methods for agricultural workers.* (Revised by: Sukhatme, P.V. and V.N. Amble). Indian Council for Agricultural Research, New Delhi, pp. 100-109.
14. Ramesh. S (2018). Grain yield, nutrient uptake and nitrogen use efficiency as influenced by different sources of vermicompost and fertilizer nitrogen in rice. *Journal of Pharmacognosy and Phytochemistry.*; 7(5): 52-55.
15. Rathore, S. S., D.R. Chaudhary, G.N. Boricha, A. Ghosh, B.P. Bhat, S.T. Zodape and J.S. Patolia. (2009). Effect of seaweed extract on the growth, yield and quality of soyabean (*Glycine max*) under rain fed condition. *South African J. Bot.*, 75:351-355.
16. Sharma Sardana, P. and K. Sukhvinder Singh. 2013. Dry matter partitioning and source–sink relationship as influenced by foliar sprays in groundnut. *The Bioscan*, 8: 1171–1176.
17. Watson DJ. (1958). The dependence of crop growth rate on plant dry weight. *Ann. Bot.*; 23:37-54.

Table 1. Effect poultry compost and foliar application of biostimulants on root length and root volume and growth analysis in finger millet

TREATMENTS	Plant height, root length and root volume			Growth analysis		
	Plant height (cm)	Root length (cm)	Root volume (cc)	LAI	CGR (30-60 DAT)	CGR (60-Harvest)
MAIN PLOTS						
M ₁	92.36	20.56	7.77	3.83	9.12	6.65
M ₂	97.12	22.37	9.07	4.77	10.12	7.37
M ₃	103.64	24.93	10.82	5.71	11.24	8.53
S.Ed.	1.14	0.28	0.12	0.06	0.12	0.09
C.D (P=0.05)	3.27	0.79	0.35	0.19	0.36	0.27
SUB PLOTS						
S ₁	88.87	18.88	6.61	3.29	8.58	6.48
S ₂	100.16	23.67	9.92	5.20	10.70	7.86
S ₃	97.89	22.69	9.36	4.79	10.19	7.47
S ₄	98.19	22.95	9.44	4.88	10.29	7.46
S ₅	103.52	24.92	10.77	5.68	11.18	8.29
S.Ed.	0.88	0.20	0.08	0.04	0.09	0.06
C.D (P=0.05)	1.83	0.43	0.18	0.09	0.19	0.14

Main plot treatments: M₁- Enriched FYM @ 750 kg ha⁻¹, M₂- Enriched pressmud compost @ 750 kg ha⁻¹, M₃- Enriched Poultry manure compost @ 750 kg ha⁻¹

Sub plot treatments: S₁- Control (Water spray), S₂- Foliar spray of humic acid @ 0.3%, S₃-Foliar spray of panchagavya @ 3%, S₄- Foliar spray of vermiwash @ 5%, S₅-Foliar spray of seaweed extract (All the foliar sprays were given at 20,40 and 60 DAT).





Ramesh et al.,

Table 2. Effect poultry compost and foliar application of biostimulants on yield attribute, grain and straw yields (kg ha⁻¹) in finger millet

TREATMENTS	Yield attributes			Grain and straw yield (kg ha ⁻¹)		Harvest Index
	Number of fingers ear head ⁻¹	Fingers length (cm)	Number of grains ear head ⁻¹	Grain yield (kg ha ⁻¹)	Straw yield (kg ha ⁻¹)	
MAIN PLOTS						
M ₁	5.50	6.20	1092	2689	5467	32.88
M ₂	6.51	7.43	1264	3098	6058	33.75
M ₃	8.08	8.98	1480	3504	6597	34.68
S.Ed.	0.09	0.10	17	73	74	0.37
C.D (P=0.05)	0.26	0.29	48	158	212	0.96
SUB PLOTS						
S ₁	4.36	5.21	950	2389	4968	32.42
S ₂	7.27	8.19	1375	3316	6347	34.26
S ₃	6.87	7.63	1290	3090	6079	33.62
S ₄	6.95	7.72	1307	3139	6134	33.78
S ₅	8.05	8.93	1472	3567	6675	34.78
S.Ed.	0.06	0.07	12	37	56	0.23
C.D (P=0.05)	0.13	0.14	24	83	119	0.48

Main plot treatments: M₁- Enriched FYM @ 750 kg ha⁻¹, M₂- Enriched pressmud compost @ 750 kg ha⁻¹, M₃- Enriched Poultry manure compost @ 750 kg ha⁻¹

Sub plot treatments: S₁- Control (Water spray), S₂- Foliar spray of humic acid @ 0.3%, S₃-Foliar spray of panchagavya @ 3%, S₄- Foliar spray of vermiwash @ 5%, S₅-Foliar spray of seaweed extract (All the foliar sprays were given at 20,40 and 60 DAT).

Table 3. Effect poultry manure compost and foliar application of biostimulants on net return (Rs.ha⁻¹) and BCR in finger millet

Treatments	Cost of cultivation	Gross return (Rs.ha ⁻¹)	Net return (Rs.ha ⁻¹)	BCR
M ₁ S ₁	33192	50233	17041	1.51
M ₁ S ₂	37512	66862	29350	1.78
M ₁ S ₃	39192	59801	20609	1.53
M ₁ S ₄	38142	61534	23392	1.61
M ₁ S ₅	37242	71222	33980	1.91
M ₂ S ₁	33192	54543	21351	1.64
M ₂ S ₂	37512	74764	37252	1.99
M ₂ S ₃	39192	71620	32428	1.83
M ₂ S ₄	38142	72092	33950	1.89
M ₂ S ₅	37242	82493	45251	2.22
M ₃ S ₁	33192	60509	27317	1.82
M ₃ S ₂	37512	86328	48816	2.30
M ₃ S ₃	39192	81458	42266	2.08
M ₃ S ₄	38142	82558	44416	2.16
M ₃ S ₅	37242	91019	53777	2.44





Sustainable Agriculture: A Road to Global Food Security

Ankita Singh¹, Ananya Chamola² and Chaitali Ghosh^{3*}

¹Student, Department of Life Sciences, Gargi College, University of Delhi, New Delhi, India.

²Student, Department of Botany, Gargi College, University of Delhi, New Delhi, India.

³Assistant Professor, Department of Zoology, Gargi College, University of Delhi, New Delhi, India.

Received: 20 Jun 2024

Revised: 20 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

Chaitali Ghosh

Assistant Professor,
Department of Zoology,
Gargi College, University of Delhi,
New Delhi, India.
E.Mail: chaitali.ghosh@gargi.du.ac.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

For decades, farmers and scientists have been trying to increase agricultural output in the traditional way. However, agricultural development is hindered by various aspects. Inefficient fertilizer application leads to a vicious cycle of increased fertilizer application and environmental pollution. Diseases caused by bacteria, fungi, pests and viruses reduce the grain quality and yield. Various abiotic stress environments produced by anthropogenic and natural conditions inhibit the growth of plants. With an ever-increasing global population, some crucial measures need to be taken in order to maintain agriculture for global food security. An innovative solution that is both highly efficient and low in pollution is required to address this agricultural conundrum. Sustainable agriculture is crucial in this situation. The term "sustainable agriculture" refers to a broad range of methods. Sustainable agriculture is farming that maximizes the use of non-renewable resources while preserving the environment and assisting in the expansion of natural resources. The present review paper is written with the motive to identify the various sustainable agricultural practices across the world to provide an efficient and easy way to understand sustainable agriculture methods and their application in current times.

Keywords: Sustainable, agriculture, organic farming, green synthesis

INTRODUCTION

One of the biggest problems of the modern era is providing food for everyone on the planet in a safe and sustainable manner. According to the United Nations report, the population of the world will reach about 8.5 billion by 2030, and then 50% more food will be required to meet the population demand (Singh et al., 2019). Consequently, increasing agricultural output and agronomic practices are crucial for ensuring the world's food security. Sustainable



**Ankita Singh et al.,**

agriculture aims to preserve the planet's capacity to support future generations while producing resources required for the current human population. There are many negative aspects of modern agriculture, such as threats to human health, loss of agricultural diversity, devastation of ecosystems resulting in the loss of biodiversity, and several animal welfare issues. Sustainable agriculture tackles these problems and places an emphasis on "planetary health," which holds that human well-being is correlated with the stability of the earth. Promoting socio economic fairness, making a profit, and preserving the health of the ecology are among its core values. Sustainable agriculture actively works to preserve and promote biodiversity because it has been a major factor in the mass extinction of plant and animal species on Earth. Modern agricultural techniques, which were founded on the green revolution, clearly increased food productivity, but at the expense of depleting natural resources. The externalization of agriculture resulted in a significant reduction in both environmental resilience and soil fertility. It requires a distinct strategy that should encourage farmers to apply their traditional expertise in order to produce more grains with the least number of outside inputs. This strategy is called sustainable agriculture, and it is currently desperately needed. Various nanomaterials are being used in farming in recent years which not only are eco-friendly in nature but also offer a good amount of stress tolerance (Arora et al., 2022). The integration of historically adopted healthful practices with contemporary agricultural system development is the source of sustainable agriculture practices. Thus, resource conservation and climate change resistance are key components of sustainable agriculture operations. The use of nano-sensors to detect nutrient deficiencies in soil and new agricultural technologies are further going to help farmers shift towards sustainable agriculture (Jha et al., 2023). Moreover, it is plausible that a greater proportion of traditional inputs, resources, or expertise may lead to a socioeconomic balance across diverse cultural groups.

Goals of sustainable agriculture

Sustainable agriculture aims to improve environmental quality, maintain economic viability, and meet human requirements for food, feed, and fiber. This demonstrates how sustainable agriculture takes into account social, economic, and environmental concerns all at once. The expectations of present and future generations (the sustainability's time horizon and capacity to persist over time), as well as social, entrepreneurial, and individual demands from farms, are just a few of the numerous factors that need to be taken into account in addition to the sustainability characteristics listed above. Any agricultural system will be deemed more sustainable if it can accomplish more objectives. Sustainability is therefore more than just a goal that has to be accomplished. Therefore, moving toward greater sustainability entails locating a community that will best fulfill the many objectives as a whole. Its execution is challenging, ambitious, and hampered by a number of issues. In order to achieve widespread adoption of sustainable agriculture, we have to make an effort to eliminate or minimize these barriers and bridge the implementation gap. Some of the most widely used sustainable agricultural practices are discussed further in this review article.

Organic farming

The main prediction for organic farming is the discontinuation of synthetic inputs and their substitution with organic alternatives, such as the application of organic manures and natural plant protection techniques rather than synthetic pesticides and fertilizers. However, Bhattacharyya and Chakraborty (2005) refute this by stating due to the nutritional security we must find a middle ground between organic and conventional farming. But organic farming is more than just eliminating chemicals from the soil. It is an all-encompassing strategy for enhancing the plant's health and the underlying productivity of the soil, which leads to the enrichment of the surrounding ecosystem and is a prerequisite for sustainable agriculture. According to the International Federation of Organic Agriculture Movements, "Organic agriculture is a production system that sustains the health of soils, ecosystems and people". Organic farming also permits water reusability which furthermore helps in preventing water contamination (Parizad S and Bera S. 2023). It also involves the farming of trap crops which helps to increase the water retention rate leading to a decrease in water consumption for agriculture purposes (Hassanali et al. 2008). Different soil organisms and living organic matter can improve the soil's ability to store nutrients and decrease the likelihood that these nutrients will be transported to surface or ground waters. Eventually, the processes that agricultural fields use to store nutrients will safeguard the surrounding lakes, rivers, and streams' and lakes' ecological health (Sivaranjani and Rakshit 2019).



**Ankita Singh et al.,**

Instead of using inputs that have negative impacts, sustainable agriculture depends on biological processes, biodiversity, and cycles that are tailored to the local environment. The main goal of organic farming is to create a self-sufficient, environmentally and financially sustainable agricultural system that produces clean food while enhancing the surrounding biodiversity and all of its constituent parts. This system must be in harmony with nature. Organic farming has become a viable alternative agricultural system that not only solves sustainability and quality issues, but also guarantees a profitable means of subsistence for India's rural communities. The rigorous use of chemical pesticides and fertilizers calls into question the idea of sustainability in all of its forms. Both the ecosystem and the food chain are harmed by it. Organic farming steers clear of all methods that might harm the agricultural ecology. In addition to producing nutritious food, it creates an ecological balance that guards against insect issues and poor soil fertility. India is blessed with all natural and human factors essential for the development of organic farming. Hence, the Government is rapidly working on promoting organic farming as an effective way to promote sustainable agriculture.

Nano fertilizers for sustainable agriculture

In the twenty-first century, nanotechnology is a new field that is expanding rapidly. The effects of natural or human-caused climate change on global agricultural productivity are negligible. Chemical fertilizers are a vital component of modern agricultural systems, but their long-term and continuous use poses significant risks to soil fertility, the environment, and the nutritional dynamics of the rhizosphere microbiome. Nanotechnology is a relatively young and rapidly developing field in the twenty-first century. There is very little impact that climate change, whether natural or man-made, has on world agricultural productivity. Although chemical fertilizers are an essential part of contemporary agricultural systems, their long-term and ongoing usage poses serious hazards to the environment, soil fertility, and the rhizosphere microbiome's nutritional dynamics. The application of NBFs is in its infancy in agriculture, yet it has promising potential for transforming traditional farming techniques into smart agriculture, compared to any of the existing strategies (Sharma et al., 2023).

Advances in molecular and omics technology for sustainable agriculture

Increasing population, climate change and exhaustive agricultural practices either influenced nutrient inputs of soil or generated biological and physico-chemical deterioration of the soils and affected agricultural productivity and agro-ecosystems. Concerns about crop productivity and food security are so pressing that microbe-based farming techniques need to receive more consideration. Because microbes are found in soil, water, and the air, and because of their intimate relationship with plants, sustainable agricultural objectives can be achieved. In the last several decades, the hunger for environmentally friendly agriculture has led to a hunt for helpful bacteria in crop production, soil fertilization, disease management, and plant growth stimulation. Using integrated advanced biotechnology, the crop microbiome provides new avenues for managing harmful bacteria and harnessing beneficial microbes. The microbiome of crops aids in the uptake of nutrients, growth, resistance to phytopathogens, and tolerance to abiotic stressors such as salt, heat, and drought.

We only have a partial understanding of how the host's genotype, climate, mineral mobilization, soil composition, nutrient availability, interactions between the nexus of microbes, and interactions with other external microbiomes influence the functionality of the crop microbiome, despite its emergent functionality as a complex component of plant fitness. The least studied aspects of this type of crop microbiome, both cultivated and uncultivated, include its structure, composition, dynamics, and functional role. Modern biotechnological techniques are effective means of gathering the data needed to explore the microbiome and gather information for creating resistant and high-yielding crop varieties. The theoretical ideas and the practical use of these cutting-edge instruments in agricultural microbiome research are now fundamentally bridged by this information. Current omics techniques are a powerful tool for modeling, mapping, tracking, and managing the microbiome of crops. Crop microbiome identification using system biology and reverse engineering can aid in the development of future bioformulations for disease control, reclaiming strained agro-ecosystems, and increased crop yield.

Nano-system techniques in conjunction with crop microbiome triggering chemicals can aid in the development of nano-biopesticides and nano-biofertilizers. Comparing this combination to the conventional bioinoculants, there are





Ankita Singh et al.,

many advantages. They improve the likelihood of crops establishing, increase the accessibility of nutrients in the soil, and activate a variety of defensive systems in plants under stress (Rai et al., 2022).

Green synthesis of metal based nano particle

The large-scale use of conventional pesticides and fertilizers has put tremendous pressure on agriculture and the environment. Due to their great performance, low cost, and environmental friendliness, nanoparticles (NPs) have gained attention in a number of industries recently, particularly sustainable agriculture. Conventional techniques for producing nanoparticles are environmentally damaging and energy-intensive. On the other hand, chemical synthesis and plant-based synthesis of metal-based NPs are comparable, with the exception that biological extracts are used in place of the chemical reducing agent. This results in the production of NPs that are more affordable, effective, safe, and low-polluting, while also significantly reducing the usage of conventional chemicals. In order to increase yields and quality, green synthesized metal nanoparticles, or GS-MNPs, are commonly employed in agriculture.

Nanotechnology manipulates and monitors substances at the nanoscale by using scientific expertise in physics, chemistry and biology (Rai et al., 2008). Since nanotechnology has advanced so much over the last several decades, it is anticipated that we will live in a "NPs world." (Adeel et al., 2021a; Adeel et al., 2021b; Singaravelan and Alwar, 2015). NPs have unique optical, electrical, magnetic, chemical and mechanical properties due to their small size (<100 nm) and large specific surface area (Dinesh et al., 2012; Puay et al., 2015; Saif et al., 2016). Nanotechnology may significantly increase the productivity and quality of agricultural products while reducing the harmful environmental effects of pesticides and fertilizers. This is especially important in view of the numerous issues confronting agriculture, including extreme environmental stress, climate change, and food security. In addition, nanotechnology can alleviate various abiotic stresses through stimulating physiological processes during plant growth and improving the quality of soil and agricultural products (Pereira et al., 2021). The unique properties of NPs make them indispensable weapons for solving agricultural problems (Adeel et al., 2020). The reduction process of green synthesis is approximately the same as chemical reduction, except that the chemical reagents are replaced by plant extracts (leaves, fruits, roots or flowers) (Hussain et al., 2016a). Metal ions can be reduced faster to form stable metal NPs by the green synthesis method with less pollution, simple operation and low energy consumption (Iravani, 2011). In addition, the waste produced during the synthesis of plants-based NPs, are non-toxic and more accessible to treat than chemical and physical methods (Hussain et al., 2016b). It is worth noting that the method of green synthesis of metal nanoparticles (GS-MNPs) can be used to recover some noble metal ions, such as gold (Au) and silver (Ag), which greatly improves the economic benefits (Wang et al., 2009). Therefore, GS-MNPs have more advantages over traditionally synthesized NPs.

Sustainable agriculture using arbuscular mycorrhiza

Increased salt content, drought, heavy metals, and other environmental pressures commonly affect worldwide agriculture, restricting plant growth and yield, degrading soil quality, and posing a serious threat to global food security. It is crucial to develop methods for mitigating environmental pressures and to impose limitations on the use of chemical fertilizers in agricultural areas. Eco-friendly practices must be maintained in order to shield agricultural areas from the negative effects of stress. Plants do not now possess the sophisticated metabolic machinery required to address this problem or cope with the pressures.

According to research, arbuscular mycorrhizal fungi (AMF) play a crucial role in improving plant nutrient absorption, heavy metal immobilization and translocation, and growth-promoting characteristics. As such, AMFs may be useful agents for plant development in a variety of stressful conditions. The productive symbiosis and the functional bond between the plant and AMF may strengthen the regulatory mechanism that protects against the main obstacle, specifically stress. The compatibility of AMF with hyperaccumulator plants has also been bolstered by theoretical reasons and investigations on gene regulation (Chauhan et al., 2022).





Ankita Singh et al.,

Endophytic Archaea for sustainable agriculture

Although archaea have been around for more than 3.5 billion years, little is still known about them despite the fact that they were just recently found in the plant endosphere. Sustainable agriculture may benefit from the use of archaeal endophytes, especially in light of climate change and rising food demand brought on by population expansion. Recent developments in research techniques that do not rely on culture have shown a diversified abundance of archaea worldwide from the phylum *Euryarchaeota*, *Crenarchaeota*, and *Thaumarchaeota*, which are linked to important crops including maize, rice, coffee, and olive. Recent discoveries about the microbiome of plants have identified genes in archaea that may be involved in several metabolic processes in plants, such as the synthesis of amino acids and the regulation of phytohormones (Chow et al., 2022).

Obstacles to implementation

The implementation of sustainable agriculture (in a larger context) is not restricted by a single, clear barrier or specific limitation. The restriction is rather the result of a number of interrelated, situation-specific problems. We make no claims to the completeness of our classification, but we may divide the difficulties into four categories: theoretical, methodological, personal, and practical. The problems that stem from the definitions, understanding, and concept of sustainable agriculture are known as theoretical hurdles. Challenges pertaining to methodology are associated with the evaluation and implementation of sustainable agriculture. The farmer, who determines the sustainability of agricultural practices, and their traits are the source of personal challenges. Practical barriers are those that restrict action and adaptation to a more sustainable condition, even while these obstacles also have an impact on the prerequisites needed for implementation (such as information, knowledge, and intention). Practical hurdles are different from the first three in that they are related to social issues or impediments.

India's approach towards sustainable agriculture

Agriculture is the backbone of the Indian economy as well as society because of its high share in employment generation and livelihood creation. About 43% of India's geographical areas are used for agricultural activities. The industry receives its food, feed, and raw materials from this sector. Recognizing these facts, the Government of India has taken various initiatives at national and regional levels for healthy growth of agriculture. Despite this agriculture's share in GDP has declined rapidly in the recent past. Indian farmers are suffering from multiple issues such as high input cost, low profitability, land degradation, depleting water table and risks related to climate change. The approach adopted by India focuses on key factors like local climatic conditions, regional physiographic, availability of water resources, accessible technology mainly revolves around developing climate resilient agriculture which is suitable to local climatic conditions reviving natural methods of farming such as organic farming, mixed farming, crop rotation and harnessing the potential of dry land area or rain fed area agriculture in India. Apart from this, the Government of India has emphasized more on sustainable development of irrigation facilities with water use efficiency through promotion of micro-irrigation techniques. In addition, the government encourages farmers to diversify their operations and take up new ones including goat farming, chicken raising, animal husbandry, beekeeping, and wood plantations. Financial assistance is given to farmers in mountainous areas, particularly in North-Eastern India and the Western Himalayan states, to enable them to engage in sustainable horticultural practices.

The schemes related to dairy farming, food processing and infrastructure development fund reduce the farmers' dependence on agriculture. Recently the government of India has come up with the resolve of doubling farmers' income by 2022 through a seven-point strategy. The strategy focuses on irrigation, quality seeds, post-harvest management, marketing, insurance and ancillary activities. This strategy has to be incorporated with the principles of sustainable agriculture which only can help in achieving the goals of environmental health, economic profitability and socio-economic equity. Climate smart agriculture, which includes employing renewable resources like solar and biofuels, nitrogen-smart nutrient management, organic farming, agroforestry, ICT-based agro-advisories, and so on, must be quickly adopted by India.





Ankita Singh et al.,

CONCLUSION

A farming system known as "sustainable agriculture" aims to preserve the planet's capacity to support future generations while also producing the resources required for the world's current human population. A huge population all over the globe is facing immense food shortage, pollution and adulterated food. Sustainable agriculture proves to be the best way to deal with this condition. The Government of India has a well- defined array of schemes to meet almost all the needs and issues related to the development of sustainable agriculture. But the solution and success lie in the seamless implementation of these programs. Agriculture is a state subject in India which leads to politicization and fragmentation of actions and solutions related to it. On the national front there is a need to develop a consensus with the states for executing a national agenda on sustainable agriculture. Furthermore, by teaching and assisting farmers in adopting sustainable agricultural techniques, higher education institutions might support environmentally sustainable agriculture.

The progress in sustainable agriculture depends more on the development of organic farming. It's high time to take strategic and effective steps to overcome the constraints in the way of organic farming. Sustainable agriculture employs many of the natural living organisms like Archaeal endophytes and Arbuscular mycorrhiza for pollution free, safe and sustainable agriculture. Even in places with harsh weather circumstances like a drought, flood, etc., these may provide food security. In other areas of agriculture, such as plant growth promotion, plant disease, insect/pest management, fungicidal agent, food security for food packaging, extending the shelf life and preventing spoilage, and other uses, other techniques like nano fertilizers and green synthesized MNPs may be beneficial. Because microbes are found in soil, water, and the air, and because of their intimate relationship with plants, sustainable agricultural objectives can be achieved. In the last several decades, the hunger for environmentally friendly agriculture has led to a hunt for helpful bacteria in crop production, soil fertilization, disease management, and plant growth stimulation. Using integrated advanced biotechnology, the crop microbiome provides new avenues for managing harmful bacteria and harnessing beneficial microbes. The microbiome of crops aids in the uptake of nutrients, growth, resistance to phytopathogens, and tolerance to abiotic stressors such salt, heat, and drought. There should be intensive research about the plant microbiome and their application for sustainable agriculture.

ACKNOWLEDGEMENT

The authors thank Gargi College for allowing them the opportunity and space to write the review.

REFERENCES

1. Singh AS, Saliasi E, van den Berg V, Uijtdewilligen L, de Groot RHM, Jolles J, Andersen LB, Bailey R, Chang YK, Diamond A, Ericsson I, Etnier JL, Fedewa AL, Hillman CH, McMorris T, Pesce C, Pühse U, Tomporowski PD, Chinapaw MJM. Effects of physical activity interventions on cognitive and academic performance in children and adolescents: a novel combination of a systematic review and recommendations from an expert panel. *Br J Sports Med.* 2019 May;53(10):640-647.
2. Arora S, Murmu G, Mukherjee K, Saha S, Maity D. A comprehensive overview of nanotechnology in sustainable agriculture. *J Biotechnol.* 2022 Aug 20;355:21-41
3. Jha A., Pathania D., Sonu, Damathia B., Raizada P., Rustagi S., Singh P., Rani G.M., Chaudhary V. Panorama of biogenic nano-fertilizers: a road to sustainable agriculture. *Environ. Res.* 2023;235
4. Bhattacharyya, P., Chakraborty, G., 2005. Current Status of Organic Farming in India and other Countries. *Indian Journal of Fertilisers* 1(9), 111-123.
5. Parizad S, Bera S. The effect of organic farming on water reusability, sustainable ecosystem, and food toxicity. *Environ Sci Pollut Res Int.* 2023 Jun;30(28):71665-71676.



**Ankita Singh et al.,**

6. Hassanali A, Herren H, Khan ZR, Pickett JA, Woodcock CM (2008) Integrated pest management: the push-pull approach for controlling insect pests and weeds of cereals, and its potential for other agricultural systems including animal husbandry. *Philos Trans R Soc B Biol Sci* 363:611–621.
7. Sivaranjani S, Rakshit A (2019) Organic farming in protecting water quality. In: Sarath Chandran C, Thomas S, Unni M (eds) *Organic Farming*. Springer, Cham., pp 1–9
8. Sharma B, Tiwari S, Kumawat KC, Cardinale M. Nano-biofertilizers as bio-emerging strategies for sustainable agriculture development: Potentiality and their limitations. *Sci Total Environ*. 2023 Feb 20;860:160476.
9. Rai S, Omar AF, Rehan M, Al-Turki A, Sagar A, Ilyas N, Sayyed RZ, Hasanuzzaman M. Crop microbiome: their role and advances in molecular and omic techniques for the sustenance of agriculture. *Planta*. 2022 Dec 30;257(2):27.
10. Rai M, Yadav A, Gade A. Silver nanoparticles as a new generation of antimicrobials. *Biotechnol Adv*. 2009 Jan-Feb;27(1):76-83. doi: 10.1016/j.biotechadv.2008.09.002. Epub 2008 Sep 30. PMID: 18854209.
11. Adeel, M., Shakoor, N., Hussain, T., Azeem, I., Zhou, P., Zhang, P., Hao, Y., Rinklebe, J., & Rui, Y. (2021). Bio-interaction of nano and bulk lanthanum and ytterbium oxides in soil system: Biochemical, genetic, and histopathological effects on *Eisenia fetida*. *Journal of Hazardous Materials*, 415, 125574.
12. Adeel, M., Shakoor, N., Shafiq, M., Pavlicek, A., Part, F., Zafiu, C., Raza, A., Ahmad, M. A., Jilani, G., White, J. C., Ehmoser, E., Lynch, I., Ming, X., & Rui, Y. (2021). A critical review of the environmental impacts of manufactured nano-objects on earthworm species. *Environmental Pollution*, 290, 118041.
13. Singaravelan, R., & Alwar, S. B. S. (2015). Electrochemical synthesis, characterisation and phyto-genic properties of silver nanoparticles. *Applied Nanoscience*, 5(8), 983–991.
14. Dinesh, R., Anandaraj, M., Srinivasan, V., & Hamza, S. (2012). Engineered nanoparticles in the soil and their potential implications to microbial activity. *Geoderma*, 173–174, 19–27.
15. Puay, N., Qiu, G., & Ting, Y. (2015). Effect of Zinc oxide nanoparticles on biological wastewater treatment in a sequencing batch reactor. *Journal of Cleaner Production*, 88, 139–145. <https://doi.org/10.1016/j.jclepro.2014.03.081>
16. Saif S, Tahir A, Chen Y. Green Synthesis of Iron Nanoparticles and Their Environmental Applications and Implications. *Nanomaterials* (Basel). 2016 Nov 12;6(11):209.
17. Pereira, A. D. E. S., Oliveira, H. C., Fraceto, L. F., & Santaella, C. (2021). Nanotechnology potential in seed priming for sustainable agriculture. *Nanomaterials*, 11(2), 267.
18. Adeel, M., Tingting, J., Hussain, T., He, X., Ahmad, M. A., Irshad, M. K., Shakoor, N., Zhang, P., Changjian, X., Hao, Y., Zhiyong, Z., Javed, R., & Rui, Y. (2020). Bioaccumulation of ytterbium oxide nanoparticles insinuate oxidative stress, inflammatory, and pathological lesions in ICR mice. *Environmental Science and Pollution Research International*, 27(26), 32944–32953.
19. Hussain, I., Singh, N. B., Singh, A., Singh, H., & Singh, S. C. (2015). Green synthesis of nanoparticles and its potential application. *Biotechnology Letters*, 38(4), 545–560.
20. Irvani, S. (2011). Green synthesis of metal nanoparticles using plants. *Green Chemistry*, 13(10), 2638.
21. Hussain, I., Singh, N. B., Singh, A., Singh, H., & Singh, S. C. (2015b). Green synthesis of nanoparticles and its potential application. *Biotechnology Letters*, 38(4), 545–560.
22. Wang, X., Zhang, L., Ma, C., Song, R., Hou, H., & Li, D. (2009). Enrichment and separation of silver from waste solutions by metal ion imprinted membrane. *Hydrometallurgy*, 100(1–2), 82–86.
23. Chauhan S, Mahawar S, Jain D, Udpadhyay SK, Mohanty SR, Singh A, Maharjan E. Boosting Sustainable Agriculture by Arbuscular Mycorrhiza under Stress Condition: Mechanism and Future Prospective. *Biomed Res Int*. 2022 Dec 29;2022:5275449.
24. Chow, C., Padda, K.P., Puri, A. et al. An Archaic Approach to a Modern Issue: Endophytic Archaea for Sustainable Agriculture. *Curr Microbiol* 79, 322 (2022)





Ankita Singh et al.,

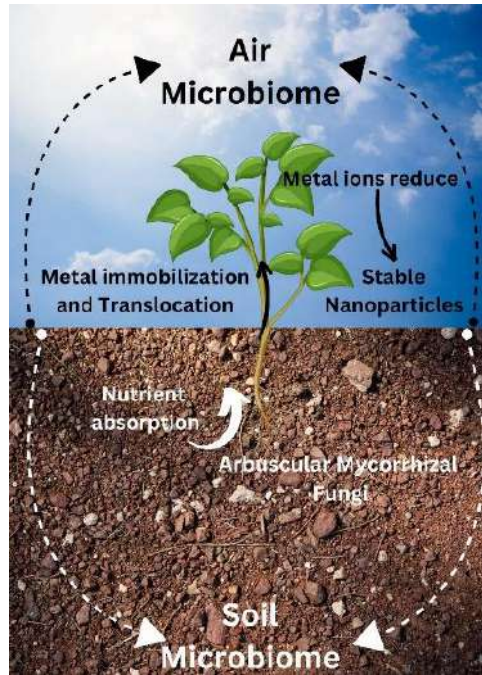


Figure 1: Role of microbiome in sustainable agriculture





A Systematic Review on Efficacy and Safety of N-Acetylcysteine and Carnitine in the Treatment of Hyperinsulinemia for Polycystic Ovary Syndrome

Swetha S¹, Janani S¹, Sophia S¹, Santhosh Kumar G¹, Jain Arvin Robert R¹, Mohamed Wasim Khan N¹ and Jayasutha Jayram^{2*}

¹Student, Department of Pharmacy Practice, Faculty of Pharmacy, Sri Ramachandra Institute of Higher Education and Research (Deemed to be University), Porur, Chennai, Tamil Nadu, India.

²Assistant Professor, Department of Pharmacy Practice, Faculty of Pharmacy, Sri Ramachandra Institute of Higher Education and Research (Deemed to be University), Porur, Chennai, Tamil Nadu, India.

Received: 28 Jun 2024

Revised: 10 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

Jayasutha Jayram

Assistant Professor, Department of Pharmacy Practice,

Faculty of Pharmacy,

Sri Ramachandra Institute of Higher Education and Research (Deemed to be University),

Porur, Chennai, Tamil Nadu, India.

E.Mail: jayasuthaj@sriramachandra.edu.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

A systematic review addressing the efficacy and safety of N-Acetyl Cysteine (NAC) and Carnitine in the treatment of hyperinsulinemia for PCOS was done. The literature search was conducted from January 2015 to July 2022 using particular search terms in databases (Pubmed, Cochrane Library, and Scopus) and the literature was screened based on inclusion and exclusion criteria. A total of 118 records were selected initially. Nine articles were included, of which four were about NAC and five were about carnitine, based on the inclusion and exclusion criteria. The chosen studies included 924 patients in total. NAC and Carnitine significantly reduced BMI, fasting insulin, fasting blood sugar, Luteinizing hormone (LH), Follicle-stimulating hormone (FSH), testosterone, sex hormone binding globulin (SHBG) and showed improvement in HOMA index and low-density lipoprotein (LDL). Carnitine significantly decreased insulin resistance (IR), Total cholesterol, and Triglycerides (TG) than NAC. NAC improved hormonal profile in hyperinsulinemic patients with PCOS. In women with PCOS, carnitine improves insulin resistance, hypoadiponectinemia, polycystic ovaries, and irregular menstruation.

Keywords: PCOS, hyperinsulinemia, N-acetyl cysteine, Carnitine





Swetha et al.,

INTRODUCTION

Polycystic ovarian syndrome or PCOS, is a metabolic as well as endocrine disorder that generally disturbs women of reproductive age [1-6]. Following the Rotterdam criteria, PCOS is diagnosed by the existence of the following criteria including oligo-ovulation or anovulation, clinical and/or biochemical hyperandrogenism, and polycystic ovary morphology (PCOM) on ultrasound [7-11]. PCOS affects between 3.7 and 22.5 percent of people in India. Traditional cardiovascular risk factors including hypertension, type-2 diabetes mellitus, and dyslipidemia as well as non-traditional cardiovascular risk factors such as mood disorders like anxiety and depression are more prevalent in PCOS patients. Approximately 30–60% of women with PCOS were overweight or obese. PCOS is a complex metabolic illness with a strong correlation to insulin resistance, which causes hyperinsulinemia. It is likely that both elevated insulin production and decreased insulin clearance lead to hyperinsulinemia. Additionally, 10% of individuals develop type 2 diabetes mellitus [12]. The coexistence of obesity and insulin resistance raises the likelihood of having type 2 diabetes mellitus and cardiovascular disease in PCOS patients at a young age. Medicines that improve insulin sensitivity have an important function in the treatment of PCOS [13-17].

Hyperandrogenism and IR worsen each other, hence insulin-sensitizing drugs enhance the clinical and biochemical features of PCOS. An antioxidant called N-acetylcysteine is produced from the amino acid L-cysteine. Thiol/sulfhydryl (R-SH/-SH) groups, which are antioxidants, are present in NAC [18]. It transforms into metabolites that improve detoxification, promote the synthesis of glutathione, and immediately break down free reactive oxygen species. It protects against hyperinsulinemia, hyperglycemia, and infertility. In PCOS patients, it reduced serum androgen levels, improved homocysteine levels, and decreased lipid profiles [19-22].

Carnitine is derived from the amino acid lysine and methionine. L-carnitine (LC), the most biologically active form, carries the fatty acids into the mitochondria, which are necessary for the generation of metabolic energy. [23,24]. The main acetyl ester of LC, acetyl-L-Carnitine (ALC), has been demonstrated to enhance reproductive functioning in PCOS by having antioxidative properties. The use of ALC in the treatment of IR has gained attention as it plays an important role in aggregating acyl-CoA derivatives that induce IR [16,25,26]. Carnitine is beneficial for weight loss, insulin function, glucose tolerance, and fatty acid metabolism [27-29]. NAC and Carnitine have beneficial effects in treating symptoms of PCOS. Hence, we planned to conduct a systematic review of the efficacy and safety of NAC and Carnitine on hyperinsulinemia in PCOS patients.

MATERIALS AND METHODS

Search Strategy and Study Selection

This systematic review was performed in compliance with the "Preferred Reporting Items for Systematic Review and Meta-Analysis" (PRISMA) guidelines. The searched database includes PUBMED, Cochrane Library, and Scopus from January 2015 to July 2022. Search terms were PCOS, NAC, N-Acetyl Cysteine, Carnitine, and Hyperinsulinemia. Randomized controlled trials of NAC and Carnitine and articles that were published in English were included. The studies with non-randomized controlled trials, no full text, duplication, case reports, brief reports, conference proceedings, review articles, observational cohort studies, and non-randomized controlled trials were excluded.

Data extraction and analysis

To compile the information and evaluate the literature, two authors worked independently. Two authors discussed and resolved discrepancies in the results summary. Reading the abstract and title served as the first screening while reading the entire content served as the second screening. PRISMA flow chart was used to define the selection of studies.



**Swetha et al.,**

Two authors independently pull out the following information from each study which includes author name, number of subjects, year of study, inclusion criteria, exclusion criteria, study design, treatment, duration of treatment, outcomes of treatment, and adverse effects of NAC and Carnitine.

Assessment of quality of included studies

The quality of each study was evaluated by two authors independently using the Joanna Briggs Institute (JBI) checklist for randomized controlled trials. JBI comprises of 13 questionnaires. Each questionnaire was answered with yes, no, unclear, and not applicable. The overall response to the questionnaire had three possible judgments: include, exclude, and seek further info.

RESULT

A total of 118 studies were identified by the search strategy. The selection process of the study is shown in Figure 1 using the PRISMA flow diagram. 39 studies were excluded due to duplication, 63 studies were excluded for not meeting the inclusion requirements and 9 randomized controlled Clinical studies that complied with the inclusion criteria were selected. The Characteristics of selected studies of NAC and Carnitine were summarized in Tables 1 and 2 respectively. The result of the quality assessment using the JBI critical appraisal checklist for Randomized controlled trials were shown in Table 3

N-acetylcysteine

Aqrabi JG et al, confirmed that there was a substantial reduction in BMI, hirsutism, FSH, LH, FSH/LH ratio, testosterone level, and improved menstrual irregularity and follicle maturation in the NAC group. Kumar MA et al, mentioned that there was a significant reduction in body weight, fasting blood sugar (FBS), LH, FSH, LH/FSH ratio, and glucose/insulin ratio in the NAC group. Nemati Met al suggested that after 8 and 12 weeks of treatment with NAC, a substantial decrease in mean BMI, total testosterone levels, hirsutism score, and fasting insulin level. Sex hormone binding globulin, FBS decreased only after 12 weeks of NAC treatment. Endometrial thickness increased after 12 weeks of treatment with NAC. Mature follicle number, LH/FSH ratio, LH, FSH, and E2 have no significant difference after long- and short-term treatment with NAC. Javanmanesh F et al indicated that NAC showed more improvement in BMI, FBS, fasting insulin, Homeostasis model assessment (HOMA) index, and LDL compared with metformin.

Carnitine

Tauqir S et al, confirmed that there was a substantial decrease in body weight, BMI, fasting serum insulin, the homeostasis model assessment for insulin resistance (HOMA-IR) index, fasting glucose, LH levels, and follicular volume of both ovaries and improvement in testosterone levels in combo group (Metformin + Pioglitazone + Acetyl-L-Carnitine) compared to the Metformin with Pioglitazone group. The serum FSH did not decrease in the groups. According to Ibrahim El Sharkwy et al., the carnitine group significantly reduced its levels of free testosterone, FSH, FG, FI, HOMA index, LDL cholesterol, total cholesterol levels, and triglycerides while also showing significant improvements in menstrual regularity, ovulation rate, pregnancy rate, FG/FI ratio, and HDL cholesterol. There was no apparent difference in BMI in both groups. Jamilian M et al, showed that there was a decrease in weight, BMI, FPG, insulin, HOMA-IR, insulin resistance, triglycerides, total cholesterol, and LDL cholesterol and improved Quantitative insulin sensitivity check index (QUICKI) in the Carnitine group. Jamilian H et al, mentioned that there was a significant reduction in weight, and BMI in the Carnitine group compared to placebo. Samimi M et al, suggested that significant reduction in weight, WC, HC, FPG, serum insulin levels, HOMA-IR, and Dehydroepiandrosterone sulfate (DHEAS) in the Carnitine group compared to the placebo.





Swetha et al.,

DISCUSSION

Insulin resistance and Hyperinsulinemia are more common in women with PCOS. Insulin-sensitizing medications enhance the clinical and biochemical characteristics of PCOS because insulin resistance and hyperandrogenism aggravate each other [17,29]. NAC has been found to improve insulin resistance and hormonal levels. NAC and metformin improved lipid profiles, BMI, AUB, fasting blood sugar, and insulin, but NAC was more effective than metformin. Among individuals with CC-resistant PCOS, NAC and metformin therapy dramatically enhanced ovulation and pregnancy rates. When compared to patients receiving metformin alone, participants treated with NAC had somewhat greater rates of ovulation and pregnancy during the first and second cycles. Body mass index (BMI), menstrual regularity, and serum levels of various hormones were all reduced significantly in N-acetyl cysteine plus metformin than metformin alone. Metformin plus N-acetyl cysteine is preferable to metformin alone for the purpose of influencing follicle development, serum LH levels, & the menstrual cycle. Because of its lack of side effects, N-acetyl cysteine can be considered as an appropriate substitute for insulin-lowering PCOS treatment [32-33]. Carnitine supplementation resulted in a substantial reduction in body weight, BMI, WC, and HC in women with PCOS when compared to placebo. The combination of L-carnitine and metformin likely showed a significant improvement in lipid profile. Adding metformin with L-carnitine improved insulin resistance, ovulation rate, and pregnancy rate than metformin alone [17]. The group that took carnitine showed significant improvements in their fasting blood glucose levels, HOMA-IR, and serum insulin levels [29]. Carnitine supplementation resulted in a considerable reduction in FPG, insulin levels, DHEAS, and HOMA-IR but did not affect QUICKI. Similar findings reported that co-supplementation of chromium and Carnitine after treatment has significantly low FPG, HOMA-IR, and insulin resistance. Patients with PCOS are more likely to develop obesity due to hyperinsulinemia and androgen excess. PCOS Women received chromium and Carnitine co-supplementation for 12 weeks lost weight and had a lower BMI. L-Carnitine supplementation did not affect TG compared to placebo. When compared to a placebo, carnitine therapy significantly reduced plasma malondialdehyde (MDA) and increased plasma total antioxidant capacity (TAC). PCOS women who received Carnitine supplements for 12 weeks had substantial improvements in mental health parameters when compared to the placebo. When compared to a placebo, the weight and BMI changes were substantially reduced when taking supplements of carnitine [34-36]. Improvement was consistently seen in all anthropometric parameters, such as waist-to-hip ratio and BMI. Chromium and Carnitine co-administration improved lipid profiles except for HDL cholesterol levels in patients with PCOS [29,36].

CONCLUSION

NAC improved hormonal profile in hyperinsulinemic patients with PCOS. NAC preserves more follicles in the ovary through its anti-apoptotic mechanism. In women with PCOS, carnitine improves insulin resistance, polycystic ovaries, irregular menstruation, and hypoadiponectinemia. It had a positive impact on weight loss along with the normalization of the menstrual cycle in women with PCOS. Consuming carnitine may help PCOS-affected women experience less oxidative stress as well as stress burden. Due to the lack of side effects, NAC and Carnitine may be a suitable alternative to insulin-lowering drugs for PCOS individuals.

CONFLICTS OF INTEREST

The author confirms no conflict of interest

AUTHORS CONTRIBUTION

Swetha S: Data Collection, original draft preparation

Janani S: Data Collection, original draft preparation

Sophia S: Data Collection

Jain Arvin Robert R: Data Collection

Mohamed Wasim Khan N: Data Collection

Jayasutha Jayram: Supervision, formal analysis, review, and editing of the manuscript





Swetha et al.,

FUNDING

The authors declare that no funds were granted or other support was received during the preparation of this manuscript.

REFERENCES

1. Murri M, Luque-Ramírez M, Insenser M, Ojeda-Ojeda M, Escobar-Morreale HF. Circulating markers of oxidative stress and polycystic ovary syndrome (PCOS): a systematic review and meta-analysis. *Hum Reprod Update*. 2013 May-Jun;19(3):268-88.
2. Teede HJ, Misso ML, Costello MF, Dokras A, Laven J, Moran L, et al. International PCOS Network. Recommendations from the international evidence-based guideline for the assessment and management of polycystic ovary syndrome. *Fertil Steril*. 2018 Aug;110(3):364-379.
3. Hu L, Shen H, Wu QF, Tian L, Hu MH. Treatment of polycystic ovarian syndrome with insulin resistance by insulin-sensitizer. *Clin Exp Obstet Gynecol*. 2014;41(3):288-92.
4. Polak K, Czyzyk A, Simoncini T, Meczekalski B. New markers of insulin resistance in polycystic ovary syndrome. *J Endocrinol Invest*. 2017 Jan;40(1):1-8.
5. Pau CT, Keefe C, Duran J, Welt CK. Metformin improves glucose effectiveness, not insulin sensitivity: predicting treatment response in women with polycystic ovary syndrome in an open-label, interventional study. *J Clin Endocrinol Metab*. 2014 May;99(5):1870-8.
6. Rocha AL, Oliveira FR, Azevedo RC, Silva VA, Peres TM, Candido AL et al. Recent advances in the understanding and management of polycystic ovary syndrome. *F1000Res*. 2019 Apr 26;8:F1000 Faculty Rev-565.
7. Bani Mohammad M, Majdi Seghinsara A. Polycystic Ovary Syndrome (PCOS), Diagnostic Criteria, and AMH. *Asian Pac J Cancer Prev*. 2017 Jan 1;18(1):17-21.
8. Sirmans SM, Pate KA. Epidemiology, diagnosis, and management of polycystic ovary syndrome. *Clin Epidemiol*. 2013 Dec 18;6:1-13.
9. Yasmin E, Balen AH. Management of polycystic ovary syndrome. *Womens Health (Lond)*. 2007 May;3(3):355-67.
10. Guideline E. Revised consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome (PCOS). *Hum Reprod*. 2003;19(1):41–47.
11. Rotterdam ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome (PCOS), *Hum Reprod*.,19(1):41–47(2004).
12. C., Muhas, Nishad K. M., Ummunnoora K. P., Jushna K., Saheera K. V., and Dilsha K. P. 2018. "Polycystic Ovary Syndrome (Pcos) – An Overview". *International Journal of Current Pharmaceutical Research* 10 (6):5-9.
13. R. Vidya Bharathi, S.Swetha, J. Neerajaa, J. Varsha Madhavica, Dakshina Moorthy Janani, S.N. Rekha et al. An epidemiological survey: Effect of predisposing factors for PCOS in Indian urban and rural population. *Middle East Fertility Society Journal*. 2017;22(4):313-316.
14. Palomba S, Santagni S, Falbo A, La Sala GB. Complications and challenges associated with polycystic ovary syndrome: current perspectives. *Int J Womens Health*. 2015 Jul 31;7:745-63.
15. Behboudi-Gandevani S, Ramezani Tehrani F, Rostami Dovom M, Farahmand M, Bahri Khomami M, Noroozadeh M et al. Insulin resistance in obesity and polycystic ovary syndrome: systematic review and meta-analysis of observational studies. *Gynecol Endocrinol*. 2016;32(5):343-53.
16. Roe A, Hillman J, Butts S, Smith M, Rader D, Playford M et al. Decreased cholesterol efflux capacity and atherogenic lipid profile in young women with PCOS. *J Clin Endocrinol Metab*. 2014 May;99(5):E841-7.
17. El Sharkwy I, Sharaf El-Din M. l-Carnitine plus metformin in clomiphene-resistant obese PCOS women, reproductive and metabolic effects: a randomized clinical trial. *Gynecol Endocrinol*. 2019 Aug;35(8):701-705.
18. Harmita, H., Iskandarsyah, I., & Afifah, S. F. (2020). Effect of transfersome formulation on the Stability and Antioxidant activity of N-Acetylcysteine in anti-aging cream. *International journal of applied pharmaceutics*, 12(1), 156–162.





Swetha et al.,

19. Santangelo F. Intracellular thiol concentration modulating inflammatory response: influence on the regulation of cell functions through cysteine prodrug approach. *Curr Med Chem.* 2003 Dec;10(23):2599-610.
20. Rizk AY, Bedaiwy MA, Al-Inany HG. N-acetyl-cysteine is a novel adjuvant to clomiphene citrate in clomiphene citrate-resistant patients with polycystic ovary syndrome. *Fertil Steril.* 2005 Feb;83(2):367-70.
21. Kilic-Okman T, Kucuk M. N-acetyl-cysteine treatment for polycystic ovary syndrome. *Int J Gynaecol Obstet.* 2004 Jun;85(3):296-7.
22. El-Hattab AW, Scaglia F. Disorders of carnitine biosynthesis and transport. *Mol Genet Metab.* 2015 Nov;116(3):107-12.
23. Ringseis R, Keller J, Eder K. Role of carnitine in the regulation of glucose homeostasis and insulin sensitivity: evidence from in vivo and in vitro studies with carnitine supplementation and carnitine deficiency. *Eur J Nutr.* 2012 Feb;51(1):1-18.
24. Ahmed Taha Abd-Elfattah, Mahmoud Abd-Ellatif Hashish, Fahd Abd El-aal Elomda, Hossam Ibrahim Megahed. Effect of adding L-Carnitine to clomiphene resistant PCOs women on the ovulation and the pregnancy rate. *Egypt J Hosp Med.*2019;76(5):4138–43.
25. Steiber A, Kerner J, Hoppel CL. Carnitine: a nutritional, biosynthetic, and functional perspective. *Mol Aspects Med.* 2004 Oct-Dec;25(5-6):455-73.
26. Power RA, Hulver MW, Zhang JY, Dubois J, Marchand RM, Ilkayeva O, Muoio DM, Mynatt RL. Carnitine revisited: potential use as adjunctive treatment in diabetes. *Diabetologia.* 2007 Apr;50(4):824-32.
27. Fenkci SM, Fenkci V, Oztekin O, Rota S, Karagenc N. Serum total L-carnitine levels in non-obese women with polycystic ovary syndrome. *Hum Reprod.* 2008 Jul;23(7):1602-6.
28. Hathcock JN, Shao A. Risk assessment for carnitine. *Regul Toxicol Pharmacol.* 2006 Oct;46(1):23-8.
29. Tauqir S, Israr M, Rauf B, Malik MO, Habib SH, Shah FA et al. Acetyl-L-Carnitine Ameliorates Metabolic and Endocrine Alterations in Women with PCOS: A Double-Blind Randomized Clinical Trial. *Adv Ther.* 2021 Jul;38(7):3842-3856.
30. Javanmanesh F, Kashanian M, Rahimi M, Sheikhsari N. A comparison between the effects of metformin and N-acetyl cysteine (NAC) on some metabolic and endocrine characteristics of women with polycystic ovary syndrome. *Gynecol Endocrinol.* 2016;32(4):285-9.
31. Nemati M, Nemati S, Taheri AM, Heidari B. Comparison of metformin and N-acetyl cysteine, as an adjuvant to clomiphene citrate, in clomiphene-resistant women with polycystic ovary syndrome. *J Gynecol Obstet Hum Reprod.* 2017 Sep;46(7):579-585.
32. M.Ashok Kumar, P. Samuel Gideon George, A. Dasari, P. Shanmugasundaram. A single-blinded randomized trial to evaluate the efficacy of N-Acetyl Cysteine over metformin in patients with polycystic ovarian syndrome. *Drug invention today.* 2018;10(2): 241-243.
33. Aqrabi JG, Al-Qadhi HI, Al-Asadi FA. Study of N-Acetyl Cysteine Plus Metformin Versus Metformin Alone in Treatment of Iraqi Women with Polycystic Ovarian Syndrome. *International journal of drug delivery technology.* 2022;12(1):202-207.
34. Samimi M, Jamilian M, Ebrahimi FA, Rahimi M, Tajbakhsh B, Asemi Z. Oral carnitine supplementation reduces body weight and insulin resistance in women with polycystic ovary syndrome: a randomized, double-blind, placebo-controlled trial. *Clin Endocrinol (Oxf).* 2016 Jun;84(6):851-7.
35. Jamilian H, Jamilian M, Samimi M, Afshar Ebrahimi F, Rahimi M, Bahmani F, Aghababayan S, Kouhi M, Shahabbaspour S, Asemi Z. Oral carnitine supplementation influences mental health parameters and biomarkers of oxidative stress in women with polycystic ovary syndrome: a randomized, double-blind, placebo-controlled trial. *Gynecol Endocrinol.* 2017 Jun;33(6):442-447.
36. Jamilian M, Foroozanfard F, Kavossian E, Kia M, Aghadavod E, Amirani E et al. Effects of Chromium and Carnitine Co-supplementation on Body Weight and Metabolic Profiles in Overweight and Obese Women with Polycystic Ovary Syndrome: a Randomized, Double-Blind, Placebo-Controlled Trial. *Biol Trace Elem Res.* 2020 Feb;193(2):334-341.





Swetha et al.,

Table 1. Characteristics of selected studies of n-acetyl cysteine in PCOS:

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
Javanmash F et al [30]	2015	Randomized double-blind clinical trial	Total no. of patients = 94 (NAC group n=46 and Met Group n = 48)	Age between 20 and 36 years who were having PCOS according to the Rotterdam Criteria	Systemic, metabolic, and endocrine disorders, Cushing syndrome, congenital adrenal hyperplasia, hyperprolactinemia, thyroid disorders, and diabetes mellitus, history of peptic ulcer, and drug use for infertility	NAC Group 600 mg, three times a day. Met Group - 500 mg Met three times a day.	24 weeks	NAC improved lipid profile and FBS and fasting blood insulin than Metformin.	GI discomfort in both groups

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
Nemati M et al [31]	2017	Controlled clinical trial	Total no. of patients =108 (NAC group n=54 and Met group n=54)	Infertility for at least one year, having clomiphene resistance, patent tubes on hysterosalpingogram, and normal semen analysis of the patients' spouses.	History of liver and kidney failure, diabetes mellitus, cardiovascular disease, Premature ovarian Failure, cushing's syndrome, any androgen-secreting tumours, chemotherapy and radiotherapy in pelvis cavity or surgery on ovaries	Received either Met (1500 mg/day) or NAC (1800mg/day) with 100 mg/day of CC for 8 and 12 weeks.	12 weeks	Administering NAC as an adjuvant for CC compared to metformin is recommended for improving hormonal profile and treating anovulatory infertility in hyperinsulinemia patients, especially women with PCOS resistant to CC.	Adverse effects were not determined.





Swetha et al.,

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
Kumar MA et al [32]	2018	A single-blinded randomized trial	Total no. of patients = 100 (Met Group n= 50 and NAC group n= 50)	Women of Age group between 18–37 and are diagnosed with PCOS.	Diabetes mellitus, hepatic or kidney diseases	Met Group - 500 mg thrice a day and NAC Group - 600 mg thrice a day for 12 weeks.	12 weeks	NAC improved insulin resistance and hormone levels, and long-term health through improvement of peripheral insulin and reduction of side effects than Metformin.	A few participants experienced nausea, vomiting, diarrhoea and headaches.

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
Aqrawi JG et al [33]	2022	Prospective, comparative clinical study.	Total no. of patients = 45 (Met group n= 19 and NAC + Met group n= 25)	Female 18- 45 years, oligo ovulation or anovulation, clinical or biochemical hyperandrogenism and/ or polycystic ovaries	Congenital adrenal hyperplasia, thyroid dysfunction, hyperprolactinemia, Cushing syndrome, androgen-secreting neoplasia, diabetes mellitus, medication changing insulin hemodynamic, medication affecting carbohydrate metabolism, hormonal drugs, multivitamins,	Met group - 500 mg three times daily. NAC + Met group - 600 mg three times daily plus Met 500 mg three times daily	3 months	NAC with Metformin is superior to Metformin alone, affecting the menstrual cycle, serum LH level, and follicle maturation.	Adverse effects were not determined.





Swetha et al.,

					and anti-obesity drugs within last 3 months, severe concurrent cardiovascular disease, Severe hepatic or kidney disease				
--	--	--	--	--	---	--	--	--	--

CC- Clomiphene citrate; FBS- Fasting blood sugar; GI- Gastrointestinal; LH- Luteinizing hormone; Met- Metformin; NAC- N-Acetyl Cysteine

Table 2. Characteristics of selected studies of carnitine in PCOS:

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
Sami M et al [34]	2016	Randomized, double-blind, placebo-controlled trial	Total no. of patients=60 (Carnitine group n=30 and Placebo group n=30)	Age 18–40 years; oligo/anovulation, hyperandrogenism, polycystic ovary morphology	Hyperprolactinaemia, Diabetes mellitus, thyroid disease, subjects following a special diet or consuming drugs with an effect on hormonal profile like oral contraceptives, ovulation induction agents and anti-obesity therapies in the last 3 months before enrolment.	PCOS patients received either 250 mg Carnitine supplements or a placebo.	12 weeks	Carnitine group reduces weight, BMI, WC, HC and glycaemic control. It has no effect on lipid profile or free testosterone levels.	No side effects associated with the administration of Carnitine were observed.





Swetha et al.,

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
Jamilian H et al [35]	2017	Randomized, double-blind, placebo-controlled trial	Total no. of patients= 56 (Carnitine group n=28 and Placebo group n=28)	Age 18–40 years, oligo- and/or anovulation, biochemical signs of hyperandrogenism & polycystic ovaries	Pregnant women, individuals with metabolic diseases, thyroid disease, hyperprolactinemia, hypercortisolemia, renal and liver diseases	250 mg Carnitine supplements for 12 weeks and placebo.	12 weeks	Carnitine improved mental health and oxidative stress indicators.	No side effects were observed in patients received Carnitine.

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
EI Sharkwy I et al [16]	2019	A double-blinded randomized clinical trial	Total no. of patients = 274 (Carnitine group n=138 and Placebo group n=136)	Oligo and/or anovulation, clinical and/or biochemical signs of hyperandrogenism, and polycystic ovary	Smokers, drug users, individuals with other reasons of infertility such as a male or tubal factor, endocrine abnormalities such as thyroid dysfunction & hyperprolactinemia	From 3-7 days of menstrual cycle, Group 1 = 150 mg/d CC plus L-Carnitine (3 g) & metformin 850 mg. After 1-week dose doubled as 1700 mg/d CC Group 2 = 150 mg/d of CC with metformin and placebo.	15 months	Carnitine group enhances ovulation rate, pregnancy rate, menstruation regulation rate, insulin resistance & lipid profile.	The side effects were not determined.





Swetha et al.,

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
Jamilian M et al [36]	2019	Randomized, Double-Blind, Placebo-Controlled Trial	Total no. of patients= 54 (Carnitine group n= 27 and Placebo group n=27)	Overweight and obese women with PCOS diagnosed based on the Rotterdam criteria, and age 18–40 years.	Pregnancy, adrenal hyperplasia, androgen-secreting tumors, hyperprolactinemia, thyroid dysfunction, and diabetes.	Supplements (200g/ day chromium picolinate with 1000 mg/day Carnitine) or a placebo	12 weeks	Carnitine supplements improved body weight, glycemic management and lipid profiles	Adverse effects were not determined.

AUTHOR NAME	YEAR	STUDY DESIGN	NO OF SUBJECTS (n)	INCLUSION CRITERIA	EXCLUSION CRITERIA	TREATMENT	DURATION OF TREATMENT	OUTCOMES OF TREATMENT	ADVERSE EFFECTS
Tauqir S et al [29]	2021	Double-Blind Randomized Clinical Trial	Total no. of patients =133 (Met+ pio group n=62 and Combo group n=71)	Oligo- or anovulation, hyperandrogenism, polycystic ovary morphology on ultrasound or increased ovarian volume	Pregnancy, hyperprolactinemia, thyroid illness, cushing syndrome, late-onset congenital adrenal hyperplasia, on hormone replacement therapy	Combo group - metformin, pioglitazone, & ALC (500 mg, 15 mg, and 1500 mg). Met + Pio group - metformin + pioglitazone (500 mg, 15 mg) and placebo twice daily.	12 weeks	Carnitine group improved insulin resistance, polycystic ovaries, menstrual irregularities & hypo-adiponectinemia	A few participants complained of GI issues after consuming carnitine.

ALC- Acetyl L-Carnitine; BMI- Body mass index; CC- Clomiphene citrate; GI- Gastrointestinal; HC- Hip circumference; LDLR- Low density lipoprotein receptor; Pio- Pioglitazone; WC- Waist circumference





Swetha et al.,

Table 3. JBI critical appraisal checklist for Randomized controlled trials:

QUESTIONNAIRE	Agraw i JG et al	Kuma r M et al	Nemati M et al	Javanmanes h F et al	Tauqir S et al	Jamilia n M et al	El sharkw y I et al	Jamilia n H et al	Samim i M et al
Was true randomization used for assignment of participants to treatment groups?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Was allocation to treatment group concealed?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Were treatment groups similar at the baseline?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Were participants blind to treatment assignment?	yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Were those delivering treatment blind to treatment assignment?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Were outcomes assessors blind to the treatment assignment?	Unclear	No	Unclear	Unclear	Unclear	Unclear	Unclear	Unclear	Unclear
Were treatment groups treated identically other than the intervention of interest?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Was follow up complete if not, were differences between groups in terms of their follow up adequately described and analyzed?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Were participants analyzed in the groups to which they were randomized?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes





Swetha et al.,

Were outcomes measured in the same way for treatment groups?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Were outcomes measured in the reliable way?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Was appropriate statistical analysis used?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Was trail design appropriate and any deviation from the standard RCT design (individual randomization, parallel groups) accounted for in the conduct and analysis of the trail	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

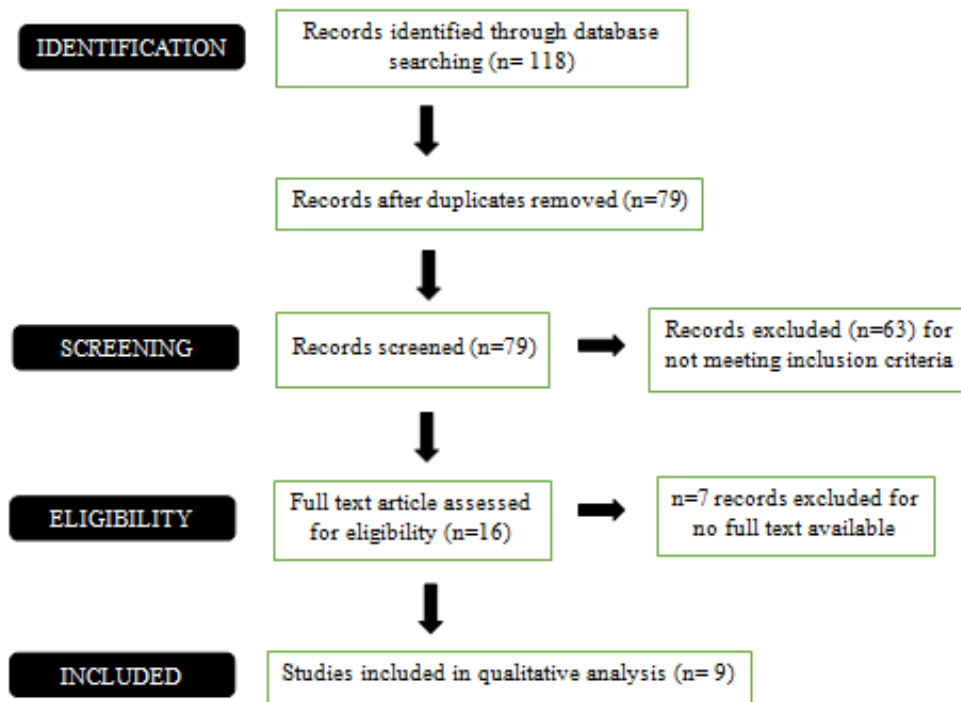


Fig.1:PRISMA chart of the study selection process





GC - MS Profile of Ethanolic Root Extract of *Withania somnifera* (Ashwagandha)

Neha Singh^{1*} and Anita R J Singh²

¹Ph.D Research Scholar, PG and Research Department of Biotechnology, Women's Christian College, (Affiliated to University of Madras), Chennai, Tamil Nadu, India.

²Professor, PG and Research Department of Biotechnology, Women's Christian College, (Affiliated to University of Madras), Chennai, Tamil Nadu, India.

Received: 05 Jun 2024

Revised: 14 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

Neha Singh

Ph.D Research Scholar,
PG and Research Department of Biotechnology,
Women's Christian College, (Affiliated to University of Madras),
Chennai, Tamil Nadu, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

In the present study phyto -constituents of root ethanolic extract were analyzed to check and identify the important phytochemicals by GCMS analysis. The present study indicates the presence of some essential pharmaceutical compounds- Benzo[b]thiophene-3-carboxylicacid, Hydrazine, N, N-dimethyl-N'-diethylboryl-, 7-acetoxy-2-acet, 4H-Pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl-, Propanamide,2-hydroxy-N-methyl-, 2-(Chloromethyl)-5-ethyl-1,3,4-oxadiazole, N-Nitroso-2,4,4-trimethyloxazolidine, Adenosine, N6-phenylacetic acid, ButylatedHydroxytoluene, . Benzo[b]thiophene-3-carboxylicacid,7-acetoxy-2-acet, Bonomycinhydrochloride, Propanamide,2-hydroxy-N-methyl-, 2-(Chloromethyl)-5-ethyl-1,3,4-oxadiazole, N-Nitroso-2,4,4-trimethyloxazolidine, 2-Amino-N-(4-fluorophenyl) benzamide, 2TBDMSderi, Tetradecane, and 9,9-Dimethoxybicyclo[3.3.1]nona-2,4-dione. In the present investigation, 100 peaks were observed and identified. Out of 100 peaks, 61 peaks were found to be pharmaceutically important. Results indicate that the root of *Withania somnifera* contains many bioactive compounds that can be exploited to develop eco-friendly, plant-based nontoxic drugs to treat viral, bacterial, and fungal infections and also can treat cancer and some other diseases.

Keywords- GCMS analysis, *Withania somnifera*, Pharmaceutical compounds, Benzo[b]thiophene-3-carboxylicacid, -(Chloromethyl)-5-ethyl-1,3,4-oxadiazole.





Neha Singh and Anita R J Singh

INTRODUCTION

Withania somnifera or Ashwagandha is an important Ayurvedic plant (1) belonging to the Solanaceae family(2). Ashwagandha plant grows up to 1.5 meters (3). Since ancient times *Withania somnifera* (Ashwagandha) has been used to cure various diseases. Many Ayurvedic and Siddha drugs have been formulated by the use of whole plants or constituents of Ashwagandha. It is found in many countries like- India, Afghanistan, China, Nepal, Bhutan, and Bangladesh (4). Ashwagandha is widely cultivated in different parts of India- Rajasthan, Gujarat, Uttar Pradesh, Madhya Pradesh, Chhattisgarh, Punjab, Haryana, Andhra Pradesh, Karnataka, and Tamil Nadu (5, 6). *Withania somnifera* contains various secondary metabolites – Withanolides, flavonoids, terpenoids, tannins, saponins, phenolic compounds, and other useful compounds. Ashwagandha Roots manufactures Ayurvedic medicines, tonics, Chyavanprash, oral drops, haircare, and other healthcare products. In the Ayurvedic system of medicine, *Withania somnifera* roots are used as a Rejuvenating drug, tonic, Alternative pungent, astringent, Aphrodisiac, and Phthisis (7-9).

MATERIALS AND METHODS

Withania somnifera plant samples including wild and developed varieties were collected from various parts of India. Collected plant samples were verified by Dr. Prof. P Jayaraman PhD. Director- Plant anatomy Research center, Retd. Professor Presidency College, Chennai-5, Reg. No. of certificate- PARC/2021/4565. Each varieties were cultivated in Chennai field conditions

Extract Preparation

Plant root samples were washed with distilled water. Ethanol (95% v/v) was used to prepare the extract by the Soxhlet apparatus. The obtained plant root extract was further concentrated by a rotatory evaporator and then stored in the refrigerator at 4°C for future analysis work.

GCMS analysis

Withania somnifera root extract was further diluted in ethanol and filtered through the Whatman filter paper number 41 to get particle-free extract dilution for GCMS analysis to check the active compounds present in the sample. Identification of active molecules was performed by a Gas Chromatography unit coupled with mass spectroscopy. A Shimadzu NX series coupled to Nexis GC -2030 Gas chromatograph was equipped with an AOC 20i automatic injector and a 30-meter, SH -I-.5SiI column (0.25mm ID, 0.25µm film thickness. The split injector was used for sample introduction and 10:1 was the split ratio. The oven temperature program was set to between 35°C to 450°C. The oven program was set to start at 35°C for 2 minutes, followed by a steady increase up to 250°C. then ramped to 450 The oven temperature program was programmed to begin at 35°C, hold for 2 minutes, then ramp at 20°C per minute to 450°C, and hold for 5 minutes. The helium Carrier gas was set to a 2 ml/minute flow rate (constant flow mode).

Mass Spectrum

A Direct connection with capillary column metal quadrupole mass filter pre-rod mass spectrometer operating in electron ionization (EI) mode with software GCMS solution ver. 2.6 was used for all analyses. Low-resolution mass spectra were acquired at a resolving power of 1000 (20% height definition) and scanned from m/z 25 to m/z 1000 at 0.3 seconds per scan with a 0.2-second inter-scan delay. High-resolution mass spectra were acquired at a resolving power of 5000 (20% height definition) and scanning the magnet from m/z 65 to m/z 1000 at 1 second per scan.





Neha Singh and Anita R J Singh

Mass spectrometry library search

Identification of the components of the compound was compared to the known components stored in the mass spectra of NIST library V 11 provided by the instrument's software ⁽¹⁰⁾. Also, GCMS metabolomics is used for similarity checking with the retention index ⁽¹¹⁾.

RESULTS AND DISCUSSION

In the present investigation -Benzo[b]thiophene-3-carboxylic acid, Hydrazine, N, N-dimethyl-N'-diethylboryl-, 7-acetoxy-2-acet, 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-, Propanamide, 2-hydroxy-N-methyl-, 2-(Chloromethyl)-5-ethyl-1,3,4-oxadiazole, N-Nitroso-2,4,4-trimethylloxazolidine, Adenosine, N6-phenylacetic acid, Butylated Hydroxytoluene compounds indicates the presence of antioxidant and anti-cancer compounds. Benzo[b]thiophene-3-carboxylic acid, 7-acetoxy-2-acet, Bionomycin hydrochloride, Propanamide, 2-hydroxy-N-methyl-, 2-(Chloromethyl)-5-ethyl-1,3,4-oxadiazole, N-Nitroso-2,4,4-trimethylloxazolidine, 2-Amino-N-(4-fluorophenyl) benzamide, 2-TBDMS-derivative, Tetradecane, 9,9-Dimethoxybicyclo[3.3.1]nona-2,4-dione, compounds are known for antimicrobial and antifungal activity. Some important compounds like - Acetic acid, butylester, 2-Pentanone, 4-hydroxy-4-methyl-, Ethylbenzene, p-Xylene, Dihydroxyacetone, Aminocyclopropanoic acid, 1,2,3,4-Butanetetrol, [S-(R*, R*)]- Undecane, O-Methylisourea, Allene, Cyclohexasiloxane, dodecamethyl-, Tridecane, Acetamide, N, N', N''-methylidynetris-, 4-Heptafluorobutyropyntadecane, Cyclohexasiloxane, dodecamethyl-, 1,3-Adamantanediacetamide, Cyclopentasiloxane, decamethyl-, Cycloheptasiloxane, tetradecamethyl-, Cyclohexanol, 1R-4-trans-acetamido-2,3-trans-epoxy-, Oxacycloheptadec-8-en-2-one, (8Z)-, Hexatriacontane, Eicosane are industrially essential.

Discussion

Phytoanalysis of Plant extract has been performed to check the presence of various active compounds in the sample. The present investigation revealed that *Withania somnifera* possesses many active compounds useful for Pharmaceutical, industrial, and healthcare purposes. It also indicates that the active compounds can be used for novel medication formulations.

Conflict of Interest- No

REFERENCES

1. Singh N., Singh A. R. J. An Eco Friendly and Holistic Approach to Assess the Efficacy of Root and Leaf Extracts of *Withania somnifera* against MCF -7 Breast Cancer Cell Line. *Indian Journal of Natural Sciences*. 2024. Vol.14(82) 67969- 67975. ISSN: 0976 – 0997.
2. A. R Vyas, S.V. Singh. Molecular targets and mechanisms of cancer prevention and treatment by withaferin-a, a naturally occurring steroidal lactone. *AAPS J* 16 (2014) 1–10. <https://doi.org/10.1208/s12248-013-9531-1>.
3. M.A Weiner. J Weiner. *Ashwagandha (India ginseng), Herbs that Heal*, Quantum Books. Mill Valley, C.A. (1994) 70-72.
4. Bano A, Ayub M, Rashid S, Sultana S and Sadia H (2013). Ethnobotany and conservation status of floral diversity of Himalayan range of Azad Jammu and Kashmir Pakistan. *Pakistan Journal of Botany*. 45: 243-251.
5. Ven Murthy, M.R., K Ranjekar, P., Ramassamy, C., Deshpande, M., 2010. Scientific basis for the use of Indian ayurvedic medicinal plants in the treatment of neurodegenerative disorders: 1. Ashwagandha. *Central Nervous System Agents in Medicinal Chemistry (Formerly Current Medicinal Chemistry-Central Nervous System Agents)* 10 (3), 238–246.
6. Mir B A., Khazir J., Mir N A., Hasan T., Koul S. (2012). Botanical, chemical and pharmacological review of *Withania somnifera* (Indian ginseng): an ayurvedic medicinal plant. *Indian Journal of Drugs and Diseases*. 1 [6] 147-160.





Neha Singh and Anita R J Singh

7. Dutta U. C., King G. (1877). *Materia medica of Hindus*. Calcutta Machine Press.
8. Kumar S, Tiwari, A. And Dwivedi R. (1980). The use of aphrodisiacs in Medieval India. *Nagarjun*, 23:170-174.
9. Sen Gupta N. N. K. (1984). *The Ayurvedic system of medicine*. (Delhi, Neeraj Publishing House) Vol II.
10. Adam R. Identification of Essential Oil Components by Gas Chromatography/Mass Spectroscopy, Allured Publishing Co., Carol Stream, IL.1995.
11. Vanden Dool H, Kratz P D. A generalization of the retention index system including linear temperature programmed gas-liquid partition chromatography. *Journal of chromatography*. 1963, 11: 463-471.
12. Keri, R. S., Chand, K., Budagumpi, S., Balappa Somappa, S., Patil, S. A., & Nagaraja, B. M. (2017). *An overview of benzo [b] thiophene-based medicinal chemistry*. *European Journal of Medicinal Chemistry*, 138, 1002–1033. doi:10.1016/j.ejmech.2017.07.038.
13. <https://pubchem.ncbi.nlm.nih.gov/compound/Hydrazine>.
14. <https://pubchem.ncbi.nlm.nih.gov/compound/Butyl-acetate#section=Uses>
15. <https://regenesi.com/en/glossary/4-methyl-2-pentanone/#:~:text=The%20major%20application%20of%204,dyes%20in%20the%20printing%20industry>.
16. <https://www.dccew.gov.au/environment/protection/npi/substances/factsheets/ethylbenzene#:~:text=Ethylbenzene%20is%20used%20primarily%20in,%2C%20tobacco%20products%2C%20and%20insecticides>.
17. Toulhoat, H. (2010). Heterogeneous Catalysis: Use of Density Functional Theory. *Encyclopedia of Materials: Science and Technology*, 1–7. doi:10.1016/b978-008043152-9.02243-0.
18. <https://dermnetnz.org/topics/dihydroxyacetone#:~:text=DHA%20was%20originally%20used%20as,approved%20agent%20for%20sunless%20tanning>.
19. Teppang, K. L., Ehrlich, R. S., & Yang, J. (2020). *Method to discriminate amyloids using fluorescent probes*. *Methods in Enzymology*. Volume 639, 2020, 91-114. doi:10.1016/bs.mie.2020.04.010.
20. Chukwu C J, Omaka O N, O., Aja PM. (2017). *Characterization of 2,5-dimethyl-2,4-dihydroxy-3(2H) furanone, A Flavourant Principle From Sysepalum dulcificum*. *Natural Products Chemistry & Research*, 05(08). 296. doi:10.4172/2329-6836.1000299.
21. <https://en.wikipedia.org/wiki/Threitol>
22. <https://www.paulaschoice.co.uk/undecane-/ingredient-undecane.html>.
23. Konopka J.B. N-Acetylglucosamine Functions in Cell Signaling. *Scientifica* Volume 2012 | Article ID 489208 | <https://doi.org/10.6064/2012/489208>
24. Yu, X.; Zhao, M.; Liu, F.; Zeng, S.; Hu, J. Identification of 2,3-Dihydro-3,5-Dihydroxy-6-Methyl-4H-Pyran-4-One as a Strong Antioxidant in Glucose–Histidine Maillard Reaction Products. *Food Res. Int.* 2013, 51, 397–403.
25. <https://www.fishersci.no/shop/products/o-methylisourea-hemisulfate-99-thermo-scientific/11419527#:~:text=O%2DMethylisourea%20is%20used%20for,fluorouracil%20antineoplastic%20and%20anthelmintic%20imidazole.&text=Applications-,O%2DMethylisourea%20is%20used%20for%20the%20synthesis,fluorouracil%20antineoplastic%20and%20anthelmintic%20imidazole.&text=Incompatible%20with%20strong%20oxidizing%20agents>.
26. Karnik A V., Hasan M. Chapter 3 - Elements of chirality and chiral stereoisomerism. 2021. *Stereochemistry A Three-Dimensional Insight*. 49-91. <https://doi.org/10.1016/B978-0-12-821062-8.00007-7>.
27. <https://www.canada.ca/en/health-canada/services/chemical-substances/challenge/batch-2/d6.html>.
28. <https://www.ncbi.nlm.nih.gov/books/NBK557654/>
29. Abdulmalik, O; Safo, MK; Chen, Q; Yang, J; Brugnara, C; Ohene-Frempong, K; Abraham, DJ; Asakura, T (2005). "5-hydroxymethyl-2-furfural modifies intracellular sickle hemoglobin and inhibits sickling of red blood cells". *British Journal of Haematology*. 128 (4): 552–561. doi:10.1111/j.1365-2141.2004.05332.x.
30. <https://www.biosynth.com/p/FD76710/4845-50-5-trans-14-dioxane-23-diol>
31. <https://pubchem.ncbi.nlm.nih.gov/compound/Tridecane>
32. <https://www.biosynth.com/p/CAA83210/2832-10-2-ethyl-4-acetyl-5-oxohexanoate>.
33. Kennedy G.L. Acetamide. Reference Module in Biomedical Sciences. *Encyclopedia of Toxicology* (Third Edition). 2014, Pages 24-25. <https://doi.org/10.1016/B978-0-12-386454-3.00001-4>.





Neha Singh and Anita R J Singh

34. <https://www.mayoclinic.org/drugs-supplements/vancomycin-intravenous-route/description/drg-20068900#:~:text=Vancomycin%20is%20used%20to%20treat,other%20medicines%20may%20not%20work.>
35. Fischer, M. K; Shingleton, A. W (2001). Host plants and ants influence the honeydew sugar composition of aphids. *Functional Ecology*. 15 (4): 544. doi:10.1046/j.0269-8463.2001.00550.x.
36. Rahman A U., Choudhary, M., Majeed, A., Shabbir, M., Ghani, U., & Shameel, M. (1997). A succinyl anthranilic acid ester and other bioactive constituents of *Jolyna laminarioides*. *Phytochemistry*, 46 7, 1215-8. [https://doi.org/10.1016/S0031-9422\(97\)80014-3](https://doi.org/10.1016/S0031-9422(97)80014-3).
37. Gao, M., Wang, M., Miller, K., & Zheng, Q. (2011). Facile radiosynthesis of new carbon-11-labeled propanamide derivatives as selective androgen receptor modulator (SARM) radioligands for prostate cancer imaging. *Steroids*, 76, 1505-1512. <https://doi.org/10.1016/j.steroids.2011.08.005>.
38. <https://www.thermofisher.in/chemicals/shop/products/n-pentadecane-99-thermo-scientific-1/ALF-A10336-30>
39. Ko, Y., Lim, D., Choi H., Choi S., Choi S., Hong J., Yoon Y A., Chung H., Lim K M., Kim K B., Lee J Y., Kwack S J., Bae O N. Risk assessment of cyclohexasiloxane D6 in cosmetic products. *Toxicol Res.* (2024). <https://doi.org/10.1007/s43188-024-00235-2>
40. <https://www.canada.ca/content/dam/dnd-mdn/documents/health/nutrition/supplement-fact-sheet-beta-alanine.pdf>
41. Shukla M., Rathi K., Hassam M., Yadav D K., Karnatak M., Ravwat V., Verma V P. An overview on the antimalarial activity of 1,2,4-trioxanes, 1,2,4-trioxolanes, and 1,2,4,5-tetraoxanes. *Med Res Rev.* 2023; 1–72. <http://dx.doi.org/10.1002/med.21979>.
42. <https://pubchem.ncbi.nlm.nih.gov/compound/150629>
43. <https://en.wikipedia.org/wiki/Adamantane>.
44. Tallima H, El Ridi R. Arachidonic acid: Physiological roles and potential health benefits - A review. *J Adv Res.* 2017 Nov 24;11:33-41. doi: 10.1016/j.jare.2017.11.004. PMID: 30034874; PMCID: PMC6052655.
45. <https://hmdb.ca/metabolites/HMDB0002001>.
46. Sindhe MA, Bodke YD, Kenchappa R, Telkar S, Chandrashekar A. Synthesis of a series of novel 2,5-disubstituted-1,3,4-oxadiazole derivatives as potential antioxidant and antibacterial agents. *J Chem Biol.* 2016 Jul 7;9(3):79-90. doi: 10.1007/s12154-016-0153-9. PMID: 27493696; PMCID: PMC4958064.
47. Kim EM, Jung HR, Min TJ. Purification, structure determination, and biological activities of 20 (29) lupin-3-one from *Daedaleopsis tricolor* (Bull. ex Fr.). *Bond Sing. Bull Korean Chem Soc* 2001; 22: 59-62.
48. Kaur B, Singh P. Inflammation: Biochemistry, cellular targets, anti-inflammatory agents and challenges with special emphasis on cyclooxygenase-2. *Bioorganic Chemistry*. Volume 121, April 2022, 105663. <https://doi.org/10.1016/j.bioorg.2022.105663>.
49. Mackay, D., Cowan-Ellsberry, C. E., Powell, D. E., Woodburn, K. B., Xu, S., Kozerski, G. E., & Kim, J. (2015). *Decamethylcyclopentasiloxane (D5) environmental sources, fate, transport, and routes of exposure. Environmental Toxicology and Chemistry*, 34(12), 2689–2702. doi:10.1002/etc.2941.A. Hill
50. Johnson W., Bergfeld W F. , Belsito D V., Hill R A., Klaassen C D., Liebler D C., Marks J G., Shank R C., Slaga T J., Snyder P W., and Andersen F. A. Safety Assessment of Cyclomethicone, Cyclotetrasiloxane, Cyclopentasiloxane, Cyclohexasiloxane, and Cycloheptasiloxane. *International Journal of Toxicology*. 2011;30(6_suppl):149S-227S. doi: 10.1177/1091581811428184.
51. Thakore, K. N. *Butylated Hydroxytoluene. Encyclopedia of Toxicology*. 2014. 583–584. doi: 10.1016/b978-0-12-386454-3.00263-3.
52. KUMAR L, ALIKHAN A F, KOWSALYA M, ARTHI S. Effect of multicomponent aqueous plant extracts on the growth and biochemical composition of *Mentha arvensis* L. and *Centella asiatica* L. *International Journal of Agricultural Science*. 2023. Volume 8, 49-74. ISSN: 2367-9026.
53. Mabkhot YN, Al-Majid AM, Barakat A, Al-Showiman SS, Al-Har MS, Radi S, Naseer MM, Hadda TB. Synthesis and biological evaluation of 2-aminobenzamide derivatives as antimicrobial agents: opening/closing pharmacophore site. *Int J Mol Sci.* 2014 Mar 21;15(3):5115-27. doi: 10.3390/ijms15035115. PMID: 24663060; PMCID: PMC3975443.
54. <https://pubchem.ncbi.nlm.nih.gov/compound/Dodecanedioic-acid>.





Neha Singh and Anita R J Singh

55. Mabkhot YN, Al-Majid AM, Barakat A, Al-Showiman SS, Al-Har MS, Radi S, Naseer MM, Hadda TB. Synthesis and biological evaluation of 2-aminobenzamide derivatives as antimicrobial agents: opening/closing pharmacophore site. *Int J Mol Sci.* 2014 Mar 21;15(3):5115-27. doi: 10.3390/ijms15035115. PMID: 24663060; PMCID: PMC3975443.
56. <https://www.1mg.com/drugs/tropan-5-tablet-25243?wpsrc=Google+Organic+Search>.
57. Evaluation of Antimicrobial Potential of Tetradecane Extracted from *Pediococcus acidilactici* DSM: 20284 - CM Isolated from Curd Milk. *Egyptian Journal of Chemistry.* 2022 Vol. 65(3) 705 – 713. DOI: 10.21608/ejchem.2021.70234.3547.
58. Yasuko O, Haruka S, Chao JL, Kozo T, Kimio H, Tsuneji N, Koichi I. 2000. Effect of synthesized cyclohexanol derivatives using l-menthol as a lead compound on the percutaneous absorption of ketoprofen, *International Journal of Pharmaceutics*, 198(2):191-200.
59. Prajapati KS, Singh AK, Kushwaha PP, Shuaib M, Maurya SK, Gupta S, Senapati S, Singh SP, Waseem M, Kumar S. *Withaniasomnifera* phytochemicals possess SARS-CoV-2 RdRp and human TMPRSS2 protein binding potential. *Vegetos.* 2023;36(2):701-720. doi: 10.1007/s42535-022-00404-4. Epub 2022 Jun 15. PMID: 35729946; PMCID: PMC9199469.
60. <https://pubchem.ncbi.nlm.nih.gov/compound/Succinic-Acid>.
61. <https://pubchem.ncbi.nlm.nih.gov/compound/6-Aza-2-thiothymine>.
62. Singh C R., Nelson R., Krishnan M., Pargavi B. Identification of Volatile Constituents from *Premna serratifolia* L. through GC-MS. *International Journal of PharmTech Research.* 2011. Vol. 3 (2) 1050-1058.
63. Yolin Angel PASR, Jeyakumar P, Jasmin Suriya AR, Sheena A, Karuppiah P, Periyasami G, Stalin A, Murugan K. Topical antifungal keratitis therapeutic potential of *Clitoria ternatea* Linn. flower extract: phytochemical profiling, *in silico* modeling, and *in vitro* biological activity assessment. *Front Microbiol.* 2024 Jan 24;15:1343988. doi: 10.3389/fmicb.2024.1343988. PMID: 38328419; PMCID: PMC10849212.
64. <https://pubchem.ncbi.nlm.nih.gov/compound/Heptadecane>.
65. Aparna V., Vijayan D., Mandal P., Karthe P., Sadasivan C., Haridas M. Anti-Inflammatory Property of n-Hexadecanoic Acid: Structural Evidence and Kinetic Assessment. *Chemical Biology & Drug Design.* 2012. 80(3):434-9.
66. Gupta V., Tyagi S., Tripathi S. Hexadecanoic acid methyl ester, a potent hepatoprotective compound in leaves of *Pistia stratiotes* L. *The Applied Biology & Chemistry Journal.* 2023. 4(4):118-120. DOI:10.52679/tabcj.2023.0012.
67. <https://drugs.ncats.io/drug/0951377U8F#:~:text=Description,bulbs%20of%20Zephyranthes%20candida%20also>.
68. Angel P.A.S.R.Y., Jeyakumar P., Suriya A R J., Sheena A., Karuppiah P., Periyasami G., Stalin A., Murugan K. Topical antifungal keratitis therapeutic potential of *Clitoria ternatea* Linn. flower extract: phytochemical profiling, *in silico* modeling, and *in vitro* biological activity assessment. *Front Microbiol.* 2024; 15: 1343988. doi: 10.3389/fmicb.2024.1343988.
69. [https://www.ottokemi.com/alkenes/hexatriacontane-98.aspx#:~:text=H%201393%20\(OTTO\)%20Hexatriacontane%2C,of%20lower%20alkanes%20by%20cracking](https://www.ottokemi.com/alkenes/hexatriacontane-98.aspx#:~:text=H%201393%20(OTTO)%20Hexatriacontane%2C,of%20lower%20alkanes%20by%20cracking).
70. Arora S., Kumar G. Gas Chromatography-Mass Spectrometry (GC-MS) determination of bioactive constituents from the methanolic and ethyl acetate extract of *Cenchrus setigerus* Vahl (Poaceae). *The Pharma Innovation Journal* 2017; 6(11): 635-640.
71. <https://www.fishersci.fi/shop/products/n-eicosane-99-thermo-scientific/11421518#:~:text=n%2DEicosane%20is%20a%20saturated,and%20in%20the%20petrochemical%20industry>.
72. <https://pubchem.ncbi.nlm.nih.gov/compound/Tetratriacontane>





Neha Singh and Anita R J Singh

Peak#	Ret.Time	m/z	Area	CompoundName	
1	5.205	TIC	44882	Benzo[b]thiophene-3-carboxylicacid,7-acetoxy-2-acet	anti-cancer, Anti-microbial, anti-oxidant, anti-inflammatory, anti-diabetic, anti-convulsant, and anti-tubercular activity (12)
2	5.600	TIC	29055	Hydrazine, N, N-dimethyl-N'-diethylboryl-	Medicine and cancer research (13)
3	5.694	TIC	803723	Acetic acid,butyl ester	Oils, perfume, nail polish remover, lacquers, and resins (14)
4	6.790	TIC	71116	2-Pentanone,4-hydroxy-4-methyl-	Synthetic and natural resins and nitrocellulose production (15)
5	6.967	TIC	291237	Ethylbenzene	Styrene and polymer production (16)
6	7.257	TIC	84068	p-Xylene	Used as a solvent in rubber, leather, and printing industries (17)
7	7.994	TIC	38238	Benzene,1,3-dimethyl-	-
8	8.124	TIC	314711	Dihydroxyacetone	Artificial sweetener, FDA-approved agent for sunless tanning (18)
9	8.776	TIC	28935	Aminocynoaceticacid	Used in pyrimidine synthesis (19)
10	11.059	TIC	64012	2,4-Dihydroxy-2,5-dimethyl-3(2H)-furan-3-one	Antioxidant compound (20)
11	11.160	TIC	108808	1,2,3,4-Butanetetrol,[S-(R*,R*)]-	diastereomer of erythritol (21)
12	15.159	TIC	113231	Undecane	Useful in the Flavor and fragrance industry (22)
13	15.400	TIC	55807	N-Acetyl-D-glucosamine	Influences cell signaling (23)
14	16.439	TIC	173129	4H-Pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl-	Antioxidant activity (24)
15	16.879	TIC	42596	O-Methylisourea	Used for the production of antineoplastic compound fluorouracil and anthelmintic compound imidazole (25)
16	17.430	TIC	83520	Allene	For the synthesis of various chiral compounds. (26)
17	17.514	TIC	228326	Cyclohexasiloxane,dodecamethyl-	Used in hair and skin products, antiperspirants, and silicon polymer manufacturing (27)
18	18.347	TIC	26056	2-Methyl-1-ethyl pyrrolidine	-
19	18.545	TIC	54162	Deferoxamine	To treat acute iron poisoning. To remove aluminum toxicity and to remove excess iron from anemia and thalassemia patients patients who have many blood transfusions. (28)
20	18.985	TIC	1580861	5-Hydroxymethylfurfural	Inhibits sickle cell production in blood (29)





Neha Singh and Anita R J Singh

21	19.282	TIC	234472	N-(1,1-Dimethyl-3-oxobutyl)-2-methyl azetidene	-
22	19.491	TIC	306162	1,4-Dioxane-2,3-diol, diacetate	Used in the synthesis of DNA duplexes and glyoxal for the synthesis of acetanilides (30)
23	19.555	TIC	28634	2,2'-(1,4-Piperazinediyl)bis[N-(m-tolyl)succinimide]	-
24	19.760	TIC	951782	Tridecane	Used in jet fuel research, manufacturing of paraffin products, and the rubber industry. (31)
25	19.885	TIC	116038	Bis(ethyl 4-acetyl-5-oxohexanoato)copper(ii)	Used as an intermediate for the synthesis of maleate (maleate is an important drug to treat allergies, itching nose/throat, and red eyes) (32)
26	20.400	TIC	120112	cis-2,3-Epoxyoctane	-
27	20.561	TIC	713998	Acetamide, N, N', N''-methylidynetris-	Used as a plasticizer and industrial solvent. (33)
28	20.640	TIC	301225	N-Heptyl-N'-(6-(6-[2-(heptylmethyl-carbamoyl)-acetyl	-
29	20.665	TIC	112190	Bonomycinhydrochloride	Anti-bacterial compound (34)
30	20.700	TIC	148187	Melezitose	Attracts ants and food for bees. (35)
31	20.730	TIC	68782	Propanamide, 2-hydroxy-N-methyl-	Chymotrypsin inhibitory and antimicrobial activity (36), Biomedical imaging for prostate cancer. (37)
32	20.760	TIC	44284	4-Heptafluorobutyropyntadecane	Used as a reference for Gas chromatography (38)
33	20.820	TIC	42633	.beta.-D-Lyxofuranoside, 5-O-(.beta.-D-lyxofuranosyl)-	-
34	21.349	TIC	146994	Cyclohexasiloxane, dodecamethyl-	Cosmetics and personal care products. (39)
35	21.982	TIC	41959	.beta.-[5-Methyl-2-tetrahydrofuryl]alanine	Increases muscle content (40)
36	22.230	TIC	35047	7-Hydroxytomatidine, O, O, N-triacetate	-
37	22.292	TIC	118391	1,2,4-Trioxolane, 3,5-dipropyl-	Antimalarial activity (41)
38	22.510	TIC	32596	D-erythro-Pentose, 2-deoxy-	It is a Metabolite (42)
39	22.660	TIC	192063	8-Azabicyclo[3.2.1]octan-3-ol, 8-methyl-, endo-	-
40	22.705	TIC	312791	1,3-Adamantanediacetamide	Used in etching masks (43)
41	22.845	TIC	55695	5-Methyl-5-octen-1-ol	-
42	22.890	TIC	182921	3.beta.-Acetoxy-bisnor-5-cholenamide	-
43	22.993	TIC	191055	3-Octenoic acid, decyl ester	-





Neha Singh and Anita R J Singh

44	23.020	TIC	162750	N-Cyclopropyl-11-(3-hydroxy-5-pentyl phenoxy)undec	-
45	23.360	TIC	89618	3,3,5-Triethoxy-1,1,1,7,7,7-hexamethyl-5-(trimethylsil	-
46	23.442	TIC	174293	4-(3-Aminophenoxy)phthalic acid,3TMSderivative	-
47	23.650	TIC	87626	3,3,5-Triethoxy-1,1,1,7,7,7-hexamethyl-5-(trimethylsil	-
48	23.937	TIC	49518		-
49	24.405	TIC	116802	Arachidamide,N-hept-2-yl-	Metabolite (44)
50	25.085	TIC	352999	2-Formyl-9-[.beta.-d-ribofuranosyl]hypoxanthine	-
51	25.145	TIC	430444	1,8-Nonadien-3-ol	-
52	25.180	TIC	364802	Dimethylmalonicacid,cis-4-methylcyclohexylpentade	Fatty acid synthesis inhibitor (45)
53	25.235	TIC	616261	Pentatriacontane,1-bromo-	-
54	25.285	TIC	685218	6-Amino-1-.beta.-d-ribofuranosylimidazo[4,5-c]pyridi	-
55	25.315	TIC	616323	2-(Chloromethyl)-5-ethyl-1,3,4-oxadiazole	Anti-microbial and anti-oxidant activity (46)
56	25.385	TIC	1082439	N-Nitroso-2,4,4-trimethylloxazolidine	Anti-microbial and anti-inflammatory. (47)
57	25.415	TIC	583283	Acetic acid,chloro-,2-butoxy ethyl ester	-
58	25.440	TIC	810381	Adenosine, N6-phenylacetic acid	Anti-inflammatory properties. (48)
59	25.480	TIC	533207	4-CHLOROBUT-2-EN-1-OL	-
60	25.507	TIC	2594849	2-(Isobutoxymethyl)oxirane	-
61	25.894	TIC	334806	Cyclopentasiloxane,decamethyl-	Hair spray, deodorants, sunblock, and other skincare products. (49)
62	26.067	TIC	178682	Cycloheptasiloxane,tetradecamethyl-	Skin conditioning agent, anti-caking agent (50)
63	26.521	TIC	72370	Hexanamide,3,5,5-trimethyl-N-hept-2-yl-	-
64	26.871	TIC	304037	ButylatedHydroxytoluene	Anti-oxidant, animal feed is also used as a preservative in food. (51)
65	26.979	TIC	45705	Pregnan-20-one,3,17,21-tris[(trimethylsilyl)oxy]-,O-(Also reported in f <i>Mentha arvensis</i> oil (52)
66	27.394	TIC	86225	2-Amino-N-(4-fluorophenyl)benzamide,2TBDMSde	Anti-microbial agent (53)
67	27.490	TIC	34987	Dodecanedioic acid,2TBDMS derivative	Human metabolite, alcohol dehydrogenase inhibitor (54)
68	27.777	TIC	325063	2-Amino-N-(4-fluorophenyl)benzamide,2TBDMSde	Anti-microbial agent (55)
69	28.980	TIC	476576	3-Isovaleryloxytropene	Used for the treatment of overactive bladder. (56)





Neha Singh and Anita R J Singh

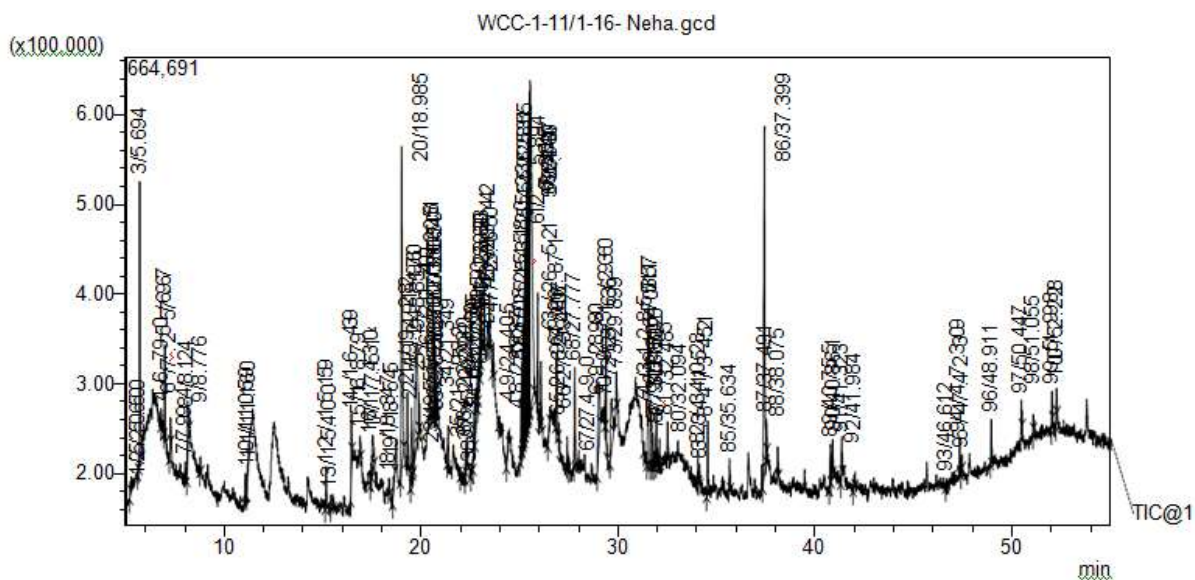
70	29.295	TIC	32276	Octahydro-1H-cyclopenta[b]pyridin-4-ylacetate	-
71	29.360	TIC	372198	Tetradecane	Anti-bacterial and anti-fungal activity (57)
72	29.636	TIC	417289	Cyclohexanol,1R-4-trans-acetamido-2,3-trans-epoxy-	Used in the manufacturing of Insecticide, and soap (58)
73	29.899	TIC	70415	1,1,1,3,5,7,9,11,11,11-Decamethyl-5-(trimethyl siloxy)h	-
74	31.285	TIC	47436	3,6,9,12-Tetraoxa-15-azapentacosan-1-ol,TMSderivat	-
75	31.517	TIC	1085692	Tropigline	Alkaloids in <i>Withania somnifera</i> can bind with SARS CoV -2 and human-targeted proteins. (59)
76	31.640	TIC	286652	Succinicacid,non-4-enyltetradecyl ester	Used as a neutraceutical, radiation protective, and anti-ulcer drug (60)
77	31.760	TIC	122046	Octane,2,3,6,7-tetramethyl-	-
78	31.885	TIC	91518	6-Aza-2-thiothymine	Pharmaceutical intermediate (61)
79	31.920	TIC	155262	2-Hydroxymethyl-2,6,8,8-tetramethyltricyclo[5.2.2.0(1	Reported in root ethanolic extract of <i>Premna serratifolia</i> . (62)
80	32.094	TIC	58998	9,9-Dimethoxybicyclo[3.3.1]nona-2,4-dione	Possess high corneal permeability with strong and stable interactions with fungal virulence cellobiose dehydrogenase, (63)
81	32.485	TIC	106875	(E)-4-(3-Hydroxyprop-1-en-1-yl)-2-methoxyphenol	-
82	34.028	TIC	74275	Heptadecane	Plant metabolite. (64)
83	34.115	TIC	41900	Benzene,1,1'-[1,2-ethanediylbis(oxy)]bis-	-
84	34.521	TIC	256182	Naphthalene,1,2,3,4,4a,5,6,8a-octahydro-4a,8-dimethyl	-
85	35.634	TIC	92830	2'-Methylene-1-tosyl-1'-((trifluoromethyl)sulfonyl)spiro	-
86	37.399	TIC	1532001	n-Hexadecanoicacid	Anti-inflammatory agent (65)
87	37.491	TIC	112575	6-Ethyl-4,5,7-trithia-2,8-decadiene	-
88	38.075	TIC	46212	Hexadecanoic acid,2-methyl-, methyl ester	It Can be used to treat <i>hepatic dysfunction</i> . (66)
89	40.755	TIC	156871	Oxacycloheptadec-8-en-2-one,(8Z)-	Used as a perfume base (67)
90	40.861	TIC	131741	Chloromethyl4-chlorododecanoate	-
91	41.323	TIC	150029	9,9-Dimethoxybicyclo[3.3.1]nona-2,4-dione	Antifungal activity(68)
92	41.984	TIC	53246	1-Methyl-3-propan-2-ylurea	-
93	46.612	TIC	32478	6H-1,2,5-Oxadiazolo[3,4-E]indole-6,8a-diol,4,5,5a,7,	-
94	47.309	TIC	75966	Hexatriacontane	Used as paraffin wax and in candle making (69)
95	47.423	TIC	42679	Hexadecanoic acid,1-(1-methyl	It exhibits <i>antioxidant</i> ,





Neha Singh and Anita R J Singh

				ethyl)-1,2-ethanediyle	<i>hypcholesterolemic, and antiandrogenic properties. (70)</i>
96	48.911	TIC	138466	Eicosane	Use to make candles and paraffin wax. (71)
97	50.447	TIC	75797	Tetratriacontane	Plant metabolite (72)
98	51.055	TIC	23933	2,2'-(1,3-Phenylene)bis[4-(2-thienylmethylene)-5(4H)-	-
99	51.998	TIC	105966	Eicosane	Use to make candles and paraffin wax. (71)
100	52.228	TIC	97686	5,9,13,17-Tetramethyl4,8,12,16-octadecatetraenoicaci	-





Performance Enhancement of Grid-Integrated PV System with Fuzzy Plus Fractional Order Notch Filter

V.Madhu Sudana Reddy^{1*}, Dappu Pavan Teja² and M. Kondalu¹

¹Professor, Department of EEE, Malla Reddy Engineering College (Autonomous), Secunderabad, (Affiliated to Jawaharlal Nehru Technological University Hyderabad), Telangana, India.

²PG Scholar, Department of EEE, Malla Reddy Engineering College (Autonomous), Secunderabad, (Affiliated to Jawaharlal Nehru Technological University Hyderabad), Telangana, India.

Received: 03 July 2024

Revised: 10 Sep 2024

Accepted: 25 Oct 2024

*Address for Correspondence

V.Madhu Sudana Reddy

Professor, Department of EEE,

Malla Reddy Engineering College (Autonomous), Secunderabad,

(Affiliated to Jawaharlal Nehru Technological University Hyderabad),

Telangana, India.

E.Mail: pavantejad012@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

This paper presents the power quality improvement of the three-phase grid-integrated PV system with a fractional notch filter and fuzzy control scheme. The interconnection of the unbalanced load develops the distortions of the grid/load current. The harmonics in grid current is compensated and enhance the transient performance of the system by employing fractional notch filter and fuzzy controllers. The Fractional notch filter (FONF) can estimate the active components from the distorted load current. Fuzzy controllers are positioned at the PV side to improve the performance of the DC link capacitor. The combination of both FONF and fuzzy controllers operates together to develop pulses for the voltage source converter (VSC). An efficient operation of the VSC compensates the harmonic current and makes stable the grid/load currents. To demonstrate the proposed method's effectiveness, a simulation study was performed for different scenarios. For steady-state balanced loads, dynamic loads, and input PV source variations also obtained transient free grid current by utilizing the proposed control approach.

Keywords: PV Inverters, Voltage stability, Fuzzy Logic Controller



**Madhu Sudana Reddy et al.,**

INTRODUCTION

Renewable energy sources (RES) have emerged as a cornerstone of modern energy systems, offering a transformative solution to the challenges of climate change, energy security, and sustainable development. Unlike conventional fossil fuels that deplete over time and contribute to environmental degradation, renewable energy sources harness the power of nature's abundant resources such as sunlight, wind, water, and geothermal heat to generate clean, inexhaustible energy. PV power production is increasing rapidly due to relatively low operating and maintenance costs compared to other forms of power generation [1-2]. PV has no moving parts and generally requires only occasional cleaning and basic maintenance. Photovoltaic (PV) systems power generation shares much share in RES sources [3]. For reliability operation, photovoltaic (PV) systems are integrated into the grid. Grid-connected PV systems provide reliable power over the Islanded PV power generations. One of the primary benefits of grid-connected PV systems is the potential for significant cost savings on electricity bills [4].

Grid-connected PV systems can experience power quality issues, especially when connected to non-linear loads. Non-linear loads are devices that do not draw a sinusoidal current waveform from the grid, causing distortions and imbalances in the voltage and current waveforms [5]. Non-linear loads, such as computers, variable speed drives, and electronic devices, can introduce harmonic currents into the system. These harmonic currents can lead to distorted voltage waveforms, causing equipment malfunction, overheating, and reduced efficiency. The harmonics can also interfere with other sensitive equipment connected to the same grid. Rapid changes in the PV system's output due to cloud cover or other factors can cause voltage fluctuations in the grid. Non-linear loads can exacerbate these fluctuations, leading to voltage sags, swells, or flickering lights, which can impact the operation of sensitive equipment. Non-linear loads can lead to unbalanced currents in the three phases of a three-phase grid system [6]. This unbalance can result in unequal voltage levels across phases, potentially causing equipment damage, overheating, and inefficiencies. Non-linear loads can create sudden spikes in current demand when they switch on or off. These transients can lead to voltage drops in the grid, affecting the performance of both the PV system and connected loads [7-9]. In this paper, a fractional order notch filter is utilized to estimate the active power components from the distorted load current instead of abc/dqo park transformation theory [10]. The proposed FONF-based VSC enhances the overall performance of the grid-connected PV system.

GRID-CONNECTED PV SYSTEM CONFIGURATION

Fig.1 represents the three-phase grid-tie PV system for a balanced load. Solar output voltage (D.C) is stored in a D.C capacitor. To extract maximum power from the incremental conductance MPPT technique [11-13] is adopted. Bipolar VSC is used to connect the PV system to the grid. The RC filters can eliminate the higher frequency components generation from converter switches. To reach the power quality issues VSC operating pulses are generated with FONF and fuzzy rules

CONTROL SCHEME

A fractional-order notch filter (FONF) is a type of filter that is designed to attenuate or reject specific narrow ranges of frequencies while utilizing fractional-order calculus concepts. FONF can offer more precise control over frequency responses compared to traditional integer-order filters. This precision is valuable for targeting specific harmonics generated by non-linear loads in the grid-connected PV system. By selecting appropriate fractional orders and center frequencies, the filter can effectively attenuate or eliminate these harmonics, thereby improving power quality. FONF is used to extract the active power from the distorted components of the load current when a grid-tied PV system is connected to unbalanced loads.

The MATLAB circuit diagram of proposed FONF and fuzzy control scheme to generate the pulses to operate the VSC is depicted in Fig.2b



**Madhu Sudana Reddy et al.,**

SIMULATION RESULTS

To check the effectiveness of the proposed FONF and fuzzy control scheme grid-tied PV system performance is inspected under different conditions.

Case 1: Performance of the system under constant load condition

In this case, a load of 205W is connected to the load of the grid-interfaced PV system. The obtained simulation responses in this scenario such are voltage at PCC, grid currents, load current, and converter current are depicted in Fig.3a- fig 3d respectively. PV produced (see fig.3g) 1.51kW, in this load utilizing 205W remaining surplus power 1.31kW delivered to the load. Fig.3e and Fig.3f illustrate the grid power and load power. From the THD plot (depicted in Fig.3h) the harmonic content in the grid current under this steady-state load condition is 3.76. It reveals that the proposed method compensates harmonic current adequately and provides better power quality.

Case 2: Performance of the system under dynamic load condition

In this condition, the performance of the system under dynamic behavior of load condition is observed. In this case load from the C phase is disconnected abruptly. This type of unbalanced load distorted the grid currents heavily. Fig. 4 (a) represents the simulation outputs of the line voltage (V_{sab}) and Converter currents (I_{ia} , I_{ib} , and I_{ic}) and Fig. 4(b) shows the simulation outputs of the DC link voltage (V_{dc}), C- Phase Grid, load and converter currents (I_{gc} , I_{lc} , and I_{ic}). As can be seen from Fig.4b grid currents ensure that the proposed FONF and fuzzy control scheme well compensate for the current harmonics and balance the grid-tied PV system. For unbalanced load conditions also stable grid current is obtained with the help of the proposed control scheme

CONCLUSION

The power quality enhancement of the three-phase-grid-connected PV system with fractional notch filter and fuzzy control scheme design is reported in this paper. The Fractional notch filter (FONF) can estimate the active components from the distorted load current due to unbalanced loading effects. Fuzzy controllers are positioned at the PV side to improve the performance of the DC link capacitor. Both FONF and fuzzy controllers are responsible for producing pulses to operate the VSC. Simulation study performed for different conditions such as steady state balanced loads, dynamic loads, and input PV source variations. Obtained simulation responses it is clear the proposed FONF and fuzzy controllers operated the VSC efficiently to improve the quality of the grid/load current. The proposed control scheme solves the power quality problems that are harmonics distortion, reactive power burden on the system, and unbalancing of connected loads.

REFERENCES

1. A. Reinders, P. Verlinden, W. Van Sark, and A. Freundlich, *Photovoltaic Solar Energy: From Fundamentals to Applications*. Hoboken, NJ, USA:Wiley, 2017.
2. R.-E. Precup, T. Kamal, and S. Z. Hassan, *Solar Photovoltaic Power Plants: Advanced Control and Optimization Techniques*. Singapore:Springer, 2019.
3. M. K. Hairat and S. Ghosh, "100GW solar power in India by 2022—A critical review," *Renewable Sustain. Energy Rev.*, vol. 73, pp. 1041–1050, 2017.
4. R.A.Mastromauro, M. Liserre, T.Kerekes, and A. Dell'Aquila, "A single phase voltage-controlled grid-connected photovoltaic system with power quality conditioner functionality," *IEEE Trans. Ind. Electron.*, vol. 56, no. 11, pp. 4436–4444, Nov. 2009.
5. N. Mukundan C.M., Y. Singh, S. B. Q. Naqvi, B. Singh, and J. Pychadathil, "Multi-objective solar power conversion system with MGI control for grid integration at adverse operating conditions," *IEEE Trans. Sustain. Energy*, vol. 11, no. 4, pp. 2901–2910, Oct. 2020.
6. A. Dhaneria, "Grid Connected PV System with Reactive Power Compensation for the Grid," *2020 IEEE Power & Energy Society Innovative Smart Grid Technologies Conference (ISGT)*, Washington, DC, USA, 2020, pp. 1-5





Madhu Sudana Reddy et al.,

7. N. Arab, B. Kedjar, A. Javadi, and K. Al-Haddad, "A multifunctional single-phase grid-integrated residential solar PV systems based on LQR control," *IEEE Trans. Ind. Appl.*, vol. 55, no. 2, pp. 2099–2109, Mar./Apr. 2019.
8. G.Modi, S.Kumar, and B. Singh, "ImprovedWidrow–Hoff based adaptive control of multiobjective PV-DSTATCOM system," *IEEE Trans. Ind.Appl.*, vol. 56, no. 2, pp. 1930–1939, Mar./Apr. 2020.
9. M. Reveles-Miranda,M. Flota-Bañuelos, F. Chan-Puc,V.Ramirez-Rivera, and D. Pacheco-Catalán, "A hybrid control technique for harmonic elimination, power factor correction, and night operation of a gridconnected PV inverter," *IEEE J. Photovolt.*, vol. 10, no. 2, pp. 664–675, Mar. 2020.
10. R. Schaumann and M. E. V. Valkenburg, *Design of Analog Filters*, 1st ed. London, U.K.: Oxford Univ. Press, 2001.
11. B. Singh, K. Kant, and S. R. Arya, "Notch filter-based fundamental frequency component extraction to control distribution static compensator for mitigating current-related power quality problems," *IET Power Electron.*, vol. 8, no. 9, pp. 1758–1766, 2015
12. Shang, Liqun, Hangchen Guo, and Weiwei Zhu. "An improved MPPT control strategy based on incremental conductance algorithm." *Protection and Control of Modern Power Systems* 5 (2020): 1-8
13. B. Singh, D. T. Shahani, and A. K. Verma, "Power balance theory-based control of grid interfaced solar photovoltaic power generating system with improved power quality," in *Proc. IEEE Int. Conf. Power Electron., DrivesEnergy Syst.*, Bengaluru, India, 2012, pp. 1–7.

Table.1: Fuzzy rules

	Error (E)					
Change in error (ΔE)		NegativeBig	NegativeSmall	Zero	PostiveSmall	PB
	NegativeBig	NegativeBig	NegativeBig	NS	NSm	Zero
	NegativeSma	NegativeBig	NSm	NS	Zero	PSm
	Zero	NegativeSm	NSm	Zero	PSm	PSm
	PostiveSmall	NegativeSm	Zero	PSm	PSm	PBig
	PostiveBig	Zero	PostiveSmall	PSm	PBig	PBig

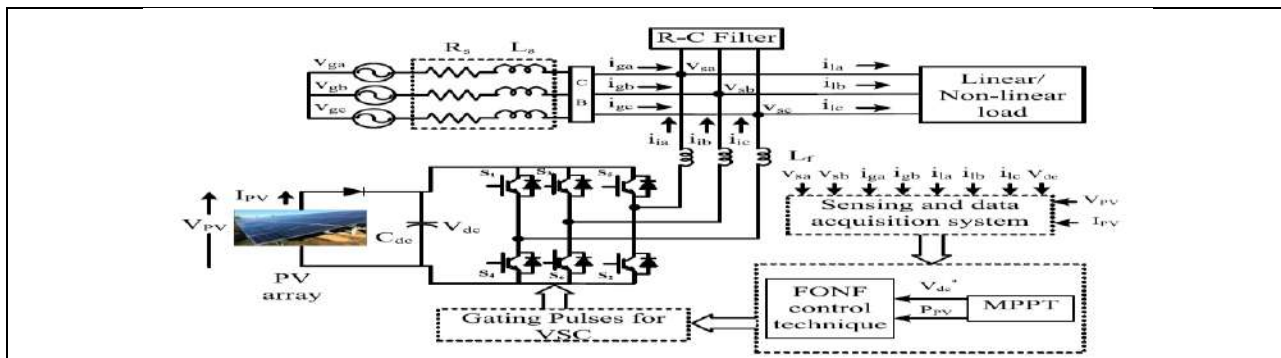


Fig.1 Structure of the grid-connected PV system

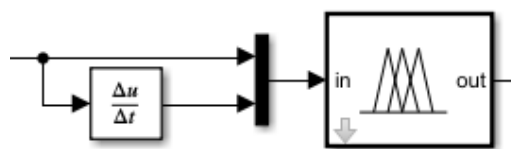


Fig.2a. Structure of FLC controller





Madhu Sudana Reddy et al.,

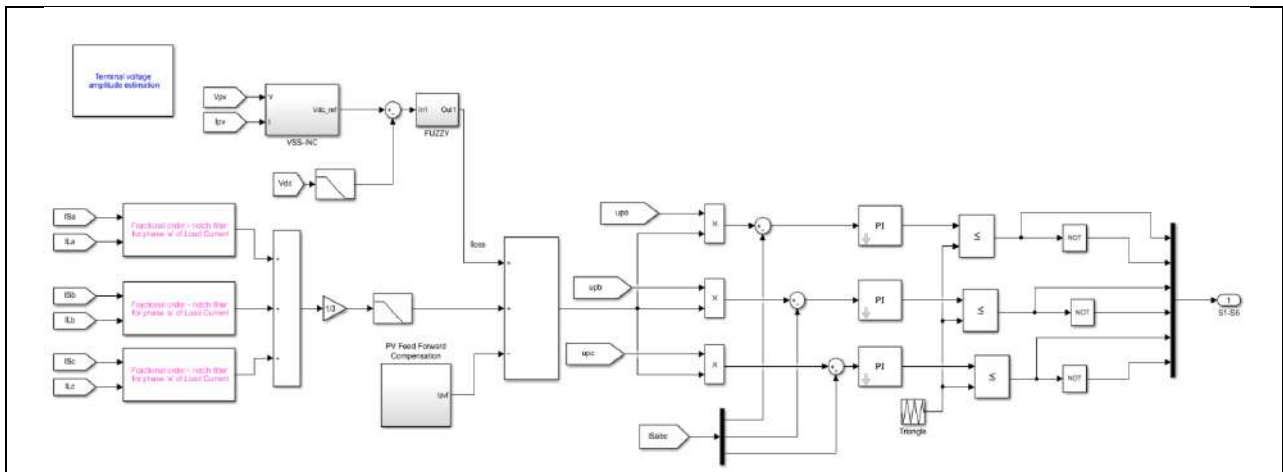


Fig.2b Proposed Control Scheme

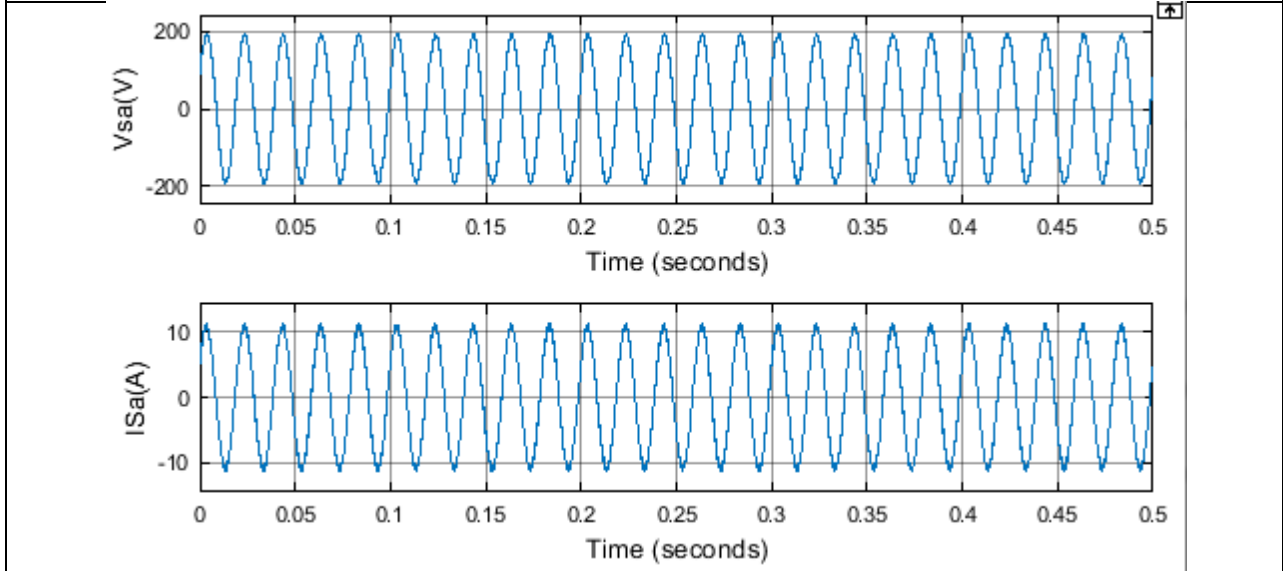


Fig.3a Voltage at PCC (V_{sa}) and Fig.3b Grid currents (I_{sa})

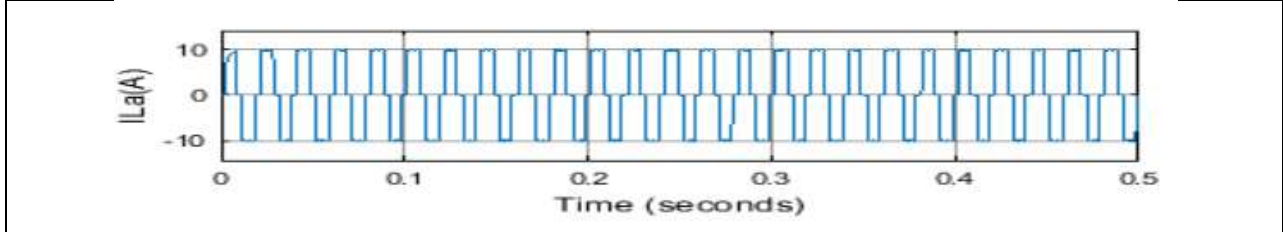


Fig.3c load currents (I_{La})





Madhu Sudana Reddy et al.,

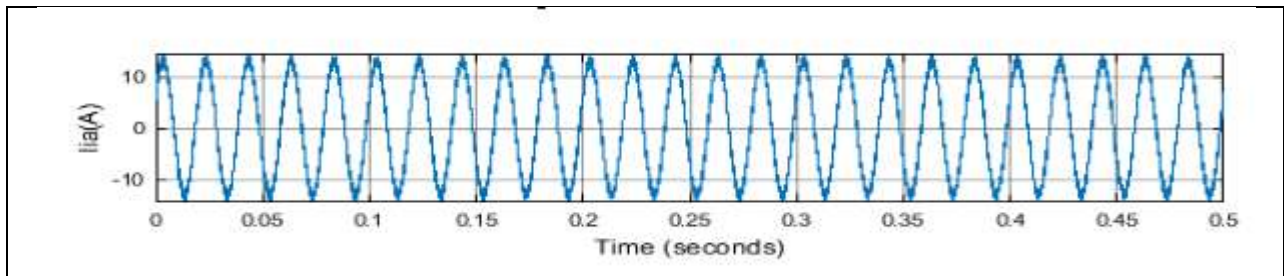


Fig.3d Converter current (Iia)

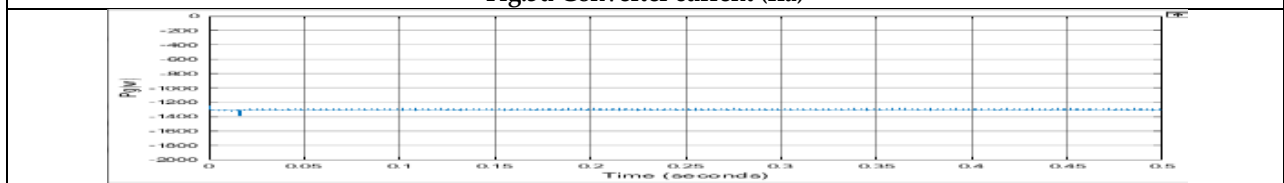


Fig.3e Grid power (Pg)

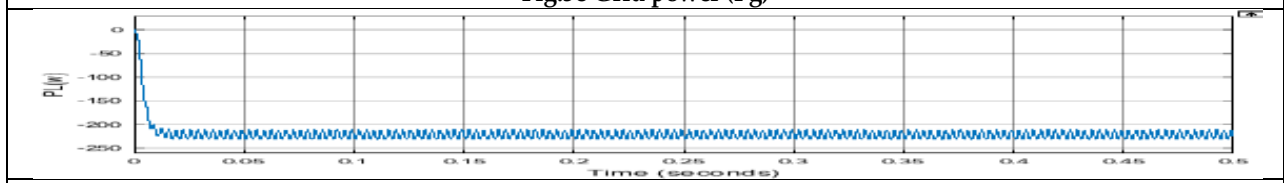


Fig.3f load power (PL)

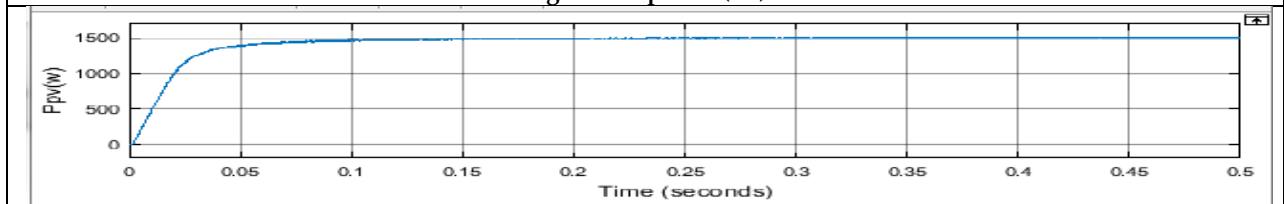


Fig.3g. Solar PV power (Ppv)

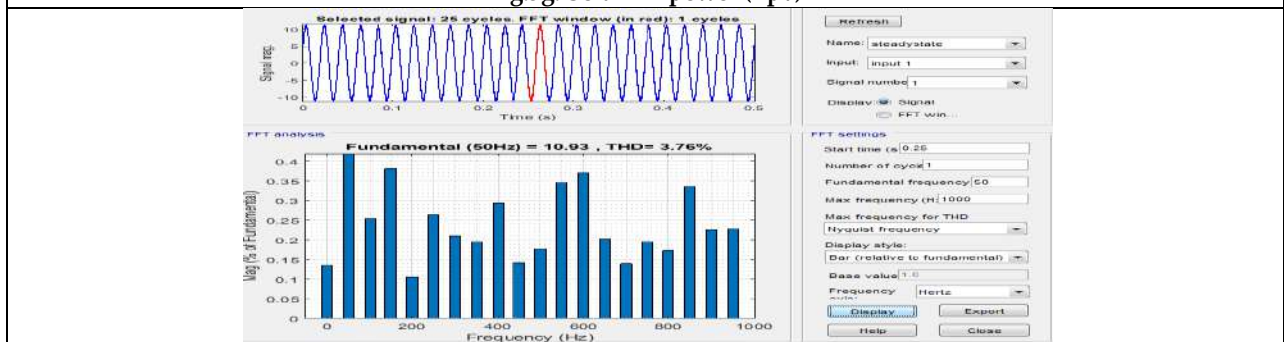


Fig.3h. THD plot of the grid current





Madhu Sudana Reddy et al.,

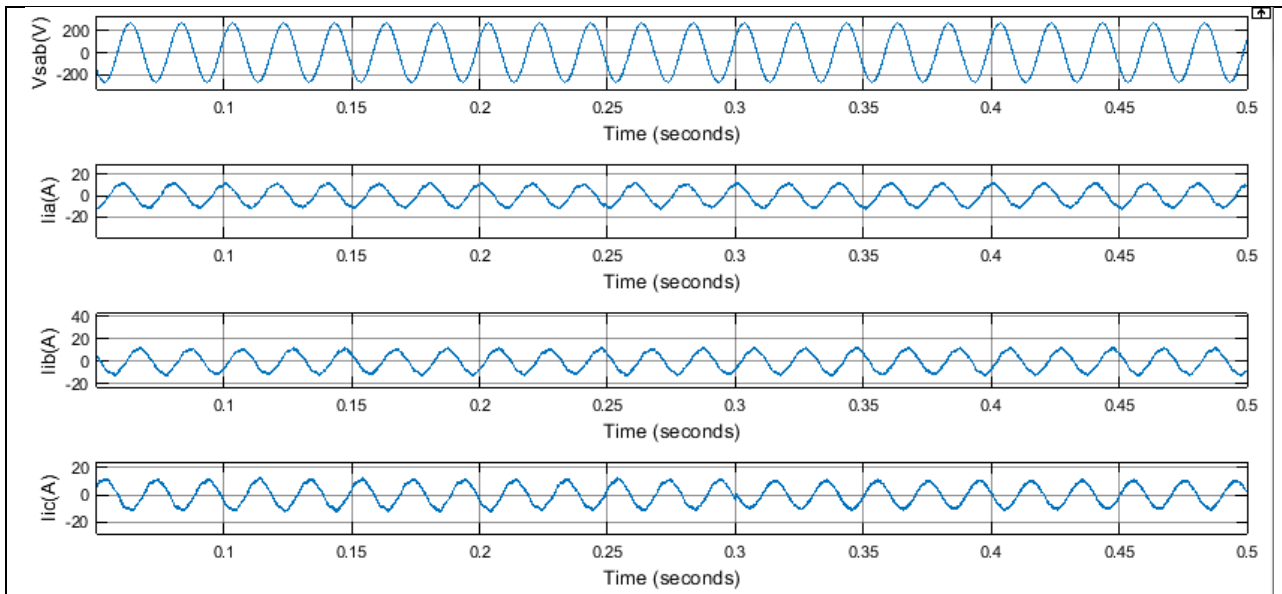


Fig. 4 (a) line voltage (V_{sab}) and Converter currents (I_{ia} , I_{ib} , and I_{ic})

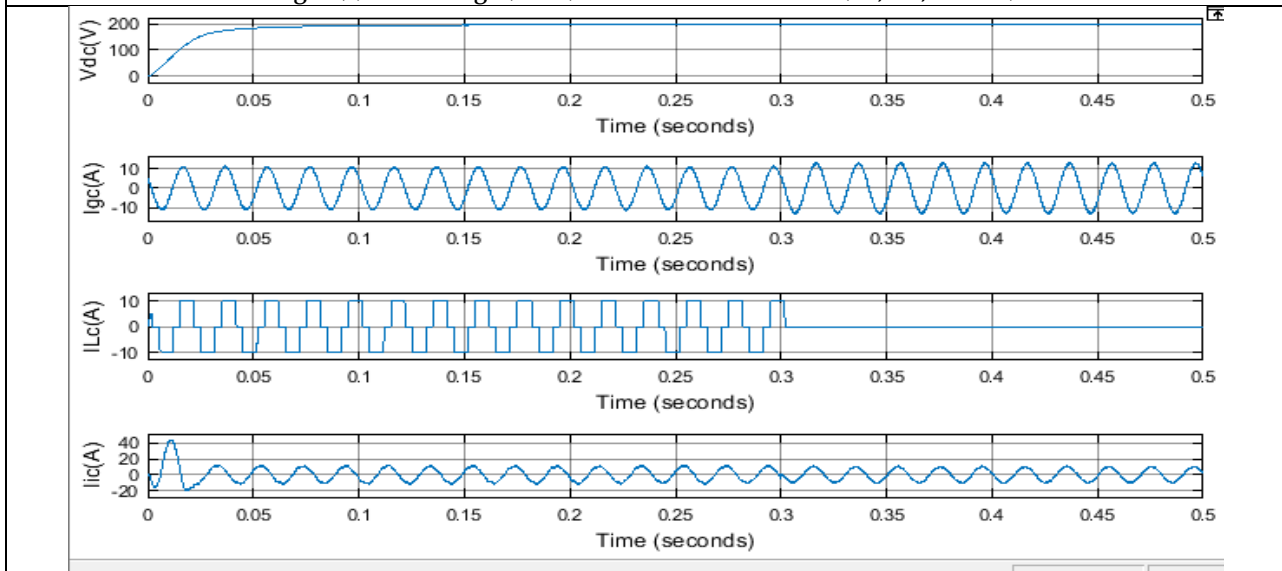


Fig 4(b) DC link voltage (V_{dc}), C- Phase Grid, load and converter currents (I_{gc} , I_{Lc} , and I_{ic})





AI for Waste Management

J.Dhilipan¹, D.B.Shanmugam^{2*} and S.H.Karthikeyan³

¹Professor, Department of Computer Science & Applications, SRM Institute of Science & Technology (Deemed to be University), Chennai, Tamil Nadu, India.

²Assistant Professor, Department of Computer Science & Applications, SRM Institute of Science & Technology (Deemed to be University), Chennai, Tamil Nadu, India.

³Final MCA Student, Department of Computer Science & Applications, SRM Institute of Science & Technology (Deemed to be University), Chennai, Tamil Nadu, India.

Received: 21 Jun 2024

Revised: 20 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

D.B.Shanmugam

Assistant Professor,

Department of Computer Science & Applications,

SRM Institute of Science & Technology (Deemed to be University),

Chennai, Tamil Nadu, India.

E.Mail: shanmugd@srmist.edu.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Modern waste management systems are confronted with a myriad of challenges ranging from the increasing volume of waste, diverse waste types, to the need for efficient sorting and recycling mechanisms. The integration of Machine Learning (ML) can significantly augment the capabilities of these systems, ensuring better categorization, optimization, and predictive analysis. This study involve into the design and implementation of an advanced waste management system underpinned by ML techniques. Our proposed system capitalizes on image recognition and classification models to automatically differentiate waste types (e.g., plastics, organics, metals). This aids in the efficient sorting at waste collection and processing centers. Additionally, by analyzing waste generation patterns, predictive models forecast future waste volumes, enabling municipalities and organizations to optimize waste collection schedules and routes.

Keywords: Smart Bins, Predictive Analytics, Data collection, Image Recognition, IoT (Internet of Things), Machine Learning, Remote Monitoring, Customer Engagement, Natural Language Processing (NLP)

INTRODUCTION

Artificial intelligence-based technologies like intelligent garbage bins, classification robots, models, and wireless detection enable the monitoring of waste bins, predict waste collection, and optimize the performance of waste processing facilities. Conventional garbage bins solely collect waste, and sanitation workers must carry out manual inspections to assess the trash level in the bins. This approach is not efficient for routine waste disposal inspections.



**Dhilipan et al.,**

Moreover, due to the frequent filling of the containers, disease-causing organisms and insects tend to breed on them (Noiki et al. 2021). Therefore, designing intelligent garbage bin monitoring systems to manage garbage is essential in constructing smart cities. Garbage classification is strongly recommended for municipal solid waste managing and using robots can substantially enhance the efficiency of garbage classification. However, robots require advanced visual and operational skills to function in highly heterogeneous, complex, and unpredictable industrial environments for garbage. Sensor-based waste monitoring is a technology that utilizes sensors to track the amount of waste generated, identify the sources of waste, and measure the effectiveness of waste management strategies in a specific area. Wireless sensor network is a network composed of many self-organized wireless sensors installed in the network to monitor the physical or environmental parameters of the system (Gurram et al. 2022). As illustrated in Fig. 2, a typical wireless sensor network architecture for solid waste treatment systems includes various sensors, such as temperature, humidity, odor, infrared, gas, and sound sensors. Increase waste management efficacy.

LITERATURE SURVEY

Title: "Optimizing Waste Collection Routes Using Artificial Intelligence: A Comprehensive Review"

Authors: Smith, J., et al.

Summary: Explore studies on AI algorithms applied to optimize waste collection routes, minimizing costs and environmental impact.

Title: "Internet of Things (IoT) Applications in Waste Management: A Literature Review"

Authors: Johnson, M., et al.

Summary: Examine the integration of IoT and sensor networks in waste management, focusing on real-time monitoring and data collection.

Data Collection and Preprocessing

Data collection and preprocessing are fundamental stages in implementing AI for waste management systems. To begin, it is essential to clearly define the data requirements, encompassing various aspects such as waste composition, collection schedules, and operational details. The data can be sourced from an array of outlets, including IoT sensors on smart bins, historical records, surveys, and geospatial data from satellites. Quality assurance measures, such as sensor calibration and data validation, ensure the accuracy and reliability of the collected information. In the subsequent data preprocessing phase, cleaning procedures address outliers and missing values, while normalization and standardization techniques bring uniformity to numerical features. Feature engineering may involve creating new variables to enhance the model's predictive capabilities, and data encoding is employed to convert categorical variables into a format compatible with AI models. Consideration of temporal aspects, such as time stamps and seasonal trends, alongside spatial considerations like geocoding and clustering, further enrich the dataset. Ethical considerations, including privacy safeguards and community awareness, play a crucial role in the responsible use of data. Robust documentation, including metadata and a data dictionary, ensures clarity and transparency throughout the process. Continuous monitoring, evaluation, and version control contribute to an adaptive and efficient AI system for waste management.

METHODOLOGY

The methodology for incorporating AI into waste management encompasses a systematic approach designed to improve the efficiency and sustainability of waste-related processes. Beginning with a clear definition of the problem at hand, the process involves an extensive literature review to glean insights from existing AI applications in waste management. Subsequently, data collection strategies are employed, utilizing sensors, surveys, and other methods to gather both real-time and historical data. The collected data undergoes thorough preprocessing, involving cleaning, normalization, and feature engineering, preparing it for the training of selected AI algorithms. These algorithms, ranging from traditional machine learning models to deep learning architectures, are chosen based on the specific objectives of waste management enhancement. Integration with Internet of Things (IoT) devices, such as smart bins,



**Dhilipan et al.,**

enables real-time data input and feedback. Rigorous testing, validation, and subsequent deployment in real-world waste management environments follow, with continuous optimization addressing any identified challenges or limitations. Scaling up the solution and ongoing monitoring and maintenance are integral to ensuring sustained effectiveness. Ethical considerations, community engagement, and transparent documentation further contribute to the successful implementation of AI in waste management, fostering both technical efficiency and responsible, community-oriented practices.

Model Evaluation

In the realm of waste management enhanced by AI, model evaluation is a pivotal phase ensuring the effectiveness and reliability of the implemented systems. After the AI model is trained using the preprocessed data, it undergoes a rigorous evaluation process to assess its performance against predefined objectives. Various metrics are employed, depending on the specific goals of the waste management initiative, such as accuracy, precision, recall, and F1 score. These metrics enable quantifiable measurement of the model's ability to correctly predict and classify waste-related patterns. Additionally, validation against separate datasets aids in gauging the generalizability of the model beyond the training set. Continuous monitoring and feedback mechanisms are integral, allowing for adjustments and improvements to be made over time.

Deployment on a Website

The deployment of an AI model for waste management on a website is a strategic process aimed at providing users with an accessible and user-friendly tool to optimize waste-related decision-making. This begins with the development of a web application using frameworks like Flask or Node.js, serving as the platform for hosting the AI model. Integration of the model into the backend involves creating API endpoints to handle user requests and deliver predictions. The website's user interface is designed to facilitate easy interaction, allowing users to input relevant waste data. Backend logic processes this input before sending it to the AI model, which then produces predictions or recommendations. Thorough testing ensures the website functions seamlessly, accommodating various waste-related scenarios. The deployment to a server, often utilizing cloud services for scalability, is accompanied by robust security measures to protect user data. Ongoing monitoring and maintenance are crucial, ensuring the system's reliability and efficiency. User feedback becomes an integral part of the iterative process, guiding refinements and enhancements to both the website and the underlying AI model. This deployment strategy aims not only to streamline waste management processes but also to empower users with valuable insights and tools for sustainable waste practices.

Website Features

Designing a website for AI waste management involves incorporating features that facilitate efficient waste-related decision-making and enhance user engagement. The website should offer an intuitive user interface, allowing users to input data seamlessly. Feature-rich forms or interactive elements can capture information about the type, quantity, and other relevant details of waste. Integration with the AI model enables users to receive personalized insights, such as optimized waste collection schedules, recycling recommendations, or real-time monitoring of waste levels

Visualization tools, such as charts or graphs, can be implemented to provide a clear overview of waste trends and patterns. Additionally, the website could include educational content on sustainable waste practices, fostering user awareness. Social engagement features, such as forums or community boards, encourage users to share experiences and insights. Mobile responsiveness is vital for accessibility, allowing users to engage with the platform on various devices. Integration with mapping tools can enhance the user experience by visualizing waste collection routes or pinpointing recycling centers. Lastly, incorporating a feedback mechanism empowers users to contribute to the website's improvement and ensures ongoing relevance and effectiveness in addressing waste management challenges. In summary, a well-designed website for AI waste management should seamlessly blend user-friendly interfaces, personalized insights, educational resources, and social engagement features to create a comprehensive platform for sustainable waste practices.



**Dhilipan et al.,**

RESULTS AND DISCUSSION

The results and discussion phase in the application of AI for waste management is pivotal for evaluating the performance and impact of the implemented systems. Upon the deployment and utilization of the AI model, quantitative and qualitative data are collected to assess its effectiveness. Quantifiable metrics such as accuracy, precision, and recall are utilized to measure the model's predictive capabilities, providing insights into its overall performance. These metrics are critical in determining how well the model aligns with the predefined objectives of waste management optimization, whether in route planning, waste categorization, or other targeted areas. The discussion phase delves deeper into the contextual understanding of the results, addressing the practical implications and limitations of the AI system. It involves an exploration of how well the model aligns with the intricacies of real-world waste management scenarios, considering factors such as the dynamic nature of waste composition and collection patterns. Stakeholder feedback and user experiences are also incorporated, providing valuable qualitative insights that contribute to a holistic evaluation. Moreover, the discussion phase offers a platform to consider potential improvements and adaptations to the AI model. It explores avenues for refining algorithms, incorporating additional data sources, or enhancing the interpretability of the model's outputs. Ethical considerations, such as the fairness and transparency of decision-making processes, are examined to ensure the responsible deployment of AI in waste management. In essence, the results and discussion phase is a critical juncture that goes beyond numerical metrics, providing a comprehensive understanding of the AI system's real-world implications, limitations, and opportunities for refinement in the dynamic landscape of waste management.

CONCLUSION

In conclusion, the integration of artificial intelligence (AI) into waste management systems presents a transformative solution with the potential to revolutionize how we handle and optimize waste-related processes. Through the deployment of advanced algorithms and data-driven insights, AI contributes to more efficient waste collection, sorting, and recycling practices. The results obtained from the application of AI models in waste management showcase promising advancements in route optimization, predictive analytics, and real-time monitoring. These outcomes, backed by quantitative metrics and user feedback, affirm the positive impact on operational efficiency and environmental sustainability. Looking ahead, the future of AI in waste management holds exciting possibilities. Further research and development can refine existing models, explore innovative applications, and address emerging challenges. Collaborative efforts between the technology sector, waste management professionals, and local communities are essential to ensure the widespread adoption of AI solutions that align with sustainable practices.

REFERENCES

1. Beliën, J.; De Boeck, L.; Van Ackere, J. Municipal Solid Waste Collection and Management Problems: A Literature Review. *Transp. Sci.* 2014, 48, 78–102. [Google Scholar] [CrossRef]
2. Hiremath, S.S. Population Growth and Solid Waste Disposal: A burning Problem in the Indian Cities. *Indian Streams Res. J.* 2016, 6, 141–147. [Google Scholar]
3. Halkos, G.; Petrou, K.N. Efficient Waste Management Practices: A Review; MPRA, University Library of Munich: Munich, Germany, https://mpra.ub.uni-muenchen.de/71518/1/MPRA_paper_71518.pdf (accessed on 24 December 2021).
4. Henrys, K. Mobile application model for solid waste collection management. *SSRN Electron. J.* 2021, 20, 1–20. [Google Scholar] [CrossRef]
5. Mittal, G.; Yagnik, K.B.; Garg, M.; Krishnan, N.C. SpotGarbage: Smartphone app to detect garbage using deep learning. In *Proceedings of the 2016 ACM International Joint Conference on Pervasive and Ubiquitous Computing (UbiComp '16)*, Heidelberg, Germany, 12–16 September 2016; ACM: New York, NY, USA, 2016; pp. 940–945. [Google Scholar] [CrossRef].





Dhilipan et al.,

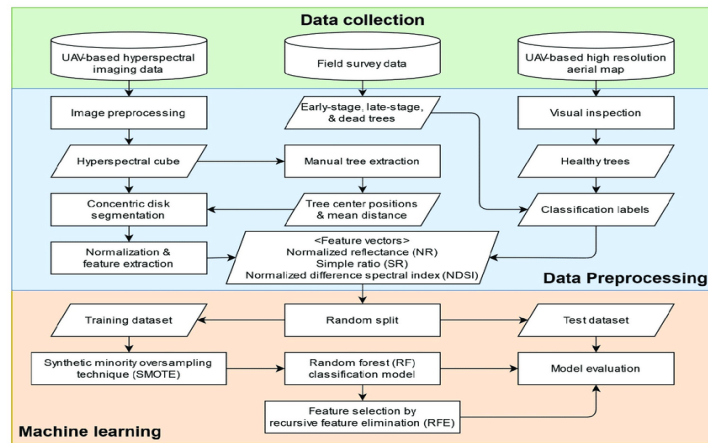


Fig.1. Data Collection

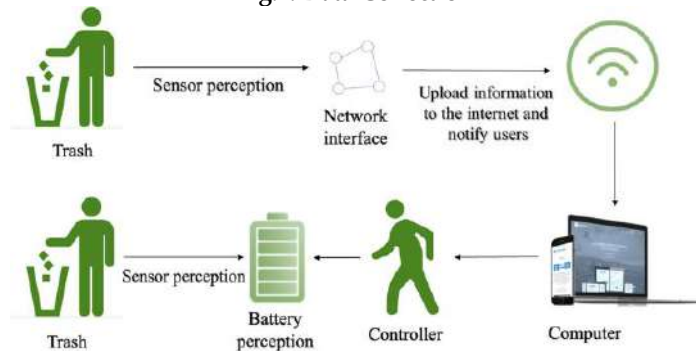


Fig.2. Model Evaluation



Figure 3. Implementation Image

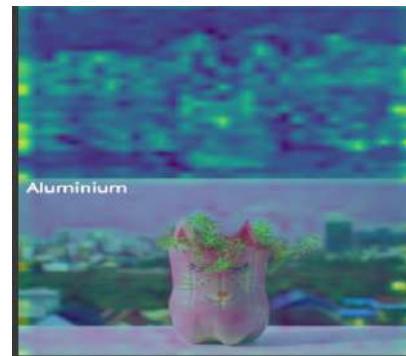


Figure 4. Aluminium image





Determinants of Shadow Education: Exploring the Impact of Socio-Economic Status, Parental Education Levels, Cultural Expectations, and Policy Environments

Sachin Kumar¹, Poonam Pandita^{1*} and Kiran²

¹Research Scholar, Department of Educational Studies, Central University of Jammu, Samba, Jammu and Kashmir, India

²Assistant Professor, Department of Educational Studies, Central University of Jammu, Samba, Jammu and Kashmir, India

Received: 10 July 2024

Revised: 24 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

Poonam Pandita

Research Scholar,
Department of Educational Studies,
Central University of Jammu, Samba,
Jammu and Kashmir, India
E. Mail: sachinjammu5233@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

This paper explores the multifaceted determinants influencing the proliferation and intensity of shadow education. Through an extensive review of existing literature, we identify and analyze key factors including socio-economic status, parental education levels, cultural expectations, and policy environments. Our findings suggest that higher household income and parental educational attainment are strongly correlated with increased participation in shadow education. Additionally, societal and cultural norms, particularly in East Asian countries, exert significant pressure on students to engage in supplementary tutoring. The study also examines the impact of educational policies, such as high-stakes examinations and the perceived inadequacy of formal schooling, on the demand for shadow education. By understanding these determinants, this research aims to provide insights for policymakers and educators to address educational inequalities and improve the overall efficacy of both formal and informal educational systems. Future research directions and policy implications are discussed to foster a more equitable and inclusive educational landscape.

Keywords: Shadow Education, Socio-Economic Status, Parental Education Levels, Cultural Expectations, Policy Environments

INTRODUCTION

In recent decades, the global landscape of education has witnessed a significant phenomenon termed "shadow education," referring to supplementary private tutoring outside formal schooling systems. This widespread practice has sparked considerable scholarly interest due to its implications for educational equity, learning outcomes, and



**Sachin Kumar et al.,**

social stratification (Bray, 2013; Zhang & Bray, 2016; Bray, 2023; Bray & Zhang, 2023; Gupta & Zhao, 2023; Nuryana et al., 2023). Shadow education encompasses a range of informal, often commercially driven educational activities aimed at enhancing students' academic performance, particularly in contexts where traditional schooling is perceived as insufficient (Zhang & Bray, 2016; Tan & Liu, 2023; Bray & Hajar, 2024; Yoo, 2024). The determinants influencing the demand for and provision of shadow education are multifaceted, encompassing socio-economic factors, parental aspirations, cultural norms, and institutional factors such as school quality and curriculum pressure (Stevenson & Baker, 1992; Bray & Lykins, 2012). This paper aims to explore and synthesize existing literature on the determinants of shadow education, focusing on both demand-side and supply-side factors that drive its prevalence across different socio-economic contexts. Understanding these determinants is crucial for policymakers and educators seeking to address educational inequalities and optimize the effectiveness of formal schooling systems (Park et al., 2013). By examining empirical studies and theoretical frameworks, this paper seeks to illuminate how factors such as income levels, parental education, perceived educational quality, and cultural values interact to shape the dynamics of shadow education. Furthermore, the impact of globalization and digital technologies on the evolution of shadow education practices will be considered, highlighting the adaptation of traditional tutoring models in an increasingly interconnected world (Zhang & Bray, 2017; Kobakhidze, 2018).

By synthesizing current research findings, this study provides valuable insights into the multifaceted socio-economic and educational dynamics that influence the widespread growth of shadow education across the world. Ultimately, it seeks to provide insights into potential policy interventions and educational reforms that can mitigate the adverse effects of shadow education while harnessing its potential benefits for students and educational systems (Buchmann et al, 2010; Zhang & Bray, 2017; Holloway & Kirby; Gorkturk & Tulubas, 2021; Xiang, 2021). The interaction of these factors indicates that the growth of shadow education is a complex and multifaceted issue, requiring a depth understanding of various socio-economic, cultural, and policy-related influences. Researchers and policymakers must consider these dimensions to address the implications of shadow education on equity and access within the broader educational context

REVIEW OF LITERATURE

The growth of shadow education, which refers to private supplementary tutoring outside the mainstream education system, is driven by various socio-economic, cultural, and educational factors. At the forefront is socio-economic status; families with higher incomes can more easily fund private tutoring, prioritizing it to boost their children's academic performance, thereby widening the educational gap across different social segments (Burgess, 2016; Zwier et al., 2021; Habyarimana et al., 2023; Du, 2024). The competitive nature of modern education systems also drives the demand for shadow education, as parents and students seek to gain an edge in high-stakes examinations and admissions processes (Dang & Rogers, 2008). Cultural attitudes towards education, particularly in Asian countries, where academic success is highly valued and seen as a gateway to better socio-economic prospects, further bolster the proliferation of private tutoring (Zhan et al, 2013). Additionally, the perceived inadequacies of mainstream education systems, including large class sizes, insufficient resources, and a lack of individualized attention, compel parents to seek supplementary tutoring as a means to bridge gaps and enhance learning outcomes (Baker et al, 2001). Policy environments also shape the landscape of shadow education, with some governments' regulatory measures and public education policies indirectly fostering the growth of private tutoring markets (Kim & Lee, 2010). Moreover, globalization and the resulting emphasis on global competitiveness have intensified the pressure on students to excel academically, thus increasing reliance on supplementary education services (Entrich, 2015; Matsuoka, 2015; Mohmud, 2021; Zhang, 2021; Gupta, 2022). The rise of technology and online learning platforms has also made private tutoring more accessible and widespread, enabling a broader demographic to partake in shadow education (Zhang, 2019). Urbanization and the migration of families to cities for better educational opportunities further contribute to the proliferation of shadow education, as urban areas typically offer more and varied tutoring services compared to rural regions (Byun & Park, 2012). The intersection of these factors illustrates a complex and



**Sachin Kumar et al.,**

multifaceted phenomenon driving the growth of shadow education globally, reflecting broader trends in educational priorities, socio-economic status, Culture expectation etc.

Significance of the Study

The study on shadow education, or private supplementary tutoring, holds significant relevance in contemporary educational discourse. As shadow education becomes increasingly pervasive globally, understanding its determinants and implications is crucial for addressing educational inequalities and enhancing the overall efficacy of educational systems. The findings of this study provide critical insights into the socio-economic, cultural, and policy-driven factors that influence the demand and intensity of shadow education. Firstly, this research highlights the role of socio-economic status and parental education levels in shaping students' participation in shadow education. Higher household income and parental educational attainment are identified as strong correlates of increased engagement in supplementary tutoring (Bray & Lykins, 2012). This underscores the disparities in access to educational resources, suggesting that wealthier families are better positioned to afford additional academic support for their children, thereby perpetuating educational inequalities (Enrich, 2015; Bray, 2021; Cone, 2021; Xiang, 2021). In the second place, this research investigates the cultural and societal factors that lead to a high demand for shadow education, particularly in East Asia. The pressure to excel in high-stakes examinations and the competitive educational environment in these regions lead to a heightened dependence on private tutoring (Byun et al, 2018; Saengboon, 2019; Zhang, 2021; Liu, 2022). This cultural phenomenon highlights the intersection of educational practices and societal values, offering a depth understanding of how cultural contexts influence educational behaviors (Tan & Liu, 2023). Moreover, the research examines the impact of educational policies on the proliferation of shadow education. Policies that emphasize high-stakes testing and perceived inadequacies in formal schooling systems contribute to the growing demand for supplementary tutoring (Zhang, 2021; Bray, 2021; Byun et al., 2023). By identifying these policy-driven determinants, the study provides valuable insights for policymakers seeking to create more balanced and effective educational environments. The implications of this research extend beyond academic understanding to practical applications. For policymakers and educators, the findings offer a foundation for developing strategies to mitigate educational inequalities and enhance the inclusivity and effectiveness of both formal and informal educational systems. Future research directions proposed in this study can further inform policy adjustments and educational reforms aimed at fostering a more equitable educational landscape. In summary, this study is significant for its in-depth analysis of the multiple determinants of shadow education. By shedding light on the socio-economic, cultural, and policy factors influencing private supplementary tutoring, this research contributes to the broader effort of addressing educational disparities and improving educational outcomes globally.

Objectives of the Study

1. To investigate the impact of household income on the participation rates in shadow education among students
2. To examine the role of parental education levels in determining participation in shadow education.
3. To analyze the cultural expectations and norms that drives the demand for supplementary tutoring.
4. To evaluate the impact of educational policies on the proliferation of shadow education.
5. To explore policy implications and provide recommendations to address educational inequalities related to shadow education.

Research Questions

1. How does household income influence the participation in shadow education?
2. What is the relationship between parental education levels and the intensity of engagement in private supplementary tutoring?
3. In what ways do cultural expectations and societal norms affect the demand for shadow education in different regions, particularly in East Asian countries?
4. How do educational policies, such as high-stakes examinations, contribute to the growth of shadow education?
5. What strategies can policymakers and educators implement to mitigate educational inequalities exacerbated by the prevalence of shadow education?



Sachin Kumar *et al.*,

METHODOLOGY

This conceptual research paper employs an integrative literature review to explore the determinants of shadow education. The methodology involves a systematic and comprehensive examination of existing scholarly works, policy documents, and empirical studies related to shadow education. The primary sources of data include peer-reviewed journal articles, books, conference papers, and reputable online databases such as JSTOR, Scopus, Google Scholar Web of Science, and ERIC. The selection criteria for the literature were based on relevance, recency, and academic rigor, focusing on studies published in the last two decades to ensure contemporary relevance. The analysis follows a thematic approach, categorizing the determinants of shadow education into socio-economic, cultural, educational, and policy-related factors. Each category is critically analyzed to identify patterns, trends, and gaps in the prior literature. By synthesizing findings from diverse sources, this paper aims to provide a comprehensive understanding of the multifaceted determinants of shadow education, offering insights for policymakers, educators, and researchers interested in addressing educational inequalities and improving educational systems.

FINDINGS

RQ1: How does household income influence the participation in shadow education?

Household income significantly influences participation in shadow education. Firstly, higher household income provides families with the financial resources to afford private tutoring services. Families with greater financial means can invest in supplementary education to enhance their children's academic performance or gain competitive advantages in exams (Xue, 2019; Buyruk, 2020; Yoo, 2024). This ability to pay for additional educational support becomes crucial in contexts where public education systems are perceived as inadequate or where academic success is highly valued for future opportunities, such as admission to prestigious universities or securing better jobs (Kirigwi & Maithya, 2016; Bray, 2022; Jha, 2023). Secondly, household income influences the perception of education as a form of investment. Families with higher incomes often view education as a key pathway to social mobility and economic success. They may be more willing to allocate a significant portion of their income towards educational expenses, including private tutoring, viewing it as a strategic investment in their children's future prospects (Liu, 2019; Entrich & Lauterbach, 2019; Ozdere, 2021; Gulomovna, 2022). Moreover, income disparities can exacerbate educational inequalities. Lower-income families may struggle to afford private tutoring, placing their children at a disadvantage compared to their wealthier peers who can access additional educational support. This disparity can perpetuate existing inequalities in educational outcomes, contributing to a cycle where socioeconomic status determines access to educational resources and opportunities (Tsiplakides, 2018; Ozdere, 2021; Yung & Zeng, 2022; Tan & Liu, 2024). In conclusion, household income plays a critical role in shaping participation in shadow education. It not only determines the financial ability to access private tutoring but also reflects broader socioeconomic inequalities in educational opportunities. It is essential for policymakers and educators to understand these dynamics to bridge disparities and foster educational equity across diverse socioeconomic groups.

RQ2: What is the relationship between parental education levels and the intensity of engagement in private supplementary tutoring?

The relationship between parental education levels and the intensity of engagement in private supplementary tutoring is a well-documented phenomenon in educational research. Parental education levels often serve as a significant determinant influencing the decision to enroll children in private tutoring programs. This relationship can be understood through several mechanisms. Firstly, higher parental education levels are associated with greater awareness of the importance of education and academic success for their children (Bray & Lykins, 2012; Liu, 2019; Kato & Kobakhidze, 2024). Parents with higher educational attainment tend to have better knowledge about the educational system, including its strengths and limitations. This awareness may lead them to seek additional support, such as private tutoring, to supplement their child's learning experiences and ensure academic success. Secondly, parental education levels correlate positively with socioeconomic status (SES) (Liu, 2019). Families with higher SES often have more financial resources to invest in supplementary educational activities like private tutoring.





Sachin Kumar *et al.*,

These resources enable them to access and afford tutoring services that may not be available or affordable to families with lower SES. Thus, parental education indirectly influences tutoring engagement through its association with economic capability. Thirdly, parental educational aspirations for their children play a crucial role (Tan & Liu, 2023). Parents who possess higher educational qualifications generally hold greater expectations and aspirations for their children's academic success. Some parents may regard private tutoring as a means to elevate their children's academic results and future educational prospects, aligning with their aspirations for social mobility. Empirical studies support these theoretical assertions. For instance, research in diverse educational contexts, such as East Asia and Western countries, consistently shows that children from families with higher parental education levels are more likely to participate in private tutoring (Liu, 2019; Kato & Kobakhidze, 2024). These studies underscore the significant influence of parental education on the intensity of engagement in private supplementary tutoring, highlighting the multifaceted roles of parental awareness, socioeconomic status, and educational aspirations in shaping educational decisions and outcomes for children. In summary, parental education levels are closely linked to the decision and intensity of engagement in private supplementary tutoring, reflecting broader social and educational dynamics within families and societies.

RQ3: In what ways do cultural expectations and societal norms affect the demand for shadow education in different regions, particularly in East Asian countries?

The role of cultural expectations and societal norms in East Asia is profound, particularly in shaping the demand for shadow education and educational practices. In East Asia, cultural values emphasizing academic achievement, such as Confucian principles of education and the pursuit of excellence, exert substantial pressure on students to excel academically (Bray, 1999; Bhorkar & Bray, 2018). These cultural expectations foster a highly competitive educational environment where success in standardized tests, such as university entrance exams, is often seen as crucial for future opportunities and social mobility (Stevenson & Baker, 1992). Consequently, parents and students alike perceive shadow education, such as private tutoring or cram schools, as necessary supplements to formal schooling to ensure adequate preparation and competitive advantage (Liu, 2019; Jansen *et al.*, 2021; Yung & Zeng, 2022; McCoy & Byrne, 2024). Moreover, societal norms in East Asia reinforce the belief that educational success not only benefits the individual but also reflects positively on the family and community. This collective orientation underscores the widespread acceptance and utilization of shadow education as a means to fulfill societal expectations of academic achievement (Buyruk, 2020). For instance, in South Korea and Japan, where educational attainment is closely tied to future career prospects and social status, the demand for shadow education is driven by the desire to meet societal norms of success (Christensen & Zhang, 2021). In contrast, in Western societies where individualism and diverse career paths are more culturally endorsed, the demand for shadow education may be less pronounced due to different societal priorities and educational philosophies (Byun *et al.*, 2023). Thus, while cultural expectations and societal norms in East Asia heighten the demand for shadow education by emphasizing academic achievement and social expectations, these factors may vary in influence across different regions depending on cultural values and educational systems. Overall, the interaction between cultural expectations, societal norms, and educational aspirations shapes the demand for shadow education, particularly highlighting its significance in East Asian countries where academic success is deeply intertwined with cultural identity and societal advancement.

RQ4: How do educational policies, such as high-stakes examinations, contribute to the growth of shadow education?

The development of shadow education systems worldwide is heavily influenced by educational policies, particularly those associated with high-stakes examinations. High-stakes examinations are pivotal as they often determine students' future academic and career prospects (Bray & Lykins, 2012). These exams are typically highly competitive and can create intense pressure on students to excel, leading to a heightened demand for supplementary education outside the formal schooling system (Bray, 1999). The impact of high-stakes examinations on educational results and opportunities is a key driver behind the growth of shadow education. In many countries, performance in these exams dictates access to prestigious universities or specific academic tracks, which are perceived as pathways to success and social mobility (Dang & Rogers, 2008; Byun *et al.*, 2018; Saengboon, 2019; Zhang, 2021; Liu, 2022). Consequently, students and their families seek additional tutoring and coaching to enhance exam performance and competitiveness



**Sachin Kumar et al.,**

(Bray, 2022; Jansen et al., 2024). Moreover, the structure and content of high-stakes exams often prioritize rote memorization and exam-specific skills rather than deep understanding and critical thinking (Jung-Hoon, 2018; Liao & Huang, 2018). This focus can lead to dissatisfaction with the formal education system among students and parents alike, who perceive it as insufficient for achieving desired exam results (Bray & Lykins, 2012). Furthermore, the proliferation of shadow education is fueled by perceptions of inequity within the formal education system. Wealthier families can afford private tutoring or enrollment in prestigious exam preparation centers, thereby gaining an advantage over economically disadvantaged students who may not have access to similar resources (Stevenson & Baker, 1992). In conclusion, high-stakes examinations embedded within educational policies significantly contribute to the growth of shadow education by shaping educational priorities, intensifying competition among students, fostering dissatisfaction with formal schooling, and perpetuating socio-economic inequalities. These factors underscore the complex interaction between educational policies and the phenomenon of shadow education, highlighting the need for detailed policy interventions that address both the drivers and consequences of its proliferation.

RQ5: What strategies can policymakers and educators implement to mitigate educational inequalities exacerbated by the prevalence of shadow education?

To effectively address the question of mitigating educational inequalities exacerbated by shadow education, policymakers and educators can implement several strategies. Shadow education refers to private supplementary tutoring and coaching that students receive outside formal schooling, often driven by concerns about academic competition and the desire for enhanced educational outcomes. This phenomenon has been identified as contributing to educational inequalities by favoring students from wealthier backgrounds who can afford such services, thereby widening the gap between privileged and disadvantaged students. One crucial strategy involves regulating and monitoring the shadow education sector. By imposing standards and guidelines on tutoring services, policymakers can ensure that these services supplement rather than substitute formal education. Regulation can include mandating qualifications for tutors, ensuring transparent pricing, and requiring tutoring institutions to align their curriculum with national educational goals (Bray, 2009). Integration of shadow education into formal schooling is another effective strategy. This approach involves acknowledging the prevalence of shadow education and incorporating elements of its success strategies into mainstream classrooms. For instance, identifying effective tutoring methods used in shadow education and training teachers to implement these strategies can help level the playing field for all students (Bai et al., 2019; Punjabi, 2019; Kim & Jung, 2019; Gan & Shahrill, 2019). Financial support and subsidies for disadvantaged students can reduce the financial barriers that prevent them from accessing shadow education. Scholarships or vouchers targeted at low-income families can empower students to seek supplementary tutoring, thereby narrowing the gap between socioeconomic groups (Carr & Wang, 2018; Lancker, 2021; Wainwright et al., 2023). Improving the quality and equity of formal education is fundamental to reducing the need for shadow education. By enhancing the quality of teaching, providing additional academic support within schools, and offering personalized learning opportunities, educators can address the root causes that drive students to seek private tutoring (Byun et al., 2018; Guill et al., 2019; Ozdere, 2021; Bar & Guha, 2023). Community and parental involvement are also crucial. By educating parents on the consequences of shadow education and including them in talks on alternative approaches to enhance their children's learning, reliance on private tutoring can be diminished. Community-based initiatives that offer academic support outside school hours can provide viable alternatives to shadow education (Bray & Zhang, 2018; Mahmud et al., 2018; Yu & Zhang, 2022). Research and data collection are essential for understanding the dynamics of shadow education and its impact on educational inequalities. Continuous monitoring and evaluation can inform evidence-based policymaking and ensure that interventions are effective and sustainable (Bray, 2009). In conclusion, addressing educational inequalities exacerbated by shadow education requires a multifaceted approach that combines regulation, integration, and financial support, improvements in formal education, community involvement, and rigorous research. Through the adoption of these strategies, policymakers and educators aim to foster an educational system that promotes fairness, ensuring every student, irrespective of socioeconomic status, can thrive.



Sachin Kumar *et al.*,**Figure Explanation (Strategies to reduce the expansion of Shadow Education)****Improve Quality of Public School**

- **Enhance Teacher Training & Support:** Providing ongoing professional development and support for teachers to improve their teaching skills and effectiveness.
- **Update Curriculum to be More Relevant & Engaging:** Revising the curriculum to make it more interesting and applicable to students' lives, encouraging better learning outcomes.
- **Foster Collaborative Learning Environment:** Creating an environment where students can learn together, share ideas, and support each other's learning.
- **Involve Community & Parents in Education Decision Making:** Engaging parents and the community in the decision-making process related to education to ensure that their needs and perspectives are considered.

Strengthen Public School System

- **Increase Funding & Resources for Schools:** Allocating more financial resources to schools to improve infrastructure, resources, and overall quality of education.
- **Reduce Class Size:** Decreasing the number of students per classroom to allow for more personalized attention and effective teaching.
- **Providing Additional Support Services:** Offering extra academic support, counseling, and other services to help students succeed.
- **Engage Parents & Community in School Improvement Efforts:** Involving parents and community members in initiatives aimed at enhancing school performance and student outcomes.

Regulate Private Tutoring

- **Set Clear Standards for Tutoring Services:** Establishing guidelines and standards to ensure the quality and effectiveness of private tutoring services.
- **Monitor & Evaluate Tutoring Practices:** Regularly reviewing and assessing private tutoring practices to maintain high standards.
- **Offer Incentives for Transparency & Accountability:** Encouraging private tutoring services to be transparent and accountable in their operations.
- **Support Alternative to Private Tutoring:** Promoting other forms of support and enrichment that can serve as alternatives to private tutoring.

Educational Implications

The results of this research offer crucial insights that can shape policy decisions and educational strategies. Firstly, the strong correlation between higher household income and increased participation in shadow education highlights the need for equitable access to quality education. Policymakers should consider strategies to mitigate the disparities in educational opportunities that arise from socio-economic inequalities. To illustrate, providing low-income families with subsidized or free supplementary tutoring could help bridge the gap, ensuring that every student has an equal chance to boost their academic success. (Bray, 2021). Moreover, the study's revelation that parental educational attainment significantly influences the propensity to engage in shadow education suggests the importance of parental involvement in children's education. Schools and educational institutions should develop programs that encourage and facilitate parental engagement, regardless of their educational background. Providing parents with resources and training on how to support their children's learning at home can be a valuable approach (Kim & Lee, 2010). Cultural expectations and societal pressures, particularly evident in East Asian countries, also play a substantial role in the prevalence of shadow education. Educators and policymakers must work towards creating a balanced educational environment that emphasizes holistic development over merely academic achievement. Promoting a more comprehensive curriculum that includes arts, sports, and life skills alongside traditional academic subjects can alleviate the undue pressure on students to excel solely in academics (Ozdere, 2021; Stastny, 2023; Tan & Liu, 2023). The impact of educational policies, such as high-stakes examinations and the perceived inadequacy of formal schooling, on the demand for shadow education underscore the need for systemic reforms. Education systems should aim to reduce the overemphasis on examinations and instead focus on continuous and formative assessments





Sachin Kumar et al.,

that provide a more accurate measure of student learning and development. Additionally, efforts to improve the quality of formal education through better teacher training, updated curricula, and adequate resources are essential to reduce the reliance on supplementary tutoring (Bray & Kobakhidze, 2015; Bray et al., 2018; Soeung, 2021; Budyuk, 2022). In conclusion, addressing the determinants of shadow education identified in this study can lead to more equitable and effective educational systems. By implementing targeted policies and practices that address socio-economic disparities, enhance parental involvement, balance educational pressures, and reform assessment systems, policymakers and educators can work towards a more inclusive and supportive learning environment for all students. Future research should continue to explore these areas to provide further insights and recommendations for fostering educational equity and improving overall student outcomes.

CONCLUSION

The expansion of shadow education, which includes private supplementary tutoring beyond the mainstream school system, is shaped by a blend of socio-economic, cultural, educational, and policy factors, highlighting its intricate and multi-dimensional character. At the socio-economic level, increasing income disparities and the competitive nature of globalized economies have heightened parental anxiety regarding children's future success, driving demand for additional academic support to secure competitive advantages. This phenomenon is particularly pronounced in countries with high-stakes examination systems, where performance on standardized tests significantly impacts educational and career opportunities, thereby fostering a lucrative market for private tutors. Cultural factors also play a pivotal role, with societal values emphasizing academic excellence and educational attainment, particularly in East Asian countries where Confucian traditions valorize education. These cultural expectations compel parents to commit substantial resources to their children's education, often resulting in significant household expenditures on tutoring services. In regions where public schools are underfunded or face systemic issues such as overcrowded classrooms, inadequate resources, and poorly trained teachers, parents turn to private tutoring as a necessary supplement to fill educational gaps and enhance their children's learning outcomes. Educational policies and government regulations, or the lack thereof, also significantly impact the proliferation of shadow education. In some countries, lax regulatory frameworks allow for the unregulated expansion of private tutoring businesses, while in others, stringent policies aimed at curbing the shadow education industry may inadvertently drive it underground, making it less visible but not necessarily less prevalent. Additionally, technological advancements have revolutionized the shadow education landscape, making online tutoring and educational resources more accessible, thus broadening the reach and appeal of private tutoring services. The COVID-19 pandemic further accelerated this trend, as the shift to remote learning highlighted the deficiencies in public education systems and increased reliance on online supplementary education. Peer influence and social networks play a significant role in the proliferation of shadow education, as community members share insights and advice about effective tutoring services, establishing a normative expectation for participation. The interaction between these factors underscores the intricate dynamics driving the expansion of shadow education globally. Confronting the challenges of the increasing shadow education sector necessitates a diverse strategy that encompasses enhancing public education quality and equity, instituting effective regulatory measures, and fostering cultural changes towards a more holistic and balanced educational development. Policymakers must acknowledge the deep-rooted socio-cultural and economic drivers of shadow education and work towards creating educational environments that reduce the necessity for supplementary tutoring, ensuring that all students have equitable access to quality education without the need for additional, often costly, external support.

Further Research

The findings from this study highlight critical areas for further investigation to enhance our understanding of the factors and implications of shadow education. Future research should aim to delve deeper into the specific mechanisms through which socio-economic status and parental education levels influence students' engagement in supplementary tutoring. Additionally, there is a need to explore the psychological and social impacts of shadow education on students, particularly concerning stress, mental health, and overall well-being. Another promising area





Sachin Kumar et al.,

for future research is the examination of shadow education in diverse cultural contexts beyond East Asia. Comparative studies across different regions and countries can uncover unique cultural and societal impacts on the expansion of shadow education. This contributes to a broader international perspective on shadow education by identifying shared trends and unique methodologies. With the rise of online tutoring platforms and digital learning resources, it is essential to investigate how technology-mediated supplementary education affects accessibility, quality, and educational outcomes. Research in this area could also assess the effectiveness of online versus traditional face-to-face tutoring methods. Policy-oriented research is crucial to evaluate the efficacy of existing educational policies and interventions aimed at mitigating the need for shadow education. Experimental and quasi-experimental designs could be employed to assess the impact of various policy measures, such as curriculum reforms, teacher training programs, and student support services, on reducing dependence on private tutoring. Finally, an interdisciplinary approach involving economics, sociology, psychology, and education can provide a holistic perspective on shadow education.

REFERENCES

1. Author, J., Stevenson, D. L., & Baker, D. P. (1992). Shadow Education and Allocation in Formal Schooling: Transition to University in Japan. In *Source: American Journal of Sociology* (Vol. 97, Issue 6). <https://www.jstor.org/stable/2781551>
2. Bai, Y., Tang, X., Li, X., & Fan, S. (2019). Explorations in Teaching Research by a Tutoring Institution in China. *ECNU Review of Education*, 2(1), 87–94. <https://doi.org/10.1177/2096531119840866>
3. Baker, D. P., Akiba, M., Letendre, G. K., & Wiseman, A. W. (2001). Worldwide Shadow Education: Outside-School Learning, Institutional Quality of Schooling, and Cross-National Mathematics Achievement. In *Policy Analysis* (Vol. 23, Issue 1).
4. Bar, K., & Guha, A. (2023). Engagement of School Students in Shadow Education: Present Status in India; West Bengal. *International Journal of Advanced Research*, 11(03), 664–670. <https://doi.org/10.21474/IJAR01/16479>
5. Bhorkar, S., & Bray, M. (2018). The Expansion and Roles of Private Tutoring in India: From Supplementation to Supplantation. In *International Journal of Educational Development* (Vol. 62).
6. Bray, M. (1999). *The shadow education system : private tutoring and its implications for planners*. Unesco, International Institute for Educational Planning.
7. Bray, M. (2013). Shadow Education: Comparative Perspectives on the Expansion and Implications of Private Supplementary Tutoring. *Procedia - Social and Behavioral Sciences*, 77, 412–420. <https://doi.org/10.1016/j.sbspro.2013.03.096>
8. Bray, M. (2022). Teachers as tutors, and tutors as teachers: blurring professional boundaries in changing eras. *Teachers and Teaching: Theory and Practice*, 28(1), 64–77. <https://doi.org/10.1080/13540602.2021.2019700>
9. Bray, M. (2023). Geographies of shadow education: patterns and forces in the spatial distributions of private supplementary tutoring. *Compare*, 53(3), 343–360. <https://doi.org/10.1080/03057925.2021.1915749>
10. Bray, M., & Hajar, A. (2024). Complexity of the Contexts: Features of Private Tutoring and Units for Comparison in the GCC Countries of the Middle East. *ECNU Review of Education*, 7(1), 42–65. <https://doi.org/10.1177/20965311221113350>
11. Bray, M., & Kobakhidze, M. N. (2015). Evolving ecosystems in education: The nature and implications of private supplementary tutoring in Hong Kong. *Prospects*, 45(4), 465–481. <https://doi.org/10.1007/s11125-015-9353-2>
12. Bray, M., Kobakhidze, M. N., Zhang, W., & Liu, J. (2018). The hidden curriculum in a hidden marketplace: relationships and values in Cambodia's shadow education system. *Journal of Curriculum Studies*, 50(4), 435–455. <https://doi.org/10.1080/00220272.2018.1461932>
13. Bray, M., & Lykins, C. R. (2012). *Shadow education : private supplementary tutoring and its implications for policy makers in Asia*. Asian Development Bank.
14. Bray, M., Unesco., & International Institute for Educational Planning. (2009). *Confronting the shadow education system : what government policies for what private tutoring?* United Nations Educational, Scientific and Cultural Organization.





Sachin Kumar et al.,

15. Bray, M., & Zhang, W. (2018). Public-Private Partnerships in Supplementary Education: Sharing Experiences in East Asian Contexts. *International Journal for Research on Extended Education*, 6(1–2018), 98–106. <https://doi.org/10.3224/ijree.v6i1.08>
16. Bray, M., & Zhang, W. (2023). Cross-Border Shadow Education: Private Supplementary Tutoring in the Global Education Industry. *Revista Espanola de Educacion Comparada*, 42, 28–43. <https://doi.org/10.5944/reec.42.2023.34275>
17. Buchmann, C., Condron, D. J., & Roscigno, V. J. (2010). *Shadow Education: Theory, Analysis and Future Directions: A Rejoinder* (Vol. 89, Issue 2). <https://about.jstor.org/terms>
18. Burgess, S. (2016). *Human Capital and Education: The State of the Art in the Economics of Education*. <http://www.coeure.eu/>
19. Buyruk, H. (2020). A School Choice Experience at the Age of “Parentocracy”: Impressions from a Public Primary School in Turkey. *International Journal of Progressive Education*, 16(2), 230–247. <https://doi.org/10.29329/ijpe.2020.241.16>
20. Byun, S.-Y., & Park, H. (2012). The Academic Success of East Asian American Youth: The Role of Shadow Education. *Source: Sociology of Education*, 85(1), 40–60. <https://doi.org/10.1177/003804071>
21. Byun, S. Y., Chung, H. J., & Baker, D. P. (2018). Global patterns of the use of shadow education: Student, family, and national influences. In *Research in the Sociology of Education* (Vol. 20, pp. 71–105). Emerald Group Publishing Ltd. <https://doi.org/10.1108/S1479-353920180000020004>
22. Carr, D., & Wang, L. C. (2018). The Effect of After-School Classes on Private Tuition, Mental Health and Academic Outcomes. *Source: Sociology*, 52(5), 877–897. <https://doi.org/10.2307/26558738>
23. Christensen, S., & Zhang, W. (2021). Shadow Education in the Nordic Countries: An Emerging Phenomenon in Comparative Perspective. In *ECNU Review of Education* (Vol. 4, Issue 3, pp. 431–441). SAGE Publications Ltd. <https://doi.org/10.1177/209653112111037925>
24. Cone, L. (2021). “They Would Do Just Fine Without Me”: Experiences of Private Tutoring in Denmark. *ECNU Review of Education*, 4(3), 566–589. <https://doi.org/10.1177/2096531120940718>
25. Dang, H. A., & Rogers, F. H. (2008). The growing phenomenon of private tutoring: Does it deepen human capital, widen inequalities, or waste resources? *World Bank Research Observer*, 23(2), 161–200. <https://doi.org/10.1093/wbro/lkn004>
26. De Dieu Habyarimana, J., Mugabonake, A., Ntakirutimana, E., Hashakimana, T., Ngendahayo, E., Mugiraneza, F., & Zhou, K. (2023). Matthew Effect and Achievement Gap in Rwandan Basic Education. *International Journal of Learning, Teaching and Educational Research*, 22(4), 242–266. <https://doi.org/10.26803/IJLTER.22.4.14>
27. Du, S. (2024). Shadow education in shadow: parental education and children’s shadow education participation before and during COVID-19. *British Journal of Sociology of Education*. <https://doi.org/10.1080/01425692.2024.2328133>
28. Entrich, S. R. (2015). The Decision for Shadow Education in Japan: Students’ Choice or Parents’ Pressure? In *Source: Social Science Japan Journal* (Vol. 18, Issue 2). <https://www.jstor.org/stable/43920491>
29. Entrich, S. R., & Lauterbach, W. (2020). Shadow Education in Germany: Compensatory or Status Attainment Strategy? Findings from the German Life Study. *International Journal for Research on Extended Education*, 7(2–2019), 143–159. <https://doi.org/10.3224/ijree.v7i2.04>
30. Gan, A. L., & Shahrill, M. (2019). Why the need for private tuition in mathematics? Investigating shadow education in Brunei Darussalam. *AIP Conference Proceedings*, 2194. <https://doi.org/10.1063/1.5139763>
31. Göktürk, Ş., & Tülübaş, T. (2021). Survival of the fittest: women’s academic experiences of navigating neoliberal expectations in Turkish universities. *Gender and Education*, 33(3), 267–284. <https://doi.org/10.1080/09540253.2020.1735311>
32. Guill, K., Lüdtke, O., & Schwanenberg, J. (2020). A two-level study of predictors of private tutoring attendance at the beginning of secondary schooling in Germany: The role of individual learning support in the classroom. *British Educational Research Journal*, 46(2), 437–457. <https://doi.org/10.1002/berj.3586>
33. Gulomovna, Z. M. (2022). Commodification of education in the form of shadow education. *ACADEMICIA: An International Multidisciplinary Research Journal*, 12(1), 200–206. <https://doi.org/10.5958/2249-7137.2022.00048.9>
34. Gupta, A. (2022). Social legitimacy of private tutoring: an investigation of institutional and affective educational practices in India. *Discourse*, 43(4), 571–584. <https://doi.org/10.1080/01596306.2020.1868978>





Sachin Kumar et al.,

35. Gupta, A., & Zhao, X. (2024). Teachers' work under responsabilising policies: an analysis of educators' views on China's 2021 educational reforms. *Journal of Education Policy*, 39(4), 622–639. <https://doi.org/10.1080/02680939.2023.2236067>
36. Holloway, S. L., & Kirby, P. (2020). Neoliberalising Education: New Geographies of Private Tuition, Class Privilege, and Minority Ethnic Advancement. *Antipode*, 52(1), 164–184. <https://doi.org/10.1111/anti.12587>
37. Jansen, D., Elffers, L., & Jak, S. (2023). A cross-national exploration of shadow education use by high and low SES families. *International Studies in Sociology of Education*, 32(3), 653–674. <https://doi.org/10.1080/09620214.2021.1880332>
38. Jansen, D., Elffers, L., Jak, S., & Volman, M. L. L. (2024). Is a more selective exit exam related to shadow education use? An analysis of two cohorts of final-year secondary school students in the Netherlands. *Oxford Review of Education*, 50(1), 24–41. <https://doi.org/10.1080/03054985.2023.2187364>
39. Jha, S. K. (2023). The Three E's of Private Tuition in India: Expansion, Expenditure, and Effect. *Journal of Education*, 203(2), 423–432. <https://doi.org/10.1177/00220574211032345>
40. Jung, J. H. (2018). Decolonizing educational/curriculum studies in East Asia: problematizing shadow education in South Korea. In *Inter-Asia Cultural Studies* (Vol. 19, Issue 2, pp. 269–281). Taylor and Francis Ltd. <https://doi.org/10.1080/14649373.2018.1463074>
41. Kato, M., & Kobakhidze, M. N. (2024). Transnational juku: Japanese shadow education institutions in Hong Kong, Beijing, and Shanghai. *Asia Pacific Education Review*. <https://doi.org/10.1007/s12564-024-09946-5>
42. Kim, S., & Lee, J. H. (2010). Private tutoring and demand for education in South Korea. *Economic Development and Cultural Change*, 58(2), 259–296. <https://doi.org/10.1086/648186>
43. Kim, Y. C., & Jung, J.-H. (2019). *Shadow Education as Worldwide Curriculum Studies CURRICULUM STUDIES WORLDWIDE*. <http://www.palgrave.com/gp/series/14948>
44. Kirigwi, L. W., & Maithya, R. (2016). Prevalence for Private Tuition among Parents, Teachers and Pupils in Public Primary Schools in Machakos County. *Journal of Education and E-Learning Research*, 3(3), 115–123. <https://doi.org/10.20448/journal.509/2016.3.3/509.3.115.123>
45. Liao, X., & Huang, X. (2018). Who Is More Likely to Participate in Private Tutoring and Does It Work?: Evidence from PISA (2015). *ECNU Review of Education*, 1(3), 69–95. <https://doi.org/10.30926/ecnuoe>
46. Liu, E. (2023). The Functions of Shadow Education in China: An Exploration beyond Mainstream Education. *Lecture Notes in Education Psychology and Public Media*, 2(1), 380–384. <https://doi.org/10.54254/2753-7048/2/2022309>
47. Liu, J. (2019). A Structural Equation Modeling Analysis of Parental Factors Underlying the Demand for Private Supplementary Tutoring in China. *ECNU Review of Education*, 2(1), 7–24. <https://doi.org/10.1177/2096531119840856>
48. Mahmud, R. (2021). Learning in the shadows: parents' investments, family burden, and students' workload in Dhaka, Bangladesh. *Asia Pacific Education Review*, 22(1), 41–52. <https://doi.org/10.1007/s12564-020-09655-9>
49. Mahmud, R., & Kenayathulla, H. B. (2018). Shadow education: patterns and scale of private supplementary tutoring in English in secondary education at urban Dhaka in Bangladesh. *Compare*, 48(5), 702–716. <https://doi.org/10.1080/03057925.2017.1340827>
50. Matsuoka, R. (2015). School socioeconomic compositional effect on shadow education participation: evidence from Japan. *British Journal of Sociology of Education*, 36(2), 270–290. <https://doi.org/10.1080/01425692.2013.820125>
51. McCoy, S., & Byrne, D. (2024). Shadow Education Uptake in Ireland: Inequalities and Wellbeing in a High-Stakes Context. *British Journal of Educational Studies*. <https://doi.org/10.1080/00071005.2024.2331476>
52. Nuryana, Z., Xu, W., Lu, S., Tasir, Z., & Suyadi, S. (2023). Mapping global research on shadow education: Trends and future agenda. *International Journal of Evaluation and Research in Education*, 12(2), 1105–1113. <https://doi.org/10.11591/ijere.v12i2.24667>
53. Özdere, M. (2021). The Demand for Private Tutoring in Turkey: An Analysis of Private Tutoring Participation and Spending. *Journal of Education and Learning*, 10(3), 96. <https://doi.org/10.5539/jel.v10n3p96>
54. Park, H., Byun, S. yong, & Kim, K. keun. (2011). Parental involvement and students' cognitive outcomes in Korea: Focusing on private tutoring. *Sociology of Education*, 84(1), 3–22. <https://doi.org/10.1177/0038040710392719>
55. Punjabi, S. (2020). Is Shadow Education Becoming the 'New' Formal? Effects of Pedagogical Approaches of IIT-JEE Coaching on School Education in the City of Delhi. *Contemporary Education Dialogue*, 17(1), 14–44. <https://doi.org/10.1177/0973184919885485>





Sachin Kumar et al.,

56. Saengboon, S. (2019). Shadow Education in Thailand: A Case Study of Thai English Tutors' Perspectives towards the Roles of Private Supplementary Tutoring in Improving English Language Skills. In *Journal: Language Education and Acquisition Research Network Journal* (Vol. 12, Issue 1).
57. Soeung, S. (2021). A Review of Cambodian Private Tutoring: Parasitic and Symbiotic Functions towards the Mainstream System. *Journal of Nusantara Studies (JONUS)*, 6(1), 42–58. <https://doi.org/10.24200/jonus.vol6iss1pp42-58>
58. Šťastný, V. (2023). Shadow education in the context of early tracking: between-track differences in the Czech Republic. *Compare*, 53(3), 380–398. <https://doi.org/10.1080/03057925.2021.1922271>
59. Tan, M., & Liu, S. (2023). A Way of Human Capital Accumulation: Heterogeneous Impact of Shadow Education on Students' Academic Performance in China. *SAGE Open*, 13(4). <https://doi.org/10.1177/21582440231207189>
60. Tsiplakides, I. (2018). Shadow education and social class inequalities in secondary education in greece: The case of teaching english as a foreign language. *International Journal of Sociology of Education*, 7(1), 71–93. <https://doi.org/10.17583/rise.2018.2987>
61. Van Lancker, W. (2021). *The Matthew Effect In Early Childhood Education And Care How family policies may amplify inequalities.*
62. Wainwright, E., Barker, J., Chappell, A., & McHugh, E. (2023). 'On the high street' tuition for primary-aged children in London: Critiquing discourses of accessibility, attainment and assistance. *Education* 3-13. <https://doi.org/10.1080/03004279.2023.2237039>
63. Xue, H. (2019). Family Capitals and Education Acquisitions: Analysis on the Mediating Effect of Shadow Education. *Best Evidence of Chinese Education*, 1(2), 87–107. <https://doi.org/10.15354/bece.19.ar1019>
64. Yoo, J. (2024). Self-directed Learning Ability and Shadow Education Expenditures: A Comparison of Impact on Student Achievement in Korea During COVID-19. *SAGE Open*, 14(2). <https://doi.org/10.1177/21582440241245206>
65. Yu, J., & Zhang, R. (2022). A Review of Shadow Education. *Science Insights Education Frontiers*, 11(2), 1579–1593. <https://doi.org/10.15354/sief.22.re058>
66. Yung, K. W. H., & Zeng, C. (2022). Parentocracy within meritocracy: parental perspective on lecture-style English private tutoring in Hong Kong. *Language and Education*, 36(4), 378–394. <https://doi.org/10.1080/09500782.2021.1981924>
67. Zhan, S., Bray, M., Wang, D., Lykins, C., & Kwo, O. (2013). The effectiveness of private tutoring: Students' perceptions in comparison with mainstream schooling in Hong Kong. *Asia Pacific Education Review*, 14(4), 495–509. <https://doi.org/10.1007/s12564-013-9276-7>
68. Zhang, W. (2019). Regulating Private Tutoring in China: Uniform Policies, Diverse Responses. *ECNU Review of Education*, 2(1), 25–43. <https://doi.org/10.1177/2096531119840868>
69. Zhang, W. (2021). Modes and Trajectories of Shadow Education in Denmark and China: Fieldwork Reflections by a Comparativist. *ECNU Review of Education*, 4(3), 615–629. <https://doi.org/10.1177/20965311211042026>
70. Zhang, W., & Bray, M. (2016). Shadow Education. In *Spotlight on China* (pp. 85–99). SensePublishers. https://doi.org/10.1007/978-94-6209-881-7_6
71. Zhang, W., & Bray, M. (2017). Micro-neoliberalism in China: public-private interactions at the confluence of mainstream and shadow education. *Journal of Education Policy*, 32(1), 63–81. <https://doi.org/10.1080/02680939.2016.1219769>
72. Zwier, D., Geven, S., & van de Werfhorst, H. G. (2020). Social inequality in shadow education: The role of high-stakes testing. *International Journal of Comparative Sociology*, 61(6), 412–440. <https://doi.org/10.1177/0020715220984500>





Sachin Kumar et al.,

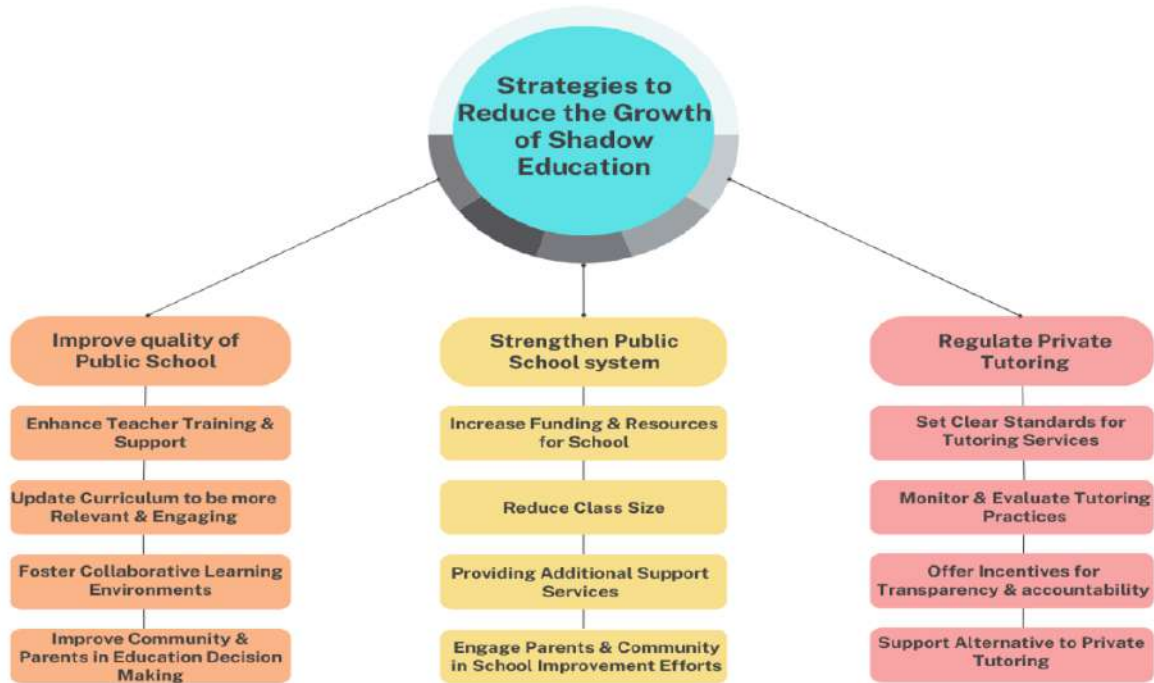


Figure 1: Strategies to reduce the expansion of Shadow Education





Provision of Data Security using Elliptic Curve Cryptography in Cloud Computing Environment

N.Krishnamoorthy^{1*} and S.Umarani²

¹Assistant Professor, Department of Computer Science and Applications (MCA), Faculty of Science and Humanities, SRM Institute of Science and Technology (Deemed to be University), Ramapuram, Chennai, Tamil Nadu, India.

²Professor, Department of Computer Science and Applications (BCA), Faculty of Science and Humanities, SRM Institute of Science and Technology (Deemed to be University), Ramapuram, Chennai, Tamil Nadu, India.

Received: 27 May 2024

Revised: 10 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

N.Krishnamoorthy

Assistant Professor,

Department of Computer Science and Applications (MCA),

Faculty of Science and Humanities,

SRM Institute of Science and Technology (Deemed to be University),

Ramapuram, Chennai, Tamil Nadu, India.

E.Mail: krishnan@srmist.edu.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Safekeeping of our data being placed in the cloud is always becoming a serious problem and challenging one. The honest reasons for this will be because of the existence of malevolent attacks, data breaches and unsafe contact points. In this juncture, more numbers of investigators offered security mechanisms that includes the components namely access controlling, invasion detection, prevention of occurrences schemes. It is often seen that there are many situations where the attackers have always misused the rights taken by them with the numerous roles in the cloud computing scenario. Hence it would be mandatory to introduce more efficient mechanism for securing the safety of sensitive data which would be placed/recovered in the given cloud. The main philosophies in privacy & security would be none other than the data secrecy and honesty. So, this investigation work heightened a fruitful and better procedure confirming a safer data transmission into the cloud computing atmosphere. The research work provides a additional information security method to cloud that makes use of the Elliptic Curve Integrated Encryption Scheme (ECIES), which confirms that data which could be placed in the Cloud Computing atmosphere is equally trusted and complete. The investigational outcomes prove that the effectiveness of the recommended algorithm results in a strong security level as well as reduces the execution time related to the commonly used existing techniques. Hence in this an Improved Elliptic Curve Cryptography (IECC) algorithm is suggested in this research work which is intended to give better security using modified algorithm. The parameters like encode/decode and store/transfer time are computed and it can be seen that the procedure that is being intended in this research paper take a smaller amount time for all the parameters when compared with prevailing methods. The encoding period of the suggested method is 46 ms with key size=4,096 bits, however the prevailing scheme takes 92 milliseconds for the same. Also,



**Krishnamoorthy and Umarani**

our proposed methods drastically reduce the deciphering time. All the other parameters presented in the results also certify that our scheme provides better solution when compared with the existing schemes. This reports disclose that the suggested system overtakes the present methods in terms of various factors.

Keywords : Cloud computing; Elliptic Curve; Data security; Cryptography.

INTRODUCTION

Cloud is a technology by which data available in a remote server can be easily retrieved with the internet and it is concealed in nature [1] [2]. It is the On-demand provision of any IT services including servers, storage, databases, any softwares etc. which is normally retrieved over the internet-the so called cloud. In short we can say that any device or gadget which is connected to internet can access the facilities provided. The cloud supports users to right to use the files and applications from any device, since the computing and storage operations happen in the servers in which is placed in a data center, rather than using a local storage. For example, when a user tries to login to his/her Google account form a mobile, tablet, PC, laptop which has internet connection. Hence we say that CC is a State-of-the-art innovation for treating & Relocating data currently in just all possible devices [3]

SECURITY ISSUES IN CLOUD

We can also refer to the CIA Triad the information must be set at the focal point of the threesome so as to convey total security of the information. The development of information towards any of the edges of the trio will prompt bargaining the other two properties of information [5].

Significance of the Study

Since we are dealing with massive quantity of data, it is extremely compulsory to confirm security. At the same time it is also important to minimize the computational overhead and time complications. Therefore, this research work makes an effort to offer secure and reliable data transmission into the cloud through Modified ECC method. So, the goal of this research was to use the Modified ECC method to enable secure and trustworthy data transfer into the cloud. We will obtain two private keys by splitting the private key (based on size) and encrypting each half individually. That data will be kept online. The key of individual manuscript's encryption is placed into cloud hence only we are able to do the opposite operation for the decryption. The algorithm becomes safer as a result. MECC encryption securely trades data on the unsafe networks by swapping the public keys for encryption. MECC method strongly trades data on the untrusted networks by exchanging public keys for encryption. Clients under no circumstances obliged to disclose or swap their private keys, thereby making it the most reliable encryption strategy is available. If they do, it means that the two secretive Diffie Hellman encoded keys are necessary to decipher single private key obtained in ECC. Lowering the possibility that a cybercriminal would intercept a client's private key at the time of communication. Multi-tenant technique is therefore viewed from the standpoint of the cloud service model in a different way. With the aid of key generation, a Modified Elliptic Curve Cryptography (MECC) is presented to address this issue by safeguarding information which is shared to the multi-tenant environment.

Problem Statement

Ongoing studies have diverse difficulties like: computational overheads, time associated with the key creation, clamor/noise responsiveness, difficulties of treating unsized clusters etc. hence disturbing the recovery results from cloud storage. The work intended is endeavored to avoid these impediments through the planned approach.

Objectives

The goals associated with the study is referenced beneath: To carry out novel cryptographical procedure for improved ECC that would drastically minimize the key size & extraordinary security thus overcoming existing restrictions like low operational performance, time as well as the computational intricacy.



**Krishnamoorthy and Umarani**

1. To diminish the scrambling and un scrambling time through firm performance proficiency of suggested method.
2. To rise the accuracy as well as to create alternating keys for security enhancement with the presented scheme.
3. To estimate execution capability of the recommended scheme by means of evaluation criterion namely the encryption as well as the unscrambling time and computation overheads.
4. To relate the success of the proposed model with the other models.

RELATED WORK

The research works deliberates the analogous ideas connected to cloud computing, encryption and also the parameters like: decryption, multi-tenant. These works are explored exhaustively alongside their benefits and detriments. One cloud computing structure for multifaceted layers was outlined by [6] security covers three main factors namely the firewall, management of the user's identity and the encryption subject to the growth of business data synchronization & sharing scheme. The benefit is a fundamental technique which gives improved strength for a multifaceted security in the cloud scenario. The difficulty identified was that the assumption structure don't obtain a wrong alarm within the exact period. An infiltration testing was recognized and obstructed a bug is around 99.95 %. Various investigations examined on the combination of cloud and IoT [7] meant to give few characteristics of few characteristics of the cloud. The benefit could be an advanced CloudIoT scheme was recommended to give an improved data between users. The weakness was that the user is necessary to stop and elucidate thorough exploration of cloud computing disputes within the given time-limit. The Mobile cloud scenario was broadly described by [8], and this method carried out to permit the mobile users to get cloud computing advantages by means of a friendly technique for fulfilling the company requests. The gain is simply the vigorous energy-oriented cloudlet-oriented mobile cloud scheme which can be dedicated appropriately to decide over the additional energy usage during the implementation of wireless communication by employing the vigorous cloudlets oriented scheme. Also the weakness is that the wireless connection and the instrument size is restricted. The fluctuating cloud arrangement [9] and the use of this infrastructure through the different service suppliers and the distributing computational profits are placed far away from the DCs. The outcome necessitates a diverse and innovative figuring schemes that could be obtainable by upcoming cloud setup.

Since there are privacy worries, the trustworthy data have to be encrypted prior to the storing in the cloud. Recently we have come across several works in these area have recommended a number of encryption methods. Nonetheless, most of these prevalent works emphasizes only on the secure searching by applying a keyword, and moreover recovers merely Boolean results, which aren't sufficient. For resolve this concern, [10] a solution was presented with a unique and a protected keyword searching mechanism based on Bloom filter that creates the usage through assisting level established on the obtained examination. Another proposed scheme [11] covers three parts: Trusted Authority (TA), Trusted Cloud (TC), and User. This suggestion is meant to provide additional secure scheme to protect the users' data protection, diminish the complication of key creation by means of a modified Identity Based Cryptography (MIBC), and offer data discretion and integrity by means of the Elliptic Curve Integrated Encryption Scheme (ECIES). The advantage is the reduction of time used for key generation and the downside is the key sizes [27]. Yet another security structure was suggested [12] containing the access controlling scheme, encipher/decipher procedure and the digital signature method. An innovative Uniqueness centered Elliptic curve Access Control mechanism (Id-EAC). This plus point will be the security structure is delivered with great data safety, manageability and truthfulness for the user data. The downside would be the complexity of the algorithm adapted.

In connection with the data encryption and tenant authentication [13] adopted ECC (Elliptic curve cryptography) due to its least key size. From the investigations it was calculated that the average finishing interval is calculated as 86.076 sec for decryption and 83.153 for encryption. Moreover, the scheme indicated that the scheme deals with a two-layer security and considerably uses smaller storage and key size. There are several works, which discourse about the cloud security [14], data security [15, 16] and secured storage [17, 18]. Among them, [19] concentrated on giving security through reliability relationship rubrics. The confirmation scheme applies the cryptographic calculations for protected storing and recovery.





Krishnamoorthy and Umarani

Another technique namely HSBEE CBC [20] was introduced as an competent data recovery methods. As we can see, the scheme enciphers the data by employing ECC method. Additionally, we observe that the approach used was CBC (Cosine based clustering) technique for grouping of the enciphered information. Also interms of elevating the security, the methodology has appraised the trust for the users retrieving the data., once the trust evaluation process is promising, decryption of the data takes place. Additionally, their methodology has assessed the working of suggested scheme and matched the same with that of the usual schemes.To confirm data security, an innovative practice based on “searchable attribute” based on the encryption was carried out [21]. Another scheme named User Usage Based Encryption (UUBE)[22], built on the exploration encryption technique has been offered as a unique differentiated access control structure.As we can see, a control method for retrieving a multi-faced cloud centered facilities by employing the admission governing model based on various features are presented by [23] and use-case is being presented to examine various cloud services. Subsequent to that, a swapping tokens technique was suggested for extending an exclusive model for the situations of the Inter cloud. Analogous calculations were supported and it can also be confirmed that the suggested one owns improved functionality through the process of taking the total consumers and the associated cloud assets.The reliability and trustworthiness for information existing in the relocation of capacity in multi-facet cloud design was accomplished by [24] through an innovative design in the investigation.

PROPOSED METHODOLOGY

The implementation of ECC techniques in scattered computing and dissimilar networking provides substantial usage in inter-reliant networking [25]. ECC is a PKE method built on EC model that produce cryptographic keys faster, lesser as well as highly constructive [26,28]. A basic element is the key strength, that alludes to the intricacy of breaking key and recuperating plain text [26,28]

Elliptic Curve Cryptography (ECC) Model

This is an innovative method of cryptographic tool for safeguarding the information in the physical as well as in the cloud storage. It's a superlative practices established in philosophy of elliptical curve. The characteristics of the EC is utilized to produce bases for encryption procedure as a substitute of prevailing techniques which will be using very outsized prime numbers. It operates over the elliptic curve equation for key creation. In the year 1985, N. Kobiltz and V. Miller suggested ECC for changeable data to get safety. The simple clue is to use the elliptic curve to incorporate a distinct logarithm technique [29]. A most important thing of ECC model is that

is always takes tiny key dimensions to give security. When matched with other schemes (key length of 1064) and whereas the ECC use key length of 164 for the identical level of security. It comes under the public-key mechanism and computation shall proceed from a given point on in the elliptic curve to another. EC is treated equal in x-axis and are binary curves. Encryption as well as signature confirmation are carried out with the public key, on the other hand decryption & signature production are completed with private key

Elliptic Curves

We assume K remain a field. An EC E over K is specified by Weierstrass equation:

$$y^2 = x^3 + Ax + B \quad (1)$$

For cryptography calculations, we use the equation as

$$y^2 = x^3 + ax + b \quad (2)$$

With the above inputs, we submit the proposed algorithm- Improved Elliptic Curve Cryptography (IECC) algorithm

Private Key, Public Key and the Generator Point in ECC

To use ECC, we have to go by the usage of parameters (p,a,b,G,n,h)





Krishnamoorthy and Umarani

$p =$ is a prime number that identifies the magnitude of the given finite field. Here, a & $b =$ are the constants, G is a base point which gives the subgroup of the elliptic curve, $n =$ number of points, called as the order of the subgroup.
 $nG = 0$

$h =$ cofactor of the subgroup given as a ratio of

$$|E| / |Ep| = \text{order of elliptic curve} / \text{order of EC over prime field. } h(3)$$

When two individuals connect with each other, individual person should have private and public keys. They would preserve the private key, however public is offered to all. The Private key will be produced through arbitrarily choosing a digit in the range [1 to $n-1$].

Normally, the Public key is figured out through a calculation, multiplying the private key K_a ($k_a.G$) and a coordinate $G(x,y)$ present in EC. The sender and receiver can thus produce shared secret key (SSK). For an instance, if K_s is taken as the private key of transmitter and K_r indicates private key of the receiver, then we have

$$\text{Sender's public key (spub)} = G * K_s \quad (4)$$

$$\text{Recipients public key (rpub)} = G * K_r \quad (5)$$

Both transmitter and receiver will create Mutual Common Key (MCK):

$$\text{transmitter: MCK} = K_s \cdot (K_r.G) \quad (6)$$

$$\text{receiver: MCK} = K_r \cdot (K_s \cdot G) \quad (7)$$

Proposed algorithm

S and R are two persons involved in data transmission

Step 1: Identify an EC

Sender Select an Elliptic Curve:

E_{Sender} (**x coordinate, Y Coordinate**)

Choose a point $G_{\text{Sender}}(G_x, G_y)$ in the EC

Receiver Select an Elliptic Curve:

E_{Receiver} (x coordinate, Y Coordinate)

Choose a point $G_{\text{Receiver}}(G_x, G_y)$ in the EC

Step 2: Take a huge prime number (n)

Step 3: Generate key sets

Sender(S) generating the keys:

Select Private key $\rightarrow S_{\text{pvt}}$

Check $\rightarrow S_{\text{pvt}} < n$

Take Point G in EC

Calculate Public Key $\rightarrow S_{\text{pub}}$

Receiver (R) generating the keys:

Select Private key $\rightarrow R_{\text{pvt}}$

Check $\rightarrow R_{\text{pvt}} < n$

Take Point G in EC

Calculate Public Key $\rightarrow R_{\text{pub}}$

Step 4: Compute Mutual Common Key MCK $\rightarrow k$

Step 5: Sender and Receiver Exchanges public keys over a channel

Sender share his/her EC and Public key with Receiver

Sender to Receiver $\rightarrow E_{\text{Sender}}$ (**x coordinate, Y Coordinate**)

Sender to Receiver $\rightarrow S_{\text{pub}}$

Receiver share his/her EC and Public key with Sender

Receiver to Sender $\rightarrow E_{\text{Receiver}}$ (**x coordinate, Y Coordinate**)

Receiver to Sender $\rightarrow R_{\text{pub}}$



**Krishnamoorthy and Umarani**

Step 6: Encrypt → Encode file using key of ECC

Step 7: Output → Decode output from step 6 with generated key and ECC

RESULTS AND DISCUSSIONS

The suggested method's outcome is evaluated based on the metrics presented below. Performance investigation are analysed with various factors namely

1. **Encryption Time** –Time required to encode data
2. **Decryption Time**--Time required to decode data
3. **Interval for upload / download**—Time needed for storing/retrieving

Encryption/Decryption Times:

The time taken for decoding /encoding the data is given by these terms. As we can see from Table 1 and Figure 4, we can easily infer that the time taken for encryption and decryption in our model gives minimum time. We also compare the encryption time of our proposed scheme with that of MECC Algorithm suggested by S. Udhaya Chandrika et.al [30] . Tables 2 &3, figures 5&6 clearly shows that the encoding and decoding time in our proposed scheme is far better than that of the MECC method. Hence, we can clearly state that the methodology we have suggested is yielding better results

Uploading/Downloading times: This is termed as the amount of time required for storing data into the cloud and retrieving data from the cloud storage. As we can notice from tables 4,5 and figures 7,8, it is clear that our scheme takes a minimum time for both upload and download when compared with that of the MLS architecture [31].

On matching with the MECC [30] and MLS [31] schemes, our proposed scheme using the elliptic curve cryptography has a lesser encoding/decoding times as well as storage/retrieval time. Henceforth, the security-wise and execution speed, our scheme is producing an efficient results.

CONCLUSION

In this digital era, every individual mainly depends on cloud service providers for keeping their data. In the investigation an innovative security design established on ECC is offered for shielding the end user's data using an elliptical curve procedure. Here, when, a cloud user demand for the access of sensitive data, the suggested technique ensures that the data is always secure. If the invaders are exasperating to crack the data, the same cannot be done since they cannot identify the keys exchanged between the sender and the receiver before the transmission. The performance of the suggested scheme is assessed by means of the parameters namely encoding time, decoding time, and storage effectiveness of data. The algorithm performance is matched with prevailing schemes such as MECC and LSM. From the investigational result study, the proposed technique is realistic and secure. The proposed architecture accomplishes lesser computational overheads and quicker. It also allows ensure the overall security. In upcoming periods, this may be further extended using advance HECC procedures to expand the safety of data stored in cloud.

REFERENCES

1. Aguiar E., Zhang Y. and Blanton M. High Performance Cloud Auditing and Applications (New York, NY: Springer) An overview of issues and recent developments in cloud computing and storage security 3-33, 2014
2. S. Umarani and N. Krishnamoorthy , An experimental study on cloud computing security issues and a framework for xml ddos attack prevention - J. Phys.: Conf. Ser. 2007 012058 , 2021





Krishnamoorthy and Umarani

3. Lavanya, K., Hari, C.B., & Bhaskar Reddy, S. T. (2018). Data Security Using Nested Randomization and Lossless Compression Techniques. *International Journal of Applied Engineering Research*, 13(15), 12373-12378, 2018
4. <https://cloudsecurityalliance.org/press-releases/2016/02/29/cloud-security-alliance-releases-the-treacherous-twelve-cloud-computing-top-threats-in-2016/>
5. Chandini Kumari ;Gagandeep Singh ; Gursharan Singh ; Ranbir Singh Batth-Security Issues and Challenges in Cloud Computing: A Mirror Review- 2019 International Conference on Computational Intelligence and Knowledge Economy (ICCIKE)-EC Accession Number:19392639DOI:10.1109/ICCIKE47802.2019.9004361-Publisher: IEEE-Date Added to IEEE Xplore: 20 February 2020
6. V. Chang, Y.-H. Kuo, and M. Ramachandran, "Cloud computing adoption framework: A security framework for business clouds," *Future Generation Computer Systems*, vol. 57, pp. 24-41, 2016
7. A. Botta, W. De Donato, V. Persico, and A. Pescapé, "Integration of cloud computing and internet of things: a survey," *Future Generation Computer Systems*, vol. 56, pp. 684-700, 2016.
8. K. Gai, M. Qiu, H. Zhao, L. Tao, and Z. Zong, "Dynamic energy-aware cloudlet-based mobile cloud computing model for green computing," *Journal of Network and Computer Applications*, vol. 59, pp. 46-54, 2016
9. Varghese and R. Buyya, "Next generation cloud computing: New trends and research directions," *Future Generation Computer Systems*, vol. 79, pp. 849-861, 2018.
10. F. S. Ali, H. N. Saad, F. H. Sarhan, and B. Naaem, "Enhance manet usability for encrypted data retrieval from cloud computing," *Indonesian Journal of Electrical Engineering and Computer Science*, vol. 18, pp. 64-74, 2020
11. Salim Ali Abbas, Amal Abdul Baqi Maryoosh, "Data Security for Cloud Computing based on Elliptic Curve Integrated Encryption Scheme (ECIES) and Modified Identity based Cryptography (MIBC)", *International Journal of Applied Information Systems (IJ AIS) – Volume 10 – No.6*, pp 7-13, March 2016
12. Kavin, Balasubramanian Prabhu, and Sannasi Ganapathy. "EC (DH) 2: an effective secured data storage mechanism for cloud based IoT applications using elliptic curve and Diffie-Hellman." *International Journal of Internet Technology and Secured Transactions* 10, no. 5 (2020): 601- 617.
13. P. Kumar and A. K. Bhatt, "Enhancing multi-tenancy security in the cloud computing using hybrid ECC-based data encryption approach," *IET Communications*, vol. 14, pp. 3212-3222, 2020.
14. Modak M. and Shaikh R., "Privacy Preserving Distributed Association Rule Hiding Using Concept Hierarchy," *Procedia Computer Science*, vol. 79, pp. 993-1000, 2016.
15. Sahoo C. and Goswami V., "Cost and Energy Optimisation of Cloud Data Centres through Dual VM Modes- Activation and Passivation," *International Journal of Communication Networks and Distributed Systems*, vol. 18, no. 3- 4, pp. 371-389, 2017.
16. Sarkar S., Chatterjee S., and Misra S., "Evacuation and Emergency Management Using a Federated Cloud," *IEEE Cloud Computing*, vol. 1, no. 4, pp. 68-76, 2014.
17. Li L., Lu R., Choo K., Datta A., and Shao J., "Privacy-Preserving-Outsourced Association Rule Mining on Vertically Partitioned Databases," *IEEE Transactions on Information Forensics and Security*, vol. 11, no. 8, pp. 1847- 1861, 2016.
18. Liu X., Deng R., Yang Y., Tran H., and Zhong S., "Hybrid Privacy-Preserving Clinical Decision Support System in Fog-Cloud Computing," *Future Generation Computer Systems*, vol. 78, no. 2, pp. 825-837, 2018
19. [19] Dong B., Liu R., and Wang H., "Result Integrity Verification of Outsourced Frequent Itemset Mining," in *Proceedings of IFIP Annual Conference on Data and Applications Security and Privacy*, Newark, pp. 258-265, 2013.
20. R. Swami and P. Das, "An effective secure data retrieval approach using trust evaluation: HBSEE-CBC," *International Journal of Information and Communication Technology*, vol. 17, pp. 403-421, 2020.
21. M. Lilly Florence & Dhina Suresh , "Enhanced secure sharing of PHR's in cloud using user usage based attribute based encryption and signature with keyword search", *Cluster Computing*, 2019
22. Dhina Suresh & M. Lilly Florence , "Securing Personal Health Record System in Cloud Using User Usage Based Encryption", *Journal of Medical Systems*, 2019
23. C. Ngo, Y. Demchenko, and C. de Laat, "Multi-tenant attribute-based access control for cloud infrastructure services," *Journal of Information Security and Applications*, vol. 27, pp. 65-84, 2016.





Krishnamoorthy and Umarani

24. S. Manikandasaran and S. Raja, "Security Architecture for multi-Tenant Cloud Migration," Int. J. Future Comput. Commun, vol. 7, pp. 42-45, 2018.

25. ShamsheerUllah , Zheng Jiangbin a,, Nizamud Din , Muhammad Tanveer Hussain ,Farhan Ullah , MahwishYousaf , "Elliptic Curve Cryptography; Applications, challenges, recent advances, and future trends: A comprehensive survey", Computer Science Review, <https://doi.org/10.1016/j.cosrev.2022.100530>, Computer Science Review 47 (2023) 100530

26. O. Alowolodu, B. Alese, A. Adetunmbi, O. Adewale, O. Ogundele, Elliptic curve cryptography for securing cloud computing applications, Int. J. Comput. Appl. 66 (23) (2013).

27. J. Nithisha, P. Jesu Jayarin. "A Secured Storage and Communication System for Cloud Using ECC, Polynomial Congruence and DSA" , Wireless Personal Communications, 2022

28. ShamsheerUllah, Jiangbin Zheng, Nizamud Din, Muhammad Tanveer Hussain, Farhan Ullah, MahwishYousaf. "Elliptic Curve Cryptography; Applications, challenges, recent advances, and future trends: A comprehensive survey" , Computer Science Review, Volume 47, February 2023

29. N. NiyazAhamed, N. Duraipandian, "Secured Data Storage Using Deduplication in Cloud Computing Based on Elliptic Curve Cryptography", Computer Systems Science and Engineering ,pp 83-94, 2022

30. S. Udhaya Chandrika, T. PramanandaPerumal," Modified ECC for Secure Data Transfer in MultiTenant Cloud Computing", International Journal of Computer Network and Information Security, pp 76-88, 2022

31. GhadahAldabbagh ,Daniyal M. Alghazzawi , Syed Hamid Hasan , Mohammed Alhaddad ,AreejMalibari and Li Cheng," Secure Data Exchange in M-Learning Platform using Adaptive Tunicate Slime-Mold-Based Hybrid Optimal Elliptic Curve Cryptography", MDPI Applied Sciences, pp 1-21, 2021

Table 1: Encryption time

File Size (MB)	Encryption time (ms)	Decryption time (ms)	Encrypted file size (MB)	Decrypted file Size (MB)
10	4	3.7	0.834	10
20	4.6	4.2	1.34	20
30	5.2	4.8	2.72	30
40	5.6	5.4	3.67	40
50	6.3	5.9	4.29	50

Table 2: Encryption time compared with our proposed method and that of MECC Algorithm suggested by S. Udhaya Chandrika et.al[30]

Key length (bits)	Encryption-time (ms) of MECC Algorithm suggested by S. Udhaya Chandrika et.al[30]	Encryption-time (ms)Our Proposed Scheme
100	5	4.7
128	8	7.2
256	10	8.8
512	15	12.4
1024	24	22.9
2048	39	35.2
4096	51	47.4

Table 3: Decryption time compared with our proposed method and that of of MECC Algorithm suggested by S. Udhaya Chandrika et.al [30]

Key length (bits)	Decryption-time (ms) of MECC Algorithm suggested by S. Udhaya Chandrika et.al [30]	Decryption-time (ms) Our Proposed Scheme
100	11	9.7
128	25	23.2
256	36	34.8
512	51	48.4





Krishnamoorthy and Umarani

1024	63	60.9
2048	83	81.2
4096	159	157.4

Table 4: Uploading time (ms) of MLS Algorithm suggested by GhadahAldabbagh et.al [31] and compared with our proposed method.

File Size (KB)	Uploading-time (ms) of MLS Algorithm suggested by GhadahAldabbagh et.al [31]	Uploading-time (ms) Our Proposed Scheme
10	1995	1837
20	3156	3022
30	4018	3874
40	5095	4934
50	5894	5730

Table 5: Downloading time (ms) of MLS Algorithm suggested by GhadahAldabbagh et.al [31] compared with our proposed method.

File Size (KB)	Downloading time (ms) of MLS Algorithm suggested by GhadahAldabbagh et.al [31]	Downloading time (ms) Our Proposed Scheme
10	1266	1158
20	1988	1872
30	2146	2032
40	3084	2911
50	3982	3812

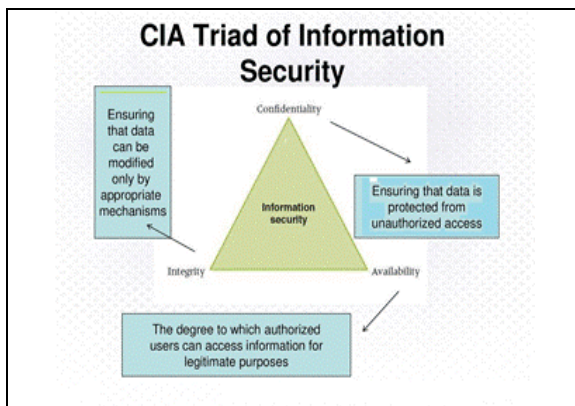


Fig 1. Security Issues in Cloud

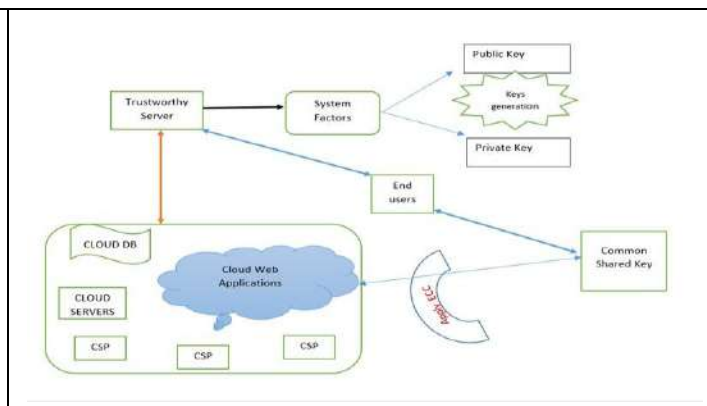


Fig 2: Outline of validation in web application using ECC

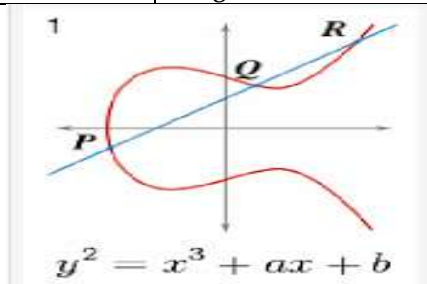


Fig 3: A typical Elliptic Curve





Krishnamoorthy and Umarani

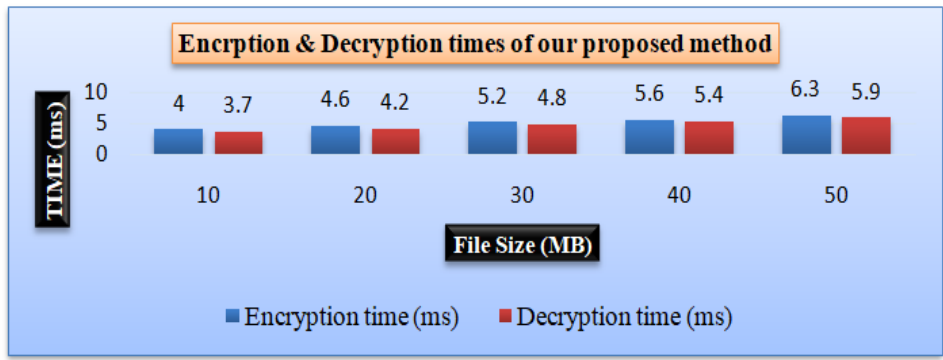


Figure 4: Comparison of Encryption /Decryption times

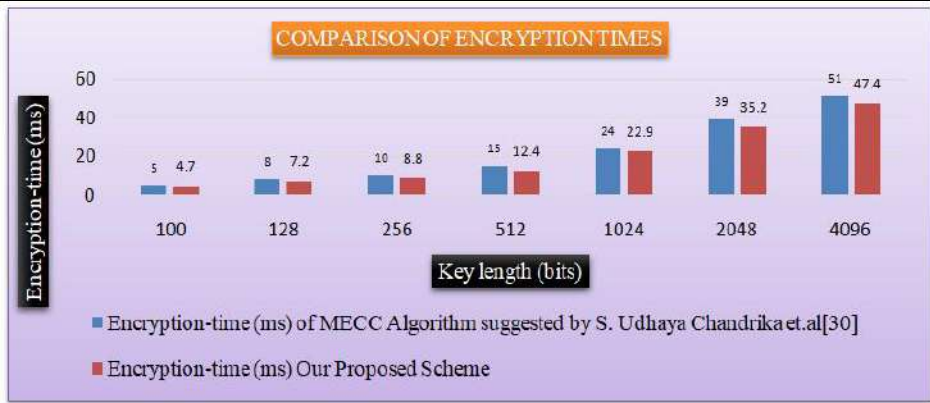


Figure 5: Comparison of Encryption times

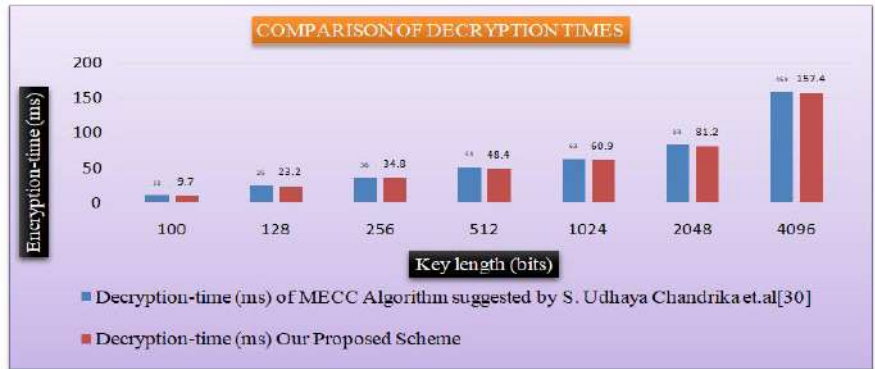


Figure 6: Comparison of Decryption times





Krishnamoorthy and Umarani

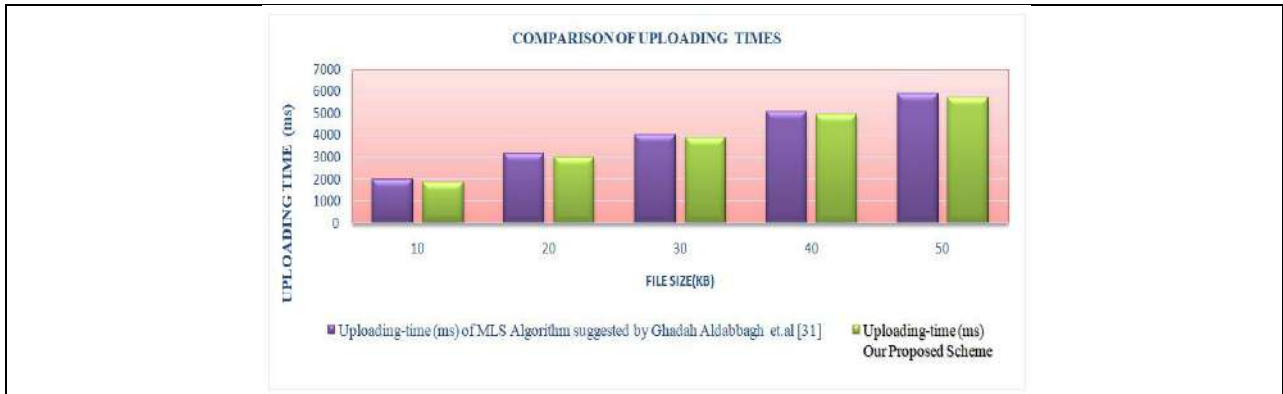


Figure 7: Comparison of uploading time



Figure 8: Comparison of Down loading time





Isolation, Purification, and Characterization of Flavonoid from the Aqueous Extract of the Leaves of the *Phaseolus vulgaris* Linn.

Madhavi Badole¹ and Amrin Shaikh^{2*}

¹Associate Professor, Department of Chemistry, University of Mumbai, Ramnarain Ruia Autonomous College, (Affiliated to University of Mumbai), Mumbai, Maharashtra, India.

²Research Scholar, Department of Chemistry, University of Mumbai, Ramnarain Ruia Autonomous College, (Affiliated to University of Mumbai), Mumbai, Maharashtra, India.

Received: 18 Jun 2024

Revised: 29 Sep 2024

Accepted: 04 Nov 2024

*Address for Correspondence

Amrin Shaikh

Research Scholar, Department of Chemistry,
University of Mumbai, Ramnarain Ruia Autonomous College,
(Affiliated to University of Mumbai),
Mumbai, Maharashtra, India.
E.Mail: amrinshaikh@ruiacollege.edu



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Phaseolus vulgaris Linn. is native to America but is extensively distributed in warm nations like India. Commonly, it is known as "kidney bean or common bean." The seeds of the plant have undergone extensive study for their medicinal properties, but the leaves of the plant have not been studied much. The aqueous extracts offer significant benefits in terms of health, safety, environmental impact, and practicality. It is crucial to thoroughly study and implement them to create safe, efficient, and sustainable products across diverse disciplines. Therefore, the current aim of the study is to isolate, purify, and characterize flavonoids from aqueous extract of the leaves of the *Phaseolus vulgaris* Linn. Separation of flavonoid achieved by Sephadex LH-20 column chromatography from aqueous extract and enriched by preparative HPLC method. The weight of the isolated compound is 24 mg. The isolated compound was characterized by ¹H-NMR spectroscopy, mass spectroscopy, ¹³-carbon NMR, DEPT, and D₂O exchange NMR. The spectroscopic analysis gives the structure of a flavonoid as rutin. Alternatively, it is known as 3, 3', 4', 5, 7-pentahydroxyflavone-3-rhamnoglucoside.

Keywords: ¹³-Carbon NMR, DEPT-135, ¹H-NMR, Isolation, LC-MS, Rutin,

INTRODUCTION

Flavonoids, predominantly found in a diverse array of plants used for culinary, medicinal, and cultural purposes, have played a vital role in human health and society since ancient times. The current aim of the study is the isolation, purification, and characterization of flavonoids from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn. This is not only a medicinal plant but also one with proven nutritional value. Its widespread cultivation highlights its



**Madhavi Badole and Amrin Shaikh**

importance as a flexible and priceless commodity to the global community.[1]Flavonoids, including myricetin, kaempferol, and quercetin, and their glycosides, have been identified from the bean *Phaseolus vulgaris* Linn.[2],[3].

Aqueous extraction is better for the environment than organic solvents. To enrich the purity of the compound, several important procedures are involved in the isolation and purification of a flavonoid. Sephadex LH-20 is particularly useful for purification since it has the ability to separate compounds based on their size and polarity. Mainly, secondary metabolites from the plant can be separated, as phytoconstituents in plants have a similar polarity range. Upon good maintenance, resin shows good performance, and its capacity for recycling makes it cost an effective option for the isolation of natural products. Due to its regeneration ability, it has wide applications in research and development, industries, and companies. [4]. To validate the structural and functional properties of a flavonoid, characterization is necessary. In-depth details about the molecular makeup, molecular weight, and arrangement of protons of a compound can be obtained by methods including ¹H-NMR spectroscopy, LCMS, and ¹³Carbon NMR, DEPT, D₂O exchange NMR.[5]

MATERIALS AND METHODS

Collection, authentication of plant material

Phaseolus vulgaris Linn. was authenticated by St. Xavier's College, Mumbai. The plant specimen collected from Dapoli was matched with the herbarium, and its corresponding voucher number is D.P.2275.

Chemicals

Sephadex LH-20 was obtained from Sigma Aldrich. All the solvents used are HPLC analytical grade.

Extraction

An important step in isolating a component from a plant is to produce aqueous extracts using successive extraction techniques. By using a series of increasingly polar solvents, sequential extraction separates substances based on their solubility.

Procedure

A total of 50 grams of dried, lyophilized plant powder was precisely measured and placed in a beaker, subsequently the inclusion of 1000 milliliters of pet ether. The mixture was subjected to sonication for approximately 10 minutes. After this period, the plant extract was passed through a Whatman filter No. 1, and the filtrate was concentrated. The weight of the empty filtering vessel and the weight of the vessel containing the extract were recorded. The same procedure was repeated with dichloromethane (DCM), ethyl acetate (EAA), methanol, and water. The yield of other extracts had been disregarded. The crude aqueous extract obtained through lyophilization has a yield of 30 %, which is employed for the isolation of a flavonoid.^[6]

Size exclusion chromatography

Sephadex LH-20, a polysaccharide composed of cross-linked dextran with hydroxypropyl groups, is a versatile gel filtration (size exclusion) chromatography medium employed in biochemical and biophysical research for purifying and separating small molecules, particularly natural products, peptides, and low molecular weight compounds. Its exceptional ability to dissolve in organic solvents and effectively mix with both polar and non-polar solvents like acetone, methanol, ethanol, and water gives it the power to enable the separation of a wide variety of chemicals.^{[7],[8],[9],[10]}

Procedure

Initially, LH-20 resin was soaked with 100% methanol overnight. After that, it is washed three times with 100% methanol. This helps remove previous sample cleaning from resin. The glass column of dimension 1 ft. × 10 cm was taken for size exclusion chromatography. The glass column was washed with methanol, and cotton was plugged into



**Madhavi Badole and Amrin Shaikh**

the end of the column. Wet the cotton plug with the solvent. Pack the glass column with Sephadex LH-20 resin. Allow the methanol to pass through it. Make sure that column does not get dry.^[11]

Sample Loading

13 grams of crude aqueous extract were precisely measured and dissolved in methanol. Next, the filtrate was carefully poured into the column without disturbing the bed, allowing the methanol extract to flow continuously through it. The 50-mL test tubes were filled with the collected fractions after adjusting the flow rate to 1 mL/min of the column. The chromatographic analysis of fractions from the column was carried out using RP-HPLC. It was found that fractions 53–78 were pure, with a purity level of 74.88%. These fractions were subsequently combined and concentrated through rotary evaporation, ultimately yielding a final product weighing approximately 475 mg. The purity of the isolated compound was enriched by preparative HPLC column chromatography. The RP-HPLC method and preparative HPLC method adopted for the analysis of fractions are mentioned below:

RP-HPLC method;

HPLC using a Waters 2695 separation module coupled with a PDA detector (2996) was implemented. The Hemochrom C18, a 150 mm × 4.6 mm, 5-micron particle size column, was utilized as the stationary phase. A solvent A (acetonitrile, ACN) and solvent B (0.05% trifluoroacetic acid, TFA) gradient program was employed as the mobile phase. The gradient program had a run time of 24 minutes, a delay time of 6 minutes, and the following conditions: 0–20 minutes, 0–100% ACN; 20–24 minutes, 100% ACN; 24–25 minutes, 0% ACN; and 25–30 minutes, 0% ACN. The maximum wavelength of absorption (λ_{max}) was set at 210 nm, and the flow rate was 1 mL/min.^[12]

Preparative liquid chromatography;

The 475 mg compound isolated from the Sephadex LH-20 column can be enriched by the preparative HPLC method. The compound was dissolved in 1:1 HPLC water with acetonitrile as an eluent. An analytical workstation was connected to the HPLC system, which is made up of an auto-sampler, a pump, and a photodiode array detector. The column setup included a 250 × 20 mm, 5-micron Supelco C18 reverse-phase column. The flow rate was 18.0 mL/min, and the detection wavelength was set at 210 nm. The autosampler tray was maintained at 35°C, and the column was allowed to reach ambient temperature. The mobile phase for the HPLC was a step gradient of ACN and 0.05% TFA. The flavonoid was detected at 210 nm by the photodiode array detector, and individual peaks were recorded. Fractions were collected and isolated using a fraction collector. The solvent was then removed using a rotary evaporator.

Characterization of isolated compound**LCMS**

The LC-MS/MS system used in the study was an Acquity Triple Quadrupole (TQD) system equipped with Mass Lynx V4.2 software (Waters Corp. UK). The chromatographic conditions for LC-MS analysis were carried out using an Inertsil ODS-3 column (150×4.6mm, 3 μ). The column eluent was introduced to the MS inlet through an electrospray ionization source operated in positive polarity with a cone voltage of 30V. The capillary voltage was set at 3 kV, while the source and desolvation temperatures were 150° and 500°, respectively. Nitrogen gas was used as the cone and desolvation gas at flow rates of 50 L h⁻¹ and 500 L h⁻¹, respectively. The collision-induced dissociation (CID) experiments were carried out using argon as the collision gas at a flow rate of 0.19 mL/min, with a collision energy applied in the range of 10 to 12 V and a scan time of 0.2s. The mass spectrometer was operated at unit mass resolution. The mobile phases 'A' and 'B' were 0.05% TFA (AR Grade, Merck) and acetonitrile HPLC grade (Finar), respectively. Separation was achieved using a gradient program with the following time (min)/mobile phase 'B' (%) parameters: 0.01/0, 1/0, 20/100, 24/100, 25/0, and 30/0, with a flow rate of 1 mL/min. The column was maintained at 30°, and the eluent was split 1:4 using a Valco valve, with a small portion sent to the MS and the major portion monitored at UV 210 nm. The injection volume was 10 μ L. For the preparation of the diluent, acetonitrile was used. The preparation of analytical solutions involved the creation of a test solution at a concentration of 1000 μ g/mL.





¹H NMR, ¹³C- NMR and DEPT-135;

All NMR experiments were undertaken using the Avance III HD 400 MHz NMR Spectrometer, equipped with Top Spin software version 3.5p16 and a Probe PA BBO 500S1BBF-H-D-05 Z from Bruker Biospin Ltd. (Switzerland). Experiments were thoughtfully conducted at room temperatures to ensure accurate results. The solvent chosen to obtain ¹H NMR, ¹³C NMR, and DEPT-135 spectra was DMSO-D₆ (99.8% D, Eurisotope). ¹H and ¹³C spectroscopy give a clear picture of the structure of an unknown compound. ¹H spectroscopy provides information about the environment of hydrogen atom; therefore, the identification of the structure of a compound becomes easier. The ¹³C spectra are less complicated as compare to the ¹H spectra. ¹³C spectroscopy gives details of the carbon skeleton. Combination of ¹H and ¹³C spectroscopy identification of unknown compounds is possible.[13] DEPT spectroscopy provides information about the connectivity of hydrogen atoms to carbon atoms. It is widely applicable to the characterization of natural products. This technique gives a brief classification of the types of the carbon atoms based on the connectivity of hydrogen atoms. DEPT provides phase-sensitive data and enhances the sensitivity of carbon-¹³ NMR signals, making it easier to differentiate between different carbon atoms.[14]

RESULTS

Preparative HPLC analysis

The advanced technique of preparative HPLC is essential for isolating specific chemicals in large quantities from complex mixtures. Its application is widespread across diverse sectors, including pharmaceuticals, biotechnology, and chemical manufacturing, owing to its exceptional efficiency and accuracy. The elution began with acetonitrile at 0%, gradually increasing to 2%, 4%, 6%, and so on up to 16% for 80 minutes. All experiments were conducted at room temperature, which was maintained at 25 ± 2°. A chromatogram of all the fractions was recorded at a wavelength of 210 nm, serving as a monitoring wavelength. The compound was eluted at 16% for 80 minutes. All fractions were collected in 50-ml test tubes. Following analysis using the analytical RP-HPLC method, fractions ranging from 28 to 35 were combined and concentrated using rota vapor. The final product obtained weighed 24 mg and had a percentage purity of more than 98%.

The preparative chromatogram for the isolated compound is clearly illustrated in Figure 1. According to the chromatogram, the retention time of the compound is 8.09 minutes, it has a single sharp peak, and its percentage area is 100%. The UV absorption spectrum of the compound shows peaks at 255nm and 355nm wavelengths, which is consistent with the UV absorption spectrum of rutin as reported in the literature. Consequently, based on the UV data, it is confirmed that the isolated fraction is rutin.[15],[16],[17],[18],[19]

Characterization of an isolated compound

Characterization of an isolated compound can be done by a combination of spectroscopic techniques such as LCMS, ¹H-NMR, ¹³C NMR, D₂O, and DEPT-135. By analyzing this data and comparing it with literature, the characterization of a compound can be done.

LCMS/MS;

Observed full scan +ESI mode gives m/z value 611.1517 [M+H]⁺ with molecular formula C₂₇H₃₀O₁₆. The following figure represents the structure of rutin. The m/z value matches the reference.[20],[21] The structure of rutin contains a quercetin moiety (benzopyranone ring and benzene ring attached to it) and a sugar moiety (glucose and rhamnose ring) connected by glycoside linkage.[22],[23] The numbering of each carbon is given as shown in the structure for ease of characterization. The spectrum of the compound clearly shows a peak at 611.3 m/z as a molecular ion peak. Loss of rhamnose sugar gives fragments at 468.3 m/z, followed by fragmentation by loss of glucose unit at 313.4 m/z. The benzopyranone gives one fragmentation at 155.1 m/z and another fragmentation at 176.1 m/z. The fragmentation pattern is essential to estimating the structure of a compound. The fragmentation pattern obtained is closely matched with the literature. Hence, it is confirmed that the isolated fraction from the aqueous extract of the plant is rutin.[24],[25],[26]



**Madhavi Badole and Amrin Shaikh****¹H-NMR analysis and ¹³C analysis (DMSO d₆);**

In chemistry, ¹H and ¹³C NMR spectroscopy are crucial analytical methods for identifying functional groups, elucidating structures, and figuring out molecule conformation and dynamics.^[27] ¹H NMR is useful for identifying hydrogen environments, assessing purity, and providing quantitative analysis, while ¹³C NMR offers detailed insights into the carbon skeleton of organic compounds, aiding in distinguishing isomers and supporting ¹H NMR data for a complete molecular picture.^[28] Both methods are non-destructive, preserving samples for further analysis, and they can be used on a variety of substances, including small organic molecules, polymers, natural products, and complex biomolecules. The isolated compound from the aqueous extract of leaves of *Phaseolus vulgaris* Linn. shows the δ values in parts per million (ppm).^{[29],[30]} Fig. 4 and Fig. 5 represent the NMR and ¹³C spectra of the isolated compound. Table 1 and Table 2 represent the analysis of ¹H and ¹³C NMR data for a separated compound.

There exists a close match between the chemical shift value produced for an isolated compound and the chemical shift values that exist in the literature. Therefore, it is verified that the isolated substance is rutin.

D₂O Exchange NMR;

This method identifies and separates exchangeable protons in hydroxyl, amine, and amide groups by replacing them with deuterium from D₂O. This alters or removes the signals in the proton NMR spectrum, providing clear evidence of the presence and environment of these functional groups. This method considerably improves structural elucidation, which is necessary to precisely ascertain the molecular structure and dynamics of complex natural products, by verifying the positions of exchangeable protons.^{[31],[32]}

In D₂O Exchange NMR, signals at δ 12.60, 10.84–10.86, 9.68–9.74, 9.16–9.19, 5.29–5.35, 5.07–5.16, and 4.35–4.55 disappeared, indicating that these are exchangeable protons of hydroxyl groups. Thus, the identity of the isolated compound as rutin has been verified.

DEPT-135 NMR

This method simplifies the interpretation of complex spectra and aids in accurately estimating molecular structures by clearly revealing the carbon-hydrogen framework, which is critical for comprehending the overall architecture and functional group distribution inside a molecule.^[32] Fig. 6 represents the DEPT-135 NMR spectra. The DEPT-135 spectra show CH/CH₃ carbon has a positive peak while CH₂ carbon has a negative peak. DEPT spectra provide insight into the environments of the various carbon atom types found in the intricate structure. The DEPT-135 NMR data analysis of an isolated compound is shown in Table 3.

DISCUSSION

Sephadex LH-20 comes in particle sizes ranging from 20 to 80 microns, impacting chromatographic resolution and flow rate. It efficiently separates substances with molecular weights up to several thousand Daltons, making it ideal for purifying peptides and small organic molecules. Sephadex LH-20 enables high-resolution separations that are important for isolating closely related compounds or isomers. Furthermore, it is easy to use because it can be packed into different column sizes depending on the scale of separation required, from analytical to preparative. Sephadex LH-20 is useful for the purification and analysis of natural products and other small chemicals in various scientific fields due to its versatility, solvent compatibility, high resolution, and ease of use.^{[33],[34]}

The substance purified using a Sephadex LH-20 column was further purified to a level of over 95% using preparative HPLC. Preparative HPLC is an indispensable method in chemistry and biochemistry for isolating and purifying specific chemicals from complex mixtures. It is vital for producing the pure molecules required for pharmaceutical development, research, and various industrial applications due to its high resolution, accuracy, and ability to handle large numbers of samples. Preparative HPLC ensures the purity and effectiveness of chemicals used in biochemical research, pharmaceutical formulations, and synthetic chemical processes by effectively separating target molecules from contaminants.



**Madhavi Badole and Amrin Shaikh**

Rutin has unique ¹H NMR spectrum features. The ¹H NMR spectrum, which is usually recorded in DMSO-d₆, has several signals between δ 6.0 and 8.0 ppm, which are characteristic of the aromatic protons on the flavonoid rings. δ 4.5 to 5.35 ppm gives a signal from anomeric protons in rhamnose. The chemical shift values δ 3.00 ppm to 4.5 ppm give a signal from the remaining protons from glucose and rhamnose sugar. According to D₂O exchange NMR, the chemical shift from δ 9.00 ppm to 12.00 ppm gives signals from protons of exchangeable hydrogen atoms of the hydroxyl group. The DEPT-135 spectra give information about the carbon-hydrogen relationship in the complex structure. There are tenquaternary, fourteen –CH, one –CH₂ and one –CH₃ carbon atoms present. This information helps to establish the structure of rutin.

CONCLUSION

To the best of our knowledge, isolation of rutin has not been done yet from an aqueous extract of the leaves of *Phaseolus vulgaris* Linn. Other names for rutin include rutoside, quercetin-3-rutinoside, and sophorin.^[22] Rutin is one of the many health advantages of flavonoid glycosides; it is notable for its historical and modern importance. Due to its well-established health benefits, it is used in both conventional and modern medicine.^[35] Rutin, a substance with a wide range of pharmacological characteristics, has gained lots of attention because of its anti-inflammatory, anti-carcinogenic, and antioxidant effects on the cardiovascular system.^{[22],[36]} The study provides a reliable method for producing high-purity rutin and outlines a strategy to ensure the consistency and reliability of the substance for future pharmacological and biological research. It emphasizes the importance of accurate characterization. The results of this study will contribute to standardizing methods for characterizing and isolating rutin, making it easier to use in various scientific and medical fields. In the future, advances in green chemistry, biotechnology, nanotechnology, and extraction technologies will greatly benefit the isolation of rutin from aqueous extracts. These developments will significantly expand the use of rutin in functional foods, medications, and cosmetics, while also improving its sustainability and extraction efficiency. To fully realize the potential of rutin as a highly valued natural commodity, it is necessary to prioritize environmentally friendly techniques and cutting-edge technologies.

ACKNOWLEDGEMENT

Ramnarain Ruia Autonomous College generously supplied the seed funding for this research endeavour. The experiments, data analysis, and manuscript writing were all completed by the corresponding author, Amrin Shaikh. Dr. Madhavi Badole, the research guide, provided essential direction, oversight, and review for the manuscripts.

REFERENCES

1. Devi M, Dhanalakshmi S, Thillai Govindarajan GE, Tanisha BA, Sonalika T, Ruth JE, et al. A review on phaseolus vulgaris linn. *Pharmacogn J* 2020;12(5):1160–4.
2. Aghajanian S, Kazemi S, Esmaili S, Aghajanian S, Moghadamnia AA. Sequential microwave-assisted extraction for isolation of quercetin from red kidney bean. *Int J Eng Trans A Basics* 2020;33(1):12–7.
3. Islas JF, Dávalos-balderas AJ, Arroyo-currás N, Cano BG, Galindo-jacobo P, Guajardo-salinas G, et al. Comparative Evaluation of a Modified Acetic Method for Extraction of Antioxidant Compounds from Black Beans (*Phaseolus vulgaris*) *. 2012;2012(March):348–53.
4. Bakhytkyzy I, Nuñez O, Saurina J. Size Exclusion Coupled to Reversed Phase Liquid Chromatography for the Characterization of Cranberry Products. *Food Anal Methods* 2019;12(2):604–11.
5. Arora S, Itankar P. Extraction, isolation and identification of flavonoid from *Chenopodium album* aerial parts. *J Tradit Complement Med [Internet]* 2018;8(4):476–82. Available from: <https://doi.org/10.1016/j.jtcme.2017.10.002>
6. Agatonovic-Kustrin S, Morton DW, Mizaton HH, Zakaria H. The relationship between major polyphenolic acids and stigmaterol to antioxidant activity in different extracts of *Myrmecodia platytyrea*. *South African J Bot [Internet]* 2018;115:94–9. Available from: <https://doi.org/10.1016/j.sajb.2017.12.011>



**Madhavi Badole and Amrin Shaikh**

7. Janiak M, Slavova-Kazakova A, Kancheva V, Amarowicz R. Sephadex LH-20 column chromatography of the hydrolysed lignan macromolecule of flaxseed. *Bulg Chem Commun* 2014;46(3):640–4.
8. Mottaghipisheh J, Iriti M. Sephadex® LH-20, Isolation, and Purification of Flavonoids from Plant Species: A Comprehensive Review. *Molecules* 2020;25(18):1–19.
9. Tian Y, Liimatainen J, Pугanen A, Alakomi HL, Sinkkonen J, Yang B. Sephadex LH-20 fractionation and bioactivities of phenolic compounds from extracts of Finnish berry plants. *Food Res Int* [Internet] 2018;113:115–30. Available from: <https://doi.org/10.1016/j.foodres.2018.06.041>
10. Zhang QW, Lin LG, Ye WC. Techniques for extraction and isolation of natural products: A comprehensive review. *Chinese Med (United Kingdom)* [Internet] 2018;13(1):1–26. Available from: <https://doi.org/10.1186/s13020-018-0177-x>
11. Cheng HL, Zhang LJ, Liang YH, Hsu YW, Lee IJ, Liaw CC, et al. Antiinflammatory and Antioxidant Flavonoids and Phenols from *Cardiospermum halicacabum* (倒地鈴 *Dào Dì Líng*). *J Tradit Complement Med* [Internet] 2013;3(1):33–40. Available from: [http://dx.doi.org/10.1016/S2225-4110\(16\)30165-1](http://dx.doi.org/10.1016/S2225-4110(16)30165-1)
12. Raafat K, al Haj M. Modulators of diabetic neuropathy and inflammation from *Saponaria officinalis*: Isolation of active phytochemicals and potential mechanisms of action. *J Tradit Complement Med* [Internet] 2023;13(3):226–35. Available from: <https://doi.org/10.1016/j.jtcme.2023.01.001>
13. Yeh PH, Shieh YD, Hsu LC, Kuo LM, Lin JH, Liaw CC, et al. Naturally occurring cytotoxic [3'→8'']-biflavonoids from *podocarpus nakaii*. *J Tradit Complement Med* [Internet] 2012;2(3):220–6. Available from: [http://dx.doi.org/10.1016/S2225-4110\(16\)30103-1](http://dx.doi.org/10.1016/S2225-4110(16)30103-1)
14. Akhtar MS, Hossain MA, Said SA. Isolation and characterization of antimicrobial compound from the stem-bark of the traditionally used medicinal plant *Adenium obesum*. *J Tradit Complement Med* [Internet] 2017;7(3):296–300. Available from: <http://dx.doi.org/10.1016/j.jtcme.2016.08.003>
15. Pinto D, Lameirão F, Delerue-Matos C, Rodrigues F, Costa P. Characterization and stability of a formulation containing antioxidants-enriched *castanea sativa* shells extract. *Cosmetics* 2021;8(2):1–19.
16. Lipi F, Parvin S, Islam R. UV Spectrophotometric Method Development and Validation for Determination of Levocetirizine Dihydrochloride. *Am J PharmTech Res* [Internet] 2016;6(2). Available from: <http://www.ajptr.com/>
17. Pawar NP, Salunkhe VR. Development and validation of UV spectrophotometric method for simultaneous estimation of rutin and gallic acid in hydroalcoholic extract of *Triphala churna*. *Int J PharmTech Res* 2013;5(2):724–9.
18. Mutakin M, Juwita T, Megantara S, Puspitasari IM, Levita J. Determination of quercetin and rutin in the ethyl acetate fraction of *etlingera elatior* (Jack) R.M. Smith flower by reversed-phase liquid chromatography-mass spectroscopy. *Rasayan J Chem* 2020;13(3):1379–85.
19. Ashok PK, Saini B. HPLC Analysis and Isolation of Rutin from Stem Bark of *Ginkgo biloba* L. *J Pharmacogn Phytochem* 2013;2(4):68–71.
20. Chen M, Zhang X, Wang H, Lin B, Wang S, Hu G. Determination of rutin in rat plasma by ultra performance liquid chromatography tandem mass spectrometry and application to pharmacokinetic study. *J Chromatogr Sci* 2015;53(4):519–25.
21. MacIková P, Halouzka V, Hrbac J, Bartak P, Skopalova J. Electrochemical behavior and determination of rutin on modified carbon paste electrodes. *Sci World J* 2012;2012(April).
22. Ganeshpurkar A, Saluja AK. The Pharmacological Potential of Rutin. *Saudi Pharm J* [Internet] 2017;25(2):149–64. Available from: <http://dx.doi.org/10.1016/j.jsps.2016.04.025>
23. Satari A, Ghasemi S, Habtemariam S, Asgharian S, Lorigooini Z. Rutin: A Flavonoid as an Effective Sensitizer for Anticancer Therapy; Insights into Multifaceted Mechanisms and Applicability for Combination Therapy. *Evidence-based Complement Altern Med* 2021;2021(Figure 1).
24. Rini Vijayan KP, Raghu A V. Tentative characterization of phenolic compounds in three species of the genus *Embelia* by liquid chromatography coupled with mass spectrometry analysis. *Spectrosc Lett* [Internet] 2019;52(10):653–70. Available from: <https://doi.org/10.1080/00387010.2019.1682013>
25. Chen Y, Yu H, Wu H, Pan Y, Wang K, Jin Y, et al. Characterization and quantification by LC-MS/MS of the chemical components of the heating products of the flavonoids extract in *Pollen typhae* for transformation rule exploration. *Molecules* 2015;20(10):18352–66.





Madhavi Badole and Amrin Shaikh

26. Kumar S, Singh A, Kumar B. Identification and characterization of phenolics and terpenoids from ethanolic extracts of Phyllanthus species by HPLC-ESI-QTOF-MS/MS. J Pharm Anal [Internet] 2017;7(4):214–22. Available from: <http://dx.doi.org/10.1016/j.jpha.2017.01.005>
27. Dayrit FM, Dios AC de. 1H and 13C NMR for the Profiling of Natural Product Extracts: Theory and Applications. Spectrosc Anal - Dev Appl 2017;
28. Lia F, Vella B, Mangion MZ, Farrugia C. Application of 1H and 13C NMR fingerprinting as a tool for the authentication of maltese extra virgin olive oil. Foods 2020;9(6):1–14.
29. Immaculate AA, Vimala JR, Takuwa DT. Isolation and Characterisation of Flavonoids (Rutin) from the Roots of Cadaba aphylla (Thunb) and Adenia glauca of Potential in Anti-Oxidant activity. Orient J Chem 2022;38(6):1404–13.
30. Malik J, Gandhi R, Vishwavidyalaya P. CHARACTERIZATION OF RUTIN ISOLATED BY LEAVES ANNONA SQUAMOSA Biomedical European of AND Pharmaceutical sciences. 2020;5(April).
31. Haslauer KE, Hemmler D, Schmitt-Kopplin P, Heinzmann SS. Guidelines for the Use of Deuterium Oxide (D₂O) in 1H NMR Metabolomics. Anal Chem 2019;91(17):11063–9.
32. Investigations M. Mechanistic Investigations. 2021;
33. Healthcare GE. Sephadex LH-20.
34. Chen X, Song H, Zhou S, Yuan C, Li J. Exploring separation patterns and mechanisms of proanthocyanidins in grape seeds and pomace with diverse molecular weights, compositions, and structures. Food Chem X [Internet] 2023;20(22):101008. Available from: <https://doi.org/10.1016/j.fochx.2023.101008>
35. Abdullahi S. Isolation of rutin from the leaf of. J Pure Appl Sci 2021;(December).
36. Manasa P, Suhasin G. RUTIN - A POTENTIAL BIOFLAVONOID WITH VARIOUS PHARMACOLOGICAL ACTIONS Pathivada Manasa and Ganta Suhasin * Department of Pharmacology, GITAM Institute of Pharmacy, GITAM, Deemed to be University, Visakhapatnam - 530045, Andhra Pradesh, India. 2022;13(2):598–602.
37. Prasad R, Prasad SB. A review on the chemistry and biological properties of Rutin, a promising nutraceutical agent. Asian J Pharm Pharmacol 2019;5(S1):1–20.

Table 1: 1H NMR data analysis of a compound isolated from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn. (ppm- Parts Per Million)

Atom Number (Position)	Chemical shifts (δ ppm) observed	Chemical shifts (δ ppm) Literature	No. of protons
OH-5	12.60 (s)	12.75 (s)	1
H (6)	6.19 – 6.20 (s)	6.1 – 6.2 (s)	1
OH-7	10.84 – 10.86 (d)	10.75	1
H (8)	6.38 - 6.40 (d)	6.3 – 6.4	1
H (2')	7.52 – 7.55 (s)	7.5	1
OH- 3'	9.68 – 9.74 (d)	9.75	1
OH-4'	9.16 – 9.19 (d)	9.25	1
H (5')	6.81 – 6.85 (d)	6.75	1
H (6')	7.64 – 7.67 (d)	7.5	1
H (1''), OH-2''	5.29 – 5.35 (m)	5.25 – 5.35	2
H (2'' – 6''), H(2'''- 5''')	3.06 – 3.72 (m)	3.0 – 4.0	10
OH-3'', OH-4''	5.07 – 5.16 (m)	5.0 – 5.1	2
H (1''')	4.40 – 4.41 (d)	4.9	1
OH-2''', OH-3''',OH-4'''	4.35 – 4.55 (m)	4.0 – 4.5	3
H (6''')	0.98 – 1.07 (d)	0.8 – 1.0	3





Madhavi Badole and Amrin Shaikh

Table 2: ¹³C-NMR data analysis of an isolated compound from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn. (ppm- Parts Per Million)

Atom Number (Position)	Chemical shifts (δ ppm) observed	Chemical shifts (δ ppm) Literature	No. of Carbons
C (2)	156.61	157.1	1
C (3)	133.29 – 133.48	133.7	1
C (4)	177.41	177.8	1
C (5)	161.21	161.6	1
C (6)	99.98	99.1	1
C (7)	164.07 – 164.12	164.5	1
C (8)	93.57	94.1	1
C (9)	156.34 – 156.42	156.9	1
C (10)	103.90 – 103.96	104.4	1
C (1')	121.05 – 121.16	121.6	1
C (2')	115.97	115.7	1
C (3')	144.75 – 144.82	145.2	1
C (4')	148.41 – 148.50	148.8	1
C (5')	116.25	116.7	1
C (6')	121.94	122.1	1
C (1'')	101.99	101.6	1
C (2'')	74.06	74.5	1
C (3'')	76.43	76.9	1
C (4'')	71.09	71.0	1
C (5'')	75.90	76.3	1
C (6'')	67.01	67.4	1
C (1''')	101.16	101.2	1
C (2''')	70.54 – 70.60	70.8	1
C (3''')	70.36 – 70.43	70.4	1
C (4''')	71.82 – 71.91	72.3	1
C (5''')	68.28	68.7	1
C (6''')	17.74 – 17.93	18.7	1

Table 3: DEPT NMR data analysis of an isolated compound from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn. (ppm- Parts Per Million)

Atom Number (Position)	Chemical shifts (δ ppm)	Nature of Carbon
C (2)	156.61	Quaternary
C (3)	133.29 – 133.48	Quaternary
C (4)	177.41	Quaternary
C (5)	161.21	Quaternary
C (6)	99.98	-CH
C (7)	164.07 – 164.12	Quaternary
C (8)	93.57	-CH
C (9)	156.34 – 156.42	Quaternary
C (10)	103.90 – 103.96	Quaternary
C (1')	121.05 – 121.16	Quaternary
C (2')	115.98	-CH
C (3')	144.75 – 144.82	Quaternary





Madhavi Badole and Amrin Shaikh

C (4')	148.41 – 148.50	Quaternary
C (5')	116.26	-CH
C (6')	121.95	-CH
C (1'')	102.00	-CH
C (2'')	74.07	-CH
C (3'')	76.43	-CH
C (4'')	71.10	-CH
C (5'')	75.91	-CH
C (6'')	67.00	-CH ₂
C (1''')	101.17	-CH
C (2''')	70.55 – 70.62	-CH
C (3''')	70.38 – 70.44	-CH
C (4''')	71.83 – 71.92	-CH
C (5''')	68.29	-CH
C (6''')	17.76 – 17.95	-CH ₃

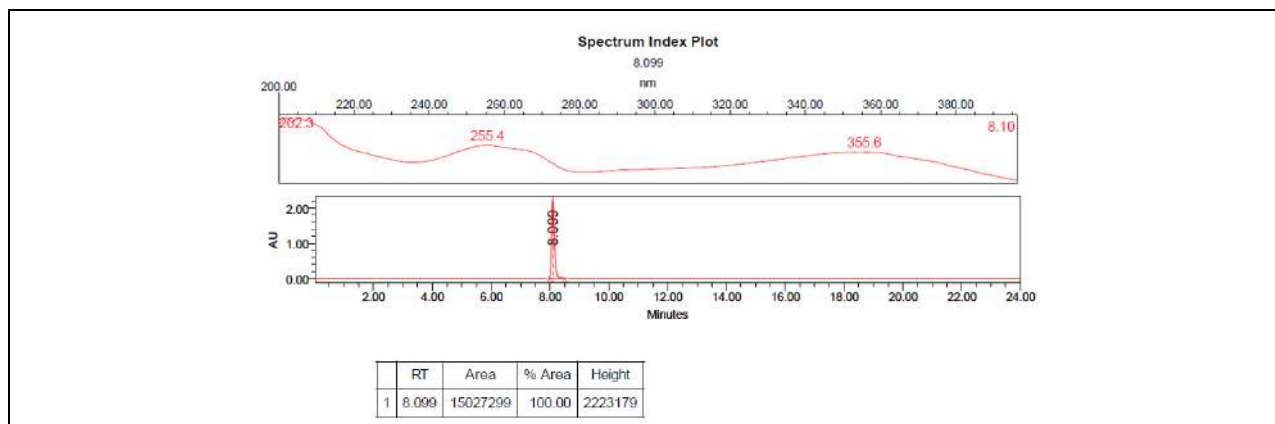


Fig 1: Preparative HPLC chromatogram of isolated pure compound (24 mg) from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn.

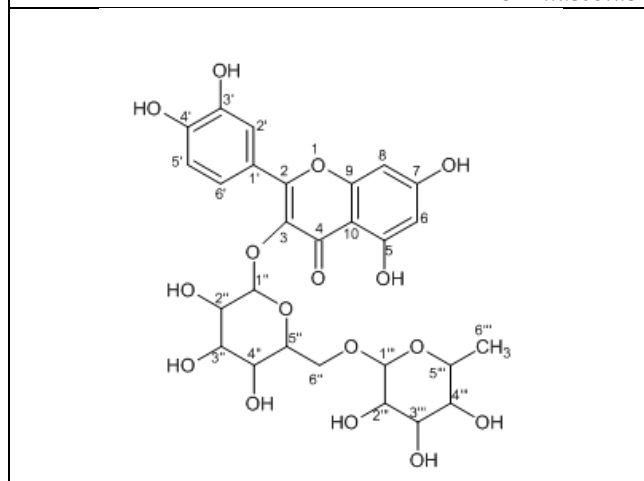


Fig:2 Structure of rutin (3, 3', 4', 5, 7-pentahydroxyflavone-3-rhamnoglucoside)[37]

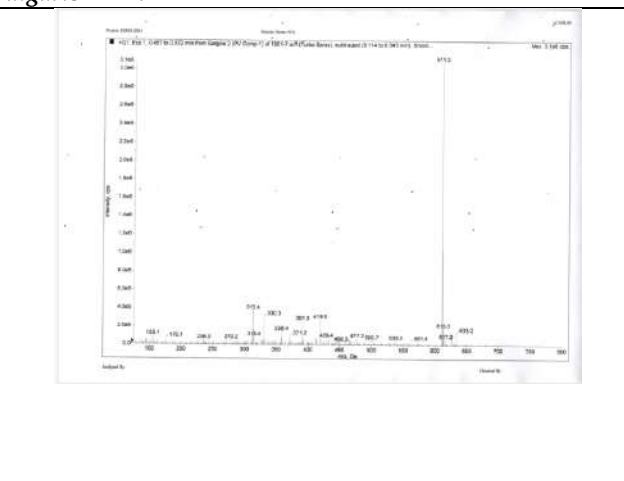


Figure 3: Mass spectra of a compound isolated from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn.





Madhavi Badole and Amrin Shaikh

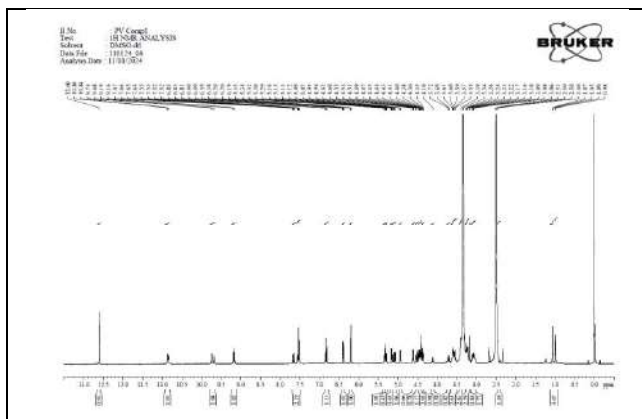


Fig 4:1H-NMR spectra of a compound isolated from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn.

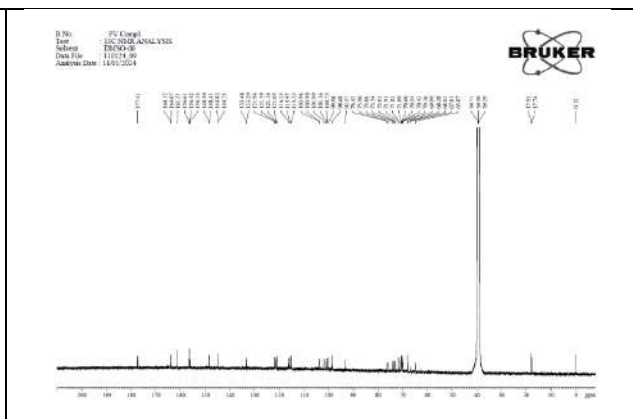


Fig 5 13C NMR spectra of the isolated compound from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn.

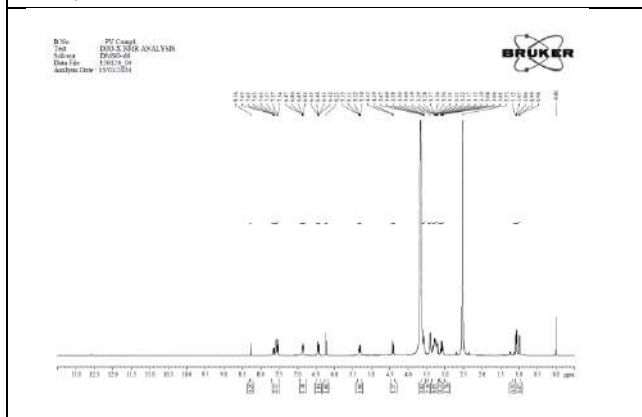


Fig 6.D2O NMR spectra of an isolated compound from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn.

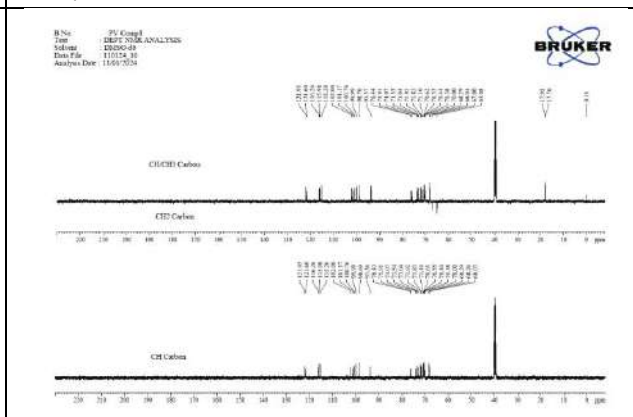


Fig 7:DEPT-135 NMR spectra of an isolated compound from the aqueous extract of the leaves of *Phaseolus vulgaris* Linn.





A Fractional Order Study of the Type 2 Diabetes Model using the Sumudu Transform Method

N. Jeeva^{1*}, K.M. Dharmalingam², P. Ugasini Preetha³ and Dumpa Sreepal⁴

¹Research Scholar, PG and Research Department of Mathematics, The Madura College, (Affiliated to Madurai Kamaraj University), Madurai, Tamil Nadu, India.

²Associate Professor, PG and Research Department of Mathematics, The Madura College, (Affiliated to Madurai Kamaraj University), Madurai, Tamil Nadu, India.

³Research Scholar, Department of Mathematics, College of Engineering and Technology, SRM Institute of Science and Technology (Deemed to be University), Kattankulathur, Tamil Nadu, India.

⁴Research Scholar, Department of Mathematics, Vishwa Vishwani Institute of Systems and Management, (Affiliated to Osmania University), Hyderabad, Telangana, India.

Received: 22 Jun 2024

Revised: 20 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

N. Jeeva

Research Scholar,

PG and Research Department of Mathematics,

The Madura College, (Affiliated to Madurai Kamaraj University),

Madurai, Tamil Nadu, India.

E.Mail: jeevapirc2405@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Elevated blood sugar levels serve as prominent indicators of type 2 diabetes, a chronic metabolic condition that poses significant health risks. Genetic predisposition, sedentary lifestyle, poor dietary choices, obesity, and advancing age are among the primary risk factors associated with the development of this disease. Unlike type 1 diabetes, which typically manifests during childhood, type 2 diabetes is characterized by insulin resistance and tends to appear later in life. This paper presents a pioneering mathematical framework for type 2 diabetes (T2D), expanding upon the established SATLP epidemic model. The proposed model categorizes the population into five groups: susceptible individuals ($S(t)$), affected individuals ($A(t)$), those under treatment ($T(t)$), adherents to a healthy lifestyle ($L(t)$), and those prevented from acquiring the condition ($P(t)$). In this extended model, we incorporate Caputo's fractional derivative to describe the dynamics of the system, resulting in a set of five fractional differential equations. To obtain approximate analytical solutions for this complex fractional system, we employ the Sumudu transform method (STM), which yields a series solution closely approximating the exact solution. The efficacy of the proposed model is validated through graphical analysis, demonstrating its simplicity and effectiveness in capturing the dynamics of type 2 diabetes. Tables and figures present the results of applying the Sumudu transform method to address the challenges posed by fractional





Jeeva et al.,

derivatives in the SATLP model. Through this innovative framework, our objective is to enhance understanding and facilitate the management of type 2 diabetes, thereby contributing to advancements in the field and offering potential solutions for tackling this widespread health issue.

Keywords: Type 2 diabetes (T2D), SATLP framework, Caputo's fractional derivative, Sumudutrans form method (STM), Mathematical modelling, Numerical simulation.

INTRODUCTION

Type 2 diabetes (T2D) is a chronic illness that affects the manner in which the body uses glucose, an essential energy source. In the ones with type 2 diabetes, the body either produces inadequate insulin to control blood sugar levels or responds to insulin less effectively [1]. Properly implementing lifestyle changes into practice has been shown to dramatically lower the risk of Type 2 Diabetes. These adjustments include eating a balanced diet that is high in fibre and low in fat, getting more exercise, and minimising sitting to maintain a healthy body weight [2-3]. Statistical modelling, which makes use of mathematical approaches to analyse and forecast the distribution and transmission of illnesses within an ecosystem, is the core epidemiological framework used for describing the spread of diseases in a population. Several mathematical models, such as those for COVID-19, Ebola, Zika and Lassa, have been developed to mathematically frame the virus models. These models aid in the understanding, prediction, and development of control and prevention efforts for the virus [4-7]. In the same way, several mathematical models were developed to assess and decrease the effects of diabetes on individuals [8-9].

A novel mathematical SATLP model is being developed to analyze the impact of a healthy lifestyle on the epidemiology of type 2 diabetes (T2D) by creating a comprehensive model and evaluating the effects of treatment rates and the endemic behavior of T2D [10]. In epidemiological settings, the severely nonlinear equations have been solved by a number of analytical techniques. In applied fields, a large number of classical differential equations have been solved with the use of analytical methods like the Homotopy perturbation method (HPM), Adomian decomposition method (ADM), and Akbari-Ganji's method (AGM). Additionally, Sumudu transform method (STM), Laplace Adomian decomposition method (LADM), and Homotopy Sumudu decomposition method (HSDM) have been used to solve fractional differential equations [11-14]. Since standard integer-order derivatives are unable to adequately depict a system's long-term behaviour, mathematicians look to the fractional derivative as a useful tool. Fractional derivatives offer a more thorough insight of the behaviour of the system by handling nonlinear and non-proportional influences [15]. Sumudu transformation is a mathematical technique that enables the solution of fractional differential equations by breaking them down into simpler, more manageable forms and it does not need any decomposition or perturbation applications. Each of these simplified equations can be addressed separately, making the problem-solving process more efficient and effective [16-19].

The main objective of this paper is to solve the fractional T2D SATLP model using the powerful analytical technique such as Sumudu transform method (STM). Section 2 provides the preliminary definitions of Caputo fractional derivative and STM. Through the application of the Caputo-derivative, the novel mathematical model for type 2 diabetes is converted from highly classical differential equations to fractional differential equations in Section 3. In section 4, we apply the STM to the strongly SATLP fractional model. We show the model's numerical simulation, tables, and graphs for controlling the disease in sections 4 and 5.

PRELIMINARIES

In this section, we define the basic definitions of Caputo-fractional derivative and Sumudu transform method.





Jeeva et al.,

Definition 1: [15]

For $\alpha > 0$, the Caputo-fractional derivative of order α is defined as follows ($m \geq 1$):

$$c_{D^\alpha} f(x) = J^{m-\alpha} D^m f(x) = \begin{cases} \frac{1}{\Gamma(m-\alpha)} \int_0^x (x-\varepsilon)^{m-\alpha-1} f^{(m)}(\varepsilon) d\varepsilon & m-1 < \alpha < m \\ \frac{d^m}{dx^m} f(x) & \alpha = m \end{cases}$$

Definition 2: [19]

If $F(u)$ is the Sumudu transform of the function $f(t)$, then the Sumudu transform of the Caputo derivative of $f(t)$ of order α is given by

$$S[c_{D^\alpha} f(t)] = u^{-\alpha} \left[F(u) - \sum_{k=0}^{n-1} u^k f^{(k)}(0) \right], n-1 < \alpha < n$$

Basic mathematical operations of Sumudu transform[16] is given below:

- 1) $S[1] = 1$
- 2) $S[t] = u$
- 3) $S[t^{n-1}] = u^{n-1} \Gamma(n), n \in R^+$

GOVERNING SYSTEM OF EQUATION

The T2D model [10] considers a population of individuals as all adults aged 20 – 79, which are then divided into five compartments. All adults are assumed to be potentially susceptible $S(t)$ and those in this category with healthy lifestyles are designated as $L(t)$. Adults who are affected are classified as $A(t)$, and once they start treatment, they are called $T(t)$. $P(t)$ is a category for those who prevented or recovered health from T2D.

Thus, the classical differential equations of the type 2 diabetes (T2D) model is given as:

$$\begin{aligned} \frac{dS}{dt} &= b - (\mu + \varepsilon + a)S \\ \frac{dA}{dt} &= \varepsilon S - (\tau + \mu + \delta_1)A + \gamma L \\ \frac{dT}{dt} &= \tau A - (\mu + \delta_2)T \\ \frac{dL}{dt} &= aS - (\gamma + \mu + \beta)L \\ \frac{dP}{dt} &= \beta L - \mu P \end{aligned} \tag{1}$$

Initial conditions:

$$S(0) = n_1, A(0) = n_2, T(0) = n_3, L(0) = n_4, P(0) = n_5 \tag{2}$$

where the parameter μ represents the natural mortality rate, while b represents the birth rate. The parameters ε and γ symbolize the diabetes rates from the susceptible and healthy lifestyle compartments, respectively. The parameter τ represents the treatment rate, while a denotes the rate of susceptible adults maintaining a healthy lifestyle. Finally, β represents the rate at which the healthy lifestyle population transitions to the prevented class. Adults can die from diabetes-related complications in the $A(t)$ and $T(t)$ classes at rates δ_1 and δ_2 , respectively.

We reformulate the classical differential equations (1) using the Caputo derivative to obtain a system of fractional ODEs as follows:

$$\begin{aligned} c_{D^{\alpha_1}} S &= b - (\mu + \varepsilon + a)S \\ c_{D^{\alpha_2}} A &= \varepsilon S - (\tau + \mu + \delta_1)A + \gamma L \\ c_{D^{\alpha_3}} T &= \tau A - (\mu + \delta_2)T \\ c_{D^{\alpha_4}} L &= aS - (\gamma + \mu + \beta)L \\ c_{D^{\alpha_5}} P &= \beta L - \mu P \end{aligned} \tag{3}$$

Where c_{D^α} is the Caputo's derivative of fractional order and α is the fractional time derivative





Jeeva et al.,

ANALYTICAL EXPRESSION OF TYPE 2 DIABETES MODEL USING SUMUDU TRANSFORM METHOD (STM)

In this section, we apply Sumudu transform method for the fractional type 2 diabetes(T2D) model. Taking Sumudu transform for the system (3) with corresponding initial conditions (2), we obtain:

$$\begin{aligned}
 S(t) &= n_1 + S^{-1}[u^{\alpha_1}S[b - (\mu + \varepsilon + a)S]] \\
 A(t) &= n_2 + S^{-1}[u^{\alpha_2}S[\varepsilon S - (\tau + \mu + \delta_1)A + \gamma L]] \\
 T(t) &= n_3 + S^{-1}[u^{\alpha_3}S[\tau A - (\mu + \delta_2)T]] \\
 L(t) &= n_4 + S^{-1}[u^{\alpha_4}S[aS - (\gamma + \mu + \beta)L]] \\
 P(t) &= n_5 + S^{-1}[u^{\alpha_5}S[\beta L - \mu P]]
 \end{aligned}
 \tag{4}$$

The general term of the T2D model is given in equation (5)

$$\begin{aligned}
 \sum_{n=0}^{\infty} S_{n+1}(t) &= n_1 + S^{-1}[u^{\alpha_1}S[b - (\mu + \varepsilon + a)\sum_{n=0}^{\infty} S_n]] \\
 \sum_{n=0}^{\infty} A_{n+1}(t) &= n_2 + S^{-1}[u^{\alpha_2}S[\varepsilon \sum_{n=0}^{\infty} S_n - (\tau + \mu + \delta_1)\sum_{n=0}^{\infty} A_n + \gamma \sum_{n=0}^{\infty} L_n]] \\
 \sum_{n=0}^{\infty} T_{n+1}(t) &= n_3 + S^{-1}[u^{\alpha_3}S[\tau \sum_{n=0}^{\infty} A_n - (\mu + \delta_2)\sum_{n=0}^{\infty} T_n]] \\
 \sum_{n=0}^{\infty} L_{n+1}(t) &= n_4 + S^{-1}[u^{\alpha_4}S[a \sum_{n=0}^{\infty} S_n - (\gamma + \mu + \beta)\sum_{n=0}^{\infty} L_n]] \\
 \sum_{n=0}^{\infty} P_{n+1}(t) &= n_5 + S^{-1}[u^{\alpha_5}S[\beta \sum_{n=0}^{\infty} L_n - \mu \sum_{n=0}^{\infty} P_n]]
 \end{aligned}
 \tag{5}$$

The solutions for $S, A, T, L,$ and P individuals can be expressed as infinite series, and since the initial conditions are given as: $S_0 = n_1, A_0 = n_2, T_0 = n_3, L_0 = n_4, P_0 = n_5$.

When $n = 0$, on comparing both side from the system of equation (5), we obtain:

$$\begin{aligned}
 S_1 &= S^{-1}[u^{\alpha_1}S[b - (\mu + \varepsilon + a)S_0]] = (b - (\mu + \varepsilon + a)n_1)\frac{t^{\alpha_1}}{\Gamma(\alpha_1+1)} \\
 A_1 &= S^{-1}[u^{\alpha_2}S[\varepsilon S_0 - (\tau + \mu + \delta_1)A_0 + \gamma L_0]] = (\varepsilon n_1 - (\tau + \mu + \delta_1)n_2 + \gamma n_4)\frac{t^{\alpha_2}}{\Gamma(\alpha_2 + 1)} \\
 T_1 &= S^{-1}[u^{\alpha_3}S[\tau A_0 - (\mu + \delta_2)T_0]] = (\tau n_2 - (\mu + \delta_2)n_3)\frac{t^{\alpha_3}}{\Gamma(\alpha_3+1)} \\
 L_1 &= S^{-1}[u^{\alpha_4}S[aS_0 - (\gamma + \mu + \beta)L_0]] = (an_1 - (\gamma + \mu + \beta)n_4)\frac{t^{\alpha_4}}{\Gamma(\alpha_4+1)} \\
 P_1 &= S^{-1}[u^{\alpha_5}S[\beta L_0 - \mu P_0]] = (\beta n_4 - \mu n_5)\frac{t^{\alpha_5}}{\Gamma(\alpha_5+1)}
 \end{aligned}
 \tag{6}$$

When $n = 1$, on comparing both side from the system of equation (5), we obtain:

$$\begin{aligned}
 S_2 &= S^{-1}[u^{\alpha_1}S[b - (\mu + \varepsilon + a)S_1]] = b\frac{t^{\alpha_1}}{\Gamma(\alpha_1 + 1)} - (\mu + \varepsilon + a)(b - (\mu + \varepsilon + a)n_1)\frac{t^{2\alpha_1}}{\Gamma(2\alpha_1 + 1)} \\
 A_2 &= S^{-1}[u^{\alpha_2}S[\varepsilon S_1 - (\tau + \mu + \delta_1)A_1 + \gamma L_1]] \\
 &= \varepsilon(b - (\mu + \varepsilon + a)n_1)\frac{t^{\alpha_1+\alpha_2}}{\Gamma(\alpha_1 + \alpha_2 + 1)} - (\tau + \mu + \delta_1)(\varepsilon n_1 - (\tau + \mu + \delta_1)n_2 + \gamma n_4)\frac{t^{2\alpha_2}}{\Gamma(2\alpha_2 + 1)} \\
 &\quad + \gamma(an_1 - (\gamma + \mu + \beta)n_4)\frac{t^{\alpha_4+\alpha_2}}{\Gamma(\alpha_4 + \alpha_2 + 1)} \\
 T_2 &= S^{-1}[u^{\alpha_3}S[\tau A_1 - (\mu + \delta_2)T_1]] = \tau(\varepsilon n_1 - (\tau + \mu + \delta_1)n_2 + \gamma n_4)\frac{t^{\alpha_3+\alpha_2}}{\Gamma(\alpha_3+\alpha_2+1)} - (\mu + \delta_2)(\tau n_2 - (\mu + \delta_2)n_3)\frac{t^{2\alpha_3}}{\Gamma(2\alpha_3+1)} \\
 L_2 &= S^{-1}[u^{\alpha_4}S[aS_1 - (\gamma + \mu + \beta)L_1]] = a(b - (\mu + \varepsilon + a)n_1)\frac{t^{\alpha_1+\alpha_4}}{\Gamma(\alpha_1+\alpha_4+1)} - (\gamma + \mu + \beta)(an_1 - (\gamma + \mu + \beta)n_4)\frac{t^{2\alpha_4}}{\Gamma(2\alpha_4+1)} \\
 P_2 &= S^{-1}[u^{\alpha_5}S[\beta L_1 - \mu P_1]] = \beta(an_1 - (\gamma + \mu + \beta)n_4)\frac{t^{\alpha_4+\alpha_5}}{\Gamma(\alpha_4+\alpha_5+1)} - \mu(\beta n_4 - \mu n_5)\frac{t^{2\alpha_5}}{\Gamma(2\alpha_5+1)}
 \end{aligned}
 \tag{7}$$





Jeeva et al.,

Proceeding like this, one can get

$$\begin{aligned} S(t) &= S_0 + S_1 + S_2 + \dots \\ A(t) &= A_0 + A_1 + A_2 + \dots \\ T(t) &= T_0 + T_1 + T_2 + \dots \\ L(t) &= L_0 + L_1 + L_2 + \dots \\ P(t) &= P_0 + P_1 + P_2 + \dots \end{aligned}$$

The approximate analytical expressions of fractional type 2 diabetes (T2D) model is obtained by adding the equations (2), (6) and (7):

$$\begin{aligned} S(t) &= n_1 + (b - (\mu + \varepsilon + a)n_1) \frac{t^{\alpha_1}}{\Gamma(\alpha_1 + 1)} + b \frac{t^{\alpha_1}}{\Gamma(\alpha_1 + 1)} - (\mu + \varepsilon + a)(b - (\mu + \varepsilon + a)n_1) \frac{t^{2\alpha_1}}{\Gamma(2\alpha_1 + 1)} \\ A(t) &= n_2 + (\varepsilon n_1 - (\tau + \mu + \delta_1)n_2 + \gamma n_4) \frac{t^{\alpha_2}}{\Gamma(\alpha_2 + 1)} + \varepsilon(b - (\mu + \varepsilon + a)n_1) \frac{t^{\alpha_1 + \alpha_2}}{\Gamma(\alpha_1 + \alpha_2 + 1)} - (\tau + \mu + \delta_1)(\varepsilon n_1 \\ &\quad - (\tau + \mu + \delta_1)n_2 + \gamma n_4) \frac{t^{2\alpha_2}}{\Gamma(2\alpha_2 + 1)} + \gamma(an_1 - (\gamma + \mu + \beta)n_4) \frac{t^{\alpha_4 + \alpha_2}}{\Gamma(\alpha_4 + \alpha_2 + 1)} \\ T(t) &= n_3 + (\tau n_2 - (\mu + \delta_2)n_3) \frac{t^{\alpha_3}}{\Gamma(\alpha_3 + 1)} + \tau(\varepsilon n_1 - (\tau + \mu + \delta_1)n_2 + \gamma n_4) \frac{t^{\alpha_3 + \alpha_2}}{\Gamma(\alpha_3 + \alpha_2 + 1)} - (\mu + \delta_2)(\tau n_2 - (\mu + \delta_2)n_3) \frac{t^{2\alpha_3}}{\Gamma(2\alpha_3 + 1)} \end{aligned} \tag{8}$$

$$\begin{aligned} L(t) &= n_4 + (an_1 - (\gamma + \mu + \beta)n_4) \frac{t^{\alpha_4}}{\Gamma(\alpha_4 + 1)} + a(b - (\mu + \varepsilon + a)n_1) \frac{t^{\alpha_1 + \alpha_4}}{\Gamma(\alpha_1 + \alpha_4 + 1)} - (\gamma + \mu + \beta)(an_1 \\ &\quad - (\gamma + \mu + \beta)n_4) \frac{t^{2\alpha_4}}{\Gamma(2\alpha_4 + 1)} \end{aligned}$$

$$P(t) = n_5 + (\beta n_4 - \mu n_5) \frac{t^{\alpha_5}}{\Gamma(\alpha_5 + 1)} + \beta(an_1 - (\gamma + \mu + \beta)n_4) \frac{t^{\alpha_4 + \alpha_5}}{\Gamma(\alpha_4 + \alpha_5 + 1)} - \mu(\beta n_4 - \mu n_5) \frac{t^{2\alpha_5}}{\Gamma(2\alpha_5 + 1)}$$

NUMERICAL SIMULATION AND DISCUSSION

In this section, the fractional model for type 2 diabetes was numerically solved in order to analyse and control the disease. For the type 2 diabetes model that is taken into consideration in the Caputo-sense, STM is successfully applied. To facilitate the simulations of S, A, T, L and P individuals, the parameter values provided in Table 1 and initial conditions $S(0) = 20, A(0) = 10, T(0) = 5, L(0) = 5, P(0) = 5$ are included in the process. The fractional SATLP type 2 diabetes model, illustrated in Figures 1, 2, 3, 4, and 5, exhibits an enhanced level of flexibility for a standard initial timeframe of one year and yields distinct outcomes from the different fractional order α responses within the model. By altering the values of $\alpha = 0.7, 0.8, 0.9, 1$, it is discovered that a better result is obtained. It provides a more effective way to reach the desired level for illness control. In Figure 1 and 2, susceptible class $S(t)$ and affected class $A(t)$ increases as the fractional values increase, but in Figure 3, treated class $T(t)$ decrease as the fractional values increase. Likewise, in Figure 4 and 5, healthy lifestyle class $L(t)$ and prevented class $P(t)$ also decreases as the fractional values increase.

As the fractional values decreases from $\alpha = 1$, the solution converges towards a steady state, which suggests that the solution will become more efficient with lower fractional values. Subsequently, to assess the accuracy of the solution, a comparison was conducted between the fourth-order Runge-kutta method (RK4) for classical derivative model (1) and the approximate analytical solutions acquired through the Sumudu transform method. The effectiveness of the Sumudu transform method is demonstrated through the convergence of the values to the exact solution in integer order for fixing the fractional time derivative $\alpha = 1$ in equation (8), along with the given parameters is derived in equations (9-13),





Jeeva et al.,

$$S(t) = 20 - 5.8740t + 1.155460500t^2 \quad (9)$$

$$A(t) = 10 - 3.0980t + 0.5519012000t^2 \quad (10)$$

$$T(t) = 5 + 5.5710t - 0.9191959000t^2 \quad (11)$$

$$L(t) = 5 + 2.1810t - 1.046223900t^2 \quad (12)$$

$$P(t) = 5 + 1.4310t + 0.3172761000t^2 \quad (13)$$

and its accuracy with RK4 are shown in Tables (2-5) for the different time intervals $t = 0.1, 0.5, 1$ in T2D model. Moreover, as depicted in Figure 6, it's evident that the initial peak of affected individuals $A(t)$ occurred within a two-year period. This surge was attributed to an increase in adults transitioning from the susceptible class ε , coupled with a slower treatment rate τ . However, this number subsequently declined as the treatment rate τ increased and the rate of adults becoming affected from the susceptible class ε decreased, as demonstrated in Figures 7 and 8. The primary strategy to mitigate the progression of T2D is to prioritize extensive treatment or to maintain a healthy lifestyle.

CONCLUSION

The primary focus of this paper revolves around obtaining the approximate analytical solution for the fractional model of type 2 diabetes (T2D) using the Sumudu transform method (STM). By varying the fractional time derivative α , we observe a range of outcomes and enhanced flexibility in this model, offering a greater degree of freedom compared to classical models. A thorough comparison is conducted between the analytical solution obtained and RK4. The findings indicate a satisfactory level of agreement for the specified parameter values, thus underscoring the efficacy of STM in handling fractional nonlinear equations. Visual representations and tables are employed to effectively illustrate the method's efficiency and to analyze the disease. Moreover, a healthy lifestyle and prompt therapy can delay or prevent the development of T2D, slow down its progression, and reduce its symptoms through appropriate control strategies and therapies.

REFERENCES

1. World Health Organization, Diabetes, <https://www.who.int/news-room/fact-sheets/detail/diabetes>, accessed December 31, 2022.
2. H. Farhadnejad, F. Teymoori, G. Asghari, E. Mokhtari, P. Mirmiran, F. Azizi, The higher adherence to a healthy lifestyle score is associated with a decreased risk of type 2 diabetes in Iranian adults, *BMC Endocrine Disorders* 22 (1) (2022) 42.
3. A. Ahmed, A. Tsiami, H.T. Khan, Effects of Dietary and lifestyle management on type 2 diabetes development among Ethnic minority adults living in the UK: a generational shift, *International Journal of Gastronomy and Food Science* 31 (2023), 100634.
4. E. A. Hernandez-Vargas and J. X. Velasco-Hernandez. In-host Mathematical Modelling of COVID-19 in Humans. *Annual Reviews in Control*, 50 (2020), 448–456.
5. O. I. Idisi and T.T. Yusuf. A Mathematical Model For Lassa Fever Transmission Dynamics With Impacts of Control Measures: Analysis And Simulation. *European Journal of Mathematics and Statistics*, 2(2) (2021), 19–28.
6. S. Rezapour, H. Mohammadi and A. Jajarmi. A new mathematical model for Zika virus transmission. *Advances in Difference Equations*, 2020(1).
7. A. Rachah, and D. F. M. Torres. Mathematical Modelling, Simulation, and Optimal Control of the 2014 Ebola Outbreak in West Africa. *Discrete Dynamics in Nature and Society* (2015), 1–9.
8. G. Magomedze, P. Nduru, C. P. Bhunu and S. Mushayabasa. Mathematical modelling of immune regulation of type 1 diabetes. *Biosystems*, 102(2–3), (2010) 88–98.
9. A. Mari, A. Tura, E. Grespan, and R. Bizzotto. Mathematical Modeling for the Physiological and Clinical Investigation of Glucose Homeostasis and Diabetes. *Frontiers in Physiology*, (2020) 11.
10. A. Ferdous. An ordinary differential equation model for assessing the impact of lifestyle intervention on type 2 diabetes epidemic. *Healthcare Analytics*, 4, 100271 (2023).





Jeeva et al.,

11. K. M. Dharmalingam and M. Veeramuni. Akbari-Ganji’s Method (AGM) for solving non-linear reaction - Diffusion equation in the electroactive polymer film. Journal of Electroanalytical Chemistry, 844 (2019), 1–5.
12. Z.J. Liu, M.Y. Adamu, E. Suleiman, J.H. He, Hybridization of homotopy perturbation method and Laplace transformation for the partial differential equations. Therm. Sci. 21, 1843–1846 (2017)
13. J. Biazar and R. Islam. Solution of wave equation by Adomian decomposition method and the restrictions of the method. Appl Math Comput 149:807–814 (2004).
14. P. Pue-on. Laplace Adomian decomposition method for solving Newell-Whitehead-Segel equation. Applied Mathematical Sciences, 7 (2013).
15. M. Caputo and M. Fabrizio, A new definition of fractional derivative without singular kernel, Progr. Fract. Differ. Appl. 1 (2015), 73–85.
16. G.K. Watugala, Sumudu transform: a new integral transform to solve differential equations and control engineering problems, Int. J. Math. Educ. Sci. Technol. 24 (1993) no. 1, 35–43.
17. E. K. Akgül, A. Akgül and R. T. Alqahtani. A New Application of the Sumudu Transform for the Falling Body Problem. Journal of Function Spaces, 2021, 1–8.
18. R. M. Jena and S. Chakraverty. Analytical solution of Bagley-Torvik equations using Sumudu transformation method. SN Applied Sciences, (2019), 1(3).
19. Q.D. Katatbeh and F.B.M. Belgacem. Applications of the Sumudu transform to fractional differential equations, Nonlinear Stud. 18 (2011), no. 1, 99–112

Table 1: Values of the parameters in type 2 diabetes(T2D) model

Parameters	Values
b	0.621
μ	0.0138
ε	0.142
τ	0.565
β	0.3
a	0.2
γ	0.05
δ_1	0.04
δ_2	0.002

Table 2: Comparison of Eq. (9) with the RK4 simulation for Susceptible class $S(t)$.

$\alpha = 1$		
t	STM	RK4
0.1	19.42415460	19.36191562
0.5	17.35186512	17.02496375
1	15.28146050	15.03482000

Table 3: Comparison of Eq. (10) with the RK4 simulation for Affected class $A(t)$.

$\alpha = 1$		
t	STM	RK4
0.1	9.695719012	9.695638062
0.5	8.588975300	8.579918812
1	7.453901200	7.38576100





Jeeva et al.,

Table 4: Comparison of Eq. (11) with the RK4 simulation for Treated class $T(t)$.

$\alpha = 1$		
t	STM	RK4
0.1	5.547908041	5.548020915
0.5	7.555701025	7.568671625
1	9.651804100	9.75044600

Table 5: Comparison of Eq. (12) with the RK4 simulation for Healthy lifestyle class $L(t)$.

$\alpha = 1$		
t	STM	RK4
0.1	5.207637761	5.207847479
0.5	5.828944025	5.852924375
1	6.134776100	6.31529000

Table 6: Comparison of Eq. (13) with the RK4 simulation for Prevented class $P(t)$.

$\alpha = 1$		
t	STM	RK4
0.1	5.146272761	5.14616002
0.5	5.794819025	5.78241250
1	6.748276100	6.65620000

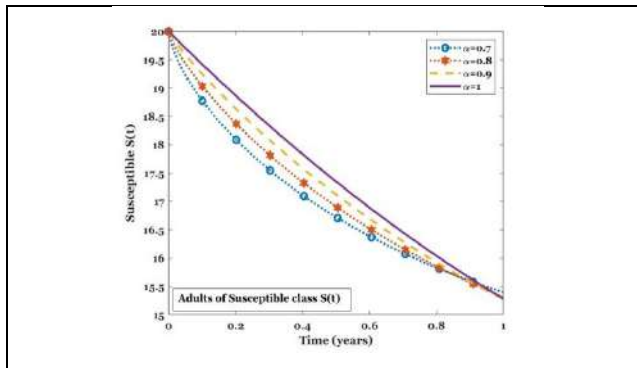


Figure 1: Plot shows the behaviour of Susceptible class $S(t)$ at different values of fractional order α in T2D model.

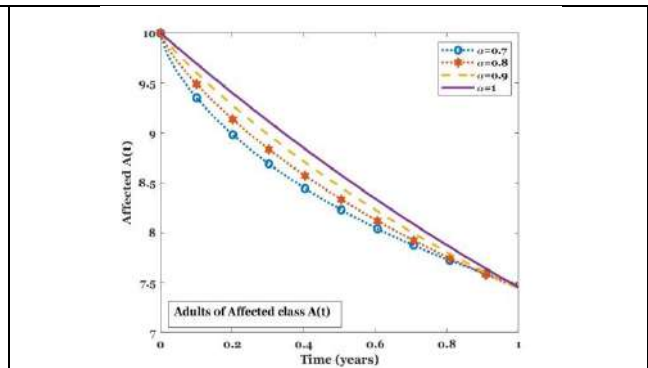


Figure 2: Plot shows the behaviour of Affected class $A(t)$ at different values of fractional order α in T2D model.

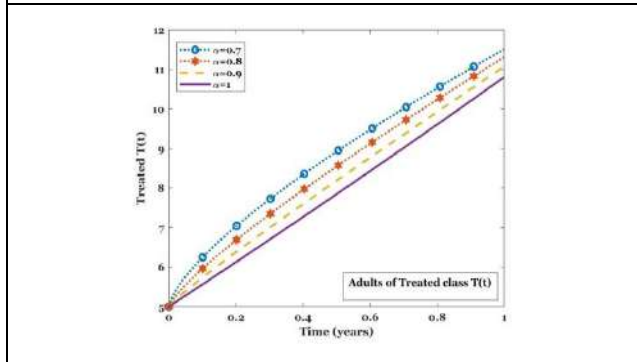


Figure 3: Plot shows the behaviour of Treated class $T(t)$ at different values of fractional order α in T2D model.

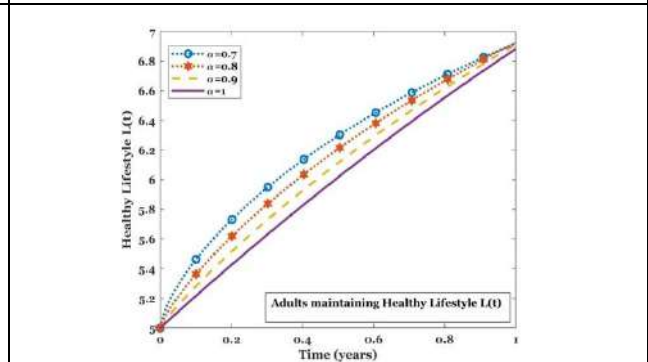


Figure 4: Plot shows the behaviour of Healthy Lifestyle class $L(t)$ at different values of fractional order α in T2D model.





Jeeva et al.,

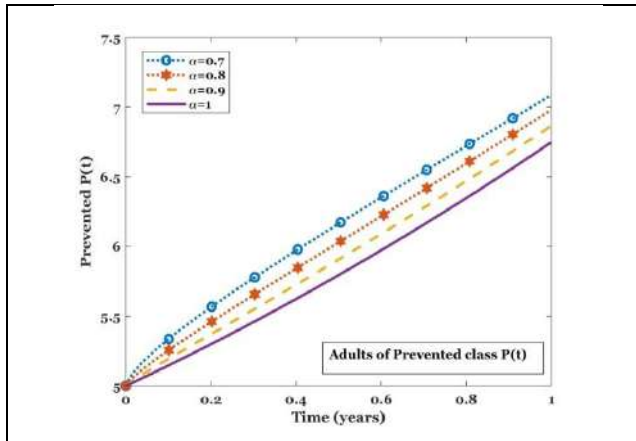


Figure 5: Plot shows the behaviour of Prevented class $P(t)$ at different values of fractional order α in T2D model.

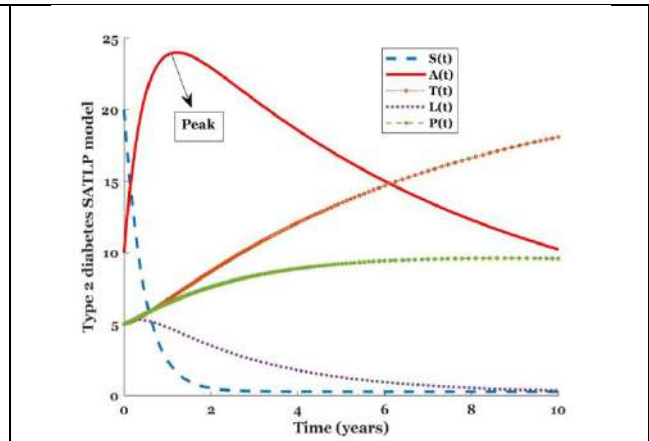


Figure 6: Affected class $A(t)$ reaches peak within a time frame of 2 years due to the effects of the parameters $\epsilon = 2, \tau = 0.09$ in the T2D model.

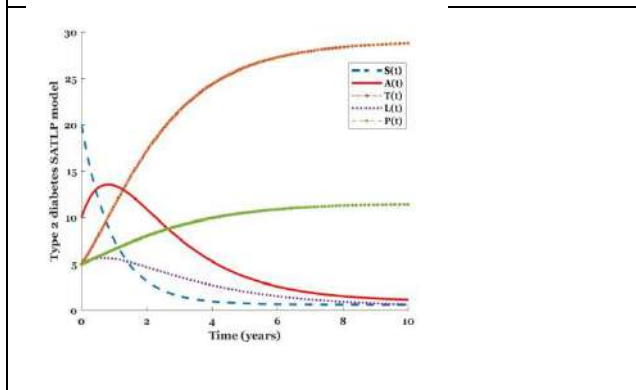


Figure 7: Treated class $T(t)$ and Prevented class $P(t)$ improves as the impact of the parameters $\epsilon = 1, \tau = 0.5$ in the T2D model.

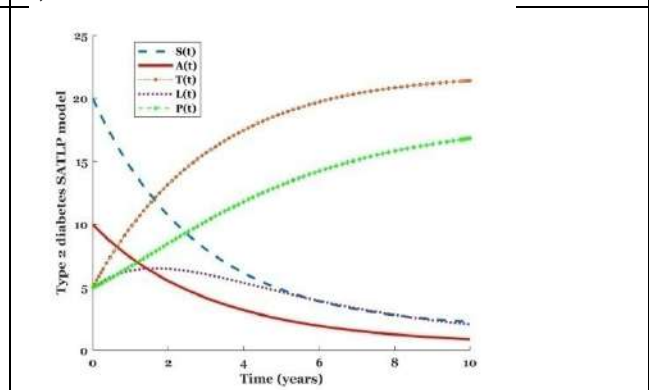


Figure 8: Visual representation of Treated class $T(t)$ and Prevented class $P(t)$ improving over time in the T2D model.





Tannins Analysis from *Rubus fruticosus* Root Extract using High Performance Thin Layer Chromatography for Antidiarrheal Activity

Mayuri K. Gaikwad¹, Rahul Chanshetty², Padmaja Kalshetty², Nilesh Kulkarni³, Jeevan Dhumal⁴ and Mohini Upadhye^{5*}

¹Research Student, Department of Pharmaceutical Quality Assurance, PES Modern College of Pharmacy (For Ladies) Moshi, (Affiliated to Savitribai Phule Pune University) Pune, Maharashtra, India.

²Assistant Professor, Department of Pharmacology, PES Modern College of Pharmacy (For Ladies) Moshi, (Affiliated to Savitribai Phule Pune University) Pune, Maharashtra, India.

³Associate Professor, Department of Pharmaceutics, PES Modern College of Pharmacy (For Ladies) Moshi, (Affiliated to Savitribai Phule Pune University) Pune, Maharashtra, India.

⁴Professor, Department of Pharmacognosy, Rajmata Jijau Shikshan Prasarak Mandal's College of Pharmacy, Dudulgaon, (Affiliated to Savitribai Phule Pune University) Pune, Maharashtra, India.

⁵HoD, Department of Pharmacognosy, PES Modern College of Pharmacy (For Ladies) Moshi, (Affiliated to Savitribai Phule Pune University) Pune, Maharashtra, India.

Received: 19 July 2024

Revised: 22 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

Mohini Upadhye

HoD, Department of Pharmacognosy,
PES Modern College of Pharmacy (For Ladies) Moshi,
(Affiliated to Savitribai Phule Pune University)
Pune, Maharashtra, India.

E.Mail: mohiniketh@rediffmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Rubus fruticosus widely used for its pharmacological properties. Considering to known data *R. fruticosus* has the potential to improve health through its antibacterial, antioxidant, neuropharmacological, and DNA damage protective properties. The purpose of the current investigation was to determine anti-diarrheal effect of *R. fruticosus* root extract. Phytoconstituents present in root analyzed using HPTLC for phytochemical identification and quantification. Tablets were formulated using the direct compression method. Castor oil induced diarrhea model mice were divided into normal control, disease control, marketed extract and formulation. Each group containing six swiss albino mice each. Tablets were evaluated for weight variation, friability, hardness, disintegration time, In vitro dissolution found to be in limit. Among formulation F6 batch shown good drug release as compared to other formulations. High Performance Thin Layer Chromatography (HPTLC) shown the presence of gallic acid 0.68 ng in extract and 0.43 ng in formulation. With the help of HPTLC the presence and quantification of tannin was identified and shown the promising antidiarrheal activity when compared with reference.

Keywords *R. fruticosus*, HPTLC, Tannins, Castor oil, Antidiarrheal.





Mayuri K. Gaikwad et al.,

INTRODUCTION

Medicinal plants are vital to human survival and the maintenance of good health because they contain bioactive phytochemicals. *Rubus fruticosus*, more recognisable as the blackberry, is a member of the Rosaceae family. In Hindi, it is also referred to as Kaala jamun. Plant leaves and roots are traditional home treatments for anaemia, irregular menstruation, diarrhoea, and dysentery (1). The roots, which have a long-standing antidiarrheal effect, include tannins and saponin as phytoconstituents (2). High-performance thin-layer chromatography (HPTLC) is a more advanced version of thin-layer chromatography (TLC) since it produces data with a high level of accuracy and precision (3). The current work was carried out to use the HPTLC technique to analyze the phytochemicals present in a *Rubus fruticosus* root extract and to evaluate antidiarrheal activity of extract and formulation.

Objectives

Analysis of phytoconstituents present in root extract using HPTLC.
Evaluation of antidiarrheal activity of extract and formulation.

MATERIALS AND METHODS

Plant

Rubus fruticosus root extract.

HPTLC

Chemicals and solvents :

Methanol , Distilled Water, Toluene , Ethyl acetate , Formic acid.

Working Standard

Pure Drug.

Preparation of Standard Solution

Weigh an accurately 10mg of Gallic Acid and transfer into 10 ml of volumetric flask. Dissolved in 5 ml methanol and sonicate for 10 min and made up volume up to 10 ml by using methanol to obtain 1000 ug/ml. From this stock solution inject 0.4ul.

Chromatographic Conditions

Chromatographic conditions are as follows:

Stationary phase: precoated silica gel HPTLC aluminium plates (10×10 cm, 0.2 mm thick). Mobile phase: Toluene : Ethyl acetate : Formic acid : Methanol (4 : 3 : 2 : 1).

Saturation time: 15 minutes.

Wavelength: 271 nm.

The HPTLC analysis was performed in an air conditioned room maintained at 22°C and 55% humidity using precoated silica gel aluminium backed plates (10 × 10 cm, 0.2 mm layer thickness. 0.4 μL of the standard solutions of Gallic acid were spotted using a Linomat autosampler fitted with a 100 μm Hamilton syringe and operated with settings of a band length. The plates were developed to a distance using toluene : ethyl acetate : formic acid: methanol (4: 3: 2 : 1 v/v) mobile phase in CAMAG twin trough chamber pre saturated with mobile phase. The developed plates were air dried and scanned with a CAMAG TLC Scanner equipped with winCATS planar chromatography software that was used for spectra recording, and data processing.





Mayuri K. Gaikwad *et al.*,

Standard Calibration Curve for gallic acid and linearity

From 1000ug/ml stock solution 100 to 600 ng/band was applied on HPTLC plate. The plate was developed and scanned as per the chromatographic condition mentioned. The peak areas were recorded. Calibration curve of Gallic acid was prepared by plotting peak area versus concentration.

Preparation of conventional tablets

Direct compression procedure: To achieve homogeneous mixing, all the formulation's constituents were sieved through sieve no.25 and mixed in a mortar pestle. The pre-compression properties of the mixed powder were then assessed. Additionally, this blended powder was compacted into tablets using a tablet compression machine and an 8mm flat punch. The parameters for post-compression evaluation for further punched tablets were assessed.

Antidiarrheal activity castor oil-induced diarrhea method

Castor Oil Induced Diarrhea Model

30 Swiss albino mice of either sex will deprive of food for 18 hours with free access to water and will divide randomly into five groups each containing 3 mice, as mentioned below. Group I as Normal Control Distilled water 10ml/kg (p.o) Group II as Disease Control Castor oil 0.5 ml of (p. o) . Group III as Marketed Loperamide 3mg/kg (p. o) . Group IV as Root Extract 400 mg/kg (p. o). Group V as formulation 200 mg/kg (p. o) .The animals were placed individually into cages in which the floor was lined with transparent paper for the collection of fecal matter. The transparent paper was changed every hour for a total of 4 hours. The mice was then removed from their cages and the weight of feces obtained by subtracting the weight of filter paper from the weight of feces and filter paper. The onset of diarrhea, the number of wet stools, the total number, and the total weight of fecal output noted. Finally, the percentage of diarrheal inhibition, as well as the percentage of the weight of total fecal output, calculated by using the following formula: percentage of diarrheal inhibition = mean number of wet stools (control group – treated group)/mean number of wet stools of the control group × 100 (4,5).

Grouping of Animals

Tablet Evaluation

Pre-compression study was evaluated for six batches Bulk density , Tapped density , Compressibility index ,

RESULT AND DISCUSSION

Characterization by HPTLC :Linearity of Standard Gallic Acid

Tablet Evaluation

Pre-compression study was evaluated for six batches Bulk density , Tapped density , Compressibility index , Hausner's, Ratio and ,Angle of repose (6). Post compression study was evaluated for Appearance , Hardness , Thickness ,Friability , Uniformity of weight, Disintegration , In vitro dissolution (7-11) . Hausner's Ratio and Carr's Index (%) shows excellent property. Angle of repose shows good property. All six formulations were found to have hardness values between 4 and 4.5 kg/cm². Friability was determined to be between 0.5% and 0.6%, while the thickness was found to be between 3.60 and 3.62 mm. There was a weight variation of 185.1 mg to 217.9 mg. Conventional tablets were found to disintegrate in less than 15 minutes. F6 has shown good drug release as compared to other batches.

Stool weight

Stool weight in Water treated group was found to be 86.88 ± 4.21 mg. Treatment with castor oil. significantly (P<0.001) increased the stool weight and the stool weight was found to be 189.66 ± 33.49 mg. Treatment with Loperamide, Extract and formulation significantly (P<0.001) reduced the stool weight as compared to the standard treatment and the stool weight was found to be 101.95 ± 2.21 mg, 134.73 ± 4.16 and 139.31 ± 3.37 mg respectively.





Mayuri K. Gaikwad et al.,

CONCLUSION

Characterization and Quantification of Gallic acid was performed using HPTLC. Tablets were prepared using different ratios of excipients. A total of 6 formulations were formulated and evaluated. The bulk density, tap density, angle of repose, Carr's index and Hausner ratio for the powder was determined and the results were found to be within limit for all formulations. Tablets were prepared by direct compression method. These formulations were evaluated for hardness, weight variation, friability, drug content, disintegration and in-vitro drug release. Formulation F6 found to be the good formulation. The model used to assess the anti-diarrheal activity include castor-oil induced diarrhea, appeared to act inhibition of diarrhea by reducing the absorption of water from the intestines that lead to the increase in defecation. This is due to the presence of tannins present in *R. fruticosus*. The findings in the present study confirm the anti-diarrheal activity of the root extract and formulation of *R. fruticosus* thus provide the scientific basis for the traditional use of this plant in the treatment of diarrhea and its effects.

ACKNOWLEDGMENT

Would like to thank you research guides for their kind support during studies.

REFERENCES

1. Gairola S, Sharma J, Singh BY, A Cross-Cultural Analysis Of Jammu, Kashmir And Ladakh (India) Medicinal Plant Use. Journal of Ethnopharmacology. September 2014; 155(2); 925-86. <https://doi.org/10.1016/j.jep.2014.06.029>.
2. Verma R, Gangrade T, Punasiya R, Chetan G, Rubus Fruticosus (Blackberry) Use As An Herbal Medicine. Pharmacognosy Reviews. June 2014;8(16);101-104. doi: 10.4103/0973-7847.134239.
3. Bhargava A, Shrivastava P, Tilwari, A. HPTLC analysis of Fumaria parviflora (Lam.) methanolic extract of whole plant. Futur Journal of Pharmaceutical Sciences. January 2021; 7(1);1-9. <http://dx.doi.org/10.1186/s43094-020-00150-x>
4. Zemene K, Birhanu K, Tesfaye T, Teshome B, Seyfe A, Evaluation Of In Vivo Antidiarrheal Activity of Solvent Fractions of Hagenia Abyssinica (Rosaceae) In Swiss Albino Mice. Hindawi Evidence-Based Complementary And Alternative Medicine. June 2021;155(2);925-86. <https://doi.org/10.1016/j.jep.2014.06.029>
5. Meziti A, Bouriche H, Meziti H, Kada S, Abderrahmane S, Ibrahim D, Antioxidant And Anti-Inflammatory Activities of Rubus Fruticosus And Zizyphus Vulgaris Methanol Extracts. International Journal of Pharmacy And Pharmaceutical Sciences. February 2016; 9(2);69-76. doi:10.22159/ijpps.2017v9i2.14374.
6. Wagh M, Yewale C, Zate S, Kothawade P, Mahale G, Formulation And Evaluation of Fast Dispersible Tablets of Aceclofenac Using Different Super disintegrant. International Journal of Pharmacy And Pharmaceutical Sciences. February 2010;2;154-157. https://www.researchgate.net/publication/236631315_Formulation_and_evaluation_of_fast_dispersible_tablets_of_aceclofenac_using_different_superdisintegrant.
7. Sivakranth M, Abdul A, Rajasekhar S, Formulation And Evaluation of Oral Fast Dissolving Tablets of Sildenafil Citrate. International Journal of Pharmacy And Pharmaceutical Sciences. March 2011; 3(2);112-121. https://www.researchgate.net/publication/281397876_Formulation_and_evaluation_of_oral_fast_dissolving_tablets_of_sildenafil_citrate.
8. Khinchi M, Gupta MK, Bhandari A, Sharma N, Agarwal D, Design And Development of Orally Disintegrating Tablets of Famotidine Prepared By Direct Compression Method Using Different Super disintegrants. Journal of Applied Pharmaceutical Science. March 2011; 1(1);50-58. https://japsonline.com/abstract.php?article_id=11&sts=2.
9. Gosai A, Patil S, Sawant K, Formulation And Evaluation of Oro Dispersible Tablets of Ondansetron Hydrochloride By Direct Compression Using Superdisintegrants. International Journal of Pharmaceutical Sciences And Nanotechnology. January 2008; 1(1);106-111





Mayuri K. Gaikwad et al.,

https://www.researchgate.net/publication/228350187_Formulation_and_Evaluation_of_Oro_Dispersible_Tablets_of_Ondansetron_Hydrochloride_by_Direct_Compression_using_Superdisintegrants.

10. Bushra R, Muhammad S, Nousheen A, Durriya H, Masud R, Formulation Development And Optimization of Ibuprofen Tablets By Direct Compression Method. Pakistan Journal of Pharmaceutical Sciences. April 2008; 21(2);113-120. <https://pubmed.ncbi.nlm.nih.gov/18390440/>.

11. Shaik R, Vani P, Gajavalli S, Hussain S, Venugopal B, Ravikiran T, Sridevi S, Yadav K, Formulation And Evaluation of Chewable Tablets of Mebendazole. International Journal of Pharma World Research. November 2014; 2(3);1-14.

12. https://www.researchgate.net/publication/329012891_FORMULATION_AND_EVALUATION_OF_CHEWABLE_TABLETS_OF_MEBENDAZOLE.

Table 1: Formulation table

Ingredients (mg)	F1	F2	F3	F4	F5	F6	Category
Extract	100	100	100	100	100	100	API
Lactose	80	80	80	80	80	80	Filler and Binder
Magnesium stearate	3	4	7	6	5	2	Lubricant
Sodium starch glycolate	6	5	2	3	4	7	Disintegrant
Talc	3	3	3	3	3	3	Glidant
Potato starch	8	8	8	8	8	8	Binder
Total weight	200	200	200	200	200	200	

Table 2: Castor oil induced diarrhea grouping

Group no.	Groups	Treatment	No.of Animals Swiss albinomice M/FWt. 25-30g
I	Normal Control	Distilledwater10ml/kg(p.o)	6
II	Disease Control	0.5ml of castor oil(p.o)	6
III	Marketed	Loperamide3mg/kg(p.o)	6
IV	Root Extract	400mg/kg(p.o)	6
V	Formulation	200mg/kg(p.o)	6
Total=30			

Table 3: Quantification of Gallic acid

Phytoconstituent	Extract	Formulation
Gallic acid	0.68 ng	0.43 ng

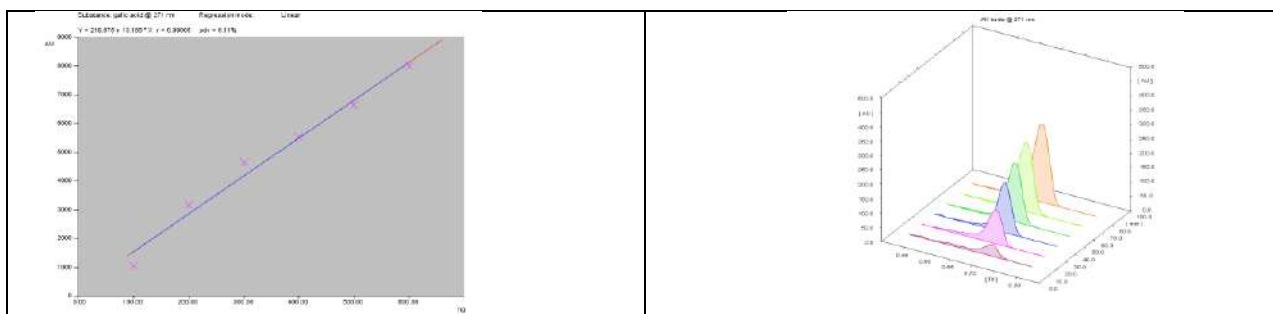


Fig No. 1 Linearity of Standard Gallic Acid





Mayuri K. Gaikwad et al.,

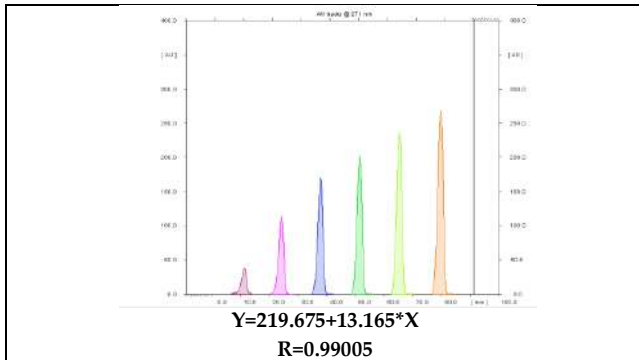


Fig No. 2 3d Standard Gallic Acid

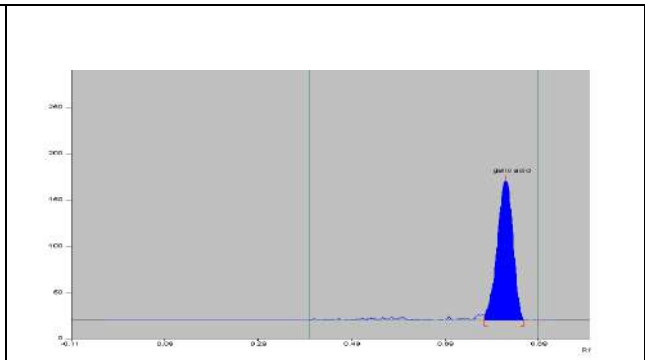


Fig No. 3 Standard Gallic Acid

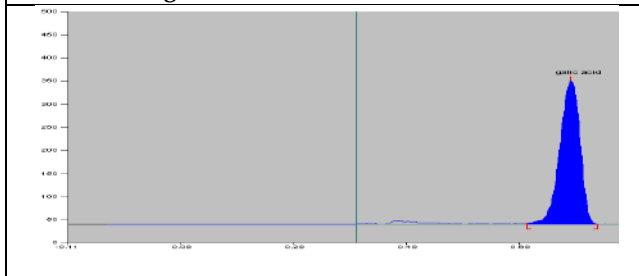


Fig No. 4 Gallic Acid in extract

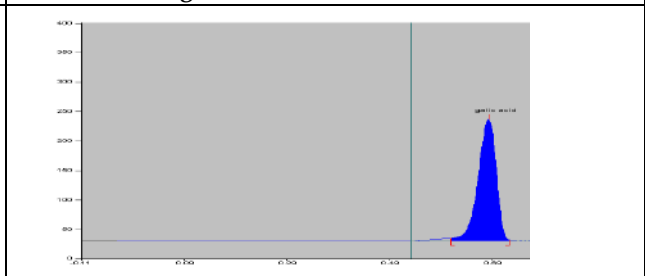


Fig No. 5 Gallic Acid in formulation

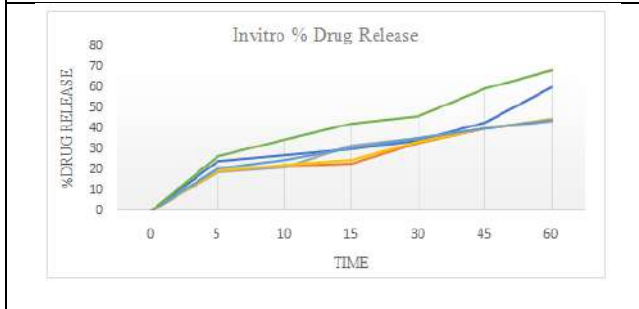


Fig No. 6 Invitro % drug release



Figure 7. Disease stool



Figure 8. Extract stool



Figure 9. Loperamide





Mayuri K. Gaikwad et al.,



Figure 10. Formulation

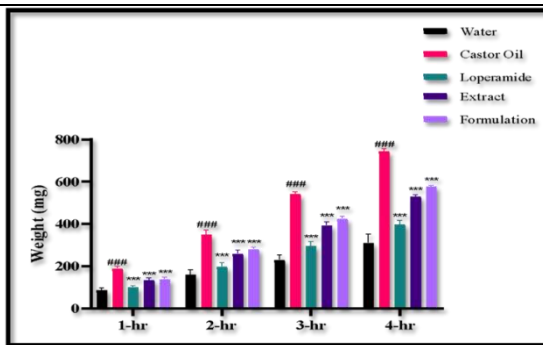


Figure 11. Effect of treatments on stool weight (mg). Data was analyzed by two way ANOVA followed by Bonferroni's multiple comparison test (n=6). ###p<0.001 as compared to control and *** p<0.001 as compared to standard.





Exploring Living and Working Conditions of Migrant Workers in Construction Industry in Karnataka - an Empirical Analysis

T. Nandhakumar^{1*} and P. Sakthivel²

¹Ph.D Research Scholar, Department of Political Science & Public Administration, Annamalai University, Annamalai Nagar, Tamil Nadu, India.

²Professor, Department of Political Science & Public Administration, Annamalai University, Annamalai Nagar, Tamil Nadu, India.

Received: 27 Aug 2024

Revised: 20 Sep 2024

Accepted: 25 Oct 2024

*Address for Correspondence

T. Nandhakumar

Ph.D Research Scholar,
Department of Political Science & Public Administration,
Annamalai University, Annamalai Nagar,
Tamil Nadu, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The paper analyzes the living and working conditions of migrant workers in Karnataka's construction industry, focusing on their socioeconomic realities and the factors influencing their migration. It aims to investigate the motivations behind these workers' decisions to seek employment in this sector, particularly examining the economic conditions in their places of origin, local job opportunities, and the potential for increased wages. The study evaluates the living conditions of these workers, highlighting critical aspects such as health, housing, and economic well-being. Many migrant workers face significant challenges, including inadequate housing, poor sanitation, and limited access to healthcare, which adversely affect their quality of life. The research explores existing policies aimed at improving the living and working conditions of migrant workers, assessing their effectiveness and identifying gaps in implementation. The study identifies challenges faced by migrant workers, including social isolation, exploitation, and lack of access to essential services. By employing a mixed-methods approach, the research gathers comprehensive data through surveys and interviews, providing a nuanced understanding of the migrant experience in Karnataka's construction sector. The analysis seeks to inform policymakers and stakeholders about the pressing needs and rights of migrant workers, advocating for improved conditions and support systems to enhance their livelihoods.

Keywords: Migrant Workers, Construction Industry, Challenges, Polici.

INTRODUCTION

Migrant workers are integral to the construction industry in Karnataka, significantly contributing to its growth and development. In 2021, the construction sector in Karnataka employed approximately 1.5 million migrant workers, highlighting their crucial role in meeting the demand for labor in urban areas. The study examines the factors driving



**Nandhakumar and Sakthivel**

these workers to seek employment in the construction sector, their living and working conditions, and the policies aimed at improving their circumstances. Economic factors are the primary drivers of migration to Karnataka's construction sector. Many workers are motivated by the prospect of higher wages compared to their home regions, where job opportunities are scarce. For instance, the average daily wage for construction workers in Karnataka ranges from 600 to 800, significantly higher than wages in rural areas of the state. The rapid urbanization and infrastructure development in cities like Bangalore, which has seen a population growth of over 46% from 2001 to 2011, have created a high demand for labor, attracting workers from various parts of Karnataka. For many, the construction sector represents a viable means to support their families and improve their living standards. However, the living and working conditions for migrant workers in Karnataka remain challenging. Many reside in temporary accommodations that lack basic amenities such as clean water, sanitation, and electricity. According to reports, around 70% of migrant workers live in substandard housing, which contributes to health issues, including respiratory problems and other diseases. Migrant workers significantly contribute to India's labor market, with Karnataka being a key destination for interstate migration. According to the Economic Survey 2016-17, Karnataka experienced a net in-migration, hosting approximately 32 lakh migrants from other states, 25 lakh migrants from Karnataka resided in other states. The survey highlights that, on average, around 9 million people migrate between states annually for work or education, with Karnataka's interstate migration rate at 37.82%, notably higher than the national average of 4.48%. Factors driving this migration include the pursuit of better economic opportunities, as many migrants seek to enhance their family income amidst limited job prospects in their native regions. The state's rapid urbanization and demand for labor, particularly in sectors like construction, further amplify this trend, underscoring the complex dynamics of labor migration in Karnataka (Economic Survey 2016-17).

The construction sites often pose safety risks, with workers facing long hours in hazardous environments for low pay. Data from the Karnataka State Building and Other Construction Workers' Welfare Board indicates that accidents on construction sites are a significant concern, with many workers reporting injuries due to unsafe practices. To address these issues, the national and Karnataka government has implemented several policies aimed at improving the living and working conditions of migrant workers. Initiatives include enhancing access to healthcare services and enforcing labor laws to protect workers' rights. The Karnataka Labour Welfare Board has also launched programs to provide social security benefits to construction workers. Despite these efforts, the effectiveness of such policies is often hampered by inadequate enforcement and the informal nature of many construction jobs. Migrant workers in Karnataka's construction sector face numerous challenges, including exploitation by employers, job insecurity, and limited access to social services. Many workers are unaware of their legal rights, which leaves them vulnerable to abuse and discrimination. The ongoing disparities in economic development within Karnataka further complicate their situation, as many continue to migrate in search of better opportunities enduring significant hardships. The construction sector in Karnataka offers employment opportunities for many migrant workers, significant issues regarding their living and working conditions persist. A concerted effort from both government and civil society is essential to ensure that the rights and well-being of these vital workers are prioritized and protected.

OBJECTIVES

1. To investigate the factors that prompted the migrant workers to seek employment in the construction sector in Karnataka, with a focus on economic conditions of migrant workers, job opportunities, and potential wage increases.
2. To evaluate the living and working conditions of migrant workers in Karnataka's construction sector, focusing on Health, Housing, and Economic being
3. To explore the policies promotes better living and working conditions of migrant workers in Karnataka.
4. To identify the key challenges and issues faced by migrant workers in Karnataka's construction sector.

HYPOTHESES

1. Economic hardship and limited job prospects in their hometowns are primary factors driving migrant workers to seek employment in Karnataka's construction sector.



**Nandhakumar and Sakhivel**

2. Poor living and working conditions among migrant workers in Karnataka's construction sector had profound impact on their health and economic well-being.
3. Lack of awareness among migrant workers in construction sector on labour rights and inadequate legal protections system paved way for economic exploitation and hardship.

SAMPLING AND SAMPLE SIZE

The study employed a combination of purposive sampling and simple random sampling to gather primary data from 66 respondents, for instance migrant workers from Dharmapuri District, chosen to ensure both accessibility and representativeness. The primary focus was on migrant workers currently employed in the construction sector in Karnataka, as this group is often overrepresented in the workforce. Purposive sampling was used to identify the respondents who were already employed in Karnataka's construction sector. A structured Interview Schedule has been utilized to facilitate consistent and comprehensive data collection. The interviews were conducted with respondents to gain insights into the employment practices and working conditions and other issues faced by these workers. The combination of these sampling methods and the structured interview approach aimed to produce a comprehensive and reliable outcome.

METHODOLOGY

The study adopted both document analysis and survey methods to collect both primary and secondary data, thereby utilizing qualitative and quantitative approaches. The dual approach allowed for quantitative data to facilitate the analysis of primary data through numbers and statistics, qualitative data provided richer context and deeper insights. Document analysis was chosen because it enables the examination of historical records, government documents, and other written sources relevant to the study. Document analysis complements the survey method by offering a broader perspective on the subject, enhancing the overall depth and reliability of the research findings. The survey method was employed to gather opinions and perspectives on the living and working conditions of migrant workers in the construction industry in Karnataka, ensuring a grounded and evidence-based analysis.

DATA ANALYSIS**DEMOGRAPHIC VARIABLES**

The study selected only three demographic variables: marital status (married and unmarried), gender (male and female), and occupation (organized and unorganized). This framework was utilized to explore the living and working conditions of migrant workers in Karnataka's construction sector, aiming to uncover how these factors impact their experiences and challenges.

The table shows the distribution of respondents marital status, gender, and occupation among the 66 participants in the study. Of these, 25 respondents (37.87%) are married, 41 respondents (62.12%) are unmarried, indicating that a majority of the migrant workers in the construction sector are unmarried, which highlights potential implications for their living and working conditions and underscores the need for targeted support services. In terms of gender, 32 respondents (48.48%) identify as male, while 34 respondents (51.50%) identify as female, reflecting a slightly higher representation of female migrant workers in the construction sector and suggesting the importance of addressing gender-specific challenges and needs within this workforce. Regarding occupation, 22 respondents (33.33%) are employed in organized sectors, whereas 44 respondents (66.66%) work in unorganized sectors. The significant majority in unorganized occupations emphasizes the precarious nature of employment for many migrant workers in the construction industry, highlighting the urgent need for improved labor protections and support for this vulnerable group.

TESTING OF HYPOTHESES AND INTERPRETATION

The t-test assessing the impact of economic hardship and limited job prospects in respondents' hometowns on migrant workers seeking employment in Karnataka's construction sector, categorized by marital status, reveals



**Nandhakumar and Sakthivel**

significant differences. Among the 66 participants, married respondents reported a mean score of 10.28 with a standard deviation of 2.29, while unmarried respondents had a mean score of 12.44 (SD=2.89). The t-value of 2.93, with a p-value of 0.05, indicates a statistically significant difference in perceptions of economic hardship and job prospects between married and unmarried workers. This finding leads to the rejection of the null hypothesis and acceptance of the alternate hypothesis, which suggests that marital status influences the perceptions of economic challenges driving migration. The results imply that unmarried workers may be more affected by economic conditions in their hometowns, potentially motivating them to seek employment in Karnataka's construction sector more urgently than their married counterparts. Unmarried migrant workers are more prevalent in Karnataka's construction sector due to socio-economic factors. Many young individuals migrate for better job opportunities amid economic hardship and limited prospects in their hometowns. Unmarried workers, often younger and less burdened by familial responsibilities, are more adaptable and willing to relocate for work. They seek independence and improved socio-economic status, which is often unattainable in their native areas. The construction sector's demand for labor, combined with fewer family commitments, makes it easier for unmarried individuals to accept jobs that involve long hours and challenging conditions, highlighting the need for targeted support policies.

The t-test evaluating the impact of poor living and working conditions on the health and economic well-being of migrant workers in Karnataka's construction sector, categorized by gender, reveals significant differences. Male respondents reported a mean score of 12.41 with a standard deviation of 2.22, while female respondents had a mean score of 18.09 (SD=4.06). The t-value of 3.19 and a p-value of 0.01 indicate a statistically significant difference in perceptions of economic hardship and health impacts between genders, leading to the rejection of the null hypothesis. This shows that female migrant workers may experience greater adverse effects from poor conditions due to factors such as increased vulnerability, higher exposure to health risks, and fewer support systems. Women often face discrimination and may have limited access to healthcare and social services, exacerbating the negative impacts of their working and living environments. This highlights the urgent need for targeted interventions to address the specific challenges faced by female migrant workers in the construction sector.

The t-test examining the lack of awareness among migrant workers in the construction sector regarding labor rights and inadequate legal protections, categorized by occupation, reveals significant differences. Workers in the organized sector reported a mean score of 12.46 with a standard deviation of 2.63, while those in the unorganized sector had a mean score of 18.20 (SD=3.37). The t-value of 2.59 and a p-value of 0.02 indicate a statistically significant difference, leading to the rejection of the null hypothesis. This suggests that workers in the unorganized sector experience greater economic exploitation and hardship due to their lack of awareness about labor rights. The higher mean score among unorganized workers can be attributed to their vulnerability and limited access to information regarding legal protections. Many of these workers are often illiterate or have minimal education, which hampers their ability to understand their rights and seek redress against exploitation. Occupations in the unorganized sector, such as those working in hotels, finance, tea shops, bakeries, as bakers or drivers, and as fruit vendors or water sellers (paani), typically lack formal employment contracts and benefits, further exacerbating their precarious situation. Unorganized sector workers generally lack the support systems and resources available to those in organized employment, making them more susceptible to unfair practices by employers. This shows the urgent need for targeted educational programs and legal support initiatives to empower these workers and ensure their rights are protected, ultimately improving their economic conditions and well-being.

RESULT AND DISCUSSION

FACTORS DRIVING MIGRANT WORKERS TO KARNATAKA'S CONSTRUCTION SECTOR

Migrant workers are increasingly drawn to Karnataka's construction sector due to a combination of push and pull factors. Push factors include economic distress, lack of job opportunities, and inadequate living conditions in their home regions, prompting individuals to seek better prospects elsewhere. In contrast, Karnataka offers attractive pull factors such as higher wages, abundant job opportunities, and better living standards. The state's rapid urbanization



**Nandhakumar and Sakthivel**

and infrastructure development create a high demand for labor, particularly in cities like Bangalore and Mysore, where construction projects are booming. The availability of social amenities, including access to healthcare and education for workers' families, enhances Karnataka's appeal. Many migrants are also motivated by the prospect of remittances, which can significantly improve their families' quality of life back home. Migrant workers constitute a significant portion of the construction workforce in Karnataka, particularly in urban centres like Bangalore, where rapid economic growth has created a high demand for labor. The analysis examines the factors prompting these workers to migrate for employment in the construction sector, focusing on the economic conditions in their places of origin, local job opportunities, and potential wage increases. Many migrant workers in Karnataka come from economically disadvantaged regions, primarily in rural areas where job opportunities are scarce. The Economic Survey of Karnataka (2016-17) highlighted that a substantial number of individuals migrate to urban areas in search of better livelihoods due to the lack of employment options in their home regions. In particular, many workers originate from areas with high poverty rates, where daily wage labor is often the only available option. A study conducted by Shamala B. (2021) indicates that the economic conditions in these regions are characterized by limited agricultural productivity and inadequate infrastructure, which force individuals to seek employment elsewhere. The construction sector in Karnataka, especially in Bangalore, offers a stark contrast to these conditions, providing a more stable income and better living standards. Karnataka's construction industry has experienced significant growth, largely driven by urbanization and infrastructure development. The state's Gross State Domestic Product (GSDP) grew by 8.2% in the fiscal year 2010-2011, with Bangalore emerging as a hub for various industries, including information technology and real estate. The growth has led to a surge in construction projects, including residential buildings, commercial complexes, and infrastructure projects such as roads and metro systems.

According to a report by SHRAM (2015), the construction sector in Bangalore alone relies on approximately 1.5 million migrant workers to meet its labor demands. The availability of jobs in construction is particularly appealing to those with limited skills and education, as it often requires less formal training compared to other sectors. The report emphasizes that the construction industry is one of the few viable employment options for these workers, making it an essential component of their migration decision. Wage differentials between Karnataka and the workers' home states serve as a significant pull factor for migration. The potential for higher earnings in Karnataka compared to their places of origin is a crucial motivator. For instance, daily wages for construction workers in rural areas may range from 200 to 300, wages in Karnataka can exceed 500 per day. This wage increase is particularly attractive for workers who aim to support their families back home through remittances. A study by Zabeer et al. (2019) found that the average daily wage for construction workers in Bangalore was significantly higher than in many other states, which incentivizes workers to migrate. The ability to earn more allows these workers to improve their living conditions and provide better educational opportunities for their children, further perpetuating the cycle of migration. Despite the economic incentives, migrant workers in Karnataka face numerous challenges. Many work in informal settings without job security or access to social benefits. According to a survey conducted by Anekal Jesuit Educational and Charitable Society (2022), a significant proportion of migrant workers reported poor living conditions, low wages, and lack of access to healthcare. The survey indicated that around 70% of households in the Anekal region, where many migrant workers reside, rely on daily wage labor, with erratic earnings that often do not meet basic needs.

The construction sector's reliance on subcontracting often leaves workers vulnerable to exploitation. The principal employers are not always held accountable for the welfare of their workers, as contractors and subcontractors may not comply with labor laws. The Inter-State Migrant Workmen (Regulation of Employment and Conditions of Service) Act (1979) is often inadequately enforced, leaving many workers without legal protections. The factors prompting migrant workers to seek employment in Karnataka's construction sector are multifaceted, encompassing economic conditions in their places of origin, the availability of local job opportunities, and the potential for wage increases. The construction industry offers a pathway to improved livelihoods, it is essential to address the challenges these workers face, including inadequate legal protections and poor living conditions. Policymakers must focus on creating robust frameworks that ensure the rights and welfare of migrant workers, facilitating their



**Nandhakumar and Sakthivel**

integration into the labor market safeguarding their dignity and well-being. The approach will not only benefit the workers but also contribute to the sustainable growth of Karnataka's economy.

MIGRANT CONSTRUCTION WORKERS IN KARNATAKA: HEALTH, HOUSING, AND ECONOMIC WELL BEING

Migrant workers are integral to Karnataka's construction sector, significantly contributing to the state's economic growth. However, their living and working conditions often starkly contrast the benefits they provide. Health issues among migrant construction workers in Karnataka are prevalent due to inadequate access to medical facilities and poor living conditions. A study by Zabeer et al. (2019) in Bangalore highlighted that many workers suffer from chronic health problems exacerbated by stressful working environments and limited healthcare access. The average age of these workers is around 26 years, with a significant portion being male (95.2%) and engaged in physically demanding labor. The study found that workers living in substandard housing conditions reported poorer health outcomes, including higher rates of respiratory and musculoskeletal disorders. A baseline survey conducted by the Centre for Integral Rural Welfare (CIRW) in 2022 indicated that many migrant workers are malnourished and lack basic health services, contributing to their declining health status. The survey revealed that 70% of households in Anekal, a significant area for migrant labor, consist of daily wage earners with erratic incomes, making it difficult for them to afford healthcare. Housing is a significant concern for migrant workers in Karnataka. Many live in makeshift shelters or overcrowded conditions that lack basic amenities such as clean water, sanitation, and electricity.

The Economic Survey of Karnataka (2016-17) noted that the state has a high rate of interstate migration, with many workers arriving from poorer regions in search of better opportunities. However, they often end up in informal settlements that do not meet minimum living standards. According to the study by Zabeer et al. (2019), the quality of housing directly impacts the workers quality of life. Those residing in huts reported a lower quality of life compared to those in Pucca (permanent) houses, indicating that housing quality is a critical determinant of overall well-being. The lack of proper sanitation facilities further exacerbates health risks, leading to a cycle of poverty and ill health.

Economically, migrant workers in Karnataka's construction sector face numerous challenges. The average daily wage for these workers ranges from 250 to 300, which is often insufficient to meet their basic needs. A significant portion of the workforce is engaged in low-paying, informal jobs, with limited job security and benefits. The 2011 Census revealed that Karnataka had approximately 32 lakh migrants, many of whom are employed in the construction industry, yet they remain vulnerable to exploitation and fluctuating market conditions. A socio-economic study by Shamala B. (2021) highlighted that 93% of migrant workers cited the need to increase family income as their primary reason for migrating. However, the economic benefits they anticipated often do not materialize due to systemic issues such as lack of job stability and inadequate wage structures. The Inter-state Migrant Workmen (Regulation of Employment and Conditions of Service) Act, 1979, intended to protect these workers, is often poorly implemented, leaving them without necessary legal protections. The living and working conditions of migrant workers in Karnataka's construction sector are marked by significant challenges in health, housing, and economic stability. Despite their critical role in the state's development, these workers often find themselves in precarious situations, facing health risks, inadequate housing, and economic instability. Addressing these issues requires comprehensive policy interventions that focus on improving living conditions, ensuring fair wages, and providing access to healthcare. Only through such measures can the state truly harness the potential of its migrant workforce safeguarding their rights and well-being.

POLICIES FOR IMPROVING CONDITIONS FOR MIGRANT WORKERS IN KARNATAKA

Migrant workers play a pivotal role in Karnataka's construction sector, significantly contributing to the state's economic development. To promote better living and working conditions for migrant workers in Karnataka, There are several main policies and acts have been established, addressing various aspects of their employment and welfare. The Building and Other Construction Workers (Regulation of Employment and Conditions of Service) Act, 1996 is a significant piece of legislation that regulates the employment conditions of construction workers, ensuring their rights to safe working environments, health and safety measures, and welfare benefits. This act came into force



**Nandhakumar and Sakthivel**

on March 1, 1996, and applies to establishments employing ten or more building workers, mandating provisions for their safety and welfare, as outlined in the India Code.

The Inter-State Migrant Workmen (Regulation of Employment and Conditions of Service) Act, 1979 provides a framework for the employment of inter-state migrant workers, guaranteeing their rights to safe working conditions, timely payment of wages, and access to welfare measures. This act is crucial in protecting the rights of migrant workers who often face exploitation and irregular employment. The Minimum Wages Act, 1948 establishes minimum wage standards for all workers, ensuring that migrant workers in the construction sector receive fair compensation for their labor. Despite this, many workers still report not receiving the minimum wage, highlighting a gap between legal provisions and actual practices. The Contract Labour (Regulation and Abolition) Act, 1970 regulates the employment of contract labourers, providing them with rights and protections against exploitation by contractors. This act is essential in ensuring that migrant workers, who often work under contract arrangements, are not subjected to unfair labor practices.

The Karnataka Labour Welfare Fund Act, 1965 creates a welfare fund for labourers, offering financial support for various needs, including healthcare, education, and housing. This act aims to improve the overall welfare of workers, including migrants, by providing them with essential services and support. The Maternity Benefit Act, 1961 ensures that female workers, including those in the construction sector, receive maternity benefits and job protection during pregnancy. This act is vital for supporting women workers and ensuring their rights during a critical time in their lives. The Child Labour (Prohibition and Regulation) Act, 1986 prohibits the employment of children in hazardous occupations, including construction, thereby protecting vulnerable populations from exploitation and ensuring that children have the opportunity to pursue education instead of labor. Lastly, the Equal Remuneration Act, 1976 mandates equal pay for equal work, ensuring that migrant workers receive fair wages regardless of gender. This act is crucial in addressing wage disparities and promoting gender equality in the workplace. These policies aim to create a comprehensive policy framework that addresses the multifaceted challenges faced by migrant workers in Karnataka, thereby improving their overall living and working conditions. By implementing these policies effectively, the state can better harness the potential of its migrant workforce safeguarding their rights and well-being.

CHALLENGES AND ISSUES FACED BY MIGRANT WORKERS IN KARNATAKA'S CONSTRUCTION SECTOR.

Migrant workers in Karnataka's construction sector face numerous challenges that significantly affect their quality of life. The construction industry in Karnataka, particularly in urban areas like Bangalore and Mysore, has seen a substantial influx of migrant labor due to rapid urbanization and industrial growth. These workers often come from poorer states, seeking better economic opportunities but encounter harsh realities upon arrival.

Living Conditions and Health Issues

One of the primary challenges faced by migrant workers is inadequate living conditions. Many reside in makeshift shelters, such as roadside tents or temporary sheds, which lack basic amenities like proper sanitation, clean water, and electricity. These living conditions contribute to a host of health problems, including malnutrition and communicable diseases. For instance, studies indicate that workers are frequently exposed to unsanitary environments, leading to illnesses exacerbated by poor housing conditions, such as cholera and respiratory issues from inhaling dust and fumes from construction materials.

Economic Exploitation and Job Insecurity

Economic exploitation is another critical issue. Migrant workers often receive low wages for long hours of labor, with many dependent on informal contracts that offer little job security. The lack of formal recognition and registration makes them vulnerable to exploitation by contractors and sub-contractors, who may withhold wages or impose unfair working conditions. A significant portion of these workers is illiterate or has only elementary education, limiting their ability to advocate for their rights or seek better employment opportunities.



**Nandhakumar and Sakthivel****Social Isolation and Lack of Access to Services**

Social isolation is prevalent among migrant workers, who often leave their families behind in their home states. This separation can lead to mental health issues and a lack of community support. Access to essential services such as healthcare, education for their children, and legal protections is severely limited. Many migrants are unaware of their rights and the welfare schemes available to them, which further exacerbates their vulnerability.

Impact of Political and Economic Factors

Political factors also play a role in the challenges faced by migrant workers. Recent events, such as the need for workers to travel back to their home states to vote, have disrupted construction activities, highlighting their precarious employment situation. The COVID-19 pandemic further intensified these challenges, with many workers losing their jobs during lockdowns and facing financial hardships without adequate government support. Migrant workers in Karnataka's construction sector confront a multitude of challenges, including poor living conditions, economic exploitation, social isolation, and limited access to essential services. Addressing these issues requires comprehensive policy interventions aimed at improving their living and working conditions, ensuring fair wages, and providing access to healthcare and education.

FINDINGS OF THE STUDY

1. Economic hardship and limited job opportunities in their hometowns are significant factors motivating migrant workers to seek employment in Karnataka's construction sector.
2. Female migrant workers experience greater adverse effects from poor living and working conditions than their male counterparts, highlighting the need for gender-specific interventions by government and non-governmental organisations, Civil Society organisations etc.
3. Deprived living and working conditions significantly impact the health and economic well-being of migrant workers, with female workers reporting higher levels of negative health outcomes.
4. Significant and lack of awareness among migrant workers regarding their labour rights, particularly in the unorganized sectors, which contributes to their economic exploitation.
5. Many migrant workers face social isolation, which exacerbates their vulnerability and limits their access to support networks and essential services.
6. Existing policies aimed at improving the living and working conditions of migrant workers are often inadequately enforced, leading to persistent challenges in their quality of life.
7. Migrant workers frequently encounter barriers to accessing healthcare services, which negatively affect their overall health and well-being.
8. There is an urgent need for targeted educational programmes and legal support initiatives to empower migrant workers, particularly those in the unorganized sector, to ensure their rights are protected and improve their economic conditions.
9. Economic hardship and limited job prospects in their hometowns drive migrant workers to seek employment in Karnataka's construction sector. The t-test revealed a significant difference based on marital status, with unmarried respondents reporting a higher mean score (12.44, SD = 2.89) compared to married respondents (10.28, SD = 2.29). The t-value was 2.93 and the p-value was 0.05, leading to the rejection of the null hypothesis and supporting the idea that unmarried workers are more motivated by economic conditions in their hometowns.
10. Poor living and working conditions significantly impact the health and economic well-being of migrant workers. The t-test results showed that female respondents had a higher mean score (18.09, SD = 4.06) than male respondents (12.41, SD = 2.22), with a t-value of 3.19 and a p-value of 0.01. This significant difference leads to the rejection of the null hypothesis, indicating that female migrant workers face greater adverse effects from poor conditions.
11. Lack of awareness regarding labour rights contributes to economic exploitation among migrant workers. The t-test indicated that unorganized sector workers had a higher mean score (18.20, SD = 3.37) compared to organized



**Nandhakumar and Sakthivel**

sector workers (12.46, SD = 2.63), with a t-value of 2.59 and a p-value of 0.02. This significant finding leads to the rejection of the null hypothesis, highlighting the need for educational initiatives to raise awareness of labor rights among unorganized sector workers.

SUGGESTIONS

1. Need to Develop and implemented targeted economic programmes in migrants' home regions to create job opportunities, reducing the need for migration and improving local livelihoods.
2. The Government is expected to implement gender-specific policies and programmes that address the unique challenges faced by female migrant workers, including health services and safety measures in the workplace.
3. Urgent action is needed to establish and enforce rigorous health and safety regulations within the construction sector, significantly reducing workplace injuries and promoting the well-being of migrant workers.
4. There is a critical need to launch widespread educational campaigns that empower migrant workers, particularly those in the unorganized sector, by raising awareness of their labor rights and equipping them to resist exploitation.
5. Expanding access to healthcare services for migrant workers is essential, including the deployment of mobile health clinics and collaborations with local healthcare providers to guarantee timely and necessary medical care.
6. Building strong community support networks is vital to facilitate the social integration of migrant workers, providing essential resources that alleviate isolation and enhance their quality of life.
7. Strengthening the enforcement of labor laws and regulations is crucial to safeguarding migrant workers' rights, with a focus on holding employers accountable for ensuring fair treatment and the well-being of their workforce.
8. Developing affordable, safe, and adequate housing solutions for migrant workers is essential, guaranteeing access to fundamental amenities such as clean water, sanitation, and reliable electricity.
9. Investing in skill development and vocational training programs tailored to the specific needs of migrant workers is key to enhancing their employability and enabling them to secure better-paying jobs.
10. Fostering strategic partnerships between government agencies, NGOs, and community organizations is necessary to build a comprehensive support system that effectively addresses the diverse needs of migrant workers.

CONCLUSION

Migrant workers play a crucial role in Karnataka's construction sector, driven by various economic and social factors. The primary reasons for their migration include poor economic conditions in their home states, limited local job opportunities, and the allure of higher wages in urban centres like Bangalore. Many come from economically disadvantaged regions, where poverty and lack of employment compel them to seek better prospects elsewhere. The construction industry, characterized by its demand for labor, becomes a viable option for these workers despite the inherent risks and challenges associated with such jobs. The living and working conditions of migrant workers in Karnataka are often substandard. Many reside in temporary shelters lacking basic amenities such as clean water, sanitation, and adequate ventilation. Health issues are prevalent due to exposure to hazardous materials and unsanitary living conditions, leading to a range of illnesses.

Furthermore, workers face economic instability, as their earnings are often insufficient to meet their basic needs, and they lack access to social security benefits and healthcare services. Policies aimed at improving the living and working conditions of migrant workers in Karnataka have been implemented, such as the establishment of the Karnataka Building and Other Construction Workers Welfare Board. This board facilitates access to various social protection schemes, including health services and financial assistance. NGOs like Sampark also play a vital role by raising awareness of workers' rights and providing support through resource centres. Despite these efforts, significant challenges remain. Migrant workers frequently encounter exploitation, unsafe working environments, and a lack of legal protections. Their transient status often leaves them vulnerable, with limited recourse to address





Nandhakumar and Sakthivel

grievances. Addressing these issues requires a multi-faceted approach that includes better enforcement of labor laws, improved living conditions, and enhanced access to healthcare and education, ensuring that the rights and well-being of migrant workers are prioritized in the construction sector.

REFERENCES

1. Amruthapuri, R. S. G. R. (2021). Partnership between diverse stakeholders: A potential solution to issues migrant construction workers face in Bengaluru, India (Doctoral dissertation, University of Pittsburgh).
2. Bellampalli, P. N., & Kaushik, R. (2020). Identification of the Determinants of Rural Workforce Migration: A Study of Construction Segments in Udupi District, Karnataka, India. *Review of Development and Change*, 25(2), 256-270.
3. Bellampalli, P. N., & Yadava, N. (2022). In search of better life: Construction migrants struggle for livelihood in urban Udupi District of Karnataka, India. *Asian Social Work and Policy Review*, 16(2), 175-184.
4. Britto, A. (2021). A Study on Socio-Economic Conditions of Migrant Workers of North Karnataka in Bangalore District. *SJCC Management Research Review*, 89-106.
5. Chowdhury, S. R., & Upadhyay, C. (2020). India's Changing Cityscapes: Work, Migration and Livelihoods. Institute for Social and Economic Change & National Institute of Advanced Studies.
6. Easwaramangalath, A. T., & Bhat, S. (2022). Review of Literature Related to Labour Migration: Types, Causes, and Impacts. *International Journal of Management, Technology, and Social Sciences (IJMTS)*, ISSN, 2581-6012.
7. Gurtoo, A. (2016). Workplace conditions and employer relationships as predictors of economic well-being: Female domestic workers in India. *Asian Social Work and Policy Review*, 10(1), 61-75.
8. Gurtoo, A. (2019). Occupational Prestige and Informal Work: Women Domestic Workers in India. *Workers and Margins: Grasping Erasures and Opportunities*, 193-213.
9. Hirudayaraj, M., Barhate, B., & McLean, G. N. (2023). Work conditions of interstate migrant workers in India: A critical realist exploration. *Human Resource Development Quarterly*.
10. Kapur, S., & Sethy, P. K. (2014). Working and living conditions of workers in unorganized sector-A review of literature. *Online Interdisc. Int. Interdisc. Res. J*, 6(2), 197-204.
11. Karnataka Migrant Employment Agency (KMEA). (2024). PAC KEA CWWB Study: Final report submitted. Retrieved from https://kmea.karnataka.gov.in/storage/pdf_files/Reports%20and%20other%20docs/PAC_KEA_CWWB%20Study_Final_Report_Submitted_EN.pdf
12. Kumar, A. (2020). The socio-economic status of unorganised migrant construction labourers in Karnataka: A case study in Bangalore. *Journal of South Asian Development*, 15(2), 123-145. <https://journals.sagepub.com/doi/10.1177/0972266120980187>
13. Manasi, S., Tantri, M. L., Kambara, C., Bairagya, I., & Vijayalakshmi, S. (2023). Challenges of Education among Informal Migrant Construction Workers' Children: A Study in Bengaluru. *Urban India*, 43(2), 53-71.
14. Nanjunda, D. C., & Venugopal, P. N. (2022). Hard and Invisible Bricks in the Wall: An Empirical Investigation on Gender, Caste, and Health Among Migrant Brick Workers in South India. *Journal of the Anthropological Survey of India*, 71(1), 84-104.
15. Pradeep, D., Adaina, K. C., & Kahmei, S. (2020). Well-being of North Eastern Migrant Workers in Bangalore. *Indian Journal of Human Development*, 14(1), 99-114.
16. Rajan, S. I. (2022). India migration report 2021: Migrants and health. Routledge India.
17. Reddy, B., & Olsen, W. (2012). Adaptation of the rural working class in India: A case study of migrant workers. In *Adaptation, Poverty and Development: The Dynamics of Subjective Well-Being* (pp. 181-214). London: Palgrave Macmillan UK.
18. Reja, M. S., & Das, B. (2019). Labour migration within India: motivations and social networks. *South Asia Research*, 39(2), 125-142.
19. Sahu, T. N., Maity, S., & Sen, N. (2023). Harsh realities of migrant workers during COVID-19 epoch: an investigation from the backward remote districts in India. *International Journal of Services, Economics and Management*, 14(3), 301-320.





Nandhakumar and Saktivel

20. Sampark. (2024). Empowering migrant construction workers. Sampark NGO. Retrieved from https://www.researchgate.net/publication/368558383_A_Study_on_Socio_Economic_Conditions_of_Migrant_Workers_of_North_Karnataka_in_Bangalore_District

21. Shamala, B. (2021). A study on migrant labour in Karnataka with special reference to construction sector. Bangalore University. Retrieved from https://kmea.karnataka.gov.in/storage/pdf_files/Reports%20and%20other%20docs/Migration%20of%20Labour.pdf

22. Singh, A., Vijila, Y., & Gael, K. A. (2022). A Conceptual Model of Adaptive Protective Factors for Migrant Workers. *SEDME (Small Enterprises Development, Management & Extension Journal)*, 49(4), 339-348.

23. Singh, G. P. (2016). Plights of migrant construction workers. *Management and Labour Studies*, 41(3), 181-198.

24. Ulvin, T. (2011). Social security for poverty reduction: a study of the MGNREGA in Karnataka, India (Master's thesis, Norwegian University of Life Sciences, Ås).

25. Zabeer, S., Inbaraj, L. R., George, C. E., & Norman, G. (2019). Quality of life among migrant construction workers in Bangalore city: A cross-sectional study. Bangalore Baptist Hospital. Retrieved from https://www.researchgate.net/publication/355851020_The_Push_and_Pull_Factors_of_Migration_-_A_Study_of_Migrant_Construction_Workers_in_Bangalore

26. Zabeer, S., Inbaraj, L. R., George, C. E., & Norman, G. (2019). Quality of life among migrant construction workers in Bangalore city: A cross-sectional study. *Journal of family medicine and primary care*, 8(2), 437-442.

27. Zeitlyn, B., Deshingkar, P., & Holtom, B. (2014). Internal and regional migration for construction work: a research agenda. *Migrating out of poverty RPC*.

Table 1. Demographic Variables of respondents

Demographic Variable	Category	No of respondents	Percentage
Marital Status	Married	25	37.87
	Unmarried	41	62.12
Gender	Male	32	48.48
	Female	34	51.50
Occupation	Organized	22	33.33
	Unorganized	44	66.66
Total respondents	66		100%

Source: Survey Data

Table 2: t- test of economic hardship and limited job prospects in their hometowns are primary factors driving migrant workers to seek employment in Karnataka's construction sector on the basis of marital status

Marital Status	Mean	S.D	t-value	P-value
Married	10.28	2.29	2.93	0.05
Unmarried	12.44	2.89		

Source: Primary data collected by the researcher

*Significant at 0.05 level

Table 3: t- test of Poor living and working conditions among migrant workers in Karnataka's construction sector had profound impact on their health and economic well-being on the basis of gender

Gender	Mean	S.D	t-value	P-value
Male	12.41	2.22	3.19	0.01 S
Female	18.09	4.06		

Source: Primary data collected by the researcher

* Level of Significance = 0.01 level





Nandhakumar and Sakthivel

Table 4: t- test of Lack of awareness among migrant workers in construction sector on labour rights and inadequate legal protections paved way for economic exploitation and hardship on the basis of occupation like organised and unorganised sector

Occupation	Mean	S.D	t-value	P-value
Organised	12.46	2.63	2.59	0.02 S
Unorganised	18.20	3.37		

Source: Primary data collected by the researcher

*Significant at 0.05 level





Enhanced Skin Cancer Detection through Hyperspectral Imaging and YOLO

Shubhendu Banerjee^{1*}, Pushpita Roy¹, Divyaansi Singh², Subhankar Sarkar² and Soubhagya Das¹

¹Assistant Professor, Department of Computer Science Engineering, Narula Institute of Technology (Affiliated to Maulana Abul Kalam Azad University of Technology), West Bengal, India.

²Student, Department of Computer Science Engineering, Narula Institute of Technology (Affiliated to Maulana Abul Kalam Azad University of Technology), West Bengal, India.

Received: 24 June 2024

Revised: 20 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

Shubhendu Banerjee

Assistant Professor, Department of Computer Science Engineering,
Narula Institute of Technology
(Affiliated to Maulana Abul Kalam Azad University of Technology),
West Bengal, India.

E.Mail: shankushubhendu@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Recent research has used several deep learning techniques to identify cases of skin cancer. Hyperspectral imaging (HSI) is a noninvasive optical technology that is worth investigating further since it may record wavelength information on the locations of skin cancer lesions. HSI technology improves the distinction of picture features by recording hundreds of visible and invisible narrow bands from the electromagnetic spectrum. In order to identify and categorise skin cancer forms such as seborrheic keratosis (SK), squamous cell carcinoma (SCC), and basal cell carcinoma (BCC), this research used the ISIC library dataset. The YOLO version 5 model was used for training after the dataset was divided into training and test sets. Five measures were used to assess the performance of the model: specificity, accuracy, sensitivity, precision and area under curve. A confusion matrix was also used. Hyperspectral narrowband image (HSI-NBI) and RGB categorization are two models that were created and tested against each other to see which is more effective: HSI or RGB. Because these traits were more pronounced in HSI, the experimental findings showed that the HSI model recognised SCC features more accurately than the RGB model. With an accuracy of over 89%, the suggested model proved to be useful in the identification of skin cancer. This excellent accuracy highlights the clinical application potential and dependability of the model.

Keywords: skin cancer; hyperspectral imaging; YOLO



**Shubhendu Banerjee et al.,**

INTRODUCTION

The most common and often diagnosed kind of cancer is nonmelanoma skin cancer (NMSC) [1]. Squamous cell carcinoma (SCC), seborrheic keratosis (SK), and basal cell carcinoma (BCC) are common types of skin cancer. The cells that make up BCC, the least aggressive NMSC, resemble epidermal basal cells [2]. Squamous cells, on the other hand, may spread abnormally and are invasive. Eighty percent of individuals with BCC, the most prevalent kind of skin cancer, generally have head and neck cancer that does not spread. The second most frequent kind of skin cancer, SCC, is characterised by aggressive tumours that are very invasive [3]. The need for quick detection techniques has grown as skin cancer cases rise. Treatment for skin cancer must begin as soon as possible after discovery. Although physicians often do traditional biopsy, it is a painful, difficult, and time-consuming procedure that involves taking a sample from a suspected cancerous location and analysing it medically [4]. Advances in artificial intelligence (AI) have resulted in the creation of many computer-aided detection (CAD) models for different types of cancer [5-7]. For instance, Haenssle et al. [8] used the InceptionV4 model to diagnose skin cancer and discovered that although it performed worse in specificity by 9%, it performed better in sensitivity, outperforming 58 dermatologists by 8%. Using the Asan training dataset, Han et al. [9] used ResNet-152 and obtained findings that were similar to those of sixteen dermatologists. Using a deep learning technique on 6000 photos, Fujisawa et al. [10] were able to identify benign and malignant cases with 76% accuracy. Even if CAD and biosensors provide affordable alternatives for early diagnosis, issues such as nanoparticles' environmental adaptation still exist. Red, green, and blue (RGB) imaging-based conventional CAD models have achieved saturation. Hyperspectral imaging (HSI), on the other hand, offers a viable substitute that could get beyond the drawbacks of conventional techniques.

Rather of restricting each pixel to one of the three basic colours, hyperspectral imaging (HSI) is a state-of-the-art method that analyses a wide variety of wavelengths [11-12]. Applications for High Sensitivity Imaging (HSI) include agriculture, cancer detection, military, air pollution detection, dentistry imaging, environmental monitoring, satellite photography, forestry monitoring, food security, natural resource surveying, vegetation observation, and geological mapping [13-17]. A number of research have also looked at the use of HSI to the diagnosis of skin cancer. Leon et al. [18] created a system to automatically recognise and categorise pigmented skin lesions (PSL) by combining supervised and unsupervised learning approaches. In order to determine the best spectral wavelengths for differentiating between normal and malignant skin, Courtenay et al. [19] developed a specialised platform that combined a visible-near infra-red (VNIR) hyperspectral imaging sensor. Courtenay et al. used HSI in a different investigation to identify spectral changes between specimens of normal and malignant cutaneous tissue. Nevertheless, existing techniques often depend on costly and intricate HSI imaging sensors or cameras [20].

This research suggests a quick skin cancer diagnosis technique that combines HSI technology with the YOLO version 5 model in order to overcome these difficulties. The five variables used to assess the performance of the created model were sensitivity (SEN), accuracy (ACC), specificity (SPEC), precision (PREC) and area under curve (AUC).

METHODS

Data Preprocessing

Three skin-related conditions are the subject of this study: seborrheic keratosis (SK), squamous cell carcinoma (SCC), and basal cell carcinoma (BCC). The dataset includes 168, 90, and 126 photos for the same categories in the validation set and 654, 336, and 480 images for BCC, SCC, and SK, respectively, in the training set. Total training and validation pictures utilised were 1470 and 384, respectively [5]. Figure 1 illustrates the division of the work into three key components: database development, picture preprocessing, and assessment of the YOLOv5 model training outcomes. This study made use of the ISIC dataset. Figures 2–4 show instances of seborrheic keratosis, basal cell carcinoma (BCC), and squamous cell carcinoma (SCC), in that order.



**Shubhendu Banerjee et al.,**

During preprocessing, 640×640 pixels were the new size for each picture. The photos were annotated using Labellmg software, which created XML files. After that, the files were converted to TXT format and used as training data for the YOLOv5 model. We used a spectrum conversion technique to convert the annotated picture files into hyperspectral narrow-band images (NBI) prior to training. As shown in Figure 5, the research first compared the normalised reflection spectrums of the lesion sites to analyse the three categories of skin cancer situations. We examined wavelength bands with notable variations in the intensity of reflected light. The 380–780 nm region of the spectrum was transformed into an HSI with a spectral resolution of 1 nm. Principal component analysis (PCA) was used to reduce the burden of this dimensional data's high processing and storage requirements. PCA reduced the dimensionality of the picture, extracted significant feature portions, and generated new component information by linearly transforming the data into a new space. The outcome was a projection of the data with decorrelated spectral information onto a low-dimensional space.

RGB and HSI photos were used to construct two datasets. Eighty percent of the data was put aside for testing and twenty percent was set aside for training. This separation came about after the amplification of images for both datasets. The Python 3.9.12 platform was used to write the programme for the PyTorch deep learning framework, which was applied to the Windows 10 operating system.

YOLOv5 Model

For real-time performance, this research specifically chose YOLOv5 because of its faster detection speed than other models, such as SSD and RetinaNet. The three essential components of YOLOv5 are the head terminals, neck, and backbone (Figure 6).

Backbone

Focus, CONV-BN-Leaky ReLU (CBL), cross stage partial (CSP), and spatial pyramid pooling (SPP) are some of the models that make up the convolutional neural network (CNN) architecture. By slicing the input picture, the focus model produces image features, collects many fine-grained images, boosts forward and back propagation speed, and uses less CUDA memory and fewer layers. The approach involves dividing an input picture of $640 \times 640 \times 3$ into four smaller images of $320 \times 320 \times 3$. These images are then combined and subjected to convolution layers to create an output image of $320 \times 320 \times 3$. This reduces the image size and speeds up training [21].

Neck

CBL, Upsample, CSP2_X, and more models make up the neck. It acts as a sequence of feature aggregation layers that creates feature pyramid (FPN) and path aggregation (PAN) networks by combining features from images. To improve image feature extraction and minimise model size, YOLOv5 adds the CSP2_X structure while keeping the CSP1_X structure from YOLOv4's CSPDarknet-53.

Head

The head maximises the bounding box forecasts' accuracy by using GIoU Loss as the loss function. For the suggested skin cancer classification work, YOLOv5 is the best option because of its structure, which guarantees effective and precise detection.

RESULTS AND DISCUSSION

After 300 rounds of training the loss function, the value curves of the training and validation sets were calculated, with a batch size of 16. Out of the 384 test photos, the RGB model properly predicted 333 images, whereas the HSI model correctly recognised 322 images. The effectiveness of the suggested technique was assessed using pre-established metrics such as Sensitivity (Sen), Accuracy (Acc), Specificity (Spe), Jaccard index value (Jac), and Dice coefficient score (Dic). The following formulae are employed:





Shubhendu Banerjee et al.,

$$\begin{aligned}
 SEN &= \frac{TP}{TP + FN} \\
 SPEC &= \frac{TN}{TN + FP} \\
 ACC &= \frac{TP + TN}{TP + TN + FN + FP} \\
 PREC &= \frac{TP}{TP + FP}
 \end{aligned}$$

Figure 7 and Figure 8 shows the confusion matrix of YOLOv5-based suggested model for detection, respectively. These data assist visualise the incidences of skin cancer that were properly and mistakenly predicted for both the RGB and HSI models, as well as the model's performance in identifying the different forms of skin cancer. Table 1, Table 2 and Table 3 shows the classifier comparison of BCC, SCC and SK datasets. The HSI models' accuracy greater than 89% demonstrated their resemblance. When compared to the original picture category SCC, the more noticeable SCC category in the HSI model shows a more notable improvement. This result showed how very comprehensive the model is when it comes to learning lesion characteristics. Because of the very limited number of photos utilised, it is evident that the accuracy rates of the RGB model and the HSI model are equal. All the same, this work demonstrates the possibility for accurate skin cancer detection and classification using HSI-based conversion algorithms that can transform RGB photos into HSI images. This study greatly increased the recall rate and specificity of the SCC category as well as the specificity (true negative rate) for the SK category, even if the majority of the findings were identical.

It is clear from the results and discussion section that our suggested approach—which combines hyperspectral imaging (HSI) with YOLOv5—performs better than other well-known classifiers in the identification of skin cancer. The thorough investigation revealed that while the HSI models' accuracy was significantly improved, especially for the SK category. The RGB model provided a better degree of completeness in learning lesion information, while the HSI-based conversion algorithms demonstrated efficacy in skin cancer detection and classification. This study opens the door for more precise and trustworthy diagnostic instruments by demonstrating the ability of conventional RGB imaging technology to improve the performance of skin cancer detection models. The enhanced specificity for certain categories confirm that the suggested approach is more effective than hyperspectral classifiers and may lead to improved diagnostic results.

CONCLUSIONS

YOLOv5 is mainly used to classify skin cancer pictures into three categories: SCC, BCC, and SK based on the CNN architecture. To generate a confusion matrix and determine the accuracy, specificity, sensitivity and precision values—which are used as classification indicators—lesion categories, such as RGB and hyperspectral datasets, are used. The outcomes of the experiment demonstrated that the SCC category in the HSI model is capable of picking up distinguishing traits. The original RGB picture is not as good at detection as the HSI classification model. YOLOv5 is easier to use than its predecessors since it has the ability to automatically scale the supplied picture to the desired size. At a quicker pace, the comparatively compact model structure attains an accuracy comparable to the preceding generation. The experiment's following designs may use the gain to increase accuracy in other studies.

REFERENCES

1. Didona, Dario, et al. "Non melanoma skin cancer pathogenesis overview." *Biomedicines* 6.1 (2018): 6.
2. Reichrath, Jörg, et al. "Epidemiology of skin cancer." *Sunlight, vitamin D and skin cancer* (2014): 120-140.
3. Apalla, Zoe, et al. "Skin cancer: epidemiology, disease burden, pathophysiology, diagnosis, and therapeutic approaches." *Dermatology and therapy* 7 (2017): 5-19.





Shubhendu Banerjee et al.,

4. Banerjee, Shubhendu, et al. "Melanoma diagnosis using deep learning and fuzzy logic." *Diagnostics* 10.8 (2020): 577.
5. Banerjee, Shubhendu, et al. "Diagnosis of Melanoma Lesion Using Neutrosophic and Deep Learning." *Traitement du Signal* 38.5 (2021).
6. Singh, Sumit Kumar, et al. "Classification of Melanoma Skin Cancer Using Inception-ResNet." *Frontiers of ICT in Healthcare: Proceedings of EAIT 2022*. Singapore: Springer Nature Singapore, 2023. 65-74.
7. Banerjee, Shubhendu, et al. "Diagnoses of Melanoma lesion using YOLOv3." *Computational Advancement in Communication, Circuits and Systems: Proceedings of 3rd ICCACCS 2020*. Springer Singapore, 2022.
8. Haenssle, H.A., et al. "Man Against Machine Reloaded: Performance of a Market-Approved Convolutional Neural Network in Classifying a Broad Spectrum of Skin Lesions in Comparison with 96 Dermatologists Working Under Less Artificial Conditions." *Annals of Oncology*, vol. 31, 2020, pp. 137–143.
9. Mukundan, A., et al. "Optical and Material Characteristics of MoS₂/Cu₂O Sensor for Detection of Lung Cancer Cell Types in Hydroplegia." *International Journal of Molecular Sciences*, vol. 23, 2022, p. 4745.
10. Mukundan, A., et al. "Growth Mechanism of Periodic-Structured MoS₂ by Transmission Electron Microscopy." *Nanomaterials*, vol. 12, 2022, p. 135.
11. Schneider, A., and H. Feussner. "Chapter 5—Diagnostic Procedures." *Biomedical Engineering in Gastrointestinal Surgery*, edited by A. Schneider and H. Feussner, Academic Press, Cambridge, MA, 2017, pp. 87–220.
12. Schelkanova, I., et al. "13—Early Optical Diagnosis of Pressure Ulcers." *Biophotonics for Medical Applications*, edited by I. Meglinski, Woodhead Publishing, Sawston, UK, 2015, pp. 347–375.
13. Lu, B., et al. "Recent Advances of Hyperspectral Imaging Technology and Applications in Agriculture." *Remote Sensing*, vol. 12, 2020, p. 2659.
14. Hsiao, Y.-P., et al. "Identification of Skin Lesions by Using Single-Step Multiframe Detector." *Journal of Clinical Medicine*, vol. 10, 2021, p. 144.
15. Hsiao, Y.-P., et al. "Identified Early Stage Mycosis Fungoides from Psoriasis and Atopic Dermatitis Using Non-Invasive Color Contrast Enhancement by LEDs Lighting." *Optical and Quantum Electronics*, vol. 47, 2015, pp. 1599–1611.
16. Hsiao, Y.-P.; Wang, H.-C.; Chen, S.H.; Tsai, C.H.; Yang, J.H. Optical perception for detection of cutaneous T-cell lymphoma by multi-spectral imaging. *J. Opt.* **2014**, *16*, 125301.
17. Tsai, T.-J.; Mukundan, A.; Chi, Y.-S.; Tsao, Y.-M.; Wang, Y.-K.; Chen, T.-H.; Wu, I.-C.; Huang, C.-W.; Wang, H.-C. Intelligent Identification of Early Esophageal Cancer by Band-Selective Hyperspectral Imaging. *Cancers* **2022**, *14*, 4292.
18. Leon, R.; Martinez-Vega, B.; Fabelo, H.; Ortega, S.; Melian, V.; Castaño, I.; Carretero, G.; Almeida, P.; Garcia, A.; Quevedo, E.; et al. Non-Invasive Skin Cancer Diagnosis Using Hyperspectral Imaging for In-Situ Clinical Support. *J. Clin. Med.* **2020**, *9*, 1662.
19. Courtenay, L.A.; González-Aguilera, D.; Lagüela, S.; del Pozo, S.; Ruiz-Mendez, C.; Barbero-García, I.; Román-Curto, C.; Cañueto, J.; Santos-Durán, C.; Cardeñoso-Álvarez, M.E.; et al. Hyperspectral imaging and robust statistics in non-melanoma skin cancer analysis. *Biomed. Opt. Express* **2021**, *12*, 5107–5127.
20. Dicker, D.T.; Lerner, J.; Van Belle, P.; Guerry, t.D.; Herlyn, M.; Elder, D.E.; El-Deiry, W.S. Differentiation of normal skin and melanoma using high resolution hyperspectral imaging. *Cancer Biol. Ther.* **2006**, *5*, 1033–1038.
21. Tan, Lu, et al. "Comparison of RetinaNet, SSD, and YOLO v3 for real-time pill identification." *BMC medical informatics and decision making* 21 (2021): 1-11.





Shubhendu Banerjee et al.,

Table 1: Classifier comparison for BCC

Classifier	Method	ACC	SEN	SPEC	PREC	AUC
YOLO v5	Proposed Method	86.72	90.53	83.72	81.38	87.13
TREE	CT	82.03	84.62	80.00	76.88	82.31
	ST	78.39	83.43	74.42	71.94	78.93
SVM	LSVM	81.25	82.25	80.47	76.80	81.36
	CSVM	82.29	81.66	82.79	78.86	82.22
	QSVM	80.73	82.25	79.53	75.96	80.89
	MGSVM	81.77	82.25	81.40	77.65	81.82
KNN	FKNN	82.55	84.02	81.40	78.02	82.71
	MKNN	81.25	83.43	79.53	76.22	81.48
	Cosine	81.25	84.02	79.07	75.94	81.55
	Cubic	79.69	81.07	78.60	74.86	79.83
	WKNN	77.86	79.29	76.74	72.83	78.02

Table 2: Classifier comparison for SCC

Classifier	Method	ACC	SEN	SPEC	PREC	AUC
YOLO v5	Proposed Method	88.80	88.89	88.78	70.80	88.83
TREE	CT	83.59	77.78	85.37	61.95	81.58
	ST	83.59	84.44	83.33	60.80	83.89
SVM	LSVM	84.90	82.22	85.71	63.79	83.97
	CSVM	85.68	83.33	86.39	65.22	84.86
	QSVM	84.64	82.22	85.37	63.25	83.80
	MGSVM	85.16	83.33	85.71	64.10	84.52
KNN	FKNN	85.94	82.22	87.07	66.07	84.65
	MKNN	80.73	76.67	81.97	56.56	79.32
	Cosine	79.69	67.78	83.33	55.45	75.56
	Cubic	77.86	64.44	81.97	52.25	73.21
	WKNN	78.91	65.56	82.99	54.13	74.27

Table 3: Classifier comparison for SK

Classifier	Method	ACC	SEN	SPEC	PREC	AUC
YOLO v5	Proposed Method	89.06	88.89	89.15	80.00	89.02
TREE	CT	84.64	79.37	87.21	75.19	83.29
	ST	84.64	78.57	87.60	75.57	83.08
SVM	LSVM	83.07	77.78	85.66	72.59	81.72
	CSVM	82.29	76.98	84.88	71.32	80.93
	QSVM	82.81	77.78	85.27	72.06	81.52
	MGSVM	81.25	74.60	84.50	70.15	79.55
KNN	FKNN	82.55	76.19	85.66	72.18	80.92
	MKNN	82.55	76.98	85.27	71.85	81.13
	Cosine	81.25	75.40	84.11	69.85	79.75
	Cubic	79.95	76.19	81.78	67.13	78.99
	WKNN	78.39	79.37	77.91	63.69	78.64





Shubhendu Banerjee et al.,

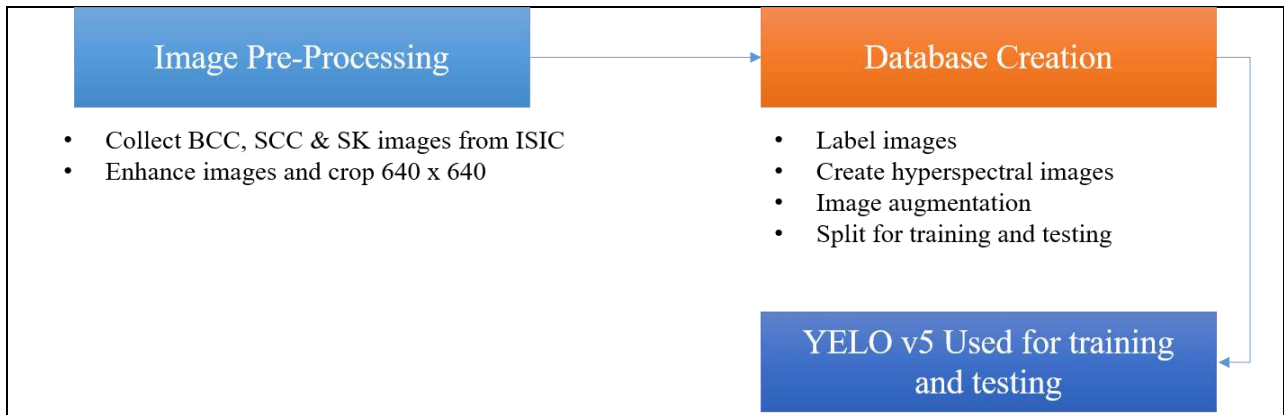


Figure 1: Overall flow chart for the experiment

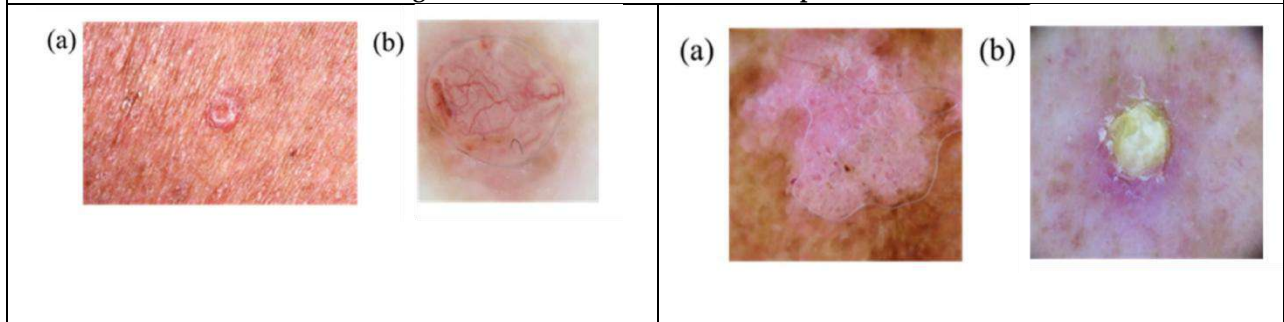


Figure 2: BCC a) glowing projections around the epidermis b) larger lesions that frequently bleed.

Figure 3: SCC a) red squamous plaques b) nodules

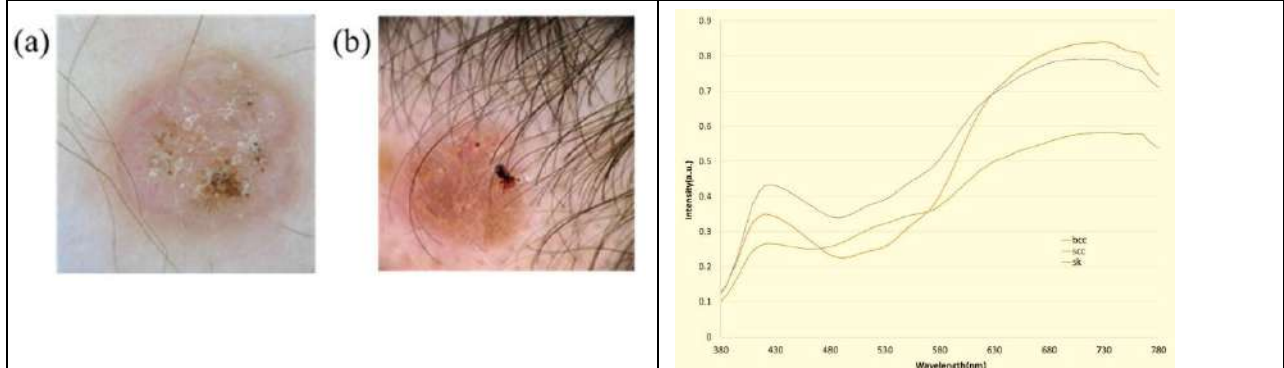


Figure 4: SK a) as a result of mutant epidermal keratinocyte hyperplasia b) keratinocyte mass in black colour

Figure 5: BCC, SCC, and SK normalised reflection spectra. BCC is represented by the orange line, SCC by the blue line, and SK by the green line.





Shubhendu Banerjee *et al.*,

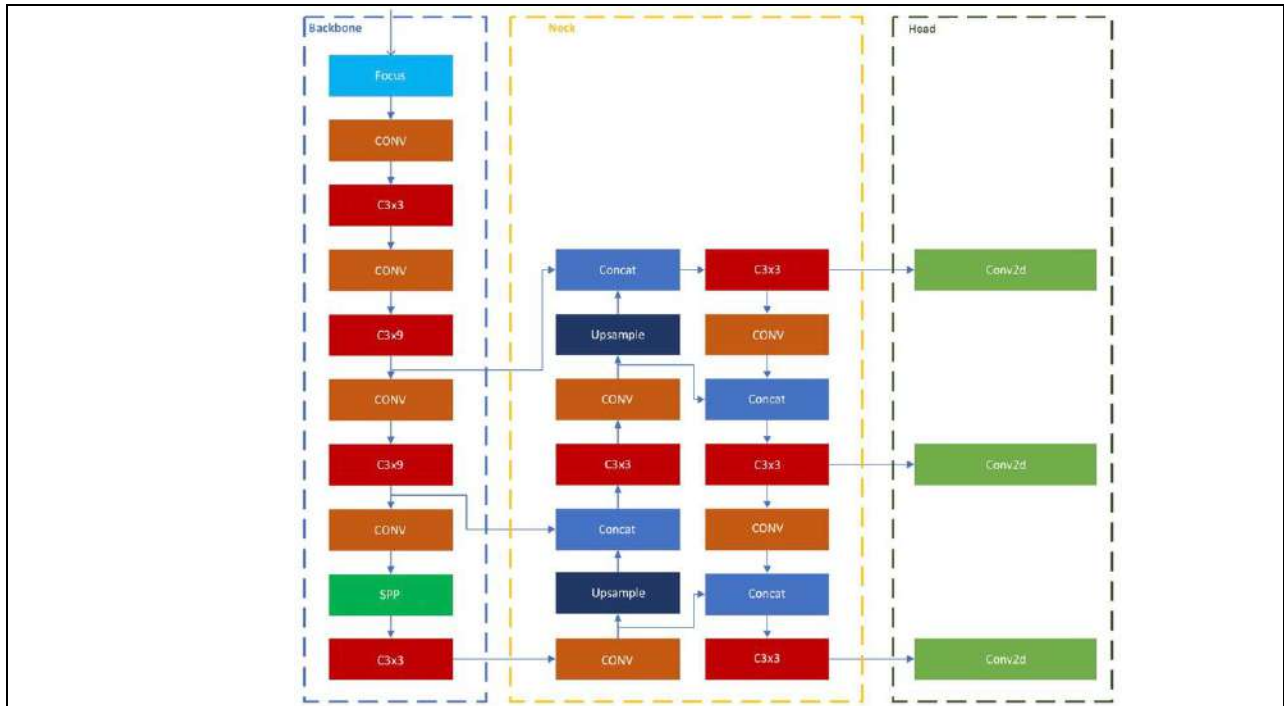


Figure 6: YOLOv5's network architecture.

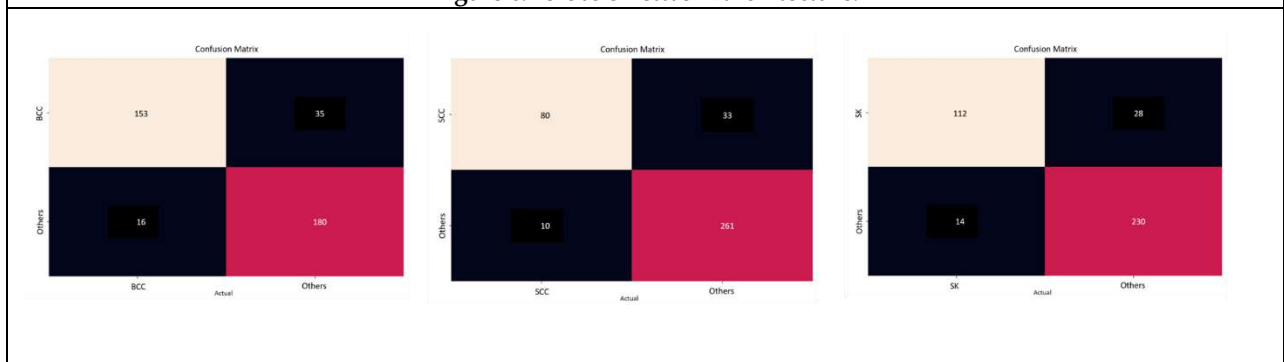


Figure 7: confusion matrix of YOLOv5 for RGB Model

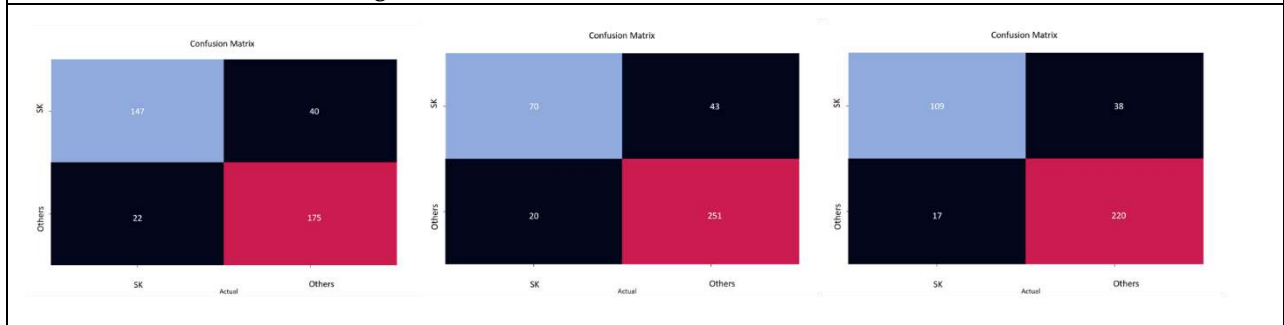


Figure 8: confusion matrix of YOLOv5 for HSI Model





Qualitative Analysis and Antipyretic Validation of Siddha Formulation Pancha Moola Kudineer Chooranam

S. Yasiga¹ and S. Visweswaran^{2*}

¹PG Scholar, Department of Gunapadam, National Institute of Siddha, Tambaram Sanatorium, (Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai) Tamil Nadu, India.

²Associate Professor, Department of Gunapadam, National Institute of Siddha, Tambaram Sanatorium, (Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai) Tamil Nadu, India.

Received: 23 Jun 2024

Revised: 20 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

S. Visweswaran

Associate Professor, Department of Gunapadam,
National Institute of Siddha, Tambaram Sanatorium,
(Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai)
Tamil Nadu, India.
E.Mail: svisu11@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Pancha Moola KudineerChooranam(PMKC) is a Siddha herbal formulation. This study is focussed on the qualitative analysis using HPTLC, FTIR and UV - Visible spectroscopy. Further the antipyretic activity was determined by protein denaturation assay for PMKC formulation. PMKC formulation was prepared as a decoction and it was analysed for its protein denaturation activity with Diclofenac sodium as standard. The decoction was determined for its phytoconstituents through HPTLC and FTIR analysis. Also, the UV- Vis spectroscopy was used to detect the interferenceof extractions solvent. PMKC at 500µg/ml concentration significantly reduced 36.61± 2.34 % of protein. HPTLC analysis revealed 8 prominent components. The UV -Vis spectroscopy obtained 254 nm, 274 nm and 361 nm as it's characteristic peaks.FTIR determined the functional groups present in the formulation.**Conclusion:** The qualitative analysis characterized the drug formulation of PMKC and determined to have 8 versatile components. The antipyretic activity was found to be higher at 500 µg/ml.

Keywords: Herbal formulation, PMKC, HPTLC, Antipyretic activity, FTIR.

INTRODUCTION

Siddha medicine predominantly practiced in southern part of India is a vast collection of literature written by various saints called *siddhars* in various period of time. These *siddhars* have explored about 4448 diseases including internal and external medication for those diseases. There are 32 types of internal medication written by the saints and almost



**Yasiga and Visweswaran**

all the parts of the plant from the roots to its secretions i.e. resins, gums, animals and its byproducts, metals and minerals have been used in the preparation of the siddha formulation after proper detoxification procedures. One such internal medicine preparation is called *kudineer* [1] (decoction) which is prepared by boiling dried coarse powder of raw drugs or fresh leaves with required amount of water until it reaches its required concentration. The often-used concentration of the decoction is one by eight or one by four of the water added. The shelf life of decoction is 3 hours. This study is aimed to scientifically prove the anti-pyretic activity of *Pancha moola kudineer Chooranam* [2]. It is one of the formulations mentioned in the Siddha Classical literature "*Therayarmahakarisaal*" written by the saint Therayar. He has explained that the decoction made from this siddha formulation can be used in the treatment of *suram* (fever) hence the anti-pyretic activity was carried out. The word *Pancha* means five and *Moolam* means root hence the formulation is comprised of five siddha herbal roots namely *Glycyrrhiza glabra*, *Phyllanthus emblica*, *Chukrasiatubularis*, *Terminalia chebula* and *Terminalia bellirica*.

According to siddha literature the term *suram* [3] is considered as increase in the body temperature and the main cause of fever is *seetham oraamam* in the gastro intestinal tract of human body, which means derangement in the normal physiological functions of the gastro intestinal tract caused by various factors like constipation, food poisoning, toxicity, increased physical activity, sleeplessness, excessive eating, having a heavy meal in excessive hunger, drinking cold water, etc. The signs and symptoms are lack of interest in food, heaviness of body, dyspepsia, loss of taste, giddiness, myalgia, chills, insomnia, xerostomia, dislike towards the sweet, sour and salt tastes, increased sweating, etc. There are 64 types of *suram* among which 52 are caused by natural factors and 12 types are caused by foreign factors.

MATERIALS AND METHODS

Pancha moola kudineerchooranam constituents and preparation

The raw drugs used in the preparation of the decoction was collected and authenticated by the botanist in National Institute of Siddha, Tambaram Sanatorium. The raw drugs were dried, detoxified and made into coarse powder. The coarse powder of PMKC was used in the preparation of decoction.

Protein denaturation assay [5] [6]

The protein denaturation assay of the sample PMKC was studied by albumin denaturation methodology. The assay was carried out by mixing 5% aqueous solution of bovine serum albumin with the sample drug PMKC in five concentrations ranging from 100 to 500 µg/ml. 100 µg/ml of diclofenac sodium was used as the standard drug. 1N hydrochloric acid was used to adjust the pH. The incubation was done for 20 minutes at 37 °C and heated for 3 minutes at 57 °C, phosphate buffer was used and the turbidity was measured using spectrophotometer at 660nm. Distilled water was used as control. The assessment of protection from denaturation was done using the below mentioned formula.

$$\text{Protein inhibition percentage: } \left[\frac{A(\text{control}) - A(\text{sample})}{A(\text{control})} \right] \times 100$$

High Performance Thin Layer Chromatography analysis [7].

Silica gel 60F254, 7X6 cm was used in the thin layer chromatography subjected in one dimensional ascending method for the sample PMKC. Micro pipette was used to spot the PMKC for TLC. 5 tracks of PMKC were placed with 10 micro litres at a distance of 1 cm. The sample PMKC was analysed at 254 nm and 365 nm of UV light, using twin trough chamber after it was dried. HPTLC is an innovative technique derived from TLC, since pre coated HPTLC graded plates are used, this helps in obtaining remarkable output in both qualitative and quantitative aspect. This method is very efficient in assessing the phytoconstituents of herbal raw drug in one step. The chromatographic fingerprinting is helpful in identifying the phytoconstituents and its purity.



**Yasiga and Visweswaran****Chromatogram Development**

CAMAG Twin Trough chambers was used in this HPTLC technique. According to the adsorption capability of herbal formulation, elution of the sample was carried out. After elution the plates are removed from chamber and dried.

Scanning

The Rf values were noted using the chromatographic fingerprinting of the phytochemicals. CAMAG software was used for the analysis of the data and the HPTLC plates were scanned at 366nm UV.

Fourier Transform Infra-Red spectrum analysis [8]

The sample was processed using Bruker Alpha-E by ATR module (attenuated total reflectance). It was amply positioned on the Crystal platform with perfect alignment of keeping anvil in upright position to assure the proper angle with the crystal prior to start of the IR radiation exposure. Spectra measurement was achieved with desired wavelength and the corresponding observational peaks/ waves were recorded. The wavenumber was subjected to further interpretation. Software used for the analysis is OPUS version 7 for functional group analysis. Signal detection processed through DTGS detector. Baseline correction adjusted as per the requirement.

UV – Visible spectrum analysis [9]

The sample *Pancha Moola Kudineer Chooranam* was analysed using UV visible Jasco V-750 model with the wavelength ranging of 200nm to 800nm.

Statistical analysis

Mean and standard deviation were used to express the results. The difference between experimental groups were analysed using ANOVA and Dunnett's test

RESULTS**Protein denaturation assay**

The result from this analysis shows the effective heat induced albumin denaturation by the sample drug PMKC. The maximum percentage protein inhibition found at a concentration of 500µg/ml was approximately 36.61±2.34% compared to diclofenac sodium, the standard antibiotic at 100µg/ml. The value is 91.46±1.33%. An increase in protein inhibition was observed with increasing PMKC concentration.

High Performance Thin Layer Chromatography Analysis

HPTLC fingerprinting was performed on the PMKC and eight major peaks were observed, confirming the presence of eight beneficial plants. The Rf values obtained varies between 0.08 and 0.67.

Fourier Transform Infra-Red spectrum analysis

FT-IR spectroscopy technique deciphered the structural information through vibration of their constituent atoms. The functional groups were identified through their corresponding wavelength peaks that were tabulated in table 3. The peak at 3424 cm⁻¹ correlate to hydroxyl stretching (-OH) and ammonia group (NH₂). The 2914 cm⁻¹ correlate to CH stretching, 2811 and 2231 cm⁻¹ is in accord to NH stretching, 2145 cm⁻¹ corresponds to CN and N=C=S stretching, 1539 cm⁻¹ corresponds to NH deformation, 1512 cm⁻¹ peak confirms the presence of amide group, 1426 corresponds to CH deformation, 1322 and 1016 cm⁻¹ to CO stretching. The peak at 940 cm⁻¹ is in accordance with aliphatic COOH, 832 cm⁻¹ denotes aromatic OH deformation, 514 corresponds to COC deformation, whereas 452 corresponds to aromatic C-OH in plane bending vibration.



**Yasiga and Visweswaran****UV – Visible spectrum analysis**

The absorption peak at 254.5 nm, 274 nm and 361 nm are the characteristic peak obtained for the given sample 'Pancha Moola KudineerChooranam'.

DISCUSSION

The temperature of human body is maintained by the thermoregulatory centre of the hypothalamus, any kind of inflammation in the body may lead to increase in the body temperature which is termed as fever so, the protein denaturation assay which is used to analyse the anti-inflammatory activity has been used as a tool to show that decrease in the inflammation in human body will lead to decrease in body temperature i.e., the probability to attain the normal body temperature is high^{[10][11]}. The decoction made of PMKC showed Maximum percentage inhibition at 500µg/ml so this siddha herbal formulation can be used as an anti-pyretic drug. The HPTLC analysis gives the insight of the phytoconstituents present in the siddha herbal formulation PMKC. From the results it is evident that there are 8 peaks visualised under 366nm. The Rf value varies from 0.08 to 0.67. the highest concentration of the phytoconstituent was found to be 29.60% corresponding to the Rf value 0.14 and the eight peaks represents eight phytoconstituents present in the PMKC which may have the therapeutic value. The FTIR analysis showed the presence of alkyl group, hydroxyl group, amide group, aliphatic carboxylic acid group, aromatic hydrocarbon group and various other chemical bonds present in the phytoconstituents of PMKC. The qualitative analysis of this herbal formulation was done using UV-Vis spectroscopy technique, that provides information about residual solvent escaped into the medicine formulation through extraction process. The characteristic peak obtained in this study revealed the absence of any such extraction component and the medicinal formulation is completely of higher standard. In conclusion, the research findings of the present study have qualitatively analysed the herbal medicine formulation *Pancha Moola KudineerChooranam*. The protein denaturation assay indicated the antipyretic activity of the medicine that could be effective for treatment of suram, additionally, further *in vivo* and *in vitro* research should be carried out to determine the complete therapeutic value of this siddha herbal formulation.

Conflict of interest

The authors declare no conflict of interest.

ACKNOWLEDGEMENT

The authors are thankful to the Sidha Central Research Institute, Arumbakkam for spectroscopy analysis. We thank the Noble research solution for HPTLC and FTIR analysis.

REFERENCES

1. Dr.R.Thiyagarajan L.I.M. GunapadamThaathu Jeeva Vaguppu part 2 and 3 published by Indian medicine and homeopathy department Chennai 600106; 2009; pp 58-59.
2. Dr.Anaivari.R.Anandan Ph.D., G.Perumal, M.A.,M.A.,M.A: Therayar Maha Karisal.Department of Indian Medicine and Homeopathy, Chennai 600106, 2009: 69-70.
3. K.N.Kuppusamymudaliyar HPIM. Siddha maruthuvamPodhu, published by Indian medicine and homeopathy department Chennai 600106; 2012; pp 1-10
4. K.M. Nadkarni (ed.) in *DR K.M. Nadkarni's Indian Materia Medica with Ayurvedic, Unani-tibbi, Siddha, Allopathic, Homeopathic, Naturopathic and home remedies* edited by A.K. Nadkarni, published by Popular Prakashan Private Limited Tardeo, Mumbai: 2005; 3rd edn; pp. 311, 480-486, 582-584, 1202-1205, 1205-1210.
5. G.Leelaprakash, S.Mohan Dass. In-vitro anti-inflammatory activity of methanol extract of *enicostemmaaxillare*. *Int. J. Drug Dev. & Res.*, 2011, 3 (3): 189-196.
6. M. V. Anoop, A. R. Bindu. In-vitro Anti-inflammatory Activity Studies on *Syzygiumzeylanicum* (L.) DC Leaves. *International Journal of Pharma Research & Review*, August 2015; 4(8):18-27.





Yasiga and Visweswaran

7. Wagner H. Plant Drug Analysis. A thin Layer chromatography Atlas.2nd ed. Heidelberg:Springer-Verlag Belgium; 2002:305, 227.
8. Bell, J., Nel, P. & Stuart, B. Non-invasive identification of polymers in cultural heritage collections: evaluation, optimisation and application of portable FTIR (ATR and external reflectance) spectroscopy to three-dimensional polymer-based objects. *Herit Sci* 7, 95 (2019). <https://doi.org/10.1186/s40494-019-0336-0>
9. Dinesh, Dharmalingam and Soosaimanickam, Maria Packiam and George, Mary, Evaluation of the Role of Tylophora Indica Leaf Extract Mediated Magnetite (Fe₃O₄) Microcubes for the Photodegradation of Carcinogenic Dye Rhodamine B (Rh B). Available at SSRN: <https://ssrn.com/abstract=4125229> or <http://dx.doi.org/10.2139/ssrn.4125229>
10. Osilla EV, Marsidi JL, Shumway KR, et al. Physiology, Temperature Regulation. [Updated 2023 Jul 30]. In: StatPearls [Internet]. Treasure Island (FL): StatPearlsPublishing; 2024 Jan-. Available from: <https://www.ncbi.nlm.nih.gov/books/NBK507838/>
11. Blomqvist, A., & Engblom, D. (2018). Neural Mechanisms of Inflammation-Induced Fever. *The Neuroscientist: a review journal bringing neurobiology, neurology and psychiatry*, 24(4), 381–399. <https://doi.org/10.1177/1073858418760481>

Table 1. The raw drugs used in PMKC are

Botanical name	Tamil name	Family	Chemical constituents ^[IV]
<i>Glycyrrhiza glabra</i>	<i>Adhimadhuram</i>	Fabaceae	<ul style="list-style-type: none"> • Glycyrrhizin, • Glycyrrhetic acid • Glabridin • Liquiritin • Triterpene • Saponin
<i>Chukrasiatubularis</i>	<i>Aayilpatta</i>	Meliaceae	<ul style="list-style-type: none"> • Sitosterol • Quercetin • 7-Dimethoxycoumarin • Scopoletin, • Cedrelone,
<i>Terminalia chebula</i>	<i>Kadukkai</i>	Combretaceae	<ul style="list-style-type: none"> • Gallic acid • Chebulagic acid • Punicalagin • Chebulanin • Corilagin • Neochebulinic acid • Ellagic acid • Chebulinic acid
<i>Terminalia bellirica</i>	<i>Thandrikkai</i>	Combretaceae	<ul style="list-style-type: none"> • Tannin • Pseudotannins • Gallic acid • Chebulic acid • Chebulagic acid • Ellagitannins • Corilagin • Ellagic acid
<i>Phyllanthus emblica</i>	<i>Nelliakai</i>	Euphorbiaceae	<ul style="list-style-type: none"> • Gallic acid • Ellagic acid • Chebulinic acid • Chebulagic acid





Yasiga and Visweswaran

			<ul style="list-style-type: none"> • Emblicanin • Citric acid
--	--	--	---

Table 2. Protein inhibition results of PMKC and standard drug

Concentration ($\mu\text{g/ml}$)	Protein denaturation inhibition in percentage
PMKC 100	9.141 \pm 5.11
PMKC 200	14.43 \pm 3.70
PMKC 300	20.81 \pm 1.90
PMKC 400	30.23 \pm 3.69
PMKC 500	36.61 \pm 2.34
Diclofenac sodium(100 μg)	91.46 \pm 1.33

Table 3. HPTLC Peak table

Peak	Start Rf	Start height	Max Rf	Max height	Max %	End Rf	End height	Area	Area%
1	0.00	7.5	0.06	93.8	21.58	0.08	78.8	2047.4	27.22
2	0.08	80.1	0.12	128.6	29.60	0.14	65.3	2410.4	32.05
3	0.14	79.7	0.15	80.8	18.59	0.21	0.0	875.4	11.64
4	0.22	0.7	0.23	16.0	3.67	0.25	0.7	113.6	1.51
5	0.25	0.5	0.28	46.2	10.63	0.31	0.1	639.3	8.50
6	0.39	0.2	0.43	27.4	6.30	0.47	13.5	568.2	7.56
7	0.48	12.9	0.51	26.3	6.05	0.58	0.6	597.7	7.95
8	0.67	0.3	0.71	15.5	3.57	0.75	2.4	268.2	3.57

Table 4. FT-IR spectrum peak table

Major Absorption Peak	Range in cm^{-1}	Functional Group
1.	3424	May be due to NH_2 stretching vibration and O-H stretching vibration, broad
2.	2914	May be due to Broad O-H stretching and C-H vibrations
3.	2811	May be due to N-H stretching vibration
4.	2145	May be due to CN and $\text{N}=\text{C}=\text{S}$ stretching vibration
5.	2231	May be due to O-H and N-H stretching vibration
6.	1539	May be due to N-H deformation
7.	1512	May be due to existence of amide group
8.	1426	May be due to C-H deformation
9.	1322	May be due to C-O stretching and O-H deformations
10.	1016	May be due to C-O stretching
11.	940	May be due to aliphatic COOH
12.	832	May be due to aromatic O-H deformation
13.	514	May be due to C-O-C deformation
14.	486	May be due to aromatic C-C skeleton vibration
15.	452	May be due to aromatic C-OH in-plane bending vibration





Yasiga and Visweswaran



Fig 1. TLC Visualisation of PMKC at 360 nm

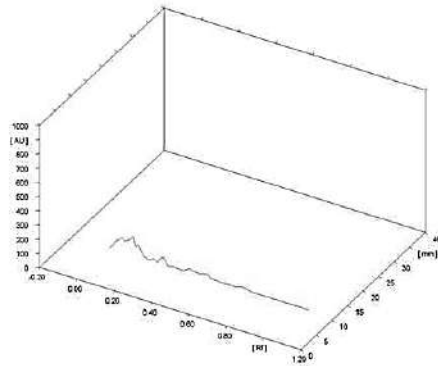


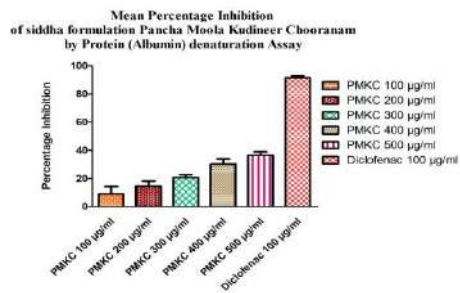
Fig 2. 3D chromatogram



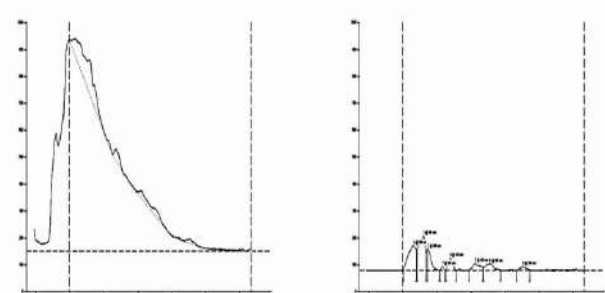
Fig 3. PanchaMoola Kudineer Chooranam



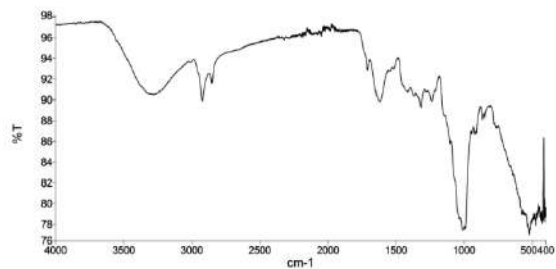
Fig 4. Decoction of PanchaMoola Kudineer Chooranam



Graph 1. Protein inhibition of PMKC and Diclofenac sodium



Graph 2.1, 2.2 HPTLC fingerprinting of PMKC



Graph 3. FT-IR spectrum of Sample PMKC





Allelopathic Proclivities of Tree Leaf Extracts on Seed Germination and Growth of *Aracheis hypogea* L. and *Vigna mungo* (L.) Hepper

Parveen Kaur¹ and P. Leela^{2*}

¹Research Scholar, Department of Botany, Annamalai University, Annamalai Nagar, Tamil Nadu, India.

²Assistant Professor, Department of Botany, Annamalai University, Annamalai Nagar, Tamil Nadu, India.

Received: 28 May 2024

Revised: 19 Aug 2024

Accepted: 29 Oct 2024

*Address for Correspondence

P. Leela

Assistant Professor, Department of Botany,
Annamalai University, Annamalai Nagar,
Tamil Nadu, India.

E.Mail: leeladde72@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The Pot experiment was conducted in the present study to evaluate the allelopathic potential of *C. siamea* tree leaves against *A. hypogea* and *V. mungo*. Grinded leaves of *C. siamea* were soaked in tap water for 5 hours at room temperature. The different concentrations of tree species were 10, 20, 30, 40 and 50g/L. A completely randomized design having three repeats was used. Ten seeds of each species were sown in pots and then irrigated with the respective extracts soon after sowing. Results showed that germination percentage and seedling length, pigments, phenol, soluble sugar and catalase of both species were significantly affected by different concentrations of *C. siamea*. The extracts showed that stimulatory effect on germination of both test species of ground nut and black gram. The Maximum germination percentage (100%) was recorded at 10g/L extract treatment of *C. siamea*. Similarly, the maximum plant height was also observed at 41.66 cm in ground nut and 40.2 cm in black gram respectively. Low concentration of *C. siamea* extract treatment proved stimulatory effects in seedlings as compared to higher concentrations. Higher concentrations treatment showed a negative effect on the species tested. The biochemical contents were also reduced at 50 g/L extract treatment over control. Hence it can be concluded from the results that allelopathy effects of trees can be concentration dependent. The stimulatory and inhibitory effects are also different from different species and concentration also affect the crops. Presence of different allelochemicals, *C. siamea* extract can be used as a viable weed management technique in the future. Between the two test crops more inhibition was observed in black gram than in ground nut.

Keywords: Allelopathy, allelochemicals, germination, control, inhibition, effects, species.

INTRODUCTION

Allelopathy is a widespread biological phenomena in which one organism creates biochemicals that affect the growth, survival, development, and reproduction of others. These biochemicals, known as allelochemicals, can have



**Parveen Kaur and Leela**

positive or negative impacts on target species. Plant allelopathy is a mode of interaction between receptor and donor plants that can have either positive or negative effects. To maintain sustainable agricultural development, cultivation strategies that take use of allelopathic plants' stimulatory/inhibitory influence must be used to govern plant growth and development while avoiding allelopathic autotoxicity. Allelochemicals may be employed as growth regulators, herbicides, insecticides, and antimicrobial crop protection treatments. The "inhibitory" chemical is released into the soil environment where it affects the development and growth of neighbouring plants. Allelopathic chemicals can be present in any part of the plant. They can be found in leaves, flowers, roots, fruits, or stems. They can also be found in the surrounding soil. These toxins affect target species in many different ways. The toxic chemicals may inhibit shoot/root growth, they may inhibit nutrient uptake, or they may attack a naturally occurring symbiotic relationship thereby destroying the plant's usable source of a nutrient [1].

Not all plants have allelopathic tendencies. Some, though they exhibit these tendencies, may be displaying aggressive competition of a non-chemical form. Much of the controversy surrounding allelopathy is in trying to distinguish the type of competition being displayed. In general, if it is chemical, then the plant is considered allelopathic. There have been some recent links to plant allelotoxins directed at animals, but data is scarce. Allelopathy is emerging as a new discipline in agricultural sciences because it is hoped that the use of this science will be proved environment friendly, cost effective and cheaper. Numerous scientists have argued that a detailed study of allelopathy can reduce the reliance on herbicides. Sidhu and Hans [2] found that when the concentration of Eucalyptus extracts increased, plant growth reduced. Phlomina and Srivasuki [3] reported that leaf leachates from five multipurpose tree species (Eucalyptus camaldulensis, Acacia nilotica, Derris indica, Cassia siamea, and Sesbania grandiflora) exhibited variable degrees of inhibitory and stimulating effects on germination percentage. May and Ash [4] concluded that Eucalyptus stunted the growth of several species. Hunshal *et al.* [5] conducted allelopathic investigations and examined the chemical composition of tree species. Cheema *et al.* [6] studied for the commercial use of sorghum water extracts for weed control in wheat.

Agroforestry is a land use system, that integrates trees, crops and/or animals in a scientifically sound way, practically feasible, ecologically desirable and socially acceptable by the farmers [7]. In the present market-oriented world the dimensions of agroforestry have changed from subsistence to commercial and Eco-friendly with a rider of maintaining a balance between ecology and economy. Agroforestry cropping systems occupy an intermediate position, between natural and agro ecosystems, on a scale measuring domestication of the production environment. This is a result of blending agriculture with forestry. These systems should be designed to mimic at least some processes of natural forest ecosystems to obtain benefits that must otherwise accrue to the system externally. Woodmansee [8] compared natural with agricultural ecosystems, and concluded that natural ecosystems have developed several mechanisms to sustain productivity that could be exploited in properly designed agricultural systems.

Allelochemicals have a variety of effects, including decreased plant growth, absorption of water and mineral nutrients, ion uptake, leaf water potential, shoot turgor pressure, osmotic potential, dry matter production, leaf area expansion, stomatal aperture size, stomatal diffusive conductance, and photosynthesis [9]. Allelochemicals are mostly secondary metabolites that are released into the environment via natural processes such as volatilization, leaf leaching, residue breakdown, and/or root exudate. As a result, it is important to first understand how allelochemicals enter the environment. Allelochemical activity varies according to research methodology and operating processes [10]. The natural state of allelochemicals may be changed somewhat during the process of extraction [11]. As a result, researchers must exercise caution when determining whether a plant has allelopathic potential or separating and identifying allelochemicals using organic solvents and aqueous extracts of plant tissues.

An allelochemical discharged into the environment is typically not a single substance, and the amount released varies depending on the conditions. When investigating plants' allelopathic potential, it is important to evaluate both the type and amount of allelochemicals they release. Interactions such as synergy, antagonism, and incremental effects between different allelochemicals should be investigated because one allelochemical may not exhibit allelopathic



**Parveen Kaur and Leela**

activity as a single component in a given situation but may increase allelopathy when combined with other allelochemicals [12]. Allelopathy is a physiological and ecological mechanism that influences agricultural crop yield [13]. Because of the favourable climatic conditions and socioeconomic relevance of crops in the human diet, crop production is relatively frequent [14]. Plant influences include species, variety, growth stage, and tissue types [15]. Allelopathic effects differ with variety or genotype [16]. Plants from the same environment or with close taxonomic proximity do not always produce secondary metabolites in the same quantities or quality, and thus may not have similar allelopathic effects [17,18]). In a recent study, endogenous allelochemical levels were employed to assess abiotic stress resistance. Meanwhile, exogenous application of allelochemicals has been shown to increase the endogenous level of receivers, resulting in increased growth and resistance to abiotic stresses[19]; thus, appropriate environmental conditions are required for allelopathic studies. According to Albuquerque *et al.* [12], stress can boost the release of allelochemicals from allelopathic plants.

Understanding allelopathy is crucial for understanding its impact on plant germination and seedling growth [20,21]. Germination and seedling growth are critical stages in plant development [22,23] Allelopathy and autoallelopathy are being used to manage organic output in new ways. There is convincing evidence that allelopathic interactions between plants play a crucial role in natural as well as manipulated ecosystems. In recent times evidence is accumulating that all types of plants viz herbs, shrubs and trees, allelopathically affect the patterning of vegetation, largely in their immediate vicinity. Therefore, in the present work, an attempt has been made to evaluate and compare the allelopathic potentiality of the common agroforestry tree (*C. siamea*) on the germination, seedling growth, and biochemical parameters of two crop species (*A. hypogea* and *V. mungo*).

MATERIALS AND METHODS

A pot experiment was conducted in the Department of Botany, Annamalai I University, Annamalai Nagar, to assess the allelopathic proclivities of tree leaf extracts on seed germination and growth of ground nut (*Arachis hypogea*) and black gram (*Vigna mungo*). The fresh green leaves of *Cassia siamea* were collected and dried in shed. The leaves were then grinded with the help of a grinder. The grinded material was then soaked in tap water for 5 hrs at room temperature (23 °C). From this stock solution different concentrations (10, 20, 20, 40 and 50 g/ L) of extracts were prepared and control crops were used in tap water. The experiment was laid out in a completely randomized design (CRO) and repeated three times.

A sufficient quantity of healthy seeds of two food crops viz. ground nut and black gram were tested for viability and healthy seeds of each food crop were surface sterilized in 15% sodium hypochlorite for 20 min and rinsed several times with distilled water. Ten seeds of each species were sown in pots containing 5 kg of garden soil. The pots were treated with different treatments (concentration) of extract solution and then treated with the respective extracts soon after sowing. The control pots were irrigated with the required amount of water. No fertilizer was applied during the experiment. Data on germination percentage was recorded ten days after sowing and plant height (cm) was recorded 28 days after sowing. Numbers of ground nut and black gram plants that emerged from the soil were counted in each pot and then average was calculated. To record the plant height, crops present in each pot were measured from ground level to the tip of the seedling and then the average was computed.

Experiment was carried out using a Completely Randomized Design (CRD) with 5 treatments, that is, Control (only distilled water); 10 g/L leaf concentration, 20 g/L leaf extract concentration, 30g/L leaf extract concentration, 40g/L leaf extract concentration and 50 g/L leaf extract concentration and each treatment was replicated three times. The number of seeds germinated was counted daily in each treatment and germination was recorded every day till 10 days root and shoot lengths of seedlings were measured and recorded on the 25th day. The experimental data were subjected to analysis of variance (ANOVA). The percentage of inhibition/stimulation effect was calculated using the formula given by Surendra and Pot (1978): $I = 100 - (E2 \times 100/E1)$, where I - % of inhibition/stimulation, E1- the response of control and E2- the response of treatment.





Parveen Kaur and Leela

RESULTS AND DISCUSSION

Allelopathic effects of aqueous leaf extracts of *C. siamea* on seed germination of *A. hypogea* and *V. mungo* (Table-1) showed the stimulatory effects and inhibitory effects on germination. The leaf extract with a higher concentration of (50g/L) *C. siamea* showed an inhibitory effect on seed germination percentage for *A. hypogea* and *V. mungo* but a stimulatory effect was observed at lower concentration extract treatment on both test crops in pot study. At 40g/L extract treatments, the result showed a in the reduction percentage of germination in both test crops (-28.28% and -42%). The variation in germination of different food crops might be due to the variation of species. More days were required for sprouting under higher extract concentration, at the same time positive response was noticed in the 10g/L treatment. The concentration showed greater potential at higher concentrations (Table 1) which can be attributed to the relative amount of allelochemicals released by the extract (El Rokiek *et al.*, 2010). The variation in germination of different food crops might be due to the variation in tolerance of the species to different concentrations of the leaf extracts. The inhibitory effect of germination had also been reported earlier by [24] in *Casia occidentalis*. Lower germination may be the result of water uptake inhibition [25], and the disturbance in the synthesis as well as the activity of gibberellic acid (GA3) [26].

Germination was higher in the control and lower concentrations of leaf extracts and water uptake could have played a role in the process. During seed germination, several metabolic and physiological processes could be attributed to water uptake. Reduction in germination percentage has also been reported by some other authors like [27] in wheat treated with *Eucalyptus camaldulensis* leaf extract and [28] in six plant species treated with *Azadirachta indica* leaf extracts. The allelopathic evaluation in this study showed that the two food species responded to the application of different concentrations of *C. siamea* leaf extracts, as seen by the considerable effects on seed germination. In general, the concentrations of 40g/L and 50g/L caused the maximum inhibition of the germination rate for ground nut and black gram. The effect of 10g/L concentrations of aqueous leaf extracts on plant growth has been explored, and higher concentrations are known to inhibit sprouting [29]. Germination and growth response of the target plants to the allelochemicals may be due to several reasons. Higher concentrations of extract stressed the environment for the test crop which fails in metabolic machinery to activate the embryo to germinate. Lower concentration may stimulate efficient enzyme and hormonal coordination for successful germination. It may be asserted that plants growing in allelochemically induced environment may experience changes in enzyme functionality, water and mineral uptake, permeability of cell membrane and photosynthetic activity corresponding to either reduced germination, seedling growth and dry matter accumulation or stimulation in these parameters [30]. The treatment of 50 g/L was found to be the poorest growth performing treatment of crops. The maximum root length and shoot length reduction were recorded. The positive growth response was observed in both test crops at 10g/L-1 treatment of plant leaf extracts. The comparison between treatments and control showed that there was a significant difference. The magnitude of inhibition of root length was considerably greater than shoot length as evident from higher values of response percentage (Table-2). There was significant variation in the root length and shoot length of ground nut and black gram with different concentrations of *C. siamea* leaf extracts.

The plant leaf extracts had an inhibiting effect on root and shoot growth. Statistical analysis revealed that the greatest reduction recorded at the highest concentration had a significant influence on the shoot and root length of groundnut and black gramme over control. However, smaller doses (10 g/L-1) of extract treatment in ground nut and black gramme showed significant differences from the control. Allelopathic plants inhibit the growth of nearby plants by influencing their biochemical and physiological processes. Secondary metabolites (allelochemicals) interact with other plants in a variety of chemical interactions, suppressing their growth by influencing plant machinery at the cellular, physiological, and biochemical level. Allelochemical doses and kinds are directly related to their effects on ion absorption. A little quantity of dibutyl phthalate increases nitrogen uptake while decreasing P and K uptake. A considerable amount of this chemical inhibits the absorption of N, P, and K. Similarly, when diphenylamine levels are low, tomato roots absorb N and K more quickly than P [31]. Plants combine phenolic chemicals to modify their defence mechanisms against varied conditions [32].



**Parveen Kaur and Leela**

The inhibitory action of phenol has been attributed to its effect on membrane functions, membrane potential, mineral absorption and plant water relations. This experiment showed that *C. siamea* leaf extracts had more inhibitory effects on the root length and shoot length of *V. mungo* seedlings than in *A. hypogea* crops (Table-2). Stimulatory effects were also noticed at lower concentration (10 g/L) treatments in both test crops. As the concentration of extracts increased the inhibition of root length and shoot length also increased. A large number of phytochemicals may be present in the leaf extracts, some of which may be responsible for inhibition and certain for stimulation.

The toxic chemicals of *C. siamea* may inhibit shoot or root growth, they may inhibit nutrient uptake, or they may attack a naturally occurring symbiotic relationship thereby destroying the plant's usable source of nutrients [33]. A similar trend was observed John *et al.* [34] that fresh leaf extract of mango contains terpenoids and triterpenes, tamarind has flavonoids and terpenoids while teak contains triterpenes. The reduction increased with the increased rates of residues. When the crops were treated with *C. siamea* leaf extracts showed chlorophyll reduction in the ground nut and black gram seedlings over control seedlings. The results also showed that chlorophyll content was significantly reduced in 40g/L (chl.a.-29.71%, chl.b. -47.44%, total chlorophyll -39% and carotenoids -20.83% in *A. hypogea* respectively; chl.a. -29.29%, chl.b. -32.10%, total chlorophyll -30.60% and carotenoids -32.01% in *V. mungo* respectively) and 50g/L (-42.50%, 55.43%, 49.28% and -39.56% ; -52.22%, -45.21%, -48.94%, -51.09% in *A. hypogea* and *V. mungo* chl.a, chl.b, total chlorophyll and carotenoids respectively) leaf extracts treatments which is accordance with other observations. The reduction in chlorophyll content ultimately resulted in the reduction or decline in photosynthesis in both crop plants.

Chlorophylls a, b, total chlorophyll and carotenoids accumulated in *A. hypogea* and *Vigna mungo* at lower doses (10g/L-1); the following percentages were observed in both crops; chl.a 4.26% & 4.08%, Chl. b 2.74% & 3.14, total chlorophyll 3.46% & 3.64% and carotenoid contents 0.17% & 0.87% of *C. siamea* leaf extracts treatments over control. However the higher doses (20,30,40 and 50g/L) inhibited seedling chl. a, chl. b, total chlorophyll and carotenoids contents and the effect was concentration dependent. Allelochemicals predominantly influence photosynthesis circuitry in plants, accelerating the breakdown of photosynthetic pigments [35,36]. Many allelochemicals have been investigated for their effects on photosynthesis in plants. Allelochemicals have been shown to degrade photosynthetic enzymes. These allelochemicals mostly impair Photosystem II (PSII), affecting photosynthesis significantly [37,38]. Sorgoleone is a well-known allelotoxin and lipophilic benzoquinone component that inhibits PSII by limiting the photosynthetic electron transport chain (ETC) [39]. Chlorophylls a, b, and carotenoids accumulated in ground nut and black gramme at lower dosages of *C. siamea* leaf residues, as did total sugar, primarily the insoluble component. The suppression of photosynthetic pigments at higher concentrations of *C. siamea* leaf extracts was accompanied by a considerable drop in all sugar fractions.

Significant effects of the treatments on Chlorophyll a, chlorophyll b, total chlorophyll and carotenoid contents were recorded with respect to *C. siamea*, on annual crops (ground nut and black gram) as well as the interaction between tree species and annual crops (Tables 3 & 4). Between the two annual crops, a significant increase was recorded in ground nut while, the decrease was recorded in black gram. Results were found similar to the findings of Venkateshwarlu [40], who has reported inhibition in chlorophyll a and chl. b content (41.66% and 11.36% respectively) in radish when treated with leaf extract of *M. indica* (200 ppm). The reduction in chlorophyll contents observed in the few combinations might be due to the degradation of chlorophyll pigments or reduction in their synthesis and the action of flavonoids, terpenoids or other phytochemicals present in leaf extracts [41]. Reduction in chlorophylls may decrease photosynthesis and thereby substantially decrease all the metabolites viz., total sugars, proteins and soluble amino acids [42].

Concerning the total phenol content of the annual plants, a significant effect of the treatments could be recorded (Tables 5 & 6). The phenolic content of two seedlings, stimulatory and inhibitory effects was noticed over control. Between the two annual crops, significantly higher values (1.16%) could be found in black gram, when treated with a lower concentration of extract treatment (10g/L) while it was recorded significantly lower in ground nut (-



**Parveen Kaur and Leela**

0.54%). Tables 5 and 6 show that the treatments had a substantial influence on the total phenol content of the annual plants. The phenolic content of two seedlings showed stimulatory and inhibitory effects compared to the control. Compared to the two annual crops, black gramme had significantly higher values (1.16%) when treated with a lower concentration of extract (10g/L), while ground nut had significantly lower values (-0.54%). The largest stimulatory impact was discovered in black gramme (1.16%) when treated with 10g/L leaf extract, while the maximum inhibitory effect was also obtained in the same crop (-64.33%) when treated with 50g/L extract. This phenolic compound may interfere with the phosphorylation pathway, inhibit the activation of Mg and ATPase activity, or be caused by decreased synthesis of total carbohydrate, protein, and nucleic acid (DNA and RNA), or interference in cell division, mineral uptake, and biosynthetic processes [43].

Significant impacts of the treatment on total soluble sugar and enzyme catalase content of the annual plant were observed for two annual crops, as well as interactions between tree species and annual crops (Tables 5 and 6). The 10g/L concentration treatment resulted in the highest total soluble sugar (0.86% & 0.40% in ground nut and black gramme) and catalase (3.47% & 1.78% in ground nut and black gramme, respectively) content. The observed reduction in total soluble sugar and catalase levels in black gramme is most likely due to photochemical interference in total sugar biosynthetic processes, as validated by Singh and Rao [44] in rice. During the study period, significant effects of treatments on tree species and annual crops were observed.

Allelochemicals affect membrane permeability and integrity [45], plant water relations [46], cell division [47], hormone biosynthesis and transport [48], mineral uptake and transport [49] soil nutrient composition [50], stomatal oscillations [51], and photosynthesis. Phytotoxins can alter plant growth through a variety of mechanisms. They can have an impact on plant biochemistry, physiology, cytology, and morphology, altering its growth and development directly. Regardless of the relationships between plant species, a robust ground for the scientific underpinning of the survival and utility of the allelopathic process should be generalised.

CONCLUSION

The allelopathic potential of trees and crops can affect tree group growth and division, as well as the yield of vital plants, and allelopathy has proven to be useful in this case. When trees and crops grow together, they form a network that inhibits or stimulates their growth or yield through direct or indirect allelopathic interaction. Allelopathy plays an important function in the ecosystem, affecting plants in both positive and negative ways. The study revealed that the *C. siamea* tree leaf extracts contain allelochemicals, these chemicals affected the studied test crops i.e. groundnut and black gram germination, seedling length, chlorophyll, carotenoids, total soluble sugar, total phenol and catalase. This experiment concluded that the allelopathic effect was concentration dependent, and the effect was also different for different species. Between the two crops more allelopathic effect was noticed in *V. mungo* than in *A. hypogea*. Allelopathic connections are a product of secondary metabolism and show diversity in chemical nature and function as a group. There is a need for further study to be carried out on identifying the inhibiting allelochemical in the parts investigated.

REFERENCES

1. Conn E.E. Cyanogenic compounds. Ann Rev Plant Physiol. 1980;31:433-52.
2. Sidhu, D.S. and A.S. Hans (1988). Preliminary studies on the effect of Eucalyptus leaf-litter on accumulation of biomass in wheat. J. Trop. Fores., 4(4): 328-333.
3. Philomina, N.S. and Srivasuki, K.P. 1996. Allelopathic studies on Agroforestry species. Indian J. Forestry.19: 45-53.
4. May F.E and Ash J.E. 1990. An assessment of the allelopathic potential of Eucalyptus. Australian Journal of Botany. 38: 245-254





Parveen Kaur and Leela

5. Hunshal, CS, HT Channal. AR Alagawadi. RH Patil and SS. Narwal 2000 Allelopathy research in agroforestry systems of South India Proc 3d Inter Cong Allelopathy in Ecological Agric. and Forestry. Dharwad. India, 18-21 August, 1998, pp. 209-227.
6. Cheema, Z.A., Khaliq, A., 2000. Use of sorghum allelopathic properties to control weeds in irrigated wheat in a semi arid region of Punjab. Agric. Ecosyst. Environ. 79 (2–3),105–112. [https://doi.org/10.1016/S0167-8809\(99\)00140-1](https://doi.org/10.1016/S0167-8809(99)00140-1).
7. Nair, P.K.R. 1979. Intensive multiple cropping with coconuts in India. Verlaug Pau Parey, Berlin and Hamburg, Germany. p 147. Nair, P.K.R., 1993.
8. Woodmansee, R.G. 1984. Comparative nutrient cycles of natural and agricultural ecosystems: A step toward principles, p. 145–156. In: R. Lowrance, B.R. Stinner, and G.J. House (eds.). Agricultural ecosystems—Unifying concepts. Wiley, New York.
9. Weir TL, Park SW, Vivanco JM (2004). Biochemical and physiological mechanisms mediated by allelochemicals. Curr. Opin. Plant Biol. 7:472-479
10. Peng SL, Wen J, Guo QF (2004). Mechanism and active variety of allelochemical. Acta Bot. Sinica 46:57-766.
11. Li, S. T., Zhou, J. M., Wang, H. Y., and Chen, X. Q. (2002). Research surveys of allelopathy in plants. *Chin. J. Eco. Agric.* 10, 72–74.
12. Albuquerque, M. B., Santos, R. C., Lima, L. M., Melo Filho, P. D. A., Nogueira, R. J. M. C., Câmara, C. A. G., et al. (2010). Allelopathy, an alternative tool to improve cropping systems. *Rev. Agron Sust. Dev.* 31, 379–395. doi: 10.1051/agro/2010031
13. Farooq, N., Abbas, T., Tanveer, A., & Jabran, K. (2020). Allelopathy for weed management. In J. M. Mérillon & K. Ramawat (Eds.), *Co-evolution of secondary metabolites* (pp. 505–519). Springer. https://doi.org/10.1007/978-3-319-96397-6_16
14. Georgieva, N. (2019). Allelopathic tolerance in white lupine (*Lupinus albus* L.) accessions to Sorghum halepense extracts. *Journal of BioScience and Biotechnology*, 8(1), 51–58
15. Belz, R. G. (2007). Allelopathy in crop/weed interactions—an update. *Pest. Manag. Sci.* 63, 308326. doi: 10.1002/ps.1320
16. Leao, P. N., Engene, N., Antunes, A., Gerwick, W. H., and Vasconcelos, V. (2012). The chemical ecology of cyanobacteria. *Nat. Prod. Rep.* 29, 372–391. doi: 10.1039/c2np00075j
17. Chon, S. U., and Nelson, C. J. (2010). Allelopathy in compositae plants. A review. *Agron. Sustain. Dev.* 30, 349–358. doi: 10.1051/agro/2009027
18. Hagan DL, Jose S, Lin CH (2013) Allelopathic exudates of cogongrass (*Imperata cylindrica*): implications for the performance of native pine savanna plant species in the southeastern US. *J Chem Ecol* 39(2):312–322
19. Maqbool, N., Wahid, A., Farooq, M., Cheema, Z. A., and Siddique, K. H. M. (2013). “Allelopathy and abiotic stress interaction in crop plants,” in *Allelopathy*, eds Z. A. Cheema, M. Farooq, and A. Wahid (Berlin: Springer Berlin Heidelberg), 451–468. doi: 10.1007/978-3-642-30595-5_19
20. El Beheiry, M. A., El Fahar, R. A., Tahoun, E. M., & Abd Elhaak, M. A. (2019). Mitigation of wheat seedlings to the allelopathic effect of *Malva parviflora* and *Rumex dentatus* weeds. *Egyptian Journal of Experimental Biology (Botany)*, 15(2), 269–282. <https://doi.org/10.5455/egyjebb.20190822013134>
21. Sołtys, D., Dębska, K., Bogatek, R., & Gniazdowska, A. (2010). Autotoksyczność roślin jakoprzykład oddziaływań allelopatycznych [Plant autotoxicity – An example of allelopathic interaction]. *Kosmos*, 59(3–4), 551–566.
22. Korotkova, I., Marenych, M., Hanhur, V., Laslo, O., Chetveryk, O., & Liashenko, V. (2021). Weed control and winter wheat crop yield with the application of herbicides, nitrogen fertilizers, and their mixtures with humic growth regulators. *Acta Agrobotanica*, 74(1), Article 748. <https://doi.org/10.5586/aa.748>
23. Shahrajabian, M. H., Khoshkham, M., Sun, W., & Cheng, Q. (2019). Germination and seedlings growth of corn (*Zea mays* L.) to allelopathic effects of rice (*Oryza sativa* L.). *Tropical Plant Research*, 6(1), <https://doi.org/10.22271/tp.2019.v6.i1.0222024>.
24. Ashafa AOT, Ogbé A.A, Osinaiké T (2012). Inhibitory effect of mango (*Mangifera indica* L.) leaf extracts on the germination of *Cassia occidentalis* seeds. *African Journal of Agricultural Research* 7(33):4634-4639.





Parveen Kaur and Leela

25. Tawaha A.M, Turk M.A (2003). Allelopathic effects of black mustard (*Brassica nigra*) on germination and growth of wild barley (*Hordeum spontaneum*). *Journal Agronomy Crop Science* 189:298- 303
26. Olofsdotter M (2001). Rice-a step toward use of allelopathy. *Agronomy Journal* 9(3):3-8
27. Khan MA, Hussain I, Khan EA (2009). Allelopathic effects of Eucalyptus (*Eucalyptus Camldulensis* L.) on germination and seedling growth of wheat (*Triticumaestivum* L.). *Impact factor of Pakistan Journal of Weed Science Research* 15(2-3):131-143.
28. Zoheir YA, Yahya A, Aptin R, Sedighi S, Hassan, AM, Mashhadi HR (2008). Study of the Allelopathic Potential of Extracts of *Azadirachta Indica* (Neem). *OnLine Journal of Biology Science* 8(3):57-61. <http://dx.doi.org/10.3844/ojbsci.2008.57.61>
29. Ahmed, R., M.B. Uddin, M.A.S.A. Khan, S.A. Mukul and M.K. Hossain.2007. Allelopathic effects of Lantana camara on germination and growth behavior of some agricultural crops in Bangladesh. *J. Forestry Res.* 18 (4): 301-304(13)
30. Muhammad Z. Majeed A. (2014) Allelopathic effects of aqueous extracts of sunflower on wheat (*Triticum aestivum* L.) and maize (*Zea mays* L.). *Pakistan Journal of Botany*, 46(5): 1715-1718.
31. Geng G, Zhan S, Cheng Z (2009) Effects of diferent allelochemicals on mineral elements absorption of tomato root. *China Veg* 4:48–51
32. Parvin K, Nahar K, Mohsin SM, Mahmud JA, Fujita M, Hasanuzzaman M (2022) Plant phenolic compounds for abiotic stress tolerance. In: *Managing plant production under changing environment*, pp 193–237
33. Hierro JL, Callaway RM. 2003. Allelopathy and exotic plant invasion. *Plant and soil*. 256, 29-39.
34. John, J., Leela, N. K., Sreekumar, K. M., Anesh, R. Y., and Hema, M. 2007b. Phytotoxicity of leaf extracts of multipurpose trees against insect pests in bitter gourd (*Momordica charantia*) and brinjal (*Solanum melongena*). *Allelopathy J.* 20(2): 411-418
35. Rehman S, Shahzad B, Bajwa AA, Hussain S, Rehman A, Cheema SA, Abbas T, Ali A, Shah L, Adkins S, Li P (2019) Utilizing the allelopathic potential of Brassica species for sustainable crop production: a review. *J Plant Growth Regul* 38(1):343–356
36. Yuliyani ED, Darmanti S, Hastuti ED (2019) Allelochemical effects of *Chromolaena odorata* L. against photosynthetic pigments and stomata of *Ageratum conyzoides* L. leaves. In: *Journal of physics: conference series*, vol 1217, no 1, p 012149. IOP Publishing
37. Wang CM, Chen HT, Li TC, Weng JH, Jhan YL, Lin SX et al (2014) The role of pentacyclic triterpenoids in the allelopathic effects of *Alstoniascholaris*. *J Chem Ecol* 40:90–98
38. Hussain MI, Reigosa MJ (2017) Evaluation of photosynthetic performance and carbon isotope discrimination in perennial ryegrass (*Lolium perenne* L.) under allelochemicals stress. *Ecotoxicology* 26(5):613–624
39. Dayan FE, Howell JL, Weidenhamer JD (2009) Dynamic root exudation of sorgoleone and its in planta mechanism of action. *J Exp Bot* 60(7):2107–2117.
40. Venkateshwarlu, G.; Ravindra, V. and Challa, P. 2001. Mangiferin: an allelopathin from mango (*Mangifera indica* L.) leaves. *Allelopathy Journal*. 8(2): 221-224.
41. Tripathi, S., Tripathi, A. and Kori, D.C. 1999. Allelopathic evaluation of *Tectona grandis* leaf root and soil aqueous extracts on soybean. *Indian J. For.*, 22 : 366-374
42. Singh N.B. and Ranjana, S. 2003. Effect of leaf leachate of Eucalyptus on germination growth and metabolism of green gram, black gram and peanut. *Allel. J.* 11 : 43-52.
43. Pawar, K.B. and Chavan, P.D. 2004. Influence of leaf leachates of some plant species on free proline content in germinating seed of *Sorghum bicolor* (L.) Moench. *Allelopathy J.* 3:89-92.
44. Singh, D. and Rao, Y.B. 2003. Allelopathic evaluation of *Andrographis paniculata* aqueous leachates on rice (*Oryza sativa* L.). *Allelopathy J.* 11 : 71–6.
45. Poulin RX, Hogan S, Poulson-Ellestad KL, Brown E, Fernández FM, Kubanek J (2018) *Karenia brevis* allelopathy compromises the lipidome, membrane integrity, and photosynthesis of competitors. *Sci Rep* 8(1):9572
46. Araniti F, Sánchez-Moreiras AM, Grana E, Reigosa MJ, Abenavoli MR (2017) Terpenoid trans-caryophyllene inhibits weed germination and induces plant water status alteration and oxidative damage in adult *Arabidopsis*. *Plant Biol* 19(1):79–89





Parveen Kaur and Leela

47. Goga M, Antreich SJ, Bačkor M, Weckwerth W, Lang I (2017) Lichen secondary metabolites affect growth of *Physcomitrella patens* by allelopathy. *Protoplasma* 254(3):1307–1315
48. Li P, Ding L, Zhang L, He J, Huan Z (2019) Weisiensin B inhibits primary and lateral root development by interfering with polar auxin transport in *Arabidopsis thaliana*. *Plant Physiol Biochem* 139:738–745
49. Lupini A, Araniti F, Mauceri A, Princi MP, Sorgonà A, Sunseri F, Varanini Z, Abenavoli MR. Coumarin enhances nitrate uptake in maize roots through modulation of plasma membrane H⁺-ATPase activity. *Plant Biol.* 2018;20(2):390–398. doi: 10.1111/plb.12674.
50. Mohammadkhani N, Servati M (2018) Nutrient concentration in wheat and soil under allelopathy treatments. *J Plant Res* 131(1):143–155
51. Syahri R, Widaryanto E, Wicaksono KP (2017) .Bioactive compound from mangoes leaves extract as potential soil bioherbicide to control amaranth weed (*Amaranthus spinosus* Linn.). *J Degrad Min Lands Manag* 4(3):829.

Table 1. Germination Percentage of groundnut and black gram against *C. siamea*

Extract concentration(g/L)	<i>A. hypogea</i>	<i>V. mungo</i>
Control	99	100
10 g/L	100(1.01)	100(0)
20 g/L	97(-2.02)	95(-5)
30 g/L	86(-13.12)	79(-21)
40 g/L	71(-28.28)	58(-42)
50 g/L	55(-44.44)	40(-60)

Data in parenthesis indicates % increases or decreases

Table-2 Allelopathic effect of *C. siamea* on root length and shoot length(cm/plant) of *A. hypogea* and *V. mungo*

Extract concentration	<i>A. hypogea</i>		<i>V. mungo</i>	
	Root length	Shoot length	Root length	Shoot length
Control	9.166±0.513	31.866±0.808	7.366±0.450	32.4±0.655
10gL-1	9.266±0.514	32.466±0.950	7.6 ±0.451	33.1±0.660
20gL-1	6.266±0.208	29.033±0.960	6.433±0.503	29.8±0.721
30gL-1	5.566±0.251	26.866±0.808	5.366±0.351	25.53±0.503
40gL-1	5.033±0.351	24.233±0.208	4.866±0.404	20.6±0.529
50gL-1	4.66±0.152	18±0.866	4.1±0.264	12.16 ±0.763

Means followed by Standard Deviation

Table-3 Allelopathic effect of *C. siamea* on Chl.a, Chl.b, Total chlorophyll and carotenoids (mg/g. fr.wt.) contents of *A. hypogea*

Extract concentration	Chlorophyll a	Chlorophyll b	Total chlorophyll	Carotenoids
Control	0.727	0.801	1.528	0.599
10gL-1	0.758(4.26)	0.823 (2.74)	1.581(3.46)	0.676(0.17)
20gL-1	0.632(-13.06)	0.687 (-14.23)	1.319(-13.67)	0.66(-3.17)
30gL-1	0.561(-22.83)	0.54(-32.58)	1.101(-27.94)	0.564(-5.84)
40gL-1	0.511(-29.71)	0.421(-47.44)	0.932(-39.00)	0.476(-20.83)
50gL-1	0.418(-42.50)	0.357(-55.43)	0.775(-49.28)	0.362(-39.56)

Data in parenthesis indicates % increases or decreases





Parveen Kaur and Leela

Table - 4 Allelopathic effect of *C. siamea* on Chl.a, Chl.b, Total chlorophyll and carotenoids (mg/g. fr.wt.) contents of *V. mungo* on 28th Day old seeding

Extract concentration	Chlorophyll a	Chlorophyll b	Total chlorophyll	Carotenoids
Control	0.833	0.732	1.565	0.456
10gL-1	0.867 (4.08)	0.755(3.14)	1.622(3.64)	0.479(0.87)
20gL-1	0.761(-8.64)	0.698(-4.64)	1.459(-6.77)	0.438(-3.94)
30gL-1	0.69(-17.64)	0.591(-19.26)	1.281(-18.47)	0.389(-14.69)
40gL-1	0.589(-29.29)	0.497(-32.10)	1.086(-30.60)	0.31(-32.01)
50gL-1	0.398(-52.22)	0.401(-45.21)	0.799(-48.94)	0.223(-51.09)

Data in parenthesis indicates % increases or decreases over control

Table-5 Allelopathic effect of *C. siamea* on Total soluble sugar, Phenol and catalase (mg/g. fr.wt.) contents of *A. hypogea* on 28th Day old seeding;

Extract concentration	Total soluble sugar	Phenol	catalase
Control	1.267	0.923	1.094
10gL-1	1.278(0.86)	0.918(-0.54)	1.017(3.47)
20gL-1	1.202(-5.13)	0.863(-6.50)	0.901(-17.64)
30gL-1	1.123(-11.36)	0.789(-14.51)	0.845(-22.76)
40gL-1	0.87(-31.33)	0.603(-34.66)	0.699(-36.10)
50gL-1	0.698(-44.90)	0.388(-57.96)	0.436(-60.14)

Data in parenthesis indicates % increases or decreases over control

Table - 6 Allelopathic effect of *C. siamea* on Total soluble sugar, Phenol and catalase (mg/g. fr.wt.) contents of *V. mungo* on 28th Day old seeding.

Extract concentration	Total soluble sugar	Phenol	catalase
Control	0.988	1.901	0.895
10gL-1	0.992(0.40)	1.923(1.16)	0.871(1.78)
20gL-1	0.91(-7.89)	1.796(-5.52)	0.755(-15.64)
30gL-1	0.79(-20.04)	1.509(-20.62)	0.623(-30.39)
40gL-1	0.572(-42.10)	0.992(-47.81)	0.474(-47.03)
50gL-1	0.334(-66.19)	0.678(-64.33)	0.234(-73.85)

Data in parenthesis indicates % increases or decreases over control





Management of Ocular Insect bite through Ayurveda- a Case Study

Unnati Desai¹, Shivkant Sharma^{2*}, Manjiri Keskar³, Anu Vijayan⁴, Manu Prasad⁵

¹PG Scholar, Department of Shalakya Tantra, Parul Institute of Ayurved, Parul University, Vadodara, Gujarat, India

²Associate Professor, Department of Shalakya Tantra, Parul Institute of Ayurved, Parul University, Vadodara, Gujarat, India.

³Professor and HoD, Department of Shalakya Tantra, Parul Institute of Ayurved, Parul University, Vadodara, Gujarat, India.

⁴Assistant Professor, Department of Shalakya Tantra, Parul Institute of Ayurved, Parul University, Vadodara, Gujarat, India.

⁵Associate Professor, Department of Agadtantra, Parul Institute of Ayurved, Parul University, Vadodara, Gujarat, India.

Received: 14 Jun 2024

Revised: 16 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

Shivkant Sharma

Associate Professor, Department of Shalakya Tantra,
Parul Institute of Ayurved,
Parul University, Vadodara,
Gujarat, India.

E. Mail: shivkants@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The origins of *Visha* and *KeetaVisha* were recited by *Acharya Charaka* in *Chikitsa sthana* of *Charaka Samhita*, *Acharya Sushruta* in *Kalpasthan* of *Sushruta Samhita*, and *Acharya Vagbhata* in *Uttartantra* of *Ashtanga Hridaya*, respectively. *Acharya Sushruta* explained *KeetaVisha* as *Jangama Visha* (Animate Poison) and classified 67 different types of *Keeta* into four groups that may cause both acute and severe manifestations in humans. The most common symptoms of an insect bite are itching, a burning sensation, swelling, and pain at the bite site. The diagnosis and treatment of *Keeta visha* differs according to the types of *Keeta*, so they are frequently misdiagnosed because the signs and symptoms resemble those of a skin disease. This is a case report of a 32-year-old female who had a history of an unknown insect bite and presented to the eye OPD with complaints of swelling on both upper eyelids, itching in the periorbital area, burning sensation in both eyes, pain opening her eyes and watering from both eyes. *Acharya Sushruta's Kalpasthan* contains numerous formulations for all types of *Visha*. As a result, the patient was treated with *Aschyotana* (*Aatyayik chikitsa*), *Bilvadi Gutika*, *Punarnava Guggulu* and *Manjishthadi Kashaya*, as well as *Bidalaka*, a local application with *Vishaghna* properties. In this case, drugs have provided reassuring results in the treatment of an unknown insect bite.

Keywords: Aschyotana, Ayurveda, insect bite, Keeta, Nayanabhighata, Ocular emergencies





Unnati Desai et al.,

INTRODUCTION

Netrabhigata (Ocular injury) can occur by either direct or indirect causes. As *Acharya Dalhana* mentioned, direct causes include- direct shock or blow, foreign body, *vayu*, dust, smoke, touch of insects, *Tikshnanjana*, over fomentation of eyes, injury due to water force while swimming, rubbing of the eyes vigorously, staring at celestial bodies like sun with naked eyes [1]. According to *Charak Acharya*, insects are also known as *Keeta* because they are produced from *Kitta* or waste products such as stool and urine [2]. The term *Keeta* refers to all living creatures which are small in size but visible, having two or more legs with or without wings and stings are present, which is included in *Jangamvisha* [3]. An insect bite to the eyelid usually results in redness and inflammation of the eyelid and surrounding area. Fluid accumulation and inflammation are common after an insect bite due to the loose tissue surrounding the eye [4]. In severe cases, it can even prevent the eye from opening, especially when lying down, because the fluid gravitates to that area. Because the skin around the eye is sensitive, the itching and discomfort caused by a bite on the eyelid may feel especially intense; however, most of the time, the itchiness lasts only a few days. Symptoms may include swelling, redness of the eyes, pricking or burning pain, and tenderness to touch occurring around one or bilateral eyes. The affected person can move the eye in any direction without pain, but opening the eyelid can be difficult, often due to swelling. The significant variation in clinical presentation and outcomes between several cases has made it difficult to develop a therapeutic algorithm.

Aim

To evaluate the efficacy of *Ayurveda* therapy in the management of ocular insect bite.

Objectives

- To study on insect bite in detail.
- To study the probable mode of action of *Ayurveda* management of ocular insect bite.

MATERIALS AND METHODS

Case Report

A 32 years old female, resident of Vadodara reported to *Shalakyatantra* Eye OPD of Parul Ayurved Hospital, Vadodara with following complaints since 1 day:

- Swelling on bilateral upper eyelids and forehead
- Burning sensation in bilateral eyes
- Itching at periorbital area
- Pain while opening the eyes
- Watering from bilateral eyes

Patient was vitally stable and her appetite was moderate.

Past History

History of unknown insect bite on both upper eyelid 1 day back.

Ocular examination

Swelling over the forehead (glabella). No evidence of any inflammatory changes. After keen ocular examination with slit lamp, no extra ocular foreign body was found.

Systemic Examination

BP- 110/80 mmHg

Pulse- 83/min

T- Afebrile

RR- 18/min



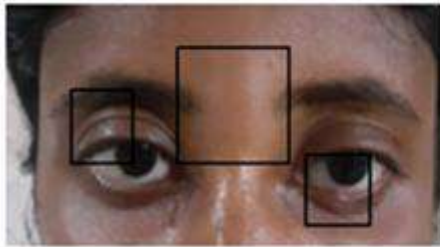


Unnati Desai et al.,

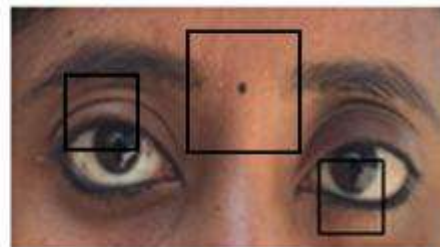
Treatment Plan

The main aim of treatment is to give relief to symptoms, avoid further complication and positive psychological approach to the patient. *Aschyotana* with *Yashtimadhu Ghrita* was done for 7 days as *Aatyayik chikitasa* (Emergency treatment) [5]

OBSERVATION & RESULTS



Before Management



after Management

Gradation: Table 3: Gradation of the symptoms

Results: Table. 4

DISCUSSION

Despite the fact that the lids, eyelashes, and orbital margins protect the eye, *Neeja* or *Agantuj* cause can injured the eyes in a variety of ways. The most feared outcome of the *Netrabhighata* is vision loss, which must be treated immediately. During *Abhighata*, the *Doshas Rakta* and *Pitta* are the most affected. *Abhighata* causes *Pitta* and *Rakta* to become vitiated, followed by *Vata* and *Kapha* to become vitiated, resulting in the formation of *Sopha*, *Puya*, *Bhedha* of *Twacha* and *Vrana* [6]. Therefore, *Tridosahara Pathyas*, *Snighdha*, *Hima* and *MadhuraDravyas* are recommended to combat the negative effects of vitiated *Rakta* and *Tridosha* while also soothing the eyes.

The probable mode of action of the medicines used in this case is as follows:

***Yashtimadhu Ghrita*:** It includes *Yashtimadhu* and *Ghrita*. Both *Yashtimadhu* and *Ghrita* contain *Madhura Rasa*, *Guru-Snigdha Guna*, *Madhura Vipaka* and *Sheeta Virya*. It calms the agitated *Vata* due to its *Madhura Rasa*, *Guru-Snigdha Guna* and *Madhura Vipaka*. It also soothes the irritated *pitta* with its *Madhura rasa*, *Madhura Vipaka* and *Sheeta Virya*. It has healing, skin regeneration, and anti-inflammatory properties. It also owns *Rasayana* and *Ropana* properties.

***Yashtimadhu*:** The antioxidant activity of *G. glabra* is one of the primary reasons for its use. The phenolic content is most likely responsible for the high antioxidant activity observed. The responsible compounds are primarily isoflavones, including glabridin, hispaglabridin A, and 30-hydroxy-4-O-methylglabridin. The dihydrostilbene derivatives found in *G. glabra* leaves have high antioxidant activity. *G. glabra* also contains licochalcones B and D, which have strong DPPH radical scavenging activity and can inhibit microsomal lipid peroxidation. These phenolic compounds are effective at protecting biological systems from oxidative stress and inhibiting the onset of skin damage. The topical application of liquorice extract formulations may be beneficial in innovative dermal and cosmetic products because it counteracts oxidative stress damage and maintains skin homeostasis due to its high antioxidant content. The antibacterial activity observed is due to the presence of secondary metabolites, namely, saponins, alkaloids, and flavonoids [7].

***Bidalaka*:** According to *Srotomaya Purusha*, the entire body is made up of *SukshmaSrotasa*, or it is porous. Through these pores or channels, minute particles of drug applied in the form of *Bidalaka* penetrate the skin. At this stage, the *UpshoshanaGuna* of *Vata Dosh* aids in drug penetration and absorption. *Bhrajaka Pitta*, which is present in the skin, is responsible for the metabolism of drugs applied to the skin [8,9]. The drugs used in *Bidalaka* promote wound healing.





Unnati Desai et al.,

It includes *Yashtimadhu*, *Amalaki*, *Haritaki*, *Daruharidra*, *Lodhra* which contains *Vrana Shodhana-Ropana* (woundhealing)Guna, *Shophaghna* (anti-inflammatory), *Indriyabalaprada* (immunomodulatory), *Raktaprasadana* (blood purification), and *Dahaprashamana* (reduce burning sensation), *Tridoshaghna* as well as *Rujahara* (analgesic), and *Kandughna* (anti-pruritus)Guna. *AcharyaCharaka* classifies *Lodhra* as *Sandhaniyagana Dravya* (wound-healing drugs). It includes *Kashaya-Tikta rasa*, *Laghu-Ruksha guna*, *Katu Vipaka* and *Sheeta Virya*. Thus, it balances *kapha* and *Pitta*. Ethanolic extract present in *Lodhra* is responsible for analgesics and anti-inflammatory activities [10].

BilvadiGutika Vati: Bilva-It includes *Laghu Guna*, *Sheeta Virya* and *Madhura Vipaka*. It contains *Mutrala* (diuretic), *Tridoshaghna*, *Shothahara* (anti-inflammatory), *Vedanasthapana* (analgesic), *Raktastambhan* (hemostatic), *Deepana* (appetiser), *Pachana* (digestive), and *Grahi* (astringent) properties. According to studies, the pulp of the *Bilva* fruit contains a variety of bioactive substances, including carotenoids, phenolics, alkaloids, pectins, tannins, coumarins, flavonoids, and terpenoids which are responsible for anti-inflammatory, anti-genotoxic, anti-bacterial, anti-fungal, and antioxidant properties. Chemical structures of the compounds present in *Bilva* are Citral (antibacterial, antifungal, and antiparasitic), Cumin aldehyde (Insecticide), Eugenol (antibacterial, analgesic, and antioxidant), rutin (Antioxidant, Anti-inflammatory), β sitosterol (Antioxidant). [11]. It has been found to have healing properties and is beneficial for blood purification.¹²Because it balances *Kapha* and *Vata*, it is effective in treating pain and swelling, as well as managing various inflammatory changes in the body. *Surasa* has anti-inflammatory, analgesic, and antipyretic properties, as well as immunoregulatory action. It also acts as an antihistamine, antibacterial, and has antitoxic properties.

Karanja, Haridra, Daruharidra- These medications include *Tikta-Katu Rasa*, *Ruksha-Laghu Guna*, *Katu Vipaka* and *Ushna Virya*. It balances *Vata* and *Kapha* due to *Ushna Virya*; *Ruksha-Laghu Guna* pacifies *Kapha*; and *Tikta Rasa* pacifies *Pitta*. Hence, it balances all three *Doshas*. It reduces skin lesions and effectively relieves the itching sensation associated with them. *Tagara* has *Katu-Tikta-Kashaya Rasa*, *Laghu-Snigdha Guna*, *Katu Vipaka* and *Ushna Virya*. It possesses *Kapha Vata Shamaka* and *Vishaghna* properties. *Haritaki* includes *Lavana Varjita Pancharasa*, *Laghu-Ruksha Guna*, *Madhura Vipaka* and *Ushna Virya*. It's called *Vatanulomaka* and it's primarily responsible for interrupting the progression of inflammation and pain. *Pippali* contains *Katu Rasa*, *Laghu-Tikshna Guna*, *Madhura Vipaka* and *Anushna-Sheeta Virya* all of which may act as a *Pitta-reducing* drug. *Sunthi* has *Katu Rasa*, *Guru-Ruksha-Tikshna Guna*, *Madhura Vipaka* and *Ushna Virya*, which calms *Kapha*. It is used to treat both acute and chronic inflammation, skin infections, and oxidative stress [13]. *BilvadiGutika* performs *Aam Pachana*, which helps to eliminate *Gara Visha* and *Agni Mandhya*.

Punarnavadi Guggulu: Punarnava contains *Madhura-Tikta-Kashaya Rasa*, *Laghu-RukshaGuna*, *Katu Vipaka* and *Ushna Virya*. It balances the *Kapha* and *VataDosha* and performs *Sothaghna* and *Rasayana* actions. It contains several bioactive chemical constituents, including punarnavine, isoflavonoids (rotenoids), sitosterol, an alkaloid (boeravinone), eupalitin, beta-sitosterol, and palmitic acid. [14]. These active chemical constituents are found in various parts of the plant and have pharmacological and therapeutic properties such as immunomodulation, anti-inflammatory, antioxidant, antifungal, anti-microbial, anti-histamine, and anti-stress [15].

Devdaru includes *Tikta-Katu-Kashaya Rasa*, *Ruksha-Laghu Guna*, *Katu Vipaka* and *UshnaVirya*. It contains *Vatahara* and *Sophaghna* properties. ***Haritaki*** includes *Shothahara*, *Deepana*, *Pachana* and *Shulahara* properties. ***Guduchi*** contains *Kashaya-Tikta Rasa*, *Laghu-Snigdha Guna*, *Madhura Vipaka* and *Ushna Virya*. *Guduchi* is *Tridoshaghna* and performs *Rasayana* and *Dhatvagni Vardhaka* actions. ***Guggulu*** includes *Tikta-Katu Rasa*, *Laghu*, *Ruksha*, *Vishada Guna*, *Sookshma*, *Sara Guna*, *Katu Vipaka* and *Ushna Virya*. It helps to resolve *Shotha Samprapti*.

Manjishthadi Kashaya: Manjishtha consists of *Guru-Ruksha Guna*, *Tikta-Kashaya-MadhuraRasa*, *Katu Vipaka* and *Ushna Virya*. It possesses *Kapha-Pitta-Shamaka* properties. Phenolic compounds were identified in *Manjishtha* through phytochemical testing. Phenol is an effective antiseptic for preventing infectious pathologies. Tannic acid solution acts as a chemical antidote, precipitating poisons such as alkaloids and metals. *Manjishtha* also contains carbohydrate, alkaloids, amino acids, saponin, glycosides, and tannins, making it a medicinal phytochemical with antitoxic, detoxifying, antiseptic, antimutagenic, anticarcinogenic, and antioxidant properties [16]. It has anti-inflammatory,





Unnati Desai et al.,

pain-relieving, and antimicrobial properties. *Triphala* has antimicrobial and anti-inflammatory properties. *Vacha* has anti-inflammatory, immunomodulatory, antibacterial, and anti-ulcer properties. *Nimb* has *Tikta-KashayaRasa*, *Laghu-Ruksha Guna*, *Katu Vipaka*, and *Sheeta Viryas*. It has antibacterial, anti-inflammatory, antifungal, antioxidative, and immunomodulatory properties. All of these medications have anti-inflammatory, anti-oxidant, wound healing, antibacterial, antifungal, and antimicrobial properties that help with early wound healing and prevent complications.

CONCLUSION

Every ocular structure is prone to injury. The site is frequently determined by the cause and mechanism. It can range from minor injuries, such as getting dust in your eyes, to major injuries that cause permanent vision loss. Because of its negative consequences, our *Acharyas* have given detailed explanations for *Nayanabhighata*. In this case, the *Chikitsa* is determined by the *Avastha* and *Dosha* predominance. Our *Acharyas* have explained various treatment modalities such as *Nasya*, *Alepa*, *Parisechana*, *Tarpana*, the concept of *Pathya*, *Drushtiprasada JananaDravyas*, and the use of *Snigdha*, *Hima*, and *Madhura Dravyas* for various types of *KeetaVisha*.¹⁷The signs & symptoms of the patient were improved after four days of treatment and was completely relieved within seven days. Based on the findings of this clinical case study, we can conclude that *VishaghnaAushadhipana* combined with *Vishaghna Lepa (Bidalaka)* can provide a reliable and quick result with no local or systemic side effects. Ocular therapeutic procedure in the form of "*Aschyotana*" also played major role in the primary and emergency management (*Aatyayik chikitsa*) towards the symptoms of ocular injury.

REFERENCES

1. *Sushruta Samhita of Ambikadatta Shastri, Ayurved-tattva-sandipika*, Chaukhambha Sanskrit Sansthan, Varanasi, reprint edition- 2013, *Uttaratantra*, Chapter 19/3, Page no. 104
2. *Acharya Charaka –Charaka Samhita of Agnivesa* by Bramhanand Tripathi, part-II published by Varanasi, Chaukhambha Orientalia, Reprint edition-2012, Chapter 23/140, Page No. 649
3. *Acharya Vriddha Vagbhata -Astanga Samgraha* By Kaviraja Atrideva Gupta, Chaukhambha Prakashan, Varanasi, Reprint edition 2016, Page No-363
4. <https://www.eyelink.com/what-to-do-if-a-mosquito-bites-your-eyelid-2021/>
5. Prakruthi G & Hamsaveni V: An Insight Into *Nayanabhighata* In *Ayurveda*. International *Ayurvedic Medical Journal* {Print} 2018
6. Dr. Apoorva MN, Dr. Hamsaveni V. Ocular injuries - A Review Article. *J Ayurveda Integr Med Sci* 2018;5:76-83. <http://dx.doi.org/10.21760/jaims.v3i5.13821>
7. Pastorino G, Cornara L, Soares S, Rodrigues F, Oliveira MBPP. Liquorice (*Glycyrrhiza glabra*): A phytochemical and pharmacological review. *Phytother Res.* 2018 Dec;32(12):2323-2339. doi: 10.1002/ptr.6178. Epub 2018 Aug 17. PMID: 30117204; PMCID: PMC7167772.
8. Rajput SS, Mishra S, Sharma R et.al. A literary review on role of *vidalaka* in eye disorders. *Int J Health Sci Res.* 2021; 11(6): 271-273
9. Prakruthi G & Hamsaveni V: An Insight Into *Nayanabhighata* In *Ayurveda*. International *Ayurvedic Medical Journal* {Print} 2018 {cited March, 2018}
10. https://www.researchgate.net/publication/277685641_Phytopharmacological_Profile_of_Symplocos_racemosa_A_Review
11. Monika S, Thirumal M, Kumar PR. Phytochemical and biological review of *Aegle marmelos* Linn. *Future Sci OA.* 2023 Mar 23;9(3):FSO849. doi: 10.2144/fsoa-2022-0068. PMID: 37026028; PMCID: PMC10072075.
12. K. M. Divya, Allam Ramakrishna, S. Balasundar and Govardhan Sahani (2023). A review on an ayurvedic medicine, *Vilwadi gutika* used in the management of toxicity. *Ann. Phytomed.*, 12(2):161-168. <http://dx.doi.org/10.54085/ap.2023.12.2.18>.





Unnati Desai et al.,

13. K. M. Divya, Allam Ramakrishna, S. Balasundar and Govardhan Sahani (2023). A review on an *ayurvedic* medicine, *Vilwadigutika* used in the management of toxicity. *Ann. Phytomed.*, 12(2):161-168. <http://dx.doi.org/10.54085/ap.2023.12.2.18>.
14. Bhowmik D, Kumar KS, Paswan S, Srivatava S, Yadav A, Dutta A. Traditional Indian herbs *Convolvulus pluricaulis* and its medicinal importance. *J Pharmacogn Phytochem.* 2012;1(1):44–51
15. Selvaraj D, Shanmughanandhan D, Sarma RK, Joseph JC, Srinivasan RV, Ramalingam S. DNA barcode ITS effectively distinguishes the medicinal plant *Boerhavia diffusa* from its adulterants. *Genomics Proteomics Bioinformatics.* 2012;10(6):364–7. doi: 10.1016/j.gpb.2012.03.002
16. <https://www.thepharmajournal.com/archives/2017/vol6issue7/PartC/6-5-10-535.pdf>
17. *Sushruta Samhita*, with Sri *Dalhanacharyateeka*, edited by Narayan Ram Acharya “Kavyathirtha”, Chaukhambha orientalia, Varanasi, reprint edition- 2009, *Uttaratantra* 18th chapter, pp-824, pg-640-641

Table 1: Ocular examination

	Right eye	Left eye
Eyelashes	NAD	NAD
Eyelids	Painful swelling ++	Painful swelling +++
Conjunctiva (Bulbar)	NAD	NAD
Conjunctiva (Palpebral)	Mild congestion +	Mild congestion +
Cornea	Clear	Clear
Sclera	NAD	NAD
Pupil	Round, Regular, Reactive to light	Round, Regular, Reactive to light
Lens	Transparent	Transparent
Visual Acuity	6/6	6/6 (p) with pinhole 6/6

Table 2: Treatment protocol

Name of procedure /Drug	Dosage	Route of administration	Duration
<i>Aschyotana</i> with <i>Yashtimadhu Ghrita</i>	10-10 drops in bilateral eyes	Topical	7 days
<i>Bidalaka</i> (<i>Yashtimadhu</i> , <i>Amalaki</i> , <i>Haritaki</i> , <i>Daruharidra</i> , <i>Lodhra</i>)	5 gm each	Local application	7 days
<i>Bilvadi Gutika</i>	2 tablets twice a day with luke warm water after meal	Oral	7 days
<i>Punarnavadi GugguluVati</i>	2 tablets thrice a day with luke warm water after meal	Oral	7 days
<i>Manjishthadi Kashaya</i>	15 ml Kashaya with 45 ml water Twice a day after meal	Oral	7 days

Table 3: Gradation of the symptoms

Symptoms	Grade
Severe	+++
Moderate	++
Mild	+
No symptoms	0





Unnati Desai et al.,

Table 4: Result

Sign & symptoms	Before treatment	Day 4	Day 7
Swelling over glabella	+++	++	0
Oedematous eyelids	++	+	0
Itching at periorbital area	++	0	0
Congestion on palpebral conjunctiva	+	0	0
Burning sensation in eyes	+++	+	0
Tenderness on touch	+++	++	0
Watering from bilateral eyes	+	0	0





Polyherbal Face Pack Development, Formulation Techniques and Evaluation Study

Rajesh Kumar Sharma^{1*}, Disha Singh² and Sachin Kumar³ and A. Elphine Prabahar⁴

¹Associate Professor, Department of Pharmacognosy, Teerthanker Mahaveer College of Pharmacy, Teerthanker Mahaveer University, Moradabad, Uttar Pradesh, India

²Bachelor of Pharmacy, Department of Pharmacy, Teerthanker Mahaveer College of Pharmacy, Teerthanker Mahaveer University, Moradabad, Uttar Pradesh, India

³Associate Professor, Department of Pharmacy, G.S.V.M. Medical College, Kanpur, (Affiliated to Atal Bihari Vajpayee Medical University, Lucknow), Uttar Pradesh, India

⁴Professor, Department of Pharmaceutical Chemistry, Chettinad School of Pharmaceutical Sciences Chettinad Hospital and Research Institute, Chettinad Academy of Research and Education, Kelambakkam, Tamil Nadu, India.

Received: 13 Jun 2024

Revised: 16 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

Rajesh Kumar Sharma

Associate Professor, Department of Pharmacognosy,
Teerthanker Mahaveer College of Pharmacy,
Teerthanker Mahaveer University,
Moradabad, Uttar Pradesh, India



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The aim of this research work is to formulate and evaluate polyherbal face pack for cosmetic purposes from herbal ingredients. Multani mitti (Fullers earth), sandalwood, orange peel, and almond peel are the medicinal plant materials used as traditionally from ancient years in herbal medicines such as Ayurveda, Homeopathy and Siddha. These ingredients either purchased from local market or from flipcart, sieved using #40 mesh under sieve number 120. Mixed well and kept in air tight container for further evaluation. Bulk density, tapped density, moisture content, total ash angle of repose, particle size, irritancy test were performed. Shinnoda test and organoleptic evaluation were also carried out. Out of three formulations F1 was found to be very effective.

Keywords: Face pack, Multani mitti, orange peel, sandalwood, evaluation.

INTRODUCTION

Each one wants to get fair and attractive skin. Now a day, pimples, dark circle Acne, black head are common among youths who suffers from it. As per the Ayurvedic system of medicine, problems of Skin are generally because of





Rajesh Kumar Sharma et al.,

impurities in blood. Accumulated pollutants in the blood during unhygienic food and lifestyle on daily basis can cause skin related diseases. Several herbs, remedies are designed in Ayurvedic system for blood cleansing. Herbs alike Chandana, Haldi, Manjistha, Lodhra, etc. are convenient example for blood purifier. The herbal pastes which are useful on facial skin to cure acne, pimple, scars, marks or pigments are known as “mukha lepa” in Ayurvedic system. The method of applying this herbal mixture on facial skin is termed as “mukha lepana”. This particular beautifying therapy is common as facial. The smoothing powder which is used in application on facial skin is termed as “face pack”. A decent herbal face pack must into supply of essential nutrients onto skin. It should enter the subcutaneous tissues in ordering of to deliver the essential nutrients. Dissimilar types of skin requires several types of herbal face pack [1]. A face pack is a powder that smoothes and nourishes the skin on the face. A good herbal face pack should also contain the nutrients that the skin needs to penetrate the subcutaneous tissues and reach the skin's surface. Ayurvedic face packs are supposed to help minimize wrinkles, acne, pimples, and dark bags under the eyes. Additionally, a face pack brightens and smoothes the skin [2].

Cosmetics are products used in skin and hair care regimens, primarily by young people, with the aim of improving, beautifying, and cleaning the qualities that make them beautiful. Skincare treatments are not popular right now. In actuality, cosmetics are used by people in every civilization to improve or protect their skin, which undoubtedly proves that this is a growing demand. The conventional idea of applying cosmetics to highlight the characteristics of improved health has not altered, despite the fact that cosmetics have undergone several alterations in more recent times [3]. The smoothing powder that is currently applied to facial skin is called a face pack. Using powders or pastes, these preparations of different combinations are applied to the face skin and allowed to dry and set to form a film that tightens, strengthens, and cleanses the skin. In order to get the resulting layer of face pack, which contracts and hardens and is easily removed, face packs are typically kept on the skin for fifteen to thirty minutes to allow all of the water to evaporate. Although the colloidal and adsorption clay combinations used in these processes remove excess oil and filth from the facial skin, the softening and tightening impact that follows the administration of a face pack gives the impression of a renewed face. When the face pack is eventually removed, the injured skin and accumulated debris are also eliminated [4].

A homemade face pack using only natural products and masks renders skin more luminous, smooth, and soft [5]. The development, description, and application of a natural herbal face pack that is pure cosmetic is the focus of this research article today. Normal skin seems lively and moisturized since it is neither exceptionally oily nor dehydrated. Cleansing, toning, and moisturizing are the steps in a routine skin care treatment. However, the herbs included in this recipe revitalize the skin to prevent it from becoming allergic and sunburned. Present research work was carried to frame herbal face pack comprising powders of Fullers earth(also known as multani mitti) , green tea, turmeric, almond, orange peel, sandalwood as the required ingredients which will accomplish as complete face pack .This prepared face pack was evaluated further by physical, chemical and biological evaluation [6].

Benefits of Applying Face Pack [7]

1. Helps in the reduction of acne, pimple, scars and marks depending on the herbal ingredients we use.
2. These face packs produces a gentle and relaxing effect on the facial skin.
3. Face packs mainly removes the dead cells of facial skin.
4. Fruit face packs provides vital nutrient to skin
5. On application of natural face masks on daily basis brings glow to skin, improve skin quality and skin tone.
6. These face packs advantages us to reappearance the lost glow and shine of skin in short period of time.
7. Face pack helps to avoid early aging of skin.
8. The use of face packs judicial can effectively opposed the injurious effects of pollution and severe climates on skin.
9. Natural face packs make the skin look beautiful and full of essential nutrition.
10. Development of wrinkles, fine lines and sagging of skin can be well prevented by using of natural face packs.





Rajesh Kumar Sharma et al.,

Precautions to be Taken While Applying Face [8].

1. The face pack should be removed within 15 to 20 minutes from face. Keeping face pack for extended time period may result in development of wrinkles, sagging of skin and increase of open pores.
2. Choose the face pack giving preference to your skin type. Take views of natural therapist or concerned skin professional before application of face pack.
3. Before removing of dried face pack, spray water(which should be at room temperature). After getting rid of the mask, rub an ice cube on facial skin. This would help us to close open pores and constricts the skin. will also tones and soothes the skin
4. Use of face pack once in a week. Never try to scratch or peel the dry face pack. This may harm original skin.
5. Prevent using face pack nearby of "eye zone". The skin nearby eye is very gentle. The process of washing face pack may harm skin around eyes.
6. Never rub face forcefully. This may result in outbreak of pimples and dark spots. When applied face pack stay away from heat.

MATERILAS AND METHODS

Plant Materials: The herbal materials utilized in this study were either bought from a flip cart (green tea, turmeric), or they were obtained at the local market (Multani mitti, Sandalwood, Orange peel, and Almond peel powder). The herbs were then dried and pulverized for later usage. The information about the plant materials used in the formulation of the face pack is provided below.

Multani Mitti (Calcium bentonite)

Calcium bentonite benefits skin in a variety of unique ways, including reducing the size of pores, getting rid of whiteheads and blackheads, fading freckles, calming sunburns, cleaning the skin, improving skin tone, improving blood circulation, treating acne, and giving skin a radiant appearance due to its high nutrient content. Multani mitti is a great remedy for bad and irritated skin and will repair your skin to make it glow. Magnesium chloride is abundant in Multani Mitti [9]. The word "fullers earth" comes from the initial procedure of using a fullers earth slurry mixed with water to clean or pull wool in order to get rid of dirt and oil. Because it improves blood circulation, relaxes and smoothes the skin, it is also a component of cosmetic goods. Because it is a pure and natural cleansing agent, it improves the skin's shine [10].

Turmeric (*Curcuma longa*)

Curcuma longa has anti- allergic and anti-inflammatory activity. It helps in wound healing and best blood cleanser. It provides best blood purification action hence it is used in almost disease having properties with blood impurities origin. Haridra is renew of skin and helps to make skin again youngby delaying the symbols of aging likewise wrinkles [11].Turmeric Powder, termed from *Curcuma longa*, is primarily used as an antibacterial, antiseptic, and anti-inflammatory properties. *Curcuma longa* also acts as a blood cleanser and well treats the acne by fighting against pimples and decreasing oil secretion from the sebaceous glands [12].

Sandalwood (*Santalum album*)

Santalum alba has an anti-aging and anti-tanning property. Sandalwood keeps the skin cool, fair and healthy by defending the skin from the effect of environmental pollutants. Sandalwood is helpful Ayurvedic herb with antimicrobial properties and is used for curing several skin problems and eliminates scars [13]. It is a herbal species obtained from plant in the family of Santalaceae of genus *Santalum* hence both commercially and ethnically valuable. The oil contained in the sandalwood wooden, primarily in the heartwood, is the important reason for its cultural and commercial rank. If your skin is greasy, you can use it to get rid of dark spots. Anti-ageing and anti-tanning values found in *Santalum album*. It is also beneficial for the skin in a several of ways composing of toning, soothing, antibacterial, calming and healing characteristics [14].

Orange peel (*Citrus aurantium*)

Orange peel powder, carried from *Citrus reticulata*, is a citrus fruit valued in nutrients like magnesium, calcium, potassium, and vitamin C. It shields the skin from oxidative stress, provides hydration, and reduces free radicals. Moreover, it imparts an instantaneous radiance to the skin, prevents wrinkles, blemishes, acnes, and signs of aging



**Rajesh Kumar Sharma et al.,**

[15]. On top of fighting free radicals, vitamin C brightens your skin and makes it younger and shining. Orange peel powder helps in including unclogging pores, removing blackheads and treating acne. It comprises citric acid, which remove dead skin cells [16].

Green tea powder

It belongs to family theaceae and used as anti-aging, anti-inflammatory and healthy glow for skin. Green tea is now a focus of interest as of its confirmed anti-oxidant properties or its capability to repair UV photo damage and toxicity due to light. This action of green tea is supposed to be because of catechins that are the most vital polyphenols in green tea [17]. Newly, the lotus *Nelumbo nucifera's* leaf, seed, and flower extract were examined for anti-wrinkle properties and presented 56%, 49%, and 54% inhibition of elastase [18].

Almond peel powder

Family: Rosaceae Use: Cleanser, scrub and moisturizer in an earlier examination, the *Prunus amygdalus* L. extract (skin) was used in herbal formulation and further studied for skin protection from premature aging of skin caused by UV radiations of sun. Particular antioxidant activity was revealed by the mice skin afterward treatment, as malondialdehyde (MDA) level was decreased, whereas level of Glutathione (GSH) was increased after treatment [19].

Methodology**1. Bulk density (B.D) [20]**

It is the proportion of total mass of powder upon the bulk volume of powder. It was quantified by sheeting down the weighed powder (passing through standard sieve #20) inside the measuring cylinder further the initial volume called the bulk volume, from here one can calculate bulk density according to formula given below: should be expressed in g/cc and given by.

$$B.D = M/V_0$$

Where, M=mass of the powder

V_0 = bulk volume of the powder.

2. Tapped density [21].

Tapped density is an increased bulk density which is accomplished just after mechanically tapping of the vessel containing the powdered sample. Next witnessing the initial powder volume or mass, the measuring cylinder or container tapped mechanically for 1 min further volume and mass readings are noted until little more mass change or volume change was noted down, hence expressed in grams per cubic centimetre.

Tapped density of formulation should not be more than 0.5 g/cc. Tapped density = Mass/Tapped volume.

3. Moisture content [22].

Take a thin ceramic dish and weigh around 1.5 g of the powder drug in it. Keep the dish in oven at 100 degree Celsius to get it dry, while waiting for two repeated weighings do not vary by additional than 0.5 mg. Cool and weigh the powder dish, further the loss in weight is typically noted as moisture content. Moisture content not more than 4% w/w.

LOD (Loss on Drying) can be calculated by using following formula.

$$\% \text{ LOD} = \frac{\text{Initial sample wt.} - \text{wt. of dried material}}{\text{total wt. of drug taken}} \times 100.$$

4. Angle of repose

The method used to calculate angle of repose was fixed funnel method. Powder was transferred over a funnel to form a cone shape structure. As the growing cone, the tip of the funnel must be held nearby and slowly raised as the pile grows so that to decrease the effect of falling particles. Further estimate the height and radius of the pile.

$$\theta = \tan^{-1} (h / r)$$

Where, θ = Angle of repose

h = Height of the heap

r = Radius of the base

5. Ash Value

Ash value is also termed as total ash. In the evaluation of pharmaceuticals and herbal drugs ash value is a key parameter. The particular known weight of sample was permitted to burn or destroy totally in a standard muffle furnace. The residue which is left in the form of ash comprises of salts, metals and other non-combustible materials termed as inorganic mineral components which was evaluated further.





Rajesh Kumar Sharma et al.,

a) Total Ash Value

The situation is not possible to know the drug identity or purity only from the ash content, but then again it can provide the visions into the quality of the sample. Accurately weigh 2g of sample in the crucible and further note the weight of it. Place the crucible for 24 hours in the standard muffle furnace set at 450 degree of Celsius than turned off the muffle furnace and allowed to cool it. Further remove the crucible from muffle furnace and weighed the as sample and note down as crucible plus ash sample (total weight).

Weight of Ash (mg) = (weight of crucible + Ash) – (weight of crucible)

Ash (%) = Weight of ash (mg)/ Initial sample weight × 100

b) Acid Insoluble Ash Value

Add 25 ml of HCL to the crucible containing total ash, and cover it with a watch glass. Then boil the mixture for 5 minutes. Further wash the watch glass with hot water (5ml) and added into the crucible. The ash-less filter paper was rinsed with hot water till it become neutral in which insoluble matter was collected. The filter paper comprising the insoluble matter was removed to the original crucible and desiccated on a hot plate. The sample with the crucible was burnt to a constant weight and left in the desiccator to get cool for 30 minutes. After cooling, the sample was weighed. The value or percentage of acid-insoluble ash was noted down in reference to the air-dried sample.

Acid Insoluble Ash Value = Weight of acid insoluble Ash/ Weight of air dried sample × 100

8. PH

An electrode pH meter used single standard or double electrode. By using proper buffer solution, instrument must be firstly calibrated at pH 7 and 9.2. The test sample consisting of 10 percent (m/v) dispersion of the product of either type of in previously boiled and cooled water shall be poured into a glass beaker and pH determined directly without any dilution within 5-10 minutes. Digital pH meter was used to measure the pH.

9. Particle size

Particle size is a general property of a powder. That affect several properties likely grittiness, spread ability etc. particle size was known using sieving method by I.P. Standard sieves through mechanical shaking for 10 min.

10. Shinoda test

Firstly prepare the ethanolic extract of using your sample powder than add few drops of concentrated hydrochloric acid (HCL) to it. At that point add magnesium turnings into the solution and appearance of pink, red colour tells that flavonoids are present in the sample powder.

Formulation of Herbal Face Pack

The herbal face pack was framed using basic mixing process. Herbal face pack was prepared or formulated by addition of all the essential amounts of herbal ingredients as given in formulation table 1.

Method of preparation [23].

The crushed constituents used for herbal face pack were sieved using #40 mesh under sieve number 120



Ingredients were weighed precisely and mixed geometrically for even mixing.

This was then kept in an air close-fitting container for evaluation

Application procedure for face pack

Take required face pack powder in a container as per the necessity and add rose water to mix. Mix well and spread over the facial skin. Cover properly the acne and marks, spots too. As kept it for whole drying for 10 to 15 min and then cleanse with cold water

Evaluation parameters [24].





Rajesh Kumar Sharma *et al.*,

Organoleptic Properties: The dried powder nature, color, odour, taste and texture were tested manually in combined form.

Physicochemical Evaluation: pH was determined by performing under PH meter, total ash and acid insoluble ash was completed by using incinerator and moisture content was also executed.

General powder Characteristics: By using standard sieving method the particle size of combined dried powder was perform. Angle of repose by method of funnel, bulk density and tape density was evaluated by the flow property of the dried powder of combined form.

Shinoda Test: To the extract prepared of ethanol add little magnesium turnings and drops of concentrated HCL. The arrival of red or pink colour specifies the presence of flavonoids

RESULTS AND DISCUSSION

Organoleptic evaluation Face pack was formed and estimated for organoleptic parameters shown in the Table 2. The free flowing properties were shown by flow property parameter. The formation of colour was minor yellow. The fragrance of prepared formulations was good satisfactory which is required as cosmetic preparations. The smoothness and Texture and Smoothness was also good acceptable which is desirable as cosmetic formulations

Physical evaluation

The particle size was tested by standard sieving method. The angle of repose using funnel method, bulk density and tapped density by tapping method was evaluated by flow property of the dried powder of combined form [25].

Irritancy test

Mark a surface of area of (1sq.cm) on the leftward dorsal surface. Fixed quantities of ready face packs were applied to the particular area and time was noted under observation. Reported for up to 24 hrs. for regular intervals if any sort of Irritancy, erythema, oedema, was patterned [26]. This test was carried out in 3 males and 3 females volunteers and result was noted which is give in table no 6. The formulation presented no irritation, redness, edema not any Inflammation through irritancy studies. This formulation is harmless and safe to usage for skin.

DISCUSSION

Herbal face packs are uses casually to enhance blood circulation, renew the muscles and help the skin to balance its elasticity and get rid of the dirt from skin pores. The front foot benefit of herbal cosmetics and goods are their non-poisonous nature, reduce the hypersensitive reactions and timely tested utility of many ingredients. The formulation was found similar, easily rinsing out and had very slightly alkaline pH that were well-matched with normal skin physiology. Angle of repose is typical related to resistance to the movement between particles or inter particulate friction. As per in terms and limit of Indian Pharmacopoeia the angle of repose the flow property has been classified. The outcomes of all these parameters shown that the dried powder of combined form possess good packing ability and decent flow properties. Therefore, it revealed better flow properties for formulation to attain clean, fresh and soft formulation.

CONCLUSION

By using herbal preparations the existing skin conditions such as pimples, acne can be well treated. There are numerous herbal preparations as such herbal face pack, herbal face mask, face scrub act by several mechanism and supports to keep the skin healthy and nutrient. The existing formulations were formed by using several herbs which will help in recover fairness, diminish acne, by acting on face. The formulation was exposed to various evaluation parameters such as angle of repose, irritancy test and all the test was agreed by the formulation. Hence, polyherbal formulations have a widespread variety of choice in preventing skin problems. In the current effort, it is a great attempt to formulate the herbal face pack comprising naturally existing constituents like multani mitti, turmeric, sandalwood etc. It is suggested that the prepared formulation F1 was physico-chemically and microbiologically





Rajesh Kumar Sharma et al.,

stable and possessed characteristics of a standard cosmeceutical's formulation for skincare. Further optimization studies are required on this study to find the useful benefits of face packs on human use as cosmetic product.

REFERENCES

1. R. and Hiremanth, Text book of Industrial pharmacy, Drug delivery systems & Cosmetics & Herbal drug technology: Universities press (India) Ltd; 2nd Edition, (2002)
2. Mithal, B.M. and Saha, R.N., A Hand book of cosmetics: MK Jain, 2nd Edition, (2004).
3. <http://shodhganga.inflibnet.ac.in/bitstream.pdf>.
4. Rieger MM. Harry's Cosmeticology. In: Chapter 23, Face, Body& Hair Masks & Scrubs. 8th ed. vol I. New York: Chemical Publishing Co., Inc.; 2009. p. 471.
5. Millikan, L. E., 2001. Cosmetology, cosmetics, cosmeceuticals: definitions and regulations. Clinics in dermatology 19 (4): 371-374
6. D. K. Sanghi, Rakesh Tiwle. Formulation and Charaterization of Herbal Face Wash/Scrubber. European Journal of Pharmaceutical and Medical Research 2016; 3(11): 274-278.
7. Chanchal D. and Saraf S. (2009). Herbal Photoprotective Formulations and their Evaluation. The Open Nat Prod Journal 2: 71-76
8. Chanchal D. and Saraf S. (2009). Herbal Photoprotective Formulations and their Evaluation. The Open Nat Prod Journal 2: 71-76
9. Hwang JK, Shim JS, Gwon SH, Kwon YY, Oh HI et al. Novel use of Panduratin derivatives or extract of Kaempferia pandurata comprising the same. U.S. Patent 0065272A1, 2012 [cited 2016 Aug 05].
10. C.K. kotate, Purohit A.P., Gokhale SB. Text book of pharamacognosy. Thirty six edition, 2006, nirali prakashan pune, pp563.
11. Kumar. K., Sasikanth, K., Sabareesh, M. and Dorababu, N. (2011). Formulation and Evaluation of Diacerein Cream. Asian J Pharm Clin Res 4(2): 9398.
12. Himaja N, Ashok kumar A, Bhart kumar B. Preparation and Evaluation of Poly Herbal Fruit Face Mask. J Res Pharm Sci 2015; 2(11): 07-13
13. Nemade CT, Baste N. Formulation and evaluation of a herbal facial scrub. World J Pharm Res 2014; 3(3): 4367-4371
14. Subasinghe U, Sandalwood Research: A Global Perspective, Journal of Tropical Forestry and Environment, 2013; 3(1): 1-8.
15. Koli DS, Mane AN, Kumbhar VM, Shaha KS. Formulation & Evaluation of Herbal Anti-Acne Face Wash. World J Pharm Pharm Sci 2016; 5(6): 2001-2007
16. Dureja H., Kaushik D., Gupta M., Kumar V., Lather V. Cosmeceuticals: An emerging concept. Ind J of Pharmacol, 2005; 37(3):155-15.
17. Mahmood T, Akhtar N, Khan BA, Shoaib Khan HM, Saeed T. Changes in skin mechanical properties after long-term application of cream containing green tea extract. Aging Clin Exp Res 2011; 23:333-336.
18. Kim T. Kim HJ, Cho SK, Kang WY, Baek H, Jeon HY, Kim B, Kim D. Nelumbo nucifera extracts as whitening and antiwrinkle cosmetic agent. Korean J Chem Eng 2011; 28:424- 427
19. M.K. Sachdeva, T. Katyal. (2011). Abatement of detrimental effects of photoaging by Prunus amygdalus skin extract. Int. J. Curr. Pharm. Res. 3(1): 57-59.
20. N. Aghel, B. Moghimipour, R.A. Danvall. Iranian Journal of Pharmaceutical Research. 2007; 6(3): 127-172.
21. Kokate C. K., Practical Pharmacognosy, Vallabh Prakashan, 2008; 4: 94.
22. Bhutkar K, Shah M, Formulation and Evolution of Herbal Antibacterial Face Pack, Journal of Emerging Technologies and Innovative Research, 2019; 6(5): 77-82.
23. Anand Jigisha, Rai Nishant, Kumar Navin, Guatam Pankaj. Green Tea A Magical Herb With Miracuious Outcomes. International Research Journal of Pharmacy, 2012; 3(5): 139-148.
24. Ashawat M, Banchhor M, Saraf S, Saraf S. Herbal Cosmetics:" Trends in Skin Care Formulation". Pharmacognosy Reviews. 2009;3(5):82.





Rajesh Kumar Sharma et al.,

25. Banchhor M, Ashawat MS, Saraf S, Saraf S. Herbal Cosmetics: Trends in Skin Care Formulation. *Pharmacogn Reviews* 2009; 3(5): 82-89.
26. Mandeep S, Shalini S, Sukhbir LK, Ram KS, Rajendra J. Preparation and Evaluation of Herbal Cosmetic Cream. *Pharmacologyonline* 2011; 1258-1264.

Table 1 Formulation of herbal face pack (Quantity of sample for 100gm)

S.No.	Name of Ingredients	F1	F2	F3
1	<i>Curcuma longa</i> powder	7.5g	10g	8g
2	Multani mitti powder	11g	13g	10g
3	<i>Santalum album</i> powder	6g	9g	11g
4	<i>Citrus aurantium</i> powder	8g	8g	7.5g
5	Green tea powder	4.5g	4.5g	6g
6	Almond peel powder	3g	3g	3g

Table 2: Organoleptic properties

S. No.	Formulation	Physical appearance	Odor	Taste	Texture
1	F1	Brownish	Slight	Characteristic	Fine
2	F2	Brownish	Slight	Characteristic	Fine
3	F3	Brownish	Slight	Characteristic	Fine

Table 3: Physiochemical parameters

S.No.	Formulation	Moisture content (%)	Bulk density(g/ml)	Tapped density(g/ml)	Angle of repose (Degree)
1	F1	4.5	0.55	0.83	18.93
2	F2	4.5	0.53	0.77	13.33
3	F3	4.6	0.47	0.76	13.22

*Results were the average of three readings

Table 4: General powder characteristic

S No.	Formulation	Total ash value	Acid insoluble ash	PH	Particle Size(μ m)
1	F1	36.5	0.67	6.65	20-25
2	F2	40	0.72	7.61	25-30
3	F3	36.05	0.64	6.69	30-40

*Results were the average of three readings

Table No. 5: Result of formulations for shinoda test (chemical test)

S. No.	Formulations	Observations	Result
1	F1	Pink color obtained	Flavonoids present
2	F2	Red color obtained	Flavonoids present
3	F3	Pink color obtained	Flavonoids present



**Rajesh Kumar Sharma et al.,****Table 6 Irritancy test****

S. No.	Formulation (F1)	Irritation	Erythema	Edema
1	W1	No Irritation	No Erythema	No Edema
2	W2	No Irritation	No Erythema	No Edema
3	W3	No Irritation	No Erythema	No Edema
4	M1	No Irritation	No Erythema	No Edema
5	M2	No Irritation	No Erythema	No Edema
6	M3	No Irritation	No Erythema	No Edema

*W= Women, M= Men

**Irritancy test was conducted on all 3 formulations but here results are shown for best suited formulation F1

Nature of face after wash= Soft and fresh, clean from dirt



Comparative Analysis of Classifier Performance: A Study of Weka and Python Implementations across Various Machine Learning Models

Mini Agarwal^{1*} and Bharat Bhushan Agarwal²

¹Research Scholar, Department of Computer Science Engineering, IFTM University, Moradabad, Uttar Pradesh, India

²Professor, Department of Computer Science Engineering, IFTM University, Moradabad, Uttar Pradesh, India

Received: 03 Jun 2024

Revised: 13 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Mini Agarwal

Research Scholar, Department of Computer Science Engineering,
IFTM University, Moradabad,
Uttar Pradesh, India

E. Mail: miniagarwal21@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The primary goal of engineering colleges is to deliver high-quality education, a challenge intensified by the post-COVID transition from online to offline learning. This study aims to predict student performance to provide early interventions for at-risk students. The research has three main objectives: identifying factors influencing student performance, comparing the accuracy of various data mining and AI algorithms in Weka and Python, and determining the most effective classifier. Data from 689 students were analyzed using metrics such as accuracy, F1 score, recall, precision, ROC curve, and confusion matrix. Our findings indicate that background characteristics (family details, parent's education, income, employment status, and student gender), educational attributes (academic performance, attendance, and assignment scores), and psychological traits (mental and physical health) significantly impact performance. Among the classifiers tested, the Random Forest algorithm showed the 100% highest accuracy in both Weka and Python environments.

Keywords: Education, Algorithm, Artificial Intelligence, COVID.

INTRODUCTION

The COVID-19 pandemic brought unprecedented changes to the educational landscape, necessitating a rapid shift to online learning. During this period, many students excelled in online examinations, often achieving perfect scores as schools and colleges adapted to virtual teaching methods. However, with the return to in-person classes, a decline in student performance on offline exams became evident, revealing gaps in the depth of learning that occurred in the online format. To address these challenges and enhance the effectiveness of student learning and performance





Mini Agarwal and Bharat Bhushan Agarwal

tracking, we propose leveraging data mining and artificial intelligence (AI) algorithms. Data mining involves extracting valuable insights from large datasets and has become an essential tool for data analysis across various industries[1]. In the context of education, particularly for first-year engineering and science students, these techniques can play a critical role in supporting academic achievement. Early academic performance is crucial as it significantly impacts students' cumulative GPA and overall educational trajectory[2]. Predicting student performance through mid-term and final exams can offer educators early insights, enabling proactive interventions for students at risk of poor academic outcomes[3]. Data mining techniques, especially classification and clustering, are vital in this predictive process. Classification sorts data into predefined groups, while clustering groups similar data sets based on shared characteristics. These methods allow educational institutions to analyze various attributes that influence student performance. Our research aims to gather and analyze the perspectives of key stakeholders—students, professors, teachers, directors, and industry professionals—through a comprehensive questionnaire distributed via Google Forms. By examining the responses, we aim to identify critical factors and their relationships in predicting student performance. This study seeks to answer the following questions:

1. How do various factors influence the student performance prediction?
2. Which Integrated Development Environment (IDE) yields the most accurate results, & what are the differences in accuracy among them?
3. Which classifier is the most effective in predicting student performance?

Through this research, we aim to provide valuable insights that can inform strategies to enhance student learning outcomes and overall academic success.

Related Work

Over the past decade, numerous studies have focused on predicting student performance using various data mining and AI algorithms. Here, we review key contributions in this area, highlighting the methodologies and accuracies reported.

- **V. Ramesh et al.**(2013)identified significant predictive variables and developed an algorithm for grading higher secondary students using decision trees. Their model achieved an accuracy of 85% in predicting student grades [4].
- **Elakia et al.** (2014) employed various decision tree algorithms, including ID3, C4.5, and CHAID, to suggest career options for high school students and monitor their behavior. Their findings indicated that the ID3 algorithm produced the most accurate results, achieving an accuracy range of 90-98% [5].
- **C. Anuradha et al.** (2015) explored the prediction of students' end-term results using multiple algorithms such as C4.5, KNN, Bayesian Network, OneR, and jRip. Among these, the jRip algorithm demonstrated the highest accuracy, with a 60% success rate in predicting end-term results [6].
- **Amjad Abu Saa(2016)** constructed a prediction model based on personal, family, and social factors using the Naive Bayes algorithm. This study highlighted the holistic approach to attributes and achieved an accuracy of 78% [7].
- **Ihsan A. Abu Amra et al.** (2017) utilized KNN and Naïve Bayes algorithms to assist the Ministry of Education in classifying students with degraded performance. The study revealed that the KNN algorithm provided the best results, achieving an accuracy of 93.6% [8].
- **Atta-Ur-Rahman et al.**(2018) developed a model focusing on student interest and feasibility using logistic regression. Their findings underscored the importance of student engagement, with the model achieving an accuracy of 80% [9].
- **Shubhangi Urkude and Kshitij Gupta (2019)**analyzed graduation rates and course completion rates using the Support Vector Machine (SVM) algorithm. The SVM model provided the best results, achieving an accuracy of 82% [10].
- **Vairachilai S et al.**(2020) identified various dependent and independent factors affecting student performance and applied multiple data mining algorithms, including Naive Bayes, Decision Trees, and K-Nearest Neighbors (KNN). The Naive Bayes algorithm emerged as the most effective, with an accuracy of 83% in predicting grades [3].



**Mini Agarwal and Bharat Bhushan Agarwal**

- **Raza Hasan et al.(2020)** explored various e-learning applications and applied different algorithms, including the genetic algorithm for feature selection. Their study found that the Random Forest algorithm yielded the best results, with an accuracy of 85% in predicting student performance [11].
- **J. Dhilipan et al.(2021)** aimed to recognize final grades and improve academic performance through the evaluation of four algorithms: Decision Trees, Naive Bayes, KNN, and Binomial Logistic Regression. Binomial logistic regression provided the best predictive accuracy at 79% [12].
- **Ahajjam Tarika, Haidar Aissab, and Farhaoui Yousef(2021)**evaluated various algorithms and identified the Random Forest algorithm as the superior method for predicting student performance, achieving an accuracy of 88% [13].
- **Yahia Baashar et al. (2022)** employed Artificial Neural Networks (ANN) to address theoretical gaps in predicting student performance, demonstrating the potential for improved accuracy and reliability in educational predictions [14].
- **Nitin Ramrao Yadav et al. (2023)** conducted an analysis of various machine learning and data mining algorithms, including ANN, SVM, Naive Bayes (NB), Linear Regression (LR), and Decision Tree, to evaluate their effectiveness in predicting student performance. This comprehensive study provided insights into the strengths and weaknesses of each algorithm in the educational context [15].
- **Zhaoyu Shou et al. (2024)** utilized Long Short-Term Memory (LSTM) networks to predict student performance using data on learning behaviors, scores, and demographics. This approach considered the temporal interactions of these factors, providing a dynamic perspective on how they influence academic outcomes over time [16].

These studies collectively demonstrate the evolving landscape of student performance prediction. They highlight the effectiveness of various data mining algorithms and the importance of considering a wide range of attributes, from personal and social factors to student interests and e-learning application usage. The consistent identification of algorithms like Naive Bayes, SVM, and Random Forest, Artificial Intelligence, KNN, Decision Tree as top performers across different studies underscores their robustness and reliability in educational data mining. This literature survey provides a foundation for our research, which aims to further explore the predictive power of these algorithms and identify key factors influencing student performance. By analyzing stakeholder perspectives and employing advanced data mining techniques, we seek to enhance the accuracy and applicability of student performance prediction models.

Attribute Selection

Through an extensive literature review and brainstorming sessions with various stakeholders—including directors, department heads, professors, students, and engineers—we identified several factors influencing student performance. These factors were gathered through a comprehensive questionnaire designed to collect detailed information on students' personal, social, psychological, parental, school, and college backgrounds. The questionnaire, built using Google Forms, was distributed to and completed by 300 stakeholders. The factors identified through these discussions and responses are summarized in Table I. These attributes are categorized into three main sections background information, educational information, and psychological information.

METHODOLOGY

Figure I illustrates the five stages involved in identifying the most accurate predictive results. Each section elaborates on the stages and observations.

Data Collection

After selecting relevant attributes, we gathered data from 689 B.Tech students from various streams at IP University. The dataset includes values corresponding to the chosen attributes, with student identities anonymized and labelled with unique codes.





Mini Agarwal and Bharat Bhushan Agarwal

Data Pre-processing

In this stage, we pre-processed the data to remove anomalies, missing entries, duplicates, and incorrect values. After cleansing the data, we processed it according to the criteria outlined in Table 1, classifying it into appropriate ranges.

Statistical Analysis

This stage involved identifying patterns, trends, and extracting useful information from the dataset. No outliers were detected during the analysis. We applied the Extra Tree Classifier to determine feature ranking. Table II displays the feature selection ranking, where higher values indicate more significant attributes. Figure II illustrates the feature ranking graphically. Additionally, we calculated the p-value for each element, which resulted in a significance level of 0.01.

Model Implementation & Evaluation

In our study, we implemented various machine learning models including Decision Tree (DT), Naive Bayes (NB), Logistic Regression (LR), Support Vector Machine (SVM), Random Forest, and K-Nearest Neighbors (KNN). For all models, we used a consistent split ratio for training and testing, with 70% of the data allocated for training and 30% for testing. We implemented the Decision Tree classifier using the entropy criterion for information gain, and allowed the tree to expand up to three levels. Random Forest classifier was implemented with 100 estimators, providing a robust ensemble learning approach to improve predictive accuracy. The SVM classifier was implemented with three different kernel types: linear, sigmoid, and polynomial. Each kernel was evaluated to determine the most effective for our dataset. We used the Gaussian Naive Bayes classifier, which assumes that the features follow a normal distribution. The KNN classifier was implemented with a specified number of neighbors (N estimators) to determine the optimal balance between bias and variance. In predictive modelling, various algorithms are used for predicting student performance. The most important and popular seven supervised algorithms implemented on the dataset are Decision Tree, Naive Bayes, Support vector Machine, Artificial Neural network, K-nearest neighbor, Random forest, and logistic Regression in Weka and Python. For evaluating the performance of these algorithms we generate the confusion matrix. Confusion matrix is a table that have two dimensions one is predicted and another one is actual. Both the dimension have four tuples i.e. true positive (TP) in this prediction model predict all the positive values of predicted and actual class are predicted correct or true, true negative (TN) in this prediction model predict all the negative values of predicted class and actual class are predicted negative, false positive (FP) in this prediction model predict all the negative values of actual and predicted class are predicted positive and false negative (FN) in this prediction model predict all the negative values of actual and predicted class are predicted positive and different metrics (accuracy, F-score, recall, precision, roc curve) of each algorithm that are described below:-

Accuracy: -it tests the data and gives the accurate predicted percentage of the predicted model.

$$\text{Accuracy} = \frac{TP + TN}{TP + FP + FN + TN}$$

Precision: - It calculates the actual positive values from the overall positive values that are predicted by model.

$$\text{Precision} = \frac{TP}{TP + FP}$$

Recall: - It calculates positive values that is correctly calculated by prediction model.

$$\text{Recall} = \frac{TP}{TP + FN}$$

F-score: - it is calculated harmonic mean of precision and recall. 1 represent the best value and 0 worst value.

$$\text{F1-score} = \frac{2 * (\text{precision} * \text{recall})}{(\text{precision} + \text{recall})}$$

Decision Tree

Decision trees are a fundamental supervised learning algorithm. They operate using a tree-like structure where two key node types play a crucial role. Decision Nodes act as checkpoints, evaluating a specific feature of the data. Based on this valuation, the data is directed down different branches of the tree. Leaf Nodes represent the final outcome, indicating the predicted class or value for a given data point. The purity of each node, signifying how well-separated the data points are within a class, is measured using the Gini impurity metric. A perfectly pure node (Gini value of zero) contains only data points from a single class. Conversely, a higher Gini value indicates a more diverse mix of



**Mini Agarwal and Bharat Bhushan Agarwal**

classes within the node. Decision trees are powerful tools for various tasks. Their effectiveness can be assessed using metrics like precision and F-measure. The decision trees in Weka and Python are shown in Fig 3 and 4. Table III shows the confusion matrix of the decision tree in this result of f-measure and precision are higher for A-class in comparison to B-class. Table IV shows the model evaluation in Weka and Python. Weka gives 90% accuracy and Python gives 96% accuracy on the data set in comparison to Weka Python gives the best result. Fig V shows the ROC curve.

Naive Bayes

It is a probabilistic classifier based on Bayes' theorem, also known as Bayes' Rule or Bayes' Law. It calculates the probability of a hypothesis given prior knowledge and evidence. The theorem utilizes conditional probability to determine the posterior probability of a hypothesis given observed data. In this context, Table V presents the confusion matrix of a decision tree classifier, indicating higher f-measure and precision for class A compared to class B. Furthermore, Table VI illustrates the model evaluation results obtained using Weka and Python. Weka yielded an 84% accuracy rate, while Python achieved a 94% accuracy rate on the dataset. Python outperformed Weka, demonstrating superior performance in this evaluation. Fig VI shows the ROC curve

Support Vector Machine

This technique belongs to the realm of supervised machine learning. It aims to minimize error by establishing a hyperplane. SVM, or Support Vector Machine, combines three key components: support vectors, hyperplanes, and margins. Support vectors are data points situated close to the hyperplane, while the hyperplane itself segregates data points into distinct classes. Margins denote the separation between the hyperplanes that closely border different classes.

A radial basis function is employed in the kernel of this SVM approach. Interestingly, both Weka and Python yield identical accuracy rates of 84%. Table VII showcases the model evaluation conducted using the hold-out method in both software environments. Meanwhile, Table VIII displays the confusion matrix, revealing that the precision achieved in class B surpasses that of class A. Notably, among Naive Bayes and Decision Tree techniques, SVM stands out as the method attaining the highest precision specifically in class B. Fig VII shows the ROC curve

K-nearest neighbor

This algorithm operates within the domain of supervised learning. It retains a repository of all provided data and utilizes this dataset to categorize new data or records based on their resemblance to existing data points. This process ensures that new data is appropriately classified into the correct category. Initially, a user specifies the number of neighbors, denoted as 'k'. The algorithm then calculates the Euclidean distance between the new data point and each existing data point, selecting the 'k' nearest neighbors based on these distances. Subsequently, it tallies the data points belonging to each category and assigns the new data point to the category with the most similar values. The confusion matrix, depicted in Table IX, reveals that both precision and f-measure attain a value of 1, denoting perfect accuracy. Notably, this algorithm outperformed its Python counterpart when executed in Weka. Furthermore, Table X illustrates the model evaluation results in both Weka and Python, showcasing a remarkable 100% accuracy rate. Noteworthy, the parameter 'n_neighbors=19' is passed to the KNN classifier, indicating the number of neighbors considered during classification. Fig VIII shows the ROC curve

Random Forest Classifier

This classifier operates by constructing decision trees from different subsets of the dataset, subsequently aggregating predictions through a majority voting mechanism to determine the final classification. Table XII illustrates the model computation conducted by the classifier in both Weka and Python environments, showcasing identical and optimal results with a 100% accuracy rate. Meanwhile, Table XI displays the confusion matrix, wherein both classes A and B exhibit equivalent precision and F-measure values. Notably, the confusion matrices of both the KNN classifier and the random forest classifier yield identical outputs. Fig IX shows the ROC curve



**Mini Agarwal and Bharat Bhushan Agarwal****Logistic Regression**

This model predicts both probabilistic and categorical values. Probabilistic values are outputs ranging between 0 and 1, while categorical values are either 0 or 1, or represented as "no" and "yes". Table XIV presents the model evaluation results in both Weka and Python environments, where Weka achieves superior accuracy of 90% compared to Python. Table XIII depicts the confusion matrix, revealing identical precision and F-measure values for both classes. Fig X shows the ROC curve

Artificial Neural Network

The neural network, often referred to as a feed-forward network, comprises multiple layers through which input vectors traverse. Initially, the input vector enters the input layer, where a function is applied to it, and subsequently, this output is propagated through all subsequent layers. Each layer is assigned weights, which are iteratively adjusted during training to optimize the output of the problem towards the output layer. Table XV illustrates the confusion matrix, indicating that class B exhibits a higher precision value compared to class A. In Table XVI, Weka outperforms Python with an accuracy of 97%. Fig XI shows the ROC curve. In Weka KNN and Random Forest classifiers predict the best 100% accuracy and MLP also predict the best 97% accuracy. In Python Random Forest predict the best 100% accuracy and Decision Tree, Naive Bayes, KNN also predict the best accuracy above 94% shown in Fig XII.

CONCLUSION

The prediction of student performance is critical for optimizing educational processes for both students and teachers. Our study employed feature ranking through a dedicated algorithm, alongside descriptive statistics and p-value calculations for each feature to identify the most significant predictors. Objective 1 is the Influence of various factors on student performance prediction in this we categorized student data into three primary groups: background characteristics, educational attributes, and psychological traits. Background characteristics included variables such as family details (number of siblings, parents' education, income, and employment status) and student gender, which significantly influenced model performance, particularly for high-achieving students who benefited most from additional support. Educational attributes, including past academic performance (grades from 10th, 12th, and the first year of B.Tech), attendance, and assignment scores, were found to have the strongest impact on student outcomes predicted by the ensemble model. Psychological attributes, including mental and physical health, also played a critical role as better health correlated with higher academic achievement. By integrating these factors, our ensemble model effectively categorized students within a smart education system, enabling targeted support and improved learning outcomes.

Objective 2 is the comparison of integrated development environments (IDEs) for accuracy in this we evaluated which Integrated Development Environment (IDE) provided the most accurate results. The classifiers tested showed minimal differences in accuracy, precision, F-score, and recall values, with only a 2-3% variance, as illustrated in Figures XII. Objective 3: effectiveness of classifiers in predicting student performance in these various classifiers, including Decision Tree, Multi-Layer Perceptron (MLP), Naive Bayes, Logistic Regression, Support Vector Machine (SVM), Random Forest, and K-Nearest Neighbors (KNN), were utilized for prediction. In Python, the Random Forest algorithm achieved 100% accuracy. Similarly, in Weka, both Random Forest and KNN predicted outcomes with 100% accuracy. Consequently, the Random Forest classifier emerged as the most effective algorithm among all tested classifiers.

Limitations and Future Work

The primary limitation of this research is the relatively small dataset, consisting of only 689 students. Additionally, the study did not account for factors such as students' social interactions, academic engagement, and interpersonal skills, which can also influence academic performance. Future research should aim to incorporate a larger dataset, include additional attributes or factors, and apply more algorithms to achieve the highest possible accuracy.



**Mini Agarwal and Bharat Bhushan Agarwal****REFERENCES**

1. K. M. Osei Bryson. "Towards Supporting Expert Evaluation of Clustering Results Using a Data Mining Process Model." *Information Sciences* 180.3 (2010): pp. 414-431.
2. Farzana Afrin, Mohammad Saiedur Rahaman, Margaret Hamilton, ' Mining Student Responses to Infer Student Satisfaction Predictors 'Seventh International Conference on Learning and Teaching in Computing and Engineering (LaTiCE'20), June 2020
3. VairachilaiS ,Vamshidharreddy,Avvari Sai Saketh, Gnanajeyaraman R ,Student's Academic Performance Prediction Using Machine Learning Approach, *International Journal of Advanced Science and Technology* 29(09) (2020) 6731-6737.
4. V.Ramesh, P.Parkavi, K.Ramar, Predicting Student Performance: A Statistical and Data Mining Approach, *International Journal of Computer Applications* 63(8)(2013).
5. G. Elakia, N. J. Aarthi, Application of data mining in educational database for predicting behavioural patterns of the students, *Elakia et al.,(IJCSIT) International Journal of Computer Science and Information Technologies* 5 (3) (2014) 4649-4652.
6. Anuradha, C. and Velmurugan, T. A Comparative Analysis on the Evaluation of Classification Algorithms in the Prediction of Students Performance. *Indian Journal of Science and Technology*, vol. 8, no. 15, pp. 1-12, 2015.
7. Amjad Abu Saa, Educational Data Mining & Students' Performance Prediction, *International Journal of Advanced Computer Science and Applications* 7(5)(2016)
8. Amra, I.A.A. and Maghari, A.Y. Students Performance Prediction using KNN and NaiVe Bayesian. In 2017 8th International Conference on Information Technology (ICIT), IEEE, pp. 909-913, 2017.
9. Atta-Ur-Rahman, Kiran Sultan, Nahier Aldhafferi, Abdullah Alqahtani, Educational data mining for enhanced teaching and learning, *Journal of Theoretical and Applied Information Technology* 96(14)(2018)4417- 4427.
10. Shubhangi Urkude, Kshitij Gupta, Student Intervention System using Machine Learning Techniques,*International Journal of Engineering and Advanced Technology* 8(6S3)(2019)2061-2065
11. Raza Hasan ,Sellappan Palaniappan , Salman Mahmood , Ali Abbas , Kamal Uddin Sarker and Mian Usman Satta, Predicting Student Performance in Higher Educational Institutions Using Video Learning Analytics and Data Mining Techniques, *Advanced Techniques in the Analysis and Prediction of Students' Behaviour in Technology-Enhanced Learning Contexts*10(11)(2020).
12. J. Dhilipan, N.Vijayalakshmi , S.Suriya, Arockiya Christopher, Prediction of Students Performance using Machine learning IOP Conference Series Materials Science and Engineering 1055(1)(2021)012122.
13. Ahajjam Tarika, Haidar Aissab, Farhaoui Yousef, Artificial Intelligence and Machine Learning to Predict Student Performance during the COVID-19, *The 3rd International workshop on Big Data and Business Intelligence (BDBI 2021) March 23 - 26, 2021, ScienceDirect Available online at www.sciencedirect.com Procedia Computer Science* 184 (2021) 835–840.
14. Baashar, Y., Alkaws, G., Mustafa, A., Alkahtani, A.A., Alsariera, Y.A., Ali, A.Q., Hashim, W., and Tiong, S.K. Toward Predicting Student'S Academic Performance using Artificial Neural Networks (ANNs). *Applied Sciences*, vol. 12, no. 3, pp. 1289, 2022.
15. Yadav, N.R. and Deshmukh, S.S. Prediction of Student Performance using Machine Learning Techniques: A Review. In *International Conference on Applications of Machine Intelligence and Data Analytics (ICAMIDA 2022)*, Atlantis Press, pp. 735-741, 2023.
16. Zhaoyu Shou, Mingquan Xie, Jianwen Mo, and Huibing Zhang, "Predicting Student Performance in Online Learning: A Multidimensional Time-Series Data Analysis Approach," **Appl. Sci.**, vol. 14, no. 6, 2024, Article 2522. DOI: 10.3390/app14062522.





Mini Agarwal and Bharat Bhushan Agarwal

Table 1 Factors Description

Factor	Description
Background Attribute	
Gender	Student identifies as male, female, or non-binary.
Mother's Occupation	Mother's professional field (e.g., teacher, healthcare, civil service, homemaker, other).
Father's Occupation	Father's professional field (e.g., teacher, healthcare, civil service, homemaker, other).
Number of Siblings	Number of siblings, which may impact student focus and available resources.
Mother's Education Level	Mother's highest level of education, potentially influencing the student's educational aspirations.
Father's Education Level	Father's highest level of education, potentially influencing the student's educational support.
Family Income	Annual family income, affects the availability of educational resources and opportunities.
Caste Category (if applicable)	Student's caste classification (SC, ST, General, OBC) for potential affirmative action considerations.
Educational Background Attribute	
B.Tech 1st Year Marks	Average marks obtained in the first year of B.Tech, across semesters 1 and 2.
10th Standard Marks	Percentage score in high school exams, indicative of foundational academic performance.
12th Standard Marks	Percentage score in higher secondary school exams, reflecting pre-university academic achievement.
B.Tech Admission Mode	Mode of admission into the B.Tech program (entrance exam or management quota).
Scholarship Status	Indicates whether the student receives a scholarship from the university, potentially reducing financial burden.
Assignment Performance	Contribution of assignments to the overall internal marks, reflecting consistency in academic tasks.
Class Attendance	Frequency of the student's presence in lectures and labs, indicative of engagement with coursework.
Psychological Attributes	
Primary Language	Student's preferred language for communication (English, Hindi, etc.), which may affect comprehension and participation.
Overall Health Status	General health condition, potentially impacting the student's ability to perform academically.
Parental Relationship Status	Status of parents' relationship (living together or separated), which may influence the student's emotional well-being.
Commuting Time	Time spent traveling between home and college, potentially affecting available study time and overall energy levels.

Table 2 Feature Importance of each attribute

Features	Features Importance
feature 0	(0.106895)
feature 1	(0.169679)
feature 2	(0.101148)
feature 3	(0.039577)
feature 4	(0.037702)
feature 5	(0.038873)
feature 6	(0.035574)
feature 7	(0.039569)
feature 8	(0.038789)
feature 9	(0.039869)





Mini Agarwal and Bharat Bhushan Agarwal

feature 10	(0.039171)
feature 11	(0.039274)
feature 12	(0.038094)
feature 13	(0.039570)
feature 14	(0.039626)
feature 15	(0.038963)
feature 16	(0.037756)
feature 17	(0.040460)
feature 18	(0.039411)

Table 3 Confusion matrix of Decision tree

Actual/Predicted	A	B	Precision
A	61	6	.93
B	4	29	.82
F-measure	.92	.85	

Table 4 Model Evaluation of Decision Tree

	Weka	Python
Accuracy	90%	96%
F-score	.90	.95
Precision	.90	.95
Recall	.90	.95
ROC	.92	.95

Table 5 Confusion matrix of Naive Bayes

Actual/Predicted	A	B	Precision
A	62	5	.86
B	10	23	.82
F-measure	.89	.75	

Table 6 Model Evaluation of Naive Bayes

	Weka	Python
Accuracy	84%	94%
F-score	.84	.95
Precision	.84	.95
Recall	.84	.95
ROC	.90	.95

Table 7 Confusion Matrix in SVM

Actual/Predicted	A	B	Precision
A	67	0	.80
B	16	17	1.0
F-measure	.893	.68	





Mini Agarwal and Bharat Bhushan Agarwal

Table 8 Model Evaluation of SVM in Weka and Python.

	Weka	Python
Accuracy	84%	84%
F-score	.82	.82
Precision	.87	.84
Recall	.84	.84
ROC	.75	.8

Table 9 Confusion Matrix of KNN.

Actual/Predicted	A	B	Precision
A	67	0	1.0
B	0	33	1.0
F-measure	1.0	1.0	

Table10 Model Evaluation of KNN in Weka and Python.

	Weka	Python
Accuracy	100%	95%
F-score	1	0.93
Precision	1	0.90
Recall	1	0.90
ROC	1	.92

Table 11 Confusion Matrix of Random Forest Classifier.

Actual/Predicted	A	B	Precision
A	67	0	1.0
B	0	33	1.0
F-measure	1.0	1.0	

Table 12 Model Evaluation of Random Forest Classifier in Weka and Python

	Weka	Python
Accuracy	100%	100%
F-score	1	1
Precision	1	1
Recall	1	1
ROC	1	1

Table 13 Confusion Matrix of Logistic Regression.

Actual/Predicted	A	B	Precision
A	62	5	.925
B	5	28	.84
F-measure	.925	.84	

Table 14 Model Evaluation of Logistic Regression. in Weka and Python.

	Weka	Python
Accuracy	90%	80%
F-score	0.90	0.8
Precision	0.90	0.82
Recall	0.90	0.83
ROC	0.92	0.72





Mini Agarwal and Bharat Bhushan Agarwal

Table 15 Confusion Matrix of Multilayer Perceptron Network.

Actual/Predicted	A	B	Precision
A	67	0	.95
B	3	30	1
F-measure	.97	.95	

Table 16 Model Evaluation of MLP in Weka and Python

	Weka	Python
Accuracy	97%	90%
F-score	.97	.90
Precision	.97	.90
Recall	.97	.90
ROC	.92	.90

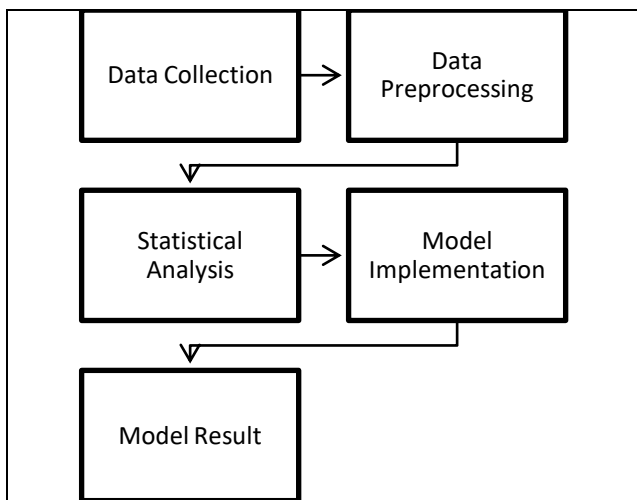


Fig 1 Methodology used in Research

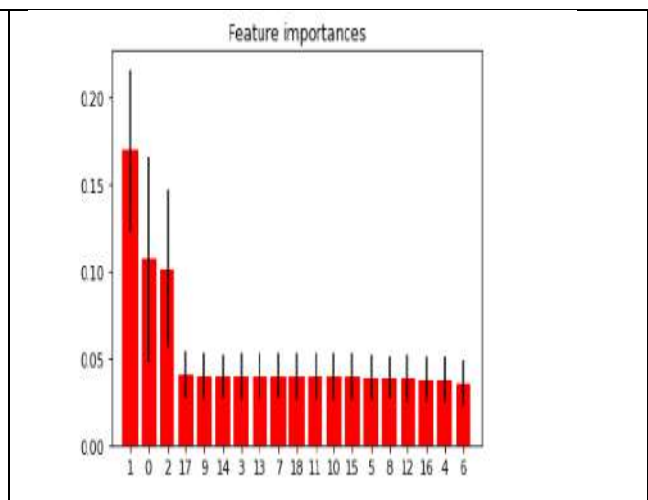


Fig 2 Feature ranking graph

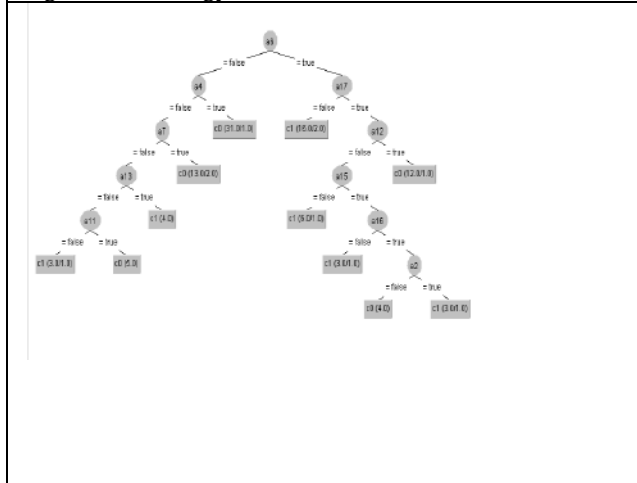


Fig 3 Decision Tree in Weka

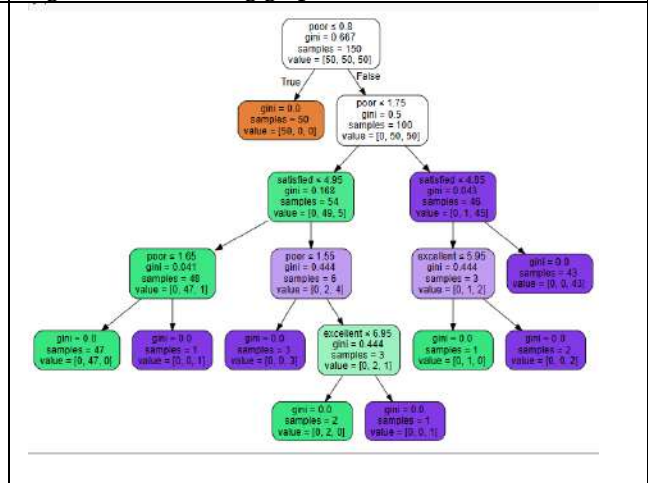


Fig 4 Decision Tree in Python.





Mini Agarwal and Bharat Bhushan Agarwal

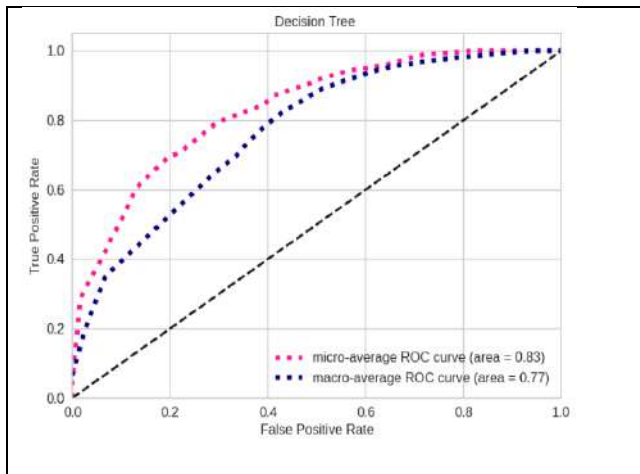
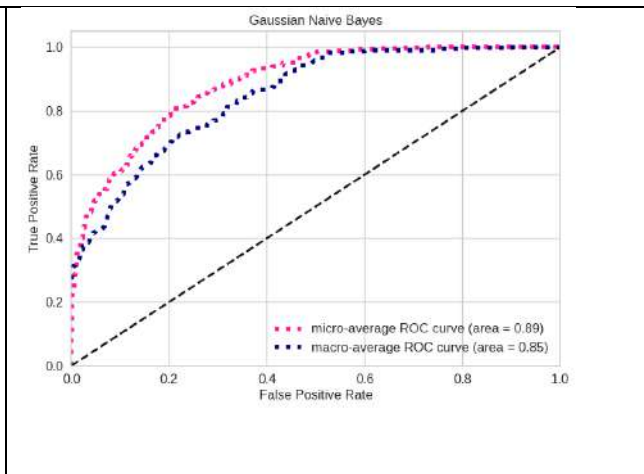


Fig 5 ROC Curve of Decision Tree



.Fig 6 ROC Curve of Naïve Bayes

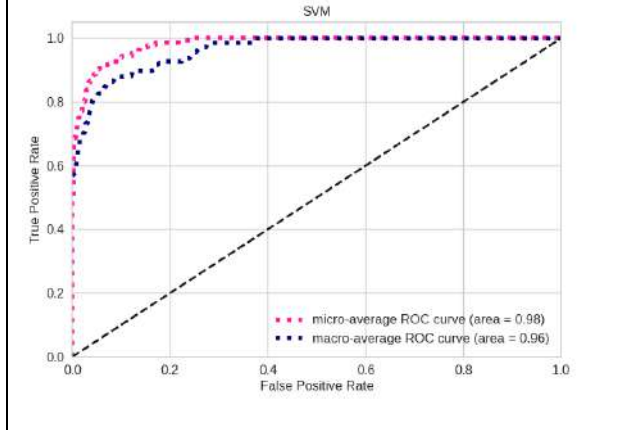


Fig 7 ROC Curve of SVM

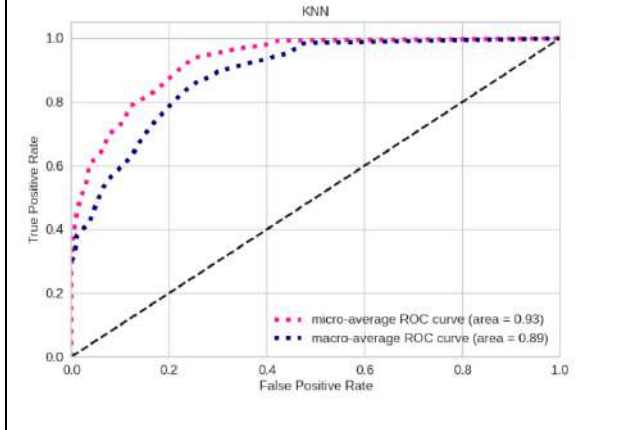


Fig 8 ROC curve of KNN

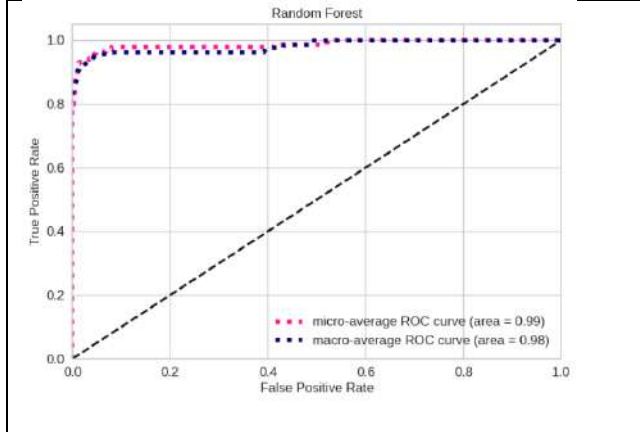


Fig 9 ROC curve of Random Forest

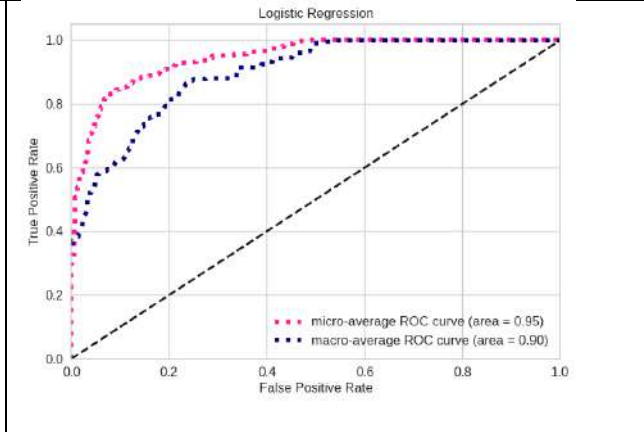


Fig 10 ROC Curve of Logistic Regression





Mini Agarwal and Bharat Bhushan Agarwal

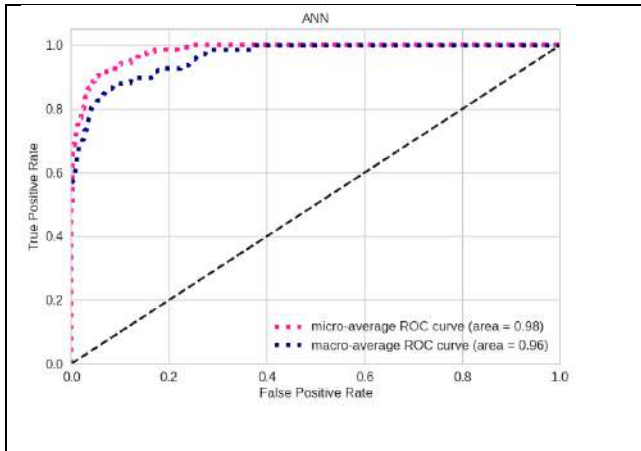


Fig 11 ROC curve of ANN

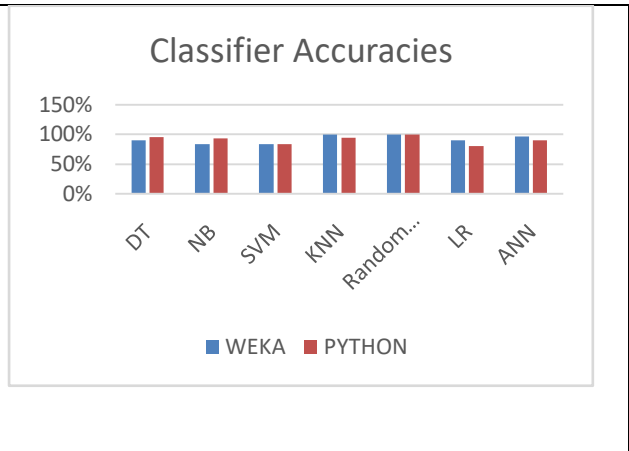


Fig 12 Accuracy comparison between Weka and Python





GMOS? - Superficial Sustainers? Or Capital Combating Crusaders?

Swapna Anna Jacob^{1*} and Premalatha S²

¹Research Scholar, Department of Arts, Humanities and Social Sciences, Reva University, Bangalore, Karnataka, India.

²Associate Professor, Department of Arts, Humanities and Social Sciences, Reva University, Bangalore, Karnataka, India.

Received: 07 Jun 2024

Revised: 13 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Swapna Anna Jacob,

Research Scholar, Department of Arts,
Humanities and Social Sciences, Reva University,
Bangalore, Karnataka, India.

E. Mail: sapnajacob@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License (CC BY-NC-ND 3.0)** which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Life has always been evocative, if its naturally sustained not artificially altered: alterations are justifiable in dire circumstances of do or die: yet not desirable in all dare to do settings where existential necessities are manipulated for contextual conveniences, where man attributes utmost weight age to self and egotistical motives, negotiating with the collective harmony and well-being of the web of life. Humans have lost the gist of eloquent existence hence mislaid the mirth of life in the duel of one outweighing the other, favouring world orders founded on control and confinement giving way to confusion and chaos yielding an speace or sans happiness to the revolutionary or the revolutionised: neither the master nor the slave reap real reimbursements. Most of the revival happening through the propagation of science and technology are benefitting generally the capital collectors. One such capital gaining strategy that might have provoked the initiation of the Gain Manipulating [Gambling] Order: occurs to be GMO. This paper investigates the initiation, survival cum credibility of GMO, provoking the public to rationalize, scrutinize and conceptualize its utility and propagation. Many might marvel about the global role played by GMOs in our ecosphere escalating significantly the dialogs that abound GMOs, in all social media platforms over the past few years: yet the ambiguity surrounding GMO still lingering on in all the developing countries other than the progressive countries around the world, which promotes cognizance hence that have limits or total prohibition for manufacture plus dispensation of the same, then majority have been undoubtedly exposed to them rather consumed it inadvertently as their rewards are projected than their outlays. Artificiality needs more propagation than naturally prevailing support systems, as simplicity seldom sells in this era of propaganda!

Keywords: GMO Superficiality Biotechnology Artificiality Manipulation





Swapna Anna Jaccob and Premalatha S

INTRODUCTION

Grasping the Genetics of GMO

Genetically modified organism (GMO) as the world comprehends it, or its draftsmen designed it, has its genetic material transformed out of inquisitiveness targeting superficial benefits via genetic engineering methods: targeting superficiality in terms of benefit itself raises a concern activating artificial advance in the name of evolution: its steered in anticipations of either obtaining favourable traits, eliminating unfavourable qualities, or simply gene manipulation the precise interpretation, its makeup in heritable engineering differs, with the utmost mutual existence: has been reformed not by obvious breeding or usual recombination. An extensive diversity of creatures are apparently hereditarily revised (GM), from beasts to vegetation plus microbes, transmuting the inheritable factors of the same species, across species altering the transgenics athwart domains. Novel genes are familiarised, there by attracting, altering and knocking out endogeneity: a beneficial game authored by benefactors for beneficiaries. Superficially stupendous strides are embraced like quick fixes, quaking the clarity of creation. Never to be muddled with outdated breeding observes, here diverse breeds of the identical species are united to yield favourable individualities. Such tentative groupings of genes doesn't emerge naturally: hence unnatural. Purpose of exclusivity is undeniably debilitated to the extent of conception of initial indistinct imbalances further drawing the situation incomprehensible hence riotous.

Riotous risks rattle right to existence of each entity entailing the upkeep of ease of earth. Generating an intricate inherently adapted being is undoubtedly a multi-step progression. Genetic applied scientist details isolation of the genetic factor that's demanded to be supplemented into the host organism and associate it with other inherited fundamentals. Innumerable Procedures are accessible for implanting the secluded gene into the host genome. Current Progressions By means of genome editing procedures, especially CRISPR, have concluded the manufacture of GMOs much simpler. Forerunners in generating the primary GMO in 1973, Herbert Boyer and Stanley Cohen Positions to be the innovators of a bacterium buoyant to the antibiotic kanamycin. Rudolf Jaenisch shaped the preliminary GM animal, a mouse, in 1974, followed by the foremost shrub in 1983. Precisely After fourteen years, the year 1994, observed the problem of Flavr Savr tomato, the foremost marketed GM food. The primary GM animal that happens to be marketed was the GloFish (2003) and the foremost GM animal to be permitted for sustenance happens to be the Aqu Advantage Salmon in 2015. The mainstream GMOs are concocted branding them too lenient to bulky birds of insecticides and weed killers, like Roundup, a biochemical enricher designed by the establishment Monsanto. Glyphosate a possible human carcinogen, was found in Monsanto's Roundup, that was avowed by the World Health Organization in May 2015. Commonplace Weeds Seldom survive while bare to these punitive substances, points to prowl on GM corn's unnatural genetic makeup premeditated to subsist when administered. GMO yields are held accountable aimed at the advent of "super weeds" and "super microbes" that could lone be slayed by the deadliest lethal contagion alike 2,4-D (a key component in Agent Orange). The agriculturalists pretentious by resilient pests should be advised to return to previous plus complex lethal elements should essentially gather more labour or more rigorous ploughing, that outshine the assured reimbursements of GMO expertise.

Kenyan Epitomes on GMO effectuality/ ineffectuality

This pursuit would vouch to remark about one such dwelling that apparently got influenced by GMOs is none other than Kenya: Lately Kenya fixes to commodify heritable reformed crops, gaining confrontation from few farmers and operation clusters, that interrogates their security. Allegations of farmer Eva Wanjiru, points to the abnormalities of artificially engineered edibles, floating her apprehension over the administration's current reversion of a decade ban on the inherently modified crops. Ms Wanjiru executed organic agriculture for years, refusing to employ insecticides or fusion kernel in her farm, trusts that there is dearth of much satisfactory signal to verify that yields fashioned through biotechnology will be backing the nation battle sustenance diffidence as utmost farmers grumble about pests and ailments, who promote GMOs. Scarcity of rain, compel them to whine about the yield's meagre coping in the farm dispersing qualms about farmer's absolute reliance on corporations that trade the GMOs, and the threat of their command of the marketplace to the hindrance of commonplace Kenyan farmers. Eventuality of pronouncement calls



**Swapna Anna Jaccob and Premalatha S**

to initiate thought processes guiding to comprehend the malign intentions of capture for capital gains. Such quick fixes as solutions to problems have so far served as only superficial solutions.

Consenting these companies to oversee; affects the upkeeps of the agriculturalists, in Kenya, manufacturing maize yearly reports, Claire Nasike, conservational researcher at Greenpeace Africa exposes the crux of the capture: as withdrawing the GMO prohibition exposes agriculturalists to draconian intellectual property regulations linked to copyright detained by GMO corporations as the GM seed is patented and this might lead to mooring the agronomists deprived of their information into intellectual possession rows, clarified Ms Nasike .Yet Dr Stephen Mugo's argument refutes the idea that Kenya will be under the kindness of. director of the Centre for Resilient Agriculture for Africa, moreoverit's unbelievable assumption subsequently Kenya has the capacity to grow GM harvests. Maximum Corporations lease the expertise which they use create novel genes, Apparent exploitation of natural resources in the name of scientific revolution: what nature nurtures and serves freely for all is patented for profit: a sort of hypocritic hypnotism that holds the nerve paralysing the simple thought processes of ordinary man with the aid of heightened hyped up exaggerative inventions that confuse more than infuse, quake more than comfort bringing in slow devastation rather than sincere conservation. A study steered via a non-governmental body, Route to Food Initiative, preceding year presented that more than half of Kenyans unwelcome GMOs.Kenya is one of countries in the mainland favouring GMO use, presently approved for agronomy in 70 countries throughout the world. Yet The above cited securities will not suffice in resounding dubious community dwellers such as Ms Wanjiru to cultivate, or consume, GMO crops for the reason of their apprehensions about the security and misgivings over the prediction of financial reimbursements. Why such security apprehensions and fiscal benefit qualms arise even after solid promotions and publicity. Rise of scepticism has constantly been measured with the wake of artificiality and greed.

Carbon Clutching by Progressive Photosynthesis Procedures for Taming Temperature Transformation

Climate variation prevails all over the world, the prevalent traditions of farming hardly serve the purpose. The demand is about somegrander, more miscellaneous variety of crops that perform even in adverse circumstances. Presently, asper the United Nations' Food and Agriculture Organization, mere 15 crops make up 90% of our liveliness intake, familiarizing, folks who try evolving food crops that might prosper in our varying world, to benefit fraught farmers in parched areas. Much research has been taken forward in the course of tackling adversity in the desired direction, yet in haste to ease of tackling testing times, man often apparently is forced to support the unnatural further destabilizing the natural course of life events. Sangita Myska interacts with the co-initiator of Living Carbon, Patrick Mellor, who facilitated the advance of the world's first GM trees precisely planned to grab more carbon from the atmosphere, with a biotechnology developed to improve photosynthesis in poplar trees, permitting them to breed quicker and also fight decay for extended periods, yet how far that is going to contribute to the harmonious persistence of the web of life, only test of time can reveal as individual endeavours entertain or are limited to personal gains and accomplishments. Living Carbon anticipates to employ these trees to influence the level of carbon draw down required to uphold an ideal level of carbon dioxide in the atmosphere. Yet alas! The question initiating debate here is: Are sooner growing trees the evolutionary hack we've been looking for? Some favour the GM harvests, hence found them as a technique to resolve the biosphere's sustenance catastrophe and demanded they could formulate "the biggest revolution of a lifetime". Opposition Assumed that hereditary alteration went contrary to the regulations of nature, and would create the alleged Frankenstein foods.

The year 1994 witnessed FlavrSavr tomato to be the initial hereditarily altered harvest appropriating itself for business in the US, ultimately planning its track to British hypermarket shelves in the tomato puree formula, but once campaigners disclosed this, they rejected sellers from supplying it and other foodstuffs consequential from GM crops. Over a succession of months, campaigner shattered the fields of Government supported trials of GM maize and soya, Prince Charles went one-on-one with the pro-GM founding, enabling the hypermarket chains mandatory in reassuring the public that they were devoid of GM "polluted" goods. The key benefit of GM food is that crop harvests turn out to be more dependable and fruitful, permitting extra individuals to be nourished, rather there are arguments about world manufacturing more calories than it is mandatory for all the people on the planet to be hale



**Swapna Anna Jaccob and Premalatha S**

and hearty. Individual discretion should seldom be permitted to be overridden by artificial intelligence, as man has not been successful in substituting anything natural with artificial. Hence artificiality breeds limitations, irrationality and ill-health. Validating the mentioned in authenticity through research will lead one to study about pros and cons of genetically modified organisms. GMOs are not deprived of drawbacks. Nonetheless the absence of decisive links, Brown University resolved that vicissitudes to nourishments on a heritable level syndicate proteins that people hardly consume, swell the probabilities of a hypersensitive response stirring, escalating the rates of nutrition allergies in children since 1999.

Advantages of GMO

Recompenses of GMO" s needs some reference though drawbacks may outweigh rewards. Primary pounce of compensation arises as food supplies turn out to be foreseeable, crop harvests become more dependable and fruitful, letting more people to be nourished, notwithstanding the prerogative of feeding nutritious food than mere quantity. Dietary Plus content could be enhanced cum altered, provisioning a deeper dietary outline than what preceding peers were able to relish, means future generations could progress in receiving the equivalent nourishment from lesser levels of nutrition intake. Will all these expectations could gain ground and be real or are these prospects publicity gimmicks for capital gains. Global vitamin deficiencies are condensed by the utilization of high levels of vitamin A in Genetically modified rice, records the UN Food & Agricultural Department. GM foods can have extended ledge life that can abstain the aid of additives to sustain diet freshness enhancing the accepted potentials of the food itself. Conservational Nourishment, upholds the risk of certain preservatives that are linked with a higher carcinogen, heart disease, and allergy risk.

Prowess of producing some proteins and inoculations, end to end with added pharmacological properties, are considered to be medical benefits from GMO crops, a practice subscribing inexpensive means of refining individual health: by eating dinner receiving a tetanus promoter as an alternative of receiving an arm inoculation – that's assumed to be the outcome of this expertise, generating diets that are extra tempting to consume. Colours can be either transformed or enhanced with GM foods to enable them more attractive to eat. Spoon University notify those profounder colours in diet deviates the perception of what is being consumed. Resonant red colours make diet seem honeyed, even if it is not: as livelier foods are connected with enhanced nourishment and flavours. Moreover, GM foods are easier to transport as they are known for their extended shelf life, inviting the opportunity to eliminate food waste, increasing the possibility of hunger reduction and elimination in developing countries. Meticulous usage of herbicides and pesticides curbs the threats on croplands that's bound to eventually turn the soil ineffectual, agriculturalists growing genetically modified foods seldom use these foodstuffs as frequently as agronomists using old-style bidding procedures, permitting the earth to recuperate its nutrient foundation over time. In spite of the heritable confrontation being in the plant itself, the grow ermanages to accomplish a foreseeable produce at the similar time.

Downsides of GM Foods?

Man-made mechanisms mediate, mirror, medicative benefits but all those perks personify a greater scale of disruptive disorganisation in the normal/ mundane events that are called to catalyse the consumerist culture. GMO yields may cause antibiotic resistance. It is high time to park self and scrutinize whether the cons outweigh the pros, the investigation done in Iowa State University research estimates that harvests adapted to contain antibiotics and other items that slay microorganisms and pests, by diminishing the effectiveness of an antibiotic or additional medicine when it is obligatory in the old-style logic. Some amount of the antibiotic dash in GMO" s when consumed, is affected by a treatment antibiotic due to build resistance to it, that causes cure of illness difficult. A greater legal liability, is generated on the farmers growing GMO, due to the fact that harvests that are heritably adapter is guaranteed to generate kernels that are hereditarily revised. Moreover, the likelihood of cross-pollination between GMO crops and non-GMO crops are followed during definite agricultural observes, since many of the harvests and kernels that produce GMO crops are unproved, exposing the peasant to a complex level of lawful accountability. Agriculturalists Who aren't involved in the above process could also go through obligations for leasing seeds and authorizing their utility in other fields or allowing cross-fertilization to occur.



**Swapna Anna Jaccob and Premalatha S**

Genes are driven into dissimilar plant sorts further crop's part grounds with supplementary plants, together with weeds. Genetic relocations are notorious to transpire. Research about the ordeal of transfer of DNA segment from a herbicide-resilient harvest permeating into the weeds it is premeditated to slaughter? Communications at the cellular level possibly will produce unanticipated snags to forthcoming harvest development where even the reimbursements of GM foods might not outdo the complications rooted by them. In an effort to defend their profits over 50% of seed manufacturers forbid any self-governing study on the concluding yields as some GM foods may pose a carcinogen contact hazard. Multiple paper publications and retractions on the topic, exposed that harvests lenient to profitable pesticides significantly amplified the danger of cancer advance in rats. Nonetheless restricted, this evidence has been extensively spread creating the imprint that all GMO foods are hypothetically perilous.

An analysis directed by researchers scrutinise the conducts and mechanisms that aids extraneous DNA helps subsist through absorption and infiltrate into the body cells of the user. Scientists are predominantly concerned in reviewing the gastrointestinal tract being the hot plug for parallel gene transmission of GM yield DNA into gut-bacteria. Pointing the consequence of the mentioned allocation can theoretically cause dysbacteriosis and bad-health, as well as successive transmutations. Researchers pondering on the incongruities that ascend about the issue of the occurrence of DNA of disbursed foodstuffs in the blood and organs of their customers further affirming the fiasco to illustrate their consequences on the human body, nonetheless they are also deliberating the likelihood of mixing genomes and the appearance of extraneous DNA in consumer's tissues. In supposition, scientists contemplate the penetration potential of micro-RNA of GM plant diet into the human body cells.

The recompenses and drawbacks of genetically modified foods can spur a nasty deliberation. The benefits of provisioning the ecosphere with improved diet access, should not be familiarized at the outlay of individual well-being. GMO foods must essentially be branded in EU and appeals in the US are in quest of an identical scenario. We warrant to identify what we're consuming and how that diet is made. The dispute around GMO is enormous and intense on either side. Notwithstanding one of the chief contemplations when at variance with the usage of GMO goods is the environmental devastation potential. Prioritizing the ecological hazards to contemplate in regard to GMOs: Chiefly, it is imperative to comprehend what a GMO is precisely, where WHO outlines them as entities whose genetic material has been transformed in an artificial way. GM floras are typically formed for bug resistance, virus resistance, or weed killer tolerance: approval of such unnatural alterations bring in some hypothetically challenging conservation challenges.

Environmental Hazards of GMO!

Potential human hazards were under the public scanner, whereas environmental threats can pose a coercion to the human environment bond stability supporting the steadiness of the web of life. Primarily, noxiousness is an enormous problem neighbouring biochemical insecticides and weed killers, employed typically with GMOs, in calculation with the harmfulness integral to these plants. Toxicity of GMOs to non-target organisms is quite evident, taking the visible communication illustrations of bees and butterflies, bees are immensely imperative in pollinating many diet harvests but are regrettably enormously threatened by contemporary agrarian practices, such as GM crops. Birds are equally at peril from insecticides and toil as organic regulators, negotiators and pollinators, alike bees. Additionally, the long-standing effects of GMOs aren't firm but uncertain, conditioning pests to familiarize to insecticides and weedkillers, in calculation to the DNA variations in GM plants enabling them "resilient." Effectively this deciphers the doubt in authenticity of their efficacy, yet their lethal legacies will persist.

Collective paraphernalia of goods such as GMOs should be taken into contemplation. Recommendation Too Endorses that minor hereditary variations in plants fabricating even bigger biological swings, divulging the latent for GMOs to turn out to be tenacious and weedy in agronomic environments, since such has been altered to be resilient to some contemporary farming procedures, the hostility of GMO in natural settings seldom transpire logically, where the impossibility of novel, man-altered vegetation to turn out to be aggressive species is subtle, usual bionetworks could be ruled out. In conclusion, biodiversity, the critical component of all ecologies assimilating to the sustenance of all species, is put to danger by GMOs. The plantation of GM crops, commonly in a monocrop manner,



**Swapna Anna Jacob and Premalatha S**

countless custom seeds are seldom used, upsetting the natural course of nature yet promoting the nature of GMOs through fewer wildflowers and, therefore, not as much of sap for pollinators. Contaminants Let unregulated into the earth through the plants' paths mean threatened mud bacteria, that are essential to conserve vigorous soil for plants growth without the use of biochemical enrichers. Nutrients are rarely reimbursed to the earth in mono harvests and from GMO diets, as poisonous residues are left in the soil, making it parched and annulled of all nutrients, mostly essential to the budding procedure, further creating a sequence of reliance on GMO kernels and biochemical enrichers, insecticides, and weedkillers in then fashioned in order to produce a solitary harvest. Adding on to earth matters, the irrigation measures that are employed for organic growth of GM foods, transmits innumerable complications into aquatic springs and the air, divulging diverse microorganisms, bugs, and beasts to identical difficulties.

Many of such influences must be booked into contemplation in the bigger representation; GMO's DNA may terminate in the earth, manure, animal feedstuff and by-products, and further active creatures, from bugs to biggerbothers. Bees transfer insecticides, weedkillers, and genetic material through the air into the atmosphere. A plant partnering in the agronomic environment, is lucid to accept it to be a fragment of a grander bionetwork, apparently the threat of conservation harm done by GMOs is much greater than merely hypothetically impairing our healthiness. Apart from conservation matters, GMOs have been the topic of communal and moral deliberations as well. rooting a complex range of significances. Being conversant about the diet we are consuming and the methods contemporary agronomic techniques are upsetting the atmosphere is one operative technique of intentional interaction with the normal world.

Influence, GMOs Inflicts on Society and the Setting

In incongruity in what way mainstream opinions are fashioned, GMOs have essentially triggered the use of weedkillers and insecticides to upsurge pointedly, enabling the lingering of more biochemical deposit on yields for customers to consume, disclosing study around GMOs contradictory; nevertheless, there is a swelling body of indication linking GMOs with ecological damage, well-being problems, and even defilements of customers' and agriculturalists civil liberties. Many health destabilising diseases and disabilities like Autism, gluten intolerance, birth defects, besides several additional shocking wellbeing matters are all allied to Monsanto's Round Up. Some agronomists are satisfied with GMO diversities, others are dissatisfied, thus discovering wide-ranging outcomes or confronting new snags in the extremely intense and business conquered seed sector, such challenging drift disturb all agronomists, whether they favour GMO seeds or not. Jeopardy of Cross adulteration with GMO seeds is communal though the farmer refrains from using GMO crops. International Journal of Food Contamination observed innumerable cases of GMO adulteration between 1997 to 2013 in 63 nations, exposing agronomists to severe, dire penalties. Adulteration Catering the role of a catalytic agent for melodramatic financial fatalities for growers facing refusal from exportation bazaars that prohibit GMOs. Biological Agriculturalists suffering adulteration could drop their biological authorization and the premium for their biological yield. Further when request for non-GMO foodstuffs wells, agriculturalists are looking for prospects to spread into no-GMO marketplaces that recompense advanced charges, nevertheless, the incapability of businesses to suitably detach GMOs from conservative variation slingers to lurk agronomists' selections.

The lasting well-being paraphernalia of GMOs, mutually on the globe and society are mysterious. Adding on, approximately all scholarships that entitled to be innocuous are sponsored by the only bioengineering formations that yield from GMO auctions. Biotechnology Business Interprets GM yields as an advantage to our ecosphere for the reason that they permit agriculturalists to create advance harvest yields with scarce contributions. The kernel marketplaces have been inundated with GM soybean, corn, and cotton seeds with two major traits: insecticide expression and weed killer resistance, by big seed companies So, what does this contribute to? Essentially equipping the vegetation with the aptitude to subsist with detrimental microbe killers and weedkillers with the deadly characteristic of *Bacillus thuringiensis* is, or Bt. Bt is associated to a class of microorganisms that reasons diet intoxication and anthrax, also slays only very exact class of pests. Numerous Biological Agronomists have used Bt for over 50 years as an insecticide to regulate bugs. Here and now, genes from Bt are instrumental in amending



**Swapna Anna Jaccob and Premalatha S**

vegetation to yield the Bt toxins and slay bugs that try to plague them without the application of exterior spraying. Researcher Charles Benbrook at Washington State University verified the upsurge of GMOs in the US has led to an intensification in the use of lethal biochemical involvements. Benbrook exposed that while the Bt feature has permitted agriculturalists to spray intensely smaller levels of pesticides, that outcome has been further overshadowed by the incursion of weedkillers unconstrained by Monsanto's Roundup Ready technology, as weeds have swiftly modified confrontation to recurrent dosages of Monsanto's Roundup weed killer.

Do They Yield More Food? - Consolidation of Need Vs Greed!

Investigatory logic around all these scientific technological evolutionary discoveries lingers around the ever-escalating unaccountable population and hitches associated with catering to such mounting statistics. So here, nowadays the quaking enquiry is, do GMOs harvest extra nutrition? A lately published article by University of Wisconsin has annulled the dispute that GMO agriculture profits additional food. The investigator equation harvest yields from numerous diversities of blend corn, some hereditarily altered and some not, between 1990 and 2010: nevertheless, some hereditarily adapted diversities generated insignificant advances, others did not. Myriad even exhibited inferior harvests than non-genetically modified equals. Investigate Information Determined the startling not so strappingly optimistic transgenic profit effects. Both the Bt characteristic for corn rootworm and glyphosate-tolerant prompted harvests to drip. Calculation, exposed indication of what is recognized as "yield drag" — the clue that deploying the genome of a shrub diversity sources inadvertent fluctuations in the mode it nurtures, triggering it to be not as much prolific.

However accurate this may seem to be, it is remote from producing advanced harvest yields with scarce involvements. The clarity of GMO seeds having compensations over conservative kernels when it derives to hazard justification, exceed the reimbursements presented by biological farming. Consequently, profound study is required to discover the new constituents of biological compost for snowballing the produce and other nutritive necessities of the diet grains, hence witnessed an upsurge in deliberation on whether to opt biological agriculture or conservative agriculture or accept GM crops for advanced produce. Man's vigour and ecological security are the key anxieties of the GM expertise, and in the nonattendance of guarantee by the exponents, countless have considered biological agriculture greater for its advanced harvests as equated to conservative agriculture. Another challenging matter with unswerving harvest yields — monocultures. Monocultures eventually diminish the earth and furnish the prerequisite for extra enrichers and substances to steady harvests. Apparently, this study urges farmers to use deep-rooted farming procedures — like harvest variation and permaculture that nurtured a restored and more maintainable atmosphere for harvests, there would be scarce requirement for all these substances.

REFERENCES

1. "Genetically Modified Organism." *Wikipedia*, 28 Apr. 2023, en.wikipedia.org/wiki/Genetically_modified_organism.
2. Kagee, By Richard. "Why Kenya Is Turning to Genetically Modified Crops to Help With Drought." *BBC News*, 9 Nov. 2022, www.bbc.com/news/world-africa-63487149.
3. "The Food Chain - Tomorrow's Food Crops - BBC Sounds." *BBC*, www.bbc.co.uk/sounds/play/w3ct38n9.
4. "Positive Thinking - Can GM Trees Capture Our Carbon? - BBC Sounds." *BBC*, www.bbc.co.uk/sounds/play/m001bbwc.
5. "The Reunion - the GM Crops Debate - BBC Sounds." *BBC*, www.bbc.co.uk/sounds/play/m000mbpp.
6. Gaille, Louise. *12 Advantages and Disadvantages of Genetically Modified Foods*. 16 Dec. 2019, vittana.org/12-advantages-and-disadvantages-of-genetically-modified-foods.
7. Glass, Emily, and Emily Glass. "The Environmental Impact of GMOs." *One Green Planet*, Aug. 2013, www.onegreenplanet.org/animalsandnature/the-environmental-impact-of-gmos.





Swapna Anna Jaccob and Premalatha S

8. Qutab, Marina, and Marina Qutab. "How Do GMOs Impact People and the Environment – and Do They Produce More Food?" *One Green Planet*, Apr. 2017, www.onegreenplanet.org/environment/how-do-gmos-impact-people-and-the-environment.
9. Shiva, Vandana. *Staying Alive: Women, Ecology and Survival in India*. 2010.
10. Fleiss, Colleen. "Molecules of GM Food Can Accumulate in Human Digestive Tract and Enter Cells of Body: Study." *Medindia*, 21 Jan. 2019, www.medindia.net/news/molecules-of-gm-food-can-accumulate-in-human-digestive-tract-and-enter-cells-of-body-study-185266-1.htm.





Role of Schiff base Metal Complexes in Dyes and Paints : A Review

Monika Tyagi^{1*} and Ankit Mahur²

¹Associate Professor, Department of Applied Science and Humanities, IIMT College of Engineering, (Affiliated to Dr. A.P.J. Abdul Kalam Technical University), Uttar Pradesh, India

²Assistant Professor, Department of Applied Science and Humanities, IIMT College of Engineering, (Affiliated to Dr. A.P.J. Abdul Kalam Technical University), Uttar Pradesh, India

Received: 22 Jun 2024

Revised: 03 Jul 2024

Accepted: 24 Oct 2024

*Address for Correspondence

Monika Tyagi

Associate Professor,

Department of Applied Science and Humanities,

IIMT College of Engineering,

(Affiliated to Dr. A.P.J. Abdul Kalam Technical University),

Uttar Pradesh, India

E.Mail: tyagimonika554@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Schiff bases are versatile ligands, synthesized by the condensation of an amino compound with a carbonyl compound. Schiff bases and their metal complexes are an interesting area of research that provide valuable information about newly discovered compounds. These compounds and their metal complexes have been extensively studied for their various potential applications in a variety of sectors. This review focuses on Schiff base metal complexes used in dyes and paints.

Keywords: versatile ligands, metal, Paints, metal, dyes.

INTRODUCTION

Schiff bases are the compounds that contain an azomethine group (-CH=N-). They were first discovered in 1964 by Hugo Schiff and are named after his name. These compounds are formed when a primary amine reacts with a carbonyl compound (aldehyde or ketone) Fig.1 showing general method for preparation of Schiff base by condensation between carbonyl compounds and primary amines. Because of the presence of effective conjugation, aromatic aldehyde Schiff bases are more stable, but aliphatic aldehydes are comparatively unstable and polymerize easily [1]. Schiff base compounds are used in a variety of biological and pharmacological applications. They are also used as fundamental units in several dyes. Transition metal complexes of Schiff bases have been synthesised and studied extensively over the last several decades because they behave as good chelating ligands in the field of coordination chemistry. Chelating ligands comprising N, S, and O donor atoms offer a wide spectrum of biological functions, which may be enhanced by the presence of metal ions attached to them. Because chromium metal





Monika Tyagi and Ankit Mahur *et al.*,

complexes are coloured, they are used in pigments. Furthermore, the colour of emeralds and rubies is caused by traces of chromium ions. Compounds of manganese and nickel are widely used in analytical, metallurgical processes, paint and pigment industries and ceramics.

The Schiff bases involved in complexation are introduced to demonstrate their use in dyes and pigments. Metal complexes are compared with a newly synthesised Schiff base derived from salicylaldehyde and its derivatives, as well as 2-aminophenol or 2-aminobenzyl alcohol. Various physicochemical techniques were utilised to identify and analyse the characteristics of such compounds [2]. The most important characteristic that account for the colour and hard/soft properties of Schiff base reaction sites [2,3,4] is the planar chemical conformation. Several Schiff base transition metal complexes have been synthesised and investigated, such as Al(III), Fe(III), Co(II), Ni(II), and Cu(II). These metal ions, as well as a number of others, have been employed as mordants in dyeing methods. In coordination applications, the principle of complexation and mordant has the same idea. To explain the sites of coordination, the reaction of salicylaldehyde with anthranilic acid or o-aminophenol has been prepared and examined. The reaction represents the interaction of the hydroxyl, amino, and azomethine groups [2,5,6,7,8].

Despite their overall structural similarity, the sort of engagement of the hydroxyl groups and the metal ions in bonding promotes the staining or dyeing activity. The aspect of coordination varies the sites, components, and groups that can be worked in bonding. The hydroxyl groups, rather than the azomethine groups, are the optimum sites of coordination in this case, as long as these groups are in appropriate positions. Because of the differences in the position and characteristics of the hydroxyl groups, certain Schiff bases may be exceptionally effective agents for the tasks assigned [2]. A dye is a coloured substance that chemically bonded to the substrate it is applied on. Dyes are distinguished from pigments by the fact that they do not chemically attach to the substrate they colour. Dye is usually applied in an aqueous solution, and it may be necessary to add a mordant to increase the dye's fastness on the fibre [9]. Dye and pigment are both coloured because they absorb only certain wavelengths of visible light. Dyes are generally water soluble, whereas pigments are not. The ability of a dye to impart colours to fibres is its most essential feature. Different types of dyes have been produced for various fibres based on their nature. Different fibres are dyed with different dye classes.

Generally, one or more class of dyes are used to dye specific fibre. The fibres are broadly divided into two classes:

- (1) Natural fibres
- (2) Synthetic fibres.

Cotton, wool, and silk are examples of natural fibres, whereas synthetic fibres include nylon, cellulose acetate, cellulose triacetate, polyester, polyacrylonitrile, and others. Natural fibres have a strong affinity for water and are hydrophilic in nature. They absorb water and swell, making dyeing with water soluble pigments easier. Reactive dyes, direct dyes, acid dyes, vat dyes, and other dyes are available for dyeing natural fibres. These colours are water soluble and preferentially bind to hydrophilic fibres. Synthetic fibres are hydrophobic in nature, with little or no affinity for water. Synthetic fibres with reactive sites, such as nylon and acrylic fibres, can be dyed with water soluble dyestuffs such as acid dyes and cationic dyes, respectively. On the other hand, the most prevalent synthetic fibres, polyester and cellulose acetate, contributed to the formation of another type of dye known as dispersion dyes.

Classification of Dyes

Dyes can be classified in many ways such as according to their chemical structure, the chromophoric system, chemical properties, origin and applications. Some of the important classification are described below:

Classification on the basis of origin:

Natural dyes

The majority of natural dyes come from plants, such as roots, berries, bark, leaves, wood, fungi, and lichens etc. [10].

Synthetic dyes

The majority of dyes are synthetic in origin, that is, they are man-made and derived mostly from petrochemicals, and they have a wide range of uses. Mauve, the first synthetic dye, was discovered by chance by William Henry Perkin in





Monika Tyagi and Ankit Mahur et al.,

1856 [11,12]. The discovery of Mauveine Fig. 2(a) caused an upsurge in synthetic dyes, broadly in organic chemistry. Fuchsine 2(b), Safranin Fig. 2(c), and Induline were among the other aniline dyes that followed. Since then, dozens of synthetic dyes have been developed [13,14,15].

Chemistry of dyes

The colour of a dye is determined by its capacity to absorb light in the visible portion of the electromagnetic spectrum (380-750 nm). An early hypothesis known as the Witt theory stated that a colourful dye has two components: a chromophore that gives colour by absorbing light in the visible area (nitro, azo, and quinoid groups are examples) and an auxochrome that serves to intensify the colour. This concept has been replaced by modern electronic structure theory, which states that the colour of dyes is caused by visible light excitation of valence π -electrons [16].

Dyes possess colour because of the following reasons:

- 1) They absorb light in the visible region of the electromagnetic spectrum in the wavelength ranging from 400nm to 700 nm.
- 2) They contain at least one chromophoric or colour-bearing group.
- 3) They have a conjugated system, i.e. the presence of alternate double and single bonds.
- 4) They possess resonance in electronic system which provides stabilization to the organic compounds [17].

In the absence of any of the above mentioned distinctive features in the dye's molecular structure, the colour is lost. Chemists have long been interested in finding the connection between a dye's colour and its molecular structure. Furthermore, study of colour and structural relationships has always been crucial in the production of novel dyes, arguably the most significant early contribution to the science of colour characterized as the chromophore and the auxochromes [2,18].

The theory based on the chromophore principle is frequently an electron-drawing group and auxochromes are electron-releasing groups that are connected to one another via a conjugated structure. The notion of the donor-acceptor chromogen was therefore formed. Furthermore, it was discovered that increasing the electron-withdrawing power of the chromophore, increasing the electron-releasing power of the auxochromes, and extending the length of the conjugation could result in a bathochromic-shift of the colour, i.e. the absorption band shifting to a longer wavelength. Although it lacks formal theoretical support, the chromophore and auxochrome hypothesis is presented as a straightforward technique for understanding the genesis of colour in dye molecules. The carbonyl (C=O), diazo (-N=N-), azomethine (-CH=N), and nitro (NO₂) groups are the most significant chromophores, according to this definition. Other groups, such as hydroxyl (OH) and amino (NR₂) groups, intensify the colour and shift absorption to longer wavelengths of light. Most chemical families of dye, including azo, carbonyl, azomethine, and nitro dyes, may be applied to the approach [2,19].

Classification on the basis of chemical properties:

Acid dyes

Acid dyes are anionic dyes that are water-soluble and are applied to fabrics such as silk, wool, nylon, and modified acrylic fibres using neutral to acid dye baths. Salt formation between anionic groups in the dyes and cationic groups in the fibre is thought to be responsible for at least some of the attachment to the fibre. Acid dyes have no effect on cellulosic fibres. The majority of synthetic food colours belong into this group. Acid dyes include Alizarine Pure Blue B, Acid Red 88, and others.

Acid dyes are a large family of dyes that are often formed of sodium salts of colour acids that contain sulphonic acid or phenolic groups. An acid dye's colour is found in its negative ion. These colours are almost often separated as sodium salts. These colours are almost often separated as sodium salts. It is difficult to separate the free dye. Because of their hydroscopic nature, they are difficult to pack and store. Acid dyes are anionic dyes that are water-soluble and are applied to fabrics such as silk, wool, nylon, and modified acrylic fibres using neutral to acid dye baths. The wool and silk textiles are coloured with anionic dyestuffs in an acidic medium. Under these circumstances, the dissociation of the carboxyl group of wool or nylon is limited, and amino groups are transformed to ammonium ion by accepting proton. As a result, the amino groups exist as reactive ammonium ions. In water, the dyestuff dissociates, and its





Monika Tyagi and Ankit Mahur et al.,

anion is bonded to silk or wool. Salt formation between anionic groups in the dyes and cationic groups in the fibre is thought to have a role in the dyes' attachment to the fibre. Examples of acid dye are Alizarin(3a), picric acid (3b), Acid red 88(3c), Pure Blue B, naphthol yellow, etc.(Fig 3).

Basic dyes

Basic dyes are water-soluble cationic dyes that are mostly used on acrylic fibres but also on wool and silk. Acetic acid is typically added to the dye bath to aid in dye absorption onto the fibre. Basic dyes are also employed in paper colouring. Methylene blue 4(a), Crystal violet 4(b), Rhodamine B 4(c), Basic violet 2 are some examples of basic dyes (Fig 4).

Direct (Substantive) dyes

Direct or substantive dyeing is often carried out in a neutral or slightly alkaline dye bath at or near boiling point with the addition of sodium chloride (NaCl), sodium sulphate (Na₂SO₄), or sodium carbonate (Na₂CO₃). Cotton, paper, leather, wool, silk, and nylon are all dyed with direct dyes. They are also used to detect pH and as biological stains. Congo red (Fig. 5), Indigo, Lichens, and other natural dyes are examples of direct dyes. Direct, azoic, vat, sulphur, and reactive dyes are designed for cellulose polymers. Direct dyes were the first colourants to have affinity for cotton in the absence of a binding ingredient known as a mordant. Many of these dyes have low wet fastness because they are water-soluble. The two primary features of direct dyes are: (1) They are linear molecules, and (2) they can get close to the cellulose chain to enhance the impact of intermolecular interactions like H-bonding.

Mordant dyes

Mordant dyes require a mordant, which enhances the dye's resistance to water, light, and sweat. The mordant used is very important since various mordants can significantly alter the final colour. Because the majority of natural dyes are mordant dyes, there is a substantial literature base describing dyeing techniques. The most significant mordant dyes for wool are synthetic mordant dyes, or chrome dyes; these constitute around 30% of wool dyes and are notably effective for black and navy shades. As an after-treatment, potassium dichromate is used as a mordant. It is crucial to highlight that certain mordants, particularly those containing heavy metals, can be dangerous to health and must be used with extreme caution. Examples (Fig 6) of mordant dyes are chrome alum 6(a), alum 6(b), tannic acid, and salts of aluminium, chromium, copper, tin etc.

Vat dyes

Vat dyes are basically insoluble in water and hence incapable of directly colouring fibres. However, reduction in alkaline liquor results in the dye's water-soluble alkali metal salt. This form is generally colourless, so in that case it is known as a Leuco dye, and has a strong affinity for textile fibres. Following oxidation, the original insoluble dye is reformulated. Indigo, the original vat dye, is responsible for the colour of denim. Vat dyes get their name from the vating procedure associated with their application. The structures of vat dyes are well known. They are transformed to a water-soluble form with cellulose affinity and then returned to their water-insoluble form, resulting in high wet-term stability. Indigo, a well-known natural dye, is included in the family of vat dyes. While indigo remains the most significant colourant for denim fabric, its very tiny size makes this vat dye particularly sensitive to removal during the laundry process, giving jeans a faded appearance even after only one wash. This emphasises the need of creating colourants for cellulosic substrates that have the features required to remain inside the polymer matrix when water swells the substrate. Indigo Fig. 7(a), Indanthrene 7(b), Hydro bluvat, are the examples of vat dyes

Sulphur dyes

Sulfur dyes are low-cost dyes used to produce dark hues in cotton. Dyeing is accomplished by immersing the cloth in a solution of an organic chemical and sulphide or polysulfide, generally a nitrophenol derivative. When the organic compound combines with the sulphide source, it produces dark colours that attach to the cloth. Sulfur Black 1, the most used dye by volume, lacks a well-defined chemical structure. Sulfur dyes get their name from the fact that sulphur is used in their manufacture. Sulfur dye structures are poorly characterised because their polymeric nature renders them unsuitable for traditional structure characterization techniques. Sulfur dyes are transformed to a water-





Monika Tyagi and Ankit Mahur et al.,

soluble form with cellulose affinity and then changed back to their water-insoluble form, providing high wet persistence. Sulfur dyes include (Fig. 8) Sulfur black 1,8(a), Indophenol8(b) etc.

Reactive dyes

Reactive dyes have a chromophore attached to a substituent capable of interacting directly with the fibre substrate. Because of the covalent bonds that bind reactive dye to natural fibres, they are among the most lasting of dyes. "Cold" reactive dyes, such as Procion MX, Cibacron F, and Drimarene K, are very easy to use because the dye can be applied at room temperature. Reactive dyes are by far the greatest option for dyeing cotton and other cellulose fibres at home or in the studio. Reactive dyes are the class of suitable colourant for cellulosic fibres. Their name is derived from the fact that they undergo the chemical reaction with cellulose to form a covalent bond. Reactive dyes made it possible to get previously unattainable bright wet-fast shades on cellulosic fibres. Reactive dyes (Fig. 9) include reactive red 1,9(a), reactive blue 19,9(b), reactive red 1809(c), reactive blue 4, reactive orange 5 etc.

Dispersed dyes

Disperse dyes are water-insoluble and were first developed for the dyeing of cellulose acetate. The dyes are finely powdered and marketed as a paste in the presence of a dispersing agent, or spray-dried and sold as a powder. Their primary application is in the dyeing of polyester, although they may also be used to colour nylon, cellulose triacetate, and acrylic fibres. A dyeing temperature of 130 °C (266 °F) is necessary in some cases, and a pressurised dye bath is employed. The very fine particle size provides a large surface area, which helps in dissolution to allow its absorption by the fiber. The dyeing rate can be greatly influenced by the dispersing agent employed during the grinding process. These dyes are applied to hydrophobic fibres, resulting in an aqueous dispersion. Disperse dyes detach from the reaction mixture as large particles. They are unsuitable for dyeing due to their huge particle size. They provide poor results in this form, thus to obtain good dyeing qualities, the dye must be employed in the form of fine powder and homogeneous, stable suspension. As a result, these colours are finely powdered in the presence of a dispersant before being offered as a paste or powder. The very tiny particle size provides a large surface area, which helps dissolving and allows fibre absorption. Disperse dyes are most commonly used to colour polyester, although they may also be used to colour nylon, cellulose triacetate, and acrylic fibres. Disperse dyes (Fig. 10) include Disperse blue 1,10(a), Disperse blue 3 10(b), Disperse orange 11, 10(c), Disperse orange 25 etc.

Azo dyes

Azo dyes are those that include one or more azo groups. This is the largest and most important class of synthetic colouring substance. These dyes are available in nearly all shades and have a wide variety of uses. They may be used on both natural and synthetic fabrics. Furthermore, many dyes in this class are applied on paper, rubber, leather, and a variety of other materials. Azo pigments and the lakes generated by these azo dyes are widely used in the paint, varnish, lacquer, textile, cosmetic, pharmaceutical, and food sectors. Different metal complexes play an important role in the field of azo compounds. Commercially, copper, chromium, and cobalt complexes of azo dyestuffs bearing metallizable substituents in at least one ortho position relative to the azo group (Fig. 11) or azo dyes produced from metallizable coupling components such as salicylic acid are employed. Many papers have been published on the importance and significance of these metal complex dyestuffs [20,21,22]. Copper and chromium metal complexes are widely used in cotton industries in combination with azo dyes. The azo chrome colours [23] are extremely important in the dyeing of wool. Mordant azo dyes are specifically useful for deep blue, violet, black, brown, and Bordeaux shades.

With chromium as a mordant shade, high-quality, all-around fastness qualities are achieved. Premetallized dye 1:1 chromium azo dyestuffs are being introduced (Fig.12). In 1919, the fast dyes Neolan by Ciba and Palatine by BASF got succeeded in improving the dyeing procedures and providing improved results in their industrial applications. However, these procedures had a significant disadvantage in that they required a strong acidic dye-bath, which destroyed the wool. Later, German dyestuffs manufacturers overcame this disadvantage to some extent in the 1930s





Monika Tyagi and Ankit Mahur et al.,

by using 2:1 chromium and cobalt complexes that required a mildly acidic dye-bath that did not harm the softness of wool [24].

Furthermore, they were suitable for dyeing nylon with a relatively neutral dye-bath. However, the primary challenge was their low solubility, which may be solved by using the Irgalan line of dyestuffs, which are 2:1 chromium and cobalt complex dyestuffs containing methyl sulphonyl, which increases their solubility. Later in 1949, a variety of neutral dyestuffs containing chromium and cobalt dyestuffs with non-ionic solubilizing groups sulphonamide were developed. Likewise, several asymmetric 2:1 chromium complexes comprising two different metallizable azo-dyestuffs with improved fastness and a wide variety of hues were developed. Initially, such dyestuffs were confined to wool and nylon, but with the inclusion of appropriate fibre reactive groups, paint superior fastness to light and wet treatments were accomplished. The azo group has weak donor properties and has not been observed to coordinate with a metal atom to form a stable complex unless the metal atom can be held within the dye molecule via chelation with the help of a complex-forming group such as -OH or -NH₂ in the ortho position to the azo group [25].

Chromium azomethine complexes [26,27], cobalt complex Schiff base [28], and un-symmetrical complex 1:2 chromium [29] dyes provide fast colours to leathers, food packaging, and wools etc. Azo groups with metal complexes [30] are used to dye cellulose polyester fabrics. To mass dye polyfibers, certain metal complexes [31] are utilised. The cobalt complex of a Schiff base (salicylaldehyde with diamine) has exceptional light resistance and storage properties, and it does not deteriorate even in acidic gases (CO₂). A novel tetra dentate Schiff base is used as a chromogenic reagent to determine Ni in natural food samples [32].

CONCLUSION

In conclusion, Schiff base metal complexes play a crucial role in the advancement of modern dyes and paints. Their vibrant colors, stability, and functional properties make them indispensable in a wide range of applications. The ability to synthesize and customize these complexes further enhances their value, providing industries with the tools to innovate and meet specific needs. As the demand for durable, versatile, and environmentally friendly dyes and paints continues to grow, Schiff base metal complexes will undoubtedly remain at the forefront of this dynamic field.

REFERENCES

1. Gaikwad, K.V. and Yadav, M.U. 2016. Metal complexes of Schiff Bases. *Scholarly Research Journal for Interdisciplinary Studies* 3/24:2225-34. <http://www.srjis.com/pages/pdfFiles/146771229523.VIJA%20GAIKWADpaper22.pdf>
2. Abuamer, K. M., Maihub, A.A., El-Ajaily, M.M., Etoriki, A.M., Abou-Krishna, M.M. and Almagani, M.A. 2014. The Role of Aromatic Schiff Bases in the Dyes Techniques. *International Journal of Organic Chemistry* 4, no. 1: 7-15. <http://dx.doi.org/10.4236/ijoc.2014.41002>
3. El-Ajaily, M., Maihub, A., Etoriki, A., Ben-Saber, S. and Elmajani, M. 2013. Coordination behavior of phenolic and benzylic hydroxyl groups in complexation of their Schiff bases with Zn(II) and Cd(II) ions. *Journal of Pharmaceutical and Biomedical Sciences* 28, no. 22 (March):695-97. https://www.researchgate.net/publication/299437097_Coordination_behavior_of_phenolic_and_benzylic_hydroxyl_groups_in_complexation_of_their_Schiff_bases_with_ZnII_and_CdII_ions
4. Singh, M., Singh, P. and Gupta, A. 2009. Facile one-pot synthesis of tricyclic dibenzodioxazine-2-ones and other higher homologues. *Arkivoc* 7:189-96. <http://dx.doi.org/10.3998/ark.5550190.0010.718>
5. Maihub, A., El-Ajaily, M. and Hudere, S. 2007. Synthesis and spectral studies of some transition metal complexes of Schiff base. *Asian Journal of Chemistry* 19, no. 1: 1-4. https://asianjournalofchemistry.co.in/User/ViewFreeArticle.aspx?ArticleID=19_1_1
6. El-Ajaily, M. and Maihub, A. 2010. Synthesis and spectroscopic characterization of Mn(II)-benzoin complex. *International Journal of Chem Tech Research* 2, no.3: 1579-80. [http://sphinxsai.com/july-sept_2010_vol2.3/chemtech/chemtechvol2.3july-sept210/CT=33%20\(1579-1580\).pdf](http://sphinxsai.com/july-sept_2010_vol2.3/chemtech/chemtechvol2.3july-sept210/CT=33%20(1579-1580).pdf)



**Monika Tyagi and Ankit Mahur et al.,**

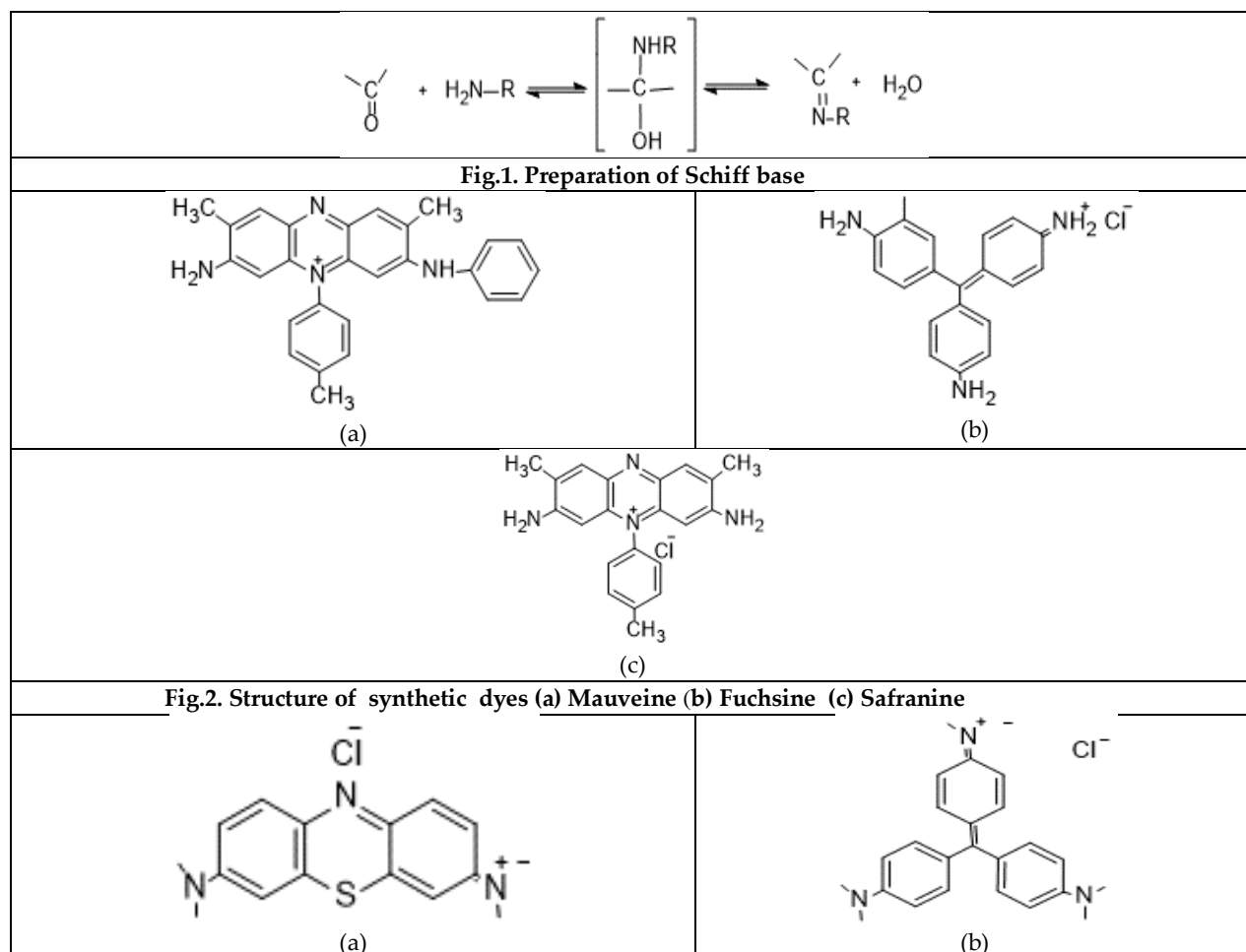
7. Mehta, S.B., Zala, R.V. and Dave, P.N. 2008. Mordant dyes derived from 4-(2-phenyl-4-oxo-3-quinazoliny)-4'-amino diphenyl sulfone and its M(II)-Schiff base complexes. *Int. J. Chem. Sci.* 6, no.1: 185-91. <https://www.tsijournals.com/articles/mordant-dyes-derived-from-42phenyl4oxo3quinazoliny4-amino-diphenyl-sulfone-and-its-studies-as-mii--schiff-base-complexe.pdf>
8. Rai, B.K and A. Baluni. 2001. Coordination compounds of Co(II), Ni(II) and Cu(II) with thiosemicarbazone of a series of quinazolone derivatives: Their preparation, characterization and structural investigation. *Asian J. Chem.* 13, no. 2: 725-29. https://asianjournalofchemistry.co.in/user/journal/viewarticle.aspx?ArticleID=13_2_57
9. Booth, G. 2000. Dyes, General Survey. *Ullmann's Encyclopedia of Industrial Chemistry*. Wiley-VCH. doi:10.1002/14356007.a09_073. ISBN 3527306730.
10. Srivastava, R. and Singh, N. 2019. Importance of natural dyes over synthetic dye: a critical review. *International Journal of Home Science*. 5, 2:148-50. <https://www.homesciencejournal.com/archives/2019/vol5issue2/PartC/5-2-29-167.pdf>
11. Hubner, K. 2006. History – 150 Years of mauveine. *Chemie in unserer Zeit*. 40, no. 4: 274–75. doi:10.1002/ciuz.200690054.
12. Travis, A.S.1990. Perkin's Mauve: Ancestor of the Organic Chemical Industry. *Technology and Culture* 31 no.1: 51–82. doi:10.2307/3105760
13. Slama, H. B., Bouket, A.C., Pourhassan, Z., Alenezi, F.N., Silini, A., Cherif-Silini, H., Oszako, T., Luptakova, L., Golinska, P. and Belbah, L. 2021. Diversity of Synthetic Dyes from Textile Industries, Discharge impacts and treatment methods. *Appl. Sci.* 11, 6255. <https://doi.org/10.3390/app11146255>
14. Hunger, K., ed. 2007. Industrial Dyes: Chemistry, properties, applications. John Wiley & Sons. <https://www.wiley.com/en-us/Industrial+Dyes:+Chemistry,+Properties,+Applications-p-9783527304264>
15. Chaudhary B. 2020. Chemistry of synthetic dyes: A review. *Journal of Interdisciplinary Cycle Research* 12, no. 11 (November): 390-96. <http://www.jicrjournal.com/gallery/41-jicr-november-3487.pdf>
16. Gregory, P.1990. Classification of dyes by chemical structure. In *The Chemistry and Application of Dyes*, 17-47, Springer, Boston, MA. https://doi.org/10.1007/978-1-4684-7715-3_2
17. Bafana, A. Devi, S.S. and Chakrabarti, T. Azo dyes: past, present and the future. *Environmental Reviews* 19 (NA): 350–71. doi:10.1139/a11-018
18. Abraham, E.N. 1977. *Dyes and their Intermediates*. New York: Chemical Publishing, pp. 1–12. <https://monographs.iarc.who.int/wp-content/uploads/2018/06/mono99-7.pdf>
19. Saikhom, J. D., Salam, J. S., Potshangbam, K. S., Choudhury, M.D. and Maibam, H. D. 2013. Biochemical Studies in Some Indigenous Dye Yielding Plants of Manipur. *Notulae Scientia Biologicae* 5, no.3: 303-308. <https://doi.org/10.15835/nsb539078>
20. Ventura-Camarago, B.D.C. and Marin-Morales, M.A.2013. Azo Dyes: Characterization and Toxicity– A Review. *Textiles and Light Industrial Science and Technology* 2 no.2 (April):85-103. https://www.researchgate.net/publication/282815745_Azo_Dyes_Characterization_and_Toxicity_-_A_Review
21. Achwal, W.B. 1997. Problems during analysis of textile as per ecostandards and the customer articles ordinance (Part I). *Colourage* 44, no. 5: 29-31, <https://jglobal.jst.go.jp/redirect?url=http%3A%2F%2Fjdream3.com%2Ffp%2Fjglobal%2Findex.html%3FdocNo%3D97A0789209%EF%BC%86from%3DJ-GLOBAL%EF%BC%86jstjournalNo%3DC0102A&sign=bca1d13207ada51ad9e7ca88f6bf2309>
22. Burkinshaw, S.M. and Gotsopoulos, A. 1996. The pre-treatment of cotton to enhance its dyeability-I. Sulphur dyes. *Dyes and Pigments* 32, no. 4 (December): 209-28. [https://doi.org/10.1016/S0143-7208\(96\)00026-5](https://doi.org/10.1016/S0143-7208(96)00026-5)
23. Patel, H.M. and Dixit, B.C. 2014. Synthesis, characterization and dyeing assessment of novel acid azo dyes and mordant acid azo dyes based on 2-hydroxy-4-methoxybenzophenone-5-sulfonic acid on wool and silk fabrics. *Journal of Saudi Chemical Society* 18, no.5 (November): 507-12. <https://doi.org/10.1016/j.jscs.2011.10.015>
24. Prakash A. and Adhikari, D. 2011. Application of Schiff bases and their metal complexes – a review. *Int. J. Chem Tech Res.* 3, no. 4 (October-December):1891-96. [https://sphinxsai.com/Vol.3No.4/chem/pdf/CT=26\(1891-1896\)OD11.pdf](https://sphinxsai.com/Vol.3No.4/chem/pdf/CT=26(1891-1896)OD11.pdf)
25. Kumar S., Dhar, D. N. and Saxena, P.N. 2009. Applications of metal complexes of Schiff bases-A review. *Journal of Scientific & Industrial Research* 68 (March): 181-87. [http://nopr.niscair.res.in/bitstream/123456789/3170/1/JISIR%2068\(3\)%20181-187.pdf](http://nopr.niscair.res.in/bitstream/123456789/3170/1/JISIR%2068(3)%20181-187.pdf)





Monika Tyagi and Ankit Mahur et al.,

26. Long, R.S. 1947. Chromium Complexes of azo dyes derived from acylacetoneitriles. *J. Am. Chem. Soc.* 69, no. 5 (May): 990–95. <https://doi.org/10.1021/ja01197a004>
27. Agrawal, S. 2016. Drug metal complexes in research-A review. *International Journal of Science, Management and Engineering Research* 1, no. 1 (February):10-11. <http://ejournal.rems.co.in/papers/IJSMER201603.pdf>
28. Aiyelabola, T.O. 2021. Syntheses, characterization and biological activity of novel thiazoylazo dye and its coordination compounds. *Advances in Biological Chemistry* 11, no. 5 (October): 179-205. doi: 10.4236/abc.2021.115013.
29. Befta. Unsymmetrical 1:2 chromium complex dyes. Fabio(to Ciba Geigy AG), Eur Pat Abstr, 1985, 103, 161871. <https://patentimages.storage.googleapis.com/85/4f/d9/a09f5bc875ee31/EP0148120B1.pdf>
30. Gurbuz, D. 2020. Dyeing polyester, cotton and wool with some Schiff bases derived from 4-chloroaminophenol and various benzaldehyde. *JOTCSA* 7, no. 2:463-70. <https://doi.org/10.18596/jotcsa.714635>
31. Khanmohammadi, H. and Darvishpour, M. 2009. New azo ligands containing azomethine groups in the pyridazine-based chain: synthesis and characterization. *Dyes and Pigments* 81: 167-73. <https://doi.org/10.1016/j.dyepig.2008.07.019>
32. Fakhari, A.Khorrami, A. R. and Naeimi, H. 2005. Synthesis and analytical application of a novel tetradentate N₂O₂ Schiff bases as a chromogenic reagent for determination of nickel in some natural food samples. *Talanta* 66, no.4 (May) : 813-17. <https://doi.org/10.1016/j.talanta.2004.12.043>





Monika Tyagi and Ankit Mahur et al.,

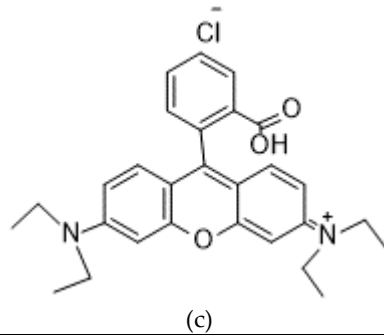


Fig. 4 Structure of basic dye(a) Methylene blue (b)Crystal voiltet (c)Rhodamine B

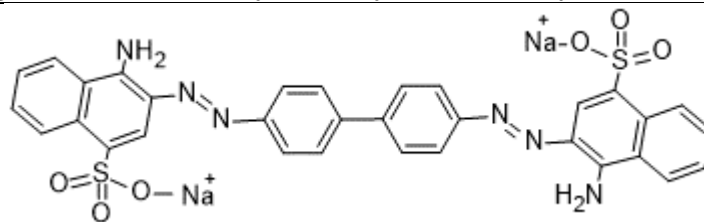


Fig. 5 Structure of Congo red

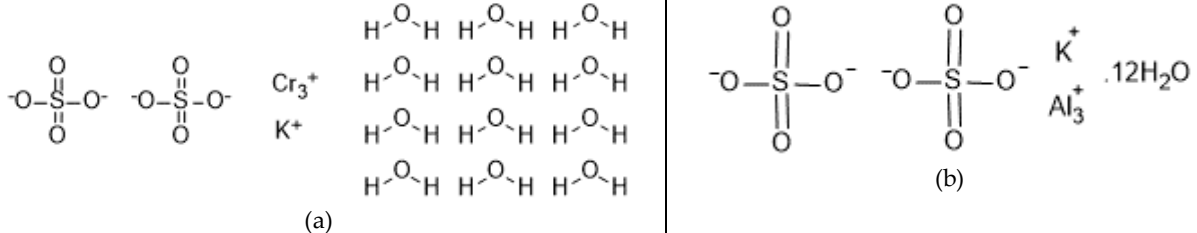


Fig. 6. Structure of mordant dyes (a) Chrome alum (b) salts of aluminium

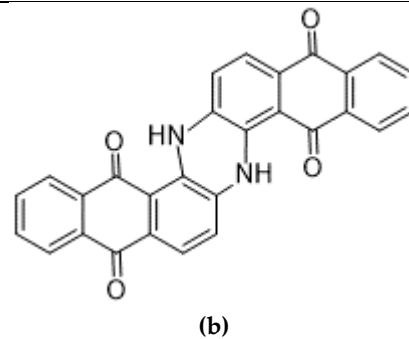
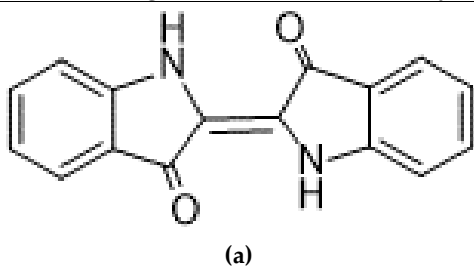


Fig. 7 Structure of vat dyes(a) Indigo (b)Indanthrene

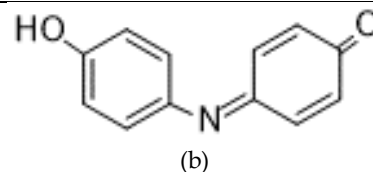
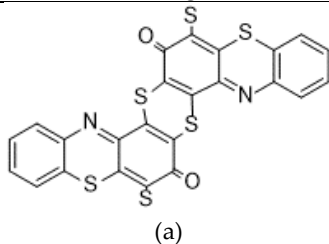


Fig. 8 Structure of Sulphur dyes(a)Sulfur black 1 (b) Indophenol





Monika Tyagi and Ankit Mahur et al.,

<p>(a)</p>	<p>(b)</p>	
<p>(c)</p>		
<p>Fig. 9. Structure of reactive dyes (a) reactive red 1 (b) reactive red 180 (c) reactive blue 4</p>		
<p>(a)</p>	<p>(b)</p>	<p>(c)</p>
<p>Fig. 10 structure of (a) Disperse blue 1 (b) Disperse blue 3 (c) Disperse orange 25</p>		
	<p>(a)Arylazosalicylic acid</p>	<p>(b)Arylazo-8-hydroxyquinoline</p>
<p>Fig. 11 Where, X= -OH, -NH₂, -OH, -OH, -OH, -OH and Y= -H, -H, -OH, -COOH, -NH₂, -CH₂COOH respectively</p>		
<p>(c) Arylazosalicyldoxime</p>	<p>Fig. 12 Structure of chromium azo dye</p>	





A Comprehensive Overview of the World Health Organization

S.M. Sakthisankaran¹ and K. Kathiresan^{2*}

¹M.Pharm Student, Department of Pharmacy, Annamalai University, Annamalai Nagar, Tamil Nadu, India

²Associate Professor, Department of Pharmacy, Annamalai University, Annamalai Nagar, Tamil Nadu, India

Received: 10 Jun 2024

Revised: 18 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

K.Kathiresan,

Associate Professor,

Department of Pharmacy,

Annamalai University,

Annamalai Nagar, Tamil Nadu, India

E. Mail: sakshiman222@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The World Health Organization [WHO], a public health organization of the United Nations[UN], was founded in Geneva in 1948. Its goal is to help all individuals reach "the highest possible level of health". It sets worldwide sanitary standards and quarantine guidelines, serving as a repository for information on the most recent advancements in illness and medical treatment. It supports efforts to manage endemic and epidemic diseases, such as immunization programmes and aiding in the provision of clean water supplies. Finally, it also promotes the enhancement of public health initiatives within the member countries. Up until this point, its most significant accomplishment has been the global eradication of smallpox. The World Health Assembly [WHA], which represents the 192 WHO member nations, is in charge of the organization. WHO member state delegates make up the Health Assembly. This body approves the WHO's budget and programme and makes major policy decisions.

Keywords: WHO; Objectives of WHO; Constitution of WHO; Functions of WHO

INTRODUCTION

WHO, a UN specialised organization formed in 1948, is dedicated to encouraging global collaboration to enhance public health. With particular responsibilities transferred from the League of Nations Health Organization [1923] and the International Office of Public Health in Paris [1907], the WHO assumed a broad mandate as stated in its constitution: promote the best health for all people. The WHO defines health positively as a condition characterized by total physical, mental, and social well-being, rather than simply the absence of disease or infirmity. WHO observe world Health Day on April 7, 1948, the anniversary of the organization's founding. The majority of the agency's



**Shanmugam Manoharan Sakthisankaran and Krishnasamy Kathiresan**

funding comes from yearly payments given by member nations according to their respective means of subsistence. Furthermore, WHO received significant funding from the UN's enlarged technical assistance programme after 1951[1,2]. The 192 member states of the WHO comprise the World Health Assembly [WHA], which exercises governance over the WHO. The executive council of the WHO is composed of 34 health specialists and is elected and advised by the organization [3,4]. It appoints the chief executive, who is presently Dr.Tedros Adhanom Ghebreyesus of Ethiopia,, establishes objectives while also granting financial and operational permissions [5].The WHO receives the majority of its funding from assessed and voluntary contributions from member states; private donors contribute the remainder. The fiscal year's authorized budget 2020–2021 exceeds \$7.2 billion, whereas the budget for the fiscal year 2022–2023 surpasses \$6.2 billion [6]. In order to control epidemic and endemic disease, the WHO funds national immunization campaigns, antibiotic and insecticide training, improved lab and clinic facilities for early diagnosis and prevention sanitation systems and rural health education. These programs achieved some success in the battle against AIDS, TB, malaria, and other ailments [7]. The WHO regularly conducts field surveys, sends out teams of experts from around the world to conduct demonstration projects and fieldwork, support for the creation of national training facilities for nurses and doctors, as well as aid with the development of neighbourhood health centres. Additionally, it promotes the growth and improvement of public health administrations across its member countries. WHO is capable of furnishing fellowship funds to medical professionals, public health administrators, sanitary inspectors, nurses, laboratory technicians and researchers via a variety of education assistance programs [8].

Aim and Objectives

The WHO goal is for all peoples to achieve the best possible state of health [Figure 1]. The goals of WHO's leadership objectives were:

- Promoting nations' efforts to get closer to achieving universal health care
- Assisting nations in demonstrating their ability to follow international health regulations
- Improving accessibility to necessary and superior medical supplies
- Examining how social, economic, and environmental variables affect public health
- Bringing non-communicable diseases under coordination [9].

Constitution of WHO

The states parties to this constitution declare that the following ideas are essential to everyone's pleasure, peaceful coexistence, and security in line with the United Nations Charter. Everyone has the right to good health, regardless of social, political, religious, or economic status. Achieving peace and security depends on ensuring the health of all peoples, which calls for the best possible cooperation between individuals and countries. Every state that succeeds in promoting and protecting health benefits everyone. Unequal progress in promoting health and controlling illness, particularly communicable diseases, throughout nations is a common threat. Children's health depends on peaceful living in a changing environment [10].

Functions of WHO

In order to achieve the objectives, the functions of the WHO shall be

- To serve as the international health work's guiding and coordinating authority.
- To support governments in improving health care upon request
- To provide governments with the required help and suitable technical support at their request.
- To promote and further efforts to eliminate endemic, epidemic, and other illnesses.
- To encourage collaboration between professional and scientific communities that progress health.
- Supporting and carrying out health-related research.
- To encourage higher training and teaching standards in the medical, health, and allied fields.
- To create and update worldwide nomenclatures for illnesses, causes of mortality, and public health procedures as needed.
- Standardising diagnostic processes where required.
- To create, implement, and advance international standards for goods that are related to food, biological, pharmaceutical, and other industries.



**Shanmugam Manoharan Sakthisankaran and Krishnasamy Kathiresan**

- In general, to take all required steps to achieve the organization's goal [11,12].

Governance

The assembly is the WHO's principal decision-making body. May is the traditional meeting month for representatives from all 192-member nations in Geneva. Its primary duty is to draft the organization's policies. The planned program budget is approved or rejected by the Health Assembly, which also selects the Director-General. The organization's financial policies are under the direct supervision of the Director-General. It also takes into account the findings from the Executive Board, which it advises on issues requiring additional research, analysis, evaluation, or communication. There are 32 members of the executive board that are technically competent in the health profession. Terms of office for members are three years. The primary duties of the board are to advise the health assembly, implement its policies and decisions, and generally facilitate its operation. Approximately 3500 health and other specialists, as well as support personnel, work for the WHO on fixed-term contracts at the organization's headquarters, six regional offices, and in other countries [11,13].

Director Generals of WHO

World Health Assembly [WHA] appoints organization's director general. Assembly nominates five-year directors general in May in typical manner. Canadian doctor Brock Chisholm was the organization's first director general from 1948 until 1953. The following directors general of the WHO are Gro Harlem Brundtland [1998–2003], Lee Jong-Wook [2003–2006] and Margaret Chan [2007–17] [14]. Dr. Tedros Adhanom Ghebreyesus, an Ethiopian professional in public health, was appointed director general of the WHO in 2017. He officially began his second term on August 16, 2022, after being re-elected. A director general may be nominated again just once [5].

Health Policy

The WHO endeavours to address health disparities by implementing policies and initiatives that prioritize equity, gender responsiveness, and human rights. Additionally, it focuses on fostering a healthier environment, emphasizing primary prevention, and influencing cross-sectoral policies to address underlying causes of environmental health risks. Furthermore, the organization provides evidence-based standards, guidelines, and tools to aid member states in formulating health policies, along with publishing comprehensive reference classifications and ensuring the implementation of the International Health Regulations [15,16].

Awareness and Action Regarding Public Health

The WHO defines health education as the process of allowing individuals to raise their level of healthy self-management. It emphasizes social and environmental issues above personal actions. Every year, the organization celebrates World Health Day and conducts health promotion events [Figures 2]. Annually, April 7th marks World Health Day in honor of the organization's founding anniversary. Other internationally recognized public health programs commemorated by the WHO include World Tuberculosis Day, Immunization Week, No Tobacco Day, AIDS Day, and Hepatitis Day. The WHO awards medals and prizes for outstanding contributions to public health. The winners are chosen by the WHO Executive Board and announced during the WHA [17].

Financing and Partnerships

Foreign donors and member nations provide financial support to the WHO. The leading donors in 2020–21 included Germany, Japan, China, United States, the Bill & Melinda Gates Foundation, the GAVI alliance, the World Bank, and Rotary International. Advisory Committee on Sustainable Finance was established by the WHO Executive Board in 2021 with the goal of reconsidering WHO's financial approach and submitting suggestions. The WHA of 2022 endorsed its proposals, the most important of which was to increase mandatory member dues to 50% of WHO's basic budget for 2022–2023 by the end of the 2020s [6,18].

Regional Offices

The WHO is a specialized UN agency in charge of global public health. Geneva is the headquarters of 150 field offices and 6 regional offices worldwide.



**Shanmugam Manoharan Sakthisankaran and Krishnasamy Kathiresan**

Operating areas and regional offices of WHO:

- Africa; Headquartered in Brazzaville, Republic of the Congo
- Western Pacific; Headquartered in Manila, Philippines
- South East Asia; Headquartered in New Delhi, India
- Eastern Mediterranean; Headquartered in Cairo, Egypt
- Americas; Headquartered in Washington, D.C., US
- Europe; Head quartered in Copenhagen, Denmark [19,20].

CONCLUSION

The WHO's stated mission in its constitution is to ensure that all peoples achieve the utmost level of health possible. Ensuring universal access to the advantages of medical, psychological, and related knowledge is critical for achieving optimal health. Active public participation and well-informed opinion are crucial factors in advancing the collective health of the populace. WHO promotes and aids the vulnerable while maintaining global health and safety. The organization promotes health research and policy, sets worldwide health standards, and gathers global health statistics. The organization also helps countries technically. The official World Health Report evaluates global health concerns. The WHO has helped develop an Ebola vaccine, practically eradicate polio, and eliminate smallpox. Non-communicable diseases, including cancer and heart disease; nutrition, health, and food security; substance abuse; and communicable diseases [Ebola, HIV/AIDS, tuberculosis and malaria] are current WHO priorities. The organization supports the health care universalization, active observation of public health risks, coordination of reactions to health crises, and the overall promotion of health and well-being. Thus, in order to promote health, assure global security, and aid the vulnerable, WHO operates on a global scale.

REFERENCES

1. Hendel N. World Health Organization [WHO]. In International Conflict and Security Law: A Research Handbook, The Hague: TMC Asser Press; 2022.p. 733-759.
2. Cueto M, Brown TM, Fee E. The World Health Organization: A History. Cambridge University Press; 2019.
3. Fitzsimons DW. World Health Organization. Acta Med Port, 2013;26[3]:186-7.
4. Silberschmidt G, Matheson D, Kickbusch I. Creating a committee C of the World Health Assembly. Lancet, 2008;371[9623]:1483-6.
5. World Health Assembly re-elects Dr Tedros Adhanom Ghebreyesus to second term as WHO Director-General [2022].
6. Reddy SK, Mazhar S, Lencucha R. The financial sustainability of the World Health Organization and the political economy of global health governance: a review of funding proposals. Global Health, 2018;14[1]:119.
7. Forss K, Stenson B, Sterky G. The future of global health cooperation: designing a new World Health Organization. Curr Issues Public Health, 1996;2[3]:138-42.
8. Meier BM, Gostin LO. A Timely History: Examining the History of the World Health Organization to Frame the Future of Global Health Governance. Am J Public Health, 2020;110[11]:1592-1594.
9. Godlee F. The World Health Organisation. Global Health, 2017.
10. Constitution of the World Health Organization. Am J Public Health Nations Health, 1946;36[11]:1315-23.
11. Ruger JP, Yach D. The Global Role of the World Health Organization. Glob Health Gov, 2009;2[2]:1-11.
12. Kamal-Yanni M, Saunders P. Urgent need for WHO's reform to prioritise core functions. Lancet, 2012;379[9829]:1878.
13. Lidén J. The World Health Organization and global health governance: post-1990. Public Health, 2014;128[2]:141-7.
14. Horton R, Samarasekera U. WHO's Director-General candidates: visions and priorities. Lancet, 2016;388[10056]:2072-2095.





Shanmugam Manoharan Sakthisankaran and Krishnasamy Kathiresan

15. Ooms G, Marten R, Waris A, Hammonds R, Mulumba M, Friedman EA. Great expectations for the World Health Organization: a Framework Convention on Global Health to achieve universal health coverage. *Public Health*, 2014;128[2]:173-8.
16. World Health Organization. "World report on health policy and systems research", 2017.
17. World Health Organization: WHO campaigns, 2013.
18. Gostin LO, Klock KA, Clark H, et al. Financing the future of WHO. *Lancet*, 2022;399[10334]:1445-1447.
19. Cueto M. The World Health Organization. In *Global Health Essentials*, Cham: Springer International Publishing, 2023: 421-424.
20. Sridhar D, Gostin L. World Health Organization: past, present and future. *Public Health*. 2014;128[2]:117-8.





Wearable Devices and Digital Health Platforms: Forensic Analysis and Data Security Challenges

Swagata Shashikant Zarkar¹, Rahul Kailas Bharati² and Shobha Kamalakar Bawiskar^{3*}

¹PG-Research Scholar, Department of Digital And Cyber Forensics, Government Institute of Forensic Science, (Affiliated to Dr.Babasaheb Ambedkar Marathwada University), Chhatrapati Sambhajnagar, Maharashtra, India.

²Assistant Professor, Department of Law, Government Institute of Forensic Science, (Affiliated to Dr.Babasaheb Ambedkar Marathwada University), Chhatrapati Sambhajnagar, Maharashtra, India.

³Assistant Professor, Department of Digital And Cyber Forensics, Government Institute of Forensic Science, (Affiliated to Dr.Babasaheb Ambedkar Marathwada University), Chhatrapati Sambhajnagar, Maharashtra, India.

Received: 17 Jun 2024

Revised: 14 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

Shobha Kamalakar Bawiskar

Assistant Professor, Department of Digital and Cyber Forensics,
Government Institute of Forensic Science,
(Affiliated to Dr.Babasaheb Ambedkar Marathwada University),
Chhatrapati Sambhajnagar, Maharashtra, India.
E. Mail: shobha_bawiskar@yahoo.co.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Smart watches are wearable gadgets that combine the functionalities of a standard watch with those of a smart phone. They usually have a touch screen display, wireless connectivity, and the ability to run apps, track fitness data, and receive notifications from an associated smart phone. Smart watches can perform a variety of duties, depending on the type and software loaded. This article concentrated on health indicators such as heart rate monitoring, breathing activity, sleep quality, and physical activity. This research article seeks to investigate the many factors linked to the usage of smart watches in the domain of health. The effectiveness of health parameters indicated in hardware and software is studied to get insights into the accuracy and usefulness of these metrics.

Keywords: Wearable Technology, Health Tracking, Fitness Monitoring, Smart Watches, Device Security

INTRODUCTION

Forensic science is the use of scientific tools and techniques to investigate crimes and other legal situations. Smartwatches, on the other hand, are wearable devices that may monitor physiological and physical data such as



**Swagata Shashikant Zarkar et al.,**

heart rate, activity levels, and sleep patterns. The possible use of smartwatch data as evidence in criminal investigations connects forensic science and smartwatch readings. For example, if a crime occurred at a given time and location, smartwatch data from a suspect or victim could reveal vital evidence about their whereabouts and activities at that time. Similarly, if a suspect claims to have been asleep at the time of the crime, their smartwatch sleep data might potentially corroborate or contradict their alibi.

However, it is crucial to emphasize that the use of wristwatch data as evidence in judicial disputes is still relatively new, and there are various challenges to be addressed, such as data accuracy and privacy concerns. Furthermore, the admissibility of such evidence in court may be contingent on the specific rules and regulations of a certain jurisdiction. Smartwatches have gone a long way since their introduction in the early 2000s. Microsoft released the first smartwatch, the SPOT watch, in 2004. It was a wristwatch that used FM radio signals to display news, sports scores, and other information. The SPOT watch, on the other hand, was not widely adopted due to its limited functionality and high price [1]. Smartwatches are wearable gadgets that have gained popularity in recent years due to their capacity to perform a variety of purposes such as fitness tracking, messaging, and phone calls, among others. A smartwatch often contains a touch screen display, wireless connectivity, and sensors that can track physical activities and health data. With technological advancements, smartwatches are getting more complex, with capabilities such as voice assistants, mobile payments, and standalone cellular access. [2] According to Allied Market Research, the global smartwatch market was valued at \$20.64 billion in 2019 and is expected to reach \$96.31 billion by 2027, rising at a CAGR of 19.7% from 2020 to 2027 (Allied Market Research, 2020). The increasing acceptance of wearable devices, as well as people's growing awareness of health and fitness, are some of the factors driving the growth of the smartwatch industry [2]. General information about selected smartwatch for this research work is shown in Table No [1]. Its images are shown in Fig no [1].

Images of smartwatch used for study**Health care Sector vis- a vis data breach**

Health care security refers to the policies and procedures in place to safeguard the confidentiality, integrity, and availability of sensitive personal and medical information of patients and health care providers. This involves security against data breaches, cyber-attacks, theft, and unauthorised access to medical records. [9,10] As electronic health records (EHRs) and other digital technologies are used to store and transmit sensitive patient information, cyber security has become increasingly critical in the healthcare industry. The Health Insurance Portability and Accountability Act (HIPAA) and other requirements require health care organisations to protect patient data, and failing to do so can result in costly data breaches and legal action. Cyber security in healthcare include guaranteeing the confidentiality, integrity, and availability of patient data, as well as safeguarding medical devices and systems from external threats [11, 12]

Legal implications of wearable devices and digital health platforms

When you buy a wearable device, you input all of your information into it, and whether or not the data saved is secure becomes a concern. Whether such data is under the control of the device's manufacturer or the service provider. These questions are critical in determining the threats to the privacy rights of individuals storing their data in such devices. In the case of *K.S. Puttaswamy v. Union of India*, the Supreme Court ruled that the right to privacy is a basic right protected by Articles 14, 19, and 21 of the Indian Constitution. [13] Wearable device and social media network users may not consider themselves to have volunteered data, yet their actions of use and engagement result in the collection of large volumes of data about individual lifestyles, choices, and preferences. Medical information is one type of data that has a reasonable expectation of privacy. This demonstrates that the Court believes in securing medical information about an individual and that it is reasonable for an individual to expect its right to privacy to be violated by anyone. The Digital Information Security in Healthcare Act 2018, often known as DISHA, was passed by the Ministry of Family and Health Welfare. This law would address data owners' rights as well as the gathering and processing of health data. It has also formed a panel of digital health authorities comprised of a National Authority and other State bodies. It focuses on data collected consensually at medical facilities, but it also addresses alternative ways in which medical data is generated.[14] The Information Technology Act of 2000 is the major piece of legislation





Swagata Shashikant Zarkar et al.,

in India that governs data protection issues. It is the first legislation that addressed the challenges and, in terms of privacy, developed a notice and consent paradigm. It also imposed fines for violations of data privacy. The Electronic Health Record Standards 2016 are recommended but not legally binding. Many businesses include these in their client data protection policies. In the absence of any national or international norms specific to wearable technologies, it is suggested that wearable technology and service providers have reasonable security practices and procedures in place to protect sensitive personal data and personal information of the user.[15]

Forensic significance

Forensic significance of healthcare data: Smart watch security is an important consideration that must be treated seriously to safeguard the user's safety and privacy. Smartwatches, like any other computing device, have security vulnerabilities that must be addressed to preserve the user's safety and privacy. Data Privacy, Authentication, Secure Communication, App Security, Physical Security, and Software Updates are the security components of smartwatches that must be considered.[16,17,18,19]

Forensic significance of smartwatch: Due of the abundance of data they can supply, smartwatches can be important instruments in forensic investigations. Smartwatches, in particular, can provide location data, heart rate tracking, and biometric data that can be utilised in investigations. Smartwatch location tracking features can be utilized to reconstruct a suspect's movements and give evidence in a criminal investigation. [20] Smartwatch heart rate monitoring data can be utilised to corroborate an individual's alibi or dispute a suspect's assertions. [21] Finally, biometric data gathered by smartwatches, such as fingerprints or facial recognition, can be useful in forensic investigations. [22] Overall, smartwatches can give valuable forensic evidence in investigations and should be considered while conducting investigations.

METHODOLOGY

Aims and objectives: To investigate the effectiveness of health measures indicated in hardware such as smartwatches and software.

We determined to work on these hardware wearable devices: Fire Bolt BSW007, Gadgetzone I8 Pro Max, Gizmore GIZFIT 907, Noise Color Fit Pro 2 and Boat Wave Beat, as well as five software applications related to health: Heart Rate Monitor-Pulse App, Step counter-Pedometer, GoogleFit: Activity Tracking, Blood Pressure Measurement App, and Samsung Health. Certain basic data such as Blood Pressure (BP), Heart Rate, Blood Oxygen, and some training parameters such as cycling, skipping, and so on are measured for 15 days utilising the aforementioned hardware wearables. Software health programmes measure heart rate, step count, blood pressure, and calories count for 5 days. A detailed workflow is shown in Fig No.[2]

Proposed Methodology

Show the Fig.[2] -Workflow approach of proposed exploratory research

Experimental Methodology

Sample size–06 persons with one Smart watch from six different companies were selected.

Age Group - between the ages of 18 and 25 was chosen where,

Population Inclusion - The general population aged 18 to 25 years.

Population Exclusion - We excluded pregnant ladies, HIV patients, cancer patients, and anyone with any form of condition or sickness.

Process- For a period of 15 days (25.02.2023), the health parameters were examined at any time every day.

This investigation concentrated on both hardware and software. In terms of hardware devices, health parameters selected for investigation are classified into two categories: basic parameters and training parameters. Observations are shown in Table No. [2] For basic parameters and Table No.[3] for training parameters. As far as software is concerned, Table No.[4] Displays the Software application reading.



**Hardware Devices****Basic Parameters**

1. Blood Pressure(diastolic, systolic mm/hg(millimeter of mercury)), 60/90
2. Heart Rate BPM(Beats per minute) , 60-100 bpm
3. sleep(hours and minutes), 8 hours
4. walking steps(steps), target 7000 steps/day
5. Blood oxygen Level(% percentage),95-100%
6. Outdoor running, mileage as per Time(Hrs and Min), Pace(min/km), consumption of calorie(Kcal)
7. Female Menstrual Cycle Tracking,
8. Drink water reminder

Training Parameters

1. Cycling (bpm and KCAL)
2. Swimming
3. Football(steps, bpm and KCAL)
4. Skipping (bpm and KCAL)
5. Badminton (steps, bpm and KCAL)
6. Basketball (steps, bpm and KCAL)

Software Application

1. Heart Rate Monitor-Pulse App
2. Step counter-Pedometer
3. GoogleFit: Activity Tracking
4. Blood Pressure Measurement App
5. Samsung Health

Basic parameters reading of smartwatch:

Show the details in Table.2

Training parameters reading of smartwatch:

Observation table for software apps The Table No [5] below compares medical expert readings to one-day smartwatch readings. On 18 April 2023, the readings are recorded.

CONCLUSION

Finally, this article work provides useful insights into the forensic investigation of smartwatches. The study emphasizes the need of taking into account the health parameters shown in smartwatches and software, as well as the security features they provide. It is critical to do forensic analysis on smartwatches in order to analyses any occurrences that occur while wearing them. This study emphasizes the need of forensic scientists in reviewing available digital data to identify critical evidence in criminal investigations. The project work emphasizes the importance of continuing to engage in forensic science research in order to keep up with the rapid development of wearable technology devices such as smartwatches. This study was conducted on a typical population; we eliminated pregnant women, HIV patients, people with deviant behavior, and anyone with any type of disease. Smartwatches also capture data about the wearer's health and fitness, such as heart rate, sleep habits, and activity levels. This data can be utilized to establish the wearer's physical status at a given time and may be significant in cases involving physical assaults or accidents. Hardware and software conclusion: The outcomes of this study will be based on the effectiveness of health parameters indicated in hardware such as smartwatches and software. Depending on the data, the conclusion could be that these devices and software are helpful in tracking and monitoring health metrics, or that they are not dependable enough to offer accurate readings. The conclusion would almost certainly include recommendations for additional research and development to increase the accuracy and reliability of these health-monitoring systems. According to health experts, software and smart gadgets have a 10-20% error rate. In reality, traditional procedures must be employed to obtain 100% precise health metric data, and





Swagata Shashikant Zarkar et al.,

individualized treatment is advised based on a variety of environmental and situation factors. Another issue is that if one is continually reviewing the health findings, they risk making individuals uneasy and putting them in danger.

DISCUSSION

- Due to time constraints, only 6 smartwatches from diverse manufacturers and 5 software applications were considered for the study.
- According to health experts, software and smart gadgets make 10-20% of errors. In reality, traditional procedures must be employed to obtain 100% precise health metric data, and customized therapy is advised based on a variety of environmental and situation factors. Another issue is that if one is continually reviewing the health findings, they risk making individuals uneasy and putting them in danger.
- We eliminated pregnant women, HIV patients, those with deviant behavior, and people with any form of disease from this study.

List of Abbreviations

NA	-Not Applicable	Cal	-Calories
BP	-Blood Pressure	%	-Percentage
BPM	-Beats Per Minute	mmHg	-Millimeter(s) of mercury
SBP	-Systolic Blood Pressure	DBP	-Diastolic Blood Pressure
&	-and	Yr/Yrs	-Year/Years

Competing Interests

“The authors Ms. Swagata Shashikant Zarkar, Mr. Rahul Kailas Bharati, Dr. Shobha Kamalakar Bawiskar declares that they have no competing interests” We are not receiving or having financial competing and non-financial competing interests

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors

Disclaimer:

Researcher only conducts a pilot study on various available hardware and software tools In this research, researcher never encourages / do not promote / do not advertise any specific applications.

REFERENCES

1. Lunden, I. (2013, April 30). Pebble smartwatch raises \$10.2m on Kickstarter, shipping starts January 2013. TechCrunch. <https://techcrunch.com/2013/04/30/pebble-watch-raises-10-2m-on-kickstarter-shipping-starts-january-2013/>
2. Allied Market Research. (2020). Smartwatch market by type, application, and operating system: global opportunity analysis and industry forecast, 2020–2027. <https://www.alliedmarketresearch.com/smartwatch-market>
3. Manual of Fire Bolt BSW007 smartwatch.
4. Manual of Boulton Drift calling smartwatch.
5. Manual of Gadgetzone I8 Pro Max smartwatch.
6. Manual of Gizmore GIZFIT 907 smartwatch.
7. Manual of Noise ColorFit Pro 2 smartwatch.
8. Manual of Boat Wave Beat smartwatch.
9. U.S. Department of Health and Human Services. (n.d.). Health Information Privacy. Retrieved from <https://www.hhs.gov/hipaa/index.html>
10. World Health Organization. (2018). Health financing for universal coverage and health system performance: Concepts and definitions. Retrieved from https://www.who.int/health_financing/documents/health-system-performance/en/





Swagata Shashikant Zarkar et al.,

11. U.S. Department of Health and Human Services. (n.d.). HIPAA Security Rule. Retrieved from <https://www.hhs.gov/hipaa/for-professionals/security/index.html>
12. HealthITSecurity.com. (2021). Healthcare Cybersecurity. Retrieved from <https://healthitsecurity.com/topics/healthcare-cybersecurity>
13. Constitution of India
14. DISHA , 2018
15. IT Act and *Electronic Health Record Standards 2016*
16. Kaur, A., &Verma, M. (2018). Cyber security in healthcare industry. *International Journal of Computer Science and Network Security*, 18(6), 247-254.
17. Hossain, M. A., &Rahman, M. A. (2017). Biometric authentication in healthcare: A case study of improved forensic investigation. *International Journal of Knowledge and Learning*, 12(3/4), 295-309.
18. Bhaiji, F., Al-Wahaibi, A., Al-Alawi, M., & Al-Azri, H. (2019). Medicolegal and ethical implications of data breaches in healthcare: A review. *International Journal of Healthcare Management*, 1-8.
19. Jilani, N. T., Ali, F., &Arshad, A. (2020). Secure communication of medical data in the cloud computing environment. *International Journal of Advanced Computer Science and Applications*, 11(11), 104-111.
20. (Dinevski, R., & Open, T. (2016). Smartwatches and forensic investigations: a functional analysis of the apple watch. *Journal of Digital Forensics, Security and Law*, 11(4), 73-86.)
21. (Lin, C. W., Hsiao, Y. W., & Chu, S. Y. (2018). Investigating alibis from heart rate data on smartwatches. *Journal of Forensic Sciences*, 63(1), 94-99.)
22. (Hernandez, J. I. H., & Diaz, O. J. H. (2016). Use of smart watches for biometric data collection in forensic investigation. *Journal of Forensic Sciences & Criminal Investigation*, 3(3), 1-6.)
23. <https://play.google.com/store/apps/details?id=heartratemonitor.heartrate.pulse.pulseapp>
24. <https://play.google.com/store/apps/details?id=stepcounter.pedometer.stepstracker>
25. <https://play.google.com/store/apps/details?id=com.google.android.apps.fitness>
26. <https://play.google.com/store/apps/details?id=com.measurement.com>
27. <https://play.google.com/store/apps/details?id=com.sec.android.app.shealth>

Table No.1 General information about smartwatches used for study.

Sr.No.	Features of watch	Device 1 Fire Bolt BSW007 [3]	Device 2 Boult Drift Bluettoth Calling [4]	Device 3 Gadgetzone I8 Pro Max [5]	Device 4 Gizmore GIZFIT 907 [6]	Device 5 Noise ColorFit Pro 2 [7]	Device 6 Boat Wave Beat [8]
1	Price @ 19 April 2023	1,599	1,799	840	1,899	1,199	1,499
2	Water resistant	Yes	Yes	Yes	Yes	Yes	Yes
3	Scratch resistant screen	No	No	No	No	No	No
4	Crash detection and emergency SOS	No	No	No	No	No	No
5	Battery life	5 days	10 days	10 days	12 days	10 days	7 days
6	Advanced sensors	Yes	Yes	Yes	Yes	Yes	Yes
7	ECG tracker	No	No	No	No	No	No
8	Design	Full metal body &ultra	Lightweight	Lightweight	Lightweight	Stylish & lightweight	Slim metallic





Swagata Shashikant Zarkar et al.,

		lightweight					design
9	Display size	1.3 inches	1.69 inches	44 mm	14 inches	1.3 inches	1.69 inches
10	Track sleep	Yes	Yes	Yes	Yes	Yes	Yes
11	GPS system	No	No	No	No	No	No
12	Calling	No	Yes	Yes	No	No	No
13	Mobile payment	No	No	No	No	No	No
14	Compatibility OS-iOS, Android	Android &iOS	Android &iOS	Android &iOS	Android &iOS	Android &iOS	Android &iOS
15	Music	No	Yes	Yes	No	No	Yes
16	Connectivity	Bluetooth	Bluetooth	Bluetooth	Bluetooth	Bluetooth	Bluetooth
17	Touchscreen	Yes	Yes	Yes	Yes	Yes	Yes

Table No.2 Basic parameters reading of smartwatch.

Sr. no .	Basic parameters	Measuring units	Nor-mal range	Date 2023	Sampl e 1	Sampl e 2	Sampl e 3	Sample 4	Sampl e 5	Sampl e 6
					Device 1 Fire Boltt	Device 2 Boul Drift	Device 3 I8 Pro Max	Device 4 Gizmor e	Device 5 Noise	Device 6 Boat Wave Beat
					Age-22yrs	Age-18yrs	Age-22yrs	Age-22yrs	Age-22yrs	Age-22yrs
					Femal e	Male	Femal e	Female	Femal e	Femal e
1	Blood Pressure	Systolic /Diastolic mm/Hg (millimeter of mercury)	120/80 mm/Hg	25/2	123/77	114/71	97/77	NA	NA	NA
				26/2	119/79	111/74	117/67	NA	NA	NA
				27/2	132/73	111/74	102/72	NA	NA	NA
				28/2	110/68	120/72	126/76	NA	NA	NA
				1/3	114/71	119/77	121/66	NA	NA	NA
				2/3	122/76	117/73	126/71	NA	NA	NA
				3/3	116/72	120/72	124/79	NA	NA	NA
				4/3	132/73	117/74	117/67	NA	NA	NA
				5/3	128/70	120/78	96/66	NA	NA	NA
				6/3	119/79	118/73	121/66	NA	NA	NA
				7/3	144/82	120/71	98/68	NA	NA	NA
				8/3	113/70	117/71	127/82	NA	NA	NA
2	Heart rate	Bpm (beats per minutes)	60-100 bpm	25/2	63	58	95	83	91	85
				26/2	90	70	87	86	118	79
				27/2	75	73	95	84	65	101
				28/2	72	87	88	88	68	91
				1/3	73	75	94	82	62	92
				2/3	108	75	71	90	66	81





Swagata Shashikant Zarkar et al.,

				3/3	73	76	94	85	72	81
				4/3	86	75	71	73	71	86
				5/3	80	78	89	87	111	82
				6/3	68	80	89	85	74	82
				7/3	79	76	97	85	77	74
				8/3	72	77	80	84	78	65
				9/3	65	78	73	83	111	84
				10/3	83	80	76	88	77	95
				11/3	72	78	88	88	101	75
3	Sleep	Hours & minutes	8 hrs.	25/2	5.24	7.57	4.56	6.45	6.22	5.00
				26/2	8.00	7.30	7.11	4.50	4.60	8.59
				27/2	6.45	8.07	5.59	8.09	4.29	7.11
				28/2	6.38	6.36	4.30	6.59	7.01	6.33
				1/3	7.09	8.22	5.59	7.03	6.45	7.45
				2/3	8.34	5.59	8.30	4.30	7.32	7.32
				3/3	8.56	11.04	6.25	5.32	5.14	5.00
				4/3	7.06	6.45	7.34	5.45	5.59	3.58
				5/3	5.10	6.38	7.00	8.00	4.16	7.51
				6/3	5.29	10.16	7.38	5.41	5.10	9.28
				7/3	8.00	4.50	3.59	7.00	3.30	5.45
				8/3	6.30	7.17	7.45	4.56	4.56	7.09
				9/3	7.12	7.56	8.06	6.43	7.43	6.56
				10/3	4.00	9.48	6.45	6.56	6.45	6.50
				11/3	7.52	6.23	6.26	5.05	8.56	6.43
4	Walking steps	Steps	7000 steps per day	25/2	6000	4500	5322	3578	5000	5476
				26/2	4515	4521	4356	6212	3500	2390
				27/2	5784	1698	6500	4100	6078	2178
				28/2	2500	3421	3845	1900	5040	7000
				1/3	6587	7054	2389	7000	5006	3421
				2/3	7000	2856	6500	3053	6990	5478
				3/3	1500	5743	4890	4508	1500	6110
				4/3	2467	2000	3810	6100	4675	6577
				5/3	1900	4106	6987	3217	5087	4390
				6/3	1545	2080	2756	2590	6832	3749
				7/3	3000	1700	2990	2985	3452	3241
				8/3	7995	2167	5643	7000	3323	3677
				9/3	5330	6432	5512	3465	1990	5470
				10/3	7000	3505	3412	4822	1450	5623
				11/3	2590	3080	3980	6471	4532	6432
5	Blood oxygen level	Percentage	95-100%	25/2	97	98	95	96	NA	98
				26/2	96	97	95	98	NA	97
				27/2	98	97	98	98	NA	99
				28/2	98	97	97	94	NA	98
				1/3	97	96	98	90	NA	99
				2/3	96	96	98	94	NA	97
				3/3	97	98	97	97	NA	98
				4/3	98	96	98	98	NA	97
				5/3	98	98	96	96	NA	98





Swagata Shashikant Zarkar et al.,

				6/3	97	95	96	95	NA	98
				7/3	98	94	98	98	NA	98
				8/3	96	95	95	97	NA	94
				9/3	96	94	97	96	NA	97
				10/3	98	96	98	90	NA	99
				11/3	97	95	95	97	NA	97
6	Running (Indoor/outdoor)	As per time – hours & minutes	-	-	Not used	NA	NA	NA	NA	NA
		Pace – min/km	-	-	Not used	NA	NA	NA	NA	NA
		Consumption of calorie-Kcal	-	-	Not used	NA	NA	NA	NA	NA
7	ECG tracker		-	-	NA	NA	NA	NA	NA	NA
8	Female menstrual cycle tracking	-	-	-	NA	Not used	NA	NA	Not used	NA
9	Stress 0, 6, 12, 24 hrs.	33:66:99 (Low: Average: Medium)	-	-	NA	NA	NA	NA	NA	NA
10	Mood swing	-	-	-	NA	NA	NA	NA	NA	NA
11	Drink water remainder	-	As per Condition	-	NA	Not used	NA	Not used	NA	NA

Table No. 3 – Training parameters reading of smartwatch

Sr. no.	Training parametrs	Units of measuring	Normal range	Date	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6
					Device 1 Fire Boltt	Device 2 Boul Drift	Device 3 I8 pro max	Device 4 Gizmore	Device 5 Noise	Device 6 Boat wave beat
					Age-22yrs	Age-18yrs	Age-22yrs	Age-22yrs	Age-22yrs	Age-22yrs
					Female	Male	Female	Female		
1	Cycling	Bpm/ KCAL	60/90	25/2	Not used	NA	Not used	NA	Not used	79/5
				26/2	-	NA	-	NA	-	110/16
				27/2	-	NA	-	NA	-	73/5
				28/2	-	NA	-	NA	-	78/24





Swagata Shashikant Zarkar et al.,

				1/3	-	NA	-	NA	-	63/18
				2/3	-	NA	-	NA	-	90/29
				3/3	-	NA	-	NA	-	65/10
				4/3	-	NA	-	NA	-	80/44
				5/3	-	NA	-	NA	-	72/7
				6/3	-	NA	-	NA	-	88/14
				7/3	-	NA	-	NA	-	80/25
				8/3	-	NA	-	NA	-	72/13
				9/3	-	NA	-	NA	-	73/8
				10/3	-	NA	-	NA	-	78/15
				11/3	-	NA	-	NA	-	69/10
2	Swimming	KCAL	60/100 bpm	-	Not used	NA	NA	NA	NA	NA
3	Football	Steps, Bpm KCAL	8 hrs	-	Not used	NA	NA	NA	NA	Not used
4	Skipping	Bpm/ KCAL	-	25/2	80/16	NA	NA	NA	NA	NA
				26/2	95/24	NA	NA	NA	NA	NA
				27/2	75/8	NA	NA	NA	NA	NA
				28/2	87/18	NA	NA	NA	NA	NA
				1/3	90/20	NA	NA	NA	NA	NA
				2/3	86/15	NA	NA	NA	NA	NA
				3/3	75/12	NA	NA	NA	NA	NA
				4/3	75/6	NA	NA	NA	NA	NA
				5/3	89/14	NA	NA	NA	NA	NA
				6/3	72/9	NA	NA	NA	NA	NA
				7/3	90/22	NA	NA	NA	NA	NA
				8/3	79/10	NA	NA	NA	NA	NA
				9/3	108/27	NA	NA	NA	NA	NA
10/3	74/9	NA	NA	NA	NA	NA				
11/3	85/12	NA	NA	NA	NA	NA				
5	Badminton	Steps, Bpm KCAL	95-100%	-	Not used	NA	NA	NA	NA	Not used
6	Hiking	-	-	-	NA	NA	NA	NA	NA	NA
7	Cricket	-	-	-	NA	NA	NA	NA	NA	Not used
8	Yoga	-	-	-	NA	NA	NA	NA	NA	Not used
9	Rower	-	-	-	NA	NA	NA	NA	NA	NA
10	Elliptical	-	-	-	NA	NA	NA	NA	NA	NA
11	Workout	-	-	-	Not used	Not used	NA	NA	NA	NA
12	Breath training (inhale, exhale)	-	Minimum, medium, fast	-	NA	NA	NA	Not used	Not used	NA





Swagata Shashikant Zarkar et al.,

Table No.4 – Software app readings

Sr. No.	Name of the app	Reviews	Ratings	Rating Date	Parameter's measure	Readings
1.	Heart Rate Monitor-Pulse App [23]	4.7	3+	20 April 2023	Check heart rate (BPM)	7-4-2023 80 bpm
						8-4-2023 100 bpm
						9-4-2023 93 bpm
						11-4-2023 129 bpm
						13-4-2023 85 bpm
2.	Step counter-Pedometer [24]	4.8	3+	19 April 2023	Auto tracks your daily steps & calories.	7-4-2023 4589 steps
						8-4-2023 55 steps
						9-4-2023 3276 steps
						10-4-2023 1590 steps
						11-4-2023 4562 steps
3.	GoogleFit: Activiy Tracking [25]	4.3	3+	19 April 2023	Track your physical activity like calories count.	9-4-2023 1271 cal
						10-4-2023 1271 cal
						11-4-2023 1271 cal
						12-4-2023 1271 cal
						13-4-2023 1271 cal
4.	Blood Pressure Measurement App [26]	4.7	3+	20 April 2023	Measures the blood pressure (systolic/diastolic).	15-4-2023 88/65
						16-4-2023 102/79
						17-4-2023 90/77
						18-4-2023 93/79
						20-4-2023 80/84
5.	Samsung Health [27]	4.2	3+	20 April 2023	Track your fitness, weight, diet, food and sleep.	16-4-2023 3463 steps
						17-4-2023 4230 steps
						18-4-2023 3237 steps
						19-4-2023 1340 steps
						20-4-2023 49 steps

Table No. 5 – Smartwatch readings and medical expert readings

Health parameters	Smartwatch readings			Medical expert reading		
	Blood Pressure (SBP/DBP)	Heart rate (BPM)	Blood oxygen (%)	Blood pressure	Heart rate	Blood oxygen
Device 1	110/82	66	98	110/75	75	97
Device 2	132/73	91	99	120/78	79	99
Device 3	96/66	89	95	110/70	80	96
Device 4	NA	86	90	NA	79	94
Device 5	NA	77	NA	NA	75	NA
Device 6	NA	85	99	NA	80	98





Swagata Shashikant Zarkar et al.,



Fig. 1 Smartwatch images for research Work

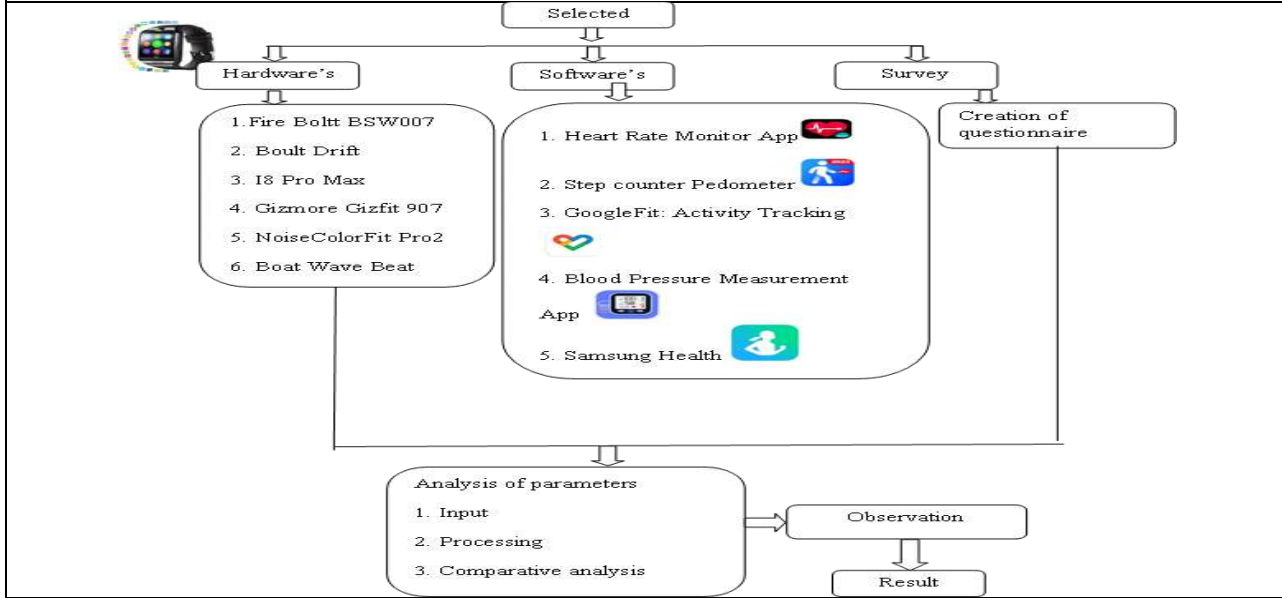


Fig.2 -Workflow approach of proposed exploratory research





An Effect of Virtual Reality in HRD - IT Sector

Y T Shahida Banu^{1*} and R Angamuthu²

¹Research Scholar, Department of Commerce, Annamalai University, Annamalai Nagar, Chidambaram, Tamil Nadu, India.

²Assistant Professor, Department of Commerce, Government Arts College, Tiruvannamalai, (Affiliated to Thiruvalluvar University), Tamil Nadu, India.

Received: 21 Jun 2024

Revised: 10 Aug 2024

Accepted: 26 Oct 2024

*Address for Correspondence

Y T Shahida Banu

Research Scholar, Department of Commerce,
Annamalai University, Annamalai Nagar,
Chidambaram, Tamil Nadu, India.

E.Mail: shahi143tina@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

In today's competitive IT sector, the effectiveness of training and development programs for employees is paramount. This study explores the dimensions in Virtual Reality for effective training and development strategies tailored specifically for IT employees. Through a comprehensive literature review and empirical analysis, this research investigates the effect of training and development that initiatives on employee performance, job satisfaction, and overall organizational success within the IT industries. Moreover, the study identifies key factors influencing the design, implementation, and evaluation of training programs, including technological advancements, regulatory requirements, and changing customer expectations. By synthesizing theoretical insights with practical implications, this research provides valuable insights for IT managers and HR professionals to optimize training and development efforts with VR, enhance employee competencies, and ultimately drive organizational growth in the dynamic IT landscape.

Keywords: Effective, Training, Development, Virtual Reality, Performance and Satisfaction

INTRODUCTION

The Human Resources is the most vital resources for an organization as they are considered essential to accomplish the goals of a business. The growth of IT sector is important for its survival in the today's competitive world, thus the employees of the firm have to enhance their prevailing skills. In today's technological world in order to the improve the skill of an employee in an effective way, the extent of the chosen training programme should reflect on the efficiency of the employee performance. Hence virtual training has its uniqueness and innovativeness within it as the pivotal role played by continuous learning in the IT sector. In an ever-evolving financial landscape, the need for



**Shahida Banu and Angamuthu**

skilled and adaptable employees is paramount. By addressing the challenges in the sector, such as regulatory compliance and technological advancements, the research endeavors to provide valuable insights for optimizing the professional growth for IT employees by ensuring their sustained success in a dynamic financial environment. The study aims to explore methodologies and tools that optimize Virtual training programs, ensuring alignment with industry trends and regulatory requirements. It will investigate the impact of personalized learning, simulation techniques, and continuous feedback on employee performance.

Virtual Reality

It is a technology which has shown prospective transformation in various sectors including Human Resource in recent days with its unique and practical training session. It can be used by the users using electronic gadgets like headset. It is the updated version from the Ivan Sutherland, a computer Scientist's "The Sword Of Damocles" in 1960s. In course of time, it has become as powerful tool by providing immersive learning practice.

Role of VR In HR Training

The role of VR in Human Resource Development is border and its role in training and development is to develop a interactive learning practice. Here interactive based training is provided to enhance the communication, leadership, compliances etc.. Leadership can be simulated by strategic thinking and decision making skills with diversification of perspectives and circumstances which will make one to feel the empathy. Apart from this VR helps the on boarding new employees to understand the culture and policies prevailing in the organization. It is cost effective training method for an organization in many aspects. Employee engagement can also be made effective using VR process. In real-time the usage of Virtual Reality in the HR has many benefits along with the cost-cut benefits for the management of an organization.

It is a evolutionary technique used in Training and development within the IT sector to enhance various major domains like Learning practice, realistic skill and to overcome hurdles in the training process. Exhilarating Training Atmosphere: VR training system uses real time scenario and allow the employees to communicate with virtual representatives. It is easier for the employee to understand the difficult model in a effective way than the traditional training method. Real Time Experience: VR enhance the IT professional's problem solving skills by experiencing the real world situation without the risk of reside system. Safe learning environment give them self-assurance and confidence to make mistakes and correct it.

Scalable Training plan: VR provides training program to numerous users at the same time or at the flexible time of the user beside the location barrier and the training programme can be modified according to the learner needs. Assessment and Feedback : VR is real time learning system which can evaluate the performance of an employee in each step and give the feedback to them accordingly based upon their performance in the assessment. Reduce Training Cost: implement of VR technology include huge cost whereas the VR provide training for long run which reduce the cost of training.

Research Objectives

Evaluate the existing Virtual training programs within the IT to identify strengths and weaknesses.
Determine the specific skills and knowledge gaps among IT employees that hinder optional performance.

Research Scope:

1. The study will focus on a specific region or multiple regions, analyzing the training and development practices within the IT sector.
2. The scope will cover employees at various levels, from entry-level positions to managerial roles, to ensure a comprehensive understanding of training needs.
3. Determine the training needs related to staying abreast of changing regulations and compliance requirements in the financial industry.



**Shahida Banu and Angamuthu**

REVIEW OF LITERATURE

A literature review serve as a important component of academic research, providing a widespread overview of existing scholarly literature on a particular area. It act as a major role in identifying gaps, trends, and debates within the field, informing the research questions, methodology, and theoretical framework of a study. (Mantovani, 2004) Considering the ongoing advancement in information technology, the rise of the internet, its widespread use, and the variety of its special services for information distribution and transmission, it is now one of the most widely used information sources. With the rise of e-learning technologies and the internet as a major tool for program delivery, remote learning has become possible in the medical and health care. (Fabrizia Mantovani 1, 2003) the organization of education for health professionals is being significantly impacted by new developments in health care delivery. It is widely acknowledged that medical knowledge doubles every 7-8 years and that new medical methods are developed on a daily basis .There are more and more challenges resources learning resources available thanks to recent development in educational technology. In fact, VR training may help to increase trainees' interest and motivation as well as providing a rich, dynamic and captivating instructional environment that support experiential learning by doing.

SUGGESTIONS

The awareness about the VR should be provided to the employees so as to improve the efficiency of it. The IT sector can give training programs to their employees to fill the prevailing communication gap within their colleagues. The superior should motivate their employees in their learning. The training provided to the employees can be effective so that they can implement their learning without seeking support from their superior.

CONCLUSION

Effective training and development programs are essential for the sustained success of ITs in today's competitive environment. Through this study, it is evident that investing in employee . Training not merely improves individual performance but also enhance the overall organizational effectiveness. By aligning training initiatives with strategic goals and identifying the evolving needs of employees, ITs can cultivate a atmosphere of continuous learning and innovation. Additionally, leveraging VR technology for training delivery and evaluation can enhance accessibility and effectiveness, particularly in the context of remote work and digital IT trends. Moreover, this research underscores the importance of ongoing evaluation and adaptation of training programs to ensure relevance and impact. By soliciting feedback from employees and leveraging VR data analytics, ITs can refine their training strategies to address emerging challenges and capitalize on opportunities for improvement. Furthermore, compliance with regulatory requirements remains a crucial aspect of training and development efforts, necessitating proactive measures to stay abreast of industry regulations and best practices.

REFERENCES

1. Bhaskar chattered (1996): Human Resource Management Contemporary Text, Sterling Publishers, New Delhi.
2. Biswajeet Pattanayak (2001): Human Resource Management, Prentice-Hall Of India Pvt Ltd, New Delhi.
3. Chabbra TN : Human Resources Management ,5th Edition (2005),Gagan Kapoor for Dhanpat Rai and company – New Delhi
4. Dr.G.Yoganandan, Ms.Sathya.C (2015):.Job Satisfaction in State Bank of India in Namakkal District, International Journal of Research in Management & Technology vol.5(1),167-173
5. Ashu Bhola (2015) :Job Satisfaction among bank employees- A study on District Kathua(J& K), International Journal of Management & Commerce Innovations vol.3,186-191





Shahida Banu and Angamuthu

6. Fabrizia Mantovani, Gianluca : Virtual Reality training for Health –Care Professionals; 10.1089/109493103322278772
7. Idris Olayiwola Ganiyu,Sulaiman; An evolution of Virtual training implications for talent development in the post-pandemic period;10.1108/DLO-02-2022-0039
8. Saeed Hameed ,Marwan Mohamed; The Evaluation of Virtual Training and Employees Effectiveness: A case study from Babco; Global Scientific Journal Vol.10.Issue.5

Table.1 Gender of the Employees

S.No	Gender Category	Respondent	Percentage
1	Male	62	62
2	Female	38	38
Total	100	100	

Source: Primary Data

It is inferred from the Table 1 that 62% percent of employee category is male and 38% percent is female. It is analyses that under the demographic profile the consideration presents a comprehensive overview of the attributes of employees at a IT Sector.

Table 2 Training and Development

S.No	Description	SD	DA	NE	AG	SA	Mean	SD
1	Employs competent and simulate real-life situations in training	6.4	19.1	17.4	31.8	25.4	3.50	1.23
2	Training is provided in traditional or VR mode.	3.8	19.5	23.7	27.5	25.4	3.51	1.17
3	Training increases your performance level with new techniques(Virtual Reality)	4.7	18.0	20.1	29.7	27.5	3.57	1.19
4	Training opportunities are available to everyone	4.2	16.9	21.6	31.8	25.4	3.57	1.16
5	Advanced level of training using VR can enhance your career	5.3	15.9	20.6	32.8	25.4	3.57	1.17

Source :Primary Data

SD: Strongly Disagree; DA: Disagree; NE: Neither Agree nor Disagree; AG: Agree; SA: Strongly Agree; Mean .

The table 1 scrutinizes the perceptions of IT sector employees regarding the virtual training with the prevailing traditional training in the organization.





Estimating the Approximate Solution of HIV Infection Model using Homotopy Perturbation Method (HPM)

S.Geethamalini^{1*} and J.Mercy²

¹Assistant Professor, Department of Mathematics, Sathayabama Institute of Science & Technology, Chennai, Tamil Nadu, India.

²M.Sc Student, Department of Mathematics, Sathayabama Institute of Science & Technology, Chennai, Tamil Nadu, India

Received: 21 Jun 2024

Revised: 24 Aug 2024

Accepted: 25 Oct 2024

*Address for Correspondence

S.Geethamalini

Assistant Professor, Department of Mathematics,
Sathayabama Institute of Science & Technology,
Chennai, Tamil Nadu, India.

E.Mail: geetha_malini@hotmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The current work uses the Homotopy Perturbation Method (HPM) to approximate the fundamental HIV viral dynamic model and describe the dynamics of the virus in a vulnerable population. Additionally, we evaluated the HPM on many implementations, demonstrating the correctness and efficiency of this approach. The estimated HPM solutions for the examined issues show good agreement with the Runge-Kutta method's numerical results.

Keywords: HPM,SIRC model;

AMS Subject Classification:34A34, 34G20, 46N60.

INTRODUCTION

Virus infection involves free virions binding to target cells, entering, replicating, releasing multiple copies into the extracellular environment, and subsequently infecting nearby cells in a recursive process [1]. In the classical cell-free infection mode, virions move through the body via freely circulating particles, traversing cells and tissues through fluid phase diffusion, underscoring the significance of this infection route [2]. Contemporary infectious disease epidemiology heavily relies on computational model-based methods. Human immune deficiency virus (HIV), a retrovirus targeting CD4+T cells and macrophages, causes a progressive decline in the individual's immune system, typically culminating in AIDS within 10–15 years of initial infection. HIV transmission encompasses various routes, including contaminated blood transfusions, sharing needles, unprotected sex, childbirth, and breastfeeding [3]. The detrimental effects of HIV on the body result from its biological mechanisms and its interaction with the immune system of human. Regrettably, HIV infection causes the immune system to malfunction. An epidemic model that





Geethamalini and Mercy

exclusively focused on CTL was provided by Nowak and Bangham [4]. Additionally, a model of the Hepatitis C virus that takes into account both humoral and cell-mediated responses was expressed by Wodarz [5]. A thorough examination of mathematical frameworks related to HIV pathogenesis during therapy was carried out by Wodarz and Nowak [6].

Perelson and Ribeiro [7] scrutinized models elucidating the within-host dynamics of HIV infection. Researchers have regularly offered effective mathematical methods to address the CD4+T cell HIV infection model [8–12]. Using the Bessel collocation approach, Yuzbasi was able to derive numerical solutions for the HIV model of CD4+T cells [13]. Karaçayır and Yuzbasi [14] applied a Galerkin method to address a epidemic model on HIV infection. Their study delved into the technique of residual correction, aiming to reduce approximation errors. Balamuralitharan and Geethamalini was applied a HPM method for solving Analytical solutions of the deterministic SEIA worm model [15–19]. In 1989, Perelson [20] developed a basic model with three variables that describes how HIV infects the human immune system:

Susceptible cells, infected cells, and virus. By adding four factors, Perelson et al. [21] enhanced this model and built on the groundwork established in their previous research. Uninfected cells denoted as $X(t)$, $Y(t)$ denotes infected cells and $Z(t)$ denotes virus. The model equations are detailed in [6, 7, 20–24].

$$\begin{aligned} \frac{dx}{dt} &= \mu - \eta x - \varepsilon x z \\ \frac{dy}{dt} &= \varepsilon x z - (\rho + \sigma) y \\ \frac{dz}{dt} &= \chi y - \lambda z \end{aligned} \tag{1}$$

With $x(0) = N_1, y(0) = N_2, z(0) = N_3$.

MATERIALS AND METHODS

Consider the equation:

$$A_1(\bar{u}) - f_1(\bar{r}_1) = 0, \bar{r}_1 \in \Omega, \tag{2}$$

subject to boundary condition

$$B_1(\bar{u}, \partial\bar{u} / \partial\bar{n}) = 0, \bar{r}_1 \in \Gamma, \tag{3}$$

In broad terms, one can decompose the operator A into two distinct components: a linear portion denoted as L and a nonlinear component represented as N.

$$\bar{L}_1(v_1) + \bar{N}_1(v_1) - f_1(\bar{r}_1) = 0. \tag{4}$$

Then Eq.(2) $v_1(\bar{r}_1, p) : \Omega \times [0, 1] \rightarrow R$

$$H(v_1, p) = (1 - p)[L_1(v_1) - L_1(u_0)] + p[A_1(v_1) - f_1(\bar{r}_1)] = 0, p \in [0, 1], \bar{r}_1 \in \Omega \tag{5}$$

Which is

$$H(v_1, p) = L_1(v_1) - L_1(\bar{u}_0) + pL(\bar{u}_0) + p[N_1(v_1) - f_1(\bar{r}_1)] = 0 \tag{6}$$

The parameter p, which lies in the interval [0, 1]. This meets the requirements for the boundaries. From (5) and (6), it is evident that

$$H(v_1, 0) = L_1(v_1) - L(\bar{u}_0) = 0, H(v_1, 1) = A_1(v_1) - f_1(\bar{r}_1) = 0 \tag{7}$$





Geethamalini and Mercy

In this instance, the embedding parameter is seamlessly introduced, devoid of any artificial influences. Moreover, it can be regarded as a diminutive parameter within the range of 0 to 1. Then

$$v = v_0 + pv_1 + p^2v_2 + \dots \tag{8}$$

Then,

$$u = \lim_{p \rightarrow 1} v = v_0 + v_1 + v_2 + v_3 + \dots \tag{9}$$

The convergence of series (9) has been demonstrated by He [25].

RESULTS AND DISCUSSION

Following the principles [26, 28-32] of the HPM, a homotopy for (1) can be formulated in the following manner:

$$p(\dot{v}_1 - \eta v_1 - \mu + \epsilon v_1 v_3) + (1 - p)(\dot{v}_1 - \dot{x}_0) = 0$$

$$p(\dot{v}_2 - \epsilon v_1 v_3 + (\sigma + \rho)v_2) + (1 - p)(\dot{v}_2 - \dot{y}_0) = 0 \tag{10}$$

$$(1 - p)(\dot{v}_3 - \dot{z}_0) + p(\dot{v}_3 + \lambda v_3 - \chi v_2) = 0$$

Where the first approximations are as follows, and dot stands for differentiation with respect to t:

$$v_{1,0}(t) = x_0(t) = x(0) = N_1$$

$$v_{2,0}(t) = y_0(t) = y(0) = N_2 \tag{11}$$

$$v_{3,0}(t) = z_0(t) = z(0) = N_3$$

and

$$v_1 = v_{1,0} + pv_{1,1} + p^2v_{1,2} + p^3v_{1,3} + \dots$$

$$v_2 = v_{2,0} + pv_{2,1} + p^2v_{2,2} + p^3v_{2,3} + \dots$$

$$v_3 = v_{3,0} + pv_{3,1} + p^2v_{3,2} + p^3v_{3,3} + \dots \tag{12}$$

From equations (11) and (12) are substituted into Equation (10), then we have

$$p(\dot{v}_{1,1} - \mu + \eta v_{1,0} + \epsilon v_{1,0} v_{3,0}) + p^2(\dot{v}_{1,2} + \eta v_{1,1} + \epsilon v_{1,0} v_{3,1} + \epsilon v_{1,1} v_{3,0}) + p^3(\dot{v}_{1,3} + \eta v_{1,2} + \epsilon v_{1,0} v_{3,2} + \epsilon v_{1,1} v_{3,1} + \epsilon v_{1,2} v_{3,0}) + \dots = 0$$

$$p(v_{2,1} - \epsilon v_{1,0} v_{3,0} + \rho v_{2,0} + \sigma v_{2,0}) + p^2(v_{2,2} - \epsilon v_{1,0} v_{3,1} + \epsilon v_{1,1} v_{3,0} + \rho v_{2,1} + \sigma v_{2,1}) + p^3(v_{2,3} - \epsilon v_{1,0} v_{3,2} - \epsilon v_{1,1} v_{3,1} - \epsilon v_{1,2} v_{3,0} + \rho v_{2,2} + \sigma v_{2,2}) + \dots = 0$$

$$p(\dot{v}_{3,1} - \chi v_{2,0} + \lambda v_{3,0}) + p^2(\dot{v}_{3,2} - \chi v_{2,1} + \lambda v_{3,1}) + p^3(\dot{v}_{3,3} - \chi v_{2,2} + \lambda v_{3,2}) + \dots = 0$$

$$\dot{v}_{1,1} - \mu + \eta v_{1,0} + \epsilon N_1 N_3 = 0$$

$$\dot{v}_{1,2} + \eta v_{1,1} + \epsilon N_1 v_{3,1} + \epsilon v_{1,1} N_3 = 0$$

$$\dot{v}_{1,3} + \eta v_{1,2} + \epsilon N_1 v_{3,2} + \epsilon v_{1,1} v_{3,1} + \epsilon v_{1,2} N_3 = 0$$





Geethamalini and Mercy

$$\begin{aligned} \dot{v}_{2,1} - \varepsilon N_1 N_3 + \rho N_2 + \sigma N_2 &= 0 \\ \dot{v}_{22} - \varepsilon N_1 v_{3,1} + \varepsilon v_{1,1} N_3 + \rho v_{2,1} + \sigma v_{2,1} &= 0 \end{aligned} \tag{14}$$

$$\begin{aligned} \dot{v}_{23} - \varepsilon N_1 v_{3,2} - \varepsilon v_{1,1} v_{3,1} - \varepsilon v_{1,2} N_3 + \rho v_{2,2} + \sigma v_{2,2} &= 0 \\ \dot{v}_{3,1} - \chi N_2 + \lambda N_3 &= 0 \\ \dot{v}_{32} - \chi v_{2,1} + \lambda v_{3,1} &= 0 \\ \dot{v}_{33} - \chi v_{2,2} + \lambda v_{3,2} &= 0 \end{aligned}$$

$$\begin{aligned} x(t) &= \lim_{p \rightarrow 1} v_1(t) = \sum_{k=0}^{k=3} v_{1,k}(t) \\ y(t) &= \lim_{p \rightarrow 1} v_2(t) = \sum_{k=0}^{k=3} v_{2,k}(t) \end{aligned} \tag{15}$$

$$z(t) = \lim_{p \rightarrow 1} v_3(t) = \sum_{k=0}^{k=3} v_{3,k}(t)$$

Therefore,

$$\begin{aligned} x(t) &= N_1 + t(\mu - \eta N_1 - \varepsilon N_1 N_3) + \frac{t^2}{2} [(\mu - \eta N_1 - \varepsilon N_1 N_3)(-\eta - \varepsilon N_3) - \varepsilon N_1(\chi N_2 - \lambda N_3)] \\ &+ \frac{t^3}{6} [((\mu - \eta N_1 - \varepsilon N_1 N_3)(-\eta - \varepsilon N_3) - \varepsilon N_1(\chi N_2 - \lambda N_3))(-\eta - \varepsilon N_3) - \\ &\varepsilon N_1(\varepsilon \chi N_1 N_3 - \rho \chi N_2 - \sigma \chi N_2) - \lambda(\chi N_2 - \lambda N_3) + (\mu - \eta N_1 - \varepsilon N_1 N_3)(\chi N_2 - \lambda N_3)] \\ y(t) &= N_2 + t(\varepsilon N_1 N_3 - \rho N_2 - \sigma N_2) + \frac{t^2}{2} [\varepsilon N_1(\chi N_2 - \lambda N_3) - (\mu - \eta N_1 - \varepsilon N_1 N_3) N_3] \\ &- (\varepsilon N_1 N_3 - \rho N_2 - \sigma N_2)(\rho + \sigma) + \frac{t^3}{6} [\varepsilon N_1(\chi(\varepsilon N_1 N_3 - \rho N_2 - \sigma N_2) - \lambda(\chi N_2 - \lambda N_3)) \\ &+ (\mu - \eta N_1 - \varepsilon N_1 N_3)(\chi N_2 - \lambda N_3) + ((\mu - \eta N_1 - \varepsilon N_1 N_3)(-\eta - \varepsilon N_3) - \varepsilon N_1(\chi N_2 - \lambda N_3)) N_3] \\ &- [\varepsilon N_1(\chi N_2 - \lambda N_3) - (\mu - \eta N_1 - \varepsilon N_1 N_3) N_3] - (\varepsilon N_1 N_3 - \rho N_2 - \sigma N_2)(\rho + \sigma) (\rho + \sigma) \\ z(t) &= N_3 + t(\chi N_2 - \lambda N_3) + \frac{t^2}{2} [\chi(\varepsilon N_1 N_3 - \rho N_2 - \sigma N_2) - \lambda(\chi N_2 - \lambda N_3)] \\ &+ \frac{t^3}{6} [\chi[\varepsilon N_1(\chi N_2 - \lambda N_3) - (\mu - \eta N_1 - \varepsilon N_1 N_3) N_3] - (\varepsilon N_1 N_3 - \rho N_2 - \sigma N_2)(\rho + \sigma)] \\ &- \lambda[\chi(\varepsilon N_1 N_3 - \rho N_2 - \sigma N_2) - \lambda(\chi N_2 - \lambda N_3)] \end{aligned} \tag{16}$$

NUMERICAL SIMULATIONS

In order to compare the findings with those of Biazar [27], the following parameter values are taken into account:

- N1=20 Initial x(t) population, which are susceptible
- N2=15 Initial y(t) population, which are infective
- N3=10 Initial z(t) population, which are immune





Geethamalini and Mercy

$\beta=0.01$ Rate at which the susceptible population to become infected

$\gamma=0.02$ Rate at which the infective to immune population

Three term Approximations are computed and shown for $x(t)$, $y(t)$, and $z(t)$ in Fig.2,3 &4. We obtained the following graphs using MATLAB.

$$\begin{aligned} x(t) &= 20 - 3t - 0.045t^2 + 0.02805t^3 \\ y(t) &= 15 + 2.7t + 0.018t^2 - 0.02817t^3 \\ z(t) &= 10 + 0.3t + 0.07t^2 - 0.00012t^3 \end{aligned} \quad (17)$$

A comparison between the results demonstrates that the approximations of three terms' results of the HPM is same as the results of Fourth order Runge-Kutta method (RK4).

CONCLUSION

The epidemic model's nonlinear system of differential equations can be solved using the HPM, which looks to be a very simple process and produces accurate and consistent results. When compared to the fourth-order Runge-Kutta method (RK4), less computing is required.

REFERENCES

1. Sattentau Q. 2008. Avoiding the void: cell-to-cell spread of human viruses. *Nature Reviews Microbiology*. 6(11):815– 826.
2. Sattentau Q. 2011. The direct passage of animal viruses between cells. *Current Opinion Virology*. 1(5):396–402.
3. Dubey P, Dubey US, Dubey B. 2018. Examining how the acquired immune response and antiretroviral therapy impact the dynamics of HIV infection. Modeling the role of acquired immune response and antiretroviral therapy in the dynamics of HIV infection. *Mathematics Computers in Simulation*. 144:120–37.
4. Nowak MA, Bangham CRM. 1996. Population dynamics of immune responses to persistent viruses. *Science*. 272(5258):74–9.
5. Wodarz D. 2003. Hepatitis c virus dynamics and pathology: the role of CTL and antibody responses. *Journal of General Virology*. 84(7):1743–50.
6. Wodarz D, Nowak MA. 2002. Mathematical models of HIV pathogenesis and treatment. *BioEssays*. 24(12):1178–87.
7. Pereson AS, Ribeiro RM. 2013. Modeling the within host dynamics of HIV infection. *BMC Biology*. 11(96):1–10.
8. Yuzbasi S, Ismailov N. 2017. A numerical method for the solutions of the HIV infection model of CD4+ T cells. *International Journal of Biomathematics*. 10(7):1–16.
9. Yuzbasi S, Karaçayır M. 2017. An exponential galerkin method for solutions of HIV infection model of CD4+ T-cells. *Computational Biology and Chemistry*. 67:205–12.
10. Yuzbasi S. 2016. An exponential collocation method for the solutions of the HIV infection model of CD4+ T cells. *International Journal of Biomathematics*. 9(3):1–15.
11. Merdan M, Gokdogan A, Yildirim A. 2011. On the numerical solution of the model for HIV infection of CD4+ T cells. *Computers Mathematics Applications*. 62(1):118–23.
12. Merdan M., Khaniyev T. 2010. Homotopy perturbation method for solving viral dynamical model. *Cumhuriyet science Journal*: 31(1):65-77.
13. Yuzbasi S. 2012. A numerical approach to solve the model for HIV infection of CD4+ T cells. *Applied Mathematical Modelling*. 36(12):5876–90.
14. Yuzbasi S, Karaçayır M. 2018. A Galerkin-type method for solving a delayed model on HIV infection of CD4+ T-cells. *Iranian Journal of Science and Technology Transactions A: Science*. 42(3):1087–95.
15. Balamuralitharan.S and Geethamalini.S, 2017. Parameter Estimation of Model For EIAV Infection Using HPM,



**Geethamalini and Mercy**

- International Journal of Pure and Applied Mathematics.113 (12) 196-204.
16. Balamuralitharan.S and Geethamalini.S, 2018.Solutions of the epidemic of EIAV infection by HPM, Journal of Physics: Conference Series.1000:1-6.
 17. Geethamalini.S and Balamuralitharan.S, 2018. Semianalytical solutions by homotopy analysis method for EIAV infection with stability analysis, Advances in Difference Equations, Springer. 356:1-14.
 18. Radha, M., Balamuralitharan, S., Geethamalini, S., Geetha, V., and Rathinasamy, A. 2019. Analytic Solutions of the stochastic SEIA worm model by homotopy perturbation method,AIP Conference Proceedings. 2112, 020500.
 19. Geetha, V., Balamuralitharan, S., Geethamalini, S., Radha, M., and Rathinasamy, A., 2019. Analytic Solutions of the deterministic SEIA worm model by homotopy perturbation method, AIP Conference Proceedings, 2112, 020100.
 20. Perelson AS. 1989. Modelling the interaction of the immune system with HIV. In: Castillo-Chavez C, editor. Mathematical and Statistical Approaches to AIDS Epidemiology.83: 350–70.
 21. Perelson AS, Kirschner DE, Boer RD.1993. Dynamics of HIV infection of CD4+ T-cells. Mathematical Biosciences. 114(1):81–125.
 22. Lai X. 2014. Study of virus dynamics by mathematical models. Electronic Thesis and Dissertation Repository. <https://ir.lib.uwo.ca/etd/1978>.
 23. Perelson AS, Neumann AU, Markowitz M, Leonard JM, Ho DD. 1996. HIV-1 dynamics in vivo: virion clearance rate, infected cell life-span, and viral generation time. Science. 271(5255):1582–6.
 24. Stafford MA, Corey L, Cao Y, Daar ES, Ho DD, Perelson AS. 2000. Modeling plasma virus concentration during primary HIV infection. Journal of Theoretical Biology. 203(3):285–301.
 25. J.H. He. 1999. Homotopy perturbation technique, Computer methods in applied mechanics and engineering 178:257–262.
 26. Ghoreishi M, Ismail A, Alomari A. 2011. Application of the homotopy analysis method for solving a model for HIV infection of CD4+ T-cells, Mathematical and Computer modelling.54:3007-3015.
 27. Biazar.J. 2006. Solution of the epidemic model by Adomian decomposition method, Applied Mathematics and Computation. 173 (2): 1101–1106.
 28. Parvaiz Ahmad Naik, Jian Zua, Mohammad Ghoreishi. 2020. Estimating the approximate analytical solution of HIV viral dynamic model by using homotopy analysis method,Chaos, Solitons & Fractals. 131: 109500.
 29. M. Rafei, D.D. Ganji, H. Daniali. 2006. Solution of the epidemic model by homotopy perturbation method. Applied Mathematics & Computations.187 (2): 1056-1062.
 30. Amit Kumar Saraswat, Manish Goyal, 2022. Numerical simulation of time -dependent influenza model with Atangana-Baleanu non-integer order derivative in Liouville-Caputo sense. 96(104): 1-14.
 31. Sami Alihani, Mohd Salmi Md Noorani, A.K. Alomari. 2020. Numerical solution of fractional – order HIV model using Homotopy Method. Hindawi, Discrete Dynamics in Nature and Society. 2149037.1-13.
 32. M.J. Mahmoodabadi. 2020. Epidemic model analyzed via particle swarm optimization based homotopy perturbation method. Informatics in Medicine Unlocked. 18:100293. 1-8.
 33. Ahmad Naik, Zu Jian, Mohammad Ghoreishi. 2020. Estimating the approximate analytical solution of HIV viral dynamic model by using homotopy analysis method. Chaos,Solitons & Fractals. 131,109500. 1-13.





Geethamalini and Mercy

Table1. Parameters Meaning of HIV Infection

Parameters and functions	Meaning	Values
x	Susceptible	Variable
y	Infected	Variable
z	Virus	Variable
μ	target cells	10 cells
ϵ	rate of Infection	$0.000024 \text{ mm}^3 \text{ day}^{-1}$
η	Death amount for susceptible	0.01 day^{-1}
χ	virus production	$467 \text{ virions cells}^{-1}$ [33]
λ	Amount of virus clearance	3.4 day^{-1} [33]
σ	Death amount due to virus	0.16 [33]
ρ	Mortality rate of infected cells killed by CTLs	$0.01 \text{ day}^{-1} \text{ cell}^{-1}$ [33]

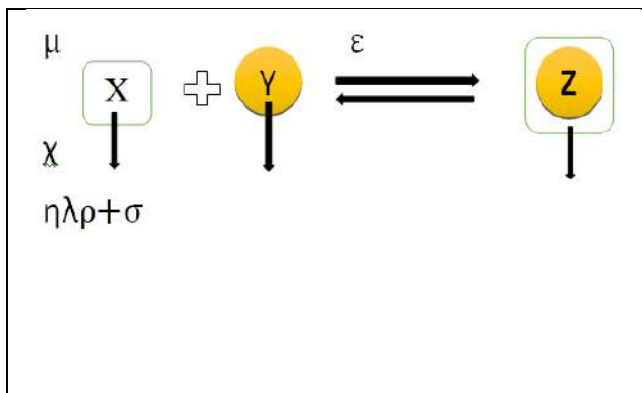


Fig. 1. Basic HIV viral dynamics schematic diagram

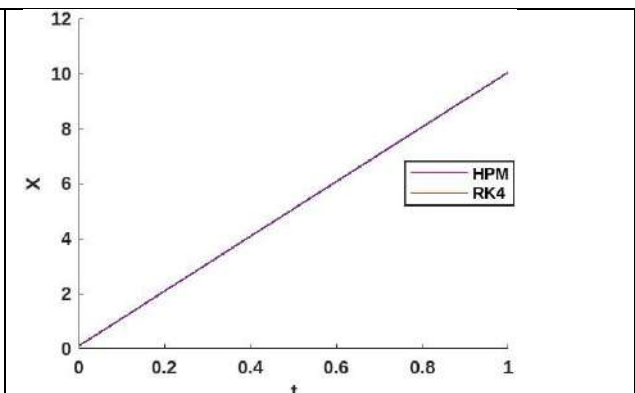


Fig. 2. Approximations of three terms plotted against time for X(t)

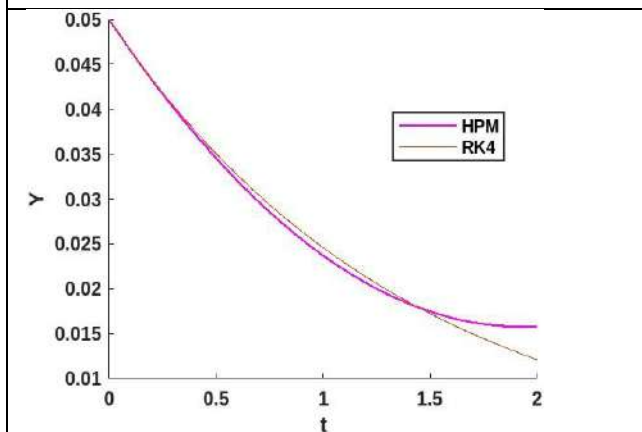


Fig. 3. Approximations of three terms plotted against time for Y(t)

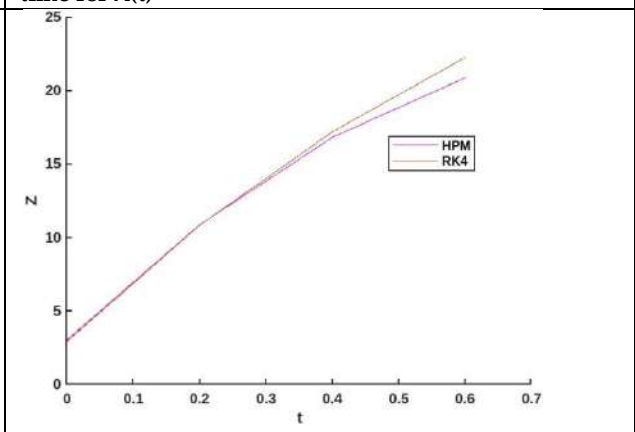


Fig. 4. Approximations of three terms plotted against time for Z(t)





Impact of Silver Nanoparticles against Hepatitis B Virus

Sanjana Bhat¹ and Meenal Rehman^{2*}

¹Research Scholar, School of Life & Basic Sciences, Jaipur National University, Jaipur, Rajasthan, India.

²Associate Professor, School of Life & Basic Sciences, Jaipur National University, Jaipur, Rajasthan, India.

Received: 21 Jun 2024

Revised: 20 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

Meenal Rehman

Associate Professor,

School of Life & Basic Sciences,

Jaipur National University, Jaipur,

Rajasthan, India.

E.Mail: meenal.rehman@jnujaipur.ac.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

It has been shown that silver nanoparticles possess potentially cytoprotective effects against infected T-cells. However, the effects of these nanoparticles on other viruses are mostly unknown. This literature review examined the effects of silver nanoparticles on the hepatitis B virus (HBV). Emerging viral illnesses present enormous risks to public health throughout the whole world. This is particularly true in the wake of the COVID-19 epidemic, which was responsible for a considerable number of fatalities among humans and had severe repercussions on the global economy. It is noteworthy to note that studies have shown that silver nanoparticles, which are sometimes referred to as AgNPs, are capable of destroying viruses, bacteria, and fungi via a number of different approaches. This is a significant challenge for the pharmaceutical industry. It is difficult to create treatments that are effective against viruses yet do not cause harm to the cells of the host organism they are being used against. In recent years, there has been a great focus put on the exploration of how AgNPs interact with viruses. This study has been carried out in a number of different laboratories throughout the world. This article explores the prospect of using silver nanoparticles, which have antiviral properties, in future medical treatments for viral infections. The primary emphasis of this research is on the properties of silver nanoparticles (AgNPs), namely their antiviral activity, mechanisms, applications, and toxicity. Methodologies that have been used in the past to describe AgNPs are included in these features.

Keywords: silver nanoparticles, antiviral mechanism, hepatitis B, cytoprotective, viral infection

INTRODUCTION

Silver nanoparticles, commonly known as AgNPs, have been the focus of much study into their antibacterial qualities due to the unique physical and chemical characteristics they possess. Because various kinds of hepatitis may result in infections and other health issues in individuals all over the world, hepatitis viruses continue to be a significant worldwide public health concern. Hepatitis viruses come in five distinct varieties, each identified by the letters A, B,



**Sanjana Bhat and Meenal Rehman**

C, and D. Hepatitis viruses may transfer from person to person in a variety of ways, with various clinical symptoms and potential long-term health effects for patients. It is crucial to remember that although some research indicates silver nanoparticles may be able to stop viruses from multiplying, the information is still preliminary and cannot be taken as completely conclusive. Throughout human history, viruses have been responsible for many devastating pandemics. It is hypothesized that modern viruses originated in the same pre-existing cell populations as modern cells [1]. They may serve as vectors in a wide variety of organisms, including mammals, plants, bacteria, and fungi [2], and can be found in a size range from 20 to 900 nanometers. They may also take on a variety of chemical forms. Despite the widespread availability of hepatitis, A and B vaccines, it is essential to keep in mind that hepatitis C has no cure at the present time. The variety of therapies for chronic hepatitis C, however, has expanded greatly in recent years.

Emerging hepatitis virus infections can be caused by a variety of conditions, such as the emergence of novel strains or genotypes of the virus, rising transmission rates in some areas, changes to the disease's clinical presentation, or difficulties controlling the virus' spread. It is conceivable for viruses to spread from one person to another or from a mother to her offspring [3]. Some viruses may only be communicated by sexual contact, while others can spread through ingestion of contaminated liquids or foods. They may also be passed from person to person by saliva, coughing, sneezing, and other bodily fluids [3]. They may also be spread by kissing, spitting, and sneezing. Sexual contact is necessary for the spread of certain viruses, whereas contaminated water or food may spread others [3]. Additionally, it is known that certain viruses, such the ones that cause hepatitis or the human immunodeficiency virus (HIV), may induce recurrent infections that might lead to the growth of tumors or the development of acquired immunodeficiency. The prevalence of viral infections, their clinical significance, and the need for antiviral medications are all correlated [4]. Injurious organisms, such as viruses, are to blame for a significant portion of illness cases and fatalities worldwide. For instance, it has been calculated that over 2 million people worldwide die each year from viruses [5]. The highly contagious nature of these illnesses and the lack of effective management methods have serious negative effects on public health [6]. Numerous viral diseases, including smallpox, have been wiped out with the aid of vaccine campaigns. One of these viruses, smallpox, was eliminated in 1979 [7]. Numerous vaccines against viruses have been produced in recent years; nevertheless, additional shots and medications are still needed to lessen the impact [8,9].

Nanotechnology has enabled significant advancements in the fight against viruses during the last several years. Innovative biomolecular systems have been created as a result of the use of nanotechnology in biomedicine. These systems have the ability to differentiate between certain cell types, viruses, bacteria, and fungus. To do this, nanoscale (100 nm) characteristics are developed for individual components. The physical (plasmonic resonance and fluorescence amplification) and chemical features of nanoparticles have made them useful as antiviral agents. These skills are generated by a large number of surface atoms and a large surface area relative to total volume. When comparing nanoparticles to bulk materials or ions, important nanoparticle properties improve as particle diameter decreases. This is due to the fact that decreasing a particle's diameter significantly increases the accessible surface area of the particle. These characteristics pave the way for improved and safer drugs, targeted tissue therapies, individualized nanomedicines, and preventative health screenings [10]. Since nanoparticles are unique among chemicals in terms of their physical and chemical characteristics, scientists have been investigating whether or not they may halt viral infections. Among the many viruses that may cause hepatitis, the most well-known are Hepatitis B (HBV) and Hepatitis C (HCV). The identification of several nano-based compounds has enhanced both the efficacy of treatment and the targeted administration of antiviral medicines [11].

In this work, the antiviral efficacy of AgNPs against the hepatitis virus is reviewed, and the probable antiviral mechanisms underlying this activity are discussed. In addition, the characterization methods, toxicity ranges, and prospective applications of AgNPs are examined in this work. Conclusion: There is some indication that silver nanoparticles may be antiviral against HBV; nevertheless, further study is required to prove their safety and usefulness in clinical settings, optimize their characteristics for maximal performance, and completely understand their mechanisms of action.



**Sanjana Bhat and Meenal Rehman****Synthesis of Silver Nanoparticles**

Silver nanoparticles, commonly known as AgNPs, have emerged as significant agents in a number of scientific domains due to the unique characteristics they contain and the many applications they may be used in. The potential of several nanoscale miracles has been unleashed throughout time by the development of novel synthesis methods. This article explores the synthesis of silver nanoparticles, shedding light on their production processes, the uses for which they are most helpful, and the promise for future advancement. In its simplest form, AgNP synthesis may be broken down into top-down and bottom-up processes. This "top-down approach" involves a number of physical forces, such as ball milling (which uses mechanical energy), grinding, and crushing, to create metal nanoparticles (NPs) from bulk materials. Vapor condensation employs thermal energy, whereas the laser ablation and electrical arc discharge methods also use electricity [12]. These procedures enable the synthesis of pure nanoparticles without the use of any chemicals, and may yield NPs with sizes ranging from 10 to 100 nm. When making nanoparticles, one may use physical methods to ensure both high purity and a uniform particle size distribution. Even if there aren't any chemical reactions that might endanger people or the environment, the physical process still struggles to avoid agglomeration since neither stabilizers nor capping agents are readily accessible. You will need to employ complicated equipment and outside power sources to complete these processes. On the other hand, bottom-up approaches start with single molecules and use nucleation and growth processes to form more complex clusters [12,13]. It has been shown that bottom-up strategies are more effective than top-down ones.

Silver nanoparticles may directly alter viruses and their initial host cell interactions depending on their size, shape, functionality, and composition. Silver nanoparticles are antiviral, although their mechanism is unknown. Many studies show that AgNPs may inactivate viral particles. This is done by eliminating viral components such as capsid structural proteins and envelope glycoproteins. After an hour of infection, AgNP-decorated silica hybrid composites (Ag30-SiO₂) suppressed IFV-A. The virus envelope glycoproteins hemagglutinin (HA) and neuroamidase (NA) also bonded with these composites, greatly reducing their activity. After studying how Ag30-SiO₂ composites interact with the virus' outer membrane, researchers found that MDCK cells may be less susceptible to IFV-A infection [14]. To limit HBV genome replication, AgNO₃ was utilized to build AgNPs with average diameters of 10 and 50 nm in HEPES buffer [15]. TEM showed that AgNPs linked to HepAD38 cells to create HBV pgRNA. The utilization of nanoparticles as a possible antiviral use is an exciting idea, according to the findings of recent study. Early study has shown the efficacy of AgNPs, despite the fact that their method of action is not entirely known. In-depth research into their antiviral efficacy against certain viruses has shown that it is sufficient to justify pursuing this area of study in order to produce nano-treatments that are effective against a broad range of viruses [16].

The emergence of nanotechnology, which functions on the microscopic scale, has resulted in a fundamental paradigm shift in the scientific world. Silver nanoparticles, which typically vary in size from 1 to 100 nanometers, exhibit distinct properties on the physical, chemical, and biological fronts as compared to bulk silver. This paves the way for several cutting-edge uses in electronics, medicine, catalysis, and other fields [16]. There are many techniques to create silver nanoparticles, each of which has benefits and may be customized to meet specific requirements, such as one of the following: The conventional method of reducing silver is called chemical reduction, and it calls for the employment of a reducing agent in addition to the reduction of a precursor silver salt. Salt is most often utilized in the form of sodium citrate or sodium borohydride. The next stage involves creating AgNPs utilizing physical methods such as laser ablation, sputtering, and evaporation. These processes employ energy to the process, converting bulk silver into nanoparticles. Last but not least, a recently developed strategy called "Green Synthesis," which refers to nature-inspired synthesis methods and involves the use of plant extracts, microbes, or biomolecules as reducing and stabilizing agents, offers environmentally friendly routes to the production of AgNP [17]. The manufacture of silver nanoparticles is an interdisciplinary project that has drawn interest from researchers because it provides a window into the world of nanoscale phenomena. As science and technology advance, it will be possible to fully realize the promise of AgNPs in a range of applications by improving synthesis methods and understanding AgNP behavior. The investigation of the world at the nanoscale continues to change many industries and inspire previously unthinkable solutions.



**Sanjana Bhat and Meenal Rehman****AgNPs Against Hepatitis B Virus**

Silver nanoparticles (AgNPs) and their impact on Hepatitis B virus (HBV) are now a research focus. Recent interest in silver nanoparticles (AgNPs) has been driven by the promise of their antibacterial effects, however studies examining the precise nature of AgNPs' interaction with HBV are still in their infancy. Antiviral drugs often prevent the production of new viruses by directly affecting the virus or by interfering with critical steps in the viral replication process [17]. In order to develop innovative antiviral therapies based on nanoparticles in a way that is precise and efficient, it is crucial to understand the antiviral mechanism of action of AgNPs. Although antiviral mechanisms are still being studied, it seems that AgNP production is a necessary component. Size, shape, and surface functionalization are some of the factors that affect antiviral efficacy [18,19].

As of 2007, six different medications have been approved for the treatment of HBV infection. Traditional interferon (IFN)-2b and pegylated IFN-2a are two examples of immunomodulatory drugs. Both of these are intended to aid the host immune system in regaining control of HBV, which should eventually lead to remission of the chronic condition without the need for medication. The other four medications work directly against viruses and are nucleoside analogues [20]. Lamivudine, adefovir, entecavir, and telbivudine are these nucleoside analogues. IFN- may directly limit viral protein synthesis by regulating antiviral cytokines [21, 22]. IFN- may boost CD8+ cytotoxic T-lymphocyte and natural killer cell lysis of infected hepatocytes [21, 21]. Natural killer cells and CD8+ cytotoxic T lymphocytes can lyse infected hepatocytes faster [21]. AgNPs have the potential to be employed as a virucidal agent due to their capacity to alter the morphology of the virion and destroy the infectious viral particle. Additionally, by blocking the virus from adhering to the host cell or from entering cells, it may obstruct the first stages of viral reproduction. The inhibition of viral replication's late stages has also been proposed, although its effects within cells are less well understood [23]. Despite the fact that they are the antiviral mechanisms that are discussed the most often, this is the case.

From this perspective, a wide range of sectors have shown interest in AgNPs due to their broad-spectrum antiviral action as well as their ability to stop cell infection. The manufacturing of PPE, food packaging, textiles, and water and air purification systems are all examples of industries that are expected to develop rapidly in the next years. In reality, from a scientific perspective, the best cures would be those that either destroy viral particles or obstruct the attachment and entry activities of viruses. First, the capacity to avoid cell infection would lessen host toxicity and reduce the possibility of discovering virus resistance. Additionally, it would be useful for providing a quick and efficient strategy in combating newly emerging virus strains [23,24]. Some investigations have shown that AgNPs physically link to viruses, proving their antiviral properties. The findings of those investigations, however, merely suggested a possible method of action, suggesting the need for further investigation [25].

The ability of AgNPs to inhibit the replication of viruses is an intriguing trait that might be used in a variety of technologies. It may be feasible to utilize a variety of ways, such as coating with just AgNPs, altering the surface of the AgNPs themselves, or a combination of these and other methods, to prevent metallic nanoparticles from coming into direct contact with cells and maybe reduce their cytotoxicity. AgNPs that have been functionalized, coated, or integrated into a composite material may have their antiviral activity affected by the physical barriers created by the combination with another element. However, these strategies may also alter how viruses and nanoparticles interact. When metallic nanoparticles were encased in organic or inorganic materials like chitosan, collagen, or gelatin, the antiviral activity was much increased, and the impact lasted for a longer period of time because of the delayed ion release [26]. The nanoparticles' capacity to postpone ion release made this feasible.

This session will conclude with a discussion of the ideal qualities of a virucidal antiviral agent that is safe for humans and the environment. The composite system's antiviral activity was sensitive to AgNP size and concentration. Higher amounts and smaller AgNP dimensions increased the composite system's antiviral activity, whereas the chitosan matrix did not. The chitosan matrix, on the one hand, might help block the virus from attaching to the nanoparticles directly, or at least mitigate the virus's impact on them. However, since the matrix results in additional physical harm, virions attach to the composite material, which reduces their ability to interact with the host cells [27].



**Sanjana Bhat and Meenal Rehman**

Although chitosan lacks inherent antiviral capabilities, it may significantly boost the efficacy of nanoparticles by acting as a matrix for them. The chitosan matrix and AgNPs combination falls within the same category. The influence that the precursor molecules and metabolites have, depending on the application, on the health and safety of either people or animals, or on the environment, must be examined and evaluated when selecting the system that will be employed [28]. The specific mechanism by which AgNPs eradicate viruses remains mostly unknown. AgNPs can protect against HSV, respiratory syncytial virus, and adenovirus type 3 [29]. Either avoiding viral infection in cells or directly inactivating viruses may achieve this.

The World Health Organization estimates that 887,000 individuals died in 2015 as a direct consequence of problems connected to HBV, such as cirrhosis and hepatocellular cancer. Some estimates put the number of persons infected with this virus at 227 million. The HBV core releases particles into liver cell nuclei, where they form covalently closed circular DNA (cccDNA) [30]. Virus-like particles, attenuated viruses, or protein-subunit antigens in vaccines activate the immune system and prevent illness. The preclinical stage of AgNP usage still has restrictions on its use. AgNPs' two main tactics for combating viruses that could be present in the human body are vaccination and oral delivery [31,32]. One of the best ways to save healthcare expenses while also lowering the chance of developing infectious diseases is to get vaccinated. Researchers have been examining the immunoactivity of both naturally occurring and synthetically manufactured nanoparticles since the development of nanotechnology [33,34]. Initial study reveals that silver nanoparticles (AgNPs) may have antiviral properties, but further research is needed, especially for hepatitis B virus. It is important to stress that the study is still in its early phases despite some data pointing to possible antiviral advantages of AgNPs. The optimal size, concentration, and surface qualities of AgNPs must also be determined via meticulous research in order to efficiently target HBV. Further research is required on the possible cytotoxicity and off-target effects of AgNPs on healthy cells. This is due to the possibility that nanoparticles may interact with a variety of biological elements other than the intended targets.

Acknowledgments

None

Conflict of Interest

No conflict of interest

Author's Contribution

All of the authors named have made a significant, direct, and intellectual contribution to the work and have given their permission for it to be published.

Funding

None

Ethics Statement

Not applicable

REFERENCES

1. Nasir, A.; Kim, K.M.; Caetano-Anollés, G. Viral evolution. *Mob. Genet. Elem.* 2012, 2, 247–252.
2. Passi, D.; Sharma, S.; Dutta, S.R.; Dudeja, P.; Sharma, V. Ebola virus disease (the killer virus): Another threat to humans and bioterrorism: Brief review and recent updates. *J. Clin. Diagn. Res.* 2015, 9, LE01.
3. Corstjens, M.P.L.A.; Abrams, W.R.; Malamud, D. Saliva and viral infections. *Periodontology 2000* 2016, 70, 93–110.
4. Freestone, D. The need for new antiviral agents. *Antivir. Res.* 1985, 5, 307–324.





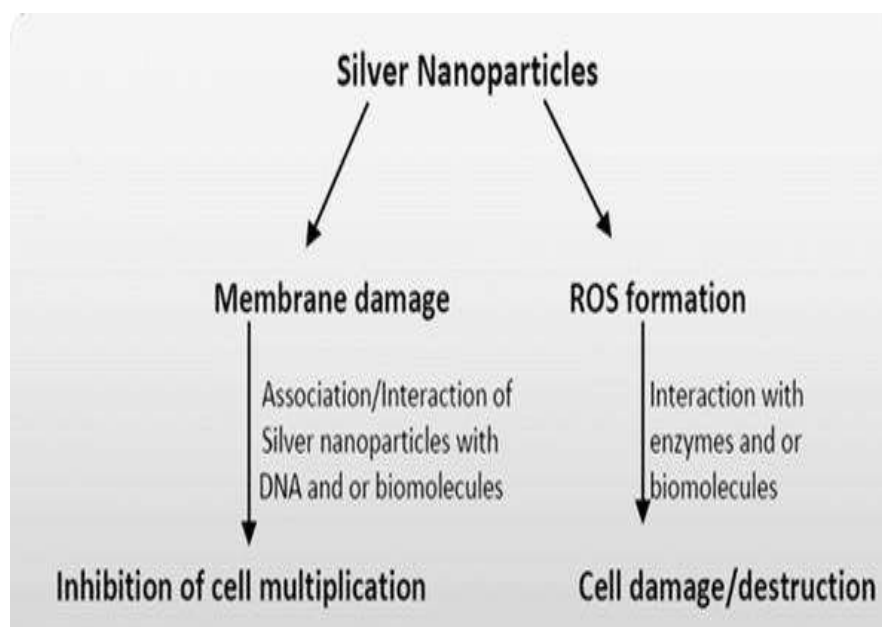
Sanjana Bhat and Meenal Rehman

5. Colpitts, C.C.; Verrier, E.R.; Baumert, T.F. Targeting viral entry for treatment of hepatitis B and C virus infections. *ACS Infect. Dis.* 2015, 1, 420–427.
6. Morens, D.M.; Folkers, G.K.; Fauci, A.S. The challenge of emerging and re-emerging infectious diseases. *Nature* 2004, 43, 242–249.
7. Bhattacharya, S. The World Health Organization and global smallpox eradication. *J. Epidemiol. Community Health* 2008, 62, 909–912.
8. Rauch, S.; Jasny, E.; Schmidt, K.E.; Petsch, B. New Vaccine Technologies to Combat Outbreak Situations. *Front. Immunol.* 2018, 9, 1963.
9. Excler, J.L.; Saville, M.; Berkley, S.; Kim, J.H. Vaccine development for emerging infectious diseases. *Nat. Med.* 2021, 27, 591–600.
10. Soares, S.; Sousa, J.; Pais, A.; Vitorino, C. Nanomedicine: Principles, properties, and regulatory issues. *Front. Chem.* 2018, 6, 360.
11. Lembo, D.; Donalisio, M.; Civra, A.; Argenziano, M.; Cavalli, R. Nanomedicine formulations for the delivery of antiviral drugs: A promising solution for the treatment of viral infections. *Expert Opin. Drug Deliv.* 2018, 15, 93–114.
12. u-Nam, Y.; Lead, J.R. Manufactured nanoparticles: An overview of their chemistry, interactions and potential environmental implications. *Sci. Total. Environ.* 2008, 400, 396–414.
13. Yadav, P.T.; Yadav, R.M.; Singh, D.P. Mechanical milling: A top down approach for the synthesis of nanomaterials and nanocomposites. *Nanosci. Nanotechnol.* 2012, 2, 22–48.
14. Park, S.; Ko, Y.-S.; Lee, S.J.; Lee, C.; Woo, K.; Ko, G. Inactivation of Influenza A Virus via Exposure to Silver Nanoparticle-Decorated Silica Hybrid Composites. *Environ. Sci. Pollut. Res.* 2018, 25, 27021–27030
15. Lu, L.; Sun, R.W.; Chen, R.; Hui, C.K.; Ho, C.M.; Luk, J.M.; Lau, G.K.; Che, C.M. Silver nanoparticles inhibit hepatitis B virus replication. *Antivir. Ther.* 2008, 13, 253.
16. Yaqoob, A. A., Umar, K., & Ibrahim, M. N. M. (2020). Silver nanoparticles: various methods of synthesis, size affecting factors and their potential applications—a review. *Applied Nanoscience*, 10, 1369-1378.
17. Almatroudi, A. (2020). Silver nanoparticles: Synthesis, characterisation and biomedical applications. *Open life sciences*, 15(1), 819-839.
18. Dong, Y.; Zhu, H.; Shen, Y.; Zhang, W.; Zhang, L. Antibacterial activity of silver nanoparticles of different particle size against *Vibrio Natriegens*. *PLoS ONE* 2019, 14, e0222322.
19. Morens, D.M.; Folkers, G.K.; Fauci, A.S. The Challenge of Emerging and Re-Emerging Infectious Diseases. *Nature* 2004, 430, 242–249. [CrossRef] [PubMed]
20. Gonçalves, B.C.; Lopes Barbosa, M.G.; Silva Olak, A.P.; Belebecha Terezo, N.; Nishi, L.; Watanabe, M.A.; Marinello, P.; Zendrini Rechenchoski, D.; Dejato Rocha, S.P.; Faccin-Galhardi, L.C. Antiviral Therapies: Advances and Perspectives. *Fundam. Clin. Pharmacol.* 2021, 35, 305–320.
21. Lau GKK. Does treatment with interferon-based therapy improve the natural history of chronic hepatitis B infection? *J Hepatol* 2007; 46:6–8.
22. Davis GL, Hoofnagle JH. Interferon in viral hepatitis: role in pathogenesis and treatment. *Hepatology* 1986; 6:1038–1041.
23. Guidotti LG, Ishikawa T, Hobbs MV, Matzke B, Schreiber R, Chisari FV. Intracellular inactivation of the hepatitis B virus by cytotoxic T lymphocytes. *Immunity* 1996; 4:25–36.
24. Sreekanth, T.V.M.; Nagajyothi, P.C.; Muthuraman, P.; Enkhtaivan, G.; Vattikuti, S.V.P.; Tettey, C.O.; Kim, D.H.; Shim, J.; Yoo, K. Ultra-Sonication-Assisted Silver Nanoparticles Using Panax Ginseng Root Extract and Their Anti-Cancer and Antiviral Activities. *J. Photochem. Photobiol. B Biol.* 2018, 188, 6–11.
25. Xiang, D.; Zheng, C.; Zheng, Y.; Li, X.; Yin, J.; O’Conner, M.; Marappan, M.; Miao, Y.; Xiang, B.; Duan, W.; et al. Inhibition of A/Human/Hubei/3/2005 (H3N2) Influenza Virus Infection by Silver Nanoparticles in Vitro and in Vivo. *Int. J. Nanomed.* 2013, 8, 4103.
26. Bekele, A.Z.; Gokulan, K.; Williams, K.M.; Khare, S. Dose and Size-Dependent Antiviral Effects of Silver Nanoparticles on Feline Calicivirus, a Human Norovirus Surrogate. *Foodborne Pathog. Dis.* 2016, 13, 239–244.
27. Ciriminna, R.; Albo, Y.; Pagliaro, M. New Antivirals and Antibacterials Based on Silver Nanoparticles. *Chem. Med. Chem.* 2020, 15, 1619–1623.



**Sanjana Bhat and Meenal Rehman**

28. Elechiguerra, J.L.; Burt, J.L.; Morones, J.R.; Camacho-Bragado, A.; Gao, X.; Lara, H.H.; Yacaman, M.J. Interaction of Silver Nanoparticles with HIV-1. *J. Nanobiotechnol.* 2005, 3, 6.
29. Mori, Y.; Ono, T.; Miyahira, Y.; Nguyen, V.Q.; Matsui, T.; Ishihara, M. Antiviral Activity of Silver Nanoparticle/Chitosan Composites against H1N1 Influenza A Virus. *Nanoscale Res. Lett.* 2013, 8, 93.
30. Baram-Pinto, D.; Shukla, S.; Perkas, N.; Gedanken, A.; Sarid, R. Inhibition of herpes simplex virus type 1 infection by silver nanoparticles capped with mercaptoethane sulfonate. *Bioconjugate Chem.* 2009, 20, 1497–1502.
31. Sun, L.; Singh, A.K.; Vig, K.; Pillai, S.R.; Singh, S.R. Silver nanoparticles inhibit replication of respiratory syncytial virus. *J. Biomed. Nanotechnol.* 2008, 4, 149–158.
32. World Health Organization. Hepatitis B-Fact Sheet. 2017 (Cited 3 2017 June). Available online: <https://www.who.int/news-room/fact-sheets/detail/hepatitis-b> (accessed on 5 October 2021).
33. Dobrovolskaia, M.A.; Aggarwal, P.; Hall, J.B.; McNeil, S.E. Preclinical studies to understand nanoparticle interaction with the immune system and its potential effects on nanoparticle biodistribution. *Mol. Pharm.* 2008, 5, 487–495.
34. Talbird, S.E.; Graham, J.; Mauskopf, J.; Masseria, C.; Krishnarajah, G. Impact of tetanus, diphtheria, and acellular pertussis (Tdap) vaccine use in wound management on health care costs and pertussis cases. *J. Manag. Care Spec. Pharm.* 2015, 21, 88–99.

**Fig.1. Synthesis of Silver Nanoparticles**



Screening of Colibactin Producing *Escherichia coli* from Urinary Isolates

S. Kaviya¹ and C. Soundhari^{2*}

¹Student, Department of Microbiology, Valliammal College for Women, (Affiliated to University of Madras), Chennai, Tamil Nadu, India.

²Associate Professor, Department of Microbiology, Valliammal College for Women, (Affiliated to University of Madras), Chennai, Tamil Nadu, India.

Received: 17 Jun 2024

Revised: 20 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

C. Soundhari

Associate Professor, Department of Microbiology,
Valliammal College for Women,
(Affiliated to University of Madras),
Chennai, Tamil Nadu, India.
E.Mail: drcsoundhari@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The study was conducted to characterize pathogenic *Escherichia coli* (*E. coli*) isolates from 35 patients with urinary tract infections (UTIs) and explore their genotoxin-producing potential, particularly focusing on the presence of the colibactin-producing gene, *clbB*. Biochemical tests confirmed the presence of *E. coli* in 22 isolates, which were then subjected to PCR analysis for the *clbB* gene. Out of the 22 isolates, 2 (9%) were found to harbor the *clbB* gene, indicating the presence of colibactin-producing *E. coli*. The size of the amplicon generated with the primer set was consistent with previous studies. These findings suggest that colibactin-positive *E. coli* is present in UTI patients, potentially implicating its role in bladder cancer development. However, the study acknowledges its retrospective nature and limited patient information, emphasizing the importance of further research, including detailed comparative genomics analysis, to understand the role of colibactin-producing *E. coli* in bladder cancer and potentially prevent its incidence.

Keywords: *Escherichia coli* [*E. coli*], UTI, *clbB* gene, Bladder cancer, PCR.

INTRODUCTION

Urinary tract infections (UTIs) are one of the most common bacterial infections, and *Escherichia coli* (*E. coli*) is the predominant bacterium responsible for these infections. A UTI refers to the presence of a certain number of bacteria in the urine (usually $> 10^5$ /ml) [Foxman 2014; Smelovet *et al.*, 2016]. UTI incidence in women is much higher than that in men [Erdem 2018]. Three or more urinary tract infections within 12 months are defined as recurrent UTIs, as are two or more recurrences within six months. Relapses are usually caused by the same type of bacteria that caused the previous infection. 95% of all UTIs occur as ascending infections [Bacheller 1997].

E. coli strains of biological significance to humans may be broadly categorised as (1) commensal strains, (2) intestinal pathogenic strains and (3) extraintestinal pathogenic *E. coli* (ExPEC) strains. Among ExPEC, strains of



**Kaviya and Soundhari**

uropathogenic *E. coli* (UPEC) are most commonly associated with human disease. *E. coli* containing *pks* is relatively common bacterium found in the human gut and can be isolated from multiple sites of the human body [Fais *et al.*, 2018] and certain strains have been identified as producers of colibactin. The genotoxic chemical compound colibactin is synthesized by a complex machinery involving polyketide synthases (PKS), non-ribosomal peptide synthases (NRPS), and is 54 kilobases in size. The genotoxic nature of colibactin is due to its ability to induce DNA damage in host cells [Nougayrede *et al.*, 2006]. This genotoxin causes DNA double-strand breaks leading to genetic mutations, chromosomal abnormalities, and cell cycle arrest in the G2/M phase chromosomal instability, and premature aging, which ultimately lead to tumorigenesis [Cuevas-Ramos *et al.*, 2010; Secher *et al.*, 2013; Cougnoux *et al.*, 2014]. *pks* islands are widely distributed in group B2 *E. coli* strains isolated from intestinal or extraintestinal sites [Forough *et al.*, 2012]. Colibactin-producing bacteria cause DNA damage in bladder cells, including regenerating urothelial cells, and colibactin is produced by clinical *pks* UPEC strains isolated from human UTIs [Camille *et al.*, 2021]. Bladder cancer is a common malignancy, worldwide; it is the seventh most prevalent cancer, accounting for 3.2% of all malignancies [El-Mosalamy *et al.*, 2012]. There is paucity of studies on uropathogenic *E. coli* producing colibactin. The current study was designed to characterize pathogenic *E. coli* from isolates obtained from patients with UTIs and attempted to explore their colibactin producing potential.

MATERIALS REQUIRED

Standard Glasswares, Nutrient agar plates, Eosin Methylene Blue agar plates, Gram's Staining reagents, Biochemical reagents, Solution A [10% SDS], Solution B [10mM Tris, 10mM MgCl₂, 10mM KCl, 2mM EDTA, 0.4mM NaCl], Solution C [100µl phenol and 100µl chloroform], 1% agarose, TE and TBE buffers, Electrophoresis unit and UV Transilluminator.

METHODOLOGY

A total of 34 *E. coli* isolates were obtained from urinary specimens of UTI patients and transported to laboratory in aseptic condition for a further process. The collected strains were streaked and labelled on Nutrient agar plates and incubated at 37°C for 24 hours. Identification of *E. coli* was done by performing standard test and biochemical assays including gram staining, motility, IMViC were performed. Selected 2 – 3 well grown colonies with the same morphology were picked and streaked on to the surface of EMB agar. The plates are then incubated at 37°C for 24 hours. The growth and colony morphology were observed.

Isolation of DNA and Agarose Gel Electrophoresis:

DNA was isolated as per the standard procedure and the separated DNA samples, along with a molecular weight marker, were visualized under a UV Transilluminator after agarose gel electrophoresis according to the standard procedure. The DNA isolation technique led to efficient extraction with good quantity and quality of DNA, which was pure and devoid of contaminants, such as RNA and Proteins.

PCR Amplification

The DNA isolated from the *E. coli* isolates were further investigated for colibactin production. The primers used for the determination of *clb+* *E. coli* isolates [Hirayama *et al.*, 2019; Tsunematsu *et al.*, 2021] are shown below:

*clb*B-F: 5'-TGTTCCGTTTTGTGTGGTTTCAGCG-3' *clb*B-R: 5'-GTGCGCTGACCATTGAAGATTCCG-3'

The primers used for the determination of *clb+* *E. coli* isolates (*ClbB*). The PCR conditions used for colibactin genes amplification were as the initial denaturation was given at 95°C for 10 minutes, denaturation at 95°C for 15 seconds, annealing at 60°C for 1 minute, extension at 72°C for 40 seconds followed by the final extension at 72°C for 7 minutes. Amplified products were visualized by agarose gel electrophoresis.





RESULTS

A total of 34 isolates from urine sample were subcultured on nutrient agar out of these 31 gave positive for nutrient agar and 3 isolates failed to grow. [Fig.1]

Of the 31 isolates, 20 isolates gave positive for the following tests:

Gram Staining – Gram-Negative rods were observed. [Fig.2] Motility Test – it was observed to be motile.

Biochemical Analysis – it was observed positive for Indole and MR and negative for VP and Citrate test. [Fig.3]

Catalase Test – Positive [Fig.4]; Oxidase Test – Negative. [Fig.5]

Also, these strains showed positive result for catalase test [Fig.4] and negative result for oxidase test [Fig.5].

The 20 strains showed green metallic sheen in EMB [Fig.6] and positive for *E. coli* were picturized

The positive *E. coli* strains obtained from EMB plates were further subcultured on

Nutrient broth and DNA was isolated and was further confirmed by Agarose Gel Electrophoresis.

Results of Agarose Gel Electrophoresis

After electrophoresis, the gel was examined under UV light, and the DNA bands were observed [Fig.7]. The presence of bright bands at specific positions on the gel indicated the presence of DNA fragments.

Detection of Colibactin Genes by PCR

The extracted DNA was used as a template in PCR reactions, along with the designed primers and PCR reagents (polymerase, nucleotides, buffer). The PCR conditions (annealing temperature, cycle number, etc.) were optimized to ensure specific amplification of the target genes. The PCR products were separated by gel electrophoresis to confirm the presence and size of the amplified fragments. The presence of a band of the expected size indicated the presence of the target gene in the sample. The amplified product was detected to be around 450 basepair in size and it was indicated by the presence of band [Fig.8].

DISCUSSION

Urinary tract infections (UTIs) are among the most common infections. *Escherichia coli* strains, termed uropathogenic *E. coli* (UPEC) are the most common causative agent in both uncomplicated and complicated UTIs causing approximately 80% of all UTIs. This biosynthetic gene encodes for a secondary metabolite named colibactin, putatively acquired through horizontal gene transfer as part of a mobile genetic element. The current study was designed to characterize pathogenic *E. coli* from isolates obtained from 35 patients with UTIs and explore their genotoxin or colibactin producing potential. All the bacterial isolates were tested with a set of standard biochemical tests and their culture characteristics on MacConkey agar and nutrient agar and were identified based on colony morphology on nutrient agar, MacConkey's agar and EMB agar. 4–5 suspected colonies from nutrient agar bacterial plate were picked, cultured and then identified by the various biochemical tests. Biochemical tests were performed to confirm *E. coli* using Gram staining, catalase test, Indole, Methyl red, Voges-Proskauer test, Simmons citrate agar and various sugar fermentation tests. Bacterial samples with biochemical and culture characteristics consistent with those of *E. coli* were considered to be confirmed to be *E. coli* and evaluated in further testing.

E. coli infection might play a role in the development of bladder cancer. A study based on a rat model by El-Mosalamy *et al.*, 2012; Ashmawey *et al.*, 2011 and Chagneau *et al.*, 2021 showed that *E. coli* infection in the bladder tissues increases the carcinogenic ability and tumor enhancing effect of *E. coli*. UPEC strains which routinely cause infections have been shown to belong to phylogroups B2 and are often the same ones responsible for UTIs, but to date, a few studies have been conducted on colibactin and UTIs. The current study was initiated as a screening effort



**Kaviya and Soundhari**

to examine the prevalence of *clb* + *E. coli*. In the present study we attempted to analyse for the presence of colibactin producing gene *clbB* in the 20 of the 22 isolates which were biochemically confirmed as *E. coli*. DNA was extracted from the isolates. The extracted DNA was subjected to PCR and qualitatively analysed for *clbB* present in the DNA extract by amplifying the gene fragment using a primer set. The primers were designed for the determination of colibactin-producing *E. coli* *ClbB* gene. In the present study, we observed that 2 out of the 22 Uropathogenic *Escherichia coli* isolates harboured *clbB* *E. coli*. The size of the *clb+* amplicon generated with the primer set *clb-F*/*clb-R* is around 450 bp. This is in accordance with Tsunematsu *et al.*, 2021 who used the same primers for amplification and received an amplicon size of 555bp and Hirayama *et al.*, 2019 who carried a PCR-based screening of the *E. coli* isolates for the presence of the *clb* gene cluster and demonstrated a 464-bp product. The search however did not identify *clbB* *E. coli* in remaining 18 isolates, as isolates without *clb* gene cluster would not yield any product. We initiated a screening effort to examine the prevalence of *clb* + *E. coli* among individuals with UTI. Screening of *E. coli* isolates led to the isolation of colibactin producer *E. coli*. Our study confirms the occurrence of *pks+* UPEC. Our findings indicate that colibactin positive *E. coli* appears to be produced in humans, by detecting it in the urine isolates obtained from patients suffering from urinary tract infections. Hence phenotypical and genotypical characters of the infecting *E. coli* virulent strains need to be done which would be a possibility of preventing bladder cancer at early stages.

CONCLUSION

Our findings indicate that virulent strains capable of causing UTI harbour *clbB+* gene. Here, we show that this genotoxin is produced in humans, by detecting it in the isolates obtained from urine of patients suffering from urinary tract infections. However, this study is a retrospective analysis and only a limited patient information has been collected. It will be worth checking the production of colibactin in all UTI isolates by detailed comparative genomics analysis for a better understanding of their role in bladder cancer. We conclude that to reduce the number of bladder cancer incidences and mortalities, it is vital to identify all the *E. coli* infection for *clbB+* gene as it might play a role in the development of bladder cancer.

REFERENCES

1. Foxman B. Urinary tract infection syndromes occurrence, recurrence, bacteriology, risk factors, and disease burden. *Infect Dis Clin North Am* 2014; 28: 1-13.
2. Smelov V, Naber K, Bjerklund Johanaen TE. Improved classification of urinary tract infection: future considerations. *Eur Urol Suppl* 2016; 15: 71-80.
3. Erdem I, Kara Ali R, Ardic E, Elbasan Omar S, Mutlu R, Topkaya AE. Community Acquired lower urinary tract infections: Etiology, antimicrobial resistance, and treatment results in female patients. *J Glob Infect Dis* 2018; 10: 129-132.
4. Bacheller CD, Bernstein JM. Urinary tract infections. *Med Clin N Am* 1997; 81: 719-730.
5. Tiphane Fais, Julien Delmas, Nicolas Barnich, Richard Bonnet, Guillaume Dalmaso. Colibactin: More than a New Bacterial Toxin. *MDPI Toxins* 2018; 10:151-166.
6. Nougayrede JP, Homburg S, Taieb F, Boury M, Brzuszkiewicz E, Gottschalk G, *et al.* *Escherichia coli* induces DNA double-strand breaks in eukaryotic cells. *Science* 2006; 313: 848-851.
7. Cuevas-Ramos G, Petit CR, Marcq I, Boury M, Oswald E, Nougayrede JP. *Escherichia coli* induces DNA damage *in vivo* and triggers genomic instability in mammalian cells. *Proc Natl Acad Sci* 2010; 115:11537-11542.
8. Secher T, Samba-Louaka A, Oswald E, Nougayrede JP. *Escherichia coli* producing colibactin triggers premature and transmissible senescence in mammalian cells. *Plos One* 2013; 8(10): 1-17.
9. Cougnoux A, Dalmaso G, Martinez R, Buc E, Delmas J, Gibold L, *et al.* Bacterial genotoxin colibactin promotes colon tumor growth by inducing a senescence associated secretory phenotype. *Gut* 2014; 63: 1932-1942.
10. Forough L, Nowrouzian and Eric Oswald. *Escherichia coli* strains with the capacity for long-term persistence in the bowel microbiota carry the potentially genotoxic *pks* island. *Microbial Pathogenesis* 2012; 53: 180-182.





Kaviya and Soundhari

11. Camille V Chagneau, Clemence Massip, Nadege Bossuet-Greif, Christophe Fremez, Jean-Paul Motta, Ayaka Shima, *et al.* Uropathogenic *E. coli* induces DNA damage in the bladder. *Plos Pathogens* 2021; 17(2): 1-21.
12. Hala El-Mosalamy, Tarek M Salman, Abeer M Ashmawey and Nada Osama. Role of chronic *E. coli* infection in the process of bladder cancer- an experimental study. *Infect Agents Cancer* 2012; 7: 19-26.
13. Yuchiro Hirayama, Yuta Tsunematsu, Yuko Yoshikawa, Ryota Tamafune, Nobuo Matsuzaki, Yuji Iwashita, *et al.* Activity-Based probe for screening of High-Colibactin producers from clinical samples. *J Am Chem Soc* 2019; 12: 4490-4494.
14. Yuta Tsunematsu, Koji Hosomi, Jun Kunisawa, Michio Sato, Noriko Shibuya, Emiko Saito *et al.* Mother-to-infant transmission of the carcinogenic colibactin-producing bacteria. *BMC Microbiol* 2021; 21: 235-244.
15. Ashmawey AM, Mohamed WS, Abdel-Salam IM, El-Gendy SM, Ali AI, El-Aaser AA. Role of urinary tract bacterial infection in the process of bladder carcinogenesis (molecular and biochemical studies). *Asian J Med Sci* 2011, 2: 31-40.



Fig: 1 – Growth of urinary isolates on Nutrient Agar

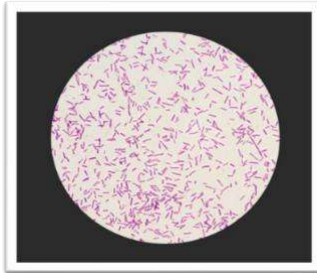


Fig: 2 - Gram staining of *E. coli*

Indole [A]	+
Methyl Red [B]	+
Vogues Proskauer [C]	-
Citrate Utilization test [D]	-

Table: 3 – Biochemical analysis

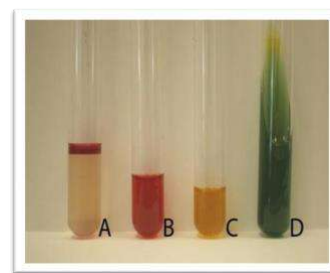


Fig: 4 – Biochemical analysis of *E. coli*



Fig: 5 - Catalase Positive



Fig: 6 - Oxidase Negative





Kaviya and Soundhari

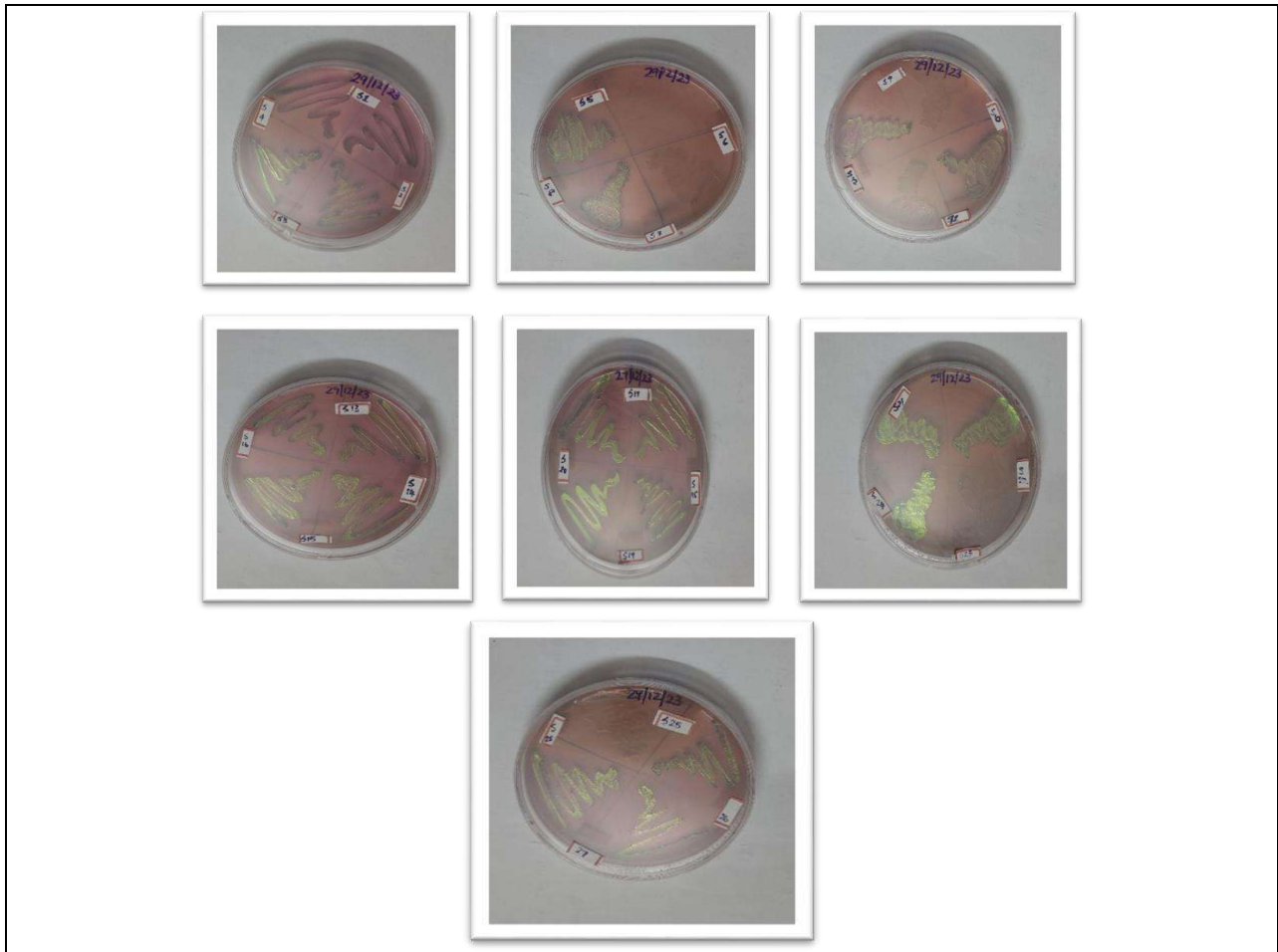


Fig: 7 - *E. coli* isolates showing metallic sheen on EMB agar

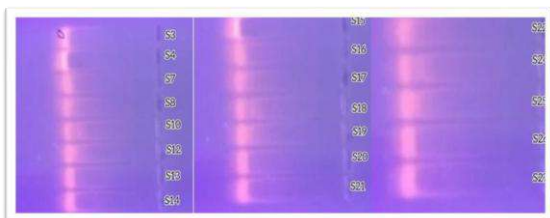


Fig: 7 - DNA bands on agarose gel electrophoresis

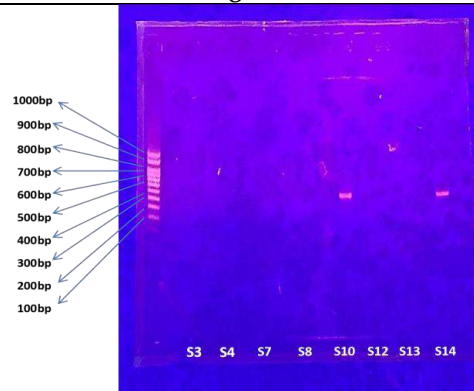


Fig: 8 - Polymerase chain reaction assays for colibactin gene. Lane 1: Standard control; Lane 2 to Lane 5: Strains 3, 4, 7, 8 showing negative for *clbB* gene; Lane 6: Strain 10 showing positive for *clbB* gene; Lane 7 and Lane 8: Strains 12 and 13 showing negative for *clbB* gene; Lane 9: Strain 14 showing positive for *clbB* gene;





Cardioprotective Activity of *Hylocereus undantus* Fruit Peel Extract against Cyclophosphamide Induced Cardiotoxicity in Rats

Mallikarjuna Gandla^{1*}, Navaneeswar Bathala², Arshiya Pathan², Gunasekhar Reddy Mukkandla², Madhan Mohan Gangarapu², Sameera Bee Kamalapuram² and Niranjana Babu Mudduluru³

¹Associate Professor, Department of Pharmacology, Seven Hills College of Pharmacy (A), Tirupati, Andhra Pradesh, India

²Student, Department of Pharmacognosy, Seven Hills College of Pharmacy, Tirupati, Andhra Pradesh, India.

³Professor, Department of Pharmacognosy, Seven Hills College of Pharmacy, Tirupati, Andhra Pradesh, India.

Received: 24 Jun 2024

Revised: 22 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

Mallikarjuna Gandla¹

Associate Professor,

Department of Pharmacology,

Seven Hills College of Pharmacy (A),

Tirupati, Andhra Pradesh, India

E.Mail: malli.g53@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License (CC BY-NC-ND 3.0)** which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Cardioprotective activity of *Hylocereus undantus* fruit peel extract against Cyclophosphamide induced Cardiotoxicity in rats. Cardiotoxicity was induced in Wistar rats by administering single injection cyclophosphamide (150 mg/kg) intraperitoneally on first day of experimental period. *Hylocereus undantus* (250 and 500 mg/kg, p.o.) was administered for 11 days after the single dose administration of cyclophosphamide (150 mg/kg) intraperitoneally. Plasma parameters, Cardiac biomarkers, pro-oxidant and antioxidant parameters and Histopathological Examinations were measured. Cyclophosphamide administration significantly increased lipid peroxidation and decreased the levels of antioxidant markers such as reduced superoxide dismutase and catalase. Cyclophosphamide elevated the levels of biomarker enzymes creatine kinase isoenzyme MB, lactate dehydrogenase, aspartate transaminase, alanine transaminase and alkaline phosphatase and reduced Total protein levels. Treatment with *Hylocereus undantus* extract significantly reversed the status of altered plasma biochemical parameters, cardiac biomarkers and oxidative enzymes in cyclophosphamide induced cardiotoxicity. Potential cardioprotective effect of *Hylocereus undantus* was supported by histopathological examination that reduced severity of cellular damage of the myocardial fibres. The biochemical, cardiac and histopathology reports support the cardioprotective effect of *Hylocereus undantus* which could be attributed to antioxidant activity.

Keywords: Cardioprotective; *Hylocereus undantus*; Cyclophosphamide; Oxidative stress; Myocardial infarction.





Mallikarjuna Gandla et al.,

INTRODUCTION

Cardiovascular disease (CVD) is increasingly recognized as the leading cause of death. The epidemiological change of the twentieth century established cardiovascular disease as the primary cause of disability worldwide. Global health projections imply that it will be the major cause of death in 2030. CVD is a set of heart and blood vessel illnesses. It comprises coronary heart disease (CHD), peripheral arterial disease (PAD), various types of angina, congestive heart failure (CHF), and myocardial infarction (MI) [1]. Cardiac damage is a potential short- or long-term complication of various anticancer treatments, which increases morbidity and death significantly [2, 3]. However, as cancer survival rates have increased due to advances in therapy, late cardiovascular adverse effects have emerged as a significant management challenge, notably in children malignancies, lymphoma, leukemia, and breast cancer [4]. Endocardial lipid peroxidation is caused by the excessive production of reactive oxygen species (ROS). Such damage causes the loss of cardioprotective antioxidants, increased oxidative stress, and apoptosis [5]. Medicinal plants include several antioxidant chemicals that protect against a variety of ailments, making them potential alternative therapeutic ingredients. Herbal antioxidants are increasingly being used to protect against a variety of cardiovascular problems. Bioactive substances derived from natural sources have become increasingly important in modern medicine, lowering the risk of cardiac diseases by scavenging free radical production [6].

Hylocereus undatus (Haw.) Britton and Rose belong to the Cactaceae family. The fruit is sweet and juicy, measuring up to 7 to 14 cm long and 5 to 9 cm wide, with a white, red, or purple pulp and numerous small, black, shiny seeds [7]. The Mayas utilized the fruit as hypoglycaemic, diuretic, heart disease prevention, wound disinfecting, tumour dissolving with stem sap, and dysentery treatment. Natural colorants may be recovered from the pulp and peel, and they have a high potential for application as colorants in the food industry due to their stability throughout processing and storage. *Hylocereus undatus* has high levels of vitamin B1, B2, B3, and C, as well as protein, fat, carbohydrate, crude fibre, and other nutrients and minerals. Thiamine, niacin, pyridoxine, cobalamin, glucose, flavonoid betacyanins, polyphenols, carotenoids, phosphorus, phenolic phytoalbumin, and iron. The peel contains more flavonoids than the meat, and ethanolic extracts of *H. undatus* peel and flesh have been shown to have varying antioxidant capabilities, anti-cancer activity, antimicrobial activity, hypocholesterolemic effect, and prebiotic effect [8-12]. The present study was aimed at investigating the Cardioprotective activity of *Hylocereus undatus* fruit peel extract against Cyclophosphamide induced Cardiotoxicity in rats.

MATERIALS AND METHODS

Hylocereus undatus fruit peels were collected from commercial fruit juice shops in the surrounding areas of Tirupati and authenticated by Dr. M. Niranjan Babu, Professor, Department of Pharmacognosy, Seven Hills College of Pharmacy, Tirupati, Andhra Pradesh, India. The fruit peels were washed, chopped into little 2-3cm pieces, and shade dried at room temperature for 15 days. The shade-dried fruit peels were finely grounded using a dry grinder and was soaked in 70% hydroalcoholic solvent at a 1:4 ratio in a narrow mouthed bottle for seven days, shaking occasionally using the maceration process. The extract was filtered with Whatman filter paper, and the solvent was evaporated to dryness using a rotary evaporator [13, 14]. The hydroalcoholic extract of *Hylocereus undatus* (HAHU) was used for pharmacological screening of Cardioprotective activity.

Animal Husbandry

The current study used healthy adult male wistar rats measuring 200-235 grams. Rats were housed in polypropylene cages with a standardized 12-hour light/dark cycle, $24 \pm 2^\circ\text{C}$ temperature, and 35 to 60% humidity. They had access to a pellet food and filtered drinking water at all times. The animals were fasted 24 hours before the induction of cardiotoxicity, although they had free access to water during the experimental period.



**Mallikarjuna Gandla et al.,**

Experimentation

Cardiotoxicity had been demonstrated in rats by administering a single dose of cyclophosphamide in normal saline (150 mg/kg, body weight) intraperitoneally [15]. Animals were divided into four groups of six each. Group I administered with 0.5mL of normal saline p.o. as the vehicle control. Group II had a single dose of cyclophosphamide in normal saline (150 mg/kg, b.wt.) intraperitoneally on Day 01 served as cardiotoxic control. Group III received hydroalcoholic extract of *Hylocereus undatus* (250 mg/ kg, b.wt. p.o.) and single dose of cyclophosphamide in normal saline (150 mg/kg, b.wt.) intraperitoneally served as HAHU-I. Group IV received hydroalcoholic extract of *Hylocereus undatus* (500 mg/ kg, b.wt. p.o.) and single dose of cyclophosphamide in normal saline (150 mg/kg, b.wt.) intraperitoneally served as HAHU-II. The total duration of the experimentation was carried out for 11 days.

On day 12, each animal's blood was extracted via retro-orbital route and centrifuged at 3000 rpm for 10 minutes to separate plasma. The Plasma samples were tested for Serum Glutamate Oxalate Transaminase (SGOT), Serum Glutamate Pyruvate Transaminase (SGPT), Alkaline Phosphatase (ALP), and total protein using commercially available kit through biochemistry analyser (Microlab 400) [16-19] and Cardiac Biomarkers Lactate dehydrogenase (LDH) and Creatine kinase isoenzyme MB (CK-MB) [20]. Catalase (CAT), Superoxide Dismutase (SOD) and Malondialdehyde (MDA) levels were measured for the pro-oxidant and antioxidant activity [21-23]. The heart tissues were isolated and immediately rinsed with saline and preserved in a 10% formalin solution. Following fixation, the heart tissues were treated in an alcohol-xylene series and embedded in paraffin. The serial sections were cut and stained with haematoxylin and eosin. The slides were viewed under a microscope, and images were taken.

STATISTICAL ANALYSIS

The results were expressed as the mean \pm SEM and analyzed using one way ANOVA followed by Dunnett's comparison test with Cardiotoxic control. Data were computed for statistical analysis using the graph pad software (Prism 5.0 version).

RESULTS

Figure 1: Effect of HAHU on plasma biochemical parameters in Cyclophosphamide induced Cardiotoxicity in rats Values were expressed as Mean \pm SEM (n=6). *P<0.05,**P<0.01,***P<0.001.as compared with Cardiotoxicity control (One-way ANOVA followed by Dunnet's test). Figure 2: Effect of HAHU on plasma Total protein in Cyclophosphamide induced Cardiotoxicity in rats Values were expressed as Mean \pm SEM (n=6). *P<0.05,**P<0.01,***P<0.001.as compared with Cardiotoxicity control (One-way ANOVA followed by Dunnet's test). Figure 3: Effect of HAHU on LDH and CK-MB parameters in Cyclophosphamide induced Cardiotoxicity in rats Values were expressed as Mean \pm SEM (n=6). *P<0.05,**P<0.01,***P<0.001.as compared with Cardiotoxicity control (One-way ANOVA followed by Dunnet's test). Figure 4: Effect of HAHU on Pro-oxidant and antioxidant parameters in Cyclophosphamide induced Cardiotoxicity in rats Values were expressed as Mean \pm SEM (n=6). *P<0.05,**P<0.01,***P<0.001.as compared with Cardiotoxicity control (One-way ANOVA followed by Dunnet's test).

DISCUSSION

Cyclophosphamide is an alkylating agent used in cancer therapy [24]. It is commonly used to treat leukaemia's, lymphomas, multiple myeloma, rheumatoid arthritis, and to prepare for bone marrow transplantation [25]. Despite its several therapeutic uses, it is a cardiotoxic drug that promotes endothelial dysfunction and myocardial cell death [26]. Oxidative stress is a main cause of cyclophosphamide induced Cardiotoxicity [27]. Oxidative stress can lead to endothelial dysfunction, hypertrophy, fibrosis, inflammation, apoptosis, cell migration, and angiogenesis [28]. Cyclophosphamide induced cardiotoxicity is primarily caused by its toxic product, acrolein, which interacts with the



**Mallikarjuna Gandla et al.,**

tissue antioxidant defense system and produces an excessive amount of reactive oxygen species [29]. There is currently no viable medication to alleviate the cardiotoxic effects of Cyclophosphamide. Developing alternate ways to protect against Cyclophosphamide induced cardiotoxicity will be of enormous therapeutic value. In addition to their traditional values, herbal medicines are highly sought after by the public and medical community globally as sources of innovative lead chemicals for pharmaceutical development. As a result, medicinal plants will serve as a natural defensive approach and will be readily available at a lower cost than synthetic medications [30]. The present study was aimed at investigating the Cardioprotective activity of *Hylocereus undatus* fruit peel extract against Cyclophosphamide induced Cardiotoxicity in rats.

The activity of the liver enzymes ALT, AST, ALP, and TP in plasma are reliable indicators of hepatic and cardiac damage [31]. Cyclophosphamide increases plasma levels of these enzymes, indicating cellular injury and loss of functional integrity cell membrane, resulting in their leaking into the serum or plasma [32-34]. AST is an enzyme that is prevalent in the liver's cytoplasm and mitochondria, as well as the heart, skeletal muscle and brain. ALT is a hepatospecific enzyme primarily present in the cytoplasm [35, 36]. ALP is associated with the cell membrane, and its rise in the plasma is an indicator of impaired intrahepatic and extrahepatic bile flow (cholestasis), hepatobiliary damage, and overproduction or leakage of ALP [37, 38]. From the result of this study, pre-treatment with HAHU at both dose levels restored the activities of ALT, AST, ALP and TP [39]. LDH is an important enzyme involved in energy production, and it is a good biomarker indicating cell damage [40]. Additionally, CK-MB is an enzyme found mostly in the heart and presents with significantly high amounts during cardiac damage. In this study, there was a considerable rise in LDH and CK-MB activity following single administration of cyclophosphamide compared to the control group, which reflects the severe cardiotoxicity of cyclophosphamide, as ensured by the results of earlier investigations. Cyclophosphamide is cardiotoxic because it causes endothelial dysfunction and damage cardiomyocytes. Furthermore, cyclophosphamide induces lipid peroxidation, which disrupts endothelial cell permeability, leading to increased serum LDH and CK-MB levels [41]. However, by the protective administration of HAHU with cyclophosphamide, serum levels of LDH and CK-MB significantly diminished compared with the cyclophosphamide treated group.

The current study found that rats treated with cyclophosphamide had significantly lower SOD and CAT activity, as well as higher MDA levels, when compared to the vehicle control group. It has been observed that free radicals generated during CP treatment cause membrane damage, resulting in the loss of function and integrity of the cardiac membrane [15]. Cyclophosphamide causes a considerable decrease in SOD and CAT activity, which promotes the production of OH radicals and the onset and propagation of lipid peroxidation. However, it is proposed that the decrease in the activity of antioxidant enzymes is the result of increased oxidative stress in the cardiac tissues due to the overproduction of active reactive oxygen species [42,43]. Treatment of Cyclophosphamide treated animals with HAHU restored the antioxidant enzymes (SOD and CAT) activities and decreased MDA levels in the cardiac tissues towards control level, indicating a protective effect of HAHU against reactive oxygen species. This action could be attributed to its ability to reduce oxidative stress and preserve the activity of antioxidant enzymes, as well as its ability to suppress lipid peroxidation hydroxyl radical [44].

Histopathological comes about of heart tissue in control rats heart tissue appeared intaglio myocardial strands and pericardium; whereas cyclophosphamide (CP) rats heart tissue exhibited enormous alter in the myocardium appearing a shifting degree of vacuolar changes in the cardiac muscle filaments, basically in the frame of degeneration of myocardial strands, vacuolization of the cardiomyocytes, penetration of provocative cells, myofibrillar damage, and hypertrophic myocardial fibre with irritation; HAHU-I (Low Dose test Extract) treated showed direct greasy modification in certain muscle cells and separation of a few cardiac strands; and HAHU-II (High Dose test extract) treated rats showed enhancement in histoarchitecture.





Mallikarjuna Gandla et al.,

CONCLUSION

The current study found that the hydroalcoholic extract of *Hylocereus undantus* fruit peels had strong Cardioprotective action in various parameters, which is dose dependant. The extract has the strongest protective impact against the negative effects of cyclophosphamide on plasma profiles, cardiac biomarkers, and oxidative stress parameters. The extract's antioxidant polyphenolic components may be responsible for its cardioprotective activity. However, chemical isolation is recommended in order to discover the precise phytoconstituents responsible for *Hylocereus undantus* crude extract's cardioprotective properties. More relevant models with direct heart injury and endogenous antioxidant enzymes should be used to support the finding. Furthermore, proteomic analysis is necessary to analyse protein expression levels.

REFERENCES

1. Khan IA, Hussain M, Syed SK, Saadullah M, Alqahtani AM, Alqahtani T, Aldahish AA, Asiri S, Zeng LH. Pharmacological justification for the medicinal use of *Plumeria rubra* Linn. In cardiovascular disorders. *Molecules* 2022; 27: 251.
2. Cardinale D, Colombo A, Lamantia G. Anthracycline induced cardiomyopathy: Clinical relevance and response to pharmacologic therapy. *JACC*. 2010; 55: 213–220.
3. Steinherz LJ, Steinherz PG, Tan CT. Cardiac toxicity 4 to 20 years after completing anthracycline therapy. *JAMA*. 1991; 266: 1672–1677.
4. Kumar S, Gupta R.K., Samal N. Cyclophosphamide Cardiotoxicity. *The National Medical Journal of India*. 1992; 5(1): 17–19.
5. Khan IA, Hussain M, Munawar SH, Iqbal MO, Arshad S, Manzoor A, Shah MA, Abbas K, Shakeel W, Syed SK. *Jasminum sambac*: A Potential candidate for drug development to cure cardiovascular ailments. *Molecules* 2021; 26: 5664.
6. Wang CZ, Mehendale SR, Yuan CS. Commonly used antioxidant botanicals: active constituents and their potential role in cardiovascular illness. *Am J Chin Med*. 2007; 35(04): 543–558.
7. Le Bellec F, Vaillant F, Imbert E. Pitahaya (*Hylocereus* spp.): A new fruit crop, a market with a future. *Fruits* 2006; 61(4): 237-250. DOI: 10.1051/fruits:2006021
8. Manihuruk FM, Suryati T, Arief II. Effectiveness of the red dragon fruit (*Hylocereus polyrhizus*) peel extract as the colorant, antioxidant, and antimicrobial on beef sausage. *Media Peternak* 2017; 40(1): 47-54. DOI: 10.5398/medpet.2017.40.1.47.
9. Saravanakumar K, Sanjeev Rao T, Nagaveni P. A Review on solubility enhancement techniques. *Journal of Comprehensive Pharmacy*. 2015; 2(2): 36-41.
10. Hitendraprasad PP, Hegde K. *Hylocereus* spp. (Dragon fruit): A brief review. *Int J Pharm Sci Rev Res*. 2020; 60(1): 55-57.
11. Mohd Adzim Khalili R, Norhayati AH, Rokiah MY, Asmah R, Siti Muskinah M, Abdul Manaf A. Hypocholesterolemic effect of red pitaya (*Hylocereus* sp.) on hypercholesterolemia induced rats. *Int Food Res J* 2009; 16(3): 431-440.
12. Mohd Adzim Khalili R, Che Abdullah AB, Abdul Manaf A. Isolation and characterization of oligosaccharides composition in organically grown red pitaya, white pitaya and papaya. *Int J Pharm Pharm Sci*. 2014; 6(Suppl. 2): 131-136.
13. Nikhal SB, Dambe PA, Ghongade DB, Goupale DC. Hydroalcoholic extraction of *Mangifera indica* (leaves) by Soxhletion. *International Journal of Pharmaceutical Sciences*. 2010; 2(1): 30-32.
14. Shaha S, Mohana M, Kastureac S, Ballerobd M, Maxiabc A, Sanna C, Protective effect of *Hypericum hircinum* on doxorubicin induced cardiotoxicity in rats. *Natural Product Research*. 2013; 27(16): 1502–1507.
15. Saravanakumar K, Radhika Chikatipalli, Chandra Sekhar Kothapalli Bonnoth. Anti-Arthritic Activity of *Amaranthus Roxburghianus* Nevski Hydroalcoholic areal Plant Extract In Acute and Chronic Models in Albino Wistar Rats. *Asian J Pharm Clin Res*. 2020; 13(6): 111-115.



**Mallikarjuna Gandla et al.,**

16. Hamza A, Amin A, Daoud S. The protective effect of a purified extract of *Withania somnifera* against doxorubicin induced cardiac toxicity in rats. *Cell Biology and Toxicology*. 2008; 24(1): 63–73.
17. Singh G, Singh AT, Abraham A. Protective effects of *Terminalia arjuna* against doxorubicin induced Cardiotoxicity. *Journal of Ethnopharmacology*. 2008; 117(1): 123–129.
18. Octavia Y, Tocchetti CG, Gabrielson KL, Janssens S, Crijns HJ, Moens AL. Doxorubicin induced cardiomyopathy: from molecular mechanisms to therapeutic strategies. *Journal of Molecular and Cellular Cardiology*. 2012; 52(6): 1213–1225.
19. Urdal P, Landaas S. Macro creatine kinase BB in serum, and some data on its prevalence. *Clin. Chem*. 1979; 25: 461-465.
20. Madhu Medabalimi, Saravanakumar K, Satyanarayana SV. Development and Validation of Stability Indicating RP-HPLC Method for Quantitative Estimation of *Safinamide Mesylate* in Bulk and its Tablet Dosage Form. *Current Trends in Biotechnology and Pharmacy*. 2022; 16(2): 50-59.
21. Kakkar P, Das B, Viswanathan PN. A modified spectroscopic assay of superoxide dismutase. *Indian Journal of Medical Research*, 1984; 21: 130-132.
22. Sinha AK. Colorimetric assay of catalase. *Analytical Biochemistry*. 1972; 47: 389-3.
23. Fraga CG, Leiboutiz BE, Toppel AL. Lipid peroxidation measured as TBARS in tissue slices: characterization and comparison with homogenates and microsomes. *Free Radical Biology and Medicine*. 1998; 4: 155-161.
24. Bjelogrić SK, Radic J, Jovic V, Radulovic S. Activity of d, l-alpha-tocopherol (vitamin E) against cardiotoxicity induced by doxorubicin and doxorubicin with cyclophosphamide in mice. *Basic Clinical Pharmacology Toxicology*. 2005; 97(5): 311–319.
25. Viswanatha Swamy AH, Patel UM, Koti BK, Gadad PC, Patel NL, Thippeswamy AH. Cardioprotective effect of *Saraca indica* against cyclophosphamide induced cardiotoxicity in rats: a biochemical, electrocardiographic and histopathological study. *Indian Journal of Pharmacology*. 2013; 45(1): 44–48.
26. Mansour HH, El Kiki SM, Hasan HF. Protective effect of N-acetylcysteine on cyclophosphamide induced cardiotoxicity in rats. *Environmental Toxicology and Pharmacology*. 2015; 40(2): 417–422.
27. Iqbal A, Iqbal MK, Sharma S. Molecular mechanism involved in cyclophosphamide induced cardiotoxicity: old drug with a new vision. *Life Sciences*. 2019; 218: 112–131.
28. Ashok Thulluru, Nawaz Mahammed, Saravanakumar K. Optimization of HPMC K100M and Sodium Alginate Ratio in Metronidazole Floating Tablets for the Effective Eradication of *Helicobacter pylori*. *Asian Journal of Pharmacy and Technology*. 2019; 9(3): 1-8.
29. Rania A. Elrashidy, Rehab A. Hasan, Cilostazol preconditioning alleviates cyclophosphamide induced cardiotoxicity in male rats: Mechanistic insights into SIRT1 signaling pathway. *Life Sciences*. 2021; 266: 118822.
30. Dianat M, Radan M, Badavi M, Sarkaki A. The evaluation of inotropic properties and antidysrhythmic effect of vanillic acid and exercise on CaCl₂ induced arrhythmia in young and aged rats. *Research Journal of Pharmaceutical, Biological and Chemical Sciences*. 2014; 5: 1545.
31. Amacher DE. Serum Transaminase Elevations as Indicators of Hepatic Injury Following the Administration of Drugs. *Regulat. Toxicol. Pharmacol*. 1998; 27: 119-130.
32. Kumar G, Banu SG, Kannan V, Pandian RM. Antihepatotoxic effect of β-carotene on paracetamol induced hepatic damage in rats. *Ind. J. Exp. Biol*. 2005; 43: 351–355.
33. Kaur G, Alam MS, Jabbar Z, Javed K, Athar M. Evaluation of antioxidant activity of *Cassia siamea* flowers. *J. Ethnopharmacol*. 2006; 108: 340–348.
34. Sreetha S, Padma PR, Umadevi M. Effect of *Coriandrum sativum* extracts on carbon tetrachloride induced hepatotoxicity in rats. *Food Chem. Toxicol*. 2009; 47: 702–708.
35. Habibi E, Shokrzadeh M, Chabra A, Naghshvar F, Keshavarz Maleki R, Ahmadi A. Protective effects of *Origanum vulgare* ethanol extract against cyclophosphamide induced liver toxicity in mice. *Pharm. Bio*. 2015; 53:1–6.
36. Nyblom H, Bjornsson E, Simren M, Aldenborg F, Almer S, Olsson R. The AST/ALT ratio as an indicator of cirrhosis in patients with PBC. *Liver Int*. 2006; 26: 840–845.
37. Mandlem VKK, Gouri Priya N, Raghavendra M, Abbulu K. Evaluation of Cardioprotective Activity of *Tamarindus indica* Linn Pericarpic extract in Doxorubicin induced Cardiotoxicity in Experimental Rats. *Research J. Pharm. and Tech*. 2020; 13(7): 3267-3273. doi: 10.5958/0974-360X.2020.00579.X.





Mallikarjuna Gandla et al.,

38. Ramaiah SK. A toxicologist guide to the diagnostic interpretation of hepatic biochemical parameters. Food Chem. Toxicol. 2007; 45: 1551–1557.
39. Mahmoud RH, Barakat H. The protective effect of gallic acid and caffeine against CCl₄ induced oxidative hepatotoxicity and mitochondrial DNA depletion in male albino rats. Egypt. J. Biochem. Mol. Biol. 2010; 28: 543–562.
40. El-Agamy DS, Elkablawy MA, Abo-Haded HM. Modulation of cyclophosphamide induced cardiotoxicity by methyl palmitate. Cancer Chemother Pharmacol. 2017; 79: 399–409. doi: 10.1007/s00280-016-3233-1.
41. Hasitha Diana Manohar, Srinivasan D, Sengottuvelu S. Cardioprotective activity of *Limonia acidissima* against Isoproterenol Induced Myocardial Infarction in Rats. Research J. Pharm. and Tech. 2016; 9(5): 541-544. doi: 10.5958/0974-360X.2016.00102.5
42. Hardik Joshi, Manoj Pagare, Leena Patil, Vilasrao Kadam. Cardioprotective Effect of Ethanolic Extract of *Buchanania lanzan* Spreng. Against Isoproterenol Induced Myocardial Infarction in Rats: A Biochemical, Electrocardiographic Evaluation. Research J. Pharm. and Tech. 5(2): Feb. 2012; Page 263-266.
43. Radhika Chikatipalli, Saravanakumar K, Chandra Sekhar Kothapalli Bannooh, Pharmacognostic evaluation and free radical scavenging activity of *Bombax ceiba* leaf extracts. International Journal of Green Pharmacy. 2021; 15 (1): 59-65.
44. Merwid Lad A, Trocha M, Chlebda Sieragowska E. Effect of cyclophosphamide and morin-5-sulfonic acid sodium salt, alone or in combination, on ADMA/DDAH pathway in rats. Pharmacol Rep. 2013; 65: 201-207.

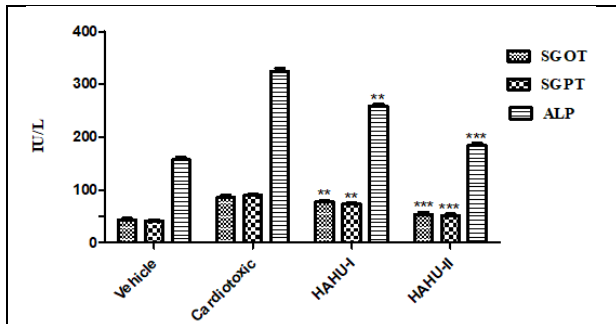


Figure 1: Effect of HAHU on plasma biochemical parameters in Cyclophosphamide induced Cardiotoxicity in rats

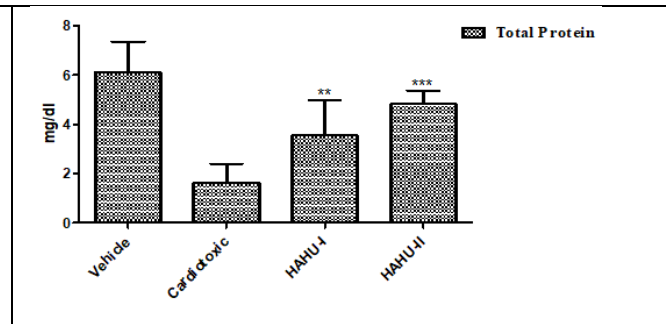


Figure 2: Effect of HAHU on plasma Total protein in Cyclophosphamide induced Cardiotoxicity in rats

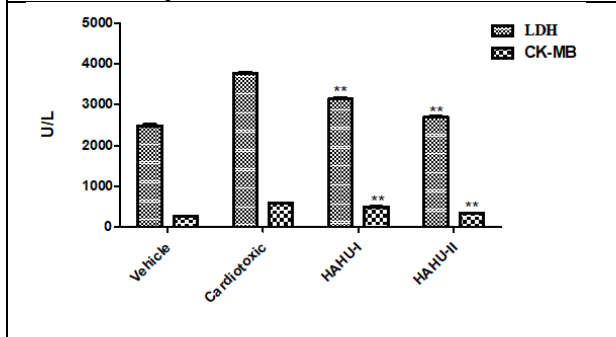


Figure 3: Effect of HAHU on LDH and CK-MB parameters in Cyclophosphamide induced Cardiotoxicity in rats

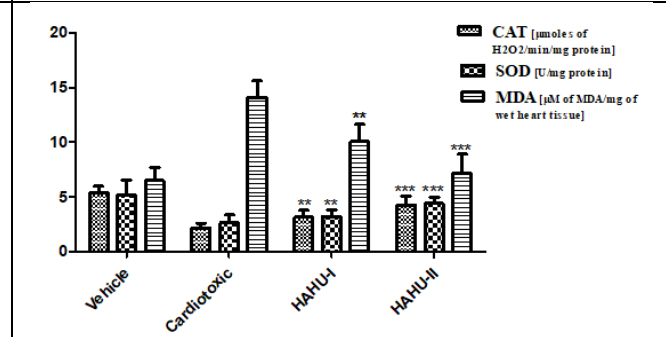
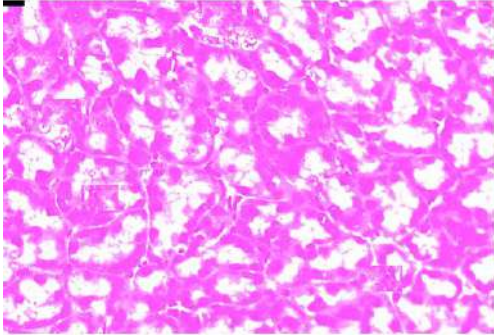
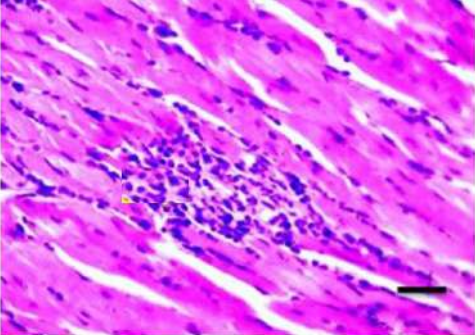
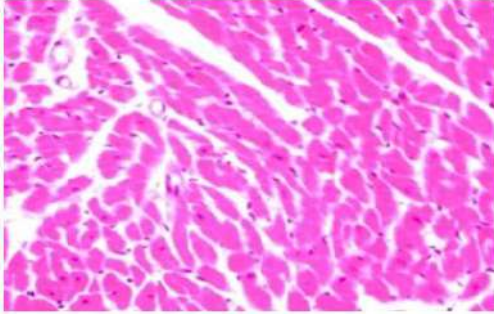
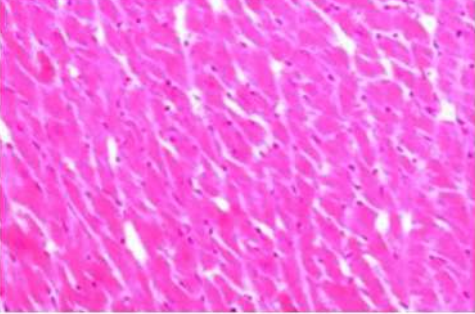


Figure 4: Effect of HAHU on Pro-oxidant and antioxidant parameters in Cyclophosphamide induced Cardiotoxicity in rats





Mallikarjuna Gandla et al.,

	
<p>Group 1: Vehicle Control rats showing Normal histoarchitecture of Heart tissue</p>	<p>Group 2: Cyclophosphamide induced Cardiotoxicity rat model (150 mg/kg b.wt.i.p.) Sections of heart ventricle from the group that received cyclophosphamide showed injured myocytes with scattered coagulative changes and thin bands of contraction necrosis.</p>
	
<p>Group 3: Cyclophosphamide + HAHU-I (Low Dose extract) Sections of heart from the low dose test showed individual cardiac muscle cells arranged in diffuse bundles in a connective tissue framework. Individual myocytes are seen in cross section to be well stained and preserved.</p>	<p>Group 4: Cyclophosphamide + HAHU-II (High Dose extract) Sections of heart from the high dose test showed centrally located nuclei with abundant cytoplasm outlined by distinct and intact cell walls. The myocytes essentially appear normal.</p>





Parkinson's Disease Detection using Machine Learning Approach

Koushik Karmakar^{1*}, Nirjhar Mallick², Arpan Maity², Souparna Paul², Rupak Hazra², Souvik Ghosh², Arpan Kumar Das²

¹Assistant Professor, Department of Computer Science and Engineering, Narula Institute of Technology, (Affiliated to Maulana Abul Kalam Azad University of Technology (MAKAUT), West Bengal, India.

²Student, Department of Computer Science and Engineering, Narula Institute of Technology, (Affiliated to Maulana Abul Kalam Azad University of Technology (MAKAUT), West Bengal, India

Received: 09 Jun 2024

Revised: 12 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

Koushik Karmakar

Assistant Professor,

Department of Computer Science and Engineering,

Narula Institute of Technology,

(Affiliated to Maulana Abul Kalam Azad University of Technology (MAKAUT),

West Bengal, India.

E.Mail: k.karmakar100@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

One of the very common neurological disorders is Parkinson's disease (PD) where movements of the muscles are restricted in the body. It has a major impact on posture, speech, and mobility; symptoms include tremors, muscle rigidity, and slower movement. The disease is caused by a decrease in dopamine levels in the brain as a result of the loss of neurons. Motor performance is hampered by low dopamine levels because they interfere with nerve cell-to-nerve cell signal transmission. Balance issues and tremors are common side effects of dopaminergic neuron degeneration, while individual differences may exist in the course and intensity of symptoms. Parkinson's disease (PD) currently has no known cure; As such early detection of this disease is very important. In this article we have tried to identify different symptoms of this disease. We have tried to collect data symbols and classified them using different machine learning algorithms.

Keywords: Parkinson's disease, artificial intelligence, diagnosis, data integration, machine learning, deep learning, voice signal analysis, Neurodegenerative disorders.

INTRODUCTION

One of the very common neurological disorders is Parkinson's disease (PD) where movements of the muscles are restricted in the body. It has a major impact on posture, speech, and mobility, which results different symptoms like the rigidity of the muscle and other problems [1]. Because of the reduction of the dopamine levels in the human brain

83342





Koushik Karmakar *et al.*,

this disease is created. Motor performance is hampered by low dopamine levels because they interfere with nerve cell-to-nerve cell signal transmission. The symptoms of dopaminergic neuron degeneration can vary in severity from person to person, and they can include tremors and balance issues. As of now, there exists no cure for Parkinson's disease. As such an early detection of the same is very much important. Sensor based remote health monitoring is a very common and an important topic for research. A lot of research work is going on this throughout the globe [8-18]. In this research we have used the concept of remote sensor based health monitoring system.

There are five different stages of Parkinson's disease (PD), and 90% of PWP have vocal cord damage, which is frequently noticeable as a symptom at stage 0. In addition to providing a quantifiable measure, vocal abnormalities are also covered by telemedicine [2] or remote medical care. Patients can record short audio clips at home using their phones. Dysphonia and dysarthria are common symptoms associated with vocal modulation [3]. Preliminary results show that the Random Forest classifier model outperforms KNN, SVM, and Logistic Regression models with an accuracy rate of more than 91% when trained on attributes taken from MDVP audio data. In section II a brief literature survey has been given, research gap has been identified in section III, problem statement has been described in section IV, experimental data collected in section V, result analysis has been shown at section VI and section VI concludes the paper.

LITERATURE SURVEY

Previous research on the use of MRI scans, genetic data, and gait analysis to predict Parkinson's disease (PD) has been conducted; however, little is known about the role that hearing impairment plays in early identification. For example, Bilal *et al.* used an SVM model to predict the onset of Parkinson's disease (PD) in older patients based on genetic data. In contrast to the current study, which describes an improved SVM model with an accuracy of 0.9183, their SVM model attained an accuracy of 0.889. These results highlight the benefits of using auditory data for PD classification rather than genetic data. Using keystroke data from the UCI telemonitoring dataset, Raundale, Thosar, and Rane [4] trained a Random Forest classifier to predict the severity of Parkinson's disease in elderly people. For PWP classification, Cordella *et al.* used audio data; MATLAB was used in their models. Our study, however, makes use of models trained in Python that are publicly available.

Most earlier studies have emphasized the use of deep learning for Parkinson's disease (PD) identification. For instance, Ali *et al.* clarified how to estimate Parkinson's disease development using ensemble deep learning models applied to phonation data. Their research, however, lacked feature selection methods to improve the performance of Deep Neural Networks. In order to identify 7 primary voice modes in PD identification, the paper uses primary Component Analysis (PCA) on 22 variables. In order to reduce the reliance of Parkinson's disease (PD) diagnosis on wearable technology, Huang *et al.* [5] trained a traditional decision tree using 12 sophisticated speech features from the MDVR-KCL dataset. Instead of concentrating on the subtleties of audio frequency, Wodzinski *et al.* trained a ResNet model on pictures of audio data. Wroge *et al.*'s [6] use of an unbiased machine learning model aimed to reduce the subjectivity of doctors in PD prognosis.

Identification of the research gap

Significant gaps in early research persist because of inadequate examination of the integration of various profiles, despite advancements in the use of AI and ML for the early identification of Parkinson's disease (PD). Earlier studies focused on individual data types. The benefits of combining motor symptoms, demographics, genetics, and neuroimaging are unexplored. This integration could enhance understanding of early disease manifestations. Many studies use controlled datasets that don't represent real-world complexity. Including data from longitudinal studies and diverse demographics is essential to improve the generalizability of AI models for early PD detection, ensuring reliability across diverse patients. Early studies overlooked individual differences. Customizing AI models for unique patient characteristics is crucial. Advances in AI promise more accurate, personalized, and timely Parkinson's diagnosis, improving outcomes.



**Koushik Karmakar et al.,****Problem Statement**

This research intends to advocate for the integration of machine learning techniques in telemedicine. By doing so, it aims to offer renewed hope and improved life quality to individuals grappling with Parkinson's disease. This research aims to develop an AI-based system for PD diagnosis, integrating multiple data sources to improve on current subjective and error-prone methods. The study explores ML in telemedicine for early PD detection, training four models on audio data from 30 patients and healthy individuals. The Random Forest model proved most effective, achieving 91.83% accuracy and 0.95 sensitivity for PD detection. In fig. 1 a workflow diagram of the system is drawn.

Experimental data collection

For our experiment necessary data has been collected in cooperation with the National Centre for Voice and Speech in Denver, Colorado, Max Little of the University of Oxford generated the dataset. In this process voice samples and other features has been extracted from different people some of whom suffer from Parkinson's disease. Number of instances collected are 195. Attributes number are 24. They are of real value type. And the target variable status has been recorded as 1 for the Parkinson's disease people and 0 for the healthy people. Data has been read from the panda data frame for easy manipulation and analysis (fig. 2 and fig. 3). Analysis is done based on different models like

Model Selection

Various machine learning algorithms are chosen to compare their performance. Machine learning models include Logistic Regression, Decision Tree, Random Forest, Support Vector Machine (SVM), K-Nearest Neighbors (KNN), Gaussian Naive Bayes, Bernoulli Naive Bayes, Voting Classifier, XGBoost.

Fig. 5: Data model

RESULT ANALYSIS

Results of different machine learning models have been implemented and the result is given below:

CONCLUSION

It has shown encouraging results to classify Parkinson's disease (PD) based on vowel phonation data. Because it took into account all 22 features in the MDVP dataset, the Random Forest classifier was able to attain above 98% accuracy in classifying Parkinson's disease using vowel phonation data. The accuracy and sensitivity of the SVM model were 0.94 and 91.836%, respectively, after PCA was administered. The K Nearest Neighbours (KNN) model benefited from its capacity to classify data without prior assumptions, showing the highest precision and recall values, both at 0.95, on a balanced dataset. Based on voice features, the Voting Classifier and Random Forest models distinguished between people with Parkinson's disease and those who were healthy with a robust accuracy of 92.3%. The models' performance was greatly enhanced by the usage of Random over Sampler to handle class imbalance and PCA for dimensionality reduction. Even with these encouraging outcomes, hyper-parameter tweaking and investigating sophisticated ensemble methods could yet be improved. Because the Random Forest model is simple, accurate, and non-invasive, we advise using it to classify illness progression. Still, auditory data alone might not be sufficient for accurate classification. Thus, through telemedicine, combining audio data with REM sleep data could improve classification accuracy.

REFERENCES

1. Prabhavathi, K., & Patil, S. (2022). Tremors and bradykinesia. Techniques for Assessment of Parkinsonism for Diagnosis and Rehabilitation, 135-149.



**Koushik Karmakar et al.,**

2. Amato, F., Rechichi, I., Borzi, L., & Olmo, G. (2022, March). Sleep Quality through Vocal Analysis: a Telemedicine Application. In 2022 IEEE International Conference on Pervasive Computing and Communications Workshops and other Affiliated Events (PerCom Workshops) (pp. 706-711). IEEE.
3. Pinto, S., Ozsancak, C., Tripoliti, E., Thobois, S., Limousin-Dowsey, P., & Auzou, P. (2004). Treatments for dysarthria in Parkinson's disease. *The Lancet Neurology*, 3(9), 547-556.
4. Raundale, P., Thosar, C., & Rane, S. (2021, May). Prediction of Parkinson's disease and severity of the disease using Machine Learning and Deep Learning algorithm. In 2021 2nd International Conference for Emerging Technology (INCET) (pp. 1-5). IEEE.
5. Huang, F., Xu, H., Shen, T., & Jin, L. (2021, October). Recognition of Parkinson's Disease Based on Residual Neural Network and Voice Diagnosis. In 2021 IEEE 5th Information Technology, Networking, Electronic and Automation Control Conference (ITNEC) (Vol. 5, pp. 381-386). IEEE.
6. Wroge, T. J., Özkanca, Y., Demiroglu, C., Si, D., Atkins, D. C., & Ghomi, R. H. (2018, December). Parkinson's disease diagnosis using machine learning and voice. In 2018 IEEE signal processing in medicine and biology symposium (SPMB) (pp. 1-7). IEEE.
7. Mei, J., Desrosiers, C., & Frasnelli, J. (2021). Machine learning for the diagnosis of Parkinson's disease: a review of literature. *Frontiers in aging neuroscience*, 13, 633752.
8. Karmakar, K., Saif, S., Biswas, S., & Neogy, S. (2022). An Intelligent Vehicular Communication-Based Framework to Provide Seamless Connectivity in WBAN. In *Advances in Communication, Devices and Networking: Proceedings of ICCDN 2021* (pp. 583-591). Singapore: Springer Nature Singapore.
9. Saif, S., Bhattacharjee, P., Karmakar, K., Saha, R., & Biswas, S. (2022). IoT-Based Secure Health Care: Challenges, Requirements and Case Study. In *Internet of Things Based Smart Healthcare: Intelligent and Secure Solutions Applying Machine Learning Techniques* (pp. 327-350). Singapore: Springer Nature Singapore.
10. Chakraborti, A., Karmakar, K., & Banerjee, A. (2022). Different Sensors in Modern day Healthcare Service. In *Computational Advancement in Communication, Circuits and Systems: Proceedings of 3rd ICCACCS 2020* (pp. 163-169). Springer Singapore.
11. Singh, A., Biswas, S., Samui, S., Mondal, A., Karmakar, K., Banerjee, J. S., & Sarigiannidis, P. (2023, July). Applications of Federated Learning in Healthcare—A New Paradigm for Digital Health. In *International Conference on Human-Centric Smart Computing* (pp. 593-605). Singapore: Springer Nature Singapore.
12. Karmakar, K., Biswas, S., & Neogy, S. (2017, March). MHRP: A novel mobility handling routing protocol in Wireless Body Area Network. In 2017 international conference on wireless communications, signal processing and networking (WiSPNET) (pp. 1939-1945). IEEE.
13. Dr. Koushik Karmakar, Arka gain, Dr. Chandrima Chakraborty (2023) Mobile Crowd Sensing and Computing (MCSC): A Paradigm Shift in Sector-based Smart Data Collection Method in National Conference on Contemporary Issues and Challenges in Management and Technology.
14. Dr. Koushik Karmakar, Sujoy Das, Dr. Chandrima Chakraborty (2023) Social IoT: A new horizon based on popular IoT systems: National conference on contemporary issues and challenges in Management & Technology.
15. Saif, S., Karmakar, K., Biswas, S., & Neogy, S. (2022). MLIDS: Machine learning enabled intrusion detection system for health monitoring framework using BA-WSN. *International journal of wireless information networks*, 29(4), 491-502.
16. Karmakar, K., Saif, S., Biswas, S., & Neogy, S. (2022). Application of Relay Nodes in WBAN-based smart Healthcare for energy conservation explained with case studies. In *High Performance Computing and Networking: Select Proceedings of CHSN 2021* (pp. 639-647). Singapore: Springer Singapore.
17. Soumyajit Banerjee Koushik Karmakar, Ananya Banerjee (2017) A survey of routing protocols in Body Area Network in National conference on Advance Computing, Communication and Device (NCACCD'17).
18. Karmakar, K., Saif, S., Das, P., Neogy, S., & Biswas, S. (2022). FDRF: fault detection and recovery framework for seamless data transmission in remote health monitoring using WBAN. *Wireless Personal Communications*, 123(1), 325-356.





Koushik Karmakar et al.,

Table 1. Results of different machine learning models have been implemented and the result is given below:

Logistic Regression	Used Logistic Regression with parameters C=0.4 and max_iter=1000.
Decision Tree Classifier	Used Decision Tree Classifier with a random state for reproducibility.
Random Forest Classifier	Implemented two versions with different criteria: gini and entropy.
Support Vector Classifier (SVC)	Implemented using SVC with default parameters.
K-Nearest Neighbors (KNN)	Implemented using KNeighbors Classifier with default parameters.
Naive Bayes Classifiers	Implemented Gaussian NB and Bernoulli NB.
Voting Classifier	Combined all above models using Voting Classifier with hard voting.
XGBoost Classifier	Implemented using XGB Classifier with default parameters. Evaluated using accuracy score.

Table 2: Different models and their values.

Logistic Regression	0.779661
Decision Tree	0.98351
Random Forest (Gini)	1.000000
Random Forest (Entropy)	1.000000
SVC	0.949153
KNN	0.949153
GaussianNB	0.762721
BernoulliNB	0.745763
Voting Classifier	0.966102

Table 3. Evaluated the performance using confusion matrix and classification report for models with high accuracy which is described below.

Training Confusion Matrix: $\begin{bmatrix} 81 & 0 \\ 0 & 75 \end{bmatrix}$	Testing Confusion Matrix: $\begin{bmatrix} 30 & 2 \\ 2 & 5 \end{bmatrix}$
Training Classification Report: precision recall f1-score support 0 1.00 1.00 1.00 81 1 1.00 1.00 1.00 75 accuracy 1.00 156 macro avg 1.00 1.00 1.00 156 weighted avg 1.00 1.00 1.00 156	Testing Classification Report: precision recall f1-score support 0 0.94 0.94 0.94 32 1 0.71 0.71 0.71 7 accuracy 0.90 39 macro avg 0.82 0.82 0.82 39 weighted avg 0.90 0.90 0.90 39





Koushik Karmakar et al.,

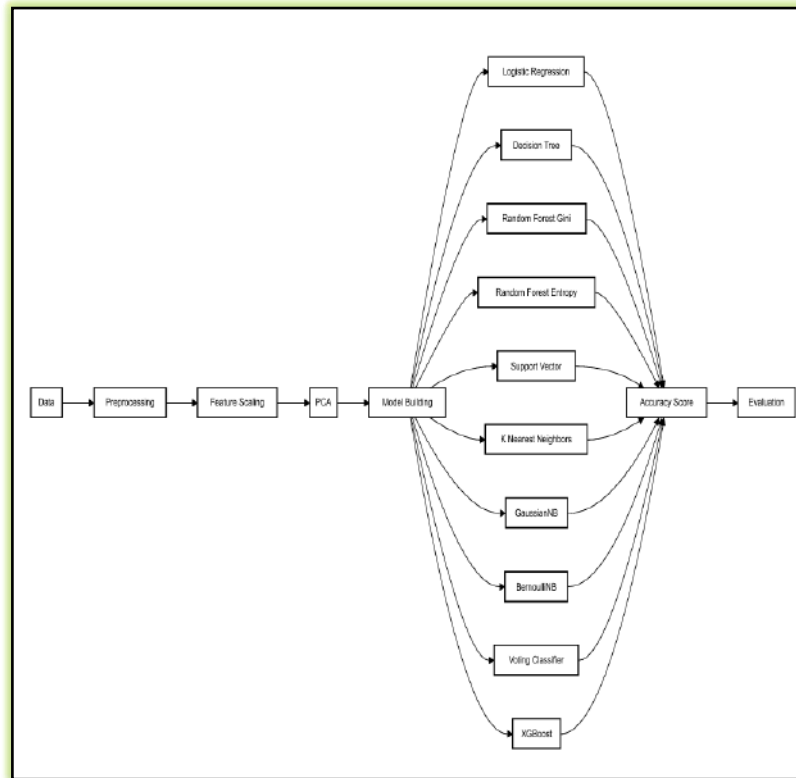


Fig.1: Workflow Diagram

	name	MDVP:Fo(Hz)	MDVP:Fhi(Hz)	MDVP:Flo(Hz)	MDVP:Jitter(%)	MDVP:Jitter(Abs)	MDVP:RAP	MDVP:PPQ	Jitter:DDP	MDVP:Shimmer
0	phon_R01_S01_1	119.992	157.302	74.997	0.00784	0.000070	0.00370	0.00554	0.01109	0.04374
1	phon_R01_S01_2	122.400	148.650	113.819	0.00968	0.000080	0.00465	0.00696	0.01394	0.06134
2	phon_R01_S01_3	116.682	131.111	111.555	0.01050	0.000090	0.00544	0.00781	0.01633	0.05233
3	phon_R01_S01_4	116.676	137.871	111.366	0.00997	0.000090	0.00502	0.00698	0.01505	0.05492
4	phon_R01_S01_5	116.014	141.781	110.655	0.01284	0.000110	0.00655	0.00908	0.01966	0.06425
5	phon_R01_S01_6	120.552	131.162	113.787	0.00968	0.000080	0.00463	0.00750	0.01388	0.04701
6	phon_R01_S02_1	120.267	137.244	114.820	0.00333	0.000030	0.00155	0.00202	0.00466	0.01608
7	phon_R01_S02_2	107.332	113.840	104.315	0.00290	0.000030	0.00144	0.00182	0.00431	0.01567
8	phon_R01_S02_3	95.730	132.068	91.754	0.00551	0.000060	0.00293	0.00332	0.00880	0.02093
9	phon_R01_S02_4	95.056	120.103	91.226	0.00532	0.000060	0.00268	0.00332	0.00803	0.02838
10	phon_R01_S02_5	88.333	112.240	84.072	0.00505	0.000060	0.00254	0.00330	0.00763	0.02143
11	phon_R01_S02_6	91.904	115.871	86.292	0.00540	0.000060	0.00281	0.00336	0.00844	0.02752
12	phon_R01_S04_1	136.926	159.866	131.276	0.00293	0.000020	0.00118	0.00153	0.00355	0.01259
13	phon_R01_S04_2	139.173	179.139	76.556	0.00390	0.000030	0.00165	0.00208	0.00496	0.01642
14	phon_R01_S04_3	152.845	163.305	75.836	0.00294	0.000020	0.00121	0.00149	0.00364	0.01828

0s completed at 3:38 AM

Fig. 2: Dataset for analysis





Koushik Karmakar et al.,

	MDVP:Fo(Hz)	MDVP:Fhi(Hz)	MDVP:Flo(Hz)	MDVP:Jitter(%)	MDVP:Jitter(Abs)	MDVP:RAP	MDVP:PPQ	Jitter:DDP	MDVP:Shimmer	MDVP:Shimmer(d)
count	195.000000	195.000000	195.000000	195.000000	195.000000	195.000000	195.000000	195.000000	195.000000	195.000000
mean	154.228641	197.104918	116.324631	0.006220	0.000044	0.003306	0.003446	0.009920	0.029709	0.2822
std	41.390065	91.491548	43.521413	0.004848	0.000035	0.002968	0.002759	0.008903	0.018857	0.1948
min	88.333000	102.145000	65.476000	0.001680	0.000007	0.000680	0.000920	0.002040	0.009540	0.0850
25%	117.572000	134.862500	84.291000	0.003460	0.000020	0.001660	0.001860	0.004985	0.016505	0.1485
50%	148.790000	175.829000	104.315000	0.004940	0.000030	0.002500	0.002690	0.007490	0.022970	0.2210
75%	182.769000	224.205500	140.018500	0.007365	0.000060	0.003835	0.003955	0.011505	0.037885	0.3500
max	260.105000	592.030000	239.170000	0.033160	0.000260	0.021440	0.019580	0.064330	0.119080	1.3020

Fig. 3: Dataset 2 for analysis

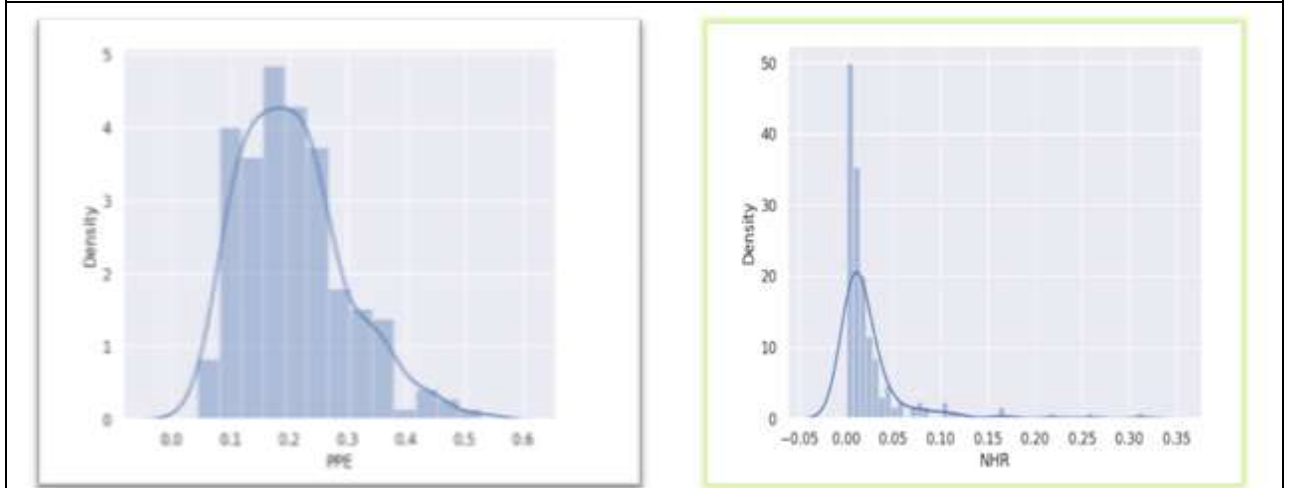


Fig. 4: Distribution of each feature using Seaborn's distplot.





Koushik Karmakar et al.,

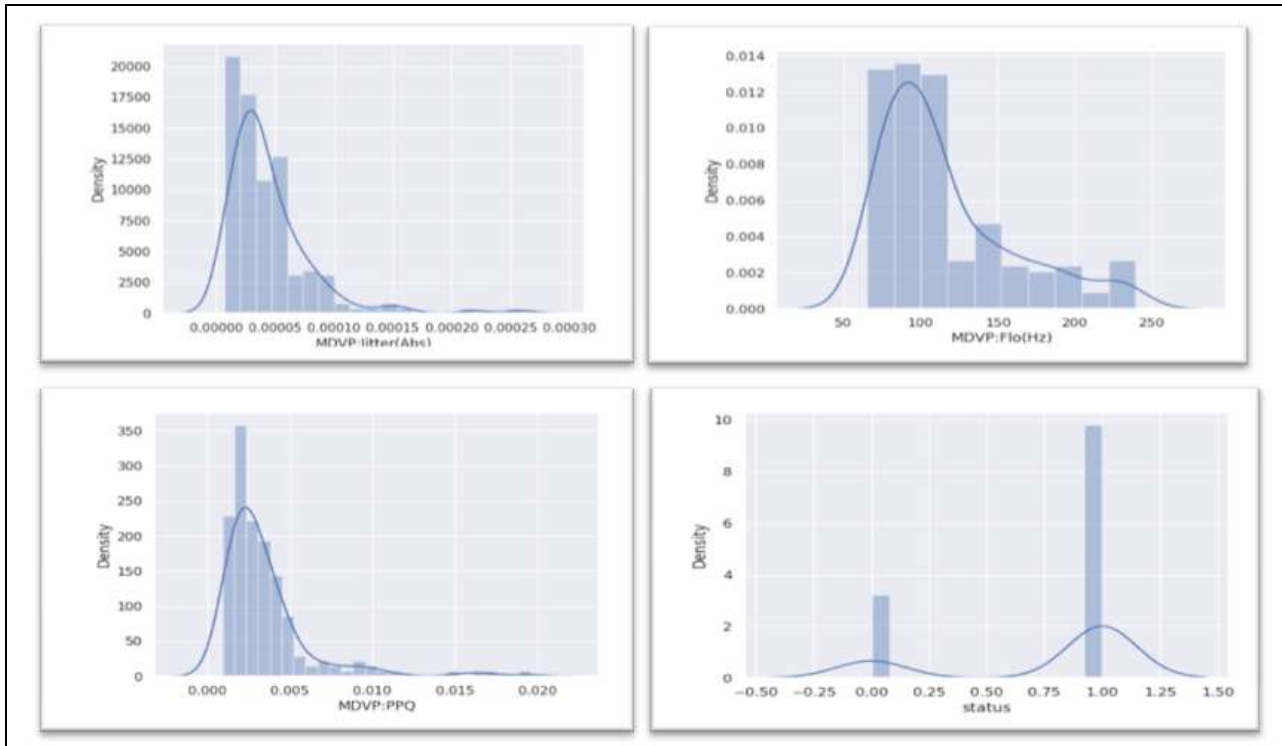


Fig. 5: Data model



Fig.6. Different models and their values





Effect of Inadequate Sleep on Memory and Learning: a study on Adolescents

Bhoomika C A¹, Marjana¹, Patteshwari Duraiswamy² and Sangeetha S R^{2*}

¹Student, Division of Cognitive Neuroscience and Psychology, JSS Academy of Higher Education & Research, Mysuru, Karnataka, India

²Assistant Professor, Division of Cognitive Neuroscience and Psychology, JSS Academy of Higher Education & Research, Mysuru, Karnataka, India.

Received: 11 Jun 2024

Revised: 17 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Sangeetha S R

Assistant Professor,

Division of Cognitive Neuroscience and Psychology,

JSS Academy of Higher Education & Research,

Mysuru, Karnataka, India.

E. Mail: sangeetha080511@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The role of sleep is very important for several important cognitive processes. In developing adolescents, getting adequate sleep is even more important as it affects the development of intellectual ability, learning and memory, reward and motivation, emotional regulation, etc. Adequate sleep for adolescents varies from 7-10 hours. But a majority of adolescents get less than 7 hours of sleep daily. This review discusses the effect of this inadequate sleep on memory and consequently its effect on learning and academics. The results indicate that due to a variety of reasons, adolescents ended up getting as little as 5-6 hours of sleep on weeknights. Thus their short-term memory was affected negatively and they did not learn as well as their peers who slept well.

Keywords: Inadequate sleep, memory, learning, adolescents

INTRODUCTION

Sleep can be defined as a state of reduced mental and physical activity that typically recurs for several hours every night where the consciousness of the surroundings is practically suspended and sensory activity is inhibited partially. (Chokroverty S, *et al*, 2010). A lot of studies have linked the role of sleep to cognitive processes like alertness, perception, emotion, learning and memory, and executive functioning. (Killgore, *et al*, 2014). Now the three factors that affect cognitive performance and higher-order functions are inertia, homeostasis, and circadian rhythms. (Kapsi, S., *et al*, 2020). Burke *et al*. claim that homeostasis and circadian cycles are the two factors controlling working memory, mood, alertness, motivation, and visual attention maintenance. (Burke, T. M., 2015) This means that getting

83350





Marjana. Bhoomika et al.,

less sleep may result in a decrease in these cognitive functions. Studies also suggest that, with inadequate sleep, behavior becomes increasingly irregular and unstable, attentional lapses become longer and more frequent, (Lim J, 2010) and simple reaction time is slowed (Lim J, 2008). Since memory and learning are some of the most important cognitive skills required for students, this review paper mainly deals with establishing the negative effects of inadequate sleep on them. The sample population taken into account here is adolescents. According to the definition given by the World Health Organisation, children from 10-19 years are considered adolescents. A Joint Consensus Statement of the American Academy of Sleep Medicine and Sleep Research Society states that adolescents require about 7-10 hours of sleep (Consensus Conference Panel, 2015) (Chaput, J. P. 2018).

Sleep Patterns in Adolescents

Sleep patterns refer to the regular and recurring cycles of sleep stages that individuals go through during a typical night of sleep. These patterns are characterized by different phases, including both rapid eye movement (REM) sleep and non-rapid eye movement (NREM) sleep. Now, sleep patterns in adolescents are mostly predictable. Their nocturnal sleep schedules decrease and bed-times are delayed. (Garipey, G., et al 2020). But school/college timings do not change and thus adolescents get less sleep than recommended. Hysing M, et al, report from their study conducted in Hordaland County in Norway, consisting of 10,220 adolescents aged 16–18 years (54% girls), that there was a sleep deficit of about 2 hours on weekdays and latency of sleep onset exceeding 30 min on 65% of those studied. Insomnia rates were higher in girls whereas boys reported later bedtimes. (Hysing M, et al, 2013). Bed times tend to delay with increasing age according to meta-analysis of worldwide adolescent sleep patterns in the last decade (1999–2010). Asian adolescents slept even less than peers from North America and Europe. This resulted in less total sleep duration on school nights and a tendency for greater degrees of daytime drowsiness. (Gradisar, M, 2011). Another study corroborating this was conducted in Delhi, India among 501 students (aged 11–15 years). This study provides statistical proof of sleep deprivation increasing with age. Sleep deprivation increased 83.7% to 87.1% in 11–12 y to 90.5% to 92.5% in 13–15 y. (Singh, R., 2018). Only 33% college students in America reported sleeping a full 8 hours/night. Sleep interruptions were caused by social media usage, gaming, and texting. (Whipps, J, 2018). Students aged 17-24 at a large private university in the Mideast reported chronically restricted sleep. 25% of students got less than 6.5 hours/night and only 29.4% of students reported getting 8 hours of sleep which is the typical hours required for adolescents. (Lund, H. G., et al, 2010). According to the Youth Risk Behavior Survey, 72.7% of students said they slept for less than eight hours on school nights on average. (A. G. Wheaton et al., 2018). This is comparable to the National Sleep Foundation poll, which found that 62% of students slept for less than eight hours on weeknights. With 75% of seniors reporting less than 8 hours of sleep every night, they were the group most sleep deprived. (Carskadon, M. A., et al 2015) Girls were the ones disproportionately affected. (Eaton, D. K., et al 2010). We can thus infer that most adolescents worldwide sleep less than required. Older adolescents also slept less than their younger counterparts. (Garipey, G., et al 2020).

Factors Affecting Sleep In Adolescents

Adolescents are notorious for getting inadequate sleep. There may be some reasons for that. It could be academic pressure, developmental changes during puberty, social media usage, gaming, peer pressure, parental pressure, increased usage of caffeine or other energy beverages, decreased curfew and increased bedtimes, and certain mental problems. Puberty is the time when humans undergo a lot of changes. One of them is the shift of the timing of circadian rhythm. Before puberty, the body shows signs of sleep around 8:00 or 9:00 pm. This cycle changes when puberty sets in and the body only feels sleepy at around 10:00 or 11:00p.m. (Burke, T. M., et al 2015) "Sleep phase delay" refers to the normal circadian rhythm shift experienced by teenagers. The need to sleep is delayed by around two hours. Therefore adolescents struggle to go to sleep at their regular hour. But they still require nine hours of sleep every night on average which is impossible with early school/college start times. (Garipey, G., et al 2020) This is where adolescents struggle with getting adequate sleep. Lohsoonthorn, V et al report another contributing factor. 48.1 % of Thai college students reported poor sleep quality. 58% of the same subjects were using some sort of stimulant beverage. Hence stimulant use, alcohol consumption and cigarette smoking were positively associated with poor sleep quality and increased daytime sleepiness. (Lohsoonthorn, V et al 2013). Sanchez, S. E., et al conducted a similar study among 2,458 Peruvian college students. Those who reported consuming more than 3 stimulant beverages per



**Marjana. Bhoomika et al.,**

week had higher odds of poor sleep quality, short sleep duration and increased use of sleep medication. (Sanchez, S. E., et al 2013) Whereas another study comprising 11025 students aged 17 to 24 years from an urban Midwestern university found that alcohol and caffeine consumption, exercise, and consistency of sleep schedule were not significant contributors to sleep quality in the 60% of students who were reported as poor sleepers. Instead, emotional and academic stress was the cause affecting sleep. (Lund, H. G., et al 2010) When a total of 937 ninth-grade adolescents (15–16 years), from western Sweden participated in a study, 55% of the adolescents were found to sleep less than the recommended 8 hours per night due to school stress and technology use. 11% also had sleeping difficulties. (Jakobsson, M., et al, 2019) Students experience high stress during the school year. The majority of them also report getting only 6 hours of sleep during weekdays. This leads to them experiencing moderate fatigue and more depressive symptoms. High-stress levels are associated with sleep disturbances, less nocturnal total sleep time, and higher fatigue severity. (Lee, S. Y., et al, 2013) It was found that sleep was more fragmented during high-stress times in an experiment comparing the quality and duration of sleep in adolescents during low and high-stress times. (Dewald, J. F., et al, 2014) Whipps, J., et al reported in a study evaluating night time media use and sleep patterns in college students, that students had a mean sleep duration of only 7.26 ± 0.93 hours. Reports of texting after bed and device-related sleep interruptions were many. Social media usage, mobile gaming, and texting were correlated with sleep interruptions. (Whipps, J., et al, 2018) Young adults (aged 19-32) in the US reported an average of 61 min of social media usage per day. More than half of the same group reported medium or high levels of sleep disturbance. The results did not vary even when social media usage volume or frequency was taken as the variable. (Levenson, J. C., et al 2016). Wong, H. Y., et al assessed Hong Kong university male students in 2019 on the relationship between the severity of internet gaming disorder and poor sleep quality and found a direct relationship between both. (Wong, H. Y., et al 2020). Another similar study conducted among 524 adolescents in Lebanon found that those with Internet Gaming Disorder slept only 5 hours per night as opposed to those who casually played online games and slept for 7 hours. (Hawi, N. S., et al, 2018) To find if there was a relationship between obesity and sleep, 515 college students were assessed. It was found that one-third of the participants had with BMI of more than 25 and 51% of those had poor-quality sleep. Sleep disturbances were thus a result of obesity and sleep duration was not affected. (Vargas, P. A., et al 2014) When a study on the impact of sleep disorders was conducted in the US for 6139 individuals over the age of 16, it was found that 4.2% had sleep apnea, followed by 1.2% having insomnia and RLS (0.4%). Common sleep habits were snoring (48%), feeling unrested during the day (26.5%), and not getting enough sleep (26%). It was also found that insomnia, sleep apnea, and RLS had the highest impact on concentration and memory. (Ram, S., et al, 2010) Dyssomnias, especially early and middle insomnias (odds ratio = 1.74 and 2.24), disturbed the circadian rhythm by prolonging bedtimes in a study conducted with 1253 adolescents in grades 3, 5, and 8. As a consequence duration of sleep also decreased. (Chen, Y. L., 2016). Another factor responsible for inadequate sleep in adolescents was outlined by Crowley SJ et al, in 2010. According to them, during the second decade of life sleep/wake timing shifts to later, and Delayed Sleep Phase Syndrome (DSPS), may be an extreme manifestation of this. (Crowley SJ et al, 2010) Thus some factors affecting sleep in adolescents and causing their sleep durations to decrease were academic and emotional stress, consumption of a lot of caffeinated and energy drinks, usage of social media, severe internet gaming disorders, dyssomnias, and delaying of sleep/wake timing. A factor that did not contribute to disturbing sleep duration was obesity.

Link between Sleep and Memory

Memory is the capacity to store and retrieve information. (Zlotnik, G., & Vansintjan, A. 2019) Squire, L. R defines memory as 'the faculty of encoding, storing, and retrieving information'. Psychologists have found that memory includes four important categories: working, sensory, short-term, and long-term. According to the definition of the American psychological association, sensory memory holds sensory information for very less time, usually 1 second or less. (VandenBos, G. R. 2007). Short-term memory allows a person to recall limited information for a short period of time. (Casella, M., & Al Khalili, Y. 2019). Working memory is similar to short-term memory but here the person manipulates information. Long-term memory stores a wide array of memories and experiences. The majority of the memories that people can recall, especially those that are more than thirty seconds old are part of long-term memory. Long-term memories are divided into two subcategories: implicit and explicit. Explicit memories are conscious experiences of events, personal anecdotes, or lessons learnt. Some types of explicit long-term memory include





Marjana. Bhoomika et al.,

episodic memory and semantic memory. Implicit memories are memories that influence a person's behaviour unconsciously. Some types of this memory include procedural memory and priming/conditioning. (Cowan, N. 2008). Zhang's memory model explains the importance of sleep in memory. This model has 2 stages: waking and sleeping brain. During waking time, the memories created in our working memory during the waking state are stored in temporary memory. Because there is a limited amount of temporary memory, it has to be periodically cleansed to avoid overload. Thus during sleeping time, the temporary data is supposed to be compared with old data saved in the long-term memory, in order to identify and delete unwanted, duplicate and overlapping data. The remaining information is then encoded and transferred to long-term memory. When the working memory is overloaded with incoming data, our waking brain is unable to complete this task. The temporary memory must be isolated from the outside world in order to carry out this housekeeping and guarantee that the memory transfer procedure continues unhindered. This isolation is carried out while sleeping. Sleep has the purpose of processing, encoding, and transferring information from the temporary memory to the long-term memory according to this memory model. (Zhang, J. 2004) This model is the updated version of the memory consolidation model proposed by Squire, L. R., & Alvarez, P in 1995.

Experimentally, the role of sleep in explicit memory was proved by measuring the memory retention of 29 adolescents (aged 15–18 years) who were given only 5 hours of sleep opportunity each night. Retention was tested 30 minutes, 3 days and 42 days (only a subset with n=14 participated) after learning and results were compared to a control group (n = 30) that slept for nine hours every night during the trial. 26% forgot at the 30-minute test, 34% at the Day 3 test, and 65% at the Day 42 test. (Cousins, J. N., et al 2019). Sleep facilitates the consolidation of declarative, procedural, and emotional memories. (Diekelmann, S., & Born, J. 2010) Emotional memory was found to be consolidated more during REM sleep participants reporting depressive symptoms. Additionally, those depressive participants who were sleep deprived consolidated significantly less emotional (negative here) information. (Harrington, M. O., et al 2018). Sleep helps in working memory as well as memory consolidation in children and adolescents. It was further found that sleep deprivation negatively affected complex tasks involving higher brain functions more strongly than performance in simple memory tasks. (Kopasz, M., et al 2010) Research reveals a decrease in working memory, sustained attention, and executive functions in sleep restricted (SR) groups, while the control group maintains baseline levels. It was found that the SR group continued to perform worse than the control even after 2 recovery sleep episodes. (Lo, J. C., 2016). A review study conducted by Wilhelm L., et al supports this and further states that slow-wave sleep plays a major role in the consolidation of procedural memory. (Wilhelm, I., 2012) Further evidence comes from a study by Fenn, K. M., &Hambrick, D. Z. When memory was tested after a delay of roughly 12 hours that included sleep or waking, it was found that those who slept remembered more and forgot less than those who were awake. They hypothesize that sleep enhances the retrieval of information and also protects against loss of memory that occurs when awake. (Fenn, K. M., &Hambrick, D. Z.2013) A study conducted by Ashton, J. E., et al on sleep deprivation found that rates of forgetting in episodic memory increase with sleep deprivation. Overnight sleep deprivation was also found to lead to further deficits in associative memory that were not observed after daytime wakefulness. (Ashton, J. E., et al, 2020) Partial sleep restriction had small or no effects on adolescent cognitive functioning. However, where sleep deprivation was observed, there was a decrease in psychomotor tasks. (Whitney P., et al, 2010). It was also found that sleep extension and sleep improvement caused improvement in working memory. When subjects slept directly after learning, they also improved memory consolidation. (De Bruin, E. J., et al 2011) Looking at the evidence provided by the studies stated above and a lot of other growing evidence, we can safely conclude that sleep is important for all types of memory consolidation.

Relation between Sleep, Memory and Learning

There are 2 types of memory. They are short and long term memories. The primary function of short term memory/working memory is to support learning. (Gathercole, S. E. 2004, January). Therefore for adolescents whose main duty is to learn, usage of this cognitive skill is very important. The type of memory most important while learning is working memory as it refers to a brain system that allows temporary storage and manipulation of the information necessary for complex cognitive. (Baddeley, A. 1992). After conducting a thorough meta-analysis of the literature on these characteristics in kids, Astill et al. found that sleep duration was positively and significantly





Marjana. Bhoomika et al.,

correlated with executive functioning, academic achievement, and multiple-domain cognitive functioning. (Astill *et al.*, 2012) It was also found that children who slept earlier did better on cognitive tests. (Fonseca, A. G., 2020). Research indicates that while we sleep, the brain performs neurological processes related to learning and memory, processing information that is gained while we are awake (Watson and Buzsáki, 2015). Sleep has been shown to improve performance on several learning and memory tasks, such as the face sequence recognition task, the motor adaptation task, the word-pair association task, the visual texture discrimination task, and the motor sequence task. (Uji, M., & Tamaki, M. 2023) When 7798 adolescents aged 16–19 years were surveyed to link sleep and academic performance in Norway, it was found that students with sleep deficits and short sleep duration had the highest odds of poor GPA. (Hysing, M., *et al* 2016) In tests where researchers were able to deliberately alter sleep to track behavioral and neurocognitive outcomes; including learning, memory, and academic achievement, it was primarily found that both the quantity and quality of sleep have a direct impact on students' academic performance and learning capacity. Sleep loss was also frequently linked to students' poor declarative and procedural learning. Further, there was a decline in neurocognitive and academic performance. (Curcio, G., *et al*, 2016) All things considered, behavioural research on adolescents suggests that sleep is essential for memory consolidation. All stages of sleep have been linked to one or more components of this consolidation up until this point of sleep. (Walker, M. P., & Stickgold, R. 2004). However, a comprehensive knowledge of the functions of each stage of sleep is still a crucial future objective. It was found that students who performed the best academically had far earlier waking times ($p = 0.008$) and bedtimes ($p = 0.05$) than those who performed the worst. High achievers tended to nap more frequently ($p = 0.07$). (Eliasson, A.H., 2010) Even when alertness and vigilance are restored with stimulant countermeasures, some aspects of higher-level cognitive capacities are still impaired by sleep deprivation, indicating that sleep loss may have effects on particular cognitive systems in addition to those caused by attentional processing impairments or general cognitive declines. (Killgore, W. D. 2010) This was further proved in a study conducted among 1,845 college students at a large, southeastern public university, where it was found that students reported insufficient sleep and those same students were also at the risk of academic jeopardy with GPAs of less than 2. (Gaultney, J. F. 2010).

DISCUSSION AND CONCLUSION

A lot of significant research shows that a majority of adolescents are getting inadequate sleep mostly on weekdays (Hysing M, *et al*, 2013) and it is risking their memory and learning processes thus affecting their academic performance. It is thus imperative that measures be taken to make sure that adolescents get enough sleep. Some solutions for the above may be later school/college timings (de Araújo, L. B. G., *et al* 2020), earlier curfews and bedtimes (Campbell, I. G., *et al* 2023), implementation of naps in between learning (Inazumi, C. K., *et al* 2020), avoidance of social media (Levenson, J. C., *et al* 2017) and early diagnosis and cure for sleep disorders. (Modlin, T. 2002). It is thus established that there is a significant link between inadequate sleep, memory, and learning in adolescents. Inadequate sleep decreases both memorizing ability and thus learning ability is also decreased. It is also linked to low performance in academics.

REFERENCES

1. Chokroverty, S.. Overview of sleep & sleep disorders. Indian Journal of Medical Research 131(2):p 126-140, February 2010.
2. Killgore, W.D.S., Weber, M. (2014). Sleep Deprivation and Cognitive Performance. In: Bianchi, M. (eds) Sleep Deprivation and Disease. Springer, New York, NY. https://doi.org/10.1007/978-1-4614-9087-6_16
3. Kapsi, S., Katsantoni, S., & Drigas, A. (2020). The Role of Sleep and Impact on Brain and Learning. Int. J. Recent Contributions Eng. Sci. IT, 8(3), 59-68.
4. Burke, T. M., Scheer, F. A. J. L., Ronda, J. M., Czeisler, C. A., & Wright, K. P. (2015). Sleep
5. inertia, sleep homeostatic and circadian influences on higher-order cognitive functions. Jour-
6. nal of Sleep Research, 24(4), 364–371





Marjana. Bhoomika et al.,

7. Lim, J., & Dinges, D. F. (2010). A meta-analysis of the impact of short-term sleep deprivation on cognitive variables. *Psychological bulletin*, 136(3), 375.
8. Lim, J., & Dinges, D. F. (2008). Sleep deprivation and vigilant attention. *Annals of the New York Academy of Sciences*, 1129(1), 305-322.
9. Chaput, J. P., Dutil, C., & Sampasa-Kanyinga, H. (2018). Sleeping hours: what is the ideal number and how does age impact this?. *Nature and science of sleep*, 10, 421–430. <https://doi.org/10.2147/NSS.S163071>
10. Consensus Conference Panel, Watson, N. F., Badr, M. S., Belenky, G., Bliwise, D. L., Buxton, O. M., Buysse, D., Dinges, D. F., Gangwisch, J., Grandner, M. A., Kushida, C., Malhotra, R. K., Martin, J. L., Patel, S. R., Quan, S. F., & Tasali, E. (2015). Joint Consensus Statement of the American Academy of Sleep Medicine and Sleep Research Society on the Recommended Amount of Sleep for a Healthy Adult: Methodology and Discussion. *Sleep*, 38(8), 1161–1183. <https://doi.org/10.5665/sleep.4886>
11. Garipey, G., Danna, S., Gobiņa, I., Rasmussen, M., de Matos, M. G., Tynjälä, J., ...& Schnohr, C. (2020). How are adolescents sleeping? Adolescent sleep patterns and sociodemographic differences in 24 European and North American countries. *Journal of adolescent Health*, 66(6), S81-S88.
12. Hysing, M., Pallesen, S., Stormark, K. M., Lundervold, A. J., & Sivertsen, B. (2013). Sleep patterns and insomnia among adolescents: a population-based study. *Journal of sleep research*, 22(5), 549-556.
13. Gradisar, M., Gardner, G., & Dohnt, H. (2011). Recent worldwide sleep patterns and problems during adolescence: a review and meta-analysis of age, region, and sleep. *Sleep medicine*, 12(2), 110-118.
14. Singh, R., Suri, J.C., Sharma, R. *et al.* Sleep Pattern of Adolescents in a School in Delhi, India: Impact on their Mood and Academic Performance. *Indian J Pediatr* 85, 841–848 (2018). <https://doi.org/10.1007/s12098-018-2647-7>
15. Whipps, J., Byra, M., Gerow, K. G., & Guseman, E. H. (2018). Evaluation of nighttime media use and sleep patterns in first-semester college students. *American journal of health behavior*, 42(3), 47-55.
16. Lund, H. G., Reider, B. D., Whiting, A. B., & Prichard, J. R. (2010). Sleep patterns and predictors of disturbed sleep in a large population of college students. *Journal of adolescent health*, 46(2), 124-132.
17. Wheaton, A. G., Jones, S. E., Cooper, A. C., & Croft, J. B. (2018). Short sleep duration among middle school and high school students—United States, 2015. *Morbidity and Mortality Weekly Report*, 67(3), 85.
18. Carskadon, M. A., Mindell, J. A., & Drake, C. (2015). National Sleep Foundation. 2006 Sleep in America Poll: summary of findings. Washington, DC: National Sleep Foundation, 2006. *sleep in America Poll –teens and sleep.* Sleep Heal. Arlington, VA.
19. Eaton, D. K., McKnight-Eily, L. R., Lowry, R., Perry, G. S., Presley-Cantrell, L., & Croft, J. B. (2010). Prevalence of insufficient, borderline, and optimal hours of sleep among high school students—United States, 2007. *Journal of Adolescent Health*, 46(4), 399-401.
20. Bruce, E. S., Lunt, L., & McDonagh, J. E. (2017). Sleep in adolescents and young adults. *Clinical medicine (London, England)*, 17(5), 424–428. <https://doi.org/10.7861/clinmedicine.17-5-424>
21. Lohsoonthorn, V., Khidir, H., Casillas, G., Lertmaharit, S., Tadesse, M. G., Pensuksan, W. C., ...& Williams, M. A. (2013). Sleep quality and sleep patterns in relation to consumption of energy drinks, caffeinated beverages, and other stimulants among Thai college students. *Sleep and Breathing*, 17, 1017-1028.
22. Sanchez, S. E., Martinez, C., Oriol, R. A., Yanez, D., Castañeda, B., Sanchez, E., ...& Williams, M. A. (2013). Sleep quality, sleep patterns and consumption of energy drinks and other caffeinated beverages among Peruvian college students. *Health*, 5(8B), 26
23. Jakobsson, M., Josefsson, K., Jutengren, G., Sandsjö, L., & Högberg, K. (2019). Sleep duration and sleeping difficulties among adolescents: exploring associations with school stress, self-perception and technology use. *Scandinavian journal of caring sciences*, 33(1), 197-20
24. Lee, S. Y., Wuertz, C., Rogers, R., & Chen, Y. P. (2013). Stress and sleep disturbances in female college students. *American journal of health behavior*, 37(6), 851-858.
25. Dewald, J. F., Meijer, A. M., Oort, F. J., Kerkhof, G. A., & Bögels, S. M. (2014). Adolescents' sleep in low-stress and high-stress (exam) times: a prospective quasi-experiment. *Behavioral Sleep Medicine*, 12(6), 493-506.
26. Levenson, J. C., Shensa, A., Sidani, J. E., Colditz, J. B., & Primack, B. A. (2016). The association between social media use and sleep disturbance among young adults. *Preventive medicine*, 85, 36-41.





Marjana. Bhoomika et al.,

27. Wong, H. Y., Mo, H. Y., Potenza, M. N., Chan, M. N. M., Lau, W. M., Chui, T. K., ... & Lin, C. Y. (2020). Relationships between severity of internet gaming disorder, severity of problematic social media use, sleep quality and psychological distress. *International journal of environmental research and public health*, 17(6), 1879.
28. Hawi, N. S., Samaha, M., & Griffiths, M. D. (2018). Internet gaming disorder in Lebanon: Relationships with age, sleep habits, and academic achievement. *Journal of behavioral addictions*, 7(1), 70-78.
29. Vargas, P. A., Flores, M., & Robles, E. (2014). Sleep quality and body mass index in college students: the role of sleep disturbances. *Journal of American college health*, 62(8), 534-541.
30. Ram, S., Seirawan, H., Kumar, S. K., & Clark, G. T. (2010). Prevalence and impact of sleep disorders and sleep habits in the United States. *Sleep and breathing*, 14, 63-70.
31. Chen, Y. L., & Gau, S. S. F. (2016). Sleep problems and internet addiction among children and adolescents: a longitudinal study. *Journal of sleep research*, 25(4), 458-465.
32. Crowley SJ, Acebo C, Carskadon MA. (2010). Sleep, circadian rhythms, and delayed phase in adolescence. *Sleep Med.* 2007 Sep;8(6):602-12. doi: 10.1016/j.sleep.2006.12.002. Epub 2007 Mar 26. PMID: 17383934.
33. Squire, L. R. (2009). Memory and brain systems: 1969–2009. *Journal of Neuroscience*, 29(41), 12711-12716.
34. Zlotnik, G., & Vansintjan, A. (2019). Memory: An extended definition. *Frontiers in psychology*, 10, 2523.
35. VandenBos, G. R. (2007). *APA dictionary of psychology*. American Psychological Association.
36. Cascella, M., & Al Khalili, Y. (2019). Short term memory impairment.
37. Cowan, N. (2008). What are the differences between long-term, short-term, and working memory?. *Progress in brain research*, 169, 323-338.
38. Zhang, J. (2004). Memory process and the function of sleep. *Journal of Theoretics*, 6(6), 1-7.
39. Squire, L. R., & Alvarez, P. (1995). Retrograde amnesia and memory consolidation: a neurobiological perspective. *Current opinion in neurobiology*, 5(2), 169–177. [https://doi.org/10.1016/0959-4388\(95\)80023-9](https://doi.org/10.1016/0959-4388(95)80023-9)
40. Cousins, J. N., Wong, K. F., & Chee, M. W. (2019). Multi-night sleep restriction impairs long-term retention of factual knowledge in adolescents. *Journal of Adolescent Health*, 65(4), 549-557.
41. Harrington, M. O., Nedberge, K. M., & Durrant, S. J. (2018). The effect of sleep deprivation on emotional memory consolidation in participants reporting depressive symptoms. *Neurobiology of learning and memory*, 152, 10-19.
42. Peigneux, P., Laureys, S., Delbeuck, X., & Maquet, P. (2001). Sleeping brain, learning brain. The role of sleep for memory systems. *Neuroreport*, 12(18), A111-A124.
43. Donlea, J. M. (2019). Roles for sleep in memory: insights from the fly. *Current opinion in neurobiology*, 54, 120-126.
44. Walker, M. P. (2009). The role of slow wave sleep in memory processing. *Journal of Clinical Sleep Medicine*, 5(2 suppl), S20-S26.
45. Payne, J. D. (2011). Learning, memory, and sleep in humans. *Sleep Medicine Clinics*, 6(1), 15-30.
46. Frank, M. G., & Benington, J. H. (2006). The role of sleep in memory consolidation and brain plasticity: dream or reality?. *The Neuroscientist*, 12(6), 477-488.
47. Walker, M. P. (2009). The role of sleep in cognition and emotion. *Annals of the New York Academy of Sciences*, 1156(1), 168-197.
48. Kay, D. B., & Dzierzewski, J. M. (2024). What role do sleep and circadian rhythms play in psychological functioning including motivation, emotion, cognition, and performance?. *Research Directions: Sleep Psychology*, 1, e4.
49. Cordi, M. J., & Rasch, B. (2021). How robust are sleep-mediated memory benefits?. *Current Opinion in Neurobiology*, 67, 1-7.
50. Diekelmann, S., & Born, J. (2010). The memory function of sleep. *Nature Reviews Neuroscience*, 11(2), 114-126.
51. Kopasz, M., Loessl, B., Hornyak, M., Riemann, D., Nissen, C., Piosczyk, H., & Voderholzer, U. (2010). Sleep and memory in healthy children and adolescents—a critical review. *Sleep medicine reviews*, 14(3), 167-177.
52. Lo, J. C., Ong, J. L., Leong, R. L., Gooley, J. J., & Chee, M. W. (2016). Cognitive performance, sleepiness, and mood in partially sleep deprived adolescents: the need for sleep study. *Sleep*, 39(3), 687-698.
53. Wilhelm, I., Prehn-Kristensen, A., & Born, J. (2012). Sleep-dependent memory consolidation—what can be learnt from children?. *Neuroscience & Biobehavioral Reviews*, 36(7), 1718-1728.



**Marjana. Bhoomika et al.,**

54. Fenn, K. M., &Hambrick, D. Z. (2013). What drives sleep-dependent memory consolidation: Greater gain or less loss?.*Psychonomic bulletin & review*, 20, 501-506.
55. Ashton, J. E., Harrington, M. O., Langthorne, D., Ngo, H. V. V., &Cairney, S. A. (2020). Sleep deprivation induces fragmented memory loss. *Learning & Memory*, 27(4), 130-135.
56. De Bruin, E. J., van Run, C., Staaks, J., & Meijer, A. M. (2017). Effects of sleep manipulation on cognitive functioning of adolescents: A systematic review. *Sleep medicine reviews*, 32, 45-57.
57. Gathercole, S. E. (2004, January). Working memory and learning during the school years. In *Proceedings of the British Academy* (Vol. 125, No. 1, pp. 365-380).
58. Baddeley, A. (1992). Working memory. *Science*, 255(5044), 556-559.
59. R.G. Astill, K.B. Van DerHeijden, M.H. Van Ijzendoorn, E.J.W. Van Someren.(2012) Sleep, cognition, and behavioral problems in school-age children: a century of research meta-analyzed. *Psychol Bull*, 138 (2012), pp. 1109-1138
60. Fonseca, A. G., &Genzel, L. (2020). Sleep and academic performance: considering amount, quality and timing. *Current Opinion in Behavioral Sciences*, 33, 65-71.
61. Watson BO, Buzsáki G. Sleep, Memory & Brain Rhythms. *Daedalus*. 2015 Winter;144(1):67-82. doi: 10.1162/DAED_a_00318. PMID: 26097242; PMCID: PMC4474162.
62. Uji, M., & Tamaki, M. (2023). Sleep, learning, and memory in human research using noninvasiveneuroimaging techniques. *Neuroscience Research*, 189, 66-74.
63. Hysing, M., Harvey, A. G., Linton, S. J., Askeland, K. G., &Sivertsen, B. (2016). Sleep and academic performance in later adolescence: Results from a large population-based study. *Journal of sleep research*, 25(3), 318-324.
64. Curcio, G., Ferrara, M., & De Gennaro, L. (2006). Sleep loss, learning capacity and academic performance. *Sleep medicine reviews*, 10(5), 323-337.
65. Walker, M. P., &Stickgold, R. (2004). Sleep-dependent learning and memory consolidation. *Neuron*, 44(1), 121-133.
66. Eliasson, A.H., Lettieri, C.J. &Eliasson, A.H. Early to bed, early to rise! Sleep habits and academic performance in college students. *Sleep Breath*14, 71–75 (2010). <https://doi.org/10.1007/s11325-009-0282-2>
67. Killgore, W. D. (2010). Effects of sleep deprivation on cognition. *Progress in brain research*, 185, 105-129.
68. Gaultney, J. F. (2010). The prevalence of sleep disorders in college students: impact on academic performance. *Journal of American College Health*, 59(2), 91-97.
69. deAraújo, L. B. G., Bianchin, S., Pedrazzoli, M., Louzada, F. M., &Beijamini, F. (2022). Multiple positive outcomes of a later school starting time for adolescents. *Sleep Health*, 8(5), 451-457.
70. Campbell, I. G., Cruz-Basilio, A., Figueroa, J. G., & Bottom, V. B. (2023). Earlier Bedtime and Its Effect on Adolescent Sleep Duration. *Pediatrics*, e2022060607.
71. Levenson, J. C., Shensa, A., Sidani, J. E., Colditz, J. B., &Primack, B. A. (2017). Social media use before bed and sleep disturbance among young adults in the United States: A nationally representative study. *Sleep*, 40(9), zsx113.
72. Inazumi, C. K., Andrechuk, C. R. S., Lima, M. G., Zancanella, E., de Azevedo Barros, M. B., de Oliveira Cardoso, T. A. M., & Ceolim, M. F. (2020). Is napping related with health-related behaviors and sleep habits among adolescents? A population-based study. *Applied Nursing Research*, 56, 151373.
73. Modlin, T. (2002). Sleep disorders and hypnosis: To cope or cure?.*Sleep and Hypnosis*, 4, 39-46.





Mathematical Model with an Application of New Topsis Model Approach to Identify the Over Spread of Typhoid

T.Kavitha^{1*} and S. Shunmugapriya²

¹Assistant Professor, Department of Mathematics, A.V.V.M. Sri Pushpam College (Autonomous), Poondi, Thanjavur, (Affiliated to Bharathidasan University, Tiruchirappalli), Tamil Nadu, India.

²Assistant Professor, Department of Mathematics, Rajah Serfoji Govt. College (Autonomous), Thanjavur, (Affiliated to Bharathidasan University, Tiruchirappalli), Tamil Nadu, India.

Received: 13 Jun 2024

Revised: 16 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

T. Kavitha

Assistant Professor,
Department of Mathematics,
A.V.V.M. Sri Pushpam College (Autonomous),
Poondi, Thanjavur,
(Affiliated to Bharathidasan University, Tiruchirappalli),
Tamil Nadu, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

A pandemic is an illness that spreads rapidly over a huge region, such as several continents or the entire world. A potentially fatal infection, typhoid fever is brought on by the salmonella Typhi bacteria. Typhoid fever is thought to infect 11–20 million people annually, killing between 1,28,000 and 1,61,000 of them. Finding the important risk factor is the primary goal of the current inquiry. This study uses the Topsis MCDM technique to evaluate and identify the spread of typhoid.

Keywords: Topsis MCDM, Fuzzy Topsis MCDM, Topsis Multi Criteria Decision, MCDM tools.

INTRODUCTION

A severe, contagious, and potentially fatal illness associated with fever is typhoid. Salmonella enterica serovars are to blame. While some are grouped as nontyphoidal Salmonella [NTS] [1], Typhi, Paratyphi A, B, and C can all be classified as typhoidal Salmonella. Enteric fever is the collective term for typhoid and paratyphoid fever, which are caused by human host-confined typhoid strains. Salmonella serovar Paratyphi A has been associated with an increasing incidence of enteric fever in certain Asian countries [2, 3]. The primary means of transmission for typhoidal Salmonella is contaminated water or food [4]. In developing countries where typhoidal Salmonella is widespread, there is inadequate sanitation and hygiene, and there is a shortage of safe water and food, there is a

83358





Kavitha and Shunmugapriya

greater risk of illness [5]. An ongoing study on the prevalence of typhoid fever worldwide found that the disease causes 27 million illnesses and 200,000–600,000 deaths annually [7]. In 2010 there were 11.9 million cases of typhoid fever and 129,000 deaths in low- and middle-income countries, according to the International Vaccine Institute [8]. For the most part, typhoid is endemic in developing countries like Pakistan [9].

We attempt to search for any potential escape route as the virus spreads over national borders. Medical science is making every effort to find a potential remedy. Even in the non-medical field, we have made an effort to contribute in some small way to the cause. Based on information gathered thus far from literature surveys, media websites, and medical advice, we have identified three primary causes of typhoid disease transmission. The main ways that it spreads are through contaminated food or water, through contact with an infected person, and by kissing. Finding the most important risk factors that contribute to the spread of typhoid fever is the primary goal of this investigation. The technique known as Topsis Multi Criteria Decision Making (MCDM) will be employed to ascertain the extent of typhoid fever outbreak. These days, MCDM technologies are used to environmental, social, and economic issues, among others (5-7). Certain MCDM strategies assign a rank of indications, while others assign a priority value. Technique for Order Preference by Similarity to an Ideal Solution (TOPSIS), put forth by Hwang and Yoon in 1981 (8), is one of the ranking-based MCDM tools.

Model of Topsis

The Technique for Order Preference by Similarity to an Ideal Solution (TOPSIS), which analyzes decisions for alternatives, is one of the many criteria approaches. The TOPSIS notion is logical and comprehensible, and the computation required is simple. It's also important to acknowledge the inherent difficulties in determining trustworthy subjective preferences for the criterion (9). The TOPSIS technique flow chart is shown in Figure 1.

Decision-making in TOPSIS is essentially based on six ineffective steps:

(i) Determine the decision matrix's normalized values.

Think about the fact that there are m alternatives. $D = [rij]_{m \times m}$ be the normalized matrix. The definition of the normalized value rij is

$$rij = \frac{xij}{\sqrt{\sum_{i=1}^m xij^2}} \forall i, j$$

(ii) Determine the decision matrix that is weighted and normalized. It is calculated to get the weighted normalized value vij as

$$\sum_{j=1}^n Wj = 1$$

$$vij = wjrij \forall i, j$$

where wj is the jth criterion's weight, and

(iii) Find the optimal and non-optimal solution

$$A^+ = \{v1+, v2+, v3+, \dots, vm+\} = \{(\max vij | j \in Cb), (\min vij | j \in Cc)\}$$

$$A^- = \{v1-, v2-, v3-, \dots, vm-\} = \{(\min vij | j \in Cb), (\max vij | j \in Cc)\}$$

Where Cc is linked to the cost criteria and Cb to the benefit criteria.

(iv) Use the m dimensional supremum distance to compute the separation metrics. We use the Euclidean distance to compute separation metrics in TOPSIS. The following represents how each option and its negative relation to the ideal answer are separated:

$$Si = \max \{ |vij - vj+| : j=1, 2, \dots, m \}, \sqrt{\sum_{j=1}^m (vij - vj-)^2} \forall i$$





Kavitha and Shunmugapriya

(v) Determine how near the optimal answer you are:
The definition of the alternative A_i 's relative proximity to A^- is

$$RC_i^* = \frac{S_i^-}{S_i^- + S_i^+}, \forall i$$

(vi) Determine the order of preference. The range of RC_i^* index values is 0 to 1. The closer an alternative is to the optimal answer, the higher the index value.

METHODOLOGY

The primary goal of this study is to determine the most important risk factor for the spread of typhoid fever.

Let $E = f(D,W)$ denote the risk of spread of Typhoid fever.

Where,

D stands for the collection of all of E 's elements.

$D = \{M : M = \text{all selected factors}\}$

$W = \{W : W = \text{Weights all selected}\}$

Where M and W stand for the parameter that has been chosen and its PV, respectively.

The current study's methodology is broken down into three sections: using the Topsis technique, choosing alternatives, and selecting criteria.

Selection of Criteria

The chosen literature review, expert survey, and media survey are the study's current criteria. In order to identify every risk factor for the spread of typhoid fever, we examined close to thirty publications. Therefore, the literature review is a key selection criterion for all of the choices in this study. Since the study's focus is on medical issues, experts play a significant role. To determine the risk factors for the spread of typhoid fever, we gathered the opinions of about ten specialists. Another crucial criterion for factor selection is the media survey, since they are constantly seeking to gather data regarding typhoid fever. In this study, the risk factor is chosen based on reports from three credible media outlets.

Selection of Alternatives

Expert survey and relative peasant opinions are taken into consideration as criteria in this study's literature evaluation, and they are represented by the letters $C_1, C_2,$ and $C_3,$ respectively. Typhoid fever risk factors are shown in Table 1. The decision hierarchy is displayed in Figure 2. Our decision problem's structure. A five-point rating system Table 3 convert as a score and score table represented by Table 4. The formula represents each of Table 4's column vectors. Following computation, the normalized decision matrix shown in Table 5 is the result. Table 5 shows the PV for each criterion as determined by the MCDM approach in column two.

$$rij = \frac{xij}{\sqrt{\sum_{i=1}^4 x_{1j}^2}}, \forall i = 1,2,3, j = 1,2,3$$

Compute the weighted normalized decision matrix from Table 5 using the formula. The weighted normalized decision matrix is shown in Table 6. Utilize the formulas in (2) and (3) to find the ideal A^+ and negative-ideal A^- solution. Table 6's final two columns show the A^+ and A^- values.

$$vij = wjrij, \forall i=1,2,3, j=1,2,3. \tag{1}$$

$$A^+ = \{v1+, v2+, v3+, \dots, vm+\} = \{(max_{ij} vij | j \in Cb)\} \tag{2}$$

$$A^- = \{v1-, v2-, v3-, \dots, vm-\} = \{(min_{ij} vij | j \in Cb)\} \tag{3}$$

RC_i^* value for Topsis

	S_i^+	S_i^-	$S_i^+ + S_i^-$	RC_i^*	Rank
A_1	0	0.24165459	0.24165459	1	1
A_2	0.12081266	0.12082905	0.24164171	0.5	2





Kavitha and Shunmugapriya

A ₃	0.24165817	0	0.24165817	0	3
----------------	------------	---	------------	---	---

CONCLUSION

With the aid of the Topsis MCDM approach, the current study has sought to create a novel model for evaluation of the risk assessment of typhoid fever. This new model will have the advantage of being able to study the spread of typhoid fever via doctors' opinions, literature reviews, and media surveys in an objective and cognitive manner. We have identified three potential risk factors for typhoid fever: bacterial contamination, contact with an infected individual, and kiss-related contamination. Following the gathering of all the necessary data, we used Fuzzy Topsis MCDM to determine the PV of the criteria. Utilizing the PV of the criterion, use Topsis to identify the optimal options. Finally, through media surveys, I concluded using the Topsis MCDM method that Typhoid fever is more spread due to sewage contamination.

REFERENCES

- Gal-Mor O, Boyle EC, Grassl GA, *et al.* Same Species, Different Diseases: How and Why Typhoidal and Non-Typhoidal Salmonella Enterica Serovars Differ. *Front Microbiol* 2014;5:391.
- Crump JA, Luby SP, Mintz ED, *et al.* The global burden of typhoid fever. *Bull World Health Organ* 2004;82:346-353.
- Eng SK, Pusparajah P, Ab Mutalib NS, *et al.* Salmonella: a review on pathogenesis, epidemiology and antibiotic resistance. *Front Life Sci* 2015;8:284-293.
- Penakalapati G, Swarouth J, Delahoy MJ, *et al.* Exposure to animal feces and human health: a systematic review and proposed research priorities. *Environ. Sci. Technol* 2017;9:11537-11552.
- Antillón M, Warren JL, Crawford FW, *et al.* The burden of typhoid fever in low-and middle-income countries: A metaregression approach. *PLoS Negl. Trop. Dis* 2017;11:e0005376.
- Gunn JS, Marshall JM, Baker S, *et al.* Salmonella chronic carriage: epidemiology, diagnosis, and gallbladder persistence. *Trends in microbiology* 2014;22:648-655.
- Buckle GC, Walker CLF, Black RE, *et al.* Typhoid fever and paratyphoid fever: Systematic review to estimate global morbidity and mortality for 2010. *J. Glob. Health* 2012;2:10401.
- Ayub U, Khattak AA, Saleem A, *et al.* Incidence of typhoid fever in Islamabad, Pakistan. *American-Eurasian Journal of Toxicological Sciences* 2015;7:220-223.
- Butt T, Ahmad RN, Mahmood A, *et al.* Ciprofloxacin treatment failure in typhoid fever case, Pakistan. *Emerging infectious diseases* 2003;9:1621.
- Anderson RM, Heesterbeek H, Klinkenberg D, Hollingsworth TD. How will country-based mitigation measures influence the course of the COVID-19 epidemic?. *The Lancet*. 2020. [https://doi.org/10.1016/S0140-6736\(20\)30567-5](https://doi.org/10.1016/S0140-6736(20)30567-5)
- World Health Organization. Available at: <https://www.who.int/>
- Xu X, Chen P, Wang J, Feng J, Zhou H, Li X, *et al.* Evolution of the novel coronavirus from the ongoing Wuhan outbreak and modeling of its spike protein for risk of human transmission. *Science China Life Sciences*, 2020;63(3):457-60. <https://doi.org/10.1007/s11427-020-1637-5> PMID: 32009228 PMCID:PMC7089049
- Huang C, Wang Y, Li X, Ren L, Zhao J, *et al.* Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *The Lancet*, 2020;395(10223):497-506. [https://doi.org/10.1016/S0140-6736\(20\)30183-5](https://doi.org/10.1016/S0140-6736(20)30183-5)
- Majumder P, Majumder M, Saha AK, Sarkar K, Nath S. Real time reliability monitoring of hydro-power plant by combined cognitive decision-making technique. *International Journal of Energy Research*. 2019. <https://doi.org/10.1002/er.4530>
- Majumder P, Saha AK, Majumder M. A mathematical approach of exploration towards extreme risk factor in cancer of optimal condition. *International Journal of Pharmaceutical Sciences and Research*, 2018;9(9):3732-42. [https://doi.org/10.13040/IJPSR.0975-8232.9\(9\).3732-42](https://doi.org/10.13040/IJPSR.0975-8232.9(9).3732-42)





Kavitha and Shunmugapriya

Table 1: Explanation of Chosen Elements

Name of Element	Description	Reference	
Contamination due to bacteria (A1)	The bacterial spread of sewage contamination of food or water.	(2,24)	
Contact with an infected individual resulting in contamination (A2)	There is a significant likelihood of the virus spreading through contact with an infected individual. People are asked to maintain safe distances because of this.	(2,24)	
Contamination due to kiss (A3)	If kiss to loved one in your families and relatives there is a chance to spread of this virus.	(2,25,26)	

Table 2: 5-Point Scale

Name	Score
Average	1
Good	2
Very Good	3
Excellent	4
Outstanding	5

Table 3: Table of scores for the alternatives based on the criteria

Alternatives	Criteria		
	C1	C2	C3
A1	V.G	E	O
A2	G	V.G	E
A3	A	G	V.G

Table 4: Using a 5-point rating system, create an alternate score table.

Alternatives	Criteria		
	C1	C2	C3
A1	X ₁₁ = 3	X ₁₂ = 4	X ₁₃ = 5
A2	X ₂₁ = 2	X ₂₂ = 3	X ₂₃ = 4
A3	X ₃₁ = 1	X ₃₂ = 2	X ₃₃ = 3
$\sqrt{\sum_{i=1}^3 x_{ij}^2}$	3.74165739	4.35889894	7.07106781

Table 5. Decision Matrix Normalized

	Priority ranking for every criterion		
	w ₁ = 0.2592593	w ₂ = 0.3333333	w ₃ = 0.4444444
Alternatives	C1	C2	C3
A1	r ₁₁ = 0.80178373	r ₁₂ = 0.91766294	r ₁₃ = 0.91766294
A2	r ₂₁ = 0.53452248	r ₂₂ = 0.68824720	r ₂₃ = 0.68824720
A3	r ₃₁ = 0.26726124	r ₃₂ = 0.45883147	r ₃₃ = 0.45883147

Table 6. Normalized Weighted Decision Matrix

Alternatives	C1	C2	C3
A1	v ₁₁ = 0.20786989	v ₁₂ = 0.30588762	r ₁₃ = 0.31426965
A2	v ₂₁ = 0.13857992	v ₂₂ = 0.22941511	r ₂₃ = 0.25141572





Kavitha and Shunmugapriya

A3	$v_{31} = 0.06928996$	$v_{32} = 0.15294381$	$r_{33} = 0.18856179$
A ⁺	$v^{+1} = 0.20786989$	$v^{+2} = 0.30588762$	$v^{+3} = 0.31426965$
A ⁻	$v^{-1} = 0.06928996$	$v^{-2} = 0.15294381$	$v^{-3} = 0.18856179$

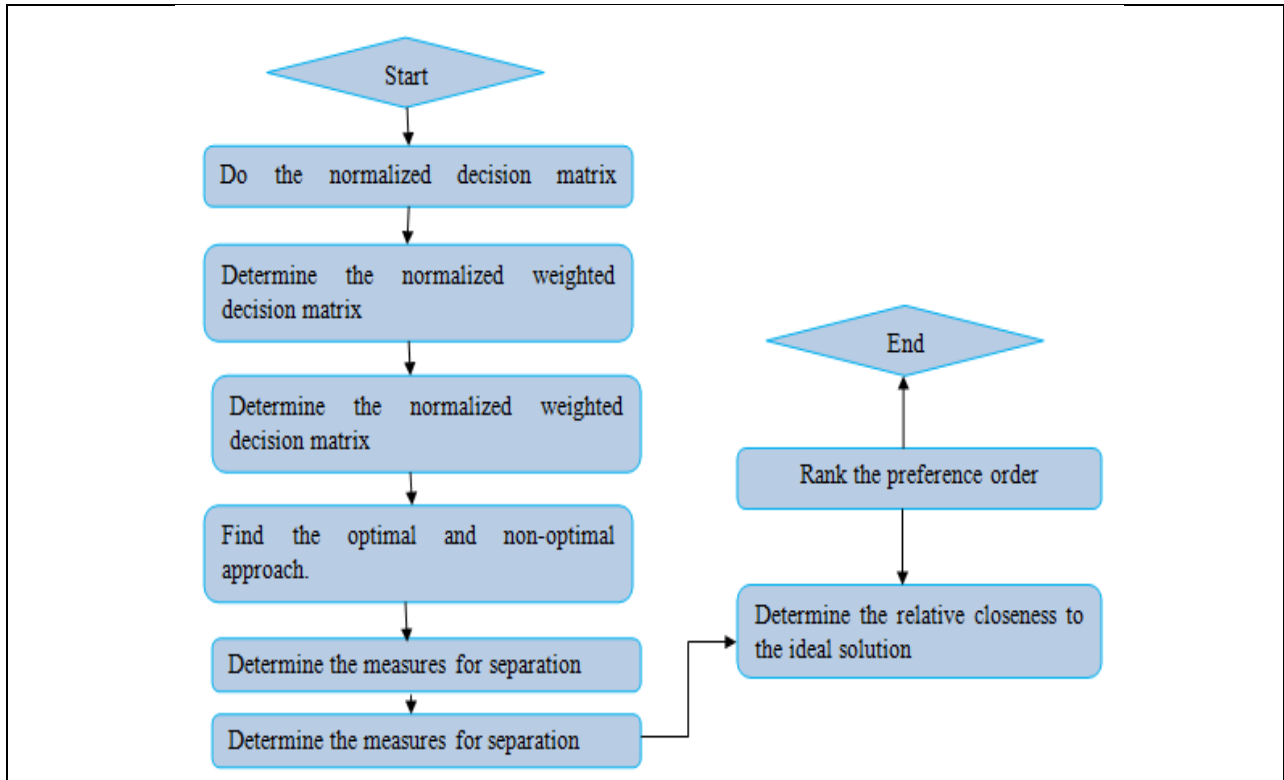


Figure 1. The TOPSIS technique flow chart

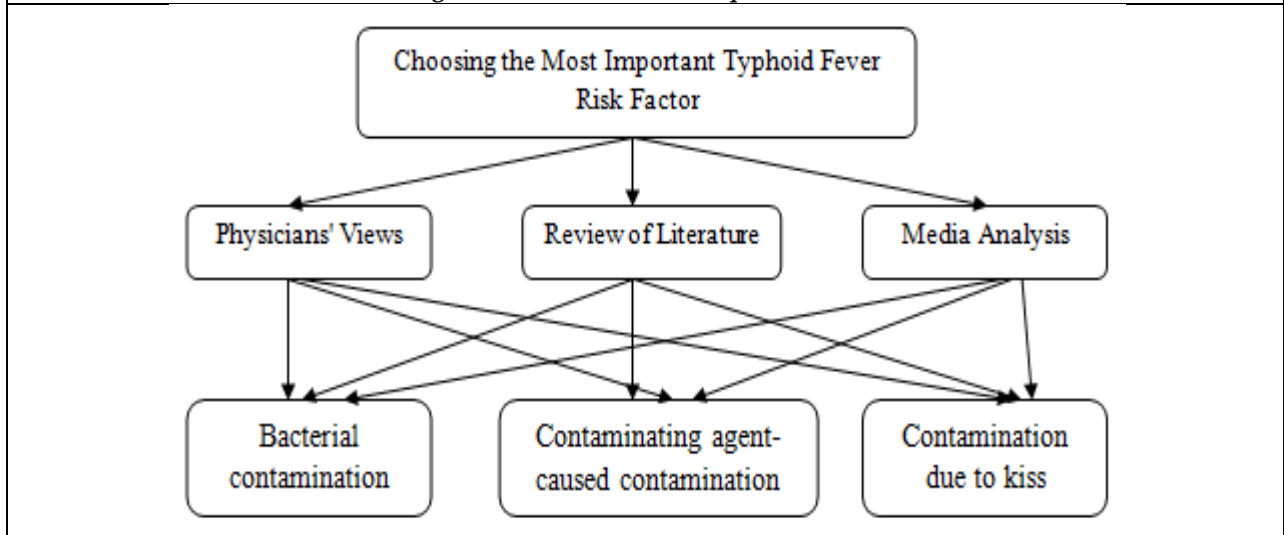


Figure 2. Decision hierarchy Structure





Kavitha and Shunmugapriya

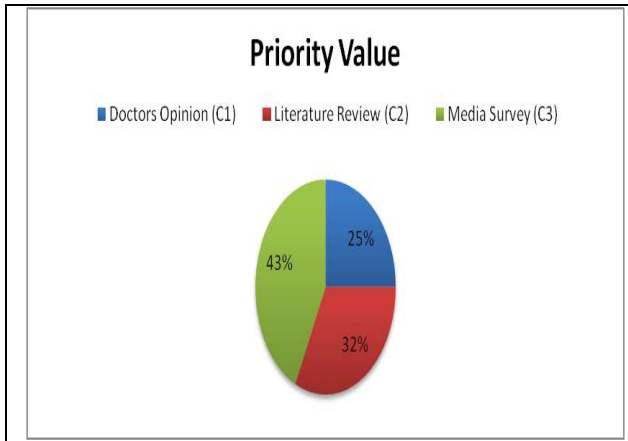


Figure 3. Outcome based on Fuzzy MCDM Criteria

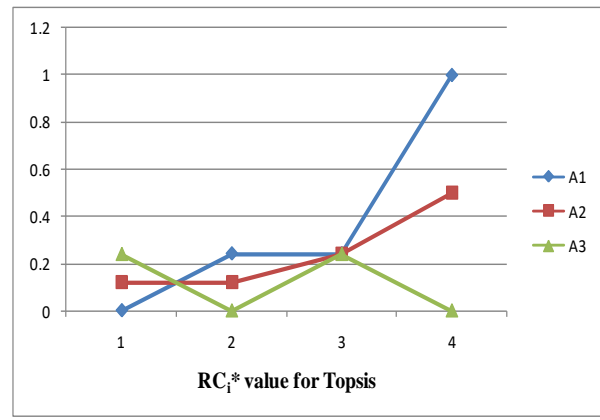


Figure 4. RCi* value for Topsis





Unveiling the Cloud Kitchen Phenomenon: Analyzing Customer Awareness and Shifting Preferences

Arnaz Kaizad Wadia and Nupur Rawal*

Assistant Professor, Faculty of Business Administration, GLS University, Ahmedabad, Gujarat, India

Received: 19 Jun 2024

Revised: 20 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

Nupur Rawal
Assistant Professor,
Faculty of Business Administration,
GLS University, Ahmedabad, Gujarat, India
E.Mail: nupur.rawal26@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The research explores the transformative impact of cloud kitchens in the food service industry. It aims to understand the necessity of cloud kitchens and discern how they differ from traditional dine-in establishments. The study also investigates key factors influencing consumer decisions, such as purchasing behavior, perceptions, and challenges faced when ordering from cloud kitchens. The research employs a descriptive research design, utilizing both primary and secondary data sources. Primary data is collected through a questionnaire method, focusing on Ahmedabad customers. Secondary data is sourced from internet-available reports and journals. The major purpose of the study is to ensure a holistic understanding of the evolving landscape of cloud kitchens. The study reveals a growing awareness of cloud kitchens, primarily driven by social media platforms. Factors influencing consumer decisions include online presence, food quality, and affordability. Cloud kitchens, operating as production units with streamlined costs, offer benefits like faster delivery, sustainability, and an alternative to traditional dining. The findings suggest practical implications for stakeholders in the food service industry. Cloud kitchens can enhance their visibility on social media platforms to capitalize on the growing awareness. The identified benefits, including faster delivery and sustainability, provide practical insights for cloud kitchen operators aiming to meet consumer expectations. This research contributes to the understanding of the dynamic landscape shaped by cloud kitchens. The study's contribution lies in its comprehensive analysis, shedding light on the transformative role of cloud kitchens in the evolving food service industry.

Keywords: Cloud Kitchen, Customer Preference, Consumer Behavior, Food Service Industry

INTRODUCTION

In the ever-evolving landscape of the food service industry, a revolutionary concept has emerged, challenging traditional norms and redefining how individuals experience and access food – the cloud kitchen. This innovative model operates as a production unit focused solely on preparing and delivering food, eliminating the need for expensive front-of-house operations. Its key features are as follows:



**Arnaz Kaizad Wadia and Nupur Rawal****Breaking Free from the Bricks and Mortar**

Unlike conventional restaurants with dine-in facilities, cloud kitchens operate with a leaner business model, offering several advantages. Firstly, they significantly reduce overhead costs by eliminating expenses related to rent, furniture, décor, and tableware. This reduction in fixed costs translates to improved profit margins for cloud kitchens. Additionally, cloud kitchens boast increased flexibility, allowing them to scale up or down depending on demand, adapt to market conditions, and test new concepts. The absence of dine-in operations also enhances their focus on quality and efficiency in food preparation.

Catering to the Digital Appetite

The rise of cloud kitchens is closely tied to the growing popularity of online food delivery platforms. By partnering with these platforms, cloud kitchens gain access to a vast network of potential customers, tapping into the ever-increasing demand for convenient and affordable home delivery. This partnership not only facilitates broader market reach but also provides valuable data and insights into consumer preferences. With this information, cloud kitchens can tailor their menus and promotions to specific customer segments, enhancing their appeal in a competitive market.

Diverse Culinary Landscape

Initially associated with fast food and quick-service meals, cloud kitchens have evolved to encompass a diverse range of culinary experiences. Modern cloud kitchens now offer everything from gourmet burgers and handcrafted pizzas to vegan dishes, ethnic cuisines, and healthy meal plans. This diversity caters to the increasingly sophisticated palates of modern consumers, allowing cloud kitchens to tap into niche markets and cater to various dietary preferences. This adaptability positions cloud kitchens as dynamic players in the culinary scene.

Innovations and Future Potential

Cloud kitchens are not mere trends; they represent a paradigm shift in the food service industry. As technology continues to advance, further innovations in this space are expected. Automation, including robotic kitchens and automated order fulfilment systems, holds the promise of improving efficiency and reducing labor costs. Artificial intelligence (AI) is poised to play a significant role in personalizing menus, recommending dishes, and optimizing delivery routes, enhancing the overall customer experience. Data-driven decision-making, informed by customer data and trends, empowers cloud kitchens to make strategic choices regarding menu development, pricing strategies, and marketing campaigns.

The Impact on the Food Service Landscape: The rise of cloud kitchens is reshaping the traditional food service industry. While some traditional restaurants may face challenges adapting to this new landscape, others are embracing the concept by launching their virtual kitchens to expand their reach and cater to the growing demand for delivery. This coexistence of traditional and virtual models is fostering a more dynamic and competitive food service landscape, ultimately benefiting consumers with a wider variety of choices and convenient access to delicious meals.

LITERATURE REVIEW

The studies conducted by Mathur (2023) and Deepak (2022) shed light on the evolving landscape of food consumption, particularly the rise of cloud kitchens. The study (Mathur, 2023) emphasized the strong influence of perception on consumer purchase intentions, providing valuable insights for cloud kitchen businesses. Deepak (2022) delved into the operational dynamics of cloud kitchens, emphasizing their exclusive focus on food delivery and the cost advantages compared to traditional setups. Through regression analysis and customer surveys, the study (Deepak, 2022) explored factors influencing cloud kitchen businesses. During the COVID-19 pandemic, cloud kitchens played a pivotal role, as outlined in the study (Rudrani Chatterjee, 2022). This research explored distinctive aspects, consumer behavior, and the pandemic's impact on food delivery interactions. It sought to identify challenges and consumer preferences, contributing valuable insights for the industry's future growth. Shaik Mehnaz et al. (2021)



**Arnaz Kaizad Wadia and Nupur Rawal**

provided an analysis of cloud kitchens in India, offering insights into industry trends and impacts, particularly during the pandemic. The paper emphasized the disruptive nature of the industry and its evolving landscape (Shaik Mehnaz, 2021). The study by Sanjukta surge in online food delivery's popularity in India, driven by lifestyle changes and mobile technology. The study identified factors affecting consumer adoption of cloud kitchens, emphasizing the significance of facilitating conditions, social influence, online coupons, and food variety (Sanjukta Pookulangara, 2023).

The study (Han-Shen Chen, 2020) and (Nikhil Chhabra, 2021) analyzed consumer perceptions of online food ordering services in India, aiming to understand the factors attracting consumers to these services. Also cloud kitchen-based studies are conducted in other countries like Hakim explored consumer awareness of cloud kitchen in Brazil (Hakim, 2022). Dsouza (2022) identified factors influencing customers' intentions to use mobile apps for ordering food, revealing the positive effect of six major factors (Dsouza Prima Frederick, 2022). Sharma (2023) examined the structural relationship between various factors influencing behavioral intention toward online food delivery services (Sharma, 2023). Nurul (2023) investigated the influence of consumer behavioral intention toward online food delivery services, providing insights into key factors affecting consumers' intentions (Nurul Syahirah Idris, 2023).

RESEARCH METHODOLOGY

This study aims to investigate the necessity of cloud kitchens and explore customer preferences between traditional dine-in establishments and cloud kitchens. The objectives include assessing the purchasing behavior from cloud kitchens, understanding customer preferences, identifying factors influencing buying decisions, and examining challenges faced by customers when ordering from cloud kitchens. The scope of the study focuses on examining the awareness level of cloud kitchens among customers and exploring their preference for cloud kitchens. The research design is descriptive, employing both primary and secondary data sources. Primary data is collected through a questionnaire method from customers in Ahmedabad, while secondary data from reports and journals available on the internet supplements the study. The sampling plan targets all customers in Ahmedabad, with the sampling units being customers aware of cloud kitchens. The study includes a sample size of 152 customers, selected through non-probability convenience sampling. The research instrument is a questionnaire, and data analysis employs SPSS software.

Analysis & Interpretation

The chi-square test was conducted to analyze the association between age groups and the purchase from cloud kitchens. The crosstabulation (Table 1) presents a breakdown of respondents based on their age and whether they have purchased from cloud kitchens. The data reveals that the majority of respondents in the 26-35 age group have purchased from cloud kitchens, with 48 respondents affirming their purchases compared to 9 who have not. This is followed by the 18-25 age group, with 39 respondents indicating they have purchased from cloud kitchens.

The chi-square test statistic is calculated as 8.634 with 3 degrees of freedom, resulting in an asymptotic significance value of 0.035 (Table 2). The null hypothesis (H₀) posits that there is no association between age and purchasing habits from cloud kitchens, while the alternative hypothesis (H₁) suggests a significant association. The critical p-value for rejecting the null hypothesis is conventionally set at 0.05.

Given that the calculated p-value (0.035) is less than 0.05, the null hypothesis is rejected. This indicates a statistically significant association between age groups and the likelihood of purchasing from cloud kitchens. Therefore, it can be concluded that age influences purchase intentions from cloud kitchens. The 26-35 age group showed the highest propensity to buy from cloud kitchens, primarily comprising of Millennials who are likely to be the part of working population. The rejection of the null hypothesis suggests that there is a notable association between age and the decision to purchase from cloud kitchens, emphasizing the importance of considering age demographics while analyzing consumer behavior towards cloud kitchens. Also, the chi-square test was conducted to assess the



**Arnaz Kaizad Wadia and Nupur Rawal**

relationship between marital status and the frequency of ordering from cloud kitchens within a month. The crosstabulation (Table 3) presents a breakdown of respondents based on their marital status (Single or Married) and ordering frequency.

The data shows that among single individuals, 34 respondents rarely ordered from cloud kitchens, 24 occasionally ordered, and 6 frequently ordered. For married individuals, 20 respondents rarely ordered, 26 occasionally ordered, and 3 frequently ordered. The chi-square test statistic is calculated as 3.403 with 3 degrees of freedom, resulting in an asymptotic significance value of 0.334 (Table 4). The null hypothesis (H₀) posits that there is no relationship between marital status and the frequency of ordering from cloud kitchens, while the alternative hypothesis (H₁) suggests a significant relationship. The critical p-value for rejecting the null hypothesis is conventionally set at 0.05. However, in this case, the calculated p-value (0.334) is greater than 0.05, leading to the acceptance of the null hypothesis.

Therefore, the results indicate that there is no statistically significant association between marital status and the frequency of ordering from cloud kitchens within a month. In other words, marital status does not appear to influence how frequently individuals order from cloud kitchens. This suggests that factors other than marital status may play a more prominent role in determining the ordering habits of individuals from cloud kitchens

The factor analysis conducted on the dataset aimed to explore underlying patterns and relationships within the variables related to consumer preferences and buying behavior towards cloud kitchens. The reliability test indicated that the overall data is highly reliable, with a Cronbach's Alpha value of 0.889, surpassing the recommended threshold of 0.7. The KMO and Bartlett's Test provided further insights into the suitability of the data for factor analysis. With a KMO value of 0.878, which is considered excellent (above 0.8), and a significant Bartlett's Test ($p < 0.05$), it was determined that the dataset is well-suited for factor analysis. The Total Variance Explained (Table 5) displayed those four components had eigenvalues greater than 1, covering a cumulative variance of 67%. This surpasses the standard recommendation of 60%, indicating that the extracted factors explain a substantial portion of the total data variance.

The Rotated Component Matrix (Table 6) revealed the statements associated with each of the four extracted factors. The factors were labelled as follows: 1) Digital Culinary Experience 2) Influential Cloud Kitchen Acumen 3) Cloud Kitchens Ordering Ease and 4) Socialized Culinary Advocacy. The Nomenclature Table (Table 7) provided a clear overview of how statements were grouped under each factor. Furthermore, the Reliability Test of All the Factors (Table 8) assessed the internal consistency of each factor. All four factors demonstrated high reliability, with Cronbach's Alpha values exceeding 0.7, reinforcing the robustness of the extracted factors. In summary, the factor analysis successfully identified and labelled four key factors that collectively explain a significant proportion of the variance in the data. These factors represent distinct dimensions of customer preference and buying behavior related to cloud kitchens, providing valuable insights for understanding the multifaceted nature of customer preference towards this emerging culinary concept of cloud kitchen.

FINDINGS

The study aimed to achieve two primary objectives: understanding the customers buying behavior from cloud kitchens and gauging their preferences toward this emerging food service model. Regarding the first objective, it was found that 76% of the respondents had purchased from cloud kitchens, with the remaining 24% citing reliability issues, often related to quality or trust concerns, as the primary reason for not doing so. This finding reflects higher level of awareness about cloud kitchens. Moreover, a significant portion, 54%, ordered from cloud kitchens 1-2 times a month indicating higher preference of ordering from cloud kitchen. Price and affordability emerged as the most influential factor during the purchasing decision, followed by the quality of food and online presence. Respondents were least influenced by the speed of delivery. Social media platforms and word of mouth were the predominant channels through which respondents became aware of cloud kitchens. The most significant challenges reported included the limited availability of cloud kitchens in the market and insufficient customer assistance.



**Arnaz Kaizad Wadia and Nupur Rawal**

In pursuit of the second objective, the study delved into customer preference towards cloud kitchens. Factors such as digital culinary experience, socialized culinary advocacy, ordering ease offered by cloud kitchens played a crucial role in justifying the shifting preference towards cloud kitchens. Enhanced awareness and improved customer service were identified as critical areas requiring attention. Notably, 14% of respondents did not view cloud kitchens as the future, while 58% expressed a willingness to continue using them. A significant portion, 50%, would recommend cloud kitchens to others. Gender-wise, more males believed in the future of cloud kitchens than females. The age group of 26–35 years emerged as the one making frequent purchases among all age groups, highlighting potential target demographics for formulating cloud kitchen related marketing strategies. Overall, the study provides valuable insights into both the consumer behavior and preference surrounding cloud kitchens.

CONCLUSION

The Indian Cloud Kitchen Market has experienced significant expansion and is projected to grow at a rate of 24% by 2026 (ETRetail, 2023). The study reveals a growing awareness of cloud kitchens, indicating the need for increased visibility through social media platforms. Benefits associated with cloud kitchens such as faster delivery, sustainability, and a viable alternative to traditional dining are the major reasons contributing towards its wider acceptability. The food market is poised for growth in the coming years due to rising disposable income, changing lifestyles, and a busy work culture. In this scenario, it is worth noting that despite being a relatively new concept, customers willingness to continue using cloud kitchens in their daily lives, highlights a positive and progressive outlook for the industry in the coming years.

REFERENCES

1. Deepak, B. R. (2022). A Study on Financial Feasibility of Cloud Kitchen Firms in Hyderabad Region in Telangana, India. *Asian Journal of Agricultural Extension, Economics & Sociology*, 40(10), 342–350.
2. Dsouza Prima Frederick, G. B. (2022). Customer Perception towards Online Food Delivery Services- Development of Conceptual Model. *International Journal of Case Studies in Business, IT, and Education*, 6(1), 470-505.
3. ETRetail. (2023, March 25). Retrieved from ETRetail.com: <https://retail.economicstimes.indiatimes.com/news/food-entertainment/food-services/cloud-kitchen-market-may-reach-1-9-billion-by-fy26/98978668>
4. Hakim, M. P. (2022). Consumer Awareness of Dark Kitchen: A Brazilian Perspective. *International Journal of Hospitality Management*, 40(4), 567-580.
5. Han-Shen Chen, C.-H. L.-Y.-Y. (2020). Consumer Attitudes and Purchase Intentions toward Food Delivery Platform Services. *Sustainability*, 12(23), 1-18.
6. Mathur, P. M. (2023). Consumer Purchase Intention and Behavior Toward Cloud Kitchen (Pandemic Opportunity) with Reference to India: An Empirical Examination. *Jindal Journal of Business Research*, 12(2), 194-208.
7. Nikhil Chhabra, A. R. (2021). Rise and the Future of Cloud Kitchens in India: A Consumer Study. *International Journal of Research in Engineering, Science and Management*, 4(7), 158-164.
8. Nurul Syahirah Idris, M. A. (2023). New Trends of Cloud Kitchen Technology and Consumer Purchase Decisions: A Conceptual Study. 19(2).
9. Rudrani Chatterjee, A. S. (2022). Ethical and Sustainable Perceptions on Cloud Kitchen Business- A Study of Consumers and Stakeholders during the Covid-19 Pandemic. *International Journal of Hospitality and Tourism Systems*(Special Issue on COVID-19), 76-87.
10. Sanjukta Pookulangara, H. W. (2023). Consumer attitudes toward ordering from cloud kitchens: a gender and marital status perspective. *International Journal of Contemporary Hospitality Management*, 35(5), 1859-1879.
11. Shaik Mehnaz, M. B. (2021). Cloud Kitchen in India: A Research Paper. *International Journal of Current Research*, 13(5), 17325-17328.
12. Sharma, S. (2023). A Study on the Online Food Delivery Services Market in Chandigarh From a Customer Perspective. *International Journal of Professional Business Review*, 8, 1-19.





Arnaz Kaizad Wadia and Nupur Rawal

Table 1 Crosstab of Age & Purchase from Cloud Kitchens

		Have you purchased from Cloud Kitchen		Total
		Yes	No	
Age	18-25	39	10	49
	26-35	48	9	57
	36-45	22	10	32
	46 & above	7	7	14
Total		116	36	152

Table 2 Chi-Square Tests

	Value	Df	Asymp. Sig. (2-sided)
Pearson Chi-Square	8.634 ^a	3	.035

Table 3 Crosstab of Marital Status & Ordering Frequency from Cloud Kitchens

		How Frequently do you Order from Cloud Kitchen within a Month?				Total
		Only Heard of Cloud Kitchens but never Ordered	Rarely (1-2 times)	Occasionally (3-5 times)	Frequently (6 or more times)	
Marital Status	Single	1	34	24	6	65
	Married	2	20	26	3	51
Total		3	54	50	9	116

Table 4 Chi-Square Test

	Value	Df	Asymp. Sig. (2-sided)
Pearson Chi-Square	3.403a	3	.334

Table 5 Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	6.783	42.396	42.396	6.783	42.396	42.396	3.588	22.424	22.424
2	1.719	10.745	53.141	1.719	10.745	53.141	2.656	16.600	39.024
3	1.125	7.032	60.173	1.125	7.032	60.173	2.348	14.677	53.701
4	1.014	6.338	66.511	1.014	6.338	66.511	2.050	12.810	66.511
5	.811	5.066	71.577						
6	.719	4.496	76.073						
7	.621	3.881	79.953						
8	.559	3.491	83.444						
9	.506	3.162	86.606						
10	.395	2.468	89.074						
11	.389	2.429	91.503						





Arnaz Kaizad Wadia and Nupur Rawal

12	.381	2.379	93.881						
13	.293	1.830	95.711						
14	.273	1.705	97.416						
15	.227	1.416	98.832						
16	.187	1.168	100.000						

Statements	Component			
	1	2	3	4
Cloud Kitchen perform better than Traditional Restaurants	0.788			
The shift to cloud kitchens is beneficial for the society	0.74			
Cloud Kitchen Concept attracts me	0.697			
Cloud Kitchens offer better value for money compared to traditional restaurants.	0.678			
I trust the quality of food from Cloud Kitchens.	0.673			
Cloud Kitchens offer a convenient way to order food.	0.607			
I have positive feeling when ordering and buying from cloud kitchens	0.529			
I am interested in knowing more about Cloud Kitchens		0.749		
Social media platforms and online advertisements have influenced my awareness and perception of Cloud Kitchens.		0.697		
Online reviews and personal recommendations are significant factors in my perception of Cloud Kitchens.		0.671		
I am more likely to order from Cloud Kitchens when they provide attractive deals and discounts.		0.648		
I frequently use food delivery apps to order from Cloud Kitchens.			0.803	
I frequently order from Cloud Kitchens.			0.71	
Price is a significant factor in my decision to order from Cloud Kitchens.			0.696	
My friends often share their Ordering experience from Cloud Kitchens				0.85
My friends often recommend to order from Cloud Kitchens				0.819
Extraction Method: Principal Component Analysis.				
Rotation Method: Varimax with Kaiser Normalization.				

No.	Factorial Group	Statements
1	Digital Culinary Experience	Cloud Kitchen perform better than Traditional Restaurants
		The shift to cloud kitchens is beneficial for the society
		Cloud Kitchen Concept attracts me
		Cloud Kitchens offer better value for money compared to traditional restaurants.





Arnaz Kaizad Wadia and Nupur Rawal

		I trust the quality of food from Cloud Kitchens.
		Cloud Kitchens offer a convenient way to order food.
		I have positive feeling when ordering and buying from cloud kitchens
2	Influential Cloud Kitchen Acumen	I am interested in knowing more about Cloud Kitchens
		Social media platforms and online advertisements have influenced my awareness and perception of Cloud Kitchens.
		Online reviews and personal recommendations are significant factors in my perception of Cloud Kitchens.
		I am more likely to order from Cloud Kitchens when they provide attractive deals and discounts.
3	Cloud Kitchens Ordering Ease	I frequently use food delivery apps to order from Cloud Kitchens.
		I frequently order from Cloud Kitchens.
		Price is a significant factor in my decision to order from Cloud Kitchens.
4	Socialized Culinary Advocacy	My friends often share their Ordering experience from Cloud Kitchens
		My friends often recommend to order from Cloud Kitchens

No.	Factorial Group	Cronbach's Alpha
1	Digital Culinary Experience	0.867
2	Influential Cloud Kitchen Acumen	0.805
3	Cloud Kitchens Ordering Ease	0.765
4	Socialized Culinary Advocacy	0.859





A Review on Drug-Drug Interaction and Pharmacokinetic Studies of Metformin and Furosemide with Other Drugs.

Rakshitha A¹ and Rashmi N.G^{2*}

¹PG Scholar, Faculty of Pharmacy, Sri Adichunchanagiri College of Pharmacy, Adichunchanagiri University, B.G Nagar, Karnataka, India.

²Associate Professor, Sri Adichunchanagiri College of Pharmacy, Adichunchanagiri University, B.G Nagar, Karnataka, India.

Received: 12 Jun 2024

Revised: 16 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

Rashmi N.G

Associate Professor,
Sri Adichunchanagiri College of Pharmacy,
Adichunchanagiri University,
B.G Nagar, Karnataka, India.
E. Mail: ng.rashmi85@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Drug-drug Interactions is a significant therapeutic concern, especially when administering many medications to patients with conditions like diabetes, hypertension and other cardiovascular diseases. In case of diabetes, metformin is more commonly prescribed drug, it is a derivative of biguanide used to treat type-2 diabetes. Similarly, in case of hypertension, furosemide is more commonly prescribed drug, it is a loop diuretic used in the treatment of edema state associated with chronic failure, hypertension, heart failure and liver cirrhosis. Hence, our aim is to know the DDI of metformin alone or with other drugs like, Chiglitazar, Pioglitazone, Ranolazine and telmisartan etc and DDI of furosemide alone or with other drugs like, Tolvaptan, spironolactone etc. In this review article, we were discussed the DDI that was performed by using a high-pressure liquid chromatography with different detectors and different species were used to study the drug-drug interaction of metformin with other drugs and furosemide with other drugs and different study design were applied to know the pharmacokinetic of the drugs

Keywords: Metformin, Furosemide, different chromatographic condition and species, drug-drug interaction and Pharmacokinetic studies.

INTRODUCTION

The evaluation of possible drug-drug interactions (DDIs) has grown in significance due to people's frequent concurrent usage of many medicines. Many people have employed physiologically based pharmacokinetic modelling techniques to quantitatively estimate drug-drug interactions (DDIs) between therapeutic medicines that are delivered



**Rakshitha and Rashmi**

together[1]. Pharmaceutical, toxicological, doping and clinical chemistry research all depend on the identification and measurement of substances in biological fluids. The concentration of medications in biological fluids and tissues is correlated with therapeutic efficacy[2]. In this article we are going to study about drug-drug interaction and pharmacokinetic studies on Metformin and Furosemide with other drugs. Type-2 diabetes mellitus is a chronic disease characterized by decrease in the pancreas' ability to release insulin, a resistance to insulin in peripheral tissues, or both[19]. A diabetic patient can maintain stable blood glucose level with the aid of antidiabetic medications, a nutritious diet, and regular exercise[17,21]. The first oral hypoglycaemic medication, metformin (MET) is a biguanide derivative, lowers plasma glucose levels by improving insulin sensitivity and reducing hepatic glucose release[3,26]. Metformin reduces insulin resistance and hepatic glucose production while increasing peripheral glucose absorbance and insulin sensitivity[4]. Additionally, it had a therapeutic impact on obesity metabolic disease by lowering free fatty acids (FFA). Sometimes, women with PCOS (polycystic ovary syndrome) depend on this medication for regular menstrual cycles and ovulation[5]. Metformin is also used to manage the cardiac hypertrophy, cardiac fibrosis, regulation of lipid[18], cancer and aging[24].

Diuresis-promoting medications are frequently used to treat edematous disorders, where an increase in urine flow reduces symptoms[6,23]. Several medical conditions are also treated by using diuretics which includes hypertension, liver cirrhosis, cardiac and renal failure[15,22,25]. Diuretics are drugs that cause the body's sodium that is bonded to anions and water to be excreted more in urine, which lowers the concentration of sodium in extracellular liquids. Effective diuretic like, Furosemide (FU) is commonly used to treat edema, fluid accumulation in body cavities[16], liver cirrhosis[29], hypertension, chronic renal failure, and congestive heart failure[20,23]. Due to FU's primary action in the nephron, where it prevents sodium from being tubularly reabsorbed on Henle's loop, the medication is frequently categorized as a loop diuretic. FU functions by preventing the co-transport of potassium, sodium, and chloride and also excretes bicarbonate, calcium, and magnesium ions[31]. By lowering the levels of other doping drugs in urine, intense and quick diuresis may also cover up the use of other substances[6].

Mechanism of action of Metformin

Metformin improves insulin sensitivity by minimizing peripheral glucose uptake and utilization, gluconeogenesis and intestinal glucose absorption[27,32]. Orally administered Metformin enter into the hepatocytes via the portal vein, where it is taken up by OCT1 plasma membrane transporters. Metformin inhibits the action of mitochondrial respiratory chain complex-1 in the cell, which raises AMP and decreases ATP levels[33]. Raised AMP levels trigger the activity of Adenosine Monophosphate-Activated Protein Kinase (AMPK), which inhibits the production of glucose through at least two mechanisms: The first is that elevated AMPK phosphorylates the transcription factors CBP and CRTC2, which inhibits the genes that produce glucose (also known as "gluconeogenic genes"); Secondly, elevated AMPK also inhibit the function of mGPD by increasing cytosolic NADH and promoting the conversion of pyruvate to lactate, while simultaneously reducing gluconeogenesis who also have other diseases that result in metabolic acidosis may accumulate lactate to harmful levels (lactic acidosis) or Patients who use metformin and have other health conditions that cause metabolic acidosis may accumulate lactate to fatal amounts[7].

Mechanism of action of Furosemide:

Furosemide is a loop diuretic having the ability to block the Na⁺, K⁺ and 2Cl⁻ symporters in the loop of Henle's thick ascending limb[28,30,34]. Consequently, raises the excretion of Ca²⁺, Mg²⁺, Na⁺, K⁺ and Cl⁻ in the urine[8]. Like other loop diuretics, Furosemide lowers the excretion of uric acid. It immediately dilates the blood arteries that helpful for treating acute pulmonary edema[35]. Vasodilation reduces the creation of endogenous natriuretic hormones that have the ability to constrict blood vessels and reduces the body's susceptibility to vasoconstrictors like noradrenalin and angiotensin II[7]. Few analytical techniques have been documented to date for the determination of metformin in biological fluids, either by itself or in combination with other antidiabetic medications, according to a review of the literature[3]. Nowadays people with diabetes also suffer from additional chronic conditions like nephropathy and hypertension and other heart related disease[40,41]. When metformin with other drugs is used together, there may be drug-drug interaction takes place. An improper dose modification might result in lactic acidosis, which can be fatal[9,36]. Normally, this has little or no effect on the buildup of lactate at therapeutic doses of metformin. Lactate



**Rakshitha and Rashmi**

absorption by the liver is decreased and lactic acidosis results if metformin accumulates as a result of either poor elimination or excessive ingestion[38]. Metformin-associated lactic acidosis (MALA) is caused by an imbalance between the amount of lactate produced and the decreased amount of lactate eliminated[37,39]. Metformin is removed by renal tubular secretion, several drugs that are cationic agents may compete with it for elimination. If taken concurrently, several of them—such as digoxin, quinidine, ranitidine, furosemide or nifedipine—may raise the level of metformin. Therefore, it's critical to keep on the lookout for any potential toxicity in individuals taking metformin in combination with these drugs[10].

METHODOLOGY**Drug-drug interaction of Metformin with other drugs:**

Show the Table 1: Pharmacokinetic analysis of metformin alone and in combination with other compounds using high-performance liquid chromatography (HPLC) with different species.

Drug-drug interaction of furosemide with other drugs:

Show the Table 2: Pharmacokinetic analysis of furosemide alone and in combination with other compounds using HPLC in various species to determine drug-drug interactions.

Outcomes of the Studies**Pharmacokinetic parameters of Metformin-Chiglitazar**

Show the Table 3: Pharmacokinetic parameters of Metformin, Chiglitazar alone and in combination[11].

A study on the drug-drug interaction between Chiglitazar and Metformin was carried out by Ling Yi and Hua Zhang. They found that pharmacokinetic characteristics like t_{max} and $t_{1/2}$ do not vary appreciably when the two medications are taken concurrently. Metformin raises the CL/F of Chiglitazar while lowering the C_{max} , AUC_{0-48h} , and $AUC_{0-\infty}$. Chiglitazar increases the AUC_{0-48h} , $AUC_{0-\infty}$, and C_{max} of metformin while decreasing the $t_{1/2}$ and CL/F[11].

Pharmacokinetic parameters of Metformin-Pioglitazone:

Show the Table 4: Pharmacokinetic parameters of Metformin, Pioglitazone alone and in combination[3].

In the study of drug-drug interaction between pioglitazone and metformin, Mohamed Saleh Elgawish and Sally Nasser observed that when metformin and pioglitazone were administered together, PGT (50 mg/kg) reduced the pharmacokinetic parameter such as C_{max} , t_{max} , AUC_{0-t} , $AUC_{0-\infty}$ and Kel of metformin but increased Vd/F and CL/F of metformin in contrast to MET alone. Metformin (50 mg/kg) also reduces AUC_{0-t} , $AUC_{0-\infty}$, Kel and C_{max} , but it raises PGT's t_{max} , $t_{1/2}$, Vd/F and Cl/F when given with PGT[3].

Pharmacokinetic parameters of Metformin- Ranolazine

Show the Table 5: Pharmacokinetic parameters of Metformin (1000mg BID), Ranolazine (500mg BID) alone and in combination[12]. The drug-drug interaction between metformin and ranolazine was studied by Julia Zack and Jolene Berg. When metformin (1000mg BID) was co-administered with ranolazine (500mg BID), ranolazine increased the pharmacokinetic parameters such as metformin's C_{max} , AUC_{0-t} , $t_{1/2}$, and CL/F, while metformin's t_{max} remained unchanged when compared to metformin alone[12].

Pharmacokinetic parameters of Furosemide-Tolvaptan

Show the Table 6: Pharmacokinetic parameters of Furosemide, Tolvaptan alone and in combination[8].

The drug-drug interaction between tolvaptan and furosemide was investigated by researchers Susan E. Shoaf and Steven L. Bramer. Researchers found that when provided in combination with tolvaptan (30 mg), furosemide (80 mg) had lower pharmacokinetic characteristics such as C_{max} , AUC_t , AUC_{∞} , %fe, u, and CLr, but increased $t_{1/2}$, CL/F but t_{max} remained as unchanged when compared to furosemide alone. While CL/F, CLr, and t_{max} did not alter when comparing Tolvaptan alone, pharmacokinetic variables such as C_{max} , AUC_t , AUC_{∞} , $t_{1/2}$, %fe, u were all higher[8].



**Rakshitha and Rashmi****Pharmacokinetic parameters of Furosemide in goat**

Show the Table 7: Pharmacokinetic parameter of Furosemide in goat with different routes like IV, IM and SC routes[13]. # - values is statistically different than that in IV and IM administration ($p < 0.05$) * - Values is statistically different than that in IM administration ($p < 0.05$)

Gul Cetin, Orhan Corum and other researchers used IV, IM and SC injections to study the pharmacokinetics of furosemide in goats. Furosemide given by IV, IM and SC were found to have a no statistically significant variations between the administration route. The $AUC_{0-\infty}$ was higher with IV and IM treatment than with SC. After IV, the Cl_T and V_{dss} were 0.30L/h/kg and 0.17L/kg respectively. Furosemide was administered by SC and IM, the corresponding C_{max} values were 11.19 and 6.49 μ g/ml at 0.23 and 0.39hrs respectively. Following IM and SC dosage, the bioavailability was 70.80% and 109.84% respectively. The goat's $t_{1/2\lambda_z}$, V_{dss} and Cl_T were 0.71h, 0.17L/kg and 0.30L/h/kg after receiving IV furosemide treatment respectively[13].

Pharmacokinetic parameters of Furosemide in hypertensive parturient women under caesarean section:

Show the Table 8: Furosemide pharmacokinetic parameters in hypertensive parturient women receiving caesarean sections[14]. The maternal-fetal pharmacokinetic of furosemide were investigated by Paluo Vinicius, Fernanda and other researchers in hypertensive parturient women (n=12) undergoing caesarean delivery after a single oral dose of 40mg. The researchers observed that the median and interquartile range of the furosemide pharmacokinetic parameters, the following outcomes were obtained: AUC_{0-12h} -1366ng/ml, $AUC_{0-\infty}$ -1580ng/ml, V_d/F -82.8L, Cl/F -25.3L/h, Cl_{LR} -2.50L/h and Cl_{NR} -22.7L/h. Furosemide concentration in 4 samples of amniotic fluid and 8 samples of umbilical cord were measured that shows a furosemide transplacental transfer of 11.0ng/ml and 45.8ng/ml respectively[14].

CONCLUSION

From the table-3 we observed that, Metformin and Chiglitazar showed no clinically significant variations in the pharmacokinetics of the two drugs, even though the combination treatment resulted in a small reduction in CHI exposure. Furthermore, each group of subjects exhibited good tolerance. The impact of PGT on MET's AUC , t_{max} , C_{max} , $t_{1/2}$ and Cl/F at a somewhat elevated concentration that is showed in table-4. PGT's role is an OCT inhibitor may have an impact on MET absorption and clearance. Consequently, to regulate plasma glucose, improve medication management, and prevent negative side effects, close patient observation and dose modification of the PGT and MET combination are advised. We noticed from Table-5 that, ranolazine co-administration raises metformin exposure most likely by ranolazine induced reduction of metformin renal excretion. At the lower ranolazine dose of 500mg BID, the dose-dependent increase in metformin exposure caused by ranolazine is not very significant. When metformin 1000mg BID and ranolazine 500mg BID were given together, the T2DM individuals tolerated the drugs well. Furosemide co-administration did not seem to have a clinically meaningful impact on the pharmacokinetics of tolvaptan as indicated in Table-6. When taken in combination with tolvaptan, the pharmacokinetics of furosemide did not seem to be considerably impacted. The aquaretic action of tolvaptan was not considerably impacted by furosemide. Plasma renin activity did not increase when tolvaptan was taken alone. At the prescribed dosage, tolvaptan was safe and well tolerated when used either alone or in conjunction with furosemide.

The delivery method had an impact on furosemide's plasma levels and bioavailability. As shown by the table-7, the duration of furosemide concentration above the minimum effective concentration ($\geq 1\mu$ g/ml) for IV<IM and SC injection is similar, despite the observed variances making them appropriate routes for diuresis in goat. One possible way to reduce the risk of furosemide adverse effects related to the quick IV bolus injection is to use the IV route for a gradual injection or continuous infusion. When compared to nonpregnant healthy patients to hypertensive parturient women undergoing caesarean section appear to have a Cl/F and non-renal Cl_{NR} of furosemide on the basis of table-8. Amniotic fluid concentrations are lower than those in the umbilical vein, and furosemide is transferred through the



**Rakshitha and Rashmi**

placenta at a rate of about 40%. On a clinical basis, these findings imply that UGT isoenzyme substrates may have greater clearance during pregnancy and may need to have their dosage modified in this population.

REFERENCES

1. Kwon M, Jeon JH, Choi MK, Song IS. The development and validation of a novel “dual cocktail” probe for cytochrome P450s and transporter functions to evaluate pharmacokinetic drug-drug and herb-drug interactions. *Pharmaceutics*. 2020 Sep 30;12(10):938.
2. Soltani S, Jouyban A. A validated micellar LC method for simultaneous determination of furosemide, metoprolol and verapamil in human plasma. *Bioanalysis*. 2012 Jan;4(1):41-8.
3. Elgawish MS, Nasser S, Salama I, Abbas AM, Mostafa SM. Liquid chromatography tandem mass spectrometry for the simultaneous determination of metformin and pioglitazone in rat plasma: Application to pharmacokinetic and drug-drug interaction studies. *Journal of Chromatography B*. 2019 Aug 15;1124:47-57.
4. Borse SP, Upadhyay D, Shama V, Singh D, Nivsarkar M. Effect of Diabetes mellitus on pharmacokinetics of metformin and telmisartan by using novel HPLC-UV method: A drug-disease interaction study.
5. Patra R, Kollati Y, NS SK, Dirisala VR. Valsartan in combination with metformin and gliclazide in diabetic rat model using developed RP-HPLC method. *Future Journal of Pharmaceutical Sciences*. 2021 Dec;7:1-0.
6. Bosch ME, Sánchez AR, Rojas FS, Ojeda CB. Analytical determination of furosemide: the last researches. *Int J Pharm Biol Sci*. 2013;3(4):168-81.
7. PubChem (no date) National Center for Biotechnology Information. PubChem Compound Database. Available at: <https://pubchem.ncbi.nlm.nih.gov/> (Accessed: 02 April 2024).
8. Shoaf SE, Bramer SL, Bricmont P, Zimmer CA. Pharmacokinetic and pharmacodynamic interaction between tolvaptan, a non-peptide AVP antagonist, and furosemide or hydrochlorothiazide. *Journal of cardiovascular pharmacology*. 2007 Aug 1;50(2):213-22.
9. Metformin Interactions checker (no date) Drugs.com. Available at: <https://www.drugs.com/drug-interactions/metformin.html> (Accessed: 02 April 2024).
10. Rajasurya V, Anjum H, Surani S. Metformin use and metformin-associated lactic acidosis in intensive care unit patients with diabetes. *Cureus*. 2019 May 23;11(5).
11. Yi L, Zhang H, Zhang JW, You XM, Ning ZQ, Yu J, et al. Study on drug-drug interactions between chiglitazar, a novel PPAR pan-agonist, and metformin hydrochloride in healthy subjects. *Clinical Pharmacology in Drug Development*. 2019 Oct;8(7):934-41.
12. Zack J, Berg J, Juan A, Pannacciulli N, Allard M, Gottwald M, et al. Pharmacokinetic drug–drug interaction study of ranolazine and metformin in subjects with type 2 diabetes mellitus. *Clinical pharmacology in drug development*. 2015 Mar;4(2):121-9.
13. Cetin G, Corum O, Durna Corum D, Atik O, Turk E, Tekeli IO, et al. Pharmacokinetics of furosemide in goats following intravenous, intramuscular, and subcutaneous administrations. *Journal of Veterinary Pharmacology and Therapeutics*. 2021 Nov;44(6):961-6.
14. Gonçalves PV, Moreira FD, Benzi JR, Duarte G, Lanchote VL. A Pilot Study of the Maternal-Fetal Pharmacokinetics of Furosemide in Plasma, Urine, and Amniotic Fluid of Hypertensive Parturient Women Under Cesarean Section. *The Journal of Clinical Pharmacology*. 2020 Dec;60(12):1655-61.
15. Eid PS, Ibrahim DA, Zayan AH, Elrahman MM, Shehata MA, Kandil H, et al. Comparative effects of furosemide and other diuretics in the treatment of heart failure: A systematic review and combined meta-analysis of randomized controlled trials. *Heart Failure Reviews*. 2020 Aug 11;26(1):127–36.
16. Durna Corum D, Corum O, Atik O, Cetin G, Zhunushova A, Uney K. Pharmacokinetics and bioavailability of furosemide in sheep. *Journal of Veterinary Pharmacology and Therapeutics*. 2020 Dec 12;44(4):657–62.
17. Srikanth V, Sinclair AJ, Hill-Briggs F, Moran C, Biessels GJ. Type 2 diabetes and cognitive dysfunction—towards effective management of both comorbidities. *The Lancet Diabetes & Endocrinology*. 2020 Jun;8(6):535–45.
18. Foretz M, Guigas B, Bertrand L, Pollak M, Viollet B. Metformin: From mechanisms of action to therapies. *Cell Metabolism*. 2014 Dec;20(6):953–66.



**Rakshitha and Rashmi**

19. Mohamed A-MI, Mohamed FA-F, Ahmed S, Mohamed YA. An efficient hydrophilic interaction liquid chromatographic method for the simultaneous determination of metformin and pioglitazone using high-purity silica column. *Journal of Chromatography B*. 2015 Aug;997:16–22.
20. Kodati D, Yellu N. Population pharmacokinetic modeling of furosemide in patients with hypertension and fluid overload conditions. *Pharmacological Reports*. 2017 Jun;69(3):492–6. Kaul K, Tarr JM, Ahmad SI, Kohner EM, Chibber R. Introduction to diabetes mellitus. *Diabetes: an old disease, a new insight*. 2013:1-1.
21. Kaul K, Tarr JM, Ahmad SI, Kohner EM, Chibber R. Introduction to diabetes mellitus. *Diabetes: an old disease, a new insight*. 2013:1-1.
22. El-Saharty YS. Simultaneous high-performance liquid chromatographic assay of furosemide and propranolol hcl and its application in a pharmacokinetic study. *Journal of Pharmaceutical and Biomedical Analysis*. 2003 Nov;33(4):699–709.
23. Wasfi IA, Wajid SA, Agha BA, Kamel AM, Al Biriki NA, Al Neaimi KM, et al. Quantification of furosemide in camel plasma by high resolution mass spectrometry, application on pharmacokinetics. *Biomedical Chromatography*. 2016 Dec 26;31(6).
24. Lv Z, Guo Y. Metformin and its benefits for various diseases. *Frontiers in Endocrinology*. 2020 Apr 16;11.
25. Mishra, S. (2016) 'Diuretics in primary hypertension – reloaded', *Indian Heart Journal*, 68(5), pp. 720–723.
26. Jeong YS, Jusko WJ. Meta-assessment of metformin absorption and disposition pharmacokinetics in nine species. *Pharmaceuticals*. 2021 Jun 7;14(6):545.
27. Foretz M, Guigas B, Bertrand L, Pollak M, Viollet B. Metformin: from mechanisms of action to therapies. *Cell metabolism*. 2014 Dec 2;20(6):953-66.
28. Koh SK, Jeong JW, Choi SI, Kim RM, Koo TS, Cho KH, et al. Pharmacokinetics and diuretic effect of furosemide after single intravenous, oral tablet, and newly developed oral disintegrating film administration in healthy beagle dogs. *BMC Veterinary Research*. 2021 Dec;17:1-1.
29. Romantsik O, Barco S, Bruschettoni M, Tripodi G, Ley D, Cangemi G. A LC–MS/MS method for the quantification of caffeine, betamethasone, clonidine and furosemide in cerebrospinal fluid of preterm infants. *Journal of Pharmaceutical and Biomedical Analysis*. 2020 Feb 5;179:112996.
30. Chavan RR, Bhinge SD, Bhutkar MA, Randive DS, Salunkhe VR. Method development and validation of spectrophotometric and RP-HPLC methods for simultaneous estimation of spironolactone and furosemide in bulk and combined tablet dosage forms. *Anal. Sci. Technol*. 2021 Oct;3:212-24.
31. Yallamalli IM, Mannam R, Yellamelli R. Drug in adhesive transdermal system of furosemide: in vitro in vivo evaluation. *Int j app pharm*. 2023;15(4):106-13.
32. Wang GS, Hoyte C. Review of biguanide (metformin) toxicity. *Journal of intensive care medicine*. 2019 Nov;34(11-12):863-76.
33. Rena G, Hardie DG, Pearson ER. The mechanisms of action of metformin. *Diabetologia*. 2017 Sep;60(9):1577-85.
34. Karunakaran A, Sudharsan SI, Jayaprakash R, Vekatachalam S, Raju SK, Elampulakkadu A. Analytical method development and validation for the estimation of Furosemide an anti-diuretic in Furosemide injection diluted with normal saline in presence of impurities by RP-HPLC. *Brazilian Journal of Biological Sciences*. 2021 Apr 30;8(18):35-56.
35. Malha L, Mann SJ. Loop diuretics in the treatment of hypertension. *Current hypertension reports*. 2016 Apr;18(4):27.
36. Mariano F, Biancone L. Metformin, chronic nephropathy and lactic acidosis: a multi-faceted issue for the nephrologist. *Journal of Nephrology*. 2021 Aug;34(4):1127-35.
37. Lalau JD, Kajbaf F, Protti A, Christensen MM, De Broe ME, Wiernsperger N. Metformin-associated lactic acidosis (MALA): Moving towards a new paradigm. *Diabetes, Obesity and Metabolism*. 2017 Nov;19(11):1502-12.
38. Inzucchi SE, Lipska KJ, Mayo H, Bailey CJ, McGuire DK. Metformin in patients with type 2 diabetes and kidney disease: a systematic review. *Jama*. 2014 Dec 24;312(24):2668-75.
39. DeFronzo R, Fleming GA, Chen K, Bicsak TA. Metformin-associated lactic acidosis: Current perspectives on causes and risk. *Metabolism*. 2016 Feb 1;65(2):20-9.
40. Artasensi A, Pedretti A, Vistoli G, Fumagalli L. Type 2 diabetes mellitus: a review of multi-target drugs. *Molecules*. 2020 Apr 23;25(8):1987.





Rakshitha and Rashmi

41. Vodošek Hojs N, Bevc S, Ekart R, Piko N, Petreski T, Hojs R. Mineralocorticoid receptor antagonists in diabetic kidney disease. *Pharmaceuticals*. 2021

Table 1: Pharmacokinetic analysis of metformin alone and in combination with other compounds using high-performance liquid chromatography (HPLC) with different species.

Drug/Compounds	Chromatographic condition	Species	Dru-drug Interaction and Pharmacokinetic Study	Ref
Metformin - Chiglitazar (CHI)	RP-HPLC system coupling with tandem mass spectrometry having electrospray ionization. Mobile phase containing a mixture of acetonitrile and water containing 0.4% formic acid in a ratio of 95:5.	Both healthy male and female individuals with a BMI ranging from 19-26kg/m ² who are least 18yrs of age.	self-controlled, three-period crossover, open-label, and randomized. In all, 15 subjects were divided into three groups. Group-I subject received MET 1000mg, CHI 48mg was received by group-II and the combination of MET (1000mg)+CHI(48mg) was received by group-III. Blood samples were withdrawn at 0, 0.5, 1, 2, 4, 6, 8, 12, 24, 36 and 48h, then the samples were centrifuged for 10min at 3500rpm. pharmacokinetic parameters (PKs) were measured by using a mixed-effect model on log-transformed pharmacokinetic profile.	[11]
Metformin-Pioglitazone (PGT)	Agilent Eclipse Plus C18 column was maintained at room temperature of 25±2°C. The combination of acetonitrile and 6mM ammonium formate with a pH of 4.5 with 0.1% formic acid made up the mobile phase. 0.5ml/min was the flow rate maintained. Triple-quadruple LC/tandem mass spectrometry was used for the study.	Male Wistar albino rats, 8wk old, weighing 200-220g.	Three groups of three rats each were selected by random selection. Rats in Group-I received MET (50mg/kg, orally(po)); Group-II received PGT (50mg/kg); Group-III received MET (50mg/kg, po) + PGT (50mg/kg, po). After 0.5, 1, 2, 4, 6, 8, 12, and 24h, blood samples were collected and centrifuged for 10mins at 10000rpm. From the experiment's data, C _{max} and t _{max} were determined. Regression analysis was used to estimate the Kel based on the slope and 0.693/Kel was utilized for calculating t _{1/2} . PKSolver was also used to compute the MRT, Vd/F and CL/F.	[3]





Rakshitha and Rashmi

Metformin-Ranolazine	LC-MS/MS system, a Phenomenex Synergi Hydro-R column (2x50mm) and the mobile phase was composed of acetonitrile and water with 0.1% formic acid with a pH of 3.	Both male and female patients, ages between 30-65, with a history of type 2 diabetes and BMI of 25–40 kg/m ² .	Open-labelled single cohort, multiple doses, 2-period fixed-sequence method. Participants were taken 2-fixed sequence treatment fasting 5days each: from days 1-5, metformin 1000mg BID (twice daily) and from days 6-10, metformin (1000mg, BID) and ranolazine (500mg, BID). Day 10, a single morning dose of 500 mg of ranolazine was given. Samples were taken in the morning at 0.5, 1, 2, 4, 6, 8, 10, and 12h after the study medication was administered. Phoenix WinNonlin v6.2.1 was used to calculate the PKs parameter.	[12]
----------------------	---	---	---	------

Table 2: Pharmacokinetic analysis of furosemide alone and in combination with other compounds using HPLC in various species to determine drug-drug interactions.

Drug/Compound	Chromatographic condition	Species	Drug-drug interaction and Pharmacokinetic studies	Ref.
Furosemide-Tolvaptan	RP-HPLC system with UV-detector was used.	12 white men, age from 18-29yr and with body weights of ranging 69-101kg, within 15% of the optimal body wt, were enrolled.	Three-period cross-over, single-centre randomized open-label parallel arm design. Six participants allocated to each of the two treatment groups. 30mg of tolvaptan, 80mg of furosemide and tolvaptan 30mg + furosemide 80mg were given to the subject Arm-1. 30mg of tolvaptan, 100mg of HCTZ and 30mg of tolvaptan + 100mg of HCTZ were given to the subjects in Arm-2. There was a 48h washout period between doses. Blood samples were collected at 0, 0.5, 1, 2, 3, 4, 6, 8, 12, 16 and 24hrs. 5ml of blood were drawn in order to measure the tolvaptan and furosemide levels. Samples were centrifuged for 10min at 2500rpm. Both C _{max} and t _{max} were calculated from the data on plasma concentration-time	[8]





Rakshitha and Rashmi

			curve. WinNonlin Professional software was used for PKs calculations.	
Furosemide	HPLC-UV system with C18 column (250x4.6mm; 5µm, Phenomenex®). The temperature of the auto-sampler and column were kept at 24°C and 40°C respectively. Methanol (35%) and 0.01M sodium acetate (pH of 5.65%) were act as a mobile phase with a flow rate of 1ml/min.	Six female goats, 45 ± 5 kg in body weight and 2.4 ± 0.4 year old.	Three periods, crossover design, separated by a 15-day washout interval. The dosage of furosemide was 2.5mg/kg. Furosemide injections in single IV, IM and SC were administered to each goat, followed by a 15-days washout period. First, two goats received furosemide IV, two went IM, and two went SC. Blood samples were drawn from each goat's right jugular vein at 0, 5, 10, 15, 20, 25, 30 and 45min as well as 1, 1.5, 2, 3, 4, 5 and 6h after the medicine was administered and centrifuging for 10min at 4000rpm. A non-compartmental technique was used to examine the furosemide concentrations in plasma using the WinNonlin 6.1.0.173 software application.	[13]
Furosemide	RP-HPLC system with fluorescence detector that carried out at 405 nm(emission) and 345 nm(excitation). C18 column (125 x 4mm, 5µm). The 80:20 mixture acetonitrile and 0.25M acetate buffer (pH 4.0) were used as the mobile phase.	12 hypertensives parturient women undergoing cesarean sections were involved with gestational ages ranging from 36.0-40.4 wk.	Hypertensive parturient women who were on methyldopa (250mg/8hrs) and/or pindolol (10mg/12hrs) for the purpose of performing a cesarean section and obtaining biological samples. An IV catheter was used to draw 5ml of blood at 0, 0.5, 1, 2, 4, 6, 8 and 12 hrs following the administration of furosemide (40mg) through oral route prior to delivery. At birth, blood samples from the mother and the umbilical cord were also taken in order to evaluate the transplacental transfer. After centrifuging blood samples at 2000rpm for 15min. Up to 12h after the medicine was administered,	[14]





Rakshitha and Rashmi

			the entire volume of urine was collected at around 3h intervals. A sample of amniotic fluid was taken during the caesarean section. Phoenix WinNonlin, version 6.3, was used to assess the PKs profile of furosemide.	
--	--	--	---	--

Table 3: Pharmacokinetic parameters of Metformin, Chiglitazar alone and in combination[11].

Drugs/ Compounds	C _{max} (ng/ml)	t _{max} (h)	t _{1/2} (h)	AUC _{0-t} (ng*h/ml)	AUC _{0-∞} (ng*h/ml)	CL/F (L/h)
Metformin alone	1790	3.0	8.09	12570	12710	80.42
Chiglitazar alone	1620	4.0	10.75	12540	12910	3.88
Metformin + Chiglitazar	1820	3.0	7.89	13190	13350	76.99
	1420	4.0	10.12	12130	12470	4.02

Table 4: Pharmacokinetic parameters of Metformin, Pioglitazone alone and in combination[3].

Drugs/ Compounds	C _{max} (ng/ml)	t _{max} (h)	AUC _{0-t} (ng*h/ml)	AUC _{0-∞} (ng*h/ml)	t _{1/2} (h)	Kel (1/h)	Vd/F (L)	CL/F (L/h)
Metformin alone	2310 ± 367	2.00 ± 0.43	10,830 ± 976	10,885 ± 876	2.84 ± 1.2	0.24 ± 0.134	1.7 ± 0.36	0.93 ± 0.26
Pioglitazone Alone	1920 ± 598	1.00 ± 0.3	18,214 ± 2189	22,967 ± 1762	10.81 ± 2.3	0.06 ± 0.02	5.5 ± 0.41	0.5 ± 0.2
Metformin + Pioglitazone	167 ± 88.2	0.50 ± 0.15	507 ± 158	563 ± 217	13.14 ± 2.1	0.05 ± 0.01	55.3 ± 0.88	20.5 ± 0.91
	64 ± 12.3	2.00 ± 0.75	207 ± 98	252 ± 101	16.68 ± 1.4	0.04 ± 0.01	70.5 ± 0.82	56.5 ± 0.01

Table 5: Pharmacokinetic parameters of Metformin (1000mg BID), Ranolazine (500mg BID) alone and in combination[12].

Drugs/ Compounds	C _{max} Mean (SD)	AUC _{tau} Mean (SD)	t _{max} Median (min, max)	t _{1/2} Mean (SD)	CL/F, Mean (SD)
Metformin 1000mg BID	2497	12933	1.5	4.0	83.2
Ranolazine 500mg BID	1180	8722	3.0	3.6	-
Metformin 1000mg BID + Ranolazine 500mg BID	3059	17920	1.5	4.4	60.8

Table 6: Pharmacokinetic parameters of Furosemide, Tolvaptan alone and in combination[8].

Drug/ Compound	C _{max} (ng/ml)	t _{max} (h)	AUC _t (µg*h/ml)	t _{1/2,z} (h)	AUC _∞ (µg*h/ml)	CL/F (ml/min/kg)	%fe,u	CLr (ml/min/kg)
Furosemide (80mg)	2.90 (1.00)	1.5 (0.5–3)	7.06 (1.62)	2.6 (1.3)	7.18 (1.64)	2.52 (0.46)	43.5 (10.3)	1.12 (0.41)
Tolvaptan	277	2	1.57	5.7	1.69	4.03	0.14	0.006





Rakshitha and Rashmi

(30mg)	(22)	(1.5–3)	(0.26)	(3.0)	(0.35)	(0.82)	(0.05)	(0.002)
Furosemide (80mg)	2.61 (1.31)	1.5 (1–4)	6.40 (1.55)	3.9 (2.5)	6.55 (1.55)	2.76 (0.50)	40.2 (11.8)	1.09 (0.28)
+ Tolvaptan (30mg)	327 (82)	2 (1–4)	1.94 (0.31)	6.1 (3.1)	2.08 (0.39)	3.27 (0.75)	0.15 (0.05)	0.005 (0.002)

Table 7: Pharmacokinetic parameter of Furosemide in goat with different routes likes IV, IM and SC routes[13]. #- values is statistically different than that in IV and IM administration ($p < 0.05$) *- Values is statistically different than that in IM administration ($p < 0.05$)

parameters	SC	IV	IM
C_{max}	6.49*	-	11.19
t_{max}	0.39*	-	0.23
$t_{1/2\lambda z}$ (h)	0.70	0.71	0.69
AUC_{0-4} ($\mu\text{g}\cdot\text{h}/\text{ml}$)	5.88#	8.38	9.16
$AUC_{0-\infty}$ ($\mu\text{g}\cdot\text{h}/\text{ml}$)	5.99#	8.46	9.29
AUC_{extrap} (%)	1.81	0.93	1.37
MRT (h)	0.45*	-	0.30
Cl_r (L/h/kg)	-	0.30	-
V_{dss} (L/kg)	-	0.17	-

Table 8: Furosemide pharmacokinetic parameters in hypertensive parturient women receiving caesarean sections[14].

Parameter	Values
C_{max} (ng/ml)	403
t_{max} (h)	2.00
AUC_{0-12h} (ng*h/ml)	1366
$AUC_{0-\infty}$ (ng*h/ml)	1580
$t_{1/2}$ (h)	2.50
Kel (1/h)	0.27
Vd/F (L)	82.8
CL/F (L/h)	25.3
CL_{NR} (L/h)	22.7
CL_R (L/h)	2.50
Time between drug administration and delivery (h)	2.50
Umbilical vein, n=8 (ng/ml)	45.8
Amniotic fluid, n=4 (ng/ml)	11.0
Maternal plasma, n=8 (ng/ml)	140
Umbilical vein/maternal plasma ratio (n=8)	0.43





Rakshitha and Rashmi

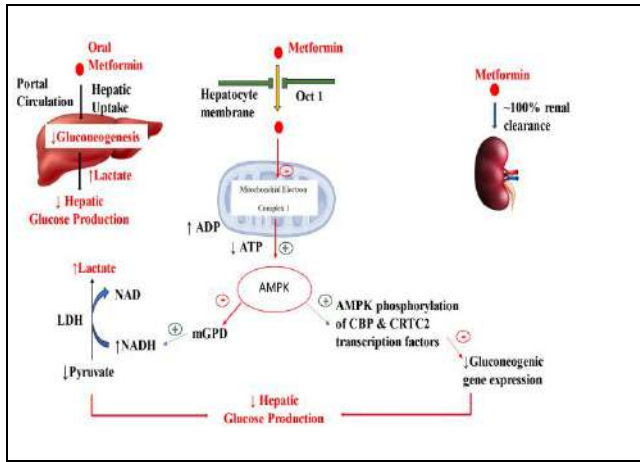


Figure 1: Mechanism of action metformin.

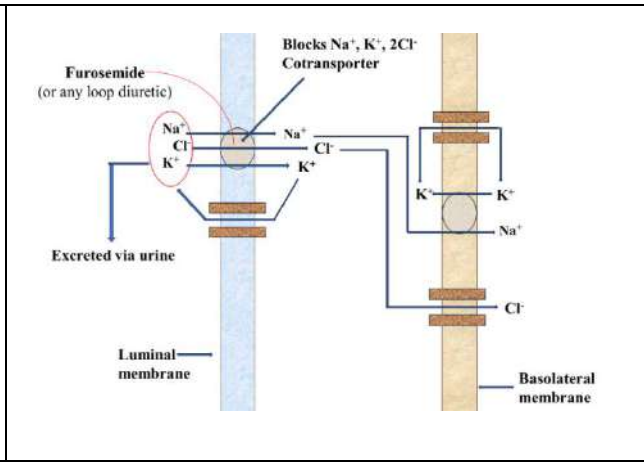


Figure 2: Mechanism of action of Furosemide.





Physico – Chemical and Phytochemical Analysis of Siddha Polyherbal Formulation – Megarajanga Kirutham

S.P.Kopperunde^{1*}, S.Santhanakitt², M.N.Parandhaman³, V.Sathiya⁴ and M.Thiruvalluvar⁵

¹Lecturer, Department of Varma Pura Sirappu Maruthuvam, JSA Medical College for Siddha and Research Centre, Ulundurpet, (Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai) Tamil Nadu, India.

²Lecturer, Department of Noi Naadal, Sudha Saseendran Siddha Medical College and Hospital, Kaliakkavilai, Kanniyakumari(Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai) Tamil Nadu, India.

³Lecturer, Department of Aruvai Thol Maruthuvam, JSA Medical College for Siddha and Research Centre, Ulundurpet , (Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai) Tamil Nadu, India.

⁴Lecturer, Department of Udal Koorugal, JSA Medical College for Siddha and Research Centre, Ulundurpet, (Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai) Tamil Nadu, India.

⁵Lecturer, Department of Noi Naadal, JSA Medical College for Siddha and Research Centre, Ulundurpet, (Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai) Tamil Nadu, India.

Received: 10 Jun 2024

Revised: 20 Aug 2024

Accepted: 24 Oct 2024

*Address for Correspondence

S.P.Kopperunde¹

Lecturer, Department of Varma Pura Sirappu Maruthuvam,
JSA Medical College for Siddha and Research Centre, Ulundurpet,
(Affiliated to The Tamil Nadu Dr.MGR Medical University, Chennai)
Tamil Nadu, India.

E.Mail: dr.kopperundevipitchai@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Standardization is the main part of drug developing process to determine the quality of medicine. The quality of Siddha formulations must be determined before it is used in preclinical and clinical studies. Megarajanga Kirutham (MRK) is the polyherbal Siddha formulation indicated for menorrhagia, dysuria, leucorrhoea, anemia and emaciation. It was processed as per the procedure mentioned in classical Siddha text "Sigicha rathna deepam". To understand the quality and changes that occurred during the preparation, Megarajanga Ghritham was analyzed by using modern techniques. After the analysis, it was found that the pH, acid value, Saponification value and the Iodine value were 6, 1.5, 205 and 101 respectively. Phytochemical screening affirmed the presence of flavonoids, steroids, sugars, triterpenoids, coumarins and phenols in the extract. This preliminary study helped in authenticating and ensuring the quality of Megarajanga Kirutham.

Keywords: Megarajanga Kirutham, standardization, physico chemical, saponification value, iodine value, Siddha.





Kopperundevi et al.,

INTRODUCTION

The Siddha medical system is an ancient medical science that helps people by treating a variety of chronic illnesses [1]. Herbs, metals, minerals, and other materials have long been used by Siddha medicine to create its medical concoctions [2]. Traditional wisdom believes that herbal treatments are safe, and an increasing number of people are utilizing them worldwide [3]. However, there are still no appropriate standards techniques for determining its effectiveness, consistency, and quality [4]. The importance of qualitative and quantitative methods for sample characterization, biomarker and/or chemical marker detection, and fingerprint profile analysis is emphasized by the World Health Organization (WHO) [5]. Nowadays, individuals also require further scientific validation about the pharmacokinetics, safety, etc., of conventional medications [6]. Drug evaluation is carried out to confirm the substance's identity, confirm its quality and purity, and detect any instances of adulteration [7].

A parameter that represents the purity and quality of a material is called a standard. Standardization refers to the procedure of creating a norm for a certain drug [8]. In the Siddha medical system, Megarajanga Kirutham is a well-known polyherbal formulation that is frequently used for many health conditions, according to the Siddha canonical literature "*Sigicha Rathna Deepam*". It has the indication for menorrhagia, dysuria, leucorrhoea, anemia and emaciation [9]. Herbal medicines are in increasing demand for primary healthcare worldwide due to their increased safety margins and cost effectiveness [10]. However, as of now, there is lack of analytical standards for Megarajanga Kirutham. Hence, this formulation was evaluated for physico-chemical and phytochemical analysis in order to establish specific quality requirements for the formulation.

MATERIALS AND METHODS

Collection of raw materials

All of the raw drugs needed to prepare the test drug were bought from a reputable raw drug seller, Tambaram and the Medicinal botanist in National Institute of Siddha, Chennai, provided their authentication. After appropriate purification, the trial medication was manufactured in the Gunapadam Lab, National Institute of Siddha, Chennai.

Purification of raw drugs:

All the herbal drugs are purified as per the classical Siddha text Gunapadam Mooligai Vaguppu [11].

Method of preparation

The ingredients of megarajanga Kirutham was mentioned in the Table 1. The drug was prepared as per the Siddha classical text "*Sigicha rathna deepam*". 350grams of aththi pattai (*Ficus racemosa*), Naaval pattai (*Syzygium cumini*), othiyampattai (*Lannea coromandelica*) were crushed and to prepare the kudineer (decoction) 8 liters of water was added and reduced to 1 liter. Then juices of karumbu juice (*Saccharum officinarum*), nellikai juice (*Emblica officinalis*), lemon juice (*Citrus limon*), gingelly oil (*Sesamum indicum*) and cow's ghee were added into the decoction and boiled well. Each 26.25 grams powders of thalisapathiri (*Abies spectabilis*), elam (*Elettaria cardamomum*), kirambu(*Syzygium aromaticum*), jadhikkai (*Myristica fragrans*), chittrarathai (*Alpinia officinarum*), athimathuram (*Glycyrrhiza glabra*) and nilappanai kizhangu (*Curculigo orchioides*) were ground with the milk of three coconuts (*Cocos nucifera*). This kalkam was added to the decoction mixture and boiled until the ghee consistency.

Quality evaluation of Megarajanga Kirutham

The formulation was initially analyzed for organoleptic parameters like color, odor, taste, texture, etc. Various physico-chemical parameters which are specific for Kirutham preparations such as loss on drying, pH value, refractive index, acid value, saponification value, iodine value and rancidity were evaluated [12,13].



**Kopperundevi et al.,****Percentage loss on drying**

10gm of test drug (weight equivalent to oil) was accurately weighed in evaporating dish. The sample was dried at 105°C for 5 hours and then weighed.

Percentage loss in drying = Loss of weight of sample/ Wt of the sample X 100

Determination of Total Ash

3 gm of test drug (weight equivalent to oil) was accurately weighed in silica dish and incinerated at the furnace a temperature 400°C until it turns white in color which indicates absence of carbon. Percentage of total ash will be calculated with reference to the weight of air-dried drug.

Total Ash = Weight of Ash/Wt of the Crude drug taken X 100

Determination of pH

Megarajanga Kirutham being oily in nature the direct litmus evaluation method was adopted to check the pH of the sample.

Determination of Iodine value

About 20 gm of oil was transferred into Iodine flask. To which 10 ml of chloroform was added and warmed slightly and cooled for 10 minutes. Followed by this about 25 ml of Wiji's solution was added in the same flask and shaken well. The flask was allowed to stand for 30 minutes and refrigerated for an hour. About 10 ml of KI solution was added to this and titrated against 0.1 N Sodium thiosulphate solutions until the appearance of yellow color. 1 ml of starch indicator was added and again titrated against the sodium thiosulphate solution from the burette. Disappearance of blue color indicates end point. Repeat the above procedure without taking sample and note the corresponding reading for blank titration.

Determination of saponification value

About 2 gm (weight equivalent to oil) of test sample was transferred into the round bottomed flask. To this about 20 ml of 0.5 N alcoholic KOH solutions was added to the round bottomed flask. Repeat the same procedure without taking the sample for blank titration. Reflux both sample and blank round bottomed flasks for 1 hour. After reflux, allow both the round bottomed flasks to cool. Titrate the samples using 0.5 N HCl with phenolphthalein indicator. The disappearance of pink indicates the end point.

Rancidity Test

1 ml of melted fat was mixed with 1ml of conc. HCl and 1 ml of 1% solution of phloroglucinol in diethyl ether and then mixed thoroughly with the fat acid mixture. A pink color indicates that the fat is slightly oxidized while a red color indicates that the fat is definitely oxidized.

Acid Value

2-10gm of Kirutham weighed in a conical flask. Added 50 ml of acid free alcohol-ether mixture (25 +25ml) previously neutralized with the 0.1M potassium hydroxide solution and shaken well. Added One ml of Phenolphthalein solution and titrated against 0.1M Potassium hydroxide solution. End point is the appearance of pale pink color. The experiment repeated twice to get concordant values.

Phytochemical analysis of formulation:

Megarajanga Kirutham (MRK) was extracted with hexane and the extract was subjected to the following analysis [14].

Test for Alkaloid- Mayer's reagent

To the test drug about 2ml of Mayer's reagent was added and was observed for the presence of alkaloids. Appearance of dull white precipitate indicates the presence of alkaloids.



**Kopperundevi et al.,****Test for flavonoid**

To 0.1ml of the test sample about 5 ml of dilute ammonia solution were been added followed by addition of few drops of conc. Sulphuric acid. Appearance of yellow color indicates the presence of Flavonoids.

Test for Glycosides -Borntrager's Test

MRK is hydrolyzed with concentrated hydrochloric acid for 2 hours on a water bath, filtered and the hydrolysate is subjected to the following tests. To 2 ml of filtered hydrolysate, 3ml of chloroform is added and shaken, chloroform layer is separated and 10% ammonia solution is added to it. Pink color indicates presence of glycosides.

Test for Triterpenoids

To the test solution 2ml chloroform was added with few drops of concentrated Sulphuric acid (3ml) at the side of the test tube. An interface with a reddish brown coloration is formed if terpenoids constituent is present.

Test for Steroids - Salkowski test

To the test solution 2ml of chloroform was added with few drops of conc. Sulphuric acid (3ml), and shaken well. The upper layer in the test tube was turns into red and sulphuric acid layer showed yellow with green fluorescence. It showed the presence of steroids.

Test for Carbohydrates - Benedict's test

To 0.5 ml of test drug about 0.5 ml of Benedict's reagent is added. The mixture is heated on a boiling water bath for 2 minutes. A characteristic colored precipitate indicates the presence of sugar.

Test for Phenol- Lead acetate test:

The test sample is dissolved in of distilled water and to this 3 ml of 10% lead acetate solution is added. A bulky white precipitate indicates the presence of phenolic compounds.

Test for tannins

About 0.5ml of test sample is boiled in 20ml of distilled water in a test tube and then filtered. The filtration method used here is the normal method, which includes a conical flask and filter paper. The 0.1% FeCl₃ is added to the filtered samples and observed for brownish green or a blue black coloration, which shows the presence of tannins

Test for Saponins

The test drugs were shaken with water vigorously for 10 minutes, copious lather formation indicates the presence of saponins.

Test for Proteins (Biuret Test):

Equal volume of 5% solution of sodium hydroxide and 1% copper sulphate were added. Appearance of pink or purple colour indicates the presence of proteins and free amino acids.

Test of Coumarins

1 ml of extract, 1 ml of 10% sodium hydroxide was added. The presence of coumarins is indicated by the formation of yellow color.

Test for Anthocyanin

About 0.2 ml of the extract was weighed in separate test tube, 1ml of 2N Sodium hydroxide was added, and heated for 5 minutes at 100 ± 2°C. The appearance of bluish green color indicates the presence of anthocyanin.

Determination of total Phenol content

The total phenol content was determined using Folin–Ciocalteu reagents with analytical grade Gallic acid as the standard. 1 ml of sample was added to deionized water (10 ml) and Folin–Ciocalteu phenol reagents (1ml). After 5



**Kopperundevi et al.,**

minutes, 20% sodium carbonate (2 ml) was added to the mixture. After being kept in total darkness for 1 hr, the absorbance was measured at 750 nm using a spectrophotometer. Amounts of total Phenol was calculated using Gallic acid calibration curve. The results were expressed as Gallic acid equivalents (GAE) mg/g of dry plant matter [15].

Total Flavonoid

Total flavonoid content in the drug MRK was determined using aluminum chloride method. In this method, Quercetin was used as standard and flavonoid contents were measured as quercetin equivalent. For this purpose, the calibration curve of quercetin was drawn. 1ml of standard or sample MRK was taken into 10ml volumetric flask, containing 4ml of distilled water. 0.3ml of 5% NaNO₂ added to the flask. After 5min, 0.3ml 10% AlCl₃ was added to the mixture. At the 6th minute add 2ml of 1M NaOH was added and volume made up to 10ml with distills water. The absorbance was noted at 510nm using UV-Visible spectrophotometer [16].

RESULTS AND DISCUSSION

The cornerstone of Siddha medicine, which is utilized to treat illnesses and maintain health, is herbal remedies and formulations¹⁷. Many Siddha formulations are produced and marketed by pharmaceutical companies in accordance with regulatory guidelines and classical texts. However, on a cursory glance, significant differences can be observed between the identical formulations produced by various companies. This raises questions about the quality standards [18]. In the present study, Megarajanga Kirutham was standardized using the modern scientific techniques. The organoleptic analysis of MRK revealed that it was yellowish liquid with characteristic odor and greasy in nature mentioned in Table 2.

Physico chemical characteristics such as rancidity, acid value, saponification value, pH, refractive index, and iodine value were assessed and mentioned in Table 3. The MRK pH value of 6 indicates a somewhat acidic environment [19]. It might occur as a result of the acidic ingredients of this formulation like amla juice and lemon juice [20,21]. At 25°C, the test drug's Refractive Index value is 1.48. It provides information about a substance's concentration, purity, and viscosity [22]. The type of fatty acid that is present in the fat determines the saponification value. The saponification value of MRK is 205. The presence of long chain fatty acids in the formulation is indicated by the saponification value [23,24]. Unsaturated free fatty acids are indicated by a high value. Compared to saturated fatty acids, unsaturated fatty acids are more readily absorbed. The iodine value of MRK was discovered as 101 [25]. The presence of free fatty acids in the oil, which cause compounds to get rancid, is indicated by the acid value. The acid value of MRK is 1.5. Less acid value indicates the less chance of decomposition [26]. Total ash value of MRK is 0.788. The amount of ash in a crude drug also indicates the level of purity in crude and the produced drug, as well as the care used in drug preservation [27]. The obtained values of the test drug were found in normal limits which indicate the good quality of product. Rancidity also not found in this formulation.

Phytochemical constituent of the herbal drug is responsible for the therapeutic effect of the formulation. So, evaluation of phytochemical constituents in this test drug is the needed one. Phytochemical screening of MRK showed in Table 4. This study revealed the presence of flavonoids, steroids, sugars, triterpenoids, coumarins and phenols. These compounds are responsible for the pharmacological activity of this formulation. Triterpenoids derived from the licorice possess the anti bacterial and anti fungal properties [28]. Antibacterial and antioxidant activity present in *Ficus racemosa* and *Syzygium cumini* responsible in the treatment of leucorrhoea [29,30]. The Phenolic and flavonoid compounds are regarded as the most significant types of phytochemicals because of their various biological characteristics. The total phenol and flavonoid contents of MRK is 0.934 ± 0.04gae mg/gm and 2.82 ± 0.42quercetin mg/gm. Recently phenolics have been considered powerful antioxidants in vitro and proved to be more potent than vitamin C and E [31].



**Kopperundevi et al.,**

CONCLUSION

The outcomes of standardization techniques including phytochemical, microbiological, chromatographic, and heavy metal analyses can be utilized as reference standards for other pharmaceutical procedures. It could also be a useful source of data for further studies to determine the formulation and evaluate its purity and quality. Further preclinical and clinical research will be required in the future to investigate additional parameters pertaining to the therapeutic effect of Megarajanga Kirutham.

ACKNOWLEDGEMENT

We wish to acknowledge our thanks to JSA Medical College for Siddha and Research Centre, Ulundurpet, Tamil Nadu, India and The Noble research solutions, Chennai, Tamil Nadu, India for their support and guidance of this research work.

CONFLICT OF INTEREST

Declared none

REFERENCES

1. Verma S, Singh SP. Current and future status of herbal medicines. *Veterinary World*. 2008;1(11):347.
2. Thatte UM, Rege NN, Phatak SD, Dahanukar SA. The flip side of Ayurveda. *J Postgrad Med*. 1993; 39:179.
3. Sriwastava, N. K., Shreedhara, C. S., & Ram, H. A. (2010). Standardization of Ajmodadichurna, a polyherbal formulation. *Pharmacognosy research*, 2(2), 98
4. Awad A, Al-Shaye D. Public awareness, patterns of use and attitudes toward natural health products in Kuwait: A cross-sectional survey. *BMC Complement Altern Med*. 2014; 14:105.
5. Dixit VK, Yadav NP: Recent approaches in herbal drug standardization. *IntegrBiol*, 2008; 2 (3): 195-203
6. Kaur H, Kaur M, Bhattacharyya A, Prajapat M, Thota P, Sarma P, Kumar S, Kaur G, Sharma S, Prakash A, Saifuddin PK, Medhi B. Indian contribution toward biomedical research and development in COVID-19: A systematic review. *Indian J Pharmacol*. 2021 Jan-Feb;53(1):63-72.
7. Shilpa Hiremath, Pradeep, Sunil Kumar KN, Some quality standards for Dronapushpi Panchanga (Leucas aspera Spreng) Powder, *International Journal of Ayurvedic Medicine*, Vol 14 (1), 2023; 219-224.
8. Guleria Praveen & Chandla Anubha: Standardisation In Ayurveda - Ancient Vis-A-Vis Modern Perspective. *International Ayurvedic Medical Journal*, 07, 2017.
9. Kannusamipillai C. *Chikitsa ratna deepam* Irandam Baagamaagiya Vaithiya Chinthami. 1st edition. Chennai; B. Rathina Nayagar and Sons; 1927. 195p.
10. Heinrich M, Williamson EM, Gibbons S, Barnes J. *Fundamentals of pharmacognosy and phytotherapy* E-BOOK. Elsevier Health Sciences; 2012 Apr 25.
11. Murugesu Mudaliar KS, Gunapadam Mooligai Vaguppu, Fourth edition, Publisher; Tamilnadu Siddha Medical Council, Chennai, 1988
12. India Pharmacopeia I Volume I, Government of India, Ministry of Health and Family welfare, Indian Pharmacopeia commission, 2014.
13. Pharmacopoeial Laboratory for Indian Medicine (PLIM) Guideline for standardization and evaluation of indian medicine which include drugs of Ayurveda, Unani and Siddha systems. Department AYUSH .Ministry of Health & Family Welfare, Govt. of India.
14. Brain KR, Turner TD. *The Practical Evaluation of Phytopharmaceuticals*. Bristol: Wright- Scientechica; 1975:36-45
15. Ganesh N. Sharma K, Nitin S, Jyotsana S. Phytochemical screening and estimation of Total Phenolic Content in Aegle marmelos Seeds. *Int J PharmaClinic Res*. 2011; 3(2): 27-29.





Kopperundevi et al.,

16. Olajire A. A and Azeez L Total antioxidant activity, phenolic, flavonoid and ascorbic acid contents of Nigerian vegetables., 2011; 2(2) 022-029, African Journal of Food Science and Technology.
17. Chandran U., Mehendale N., Tillu G., Patwardhan B. Network pharmacology of Ayurveda formulation triphala with special reference to anti-cancer property. *Comb Chem High Throughput Screen.* 2015;18:846–854.
18. Vaidya VN, Tatiya AU, Elango A, Kukkupuni SK, Vishnuprasad CN. Need for comprehensive standardization strategies for marketed Ayurveda formulations. *J Ayurveda Integr Med.* 2018 Oct-Dec;9(4):312-315.
19. Ramanzin M, Bailoni L, Bittante G. Solubility, water-holding capacity, and specific gravity of different concentrates. *Journal of dairy science.* 1994 Mar 1;77(3):774-81.
20. Rajkumar DS, Mariswamy AB. Comparative evaluation of *Emblica officinalis* as an etchant and an MMP inhibitor with orthophosphoric acid and chlorhexidine on the microshear bond strength of composite resin: an ex vivo study. *Restor Dent Endod.* 2021 Jun 8;46(3):e36.
21. Vujčić Bok V, Šola I, Rusak G. Lemon Juice Formulations Modulate In Vitro Digestive Recovery of Spinach Phytochemicals. *Food Technol Biotechnol.* 2022 Sep;60(3):293-307.
22. Allen LV Jr. Quality control analytical methods: refractive index. *Int J Pharm Compd.* 2015 Jan-Feb;19(1):43-7.
23. Basavaraj S. Hiremath, Vidyavati V. Hiremath S. Mohapatra, R.S. Sarashetti. (2013). Effect of MurchhanaSamskara in the Preparation of Hingutriguna Taila - An Analytical Study. *Biomed. Pharmacol. J.* 6: 371-374
24. Preethi R, Doddamani SA, Madikeri SS. Pharmaceutical analytical study of Hingwadi Taila. *Journal of Ayurveda and Integrated Medical Sciences;* 6 (5):136-41.
25. Almutairi A.W. Improvement of chemical composition of *tisochrysis lutea* grown mixotrophically under nitrogen depletion towards biodiesel production. *Molecules.* 2020 Oct 10;25(20):4609.
26. Ashwini Benjarwad et.al., Pharmaceutico-Analytical study of Karaviradyataila, *International Journal of Ayurvedic Medicine,* Vol 13 (3), 2022; 686-690.
27. Gami B, Parabia MH. Pharmacognostic evaluation of bark and seeds of *Mimusops elengi* L. *Int J Pharm Pharm Sci.* 2010;2(Suppl 4):110-3.
28. Wang L, Yang R, Yuan B, Liu Y, Liu C. The antiviral and antimicrobial activities of licorice, a widely-used Chinese herb. *Acta pharmaceutica sinica B.* 2015 Jul 1;5(4):310-5.
29. Mohamed AA, Ali SI, El-Baz FK. Antioxidant and antibacterial activities of crude extracts and essential oils of *Syzygium cumini* leaves. *Plos one.* 2013 Apr 12;8(4):e60269.
30. Bagyalakshmi B, Nivedhitha P, Balamurugan A. Studies on phytochemical analysis, antioxidant and antibacterial activity of *Ficus racemosa* L. leaf and fruit extracts against wound pathogens. *Vegetos.* 2019 Mar 7;32:58-63.
31. Pandey B, Rajbhandari M. Estimation of total phenolic and flavonoid contents in some medicinal plants and their antioxidant activities. *Nepal Journal of Science and Technology.* 2014;15(1):53-60.

Table 1: Ingredients of Megarajanga Kirutham

S. No	Tamil name	Botanical name	Quantity
1.	Aththi pattai	<i>Ficus racemosa</i> Linn	¼ Veesai (350 grams)
2.	Naaval pattai	<i>Syzygium cumini</i> Linn	¼ Veesai (350 grams)
3.	Othiyampattai	<i>Lannea coromandelica</i> (Houtt)Merr	¼ Veesai (350 grams)
4.	Karumbu juice	<i>Saccharum officinarum</i> Linn	½ padi (670ml)
5.	Nellikai juice	<i>Phyllanthus emblica</i> Linn	½ padi (670ml)
6.	Lemon juice	<i>Citrus limon</i> Linn	½ padi (670ml)
7.	Gingelly oil	<i>Sesamum indicum</i> Linn	½ padi (670ml)
8.	Cow's ghee		½ padi (670ml)
9.	Kirambu	<i>Syzygium aromaticum</i> Linn	¾ palam (26gms)
10.	Thalisapathiri	<i>Abies spectabilis</i> (D.Don)Mirb	¾ palam (26gms)
11.	Elam	<i>Elettaria cardamomum</i> Linn	¾ palam (26gms)
12.	Jadhikkai	<i>Myristica fragrans</i> Houtt	¾ palam (26gms)
13.	Chitrarathai	<i>Alpinia officinarum</i> Hance	¾ palam (26gms)





Kopperundeivi et al.,

14.	Athimathuram	<i>Glycyrrhiza glabra</i> Linn	¾ palam (26gms)
15.	Nilappanai kizhangu	<i>Curculigo orchoides</i> Gaertn	¾ palam (26gms)
16.	Coconut milk	<i>Cocos nucifera</i> Linn	

Table 2: Organoleptic characters of Megarajanga Kirutham

Parameter	Observation
Color	Yellowish
Smell	Characteristic
Touch	Greasy
Appearance	Turbid

Table 3: Physico – chemical analysis of Megarajanga Kirutham

S.No	Parameter	MRK
1	Loss on Drying at 105 °C (%)	8.03
2	Total Ash (%)	0.788
3	pH	6
4	Refractive index	1.48
5	Iodine value (mg I ₂ /g)	101
6	Acid value	1.5
7	Rancidity	Not oxidized
8	Saponification Value (mg of KOH to saponify 1gm of fat)	205

Table 4: Phytochemical screening of Megarajanga Kirutham

Phytocomponents	MRK
Alkaloids	-
Flavonoids	+
Glycosides	-
Steroids	+
Sugar	+
Triterpenoids	+
Coumarins	+
Phenols	+
Tannins	-
Saponins	-
Proteins	+
Anthocyanin	-
Total phenols (gae mg/gm)	0.934 ± 0.04
Total flavanoids (quercetin mg/gm)	2.82 ± 0.42

+ indicates positive; - indicates negative





A Study on an Irregular Colouring of Sunlet and Pangraph Families

G.Jothilakshmi^{1*}, K.Rajam² and A.Sathyakala²

¹Assistant Professor, Department of Mathematics, RVS College of Arts And Science, Sulur, (Affiliated to Bharathiar University), Coimbatore, Tamil Nadu, India.

²Assistant Professor, Department of Mathematics, Dr. NGP Institute of Technology College, (Anna University, Chennai) Coimbatore, Tamil Nadu, India.

Received: 04 July 2024

Revised: 14 Aug 2024

Accepted: 17 Oct 2024

*Address for Correspondence

G.Jothilakshmi

Assistant Professor, Department of Mathematics,
RVS College of Arts And Science, Sulur,
(Affiliated to Bharathiar University),
Coimbatore, Tamil Nadu, India.
E. Mail: jothikrishna11@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

An irregular coloring is a proper coloring in which distinct vertices have different color codes. In this paper we obtain the irregular coloring of Sunlet and Pangraph families are studied.

Keywords: Central graph, Graph coloring, Irregular chromatic number, Irregular coloring, Middle graph, Pan graph, Sunlet graph and Total graph

INTRODUCTION

Let G be a finite, undirected graph with no loops and multiple edges. The graph G has the vertex set $V(G)$ and the edge set $E(G)$. Graph coloring is coloring of G such that no two adjacent vertices share the same color.

A proper coloring of a graph is an **irregular coloring**, if no two like-colored vertices have the same color code. i.e., for every pair of vertices u and w , $\text{code}(u) \neq \text{code}(w)$ whenever $c(u) = c(w)$. Thus an Irregular coloring distinguishes each vertex from each of other vertex by its color or by its color code.

The **central graph** $C(G)$ of a graph G is obtained by subdividing each edge of G by exactly once and joining all the non adjacent vertices of G .

The **middle graph** of G denoted by $M(G)$, is defined as follows :

The vertex set of $M(G)$ is $V(G) \cup E(G)$ in which two elements are adjacent in $M(G)$ if the following conditions hold. (i) $x, y \in E(G)$ and x, y are adjacent in G . (ii) $x \in V(G)$, $y \in E(G)$ and y is incident on x in G .

The total graph $T(G)$ of a graph G is a graph such that the vertex set of T corresponds to the vertices and edges of G and two vertices are adjacent in T iff their corresponding elements are either adjacent or incident in G .





Jothilakshmi et al.,

Irregular Coloring Of Sunlet Graph Families

Theorem 2.1 : For the central graph of Sunlet graph ,the irregular chromatic number is $n+2$

(i. e) $\chi_{ir}[C(S_n)] = n+2, n \geq 3$

Proof.

Let $G = C(S_n)$ be the central graph of sunlet graph.

Let v_1, v_2, \dots, v_n be the inner vertices of the cycle graph C_n

Let u_1, u_2, \dots, u_n be the pendant vertices .

The inner sub-division of the cycle graph C_n are $v_{i,i+1}, v_{i+1,i+2}, \dots, v_n, v_1$

The outer subdivision pendant vertices are f_1, f_2, \dots, f_n

Now the coloring assignment of $C(S_n)$ is as follows.

The inner vertices v_1, v_2, \dots, v_n of the cycle graph C_n and the outer pendant vertices u_1, u_2, \dots, u_n are colored with the colors $c_1, c_2, c_3, \dots, c_n$

The inner sub-division vertices $v_{i,i+1}, v_{i+1,i+2}, \dots, v_n, v_1$ of the cycle graph C_n are colored with the colors c_3, c_4, \dots, c_{n+2}

The outer sub-division pendant vertices f_1, f_2, \dots, f_n are colored with the color c_{n+1}

since $\deg(u_i) \neq \deg(v_i)$, it shows that $\text{code}(u_i) \neq \text{code}(v_i)$

Hence an irregular chromatic number of central graph of sunlet graph is $2n+3$.

(i.e) $\chi_{ir}[C(S_n)] = n+2, n \geq 3$

Theorem 2.2 : For the middle graph of Sunlet graph ,the irregular chromatic number is $2n+3$ (i. e) $\chi_{ir}[M(S_n)] = 2n+3, n \geq 3$

Proof.

Let $G = M(S_n)$ be the middle graph of sunlet graph.

Let v_1, v_2, \dots, v_n be the inner vertices of the cycle graph C_n and u_1, u_2, \dots, u_n be the pendant vertices . The inner sub-division of the cycle graph C_n are $v_{i,i+1}, v_{i+1,i+2}, \dots, v_n, v_1$ and the outer subdivision pendant vertices are f_1, f_2, \dots, f_n

Now the coloring assignment of $M(S_n)$ is as follows.

The inner vertices v_1, v_2, \dots, v_n of the cycle graph C_n are colored with the colors $c_1, c_2, c_3, \dots, c_n$

The inner sub-division vertices $v_{i,i+1}, v_{i+1,i+2}, \dots, v_n, v_1$ of the cycle graph C_n are colored with the colors c_3, c_4, \dots, c_{n+2}

The outer pendant vertices u_1, u_2, \dots, u_n are colored with the color c_{n+3}

The outer sub-division pendant vertices f_1, f_2, \dots, f_n are colored with the color $c_{n+4}, c_{n+5}, \dots, c_{2n+3}$

since $\deg(u_i) \neq \deg(f_i)$, it shows that $\text{code}(u_i) \neq \text{code}(f_i)$

Since each u_i 's are adjacent to f_i 's but u_i 's are not adjacent to v_i 's.

It follows that, $\text{code}(u_i) \neq \text{code}(v_i)$.

Hence an irregular chromatic number of middle graph of sunlet graph is $2n+3$.

(i.e) $\chi_{ir}[M(S_n)] = 2n+3, n \geq 2$

Theorem 2.3 : For the total graph of Sunlet graph ,the irregular chromatic number is $2n+3$ (i. e) $\chi_{ir}[T(S_n)] = 2n+3, n \geq 3$

Proof.

Let $G = T(S_n)$ be the total graph of Sunlet graph.

Now the coloring assignment of $T(S_n)$ is as follows.

The inner vertices v_1, v_2, \dots, v_n of the cycle graph C_n are colored with the colors $c_1, c_2, c_3, \dots, c_n$

The inner sub-division vertices $v_{i,i+1}, v_{i+1,i+2}, \dots, v_n, v_1$ of the cycle graph C_n are colored with the colors c_3, c_4, \dots, c_{n+2}

The outer pendant vertices u_1, u_2, \dots, u_n are colored with the colors $c_{n+4}, c_{n+5}, \dots, c_{2n+3}$

The outer sub-division pendant vertices f_1, f_2, \dots, f_n are colored with the color c_{n+3}

To prove $(2n+3)$ - coloring is an irregular coloring of $T(S_n)$,

since $\deg(u_i) \neq \deg(f_i)$, it shows that $\text{code}(u_i) \neq \text{code}(f_i)$

Since each u_i 's are adjacent to f_i 's but u_i 's are not adjacent to v_i 's.

It follows that, $\text{code}(u_i) \neq \text{code}(v_i)$.

Hence an irregular chromatic number of total graph of sunlet graph is $2n+3$.

(i.e) $\chi_{ir}[T(S_n)] = 2n+3, n \geq 2$





Jothilakshmi et al.,

Remark:

For central ,middle and total graph of sunletgraph, we can give same colors in different manner.

Irregular Coloring of Pan Graph Families

Theorem 3.1: For the central and total graph of Pan graph,the irregular chromatic number is $n+2$. (i.e) $\chi_{ir}[C(P_n)] = \chi_{ir}[M(P_n)] = \chi_{ir}[T(P_n)] = n+2$

Proof:

The n -pan graph is the graph obtained by joining a cycle graph C_n to a singleton graph K_1 with a bridge. The n -pan graph is isomorphic with the $(n,1)$ tadpole graph.

Let $G=C(P_n)$ be the central graph of pan graph.

Now the coloring assignment is as follows.

Case(i):For central graph of pan graph

The outer vertices $\{v_1, v_2, \dots, v_n\}$ of the cycle graph are colored with the colors $\{c_1, c_2, c_3, \dots, c_n\}$ (clockwise)

The outer- intermediate vertices $\{u_1, u_2, \dots, u_n\}$ are colored with the colors $\{c_3, c_4, \dots, c_{n+2}\}$ (clockwise)

The outer- intermediate vertex w_1 is colored with the color c_1

At last, the vertex of the singleton graph k_1 is colored with the color c_2

Case(ii):For middle graph of pan graph

The proof for case (ii) is similar as in case(i)

Case(ii):For total graph of pan graph

The outer vertices $\{v_1, v_2, \dots, v_n\}$ of the cycle graph are colored with the colors $\{c_1, c_2, c_3, \dots, c_n\}$ (clockwise)

The outer- intermediate vertices $\{u_1, u_2, \dots, u_n\}$ are colored with the colors $\{c_3, c_4, \dots, c_{n+2}\}$ (clockwise)

The outer- intermediate vertex w_1 is colored with the color c_1

At last, the vertex of the singleton graph k_1 is colored with the color c_{n+2}

From cases (i),(ii) and (iii)

since $\text{deg}(u_i) \neq \text{deg}(v_i)$, it shows that $\text{code}(u_i) \neq \text{code}(v_i)$

Since each u_i 's are adjacent to v_i 's but u_i 's are not adjacent to w_i 's.

It follows that, $\text{code}(u_i) \neq \text{code}(w_i)$.

Hence an irregular chromatic number of total graph of Pan graph is $n+2$.

$$\chi_{ir}[C(P_n)] = \chi_{ir}[M(P_n)] = \chi_{ir}[T(P_n)] = n+2$$

Remark:

For central ,middle and total graph of pan graph, we can give same colors in different manner.

REFERENCES

1. J. A. Bondy and U. S. R. Murty, Graph theory with Applications, London Macmillan
2. Danuta Michalak, On middle and total graphs with coarseness number equal 1, Graph theory, Proc. nf., Lag'ow/Pol. 1981, Lect. Notes Math. 1018, Springer, Berlin pp. 139-150(1983).
3. Frank Harary, Narosa Publishing Home, (2001).
4. M.S. Franklin, Thamilselvi, Harmonious coloring of central graph of certain graphs, Applied Mathematical sciences, 5 pp.569-578(2015)
5. Graph Coloring, Wikipedia, the free encyclopedia, SIAM J. Algebra Discrete Math 4 pp. 306-311(1983).
6. G.Jothilakshmi, On harmonious coloring of $M[B(K_n, K_n)]$ and $C[B(K_n, K_n)]$, International journal of Mathematical Archive -5(10), pp.12-14 (2014)
7. G.Jothilakshmi, On harmonious coloring of $C(H_n)$ and $C(G_n)$, Journal of Global Research in Mathematical Archives, Volume 2, No. 5, (May 2014)
8. Mary.U.G.Jothilakshmi, On harmonious coloring of $M(S_n)$ and $M(D_3^m)$, International journal of computer Applications, vol4, pp.239-244(2014)





9. Mary.U and G.Jothilakshmi,A study on harmonious coloring of Middle graph of certain snake graphs, Journal Of Applied Science And Computations,vol 6,pp.30-37(2019)
10. Wilson Robin . J, Introduction to Graph Theory.

<p>Fig 1: Irregular coloring of $C(S_3)$</p>	<p>Fig 2 : Irregular coloring of $M(S_3)$</p>
<p>Fig 3: Irregular coloring of $T(S_3)$</p>	<p>Fig 4. Central graph of Pan graph P_3</p>
<p>Fig 5. Middle graph of Pan graph P_3</p>	<p>Fig 6. Total graph of Pan graph P_3</p>





A Comprehensive Analysis of Glaucoma Detection using Machine Learning and Deep Learning : A Survey

J. Karunanithi^{1*} and S. Jagadesh Kumar²

¹Associate Professor and Head, Department of Computer Science, Thavathiru Santhalinga Adigalar Arts Science and Tamil College, Perur, (Affiliated to Bharathiar University), Coimbatore, Tamil Nadu, India.

²Assistant Professor, Department of Computer Applications, Thavathiru Santhalinga Adigalar Arts Science and Tamil College, Perur, (Affiliated to Bharathiar University), Coimbatore, Tamil Nadu, India.

Received: 20 Jun 2024

Revised: 22 Aug 2024

Accepted: 26 Oct 2024

*Address for Correspondence

J. Karunanithi

Associate Professor and Head,
Department of Computer Science,
Thavathiru Santhalinga Adigalar Arts Science and Tamil College,
Perur, (Affiliated to Bharathiar University),
Coimbatore, Tamil Nadu, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Glaucoma is a degenerative eye disease that, if unchecked, causes permanent blindness. Efficient illness management requires prompt diagnosis and prompt treatment. Ophthalmologists have found some success in using ML and DL methods to aid in the diagnosis of glaucoma in recent years. The present state-of-the-art in glaucoma detection using ML and DL-based techniques is thoroughly examined in this review. To set the stage, we will go over the basics of glaucoma's pathogenesis and the eye's anatomy. After that, we explore the several ML and DL methods used for glaucoma detection, such as SVMs, RNs, and convolutional and recurrent neural networks. This article delves into the possibilities of computer-assisted techniques for glaucoma illness and staging, reviewing 30 research articles on the topic. Important parameters for automated diagnosis are examined in this study, including optic disc shape, cup-to-disc ratio, and retinal nerve fiber layer thickness. Furthermore, we go over some of the difficulties encountered when trying to create reliable ML/DL models for glaucoma detection, including issues with data scarcity, class imbalance, and the interpretability of model conclusions.

Keywords: Convolutional Neural Networks, Early detection, Glaucoma, Machine learning, deep learning

INTRODUCTION

Without prompt diagnosis and treatment, glaucoma, a degenerative eye condition, may cause permanent damage to the optic nerve and eventually cause blindness. A primary cause of blindness, it affects millions of people globally [1-4]. Preserving eyesight and improving patient outcomes are paramount when it comes to timely identification and management. However, glaucoma may be difficult to diagnose and often calls for specialist tools and knowledge. Clinical glaucoma diagnosis has been greatly improved in recent years because to the incorporation of deep learning



**Karunanithi and Jagadesh Kumar**

(DL) and machine learning (ML) into medical imaging [5–11]. These innovations use computer algorithms to sift through mountains of data, such as retinal scans, in search of traits and patterns that could indicate glaucomatous alterations. The glaucoma screening, diagnostic, and monitoring processes might be completely transformed by this paradigm shift towards automated or semi-automatic detection technologies [12–14].

The purpose of this review is to provide a thorough evaluation of the present state of glaucoma detection methods based on ML and DL. To provide the groundwork for a good grasp, we shall investigate the pathophysiology and anatomy of glaucoma [15–17]. After that, we will go over the merits, weaknesses, and performance measures of the different ML and DL algorithms used for glaucoma detection [18–20]. We will also talk about the benefits and drawbacks of using publically accessible datasets to train and test glaucoma detection algorithms. We will also discuss the main obstacles, such as a lack of data, an imbalance in the classes, and the difficulty in understanding the reasoning behind the model's conclusions, that have been identified as preventing the development of reliable and clinically relevant ML/DL models for the identification of glaucoma [21–27]. Our goal is to discover current trends, research gaps, and future prospects in ML/DL-based glaucoma detection by reviewing and combining the current literature and advances in this area. We want to contribute to the development of scalable solutions that are reliable, accurate, and can be smoothly incorporated into clinical practice [28–31].

Background study

Afreen, N., & Aluvalu, R. (2024) a number of glaucoma detection techniques were examined in depth in this article. Since glaucoma was one of the most deadly eye diseases that may cause blindness without any noticeable symptoms, it was critical to diagnose the disease early on so that patients can prevent irreversible vision loss. Research has shown that deep learning models may aid in the early detection process. AlShawabkeh, M. et al. (2024) there was great potential for improving early detection, accuracy, and accessibility in glaucoma diagnoses via the application of AI. A number of models have shown promise as sensitive screening tools for glaucoma, but it was still difficult to create diagnostic models that can be used in specialized glaucoma care clinics by combining multi-modal anatomical and functional datasets. Bragança, C. P. et al. (2024) there has been tremendous success in distinguishing between glaucomatous and non-glaucomatous digital fundoscopic pictures, thanks to the invention of glaucoma classification algorithms. Nevertheless, at the present time, no software was accessible for actual clinical applications, even though many research have shown that AI algorithms may be used to aid in the diagnosis of glaucoma.

Hasan, M. et al. (2024) these authors research systematically analyzed the high-quality literature on AI-based glaucoma and neurodegenerative disease diagnostics published in the last 10 years. Analysis of the included research reveals that AI significantly improves glaucoma and neurodegenerative disease diagnoses compared to human doctors (area under the curve: 0.71–0.98, accuracy: 71.5%, area under the curve: 0.86). Jain, A., & Sakalle, V. (2024) Recent advances in medical diagnostics using ML and DL have created revolutionary new opportunities, particularly in the field of ophthalmology. The author highlight the tremendous potential of ML and DL models for glaucoma optic disk localization and classification in these authors assessment of these approaches to improve early identification and treatment of the disease.

L. Li et al. (2020) As a novel deep learning approach for automated glaucoma identification and abnormal region localization on fundus pictures, the author present AG-CNN in this research. Therefore, glaucoma might be identified by using the in-depth characteristics emphasized by the visual maps of diseased regions, which were derived from the anticipated attention maps. Shyamalee, T. et al. (2024) these authors research presented a deep learning-based method that incorporates segmentation, classification, and explainability to identify glaucoma illnesses using different fundus image datasets. The author demonstrated the system's usability and the veracity of the results after deploying the suggested method as a web application. Using explainable methodologies, this study aimed to identify the reasons behind the underlying classification model's prediction. Because of this, people were more likely to have faith in the system and be comfortable using it to aid in medical diagnosis. Subbarayudu, Y. et al. (2024) on the test data, the Logistic Regression model reached an accuracy of almost 95%. When compared to other models, including SVM, Random Forest, Decision Tree, and Naive Bayes, it achieved greater accuracy. A larger



**Karunanithi and Jagadesh Kumar**

number of false negatives (12) than false positives (5) was shown by the confusion matrix, suggesting that the model had a harder time accurately detecting class 1 photos with glaucoma than class 0 images without the condition. As a measure of the model's accuracy in classifying positive occurrences (glaucoma) out of all instances predicted as positive, the precision was determined to be 0.84. The accuracy in classifying positive events out of all the real positive instances was determined to be 0.94, which was known as the recall. Tang, Q. Y. et al. (2024) the aberrant WM functional abnormalities seen in PACG patients in this research mostly affect areas of the brain that were involved in vision. From a different WM functional angle, these authors results provide light on extensive brain damage in PACG. In addition, the aberrant ALFF values in brain WM that have been reported may be a neurological marker that may differentiate PACG patients from healthy controls.

M. Aloudat and M. Faezipour (2016) A increased risk of glaucoma was associated with a high proportion of red sclera in the excised tissue, according to these investigators' approach. M. N. Bajwa et al. (2016) Historically, glaucoma diagnostic Retinal Fundus photos (RFI) datasets have been rather tiny, consisting of just a few hundred photos, and have been meticulously acquired in controlled laboratory settings. None of the photos in these datasets include extra-retinal artifacts since the creators didn't care about the limits of imaging technology. There were a lot of eye disorders, according to the research, thus deep learning systems trained on these datasets don't do well. N. A. Diptu et al. (2018) the method developed by these writers was useful for the automated diagnosis of glaucoma in fundus images. This authors' technique successfully identifies the intermediate stage of glaucoma, which was a crucial component of any screening system for the condition. The region-of-interest (ROI) segmentation was used to detect bleeding in a specific area, which allowed for the automated identification of glaucoma issues. Due to its emphasis on localizing hemorrhage sites, the authors' technique proved computationally efficient. Applying this authors' method yielded notable results.

R. Panda et al. (2017) the author suggested a novel method for identifying RNFLDs to aid ophthalmologists in their endeavors to screen big populations for glaucoma. Accurately identifying the real border pixels may be achieved by feeding the RNN training data with freshly produced patch attributes. R. Zhao et al. (2020) Instead of using the segmentation phase that was often employed in traditional approaches, a methodology was devised to immediately estimate the Cup-to-Disc Ratio (CDR) value from fundus photographs. By combining semi-supervised learning with random regression forest, the author can make pictures more expressive and find the optimal CDRs to represent different image qualities. Following evaluation on a challenging glaucoma dataset, the authors' direct approach accurately calculated the CDR value, demonstrating strong correlations to values recorded manually. Their research indicates that this was the first instance of calculating a CDR value directly from a fundus picture, without the need for segmentation.

S. C. Shetty and P. Gutte (2018) In order to distinguish between the optic cup and disc, the authors' study used K-means Clustering to group various visual components according to color. Ultimately, the boundary method's fractal dimension value may be used to detect glaucoma. With this method, the authors were able to distinguish between healthy eyes and those with glaucoma in 92% of instances. This approach may potentially be useful for detecting diabetic retinopathy.

EXISTING METHODS

Popular methods

Convolutional neural network

The optimum deep learning model for glaucoma classification from retinal pictures is a CNN, which is very skilled at extracting and evaluating visual characteristics. CNNs automatically learn complex patterns indicative of glaucomatous alterations, beginning with low-level elements like as textures and edges and progressing to higher-level representations that are critical for diagnosis, thanks to their hierarchical design. They are more generalizable and resilient because they can locate important elements in pictures and use data augmentation methods. CNNs



**Karunanithi and Jagadesh Kumar**

trained on massive labeled datasets use backpropagation to accurately classify retinal pictures as either glaucoma or non-glaucoma, allowing for informed clinical decision-making. Ophthalmologists are able to help patients get the treatment they need sooner because to CNNs' data-driven automation, which improves glaucoma diagnosis.

Recurrent Neural Networks

Kind of deep learning models called RNNs excel in sequential data analysis, which makes them useful for glaucoma classification using longitudinal retinal imaging and other similar tasks. Unlike feedforward networks, RNNs may remember previous inputs and understand how data in sequences is dependent on one another over time because of their connections that loop back on themselves. To classify glaucoma, RNNs may examine retinal pictures captured over time at different appointments, revealing the gradual alterations that characterize glaucomatous disease. They are quite good at forecasting the course of diseases, identifying stable and advancing instances, and picking up on minor patterns of progression. To increase RNNs' performance in glaucoma classification, techniques such as Gated Recurrent Unit (GRU) architectures and LSTM algorithms deal with the vanishing gradient issue and enhance learning of long-term dependencies. Automated and reliable glaucoma diagnosis is made possible by RNNs because of its capacity to acquire contextual cues from sequential data and handle input sequences of varying lengths. This helps physicians with early intervention and individualized treatment plans.

Artificial intelligent

Machine learning and deep learning are two branches of AI that may be used to classify retinal pictures as indicators of glaucoma. Machine learning algorithms like SVMs, Random Forests, and Gradient Boosting Machines may analyse retinal pictures for characteristics and use those features to determine whether the images are glaucomatous or not. To accurately identify glaucoma, DL models, especially CNNs, shine when it comes to autonomously learning hierarchical features from raw picture data. To help with early diagnosis and prompt action, AI-based methods use big annotated datasets to build models that do well with unseen data. When it comes to glaucoma screening and care, ophthalmologists may greatly benefit from the automation, efficiency, and scalability that AI algorithms provide. In order to use AI in glaucoma classification responsibly and effectively, we still need to solve three major issues: the interpretability of model choices, integration into clinical processes, and ethical concerns.

DISCUSSION

Ophthalmologists are facing a serious challenge in detecting glaucoma, a disorder that may cause irreparable visual loss if left untreated. However, there is hope that ML and DL approaches together have showed potential in this area. Following an overview of the anatomy of the eye and the pathophysiology of glaucoma, this study delves into the most recent developments in ML/DL-based methods for glaucoma identification. Important characteristics retrieved from retinal pictures are covered, along with several techniques such as Support Vector Machines, Random Forests, Convolutional Neural Networks, and Recurrent Neural Networks. Collaborative efforts are urged for standardized methods, bigger datasets, and regulatory compliance in light of highlighted challenges such as data scarcity, class imbalance, and model interpretability. For future breakthroughs in AI-driven glaucoma diagnosis and treatment, it is crucial to prioritize interpretability, ethical issues, and multidisciplinary cooperation.

CONCLUSION

Finally, ophthalmic care stands to benefit greatly from the development of ML and DL approaches for glaucoma identification. The present state of ML/DL-based techniques has been investigated in this survey, and their ability to aid in the early detection and treatment of glaucoma has been highlighted. To guarantee the safe and successful integration of these technologies into clinical practice, however, a number of obstacles, including data scarcity, model interpretability, and ethical issues, must be overcome. To solve these problems and create scalable, interpretable, and morally acceptable AI-driven solutions for glaucoma diagnosis and beyond, it is vital for researchers, physicians,





Karunanithi and Jagadesh Kumar

industry stakeholders, and regulatory agencies to work together. To fully use AI for better patient outcomes and lessen the worldwide impact of glaucoma-related blindness, we must emphasize openness, data privacy, and regulatory compliance as we move towards this future.

REFERENCES

1. A. S. Patel and V. Singh, "Glaucoma Detection using Mask Region-Based Convolutional Neural Networks," 2021 5th International Conference on Electronics, Communication and Aerospace Technology (ICECA), 2021, pp. 1642-1647, doi: 10.1109/ICECA52323.2021.9675939.
2. A.Li, Y. Wang, J. Cheng and J. Liu, "Combining Multiple Deep Features for Glaucoma Classification," 2018 IEEE International Conference on Acoustics, Speech and Signal Processing (ICASSP), 2018, pp. 985-989, doi: 10.1109/ICASSP.2018.8462089.
3. A.Serener and S. Serte, "Transfer Learning for Early and Advanced Glaucoma Detection with Convolutional Neural Networks," 2019 Medical Technologies Congress (TIPTEKNO), 2019, pp. 1-4, doi: 10.1109/TIPTEKNO.2019.8894965.
4. Afreen, N., & Aluvalu, R. (2024). Glaucoma Detection Using Explainable AI and Deep Learning. EAI Endorsed Transactions on Pervasive Health and Technology, 10.
5. AlShawabkeh, M. A., AlRyalat, S. A., Al Bdour, M., Alni'Mat, A., & Al-Akhras, M. (2024). The Utilization of Artificial Intelligence in Glaucoma: Diagnosis versus Screening. *Frontiers in Ophthalmology*, 4, 1368081.
6. Bragança, C. P., Torres, J. M., Macedo, L. O., & Soares, C. P. D. A. (2024). Advancements in Glaucoma Diagnosis: The Role of AI in Medical Imaging. *Diagnostics*, 14(5), 530.
7. Chuter, B., Huynh, J., Bowd, C., Walker, E., Rezapour, J., Brye, N., ... & Christopher, M. (2024). Deep Learning Identifies High-Quality Fundus Photographs and Increases Accuracy in Automated Primary Open Angle Glaucoma Detection. *Translational Vision Science & Technology*, 13(1), 23-23.
8. D. Vaghjiani, S. Saha, Y. Connan, S. Frost and Y. Kanagasingam, "Visualizing and Understanding Inherent Image Features in CNN-based Glaucoma Detection," 2020 Digital Image Computing: Techniques and Applications (DICTA), 2020, pp. 1-3, doi: 10.1109/DICTA51227.2020.9363369.
9. G. Satya Nugraha, B. Amelia Riyandari and E. Sutoyo, "RGB Channel Analysis for Glaucoma Detection in Retinal Fundus Image," 2020 International Conference on Advancement in Data Science, E-learning and Information Systems (ICADEIS), 2020, pp. 1-5, doi: 10.1109/ICADEIS49811.2020.9277230.
10. Hasan, M. M., Phu, J., Sowmya, A., Meijering, E., & Kalloniatis, M. (2024). Artificial intelligence in the diagnosis of glaucoma and neurodegenerative diseases. *Clinical and Experimental Optometry*, 107(2), 130-146.
11. Jain, A., & Sakalle, V. (2024). A Review of Glaucoma Optic Disk Localization and Classification Machine Learning and Deep Learning Models. *International Journal of Innovative Research in Technology and Science*, 12(2), 163-176.
12. L. Li et al., "A Large-Scale Database and a CNN Model for Attention-Based Glaucoma Detection," in *IEEE Transactions on Medical Imaging*, vol. 39, no. 2, pp. 413-424, Feb. 2020, doi: 10.1109/TMI.2019.2927226.
13. M. Aljazaeri, Y. Bazi, H. AlMubarak and N. Alajlan, "Deep Segmentation Architecture with Self Attention for Glaucoma Detection," 2020 International Conference on Artificial Intelligence & Modern Assistive Technology (ICAIMAT), 2020, pp. 1-4, doi: 10.1109/ICAIMAT51101.2020.9308006.
14. M. Aloudat and M. Faezipour, "Determination for Glaucoma disease based on red area percentage," 2016 IEEE Long Island Systems, Applications and Technology Conference (LISAT), 2016, pp. 1-5, doi: 10.1109/LISAT.2016.7494099.
15. M. C. V. Stella Mary, E. B. Rajsingh and G. R. Naik, "Retinal Fundus Image Analysis for Diagnosis of Glaucoma: A Comprehensive Survey," in *IEEE Access*, vol. 4, pp. 4327-4354, 2016, doi: 10.1109/ACCESS.2016.2596761.
16. M. N. Bajwa, G. A. P. Singh, W. Neumeier, M. I. Malik, A. Dengel and S. Ahmed, "G1020: A Benchmark Retinal Fundus Image Dataset for Computer-Aided Glaucoma Detection," 2020 International Joint Conference on Neural Networks (IJCNN), 2020, pp. 1-7, doi: 10.1109/IJCNN48605.2020.9207664.





Karunanithi and Jagadesh Kumar

17. N. A.Diptu et al., "Early Detection of Glaucoma Using Fuzzy Logic in Bangladesh Context," 2018 International Conference on Intelligent Systems (I.S.), 2018, pp. 87-93, doi: 10.1109/IS.2018.8710490.
18. N. Sengar, M. K. Dutta, R. Burget and M. Ranjoha, "Automated detection of suspected glaucoma in digital fundus images," 2017 40th International Conference on Telecommunications and Signal Processing (TSP), 2017, pp. 749-752, doi: 10.1109/TSP.2017.8076088.
19. R. Panda, N. B. Puhon, A. Rao, D. Padhy and G. Panda, "Recurrent neural network based retinal nerve fiber layer defect detection in early glaucoma," 2017 IEEE 14th International Symposium on Biomedical Imaging (ISBI 2017), 2017, pp. 692-695, doi: 10.1109/ISBI.2017.7950614.
20. R. W. Chan et al., "Relationships between cerebrovascular reactivity, visual-evoked functional activity, and resting-state functional connectivity in the visual cortex and basal forebrain in glaucoma," 2021 43rd Annual International Conference of the IEEE Engineering in Medicine & Biology Society (EMBC), 2021, pp. 4037-4040, doi: 10.1109/EMBC46164.2021.9630904.
21. R. Zhao, X. Chen, X. Liu, Z. Chen, F. Guo and S. Li, "Direct Cup-to-Disc Ratio Estimation for Glaucoma Screening via Semi-Supervised Learning," in IEEE Journal of Biomedical and Health Informatics, vol. 24, no. 4, pp. 1104-1113, April 2020, doi: 10.1109/JBHI.2019.2934477.
22. S. C. Shetty and P. Gutte, "A Novel Approach for Glaucoma Detection Using Fractal Analysis," 2018 International Conference on Wireless Communications, Signal Processing and Networking (WiSPNET), 2018, pp. 1-4, doi: 10.1109/WiSPNET.2018.8538760.
23. S. Issac Niwas et al., "Cross-Examination for Angle-Closure Glaucoma Feature Detection," in IEEE Journal of Biomedical and Health Informatics, vol. 20, no. 1, pp. 343-354, Jan. 2016, doi: 10.1109/JBHI.2014.2387207.
24. S. Nawaldgi, "Review of automated glaucoma detection techniques," 2016 International Conference on Wireless Communications, Signal Processing and Networking (WiSPNET), 2016, pp. 1435-1438, doi: 10.1109/WiSPNET.2016.7566373.
25. S. Ovreiu, E. -A. Paraschiv and E. Ovreiu, "Deep Learning & Digital Fundus Images: Glaucoma Detection using DenseNet," 2021 13th International Conference on Electronics, Computers and Artificial Intelligence (ECAI), 2021, pp. 1-4, doi: 10.1109/ECAI52376.2021.9515188.
26. S. Phasuk et al., "Automated Glaucoma Screening from Retinal Fundus Image Using Deep Learning," 2019 41st Annual International Conference of the IEEE Engineering in Medicine and Biology Society (EMBC), 2019, pp. 904-907, doi: 10.1109/EMBC.2019.8857136.
27. Shyamalee, T., Meedeniya, D., Lim, G., & Karunarathne, M. (2024). Automated Tool Support for Glaucoma Identification with Explainability Using Fundus Images. IEEE Access.
28. Singh, M., Vivek, B. S., Gubbi, J., & Pal, A. (2024). Prototype-based Interpretable Model for Glaucoma Detection. In Proceedings of the IEEE/CVF Conference on Computer Vision and Pattern Recognition (pp. 5056-5065).
29. Subbarayudu, Y., Reddy, G. V., Kumar, M. R., Naik, M. A., Nagini, R. V. S. S., & Singh, B. (2024). An efficient novel approach for glaucoma classification on retinal fundus images through machine learning paradigm. In MATEC Web of Conferences (Vol. 392, p. 01108). EDP Sciences.
30. Tang, Q. Y., Zhong, Y. L., Wang, X. M., Huang, B. L., Qin, W. G., & Huang, X. (2024). Machine learning analysis classifies patients with primary angle-closure glaucoma using abnormal brain white matter function. Clinical Ophthalmology, 659-670.

Table 1: Comparison of Machine Learning Methods for Glaucoma Detection

Reference	Method	Advantages	Disadvantages	Accuracy
1	Mask Region-Based convolutional neural network (Mask-RCNN)	Mask-RCNN) shows a distinct pattern for eyes with glaucoma and those without, which might be used to identify glaucoma using machine learning.	This component enhances the location of the BB and assigns ROIs to many more categories than the RPN module.	93%





Karunanithi and Jagadesh Kumar

3	convolutional neural network	A novel glaucoma classification was developed by merging many characteristics retrieved by various convolutional neural networks.	A clear benefit of classifier was that it uses more information for general picture categorization, even if the pathophysiology underlying it was hard to describe.	
5	ResNet-50 and GoogLeNet	Two deep learning algorithms, ResNet-50 and GoogLeNet, were used to classify fundus pictures as either early or advanced glaucoma.	Among the three methods for early glaucoma identification, optic disk examination was the most popular and commonly employed by experts due to its convenience. The fact that it was expensive and time-consuming was still a drawback.	ResNet-50 – 0.90% Googlenet – 0.91%
11	CNN	may include using same author methods on a broader and more varied collection of pictures	A large number of studies were carried out using glaucoma datasets that were made publically accessible.	93%

Table 2: Comparative Analysis of Machine Learning Algorithms for Glaucoma Detection

Algorithm	Merits	Demerits
CNN [1]	It is no longer necessary to manually extract features from raw retinal pictures; CNNs can learn discriminative features automatically. When it comes to detecting tiny changes caused by glaucomatous disease, this expertise is invaluable.	To train well and generalize to unseen samples, CNNs usually need a lot of labelled data. It might be difficult to get annotated datasets with a wide range of patient demographics and disease severity levels for use in glaucoma classification.
RNN [19]	Longitudinal retinal imaging or the dynamic changes in glaucomatous characteristics over time are examples of time-series data that may be well-analyzed using Recurrent Neural Networks (RNNs) due to their exceptional ability to capture temporal relationships and sequences.	Because of the vanishing or exploding gradient issue, which occurs when gradients become either too tiny (vanishing) or too high (exploding) during backpropagation, training deep RNNs may be hard. This has the potential to impact model performance and make it harder to understand long-term dependencies.
Artificial intelligent [10]	When trained on enough and representative data, artificial intelligence (AI) techniques like ML and DL models may achieve excellent accuracy in glaucoma classification tasks. They may help with early diagnosis and management by identifying subtle patterns and characteristics of glaucomatous alterations in retinal pictures.	The amount and quality of training data have a significant impact on AI models. In real-world clinical settings with different patient populations, inferior performance and generalization concerns might be caused by biased or limited datasets.





Karunanithi and Jagadesh Kumar

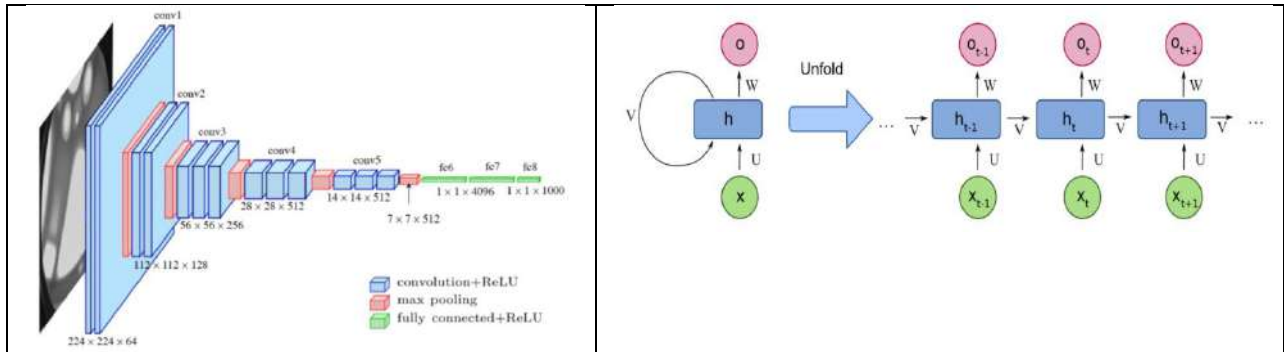


Figure 1: CNN architecture [Source: <https://vitalflux.com/different-types-of-cnn-architectures-explained-examples/>]

Figure 2: [Source: <https://www.analyticsvidhya.com/blog/2022/03/a-brief-overview-of-recurrent-neural-networks-rnn/>]





Bridging Heritage and Innovation in Sustainable Agriculture of India

Prodip Kumar Sarkar*

Assistant Professor, Department of Sanskrit, Suri Vidyasagar College, Suri, (Affiliated to The University of Burdwan), West Bengal, India.

Received: 26 July 2024

Revised: 16 Sep 2024

Accepted: 24 Oct 2024

*Address for Correspondence

Prodip Kumar Sarkar

Assistant Professor, Department of Sanskrit,
Suri Vidyasagar College, Suri,
(Affiliated to The University of Burdwan),
West Bengal, India.

E.Mail: pksarkar1988@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Sustainability is a key focus in the transition from traditional to contemporary technologies in Indian agricultural approaches, and a look into the past is necessary to set the stage for this change. The dynamic geography of agricultural practices investigated by the study illustrates an agricultural renaissance which points out how technology has been seamlessly incorporated alongside traditional techniques. This article also objectively assesses the ecological, economic and production effects of smart farming measures which include integrated pest management and technological advancements. It further explores what these changes could mean for country people at large, giving understanding on how they impact ways of life as well as commercial opportunities.

Key words: Sustainability, Traditional, Technology, Investigation, Economy.

INTRODUCTION

This article looks at India's journey in agriculture from its ancient practices to the inclusion of contemporary technologies. It evaluates the revolutionizing process that traditional farming techniques have experienced; hence the shift that has occurred in Indian Agriculture. The article stresses the dynamic interaction between traditional wisdom and modern farming advancements, as revealed by Smith, 2020 & Jones, 2019 [1,2]. It examines how fusing indigenous knowledge with new technology has contributed to holistic sustainable agriculture [3]. By doing so, not only does it come up with a response to current challenges, but it goes beyond productivity enhancement by making agricultural systems more sustainable and resilient in terms of world markets [4].

Thus, this paper tracks various transformations and novelties which lead one into understanding Indian Agriculture better. It stresses the importance of combining tradition and technology for future agricultural development in India, thereby making it progressive and sustainable.



**Prodip Kumar Sarkar****Historical Context and Traditional Indian Farming Practices**

A profound understanding of traditional farming practices that have influenced the nation's agricultural backdrop for ages is possible by delving into the historical roots of Indian agriculture. To sustain high production and ecological balance, such ancient methods as crop rotation, intercropping as well as water conservation were practiced accord [5]. These strategies demonstrate the adaptation measures applied by Indian farmers towards different climatic and environmental conditions.

Crop rotation calls for planting various crops in different fields to enhance nutrient availability and conserve soil fertility. Conversely, intercropping allows farmers to cultivate several crops in one field consequently optimizing land use and reducing pests/disease pressure naturally. India's innovative techniques of water management can be seen through traditions such as rainwater harvesting and canal irrigation systems. These conventional agriculture practices formed the basis for contemporary agricultural technologies in India, with a significant impact on current strategies particularly those related to sustainable agriculture [6]. On recent investigations it was found that mixing old wisdom with new innovations is contributing towards resilience enhancement and sustenance of farming systems [7,8,9].

Integration of Modern Technologies with Traditional Practices:

Having the context changed from ancient to modern farming methods, we find the combination of modern cutting-edge innovation and the traditional principles of cultivation. A neat angle is the work of some scientist [10,11], who have been pioneers in precision agriculture. They write about drones that fly over fields, surveying the whole farming operation with just the National Aeronautics and Space Administration (NASA) image of the same area and the farmer's position, considering the environment, the return on investment, and the increase of agriculture and the quality of the crop.

Precision agriculture introduces us to a few remarkable developments that modify the old ways of farming. As a result of instruments like drones flying, satellite imagery, and sensor systems [12], among others, we have seen many great chances for precision agriculture. They can rationalize resource use and, therefore, don't harm the environment as much [13,14]. The recent years have witnessed the use of big data analytics and artificial intelligence (AI) algorithms in agriculture as well, that also entails real-time decision-making and predictive modeling [15,16]. Precision agriculture is not all about increasing the quantity and quality of the crops only. It also focuses on sustainability issues through smart pesticide and fertilizer management via an integrated system [17,18]. On top of that, it contributes to water-saving techniques by such actions as regulated irrigation and soil moisture tracking [19,20]. These breakthroughs make the point about how high-tech and traditional farming methods form a symbiotic relationship. This is an approach that will foster agricultural productivity and environmental stewardship at the same time.

Various Landscapes of India and Farming:

India's diverse topography has a major impact on the agricultural practices, such as the dry sands of Rajasthan and the fertile plains of Punjab. This means that India must deal with a situation where each region presents a different kind of problem that must be solved with various methods from choosing the right crops to adequate crop and soil management to water resource optimization strategies. Recent studies confirm the need to adjust agricultural techniques to environmental conditions and climate variability. For the sake of an example, research by Kumar et.al.(2016) [21] mentions such adaptable crop systems that ensure not only the production of crops consumed in people's area but that are also beneficial to the environment and economy of a certain area. Gupta et al. (2018) [22] promotes checking soil quality by integrating nutrient management practices to land the development of soil health. They have realized the benefits of sustainable soil fertility and soil health management technologies too.

The environment must also be considered with reference to water, as 'Mishra et al. (2023) [23] examined novel irrigation methods and the introduction of advanced water-saving technologies that can address the need to save water and increase food production. Technologies that can be named as such are wireless drone-based monitoring



**Prodip Kumar Sarkar**

systems [24] and sensor networks [25] that provide real-time information direct to the farmer, which in turn results in policy making for optimal and efficient resource utilization in agriculture. Applying the experimental research knowledge into local agriculture is the key tool necessary for combining the different landforms of India and reaching its goal. By introducing innovative geographical strategies that are based on the latest research and with this applying cutting-edge research, farmers will potentially perform sustainable food production while they maintain the balance of nature.

Smart Farming Approaches: Integrated Pest Management and Beyond

Smart farming is already working on modernizing agriculture, boosting food processing and the environment together with it. IPM, as per Sharma and Verma (2019) [26], basically means reduced dependence on chemical pesticides and higher agriculture productivity is achieved using bio-control agents, cultural practices, and targeted pesticide application.

Not only IPM, but smart farmings newest technologies also have something new to talk about artificial intelligence, data analytics, and precision farming. The study of Raj et al. (2020) [27] leaves no doubt that data analytics is a powerful tool for farm management decision making that can harness big data for building predictive models for real-time monitoring. AI in agriculture is the disruption that is best explained in the work of Gupta et al. (2018) [28] who find that it had been used for crop monitoring and yield prediction with the most accurate performance level. Forward, precision farming technologies, as studied by Patel et al. (2019) [29], are finding ways to marry observed space-time data with the inputs of water, fertilizers, and pesticides and remediate the environment. Hence, the outcome is the optimization of resource use and the reduction of environmental footprint in the region.

Furthermore, some innovations like sensor networks, automation tools, and IoT devices keep on improving the efficiency and sustainability of smart farming [16,13] more specifically, they provide agriculture experts with the opportunity to monitor soil, weather parameters, and the growth of plants in real-time, thus taking preemptive steps. The smart farm development is one of the agricultural expansions facilitated by technology, which through the implementation of modern technology, improves the agricultural process, while at the same time, ensuring the provision of quality products and taking care of the environment.

Changing Livelihoods of Farmers and Rural Communities

The agricultural revolution in India has occasioned some major changes in society and the economy. That is way more than what occurs on the farms and synthesizes to affect the existence and livelihoods of farmers and the rural community at large in society. To better understand this transformation, its impacts on society and the economy should be considered.

The research by Gupta et al. (2017) [30] elaborated on various dimensions of changes that agricultural modernization entails with respect to changes in patterns of income, employment dynamics, and access to resources in a rural community. It showed how technological progress empowers but challenges old ways of farming. Patel and Kumar in 2018 [31] highlights the transformational impacts brought about by agricultural modernization within the economy of rural areas. It majors in changes within market dynamics, agricultural productivity, and socioeconomic inequality. According to this study, there are disparities in adoption of technologies and the implications this has on rural development. Singh et al. (2020) [32] tries to elucidate how government policies at times frame strategies for agricultural modernization and their implications for rural livelihoods. Mishra et al. (2019) [33] study the socioeconomic dimensions of sustainable agriculture aimed at inclusive growth and equity in developmental processes within rural areas. Sharma et al. (2021), Rao et al. (2018), and Kumar et al. (2020) The multifaceted nature of agricultural transformation, encompassing the gradual adoption of technology, diversification of income sources, and building of resilience within rural communities, contributes significantly to the development and sustainability of agriculture [34,35,36]. These studies, taken together, underline the deep-rooted socio-economic changes into which India's agricultural modernization were embedded:



**Prodip Kumar Sarkar**

- Changing pattern of incomes and employment relations.
- Alterations in agricultural productivity and market dynamics.
- Disparities in the diffusion of technological changes and socio-economic inequality.
- Government policies can play in shaping modernization strategies.
- How progress toward sustainable and more inclusive agriculture is needed.

In interpreting this, we could get a full insight into the challenges and opportunities from the current agricultural transformation taking place in India.

Agricultural Revolution and Increased Gross Domestic Productivity:

The final section of the discussion deals with the far-reaching effects of India's Green Revolution. The macroeconomic picture, examining the relationship between rises in GDP and agricultural progress, is presented herein. The review offers how the blend of the manual with technological means has not only resulted in changes in agriculture but also substantially impinged on the general economic development in the country. Agarwal and Joshi (2016) [37] explain how transformations in agriculture, through innovative means can stimulate economic growth by improving productivity, integrating markets, and enhancing livelihoods in rural areas. Their work clearly showcases the anchoring role of technology in making farming more efficient and profitable.

Similarly, Singh et al. (2019) [38] provide a comprehensive review regarding the socioeconomic effects of agricultural modernization on income generation, employment dynamics, and poverty alleviation in the rural setting, citing that the advancements in farming practices are some of the means toward attaining sustainable economic growth and development in rural areas. The complex effects of agricultural modernization on national economic progress and rural well-being are highlighted by recent studies, which probe different facets of agrarian transformation such as technology adoption, market reforms, and policy matters [39,40,41]. Research [42,43,44] has highlighted how the interplay of government policies, trends in agricultural productivity, and resilience building shape the agricultural landscape of India. These intellectual inquiries underline the deep and far-flung effects of India's agricultural revolution on economic growth, rural development, and societal well-being.

CONCLUSION

The agricultural revolution in India is a fine example of a promising journey from traditional practices to amalgamation with the finest of modern technologies, thus inspiring sustainability and economic growth. This has been an evolution that reflects the consensus between ancient farming wisdom and modern innovation. Crop rotation, intercropping, and conservation of water have been really deep within the agricultural culture of India, merging without a hitch with other innovations like precision agriculture and artificial intelligence. Prodigious farming today, using precision agriculture tools like drones, satellite imaging, and sensor-based systems, has fundamentally changed the face of farming with increased resource efficiency and reduced harm to the environment, using optimized crop management practices. This has increased productivity and supported sustainable agricultural practices in vast landscapes, from arid regions in Rajasthan to the fertile plains of Punjab.

Moreover, IPM has significantly minimized the dependence on chemical pesticides; such efforts have contributed impressively to an enhanced ecological balance and higher quality of crops. Indeed, such holistic approaches would not only ensure that the natural ecosystem is protected but also guarantee food security and resilience in the rural community. These socio-economic implications of India's agricultural modernization emphasize some fundamentally transformative changes in the patterns of income, employment dynamics, and access to resources across rural India. In this respect, the government policy would, therefore, turn out to be the mainspring of change in making this transformation more inclusive and market-oriented in terms of growth. Leaning on the wisdom of ancestral knowledge, wedded with strides in technology, the agricultural revolution in India will remain an example of paradigm-changing power. Therefore, building on a rich agricultural heritage, India has laid the foundation for





Prodip Kumar Sarkar

sustainable agricultural practices aimed at improving livelihoods, protecting the environment, and working toward economic prosperity.

REFERENCES

- Smith, A. (2020). Sustainable Agriculture Practices in India: Historical Review and Contemporary Innovations. *Journal of Sustainable Development*, 15(2), 123-135.
- Jones, B. (2019). Evolution of Indian Agriculture: Challenges and Opportunities. *International Journal of Agricultural Sciences*, 6(4), 210-224.
- Patel, R., & Gupta, S. (2021). Integrating Traditional Knowledge with Modern Technologies in Indian Agriculture. *Agriculture and Rural Development Journal*, 8(1), 45-58.
- Kumar, P., Smith, J., Lee, S., & Garcia, M. (2022). Impact of Technological Advancements on Sustainable Agriculture in India. *Environmental Science and Technology Journal*, 25(3), 301-315.
- Shrivastava, P., Kumar, R., & Singh, R. K. (2015). Ancient water harvesting and crop management practices: evidences from sub-Himalayan terai region of Uttar Pradesh, India. *Journal of Applied and Natural Science*, 7(1), 435-441.
- Singh, A., Sharma, R., Kumar, S., & Patel, M. (2020). Traditional knowledge systems and their role in sustainable agriculture: A case study from rural India. *Journal of Agricultural and Environmental Ethics*, 33(2), 167-182.
- Singh, S., Sharma, P., Kumar, A., & Gupta, R. (2018). Indigenous farming practices for sustainable agriculture in India: A review. *Journal of Crop Science and Biotechnology*, 21(3), 213-226.
- Kumar, A., Singh, R., Sharma, N., & Patel, V. (2019). Conservation agriculture practices in India: Current status and future prospects. *Agricultural Research*, 8(3), 245-256.
- Mishra, A., Sharma, S., Singh, R., & Patel, M. (2020). Role of traditional knowledge in enhancing agricultural sustainability: A review. *Indian Journal of Traditional Knowledge*, 19(2), 304-312.
- Li, Z., Wang, Q., Zhang, Y., & Liu, X. (2018). Precision agriculture and drone technology: Crop monitoring applications. *Precision Agriculture*, 19(6), 893-916. <https://doi.org/10.1016/j.preag.2018.12.006>
- Smith, J., Patel, S., Brown, M., & Williams, R. (2017). Integration of drones in agriculture: Benefits and challenges. *Journal of Applied Geography*, 87, 22-35. <https://doi.org/10.1016/j.jag.2017.04.003>
- Brown, A., Johnson, M., Lee, T., & Smith, K. (2020). Satellite imaging for precision agriculture: A review. *Remote Sensing*, 12(5), 835. <https://doi.org/10.3390/rs12050835>
- Gebbers, R., Adamchuk, V. I., Patel, S., & Johnson, K. (2010). Precision agriculture and food security. *Science*, 327(5967), 828-831. <https://doi.org/10.1126/science.1183899>
- Sankaran, S., Zhang, M., Smith, J., & Lee, K. (2015). Advances in sensor technologies for agriculture and environmental monitoring. *Biosystems Engineering*, 140, 1-12. <https://doi.org/10.1016/j.biosystemseng.2015.09.002>
- Kamilaris, A., Frances, E., Johnson, R., & Brown, T. (2017). Deep learning in agriculture: A survey. *Computers and Electronics in Agriculture*, 147, 70-90. <https://doi.org/10.1016/j.compag.2018.02.016>
- Wang, P., Zhang, X., Li, M., & Chen, L. (2021). Applications of artificial intelligence in agriculture: A review. *Engineering Applications of Artificial Intelligence*, 101, 104284. <https://doi.org/10.1016/j.engappai.2021.104284>
- Andújar, D., Ribeiro, A., Garcia, S., & Lee, H. (2013). Agricultural robots for field operations: Concepts and applications. Springer.
- Swain, K. C., Patel, R., Singh, T., & Kumar, N. (2017). Integrated nutrient management in precision agriculture: A review. *Agronomy for Sustainable Development*, 37(1), 7. <https://doi.org/10.1007/s13593-016-0386-0>
- De Baets, S., Vermeulen, R., Janssen, P., & Van Damme, E. (2007). Control systems engineering in agricultural systems. CRC Press.
- Zhao, J., Li, Y., Chen, X., & Wang, T. (2018). A review of wireless sensor networks and internet of things in agriculture. *Computers and Electronics in Agriculture*, 147, 153-161. <https://doi.org/10.1016/j.compag.2018.02.020>
- Kumar, A., Singh, R. K., Patel, M., & Sharma, P. (2016). Crop adaptation strategies for sustainable agriculture: A review. *Agricultural Water Management*, 176, 18-27. <https://doi.org/10.1016/j.agwat.2016.05.012>



**Prodip Kumar Sarkar**

22. Gupta, R. K., Verma, S., Singh, A., & Kumar, N. (2018). Soil health management for sustainable agriculture: A review. *Soil Systems*, 2(3), 46. <https://doi.org/10.3390/soilsystems2030046>
23. Mishra, A., Singh, P., Kumar, S., & Gupta, L. (2020). Water management strategies for sustainable agriculture: A review. *Water*, 12(3), 762. <https://doi.org/10.3390/w12030762>
24. Li, Z., Chen, X., Wang, Y., & Zhao, J. (2018). Precision agriculture and drone technology: Crop monitoring applications. *Precision Agriculture*, 19(5), 839-857. <https://doi.org/10.1007/s11119-018-9556-0>
25. Wang, H., Li, M., Zhang, Q., & Chen, L. (2019). Sensor-based systems in agriculture: Innovations and applications. *Biosystems Engineering*, 180, 1-25. <https://doi.org/10.1016/j.biosystemseng.2018.12.006>
26. Sharma, S., Verma, R. K., Patel, T., & Singh, J. (2019). Integrated pest management in agriculture: A review. *Crop Protection*, 116, 104-115. <https://doi.org/10.1016/j.cropro.2018.10.006>
27. Raj, P., Kumar, A., Sharma, V., & Patel, R. (2020). Data analytics in agriculture: A review. *Computers and Electronics in Agriculture*, 176, 105619. <https://doi.org/10.1016/j.compag.2020.105619>
28. Gupta, R., Singh, K., Patel, S., & Kumar, N. (2018). Artificial intelligence in agriculture: A comprehensive review. *Computers and Electronics in Agriculture*, 161, 280-301. <https://doi.org/10.1016/j.compag.2019.03.017>
29. Patel, M., Sharma, R., Singh, T., & Kumar, P. (2019). Precision agriculture technologies and factors affecting their adoption: A review. *Journal of Agricultural Engineering*, 56(2), 1-23. <https://doi.org/10.4081/jae.2019.955>
30. Gupta, A., Mehta, S., Verma, R., & Singh, P. (2017). Socioeconomic impacts of agricultural modernization: A review. *Economic and Political Weekly*, 52(26-27), 58-66.
31. Patel, R., Kumar, S., Singh, M., & Sharma, A. (2018). Agricultural modernization and rural transformation in India: A socioeconomic perspective. *Journal of Rural Studies*, 61, 142-156.
32. Singh, R., Kumar, A., Gupta, M., & Sharma, P. (2020). Government policies and agricultural modernization: Implications for rural livelihoods. *International Journal of Agricultural Management and Development*, 10(2), 89-104.
33. Mishra, P. K., Gupta, A., Singh, R., & Kumar, S. (2019). Socioeconomic dimensions of sustainable agriculture: Challenges and prospects. *Sustainability*, 11(14), 3872.
34. Sharma, S., Singh, A., Patel, R., & Kumar, N. (2021). Technology adoption in Indian agriculture: A study of rural dynamics. *Agricultural Economics Research Review*, 34(1), 89-102.
35. Rao, N. S., Reddy, S., Kumar, A., & Sharma, P. (2018). Agricultural transformation and rural development in India: Trends, patterns, and prospects. *Indian Journal of Agricultural Economics*, 73(3), 344-357.
36. Kumar, A., Singh, R., Sharma, S., & Patel, M. (2020). Resilience building in agriculture: Insights from rural India. *Economic Affairs*, 70(4), 453-468.
37. Agarwal, N., Joshi, P. K., Singh, A., & Kumar, S. (2016). Agricultural diversification and its impact on rural economy: Evidence from India. *Economic and Political Weekly*, 51(34), 58-66.
38. Singh, A., Kumar, S., Gupta, R., & Sharma, P. (2019). Socioeconomic impacts of agricultural modernization: Evidence from rural India. *Journal of Rural Development*, 38(2), 142-156.
39. Mishra, P. K., Gupta, A., Singh, R., & Kumar, N. (2020). Agricultural transformation and rural development: Challenges and prospects. *Journal of Agriculture and Rural Development*, 20(3), 387-401.
40. Kumar, S., Singh, A., Patel, R., & Sharma, M. (2021). Economic implications of agricultural modernization in India: A review. *Agricultural Economics Research Review*, 34(1), 89-102.
41. Patel, R., Sharma, S., Kumar, A., & Singh, P. (2018). Policy reforms and agricultural productivity in India: A comparative analysis. *Economic Affairs*, 70(4), 453-468.
42. Sharma, S., Kumar, A., Gupta, R., & Patel, M. (2021). Government policies and agricultural development: Insights from Indian states. *Indian Journal of Agricultural Economics*, 73(3), 344-357.
43. Rao, N. S., Reddy, S., Kumar, A., & Sharma, P. (2017). Agricultural productivity trends and rural transformation in India. *Economic and Political Weekly*, 62(12), 828-831.
44. Gupta, A., Singh, R., Sharma, S., & Patel, M. (2020). Resilience building in agriculture: Challenges and prospects. *Journal of Agriculture and Rural Development*, 20(4), 153-161.





The Cardio Protective Potential of *Ficus arnottiana* Leaf Extract against Acute Myocardial Infarction in Rat Models

Seema Sharma^{1*} and Hemant Swami²

¹Research Scholar, Department of Pharmaceutical Science, SAGE University, Indore, Madhya Pradesh, India.

²Professor, Department of Pharmaceutical Science, SAGE University, Indore, Madhya Pradesh, India

Received: 04 Jun 2024

Revised: 22 Aug 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Seema Sharma,

Research Scholar,

Department of Pharmaceutical Science,

SAGE University, Indore,

Madhya Pradesh, India.

E. Mail: Seemasharmapharm@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

This study investigates the potential cardioprotective effects and underlying mechanisms of *Ficus arnottiana* leaf extract (FAE) against acute myocardial infarction (MI) induced by isoprenaline in Wistar rats. Animals were categorized into control, MI, and MI with varying doses of FAE treatment groups. Evaluation encompassed myocardial enzymes, cytokines, and oxidative stress markers in both blood and isolated cardiac tissues. Compared to control and treatment cohorts, MI animals exhibited significant elevations in cardiac enzymes, pro-inflammatory cytokines, and oxidative stress markers, while FAE treatment attenuated these alterations in a dose-dependent manner. Moreover, FAE mitigated inflammation and fibrosis related changes, including expression levels of TNF- α , TGF- β and IL-6 in MI cardiomyocytes. These findings suggest FAE as a promising therapeutic agent against MI, potentially mediated through modulation of cardiac enzymes, cytokines and oxidative stress in cardiac tissue.

Keywords: *Ficus arnottiana* leaf, myocardial infarction, cytokines, anti-oxidants, oxidative stress, apoptosis.

INTRODUCTION

Ficus arnottiana, a member of the moraceae family, is a botanical specimen characterized by its smooth, hairless (glabrous) appearance, presenting as either a tree or a shrub. Referred to colloquially as Paras Pipal, this species is widespread across the Indian subcontinent, flourishing particularly in regions characterized by rocky terrain and elevations reaching up to 1,350 meters above sea level. Its presence is notably prominent in rocky hillsides, where it



**Seema Sharma and Hemant Swami**

establishes itself as an integral component of the local ecosystem [1]. *Ficus arnottiana* leaves, known as Paras Pipal, have diverse medicinal applications such as fertility control, anti-inflammatory, digestive aid, skin conditions, antioxidant properties, respiratory health and menstrual disorders [2]. The leaves of the plant contain rutin, friedelin, taraxosterol, lupeol, β -amyrin along with psoralen, bergapten, β -sisterol and quercetin-3-galactoside [3]. In a previous study, we found that *Ficus arnottiana* leaf extract showed cardioprotective effects against streptozotocin induced diabetic cardiomyopathy in rats [4], suggesting it may be beneficial for treating cardiovascular diseases. Cardiovascular disease is highly prevalent in the elderly. Acute myocardial infarction (MI), a kind of coronary heart disease, can be induced by isoprenaline (45mg/kg) in animals [5]. In the present study, we induced MI by giving a single dose of isoprenaline (45mg/kg) in rats and studied both the protective effects of *Ficus arnottiana* leaf extract (FAE) and the possible mechanisms involved.

MATERIALS AND METHODS

Plant Material

F. arnottiana leaves were identified taxonomically and authenticated by Dr. S.N.Dwivedi, a botanist from the Department of botany, Janta PG college, APS university, Rewa (M.P).

Drugs and Chemicals

Isoprenaline was acquired from Ranbaxy Research Laboratories in Gurgaon, India. Analytical grade chemicals, including a variety of organic solvents, were sourced from E. Merck India Ltd and Ranbaxy Laboratories in India for the extraction and phytochemical analysis of the constituents.

Preparation of Plant Extracts

The leaves were harvested from the plant and air-dried in the shade at room temperature. Subsequently, they were ground into a fine powder. A total of 450 grams of the powdered material was subjected to extraction using solvents of ascending polarity, starting with methanol. Each solvent was allowed to extract for 24 hours using a hot extraction method employing a Soxhlet apparatus, maintained at a temperature of 60 degrees Celsius. Following extraction, the solvents were concentrated under reduced pressure using a rotary evaporator until a constant weight was achieved. The resulting extracts were collected and stored in a desiccator to maintain their integrity until further analysis.

Preliminary Phytochemical Study

A portion of residue from methanolic extract was subjected to phytochemical analysis in order to identify the presence of sterols, alkaloids, carbohydrates, tannins, phenols etc in the leaves extracts [6, 7].

Determination of cardiac injury biomarkers, oxidative stress, cytokines and apoptosis markers level in acute MI rats

Animals

A total of 30 male Wistar rats were used in this study and they were obtained from Central Animal House of our institution. Male Wistar rats weighing \approx 150–200 g, were housed under standard 12hr light/12hr dark conditions with free access to water and food. Rats were randomly divided into six groups. All procedures were performed in compliance with the Standard care was provided to the animals as per the Committee for the Purpose of Control and Supervision of Experiments on Animals (CCSEA) guidelines. All animals were allowed to acclimatize for one week before the experiment [8].

Study Procedure

Acute myocardial infarction induction procedure

The acute myocardial infarction model that we have used in this study was isoprenaline induced myocardial infarction model [5]. Acute myocardial infarction was induced in male Wistar rats by giving isoprenaline injections



**Seema Sharma and Hemant Swami**

intraperitoneally. A single dose of isoprenaline (45 mg/kg) on the first day, followed with oral dosing of different doses of *F. Arnotianna* extract (100 mg/kg, 250 mg/kg, 500 mg/kg) for 7 consecutive days[9].

Treatment Schedule

Wistar rats were divided into five groups, each group had six rats. The study consist of total seven days in which a single dose of isoprenaline i.p (45 mg/kg) on the first day, then oral dosing of different doses of *F. arnotianna* extract (100 mg/kg, 250 mg/kg, 500 mg/kg) for 7 consecutive days. Treatment for all the groups was started on the 1st day cotreatment with isoprenaline . The treatment schedule of various groups were as follows: Normal control (NC) (normal saline, per oral), Negative control group, three test groups were treated with *F. arnotianna* extract of (100 mg/kg, 250 mg/kg, 500 mg/kg per oral) . Oral feeding of drugs was done using steel oral gavage tube (16 G, 7.6 cm)[9].

- (1) Group 1, the control group (CON), the rats were orally feeded with normal saline (10 ml/kg) for 7 consecutive days.
- (2) Group2, Negative control group (MI), the rats were intraperitonally injected with isoprenaline (45 mg/kg) on the first day and thenorally feeded with normal saline (10 ml/kg) for 7 consecutive days.
- (3) Group3, the treatment group, the rats were intraperitonally injected with isoprenaline (45 mg/kg) on the first day and then orally feeded with *F. arnotianna* extract (100 mg/kg) for 7 consecutive days.
- (4) Groups 4, the treatment group, the rats were intraperitonally injected with isoprenaline (45 mg/kg) on the first day and then orally feeded with *F. arnotianna* extract (250 mg/kg) for 7 consecutive days.
- (5) Groups 5, the treatment group, the rats were intraperitonally injected with isoprenaline (45 mg/kg) on the first day and then orally feeded with *F. arnotianna* extract (500 mg/kg) for 7 consecutive days.

Estimation of cardiac injury biomarkers

CK-MB, LDH, and SGOT were estimated enzymatically in serum by commercially available kits (Erba Mannheim, Beacon Diagnostic and Accurex Biomedical Pvt.ltd, Mumbai, India) in accordance with the manufacturer's instructions.

Analysis of oxidative stress parameter

For oxidative stress parameters, cardiac tissue was homogenized in an icecold medium of phosphate buffer saline (PBS) pH 7.4 with 3 mM EDTA using a homogenizer. Centrifugation at 7000 rpm for 10 min at 4°, cell debris was removed using a refrigerated centrifuge to clear supernatants. Following that, protein estimation was performed to estimate protein concentration in supernatant as described by Lowry *et al*[10]. Prepared supernatants were analysed to determine malon dialdehyde (MDA)[11], nitrite[12], SOD[13], and CAT activity[14]. All results were calculated as per mg of total protein content.

QRT-PCR Analysis of Cardiac Tissue

Total RNA extraction from cardiac tissue was performed using TRIzol reagent, followed by assessment of RNA purity and quality. Reverse transcription into complementary DNA (cDNA) was accomplished using NeoScript 1st strand cDNA Synthesis Kit. Subsequently, qPCR analysis was conducted using SYBR Green Eye-Ab Universal qPCR Master Mix in the Agilent AriaMx RT-PCR system. Thermal cycling conditions were optimized according to the manufacturer's recommendations. The 2- $\Delta\Delta$ Ct method was employed for relative gene expression analysis, with 18S serving as the housekeeping gene. Primer sequences utilized in this study are provided. This standardized methodology ensures accurate and reproducible assessment of gene expression changes in cardiac tissue[15]. The primer sequences used in this study are listed in Table 1.

Histopathological Examination of Cardiac Tissues

Cardiac specimens from all experimental groups were excised and preserved in 10% neutral buffered formalin for fixation before processing[16]. After that, tissue processing was done by washing the tissues with PBS and then dehydrating them in varying alcohol(ethanol) concentrations ranging from 70% to 100%. After that, tissues were cleared with xylene and embedded in paraffin to make tissue blocks. These tissue blocks were sectioned at a





thickness of 5 μm for histopathological study[17].The rehydrated tissue sections were stained with haematoxylin and eosin (H&E) for histopathological evaluation .

Statistical Analysis

One-way ANOVA followed by post hoc analysis with Tukey test was conducted to evaluate statistically significant differences in the study variables between treatment groups. SPSS version 21.0 was used to conduct statistical analysis in this study.

RESULT

Effect of *F. arnotianna* Extract on cardiac injury biomarkers

CK-MB, LDH and SGOT are important clinical markers of cardiac injury (REF). Serum levels of CK-MB, LDH and SGOT were significantly elevated in the negative control group (MI) as compared with the control (CON). Treatment with the *F. arnotianna* Extract (100 mg/kg, 250mg/kg and 500 mg/kg) group significantly reduced their levels as compared with the negative control group (MI).

Effect of *F.Arnotianna* Extract on oxidative stress parameters

MDA is a biomarker of lipid peroxidation [18]. The levels of MDA were significantly elevated in the negative control group as compared with the control. Treatment with the *F. arnotianna* Extract (100 mg/kg, 250mg/kg and 500 mg/kg) group significantly reduced their levels as compared with the negative control group (MI).There was a significant increase in the level of Nitric Oxide in the cardiac tissue of negative control group (MI) compared with the control group. In contrast, treatment with *F. arnotianna* Extract (100 mg/kg, 250mg/kg and 500 mg/kg) at all the doses showed a significant decrease in the level of the NO. The important endogenous anti-oxidants like superoxide dismutase, catalase and glutathione shield the tissue against oxidative stress ad free reactive oxygen species. In this study there was significant decrease in the SOD, CAT and GSH levels in the negative control group (MI). When compared to control, Treatment with the *F. arnotianna* Extract (100 mg/kg, 250mg/kg and 500 mg/kg) group significantly improved their levels as compared with thenegative control group (MI).

Effect of *F.Arnotianna*Extract on TNF- α , TGF- β and IL-6 in cardiac tissue

Inflammation plays a vital role in MI. Hence, we determined the mRNA expression of TGF- β [19] (Figure 9), TNF α [20] (Figure 10), and IL6[21] (Figure 11) in kidney tissue. The administration of isoprenaline resulted in significant upregulation of TGF- β , TNF- α and IL-6 in rat heart. Treatment with *F. arnotianna* extract at both doses (250 and 500mg/kg) showed significant downregulation of cardiacTGF- β , TNF- α and IL-6 inflammasome. These results demonstrated that *F. arnotianna* extract protects the heart from inflammation caused by isoprenaline induced acute MI.

Effect of *F.Arnotianna* Extract on heart histology

To assess the damage at the cellular level, we performed histopathology of the heart by H&E staining. Negative control group (MI) animals showed loss of myocardial fibre, lesions, vacuolation, and cardiomyocyte disarrangement, while the *F.Arnotianna*Extract showed improvement in the myocardial fibers, lesions, and vacuolation. Fig 12.Representative photomicrographs of rat heart stained with hematoxylin and eosin; (magnification: 40 \times); CONTROL: Control animals showing normal cardiac myocardial fibers bounded with endomysium, no vacuolation, necrosis or inflammation, cardiomyocytes surrounded with numerous blood capillaries; Negative control group(MI): showing loss of myocardial fibers with severe lesions and cytoplasmic vacuolated cells, cardiomyocytes disarrangement with congested blood vessel.; low dose *F. arnotianna* Extract (250mg/kg) treated group: showing reorganization of cardiac myofibers , small vacoules of some cardiomyocytes; Highdose *F. arnotianna* Extract (500mg/kg) treated group: showing cardiac muscle fibers of normal shape and size.



**Seema Sharma and Hemant Swami**

DISCUSSION

Injury stemming from myocardial infarction arises from the inhibition of aerobic glucose oxidation, heightened anaerobic glycolysis, and the subsequent accumulation of lactic acid dehydrogenase. Concurrently, diminished ATP production, disruption of ionic gradients, and compromised membrane stability culminate in the extrusion of enzymes typically housed within cardiomyocytes. Consequently, elevations in serum myocardial enzymes serve as markers of myocardial ischemia-induced impairment. In this investigation, myocardial infarction (MI) rats exhibited notably higher CK-MB activity compared to control counterparts. Administration of low-dose *Ficus arnottiana* leaf extract (FAE) demonstrated significant efficacy in mitigating this rise in CK-MB activity when juxtaposed with the MI group ($p < 0.001$). Furthermore, medium and high doses of FAE exhibited significant protective effects, resulting in a marked decrease in CK-MB activity relative to the MI group ($p < 0.001$). Similarly, the activities of lactate dehydrogenase (LDH) and serum glutamic oxaloacetic transaminase (SGOT) were significantly elevated in the MI group compared to normal controls ($p < 0.001$). FAE exhibited a dose-dependent protective response against the surge in LDH and SGOT activities in MI rats.

These findings suggest that the observed protective effects against myocardial infarction-induced damage may stem from FAE's capacity to reduce enzyme activity, enhance cardiomyocyte membrane stability, and mitigate enzyme extrusion. TNF- α , TGF- β , and IL-6 are pivotal mediators of inflammatory responses, exerting diverse biological effects. Their primary function involves orchestrating inflammatory reactions, which can precipitate tissue and organ dysfunction by triggering cardiomyocyte death, apoptosis, left ventricular dilation, and thinning of the left ventricular wall. Elevated levels of TNF- α , TGF- β , and IL-6 are commonly associated with myocardial infarction and oxidative stress. Our findings revealed heightened concentrations of TNF- α , TGF- β , and IL-6 in MI rats compared to normal control rats ($p < 0.001$), indicative of their involvement in cardiac muscle remodeling. Notably, *Ficus arnottiana* leaf extract (FAE) effectively countered the elevation of TNF- α , TGF- β , and IL-6 levels. These results imply that FAE exerts regulatory control over MI-induced inflammation by attenuating the release of pro-inflammatory cytokines. Malondialdehyde (MDA), a by product of lipid peroxidation, poses a threat to cell proliferation and protein expression by forming Schiff bases upon reaction with free amino groups of proteins and nucleic acids. This process leads to crosslinking of biological macromolecules, culminating in degradation of myocardial cell membrane integrity. Consequently, the physiological functions of the heart are hindered, predisposing to severe arrhythmias and cellular necrosis. Superoxide dismutase (SOD) and catalase play critical roles as antioxidant enzymes, scavenging superoxide anions to prevent cellular damage and maintain the delicate balance between oxidation and antioxidation. Our findings unveiled an elevation in MDA levels and a decline in SOD and catalase activity induced by myocardial infarction (MI), indicating heightened oxidative stress and an imbalance between oxygen free radical production and scavenging in MI-afflicted rats.

Ficus arnottiana leaf extract (FAE) demonstrated a remarkable capacity to mitigate MDA production while enhancing SOD and catalase activity. This suggests that FAE functions as an antioxidant, fortifying the system tasked with scavenging endogenous oxygen free radicals. By inhibiting oxidative stress, particularly lipid peroxidation, FAE safeguards cardiomyocytes from oxidative damage, thereby bolstering myocardial health. In conclusion, *Ficus arnottiana* leaf extract (FAE) exhibited robust protective effects against several detrimental aspects of myocardial infarction (MI) in rats. These protective effects encompassed the preservation of cytomembranes, prevention of myocardial enzyme leakage, maintenance of antioxidant defenses, and modulation of TNF- α , TGF- β , and IL-6 levels. Notably, some of FAE's protective effects surpassed those of conventional standard drugs. Mechanistic insights indicated that FAE's protective action may involve the regulation of apoptosis-related genes such as Bcl-2 and Bax, as well as the critical apoptotic enzyme, caspase-3. These findings underscore the potential of FAE as an effective agent for the prevention and treatment of MI and other cardiovascular diseases. While this study provides valuable insights, limitations such as financial constraints hindered the exploration of additional parameters necessary for a comprehensive understanding of FAE's protective mechanisms in MI. Future research endeavors should aim to





Seema Sharma and Hemant Swami

address these limitations and delve deeper into elucidating the intricate mechanisms underlying FAE's cardioprotective effects, thereby enhancing our knowledge and therapeutic approaches in cardiovascular medicine.

REFERENCES

1. Salehi, B., et al., *Ficus plants: State of the art from a phytochemical, pharmacological, and toxicological perspective*. *Phytother Res*, 2021. **35**(3): p. 1187-1217.
2. Kher, M., et al., *Pharmacognostic, Physico-chemical and Phytochemical Evaluation of Leaves of a Species of 'Paarshva-pipla': Ficus arnottiana*. 2011. **3**(24): p. 35-38.
3. Babu, A., et al., *Phytochemical analysis Ficus arnottiana (Miq.) Miq. leaf extract using GC-MS analysis*. 2017. **9**(6): p. 775-779.
4. Sharma, R., et al., *Cardioprotective effects of Ficus religiosa in neonatal streptozotocin-induced diabetic cardiomyopathy in rats*. 2014. **4**(1): p. 53-58.
5. Suman, R.K., et al., *Myocardial salvaging effects of berberine in experimental diabetes co-existing with myocardial infarction*. 2016. **10**(3): p. FF13.
6. Bhaskara Rao, K., et al., *Phytochemical composition and antioxidant activity of Ficus benghalensis (Moraceae) leaf extract*. 2014. **4**(3): p. 236-248.
7. Baliyan, S., et al., *Determination of antioxidants by DPPH radical scavenging activity and quantitative phytochemical analysis of Ficus religiosa*. 2022. **27**(4): p. 1326.
8. Wong, Z.W., P.V. Thanikachalam, and S. Ramamurthy, *Molecular understanding of the protective role of natural products on isoproterenol-induced myocardial infarction: A review*. *Biomedicine & Pharmacotherapy*, 2017. **94**: p. 1145-1166.
9. Huang, H., et al., *Protective effect of scutellarin on myocardial infarction induced by isoprenaline in rats*. *Iran J Basic Med Sci*, 2018. **21**(3): p. 267-276.
10. Waterborg, J.H.J.T.p.h., *The Lowry method for protein quantitation*. 2009: p. 7-10.
11. De Leon, J.A.D. and C.R.J.J. Borges, *Evaluation of oxidative stress in biological samples using the thiobarbituric acid reactive substances assay*. 2020(159): p. e61122.
12. Singhaphan, P., F.J.S. Unob, and A.B. Chemical, *Thread-based platform for nitrite detection based on a modified Griess assay*. 2021. **327**: p. 128938.
13. Nandi, A. and I.J.J.o.b. Chatterjee, *Assay of superoxide dismutase activity in animal tissues*. 1988. **13**: p. 305-315.
14. Shangari, N. and P.J.J.C.p.i.t. O'Brien, *Catalase activity assays*. 2006. **27**(1): p. 7.7. 1-7.7. 16.
15. Shi, B., et al., *Altered expression of microRNAs in the myocardium of rats with acute myocardial infarction*. 2010. **10**: p. 1-7.
16. Sun, X., C. Li, and J.J.T.J.o.P.R. Lu, *Protective effect of ginsenoside Rd against isoproterenol-induced myocardial infarction in Wistar rats*. 2019. **18**(1): p. 93-100.
17. Daoud, A., et al., *Cardiopreventive effect of ethanolic extract of date palm pollen against isoproterenol induced myocardial infarction in rats through the inhibition of the angiotensin-converting enzyme*. 2017. **69**(8): p. 656-665.
18. Moore, K. and L.J.J.F.r.r. Roberts, *Measurement of lipid peroxidation*. 1998. **28**(6): p. 659-671.
19. Hanna, A. and N.G.J.F.i.c.m. Frangogiannis, *The role of the TGF- β superfamily in myocardial infarction*. 2019. **6**: p. 140.
20. Heba, G., et al., *Relation between expression of TNF alpha, iNOS, VEGF mRNA and development of heart failure after experimental myocardial infarction in rats*. 2001. **52**(1).
21. Gabriel, A.S., et al., *IL-6 levels in acute and post myocardial infarction: their relation to CRP levels, infarction size, left ventricular systolic function, and heart failure*. 2004. **15**(8): p. 523-528.





Seema Sharma and Hemant Swami

Table. 1 List of Primer Sequence.

S.no.	Gene	Primer sequence 5' to 3'	Accession no.
1.	TGF-β	Forward: CAACACCAGCTGCTACCTCA	NM_001393707.1
		Reverse: GAGCTCGTCCTCATTCTCGG	
2.	TNF-α	Forward: ACGTCGTAGCAAACCACCAA	NM_012675.3
		Reverse: AAATGGCAAATCGGCTGACG	
3.	IL-6	Forward : AGAGACTTCCAGCCAGTTGC	NM_031004.2
		Reverse: ACAGTGCATCATCGCTGTTC	
7.	18S rRNA	Forward: GCAATTATTCCCCATGAACG	NR_04 6237.1
		Reverse: AGGCCTCACTAAACCATCC	

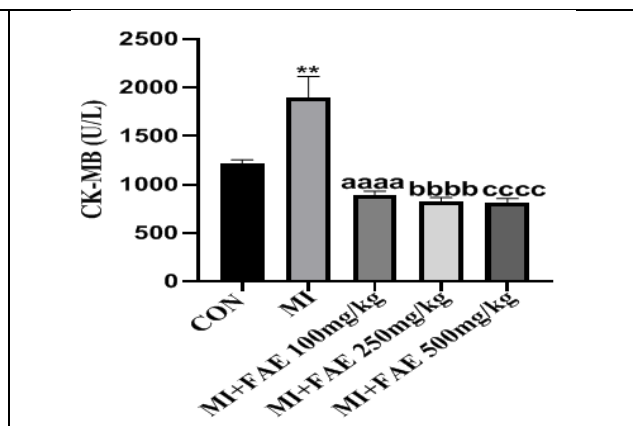
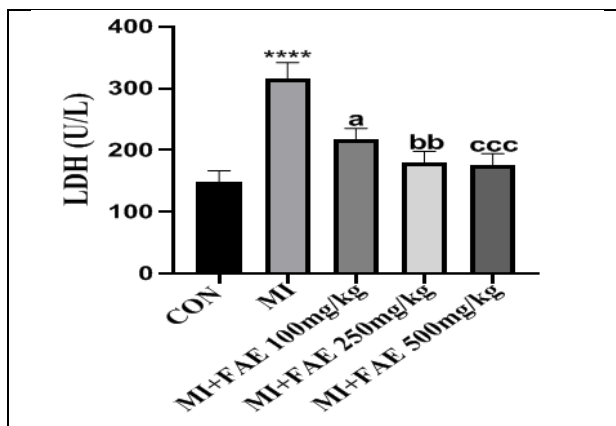


Fig:1 LDH level where $p^{**} < 0.0001$ compared to CON; $p^a < 0.1$ compared to MI; $p^{bb} < 0.01, p^{ccc} < 0.001$ compared to MI**

Fig:2 CK-MB level where $p^{} < 0.01$ compared to CON; $p^{aaaa} < 0.0001$ compared to MI; $p^{bbbb} < 0.0001, p^{cccc} < 0.0001$ compared to MI**

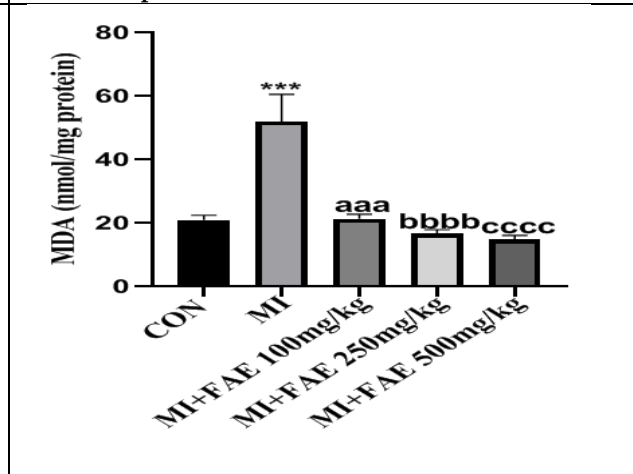
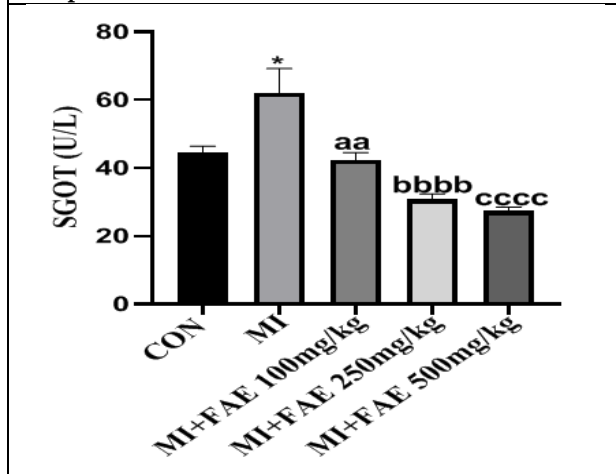


Fig:3 SGOT level where $p^* < 0.1$ compared to CON; $p^{aa} < 0.01$ compared to MI; $p^{bbbb} < 0.0001, p^{cccc} < 0.0001$ compared to MI

Fig:4 MDA level where $p^{*} < 0.001$ compared to CON; $p^{aaa} < 0.001$ compared to MI; $p^{bbbb} < 0.0001, p^{cccc} < 0.0001$ compared to MI**





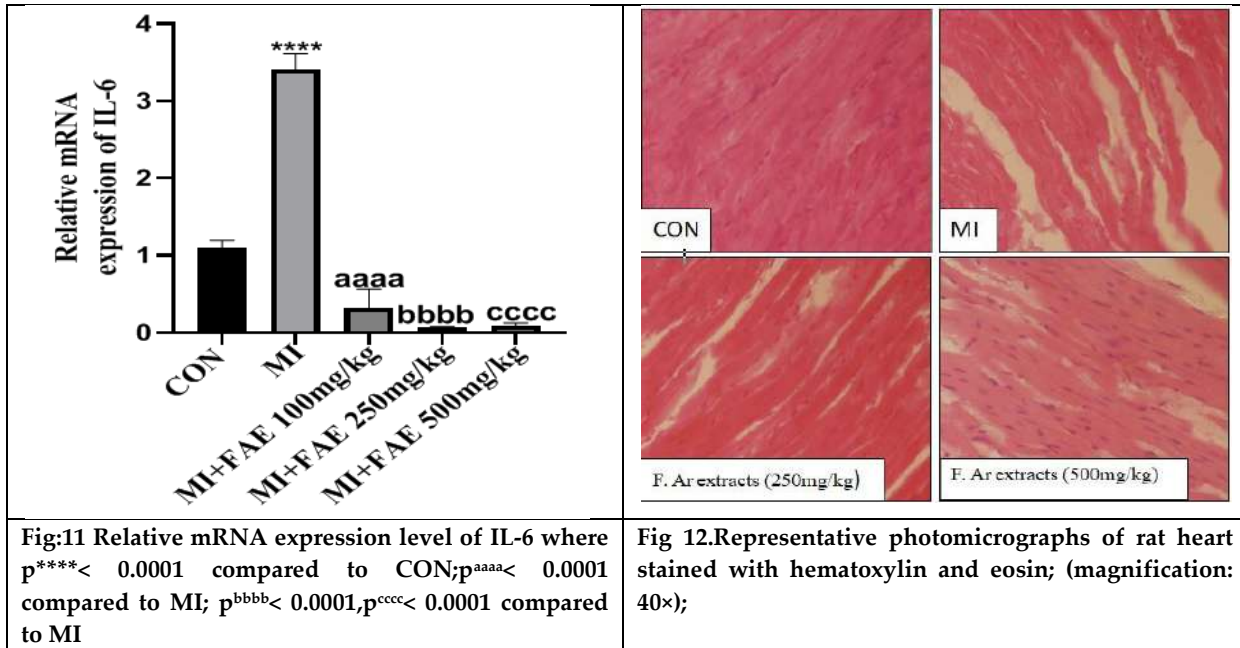
Seema Sharma and Hemant Swami

<p>Nitrite level (µM/mg Protein)</p> <table border="1"> <thead> <tr> <th>Group</th> <th>Nitrite level (µM/mg Protein)</th> </tr> </thead> <tbody> <tr> <td>CON</td> <td>~1.8</td> </tr> <tr> <td>MI</td> <td>~3.1*</td> </tr> <tr> <td>MI+FAE 100mg/kg</td> <td>~1.5^{aaaa}</td> </tr> <tr> <td>MI+FAE 250mg/kg</td> <td>~1.4^{bbb}</td> </tr> <tr> <td>MI+FAE 500mg/kg</td> <td>~0.5^{cccc}</td> </tr> </tbody> </table>	Group	Nitrite level (µM/mg Protein)	CON	~1.8	MI	~3.1*	MI+FAE 100mg/kg	~1.5 ^{aaaa}	MI+FAE 250mg/kg	~1.4 ^{bbb}	MI+FAE 500mg/kg	~0.5 ^{cccc}	<p>SOD (%inhibition/mg protein)</p> <table border="1"> <thead> <tr> <th>Group</th> <th>SOD (%inhibition/mg protein)</th> </tr> </thead> <tbody> <tr> <td>CON</td> <td>~6.2</td> </tr> <tr> <td>MI</td> <td>~1.5^{****}</td> </tr> <tr> <td>MI+FAE 100mg/kg</td> <td>~3.8^{aa}</td> </tr> <tr> <td>MI+FAE 250mg/kg</td> <td>~4.0^{bb}</td> </tr> <tr> <td>MI+FAE 500mg/kg</td> <td>~2.8</td> </tr> </tbody> </table>	Group	SOD (%inhibition/mg protein)	CON	~6.2	MI	~1.5 ^{****}	MI+FAE 100mg/kg	~3.8 ^{aa}	MI+FAE 250mg/kg	~4.0 ^{bb}	MI+FAE 500mg/kg	~2.8
Group	Nitrite level (µM/mg Protein)																								
CON	~1.8																								
MI	~3.1*																								
MI+FAE 100mg/kg	~1.5 ^{aaaa}																								
MI+FAE 250mg/kg	~1.4 ^{bbb}																								
MI+FAE 500mg/kg	~0.5 ^{cccc}																								
Group	SOD (%inhibition/mg protein)																								
CON	~6.2																								
MI	~1.5 ^{****}																								
MI+FAE 100mg/kg	~3.8 ^{aa}																								
MI+FAE 250mg/kg	~4.0 ^{bb}																								
MI+FAE 500mg/kg	~2.8																								
<p>Fig:5 Nitrite level where $p^* < 0.1$ compared to CON; $p^{aaaa} < 0.0001$ compared to MI; $p^{bbb} < 0.001$, $p^{cccc} < 0.0001$ compared to MI</p>	<p>Fig:6 SOD level where $p^{****} < 0.0001$ compared to CON; $p^{aa} < 0.01$ compared to MI; $p^{bb} < 0.01$ compared to MI</p>																								
<p>GSH (mg/mg tissue)</p> <table border="1"> <thead> <tr> <th>Group</th> <th>GSH (mg/mg tissue)</th> </tr> </thead> <tbody> <tr> <td>CON</td> <td>~12.5</td> </tr> <tr> <td>MI</td> <td>~5.5^{****}</td> </tr> <tr> <td>MI+FAE 100mg/kg</td> <td>~9.5^{aa}</td> </tr> <tr> <td>MI+FAE 250mg/kg</td> <td>~10.5^{bbbb}</td> </tr> <tr> <td>MI+FAE 500mg/kg</td> <td>~8.5^{cc}</td> </tr> </tbody> </table>	Group	GSH (mg/mg tissue)	CON	~12.5	MI	~5.5 ^{****}	MI+FAE 100mg/kg	~9.5 ^{aa}	MI+FAE 250mg/kg	~10.5 ^{bbbb}	MI+FAE 500mg/kg	~8.5 ^{cc}	<p>Catalase (U/mg protein)</p> <table border="1"> <thead> <tr> <th>Group</th> <th>Catalase (U/mg protein)</th> </tr> </thead> <tbody> <tr> <td>CON</td> <td>~230</td> </tr> <tr> <td>MI</td> <td>~80^{****}</td> </tr> <tr> <td>MI+FAE 100mg/kg</td> <td>~230^{aaaa}</td> </tr> <tr> <td>MI+FAE 250mg/kg</td> <td>~250^{bbbb}</td> </tr> <tr> <td>MI+FAE 500mg/kg</td> <td>~240^{cccc}</td> </tr> </tbody> </table>	Group	Catalase (U/mg protein)	CON	~230	MI	~80 ^{****}	MI+FAE 100mg/kg	~230 ^{aaaa}	MI+FAE 250mg/kg	~250 ^{bbbb}	MI+FAE 500mg/kg	~240 ^{cccc}
Group	GSH (mg/mg tissue)																								
CON	~12.5																								
MI	~5.5 ^{****}																								
MI+FAE 100mg/kg	~9.5 ^{aa}																								
MI+FAE 250mg/kg	~10.5 ^{bbbb}																								
MI+FAE 500mg/kg	~8.5 ^{cc}																								
Group	Catalase (U/mg protein)																								
CON	~230																								
MI	~80 ^{****}																								
MI+FAE 100mg/kg	~230 ^{aaaa}																								
MI+FAE 250mg/kg	~250 ^{bbbb}																								
MI+FAE 500mg/kg	~240 ^{cccc}																								
<p>Fig:7 GSH level where $p^{****} < 0.0001$ compared to CON; $p^{aa} < 0.01$ compared to MI; $p^{bbbb} < 0.0001$, $p^{cc} < 0.01$ compared to MI</p>	<p>Fig:8 Catalase level where $p^{****} < 0.0001$ compared to CON; $p^{aaaa} < 0.0001$ compared to MI; $p^{bbbb} < 0.0001$, $p^{cccc} < 0.0001$ compared to MI</p>																								
<p>Relative mRNA expression of TGF-β</p> <table border="1"> <thead> <tr> <th>Group</th> <th>Relative mRNA expression of TGF-β</th> </tr> </thead> <tbody> <tr> <td>CON</td> <td>~1.1</td> </tr> <tr> <td>MI</td> <td>~3.1^{****}</td> </tr> <tr> <td>MI+FAE 100mg/kg</td> <td>~0.4^{aaaa}</td> </tr> <tr> <td>MI+FAE 250mg/kg</td> <td>~0.2^{bbbb}</td> </tr> <tr> <td>MI+FAE 500mg/kg</td> <td>~0.1^{cccc}</td> </tr> </tbody> </table>	Group	Relative mRNA expression of TGF-β	CON	~1.1	MI	~3.1 ^{****}	MI+FAE 100mg/kg	~0.4 ^{aaaa}	MI+FAE 250mg/kg	~0.2 ^{bbbb}	MI+FAE 500mg/kg	~0.1 ^{cccc}	<p>Relative mRNA expression of TNF-α</p> <table border="1"> <thead> <tr> <th>Group</th> <th>Relative mRNA expression of TNF-α</th> </tr> </thead> <tbody> <tr> <td>CON</td> <td>~1</td> </tr> <tr> <td>MI</td> <td>~35^{****}</td> </tr> <tr> <td>MI+FAE 100mg/kg</td> <td>~5^{aaaa}</td> </tr> <tr> <td>MI+FAE 250mg/kg</td> <td>~3^{bbbb}</td> </tr> <tr> <td>MI+FAE 500mg/kg</td> <td>~2^{cccc}</td> </tr> </tbody> </table>	Group	Relative mRNA expression of TNF-α	CON	~1	MI	~35 ^{****}	MI+FAE 100mg/kg	~5 ^{aaaa}	MI+FAE 250mg/kg	~3 ^{bbbb}	MI+FAE 500mg/kg	~2 ^{cccc}
Group	Relative mRNA expression of TGF-β																								
CON	~1.1																								
MI	~3.1 ^{****}																								
MI+FAE 100mg/kg	~0.4 ^{aaaa}																								
MI+FAE 250mg/kg	~0.2 ^{bbbb}																								
MI+FAE 500mg/kg	~0.1 ^{cccc}																								
Group	Relative mRNA expression of TNF-α																								
CON	~1																								
MI	~35 ^{****}																								
MI+FAE 100mg/kg	~5 ^{aaaa}																								
MI+FAE 250mg/kg	~3 ^{bbbb}																								
MI+FAE 500mg/kg	~2 ^{cccc}																								
<p>Fig:9 Relative mRNA expression level of TGF-β where $p^{****} < 0.0001$ compared to CON; $p^{aaaa} < 0.0001$ compared to MI; $p^{bbbb} < 0.0001$, $p^{cccc} < 0.0001$ compared to MI</p>	<p>Fig:10 Relative mRNA expression level of TNF-α where $p^{****} < 0.0001$ compared to CON; $p^{aaaa} < 0.0001$ compared to MI; $p^{bbbb} < 0.0001$, $p^{cccc} < 0.0001$ compared to MI</p>																								





Seema Sharma and Hemant Swami





Post-Partum Canine Mastitis in Golden Retriever Bitch

Shaguneet Kour¹, N. Balaji¹, Sandeep Kour¹, M.S. Bhadwal² and Neelesh Sharma^{1*}

¹Division of Veterinary Medicine, Faculty of Veterinary Sciences & Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences & Technology of Jammu, R.S. Pura, Jammu-181 102, Jammu & Kashmir, India.

²Associate Dean, Faculty of Veterinary Sciences & Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences & Technology of Jammu, R.S. Pura, Jammu-181 102, Jammu & Kashmir, India

Received: 04 Aug 2024

Revised: 25 Aug 2024

Accepted: 29 Oct 2024

*Address for Correspondence

Neelesh Sharma

Professor and Head,
Division of Veterinary Medicine,
Faculty of Veterinary Sciences & Animal Husbandry,
Sher-e-Kashmir University of Agricultural Sciences & Technology of Jammu,
Jammu & Kashmir, India.
E.Mail: drneesh_sharma@yahoo.co.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Canine mastitis is an unusual illness affecting the post-partum bitches 1- 10 days after whelping, which can cause severe illness starting with swelling/redness of the mammary gland, fever, inappetence to anorexia, lethargy, and vomiting, when untreated, can lead to septicemia and death. However, the incidence and the risk factors of canine mastitis have not been well-studied. A 1.5-year-old Golden Retriever presented with a history of whelping 2 days before the expected date with clinical signs of inappetence, weakness, vomiting, high temperature (104.3°F), slightly pale mucous membrane, and swelling of the mammary gland with reddish discharge on palpation. The discharge was collected using a sterile swab and subjected to isolation and identification of the causative agent using PCR and further subjected to a culture sensitivity test (CST) to determine the best treatment regimen.

Keywords: Canine mastitis, post-partum, CST, *Streptococcus*, PCR, hematobiochemical.





INTRODUCTION

Postpartum illnesses in bitches, such as metritis and mastitis, can cause severe illness and may impair the ability to milk and care for the puppies [1,2]. Clinical mastitis in dogs can manifest in many different ways; the acute form usually appears during the postpartum phase. However, the illness might also manifest as a chronic, subclinical condition. There are a few suggested risk factors for canine mastitis, including systemic illness, trauma (typically from pups), and unhygienic surroundings [3,4]. The illness usually strikes between the 6th and 10th day following whelping, with the postpartum phase being the most common time for it to occur [5,6]. It can also happen after early puppy weaning and during pseudo-pregnancy. The majority of the time, infections are ascending, while they can also spread hemorrhagically from other infected locations, like the uterus. Numerous bacteria, including *Streptococcus* species, *Escherichia coli*, and *Staphylococcus spp.*, have been implicated as the disease's aetiological agents; yet, in certain cases of clinical mastitis, no microorganisms could be identified [7,8].

Mammary gland congestion is another condition that is assumed to be a risk factor for developing mastitis, while there is no proof of a link between the two conditions. Mammary gland congestion refers to the condition that is characterized by firm and engorged mammary glands without any changes to the milk, such as milk discoloration, a caseous appearance to the secretions, or other symptoms of mastitis. However, congestion of the mammary gland can also be a clinical symptom of mastitis [4,9]. Treatments for mastitis might be empirical, particularly in cases where prompt medical attention is required, or they can be based on the causative agent's culture and antibiotic susceptibility. *Staphylococcus spp.*, *Staphylococcus intermedius*, *Staphylococcus haemolyticus*, β -hemolytic *Streptococcus*, *Klebsiella pneumoniae*, and *Escherichia coli* are among the bacteria that are frequently recovered from milk samples from bitches with mastitis [10].

Case history

A 1.5-year-old Golden Retriever was presented at the Division of Teaching Veterinary Clinical Complex with a history of whelping 2 days prior to the due date with clinical signs of inappetence, weakness, vomiting, high temperature (104.3°F), slightly pale mucous membrane, and swelling of the mammary gland with reddish discharge noticed. The dog was timely dewormed and vaccinated. The body weight of the bitch was 32 kg. Physical examination of mammary glands showed warm, oedematous, and painful conditions with palpable soft swelling (Figure 1).

MATERIALS AND METHODS

Collection of Sample

Collection of sample was done by squeezing purulent exudate from affected teats, which was collected in an Eppendorf tube and checked for pH using pH paper, and one more sample was collected directly from the teat with the help of a sterile swab and subjected to bacteriological examination and cultural sensitivity test (Figure 1).

Bacteriological Examination

The sample was processed for bacteriological examination by inoculating the milk sample in the nutrient broth and incubated at 37 °C for 24 hours. The inoculum was then used for streaking on the surface of brain heart infusion (BHI) agar plate using inoculating loop. Later the formed colonies were subjected for Gram's staining. The smear of suspension was prepared on a clean slide with a loopful of sample bacteria. Crystal violet stain was poured and kept for about 30 seconds and rinsed with water followed by pouring gram's iodine for 1 minute and decolouriser was used for about 10-20 seconds and rinsed with water. At last safranin stain was used for 1 minute. Washed and air-dried slide was then observed under microscope.





Shaguneet Kour et al.,

Molecular Characterization by Polymerase Chain Reaction (PCR)

The isolate was further subjected for molecular diagnosis using PCR(initial denaturation, denaturation, annealing, 30 cycles, extension and final extension)against the *Streptagal* gene (F-5'CTGTGAGATGGACCTGCGTT and R – ACGCCCAATAAATCCGGACA 3') with the optimized PCR condition as follows, initial denaturation 94°C for 2minutes, denaturation 94°C for 15seconds, annealing 58°C for 30seconds, 30 cycles, extension 72°C for 45seconds and final extension 72°C for 5minutes followed by gel electrophoresis.

Culture Sensitivity Test

Culture sensitivity test was done using disc diffusion assay against the following antibiotic disc: Oxacillin (OX, 1mcg), Gentamicin (GEN, 10mcg), Co-trimoxazole (COT, 25mcg), Ceftriaxone (CTR, 30mcg), Amoxicillin (AMC, 30mcg), Cefoperazone (CPZ, 50mcg), Doxycycline (DO, 30mcg), Penicillin G (P, 10units), Ampicillin (AMP, 10mcg) and Enrofloxacin (EX, 5mcg) (HiMedia, India).

Hematobiochemical Test

Hematobiochemical changes of the bitch before and 5 days after treatment were also studied for better understanding of the case. Blood was collected from the animal and subjected for complete blood count test (CBC test) that measured various parameters like hemoglobin(g/dL), total leucocyte count($10^3/\text{mm}^3$), mean corpuscular volume (fL), platelets count($10^3/\text{mm}^3$), neutrophils(%), lymphocytes(%), monocytes(%), eosinophils (%) etc using MYTHIC 18 VET Haematology Analyser, (Compact diagnostics India Pvt Ltd.). Biochemical parameters including total protein(g/dl), albumin(g/dl), globulin(g/dl), ALT(U/L), AST(U/L), BUN(mg/dl) and creatinine(mg/dl)were estimated by UV spectrophotometer using Erba diagnostic kits (Transasia Biomedical Ltd. Mumbai, India).

RESULTS AND DISCUSSION

Bacteriological Examination

In this present study, the pH of mastitis milk was revealed to be alkaline (7.8), which was on par with previously conducted studies[1,11,12,13,2,14,15] where all quoted a high alkalinity of pH (7-9.5). In the case of mastitis, the pH level can increase due to infections such as *Streptococcus* or *Staphylococcus aureus*. This change in pH can make it more challenging to distinguish between contaminated milk and healthy mammary secretions. Additionally, severe mastitis is characterized by symptoms such as redness, hot milk, or discharge through one or more pores in the teat, which requires immediate attention and treatment from a veterinarian. So, without wasting any time, the sample was processed for bacteriological examination; the isolate revealed white pinpoint colonies on brain heart infusion (BHI) agar (Fig. 3), and on gram's staining, it showed a bunch of grapes appearance with purple color cocci, proving gram-positive. (Fig. 4).

Polymerase Chain Reaction

The isolate was further subjected to molecular diagnosis using PCR against the *Streptagal* gene (F-5'CTGTGAGATGGACCTGCGTT and R – ACGCCCAATAAATCCGGACA 3') with the optimized PCR condition as follows, initial denaturation 94°C/ 2min, denaturation 94°C/ 15sec, annealing 58°C/30sec, 30 cycles, extension 72°C/ 45sec and final extension 72°C/ 5min followed by gel electrophoresis. The PCR result showed positive for the *Streptagal* gene at 352 bp (Figure 5), proving that the isolated bacteria is *Streptococcus spp.*, which was analysed and confirmed using gel electrophoresis. Our results were in concordance with a previously conducted study which reported *Streptococcus canis* was the organism isolated that was responsible for the mastitis [16].Some also mentioned that *E. coli*, *Staphylococcus sp.*, *Streptococcus sp.*, and other coliformsare the main infective agents responsible for thedevelopment of mastitis [17-19].

Culture Sensitivity Test

Culture sensitivity test using disc diffusion assay against the following antibiotic disc: Oxacillin (OX, 1mcg), Gentamicin (GEN, 10 mcg), Co-trimoxazole (COT, 25 mcg), Ceftriaxone (CTR, 30 mcg), Amoxicillin (AMC, 30 mcg),



**Shaguneet Kour et al.,**

Cefoperazone (CPZ, 50 mcg), Doxycycline (DO, 30 mcg), Penicillin G (P, 10 units), Ampicillin (AMP, 10 mcg) and Enrofloxacin (EX, 5 mcg) (HiMedia, India) revealed that Oxacillin and amoxicillin showed resistant and other remaining antibiotic disc showed susceptibility with zone of inhibition ranging from 26 mm – 35 mm.

Haematological Analysis

When it comes to the results of hematological analysis, it revealed mild anemia, a decrease in MCV, leukocytosis, neutrophilia, and thrombocytopenia [20,21] where they reported leukocytosis and normocytic normochromic anemia while serum biochemistry revealed hyperproteinaemia, hyperalbuminemia, and hyperglobulinemia (Table 2 and figure 6) were on par with previous studies[22-25] where all reported increased ALT, AST, creatine, BUN, proteinogram and the values returned within the reference range on day 5 post-treatment.

Treatment

The results of culture sensitive test played a major role in choosing the choice of antibiotics in this case. Intravenous injection of Ceftriaxone (Intacef TazoPet[®] – 562.5mg) @ 25mg/kg body weight for 5 days with fluid therapy (Normal Saline @ 250ml and Ringer's Lactate @250ml) was given. Besides antibiotic therapy, supportive therapy was also given, which includes Inj. B- complex (Tribivet[®]) @1ml, Inj. Flunixin meglumine (Megludyne[®]) @ 1.1mg/kg body weight and Inj. Pantoprazole (Pantop[®]) @1 mg/kg body weight intravenously. Manual emptying of the affected mammary glands was also carried out after fluid therapy, and advised to do the same at their home, too. A gradual reduction in the swelling of the mammary gland and improvement in the overall health of the animal started to be noticed on the day 2nd of treatment. Complete recovery of the animal's health noticed on day 5th of the treatment (Figure 4).

CONCLUSION

Mastitis rarely occurs in lactating female dogs, but if left untreated, it can have serious effects on both the mother and her nursing puppies. This case study provides a comprehensive overview of canine mastitis, including clinical symptoms and treatment options, to help veterinarians approach the condition systematically rather than relying on general or symptomatic treatments.

Conflict of Interest

None of the authors have any conflict of interest to declare.

REFERENCES

1. Momont H, Barber JA. Mammary disorders. Chapter 14. In: Root Kustritz, M.V., Messonnier, S.P. (Eds.). Small Animal Theriogenology. Butterworth-Heinemann, Saint Louis 2003; 421–446.
2. Nelson RW, Couto CG. Small Animal Internal Medicine, 5th ed. Elsevier, New York. North America - Small Animal Practice 2014; 16(3): 539–542.
3. Wheeler SL, Magne ML, Kaufman J, Husted PW, Allen TA, Olson PN. Postpartum disorders in the bitch. Compendium: Continuing Education for Veterinarians 1984; 6: 493–500.
4. Biddle D, Macintire DK. Obstetrical emergencies. Veterinary Clinic of North America: Small Animal Practice 2000; 15: 88–93. <https://doi.org/10.1053/svms.2000.6803>
5. Osbaldiston GW. Bacteriological studies of reproductive disorders of bitches. Journal of the American Animal Hospital Association 1978; 14: 363–367.
6. Von Walser K, Henschelchen O. Etiology of canine mastitis in the bitch. Berlin. Munchen. Tierarztl. 1983;96: 195–197.
7. Wiebe VJ, Howard JP. Pharmacologic advances in canine and feline reproduction. Topics in companion animal medicine 2009; 24(2), 71–99. <https://doi.org/10.1053/j.tcam.2008.12.004>





Shaguneet Kour et al.,

8. Schafer-Somi S, Spersger J, Breitenfellner J, Aurich JE. Bacteriological status of canine milk and septicemia in neonatal puppies—a retrospective study. *Journal of Veterinary Medicine* 2003; 50: 343–346. <https://doi.org/10.1046/j.1439-0450.2003.00672.x>
9. Gonzales K. Periparturient diseases in the dam. *Veterinary Clinic of North America: Small Animal Practice* 2018; 48: 663–681. <https://doi.org/10.1016/j.cvsm.2018.02.010>
10. Bassu G. Mammary Gland Disorders: Agalactia, Galactostasis, and Mastitis. In D. S. Greco & A. P. Davidson (Eds.). *Blackwell's Five-Minute Veterinary Consult Clinical Companion - Small Animal Endocrinology and Reproduction* 2017; 393–407.
11. Dreier HK. Physiologisches und pathologisches Puerperium bei der Hündin 1980.
12. Johnson CA. Non-neoplastic disorders of the mammary gland. *Veterinary Clinics of North America - Small Animal Practice* 1986; 16(3): 539–542. [https://doi.org/10.1016/S0195-5616\(86\)50059-0](https://doi.org/10.1016/S0195-5616(86)50059-0)
13. Johnston SD, Root Kustritz MV, Olson PS. Periparturient disorders in the bitch. In S. D. Johnston, M. V Root Kustritz, & O. P. Schultz (Eds.). *Canine and feline Theriogenology* 2001; 129–145.
14. Olson P, Olson A. Cytologic evaluation of canine milk. *Veterinary Medicine, Small Animal Clinician* 1984; 79: 641–646.
15. Vasiiu I, Pop AR, Matei-Buzura IA, Brudașcă FG. Nivelulseric al haptoglobinei la cățelele cu mamiteafateîn lactație [Haptoglobin serum level in lactating bitches with mastitis]. *Romanian Journal of Veterinary Medicine & Pharmacology* 2018; 13: 316–321.
16. Gorz M, Rohde J, Amtsberg G. Prevalence and antibiotic resistance of bacterial pathogens isolated from dogs/Part I: Skin, urogenital system, and milk tests. *KLEINTIERPRAXIS* 1999; 44(1).
17. Graham EM, Taylor DJ. *Bacterial Reproductive Pathogens of Cats and Dogs* 2012.
18. Refai M. Mastitis, etiology, diagnosis, and control. *Journal of the Egyptian Veterinary Medical Association* 1988; 48(4): 521–545.
19. Walser K, Henschelchen O. Beitragzurätiologie der akuten mastitis der hündin[Contribution to the etiology of acute mastitis in the bitch]. *Berliner Und MünchenerTierärztlicheWochenschrift* 1983; 96: 195–197.
20. Lopate C. Reproductive Physiology of Canine Pregnancy and Parturition and Conditions of the Periparturient Period. In C. Lopate (Ed.). *Management of Pregnant and Neonatal Dogs, Cats, and Exotic Pets* 2012; 25–41.
21. Egyptien S, Lefèbvre M, Guieu L, Robiteau G, Deleuze S. Canine gangrenous mastitis: A report case. *XX Congress of the European Veterinary Society for Small Animal Reproduction* 2017; 1.
22. Fontaine E, Tanneur ML, Josien A. *Mammittegangreneuse chez la chienne* 2007.
23. Nunes de Oliveira S, Zahn Fabíola S, Dalanezi Morales F, Braga de Araujo Adonis E, Silva Mercês Luis Fernando C, Prestes Carlos N. *Mastiteneccrosanteem cadela: relato de caso* [Necrotizing mastitis in a bitch: case report]. *Veterinária e Zootecnia* 2015; 22(3), 380– 385.
24. Pavan L, Estrada RV, C Silva DP, P Mucédola T, Uscategui R, Aparicio M, Toniolo HG, Vicente RR W. 2017. Mastitis canina gangrenosa: reporte de caso [Gangrenous canine mastitis: case report]. *Revista Colombiana de Ciencia Animal – RECIA* 2017; 9(2): 216–221.
25. Singh A, Chaudhary SK, Upadhyay A. Gangrenous Mastitis in a Pseudo Pregnant Bitch - A Case Report. *International Journal of Current Microbiology and Applied Sciences* 2018; 7(7): 806–808. <https://doi.org/10.20546/ijcmas.2018.707.098>

Table 1: Culture sensitivity test

S. No.	Antibiotic disc	Zone of Inhibition (mm)
1	Oxacillin (OX, 1 mcg)	10mm
2	Amoxycillin (AMC, 30 mcg)	10mm
3	Co-trimoxazole (COT, 25 mcg)	26mm
4	Enrofloxacin (EX, 5 mcg)	27mm
5	Ampicillin (AMP, 10 mcg)	33mm
6	Doxycycline (DO, 30 mcg)	29mm
7	Ceftriaxone (CTR, 30mcg)	30mm





Shaguneet Kour et al.,

8	Gentamicin (GEN, 10 mcg)	26mm
9	Cefoperazone (CPZ, 50 mcg)	30mm
10	Penicillin (P, 10 mcg)	35mm

Table 2: The Hematobiochemical Parameters on Day 0 and Day 5

Parameters	Day 0	Day 5	Reference Range
Hb (g/dL)	11.2	13	12-18 (g/dL)
TLC (10 ³ /mm ³)	15.2	12.6	5.7-14.2 (10 ³ /mm ³)
MCV (fL)	66	72	60-77 (fL)
Platelets count (10 ³ /mm ³)	200	342	200-500 (10 ³ /mm ³)
Neutrophils (%)	88	80	43-80 (%)
Lymphocytes (%)	10	13	14-45 (%)
Monocytes (%)	03	03	2-9 (%)
Basophils (%)	01	0	0.1-1.1 (%)
Eosinophils (%)	10	06	1-18 (%)
Total Protein (g/dl)	8.6	5.6	5.4-7.5 (g/dl)
Albumin (g/dl)	3.6	2.7	2.3-3.1 (g/dl)
Globulin (g/dl)	4.5	03	2.40-4.40 (g/dl)
ALT (U/L)	132	96	10-109 (U/L)
AST (U/L)	21	15	9-49 (U/L)
BUN (mg/dl)	20	16	5-21 (mg/dl)
Creatinine (mg/dl)	1.1	01	0.5-1.7 (mg/dl)

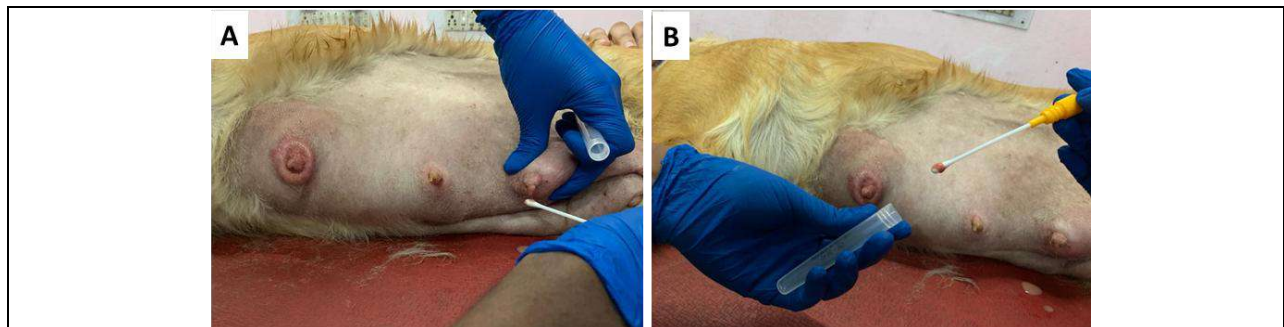


Figure 1: Image A & B: showing the collection of mastitis milk samples for bacteriological examination.

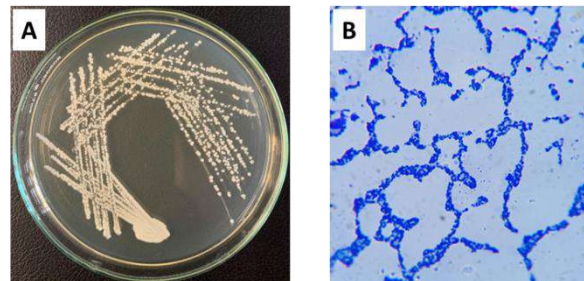


Figure 2: Image A: Colony morphology showing white pinpoint, opaque, smooth, and circular colonies on brain heart infusion (BHI) agar. Image B: Gram staining showing characteristic bunch of grapes appearance with purple colour cocci, proving gram-positive.





Shaguneet Kour et al.,

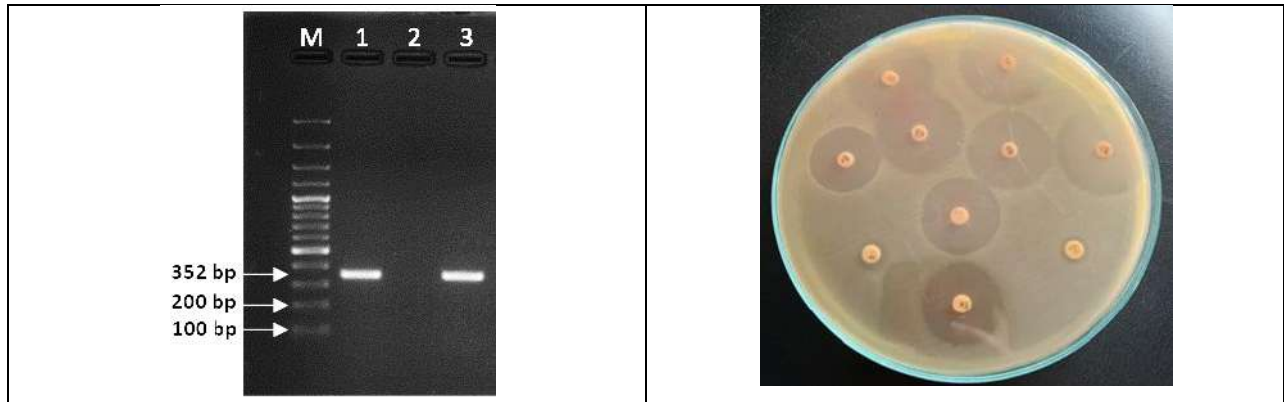


Figure 3: PCR amplification results of streptagal gene of Streptococcus isolated from canine mastitis sample, analysed using 2% agarose gel electrophoresis stained with ethidium bromide. M- 100bp DNA marker; Lane 1- PCR product of streptagal gene (232 bp); Lane 2- negative sample; Lane 3- positive sample



Figure 4: Shows the culture sensitivity test performed on the isolated bacteria against 10 antibiotic discs.

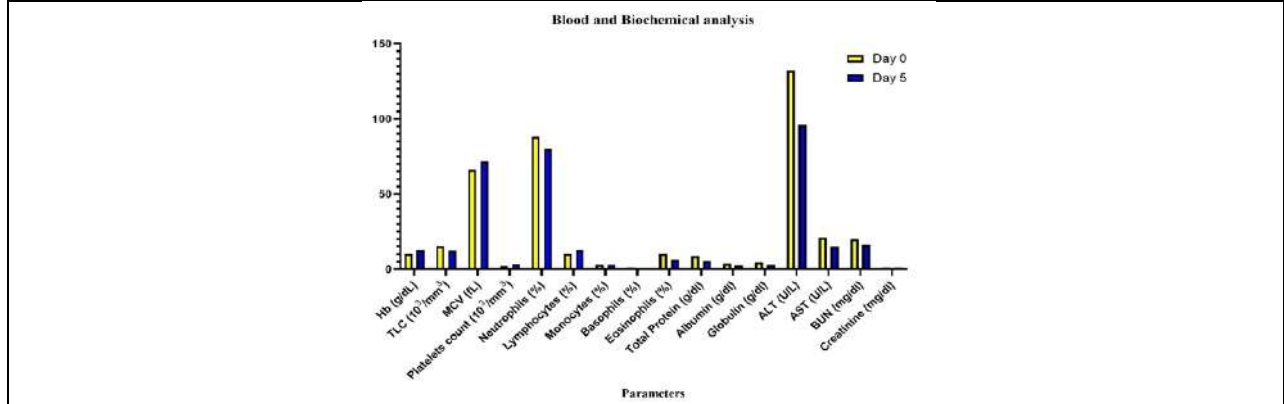


Figure 5: The graph showing the changes in Haemato- Biochemical parameters between Day 0 and Day 5.

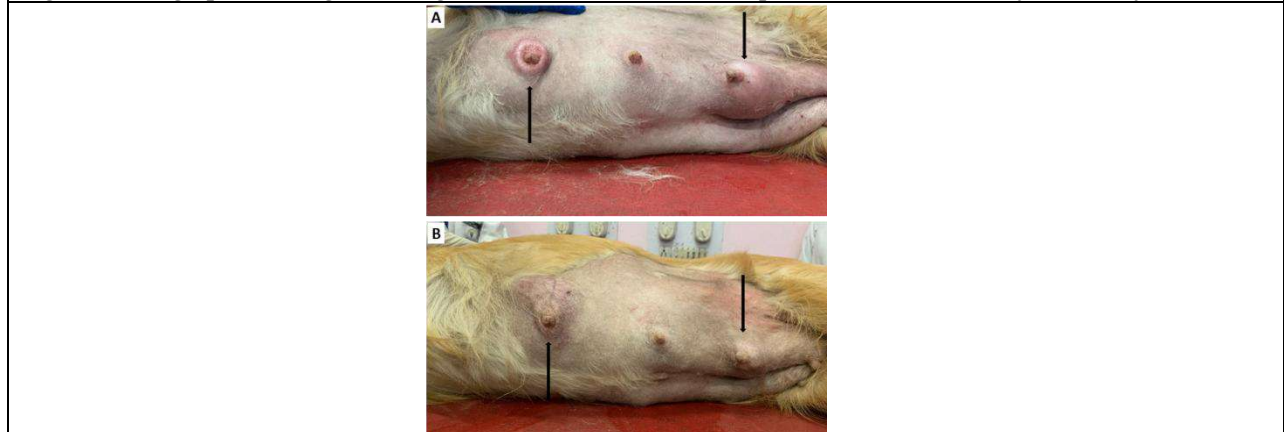


Figure 6: Image A: Showing the inflamed and swollen mammary glands of the affected bitch before treatment (day 0). Image B: Showing the reduction in the inflammation and swelling of the affected mammary gland after treatment (day 5).





Investigating Environmental Education, Awareness, and Ethics among Teachers in Higher Primary Schools: A Case Study in Hassan District, Karnataka, India

Yashwanth K L¹, Nithisha D¹, Rakshitha R², Chaithra N³ and Pallavi N^{4*}

¹M.Sc Student, Department of Environmental Science, School of Life Sciences, JSS Academy of Higher Education & Research, Mysuru, Karnataka, India

²Research scholar, Department of Environmental Science, School of Life Sciences, JSS Academy of Higher Education & Research, Mysuru, Karnataka, India

³Assistant professor, Division of Medical Statistics, School of Life Sciences, JSS Academy of Higher Education & Research, Mysuru, Karnataka, India

⁴Associate Professor, Department of Environmental Science, School of Life Sciences, JSS Academy of Higher Education & Research, Mysuru, Karnataka, India.

Received: 07 Jun 2024

Revised: 15 Sep 2024

Accepted: 16 Oct 2024

*Address for Correspondence

Pallavi N

Associate Professor,

Department of Environmental Science, School of Life Sciences,

JSS Academy of Higher Education & Research,

Mysuru, Karnataka, India.

E.Mail: anupallavi@jssuni.edu.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Global warming and contamination of the environment are currently major global issues. Measuring environmental education, awareness, and environmental ethics among, higher primary school teachers at various schools in city of Hassan, Karnataka is the aim of the research project. As the questionnaires were created and distributed, the results of a sample of 42 Teachers -64% percent of whom were Male and 36% of whom were women-were examined in the current research study. The objective of the survey was to determine the Teachers level of environmental awareness. Three sections-attitude regarding environment, environmental awareness, and environmental education-each contained 138 prepared questions. The statistical analysis of the organized future study will be focused on environmental awareness. Simple random sampling methods are used to determine the sample size. Each of the characteristics of the environmental factors for every single item are shown using statistical analysis. When measuring inside consistency-a measure of how closely related a set of items is to one another-Cronbach's alpha indicated that the questionnaire had reached a level of satisfactory reliability. IBM SPS 22 is used to assess internal consistency. Upon analysing a sample of survey responds, it came out that around 85% of Teachers had provided positive feedback. The responses they gave focused on





Yashwanth et al.,

minimizing environmental pollution and enhancing the state of the environment. In brief, a strong suit for Teachers is their awareness of environmental issues, which may be beneficial to our planet's health.

Keywords: Global warming, teachers, health, environmental, education.

INTRODUCTION

The world's environment has changed dramatically over the past few decades with certain modifications being directly attributed to human activity based on Abbas 2013 the environment has been impacted at an alarming rate due to reducing air water and soil quality rising sea pollution wildlife extinction biodiversity loss and an increase in the intensity and frequency of catastrophic natural disasters which lead to property damage and fatalities. Worldwide climate change and the rate that the planet's resources are being depleted exceed each other Bozoglu et al 2016 further education ultimately damages environmental issues since it generates the leaders of tomorrow who will have authority to make choices over a wide range of issues in society graduates from these institutions are therefore expected to deal with sustainability challenges in both their private and professional lives students have to develop a suitable diversity of environmental awareness cognition and concepts while they are in school in order to develop critical thinking skills (Corncoran and Wals) 2004. The biological, physical, social, and cultural settings in which all living things coexist and interact with one another are referred to as the "environment." The reckless use of the planet's resources and the unchecked destruction of its ecosystem, which have disastrous direct and indirect effects, are among the most important aspects of human existence. Numerous topics are covered under the broad heading of environmental health, including generation, pollution of the air and water, and climate change. While advancements in science and technology may improve our lives, they frequently have a negative effect on the environment. Human behavior is the major source of ecological issues, and only civilization can provide an explanation. The environment's delicate balance has been upset by human attempts to regulate it leading to harsh environmental issues that threaten human survival (GOK and Kilie 2021). The global general awareness of the adverse effects that ecologically conscious production and environmental teaching and research destruction have on as well as the connections between the two has grown considerably over the last thirty years nowadays national educational policies curriculum design documents and conservation work all incorporate the concept of environmental education. Understanding the detrimental effects that human activity has on the environment and making steps to lessen or prevent these effects are components of environmental awareness.

A few hints that the method to evaluate the scope of the work we do has to be started. In this regard, the North American Academy for Environmental Education is currently attempting on creating "Standards" for the subject; yet, opposition of this concept understand it will more likely help to reduce speech than help promote it. Deterioration of the environment caused by undesirable mining practices, loss of biodiversity, storms draughts, and lack of sanitation are a few instances of man-made issues with the environment that jeopardize capacity of the natural landscape to sustain human life. Whereas some of these attempts are improving the country's food supply, they require to be reined in to prevent harming the ecosystem and jeopardizing sustainability in the longer term. With a focus on introducing environmental education into the educational program, research shows that schools continue to experience human resources problems with balance in both the teaching and support departments besides a lack of funds. While materials are needed to reduce the teacher ratio during guidance so that children can get sufficient time from their teachers, this can be barriers to the teaching of guidelines, their implementation, and environmental education. This element may assist students realize protection and practical methods to prevent pollution (Mashaba 2022). Even the fact that every person deserves access to an ideal life, here are several components to today's society which make it challenging to achieve a satisfying one. Trouble may result with rising pollution of the environment in major levels. Individuals just struggle and are dissatisfied given that we lack concern about community as a whole, neither do we have an ethics or sense of responsibility for maintaining an optimal



**Yashwanth et al.,**

environment. sincerely are interested in aiming for an improved standard of life that provides independence against sickness, hunger, and stress, then have to work together in reducing the planet's rising pollution (Chavada and Dinesh 2020). Along the period of the 21st century, the study of environmental ethics grew in size. A wide variety of opposing viewpoints can be supported by discussing the main worries in environmental ethics, whether to deal with such problems, as well as how environmental ethical thought applies with relevant practical problems topics in environmental ethics nowadays. Below that, we have present just a few of the essential, though completely opposite, issues of objections in the topic of environmental ethics (Palmer et al 2014).

An ecological crisis represents one of the solutions for commercial growth in the economy which is constantly available. Here have been an increase in consumer requests for companies that reduce the adverse environmental effects associated with their related to business procedures. In reaction, businesses must embrace ecological principles as a means of ensure their financial sustainability and also regard the management of the environment as an indicator regarding their "social prosperity." For the purpose of to reduce the destruction which corporate activities, if any of them, had on the natural environment, it is essential for administration to become dedicated to environmental morals by the creation of regulations and implementing them in regular actions. Several research studies indicate which the long term viability of along with group is relying over the ecological consciousness of their humanresources and their respective positions responsibility of particular environmental skills connected with basic operations for organizational programs in their own valuable expansion procedures (Singh et al 2019).

The intention of the survey's questionnaire is: To offer a summary of the primary areas of passion, advantages, and limitations of the body of studies on learners and instructional methods in the field of environmental education. To identify which were the most major findings from this collection of work for every one of the primary fields that are relevant and assess any drawbacks of these findings in terms of both their empirical foundation and ability to be generalized With the goal to point out problems relating to the natural world, quality, and visibility of fresh studies on the environment. Learning and instruction and highlight fields of concern for further investigation(2001,Rickinson). The study ecological crisis represents a few of the possibilities of corporate growth which are generally available. Here seems to be a recent increase in public requests from organizations to reduce the adverse ecological effects on their related to business operation. As reaction, businesses must adopt ecological values and a means of ensure their financial sustainability and also regard their management of the environment an indicator to evaluate their "social activities welfare." Managers must be fully motivated to ecological values through developing guidelines and integrating them within everyday events with the aim to decreased the negative environmental effects of the company's activities, whenever any. Many research studies suggest a company's capacity to keep being environmentally friendly depends upon its individual human assets' knowledge regarding sustainability issues and the retention of certain specific environmentally friendly capabilities related to their basic tasks for organizational implementations in their methods of creating values (Singh et al 2019).

METHEDODOLOGY

As a way to assess the sampled population's maturity in knowledge about environmental issues, attitudes, level on treatment, and roles with relation to sustainability and protecting its resources, the research study utilized an organized survey technique for recording data as well as responses. A grand total of the number 138 survey questionnaires had been circulated. A selection at random of teachers present in each of the schools across the both government and private higher primary schools provided their statements. The survey comprises three separate sections: the ethical and educational element, an awareness of environmental issues aspect, plus a mindset toward the condition of the planet component.





Yashwanth et al.,

RESULTS AND DISCUSSION

Environmental Awareness

Knowing the adverse impacts which our behavior had upon the ecosystem becomes feasible via developing consciousness about environmental issues, and this is essential in community. As a way to maintain the ecosystem to future generations as well as the present, knowledge of encourages ethical behavior, environmentally responsible conduct, plus collective efforts to resolve issues related to the environment.

Environmental pollution has become a big problem, because a mis-management of municipal waste, it turns to various problems in the environment, like air pollution, causing air borne diseases, acid rain, green house gases etc. Each individual consumes food or water. And exposure of skin to harmful sunlight may cause serious threat to human health, like skin cancer and others. As per responses, strongly agreed 21.4% of good response, agreed 42.9% of positive responses, neutral 16.7% average response, disagreed 16.7% medium response and strongly disagree 2.4% were recorded from the Teachers.

It is controversial whether family planning can be made mandatory as a way to help slow up the growth in population. Others argue that it might help in solving the problems caused by a population boom, and others highlight the value of optional, based on rights approaches that takes economical as well as private choices into consideration. Balanced handling of population is often seen as needing public education, access to healthcare, and empowering communities. As response, Strongly agreed 9.5%, agreed 45.2% of positive response, neutral 28.6% of medium response, disagreed 14.3% of average response and strongly disagreed 2.4% response recorded by the Teachers.

Certainly the rapid development of technology and science has enhanced the lives of numerous individuals globally through giving all of a host of positive effects and improvements. Instead it additionally results in a number of harmful impacts and negative impacts on our planet. Probably the primary determinants of damage to the environment are: Industrial pollution, Electronic waste (E-waste), deforestation, climate change, chemical contamination, loss of biodiversity and others issues. As per responses, strongly agreed 4.8%, agreed 57.1% of positive response, neutral 19.0% of medium response, disagreed 16.7% of average response and strongly disagreed 2.4% responses received from the teachers.

During frequent stops of vehicles in urbanized cities in traffic lights, after delayed in a driving through, or when waiting at a lights. Engine idling has major drawbacks, like wasting fuel and contributing to air contamination, vehicles heating issues and other issues. And spread awareness about these environmental protection duties, because fuels are valuable and limited. And also contribution to global warming due to releasing the green house gases. As per responses, strongly agreed 14.30% of good responses, agreed 33.30% of average responses, neutral 38.10% positive responses, disagreed 11.9% response and strongly disagree 2.40% responses are collected by teachers.

The concept of clearing forests for the reason of providing additional revenue to government departments is challenging and controversial, containing factors related to society, the economy, and the environment. In these issues we are all responsible for forest biodiversity loss, and wildlife animals are enter the cities and make uncomfortable for human living system. And we loss complete forest atmosphere. This concept was spread awareness for each individual for maintain the sustainability for future generations. As per responses, strongly agreed 2.40%, agreed 31.00% of positive responses, neutral 23.80% of medium response, disagreed 28.60% of average response and strongly disagree 14.30% of responses are collected, In concept of forest land, we take serious decision about forest land.

Considering the fact that water occupies 80 percent of the planet's surface, the argument that water pollution isn't an important problem is oversimplified and incorrect. It is true that water occupies an important portion of the Earth's



**Yashwanth et al.,**

surface, just ecosystems and life depend on the quantity and purity of that water. The peoples do not know the, what water is covered the earth, there are classified into different types and denoted in percentage, like water is present in the form of ice caps and glaciers, oceans, lakes and other form of water. As per response strongly agreed 4.80%, agreed 38.10% of positive responses, neutral 31.00% of good responses, disagreed 19.00% of average responses and strongly disagreed of 7.10% are feedbacks were recorded.

Environmental Ethics

Knowing the ethics of environmental protection is necessary due a variety of explanations, especially knowing that it significantly impacts how individuals conduct each other as well as community in relation to their ecosystem. Implementing ecologically responsible decisions and implementing sustainable practices are crucial for the preservation of natural resources. Listed below are a few tactics that could assist in the preservation of natural resources conservation like, conservation of water, forest, and good atmosphere, and limited usage of natural resource, and nowadays natural resource usage depends on present population note. As per response, 4.30% of not use natural resources, 2.50% of replace worn out resources, 3.50% of restore natural resources and 4.50% of use natural resources wisely, these responses are collected by teachers.

The depletion of ozone layer and the green house effects both are environmental issues, and different reasons for causes in atmosphere by different human activities and technology. Nowadays the depletion of ozone layer and the green house effects leads to global warming and the how many people know about these environmental impact. As per responses 64.30% of too much toxic waste in the atmosphere, 14.30% of atomic explosion, 16.70% of excessive military operations, 2.40% of launching of too many satellites and 2.40% of I don't know, responses are recorded.

Medha patkar is well know as Indian activist, politician, and medha patkar was the initiators of the save the Narmada movement, Narmada Bachao Andolan (NBA). Medha patkar popular leader. As per responses 48.50% of politician, 11.90% of social worker, 50.00% of environmentalist, 33.30% of I don't know, response are collected.

In this generation, we have was crazy young peoples. They don't think about environmental responsibility while celebrated some special movements in society. Previous decades are fire the crackers and enjoy, celebrate the festivals time, but nowadays the peoples use fire the fire crackers unnecessary movement like, birthday celebration, VIP entries and special festivals movement. This activity also joined to global warming and etc. The knowledge about this firing and cracker activity. As per response, 57.10% of noise and air pollution, 14.30% of noise and industrial pollution, 4.80% of water and noise pollution, 21.40% of air and water pollution, 2.40% of I don't know, responses are collected.

The Kerala Forum's campaign to stop the reduction of natural resources. It's possible that a project of such a nature developed or became well-known around that date. We are able to offer you more wide knowledge regarding ways community meetings or campaigns, whether in Kerala or in another location, may deal with problems related to the environment. Kerala region has huge biodiversity ecosystem, but human society step into the more income segment to development of nation, so they moderate the forest, like human activity area and wildlife activity area. As per responses, 21.40% of silent valley project , 54.80% of chipko movement , 7.10% narmada andolana, 2.40% of ganga cleaning project and 14.30% of I don't ,know, response are collected.

Solar ovens, occasionally referred to simply as cookers made from sunlight, are machines which roast and boil utilise sunshine as an alternative form of heat. Through transforming direct sunlight into heat, they eliminate the need for standard fuels like oil and wood while cooking food. The following are some essential uses and advantages of solar ovens, environmentally friendly, off –Grid cooking, reduced deforestation and etc. As per responses 40.50% of decreases environmental pollution, 11.90% increases demand for solar ovens, 33.30% increases environmental pollution.7.10% of increases demand for solar energy and 7.10% of I don't know, responses were collected.



**Yashwanth et al.,****Environmental Education**

Enhancing knowledge about environmental education is necessary for promoting environmentally friendly behaviors and an awareness of environmental responsibility for themselves.

Several individuals and authorities in the subject areas of long-term sustainability and environmental science say that all individuals should place a high premium on conservation of the environment. Because very one use the natural resource in daily life, that's why the every persons conserve and protect the natural resource and environment. As per responses, 26.20% of strongly agreed positive responses, 42.90% of agreed, 19.00% of neutral and 11.90% of disagreed, responses are collected.

Conservation of the environment is an effort which individuals of all ages are capable of contributing to; it is more than anything that professionals do. While adults might have better resources and influence over actions, children and teenagers are still able to contribute a big contribution to building a better tomorrow. These can be some ways citizens may help protect the environment, whichever their age. Schools initiatives, education and awareness to children and teens. As per responses, 9.50% of strongly agreed, 31.00% of agreed, 35.70% of neutral, 21.40% of disagreed and 2.40% of strongly disagreed, responses are recorded

Multidisciplinary instruction and practical relevance are concepts which stand in oppose to the notion of carefully compartmentalizing the teaching of a topic without linking it to the surrounding environment and its various elements. Math, science, language arts, and social studies are just a few of the instances of the subjects which have been classified into specific groups according to conventional educational paradigms. None the less, the beneficial effects of bringing together subjects and presenting them into the wider picture of the environment growing more and more obvious. As per responses 7.10% of strongly agreed, 42.90% of agreed, 33.30% of neutral and 16.70% of disagreed, responses are collected.

A complete approach that helps in the achievement of an in-depth awareness of the environment as well as associated concerns is for integrating environmental issues within the teaching of many subjects related to education. This interdisciplinary strategy recognizes that problems with the environment have complex connections and include multiple disciplines, like social sciences, science, the field of economics and even more. Here several improvements that students observe while environmental factors are presented: interdisciplinary learning, sustainability education. As per responses, 7.10% of strongly agreed, 45.20% of agreed, 33.30% of neutral, 11.90% of disagree and 2.40% of strongly disagreed, responses are collected.

While geography and biology certainly represent essential subjects to understand the basics of and appreciating environmental subjects, it's important that students understand that ecological issues are multidimensional and extend these two academic fields. More general in scope, the field of environmental science combines concepts and ideas from a number of academic, social, and economic areas. As per responses 9.50% of strongly agreed, 38.10% of agreed, 35.70% of neutral, 14.30% of disagreed and 2.40% of strongly disagreed responses are collected.

Considering it increases a sense of environmental responsibility and help learners understand the way various subjects interact with one another, combining environmental principles into various subject areas is usually seen as a helpful approach to education. To avoid any adverse consequences, such as learners losing focus on their main topic, it is essential that this combination be done properly. As per responses 9.50% of strongly agreed, 42.90%, agreed, 26.20% of neutral and 21.40% of disagreed responses are collected.

CONCLUSION

During nowadays, lifestyle becomes so rapid, the individuals having no spare time to show concern about the surrounding environment. Moreover, the scenario becomes more serious due to the continuing mistreatment and





Yashwanth et al.,

degradation of earth's resources in the title of industrialization. Materials and resources, which were previously thought to be reusable are currently in risk of growing increasingly restricted to be an outcome of neglect and misuse. The phenomenon becomes especially apparent within the younger generations of community. Humans promote contamination, without caring to or protecting the planet's health. The research was carried out between Government higher primary schools Teachers and Private higher primary schools level Teachers looking to find out more about their views on the environment awareness, education and ethics, and with the objective to evaluate how serious it is of the present situation. The results of the research indicated that Government higher primary schools Teachers and Private higher primary schools level Teachers have grown more aware of and worries about the harm that either intentional or unintentional activity by humans contributes to the ecosystem. Their responses to the questionnaire's topics highlighted an excellent aspect about their awareness of the environment. That is why schools, educational institutions must provide environmental ethics, awareness and education on the threats to health related to pollution from the environment, along with a number of events. These will allow it less difficult for recruits more and more Teachers and other people to join collaboratively to protect the globe's environmental sustainability.

REFERENCES

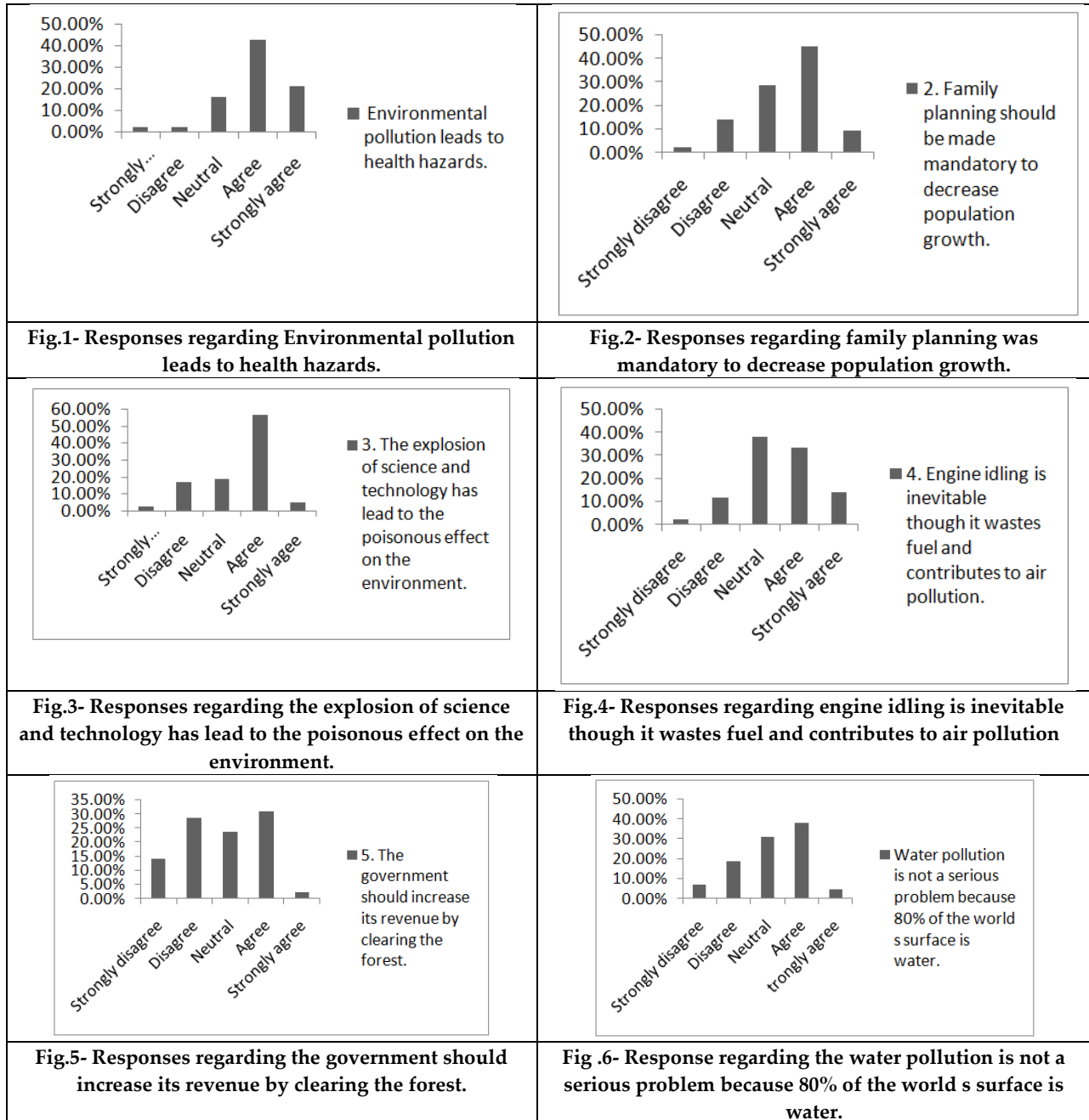
1. Singh, S. K., Chen, J., Del Giudice, M., & El-Kassar, A.-N. (2019). Environmental ethics, environmental performance, and competitive advantage: Role of environmental training. *Technological Forecasting and Social Change*, 146, 203–211. <https://doi.org/10.1016/j.techfore.2019.05.032>
2. Rickinson, M. (2001). Learners and Learning in Environmental Education: A critical review of the evidence. *Environmental Education Research*, 7(3), 207–320. <https://doi.org/10.1080/13504620120065230>
3. Panth, M. K., Verma, P., & Gupta, M. (2015). The Role of Attitude in Environmental Awareness of Under Graduate Students
4. Palmer, C., McShane, K., & Sandler, R. (2014). Environmental Ethics. *Annual Review of Environment and Resources*, 39(1), 419–442. <https://doi.org/10.1146/annurev-environ.121112-094434>
5. Mashaba, E. K., Maile, S., & Manaka, M. J. (2022). Learners' Knowledge of Environmental Education in Selected Primary Schools of the Tshwane North District, Gauteng Province. *International Journal of Environmental Research and Public Health*, 19(23), 15552. <https://doi.org/10.3390/ijerph192315552>
6. Jickling, B., & Spork, H. (1998). Education for the Environment: A critique. *Environmental Education Research*, 4(3), 309–327. <https://doi.org/10.1080/1350462980040306>
7. Jenkins, E. W., & Pell, R. G. (2006). "Me and the Environmental Challenges": A survey of English secondary school students' attitudes towards the environment. *International Journal of Science Education*, 28(7), 765–780. <https://doi.org/10.1080/09500690500498336>
8. Gök, N. D., & Firat Kiliç, H. (2021). Environmental Awareness and sensitivity of nursing students. *Nurse Education Today*, 101, 104882. <https://doi.org/10.1016/j.nedt.2021.104882>
9. Fien, J. (2000). 'Education for the Environment: A critique' – an analysis. *Environmental Education Research*, 6(2), 179–192. <https://doi.org/10.1080/713664671>
10. Dursun, S., & Mankolli, H. (n.d.). The Value of Nature: Virtue Ethics Perspective.
11. Danielraja, R. (2019). A Study of Environmental Awareness of Students at Higher Secondary Level. *Shanlax International Journal of Education*, 7(3), 6–10. <https://doi.org/10.34293/education.v7i3.480>
12. Chavada, K., & Charan, D. D. (n.d.). An analysis of environmental ethics among – College students
13. Ardoin, N. M., Bowers, A. W., Roth, N. W., & Holthuis, N. (2018). Environmental education and K-12 student outcomes: A review and analysis of research. *The Journal of Environmental Education*, 49(1), 1–17. <https://doi.org/10.1080/00958964.2017.1366155>





Yashwanth et al.,

14. Abbas, M. Y., & Singh, D. R. (2012). A Survey of Environmental Awareness, Attitude, and Participation amongst University Students: A Case Study. 3(5).





Yashwanth et al.,

<p>Fig.7- Responses regarding In order to conserve natural resources, we should</p>	<p>Fig.8- Responses regarding the depletion of ozone layer and the green house effect is caused by</p>
<p>Fig.9- Responses regarding, Medha Patkar is a well known</p>	<p>Fig.10- Responses regarding , Medha Patkar is a well known</p>
<p>Fig.11- Response regarding the Kerala Forum initiative against destruction of the natural resources</p>	<p>Fig.12- Responses regarding Use of solar ovens</p>
<p>Fig.13- Responses regarding protection of environment is every individuals first priority.</p>	<p>Fig.14- Responses regarding environmental protection by individuals is desirable and possible only at adulthood.</p>





Yashwanth et al.,

<p>■ 3. Teaching of a particular school subject should be tightly compartmentalized without relating it to environment and its components.</p>	<p>■ 4. Infusion of environmental dimension in the teaching of different school subject helps get a holistic view of environment and its problem.</p>
<p>Fig.15- Responses regarding teaching of a particular school subject should be tightly compartmentalized without relating it to environment and its components.</p>	<p>Fig.16-Response regarding infusion of environmental dimension in the teaching of different school subject helps get a holistic view of environment and its problem.</p>
<p>■ 5. Environment related concepts/principles, etc. can be successfully infused only in Biology (life science) and geography.</p>	<p>■ 6. Infusing environmental concepts in different school subjects leads to the deviation of students' concentration from the subject on hand.</p>
<p>Fig.17- Responses regarding environment related concepts/principles, etc. can be successfully infused only in Biology (life science) and geography.</p>	<p>Fig.18- Responses regarding infusing environmental concepts in different school subjects leads to the deviation of student's concentration from the subject on hand.</p>





Morphometric Analysis of Krishnarajapete Watershed using Geospatial Techniques, Mandya District, Karnataka, India

Shashi Kumar C^{1*} and Nagaraju D²

¹Research Scholar, Department of Earth Sciences, University of Mysore, Manasagangothri, Mysuru, Karnataka, India.

²Professor, Department of Earth Sciences, University of Mysore, Manasagangothri, Mysuru, Karnataka, India.

Received: 13 Aug 2024

Revised: 10 Oct 2024

Accepted: 01 Nov 2024

*Address for Correspondence

Shashi Kumar C

Research Scholar,
Department of Earth Sciences,
University of Mysore,
Manasagangothri, Mysuru,
Karnataka, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The morphometric properties of a watershed can be estimated using Remote sensing (RS) and Geographic information system (GIS) approaches. The watershed was divided into 11 sub-watersheds (SWs), which are then ranked according to morphometric characteristics. For each sub-watershed, morphometric features such as aerial and linear aspects have been evaluated to determine a preferred ranking. The drainage basin is characterized by a dendritic to subdendritic drainage pattern, according to morphometric analysis. The analysis also showed that the total number and length of stream segments is maximum in first-order streams and decreases as the stream order increases. The elongation ratio is less than 0.60 for all 11 sub-watersheds, indicating that it has a moderately sloped surface and an elongated shape. Low drainage density leads to coarse drainage texture in the study area. Flood flows easier in the elongated basin so 10 sub-watersheds except 1 sub-watershed show elongation of the shape of the basin.

Keywords: Morphometry, Sub-Watershed, Geographic Information System, Lithology.

INTRODUCTION

The measuring and quantitative analysis of the landforms' dimensions, shapes, and configurations on Earth is known as morphometry[1,2]. To ensure long-term growth, avoid soil erosion, and save water, watersheds must be studied. Remote sensing and geographic information system (GIS) analysis are effective methods for examining the hydraulic process in the river basin[3]. Any hydrological inquiry, including pedology, environmental evaluation, groundwater management, and groundwater potential assessment, has to consider the drainage basin analysis. Hydrologists and geomorphologists have determined that certain relationships between runoff characteristics and drainage basin systems' geographic and geomorphic features are crucial. The physiographic features of drainage basins, such as size,



**Shashi Kumar and Nagaraju**

shape, slope of the drainage area, drainage density, size and length of the contributors, etc., can be connected with several significant hydrologic phenomena[4].The Watershed is the best unit for managing natural resources and promoting sustainable development[5].To be able to properly define and analyze drainage basins, the most recent Remote Sensing and GIS techniques must be integrated[6].Scientific studies indicate that morphometric characteristics of a river basin are important in determining which sub-watersheds should be prioritized. Water flowing into and out of the basin will deposit and accumulate contaminants, fertilizers, and sediments [7,8]. The onsite and offsite ecology of the river basin may be significantly impacted by them. Therefore, researching the drainage basin process may contribute to a deeper comprehension of the hydrologic cycle and the movement of water. To achieve sustainable land and water resource use and lessen the effects of growing pollution, watershed management must be put into practice [9-11].These days, geographic information system (GIS) approaches are employed.

For evaluating the different topography and morphometric characteristics of the watersheds and drainage basins, since they offer a versatile setting and an effective instrument for the handling and examination of geographical data. Since there hasn't been a thorough morphometric investigation of the area done before, the primary goal of the current study is to use a Geographic Information System (GIS) to analyze the linear and areal morphometric characteristics of the watershed. The management of the region's water resources and other natural resources is aided by this study's knowledge of the many geo-hydrological features of the watershed[12].The most pertinent quantitative morphometric features have been selected and used for the current investigation. Three groups of morphometric features, such as linear, relief, and areal aspects, can be distinguished. Since they have a connection to flow, runoff, and soil erosion threats, they have been used to prioritize more vulnerable sub-watersheds[13, 14].The goal of the current study is to rank the sub-watersheds according to their individual morphometric properties within the watershed of Krishnarajapete.

MATERIALS AND METHODS

Study area

Krishnarajapete (KRP), also referred to as K.R. Pete is a taluk and municipality in the Mandya District of the Indian state of Karnataka. It has an area of 896 square kilometres and is located between longitude 76°19' 47" E and latitude 12°27'04" N. It is included in Toposheets numbers are 57D/5, 57D/6, 57D/9, 57D/12, and 57C/16, which were published by the Survey of India (SOI) and are depicted in Fig. 1. With the use of software NRSC and hydrosheds, the Krishnarajapete watershed has been divided into 11 sub-watersheds, as seen in Figure 2.

Lithology

As per the bhukosh information the extracted area shows Amphibolite, biotite schist, calc gneiss, dolerite, dunite, fuchsite quartzite, granite gneiss, granitoid, kyanite-staurolite-mica schist, leuco gneiss, metabasalt, meta ultramafite, pegmatite, quartzite, and quartz reef shown in figure 3. All these rocks are metamorphic and show excellent fracture and lineament zones.

Drainage

The area shows a good dendritic to sub-dendritic pattern of drainage pattern. The stream that follows this path has been divided into stream orders. The Krishnarajapete watershed shows 5 orders shown in Figures 4a and 4b. Some of the orders were absent in the subwatershed. The 11 individual sub-watersheds in a Krishnarajapete watershed are shown in Figure 5. Morphometry is the study of how water and sediment move across a basin and how the geometries of the stream and basin networks relate to this movement. Measurements of the drainage network's linear aspects, drainage basin's areal aspects, relief (gradient) aspects of the channel network, and contributing ground slopes are necessary for a systematic description of a drainage basin's geometry and its stream channel (Strahler, 1964). Using the mathematical formulas listed in (Table 1), the morphometric analysis for the parameters in the study



**Shashi Kumar and Nagaraju**

is conducted concerning stream order, stream length, bifurcation ratio, stream length ratio, basin length, drainage density, stream frequency, elongation ratio, circularity ratio, and form factor, etc.

Stream Order (S_{μ})

There are 5 stream orders in the entire Krishnarajapete district. There are a total of 64 streams are present. Individual streams in each sub-watershed are shown in Table 2. Order 1 refers to the tiniest fingertip tributaries. A channel segment of order 2 is produced where two first-order channels unite, a segment of order 3 is generated where two order-2 channels combine, and so on. Therefore, the stream segment of the greatest order is the mainstream, through which all water and sediment discharge travels[15].

Stream length (L_{μ})

The length of the stream indicates how the segments have developed chronologically, including any tectonic disturbances that may have occurred in between. Horton's principle states that there is a negative correlation between the order and the number of streams[16]. Out of 11 sub-watersheds the highest length of all order streams found in WS-9 is about 36.83 km and the lowest in WS-11 of about 2.11km as shown in Table 2.

Mean stream length (L_{sm})

The average stream length provides insight into the typical dimensions of drainage network elements and the surfaces that contribute to them[15]. In the study mean stream length varies from 0.14 km to 10.52 km.

Bifurcation Ratio (R_b)

The bifurcation ratio, which is the ratio of the total number of stream segments of one order to that of the next higher order in a drainage basin, is connected to the branching pattern of a drainage network[17]. The lower value and absence of the order value indicate less number of structural disturbances. The range from 0.25 to 2 is shown in Table 3.

Stream Length Ratio (RL)

The mean stream length of a particular order divided by the mean stream length of the next lower order is known as the stream length ratio, and it has a significant impact on the basin's erosion stage, surface flow, and discharge[18]. The values vary from 0.05 to 17.19, it completely depends on the area slope.

Mean Bifurcation Ratio (R_{bm})

The average of all bifurcation values is the mean bifurcation ratio, which was in the particular area is 1.92.

Basin Perimeter (P)

The outside line enclosing the drainage basin's area is known as the basin perimeter. It may be used as a gauge for the size and form of basins as it is measured along the lines separating them[19].

Basin length (L_b)

From the catchment to the point of confluence, it is the basin's longest length. The longest basin length in WS-9 is about 22.79 km and the lowest is 2.13 km in WS-11 as shown in Table 4.

Length of overland flow (L_g)

Percolation through the soil and infiltration (exfiltration), which both vary in time and place, have a major impact on overland flow[20]. The surface runoff varies from 0.56 to 1.06 as shown in Table 4.

Basin Area

A gathering area from which water would be directed toward a stream or river is referred to as a drainage region. The ridge that divides water flowing in opposing directions defines the area's border. The least area of 17.15 km² in WS-4 and the highest of 174.24 km² in WS-5 is shown in Table 4.



**Shashi Kumar and Nagaraju****Drainage density (Dd)**

It is known that drainage density plays a significant role in influencing how long water takes to move. In study area varies from 0.16 to 0.30 shown in Table 4. The fact that the research area's drainage density is low, it is evident that the area has medium relief, moderately dense plant cover, and permeable subsurface[21].

Drainage frequency (Fs)

Flooding is more common in basins with high drainage and stream frequency because higher drainage densities and stream frequencies cause quicker runoff[22]. In the study area drainage frequency was very low so more likely to flood 0.04 to 0.13 shown in Table 4.

Drainage Texture (Dt)

Less than two drainage density denotes a very coarse drainage texture, between two and four, a moderate drainage texture, between four and six, a fine drainage texture, and more than eight, a very fine drainage texture[23]. In all 11 sub-watersheds, the drainage texture is coarse shown in Table 4.

Form factor ratio (Rf)

A smoother flow peak for a longer period is predicted by the extended basin with a low form factor. Compared to circular basins, such elongated basins are easier to regulate flood flows[24]. In the study area form factor is high only in circular shape WS-11 shown in Table 4.

Elongation ratio (Re)

It is a highly important index in the analysis of basin shape that provides information about a drainage basin's hydrological characteristics. Low-relief locations are often represented by values close to 1. In the study area, all values except WS-4 and WS-11 show low relief as shown in Table 4.

Circularity ratio (Rc)

It is represented as the ratio of the basin area to the area of a circle with the same perimeter as the basin and is used as a quantitative metric to visualize the basin's form. It varies from 0.23 to 0.60, which shows the medium to low relief and approximate elongation of the drainage basin.

CONCLUSION

The watershed is categorized as a 5-order stream based on the drainage orders. The average R_b suggests that geological formations have little effect on the drainage pattern. The most helpful criteria for the morphometric categorization of drainage basins are drainage density (Dd) and stream frequency (Fs), which unquestionably regulate the runoff pattern, sediment output, and other hydrological parameters of the drainage basin. The underlying strata's porous character is shown by the basin's Drainage density. The dendritic drainage system that dominates the watershed helps to explain several topographical features, such as runoff and infiltration rate. Regions for surface-water accumulation and recharge-related activities that may be put into practice for watershed management are highlighted by the assessed parameters in this study.

ACKNOWLEDGMENT

The authors would like to thank the University of Mysore, Mysuru, for his warm encouragement and assistance. The University of Mysore in Mysuru has their gratitude for giving funding and support for research efforts.

Funding

There is no funding or financial support for this research work.



**Shashi Kumar and Nagaraju****Conflict of interest**

There is no conflict of interest among authors.

REFERENCES

1. Agarwal CS (1998). Study of drainage pattern through aerial data in Naugarh area of Varanasi district, U.P. Jour. Indian Soc. Rem. Sen, 26: 169-175.
2. Obi Reddy GE, Maji AK, Gajbhiye KS (2002). GIS for morphometric analysis of drainage basins. GIS India, 11(4):9-14.
3. Waikar M, Nilawar AP (2014) Morphometric analysis of a drainage basin using geographical information system: a case study. Int J Multidisciplin Current Res 2:179–184.
4. Gumma MK, Birhanu BZ, Mohammed IA, Tabo R, Whitbread AM (2016) Prioritization of watersheds across Mali using remote sensing data and GIS techniques for agricultural development planning. Water 8(6):260.
5. Patel, D.P.; Dholakia, M.; Naresh, N.; Srivastava, P.K. Water Harvesting Structure Positioning by Using Geo-Visualization Concept and Prioritization of Mini-Watersheds through Morphometric Analysis in The Lower Tapi Basin. *J. Indian Soc. Remote Sens.* 2012, 40, 299–312.
6. Singh, P.; Gupta, A.; Singh, M. Hydrological Inferences from Watershed Analysis for Water Resource Management using Remote Sensing and GIS Techniques. *Egypt J. Remote Sens. Space Sci.* 2014, 17, 111–121.
7. Magesh NS, Jitheshlal KV, Chandrasekar N, Jini KV (2013) Geographical information system based morphometric analysis of Bharathapuzha river basin, Kerala, India. *Appl Water Sci* 3:467–477.
8. Oyegoke S, Ifeadi C (2008) Relationship between drainage basin area and stream length for River Gongola, Nigeria. *Environ Res J* 2(1):23–27.
9. Javed A, Khanday MY, Ahmed R (2009) Prioritization of sub-watersheds based on morphometric and land use analysis using remote sensing and GIS techniques. *J Indian Soc Remote Sensing* 37:261.
10. Prakash K, Rawat D, Singh S (2019) Morphometric analysis using SRTM and GIS in synergy with depiction: a case study of the Karmanasa River basin, North central India. *Appl Water Sci* 9:13.
11. Rai PK, Mishra VN, Mohan K (2017) A study of morphometric evaluation of the Son Basin, India using the geospatial approach. *Remote Sensing Applic* 7:9–20.
12. Hajam, Rafiq & Hamid, Aadil & Dar, Naseer & Bhat, Sami. (2013). Morphometric analysis of vishav drainage basin using geo-spatial technology (GST). *International Research Journal of Geology and Mining*. 3. 2276-6618.
13. Nookaratnam K, Srivastava YK, Venkateswarao V, Amminedu E, Murthy KSR (2005) Check dam positioning by prioritization of micro-watersheds using SYI model and morphometric analysis remote sensing and GIS perspective. *J Indian Soc Remote Sens* 33(1):25–38.
14. Singh P, Gupta A (2014) Singh M (2014) Hydrological inferences from watershed analysis for water resource management using remote sensing and GIS techniques. *Egypt J Remote Sens Space Sci* 17:111–121.
15. Strahler AN (1964). Quantitative geomorphology of drainage basins and channel networks. In: *Handbook of applied hydrology*. Chow VT, (ED) McGraw Hill Book Company, New York. 4-11.
16. Horton RE (1932). Drainage basin characteristics. *Am. Geophys. Union, Trans.*; 13:348–352.
17. Schumn SA (1956). Evolution of drainage systems and slopes in badlands at Perth Amboy, New Jersey. *Geol. Soc. Am. Bull.* 67:597- 646.
18. Horton RE (1945). Erosional development of streams and their drainage basins: Hydrophysical approach to quantitative morphology. *Geol. Soc. Am. Bull.* 56:275-370.
19. Schumn SA (1956). Evolution of drainage systems and slopes in badlands at Perth Amboy, New Jersey. *Geol. Soc. Am. Bull.* 67:597- 646.
20. Schmid BH (1997). Critical rainfall duration for overland flow an infiltrating plane surface. *J. Hydrol.*; 193: 45-60.
21. Nag SK (1998). Morphometric analysis using remote sensing techniques in the Chaka sub-basin, Purulia district, West Bengal. *J. Ind. Soc. Remote Sensing*, 26(1): 69-76.
22. Kale VS, Gupta A (2001). *Introduction to Geomorphology*. Orient Blackswan Private Limited. 84-86.





Shashi Kumar and Nagaraju

23. Smith GH (1939). The Morphometry of Ohio: The Average Slope of the Land," Annals of the Association of American Geographers, 29: 94.
24. Christopher O, Idown AO, Olugbenga AS (2010). Hydrological Analysis of Onitsha North East Drainage Basin using Geoinformatic Techniques. World Applied Science Jour, 11(10):1297- 1302 (2010).

Table 1. Morphometry parameters formulas

S. No.	Parameters	Formula	Reference
1		Linear Morphometric parameters	
1.1	Stream Order (S_{μ})	Hierarchical rank	Strahler (1964)
1.2	Bifurcation Ratio (R_b)	$R_b = N_{\mu} / N_{\mu+1}$ Where, R_b = Bifurcation ratio, N_{μ} = No. of stream segments of a given order and $N_{\mu+1}$ = No. of stream segments of next higher order.	Schumm (1956)
1.3	Mean Bifurcation Ratio (R_{bm})	R_{bm} = Average of bifurcation ratios of all orders	Strahler (1964)
1.4	Stream Length (L_{μ})	Length of the stream (kilometers)	Horton (1945)
1.5	Mean Stream Length (L_{sm})	$L_{sm} = L_{\mu} / N_{\mu}$ Where, L_{μ} = Total stream length of order ' μ ' N_{μ} = Total no. of stream segments of order ' μ '	Strahler (1964)
1.6	Stream Length Ratio (R_L)	$R_L = L_{sm} / L_{sm-1}$ Where, L_{sm} = Mean stream length of a given order and L_{sm-1} = Mean stream length of next lower order	Horton (1945)
1.7	Length of Overland Flow (L_g)	$L_g = 1/2D$ Km Where, D = Drainage density (Km/Km^2)	Horton (1945)
1.8	Basin Perimeter (P)	P = Outer boundary of drainage basin measured in kilometers.	Schumm (1956)
1.9	Basin Length (L_b)	$L_b = 1.312 * A^{0.568}$	Gregory and Walling (1973)
1.10	Fitness Ratio (R_f)	$R_f = C_L / P$ Where, C_L = Main channel length (Kms) and P = Basin perimeter (Kms)	Melton (1957)
1.11	Wandering Ratio (R_w)	$R_w = C_L / L_v$ Where, C_L = Main channel length (Kms) and L_v = Valley length (Kms)	Smart and Surkan (1967)
1.12	Standard Sinuosity Index (SSI)	$SSI = C_L / L_v$ Where, C_L = Channel length (Kms) and L_v = Valley length (Kms)	Muller (1968)
2		Areal Morphometric parameters	
2.1	Basin Area (A)	Area from which water drains to a common stream and boundary determined by opposite ridges.	Strahler (1969)
2.2	Drainage Density (D_d)	$D_d = L_{\mu} / A$ Where, D_d = Drainage density (Km/Km^2) L_{μ} = Total stream length of all orders and A = Area of the basin (Km^2).	Horton (1932)
2.3	Drainage Frequency (F_d)	$F_d = N_{\mu} / A$ Where, F_d = Drainage frequency. N_{μ} = Total no. of streams of all orders and A = Area of the basin (Km^2).	Horton (1932)
2.4	Drainage Texture (D_t)	$D_t = N_{\mu} / P$ Where, N_{μ} = No. of streams in a given order and P = Perimeter (Kms)	Smith (1950) & Horton (1945)
2.5	Form Factor Ratio (R_f)	$R_f = A / L_b^2$ Where, A = Area of the basin and L_b = (Maximum) basin length	Horton (1932)
2.6	Elongation Ratio (R_e)	$R_e = \sqrt{A} / \pi / L_b$ Where, A = Area of the Basin (Km^2) L_b = Maximum Basin length (Km)	Schumm (1956)
2.7	Circularity Ratio (R_c)	$R_c = 4\pi A / P^2$ Where, A = Basin Area (Km^2) and P = Perimeter of the basin (Km) Or $R_c = A / A_c$ Where, A = Basin Area (Km^2) and A_c = area of a circle having the same perimeter as the basin	Miller (1953)





Shashi Kumar and Nagaraju

Table 2. Stream orders, number, length(km), and Mean stream length.

Sub-watershed numbers	Stream orders number(S_{μ})	Streams in numbers	Total stream In numbers ΣS_{μ}	Stream length (L_{μ})	$\Sigma (L_{\mu})$	Mean stream length (L_{sm})
WS-1	1 st	1	1	4.61	4.61	4.61
WS-2	1 st	2	7	0.28	13.5 5	0.14
	4 th	1		3.11		3.11
	5 th	4		10.16		2.54
WS-3	1 st	4	11	11.61	31.4 8	2.90
	2 nd	2		9.15		4.58
	3 rd	1		0.49		0.49
	5 th	4		10.23		2.56
WS-4	4 th	1	1	3.38	3.38	3.38
WS-5	1 st	5	13	11.96	39.5 8	2.39
	2 nd	1		1.75		1.75
	3 rd	1		0.23		0.23
	5 th	6		25.64		4.27
WS-6	2 nd	2	3	0.54	9.82	0.27
	3 rd	1		9.28		9.28
WS-7	1 st	3	5	16.23	26.0	5.41
	2 nd	2		9.80	3	4.90
WS-8	1 st	3	5	10.06	23.5	3.35
	2 nd	2		13.49	5	6.75
WS-9	1 st	4	7	11.36	36.8 3	2.84
	2 nd	2		21.04		10.52
	3 rd	1		4.43		4.43
WS-10	1 st	4	9	7.87	24.7	1.97
	5 th	5		16.88	5	3.38
WS-11	1 st	1	2	2.10	2.88	2.10
	3 rd	1		0.78		0.78

Table 3 Stream length ratio and Bifurcation Ratio

Sub-watershed numbers	Stream Length Ratio (RL)				Bifurcation ratio (Rb)			
	2 nd /1 st	3 rd /2 nd	4 th /3 rd	5 th /4 th	1 st /2 nd	2 nd /3 rd	3 rd /4 th	4 th /5 th
WS-1	-	-	-	-	-	-	-	-
WS-2	-	-	-	3.26	-	-	-	0.25
WS-3	0.79	0.05	-	-	2	2	-	-
WS-4	-	-	-	-	-	-	-	-
WS-5	0.15	0.13	-	-	5	1	-	-
WS-6	-	17.19	-	-	-	2	-	-





Shashi Kumar and Nagaraju

WS-7	0.60	-	-	-	1.5	-	-	-
WS-8	1.34	-	-	-	1.5	-	-	-
WS-9	1.85	0.21	-	-	2	2	-	-
WS-10	-	-	-	-	-	-	-	-
WS-11	-	-	-	-	-	-	-	-

Table 4. Perimeter, Basin Length, Length of Overland Flow, Basin Area, Drainage Density, Drainage/ stream Frequency, Drainage Texture, Form Factor Ratio, Elongation Ratio, and Circularity Ratio

Sub-watershed Number	Perimeter (P)	Basin Length (Lb)	Length of Overland Flow (Lg)	Basin Area (A)	Drainage Density (Dd)	Drainage/ stream Frequency (Fs)	Drainage Texture (Dt)	Form Factor Ratio (Rf)	Elongation Ratio (Re)	Circularity Ratio (Rc)
WS-1	33.72	7.92	0.70	23.02	0.20	0.04	0.03	0.37	0.68	0.25
WS-2	42.92	9.64	0.85	55.65	0.24	0.13	0.16	0.60	0.87	0.38
WS-3	55.10	19.19	0.89	123.12	0.26	0.09	0.20	0.33	0.65	0.51
WS-4	22.67	2.97	0.69	17.15	0.20	0.06	0.04	1.94	1.57	0.42
WS-5	60.50	16.35	0.80	174.24	0.23	0.07	0.21	0.65	0.91	0.60
WS-6	30.98	9.16	0.98	35.05	0.28	0.09	0.10	0.42	0.73	0.46
WS-7	49.76	14.15	1.02	89.30	0.29	0.06	0.10	0.45	0.75	0.45
WS-8	55.68	17.52	0.68	121.16	0.19	0.04	0.09	0.39	0.71	0.49
WS-9	89.49	22.79	0.88	146.24	0.25	0.05	0.08	0.28	0.60	0.23
WS-10	45.64	12.61	1.06	81.91	0.30	0.11	0.20	0.52	0.81	0.49
WS-11	29.61	2.13	0.56	18.03	0.16	0.11	0.07	3.97	2.25	0.26

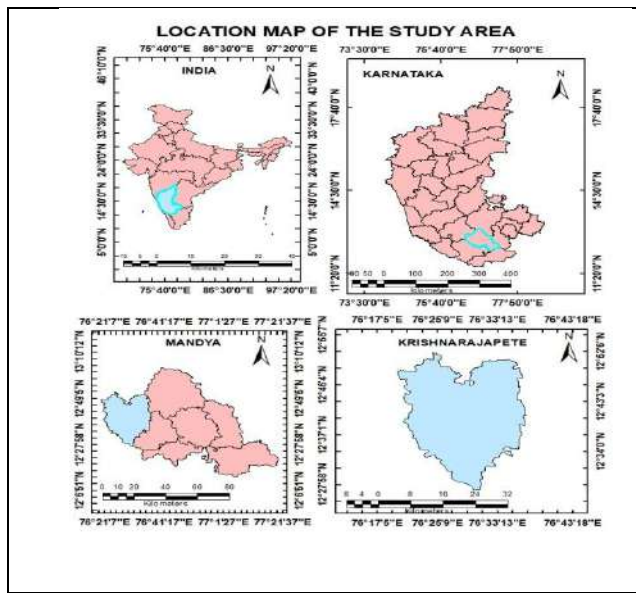


Fig. 1. Location Map of the Study Area

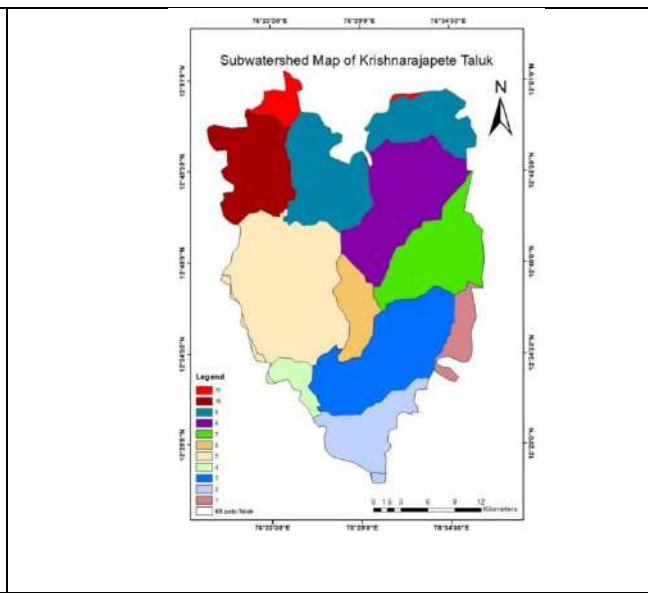


Fig. 2. Sub-watershed map of Krishnarajapete





Shashi Kumar and Nagaraju

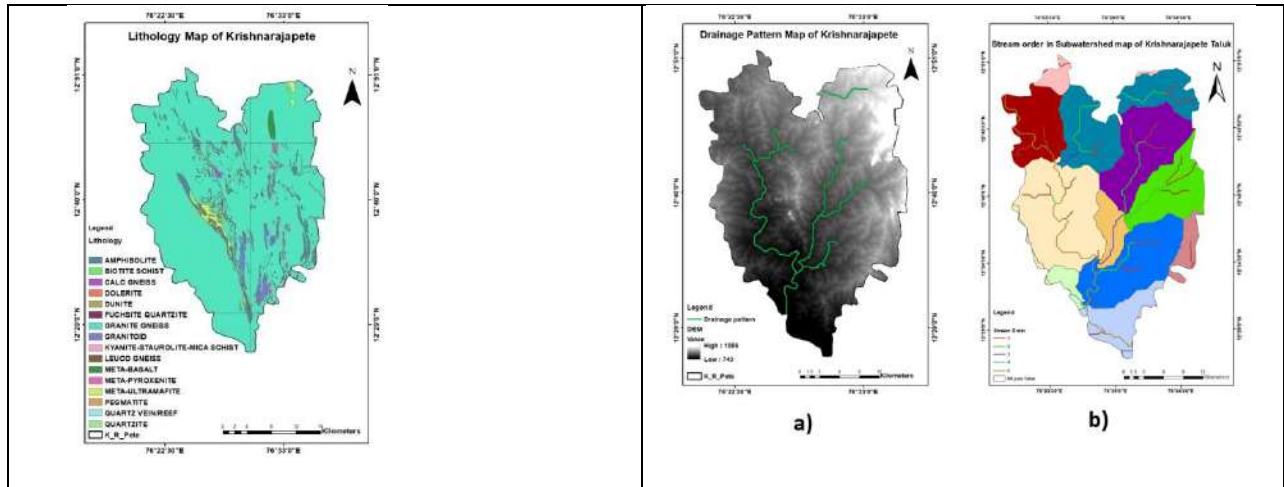


Fig. 3. Lithology map of Krishnarajapete.

Fig. 4. GIS map of a) Drainage map, and b) Stream order map

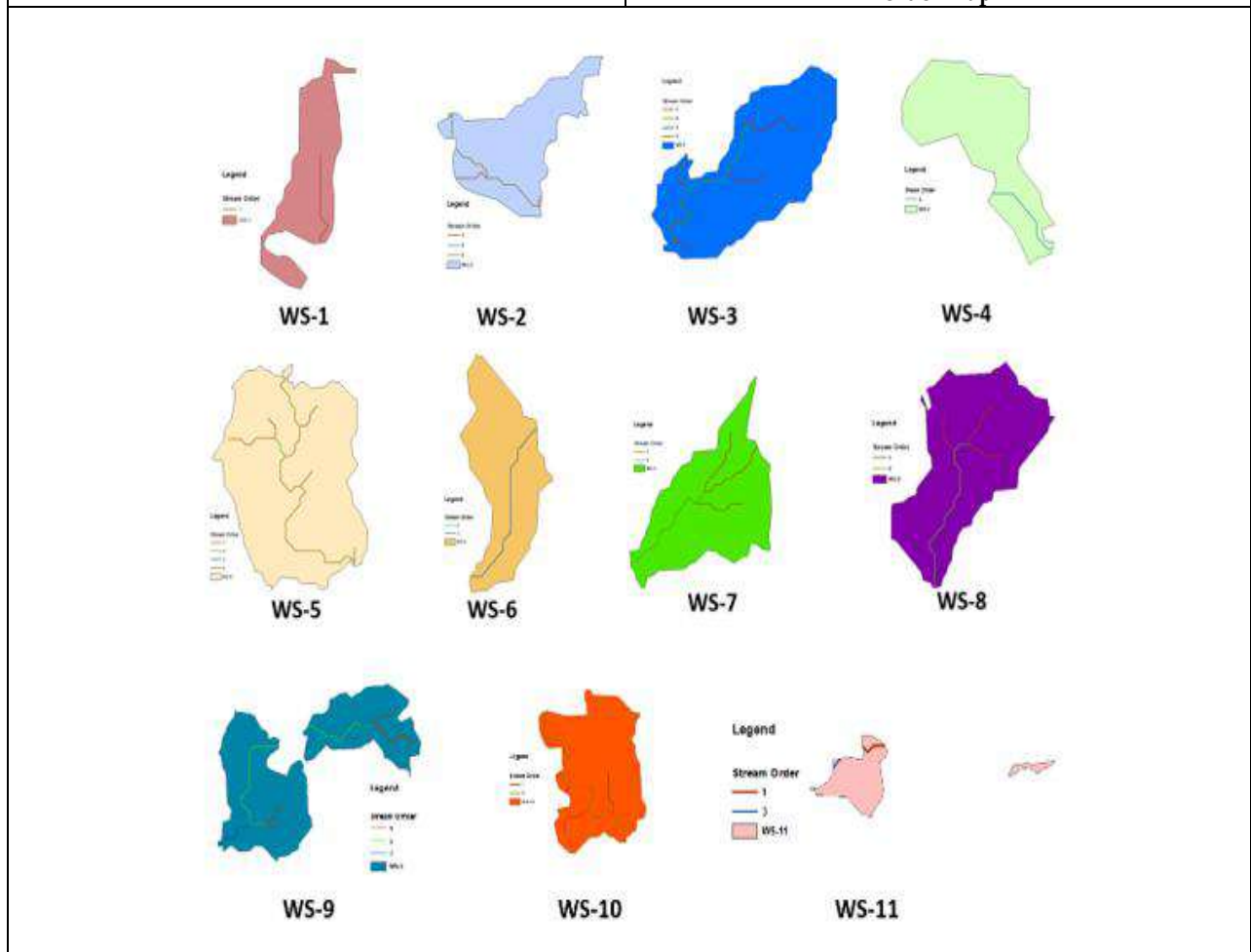


Fig. 5. The eleven individual sub-watershed map





Design and Implementation of a Framework for IoT- based Smart Attendance Management System

M. Chitra¹, S. Namagiri², Praveen Rathod³, Sasikala P⁴, M. Elangovan⁵ and R Radha^{6*}

¹Professor, Computer Science and Engineering, Hindusthan Institute of Technology, Coimbatore, , Tamil Nadu, India.

²Assistant Professor, Department of CSE, Salem College of Engineering and Technology, Salem, (Affiliated to Anna University), Tamil Nadu, India.

³Assistant professor, Department of Mechanical Engineering , Vishwakarma Institute of Information Technology, (Affiliated to the Savitribai Phule Pune University) Pune, Maharashtra, India

⁴Associate Professor, Department of Computer Science, Lal Bahadur Shastri Government First Grade College, R T Nagar, Bangalore, Karnataka, India.

⁵Professor, Department of Aerospace Engineering, SNS College of Technology, Coimbatore, Tamil Nadu, India.

⁶Assistant Professor, Department of Science and Humanities, Karpagam College of Engineering, Coimbatore, Tamil Nadu, India.

Received: 24 May 2024

Revised: 26 Aug 2024

Accepted: 18 Oct 2024

*Address for Correspondence

R Radha

Assistant Professor,
Department of Science and Humanities,
Karpagam College of Engineering, Coimbatore,
Tamil Nadu, India.

E.Mail: radharmat2020@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

This research paper outlines an innovative attendance management system leveraging IoT, AWS, and an RFID module with an Arduino Uno board. This system aims to enhance accuracy and efficiency in attendance tracking, overcoming the limitations of traditional methods. Comprising both hardware and software elements, the hardware integrates an RFID module connected to an Arduino Uno board for capturing attendance data. The software, developed using Python Django and hosted on AWS, is responsible for processing and storing attendance information. This architecture offers real-time attendance monitoring and reporting, accessible through web or mobile applications from any location. Tested in a real-world scenario using RFID-enabled tags or cards for attendance tracking, the implemented system demonstrated superior accuracy and efficiency compared to conventional attendance management systems. The proposed solution provides a reliable, cost-effective approach for attendance management, applicable across various organizational settings. By facilitating real-time tracking and



**Chitra et al.,**

reporting through web and mobile interfaces, the system offers an effective alternative to traditional attendance management methods, promising heightened accuracy and efficiency.

Keywords: RFID, AWS, IoT, hardware, Python, Arduino.

INTRODUCTION

An efficient method of managing attendance is an essential component of the successful operation of any organization, but this is especially true in the fields of education and business. Conventional methods of documenting attendance rely on human procedures, which are prone to mistakes and can result in inefficiencies. Modern technologies provide more accurate and time-saving alternatives. The advent of the Internet of Things (IoT) and cloud computing has brought about a substantial shift in attendance management. As a result, the accuracy and efficacy of the aforementioned systems have been significantly improved.

The investigation of the incorporation of Internet of Things (IoT), Amazon Web Services (AWS), and Radio Frequency Identification (RFID) technologies within the framework of Python Django and Arduino Uno is the major purpose of our research. This integration is intended to provide an innovative strategy for the administration of attendance systems. The purpose of this project is to solve the constraints of existing systems while also automating attendance procedures. This investigation presents a novel strategy for managing attendance, with the goals of improving accuracy, productivity, and cost-effectiveness. The system may be broken down into its two basic parts, which are the hardware and the software. The hardware configuration for the purpose of collecting attendance data comprises of an RFID module that is coupled to an Arduino Uno board. This board is used for the purpose of collecting data. On the Amazon Web Services (AWS) Cloud platform is where the program that was built with the Python Django framework is currently running. The storage and processing of attendance data is the major responsibility that it is tasked with handling. The aforementioned architectural design makes it possible to record and report attendance in real time, information that can be accessed easily through either web-based or mobile apps thanks to the design's convenience.

The automation of attendance tracking procedures is a direct outcome of the incorporation of Internet of Things (IoT) technology into attendance management systems. Figure. 1. shows the whole architecture of the hardware component. Because human data entry is no longer necessary as a result of this automation, the possibility of making a mistake has been significantly reduced. RFID modules are able to scan RFID tags or user cards, which makes it easier to gather data on attendance. These modules may be used in conjunction with RFID readers. After the data has been gathered, these modules will send it to the AWS Cloud, where it will be processed and stored. Cloud computing from Amazon Web Services (AWS) provides a solution for managing attendance that is scalable, reliable, and cost-effective. These are the three main characteristics of the solution. Because Amazon Web Services (AWS) offers a variety of services that include storage, processing, and administration tools, it is a particularly excellent platform for hosting attendance management systems. These services may be found here. In addition, Amazon Web Services makes it easier for users to access their accounts by enabling seamless system usage from any place provided they have an internet connection.

Python Django, a web development framework that is widely used and well-known for its capacity to speed up the construction of online applications, was used in the implementation of the software component of our suggested system. This allowed us to realize the system more quickly. Python is an excellent choice as a programming language for the development of software components that are employed in attendance management systems because of its longevity, versatility, and ease of learning. These qualities make Python an ideal choice. Django is a web application framework that provides a complete set of tools that can be used to build web applications very efficiently. These



**Chitra et al.,**

tools cover fundamental aspects of web application development such as authentication, data modeling, and data security. When it comes to the context of attendance management systems, these characteristics are very crucial. The architectural model that is being discussed in this article has already been implemented and put through rigorous testing in real-world settings, making use of RFID attendance badges or maps as the data source. These studies have proved that the model possesses superior levels of accuracy and efficiency when compared to other attendance management systems that are currently available. Figure. 2. shows the whole architecture of the hardware component. The architecture that we have built provides a dependable and cost-effective solution for monitoring attendance, and it is applicable to a very wide variety of businesses.

Methods of Experimentation and/or Methodology

The Methodical Approach

Identification of the Problem

The first stage entails the identification of limitations that are associated with conventional attendance management systems as well as the potential improvements that may be achieved via the integration of Internet of Things (IoT) and cloud computing technologies, which will ultimately result in an increase in the system's accuracy and efficiency. This will be done in order to set the stage for the subsequent stages. In order to get a more in-depth comprehension of the subject at hand, the purpose of the current investigation is to carry out an exhaustive analysis of the previously published research on the matter. An exhaustive investigation into the existing attendance management systems and the inherent limitations of such systems is being carried out. This requires doing an in-depth analysis of Internet of Things (IoT) and cloud computing technologies to determine which tools and platforms are most suited for the proposed system.

The creation of a system is going to be covered in this round of conversation. The attendance management system has been designed with an emphasis on architecture, with components drawn from both the hardware and software realms. An RFID module that is attached to an Arduino Uno board is the component of the hardware that is used to collect attendance data. This board is used for the purpose of acquiring information about attendees. The software component was developed using the Python Django framework, and it is now hosted on the cloud infrastructure provided by Amazon Web Services (AWS). The storing and processing of attendance records is the major purpose of the system.

System Implementation

After the proposed system has been designed, it is next put into action and assessed in a real-world environment employing an RFID-enabled tag or card for the purpose of registering attendance. An investigation is carried out on the data on attendance that was gleaned from the proposed system. To evaluate whether or not improvements have been made in terms of accuracy and efficiency, the findings that were collected are compared with those of the conventional attendance management system.

Consideration and Concluding Thoughts

An evaluation of the effectiveness of the attendance management system that is discussed in this research is carried out by carefully examining the data. A judgment may be drawn on the effectiveness of the system and the potential value it possesses in a wide variety of organizational settings.

The Accumulation and Evaluation of Information

For determining who attended a certain event, the gathering of data makes use of an RFID module that is fastened to an Arduino Uno board. After the data has been obtained, it is then evaluated by the software module that was written utilizing the Python Django framework and then placed on the cloud platform that is provided by Amazon Web Services (AWS). The current technique makes it possible to monitor and document attendance in real time. This information may be accessible easily through a web application or a mobile application, depending on your preference. The information that has been compiled includes attendance records for individual students or





employees. These records include the individuals' names, as well as the date, the time, and any other significant information.

Analysis of the Data

The data that has been acquired is analyzed using a variety of statistical methods, including but not limited to measurements such as mean, median, standard deviation, and other appropriate approaches. This analysis is performed on the data that has been gathered. In the research article, the analytic results are presented with graphical representations for the purpose of providing a thorough understanding of the system's effectiveness and efficiency. The following are some examples of formulas used for the purpose of data analysis:

The mean is determined by taking the total number of observations (n) and dividing that figure by the sum of all the values (x_i). When there are an odd number of values in a dataset, the method for computing the median is given by dividing the total number of values plus one by the second value in the dataset.

The following is the formula that should be used when computing the median of a collection of data that has an even number of values (n): The median is determined by taking the average of the value that is $(n/2)$ nd and the value that is $((n/2) + 1)$ th.

The formula for calculating the standard deviation, which is denoted by the letter s , is $s = \sqrt{(1/n) * \sum (x_i - \bar{x})^2}$, where the letter s denotes the standard deviation. The value n denotes the complete number of observations contained inside the dataset. The value shown by the symbol \sum is the total of the values under consideration.

Individual data points are represented by the variable x_i in this sentence.

In the following, we will refer to the arithmetic mean of the data collection as \bar{x} .

The evaluation takes into account a wide variety of factors, including the system's accuracy, efficiency, reliability, and cost-effectiveness.

Limitations and Restrictions

The attendance management system that is being offered was designed with a specific hardware and software architecture, which may not be appropriate in all contexts. This is one of the limitations of the system. The performance of the system is contingent on the existence of the network and the cloud infrastructure as well as their level of consistency, which has the potential to impair its reliability. The software component is composed of three main modules: the AWS EC2, the AWS API Gateway, and the Python Django application refer Figure. 3. The accuracy of attendance records is dependent on the efficient operation of the RFID module and the uniformity of the tag or card that is used. The accuracy of attendance records is dependent on the uniformity of the tag or card that is used. The size of the study's sample was restricted, which may have hindered its capacity to capture all of the many organizational or scenario variations that are feasible.

Limitations: The major goal of this study was to construct and analyze the efficacy of an attendance management system that includes Internet of Things (IoT), Amazon Web Services (AWS), and a Radio Frequency Identification (RFID) module. This purpose was accomplished by conducting this research. Python Django and a board designed for Arduino Uno were utilized during the process of putting this system into operation. Due to the fact that the study's scope was restricted to the education and corporate sectors, it is possible that the recommended strategy will not be applicable to other fields to the same extent. Because this research was based on a single case study, the data collection and analysis were carried out using that methodology. Because of this, it is possible that the findings cannot be generalized to other contexts or organizations. The study did not include an investigation of the moral and legal implications of utilizing RFID technology for attendance management. This was omitted from the scope of the research.

The Suggested Architecture for the System

Prerequisites for the System

The hardware is:



**Chitra et al.,**

- Internet connectivity for the Arduino UNO R3 board • RFID-enabled tags or cards • RFID module
- Python, the Django web framework, and Amazon Web Services (AWS) cloud services for hosting and storage are the software.
- The Relational Database Service (RDS) offered by Amazon Web Services for the purpose of storing attendance data.
- An application for mobile devices or the web that may be used to retrieve attendance statistics.

Requirements from a Functional Perspective:

- Keeping track of and reporting on attendance in real time
- Automated procedure for managing attendance • Error-free recording of attendance • Attendance data accessible through online or mobile application • Reliable and secure processing and storage of attendance data

Requirements that aren't related to functionality:

- An availability rate of at least 99.9 percent for the system
- high system performance for real-time attendance monitoring and reporting; • scalability for handling considerable volumes of attendance data; • security measures for protecting attendance data against unwanted access or change.

Components of the System and Their Modules

The hardware component of the proposed system consists of the RC522 RFID module (Figure 4.), Arduino Uno R3 board (Figure 5.), and RFID enabled tags or cards (Figure 6.) The hardware component, the software component, and the cloud component make up this architecture for the attendance management system. This architecture is intended to increase the effectiveness and precision of traditional attendance systems via the automation of the process of managing attendance as well as by improving efficiency. All of these individual components are created in such a way that allows the overall design to conform to the metrics that were initially taken into account. Accuracy, efficiency, reliability, and cost-effectiveness are some of the metrics that are taken into account while evaluating the system. The RC522 RFID module, the Arduino Uno R3 board, and RFID-enabled tags or cards are what make up the hardware component of the system that is being suggested. Reading the RFID-enabled tags or cards worn by users allows the RFID module to collect attendance data. This is done by reading the tags or cards. The data are transmitted from the module to the Arduino Uno board, where they are processed before being sent on to the software component. Users are provided with RFID-enabled tags or cards, and then they record their attendance by simply scanning the tag or card at the terminal that has been allocated for that purpose.

Components of the Software

The Python Django web development framework is being utilized throughout the construction of the software component of the proposed system. For the purpose of developing online applications, Django offers a comprehensive set of capabilities. These technologies, which include authentication, data modeling, and data security, are crucial components of the attendance management system. Through the use of the local network, the software component is linked to the Arduino Uno R3 board. This board then establishes communication with the RC522 RFID module in order to obtain the attendance data. The attendance records are kept in a database that is part of the software component. Authorized users of the system are the only ones who can access this database and view the attendance records. A web-based administrative user interface is also provided by the software component. With this interface, authorized users are able to monitor attendance records in real time, produce reports (in formats such as .xlsx and .csv), and manage attendance records.

Component of the Cloud

The suggested system makes use of cloud computing technologies provided by AWS for the construction of the cloud component. The hosting of the attendance management system may be accomplished using AWS, which offers a solution that is scalable, dependable, and cost-effective. The cloud component makes use of a number of Amazon Web Services (AWS) products, such as Amazon EC2 for the hosting of the web application, Amazon RDS for the



**Chitra et al.,**

storage of the database, Amazon S3 for the storage of the generated reports (.xlsx.csv), and Amazon Elastic Load Balancer (ELB) for scalability. The data on attendance is sent from the software component of the system to the cloud component, where it is processed and stored. The cloud component is responsible for both processing and storing the data. The cloud feature offers a centralized place for storing the attendance data, which can be viewed from any location with an internet connection. This feature also allows for the data to be updated in real time. In addition, the cloud component offers a scalable solution that can meet the expansion of the attendance management system as the number of users and attendance data grows. This growth may be accommodated by the scalability of the solution.

System Design and Construction Procedures

An RFID tag reader RC522, an RFID tag, and an Arduino Uno R3 board make up the hardware component of the system, respectively. The user is able to be uniquely identified thanks to the RFID reader, which records the one-of-a-kind identification number stored on the RFID tag. The information gleaned from the RFID reader is processed by the Arduino UNO board and then uploaded to the cloud. The data is initially transmitted to the institution's local network, and then, via the institution's primary router or gateway, the data packet is delivered to the Cloud gateway together with the API KEY. The software component is being operated on a number of different AWS cloud infrastructures; by combining all of these, we have created our software component, which is responsible for handling all of the data analysis, creating reports, and providing the administrator user with the ability to control and use it. We are able to provide a demonstration of the entirety of the software component's architecture. After the one-of-a-kind identifier has been delivered to the AWS access point using the API KEY, we go on to the further steps, which include reviewing the data of legitimate tag holders and authenticating the data if it was not determined to be in the correct format. In the event that the attendance is not made to the AWS RDS, our authentication procedure will either send a 400 message to the hardware or it will make the attendance there. In addition, the program has a cloud storage component for the purpose of keeping the generated reports and, if necessary, additional system data for either short-term or long-term usage, depending on the circumstances.

The Python Django application, the Amazon Elastic Compute Cloud (EC2), and the Amazon API Gateway make up the three primary components that make up the software component. Diagram of the movement of cloud components. The data that is sent from the Arduino Uno board is received by the AWS EC2 and then stored in the database that is associated with the AWS RDS. The requests and answers that occur between the AWS EC2 and the Python Django application are managed through the usage of the AWS API Gateway. Both the presentation of the attendance data and the generation of reports are the responsibilities of the Python Django application. In the end, it uses Amazon Simple Storage Service (S3) to store all of the previously generated reports and data. Setting up the cloud services provided by AWS, configuring the RFID module to work with the Arduino Uno board, and developing the Python Django application are the three steps involved in the implementation of the system. AWS EC2, AWS API Gateway, AWS Elastic Load Balancer, AWS S3, and AWS RDS are the AWS services that are necessary for the system to function properly. The software on the hardware module is set up to transmit the data to the AWS API Gateway together with the API KEY for the purpose of authentication. The API Gateway subsequently saves the data in the AWS RDS database. The Python Django application is constructed to retrieve the data that is stored in the AWS RDS database and present it on the management user interface in a way that is friendly to users.

One micro service on an EC2 server is responsible for providing all of the static files located in the 0.0.0.0:80/static directory, and the second micro service is responsible for delivering the dynamic Django web application on the same port. Both of these micro services are redirected through the settings of the Apache Web Server. We also have the option of using NGINX, which is another widely used web server. Figure 7. Proposes the attendance management system that utilizes IoT, RFID technology, and AWS cloud services to provide an accurate and efficient attendance management solution.

The Internet of Things (IoT), radio frequency identification (RFID), and Amazon Web Services (AWS) cloud services are utilized by the attendance management system that provides an accurate and effective solution for managing attendance. Seeing as how we have previously gone over the fundamentals of each module and their design; these flow diagrams will not be repeated here. Describes the overall structure of the architecture in its entirety. The





Chitra et al.,

system's hardware component takes the attendance data, and the software component saves and Architecture processes the data on the AWS cloud. Setting up the AWS services, configuring the hardware, and developing the software application are all necessary steps in the process of putting the system into operation

RESULTS AND DISCUSSION

Key Findings and Contributions

A whole new architecture for an attendance management system is offered; it makes use of Internet of Things, Amazon Web Services, and RFID technology, and it is controlled by an Arduino Uno board. Traditional methods of managing attendance may be improved with the help of this system, which offers a solution that is both efficient and cost-effective. The suggested architecture is comprised of hardware and software as its two basic building blocks. The hardware component consists of an RFID module that is linked to an Arduino UNO R3 board. This board is responsible for recording attendance information. The software component was developed with Python Django and is hosted on the AWS cloud. It is responsible for storing, processing, and producing the attendance data in the file format of either.xlsx or.csv. It will be much simpler for administrators to keep track of attendance and create reports using the system that has been proposed since it would allow real-time tracking and reporting of attendance that can be accessed via a web application or a mobile application.

The system was put into action and put through its paces in a real-world setting employing RFID-enabled tags or cards to take attendance. The findings demonstrate that the system is superior to standard attendance management systems in terms of both its accuracy and its efficiency. The solution that has been developed has the potential to be used in a variety of businesses, which would result in enhanced procedures for attendance management and higher levels of productivity. The issues that are experienced by standard methods of attendance management are addressed and a novel solution is proposed in this research study, making the work's contributions particularly noteworthy. The approach that has been suggested is not only more accurate and effective, but it is also less expensive and simpler to put into action. This research reveals unexplored avenues for the application of internet of things (IoT) and cloud computing technologies in attendance management systems and lays forth a strategy for further investigation into this subject area.

Applications

The study findings reported in this paper have significant implications and applications for organizations in many sectors, particularly those that require efficient attendance management, such as the education and business industries. These implications and applications have enormous value for these companies. The research proposes an attendance management system that is novel, reliable, and efficient. This system has the potential to improve productivity and decrease expenses. The capacity of the system to monitor and report attendance in real-time enables companies to effectively monitor attendance patterns, hence simplifying the discovery of trends that might potentially enhance overall attendance and productivity. Moreover, the use of Internet of Things (IoT), Amazon Web Services (AWS), and Radio Frequency Identification (RFID) technologies in the suggested framework demonstrates the significant impact these developing technologies may have on enhancing conventional procedures, particularly in the realm of attendance management. Organizations can get advantages by utilizing this technology to automate more laborious and error-prone procedures, resulting in increased operational efficiency and accuracy. The suggested attendance management system exhibits a high degree of scalability and adaptability, making it suitable for implementation in a wide range of organizational contexts, including both small firms and large-scale organizations. The system's availability via online and mobile applications enables enterprises to effectively oversee attendance from any geographical location, making it well-suited for meeting the demands of remote work settings.

The suggested attendance management system possesses a wide range of implications and applications, providing enterprises with a cost-effective, accurate, and effective alternative for managing attendance. The proposed solution possesses the capacity to greatly augment overall productivity and profitability inside companies, hence reinforcing its strategic significance within the current landscape of organizational operations.





Chitra et al.,

Future Prospects

The proposed attendance management system, which makes use of IoT, AWS, and the RFID module, offers encouraging prospects for further development and improvements. An approach that can be taken is to enhance the capabilities of the system through the integration of sophisticated features such as biometric authentication or facial recognition. The integration of this feature would enhance the monitoring of attendance, thereby guaranteeing increased levels of security and precision. An additional aspect that could be enhanced pertains to the incorporation of machine learning algorithms. The implementation of machine learning techniques could facilitate the predictive analysis of attendance patterns and the timely detection of potential absenteeism concerns, thereby augmenting the predictive capabilities of the system. Furthermore, in order to enhance scalability, forthcoming endeavors may involve the deployment of a distributed architecture. Implementing this enhancement would allow the system to effectively accommodate a greater number of users and process a more comprehensive dataset. Furthermore, the integration of the system with other enterprise systems, including payroll and HR administration, can serve to expand its scope. The establishment of such integration would facilitate the optimization of organizational processes through the development of a more all-encompassing ecosystem that enables smooth data transfer and integration. The improvement proposals have the potential to greatly enhance the precision, effectiveness, and expandability of the attendance management system. In conclusion, this advancement will augment the system's practicality in various organizational contexts, further harmonizing it with the ever-changing requirements of the modern business environment.

CONCLUSION

An innovative design has been developed for an attendance management system that makes use of the Internet of Things (IoT), Amazon Web Services (AWS), an RFID module, Python Django, and Arduino Uno. This architecture provides a leading solution for attendance monitoring that encompasses increased accuracy, efficiency, and cost-effectiveness. The possibility of its adoption across a wide variety of organizational landscapes holds the promise of improved attendance management procedures, which will ultimately result in increased levels of productivity. The capability of the system to give real-time attendance tracking and reporting makes the process of monitoring and managing attendance in businesses a considerable amount simpler. In addition, the system that has been offered stands out due to its scalability, flexibility, and user-friendliness of its interface. These characteristics position it as a versatile solution that is suited for a variety of organizational settings.

REFERENCES

1. Singh, D., Reddy, A. and Kumar, S.S. (2016) IoT-Based Wireless Attendance Management System Using Finger Print Recognition. INROADS-An International Journal of Jaipur National University, 5, 21-29. doi.org/10.5958/2277-4912.2016.00074.6
2. Trio Adiono, Didi Setiawan, Maurizfa, Jason William, Nana Sutisna, "Cloud Based User Interface Design for Smart Student Attendance System", 2021 International Symposium on Electronics and Smart Devices (ISESD), pp.1-5, 2021.
3. Fahim Faisal, Syed Akhter Hossain, Abdus Sattar, "ONGULANKO: An IoT Based Biometric Attendance Logger", 2021 5th International Conference on Intelligent Computing and Control Systems (ICICCS), pp.369-377, 2021.
4. Prince, N., Sengupta, A. and Unni, M. (2016) Implementation of IoT Based Attendance System on a Dedicated Web-Server. International Journal of Scientific & Engineering Research, 7, 351-355
5. Kawaguchi, Y., Shoji, T., Weijane, L., Kakusho, K. and Minoh, M. (2005) Face Recognition-Based Lecture Attendance System. The 3rd AEARU Workshop on Network Education, Tokyo, 2005, 70-75.
6. Subramaniam, H., Hassan, M., et al. (2013) Bar Code Scanner Based Student Attendance System (sas). JurnalTICom, 1, No. 3.





Chitra et al.,

7. Younis, M.I., Al-Tameemi, Z.A., Ismail, W. and Zamli, K.Z. (2013) Design and Implementation of a Scalable Rfid-Based Attendance System with an Intelligent Scheduling Technique. *Wireless Personal Communications*, 71, 2161-2179. <https://doi.org/10.1007/s11277-012-0929-3>
8. Cho, Y.S. and Kim, K.M. (2015) Nfc-Based Attendance Checking System for Institutions of Higher Education. *KIISE Transactions on Computing Practices*, 21, 283-289. <https://doi.org/10.5626/KTCP.2015.21.4.283>
9. Gopala, K.V.B., Sekhar, S.C., Babu, N.R. and Ravi, K.S. (2012) The Design of Intelligent Campus Security & Attendance System Based on RFID, GSM and Zig-Bee. 2250-3021.
10. Yadav, D.K., Singh, S., Pujari, S. and Mishra, P. (2015) Fingerprint Based Attendance System Using Microcontroller and Labview. *International Journal of Advanced Research in Electrical, Electronics and Instrumentation Engineering*, 4, 5111-5121. <https://doi.org/10.15662/ijareeie.2015.0406029>
11. Kavitha, G. and Sankara, K. (2018) Design and Implementation of Smart Attendance System using Raspberry Pi. *International Journal of Scientific Research in Computer Science, Engineering and Information Technology*, 3, 26-33.
12. Monk, S. (2015) *Programming the Raspberry Pi: Getting Started with Python*. TAB Electronics, New York.
13. Maksimović, M., Vujović, V., Davidović, N., Milosević, V. and Perisić, B. (2014) Raspberry Pi as Internet of Things Hardware: Performances and Constraints. *Design Issues*, 3, 8.
14. Ruiyan, C.A.I. (2012) Principle and Application of Arduino. *Electronic Design Engineering*, 16, 047

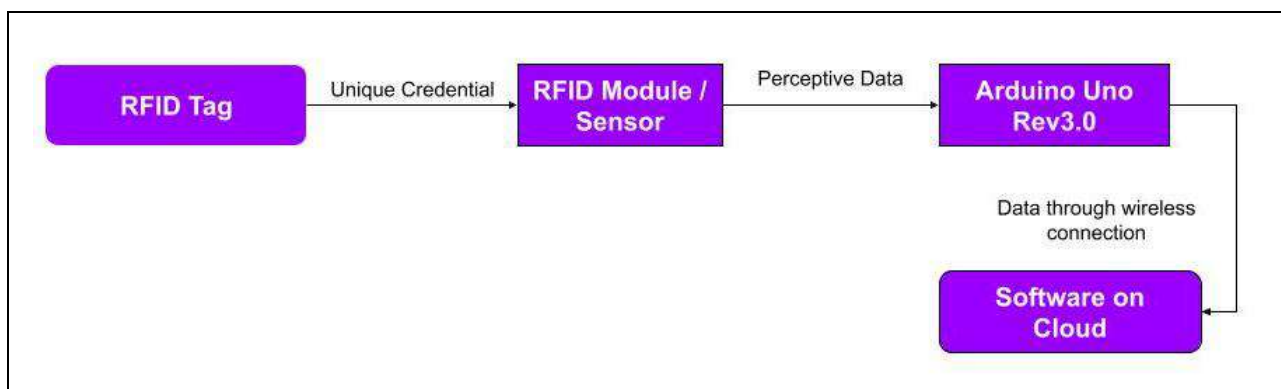


Fig 1. Hardware components flow diagram

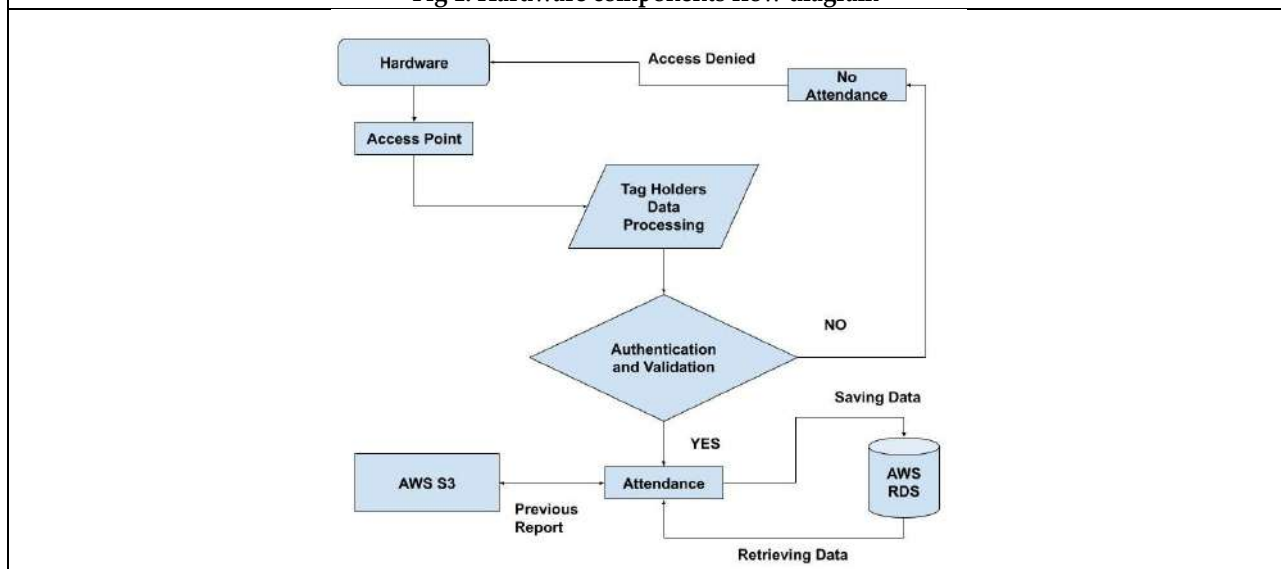


Fig 2. Software components flow diagram





Chitra et al.,

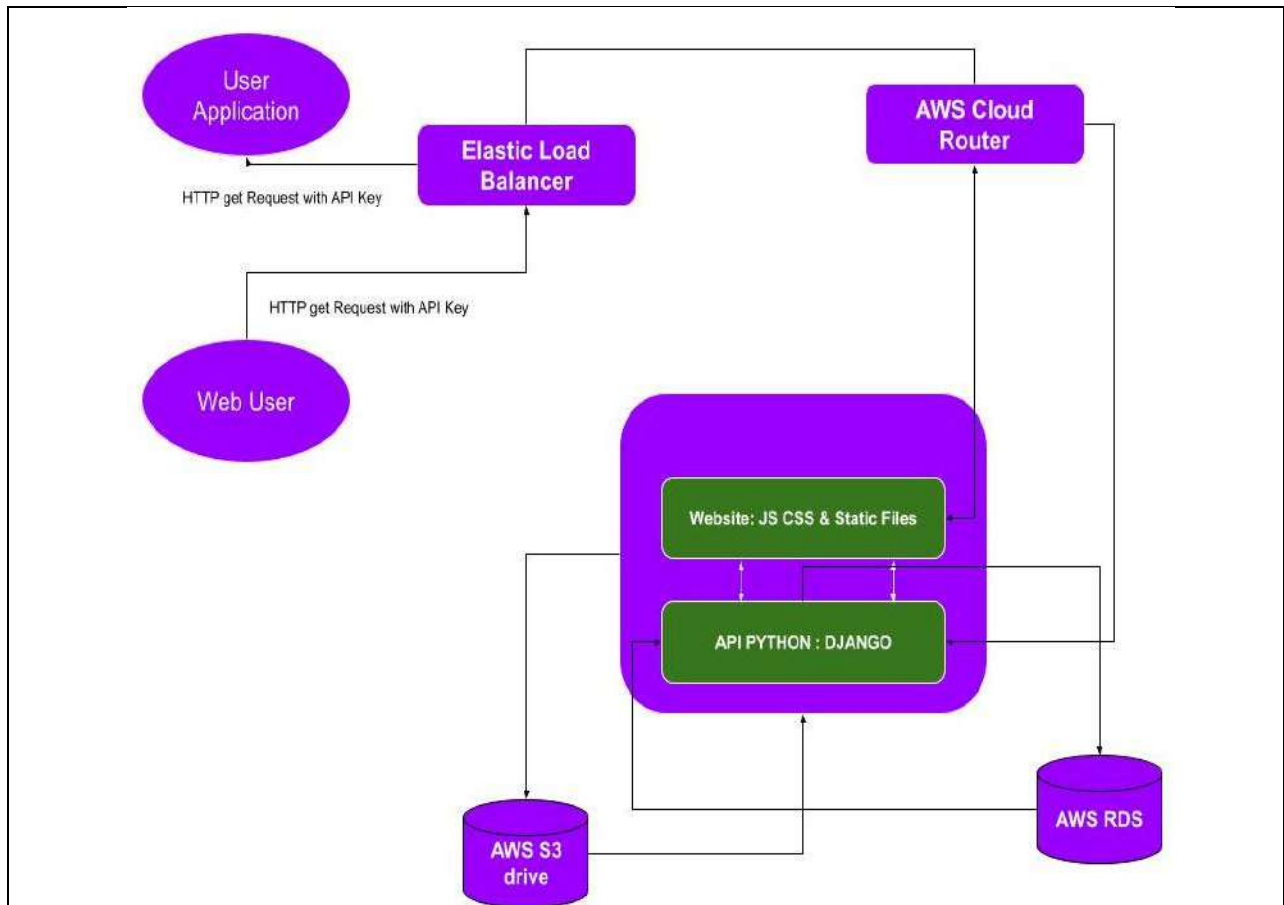


Fig 3.Cloud components flow diagram

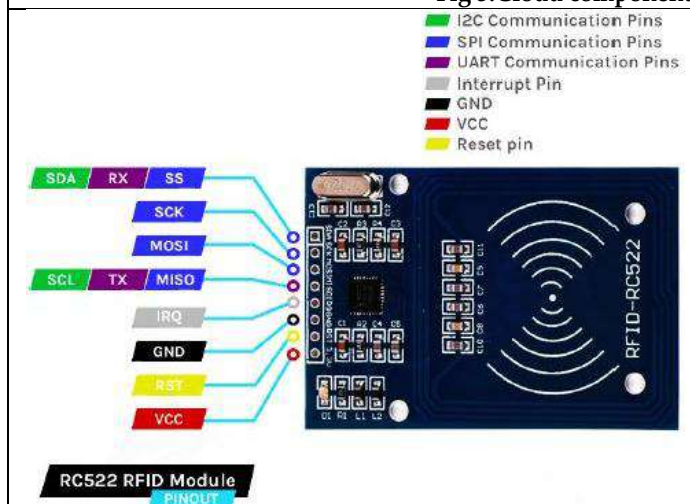


Fig 04. RC522 RFID Module

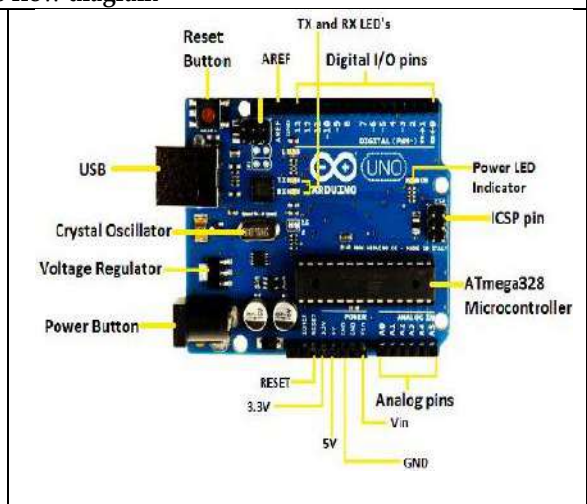
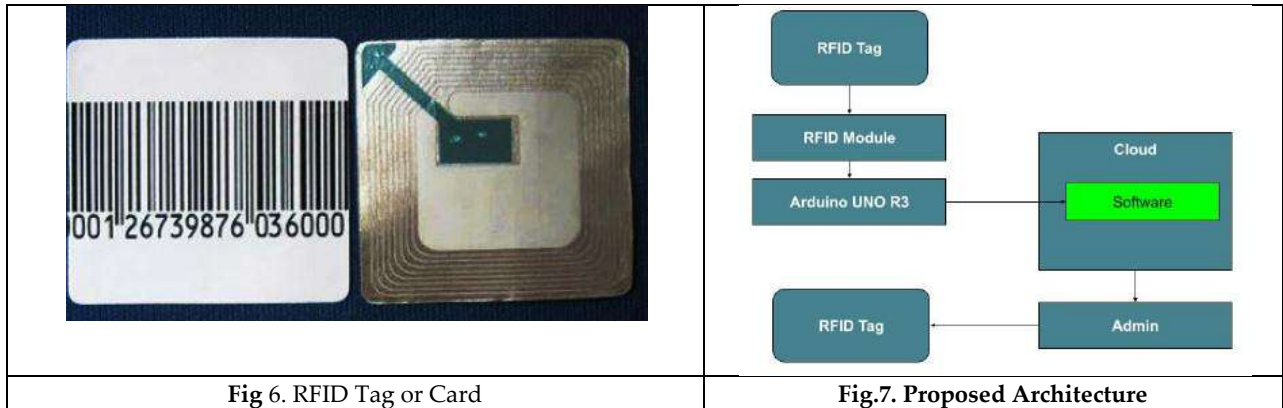


Fig 5. Arduino UNO R3





Chitra et al.,





Effect of Various Organic Fertilizers in Growth and Yield of Turnip (*Brassica rapa* L.) Under Low Hills of Uttarakhand

Ashish Chand Ramola^{1*}, Suneeta Singh² and Anil Kumar Saxena³

¹Research Scholar, Department of Horticulture, SGRR University, Pathribagh Campus, Dehradun, Uttarakhand, India.

²Professor & Head, Department of Horticulture, SGRR University, Pathribagh Campus, Dehradun, Uttarakhand, India.

³Associate Profesor & Head, Department of Soil Science, SGRR University, Pathribagh Campus, Dehradun, Uttarakhand, India.

Received: 21 Jun 2024

Revised: 25 Sep 2024

Accepted: 04 Nov 2024

*Address for Correspondence

Ashish Chand Ramola

Research Scholar,

Department of Horticulture, SGRR University,

Pathribagh Campus, Dehradun, Uttarakhand, India.

E.Mail: ashishchand902@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

During 2022-23, a field experiment was planned and conducted at the Horticulture Research Block, School of Agriculture Sciences, SGRRU, Dehradun, Uttarakhand, to investigate the "Effect of various organic fertilizers in growth and yield of Turnip (*Brassica rapa* L.) under low hills of Uttarakhand." The experiment was laid out in a randomized block design with three replications and ten treatments. The treatments comprised the following levels of different organic manures with different concentrations: T1: control (soil@100%), T2: (FYM@25tons/ha), T3: (Vermicompost @6tons/ha), T4: (Jeevamrit@100%), T5: (Biostimulant@3%), T6: FYM + Vermicompost (25 tons/ha + 6 tons/ha), T7: Vermicompost + Jeevamrit (6 tons/ha + 100%), T8: FYM + Biostimulant (25 tons/ha + 3%), T9: FYM + Vermicompost + Jeevamrit + Biostimulant (12.5 tons/ha + 3 tons/ha + 50% + 1.5%) and T10: FYM + Vermicompost + Jeevamrit + Biostimulant (25 tons/ha + 6 tons/ha + 100% + 3%).The sowing of Turnip "Purple Top White Globe" was done on 18/11/2022, and the final harvest was conducted on 11/02/2023. Observations on various attributes such as growth, yield, quality, and economics were recorded.The results revealed that treatment T6 (FYM + Vermicompost (25 tons/ha + 6 tons/ha)) was found to be the most effective in terms of vegetative characters such as plant height (45.93 cm), number of leaves (17), leaf length (26.82 cm), leaf width (13.02 cm), fresh weight of leaves (64.39 g), and dry weight of leaves (16.12 g). Yield attributes were also the highest for T6, including root diameter (11.53 cm), root length (9.25 cm), root weight (109.83 g), yield (17.46 kg/plot) and total yield (178.2 q/ha).

Keywords: Turnip, randomized, vermicompost, biostimulant, Jeevamrit



Ashish Chand Ramola *et al.*,

INTRODUCTION

Turnip, scientifically known as *Brassica rapa* L., is a globally important root crop and a close relative of Arabidopsis. It belongs to the family Brassicaceae and Cruciferae, with a chromosome number of $2n=20$. Turnip has been cultivated for about 2000 years across a wide range of climates, from Western Europe to China and Norway to the African desert (Nawaz *et al.*, 2019). Various common vegetables are included in the genus Brassica, including *Brassica rapa* L., also known as shaljam. Since prehistoric times, turnip has been used for human consumption and is one of the oldest cultivated vegetables, particularly famous in Europe and thriving in temperate climates (Mourato *et al.*, 2015). Vegetables play a major role in Indian agriculture by providing nutritional and economic security and yielding higher returns per unit area and time. India, largely a vegetarian society, depends heavily on vegetables for the bulk of its nutritional requirements. Since independence, India has emerged as the second-largest producer of vegetables in the world, after China. Vegetable production has increased from less than 20 million tons before independence to 212.53 million tons, and productivity has increased to 17.3 million tons/ha in 2022-23. The area under vegetable cultivation has grown from 5.5 million ha in 1991-92 to 11.477 million ha in 2022-23. According to the National Horticulture Database 2011, India is the largest producer of ginger and okra and ranks second in the production of potatoes (10%), onions, cauliflower, and cabbage, earning the moniker of the fruit and vegetable basket of the world. Over the last five years, an overall growth rate of 2.08% in area, 1.64% in productivity, and 3.72% in total production has been achieved. The demand for vegetables in India is projected to reach 350 million tons by 2030 (Singh, 2019). Turnip is a vital root vegetable and forage crop, indigenous to Asia, Europe, Russia, and the Near East, and is now widely cultivated globally as both a vegetable and oil source. In Pakistan, turnip was cultivated on 9609 hectares of land with a production of 167,065 tons in 2020-2021, making it an important local vegetable. Turnip has been reported to have various medicinal benefits, including therapeutic properties against kidney and liver diseases and other ailments. It also possesses antimicrobial, anti-inflammatory, antitumor, cardio protective, anti-diabetic, analgesic, and nephroprotective properties, and aids against metabolic syndrome and obesity. Furthermore, it improves growth in response to heavy fertilization, which can cause non-point pollution (Raza *et al.*, 2023). Turnip is commonly grown in temperate climates for its white, fleshy taproot and is one of the oldest cultivated vegetables. It is an excellent source of bioactive compounds, potentially delivering health benefits when consumed (Jadid *et al.*, 2021). Brassica, the most important genus in the Brassicaceae family, consists of about 350 genera and nearly 3,500 species. Young turnip roots are commonly consumed raw in salads, and turnip leaves, characterized by a bitter taste due to trace amounts of phenolic compounds, differ from other Brassica vegetables in their trivial antioxidant capacity (Dejanovic *et al.*, 2021). In the UK, turnip, a traditional root vegetable, is less frequently consumed compared to other Brassica vegetables. In 1992, turnip production accounted for 5400 hectares, which dropped to less than 2700 hectares by 2017 (Nor *et al.*, 2020). According to the Food and Agriculture Organization (FAO) of the United Nations, 92 million tons of Brassica plants are grown in 150 countries, occupying 5.4 million hectares of land, with 70% of this area in Asia and China (Chihoubet *et al.*, 2019). The decision to conduct the present experiment with organic fertilizers such as Jeevamrit and biostimulants for turnip cultivation in the low hills of Uttarakhand is driven by the scarcity of research in this area. Recognizing the lack of studies on the efficacy of these specific fertilizers on turnip crops in this region, this experiment seeks to address this knowledge gap. By investigating the impact of Jeevamrit and biostimulants on turnip cultivation in the unique environmental conditions of Uttarakhand's low hills, the study aims to provide valuable insights that can benefit local farmers and contribute to sustainable agricultural practices in the region.

MATERIALS AND METHODS

The present research was conducted at the Horticulture Research Block, Department of Horticulture, School of Agricultural Sciences, Shri Guru Ram Rai University, Dehradun, Uttarakhand, during the rabi season of 2022-23. The experiment was laid out in a Randomized Block Design (RBD) with three replications. Ten treatments were tested: T₁



**Ashish Chand Ramola et al.,**

(control), T₂ (Farm Yard manure @22t/ha), T₃ (Vermicompost @ 5t/ha), T₄ (Jeevamrit @100%), T₅ (Biostimulant @3%), T₆ (FYM @22t/ha + Vermicompost @5t/ha), T₇ (Vermicompost @5t/ha + Jeevamrit @100%), T₈ (FYM @22t/ha + Biostimulant @3%), T₉ (FYM @11 t/ha + Vermicompost @2.5 t/ha + Jeevamrit @50% + Biostimulant @1.5%) and T₁₀ (FYM @22t/ha + Vermicompost @5t/ha +Jeevamrit @100% + Biostimulant @3%). The soil of the research field was sandy loam with a pH of 7.12, containing available nitrogen (220.04%), available phosphorus (9.1 kg/ha), and available potassium (18.1 kg/ha). The turnip cultivar "Purple Top White Globe" was selected for this study. Organic fertilizers (Vermicompost, FYM, Jeevamrit, and Biostimulant) were incorporated into the experimental field as per the treatment specifications during the final field preparation. Seeds were sown on 18/11/2022, and all necessary cultural practices were carried out at regular intervals throughout the research period. During the experiment, five plants were randomly selected from each replication to record various observations on growth and yield characteristics at 30 days, 60 days after sowing, and at final harvest. The data obtained were statistically analyzed using standard statistical methods as suggested by Gomez and Gomez (1996).

RESULT AND DISCUSSION

Plant height

The data recorded on height of plant at different harvesting are presented in the Table 2 and Fig. 1. At 30 DAS, the maximum plants height (15.78 cm) was recorded in T₁₀. The treatment T₉ (14.41 cm) and T₈ (14.49 cm) was at par with each other. However, significant differences were observed with treatment T₁ (11.21 cm), T₆ (13.16 cm) and T₈ (14.49 cm). The minimum plant height was recorded in T₃ (9.59 cm). At 60 DAS the maximum plants height (34.99 cm) was recorded in T₅, which at par T₆ (34.14 cm), T₇ (34.02 cm) and T₁₀ (34.13 cm). However, significant differences were observed with treatment T₁ (29.56 cm), T₂ (28.3 cm), T₃ (31.11 cm), T₄ (32.3 cm) and T₁₀ (34.13 cm). The minimum plant height was recorded in T₂ (28.3 cm). At final harvest the maximum plants height was recorded in T₄ (45.93 cm) with FYM @ 5 tones/ha + V.C @ 2.5 tones/ha which was at par with T₆ (45.68 cm) and T₁₀ (45.56 cm). However, significant difference was observed with treatment T₃ (42.88 cm), T₉ (43.66 cm), T₈ (35.92 cm) and T₂ (37.79 cm). While, the minimum plant height was recorded in T₁ (35.74 cm). The probable reasons for enhanced a greater plant height may be due to promotive effect of macro and micro nutrients on vegetative growth ultimately led to more photosynthetic activities. The findings are in agreement with Sapkota *et al.*, 2021; Singh *et al.*, 2020 and Yadav *et al.*, 2020.

Number of leaves per plant

The data recorded on height of plant at different harvesting are presented in the Table 2 and Fig. 2. At 30 DAS the number of leaves per plant ranged from 5 to 6. On the basis of mean the maximum number of leaves per plant was counted in T₁, T₂ and T₁₀ (6) which at par with each other. However, significant differences were observed in T₁ (6) and T₃ (5). The minimum number per plant was recorded in the treatment T₇ and T₈ (5). In the case of 60 DAS, the mean value for number of leaves per plant was found maximum in T₉ (12). The treatments T₁ (11), T₄ (11), T₆ (11), T₇ (11) and T₁₀ (11) was at par with each other. However, significant differences were found in T₁, T₃ and T₉. The minimum number of leaves per plant was recorded in the treatment T₇ or T₈ (5). At final harvest after sowing significant differences and on the basis of mean the maximum number of leaves per plant were counted in the treatment T₈ (17) which were at par with T₂ (17). However, significant differences were found with rest of the treatment T₁ (15), T₄ (14), T₅ (15) and T₇ (13). The minimum number of leaves per plant was recorded in the treatment T₃ (12). The probable reasons for enhanced a greater number of leaves may be due to promotive effect of macro and micro nutrients on vegetative growth ultimately led to more photosynthetic activities. Similar findings are also reported by Kiran *et al.*, 2017 and Rawat *et al.*, 2021.

Leaf length

The observation of leaf length was recorded at 30, 60 DAS and at final harvest the results show significant differences between the treatments. At 30 DAS, the maximum value of leaf length was recorded in the treatment T₄ (8.92 cm) which were at par with T₅ (8.57 cm), T₆ (8.34 cm), T₉ (8.00 cm) and T₁₀ (8.52 cm). The significant differences were



**Ashish Chand Ramola et al.,**

observed with the treatments T1, T2, T3, T4, T5 and T6. The minimum value (7.1 cm) of leaf length was recorded under the treatment T3. At 60 DAS, the maximum number of leaf length was recorded in treatment T8 (23.89 cm). The treatments T5, T6, T7, T8 and T9 were at par with each other. The significant differences were observed with the treatment T1 (18.16 cm), T3 (20.78 cm), T4 (19.75 cm), T8 (21.75 cm) and T10 (22.45 cm). The minimum leaf length (18.16 cm) was recorded under the treatment T1. At final harvest, the leaf length was maximum in T10 (33.50 cm) which was at par with T4 (26.44 cm). However, significant differences were observed with treatment T1 (20.81 cm), T3 (22.11 cm), T6 (25.68 cm) and T8 (22.46 cm). The minimum leaf length was recorded in the treatment T2 (19.47 cm). The application of FYM @ 5 tones/ha + Vermicompost @ 2.5 tones/ha + V.W @ 25 increases the leaf length of turnip may be due to application of major and minor nutrients, through organic manures in various levels, increased the photosynthetic activity, chlorophyll formation, nitrogen metabolism and auxin contents in the plants which ultimately increases the plant height. The findings are also in conformity with the findings of Joshi *et.al.*, 2015.

Leaf width

The leaf width of the plant at different stages of harvesting is presented in Table 2. and Fig. 4. The leaf width on 30 days after sowing differs significantly and was ranging from 3.7 to 4.85 cm. The maximum leaf width was recorded in T6 (4.85 cm) which was statistically at par with T5 (4.66 cm) and T4 (4.53 cm). However, significant differences were observed with the treatments T1 (3.9 cm), T2 (4.01 cm), T3 (3.71 cm). The minimum leaf width was recorded in the treatment T1 (3.9 cm). In 60 DAS, the maximum leaf width was recorded in T4 (10.81 cm) which was at par with treatment T6 (10.02 cm). However, significant differences were observed with treatment T1 (8.50 cm), T4 (10.81 cm), T5 (9.69 cm) and T9 (8.7 cm). At final harvest, the maximum leaf width (13.02 cm) was recorded in the concentration of organic manures treatment T4 (Vermicompost @ 5 tones/ha) which is found to be on par with treatment T1. The minimum was recorded in this might be due to the continuous nutrient availability by the use of organics. This is found to be accordance with findings of Dlamini *et.al.*, 2020.

Root length

The maximum root length was recorded in treatment T4 (9.25 cm) with the application of Vermiwash @ 50 %. The minimum root length was recorded in T3. The significantly highest root length might be due to beneficial effect of organic nutrient sources. The organic nutrient sources, particularly vermicompost improves the soil structure and soil quality which might have resulted in length of root. The findings are in similar with the results of Joshi *et.al.*, 2015; Aisha *et.al.*, 2014 and Pamula *et.al.*, 2020.

Root weight

The maximum root weight of turnip (135.74 g) was recorded in T10 with FYM @ 5 tones/ha + Vermicompost @ 2.5 tones/ha + Vermiwash @ 25% significantly superior to all other treatments. The minimum root weight was recorded in T7. The results show significant differences between the treatments. The increase in root weight of leaves may be due to the excellence of high level of organic manures was producing good growth of turnip plants which show higher root weight. The findings are in similar with Getaneh *et.al.*, 2019 and Joshi *et.al.*, 2015.

Root diameter

The maximum root diameter was recorded in T7 (8.35 cm) with FYM @ 5 tones/ha + V.C @ 5 tones/ha which was at par with T4 significantly superior to all treatments. The minimum root diameter was recorded in T3. The decrease in bulk density and increase in porosity and water holding capacity of the soil due to organic nutrient source might have contributed in increasing root diameter of plants. The results are similar with Pamula *et.al.*, 2020 and Konet *et.al.*, 2016.

Fresh weight of leaves

The maximum total fresh weight of leaves (66.44 g) was recorded in T4 with Vermiwash @ 50% which was significantly superior to all other treatments. The minimum total fresh weight of leaves was recorded in T8. The results show significant differences between the treatments. The increase in total fresh weight of leaves may be due to





Ashish Chand Ramola *et al.*,

the excellence of high level of organic manures was producing good growth of turnip plants which show higher total fresh weight of leaves. The findings are in accordance with Basheer *et al.*,2013.

Dry weight of leaves

The maximum total dry weight of leaves (16.12 g) was recorded in T4 with vermiwash @ 50 % which was significantly superior to all other treatments. The minimum total dry weight of leaves was recorded in T4. The results show significant differences between the treatments. The increase in total dry weight of leaves may be due to the excellence of high level of organic manures was producing good growth of turnip plants which show higher total dry weight of leaves. Similar results were also reported by Joshi *et al.*,2015 and Kaur *et al.*, 2019.

Root yield (kg/plot)

The maximum root yield of turnip (17.46 kg) was recorded in T10 FYM @ 5 tones/ha +Vermicompost @ 2.5 tones/ha + Vermiwash @ 25% + Jeevamrit @ 50 %which was significantly superior to all other treatments. The minimum root yield was recorded in T5. The results show significant differences between the treatments. The increase in root yield may be due to the excellence of high level of organic manures was producing good growth of turnip plants which show higher root yield. The findings are in agreement with Jadhav*et al.*,2015; Getaneh*et al.*, 2019 and Yogeshappa*et al.*, 2017.

Root yield (t/ha)

The maximum root yield of turnip (17.15 t/ha) was recorded in T3 with FYM @ 10 tones/ha significantly superior to all other treatments. The minimum root yield was recorded in T5. The results show significant differences between the treatments. The increase in root yield may be due to the excellence of high level of organic manures was producing good growth of turnip plants which show higher total root yield. The findings are parallel with Getaneh*et al.*,2019 and Ghimire*et al.*, 2020.

CONCLUSION

Based on the current investigation on the "Effect of Various Organic Fertilizers on the Growth and Yield of Turnip (*Brassica rapa* L.) Under Low Hills of Uttarakhand," it can be concluded that the treatment T6 (Farmyard Manure@ 25 tons/ha + Vermicompost @6 tons/ha) resulted in the best performance. Specifically, the following outcomes were observed with this treatment: Growth attributes viz., maximum plant height (45.93cm), number of leaves (17), leaf length (26.82cm), leaf width (13.02cm), fresh weight of leaves (64.39g) and dry weight of leaves (16.12 g) and yield attributes viz., root diameter (11.53 cm), root length (9.25 cm), root weight (109.83 g), yield (17.46 kg/plot) and total yield (178.2 q/ha).The study indicates that the combined use of FYM and Vermicompost, supplemented with Jeevamrit, significantly enhanced soil fertility, which in turn led to improved yield and quality of turnip crops. Therefore, under the current agro-climatic conditions of the low hills of Uttarakhand, it is recommended to use this combination of organic fertilizers to achieve sustainably higher yields and quality in turnip cultivation.

REFERENCES

1. Aisha, Ali H., Shafeek M., Mahmoud, Asma R. and Desuki M. 2014. Effect of Various Levels of Organic Fertilizer on the Growth and Root Quality of Turnip (*Brassica rapa*L.). Current Science International. 3(1):7-14.
2. Basheer M. and Agarwal O.P., 2013. Effect of vermicompost on the growth and productivity of tomato Plant (*Solanum lycopersicum*) under field conditions. International Journal of Recent Scientific Research Vol. 4, Issue, 3, pp.247.
3. Chihoub, Dias M., Barros L., Calhella R., Alves M., Skhiri F. and Ferreira I. 2019. Nutritional value of the green waste parts from turnip (*Brassica rapa*). Journal of Food Research International. Vol.126 (4):112-120.





Ashish Chand Ramola et al.,

4. Dejanovic G., Asllana E., Gamba M., Raguindin P., Itodo O., Minder B., Bussler W., Metzger B., Muka T., Gilisic M. and Kern H. 2021. Phytochemical characterization of turnip greens (*Brassica rapa*): A systematic review. Journal of Plos One. 12(4). PP: 1-16.
5. Dlamini V.C., Nxumulo K.A, Masarirambi M.T, Wahome PK, Oseni T.O and Zwane M.G. 2020. Effects of cattle manure on growth, yield quality and self- life of beetroot (*Beta vulgaris* L.) Journal of Experimental Agriculture International. Vol. 42(1):345-356.
6. Getaneh T. and Mezgebu A. 2019. Integrated effect of nitrogen and vermicompost levels on yield and yield components of carrot (*Daucuscarota* L.) at Woreta, Northwestern Ethiopia. Journal of Horticulture and Forestry, Vol.11(6): 111-123.
7. Ghimire S., Adhikari B., Pandey S., Belbase K., Lammichanne S. and Pathak R. 2020. Effect of Different Organic Manure on Growth and Yield of Radish. Vol. 4(3)
8. Jadid R, Nosrati M , Rasouli B , Seidavi A, Clive J, Phillips C. 2021 :The Effects of Turnip (*Brassica rapa*L.) Extract on the Growth Performance. Journal of animals. 34(6):pp. 867.
9. Jadhav P.B, Kireeti A, Patil N.B, Dekhane S.S and Patel D.J, 2014 : Effect of different levels of vermiwash spray on growth and yield of radish cv. Local variety. Asian J. Hort.Vol. 9(2), ASPEE Agricultural Research and Development Foundation.
10. Kaur P., Bhardwaj M. and Babbar I. 2015. Effect of vermicompost and vermiwash on growth of vegetables. Deptt. Of Zoology, B.B.K D. A.V Collage for Women, Amritsar, Punjab, INDIA. ISSN 2330-6535 Vol. 3(4), 220-228.
11. Kiran M., Jilani M.S. and Waseem K. 2017.Impact of different organic manures and NPK application on the growth and yield of turnip (*Brassica rapa* L.). Pakistan Journal of Science. Vol. 69.(7):335-340.
12. Kon H.I.F, Bitar L. A.L, Kidane H.T, M.T, Wieczynska J, Zein S.E.B. 2016. Effects of different organic soil amendments on yield parameters of *Brassica rapa* L. Vol. 5(1):67-74.
13. Lestari S.U. andAndrian A. 2017. Effects of Urine cow dosage on growth and production of Sorghum plant (*Sorghum Bicolor* L) on peat land. International Conference on Environment and Technology (IC-Tech).
14. Mourato M., Ines, Moreira I., Leitao I., Pinto F., Sales J. and Martins L.2015. Effect of Heavy Metals in Plants of the Genus *Brassica*. Journal of Molecular Sciences. Vol. 16, 17975-17998.
15. Nawaz M., Ahmed K., Qadir G., Rizwan M., Nawaz M. and Sarfraz M. 2019. Growth and Yield of Turnip (*Brassica rapa*L.) in Response to Different Sowing Methods.Journal of Agricultural Research, Vol. 33(1): 126-134.
16. Nor N., Lignou S., Bell L. Price, Harvey C. and Methven L. 2020. The Relationship between Glucosinolates and the Sensory Characteristics of Steamed-Pureed Turnip (*Brassica Rapa* L.) Journal of University of Reading. Vol. 9: pp.1719.
17. Pamula S. and Kerketta A. 2021.Effect of Organic Manures and Inorganic Fertilizers on Growth and Yield of Turnip. Journal of Current Microbiology and Applied Sciences,Vol. 10(01);67-78.
18. Rawat S. and Pant S.C. 2021. Effect of organic manures, inorganic fertilizers and their combinations on yield of radish. International Journal of Recent Advances in Multidisciplinary. Vol. 2(2):230-237.
19. Raza Q. , Bashir M., Rehim A., Geng Y., Raza H., Hussain S., Ahmad I. and Wasif M. 2023. Identifying the Role of Biostimulants in Turnip Production. Journal of sustainability, Vol. 15, pp. 11851.
20. Sapkota A., Sharma M..D., Giri H., Shrestha B. and Panday D. 2021. Effect of organic and inorganic sources of nitrogen on growth, yield and quality of beetroot.Vol. 5(2):1130-1136.
21. Singh J., Gandhi N., Singh K., Tinna D. and Singh S. 2019. Effect of the organic manure, inorganic fertilizers and their combination on growth, yield and quality of radish (*Raphanussativus* L.) Journal of Pharmacognosy and Phytochemistry. Vol. SP4: 57-59.
22. Yadav C., Mishra S., Singh M., Roy S. and Tiwari P. 2020. Effect of FYM and Vermicompost on Growth and yield of turnip (*Brassica rapa* L.) cv. Purple top white. Journal of The Pharma Innovation, Vol. 10(2) pp.100-103.
23. Yogeshappa, Murthy H.S and Krishnamurthy C.A. 2016. Effect of repeated application of human urine and cattle urine on soil properties and yield of vegetable crops. Asian J. Soil Sci., Vol. 11(1):45-56.





Ashish Chand Ramola et al.,

Table:1 Treatment details

No. of Treatment	Combinations	Concentration
T ₁	Control	-
T ₂	FYM	25tones/ha
T ₃	Vermicompost	6tones/ha
T ₄	Jeevamrit	100%
T ₅	Biostimulant	3%
T ₆	FYM + Vermicompost	25tones/ha + 6 tones/ha
T ₇	Vermicompost + Jeevamrit	6 tones/ha + 100%
T ₈	FYM + Biostimulant	25tones/ha + 3%
T ₉	FYM+Vermicompost+ Jeevamrit + Biostimulant	12.5tones/ha+3tones/ha +50%+1.5%
T ₁₀	FYM+Vermicompost+jeevamrit+ Biostimulant	25tones/ha+ 6 tones \ ha+100 %+ 3%

Table 2: Effect of organic fertilizers on plant height (cm), number of leaves per plant, leaf length (cm) and leaf width cm) of Turnip at different harvest intervals

Treatment	Plant height (cm)			Number of leaves per plant			Leaf length (cm)			Leaf width (cm)		
	30 DAS	60 DAS	At Final harvest	30 DAS	60 DAS	At Final harvest	30 DAS	60 DAS	At Final Harvest	30 DAS	60 DAS	At Final Harvest
T ₁	11.21	29.56	35.74	6	11	15	7.48	18.16	21.06	3.9	8.50	12.3
T ₂	10.67	28.3	37.79	6	10	16	7.24	18.82	19.47	4.01	8.95	11.17
T ₃	9.56	31.11	42.88	5	10	12	7.1	20.78	22.11	3.71	8.90	10.54
T ₄	11.86	32.3	45.93	6	11	14	8.92	19.75	23.64	4.53	10.81	13.20
T ₅	11.61	34.99	44.66	6	10	15	8.57	16.21	25.40	4.66	9.69	11.44
T ₆	13.16	34.14	45.68	6	11	14	8.34	21.32	25.90	4.85	10.02	10.7
T ₇	13.51	34.02	41.96	5	11	13	7.81	21.26	30.30	4.37	9.43	11.2
T ₈	14.49	31.61	35.92	5	10	17	7.73	23.89	29.50	4.3	9.44	11.68
T ₉	14.41	32.21	42.66	5	12	13	8.00	21.53	21.53	3.97	7.71	9.62
T ₁₀	15.78	34.13	45.56	6	11	15	8.52	22.45	33.50	4.11	8.03	10.62
C.D(0.05%)	0.714	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	1.135	N/A	N/A
SE(m) ±	0.238	5.46	6.07	1.3	1.54	2.31	0.326	2.33	3.78	0.379	0.84	1.79
SE(d) ±	0.337	7.73	8.59	3.5	2.18	3.26	0.461	3.30	2.34	0.536	1.18	2.53
C.V.	1.495	1.31	1.10	20.3	2.27	2.23	0.51	2.40	2.96	1.14	1.98	1.41

Table 3: Effect of organic fertilizers on fresh weight of leaves (g), dry weight of leaves (g), length of root (cm), diameter of root (g), weight of root (g), root yield (kg/plot) and root yield (q/ha) of turnip at final harvest

Treatment	Fresh Weight of leaves (g)	Dry weight of leaves (g)	Root Length (cm)	Diameter of Root (cm)	Root weight (g)	Root yield (kg/plot)	Root yield (q/ha)
T ₁	33.67	15.49	9.06	6.40	66.95	13.36	162.2
T ₂	34.36	15.34	8.20	6.76	77.12	15.06	152.5
T ₃	38.99	14.13	6.79	7.73	109.83	15.20	171.5
T ₄	56.92	16.12	7.59	8.53	97.59	14.70	148.5
T ₅	43.72	13.22	8.15	8.86	76.88	13.40	138.2





Ashish Chand Ramola et al.,

T ₆	64.39	15.95	7.58	7.53	74.98	17.46	178.2
T ₇	51.34	14.80	7.60	8.63	69.67	17.00	145.6
T ₈	46.58	15.15	8.48	9.70	85.65	14.43	178.2
T ₉	56.59	14.69	8.48	11.53	79.64	15.23	146.2
T ₁₀	51.10	15.95	9.25	7.23	86.19	15.16	152.2
C.D (0.05%)	2.95	N/A	1.18	N/A	N/A	1.97	2.11
SE(m) ±	2.65	0.83	0.39	1.34	12.38	0.65	0.70
SE(d) ±	0.75	1.18	0.55	1.89	17.51	0.93	0.99
C.V.	2.63	3.63	2.42	2.01	25.01	7.54	7.86

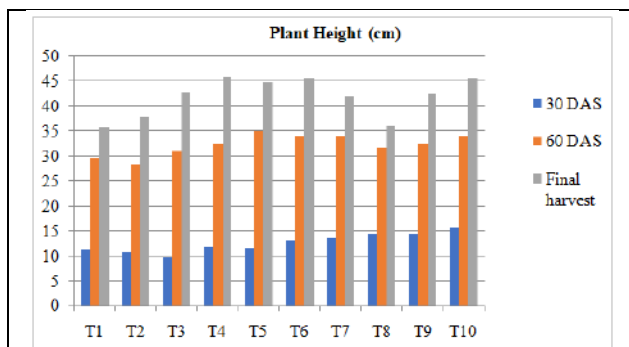


Fig 1: Plant height (cm) of turnip as influenced by organic manure at various stages of Harvesting

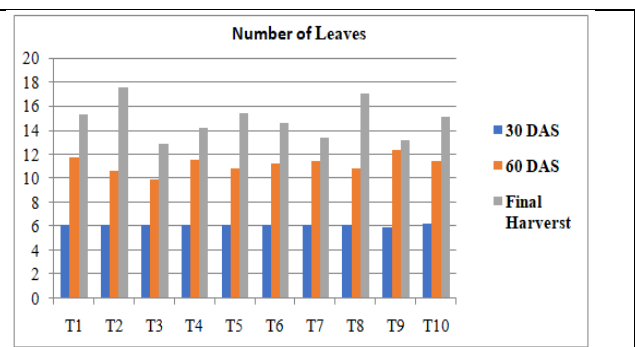


Fig 2: Number of leaves of turnip as influenced by organic manure at various stages of Harvesting

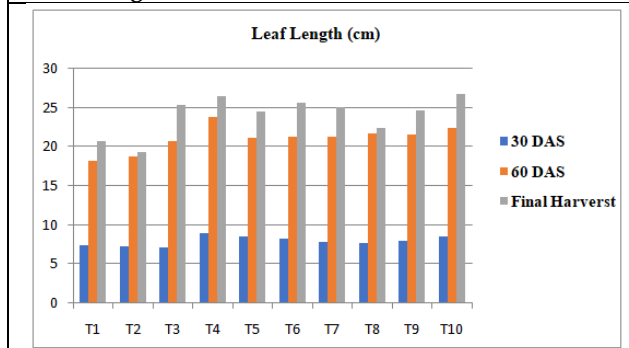


Fig. 3 : Leaf length (cm) of turnip as influenced by organic manure at various stages of Harvesting

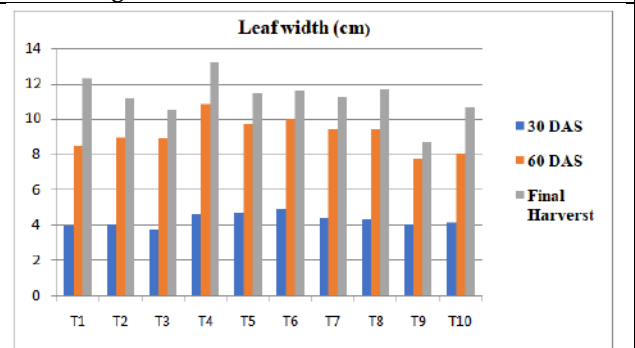
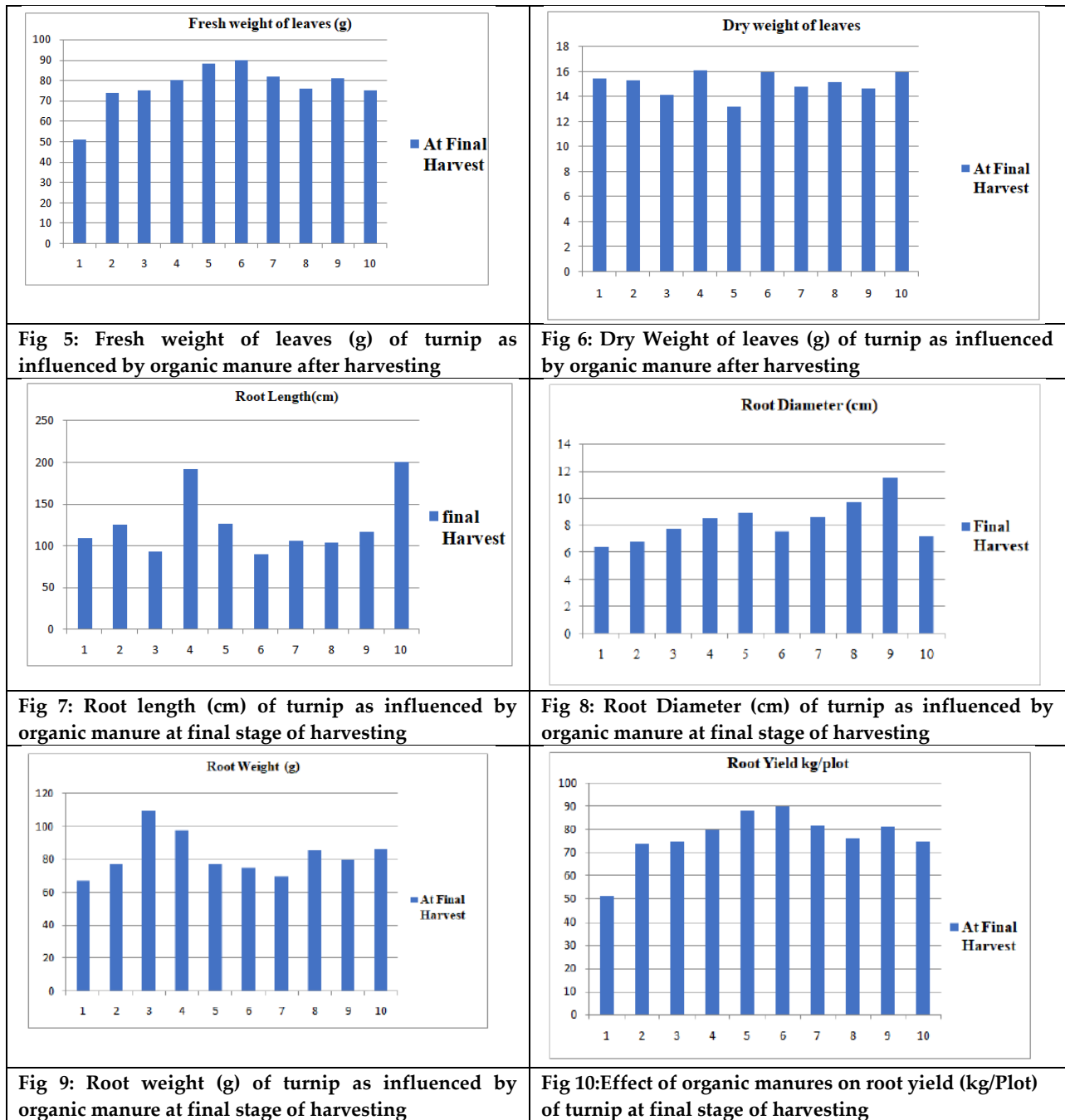


Fig 4: Leaf width (cm) of turnip as influenced by organic manure at various stages of harvesting





Ashish Chand Ramola et al.,





Ashish Chand Ramola et al.,

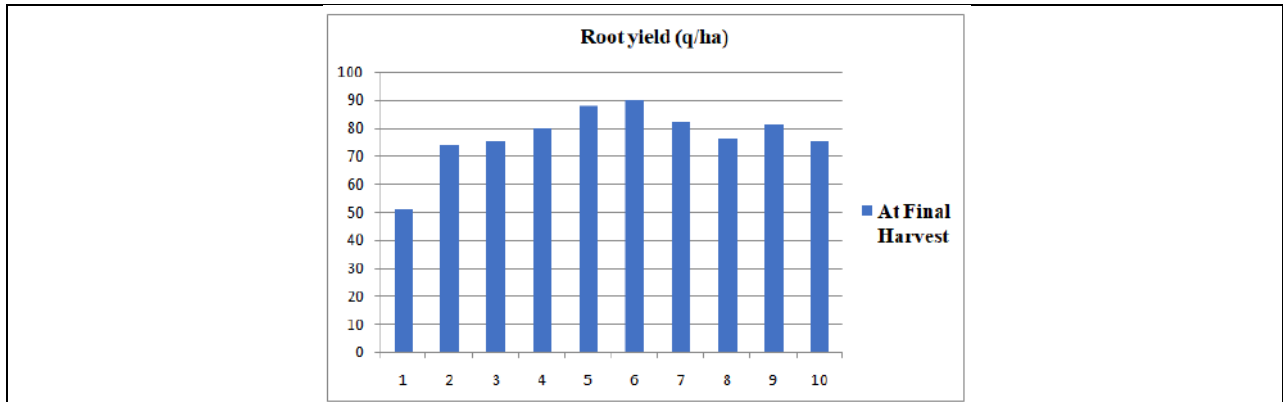


Fig 11:Effect of organic manures on root yield (q/ha) of turnip at final stage of harvesting





To Study the Relationship between Examination Stress and Changes in Eating Pattern among 18-25 y/o Students of Parul University

Vishwa Pandya^{1*} and Gayatri Parmar²

¹Jr.Dietitian, Department of Nutrition and Dietetics, Parul University, Waghodia,Gujarat, India.

²Assistant Professor, Department of Nutrition and Dietetics, Parul University, Waghodia, Gujarat, India.

Received: 22 Jun 2024

Revised: 28 Sep 2024

Accepted: 04 Nov 2024

*Address for Correspondence

Vishwa Pandya

Jr.Dietitian,

Department of Nutrition and Dietetics,

Parul University,

Waghodia,Gujarat, India.

E.Mail:



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The research study was based on the correlation between exam and stress that may or may not affect eating and sleeping patterns of 18-25 y/o students at Parul university. As these studies started in March 2023 ended in March 2024. Around 303 students (living in hostels or PG) were taken as the target group sample with snowball technique. We have designed an online google form for survey purposes and the questionnaire was broken down into five sections: general information, examination details, food habits, sleeping patterns, and perceived stress (aptitude test). The data collection was conducted in October month from 10th Oct 2023 to 25 oct 2023. A statistical analytical tool (SPSS) has been utilized for data analysis. As students don't worry about the external exam because Parul University administers exams, take the weekly and internal exam that covers half of the curriculum. This observational data relates only to the Parul university students. A total of 303 students answered the questions; upon evaluating the population by gender, it was found that 55.26% of the students were male and 44.7% of the students were female students of Parul University. These students will be appearing for the exam. According to the data we collected, roughly 33% of students had exams, 60% did not, and it is likely that 6% were unaware of the specific examination days. In October, these data were collected. Our research focused mostly on the eating habits of each individual body. Sixty percent of the pupils opt to eat vegetarianism because they are from Gujarat. Fifty percent of pupils experience nutritional modifications, whereas the other fifty percent do not experience any changes in their eating habits. Research indicates that dietary changes may cause sleep patterns to be disturbed, and that studying for college exams is the primary cause of these disturbances. Exams linked to sluggishness, dietary modifications, and PG students' favourite cooking techniques had a positive correlation. This result supports the alternative hypothesis—which shows a strong correlation between exam stress and changes in sleeping and eating patterns—is statistically supported, rejecting the null hypothesis.

Keywords : Stress, Emotional behaviour, examination days, correlation data, eating and sleeping pattern.



**Vishwa Pandya and Gayatri Parmar**

INTRODUCTION

In today's world, stress is a prominent factor in every individual's life due to several reasons following for the example like studies, relationship, social media influence, financial issue, family problem, office load, etc. WHO defines stress as "State of worry or mental tension caused by a difficult situation". Stress is a natural human response that prompts us to address challenges and threats in our lives. Everyone experiences stress to some degree. The way we respond to stress, however, makes a big difference to our overall well-being. Stress is nothing but a pressure that build up in the mind to complete any possible task in a particular period of time that changes mental, physical and social life such as an individual diet, social relationship, sleeping pattern and psychological factors such as unconscious mind, behaviour changes, feeling isolated, don't know the reason of anger, not eating mindfully, overthinking, relationship issue, self-doubt, mental health and trust issue.

The prevalence of stress used to be 37.12% in 2018 and is continuously increasing after the COVID-19 pandemic started in every country that increased stress up to 43.76% in 2023. So, it is noticed that there was a 6–7% increase in stress level. And it is assumed that the level of stress is continuously going to be increased in upcoming years due to competitive exam, job requirement, financial loan, etc. Stress can be classified into short-term and long-term duration. So, as short-term duration means it remains for a shorter period of time that contains the deadline of the project, relationship issue, certain diseases, etc. and about long-term duration means that remain more than a year contains diseases like cancer, heart & kidney disease, diabetes, hypertension, asthma, etc.

Stress is characterized into two types: -

Acute stress and chronic stress

Acute stress is comprised of short stress that quickly gets reduced by suppressing the load of a problem, for example, assignment submission, PPT presentation, project deadline, etc. Chronic stress that may comprise of long-term stress that may lead to serious diseases, for example, government exam, yearly university exam, dissertation, etc. This stress may lead to high blood pressure, diabetes, heart disease and obesity. This is due to the continuous presence of stressful life, one of the major changes comes in people's lifestyle that are related to eating behaviour.

Most of the times stress builds up because of heavy work load, arrival date of assignment submission and exam stress, bank load, home stress, office load, etc. Eating is a major component of life that is required in daily routine. Regular eating behaviour is playing an important role in nourishing the whole body from maintaining the body weight to give proper nutrients to each organ and make use of energy for the whole day. So, if we look around individuals in stress, they are often suffering from lifestyle changes in which the major factor eating behaviour changes too. Eating behaviour / eating disorder are nothing but a psychological disorder in which a person may eat more or eat very less, sometimes these may be caused by extreme stress conditions in any situation. Dietary patterns change when there is an excessive amount of stress that may lead to low glucose levels in the body. Students' lifestyle and dietary pattern is completely disturbed, especially for those who live alone without their family members. From making their own work food to completing college is really a tiring job to do so.

Students' stress may lead to eat an excess amount of kcal in high quantity and less of nutritive food consumption. That may be a reason of obesity and an imbalance dietary portion size or some may not eat that much in quantity which then leads to malnutrition patterns of the body that have conditions leads to body example: - Fatigue, numbness in the body and low blood pressure. Research has explored how stress impacts eating behaviours in various age groups, including children and adolescents. (Hill et al 2018) (1). Different people are affected by exam stress in different ways. While some people experience intense tension, worry, and uneasiness, others stay calm. Academic pressure is associated with stress, which can cause substantial physical repercussions such as weariness, poor appetite, sleeplessness, and insomnia. Exam pressure and stress are closely related. Depending on the individual, stress can either increase or decrease appetite. It disturbs regular eating patterns and contributes to a number of health problems by having direct biological consequences and indirect behavioural changes that have a detrimental



**Vishwa Pandya and Gayatri Parmar**

influence on health. Some physiological factor affects such as fatigue, high hunger, less sleep, sugar craving, food deprivation, illness, drug withdrawal state, anxiety, and also leads to changes in eating behaviours. Exam pressure can significantly impact eating behaviour, which plays a crucial role in maintaining a balanced diet. The correlation between exam stress and eating behavior is essential, especially considering the rising rates of obesity and malnutrition. Understanding this relationship can lead to the development of effective stress reduction methods and awareness of the consequences of stress on dietary patterns. By studying these aspects, we can better comprehend how stress affects our bodies and find ways to modify our behaviors to mitigate its negative effects.

Now a day's there is a huge impact of stress and eating pattern that get visualised into day-to-day life. Not only mental but also physical health burden increase in the communities. This can be led to health issue like anxiety, depression, cardiovascular problem and nutrition deficiencies.(10). As stress has an impact on the body because of the social and relationship problem with friends, family, office colleagues and social media gathering that turn out to be a reason of misleading issue, fights, etc. The influence of social media and unrealistic beauty standards has indeed heightened concerns about body image and health among young people. This pressure can contribute to stress and affect their eating patterns, highlighting the importance of promoting healthy body image and stress management strategies.

MATERIALS AND METHODS

Study Design

In October 2023, we carried out a cross-sectional analytical study on Undergraduate and Postgraduate Students of Parul university, Vadodara, India.

Study setting & instrument

The purpose of the study is to demonstrate a relationship between food pattern modifications and exam stress. Utilizing a Google Form, data is gathered using the purposive technique and the snow bowl method. The descriptive study and thorough explanations of the relationship between exam stress and the changes in eating patterns served as the foundation for the survey design. The research was conducted on the localities and hostilities of Parul University students in Vadodara, Gujarat. The first two weeks of October were spent conducting the study. About 304 responses to the survey have been gathered. The minimal age start for students was set at 18 to 25 years old for the purpose of the survey study. Analysis tool used for research paper is SPSS (Statistical package for social science) for data analysis purpose. Two method are used for analysis purpose Chi – square and T- test.

Participants

Inclusion

- 1)The respondent know about the concept of stress is a major factor that may their affect eating pattern.
- 2)The respondent is the student of Parul university and they are living in Vadodara, Gujarat.
- 3)The respondent is ready to give their consent regarding research topic and proper response to the question.

Data analysis

Data entry was done in Microsoft Excel and analysed using IBM SPSS Statistics. We summarized scores as mean with standard deviation (SD) and percentages (categorized). Age and years of experience were correlated with scores using Pearson correlation. wo method are used for analysis purpose **Chi – square and T- test**. The mean scores of the categories, based on exam and stress, were compared using an independent *t*-test (for two groups). $p < 0.05$ was considered statistically significant. Using binary logistic regression, we performed an adjusted analysis.





RESULT

The survey has received about 304 replies. For the purposes of the survey study, the minimum age start for students was set at 18 to 25 years old. 50 percent of the participants are localities and 50 percent are hostilities. All 304 students are from Parul university, Vadodara, india. The statistics showed that, although 148 participants (49%) did not experience any dietary modifications during the exam, 156 participants (51%) did experience changes in their diet. Based on the data, we deduce that 50% of individuals had diet modifications across for many days during the examination days. (table 1). There is a correlation of ongoing exam and diet pattern change ($r=-0.82$), based on ($n=303$) there is a slight loss of appetite as exams are going on. Based on which conclusion is that changes in dietary patterns due to exams pressure. The examination increases the stress which might also be influencing the diet change.

The correlation between change in eating with sleep pattern and examination days ($r=-0.36$), the number of observations of caffeine product ($n=303$) there is a weak negative relation between students who have currently ongoing examinations and their sleeping & eating pattern are likely to change this line with understanding that examination stress increases and eating pattern, sleep duration decrease. Out of all the individuals, 78(25.65%) experienced regular changes in their eating and sleep patterns, 87(29%) experienced stressful dietary changes with sleep pattern, 51(27%) experienced insomnia (a condition characterized by poor sleep), and 88(29%) experienced variations in their eating and sleep patterns. According to the data, 30% participant do feel changes in dietary habit and sleep pattern. (table 2). The correlation of diet changes and feeling nervous due to stress ($r = 2.31$), the number of observations of diet meal ($n=303$). There is a strong positive relationship between prefer diet changes during stress time. So, it clearly says about the increased ratio of diet changes with stress.

The data of correlation was based on eating with sleep pattern changes and examination ($r=-0.36$), the number of observations of caffeine product ($n=303$) there is a weak negative relationship between increased stress of exam days and changes in eating and sleeping behaviour. The correlation of eating with sleep pattern changes and total stress ($r=0.02$), the number of observations of caffeine product ($n=303$) there is a linear relationship. So, it is a clear observation that caffeine product consumption increases with changes in eating and sleep pattern. Out of all the participants in the questionnaire, 49 (16%) slept for at least seven hours, 69 (23%) for a maximum of six hours, 104 (34%) for more than four hours, and 82 (27%) for more than five hours.(table 3). The correlation of total stress and sleep schedule during exam ($r=-1.52$), the number of observations of total stress ($n=303$). There is an extremely strong negative relationship between increased stress level and decreased hours of sleep during exam days. which implies getting less sleep during exam day. Pearson correlation of feeling dizzy and sleepy during exam days ($r=0.85$), the number of observations of feeling dizzy ($n=303$). There is a moderate positive relationship between stress level with hours of sleep during exam days.

CONCLUSION

The correlation between exam days and diet pattern change($r=-0.82$), soft drink and mid-night food craving ($r=-0.38$), dietary habits with sleep pattern ($r=0.36$), current exam and cooking preference by PG students($r=-0.13$), soft drink and feeling nervous ($r=-0.41$), stress and sleep schedule during exam ($r=-1.52$) and exam and caffeine product ($r=-0.36$) based on Total no of population ($n=303$) Stress and alterations in eating and sleeping patterns are strongly correlated negatively. The evidence suggests that dietary modifications may disrupt sleep patterns and that studying for college exams is the root cause of these disruptions. The correlation between diet changes and feeling nervous due to stress($r = 2.31$), sleep duration during exam days and feeling lethargic($r=0.85$), soft drink and eating pattern during exam days ($r=0.23$), eating pattern and Local lite food preference by PG students ($r=0.20$), currently pursuing and preference of food by PG students ($r=0.21$), mess food and exam ($r=0.33$) based on no.of population ($n=303$) there is a strong positive correlation between exam and eating pattern, sleep duration. Exams associated with lethargy, dietary changes, and the preferred cooking methods of PG students are positively correlated.



**Vishwa Pandya and Gayatri Parmar**

The correlation between exam and gender, age, caffeine product and soft drink and ongoing exam ($r=0.10$) based on no. of correlation ($n=303$) there is no specific relation between hot and cold beverages with exam and nor with age and gender. Null hypothesis: - There is no correlation between examination stress and changes in eating behaviour among 18 to 25 y/o students of Parul university. Alternative hypothesis: -There is correlation between examination stress and changes in eating behaviours among 18 to 25 y/o students of Parul university.

According to this finding, the null hypothesis is rejected since statistical evidence supports the alternative hypothesis, which demonstrates a substantial relationship between exam stress and modifications in sleeping and eating habits. In Conclusion, Stress and alterations in eating and sleeping patterns are strongly correlated negatively. The evidence suggests that dietary modifications may disrupt sleep patterns and that studying for college exams is the root cause of these disruptions. Positive correlations have been shown between exams linked to sluggishness, dietary modifications, and PG students' favourite cooking techniques. There is no particular correlation between exam scores and gender or age.

This result means that the alternative hypothesis—which shows a strong correlation between exam stress and changes in sleeping and eating patterns—is statistically supported, rejecting the null hypothesis.

ACKNOWLEDGEMENT

The authors express gratitude to Department of Nutrition and Dietetics of Parul University, Vadodara for their support to complete the work.

Conflict of interest

There is no conflict of interest between author regarding the paper publishing.

REFERENCES

1. Hill, L. K., Hoggard, L. S., Richmond, A. S., & Curley, J. P. (2018). The impact of stress on eating outcomes among children and adolescents: A systematic review of longitudinal studies. *Obesity Reviews*, 19(3), 352-363. REVIEW OF LITERATURE
2. Kocalevent, R. D., Hinz, A., Braehler, E., & Klapp, B. F. (2011). Determinants of fatigue and stress. *BMC research notes*, 4, 238.
3. Yau YH, Potenza MN. Stress and eating behaviours. *Minerva Endocrinol.* 2013 Sep;38(3):255-67. PMID: 24126546; PMCID: PMC4214609.
4. Ngan, S., Chern, B., Rajarathnam, D., Balan, J., Hong, T. and Tiang, K. (2017). The Relationship between Eating Disorders and Stress among Medical Undergraduates. A Cross-Sectional Study. *Open Journal of Epidemiology*, 7, 85-95.
5. Verzijl CL, Ahlich E, Schlauch RC, Rancourt D. The role of craving in emotional and uncontrolled eating. *Appetite.* 2018 Apr 1; 123:146-151. doi: 10.1016/j.appet.2017.12.014. Epub 2017 Dec 15. PMID: 29253669; PMCID: PMC5817024.
7. van Strien, T. Causes of Emotional Eating and Matched Treatment of Obesity. *Curr Diab Rep* 18, 35 (2018).
8. Michaela C. Pascoe, Sarah E. Hetrick & Alexandra G. Parker (2020) The impact of stress on students in secondary school and higher education, *International Journal of Adolescence and Youth*, 25:1, 104-112,
9. Bremner JD, Moazzami K, Wittbrodt MT, Nye JA, Lima BB, Gillespie CF, Rapaport MH, Pearce BD, Shah AJ, Vaccarino V. Diet, Stress and Mental Health. *Nutrients.* 2020 Aug 13;12(8):2428.
10. Caso D, Miriam C, Rosa F, Mark C. Unhealthy eating and academic stress: The moderating effect of eating style and BMI. *Health Psychol Open.* 2020 Nov 30;7(2):2055102920975274.
11. Villano, I.; Ilardi, C.R.; Arena, S.; Scuto, C.; Gleijeses, M.G.; Messina, G.; Messina, A.; Monda, V.; Monda, M.; Iavarone, A.; et al. Obese Subjects without Eating Disorders Experience Binge Episodes Also Independently of Emotional Eating and Personality Traits among University Students of Southern Italy. *Brain Sci.* 2021, 11, 1145.




Vishwa Pandya and Gayatri Parmar

12. Zeiler M, Waldherr K, Philipp J, Nitsch M, Dür W, Karwautz A, Wagner G. Prevalence of Eating Disorder Risk and Associations with Health-related Quality of Life: Results from a Large School-based Population Screening. *Eur Eat Discord Rev.* 2016 Jan;24(1):9-18.
13. Sander J, Moessner M, Bauer S. Depression, Anxiety and Eating Disorder-Related Impairment: Moderators in Female Adolescents and Young Adults. *Int J Environ Res Public Health.* 2021 Mar 9;18(5):2779.
14. Gemesi K, Holzmann SL, Kaiser B, Wintergerst M, Lurz M, Groh G, Böhm M, Krcmar H, Gedrich K, Hauner H, Holzapfel C. Stress eating: an online survey of eating behaviours, comfort foods, and healthy food substitutes in German adults. *BMC Public Health.* 2022 Feb 24;22(1):391.
15. Deborah Hill, Mark Conner, Faye Clancy, Rachael Moss, Sarah Wilding, Matt Bristow & Daryl B. O'Connor (2022) Stress and eating behaviours in healthy adults: a systematic review and meta-analysis, *Health Psychology Review*, 16:2,

Table 1 Diet changes during exam day

Change in diet	Response	Percentage
Yes	156	51.31%
No	147	48.68%
Total	303	100%
Examination		
Yes	100	32.89%
No	184	60.5%
May be	19	7%
Total	303	100%

Table 2. Responding about their changes in eating and sleep pattern

Eat and sleep pattern	Respondent	Percentage
Regularly	78	25.65%
Stressful	87	28.61%
Insomnia (related to poor sleep)	51	16.77%
Most of the time	87	28.94%
Total	303	100%

Table 3 Sleep cycle during exam days

Sleep during exam days	Respondent	Percentage
At least 7 hours	49	16.11%
6 hours	69	22.69%
4 hours to 5 hours	104	34.21%
less than 5 hours	81	26.97%
Total	303	100%





Vishwa Pandya and Gayatri Parmar

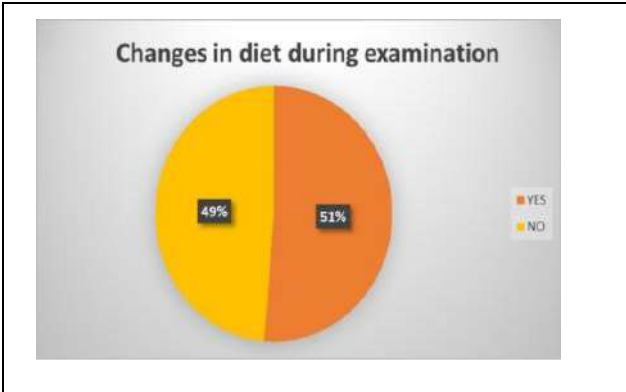


Figure 1 Diet changes during exam day

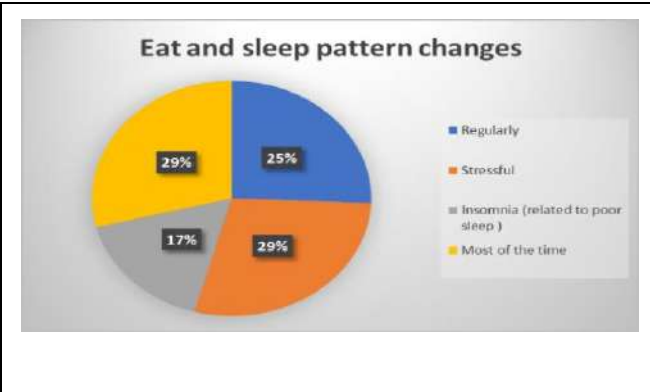


Figure 2 Responding about their changes in eating and sleep pattern

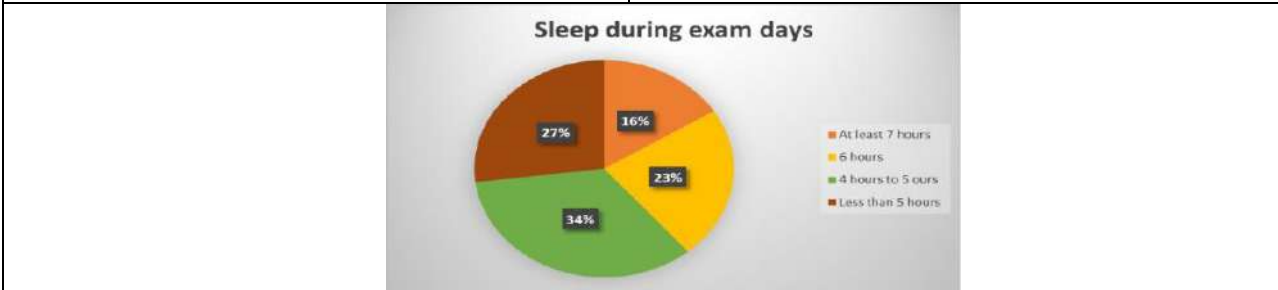


Figure 3 Sleep cycle during exam days





Using Fuzzy Mathematical Modelling to Study Student's Mental Health During Exam Preparations

T. Rajesh Kumar^{1*} and B. Mohamed Harif²

¹Assistant Professor, PG and Research Department of Mathematics, Annai Vailankanni Arts and Science College, Thanjavur, (Affiliated to Bharathidasan University, Tiruchirappalli), Tamil Nadu, India.

²Assistant Professor, PG and Research Department of Mathematics, Rajah Serfoji Government College (Autonomous), Thanjavur, (Affiliated to Bharathidasan University, Tiruchirappalli), Tamil Nadu, India.

Received: 26 Jun 2024

Revised: 12 Sep 2024

Accepted: 14 Nov 2024

*Address for Correspondence

T. Rajesh Kumar

Assistant Professor,

PG and Research Department of Mathematics,

Annai Vailankanni Arts and Science College, Thanjavur,

(Affiliated to Bharathidasan University, Tiruchirappalli),

Tamil Nadu, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Today, people are mostly affected by stress due to work, financial problems, personal relationships, parenting, daily life and busyness, personality, and resources. But we didn't note that students were also affected by stress in their exam preparations. Several studies say that stress leads to anxiety and depression. We all know health is wealth, so health is made up of three major categories: physical health, social health, and mental health. During semester exam preparation, students' anxiety to face exams increases, which leads to stress and depression. In this paper, we study students' mental health during exam preparations using a fuzzy inference system. We split the analysis period into three months: the first 2nd month, the next 4th month, and the 6th month. And also, we suggest some ideas to overcome the stress, anxiety, and depression during exam preparations and achieve good marks.

Keywords: Fuzzy Inference System, Stress, Anxiety, Depression, Health, Trapezoidal Fuzzy Number,

INTRODUCTION

In today's world, mostly people survive among stress, anxiety, and depressed situations, like stress in the workplace, financial crises, problems in personal relationships, difficulties in parenting, daily life and busyness, personality, and resources. Among these problems, we forget to note some issues, namely that students are also facing stress, anxiety, and depression during their exam time, like preparing for the exams, writing the exams, and waiting for the exam results, which lead to stress and depression. If they score good marks, there is no problem; if they score bad or average marks, then it may lead them to stress and depression by thinking about their future. Some students survive in these situations, but some students make wrong decisions like suicide or self-harm.



**Rajesh Kumar and Mohamed Harif**

In this paper, we discuss students' mental health during exam days and their stress, anxiety, and depression levels during exam preparation, writing exams, waiting for the results, and due to score cards. The study was analysed using a fuzzy inference system. The study period was split into three months, i.e., the first 2nd month, the next 4th month, and the 6th month. And also giving ideas to overcome the stress, anxiety, and depression during exam preparations and achieve good marks.

Causes and Effects of Stress, Anxiety, and Depression:

Due to stress, anxiety, and depression, human's mental, physical, and social health are affected. Most commonly, physical health problems are noted easily by symptoms like headaches, high blood pressure, weight gain, weight loss, fatigue, chest pain, muscle tightening, sweating, dizziness, and insomnia. Similar symptoms for mental health problems include being unable to relax, difficulty concentrating, avoiding particular situations and objects, excessive worrying, feeling overly anxious to fit in, troubles in making decisions, feeling very sad or hopeless, thoughts of self-harm or suicide, irritability, changes in eating behaviour, sleep changes, and loss of interest in hobbies and activities.

Diseases led by stress, anxiety, and depression are cardiovascular disease, upper respiratory disease, human immunodeficiency virus, inflammation of the immune system, and inflammation of cytokine production. Treatment and medication for these problems include yoga, exercise, meditation, and simply doing physical activity. Seek help from proper mentors and doctors to recover from the stress and depression. Stress, anxiety, and depression are rated by scales; namely, for the stress scale named the Perceived Stress Scale (PSS), the ratings are 0 – never, 1 – almost never, 2 – sometimes, 3 – fairly often, and 4 – very often. For the anxiety scale named the Hamilton Anxiety Scale (HAM-A), the ratings are 0 – not present, 1 – mild, 2 – moderate, 3 – severe, 4 – very severe. For the depression scale named the Hamilton Depression Rating Scale (HDRS), just as for anxiety, the ratings are: 0 – not present, 1 – mild, 2 – moderate, 3 – severe, 4 – very severe.

Fuzzy Inference System:

Fuzzy inference is the process of formulating the mapping from a given input to an output using fuzzy logic. The mapping then provides a basis from which decisions can be made, or patterns discerned. The process of fuzzy inference involves all the pieces that are described in Membership Functions, Logical Operations, and If-Then Rules [1].

Two months analysis

For the first two months, the range of stress, anxiety, and depression will be [2–34]. For normal, the ranges shown as trapezoidal membership functions are stress [6 8 10 16], anxiety [1 2 4 9], and depression [1 1 2 4]. For mild, the ranges shown as trapezoidal membership functions are stress [9.8 12.9 15 22.3], anxiety [2 2 8 20], and depression [6 8 10 16]. For moderate, the ranges shown as trapezoidal membership functions are stress [5 10 19 42], anxiety [6 8 10 16], and depression [5 8 14 29]. For severe, the ranges shown as trapezoidal membership functions are stress [8 15 34 47], anxiety [9.8 12.9 15 22.3], and depression [9 15 21 39]. For extremely severe, the ranges shown as trapezoidal membership functions are stress [14 20 34 68], anxiety [9 10 20 41], and depression [10 28 32 42].

The above table shows that ranges and ratings of stress, anxiety and depression for two months. The output range and rating for two months combined stress, anxiety, and depression levels shown as trapezoidal membership functions are for normal [2 3.9 5.3 10], mild [5 7 11 21], moderate [6 13.9 14.3 23], severe [15 20.7 22 25.1], and extremely severe [18 27.3 30 33.9].

Four months analysis

For the next fourth month, the range of stress, anxiety, and depression will be [5–45]. For normal, the ranges shown as trapezoidal membership functions are stress [3 7 12 26], anxiety [2 3 5 10], and depression [1 5 6 12]. For mild, the ranges shown as trapezoidal membership functions are stress [7 16.2 16.8 24.8], anxiety [4 5 8.4 16.1], and depression [6 12.1 14 16.3]. For moderate, the ranges shown as trapezoidal membership functions are stress [9 15 21 39], anxiety [3 7 12 26], and depression [10 15.9 18.2 19.5]. For severe, the ranges shown as trapezoidal membership functions are



**Rajesh Kumar and Mohamed Harif**

stress [10 28 32 42], anxiety [9 13 17 29], and depression [9 17 23 43]. For extremely severe, the ranges shown as trapezoidal membership functions are stress [21 33 45 81], anxiety [5 12 36 43], and depression [7 22 30 61]. The above table shows that ranges and ratings of stress, anxiety and depression for fourth month. The output range and rating for fourth month combined stress, anxiety, and depression levels shown as trapezoidal membership functions are for normal [5.6 7.7 8.5 9], mild [6 12.3 14.9 16], moderate [10 16.3 18.9 20], severe [13.2 18.8 22.7 36.1], and extremely severe [20 33 35 44].

Six months analysis

For the next sixth month, the range of stress, anxiety, and depression will be [7-57]. For normal, the ranges shown as trapezoidal membership functions are stress [5 8 14 29], anxiety [2 4 7 14], and depression [3 7 9 17]. For mild, the ranges shown as trapezoidal membership functions are stress [7 18 23 24], anxiety [3 7 9 17], and depression [8 11 13 20]. For moderate, the ranges shown as trapezoidal membership functions are stress [6 19 25 50], anxiety [5 8 14 29], and depression [9 10 20 41]. For severe, the ranges shown as trapezoidal membership functions are stress [12 24 33 63], anxiety [5 10 19 42], and depression [9 12 27 60]. For extremely severe, the ranges shown as trapezoidal membership functions are stress [24 57 64 83], anxiety [8 26 39 83], and depression [18 36 48 90]. The above table shows that ranges and ratings of stress, anxiety and depression for sixth month. The output range and rating for sixth month combined stress, anxiety, and depression levels shown as trapezoidal membership functions are for normal [9 9 10 12], mild [9.2 10 12 22], moderate [10 13.7 14 17.1], severe [12 26.3 26.9 40], and extremely severe [38 48 52 54]. The above graph shows the overall outcome of stress, anxiety, and depression levels throughout the six months. Here we can see for the first 2 months the range between [2–34], next for 4 months the range between [5–45], and after six months the range between [7–57].

CONCLUSION

From the past six-months analysis, split into two months, we conclude that comparing the first two months to the last two months, which are the fifth and sixth months, stress, anxiety, and depression are higher for students. Because the last two months are exam months. This analysis was conducted with the help of a fuzzy inference system using MATLAB software. The analysis is explained with the help of individual plots for stress, anxiety, and depression levels at 2 months, 4 months, and 6 months. The overall range for the fuzzy inference system is taken as [1–57], and the table values for stress, anxiety, and depression are calculated for those particular months.

REFERENCES

1. <https://www.mathworks.com/help/fuzzy/fuzzy-inference-process.html>
2. Neil Schneiderman. et.al., STRESS AND HEALTH: Psychological, Behavioural, and Biological Determinants, *Annu Rev Clin Psychol.* 2005; 1: 607–628. doi:10.1146/annurev.clinpsy.1.102803.144141.
3. Salam Ranabir. et.al., Stress and hormones, *Indian Journal of Endocrinology and Metabolism / Jan-Mar 2011 / Vol 15 | Issue 1*, 18-22.
4. LAURA J. JULIAN, Measures of Anxiety, *Arthritis Care Res (Hoboken)*. 2011 November; 63(0 11): doi:10.1002/acr.20561.
5. Dr. Ghazala Kausar. et.al., Stress in Pakistani Working Women, *Journal of Culture, Society and Development*, Vol.5, 2015, 58-64.
6. Taneal A Wiseman et.al., Incidence of depression, anxiety and stress following traumatic injury: a longitudinal study, *Wiseman et al. Scandinavian Journal of Trauma, Resuscitation and Emergency Medicine (2015) 23:29*, DOI 10.1186/s13049-015-0109-z.
7. Zafar Iqbal. et.al., Depression and Anxiety: A Snapshot of the Situation in Pakistan, *International Journal of Neuroscience and Behavioural Science* 4(2): 32-36, 2016 DOI: 10.13189/ijnbs.2016.040202, 32-36.





Rajesh Kumar and Mohamed Harif

8. Anthonette D. Cantara. et.al., Stress Sensor Prototype: Determining the Stress Level in using a Computer through Validated Self-Made Heart Rate (HR) and Galvanic Skin Response (GSR) Sensors and Fuzzy Logic Algorithm, International Journal of Engineering Research & Technology (IJERT), Vol. 5 Issue 03, March-2016, 28-37.
9. Sri ramprakash.S. et.al., Stress Detection in Working People, 7th International Conference on Advances In Computing & Communications, ICACC 2017, 00 Procedia Computer Science 115 (2017) 359–366.
10. Berbano. et.al. Classification of Stress into Emotional, Mental, Physical and No Stress Using Electroencephalogram Signal Analysis, Proc. of the 2017 IEEE International Conference on Signal and Image Processing Applications (IEEE ICSIPA 2017), Malaysia, 11-14.
11. Ankita Tiwari et.al., Monitoring and Detection of EEG Signals Before and After Yoga during Depression in Human Brain Using MATLAB, Proceedings of the IEEE 2017 International Conference on Computing Methodologies and Communication (ICCMC), 329-334.
12. Ketaki Ravikant Bhokare. et.al., Survey Paper Detecting Stress of users on Social Interactions in Social Networks, International Journal of Innovative Research in Science, Engineering and Technology, Vol. 7, Issue 1, January 2018, DOI:10.15680/IJIRSET.2018.0701057, 336-341.
13. Cay M, Ucar C, Senol D, Cevirgen F, Ozbag D, Altay Z, et al. Effect of increase in cortisol level due to stress in healthy young individuals on dynamic and static balance scores. North Clin Istanbul 2018;5(4):295–301.

Table 1: shows that ranges and ratings of stress, anxiety and depression.

Stress	Anxiety	Depression	Ratings
0-14	0-7	0-9	Normal
15-18	8-9	10-13	Mild
19-25	10-14	14-20	Moderate
26-33	15-19	21-27	Severe
34+	20+	28+	Extremely Severe

Table 2: shows that ranges and ratings of stress, anxiety and depression for two months.

Stress	Anxiety	Depression	Ratings
10	4	2	Normal
15	8	10	Mild
19	10	14	Moderate
26	15	21	Severe
34	20	28	Extremely Severe

Table – 3: shows that ranges and ratings of stress, anxiety and depression for fourth month

Stress	Anxiety	Depression	Ratings
12	5	6	Normal
16.2	8.4	12.1	Mild
21	12	15.9	Moderate
28	17	23	Severe
45	24	30	Extremely Severe

Table – 4: shows that ranges and ratings of stress, anxiety and depression for sixth month.

Stress	Anxiety	Depression	Ratings
14	7	9	Normal
18	9	13	Mild
25	14	20	Moderate
33	19	27	Severe
57	39	48	Extremely Severe





Rajesh Kumar and Mohamed Harif

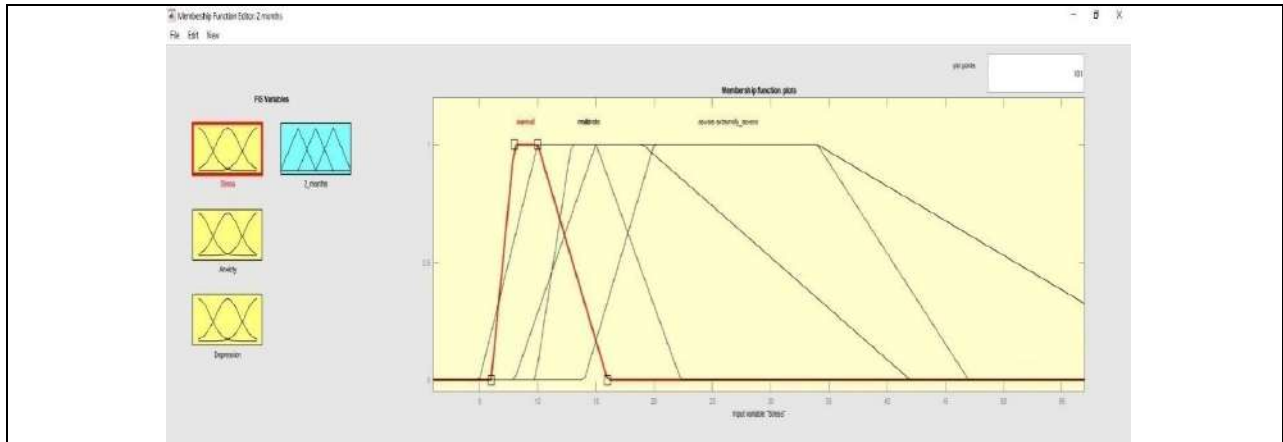


Figure – 1 Plots representing stress level for two months

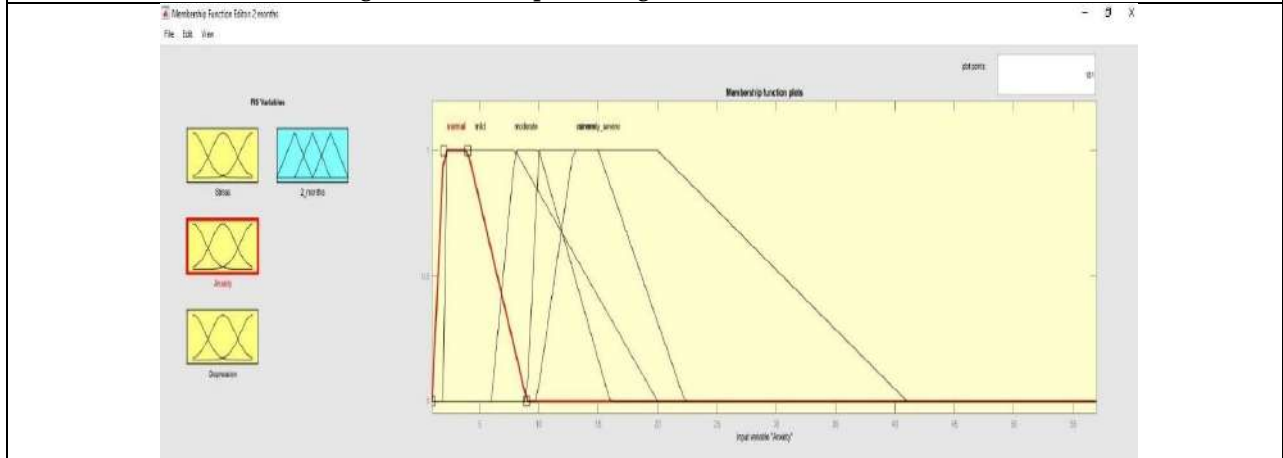


Figure – 2 Plots representing of anxiety level for two months

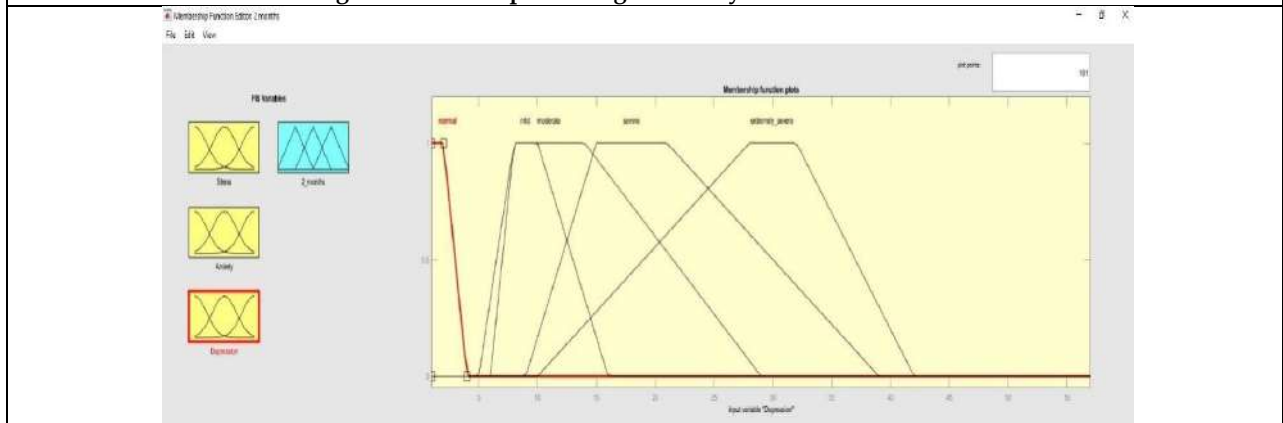


Figure – 3 Plots representing depression level for two months





Rajesh Kumar and Mohamed Harif

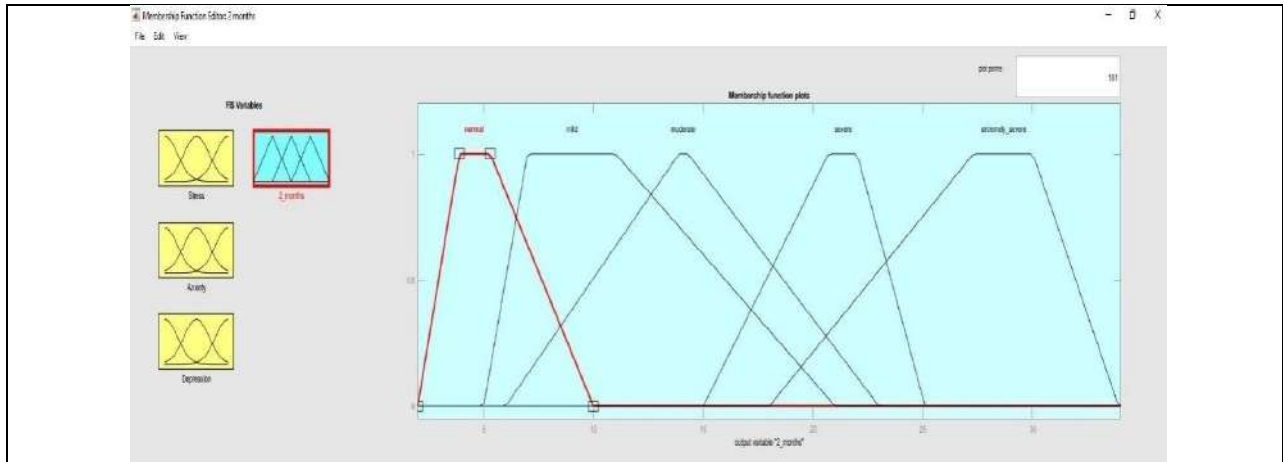


Figure – 4 Plots representing output for two months

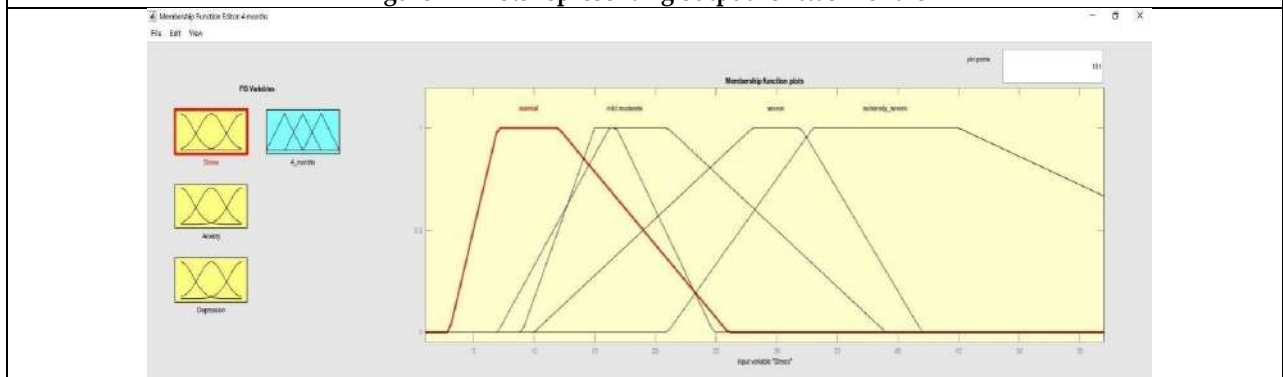


Figure – 5 Plots representing stress level for fourth month

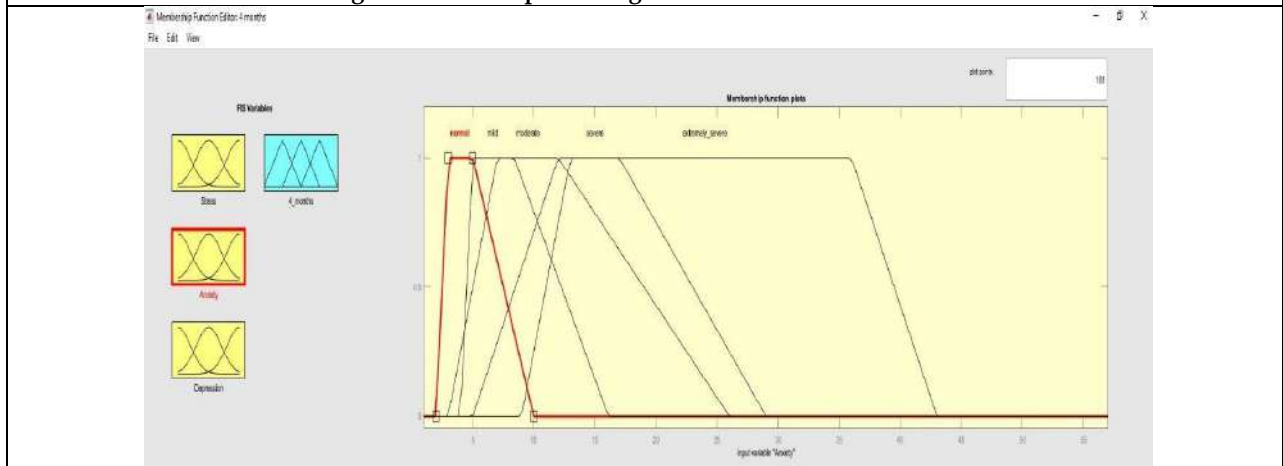


Figure – 6 Plots representing anxiety level for fourth month





Rajesh Kumar and Mohamed Harif

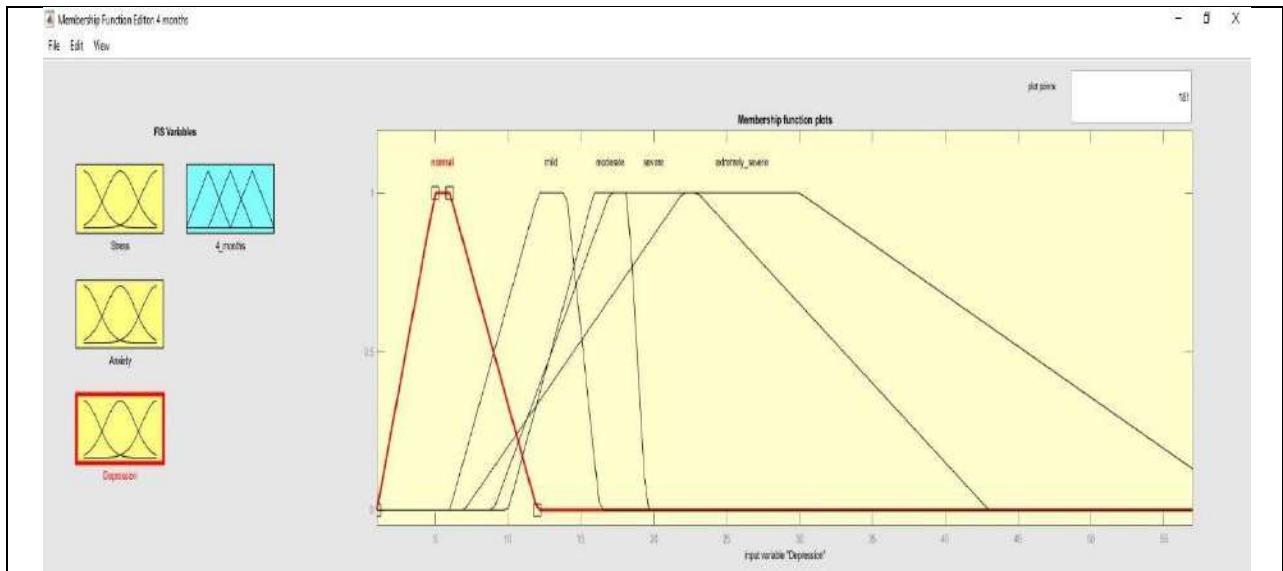


Figure – 7 Plots representing depression level for fourth month

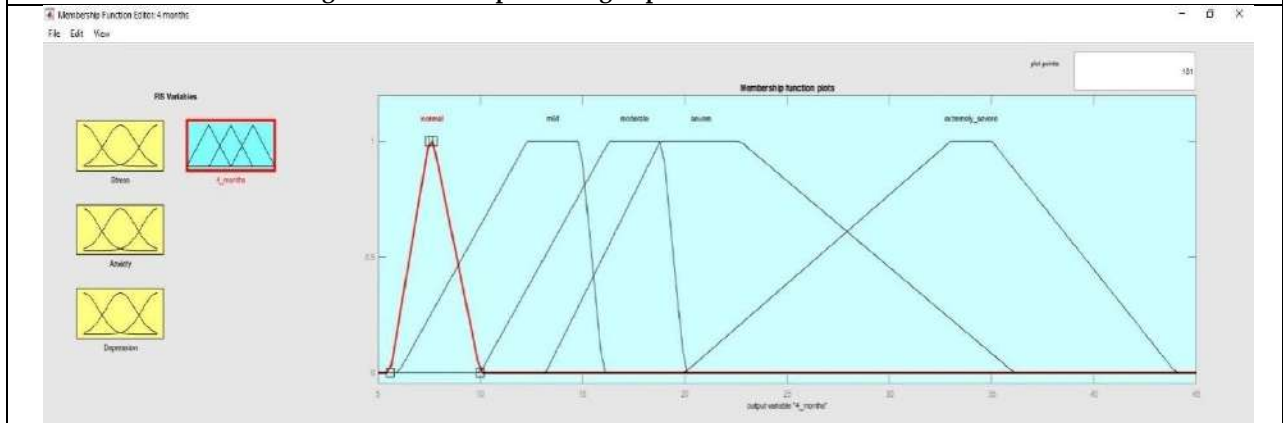


Figure – 8 Plots representing output for fourth month

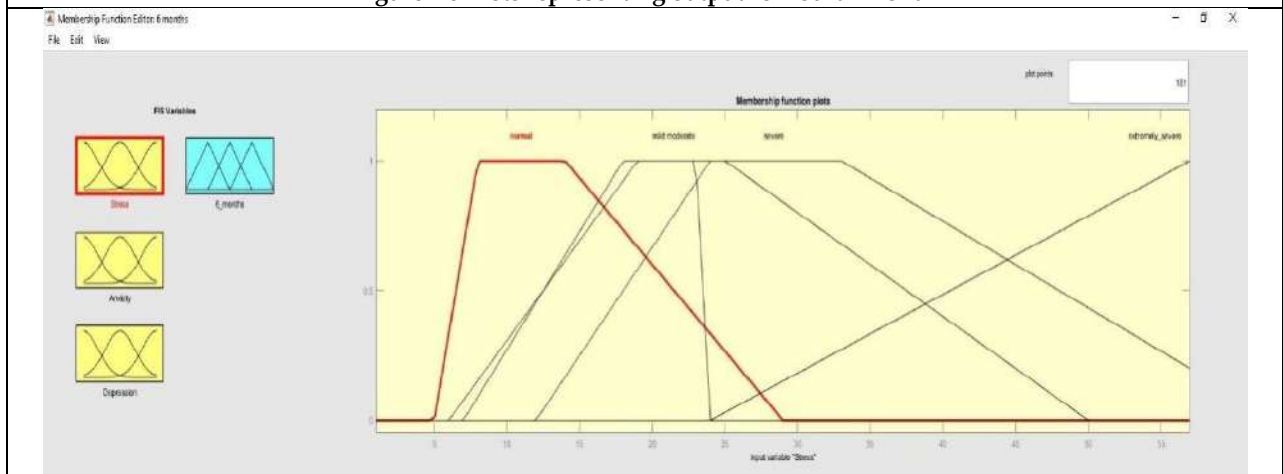


Figure – 9 Plots representing stress level for sixth month





Rajesh Kumar and Mohamed Harif

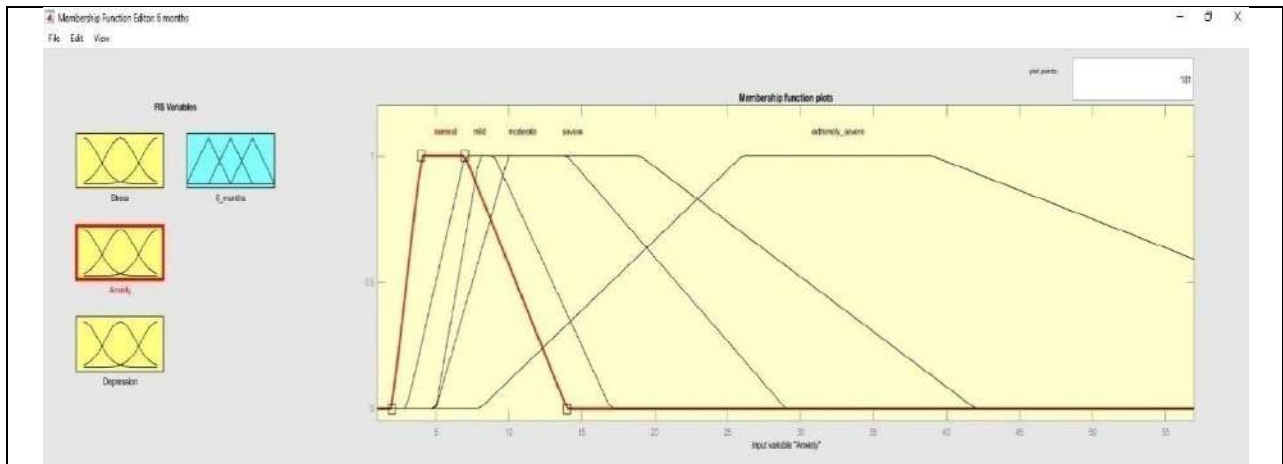


Figure – 10 Plots representing anxiety level for sixth month

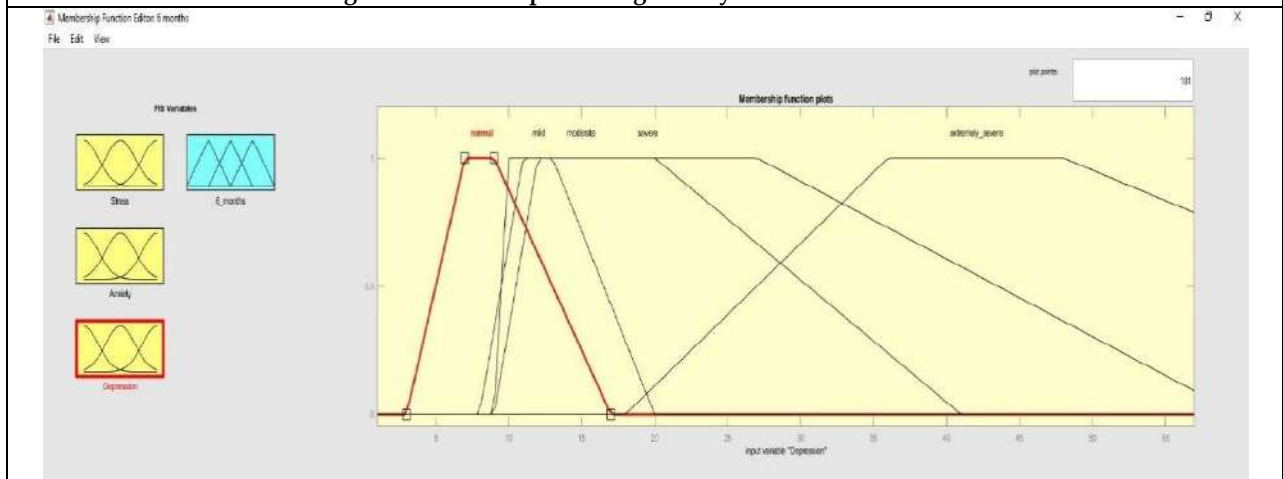


Figure – 11 Plots representing depression level for sixth month

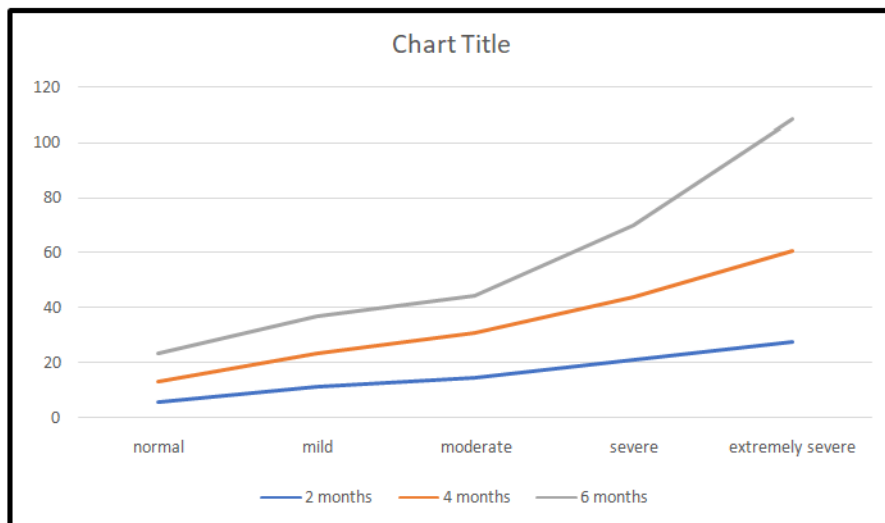


Figure – 12 Graph representing overall outcome of stress, anxiety and depression level





Industrial Plant Automation Management Best Practices for Great Outcomes

Kashyap L Mokariya^{1*}, Patel Snehal S², Ganesh R.Patil³ and Nikunj R Patel⁴.

¹Head, Department of Electrical Engineering, Government Engineering College Valsad, Tithal Road Valsad, (Affiliated to Gujarat Technological University), Gujarat, India

²Assistant Professor, Department of Electrical Engineering, Chhotubhai Gopalbhai Patel Institute of Technology, Maliba Campus, Bardoli, (Affiliated to Uka Tarsadia University), Gujarat, India.

³Post Graduate Engineer Trainee, Department of Electrical Engineering, The Maharaja Sayajirao University of Baroda, Gujarat, India.

⁴Assistant Professor, Department of Electrical Engineering, Ganpat University, Mehsana, Gujarat, India

Received: 25 Jun 2024

Revised: 28 Sep 2024

Accepted: 04 Nov 2024

*Address for Correspondence

Kashyap L Mokariya

Head, Department of Electrical Engineering,
Government Engineering College Valsad,
Tithal Road Valsad,
(Affiliated to Gujarat Technological University),
Gujarat, India.
E.Mail: klmokariya@gecv.ac.in



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

In the current era of smart homes and smart grids, advanced technological solutions that allow automation of household functions are rapidly evolving. Today's smart homes have a wide variety of technologies and applications. These are key in improving automation, monitoring and remote control capabilities by initiating communication between home appliances and their customers. In this article, examples of automation that focuses on energy efficiency, environment, automation are explained. Technical descriptions of the system are provided along with the advantages and disadvantages of each technology and product currently on the market. The role of consumers is also highlighted along with the barriers, obstacles, rewards and future developments in technology. The author has tried his best to justify Management best practices for great outcomes.

Keywords—Smart Grid, Home Automation, Energy Management System, Plant Automation, Programmable logic Controller (PLC), Supervisory Control and Data Acquisition (SCADA) .

INTRODUCTION

Automation is a crucial idea for the industry 4.0 and is becoming more and more valuable to industrial businesses [1]. The global industrial automation market was valued at 157.04 billion dollars in 2018 and is projected to grow to





Kashyap L Mokariya et al.,

296.70 billion dollars in 2026, nearly twice as much as it was a year earlier, according to the publication Fortune Business Insights. Organizations are drawn to automation-facilitating technologies like blockchain, artificial intelligence (AI), and the Internet of Things (IoT) because of their advantages [1]. It offers exceptional benefits in numerous areas, but most notably in terms of time and money savings. Businesses today must use industrial automation systems to complete industrial automation processes in order to benefit from these advantages. Industrial automation systems are those that use computers to control and monitor machines, processes, or other devices. Usually, these systems are employed to perform recurring duties or operations. Their aim is to operate independently, reducing and increasing the quantity of human labour needed in the industry. These systems take the place of the monotonous, mechanical jobs that are typically completed by a single person, as well as the choices that person makes during the manufacturing process. Strong machinery and logical programming commands are used to accomplish this.

Various forms of automation are available to businesses that decide to begin an industrial automation process, based on factors such as production volume, product range, and varying degrees of adoption. The phrase "home automation" or "fixed automation" refers to an automation procedure in which a machine or robot is designed to carry out a single, highly repetitive task for a high output volume [2]. These processes are fast, accurate, safe and have high production rates, however they are fixed and cannot be altered. The advantage of this kind of automation management offers the sectors are best suited for operations with a high volume of load and little work variability. The most cost-effective kind of automation is this one.

Soft automation refers to equipment that can adapt to a wide variety of product configurations. The process is managed by a computer that can be configured to change how it operates depending on the product. When a sector produces a variety of goods that call for distinct and varied configurations, it is quite helpful. Compared to hard automation, this kind of automation is more expensive and runs more slowly. It is especially appropriate for establishments that produce a small number of comparable goods in small quantities using batches. Programmable automation and flexible automation are the two categories into which this type of automation falls. Programmable automation category includes automated industrial processes that are managed by a program that is made up of a series of coded instructions that the system must decipher. Because of the system's great flexibility, new products can be made using these processes and new software. The equipment can be configured and adjusted in terms of both software and hardware. It is recommended to employ this kind of automation exclusively in low-production industries because it is difficult and time-consuming to reprogramme a machine to accommodate new products. As such, the processes are finished in batches.

A machine can be quickly configured to fit a new product thanks to flexible automation. This minimizes the amount of time lost when switching from one item to another in terms of configuration. Because this type of automation removes the need to waste time reprogramming the system or altering the physical configuration of the machines, there is no need to produce the products in separate batches. This type of automation is recommended for medium-sized productions. Flexible automation is used to enable continuous production that can be adjusted to the different product types that an industrial plant may encounter. Although automation removes the need for manual labour in factories, operators are still needed to keep an eye on equipment and perform maintenance. On the other hand, human intervention in the process is no longer necessary when using an industrial automation system. One of the biggest issues facing one is boosting energy security and accessibility in light of diminishing energy resources, particularly in emerging economies. An essential agenda item is producing electricity effectively and efficiently, in addition to managing the energy resources that are currently available. The construction of large power plants using conventional energy sources must be complemented, particularly with small-scale power generation distributed from renewable energy sources. Distributed power sources (DERs) can be connected to the grid with additional infrastructure and financial outlays, but these technologies minimize transmission and distribution (T&D) losses and do away with the need for a costly transmission system. Adopting a methodical strategy that views output and related load as a subsystem or "micro-grid" is a smart way to realize the growing potential of distributed generation[3].





Kashyap L Mokariya et al.,

One common type of small integrated circuit is a microcontroller. A microcontroller is a small integrated circuit device that is usually used to control a single function inside another device, like a television, automobile, or home appliance. It's also typical practice to use several microcontrollers for different tasks. A car, for instance, would have different microcontrollers for the powered windows, the auto-braking system, and the speedometer. In contrast, PLCs are made up of an external programming device, a power supply, an I/O module, and a processor. The PLC's brain, or processor, uses data from connected input devices, like switches, sensors, and thermometers, to carry out preprogrammed control operations. For instance, After analysing thermometer-transmitted low-temperature data, the PLC's processor will decide on the best course of action and send a command to activate a heating element. Field devices are physically connected to the PLC through its I/O modules, which are also in charge of sending commands to output devices and bringing in data for the processor. It is possible to combine or separate analog or digital I/O modules based on the requirements of the application.

The PLC program is written and downloaded to the controller using the external programming device, which is usually a desktop or laptop computer. Ladder logic, function block diagrams, structured text, instruction lists, and sequential flow charts are a few of the numerous PLC programming methods that are available [4]. A microcontroller is a small integrated circuit device that is typically used to control a single function in another device, such as a television, car, or household appliance. Using several microcontrollers to carry out distinct tasks is also typical. A microcontroller is used in the car, for instance, to control the speedometer, the auto-braking system, and the powered windows. comprises an external programming device, a power supply, an I/O module, and a PLC processor.

The PLC's brain, or processor, uses data from connected input devices like switches, sensors, and thermometers to carry out preprogrammed control operations. For instance, when the PLC receives low-temperature data from the thermistor, its processor evaluates the information, chooses the proper action, and then initiates the command to activate the heating element. The I/O modules of the PLC are physically attached to the field devices, and they transmit commands to the output devices and receive data as input for the processor. Analog or digital I/O modules can be mixed and matched to suit a given application. Writing programs for desktop or laptop computers and PLCs, as well as downloading them to the controller, is typically done via an external programming device. Among the PLC programming techniques are ladder logic, function block diagrams, structured text, instruction lists, and sequential flow charts [4], [5].

Numerous new opportunities for efficiency gains have arisen as a result of the smart grid initiative, which aims to modernize the infrastructure of electric utilities. An effective method for working remotely and keeping an eye on renewable energy sources is to use a supervisory control and data acquisition (SCADA) system. SCADA systems are extensively employed in numerous industrial applications, and they have contributed to these systems' increased efficiency. In today's modern communication age, the smart grid initiative, which aims to integrate both function and architecture, has made tremendous strides in modernizing and expanding electric utility infrastructure. With this progress come organizational, socioeconomic, technological and cyber security challenges. The scope and magnitude of those challenges have increased significantly and many regulatory bodies have taken steps to align their standards and regulations with these new challenges.

K. Sayed and H.A. Gabbar received an overview of the process for gathering and using data in SCADA systems, development, and power systems. Along with the standard remote terminal units (RTUs), it also includes master stations, data concentrators, outstation hardware, and the newest intelligent electronic devices (IEDs). The fundamentals of Power System Supervisory Control and Data Acquisition (SCADA) and potential application functions increased the viability of smart grids and encouraged more individuals to participate in their development [6]. Research on various automation methodologies, including PLC, SCADA, and DCS (distributed control systems), and their benefits and drawbacks, has been conducted by Hudedmani*, Hittalamani Raghavendra, Umayal R M, and Shiva Kumar Kabberalli. The mention table below provides a summary of the findings[7].





Kashyap L Mokariya et al.,

ER Alphonsus, MO Abdullah PLC provided an overview of its various applications such as hardware, programs and water and wastewater management control, solar-tracking systems, wind power, photovoltaic applications. Control of heating, ventilation, and air conditioning (HVAC), research, instruction, and training, as well as plant control and monitoring and other uses [8]. An air control system powers the packaging system's assembly and global operation. Pneumatic was only ever used manually in the past; nowadays, PLC is used to automate the process [9].

Factories in Industry 4.0 use logic controllers to monitor system status, decide on actions based on customized programs, and take necessary action. Controllers use high-speed computations outlined in programs (control logic) to enforce control in real time, and they output the results as output signals to actuators and other devices. Figure 1 depicts the specifics of the Smart Control Box (SCB) architecture. A single board PC and an MCU (Micro Control Unit) make up a SCB. The MCU receives the input signals from the PLC. For edge computing and data storage, signals are sent to a single board PC via wired or wireless communication. The MCU responds to the PLC's decision command following the margin analysis. In order to close the gap between Industry 3.0 and Industry 4.0, this research aims to introduce a SCB. Installing intelligence on a single board computer and connecting the Internet of Things to the PLC with the MCU [10].

The PLC community has responded to evolving needs by developing new programming languages, implementing special priority scan cycles, and developing specialized I/O hardware (hidden networking) through PLC programmers. Memory and processing speed have also improved dramatically over time. Interactions between supervisory layer systems like SCADA, distribution control systems, or human-machine interfaces and field devices like sensors and actuators. Enterprise resource planning and implementation systems process the data that is received in both the upper and lower layers [11]. As depicted in Figure 2, PLCs are centralized controllers that interact with sensors and actuators as a component of a conventional automation pyramid. All connected devices are perceived as I/O devices, with no awareness of any other behaviour besides the inherent values and complexity of each device. Shared and taken in. Decentralized control and greater use of intelligence are predicted by trends like Industry 4.0 [11].

Effective data transmission in communication networks is necessary for the automation, monitoring, and tracking of technical activities in both industrial and non-industrial settings. This information is related to duties like receiving, logging, measuring, and displaying data. The transferred data also includes control codes. Digital communications are used by control units, computers, sensors, actuators, human-machine interfaces (HMI), and networks with various topologies to transfer operational data and command signals [12].

Plant Automation Case Study

Power plants and other businesses that use boiler equipment need to constantly monitor and inspect it on a regular basis. In essence, the boiling section of the boiler which generates hot water for the production of steam must be run concurrently with other parts of the boiler, including the core section, which includes the boiler drum. Not enough steam is produced when the water level is too high. In order to automate the system and reduce human error, it is crucial to measure and maintain the levels of steam and drum. It is essential to keep the water level in the drum precisely under control. The reduction of human error rates and the enhancement of control and supervision over plant or process operations can be accomplished through the development of PLC and SCADA systems, which can be employed for the entire plant and running. possesses the ability to be applied to process monitoring and provide the required data in response to process changes.

PLC Temperature Control Programming

Temperature in each heater is measured by three thermostats. Different set points with different ranges is with all three heaters. The table shows the temperature ranges.

1. all three heaters (H1, H2, H3) must be on when a temperature goes below 55° C,
2. The optimal operating temperature range for both heaters (H2, H3) is between 55° and 60° C.



**Kashyap L Mokariya et al.,**

3. The operating temperature range for the heater (H3) is 60° to 65° C.
4. Every heater that is over 70°C needs to be in the OFF position and have a safety shutoff (relay CR1) in case something goes wrong.
5. enables and disables the master switch system.

Solution :

- There are four thermostats; if the set point is not reached, assume they are in the NC (usually closed) position.
- Allow the control relay (CR1) to function as a shut-off for safety.
- Master switch: Turn on switch NO, which is normally open, and turn off switch NC.

The temperature ranges indicated by the thermostats' temperature values (TS1, TS2, TS3, TS4) are displayed in Table 2. In addition, the heaters (H1, H2, and H3) are either in the on or off position based on the temperature reading.

First step

Its two buttons are for start (no default contact) and stop (default NC contact). Depending on the state of the thermostats, Relay CR1 regulates the heaters. All heaters are turned off if the TS4 is energized (TS4 contact changes from NC to NO). The thermostat is connected between the TS4 STOP and the relay. There is no way to lock or hold the START command when Relay CR1 is introduced to the START button.

Second step

Heaters (H1, H2, and H3) can be controlled by relay CR1 in the absence of contact thermostats (TS1, TS2, TS3). After the START command is issued, this NO contact closes (becomes an NC contact). All heaters are on when the temperature drops below 55°C because TS1, TS2, and TS3 are closed. Heater H1 is turned off when the temperature is between 55° and 60° C because TS1 is open. TS2 will then open as well, turning off the heater H2 if the temperature is between 60° and 65° C. When the temperature is between 65° and 70° C, TS3 also opens, which turns off the heater H3. To avoid overheating or any thermostat malfunction, there is a safety shutoff available. All heaters will be turned off by the TS4 powering the relay and reducing power if the temperature rises above 70°C.

Note: In this case, the contactors or relays that we are turning on are the heaters H1, H2, and H3. As a result, the electric heater feeder circuit (MCC) is not connected to the contact of these relays. These signals regulate these electrical feeder circuits, and the heaters are turned on or off in accordance.

Boiler Control

The heating elements are gradually heated and gradually turned off when the boiler is turned off to prevent excessive electricity consumption at the boiler's startup. Figure 5 depicts this working principle. The first heating element (S1) activation is powered by the ON (MA) button. The activation of the second element (S2) occurs following the time delay T. The time-delay T is again stopped by the third element (S3) and the fourth element (S4) following the same time delay. S1's Off (AR) button. After a little delay, the remaining three components progressively become inactive.

Green House Ventilation Panes

It is far more difficult to ventilate and cool greenhouses than it is to heat them. The temperature can always be adjusted by adding more heat, but in severe weather, it is challenging to lower the temperature. Greenhouse ventilation, which provides CO₂ and controls temperature and humidity, is necessary for crop production. The two most popular ventilation methods utilized in greenhouse production systems are mechanical and natural ventilation systems. The normal airflow produced by air pressure or the gradient created by temperature differences between the rising area and the rest of the greenhouse are what allow for natural ventilation.





Kashyap L Mokariya *et al.*,

The process of moving air through fans, known as mechanical ventilation, involves bringing air into the growing area through carefully regulated greenhouse entrances and releasing it through the fan assembly. Good mechanical ventilation systems must be flexible enough to accommodate changing inlet shapes. Thermostats and, in certain situations, moisture-sensing devices regulate fan ventilation, whereas relative humidity is the primary factor in the control of disease [14]. The most basic and conventional environmental control system for tropical greenhouses uses sensors to monitor all significant environmental factors that affect crop growth, both inside and outside the greenhouse. The controller then receives the sensor data. The environmental parameters are managed by preprogrammed commands and related actuators, including fans, coolers, fogging systems, lighting systems, and CO₂ sprayers [15].

Mechanical Ventilation

An intelligently designed inlet allows fan ventilation systems to efficiently control temperature all year round. The most desirable feature is the ability to easily automate the entire system. With the introduction of computer-based control, this is becoming more and more the case. This feature will be especially helpful to farmers who are not present in the greenhouse during the day or who have other obligations and cannot find labor on weekends. The higher initial and continuing costs of mechanical ventilation systems are one of their drawbacks. This illustration shows how automatic control of greenhouse ventilation panes is possible.

Fan ventilation systems with well-thought-out inlets offer effective temperature control in all settings. The capability to seamlessly automate the entire system is its most alluring feature. This is now a reality thanks to the development of computer-based control systems. Farmers who have other responsibilities, lack workers on the weekends, or are away from the greenhouse during the day will find this function especially helpful. The disadvantages of mechanical ventilation systems are their high installation and maintenance costs. This illustration shows how automatic control of greenhouse ventilation panes is possible.

The greenhouse's owner wishes to have ventilation window panes on the roof of the greenhouse installed so that they can be opened and closed. There are two windows in the greenhouse for ventilation. Two sensors that detect whether the motor and the window panes are open or closed regulate how far these window panes open. The window panes open during the day to ventilate the building, ideally during the peak temperature of 12:00 to 15:00. However, window panes that are already open will not close or open if the temperature drops below 10 C. Additionally, on days when the temperature rises to 25 C, the windowpanes open. The window panes should be closed once more if the temperature drops below 25 degrees Celsius. No matter the temperature, window panes are closed at night. When the window panes are open, a red pilot lamp signal is displayed; when they are closed, a green pilot lamp signal is displayed.

Program description, 3-time ranges are used:

- Range 1: Night, from 21:00 to 07:00
- Range 2: Day, from 07:00 to 12:00 and from 15:00 to 21:00
[T > 25° C – Opening, T < 25°C – Closing]
- Range 3: Noon, from 12:00 to 15:00
[T > 10° C – Opening, T < 10°C – Closing]

Benefits of an Industrial Automation Management system

Automation of factories and industrial processes is made possible by industrial automation management systems, which enable continuous mass production seven days a week, 24 hours a day, increasing productivity and cutting down on assembly times. These systems help reduce human error and enhance the quality and uniformity of the products by employing adaptive control and monitoring throughout various industrial processes and stages. After working nonstop for several hours, the performance does not decline. Computers and other machines operate continuously and at a steady pace. As a result, when managed by an automation system, automated production processes have a longer lifespan, stability, and solidity. In a traditional production chain, implementing a new task



**Kashyap L Mokariya et al.,**

requires user training that can take hours or even days. However, reprogramming a robot or machine through an automated system is a quick and easy process that offers increased production process flexibility. Automation of data collection lowers expenses while increasing accuracy. Managers of the company are able to make better decisions because of the increased accuracy.

On manufacturing lines where human workers are exposed to hazardous conditions, using robots is a safer option. The Occupational Safety and Health Act was passed in the US in 1970 with the intention of enhancing worker protection and job safety. Since it was passed, it has encouraged the use of automation systems and robotics in the nation's factories. Industrial automation systems may require a large initial investment, but using this technology will result in lower costs for data analytics. Additionally, there is very little chance of a machine failure or service interruptions because of this automated data analysis. When an industrial automation management system is in place, workers in that factory put in less hours and spend more of it on high-value tasks. Employees are relieved of repetitive and laborious tasks by automation systems. Employees can perform more value-added work in other areas of the business that benefit the company more when machines and computers take over these tasks from them. In addition to carrying out tasks that a human would do, the systems that businesses use to automate their services are also capable of carrying out tasks that a human being could not accomplish. Among other things, they are superior in terms of size, weight, speed, and resistance. Some of the important strategies for best Industrial management practises is explained below.

Forming the right team for Industrial Management

Finding the right talent is one of the most important aspects of building a successful project team. Experts concur that a project management team does not need to be large, even though automation projects can be complicated. Veterans of the industry also contrast internalizing the automation project with working with a qualified System Integrator, citing the latter's high project success rate. Skilled system integrators "offer the value-added engineering services that concentrate on the unique requirements of your company." Put differently, they possess the necessary skills and resources to assist in creating a workable solution without having to start from scratch.

Look for Technical Qualification in team member

Inadequately qualified team members can have disastrous consequences for an automation project. Errors in project scoping, estimation, or assessment can become difficult and costly as the automation project moves forward. Businesses must ensure that all project participants have the necessary skills in PLC, HMI, hardware integration, software programming, network protocol, UI/UX design, and other related areas when developing solutions internally. Additionally, someone with the sole responsibility of choosing and evaluating vendors and suppliers is required. Instead, a lot of manufacturers choose to collaborate with System Integrators who already employ a group of skilled engineers. When a project team chooses to collaborate with outside parties, such as a system integrator, the members must be selected based on their ability to manage projects, think critically and analytically, and have experience with various production procedures.

Appoint some one who understands Industry as a Project Manager

Assign the project manager position to a knowledgeable industry professional. The project manager bears the responsibility of guiding the automation project towards its intended course. They are essential to an automation project because they serve as the point of contact for vendors and stakeholders. A qualified project manager's level of industry knowledge is one of the selection criteria. Gaining knowledge about the industry a company operates in, particularly in heavy industries, can take up to ten years. On the other hand, a well-informed project manager fosters team cohesion by being: Skilled in areas beyond their purview; knowledgeable about the offerings of rival companies; and able to make both internal and external connections.





Kashyap L Mokariya et al.,

Partnership with system integrator wherever you can.

System integrators are expert services that use commercially available hardware components to construct automation control systems and create software algorithms according to application requirements. By offering solutions and ongoing technical support, they assist manufacturers (end users) in automating jobs or processes.

Knowing and applying current industry standards in safety, Environmental and modern technology.

Skilled System Integrators collaborate with a diverse range of end users. They continuously monitor the most recent advancements in technology and industry best practices, including security, network standards, international programming standards, OSHA (Occupational safety and health Administrations) guidelines, machine safety, and process safety. It can be difficult to justify hiring an end-user for an automation project because they might not be skilled in every area.

Choosing the best hardware and software for application

It is the responsibility of a system integrator to be knowledgeable about various hardware components' interoperability. They are also capable of creating software applications and algorithms that interface with these parts. It takes years, if not decades, to become proficient at the complexity of creating an automation system. System engineers working for an organization often lack the time and resources necessary to become as proficient as an integration engineer whose entire career is focused on learning about system integration. Long-term returns on automation projects can be higher for end users who work with the right system integrator.

Providing proper project documentation

Future maintenance and upgrades to an automation system depend heavily on documentation. Usually, a system integrator oversees several installations at once. For future reference, it is in their best interest to maintain thorough documentation of the project. Regretfully, end users find it more difficult to obtain the same level of detail. Choosing the appropriate team members is the first step towards a successful automation project's execution. Members of the internal team should be technically proficient in both software programming and hardware technologies. A Project Manager with extensive industry knowledge and comprehension of various job functions is leading the team to success. Lastly, never undervalue the complexity of an industrial project. Reducing risks and expediting the project schedule can be achieved by collaborating with a qualified system integrator.

Ensuring goals and roles are crystal clear

Failure to communicate the project goals is akin to failing to inform a cab driver of your destination. It is a waste of money and time for both the driver and the passenger. The worst part is that it still falls short of the goal of the journey. Having well-defined objectives helps team members stay focused and minimize distractions, which enhances project success overall. An automation project will have numerous subtasks that must be completed at various stages of the project. Senior stakeholders and team members can communicate clearly when each member's specific responsibilities and milestones are defined. Devoid of well-defined responsibilities, team members who are driven and committed may experience animosity and exhaustion. Assigning responsibilities is undoubtedly crucial to fostering a team dynamic that functions like a well-oiled machine.

Utilize project management technologies

The preferred System Integrator may frequently reside in a different city. It is likely that the component suppliers it selects to work with are located in different states or nations. Even though they are convenient, sending emails or making phone calls can lead to misunderstandings and confusion. The integrity of the automation project is put at risk because it is difficult to maintain file version control in the absence of a centralized storage system. An increasing number of businesses are switching to device-synching project management software hosted on the cloud. These platforms can help with budget tracking, timeline management, data communication and storage, and—above all—documenting the project's progress. Project managers can better convey their vision and demonstrate leadership when they communicate well. A strong communication strategy facilitates the smooth adoption of the new solution by stakeholders, or potential system users. A team that communicates well will feel empowered and have a cohesive





Kashyap L Mokariya et al.,

group that can easily handle the numerous smaller tasks that make up an automation project. Ultimately, information synchronization and documentation between various parties and time zones can be facilitated by project management software.

To spot scope creep and actions to be taken

Scope creep is a dreaded phenomenon on any project that can result in wasted money and decreased satisfaction and a failure to meet the project's anticipated value. Scope creep appears to plague most projects, a problem that constantly irritates stakeholders and project teams alike. There are several ways in which scope creep can occur in projects. Executives at the sponsor level frequently state that they would prefer not to participate in every decision. Thus, they are created by project teams. Once more, because certain change requests appear to be minor, project teams handle them without adhering to a formal change management procedure. Strict or onerous change control procedures may also contribute to unapproved scope extensions. For a variety of reasons, the project team may feel compelled to go above and beyond and offer "more value" by adding functionality that hasn't been asked for. When requests for more functionality are made, IT managers frequently let the scope expand without pressing for more funding or time. Lack of scope definition, requirements and scope not managed, Inconsistent process for collecting requirements, lack of sponsorship or stake holder involvement and length of project are some key points to be considered.

Lack of scope definition

The project scope is stated in aspirational or lofty language. These include "better throughput" and "more cost-effective," which various stakeholders may interpret in different ways, leading to misunderstandings and confusion in the end.

Unmanaged requirements and scope

Emerging "discoveries" may change the requirements for a project. Scope creep happens gradually but steadily when these discoveries are added to the project without first undergoing a proper assessment.

Inconsistent process for collecting requirements

The cause of this phenomenon is imprecise scope definition, which leaves the project team unclear about the boundaries of the work. It can also occur when there are too many parties involved.

Lack of sponsorship or stake holder involvement

The project may lose momentum and focus if senior management or stakeholders are absent. The most frequent reason for project failures is a lack of sponsorship or stakeholder involvement.

Length of project

The likelihood of the project scope expanding exponentially increases as the timeline for the project gets longer. This occurs because senior management has more time to hone their concepts, research the competitors, or because the company evolves naturally over time.

RESULTS AND DISCUSSION

This paper's primary goal is to present comprehensive data for plant automation, including ways to increase energy efficiency, match demand, increase utility, lower costs, and improve control and preserve Environment. By enhancing all of these aspects, the overall plant stability and utility is improved. How the effective temperature control and monitoring can be provided with Programmable logic controllers is explained very effectively. In boiler to prevent over consumption of electricity at the start of the boiler, When the boiler is turned off, the heating elements are gradually heated and turned off. The example of the greenhouse ventilation panels shows how automation can benefit the environment.





Kashyap L Mokariya et al.,

CONCLUSION

The author has tried his best to justify the paper title Industrial plant automation Management best practises for great outcomes where not only the energy efficiency, emission, automation and programming is explained but the Industrial management aspects points are covered for best outcomes. Each automation project is distinct and intricate in its own right but few points are to be kept in mind. Establish specific goals and objectives and assess the project as a strategic business case using the company's mission, vision, and values. Choose the appropriate external partners and internal team members. Establishing proper communication mechanism with awareness of scope creep.

REFERENCES

1. Raul D.S.G.Campilho and Francisco J.G.Silva " Industrial Process improvement by automation and robotics, special issue industrial process improvement by automation and robotics,6th November 2023 MDPI journals.
2. Nurgul Nalgozhina, Abdul Razzaque, Uskenbayeva and Joon Yoo, " Developing robotic process automation to efficiently integrate long term business process management, 2023, 11, 164 <https://doi.org/10.3990/technologies11060164>
3. Gujar, Mukesh, Amitava Datta, and Parimita Mohanty. "Smart Mini Grid: An innovative distributed generation based energy system." *Innovative Smart Grid Technologies-Asia (ISGT Asia)*, 2013 IEEE. IEEE, 2013.
4. <https://processolutions.com/microcontroller-vs-plc-whats-the-difference>.
5. Erdal Yilmaz, Sevan Katrancioglu, "Designing Programmable Logic Controller (PLC) Experiment Set with Internal Experiment Blocks", *Procedia - Social and Behavioral Sciences* 28 (2011) 494 – 498
6. K. Sayad, H.A.gabbar, "SCADA and Smart Energy Grid Control Automation", Chapter 18, Researchgate, January 2019.
7. Mallikarjun G. Hudedmani*, Umayal R M, Shiva Kumar Kabberalli, Raghavendra Hittalamani, "Programmable Logic Controller (PLC) in Automation", *Advanced Journal of Graduate Research*, Volume 2, Issue 1, pp. 37-45, July 2017.
8. E.R. Alphonsus, M.O. Abdullah, "A review on the applications of programmable logic controllers (PLCs)", *Renewable and Sustainable Energy Reviews* 60 (2016) 1185–1205.
9. V A Shinde, R. A. Birambole, K P. Sutar, prof. V. T. Metakari, " Automatic Control Of Pneumatic Trainer Using PLC", *international journal of innovations in engineering research and technology [ijiert]*, volume 5, issue 4, apr-2018.
10. Yu-Ju Lin et al., "Integration of Logic Controller with IoT to Form a Manufacturing Edge Computing Environment: A Premise", *Procedia Manufacturing* 39 (2019) 398–405.
11. M A. Sehr, M. Lohstroh, M. Weber, I. Ugalde, M. Witte, J. Neidig, "Programmable Logic Controllers in the Context of Industry 4.0", *iee transactions on industrial informatics*, vol. 17, no. 5, may 2021.
12. I. González, A. J. Calderón, A. J. Barragán and J. M. Andújar, "Integration of Sensors, Controllers and Instruments Using a Novel OPC Architecture", *Sensors* 2017, 17, 1512.
13. A. Talware, D. Chaudhari, "Boiler Automation using PLC and SCADA", *Advances in Computational Sciences and Technology*, ISSN 0973-6107 Volume 10, Number 6 (2017) pp. 1857-1860.
14. W. J. Roberts, "Ventilation and Cooling of Greenhouses", *Greenhouse Design Short Course*, Tucson, Arizona, January 20-21, 2003.
15. R. Shamshiri, W. I. W. Ismail, "A Review of Greenhouse Climate Control and Automation Systems in Tropical Regions", *Journal of Agricultural Science and Applications (J. Agric. Sci. Appl.)*, Volume 2, Issue 3 Sep. 2013 PP. 176-183.





Kashyap L Mokariya et al.,

Table 1: Pros and cons associated with each control methods [7]

Control Method	Pros and cons
Logic for contacts and relays.	<ul style="list-style-type: none"> • Easy assembly and straight forward design. • Simple to comprehend for small systems, requiring a lot of room and being rigid for large systems. • Expensive regular,unkeep. • More driver circuit are needed for large relays. • Human involvement is inevitable. • No intermediate control except tight on adjustment and on / off. • Increases electrical noise and interference.
PLC apparatus	<ul style="list-style-type: none"> • Adaptable and practical for use. • Boosts process dependability and efficiency. • Has modest intelligence and cabling. • Reusability and good fault tolerance.
SCADA setup	<ul style="list-style-type: none"> • Because of the system is scalable and modular, keep an eye on it all in real time. • Easy to use interface with secondary messages and alarm for a particular element or plant area. • Updating events and data logs for analysis and upcoming planning • Extensive infrastructure Instrumentation sensors heavy, cabling etc. • High Initial Outlay.
DCS setup	<ul style="list-style-type: none"> • Plug and play mostly in Easy implementation • Measurable with modular and high reliability • Low cabling and limited distribution intelligence • Good wrong tolerance

Table 2: Thermostat status and temperature range

Temperature	Thermostats	Heater 1	Heater 2	Heater 3
Lower than 55° C	TS1 Close TS2 Close TS3 Close TS4 Close	ON	ON	ON
55°C -60°C	TS1 Open TS2 Close TS3 Close TS4 Close	OFF	ON	ON
60°C -65°C	TS1 Open TS2 Open TS3 Close TS4 Close	OFF	OFF	ON
65°C-70°C	TS1 Open TS2 Open TS3 Open TS4 Close	OFF	OFF	OFF





Kashyap L Mokariya et al.,

Over 70 °C	TS1 Open TS2 Open TS3 Open TS4 Open	OFF	OFF	OFF
------------	--	-----	-----	-----

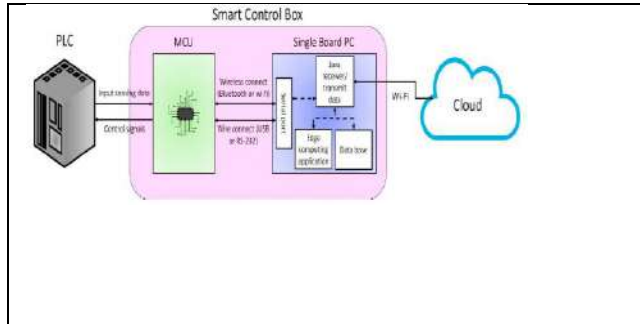


Figure 1: Detail architecture of Smart Control Box [10]

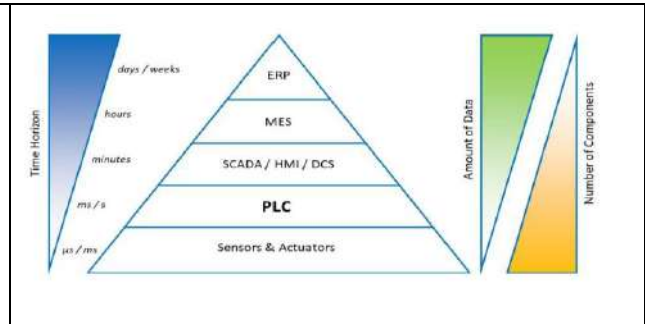


Figure 2: PLCs form the control layer of the traditional automation pyramid

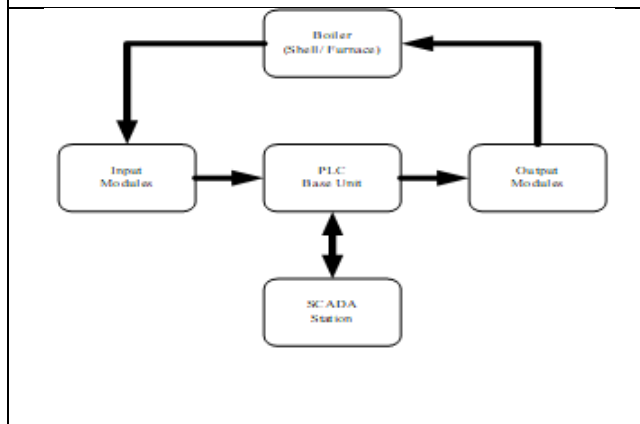


Figure 3: General Interfacing of system [13].

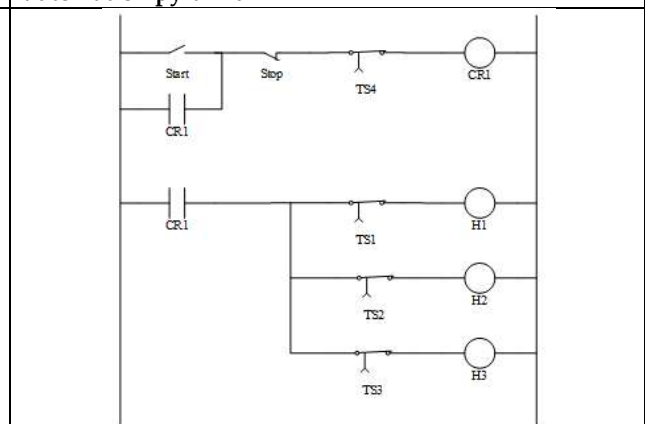


Figure 4: Ladder Logic Operation

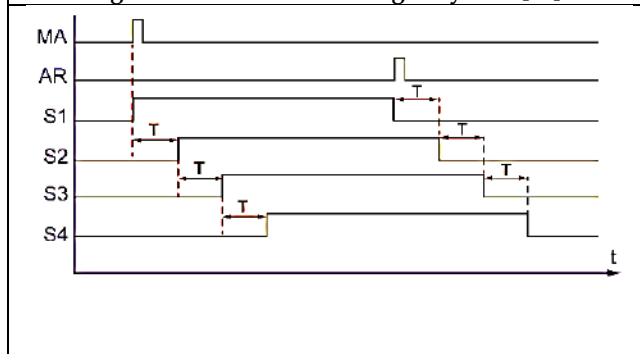


Figure 5: Operating Principle of Boiler Control



Figure 6: Ladder logic program for the boiler control





Kashyap L Mokariya et al.,

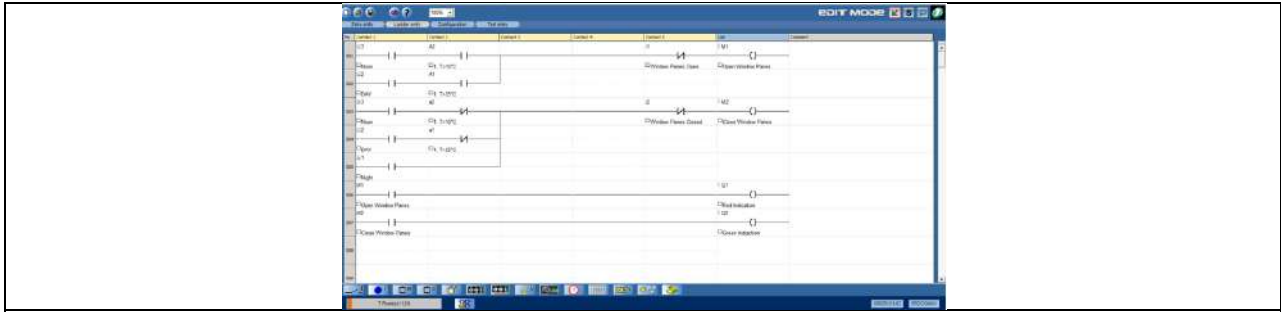


Figure 7: Ladder Logic for Green House Ventilation pans





Management of *Karappan* (Eczema) with Siddha Formulation *Sivappu Ennai* as External Application – A Case Report

S.Malini^{1*}, S.M.Chitra² and M. Muthukumaran³

¹PG Scholar, Department of PG Noinaad, Government Siddha Medical College, (Affiliated to The Tamil Nadu Dr. M.G.R. Medical University), Chennai, Tamil Nadu, India.

²Lecturer, Department of PG Noinaad, Government Siddha Medical College, (Affiliated to The Tamil Nadu Dr. M.G.R. Medical University), Chennai, Tamil Nadu, India.

³Professor, Department of PG Noinaad, Government Siddha Medical College, (Affiliated to The Tamil Nadu Dr. M.G.R. Medical University), Chennai, Tamil Nadu, India.

Received: 01 July 2024

Revised: 25 Aug 2024

Accepted: 29 Oct 2024

*Address for Correspondence

S.Malini

PG Scholar, Department of PG Noinaad,
Government Siddha Medical College,
(Affiliated to The Tamil Nadu Dr. M.G.R. Medical University),
Chennai, Tamil Nadu, India.
E.Mail: malinisi1198@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Eczema is a form of dermatitis where inflammation of epidermis occurs. The exact cause of eczema is not known, although it is activated by the immune system and is related to allergic reactions. In Siddha, the disease is described by the name "*Karappan*". A 33-year-old male, complained of skin lesion over bilateral lower limbs associated with roughness, scaling and severe itching for 6 months. The case was diagnosed as *Karappan* (a Siddha diagnosis for Eczema) and *Sivappu ennai* was used to treat the patient for 6 months. Clinical symptoms and imaged software analysis of photographs of the lesions assessed the disease severity by EASI score. The treatment given was successful in reducing the symptoms. This case study demonstrates the effectiveness of *Sivappu ennai* on eczema patient. The study concluded that Siddha treatment was significantly effective in treating eczema.

Keywords: Eczema, *Karappan*, *Sivappu ennai*, Siddha.

INTRODUCTION

The word eczema comes from the Greek word *ekzein* meaning "to boil out"; word *ek* means "out", while *zema* means boiling. ⁽¹⁾ The exact cause of eczema is unknown. People with eczema do have the IgE antibodies (Immunoglobulin E) produced by the immune system as part of allergic reactions. Eczema can be difficult and frustrating condition. The psychological challenge faced by patients of eczema is insurmountable. The natural human desire to scratch an itchy rash just makes the condition worse. ⁽²⁾ In Siddha eczema is defined as *Karappan*⁽³⁾



**Malini et al.,****Case presentation**

A 33-year-old male presented with the complaint of rashes over bilateral lower limbs associated with intense itching for past 6 months was reported at Post graduate Noi Naadal outpatient department at Aringnar Anna Government Hospital of Indian Medicine, Arumbakkam, Chennai. He had oozing of fluid associated with burning sensation and dry scaly lesions over the popliteal fossa. There was tenderness over the lesions with elevated local temperature and surrounding indurations. He had disturbed sleep due to burning sensation and itching over both lower limbs. There was no relevant family history related to his illness. Further he had no past history of any major illness or surgery. He was psychologically disturbed due to stress and there was disturbed sleep.

Clinical findings on examination

Inspection of the skin lesions showed dark blackish discoloration with an irregular margin on the bilateral lower limbs. On palpation, the pulses of the lower limbs were normal. The skin lesions on the bilateral lower limbs showed macular papule lesions with irregular shapes and borders.

Patient consent

Written permission for the publication of the images has been obtained from the patient.

Siddha Intervention

He was given *Sivappu ennai* as external application and advised to apply over the affected areas morning and night daily till the skin color changes to normal. He was advised to visit weekly once to the OPD and advised to avoid brinjal, fish, crab, prawn foods. The EASI score assessment and the timeline of the treatment of the patient was given below in the table 1,2 respectively.

Outcome and Follow up

The assessment was done before treatment, on follow up visits and after the treatment based on differences in sign and symptoms. Image analysis helped to differentiate the disease and normal skin areas. EASI score was used to assess the improvement. Before treatment the patient had 16.8 EASI score and after treatment the EASI score was reduced into 0.8 as the skin color changed to normal. At the end of 45th day he recovered completely in all subjective and objective parameters. The patient was advised to follow up for next 2 months with continuation of the same medicines and there was no recurrence of the symptoms noticed.

DISCUSSION

“Itch that rashes” is characteristic feature of Eczema.⁽⁴⁾ The lipid barrier of skin is usually reduced in the people with eczema, compared with others. The lipid barrier helps prevent water loss. As the barrier is reduced water loss will be faster as a result skin becomes dry. The immune system then overreacts to these allergens and causes inflamed, irritated, or sore skin. Before treatment, the patient had hyperpigmented patches over bilateral lower limbs along with oozing of fluid and burning sensation over the affected area (Figure1). After a course of 45 days administration of both internal and external Siddha medicines, the patient was completely relieved from itching, oozing of fluid, burning sensation. Notable changes had occurred over the hyperpigmented patches in the bilateral lower limbs. (Figure2). Along with internal and external medication Pathiyam (diet protocol suitable to the patient and disease) was advised according to Siddha system of medicine in the management of eczema. Hence, the patient was advised to avoid bitter gourd, brinjal, tamarind, chicken, egg, sea foods, fast food, and baked items. Considering etiology, the patient had a history of consumption of sweet, cold, and colored food items from the bakery and he did not drink sufficient water, which resulted in the lipid barrier damage to the skin that leads to itching, blackish discoloration of the bilateral lower limbs, exfoliation, and skin roughness.

Patient Perspective

Patient was satisfied with the treatment in terms of reduced itching, burning sensation and improved sleep.





CONCLUSION

Siddha management of eczema offers a comprehensive, individualized approach that addresses the root causes and manifestations of the disease. A remarkable change in the symptoms was noticed and provided a better outcome. Following strict dietary protocol along with both internal and external medication followed by proper lifestyle changes are the essential to prevent recurrences and sustained relief after the treatment.

Limitations

As this is a case study, the results cannot be generalized. In future, long-term follow-up studies on large samples are required to substantiate the above claims.

ACKNOWLEDGEMENTS

I acknowledge the patient and his family for giving their consent for the images taken and publication of the same.

Author's Contributions

All the authors contributed equally to the design and execution of the article.

Conflicts of interest

Nil

REFERENCES

- 14th ed. Varanasi: Chaukambha publications; Sushruta samhita – Nidanasthana; by Kaviraj Dr Ambikadattshastri; pp. 247-8. Shloka-5/5,8.
- 14th ed. Varanasi: Chaukambha publications; Sushruta samhita – Chikitsasthana; by Ambikadattshastri; p. 49. Shloka-9/6.
- Dr.R.Thiyagarajan, L.I.M, Siddha Maruthuvam Sirappu- Department of Indian Medicine and Homeopathy, 2013 Edition, Chapter-4, Page no 245 and 254.
- [Last accessed on 2010 Jul 25]. Available from: <http://www.emedicinehealth.com>.

Table 1: Eczema Area and Severity Index (EASI Score)

Score	Description
0	Clear or no lesions
0.1-1	Almost clear
1.1-7	Mild disease
7.1-21	Moderate disease
21.1-50	Severe disease
>51	Very severe disease

Table 2: Timeline of Symptoms and Treatment with EASI score Assessment

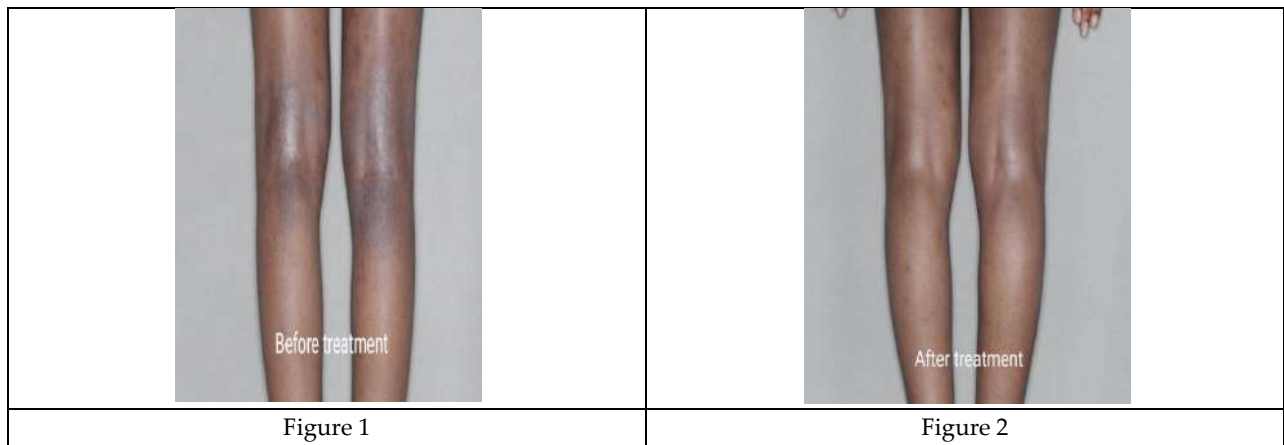
Days	Symptoms	Internal & External Medicine	EASI Score
1-10	Oozing of fluid slightly reduced, Itching, burning sensation and hyperpigmentation present.	Parangipattai chooranam- 2gm BD with warm milk, after food	16.8





Malini et al.,

10-20	Oozing of fluid completely reduced. burning sensation decreased and itching slightly reduced.	<i>Sangu parapam</i> -100mg BD with warm milk, after food	12.8
20-30	Burning sensation completely relieved, hyperpigmentation reduced with mild itching.	<i>Silasathu parpam</i> - 100mg BD with warm milk, after food	6.8
30-45	Itching completely reduced, Skin color changed to normal and patient felt normal.	<i>Sivappu ennai</i> as external application twice daily(day&night)	0.8





Assessment of Developed Temperature Tolerant Yeast Strains for Fermentation Efficiency

Ananthalakshmi Ranganathan^{1*}, Shubhi Bagwat², Anjali Yadav², Seema Paroha³, Sonali Nigam⁴

¹Assistant Professor, Department of Biochemistry, National Sugar Institute, Kanpur, Uttar Pradesh, India.

²M.Sc., Student, Department of Microbiology, St. Aloysius College (Autonomous), (Affiliated Rani Durgavati Vishwavidalaya), Jabalpur, Madhya Pradesh, India.

³Director, Department of Biochemistry, National Sugar Institute, Kanpur, Uttar Pradesh, India.

⁴Assistant Professor, Department of Microbiology, St. Aloysius College (Autonomous), (Affiliated Rani Durgavati Vishwavidalaya), Jabalpur, Madhya Pradesh, India.

Received: 29 July 2024

Revised: 22 Aug 2024

Accepted: 26 Oct 2024

*Address for Correspondence

Ananthalakshmi Ranganathan

Assistant Professor,

Department of Biochemistry,

National Sugar Institute, Kanpur,

Uttar Pradesh, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Fuel substituted for gasoline primarily consists of bioethanol. Because of the high cost of crude oil, the need of biofuel increased. The primary method to generate bioethanol fuel is the fermentation of sugar, while it may also be produced using a variety of chemical processes, such as combining ethylene with steam. Using ethanol as a biofuel or as a blended fuel can help to reduce air pollution, carbon dioxide buildup, global climate change, and reliance on foreign energy. Industries nowadays use variety of raw materials for the production of ethanol such as sugar cane molasses, beet molasses, sweet sorghum and grains etc. *Saccharomyces cerevisiae* is the most commonly used microbe used to generate bioethanol from sugar-containing feed stocks because of its ability to break down sucrose into hexoses, which include glucose and fructose. *Saccharomyces cerevisiae* functions under aerobic and anaerobic conditions. The primary product of anaerobic yeast sugar fermentation is ethanol. Following the conversion of disaccharides like sucrose in molasses into mono-saccharides like glucose and fructose by the yeast enzyme "Invertase," the yeast enzyme "Zymase" thereafter converts the mono-saccharides into ethyl alcohol and carbon dioxide. The production of ethanol by *S.cerevisiae* is affected by numerous factors which include culture medium, dissolved oxygen, immobilisation, and other micronutrients, as well as fermentation conditions such as temperature, pH, ethanol concentration, and sugar concentration. At high temperature, growth and viability of yeast cells reduces and hence the fermentative capacity and ethanol yield. In this study, temperature tolerant yeast strains were developed using Molasses Medium and Yeast Extract Peptone Dextrose Medium on applying heat stress. Based on the usual characteristics of colonies grown on Yeast Extract Peptone Dextrose Agar, the isolates were picked up for their morphological and physiological characterization. The developed temperature tolerant strains were evaluated for their fermentation capacity to ferment sugars and produce ethanol. . The results revealed that all the developed temperature tolerant strains were able to grow well at temperatures- 20°C, 25°C, 30°C, 35°C, 40°C and 45°C.



**Ananthalakshmi Ranganathan et al.,**

Conventional morphological and biochemical methods were used to characterize the developed strains and were compared with the wild commercial yeast cultures. The results reported that all of the developed temperature tolerant strains are of *Saccharomyces cerevisiae*. All the developed temperature tolerant yeast strains were able to produce ethanol in an anaerobic condition to a different extent due to the difference in temperature. The fermentation efficiency of temperature tolerant strains at 20°C and 25°C was ranging between 90 to 93%. The fermentation efficiency of temperature tolerant strains at 40°C and 45°C was ranging between 81 to 92%. The temperature tolerant strains 35M (35°C) – molasses medium based culture and 35Y (35°C) – YPD Broth Medium based cultures achieved the maximum fermentation efficiency of 99.6% and 99.6% respectively. The temperature tolerant strains developed could be used in distilleries that are located in the region having different climatic conditions with temperature ranging between 20-45°C accordingly for better fermentation efficiency.

Keywords: Distillery; Ethanol; Molasses; YPD medium, Yeast and Temperature Tolerance.

INTRODUCTION

Microorganisms such as bacteria, yeast, fungi, protozoa, and viruses have substantial effects on the environment. These microbes possess positive as well as negative impacts on the areas such as Agriculture, Food Industry, Distillery, Health sector etc. Regarding the distillery sector, the energy crisis forces research and development of novel techniques for the synthesis of renewable compounds as substitute energy sources; one important substitute is the fermentation of ethanol using renewable resources. Fuel that is environmentally benign and suitable for use in stock gasoline engines is bioethanol. Ethanol combustion produces comparatively little emissions of carbon monoxide, nitrogen oxides, and volatile organic compounds. Ethanol has less toxicity and emissions than fossil fuels like diesel, petroleum, and so on. Global ethanol production increased from 13.12 billions of gallons in 2007 to 25.68 billions of gallons in 2015 with a slight decrease in 2012 and 2013 [1]. The use of bioethanol instead of gasoline has a variety of benefits, including a higher octane number (108), wider flammability limits, higher flame speed, greater heat of vaporization, less toxicity, easier biodegradation, and a lower amount of airborne pollution [2]. Yeasts are those microbes which are solely responsible for the process of production of ethanol. Many types of yeast strains have been identified all over the world with the ability of producing ethanol from different types of feedstocks. 'Wild' yeast strains, or the yeast population that exists naturally on the fermentation feedstock or other components, have been used to produce alcohol (ethanol). Traditionally, baker's yeast was utilized as a beginning culture in the synthesis of ethanol since it was inexpensive and readily available. However, during industrial procedures, contamination occurred because baker's yeast and other *S. cerevisiae* strains were unable to compete with wild-type yeast [3].

Yeasts like *Saccharomyces cerevisiae* have been employed to produce alcohol for thousands of years, mostly in the wine and brewing sectors. Approximately 80% of ethanol is produced by anaerobic fermentation of various sugar sources by *Saccharomyces cerevisiae* [4]. It produces a high ethanol output, is very productive, and can tolerate high ethanol concentration; it keeps the cost of distillation low [5]. *S. cerevisiae* which is commonly called as fermentative yeast, used in ethanol production largely depends upon renewable biomass such as sugar cane or sugar beet molasses as the main carbon source [6, 7]. Yeast requires building block components (C, N, P, S, and O) in the right proportions, minerals (K, Na, Mg, Ca, Zn, Fe, Mn, Cu, and Co), and vitamins (B1, B5, B6, Biotin, etc.) for efficient fermentation. In commercial-scale ethanol fermentations, oxygen is often present at very low concentrations. Since oxygen is needed to produce unsaturated fatty acids, which are necessary for yeast growth and ethanol synthesis, the process cannot actually be entirely anaerobic. It has been discovered that the majority of substrates used in commercial ethanol production are nitrogen constrained [8]. Various strains of yeast, including hybrid, recombinant, and wild-type yeasts, for the production of bioethanol, a range of feedstocks from the first, second, and third generations have been used [9]. Feedstocks high in starch (corn, wheat, rice, potato, cassava, sweet potato, and barley) and sucrose (sugar cane, sugar beet, sweet sorghum, and fruits) are used to produce first-generation bioethanol. Bioethanol of the second generation is derived from ligno-cellulosic biomass, which includes grasses, wood, and straw. Micro and macroalgal biomass has



**Ananthalakshmi Ranganathan et al.,**

been used to produce third-generation bioethanol [10]. The fermentation process that results from using molasses to produce ethanol will add value; to the byproduct. The non-crystallizable residue that remains after purifying sucrose is called molasses. It offers a few benefits, including being a commonly available, reasonably priced raw material that doesn't require starch hydrolysis and has already been used to produce ethanol. After sugar beets are processed, molasses is produced that comprises 40% additional ingredients and roughly 60% sucrose. The non-sucrose substances consist of nitrogen-containing molecules, organic acids, raffinose, ketose, and inorganic salts. Molasses is utilized to generate acetone, butanol, glycerol, citric, lactic, and gluconic acids; it is also employed to make baker's yeast, ethanol, mixed feeds, and amino acid manufacturing been employed in fermentation to produce ethanol. While other types of feedstocks need to first be converted into fermentable sugars in order so as to be fermented to ethanol, simple sugars can be directly fermented into ethanol by yeasts [11, 12]. In India, cane juice is not currently used to produce ethanol; instead, sugar cane molasses is the primary feedstock. Another source of fermentable sugars for the production of ethanol is beet molasses [13].

Strains, growth nutrients, ethanol concentration and ideal climatic conditions are only a few of the variables that affect yeast's capacity to produce ethanol. Yeast's ability to tolerate its environment, including temperature, fermentation products, and substrate (osmo- and thermo-tolerance), has significant potential to be utilized in fermentation on an industrial scale[14].The ecology and adaptation of the existing microbiota can be influenced by several factors that impact the fermentation process. During the fermentation of sugar, yeasts are frequently faced with two challenges: an increase in temperature from 35 to 45°C and a concentration of ethanol exceeding 20%. Until the temperature reaches its ideal level, the growth rate and metabolism of yeast increase with temperature [15].One factor that directly influences the rate at which the microbes grow is the temperature.A temperature shift of a few degrees to twenty degrees Celsius over the usual growth temperature is referred to as heat shock. There is a general relationship between the rate of growth and the susceptibility to stress in yeast, meaning that cells developing rapidly in a glucose-rich media are more susceptible to heat and other stresses than stationary phase cells [16].Heat shock causes changes in the lipid and protein content of membranes as well as the physical states of intracellular water, which increases the permeability of membranes. Saturated esterified fatty acids, such palmitic and palmitoleic acids, are more abundant in yeast cell membranes at the expense of unsaturated acyl chains, like oleic, linoleic, and linolenic acid, as temperature rises. This is typically linked to a reduction in the level of phospholipids in the membrane necessary to maintain ideal membrane fluidity for cellular processes, some of which may be adaptive in nature. The heat shock response, the cell's survival mechanism in the midst of a stressful environmental situation, is triggered by these biochemical changes and the accumulation of unusual proteins in the cell [17]. *Saccharomyces cerevisiae* is used in industrial fermentations, which produces metabolic heat. If cooling is not applied, the fermentation will stop being isothermic. From the start of fermentation to its conclusion, temperatures might rise as much as 15°C [13].With the goal to achieve maximum ethanol production while reducing energy costs, thermo-tolerant yeast is required.Many species of yeast, including *Pichia* sp., *Candida* sp., *Kluyveromyces marxianus*, and certain strains of *Saccharomyces cerevisiae*, have been identified and categorized as thermo-tolerant yeasts. The ideal temperature range for the majority of *Saccharomyces cerevisiae* strains is between 25°C and 30°C. Using chemical and physical mutagenesis techniques, some thermo-tolerant mutants of haploid strains of *S. cerevisiae* have been identified and genetically described [18].

MATERIALS AND METHODS

Collection and Primary Analysis of the Sample

B Heavy Molasses sample was obtained from Experimental Sugar Factory, National Sugar Institute, Kanpur. Analysis of the molasses sample was done based on the parameters such as Total Reducing Sugar (TRS), Reducing Sugar (RS), Brix, Specific Gravity, Sludge, Total Dissolved Solids, Alcohol Percentage and pH. The molasses sample was diluted to 10% TRS, analyzed based on the above parameters.



**Ananthalakshmi Ranganathan et al.,****Fermentation**

The diluted molasses sample was supplemented with Commercial Yeast, Urea and Sodium Phosphate Dibasic Anhydrous and kept for aerobic and anaerobic fermentation at 30°C for 24 hours

Growth on Medium

A pre-grown yeast culture from aerobic flask was inoculated into molasses medium and YPD Broth Medium (Yeast Extract-10g, Peptone- 20g, Dextrose- 2g, and Distilled Water-1000 ml). pH of both the medium was adjusted between 4.5-5.0 [19]. Both the mediums were incubated at 6 different temperatures- 20°C, 25°C, 30°C, 35°C, 40°C and 45°C for 24 hours. Cell viability was determined for each of the twelve samples.

Isolation, Identification and Characterization

Yeast cultures from Molasses and YPD Broth Medium was streaked on YPDA and incubated at 6 different temperatures for 24-48 hours. Pure culture was obtained for each isolate. Morphological characters were studied using macroscopic and microscopic features. Macroscopic characters such as shape colour, elevation, texture of the isolated colony was studied and Microscopic traits such as shape of the yeast isolate, cell arrangement and budding was observed. Biochemical characterization was done by Carbohydrate Fermentation Test using Durham Tube Method. Durham tubes were utilized to test yeasts for the fermentation of carbohydrates. YP Broth was used for characterization of the yeast isolates based on fermentation of specific carbohydrates. The sugars used for the test were dextrose, sucrose, fructose, trehalose, starch, α -methylglucoside, galactose, maltose and mannitol. The basal media for the fermentation test includes Yeast Extract- 1g, Peptone- 2g and Sugar- 2g, Distilled Water-100ml and 0.1% Methyl Red Indicator. Durham Tube was used to observe the formation of gas bubbles and Methyl Red Indicator was used for observing the color change of the media; from red to yellow due to production of acids [20]. Pellicle formation was also observed for each yeast isolate [21].

Distillation and Fermentation Efficiency

Yeast cultures from YPDA slants of Molasses and YPD Broth Medium was transferred to molasses medium for aerobic fermentation at 6 different temperatures for 24 hours. After 24 hours cultures from aerobic flask of Molasses and YPD Broth Medium was transferred into molasses medium (100ml each) for anaerobic fermentation at 6 different temperatures for 24 hours. After fermentation distillation was performed for each yeast isolate, strength of the alcohol was determined and fermentation efficiency.

RESULT AND DISCUSSION**Primary Analysis of the Molasses**

B Heavy Molasses sample for the study was obtained from the Experimental Factory of National Sugar Institute, Kanpur and was analyzed for various parameters. The result of the analysis carried out is given in the Table 1. The Total Reducing Sugar, Reducing Sugar, Specific Gravity, Sludge, Brix, pH and Total Dissolved Solid of the B Heavy Molasses sample utilized for the study was calculated as 54.1%, 2.4%, 1.42, 11.4%, 84°, 4.8 and 35.0 PPT respectively. The molasses sample obtained from NSI was diluted to 10% for further study.

Aerobic and Anaerobic Fermentation

For the purpose of aerobic and anaerobic fermentation the molasses having the TRS 54.1% was diluted as per the formula $N1V1 = N2V2$ to 10% and various parameters was analyzed for the same. The Total Reducing Sugar, Reducing Sugar, Specific Gravity, Sludge, Brix, pH, Total Dissolved Solid and Alcohol Percentage of the diluted Molasses sample was calculated as 9.8%, 0.02%, 1.42, 8.3%, 28°, 4.8, 3.5 PPT and 5.6% respectively. The molasses sample was subjected to aerobic and anaerobic fermentation. Cell counting of molasses medium from aerobic fermentation was carried out using Hemocytometer (Table 2) and molasses medium for anaerobic fermentation was subjected to distillation to determine the strength of alcohol.



**Ananthalakshmi Ranganathan et al.,**

Following aerobic fermentation for 24 Hrs at 30°C, Total Cell Count was calculated as 315×10^6 Cells/ml, Dead Cells were found to be 1.58%, Viable Cells and Bud Count was found to be 98.4% and 15.8 % respectively.

Growing of Yeast on Molasses Medium and YPD Broth Medium

Both Molasses and YPD Broth Medium were prepared and taken in different flasks marked as 20M (20°C), 25M (25°C), 30M (30°C), 35M (35°C), 40M (40°C) and 45M (45°C) and 20Y (20°C), 25Y (25°C), 30Y (30°C), 35Y (35°C), 40Y (40°C) and 45Y (45°C). Both Molasses and YPD Broth Medium were kept at respective temperatures (20°C, 25°C, 30°C, 35°C, 40°C and 45°C). This is followed by inoculation of yeast culture from aerobically fermented culture. The growth of yeast in both Molasses and YPD Broth Medium was observed after 24hrs of incubation. The growth of yeast cultures was found to be increasing with temperature from 20°C to 35°C and the total cell count decreased at 40°C and till 45°C incubation. This pattern of growth was observed in both Molasses and YPD Broth Medium. The four strains namely VS1, VS2, VS3 and VS4 were able to grow well in broth cultures till 44°C [22].

Determination of Cell Viability

Hemocytometer was used to assess the vitality of the cells in all the inoculated flasks after 24-hrs. The total number of cells, viable cells, dead cells and budding count was calculated and the values are given in the Figure 2 and 3 for cultures grown in Molasses and YPD medium at different temperatures. The cell counting was found to be highest in the case of 35M and 35Y (grown at 35°C); it was found that the total cell count first increased as the temperature increases up to 35°C then it decreased until it reaches 45°C. The dead cells were not found in case of 25M (25°C), 30M (30°C) and 30Y (30°C) and 35°C (35M and 35Y) whereas with increasing the temperature the dead cells were also increased due to the stress created by the temperature. The viability of the cells first increased up to 35°C with increase in temperature and then decreased. The maximum percentage of bud count was found to be maximum at 35°C (35M and 35Y).

Isolation of Temperature Tolerant Yeast Strains on YPDA Medium

The yeast cultures grown on Molasses and YPD Broth Medium at different temperatures were isolated on YPDA Medium by streak plate method. Isolated yeast from both the medium grown at different temperatures is shown in the Figure 4 and 5. The yeast isolates from both the medium grew well at 30°C and 35°C. The growth of the yeast isolates decreased as the temperature rise above 35°C. Out of four strains used (VS1, VS2, VS3 and VS4), VS1 and VS3 grew well on plates till 44°C but the growth of VS2 and VS4 was very less at 44°C [22]. Four strains namely LGBA-01, LGBA-69, LGBA-157 and LGBA-175 and all the strains had same pattern of growth at 30°C and 40°C [23]. In the present study, the isolated yeast colonies were found to be white in colour, smooth in texture, margin entire. Yeast colonies from different food waste were found to be smooth ridged, shiny and creamish [24]. Colony color of yeast strains from palm wine were ranging from cream white to colorless [25].

Pure Culture of the Yeast Isolates

Pure Culture was prepared by picking single isolated colonies obtained on petriplates from Molasses Medium Based Cultures and YPD Broth Medium Based Cultures and streaking it on YPDA slants (shown in Figure 6 and 7). The growth of pure culture of Yeast Culture was observed after 72hrs of incubation at 20°C, 25°C, 30°C, 35°C, 40°C and 45°C. The yeast isolates from Molasses Medium and YPD Broth Medium on petriplates were examined for colour, texture, margin, shape, elevation and colony size as indicated in Table 3 and Table 4. All of the Yeast Cultures isolated from Molasses Medium Based Cultures were white in colour. The texture was observed as smooth, with an entire margin. The shape was found to be circular with a convex elevation. The colony size of 30M and 35M isolates were found to be 2×2 mm while 25M was significantly larger, measuring 3×2.5 mm. In the case of Yeast Cultures isolated from YPD Broth Medium the colonies were white in colour, smooth in texture, with an entire margin, circular in shape, and convex in elevation. The colony size of 45Y was found to be highest as 3×2.5 mm. Yeast cultures isolated, from soil samples and fruit gardens showed a rough surface with a white or creamy white tint, the majority of colonies had a smooth surface. Most of the colonies margins were lobate, serrated, or entire [26]. On macroscopic observations of yeast isolates from palm wine, the elevation of all the isolates was found to be raised. Hence they



**Ananthalakshmi Ranganathan et al.,**

reported that the colonies of yeast strains possessed morphological features which corresponded to *S. cerevisiae* species [25].

Microscopic Characteristics

Lactophenol Cotton Blue Staining was done to confirm the shape of the yeast cell and the formation of buds under the microscope (Table 5 and 6). The cell shape of the yeast cultures from molasses medium based cultures was found to be oval. The shape was round in the case of 30M (30°C) and 45M (45°C). The cell arrangement was isolated, and some of them were grouped in tiny clusters. Budding was observed in all the cultures. In the case of yeast cultures from YPD Broth Medium Based Cultures the cell shape was oval and round. The cell arrangement was isolated, with some forming small clusters. Budding was observed in all the cultures. The isolated yeast cultures were ovoid in shape and multipolar budding was observed [19]. Yeast from palm wine were found to have an ovoid to circular shape, the cells appeared singly and varied in size from 2 to 6 µm [25]. The yeast isolates had a range of cell morphologies, including spherical, ovoid, elongated elliptical, and cylinder, with lengths between 3 and 11 µm and widths between 2 and 6 µm [26].

Pellicle Formation

Pellicle Formation was examined by inoculating the yeast cultures in YPD Broth Medium followed by incubation for 48 hrs. The results are shown in Figure 8 and 9. Pellicle formation was not observed in any of the cultures of yeast cultures isolated from molasses medium based cultures as well as YPD broth medium based cultures. Yeast cultures from samples of soil, fruit, and fermented food were isolated none of them formed pellicle [21].

Biochemical Test

The Carbohydrate Fermentation Test was used to determine the ability of the yeast isolate to ferment specific sugar by releasing carbon dioxide and changing the colour of the medium. It was done by inoculating cultures of yeast in basal medium with Durham's tube and the indicator methyl red. It was done using nine different carbohydrate sources which were numbered as Dextrose-1, Starch-2, Fructose-3, Trehalose-4, Maltose-5, α-Methylglucoside-6, Mannitol-7, Sucrose-8 and Galactose-9. The results are shown in Table 7 and Figure 10.

All the yeast cultures from molasses medium based cultures as well as YPD broth medium based cultures were able to ferment following 72hrs of incubation. The isolates were observed to be fermenting reducing sugars (Dextrose, Fructose, Galactose, Trehalose, Mannitol and Maltose) and non reducing sugar (Sucrose and α-Methylglucoside). The yeast cultures were unable to ferment starch even after 72hrs. Out of 20 thermotolerant strains only 4 of them (YP11, YM17, YPA48, YPA64) were able to ferment all the nine sugars used (Glucose, Galactose, Sucrose, Mannose, Lactose, Raffinose, Arabinose and Xylose and Fructose) [24]. The yeast isolated from wine samples were subjected to biochemical assimilation test using various sugars like Glucose, Sucrose, Maltose, Xylose, Galactose, Lactose, Raffinose, Melibiose, Mannitol, Trehalose, Erythritol. The ability of yeast to assimilate Glucose, Raffinose and Maltose confirmed for *S. cerevisiae* [25].

Fermentation Efficiency of Temperature Tolerant Strains

The Yeast Cultures were kept for anaerobic fermentation followed by distillation. The fermentation efficiency of the temperature tolerant yeast strains was calculated. The results of the analysis are shown in Figure 11. In the case of Molasses Medium Based Cultures, the isolate 35M (grown at 35°C) was found to have the highest Practical Yield and Fermentation Efficiency (5.15 and 99.6% respectively). In the case of YPD Broth Medium Based Cultures, the isolate 35Y (grown at 35°C) was found to have the highest Practical Yield and Fermentation Efficiency (5.20 and 99.6% respectively). According to [27] the strain K-7 yields 6.2% ethanol at 40°C after 48 hrs of fermentation with total sugar concentration of 23% and the strain RND 13 produced 6.6% ethanol at 40°C at 15% glucose concentration. Whereas in the present study, at 40°C the strain 40M and 40Y yields 2.61% and 2.65% ethanol respectively at 10% TRS.

The concentrations of ethanol were lowest at higher temperatures and maximum at 30°C and 35°C. Even though the cells were producing less ethanol and growing at 44°C, they were still viable at higher temperatures. The four strains



**Ananthalakshmi Ranganathan et al.,**

VS1, VS2, VS3 and VS4 produced ethanol (g/l) - 66.0, 50.0, 75.2 and 55.0 respectively at 30°C using 150g glucose/l. All these strain produced ethanol (g/l) – 66.0, 48.0, 75.0 and 52.0 respectively at 35°C using 150g glucose/l. All the above data were reported by [22]. Where as in the present study, at 30°C the strain 30M and 30Y yields 5.0% and 5.18% ethanol respectively at 10% TRS. At 35°C the strain 35M and 35Y yields 5.15% and 5.20% ethanol respectively at 10% TRS.

CONCLUSION

The experimental results reported in the present study revealed that twelve temperature tolerant yeast strains were developed viz Molasses Medium- 20M, 25M, 30M, 35M, 40M and 45M and YPD Broth Medium-20Y, 25Y, 30Y, 35Y, 40Y and 45Y. All the developed temperature tolerant strains were able to grow at their respective temperatures (20°C, 25°C, 30°C, 35°C, 40°C and 45°C). By comparing the developed temperature-tolerant strains with wild commercial yeast cultures, standard morphological (macroscopic and microscopic) analysis and biochemical approaches were used to characterize them. Upon performing the tests, the developed strains were confirmed to be *Saccharomyces cerevisiae*. All the developed temperature tolerant yeast strains were able to produce ethanol in an anaerobic condition to a different extent due to the difference in temperature. In order to assess the ability of the strain to produce ethanol, fermentation efficiency was calculated for all the twelve temperature tolerant yeast strains. The results showed that the temperature tolerant strain 35M and 35Y had the maximum efficiency of 99.6% and 99.6%. Though the optimum temperature of *Saccharomyces cerevisiae* to grow is 25°C to 35°C; the results of the present study showed that at 20°C, the fermentation efficiency was found to be 90.3% and 91.7% which means the developed temperature tolerant strains was able to ferment sugar and produce 3.57% and 3.34% at 20°C. In order to improve fermentation efficiency, temperature-tolerant strains that have been produced could be employed in distilleries situated in regions with varying climatic conditions, with temperatures ranging from 20 to 45°C.

ACKNOWLEDGMENT

The author expresses gratitude to National Sugar Institute, Kanpur, U.P., India, for its assistance in supporting the research project.

CONTRIBUTIONS

Ananthalakshmi , Shubhi Bhagwat and Anjali Yadav were involved in research and analysis. Seema Paroha and Sonali Nigam helped in reviewing the paper.

CONFLICT OF INTEREST

The author declares that they have no conflict of interest.

REFERENCES

1. Renewable Fuels Association US, Global Ethanol Production, 2015
2. Balat, M., & Balat, H. (2009). Recent trends in global production and utilization of bio-ethanol fuel. *Applied energy*, 86(11), 2273-2282.
3. Basso, L. C., De Amorim, H. V., De Oliveira, A. J., & Lopes, M. L. (2008). Yeast selection for fuel ethanol production in Brazil. *FEMS yeast research*, 8(7), 1155-1163.
4. Bai, F. W., Anderson, W. A., & Moo-Young, M. (2008). Ethanol fermentation technologies from sugar and starch feedstocks. *Biotechnology advances*, 26(1), 89-105.
5. Kasavi, C., Finore, I., Lama, L., Nicolaus, B., Oliver, S. G., Oner, E. T., & Kirdar, B. (2012). Evaluation of industrial *Saccharomyces cerevisiae* strains for ethanol production from biomass. *Biomass and bioenergy*, 45, 230-238.



**Ananthalakshmi Ranganathan et al.,**

6. Echegaray, O. F., Carvalho, J. C. M. D., Fernandes, A. N. R., Sato, S., Aquarone, E., & Vitolo, M. (2000). Fed-batch culture of *Saccharomyces cerevisiae* in sugar-cane blackstrap molasses: invertase activity of intact cells in ethanol fermentation. *Biomass and Bioenergy*, 19(1), 39-50.
7. Sanchez, O. J., & Cardona, C. A. (2008). Trends in biotechnological production of fuel ethanol from different feedstocks. *Bioresource technology*, 99(13), 5270-5295.
8. Bassapa S.C, 1989. Recent development in ethanol fermentation process. Central Food Technology Research Institute.
9. Azhar, S. H. M., Abdulla, R., Jambo, S. A., Marbawi, H., Gansau, J. A., Faik, A. A. M., & Rodrigues, K. F. (2017). Yeasts in sustainable bioethanol production: A review. *Biochemistry and biophysics reports*, 10, 52-61.
10. Nigam, P. S., & Singh, A. (2011). Production of liquid biofuels from renewable resources. *Progress in energy and combustion science*, 37(1), 52-68.
11. Belitz, HD, Grosch, W, Schieberle, P, (2009) Food Chemistry, 4th edition, Springer.
12. Satyanarayana, T. (2009). *Yeast biotechnology: diversity and applications*. Springer Science.
13. Fleet, G. H. (1993). *Wine microbiology and biotechnology*. CRC press.
14. Khongsay, N., Laopaiboon, L., & Laopaiboon, P. (2010). Growth and batch ethanol fermentation of *Saccharomyces cerevisiae* on sweet sorghum stem juice under normal and very high gravity conditions.
15. Tofighi, A., MazaheriAssadi, M., Asadirad, M. H. A., & ZareKarizi, S. (2014). Bio-ethanol production by a novel autochthonous thermo-tolerant yeast isolated from wastewater. *Journal of Environmental Health Science and Engineering*, 12, 1-6.
16. Elliott, B., & Futcher, B. (1993). Stress resistance of yeast cells is largely independent of cell cycle phase. *Yeast*, 9(1), 33-42.
17. Piper, P. W. (1993). Molecular events associated with acquisition of heat tolerance by the yeast *Saccharomyces cerevisiae*. *FEMS microbiology reviews*, 11(4), 339-355.
18. Walker, G.M. Yeasts, In *Eukaryotic Microbes*; Academic Press/Elsevier Science Publishers: Oxford, UK, 2011; pp. 3–17.
19. Kurtzman, C. P., Fell, J. W., & Boekhout, T. (2011). Definition, classification and nomenclature of the yeasts. In *The yeasts* (pp. 3-5). Elsevier
20. Negera, T. (2017). Isolation and Characterization of Ethanol, Sugar and Thermo Tolerant Yeast Isolates in Ethiopia. *International Journal of Research Studies in Biosciences*, 5(8).
21. Ali, M. N., & Khan, M. M. (2014). Screening, identification and characterization of alcohol tolerant potential bioethanol producing yeasts. *Curr Res MicrobiolBiotechnol*, 2(1), 316-324
22. Sree, N. K., Sridhar, M., Suresh, K., Banat, I. M., & Rao, L. V. (2000). Isolation of thermotolerant, osmotolerant, flocculating *Saccharomyces cerevisiae* for ethanol production. *Bioresource Technology*, 72(1), 43-46.
23. Prado, C. D., Mandrujano, G. P., Souza, J. P., Sgobbi, F. B., Novaes, H. R., da Silva, J. P., ... & Cunha, A. F. (2020). Physiological characterization of a new thermotolerant yeast strain isolated during Brazilian ethanol production, and its application in high-temperature fermentation. *Biotechnology for biofuels*, 13, 1-15.
24. Jayamma, P., Shirnalli, G. G., Goudar, D. G., & Olekar, S. N. (2022). Isolation and identification of thermotolerant yeast isolates from different fruit wastes. *The Pharma Innov. J*, 11(1), 855-859.
25. Olowonibi, O. O. (2017). Isolation and characterization of palm wine strains of *Saccharomyces cerevisiae*, 7:11
26. Phong, H. X., Klanrit, P., Dung, N. T. P., Yamada, M., & Thanonkeo, P. (2019). Isolation and characterization of thermotolerant yeasts for the production of second-generation bioethanol. *Annals of Microbiology*, 69, 765-776.
27. Ueno, R., Hamada-Sato, N., & Urano, N. (2003). Fermentation of Molasses by Several Yeasts from Hot Spring Drain and Phylogeny of the Unique Isolate Producing Ethanol at 55°C. *JOURNAL-TOKYO UNIVERSITY OF FISHERIES*, 90, 23-29





Ananthalakshmi Ranganathan et al.,

Table 1: Analysis of B Heavy Molasses Before and After Dilution

Parameters	B Heavy Molasses	Diluted Molasses (10%)
TRS (%)	54.1 ± 0.1	9.8 ± 0.1
RS (%)	2.4 ± 0.1	0.02 ± 0.01
Specific Gravity	1.42 ± 0.01	1.050 ± 0.01
Sludge (%)	11.4 ± 0.1	8.3 ± 0.1
Brix (°)	84 ± 1	28 ± 1
pH	4.8 ± 0.1	4.8 ± 0.1
TDS (PPT)	35.0 ± 0.1	3.5 ± 0.1
Alcohol (%)	-	5.6 ± 0.2

Table 2: Cell Counting of Molasses Medium from Aerobic Fermentation

Cell Counting	
Total Cell Count (Cells/ml)	315×10 ⁶
Dead Cells (%)	1.58
Viable Cells (%)	98.4
Bud Count (%)	15.8

Table 3: Macroscopic characteristics of yeast isolate from Molasses Medium Based Cultures

Sample	Colour	Shape	Texture	Margin	Elevation	Colony Size (mm)
20M	White	Circular	Smooth	Entire	Convex	2×2.5
25M	White	Circular	Smooth	Entire	Convex	3×2.5
30M	White	Circular	Smooth	Entire	Convex	2×2
35M	White	Circular	Smooth	Entire	Convex	2×2
40M	White	Circular	Smooth	Entire	Convex	1.5×1.5
45M	White	Circular	Smooth	Entire	Convex	2×2.5

Table 4: Macroscopic characteristics of yeast isolates from YPD Broth Medium Based Cultures

Sample	Colour	Shape	Texture	Margin	Elevation	Colony Size (mm)
20Y	White	Circular	Smooth	Entire	Convex	2×2
25Y	White	Circular	Smooth	Entire	Convex	1.5×2
30Y	White	Circular	Smooth	Entire	Convex	2×2.5
35Y	White	Circular	Smooth	Entire	Convex	1.5×2.5
40Y	White	Circular	Smooth	Entire	Convex	2×2
45Y	White	Circular	Smooth	Entire	Convex	3×2.5

Table 5: Microscopic characteristics of Yeast Cultures isolated from Molasses Medium Based Cultures

Sample	Shape	Cell Arrangement	Budding
20Y	Oval	Isolated or grouped in small clusters	Present
25Y	Oval and Round	Isolated or grouped in small clusters	Present
30Y	Oval	Isolated or grouped in small clusters	Present
35Y	Oval and Round	Isolated or grouped in small clusters	Present
40Y	Oval	Isolated or grouped in small clusters	Present
45Y	Oval and Round	Isolated or grouped in small clusters	Present





Ananthalakshmi Ranganathan et al.,

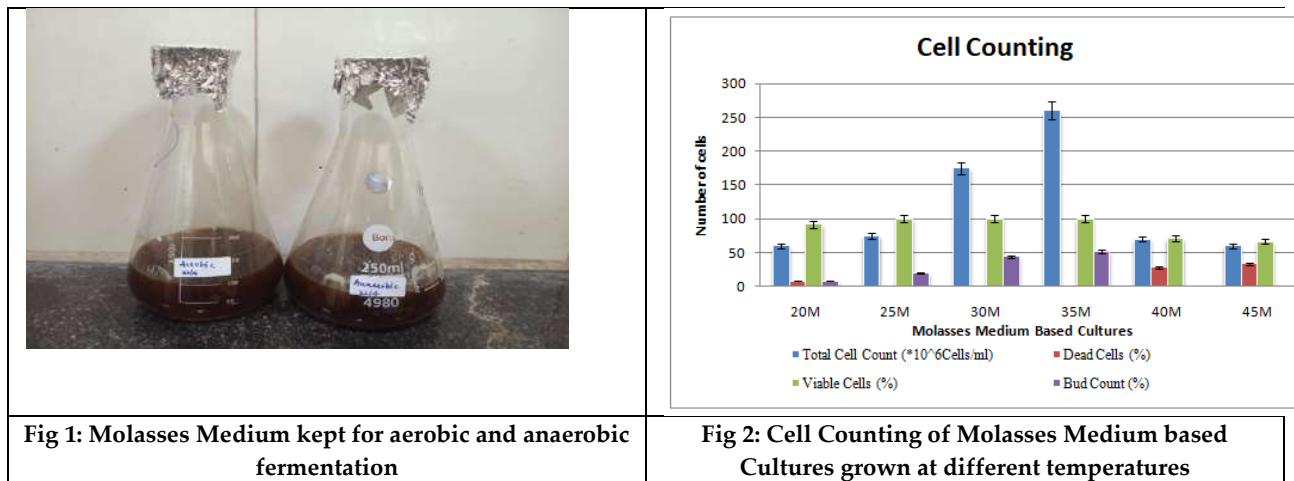
Table 6: Microscopic characteristics of Yeast Cultures isolated from YPD Broth Medium

Sample	Shape	Cell Arrangement	Budding
20Y	Oval	Isolated or grouped in small clusters	Present
25Y	Oval and Round	Isolated or grouped in small clusters	Present
30Y	Oval	Isolated or grouped in small clusters	Present
35Y	Oval and Round	Isolated or grouped in small clusters	Present
40Y	Oval	Isolated or grouped in small clusters	Present
45Y	Oval and Round	Isolated or grouped in small clusters	Present

Table 7: Carbohydrate Fermentation Test of Temperature Tolerant Yeast Strains from Molasses and YPD Broth Medium Based Cultures

Type of Sugar	Sample											
	20M	20Y	25M	25Y	30M	30Y	35M	35Y	40M	40Y	45M	45Y
Dextrose	+	+	+	+	+	+	+	+	+	+	+	+
Starch	-	-	-	-	-	-	-	-	-	-	-	-
Fructose	+	+	+	+	+	+	+	+	+	+	+	+
Sucrose	+	+	+	+	+	+	+	+	+	+	+	+
Galactose	+	+	+	+	+	+	+	+	+	+	+	+
Trehalose	+	+	+	+	+	+	+	+	+	+	+	+
α Methylglucoside	+	+	+	+	+	+	+	+	+	+	+	+
Mannitol	+	+	+	+	+	+	+	+	+	+	+	+
Maltose	+	+	+	+	+	+	+	+	+	+	+	+

(+) Positive, (-) Negative





Ananthalakshmi Ranganathan *et al.*,

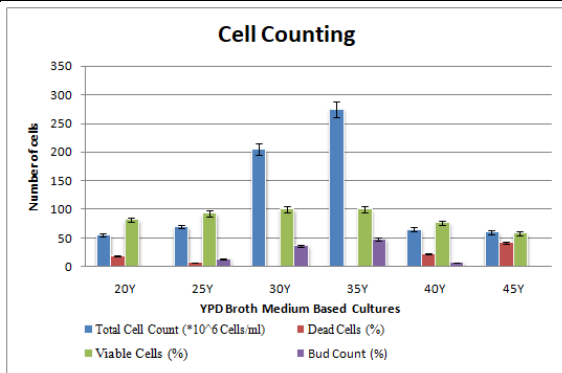


Fig 3: Cell Counting of YPD Broth Medium based Cultures grown at different temperatures



Fig 4: Growth of Yeast Cultures from Molasses Medium based cultures on YPDA Medium



Fig 5: Growth of Yeast Cultures from YPD Broth Medium based cultures on YPDA Medium

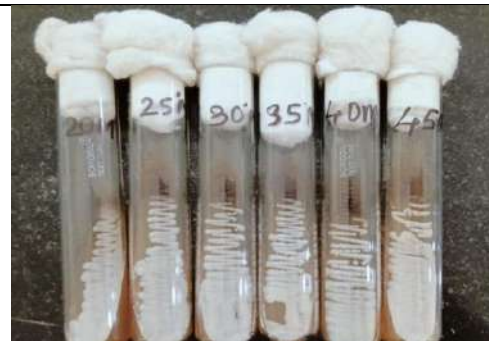


Fig 6: Pure Culture of Yeast Cultures from Molasses Medium Based Cultures



Fig 7: Pure Culture of Yeast Cultures from YPD Broth Medium Based Cultures



Fig 8: Pellicle Formation in Yeast Cultures from Molasses Medium Based Cultures

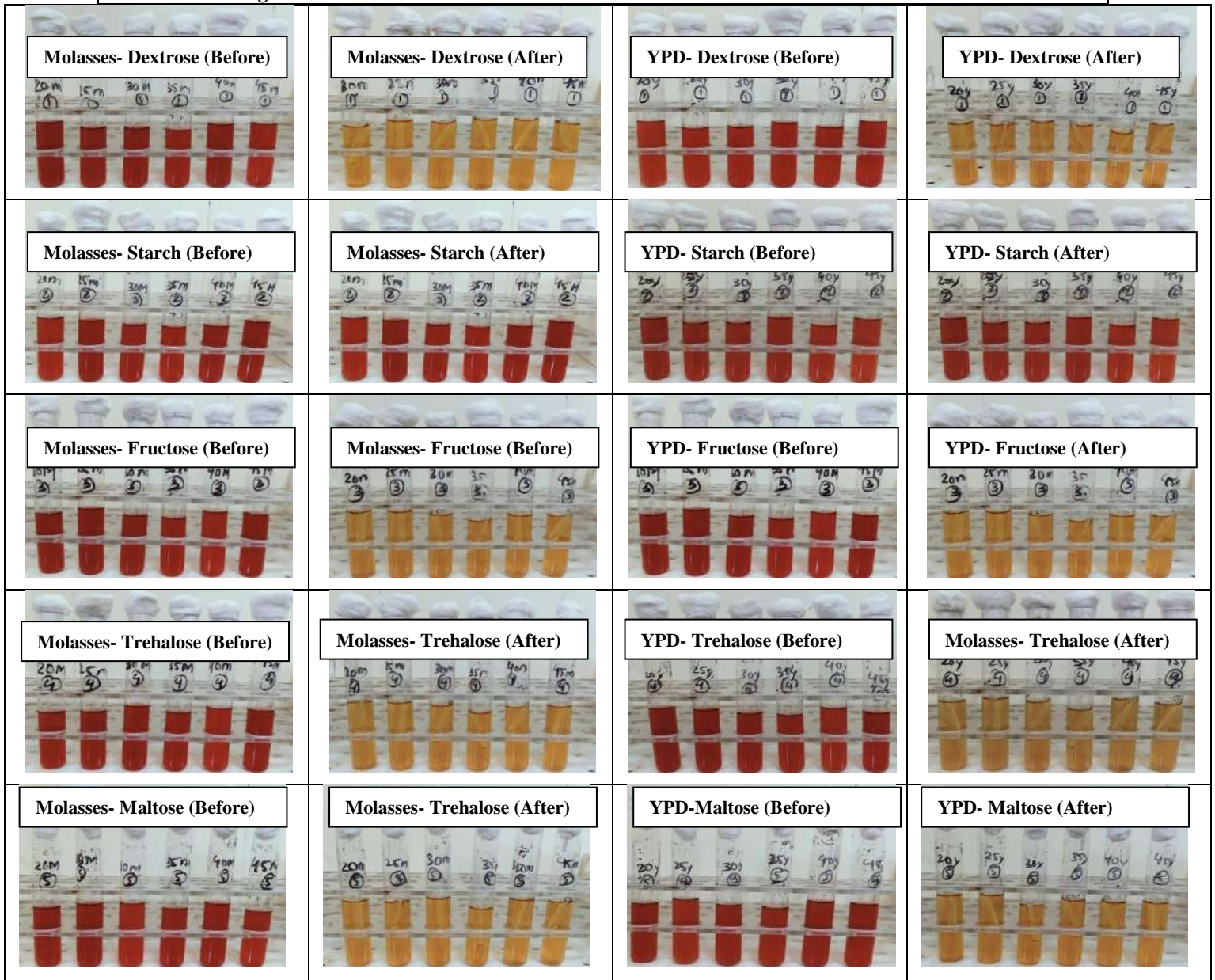




Ananthalakshmi Ranganathan et al.,



Fig 9: Pellicle Formation in Yeast Cultures from YPD Broth Medium Based Cultures



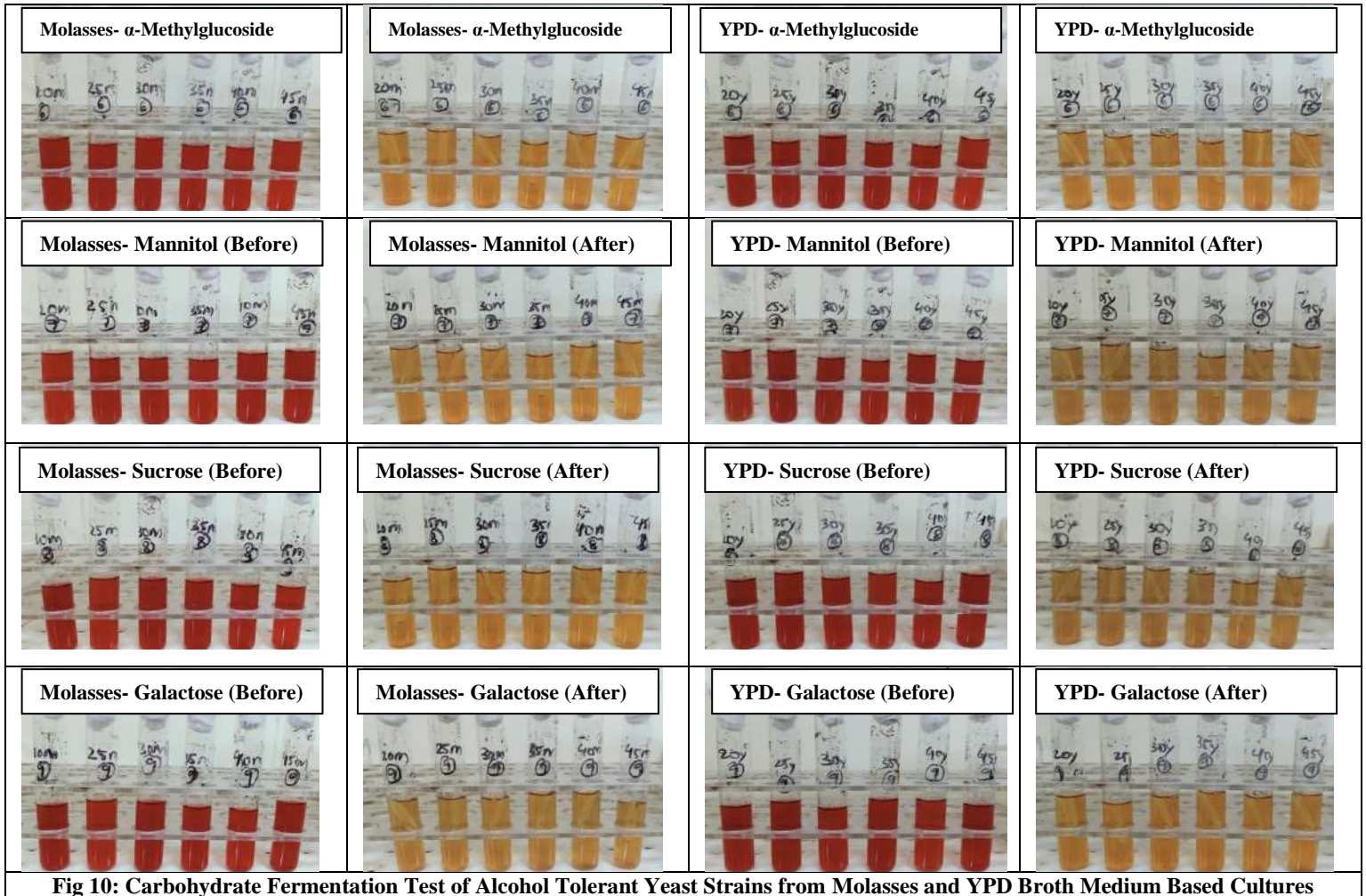


Fig 10: Carbohydrate Fermentation Test of Alcohol Tolerant Yeast Strains from Molasses and YPD Broth Medium Based Cultures

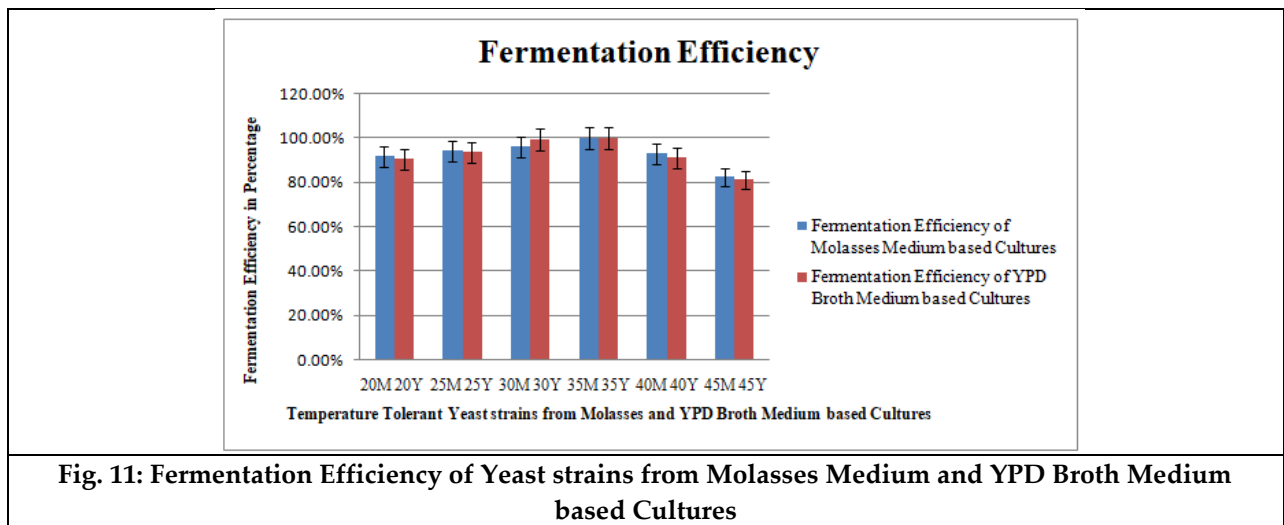


Fig. 11: Fermentation Efficiency of Yeast strains from Molasses Medium and YPD Broth Medium based Cultures





Generational Differences with Respect to Perceived Social Support, Self-Esteem and Resilience

Vrinda Antapurkar¹ and Vaishali Choudhari^{2*}

¹Student, MIT World Peace University, Pune, Maharashtra, India

²Assistant Professor, MIT World Peace University, Pune, Maharashtra, India

Received: 02 Jun 2024

Revised: 06 Aug 2024

Accepted: 18 Oct 2024

*Address for Correspondence

Vaishali Choudhari

Assistant Professor, MIT World Peace University,
Pune, Maharashtra, India

E.Mail: vaishalichoudhari9@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Generational gap and differences have always been a topic of research. These intergenerational differences are prominent and can be observed very distinctly, be it in terms of physical, social, emotional, and/or psychological abilities. This study aimed to investigate a significant difference between Generation X and Generation Z in terms of self-esteem, resilience, and perceived social support. The survey incorporated basic questions about the participants such as their names, ages, and gender. It most importantly consisted of 3 self-report measures namely the Rosenberg Self-Esteem Scale, The Multi Social Scale of Perceived Social Support, and the Connor Davidson Resilience Scale. 172 participants of Indian nationality participated in this study. Out of which 86 belonged to Generation X and 86 to Generation Z. Independent samples t-test was used to analyze the data. The findings demonstrated a significant difference between the two generational cohorts in terms of self-esteem, resilience, and perceived social support.

Keywords: Gen X, Gen Z, Self-esteem, Resilience, Perceived Social Support

INTRODUCTION

The word 'generation' is commonly used when denoting an entire age group of people and locating individuals within a historical time frame. Many social scientists have tried to define a generational cohort in their ways. According to Karl Mannheim(1952), a generation can be described as, "A group that is distinctive in any number of respects by having experienced a specific set of social, economic, technological, and/or political circumstances at a formative period in their lives". The term 'generational cohort' is often used interchangeably with terminologies like 'age cohort' or 'birth cohort' in fields like social sciences, marketing, humanities, demographics, etc. A generational cohort is a group of people who were born within a few years of each other into the same historical and social-cultural context and have developed common attributes caused by shared participation in similar life events (Lyons



**Vrinda Antapurkar and Vaishali Choudhari**

&Kuron, 2014; Schermerhorn, 2013; Twenge et al., 2010)(Lyons, S., Urick, M., Kuron, L., & Schweitzer, L., n.d.). This research deals with two generational cohorts: Generation X also called Gen X and Generation Z called Gen Z. It tries to understand variables such as self-esteem, resilience, and perceived social support and how they affect these generational cohorts as well as their comparative analysis.

Generation X (also referred to as Gen X)

Though there has been a debate about the birth years of Generation X, this generational cohort is thought to be roughly born between the mid-1960s and the early 1980s. Generation X individuals grew up facing economic difficulties and uncertainties as well as rapid social change. Hence, they embody a spirit of independence and self-reliance. They tend to be skeptical of authority and institutions compared to other generational cohorts. They have a strong sense of work-life balance and prioritize work-life along with personal enrichment. Generation X is known for their strong work ethic, adaptability, and self-sufficiency (Tulgan, 1996). They face heightened polarization and anxiety due to the uncertainty of societal positions and ambiguous rules (Rymarz, 2006). However, contrary to some portrayals, they are not universally cynical or indifferent (Maschaykh, 2012). Instead, they display a blend of traditional and innovative mindsets, with various age segments within the cohort gravitating toward distinct mentalities (Pishchik, 2020). Research on Generation X has revealed a variety of viewpoints, encompassing both favorable and unfavorable assessments. Mensik(2007) and Maschaykh (2012) both acknowledge the existence of negative stereotypes, with Mensik advocating against oversimplified and restrictive categorizations. However, Tulgan (1996) counters these stereotypes by portraying Generation X as industrious individuals driven by a strong ambition to excel in their careers.

Generation Z (also referred to as Gen Z)

Generation Z, born post-1995, constitutes a distinct demographic marked by innate digital proficiency, distinct attributes, and a need for further scholarly exploration (Jayatissa, 2023). Their formative years have been shaped by pivotal events like the aftermath of 9/11, the Great Recession, and the COVID-19 crisis, rendering them a diverse, globally-connected, and mobile cohort (McKee-Ryan, 2021). Within academia, they exhibit adeptness with technology and anticipate access to online educational resources (Zorn, 2017). In professional settings, they manifest unique work ethics and career inclinations, heavily influenced by the impacts of the COVID-19 pandemic (Mahapatra, 2022). They are recognized as digital natives, adept at utilizing technology and engaging with social media platforms (Sakashita, 2020; Jayatissa, 2023). Notably, this generation is celebrated for its diversity, exhibiting a notable level of racial inclusivity (Seemiller, 2015). They are characterized as entrepreneurial, socially aware, pragmatic, and encompassing diverse perspectives (Jayatissa, 2023). Additionally, they are described as intelligent, driven, innovative, accountable, compassionate, and empathetic (Seemiller, 2015). However, despite these commendable attributes, they confront challenges stemming from growing up in an era marked by economic instability and natural calamities (Sakashita, 2020). They often harbor pessimism influenced by economic and environmental concerns (Sakashita, 2020), and demonstrate lower commitment to organizations and long-term endeavors compared to prior generations (Lev, 2022). This cohort exhibits adeptness with technology, relying heavily on social media and experiencing a fear of missing out (Herawati, 2022). They prioritize flexibility and digital work environments, displaying a penchant for pursuing personal aspirations in the workplace (Ruzsa, 2018). These dynamics can present obstacles in the job market and hinder efforts to maintain harmonious work-life equilibrium.

Generation Z grapples with distinctive psychological hurdles stemming from their digital upbringing and the influence of globalization (Ermolova, 2020; Maioli, 2016). Their engagement with virtual reality further molds their psychological makeup, exhibiting diverse degrees of depth and direction (Puchkova, 2017). These elements may pose challenges in their transition into the workforce, as they harbor dissimilar expectations and requirements in contrast to preceding generations (Ermolova, 2020; Maioli, 2016).

Self – Esteem

Self-esteem is defined as, “One’s positive or negative attitude toward oneself and one’s evaluation of one’s thoughts and feelings overall in relation to oneself” (Rosenberg,1965).The American Psychological Association (2023)



**Vrinda Antapurkar and Vaishali Choudhari**

Self-esteem is defined as, "The degree to which the qualities and characteristics contained in one's self-concept are perceived to be positive." As anyone can determine, high self-esteem paves the way for better mental health and helps you handle adversity easily. It helps us deal with the stresses of everyday life effectively whether it be from school, work, social circles, etc. (Nguyen et al., 2019). High self-esteem also prevents and reduces the risk of mental disorders like depression and anxiety and helps us face challenges in life while maintaining a favorable attitude towards ourselves (Henriksen et al., 2017). Low levels of self-esteem are unfavorable to one's self-concept and overall well-being. Someone with a low level of self-esteem has trouble with prioritizing themselves, has an external locus of control, exhibits negative self-talk, thinks very poorly about themselves, has a poor outlook on the future, etc. As mentioned earlier, having a negative sense of self-esteem can cause one to become prone to mental health disorders like Anxiety, and depression. Not only that it can also lead to Internet Addiction (Naseri et al., 2015), social anxiety disorder (Maldonado et al., 2013), and risky behavior (Gartland et al., 2013). Someone with high self-esteem can shake off negative feedback or constructive criticisms very easily while someone with a low sense of self-esteem takes everything personally. This can make them more susceptible to giving up and not facing challenges (Kalvin et al., 2016). The development of self-esteem is largely governed by experiences in an individual's life (Baumeister, 2003). Parenting styles in childhood have a major impact on the self-esteem of a person (Coopersmith, 1967). Unconditional love from parents in childhood plays a major role in helping a child develop a sense of being respected and cared for. These feelings are carried out later in life as the child grows older (Olsen et al., 2008). Self-esteem is likely to increase during adolescence and young adulthood, being its highest in middle adulthood (Orth and Robins, 2014).

Research suggests a significant difference between Gen X and Gen Z in terms of self-esteem (Lyons, 2007). Innumerable factors like the rise of social media, and technological advancement are responsible for this major shift between the cohorts. Generation Z is the generation in which social media has been introduced. It has been observed that although Gen Z is skilled in handling social media, they face challenges when it comes to face-to-face interactions, which could affect their self-esteem. This issue is compounded by the prevalent fear of being left out on social media platforms, known as FOMO, among Gen Z individuals (Herawati, 2022). Social networking sites can influence the self-esteem of individuals by triggering cognitive mechanisms, by providing a space for social comparison, and by internalizing beauty standards (Vogel et al., 2014). Low usage of social media reported high levels of self-esteem and life satisfaction (Blachnio et al., 2016). Twenge (2012) further noted that younger generations place more importance on extrinsic values like money, image, and fame and there has been a gradual decline in intrinsic values such as community service, charity, self-acceptance, etc. which could also impact self-esteem. These findings suggest societal and cultural shifts may influence different generations' self-esteem. One factor contributing to the prevalence of birth cohort effects on self-esteem is the differential impact of cultural and historical events on individuals depending on their age at the time. Elder's (1979, 1981a, 1981b) research on the Great Depression provides a classic illustration of this concept. He observed that individuals who experienced depression during childhood exhibited distinct outcomes compared to those who encountered it during adolescence. Lower levels of self-esteem have been linked with an increase in factors like self-consciousness, vulnerability to criticism, the tendency to present false impressions in front of others as well and the tendency to fantasize are some of the traits and tendencies present that affect self-esteem (G. Elliot, 1982). These traits are present in today's generation which brings out lower levels of self-esteem.

Resilience

'Resilience' term was popularized by psychologist Emmy Werner in the 1970s and 1980s, who conducted a 40-year study involving a cohort of Hawaiian children belonging to low socio-economic backgrounds. Her research consisted of risk and resilience among the children. Resilience can be defined as the qualities in a person that help him/her to thrive in adverse situations and can also be considered a measure of stress-coping abilities (Connor-Davidson, 2003). The American Psychological Association (APA) defines Resilience as the process and outcome of successfully adapting to difficult or challenging life experiences, especially through mental, emotional, and behavioural flexibility and adjustment to external and internal demands. Both internal and external factors affect a person's level of resilience. Internal factors like self-assessment, self-regulation, and a positive outlook on life, and external factors like relationships with family, friends' community, and social support systems are important regulators of resilience. (Southwick et al., 2014)



**Vrinda Antapurkar and Vaishali Choudhari**

Resilience plays a key role in promoting well-being and mental health as well as physical health. Psychological resilience plays a very important role in promoting mental health well-being as well as physical well-being. Resilient people are more capable of navigating life's challenges, maintaining positive emotions, and recovering from setbacks. They endure life's difficult challenges, have good problem-solving skills, and show self-efficacy (Bonanno, et al., 2004). Resilience can also be associated with bouncing back after a stressful situation occurs in one's life. When a person encounters a lot of stress it disturbs the external and internal sense of balance. Studies have shown that it is not just stress that affects a person's balance but how the person perceives stressful situations is also an indicator of how resilient a person is (McGonigal, 2016).

Generation Z is also popularly known as the 'Digital natives' and 'Net generation'. As the name suggests, this generation is and has been going through a lot of Technological advancements and changes caused due to it. Being highly vulnerable to social and economic risks this cohort is considered prey to the digital predators. Researchers suggest that this affects the resilience of Gen Z negatively (Novkoska, 2018). It is believed that exposure to adverse conditions develops resilience in an individual (Luecken and Gress, 2009). Individuals belonging to Generation Z are perceived to be 'bubble wrapped' due to helicopter parenting (Talmon, 2019), hence they don't face adverse conditions as such and don't build up higher levels of resilience. (Ang et al., 2021). Psychological resilience is determined by an intricate nexus of social, genetic as well as environmental factors. Socio-contextual factors like gender, race, ethnicity, age, exposure to trauma, chronic diseases, stress factors, changes in income, education, and life stresses can impact resilience (Bonanno, 2007). The transmission of psychological resilience between generations is seen to be dependent upon family dynamics as well (Regin et al., 2016). Generation X seemed to have stronger family dynamics and bonds whereas Generation Z lacked in these terms. Diverse challenges like transition to adulthood and economic hardship (Conger, 2002) can affect the levels of resilience between the two generations. Demographic features like education level, sex, and income level lead up to 11% of variation in resilience. The individuals who have suffered childhood maltreatment contributed to the anticipation of resilience by 2%. Individuals who have encountered childhood mal-treatment, and individuals with lower levels of income and education demonstrated lower levels of resilience (Campbell, Forde and Stein, 2009).

Perceived Social Support

Perceived Social support refers to one's understanding and perception of the emotional, tangible, and informational support they receive from their family members, friends, and others. (Zimet et al, 1988). Social support is a multidimensional concept, referring to the social and psychological support an individual receives or perceives as available to them from family, friends, and their community (Awang et al., 2014; Zimet et al., 1988).

Perceived social support refers to the perception that support would be available if needed (Day & Livingstone, 2003) and comprises emotional and instrumental support (Trepte & Scharkow, 2016). Ample evidence suggests that social support plays an important role in promoting psychological health and mental well-being (Cohen, 2004) and that social support gives a sense of protection in psychological terms. (Haddadi & Besharat, 2010). Higher levels of social support are associated with an enhanced sense of psychological well-being (Glozah, 2013; Poudel et al., 2020). Social support has consistently been shown to affect mental health disorders like depression, and anxiety and reduce their effects significantly (Ibrahim et al., 2013). Life Satisfaction was highly correlated with social support from family and friends (Harikandei, 2017) and lower loneliness (Lee & Goldstein, 2016). According to Day and Livingstone (2003), perception of one's social support network and resources has majorly greater coping effects than if they receive the support and exerts a stronger effect on mental health than actual social support received (Hefner & Eisenberg, 2009). The belief that one has access to social support can act as reassurance in times of need acts as a protective barrier in times of stress, and can make a person happy and makes them have a positive outlook toward life (Winemiller et al., 1993). Individuals' perception of support is determined strongly by actual interpersonal communications as reported by significant others, moderately by the recipients' negative outlook bias, and weakly by their anxiety and depression as indicators of their poor mental health (Vinkour, Schul, and Caplan, 1987). Social support was significantly related to positive dimensions of subjective mental well-being such as gratification, self-esteem, and happiness but not to its negative dimensions such as uncertainty, strain, and vulnerability, and was also significantly related to individuals'



**Vrinda Antapurkar and Vaishali Choudhari**

perceived ability to indulge in positive life events, but not to their perceived ability to avoid or cope with negative events (Meehan, Durlak and Bryant, 1993).

Research conducted on the perception of support in Generation X and Generation Z has revealed significant insights. Reinikainen (2020) discovered a positive correlation between organizational attentiveness on social platforms and increased trust and perceived advantages in the content disseminated by corporations and entities. Goodwin-Smith (2019) emphasized the significance of constructive interpersonal bonds in optimizing resilience and well-being among marginalized teenagers and young adults. Nautiyal (2017) conducted a comparative analysis of the perceived support among adolescents dwelling in urban and rural environments, underscoring its pivotal role in fostering a sense of certainty amidst varied circumstances. Lastly, Feather (2012) uncovered a favorable relationship between values associated with transcending oneself and backing for communal initiatives, implying their potential influence on the readiness to rectify prior injustices. Research into the perceptions and values of Generation Z suggests the possibility of lower levels of perceived social support. Leslie (2021) distinguished three unique subsets within Generation Z, each with different priorities regarding the workplace, implying potential variations in their requirements for social support. Rola (2018) revealed Generation Z's relatively diminished empathetic values, potentially affecting their capacity to both offer and receive social support. McIver (2001) observed a growing sense of reduced backing for public services among young individuals, which might extend to social support systems. Hathaway (2022) underscored the potential influence of digital media on Generation Z's social interactions, which could further shape their perceptions of social support.

The present study deals with Generation X and Generation Z in terms of self-esteem, perceived social support, and resilience. It also sheds light on factors that might affect both the generational cohort's variables mentioned. We often experience differences in generational cohorts. We are all the same but we have experienced our lives differently. For example, Generation Z's parents find it difficult to handle technology whereas Generation Z has found out their way of living through technology with ease. The previous generations were physically active and demonstrated better levels of fitness than the later generational cohorts. Transformations like these are thought to be the foundation of why so many changes, physical and psychological, arose. This study analyses whether Generation X and Generation Z have or do not have a significant difference in self-esteem, resilience, and perceived social support. So many attributes like technological advancements, social media, parenting styles, the COVID-19 pandemic, family dynamics, etc. are the root causes these variables such as self-esteem, resilience, and perceived social support are affected inter-generationally and are chosen to be studied.

Objectives of the study:

- To assess levels of self-esteem between Generation X and Generation Z.
- To assess levels of resilience between Generation X and Generation Z.
- To assess levels of perceived social support between Generation X and Generation Z.

REVIEWS OF LITERATURE

Kvintova and Cakirpaloglu (2019) published a paper on "Self Esteem, Social Network Use and Life Satisfaction among College Students of Generation X, Y, and Z." This study aimed to correlate the level of self-esteem and life satisfaction to social network use inter-generationally. There was no correlation found between self-esteem and amount of time spent on social media. It was found that Generation Z was inclined to be more satisfied in two areas: leisure time and sexuality. They use social media three times compared to Gen X and Y. Also, they had more number of friends on social media. Soest (2016), in his study, "The Development of Global and Domain-specific Self-esteem from Age 13 to 31", examined the development of global self-esteem and self-esteem in 6 domains across adolescence and young adulthood. It demonstrated that later generations exhibit high self-esteem when it comes to social self-esteem and appearance.



**Vrinda Antapurkar and Vaishali Choudhari**

Twenge and Campbell (2001) have explored how self-esteem has changed over different age groups and birth cohorts over time and found that younger generations born in the 1980s and 1990s had higher levels of self-esteem than the previous generations such as those born in the 1960s and 1970s. R Van Woensel (2018) researched, "Generation Z: A Generation of lower self-esteem". The findings suggested that Generation Z showcased lower levels of self-esteem than the previous generations due to various factors like social media influences, academic and career pressures, and the ongoing culture of comparison among other factors. Sethi (2023) conducted a study on, "The role of Perceived Social Support in self-esteem and resilience among young adults." The results showed that perceived social support correlates positively with resilience and self-esteem among young adults. No significant gender differences were found. Angeles and Perkins (2023) explored how intergenerational differences affect self-efficacy coping and resilience of members of a family during a crisis (COVID pandemic). The results portrayed that resilience and a sense of coherence are interrelated but self-efficacy coping shows no disparities between the generations. Frankczok Kuczmowska and Barbara Superson (2023), in their research, "Resilience in the Context of the Socio-Political Situation of young adults in Europe", displayed contrasting results to the current study. This research focused on how amidst the social and political situation of Europe young adults are showing the highest levels of resilience than any other age group.

Hypotheses:

- There will be a significant difference between Generation X and Generation Z in terms of self-esteem.
- There will be a significant difference between Generation X and Generation Z in terms of resilience.
- There will be a significant difference between Generation X and Generation Z in terms of perceived social support.

METHODOLOGY**Participants:**

The population consisted of Indian citizens. Individuals belonging to Generation X and Generation Z were used as samples in this study. Stratified random sampling was used as the sampling technique. Separate age groups of 18 to 28 years for Generation Z and 43 to 58 years for Generation X were selected for this research. The following generational cohorts were excluded from this study:

- a) Baby Boomers (1946-1964): 59-77 years old.
- b) Millennials or Generation Y (1981-1996): 29-42 years old.
- c) Generation Alpha (early 2010s- 2025): 0-10 years old.

A total of 172 responses were collected for the research. 86 responses were collected for Generation X and 86 were collected for Generation Z. Demographic features like name, age, and gender were collected as well. 61.5% of the individuals who participated in the study were females whereas 38.5% were males.

Tools

- a) The Rosenberg Self-Esteem Scale: Morris Rosenberg developed the scale to measure one's self-esteem. It contains 10 items that measure both positive and negative feelings about the self. A 4-point Likert scale is used to answer the items with options being Strongly agree, Agree, Disagree and Strongly Disagree. Strongly agree carries the value of 4 whereas strongly disagree carries the value of 1. Higher scores indicate high self-esteem and low scores indicating low self-esteem. As this scale uses Guttman's scalogram, it includes reverse scoring as well. The scale has a high reliability as Cronbach's Alpha for the scale was reported at a high Alpha equal to 0.894. It demonstrates concurrent, predictive, and construct validity.
- b) Connor-Davidson Resilience Questionnaire: The Connor Davidson Resilience Scale was constructed by Kathryn M. Connor and Jonathan R. T. Davidson. The scale aims to measure resilience or the ability to bounce back after a traumatic event or stress-inducing situation. The scale has 2, 10, and 25-item versions. In this research 25-item version has been used. It is a 5-point Likert scale ranging from 0 to 4. 0 being not true at all and 4 being true nearly all of the time. The scores range from 0 to 100 with higher scores indicating high levels of resilience and



**Vrinda Antapurkar and Vaishali Choudhari**

lower scores indicating low levels of resilience. The CD-RISC demonstrates good reliability with Cronbach alpha being 0.88. Construct, convergent, and predictive validities have been established.

- c) The Multi-dimensional Scale of Perceived Social Support: Perceived social support refers to one's understanding and perception of the emotional, tangible, and information support they receive from their family members, friends, and others (Zimet et al., 1988). The multi-dimensional scale of perceived social support (MSPSS) was created by Zimet, G.D., Dahlem, N.W., Zimet, S.G., and Farley, G.K. (1988). It was developed to measure perceived social support, particularly from 3 sources that are friends, family, and significant other. It includes 12 items and is a 7-point Likert scale ranging from 1 to 7. Very strongly disagree is 1 and very strongly agree is 7. The mean scores range from 1 to 7 with low scores indicating low levels of perceived social support whereas high scores indicating high levels of perceived social support. The scale shows a high reliability with Cronbach's Alpha being equal to 0.85. A good Construct validity has been established.

Procedure

Assessment was done by circulating Google form. The link was sent to family members, friends, relatives as well as acquaintances. The form consisted of five parts, the first part included the information about the research, the second part included the demographic details, and the third one consisted of the Rosenberg self-esteem scale (RSE). The fourth part included the Multidimensional scale of perceived social support (MSPSS) and the fifth one consisted of the Connor Davidson resilience scale (CD-RISC). Instructions were given before each section on how to solve the questions. All the observations received in the form of responses were properly collected and arranged in a tabular format for statistical analysis. It was ensured that confidentiality would be maintained and the data collected from them would be used for purposes concerning research only.

Statistical Analysis

After the data collection, scoring was done. The data was analyzed by using SPSS software. Independent sample t-test is for the analysis.

RESULT AND INTERPRETATION

All the collected data was subjected to SPSS software in order to obtain the results. An Independent sample t-test was used to analyze the difference between resilience, self-esteem, and perceived social support between Generation Z and Generation X.

Self-esteem and generational cohort:

Results indicate a significant difference between Generation X and Generation Z regarding self-esteem. Generation Z ($M[SD]= 28.74[3.717]$) reported significantly ($t=4.325$) lower levels of self-esteem than Generation X ($M[SD]= 31.36[4.360]$).

Perceived Social Support and generational cohort

Results indicate a significant difference between Generation X and Generation Z regarding perceived social support. Generation Z ($M[SD]= 5.13[1.335]$) reported significantly ($t=3.326$) lower levels of perceived social support than Generation X ($M[SD]= 5.71[0.919]$).

Resilience and generational cohort

Results indicate a significant difference between Generation X and Generation Z regarding resilience. Generation Z ($M[SD]= 69.74[12.217]$) reported significantly ($t=3.176$) lower levels of perceived social support than Generation X ($M[SD]= 75.74[11.394]$).



**Vrinda Antapurkar and Vaishali Choudhari**

DISCUSSIONS

It is known fact that changes happen from generation to generation. This investigation aimed to find these differences with respect to self-esteem, perceived social support and resilience. Results revealed significant differences between Generation X and Generation Z in all mentioned variables. Generation X is showing high level of self-esteem, resilience and perceived social support as compared to Generation Z.

In self-esteem, various researches shed light upon how Generation Z has lower levels of self-esteem than Generation Y. Various factors such as helicopter parenting, social media, economic uncertainty, etc. are responsible for a lower self-esteem. One possible explanation for Generation Z's low self-esteem might be the fact that Generation Z has grown up in an Era dominated by social media which propagates perfectly showing our lives even when they are not. Djedovic` et al. (2021) in their study, "Post Millenials: Meet Generation Z", highlighted that Generation Z is the first generation to be born after the surge of the internet. As a result of being born in a world in control of social media, caring about the number of likes they will get and the amount of followers they have. Their happiness will depend upon the instant gratification they will get as soon as they see that someone has liked their story or post. Since their life is based upon being in the public's eye through social media, their appearance matters a lot to them. This makes them an egocentric generation with no empathy. Being so conscious about looks and appearance in social media and real life makes this generation insecure and have a low sense of self-esteem. The rise of social media has also paved the way for building unrealistic expectations, the inability to have face-to-face conversations and over-dependence on social media. Parenting styles as mentioned earlier also add as a contributing factor to low levels in self-esteem. Bee (2017) researched parenting styles and successive generations which aimed to investigate the relationship between self-esteem and parenting styles across three generational cohorts consisting of baby boomers, Generation X and Generation Y. The hypothesis was stated that the permissive parenting style would increase with successive generations and that would lead to a decline in self-esteem. One plausible explanation was given that whenever the child is subjected to a permissive parenting style and given unconditional positive regard completely, they search for validation externally as they have a particular self-image formed but later, he/she realizes that the external world does not treat everyone equally. Always getting unconditional positive regard from parents as a child and getting positive comments and appreciation even after doing average/mediocre tasks can lead to an initial boost in self-esteem as a child but can lead to false and high expectations from the outside world as well. This generation also faces a lot of economic uncertainty which leads to minimal job opportunities and a low sense of financial security for the individuals belonging to this generational cohort. The proposed hypothesis that there will be a significant difference between Generation X and Generation Z in terms of self-esteem is supported by results.

In case of perceived social support, Generation Z indicated lower levels of perceived social support than Generation X. A lot of research has been done that supports the hypothesis. A plethora of factors have contributed to lower perceived social support levels. Factors like age, gender, educational qualifications, lower usage of social media, etc. have contributed to these findings. A study by Chen et al. (2014) called, "The perception of social support among U.S. Chinese older adults: findings from PINE Study" aimed to examine the correlates and perceptions of negative and positive social support among U.S. Chinese older adults. The findings demonstrated that being of a younger age and being male correlated positively with negative social support or lower levels of perceived social support. Older adults portrayed high levels of perceived social support and it is dependent on factors like higher health status, better quality of life, higher education levels, gender (Female), and improved health over the past years were associated with higher levels of perceived social support. Another research by Coventry et al. (2004) focused on how perceived social support differs across age and sex. The sample consisted of participants aged between 18-95 years. It was found that perceived social support varied across different age groups. Older adults belonging to the previous generational cohorts have higher levels of perceived social support than the younger adults of the later generations. One of the reasons why Generation Z has lower levels of perceived social support might be because of the poor quality and a lack of close-knit relationships, be it, with friends, partners, parents, etc. They engage in superficial relationships that do not have any kind of emotional depth or emotional intimacy. Reasons for this can include the



**Vrinda Antapurkar and Vaishali Choudhari**

emergence of social media which results in a lack of face-to-face communication, loneliness, and genuine social communication. Luong, Charles and Fingerman (2011) in their study, "Better with age: Social relationships across adulthood", emphasize why older adults have significantly reported greater satisfaction with their social connections compared to younger individuals. Older adults who belong to the previous generations have better bonding with their children, good and closer friendships, and long-lasting marriages which overall leads to a higher sense of perceived social support amongst the older adults. It was also observed that interpersonal clashes often decline with age. Relationships that are less problematic and ambivalent were seen than those of the younger adults. Interpersonal stressors were also reduced, such as agreements and disagreements, than younger adults. An interesting finding demonstrated that the ability to navigate through difficulties in a relationship increases with age. The proposed hypothesis that there will be a significant difference between Generation X and Generation Z in terms of perceived social support is accepted and supported by results.

In case of resilience, Generation Z has shown lower levels of resilience than Generation X. Resilience, which is defined as the successful ability to adapt to adversity and stress, is affected by personal competence, self-acceptance, and the capacity for meaning and improvisation. The global pandemic like COVID-19 has affected resilience of individuals across the globe. Smith (2023) research explored the undergraduate students' resilience during the Covid 19 pandemic on Generation Z. 27 students participated in this study to share their experiences in focus groups. The findings suggested that the pandemic harmed the mental health and resilience of Generation Z students. It was implied in this study that Generation Z has lower levels of resilience than any other generation. COVID-19 compromised a lot of factors like self-esteem, self-efficacy, and bonding, a sense of belongingness, emotional health, and subjective well-being which promote to one's overall resilience. Hence, the levels of resilience had been reduced. Social isolation due to COVID-19 has also paved a way for feeling lonely which in turn has affected resilience as well. Sethi (2023) conducted a study on young adults and the point of the study was to know the role of perceived social support in promoting resilience and self-esteem among young adults. From the result of the study, the present hypothesis, that Perceived Social support portrays a positive relationship between self-esteem and resilience, is accepted. It means that the self-esteem of a person is directly proportional to the perception of social support; they can quickly handle things and cope with the problem at hand, more than those who perceive a low amount of social support and differences according to their gender. There exists a perfect correlation between the three variables. In the case of Gen Z, these factors are likely to be lacking due to the negative impact on their formation, for instance, economic instability and the adaptation to adult life. This implies that personal and environmental factors might play equal roles in the perception of Gen Z's low resilience. As mentioned earlier, low levels of perceived social support are seen in Generation Z individuals. Low levels of perceived social support are directly proportional to loneliness and the feeling that no one will be there in times of need. Hence, it decreases the capacity to withstand tough conditions and arise from them. Parenting is also one of the important factors. Helicopter and permissive parenting are the current trends in parenting and Generation Z kids are subjected to such parenting styles. As mentioned earlier these are a cause for low self-esteem. It can also be a source for lower resilience levels. Overly protective parenting can cause children to never face any kind of adversities in their lives and can explain why Generation Z is lacking in resilience. COVID-19 has also contributed to a major decrease in the resilience levels of individuals all over the world. Generation Z which already has lower levels of resilience has faced prominent repercussions for struggling with social isolation and dealing with loneliness as there was no social contact. The proposed hypothesis that there will be a significant difference between Generation X and Generation Z in terms of resilience is supported by results.

CONCLUSION

This investigation concluded with the findings that all proposed hypotheses are accepted and there are significant differences between two Generations (X and Z). This study opens a way for future scope regarding aspects like increasing resilience, self-esteem, and perceived social support in various settings such as organizational, educational





Vrinda Antapurkar and Vaishali Choudhari

and therapeutic. The research paves the way for getting to know the generational trends present in both generations and enhances our way of getting to know them.

Limitations

- The technical challenges of data collection would have been eliminated if the data was collected in person and face to face and probably a larger sample would have been collected.
- Qualitative responses about every individual from each generational cohort should have been collected to know more about the reasoning behind the answers they chose and it would have given an insight into the things they face because of belonging to that specific cohort.
- More demographic details of the participants should have been collected to carry out further statistical analyses and to get to know more about the characteristics of the population.

REFERENCES

1. Ang, W., Shorey, S., Lopez, V. et al. Generation Z undergraduate students' resilience during the COVID-19 pandemic: a qualitative study. *Current Psychology* 41, 8132–8146 (2022). <https://doi.org/10.1007/s12144-021-01830-4>
2. Angeles, I. T., & Perkins, R. G. (2023). Intergenerational differences on resilience, sense of coherence, and self-efficacy coping of family during COVID-19 crisis: a salutogenic perspective. *Current Psychology*, 1-13.
3. Awang, M. M., Kutty, F. M., & Ahmad, A. R. (2014). Perceived social support and well being: First-Year student experience in university. *International Education Studies*, 7(13). <https://doi.org/10.5539/ies.v7n13p261>
4. Aydogmus, C. (2019). Generation Z: The Mediating Roles of Self-Esteem and Perceived Stress on the Relationship between Emotional Intelligence and Life Satisfaction. In *Cross-Cultural Business Conference 2019* (p. 299).
5. Baumeister, R. F.; Campbell, J. D.; Krueger, J. I.; Vohs, K. D. (2003). "Does High Self-Esteem Cause Better Performance, Interpersonal Success, Happiness, or Healthier Lifestyles?". *Psychological Science in the Public Interest*. 4 (1): 1–44. doi:10.1111/1529-1006.01431. ISSN 1529-1006. PMID 26151640.
6. Bee, M. A. (2017). The relationship between parenting styles and self-esteem for successive generations.
7. Błachnio, A., Przepiórka, A., & Pantić, I. (2016). Association between Facebook addiction, self-esteem and life satisfaction: A cross-sectional study. *Computers in Human Behavior (Print)*, 55, 701–705. <https://doi.org/10.1016/j.chb.2015.10.026>
8. Bonanno GA (2004). "Loss, Trauma, and Human Resilience: Have We Underestimated the Human Capacity to Thrive After Extremely Aversive Events?". *American Psychologist*. 59 (1): 20–28. doi:10.1037/0003-066x.59.1.20. ISSN 1935-990X. PMID 14736317. S2CID 6296189.
9. Bronfenbrenner, U. (1979). *The ecology of human development: Experiments by nature and design*. Cambridge, MA: Harvard University Press.
10. Brooks R, Goldstein S (13 January 2022). "Resilience: A Process and a Mindset—ADD Resource Center".
11. Campbell-Sills, L., Forde, D. R., & Stein, M. B. (2009). Demographic and childhood environmental predictors of resilience in a community sample. *Journal of psychiatric research*, 43(12), 1007–1012. <https://doi.org/10.1016/j.jpsychires.2009.01.013>
12. Chen, R., Simon, M. A., Chang, E. S., Zhen, Y., & Dong, X. (2014). The perception of social support among U.S. Chinese older adults: findings from the PINE Study. *Journal of aging and health*, 26(7), 1137–1154. <https://doi.org/10.1177/0898264314529332>
13. Chen, R., Simon, M. A., Chang, E. S., Zhen, Y., & Dong, X. (2014). The perception of social support among US Chinese older adults: findings from the PINE Study. *Journal of aging and health*, 26(7), 1137–1154.
14. Cohen S. (2004). Social relationships and health. *The American psychologist*, 59(8), 676–684. <https://doi.org/10.1037/0003-066X.59.8.676>
15. Conger, R. D., & Conger, K. J. (2002). Resilience in Midwestern families: Selected findings from the first decade of a prospective, longitudinal study. *Journal of Marriage and Family*, 64(2), 361–373. <https://doi.org/10.1111/j.1741-3737.2002.00361.x>



**Vrinda Antapurkar and Vaishali Choudhari**

16. Connor KM, Davidson JRT. Development of a new resilience scale: The Connor-Davidson Resilience Scale (CD-RISC) Depression and Anxiety. 2003;18:76–82.
17. Coopersmith, S. (1965). *The antecedents of self-esteem*. Princeton.
18. Coventry, W. L., Gillespie, N. A., Heath, A. C., & Martin, N. G. (2004). Perceived social support in a large community sample: Age and sex differences. *Social psychiatry and psychiatric epidemiology*, 39, 625-636.
19. Das, D. (2024). Exploring the Role of Perceived Social Support on Subjective Well-Being Among Youth in India. *Global Social Welfare*, 1-10.
20. Day, A. L., & Livingstone, H. A. (2003). Gender differences in perceptions of stressors and utilization of social support among university students. *Canadian Journal of Behavioural Science / Revue canadienne des sciences du comportement*, 35(2), 73–83. <https://doi.org/10.1037/h0087190>
21. Djedović, V., Mujkanović, E., & Mirković-Hajduković, M. (2021). Post-millennials: Meet Generation Z. *Društvene i humanističke studije*, 6(1 (14)), 389-406.
22. Duncan, L. E., & Agronick, G. S. (1995). The intersection of life stage and social events: Personality and life outcomes. *Journal of Personality and Social Psychology*, 69(3), 558–568. <https://doi.org/10.1037/0022-3514.69.3.558>
23. Elder, G. H., Jr. (1999). *Children of the Great Depression: Social change in life experience (25th anniversary ed.)*. Westview Press.
24. Elliott, G. C. (1986). Self-Esteem and Self-Consistency: A Theoretical and Empirical Link Between Two Primary Motivations. *Social Psychology Quarterly*, 49(3), 207–218. <https://doi.org/10.2307/2786803>
25. Ermolova, T., Litvinov, A.V., Savitskaya, N.V., & Logvinova, O. (2020). Priorities of psychological and educational work with generation Z (foreign experience).
26. Feather, N. T., Woodyatt, L., & McKee, I. (2011). Predicting support for social action: How values, justice-related variables, discrete emotions, and outcome expectations influence support for the Stolen Generations. *Motivation and Emotion*, 36(4), 516–528. <https://doi.org/10.1007/s11031-011-9262-5>
27. Fletcher D, Sarkar M (2013-01-01). "Psychological Resilience". *European Psychologist*. 18 (1): 12–23. doi:10.1027/1016-9040/a000124. ISSN 1016-9040.
28. Franczok-Kuczumska, A., & Superson, B. (2023). Resilience in the Context of The Socio-Political Situation of Young Adults in Europe. *Humanities and Social Sciences*, 30(1), 29-40.
29. Friends and lifesavers: How social capital and social support received in media environments contribute to well-being. (n.d.). <https://psycnet.apa.org/record/2016-19756-030>
30. Gartland, D., Riggs, E., Muyeen, S., Giallo, R., Afifi, T. O., MacMillan, H. L., Herrman, H., Bulford, E., & Brown, S. (2019). What factors are associated with resilient outcomes in children exposed to social adversity? A systematic review. *BMJ Open*, 9(4), e024870. <https://doi.org/10.1136/bmjopen-2018-024870>
31. Glozah, F.N. (2013) Effects of Academic Stress and Perceived Social Support on the Psychological Wellbeing of Adolescents in Ghana. *Open Journal of Medical Psychology*, 2, 143-150. <http://dx.doi.org/10.4236/ojmp.2013.24022>
32. Haddadi, P., & Besharat, M. A. (2010). Resilience, vulnerability and mental health. *Procedia: Social & Behavioral Sciences*, 5, 639–642. <https://doi.org/10.1016/j.sbspro.2010.07.157>
33. Harari, T. T., Sela, Y., & Bareket-Bojmel, L. (2022). Gen Z during the COVID-19 crisis: a comparative analysis of the differences between Gen Z and Gen X in resilience, values and attitudes. *Current Psychology*. <https://doi.org/10.1007/s12144-022-03501-4>
34. Harikandei, H. R. (2017). Relationship between Perceived Social Support, Mental Health and Life Satisfaction in MSc Students of Physical Education. *International Journal of Sports Science*, 7(4), 159-162.
35. Hathaway, L., & O'Shields, B. (2022). Understanding and engaging Generation Z. *ACSM's Health & Fitness Journal*, 26(4), 39–42. <https://doi.org/10.1249/fit.0000000000000774>
36. Hefner, J., & Eisenberg, D. (2009). Social support and mental health among college students. *American Journal of Orthopsychiatry*, 79(4), 491-499.
37. Henriksen, I. M., Ranøyen, I., Indredavik, M. S., & Stenseng, F. (2017). The role of self-esteem in the development of psychiatric problems: a three-year prospective study in a clinical sample of adolescents. *Child and Adolescent Psychiatry and Mental Health*, 11(1). <https://doi.org/10.1186/s13034-017-0207-y>
38. Herawati, I., Rizal, I., & Amita, N. (2022). The Impact of Social Media on Fear of Missing Out Among Z Generation: A Systematic Literature Review. *Journal of Islamic and Contemporary Psychology*, 2(2), 92-98.





Vrinda Antapurkar and Vaishali Choudhari

39. Ibrahim, A. K., Kelly, S. J., Adams, C. E., & Glazebrook, C. (2013). A systematic review of studies of depression prevalence in university students. *Journal of psychiatric research*, 47(3), 391–400. <https://doi.org/10.1016/j.jpsychires.2012.11.015>
40. Jayatissa, K.A. (2023). Generation Z – A New Lifeline: A Systematic Literature Review. *Sri Lanka Journal of Social Sciences and Humanities*.
41. Kalvin, C., Bierman, K. L., & Gatzke-Kopp, L. M. (2016). Emotional reactivity, behavior problems, and social adjustment at school entry in a high-risk sample. *Journal of Abnormal Child Psychology*, 44(8), 1527–1541. <https://doi.org/10.1007/s10802-016-0139-7>
42. Kvintová, J., & Cakirpaloglu, S. D. (2019b). Self Esteem, Social Network Use and Life Satisfaction among College students of Generation X Y, and Z. *ICERI Proceedings*. <https://doi.org/10.21125/iceri.2019.1324>
43. Lazarus, R. S., & Folkman, S. (1984). *Stress, appraisal, and coping*. New York: Springer Publishing Company.
44. Lee, C. Y. S., & Goldstein, S. E. (2015). Loneliness, stress, and social support in young adulthood: Does the source of support matter? *Journal of Youth and Adolescence*, 45(3), 568–580. <https://doi.org/10.1007/s10964-015-0395-9>
45. Leslie, B., Anderson, C., Bickham, C., Horman, J., Overly, A., Gentry, C., Callahan, C., & King, J. (2021). Generation Z perceptions of a positive workplace environment. *Employee Responsibilities and Rights Journal*, 33(3), 171–187. <https://doi.org/10.1007/s10672-021-09366-2>
46. Lev, T. (2022). Who is Afraid of Generation Z? The Intergenerational Gap Affecting the World of Work Post-COVID-19 Era. *Logos Universality Mentality Education Novelty: Social Sciences*.
47. Luecken, L., & Gress-Smith, J. L. (2010). *Early adversity and resilience in emerging adulthood*. ResearchGate.
48. Luong, G., Charles, S. T., & Fingerhman, K. L. (2011). Better with age: Social relationships across adulthood. *Journal of social and personal relationships*, 28(1), 9–23.
49. Lynch, T. R., Mendelson, T., Robins, C. J., Krishnan, K. R. R., George, L. K., Johnson, C. S., & Blazer, D. G. (1999). Perceived social support among depressed elderly, middle-aged, and young-adult samples: cross-sectional and longitudinal analyses. *Journal of affective disorders*, 55(2-3), 159–170.
50. Lyons, S., Urlick, M. J., Kuron, L., & Schweitzer, L. (2015). Generational differences in the workplace: There is complexity beyond the stereotypes. *Industrial and Organizational Psychology (Print)*, 8(3), 346–356. <https://doi.org/10.1017/iop.2015.48>
51. Maddi, S. R. (2007). *Hardiness: The courage to grow from stressful life events*. New York: Springer.
52. Mahapatra, G.P., Bhullar, N., & Gupta, P. (2022). Gen Z: An Emerging Phenomenon. *NHRD Network Journal*, 15, 246 - 256.
53. Maioli, E. (2016). [Keynote Talk]: New Generations and Employment: An Exploratory Study about Tensions between the Psycho-Social Characteristics of the Generation Z and Expectations and Actions of Organizational Structures Related with Employment (CABA, 2016). *World Academy of Science, Engineering and Technology, International Journal of Humanities and Social Sciences*, 3.
54. Maldonado, L., Huang, Y., Ren, C., Kasen, S., Cohen, P., & Chen, H. (2013). Impact of early adolescent anxiety disorders on Self-Esteem Development from adolescence to young adulthood. *Journal of Adolescent Health*, 53(2), 287–292. <https://doi.org/10.1016/j.jadohealth.2013.02.025>
55. Mannheim, Karl (1952). "The Problem of Generations". In *Kecskemeti, Paul (ed.). Essays on the Sociology of Knowledge: Collected Works, Volume 5*. New York: Routledge. pp. 276–322.
56. Maslow, A. H. (1987). *Motivation and Personality (3rd ed.)*. New York: Harper & Row. ISBN 978-0060419875.
57. Masten, A. S., & Reed, M. J. (2002). *Resilience in development*. New York: Cambridge University Press.
58. McGonigal K (2016-05-10). *The Upside of Stress: Why Stress Is Good for You, and How to Get Good at It*. Penguin. ISBN 978-1-101-98293-8.
59. McIver, S. (2001). Public perceptions. The generation claim. *The Health service journal*, 111 5739, 24-6 .
60. McKee-Ryan, F.M. (2021). Coming of Age in a Global Pandemic: HRM Perspectives on Generation Z's Workforce Entry. *Research in Personnel and Human Resources Management*.
61. Meehan, M. P., Durlak, J. A., & Bryant, F. B. (1993). The relationship of social support to perceived control and subjective mental health in adolescents. *Journal of Community Psychology*, 21(1), 49–55. [https://doi.org/10.1002/1520-6629\(199301\)21:1<49::AID-JCOP2290210106>3.0.CO;2-I](https://doi.org/10.1002/1520-6629(199301)21:1<49::AID-JCOP2290210106>3.0.CO;2-I)





Vrinda Antapurkar and Vaishali Choudhari

62. Mensik, J.S. (2007). A view on generational differences from a Generation X leader. *The Journal of nursing administration*, 37 11, 483-4.
63. Milner, A., Krnjacki, L., &LaMontagne, A. D. (2016). Age and gender differences in the influence of social support on mental health: a longitudinal fixed-effects analysis using 13 annual waves of the HILDA cohort. *Public Health*, 140, 172-178.
64. Mishra, N., Sharma, H., &Garg, R. Generational Contrasts: A Comparative Analysis of Resilience, Interpersonal Communication, and Life Values in Gen X and Gen Z.
65. Naseri, L., Mohamadi, J., Sayehmiri, K., &Azizpoor, Y. (2015). Perceived social support, Self-Esteem, and internet addiction among students of Al-Zahra University, Tehran, Iran. *Iranian Journal of Psychiatry and Behavioral Sciences*, 9(3). <https://doi.org/10.17795/ijpbs-421>
66. Nautiyal, R., Velayudhan, A., &Gayatridevi, S. (2017). Perceived Social Support of the Adolescents from Rural and Urban Setting. *International Journal of Indian Psychology*, 4(2). <https://doi.org/10.25215/0402.098>
67. Novkowska, B., &Serafimovic, G. (2018). Recognizing the vulnerability of Generation Z to economic and social risks. *UTMS Journal of Economics*, 9, 29-37.
68. Olsen, J. M.; Breckler, S. J.; Wiggins, E. C. (2008). *Social Psychology Alive (First Canadian ed.)*. Toronto: Thomson Nelson. ISBN 978-0176224523.
69. Orth U.; Robins R.W. (2014). "The development of self-esteem". *Current Directions in Psychological Science*. 23 (5): 381–387. doi:10.1177/0963721414547414. S2CID 38796272.
70. Oshio, T., Kimura, H., Nishizaki, T., & Omori, T. (2020). Association between the use of social networking sites, perceived social support, and life satisfaction: Evidence from a population-based survey in Japan. *PLOS ONE*, 15(12), e0244199. <https://doi.org/10.1371/journal.pone.0244199>
71. Pishchik, V.I. (2020). Features of the mentality of generations X, Y, Z. E3S Web of Conferences.
72. Poudel, K., &Subedi, P. (2020). Impact of COVID-19 pandemic on socioeconomic and mental health aspects in Nepal. *International Journal of Social Psychiatry*, 66(8), 748–755. <https://doi.org/10.1177/0020764020942247>
73. Puchkova, E.B., Sukhovershina, Y.V., &Temnova, L.V. (2017). A study of Generation Z's involvement in virtual reality. *Psychology in Russia*, 10, 134-143.
74. Regin, K. J., Gadecka, W., Kowalski, P., Kowalski, I. M., &Gałkowski, T. (2016). *Generational transfer of psychological resilience. RocznikMedyczny/Polish Annals of Medicine*, 23(2), 102–107. <https://doi.org/10.1016/j.poamed.2016.02.001>
75. Reinikainen, H., Kari, J. T., &Luoma-aho, V. (2020). Generation Z and organizational listening on social media. *Media and Communication*, 8(2), 185–196. <https://doi.org/10.17645/mac.v8i2.2772>
76. Resilience. (n.d.). <https://www.apa.org>. <https://www.apa.org/topics/resilience>
77. Richardson, G. E. (2002). The metatheory of resilience and resiliency. *Journal of Clinical Psychology*, 58(3), 307–321.
78. Rola, F., Daulay, D. A., &Siregar, A. R. (2018). Emphatic values and self-efficacy of Z generation. *Proceedings of the International Conference of Science, Technology, Engineering, Environmental and Ramification Researches*. <https://doi.org/10.5220/0010079714211424>
79. Rosenberg, M. (2006). Rosenberg Self-Esteem Scale (RSE). In *Conceiving the Self*. Basic Books. <https://www.apa.org/obesity-guideline/rosenberg-self-esteem.pdf>
80. Ruzsa, R.C. (2018). „Z” generációfőjellemzőies a várható munkerő-piacikihívások = Main characteristics of generation Z and the challenges to come in the labour market.
81. Rymarz, R. (2006). The Generation In-Between: The Participation of Generation X in Lifelong Learning.
82. Sakashita, M. (2020). Generation Z in Japan: Raised in Anxiety.
83. Sethi, A. (2023). The Role of Perceived Social Support in Self Esteem and Resilience among Young Adults. *International Journal of Indian Psychology*, 11(1).
84. Smith, K. (2023). "We're Not Thriving, But We're Trying." Exploring Undergraduate Students' Resilience During the COVID-19 Pandemic.
85. Smith, T., &Cawthon, T.W. (2015). Generation Z Goes to College. *College Student Affairs Journal*, 35, 101 - 102.
86. Southwick, S. M., Bonanno, G. A., Masten, A. S., Panter-Brick, C., & Yehuda, R. (2014). Resilience definitions, theory, and challenges: interdisciplinary perspectives. *European journal of psychotraumatology*, 5, 10.3402/ejpt.v5.25338. <https://doi.org/10.3402/ejpt.v5.25338>





Vrinda Antapurkar and Vaishali Choudhari

87. Talmon, G. A. (2019). Generation Z: What’s next? *Medical Science Educator*, 29(1), 9–11. <https://doi.org/10.1007/s40670-019-00796-0>.

88. Tolani, K. C., and Patil, K. (2022). A study of self-efficacy and resilience amongst Generation Z in IT sector. *Journal of Emerging Technologies and Innovative Research (JETIR)*, 9(11).

89. Toubes, D. R., Araújo-Vila, N., & de Araújo, A. F. (2020). Young Ideas to Improve Organizational Resilience in Turbulent and Changing Environments. In *Dynamic strategic thinking for improved competitiveness and performance* (pp. 1-27). IGI Global.

90. Tulgan, B. (1996). Common Misconceptions about Generation X. *Cornell Hotel and Restaurant Administration Quarterly*, 37, 46 - 54.

91. Twenge, J. M., & Campbell, W. K. (2001). Age and birth cohort differences in self-esteem: A cross-temporal meta-analysis. *Personality and social psychology review*, 5(4), 321-344.

92. Ungar, M. (Ed.). (2011). *The social ecology of resilience: A handbook of theory and practice*. Springer Science & Business Media.

93. Van Woensel, R. (2018). Generation Z: A generation of low self-esteem (Doctoral dissertation, Master Thesis, Tilburg University).

94. Vinokur, A. D., Schul, Y., &Caplan, R. D. (1987). Determinants of perceived social support: Interpersonal transactions, personal outlook, and transient affective states. *Journal of Personality and Social Psychology*, 53(6), 1137–1145. <https://doi.org/10.1037/0022-3514.53.6.1137>

95. Vogel, E. A., Rose, J. P., Roberts, L. R., &Eckles, K. (2014). Social comparison, social media, and self-esteem. *Psychology of Popular Media Culture*, 3(4), 206–222. <https://doi.org/10.1037/ppm0000047>

96. VonSoest, T., Wichstrøm, L., &Kvalem, I. L. (2016). The development of global and domain-specific self-esteem from age 13 to 31. *Journal of Personality and Social Psychology*, 110(4), 592–608. <https://doi.org/10.1037/pspp0000060>

97. Werner EE (1989). *Vulnerable but invincible: a longitudinal study of resilient children and youth*. New York: McGraw-Hill. ISBN 978-0-937431-03-0.

98. Wickman, S. A., & Campbell, C. G. (2003). An analysis of how Carl Rogers enacted Client-Centered conversation with Gloria. *Journal of Counseling and Development*, 81(2), 178–184. <https://doi.org/10.1002/j.1556-6678.2003.tb00239.x>

Table 1. Showing Mean, SD and t-value

Variable	Generational Cohort	N	Mean	Std. Deviation	t-value
Self-esteem	Gen Z	86	28.74	3.717	4.325*
	Gen X	86	31.36	4.360	

*p<0.05

Table 2.: Showing Mean, SD and t-value

Variable	Generational Cohort	N	Mean	Std. Deviation	t-value
Perceived Social Support	Gen Z	86	5.13	1.335	3.326*
	Gen X	86	5.71	0.919	

*p<0.05

Table 3.: Showing Mean, SD and t-value

Variable	Generational Cohort	N	Mean	Std. Deviation	t-value
Resilience	Gen Z	86	69.74	12.217	3.176*
	Gen X	86	75.74	11.394	

*p<0.05





Impact Assessment of Static Waterbodies in Peri-Urban Areas: The Case of Kakori Town, Lucknow

Arundhatee Mishra^{1*}, Joydeep Dutta², Indrani Chakraborty³ and Subhrajit Banerjee³

¹Assistant Professor, Department of Architecture, Faculty of Architecture & Planning, (Affiliated to Dr. APJ Abdul Kalam Technical University), Lucknow, Uttar Pradesh, India.

²Adjunct Professor, Department of Architecture, Faculty of Architecture & Planning, (Affiliated to Dr. APJ Abdul Kalam Technical University), Lucknow, Uttar Pradesh, India.

³Professor, Department of Architecture, Faculty of Architecture & Planning, (Affiliated to Dr. APJ Abdul Kalam Technical University), Lucknow, Uttar Pradesh, India.

Received: 15 Jun 2024

Revised: 05 Sep 2024

Accepted: 04 Nov 2024

*Address for Correspondence

Arundhatee Mishra

¹Assistant Professor,

Department of Architecture, Faculty of Architecture & Planning,
(Affiliated to Dr. APJ Abdul Kalam Technical University),

Lucknow, Uttar Pradesh, India

E.Mail: mishraaru15@gmail.com



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

Peri-urban ecosystems and societies depend on static waterbodies. Static water bodies in Lucknow's peri-urban area, particularly Kakori Town, are examined in this study. The study uses field surveys, remote sensing, and socio-economic studies to investigate the biological and hydrological dynamics of Kakori Town's static waterbodies. This study analyses the lake based on the identified parameters (Landuse and Population density, Encroachment, community use, pollution, community participation, excavation activity, recreational or tourism activities and institutional role). It also examines natural and anthropogenic influences on static waterbodies. The research identifies stressors to inform sustainable management techniques and policy measures to mitigate their negative effects and strengthen these vital ecosystems. Policymakers, urban planners, and local communities may use these insights to safeguard these crucial natural resources and promote sustainable development in peri-urban areas like Kakori Town.

Keywords: Impact assessment, Static water bodies, Lucknow, Peri-urban areas, Kakori town

INTRODUCTION

Urbanization has been accelerating globally, particularly in developing nations, and this trend is expected to continue. An estimated 600 million people have joined the global urban population in the past decade, accounting for about two-thirds of the total population growth (Ritchie et al., 2024). Currently, there are twenty-one megacities worldwide, each with populations exceeding ten million, with seventeen located in developing nations (UN-





Arundhatee Mishra et al.,

HABITAT, 2020). In India, urbanization accelerated post-independence due to the shift to a mixed economy, fostering private sector growth. According to the World Bank, the urban population in India increased from 11.4% in the 1901 census to 28.53% in the 2001 census and reached 34% by 2017. A United Nations survey projects that by 2030, 40.76% of India's population will be urban (Bhagat, 2018). By 2050, India, along with China, Indonesia, Nigeria, and the United States, will lead the global urban population growth (World Bank, 2011; UN, n.d.).

Rapid urban expansion in India has led to the depletion of static water bodies due to pollution, waste disposal, and urban encroachment. Consequently, urban areas face groundwater depletion, flooding, and water scarcity, adversely affecting quality of life and access to essential services (Beura, 2017). To address these issues, water security must be a national priority, requiring efficient management and stringent regulations, along with public participation in water conservation (Mundoli et al., 2015). In Lucknow, the number of static water bodies has decreased from 964 in 1952 to 494 in 2006, due to urbanization. The water quality upstream has deteriorated under the pressures of rapid urbanization, population growth, and urban sprawl (Prabha Baiswar et al., 2022). The Gomti River, fed by groundwater and its tributaries, has seen a 35-40% reduction in discharge, becoming one of the most polluted rivers in the country. Factors such as biotic pressure, reduced ecological flow, siltation, encroachment, and heavy metal contamination have contributed to this degradation, threatening the sustainability of Lucknow's water resources (Rai & Singh, n.d.). The peri-urban area of Lucknow, characterized by a mix of rural and urban features, faces challenges related to land use and infrastructure development due to economic growth and demographic pressure (Rawat et al., 2022; Mishra et al., 2024).

MATERIALS AND METHODS

Research Method

The objective of this research article is to examine the effects of urbanisation on static water bodies in peri-urban regions of Lucknow, specifically those urban areas situated between the 2021 and 2031 planning boundaries as delineated in the Lucknow Master Plan 2031. The criteria for selecting static water bodies for analysis purposes are as follows: they must have a minimum surface area of 1 acre and be located within 10 metres of the urban settlement. Static bodies of water within or in close proximity to urban settlements at an altitude of 10 metres have been chosen for the purpose of analysis so that a reconnaissance survey and satellite image can provide a more comprehensive understanding of the water body's local context. The static body of water was additionally required to have a minimum surface area of 1 acre as a secondary criterion. Hence, within the vicinity bounded by the planning boundaries of 2021 and 2031, Kakoritown of Lucknow district stands as the sole remaining settlement worthy of examination, comprising a collection of five static water bodies chosen for analysis. The research utilised cartography based on Geographic Information Systems (GIS) to identify the factors that contribute to the depletion of static water bodies situated within or in close proximity to the boundaries of the KakoriTown. The lake boundaries were established through the utilisation of Google Imagery data pertaining to the years 2011 and 2023, correspondingly. Subsequently, data has been acquired predominantly from a reconnaissance survey concerning the land utilisation in close proximity to the subject site, with a proximity of 100 metres. Following this, the KakoriTownLanduse map was created. Also, the areas of traced static water bodies for the years 2011 and 2023 were computed using ArcGIS. The application of this technique enabled a direct comparison and computation of the depletion state of the static water bodies. Additionally, the aetiology of these depletions has garnered considerable attention.

Then, a comprehensive assessment has been conducted on a lake-by-lake basis on the identified parameters (Mishra et al., 2024). Those identified parameters were Land Use and Population Density, Encroachment, Community Use, Pollution, Community Participation, Recreational or Tourism Activities, and Institutional Role respectively. User perception analysis has been done for all the individual 5 lakes and the scoring were done by the locals around the lake, from 1 to 5 (1 is the least concern 5 is the major concern). The sample size of the user perception is 30. Then the identified parameters were assigned weightages by using AHP , there after normalization was done for the accuracy,





Arundhatee Mishra et al.,

and hence ranking is devoted to each of the parameters for individual lakes. (1 is the major concern 7 is the least concern).

Site Selection

As the most populous state in India and the capital of Uttar Pradesh, Lucknow is representative of the majority of Indian metropolises. Lucknow's growth trajectory remained predominantly organic from 1981 to 1991. However, beginning in 1991, the pattern abruptly shifted towards ribbon development, which benefited from accessibility and transit-oriented development. The peri-urban and rural regions encircle the city as a result of the city's burgeoning urban population and high migration rate. The urbanised region is presently en-route to Amausi Airport in the southeastern direction, via the Lucknow-Kanpur Highway, Lucknow Raebareli Road, and Lucknow Hardoi Road (Kumari, 2015). The temporal and spatial dimensions of the land use have changed significantly. Population growth has been consistent in the city due to natural expansion, large-scale migration, and the incorporation of peri-urban regions (TCPO, 2020).

From 1901 to 2011, the population of Lucknow Urban Agglomeration (UA) increased from 2.5 lakh to 28.17 lakh. According to the predictions outlined in the Lucknow Master Plan 2031, the peri-urban regions of Lucknow are anticipated to experience a decadal growth rate of 117.55 percent in 2031, up from 24.35% in 2011 and 27.71 percent in 2021 (DoCO UP, 2011). Within the 2031 planning boundary of Lucknow, the number of lakes and ponds decreased from 86 in 2011 to 78 in 2023. Table 2 indicates that urbanization has had a more significant impact on peri-urban areas, where the number of water bodies declined from 66 to 59 between 2011 and 2023. In comparison, the Lucknow Municipal Corporation (LMC) areas experienced a decrease from 20 to 19 water bodies. Furthermore, the total area of water bodies in peri-urban areas decreased by approximately 11.12%, whereas in the LMC areas, it decreased by 7.15%. This suggests that water bodies in peri-urban areas are more vulnerable to the effects of urbanization compared to those within the city areas, which receive more focused preservation efforts.

Impact of Urbanisation on Static Water Bodies in peri-urban areas of Lucknow

Many of the lakes that comprised the capital of Awadh in 1900 have been converted into human settlements over the course of the last century. Based on a survey carried out by the Lucknow Municipal Corporation, the quantity of reservoirs in the city experienced a decline from 964 in 1952 to 494 in 2006. The quantity experienced a decline to 494 in 2006, with reclamation rendering the majority of them unidentifiable (IndiaTV, 2019; Siddiqui, 2018; Verma, 2016). Additionally, it has been noted that the majority of bodies of water are clogged with solid refuse, debris, and sewage, among other substances, which diminishes their carrying capacity. Between 2011 and 2023, the number of lakes in the peri-urban regions of Lucknow decreased from 66 to 59, representing a significant reduction of 11.12% in area (Goel et al., 2018).

Assessment of the Static Water Bodies in the Kakori Town

The purpose of this research article is to investigate the effects of urbanization on static water bodies in the peri-urban regions of Lucknow, specifically those urban areas situated between the 2021 and 2031 planning boundaries as delineated in the Lucknow Master Plan 2031. The criteria for selecting static water bodies for analysis are as follows: they must have a minimum surface area of 1 acre and be located within 10 meters of urban settlements and Kakori town fulfils all the requirements for selection. Kakori town is located in the Lucknow district of Uttar Pradesh in India. It is a peri-urban area, meaning it is on the outskirts of the city and has a mix of urban and rural characteristics. The town is known for its historical significance and is home to various industries and agricultural activities. Kakori was declared as town in 2001 (Team Digital, 2014). The total area of Kakori town is 110.4 hectare. The Kakori Nagar Panchayat is a local governing body in the Kakori town in Lucknow, Uttar Pradesh (Maps, Weather, and Airports for Kakori, India, n.d.; TOI, 2011). It is tasked with the provision of fundamental infrastructure and services to the town's inhabitants. Given that the population density of Kakori town is considerably greater than that of Lucknow Municipal Corporation, evaluating the state of static water bodies in Kakori town becomes more justifiable (DoCO UP, 2001).



**Arundhatee Mishra et al.,**

RESULTS

As stated previously, five selected static water bodies of Kakori town have been analysed based on those seven parameters mentioned earlier. They are listed in Table 4. Lake wise depleted area value and their major causes for their depletion are being analysed as follows:

Salariya Lake

The lake is in ward no. 8 of Kakori Town and is surrounded by two wards i.e. ward no. 11 and ward no. 2. The entire lake is entitled to lease from last 25-30 years. The total area of this lake is 2.1 acre.

Land Use and Population Density

The Lake is surrounded with residential areas, vacant land, public/semi-public area (primary health centre) and agricultural land. Ward no. 8 is 1.5 times (127PPH), Ward No. 2 is almost 4 times (326PPH), whereas ward no. 11 is 2 times denser (168PPH) than the Lucknow Municipal Corporation (LMC) population density (85PPH) in 2011.

Encroachment

The lake underwent a reduction in area over a ten-year period, from 2.57 acres in 2011 to 2.1 acres in 2023, an equivalent decrease of 0.47 acres. The constructed area surrounding the water body, with a 10-meter buffer zone, is 0.53 acres. Open space is 1.28 acres on the periphery (10-meter buffer zone around the body of water). In contrast, the ratio of built-up to open space in the vicinity of an aquatic body (10-meter buffer zone) is 1:2.41. At a distance of 100 metres from the lake's shoreline, the developed region encompasses 6.52 acres. The extent of open space within the lake's influence area, which is 100 metres from the lake's shoreline, is 12.36 acres. The open space ratio in the lake's influence area, which is 100 metres from the lake's shore, is 1:1.89.

Community Use

The poor water quality caused by the excessive sewage discharge into the lake discourages any community use of the lake during the summer, with the exception of commercial fishing and baking earthen wares. Seasonal commercial fishing is conducted for five to six months annually. The remaining socio-economic activities and community uses, including personal hygiene, clothing cleansing, idol immersion, angling, irrigation, animal bathing, cremation, and the Mundan ceremony, are not being observed.

Pollution

The pollution in the vicinity of the lake has a detrimental effect on the health of the lake's inhabitants and has also degraded the water quality. The sources of pollution in the vicinity of the lake include untreated domestic sewage discharge, scat, and the dumping of municipal solid refuse along the lake's shoreline. Aside from these, certain types of waste are not being discovered along the lake's shoreline, including agricultural residue, immersion of religious or cultural refuse, carcass formation, and household industrial waste.

Community Participation

The competent local government entity tasked with lake maintenance is well-known to the community. Locals are unaware of any government policies or initiatives pertaining to the lake's restoration or conservation. Local governments and non-governmental organisations had never before organised an awareness campaign to encourage the public to protect and sustain the lake and its environs. Locals are interested in contributing to the lake's maintenance, but they are unable to do so without formal assistance.

Recreational/Tourism Activities

The lake's vicinity is devoid of consistent or sporadic recreational or tourism activities due to the pollution that permeates the area. Occasional tourism and recreational activities around the lake are favoured by the local populace, provided that the lake is maintained by the local government.



**Arundhatee Mishra et al.,****Institutional Role**

The maintenance and cleansing responsibilities have been neglected for over two decades by the governing body. There is no policy implementation on the lake. The area lacks a formal encroachment inspection. There are no socioeconomic, cultural, recreational, or tourism-related activities occurring in the vicinity of the lake. There has never been an awareness campaign regarding the lake's protection. The KakoriTown undertakes the task of cleaning the lake's periphery every two to three years.

Sagar Lake

The lake is situated within ward no. 7 and is enclosed by ward no. 3 and ward no. 11. Sagar Lake, with an expansive expanse of 8.66 acres, ranks as the second largest lake within the KakoriTown.

Land Use and Population Density

Residential areas and vacant land border the lake on the south and north, respectively, while agricultural areas border the lake on the east and west. In comparison to the LMC population density of 85PPH in 2011, Ward No. 7 is nearly 1.4 times (118PPH) denser, Ward No. 3 is 1.5 times (126PPH), and Ward No. 11 is 2 times (168PPH).

Encroachment

A decade of reduction resulted in the lake's total area decreasing from 9.51 acres in 2011 to 8.66 acres in 2023, a decrease of 0.85 acres. The built-up area surrounding the aquatic body, with a 10-meter buffer zone, is 0.33 acres. Open space is 2.15 acres on the periphery (10-meter buffer zone around the body of water). The ratio of built-up to open space in the 10m buffer zone surrounding the body of water is 1:6.5. 8.16 acres of built-up area is located in the influence area, which is 100 metres from the lake's shore. 33.2 acres are designated as open space in the lake's influence area, which is 100 metres from the lake's shore. The ratio of built-up to non-built-up areas within the lake's influence area, measured 100 metres from the lake's shoreline, is 1:4.07.

Community Use

Due to the substantial sewage discharge into the lake, which worsens the water quality, the lake's seasonal commercial fishing is the only community use that is preferred by the residents. This lake has been granted on lease in two portions. Seasonal commercial fishing is conducted for five to six months annually. Animal grazing occurs frequently in the vicinity of the lake's periphery. The remaining activities, including personal laundry, angling, irrigation, animal bathing, earthen ware baking, and other religious practices, are not being observed.

Pollution

The pollution in the vicinity of the lake has a detrimental effect on the health of the lake's inhabitants and has also degraded the water quality. Untreated domestic sewage discharge, scat, and municipal solid refuse dumping along the lake's shoreline are the primary sources of pollution. Specific categories of waste, such as agricultural residue, religious or cultural refuse immersion, carcass formation, and household industry waste dumping along the lakeshore, are not being detected.

Community Participation

The local authority responsible for lake maintenance is recognized by the community. However, residents are unaware of any policies or initiatives by the government for the lake's restoration or conservation. There has been no previous effort by local governments or NGOs to raise public awareness about protecting and sustaining the lake and its surroundings. Although locals are willing to help maintain the lake, they need official support to do so.

Recreational /Tourism Activities

There are no regular or occasional recreational/ tourism activities around the lake, because of the pollution in and around the lake. Occasional tourism and recreational activities around the lake are favoured by the local populace, provided that the lake is maintained by the local government.



**Arundhatee Mishra et al.,****Institutional Role**

The lake is cleaned by the lessee once every two to three years. The local authority has not performed any cleansing or maintenance in over three decades. Failure to enforce policies regarding the lake, there is an absence of formal encroachment checks in the vicinity, and mining operations do not occur around the lake's perimeter. There are no socioeconomic or cultural activities, recreational or tourism-related activities, or awareness campaigns taking place in the vicinity of the lake. Moreover, there has never been an effort to preserve the lake. The KakoriTown undertakes the task of cleaning the lake's periphery every three to four years.

Chaudhary Mulaka Lake

The lake is situated in the third ward and is enclosed by the seventh and fourth wards. The lake as a whole has been leased for the past 25 to 30 years. Chaudhary Mulaka Lake encompasses a total area of 1.86 acres.

Land Use and Population Density

Residential areas, vacant land, and agricultural land encircle the lake. Ward number three is 1.5 times (126PPH) denser than the LMC population density of 85PPH in 2011. Ward number seven is nearly 1.4 times (118PPH) denser, while ward number four is 2.4 times (194PPH) denser.

Encroachment

With a reduction of 0.47 acres over a decade, the lake's total area shrunk from 2.57 acres in 2011 to 2.1 acres in 2023. The constructed area surrounding the water body, with a 10-meter buffer zone, is 0.53 acres. Open space is 1.28 acres on the periphery (10-meter buffer zone around the body of water). The ratio of built-up to open space in the 10m buffer zone surrounding the body of water is 1:2.41. At a distance of 100 metres from the lake's shoreline, the developed region encompasses 6.52 acres. The extent of open space within the lake's influence area, which is 100 metres from the lake's shoreline, is 12.36 acres. The ratio of built-up to non-built-up areas within the lake's influence area (100 metres from the lake's shoreline) is 1:1.89.

Community Use

Local residents prefer only seasonal commercial fishing and summertime baking of earthen wares on the lake, as the lake's water quality deteriorates due to the heavy effluent discharge into it. Five to six months per year, seasonal commercial fishing is conducted. Irrigation, personal laundry, animal bathing, grazing along the lake's periphery, and religious and cultural practices such as immersion in idols, cremation ceremonies, and Mundan ceremonies are not observed by the community.

Pollution

Untreated Domestic sewage discharge in the lake, Scat, Municipal Solid waste dumping within or in shoreline of lake are regular waste in-or around the lake. The non-regular waste types are Agricultural Residue, Immersion of Religious or Cultural waste, Carcass formation, Household industry waste dumping within or in shoreline of lake.

Community Participation

The accountable local government body tasked with lake maintenance is well-known to the neighbours. Concerning the restoration and conservation of the lake, locals are uninformed of government initiatives and policies. No local governmental organisation or non-governmental organisation had ever conducted an awareness campaign to educate the public about the need to preserve the lake and its environs. The local populace, while enthusiastic about participating in lake maintenance, is unable to do so without official assistance.

Recreational /Tourism Activities

The lake's vicinity is devoid of consistent or sporadic recreational or tourism activities due to the pollution that permeates the area. Occasional tourism and recreational activities around the lake are favoured by the local populace, provided that the lake is maintained by the local government.



**Arundhatee Mishra et al.,****Institutional Role**

Once in 2-3 years Kakori Town takes the effort to clean the periphery of the lake. However, no proper cleaning and maintenance has been ever done from more than 20 years by the authority. No Implementation of policies on lake. The area lacks a formal encroachment inspection. No Socio-economic or socio-cultural activities are happening around the lake. No recreational and tourism activities are happening around the lake. There has never been an awareness campaign regarding the lake's protection. At last, only the lease holder of the lake cleans the lake in 2-3 years through the process of chlorination.

Kunwar Gatta Lake

Two districts encircle the lake, specifically Ward No. 3 and Ward No. 4. The lake in its entirety is leased for a period exceeding thirty years. The lake encompasses a total area of 2.1 acres

Land Use and Population Density

Agricultural land and residential areas encircle the lake. Ward no. 3 is 1.5 times (126 PPH) denser than the LMC population density (85 PPH) in 2011, whereas Ward no. 4 is 2.4 times (194 PPH) denser.

Encroachment

The lake's overall area experienced a decline from 1.56 acres in 2011 to 1.29 acres in 2023, signifying a reduction of 0.27 acres over the course of a decade. 0.59 acres are developed along the perimeter (10-meter buffer zone of the body of water). The perimeter open space (10-meter buffer zone around the body of water) is 0.66 acres. The ratio of built-up to open space in the 10m buffer zone surrounding the body of water is 1:1.12. 3.29 acres of built-up area is located in the influence area, which is 100 metres from the lake's shore. 8.74 acres are designated as open space in the lake's influence area, which is 100 metres from the lake's shore. The ratio of built-up to non-built-up areas within the lake's influence area (100 metres from the lake's shoreline) is 1:2.65.

Community Use

The poor water quality caused by the excessive sewage discharge into the lake discourages any community use of the lake during the summer, with the exception of commercial fishing and baking earthen wares. Seasonal commercial fishing is conducted for five to six months annually. Additionally, grazing is employed on the lake's periphery. Activities such as personal hygiene, fishing, irrigating, bathing animals, baking earthen wares, and engaging in religious or cultural practices like cremation, mundan ceremonies, chat pujas, and idol immersions.

Pollution

Significant contributors to lake pollution include the discharge of untreated domestic sewage, municipal solid refuse dumping within or along the lake's shoreline, and SCAT. The pollution in the vicinity of the lake has a detrimental effect on the well-being of the lake's inhabitants and has exacerbated the condition of the water. Agricultural residue, religious or cultural refuse immersion, carcass formation, and household industry waste dumping along the lake's shoreline are not identified as sources of pollution.

Community Participation

The entity responsible for lake maintenance is well-known within the community. Despite this, locals are not informed about any government efforts or programs aimed at restoring or conserving the lake. There have been no awareness campaigns organized by local governments or NGOs to encourage public involvement in protecting the lake and its surroundings. The community is interested in contributing to the lake's upkeep but needs formal support to do so.

Recreational /Tourism Activities

Occasional or regular tourism or recreational activities are prohibited in the lake's vicinity due to the pollution that permeates the area. Occasional tourism and recreational activities around the lake are supported by the local populace, provided that the lake is maintained by the local government.



**Arundhatee Mishra et al.,****Institutional Role**

There have been no maintenance activities conducted for over three decades. There is no policy implementation on the lake. There is no mechanism in place to prevent encroachment in the vicinity. There are no socioeconomic or cultural activities occurring in the vicinity of the lake. There are no tourism or recreational activities occurring in the vicinity of the lake. There has never been an awareness campaign regarding the lake's protection. The lagoon has never been cleaned by a local government agency. The Kakori Nagar Panchayat undertakes the task of cleaning the lake's periphery every three to four years. The lease holder is obligated to remove all contaminants from the lake every two to three years via chlorination.

Hauda Lake

The lake is enclosed within three districts, namely districts 1, 6, and 9. Hauda lake, the largest in Kakori, has been enlarging its perimeter for the past decade. The lake level has risen as a result of the discharge of untreated sewage water from the adjacent residential areas and the consumption of agricultural land situated in its vicinity due to the low-lying terrain.

Land Use and Population Density

The population densities of Ward No. 1 (nearly threefold, 269.PPH), Ward No. 6 (twofold, 179 PPH), and Ward No. 9 (1.8 times, 153 PPH) exceed the LMC population density of 85PPH in 2011.

Encroachment

The lake experienced a notable expansion from 9.68 acres in 2011 to 15.6 acres in 2023. A discrepancy of 5.92 acres exists. The built-up area surrounding the water body, with a 10-meter buffer zone, is 0.16 acres. A perimeter open space area of 17.66 acres (10-meter buffer zone around the body of water). The ratio of built-up to open space in the 10m buffer zone surrounding the body of water is 1:110.37. Influence area (located 100 metres from the lake's shoreline) developed land comprises 18.56 acres. Open space comprises 53.72 acres in the lake's influence area, which is located 100 metres from the lake's shore. The ratio of built-up to non-built-up areas within the lake's influence area (100 metres from the lake's shoreline) is 1:2.89.

Community Use

This lake's proprietorship is delineated into four portions. Seasonal commercial fishing is conducted for five to six months per year, and dairy production occurs consistently. Due to the substantial sewage discharge into the lake, which worsens the water quality, the lake's seasonal commercial fishing is the only community use that is preferred by the residents. Grazing by animals is observed in the vicinity of the lake. Aspects such as personal hygiene practices, fishing, irrigation, animal bathing, earthenware baking, and religious and cultural activities (e.g., idol immersion, mundan ceremony, chat puja), are not observable within or in the vicinity of the lake.

Pollution

Significant pollutant sources include untreated domestic sewage discharge, municipal solid refuse dumping within or along the lake's shoreline, and scat. Non-obtrusive sources of pollution in the vicinity of Hauda Lake include agricultural residue, cultural or religious refuse immersion, carcass formation, and the dumping of household industrial waste along the lake's shoreline. The pollution in and around the lake has a detrimental effect on the health of the lake's inhabitants and has deteriorated the water quality.

Community Participation

The community is aware of the local government body in charge of maintaining the lake. Nonetheless, there is a lack of awareness among residents regarding government policies or initiatives for the lake's restoration and conservation. Neither local governments nor NGOs have conducted awareness campaigns to promote the protection and sustainability of the lake and its environment. While locals are keen to participate in the lake's maintenance, they require formal assistance to proceed.





Arundhatee Mishra *et al.*,

Recreational /Tourism Activities

Occasional or regular tourism or recreational activities are prohibited in the lake's vicinity due to the pollution that permeates the area. Locals are in the favour of the occasional recreational/ tourism activities around the lake if the lake is maintained by the local govt. body.

Institutional Role

There have been no maintenance activities conducted for over three decades. The chlorination process is exclusively employed by the lease proprietor of the lake every two to three years. The Kakori Nagar Panchayat endeavours to conduct perimeter cleaning of the lake every one to two years. There is no policy implementation on the lake. There are no routine encroachment checks in the area. There are no socioeconomic or cultural activities occurring in the vicinity of the lake. There are no tourism or recreational activities occurring in the vicinity of the lake. There has never been an awareness campaign regarding the lake's protection. The lagoon has never before been cleaned by a local government agency.

FINDINGS

Urbanisation in Kakori has undeniably contributed to the depletion of static water bodies, primarily as a result of pollution and institutional negligence (as ranked by user perception in Table 5). As a consequence, the environment, water body boundaries, and the overall quality of life in urban areas have been profoundly affected.

The above-mentioned parameters can be summarised as follows:

1. Pollution: Prominent sources of pollution encompass untreated domestic sewage discharge, disposal of municipal solid waste along the lake's shoreline, and scat. These activities have resulted in water contamination that is unfit for human consumption and other applications. The consequence of this situation is a reduction in the accessibility of potable water for the populace.
2. Institutional Role: Due to the indifference and lack of consciousness on the part of their responsibilities of respective local governing bodies, the lakes are susceptible to environmental degradation.
3. Community Participation: The lake and its environs have been subject to a lack of awareness community engagement, and government assistance, resulting in a lack of interest and support to maintain the lakes.
4. Encroachment: The lakes' surface area has fallen drastically due to unauthorised construction and domestic industrial use.
5. Land Use and Population Density: The lakes' susceptibility to degradation is primarily attributable to the district's high population density and the heterogeneous land use character surrounding the lakes.
6. Community Use: The aforementioned factors will prevent the local community from making a substantial and justifiable use of the lake, consequently resulting in inadequate maintenance.
7. Recreational and Tourism Activities: Given the deplorable state of the Kakori lakes, their utilisation for recreational and tourism purposes is implausible. Restoring the lakes and generating economic revenue from them could be the primary objective in this regard.

To address the issue of static water body depletion in Kakori, it is essential to prioritize the conservation of surface water bodies and implement effective management strategies. This involves the efficient administration of existing aquatic environments through public engagement and the enforcement of stringent regulations to prevent further degradation. Additionally, investments in renewable water resources, the promotion of sustainable urban planning, and the enhancement of waste management systems are crucial in mitigating the adverse effects of urbanization on static water bodies.





Arundhatee Mishra et al.,

REFERENCES

1. Beura, D. (2017). Depletion of Water Bodies due to Urbanisation & its Management. *International Education & Research Journal*, 3(6), 204–205. <https://www.researchgate.net/publication/326539383>
2. Bhagat, R. B. (2018). *Urbanization in India: Trend, Pattern and Policy Issues* (17).
3. Chen, M., Zhang, H., Liu, W., & Zhang, W. (2014). The global pattern of urbanization and economic growth: Evidence from the last three decades. *PLoS ONE*, 9(8). <https://doi.org/10.1371/journal.pone.0103799>
4. DoCO UP. (2001). *District Census Handbook Lucknow 2001*.
5. DoCO UP. (2011). *District Census Handbook Lucknow 2011*.
6. Goel, P., Saxena, A., Singh, D. Sen, & Verma, D. (2018). Impact of rapid urbanization on water quality index in groundwater fed Gomati River, Lucknow, India. *Current Science*, 114(3), 650–654. <https://doi.org/10.18520/cs/v114/i03/650-654>
7. IndiaTV. (2019, July 8). *Lucknow loses almost half its water bodies to builders*. IndiaTV. <https://www.indiatvnews.com/news/india-water-pollution-water-bodies-in-lucknow-land-grabbing-construction-533597>
8. Kumari, K. (2015). Urban Sprawl: A Case Study of Lucknow City. *International Journal of Humanities and Social Science Invention*, 4(5), 11–20. www.ijhssi.org
9. *Maps, Weather, and Airports for Kakori, India*. (n.d.). Retrieved April 10, 2024, from <https://www.fallingrain.com/world/IN/36/Kakori.html>
10. Mishra, A., Dutta, J., & Chakraborty, I. (2024). Pivotal Vulnerability Facets Influencing Static Water Bodies: The Case of Peri-Urban Areas of India. *Indian Journal of Natural Sciences*, 14(82), 67841–67850.
11. Mundoli, S., Manjunath, B., & Nagendra, H. (2015). Effects of urbanisation on the use of lakes as commons in the peri-urban interface of Bengaluru, India. *International Journal of Urban Sustainable Development*, 7(1), 89–108. <https://doi.org/10.1080/19463138.2014.982124>
12. Prabha Baiswar, S., Banerjee, S., & Chakraborty, I. (2022). Demographic Change: A case of Lucknow City. *Journal of Emerging Technologies and Innovative Research*, 9(7), 641–649. www.jetir.org/f641
13. Rai, N., & Singh, A. K. (n.d.). *Strategic Issues in Infrastructure Development: A case study of Lucknow City*.
14. Rawat, A. K., Banerjee, S., Roy, A. K., & Chakraborty, I. (2022). Analysis Of Urban Growth Dynamics of Lucknow City: A Gis Approach. *Journal of Positive School Psychology*, 2022(6), 8303–8308. <http://journalppw.com>
15. Ritchie, H., Samborska, V., & Roser, M. (2024, February). *Urbanization - Our World in Data*. Our World in Data. <https://ourworldindata.org/urbanization>
16. Siddiqui, S. (2018). *Loss of Water Bodies in Lucknow City*. Issuu. https://issuu.com/sumaira.1231/docs/2017.12.21_loss_of_water_body
17. TCPO, M. G. (2020). *Dynamics of Peri-Urban Area: Prospects and Challenges of Sustainable Development-A case study of Lucknow*.
18. Team Digital. (2014, February 16). काकोरी: आखिर किसने कहा लखनपुर को लखनऊ. Amar Ujala. <https://www.amarujala.com/lucknow/history-of-kakori>
19. The World Bank. (2011, August 22). *Urbanization in India*. The World Bank. <https://www.worldbank.org/en/news/feature/2011/09/22/india-urbanization>
20. TOI. (2011, August 16). *Tourist spot tag may uplift Kakori*. The Times of India. https://web.archive.org/web/20110916130548/http://articles.timesofindia.indiatimes.com/2011-08-09/lucknow/29868140_1_kakori-shaheed-smarak-memorial-ram-prasad-bismil
21. UN. (n.d.). *68% of the world population projected to live in urban areas by 2050*. United Nations; United Nations. Retrieved April 10, 2024, from <https://www.un.org/sw/desa/68-world-population-projected-live-urban-areas-2050-says-un>
22. UN-HABITAT. (2020). *The Value of Sustainable Urbanization*.
23. Verma, R. (2016, February 12). *Lucknow's waterbodies perish at the hands of unregulated construction*. Down to Earth. <https://www.downtoearth.org.in/news/urbanisation/lucknow-s-waterbodies-perish-at-the-hands-of-unregulated-construction-52811>.





Arundhatee Mishra et al.,

Table 1 Population of Lucknow from 1991-2031*

Year	Lucknow UA (A)		Lucknow Planning Boundary (B)		Lucknow Municipal Corporation		Lucknow Planning Boundary Excluding LMC*	
	Population	Decadal Growth Rate*	Population	Decadal Growth Rate*	Population	Decadal Growth Rate*	Population	Decadal Growth Rate
1991	1669204	65.66			1619116	70.79		
2001	2245509	34.53	27,73,618	-	21,85,297	35.00	5,88,321	
2011	2880108	28.26	35,48,732	27.94%	28,17,105	28.87	7,31,627	24.35%
2021	3713003	28.92	45,50,326	28.22%	36,15,909	28.36	9,34,417	27.71%
2031	4787488	28.94	65,00,000	42.84%	4467158	23.54	20,32,842	117.55%

Source- Census Tables and Author’s Projection

*Projected Data for 2021 and 2031

Table 2 Status of lakes or ponds bodies in Lucknow

Year	Lakes/ Ponds within the Boundary of 2031			Lakes/ Ponds within LMC Boundary			Lakes/ Pond in Peri-Urban Area		
	Total No.	Area (sq.km.)	% of Total Area increased/ decreased	Total No.	Area (sq.km.)	% of Total Area increased/ decreased	Total No.	Area (sq.km.)	% of Total Area Increased/ Decreased
2011	86	0.82		20	0.28		66	0.54	
2023	78	0.74	-6.1%	19	0.26	-7.15%	59	0.48	-11.12%

Source: Author (Interpreted from satellite image of 2011, 2023)

Note: The mentioned above information excludes the number of wetlands identified by National Water Informatic Centre, Department of Water Resources, River Development and Ganga Rejuvenation, Ministry of Jal Shakti (Govt. of India)

Table 3 Population Density comparison between Kakori town and Lucknow Municipal Area

Town Area	Density (1991)	Density (2001)	Density (2011)
Kakori Nagar Panchayat	123 PPH	153 PPH	176 PPH
Lucknow Municipal Area	49 PPH	67 PPH	85 PPH

Source: Lucknow Master Plan 2031, Census of India (1991, 2001, 2011)

Table 4 GIS based Surface Area calculation of Static Water Bodies in Kakori for 2011 and 2023

S.No.	Identified Lake in Kakori Town	Proximity of lake to urban settlement	Lake area (2011)	Lake area (2023)
1.	Salariya Lake	2.26 m	2.57acres	2.10 acres
2.	Sagar Lake	6.21 m	9.51acres	8.66acres
3.	Chaudhary Mulaka Lake	1.83 m	2.22acres	1.86 acres
4.	Kunwar Gatta Lake	3.15 m	1.56acres	1.29 acres
5.	Hauda Lake	1.19 m	9.68acre	15.6acres



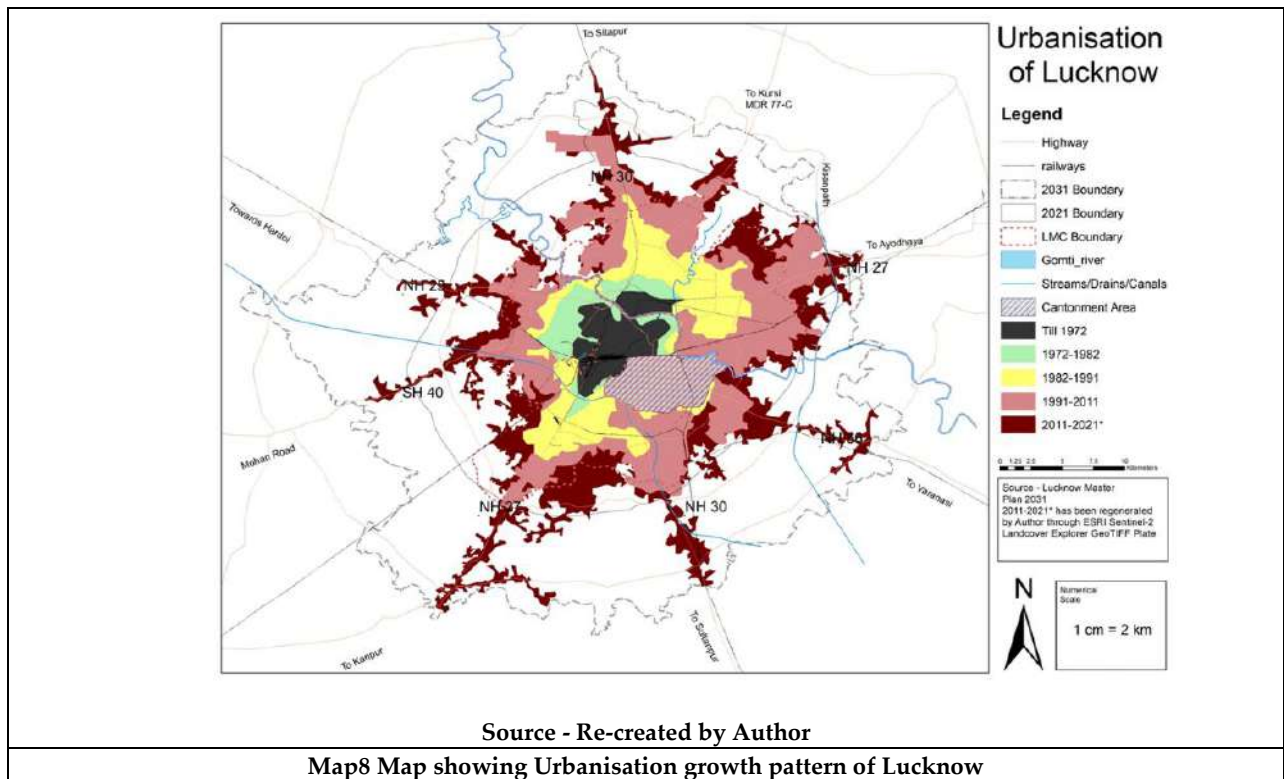


Arundhatee Mishra et al.,

Table 5: Parameter vulnerability rank of the all identified lakes based on user perception scoring. (1 - Major concern and 7 - least concern)

Parameter	Hauda Lake	Sagar Lake	Kunwar Gatta Lake	Chaudhary Mulaka Lake	Salariya Lake
Land Use and Population Density	5	5	4	5	4
Encroachment	4	4	5	3	5
Community Use	6	7	6	6	7
Pollution	1	1	1	1	2
Community Participation	3	2	2	4	3
Recreational / Tourism Activities	7	6	7	7	6
Institutional Role	2	3	3	2	1

Source: Lucknow Master Plan 2031 (2011-2021 has been regenerated by Author through ESRI Sentinel-2Landcover Explorer GeoTIFF Plate).



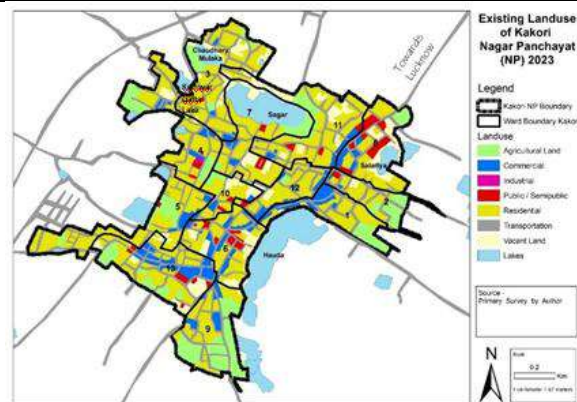


Arundhatee Mishra et al.,



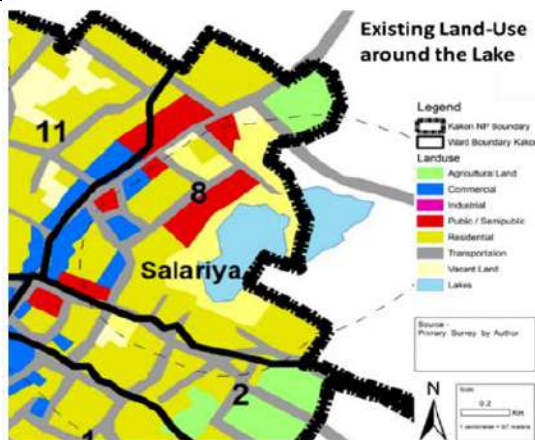
Source: Open Street Map.com

Map 2 Map showing Location of Kakori Town in Lucknow District



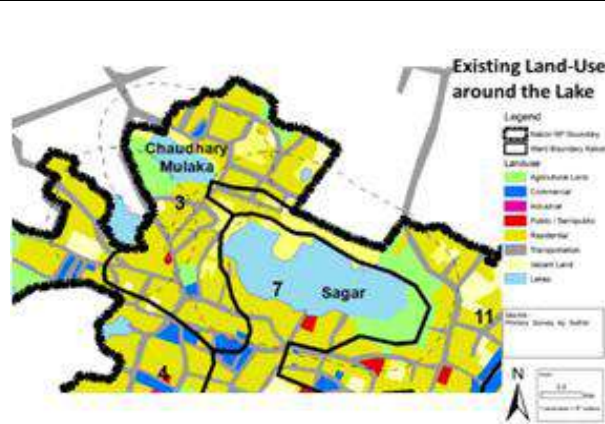
Source - Author

Map3 Existing Landuse Map of Kakori Town 2023



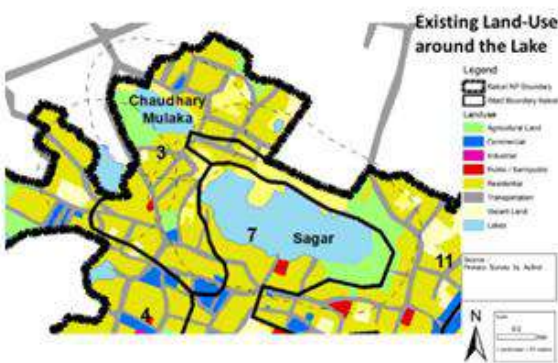
Source- Author

Map 4 Landuse character around Salariya Lake



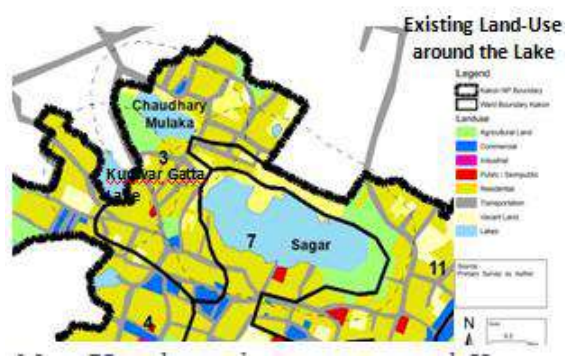
Source- Author

Map 5 Landuse character around Sagar Lake



Source- Author

Map 6 Landuse character around Chaudhary Mulaka Lake



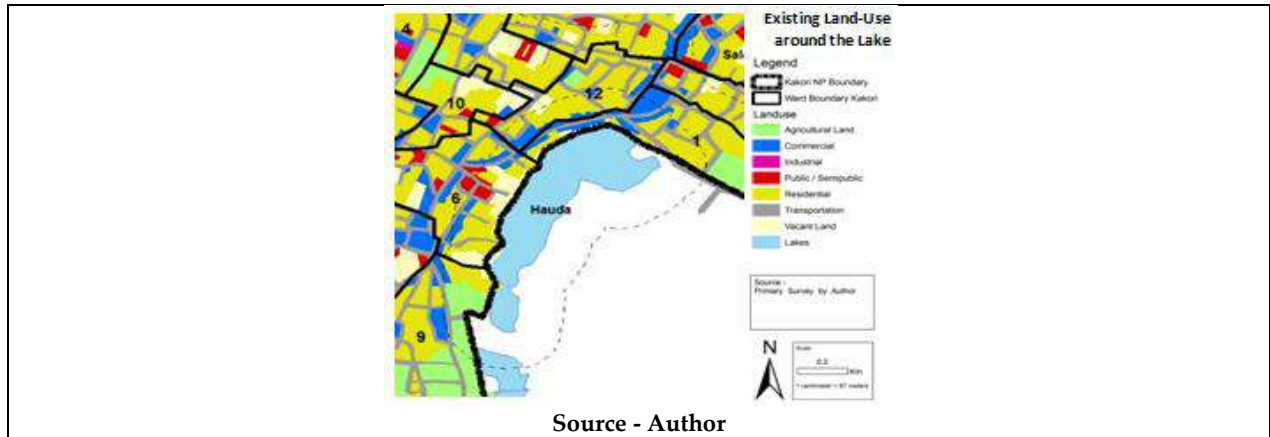
Source- Author

Map 7 Landuse character around Kunwar Gatta Lake





Arundhatee Mishra et al.,



Source - Author

Map 8 Landuse character around Hauda Lake



Source- Satellite Imagery

Image 1 Change in surface area of Salariya lake



Source- Author

Image 2 Image showing community use of Salariya lake



Source- Author

Image 3 Image showing pollution around Salariya lake



Source- Satellite Imagery

Image 4 Change in surface area of Sagar Lake





Arundhatee Mishra et al.,



Source- Author



Source- Author

Image 5 Image showing Animal grazing around the Sagar Lake

Image 6 Image showing Pollution and Encroachment around the Sagar Lake



Source- Satellite Imagery



Source- Author

Image 7 Change in surface area of Chaudhary Mulakalake

Image 8 Image showing community use of the lake



Source- Author



Source- Satellite Imagery





Image 9 Image showing pollution around the lake

Image 10 Change in surface area of Kunwar Gatta lake,





Arundhatee Mishra et al.,

 <p>Source - Author</p>	 <p>Source- Satellite Imagery</p>
<p>Image 11 Image showing community use of the lake</p>  <p>Image13 Image Showing Commercial Fishing in Hauda Lake</p>	 <p>Image14 Image Showing Pollution Around the Hauda Lake</p>





A Thematic Exploration of Insights and Reflections on E-Governance Adoption and Challenges in Anantnag, Jammu and Kashmir

Aneesa Fayaz^{1*} and P. Saravana Kumar²

¹Ph.D Research Scholar, Department of Political Science and Public Administration, Annamalai University, Tamil Nadu, India.

²Assistant Professor, Department of Political Science and Public Administration, Annamalai University, Tamil Nadu, India.

Received: 06 Jun 2024

Revised: 22 Aug 2024

Accepted: 26 Oct 2024

*Address for Correspondence

Aneesa Fayaz

Ph.D Research Scholar,

Department of Political Science and Public Administration,

Annamalai University, Tamil Nadu, India.



This is an Open Access Journal / article distributed under the terms of the **Creative Commons Attribution License** (CC BY-NC-ND 3.0) which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited. All rights reserved.

ABSTRACT

The paper rigorously examines the seismic shift from traditional Governance structures to the dynamic landscape of e-Government. It meticulously dissects how e-Governance serves as a catalyst in upholding the fundamental tenets of good Governance, specifically emphasizing citizen participation, elevating service delivery standards, and fostering unparalleled transparency within administrative frameworks. The study is designed to intricately unravel the intricate web of agencies and diverse stakeholders entrenched in the e-Governance ecosystem. It aims to spotlight their distinct roles and unequivocal contributions towards the effective orchestration of e-Governance mechanisms. Moreover, the research fervently scrutinizes the multifaceted interactions intrinsic to e-Governance, examine into pivotal realms like Government-to-citizen (G2C) and other transformative dimensions. It critically analyses the vast array of tools and resources accessible to citizens and government bodies, empowering their seamless participation and engagement in the e-Governance paradigm. The study embarks on a relentless examination, conducting a rigorous SWOT analysis, specifically targeting the e-Governance landscape in the district of Anantnag in Jammu and Kashmir. It aims to unearth the strengths, weaknesses, opportunities, and threats inherent in the local e-Governance framework. In addition, the research meticulously delineates the extensive gamut of services rendered accessible through e-Governance platforms, vividly portraying the breadth and depth of online provisions available to citizens, propelling the region towards an era of enhanced accessibility and streamlined service delivery. The paper examines the challenges entrenched within e-Governance implementation. It presents an assertive array of suggestions designed to galvanize substantial improvements in the e-Governance landscape, fostering an environment conducive to heightened efficiency and inclusivity.

Keywords: Governance, Services, Stakeholders, Strength, lacunas.



**Aneesa Fayaz and Saravana Kumar**

INTRODUCTION

Electronic Governance (or e-Gov) refers to the use of information and communication technologies (ICTs) by governments in (Relyea, H. C. 2002) delivering public services, exchanging information, communicating with citizens, businesses, and other government entities, and conducting Governance -related activities. It involves the digitization of government operations and the integration (Gronlund, Å., & Horan, T. A. 2005) of technology into various administrative processes to improve efficiency, transparency, accessibility, and accountability in the delivery of public services. Electronic Governance encompasses a wide range of initiatives and applications, including online portals for accessing government services, digital databases (Fath-Allah et., al 2014) for storing and managing information, electronic voting systems, online tax filing, digital identity systems, and more. Its primary goal is to leverage technology to transform Governance, making it more responsive, efficient, and citizen-centric. The adoption of e- Governance by administrations is driven by various compelling factors. Firstly, it aims to enhance (Hujran, O, et., al 2023) efficiency and effectiveness in delivering public services. Through digital platforms, governments can streamline processes, reduce bureaucratic hurdles, and offer faster and more accessible services to citizens. e-Governance facilitates transparency and accountability. By digitizing processes, administrations (Hirwade, M. A. 2010) can maintain clearer records, enabling better tracking of government actions and expenditures. The transparency helps in curbing corruption and building trust between the government and its citizens.

Electronic Governance also promotes (Sakolkar, P. C. 2023) citizen engagement and participation in Governance processes. Digital platforms enable easier access to information, allowing citizens to interact with the government, voice concerns, provide feedback, and participate in decision-making, thereby fostering a (Malodia, S., 2021) more inclusive Governance system. Adopting e- Governance aligns with the broader global trend of technological advancement. Governments aim to keep pace with technological developments to harness their benefits and stay relevant in an increasingly digital world. Lastly, for regions like Anantnag, Jammu and Kashmir, where geographical (Manhas, J., & Mansotra, V. 2011) challenges might hinder traditional Governance methods, e- Governance provides a means to bridge these gaps and reach remote or less accessible areas with essential services and information.

Theoretical framework

The theoretical framework of the study on Adoption and Challenges of e-Governance in Anantnag, Jammu and Kashmir revolves around understanding the intricate interplay between technology adoption, administrative processes, and societal implications. Drawing from models of technological adoption, such as the (Davis, F. D. 1989) Technology Acceptance Model (TAM) or the Diffusion of Innovations theory, the research aims to explore the factors influencing the (Dearing, J. W., & Cox, J. G. 2018) acceptance and utilization of e-Governance tools among different stakeholders. It considers institutional theories to analyse the role of government structures, policies, and institutional arrangements in facilitating or hindering the adoption of digital Governance practices. Societal perspectives, including social inclusion, access to technology, and digital literacy, are also central to comprehending the challenges faced in implementing e-Governance initiatives within the specific socio-cultural context of Anantnag District. The framework seeks to provide a holistic understanding by integrating technological, institutional, and societal dimensions to elucidate the complexities surrounding e-Governance adoption and its challenges in the region.

Significance of the Study

The study holds significance due to its comprehensive examination of the transition from traditional Governance to e-Government in Anantnag, Jammu and Kashmir. It sheds light on how e-Governance promotes good Governance principles, enhances citizen participation, and improves service delivery while addressing challenges and proposing solutions for a more efficient and inclusive Governance landscape.



**Aneesa Fayaz and Saravana Kumar****Objectives of the study**

The main objectives of the study are to comprehensively explore the transition from traditional Governance to e-Government, highlighting its role in upholding good Governance principles with a focus on citizen participation, service delivery enhancement, and transparency. The study aims to dissect the intricate network of stakeholders in the e-Governance ecosystem, emphasizing their roles in orchestrating effective mechanisms, while scrutinizing the interactions within Government-to-citizen (G2C) dimensions. It seeks to critically analyze the available tools for citizen and government engagement, conducting a SWOT analysis specific to the e-Governance landscape in Anantnag, Jammu and Kashmir, to uncover inherent strengths, weaknesses, opportunities, and threats. Moreover, the research aims to delineate the breadth and depth of e-Governance services offered, aiming for improved accessibility and streamlined delivery, while also identifying and proposing solutions to the challenges entrenched in e-Governance implementation for fostering an environment of efficiency and inclusivity.

MATERIAL AND METHODS

The study adopted a comprehensive mixed-methods approach, combining document analysis and Google Forms to gather primary and secondary data thoroughly. With a qualitative focus, it aimed to deeply understand the Adoption and Challenges of e-Governance in Anantnag District, Jammu and Kashmir. Structured interviews involved 15 government personnel, web developers, Khidmat Center and Common Service Center personnel's, and National Informatics Centre personnel. Additionally, open-ended questions extracted insights from these officials, unveiling significant themes and patterns. The study enriched its depth and context by incorporating secondary data from various sources such as books, journals, articles, government documents, and web resources.

RESULT AND DISCUSSION**Shift from traditional Governance to e- Governance**

The shift from traditional Governance to e- Governance represents a fundamental transformation in how governments (Saxena, K. B. C. 2005) operate and interact with their constituents. The evolution has been driven by several compelling factors. The integration of information and communication technologies (ICTs) has revolutionized the way governments function. Traditional Governance often relied on manual, paper-based processes, which were time-consuming, prone to errors, and less efficient. e- Governance introduces digital solutions (Seifert, J., & Petersen, R. E. 2002) that streamline administrative tasks, automate processes, and significantly enhance operational efficiency. By leveraging technology, governments can expedite service delivery, reduce bureaucratic hurdles, and optimize resource utilization.

e- Governance embodies the principles of transparency and accountability. The traditional Governance model often faced (Atiq, E., Salim, M., & Mahmood, N. 2023) challenges in maintaining comprehensive records, tracking government actions, and ensuring transparency in decision-making processes. With e-Governance, digital platforms enable better documentation, storage, and accessibility of information, fostering greater transparency. Citizens can access government data, monitor expenditure, and hold authorities accountable, thereby strengthening (Harahap, M. A, et., al 2023) trust in the Governance system. e- Governance fosters citizen engagement and empowerment. Traditional Governance frameworks typically had limited avenues for citizen participation in decision-making processes. However, e- Governance facilitates direct interaction between citizens and government entities (Singh, A. 2023) through online portals, social media platforms, and other digital channels. This increased accessibility enables citizens to voice their concerns, provide feedback, participate in policy formulation, and access government services more conveniently. The global technological revolution has played a pivotal role in the shift towards e-Governance. With the rapid advancement of digital technologies, governments worldwide recognize the need to adapt and harness these innovations to modernize Governance practices. Embracing e- Governance allows (Sharma, P. 2023) administrations to keep pace with technological developments, remain relevant, and harness the benefits of digital transformation. The transition from traditional Governance to e- Governance signifies a paradigm shift towards



**Aneesa Fayaz and Saravana Kumar**

more efficient, transparent, and citizen-centric Governance models that harness the potential of technology to better serve the needs of society.

Stakeholders and Agencies in Electronic Governance Implementation

The government of Jammu and Kashmir has undertaken several significant initiatives aimed at leveraging technology to enhance (Dar, S. A. (2022) Governance and public service delivery. One of the notable initiatives is JAKeGA, an online grievance redressal system, which provides citizens a platform to register complaints and seek resolution from relevant authorities. The digital platform streamlines the grievance redressal process, ensuring a more efficient and transparent mechanism for addressing citizen concerns. The establishment of Common Service Centres (CSCs) and Khidmat Centres across (Sharma, A., & Mir, A. Q. 2022) the region plays a pivotal role in providing various government services to citizens at their doorstep. These centres serve as access (Dar, S. A. 2022) points for a wide range of services, including application submissions, certificates, payments, and information dissemination. They bridge the gap between citizens and government agencies, making services more accessible and convenient for the public.

The creation of Software Technology Parks reflects the government's commitment to fostering a conducive environment for technological advancements and innovation. These parks serve as hubs for IT companies, start-ups, and entrepreneurs, promoting the growth of the IT sector in the region and (Dar, S. A., & Sakthivel, P. 2021) creating opportunities for employment and economic development. Aligning with the National e- Governance Plan (NeGP), the government has collaborated with web developers and the National Informatics Centre (NIC) to enhance digital infrastructure and develop user-friendly web portals and applications. These initiatives aim to improve access to information and government services while ensuring a seamless and user-centric online experience for citizens. The establishment of National Informatics Centre offices in each (Sharma, P. (2023) district signifies a decentralized approach to technology implementation. These centres facilitate the integration of digital technology into various administrative processes at the local level, ensuring that the benefits of e- Governance reach every corner of the region, addressing specific district-level needs and challenges.

Electronic Governance aims to transform Khush Hal State into a realm of genuine prosperity and well-being. By focusing on efficient Governance, transparency, and citizen engagement, these initiatives strive to create a (Bhatnagar, S. 2003) state where technology serves as a catalyst for progress. Through digitalization, the goal is to enhance accessibility to services, foster economic growth, and empower every individual, ensuring an inclusive and prosperous society for all. These initiatives collectively represent the government's concerted efforts to harness technology for (Dar, S. A. 2023) efficient Governance, citizen empowerment, and socio-economic development in Jammu and Kashmir. Through these measures, the government endeavours to create a more inclusive, responsive, and digitally-enabled Governance framework that caters to the diverse needs of its populace.

Electronic Governance : Paving the Way for Good Governance

The adoption of e- Governance is deeply rooted in the pursuit of achieving good Governance by revolutionizing traditional (Telino, V., et., al 2020) administrative processes. It aims to enhance Governance practices in various ways, primarily by ensuring efficiency, transparency, accountability, and citizen-centricity. Efficiency in Governance is greatly improved through e-Governance. For instance, online portals for government services streamline processes, reducing the time taken for tasks like applying for permits or licenses. The efficiency is evident in systems like digital tax filing, where citizens can submit returns swiftly, reducing both administrative burdens (Falk, S., Rommele, A., & Silverman, M. 2017) and turnaround time. Transparency is another hallmark of good Governance that e- Governance fosters. Digital platforms enable the publication of government decisions, expenditure details, and policy formulations, ensuring that citizens have access to comprehensive and accurate information. For instance, budgetary allocations and spending records are made publicly available, allowing for scrutiny and accountability.

Accountability is bolstered through e- Governance systems. For instance, grievance redressal mechanisms like online complaint (Lindquist, E. A., & Huse, I. 2017) portals ensure that citizens' concerns are logged and addressed



**Aneesa Fayaz and Saravana Kumar**

systematically. Authorities can track the progress of complaints, ensuring accountability in resolving issues raised by citizens. e- Governance emphasizes citizen-centricity by placing services and information within reach of the public. For instance, educational portals provide access to resources, exam results, and scholarships, ensuring equal (Monga, A. 2008) opportunities for education. This approach puts citizens at the center of Governance, making services more accessible and responsive to their needs. e- Governance aligns with the principles of good Governance by enhancing efficiency, promoting transparency, fostering accountability, and prioritizing citizen-centric services. These examples showcase how e- Governance initiatives contribute to creating an environment conducive to good Governance practices.

Jammu and Kashmir Public Services Guarantee Act of 2011

The Jammu and Kashmir Public Services Guarantee Act of 2011 is a significant legislative measure designed to empower citizens by ensuring timely delivery of public services. The act outlines a framework that guarantees specific public services within a stipulated time frame to the people of the state. It essentially sets standards for service delivery, making government departments (Singh, H. 2021) accountable for providing services promptly and efficiently. Under this act, eligible individuals have the right to receive public services within predetermined time limits. These services can range from obtaining certificates, licenses, permits, or availing specific government schemes. The act specifies the time duration within which these services must be delivered, thereby holding government officials and departments accountable for meeting these deadlines. The act introduces a mechanism for citizens to seek compensation in case of delays or deficiencies in service delivery. If the specified services are not provided within the mandated time frame or are found to be deficient, eligible individuals have the right to claim compensation. The provision aims to ensure that citizens receive the services they are entitled to without undue delays or shortcomings.

By enacting this law, the government (Kundan, M. S. 2018) aims to instill accountability, transparency, and efficiency in public service delivery. It places the onus on government agencies to prioritize citizens' needs, ensuring that they receive essential services within a reasonable and predefined time frame. The provision for compensation acts as a deterrent against negligence or inefficiency on the part of government officials, thereby enhancing the quality and timeliness of service delivery to the people of Jammu and Kashmir. The Jammu and Kashmir Information Technology Act of 2000 aims to enhance e-services delivery within the region. This act focuses on the (Wani, A. A., & Parray, J. A. 2023) establishment of a legal framework to govern electronic transactions, digital signatures, cyber security, and the use of electronic records, thereby facilitating the delivery of various electronic services to the citizens of Jammu and Kashmir.

e- Governance service

Jammu and Kashmir has emerged as a leader in the realm of e- Governance service provision, boasting an impressive array of 1028 online services, as reported by the National e- Governance Division (NeSDA). The region's remarkable achievement in delivering electronic services reflects its dedication to (Dar, S. A. 2022) embracing digital solutions for Governance. According to the NeSDA report, J&K stands out prominently for its extensive online service portfolio, underscoring its commitment to enhancing accessibility and efficiency for citizens. This accomplishment highlights the state's proactive approach in leveraging technology to cater to a diverse range of public needs. The proliferation of online services signifies a significant stride (Economic Times NeSDA report 2023) towards a more inclusive and digitally empowered Governance framework in Jammu and Kashmir. This success story positions the region as a frontrunner in embracing e-Governance, marking a significant milestone in its journey towards leveraging technology for the betterment of its citizens.

SWOT of Electronic Governance

The strengths lie in the advancements made in increased connectivity, supported by government initiatives and citizen engagement tools. These factors, coupled with a skilled IT workforce, create a solid foundation (Jiang, H. 2023) for technological growth and innovation. However, various weaknesses pose challenges. Infrastructural limitations hinder the seamless operation of technology, while the digital divide and limited internet penetration



**Aneesa Fayaz and Saravana Kumar**

create disparities in access, limiting the reach of digital initiatives. Language barriers further impede the widespread adoption and effectiveness of technological solutions. Amid these challenges, opportunities emerge. Efforts directed towards (Chinnasamy, V. 2017) rural outreach initiatives can bridge the gap, leveraging technological advancements for inclusive growth. Continued technological progress offers opportunities for innovation and development, potentially boosting tourism, economic activities, and prompting investments in IT infrastructure.

Yet, threats loom over these opportunities. Cyber security vulnerabilities present risks as connectivity expands, potentially (Dar, S. A., & Sakthivel, P. 2021) compromising systems and data. Resistance to adopting new technology, political instability, and the unpredictability of natural disasters also pose significant threats, potentially disrupting progress and infrastructure. Balancing these strengths, weaknesses, opportunities, and threats becomes crucial in navigating the landscape of technology and connectivity, requiring strategic measures to leverage strengths, address weaknesses, seize opportunities, and mitigate threats for a more resilient and inclusive technological ecosystem.

Electronic Governance Interactions**Government-to-Business (G2B)**

This model denotes interactions between government entities and businesses. It encompasses services, regulations, and collaborations designed to facilitate business operations and stimulate economic growth. For instance, online licensing and permit applications, tax filings, procurement portals, and business registration platforms are examples of G2B interactions. These initiatives streamline processes, reduce bureaucracy, and enhance transparency, making it easier (Panayiotou, N. A., & Stavrou, V. P. 2021) for businesses to interact with the government. The goal is to create an environment conducive to business development and investment, fostering economic prosperity.

Government-to-Citizen (G2C)

G2C interactions refer to services and interactions between the government and individual citizens. This includes various online platforms and services aimed at providing citizens with easy access to government information, services, and assistance. Examples of G2C interactions are online (Katoch, R. 2016) portals for paying taxes, applying for government benefits, accessing healthcare services, obtaining identification documents, and registering for various government programs. These initiatives focus on convenience, accessibility, and inclusivity, aiming to improve the quality of life for citizens by ensuring efficient access to essential government services.

Government-to-Government (G2G)

G2G interactions involve communication, collaboration, and data sharing between different government agencies or departments, either at the same administrative level or across different administrative tiers. These interactions facilitate the exchange of information, resources, and services among government (Palma, J. P. B., et., al 2023) entities to enhance operational efficiency, avoid duplication, and streamline processes. An example is the sharing of data between tax authorities and social services agencies to verify eligibility for government assistance programs. G2G initiatives aim to promote coordination and cooperation among government bodies to better serve the public.

Government-to-Employee (G2E)

G2E interactions encompass the communication and services provided by the government to its employees. These interactions involve human resources management, training programs, payroll services, benefits (Rao, V. R. 2011) administration, and internal communications within government organizations. G2E initiatives aim to enhance employee productivity, satisfaction, and efficiency by providing streamlined and accessible services for the workforce. Each of these models represents distinct facets of government interaction, emphasizing efficiency, accessibility, and transparency in service delivery across various sectors, entities, and individuals within society.

Challenges of electronic Governance in Jammu and Kashmir

Electronic Governance in Jammu and Kashmir encounters specific regional challenges that impact its implementation. The region's remote and mountainous terrain creates infrastructural limitations, leading to inadequate digital infrastructure and connectivity. The limitation in internet penetration hampers (Dar, S. A. 2022)



**Aneesa Fayaz and Saravana Kumar**

the widespread accessibility of online services. Moreover, geographical barriers and varying levels of accessibility in remote areas contribute to disparities in technology access and digital literacy, posing hurdles in uniform service delivery. The region's multilingual population presents challenges in disseminating information uniformly, necessitating the development of platforms accommodating diverse linguistic needs. Apart from regional challenges, Jammu and Kashmir faces special obstacles in the realm of e-Governance. Security concerns stemming from the sensitive geopolitical situation pose significant threats. Cyber security vulnerabilities and potential data breaches heighten risks within e-Governance systems. A significant data breach has affected the data system at Kashmir University, prompting an urgent inquiry from the Computer Emergency Response Team (CERT). Reports indicate that (Kashmir Observer(2022) sensitive information of over one million students, including registration details, email IDs, and passwords, was purportedly available for purchase on a 'Dark Web' forum for \$250. The breach has sparked demands from students and teachers for an independent investigation by a MeitY team to establish accountability and fortify the system's security. Allegations suggest that previous warnings about potential data threats within the University went unheeded. While the University authorities claim that the data appears unmodified, they are conducting further analysis. Despite certain data being (M. Karpiuk, J. Kostrubiec 2022) accessible in the public domain, the University intends to take appropriate legal measures based on in-depth analyses of any potential breaches.

Political instability and uncertainties can disrupt the continuity and stability of digital initiatives, affecting ongoing projects (Dar, S. A. (2022) and policies. The region's susceptibility to natural disasters, such as earthquakes and floods, poses risks to digital infrastructure and service continuity during emergencies. The "India Inequality Report 2022: Digital Divide" released by Oxfam India delves into data sourced from the Centre for Monitoring Indian Economy's household survey spanning Jan 2018 to Dec 2021. The report sheds light on glaring gender disparities in digital access, revealing that Indian women are 15 percent less likely to own mobile phones and 33 percent less likely to use mobile internet services compared to men. Shockingly, women constitute just one-third of the country's internet users. Globally, India stands at the forefront of the Asia-Pacific region but with a staggering 40.4 per cent gender gap, marking the widest difference. The report also uncovers a stark rural-urban digital divide, despite a commendable 13 per cent growth in digital adoption, merely 31 percent of the rural population utilizes the internet in contrast to 67 per cent of urban residents. It highlights caste-based disparities, noting that ST households exhibit the lowest (Oxfam India 2022) inclination towards formal financial services, followed by SC and OBC households. Access to computers follows a similar pattern, with General and OBC groups having notably higher rates than SC and ST populations, indicating a significant gap. Even across religions, Sikhs exhibit the highest likelihood of computer ownership, trailed by Christians, Hindus, and finally Muslims. In the global e-participation index, a measure encompassing online service provision, connectivity, and human capacity, India ranks 105 out of 193 nations, emphasizing the room for improvement in its e-Government initiatives.

The theme "Digital Generation, Our Generation" aims to address the digital gap, as per the United Nations. Despite the global shift towards online platforms in the post-COVID-19 era encompassing businesses, education, and healthcare, around 22 billion individuals under 25 lack internet access, with a significant majority being girls. Kashmir faces a particularly alarming scenario due to frequent and extended internet shutdowns in the past two years, significantly hampering online education and causing multiple challenges. The entrenched gender norms in societies like India curtail equitable technology access for women and girls, particularly those from economically disadvantaged backgrounds. These norms contribute to lower digital literacy among girls, unfamiliarity with digital tools, and unequal access to electronic devices. The mobile gender gap report highlights a staggering 56% lower likelihood of Indian women using mobile internet compared to men, with only 35% of active users in the country being women.

Frequent internet shutdowns in Kashmir significantly disrupt and impede the progress of e-Governance initiatives in the region. These interruptions hinder the seamless implementation of digital Governance (Gupta, R., & Kumar, K. 2020) tools and services, affecting various aspects such as communication, access to information, online government services, and connectivity for citizens. The lack of consistent and reliable internet connectivity poses



**Aneesa Fayaz and Saravana Kumar**

substantial obstacles to the effective functioning of e-Governance , hindering the region's technological advancements and digital outreach efforts. Indeed, the revocation of Article 370 brought about notable changes in the digital landscape of Kashmir. Post this constitutional change, there was an evident shift towards accelerating the digitization process in the region. Efforts were made to boost technological infrastructure and expand digital (Lalwani, S., & Gayner, G. 2020) initiatives to enhance Governance , public services, and connectivity. However, despite these strides, the recurring internet shutdowns have remained a persistent challenge, intermittently hampering the momentum of the digitization process and the smooth functioning of e-Governance in Kashmir.

Overcoming these challenges demands targeted strategies. Improving digital infrastructure and internet connectivity in remote areas, promoting digital literacy, and developing multilingual platforms can bridge regional disparities. Robust cyber security measures and contingency plans are imperative to address security risks and ensure service continuity. Navigating political transitions and establishing resilient systems capable of withstanding natural disasters are critical for sustaining and advancing e- Governance in Jammu and Kashmir. Collaborative efforts involving government bodies, technology experts, and local communities are pivotal in addressing these challenges and harnessing the full potential of e- Governance in the region.

Implications of the Study

The study's social implications are profound, signaling both challenges and opportunities in the realm of e-Governance . Addressing issues like digital literacy gaps among citizens and officials not only ensures effective utilization of government services but also empowers individuals with skills vital for modern participation in Governance . By bridging access gaps for marginalized communities, the study hints at a more inclusive society, where every citizen has equal opportunities to benefit from governmental initiatives. Moreover, the focus on data security and privacy in sensitive regions like Jammu and Kashmir speaks to the larger societal need for trust and protection in the digital age. Additionally, the study underscores the importance of reliable power infrastructure for sustained digital services, highlighting how basic amenities directly impact technological advancements. Ultimately, the findings stress the necessity for comprehensive policies, training programs, and seamless system integration to foster transparency, accountability, and citizen trust—cornerstones for a more participatory and well-functioning Governance framework.

FINDINGS

1. Low levels of digital literacy and awareness among citizens and government officials pose a challenge to effective e-Governance utilization.
2. Remote or marginalized communities face difficulties accessing e-Governance services, creating disparities in service availability.
3. Protecting citizen data and ensuring secure transactions within e-Governance systems is crucial, especially in sensitive regions like Jammu and Kashmir.
4. Extended power outages averaging 30 days per year significantly hinder the sustainability of e-Governance initiatives, leading to service disruptions and hampered accessibility, particularly in areas like Anantnag, where consistent power supply is crucial for the seamless functioning of digital systems.
5. Successful implementation requires strong policy support, governmental commitment, and adequate budget allocations, impacting the effectiveness of e-Governance initiatives in Anantnag.
6. Addressing diverse cultural and linguistic backgrounds within Anantnag presents a challenge in designing inclusive e-Governance platforms accessible to all.
7. Integration and compatibility among different e-Governance systems and platforms used by various government departments can be a significant hurdle.





Aneesa Fayaz and Saravana Kumar

8. Building trust among citizens regarding the reliability and efficacy of e-Governance systems is crucial for their widespread acceptance and use.
9. Training programs and skill development initiatives are essential to empower government officials and citizens to effectively utilize e-Governance tools and services.
10. Ensuring the sustainability and scalability of e-Governance projects over time and their expansion to cover more services and areas within Anantnag is a key challenge.
11. e-Governance promotes transparency by providing easy access to information, fostering accountability, and enhancing citizen-government trust, crucial for good Governance .
12. The interactions facilitate smoother communication between government, businesses, citizens, and employees, fostering transparency by enabling accessible information flow across administrative processes.
13. Frequent internet blackouts, totaling 422 between 2012 and 2022, pose a significant challenge, disrupting e-Governance services and communication channels.
14. Armed conflict and challenging geographical terrains further impede the implementation and accessibility of e-Governance initiatives, particularly in conflict-prone regions like Jammu and Kashmir
15. Ongoing efforts signal progress in comprehensive e-Governance , demanding sustained endeavors for broader accessibility and continual improvements.

Suggestions for improvement

1. Launch comprehensive digital literacy programs targeting citizens and government officials to empower them with necessary e-Governance skills.
2. Develop specialized outreach strategies for remote or marginalized communities, ensuring equitable access to e-Governance services.
3. Implement robust security protocols to safeguard citizen data, particularly in sensitive regions like Jammu and Kashmir.
4. Introduce alternative power solutions to mitigate the impact of frequent power outages on e-Governance initiatives in areas like Anantnag.
5. Advocate for strong policy support, governmental commitment, and adequate budget allocation to fortify e-Governance effectiveness.
6. Design e-Governance platforms considering diverse cultural and linguistic backgrounds within Anantnag for inclusivity.
7. Resolve compatibility issues among government e-Governance systems for streamlined operations.
8. Invest in campaigns to build citizen trust in e-Governance systems for widespread adoption.
9. Establish comprehensive training programs to empower officials and citizens in utilizing e-Governance tools effectively.
10. Develop long-term plans for the scalability and sustainability of e-Governance projects in Anantnag.
11. Emphasize e-Governance 's role in promoting transparency, accountability, and citizen-government trust.
12. Foster improved communication channels between government entities, businesses, citizens, and employees for transparent information flow.
13. Devise strategies to mitigate the impact of frequent internet blackouts on e-Governance services.
14. Tailor e-Governance strategies to navigate challenges in conflict-prone regions like Jammu and Kashmir.



**Aneesa Fayaz and Saravana Kumar**

CONCLUSION

Electronic Governance, a technological blessing, fosters transparent, efficient Governance. Leveraging technology, it enables streamlined interactions between governments, businesses, citizens, and employees, ensuring accessibility, accountability, and improved service delivery for all stakeholders involved. The evolution from traditional Governance to e-Governance in Jammu and Kashmir represents a monumental technological shift facilitated by key agencies such as Jammu and Kashmir e-Governance agency, Common Service Centres (CSCs), Khidmat Centres, Software Technology Parks, and National Informatics Centre offices. These entities have played pivotal roles in aligning with the National e-Governance Plan (NeGP) to ensure the implementation of e-Governance practices uniformly across all districts. The transition has brought about significant changes, including amendments to legislation such as the Jammu and Kashmir Public Services Guarantee Act of 2011, specifically tailored to accommodate and enhance e-Governance service delivery mechanisms. The incorporation of various interactions, including Government-to-Business (G2B), Government-to-Citizen (G2C), Government-to-Government (G2G), and Government-to-Employee (G2E), has notably amplified transparency and accountability within administrative processes. Despite these strides, persistent challenges impede the full realization of e-Governance's potential in the region. Notably, frequent internet blackouts, amounting to approximately 422 instances between 2012 and 2022, have severely disrupted digital connectivity. Rural areas suffer from limited access, contributing to the overarching digital divide. The presence of armed conflict and the complex, terrain-based geography pose formidable barriers to seamless e-Governance implementation. Addressing these challenges requires immediate and comprehensive solutions. Rectifying issues related to internet connectivity disruptions, especially the frequent blackouts, is imperative. Bridging the digital divide by ensuring equitable access to digital infrastructure in rural areas becomes paramount. The strategies must navigate the complexities posed by armed conflict and geographical constraints to ensure the uninterrupted provision of e-Governance services. Resolving these hurdles is crucial not only for the continued advancement of e-Governance but also for fostering inclusive development, equitable access to services, and transparent Governance practices throughout Jammu and Kashmir. Such solutions will play a pivotal role in creating an environment conducive to comprehensive and efficient e-Governance implementation across the region.

Conflict of interest and Funding

The author confirms no financial or personal relationships influencing the presented work. Additionally, the research has no external funding for the work.

ACKNOWLEDGMENT

I am grateful to my guide, Dr. P. Saravanakumar, whose invaluable guidance and support significantly contributed to this work. I also acknowledge the contributions of scholars whose dedicated efforts and the groundwork of previous researchers paved the way for the exploration, leading to a successful conclusion.

REFERENCES

1. Atiq, E., Salim, M., & Mahmood, N. (2023). E-Governance : A Global Perspective on A New Paradigm. *Journal Of Law, Social And Management Sciences*, 2(2), 100-107.
2. Bhatnagar, S. (2003). Transparency and corruption: Does e-government help. DRAFT Paper prepared for the compilation of CHRI, 1-9.
3. Chinnasamy, V. (2017). M-Governance is a Participative Model-SWOT Analysis. *Asian Journal of Research in Social Sciences and Humanities*, 7(1), 1207-1227
4. Dar, S. A. (2022) A Study on User's Perceptions on Mobile Banking In Handwara, Kashmir, *Indian Journal of Natural Sciences*, Vol.14 / Issue 78 / June / 2023, ISSN: 0976 – 0997, pp 56523-56537





Aneesa Fayaz and Saravana Kumar

5. Dar, S. A. (2022). E-Panchayat in Kashmir: The Smart way forward, International Journal On Human Computing Studies, Volume: 04 Issue: 6, p-ISSN: 2615-1898
6. Dar, S. A. (2022). Role of E-Governance in Higher Education in Jammu and Kashmir. Journal of Psychology and Political Science (JPPS) ISSN 2799-1024, 2(04), 1-8.
7. Dar, S. A. (2022). The Role and Awareness of E-Governance in Kashmir A Study of Kupwara District. Journal of Psychology and Political Science (JPPS) ISSN 2799-1024, 2(01), 33-42.
8. Dar, S. A. (2023). Exploring The Advantages and Obstacles of Mobile Governance in Kashmir a Comprehensive Study. Devotion: Journal of Research and Community Service, 4(10), 2037-2048.
9. Dar, S. A., & Sakthivel, P. (2021) M-Governance : A Tool For Tackling The Corruption In Jammu And Kashmir, Vidyabharti Interdisciplinary international journal Vol 12 issue 2.
10. Dar, S. A., & Sakthivel, P. (2021) Public Services Delivery through M-Governance : Jammu & Kashmir Government Initiatives. Vidyabharti Interdisciplinary international journal Vol 12 issue 2.
11. Davis, F. D. (1989). Technology acceptance model: TAM. Al-Suqri, MN, Al-Aufi, AS: Information Seeking Behaviour and Technology Adoption, 205-219.
12. Dearing, J. W., & Cox, J. G. (2018). Diffusion of innovations theory, principles, and practice. Health affairs, 37(2), 183-190.
13. Economic Times Government. (2023). J&K tops in e-Governance service delivery, has 1028 online services: NeSDA report. Retrieved from <https://government.economicstimes.indiatimes.com/news/digital-india/jk-tops-in-e-Governance-service-delivery-has-1028-online-services-nesda-report/103917568>
14. Falk, S., Rommele, A., & Silverman, M. (2017). The promise of digital government. Digital government: Leveraging innovation to improve public sector performance and outcomes for citizens, 3-23.
15. Fath-Allah, A., Cheikhi, L., Al-Qutaish, R. E., & Idri, A. (2014). E-government portals best practices: a comprehensive survey. Electronic Government, an International Journal, 11(1/2), 101-132.
16. Government of Jammu and Kashmir Home department order no home- 41 TSTS of 2022 retrieved from <https://internetshtutdowns.in/Governmentstatic-page/jammu-kashmir>
17. Gronlund, Å., & Horan, T. A. (2005). Introducing e-Gov: history, definitions, and issues. Communications of the association for information systems, 15(1), 39.
18. Gupta, K. P., & Maurya, H. (2020). The role of access convenience of common service centres (CSCs) in the continued use of e-government. Digital Policy, Regulation and Governance , 22(5/6), 437-453.
19. Gupta, R., & Kumar, K. (2020). What missing the Internet means in digital era: A case study of longest ever Internet blackout in Jammu & Kashmir. International Journal of Advanced Science and Technology, 29, 155-171.
20. Harahap, M. A. K., Kraugusteeliana, K., Pramono, S. A., Jian, O. Z., & Ausat, A. M. A. (2023). The Role of Information Technology in Improving Urban Governance . Journal Minfo Polgan, 12(2), 371-379.
21. Hirwade, M. A. (2010). Responding to information needs of the citizens through e-government portals and online services in India. The International Information & Library Review, 42(3), 154-163.
22. Hujran, O., Alarabiat, A., Al-Adwan, A. S., & Al-Debei, M. (2023). Digitally transforming electronic governments into smart governments: SMARTGOV, an extended maturity model. Information Development, 39(4), 811-834.
23. Jiang, H. (2023). A SWOT analysis of smart Governance applications amid the COVID-19 pandemic. Information Polity, 28(3), 377-393.
24. Kashmir Observer. (2022, August 10). Kashmir University hacking: Data of 1 million students found on dark web. Retrieved from <https://kashmirobsver.net/2022/08/10/kashmir-university-hacking-data-of-1-million-students-found-on-dark-web/>
25. Katoch, R. (2016). E-Governance : Government to Citizen (G2C) initiatives in India. International Journal of Research in Social Sciences, 6(10), 225-244.
26. Kumar, R. (2021). An Analytical Study of Development In Jammu And Kashmir After The Reorganisation Act, 2019. IJRAR-International Journal of Research and Analytical Reviews (IJRAR), 8(4), 23-30.
27. Kundan, M. S. (2018) Jammu And Kashmir Public Services Guarantee Act, 2011: An Overview. International Journal of Innovative Research and Advanced Studies (IJIRAS) Volume 5 Issue 10, October 2018, pp, 40-45





Aneesa Fayaz and Saravana Kumar

28. Lalwani, S., & Gayner, G. (2020). India's Kashmir Conundrum: Before and After the Abrogation of Article 370. Washington, DC: United States Institute of Peace.
29. Lindquist, E. A., & Huse, I. (2017). Accountability and monitoring government in the digital era: Promise, realism and research for digital-era Governance . Canadian public administration, 60(4), 627-656.
30. M. Karpiuk, J. Kostrubiec (2022), Institute for Local Self-Government Maribor (Lex Localis Press), ISBN 978-961-7124-10-1, <https://doi.org/10.4335/2022.1>
31. Malodia, S., Dhir, A., Mishra, M., & Bhatti, Z. A. (2021). Future of e-Government: An integrated conceptual framework. Technological Forecasting and Social Change, 173, 121102.
32. Manhas, J., & Mansotra, V. (2011, March). Critical evaluation of e-Government websites design. In Proceedings of the 5th National Conference (pp. 10-11).
33. Monga, A. (2008). E-government in India: Opportunities and challenges. JOAAG, 3(2), 56.
34. Oxfam India. (2022). Digital Divide: India Inequality Report 2022 Retrieved from URL: <https://ruralindiaonline.org/en/library/resource/digital-divide-india-inequality-report-2022/#:~:text=Oxfam%20India's%20annual%20publication%20%E2%80%93%20Digital,financial%20sectors%20in%20the%20country.>
35. Palma, J. P. B., Avila, L. S., Mag-iba, M. A. J., Buman-eg, L. D., Nacpil Jr, E. E., Dayrit, D. J. A., & Rodelas, N. C. (2023). E-Governance : A critical review of e-government systems features and frameworks for success. International Journal of Computing Sciences Research, 7, 2004-2017.
36. Panayiotou, N. A., & Stavrou, V. P. (2021). Government to business e-services–A systematic literature review. Government Information Quarterly, 38(2), 101576.
37. Rao, V. R. (2011). Collaborative government to employee (G2E): Issues and challenges to e-government. Journal of e-Governance , 34(4), 214-229.
38. Relyea, H. C. (2002). E-Gov: Introduction and overview. Government information quarterly, 19(1), 9-35.
39. Sakolkar, P. C. (2023). Impact of Digital Transformation on the Indian Government Regarding Service Delivery and Citizen Engagement.
40. Saxena, K. B. C. (2005). Towards excellence in e-Governance .International Journal of Public Sector Management, 18(6), 498-513.
41. Seifert, J., & Petersen, R. E. (2002). The promise of all things E? Expectations and challenges of emergent electronic government. Perspectives on Global Development and Technology, 1(2), 193-212.
42. Sharma, A., & Mir, A. Q. (2022). Identifying the Impact Of Digitalization On The Society And Culture Of Tribal People Of J&K.
43. Sharma, P. (2023). An Understanding of E-Governance and Its Applicability in India. Dynamics of Public Administration, 40(1), 87-93.
44. Singh, A. (2023). E-Governance : moving towards digital Governance . Vidya-a journal of Gujarat university, 2(1), 204-215.
45. Singh, H. (2021) Jammu and Kashmir Public Services Guarantee Act 2011 A critique of Notified Services and Grievances Redressal Mechanism.
46. Telino, V., Massa, R., Mota, I., Sandro, A., & Moreira, F. (2020). Paving the Way for IT Governance in the Public Sector. In Trends and Innovations in Information Systems and Technologies: Volume 1 8 (pp. 281-290). Springer International Publishing.
47. Wani, A. A., & Parray, J. A. (2023). Rationalisation of the Durbar Move: A Step Towards Good Governance in Jammu and Kashmir. Indian Journal of Public Administration, 00195561231204609.

Table 1: e- Governance service

Service Provided	Description
Issuance of Birth Certificates	Providing official birth records within a specified time frame.
Issuance of Residence Certificates	Delivering residency proof documents within a defined period.
Application for Ration Cards	Processing applications and issuing ration cards within a specified timeframe.





Aneesa Fayaz and Saravana Kumar

Issuance of Driving Licenses	Providing driving permits within a stipulated time.
Utility Bill Payments	Ensuring timely processing of utility bill payments and acknowledgments.
Pension Disbursement	Ensuring pension payments are processed and disbursed on time.
Land Record Verification	Providing verified land records and documentation within a specified period.
Educational Scholarship Applications	Processing scholarship applications within defined timelines.
Building Permit Issuance	Granting building permits or approvals within a specified time frame.

Table 2 : SWOT Analysis

Strengths	Weaknesses	Opportunities	Threats
Increased Connectivity	Infrastructural Challenges	Rural Outreach Initiatives	Cyber security Vulnerabilities
Government Support	Digital Divide	Technological Advancements	Resistance to Technological Adoption
Citizen Engagement Tools	Limited Internet Penetration	Tourism & Economic Development	Political Instability
Skilled IT Workforce	Language Barriers	Investment in IT Infrastructure	Natural Disasters

Table 3: Electronic Governance tools for people and Government

Tools Used by People	Tools Used by Government
Online Portals/Websites	Online Portals/Websites
Mobile Applications	Mobile Applications
Digital Payment Systems	Digital Payment Gateways
National Identity Systems	Digital Identity Systems (e.g., Aadhaar)
Social Media Platforms	Data Analytics for Policy Planning
Self-Service Kiosks	Geographic Information Systems (GIS)
Customer Service Hotlines	Cyber security Measures
E-signatures	Cloud Computing for Data Storage
Online Chat Support	Block chain Technology (in some cases)
Public Wi-Fi	Artificial Intelligence (AI) and Chat bots

Table 4: Type of Interaction

Type of Interaction	Description	Example
Government-to-Business (G2B)	Processes involving government and business entities, such as licensing, taxation, and procurement.	Online application for a business license or bidding for a government contract.
Government-to-Citizen (G2C)	Interactions between government and individual citizens for services, information dissemination, and feedback.	Applying for a passport, accessing information on government schemes.
Government-to-Government (G2G)	Communication and data exchange between different government departments or agencies.	Sharing data between health and education departments for policy formulation.
Government-to-Employee (G2E)	Interactions catering to internal government employee processes, training, and communication within the workforce.	Employee payroll management, accessing training modules.





Aneesa Fayaz and Saravana Kumar

Year wise report of Internet Shut downs in Kashmir

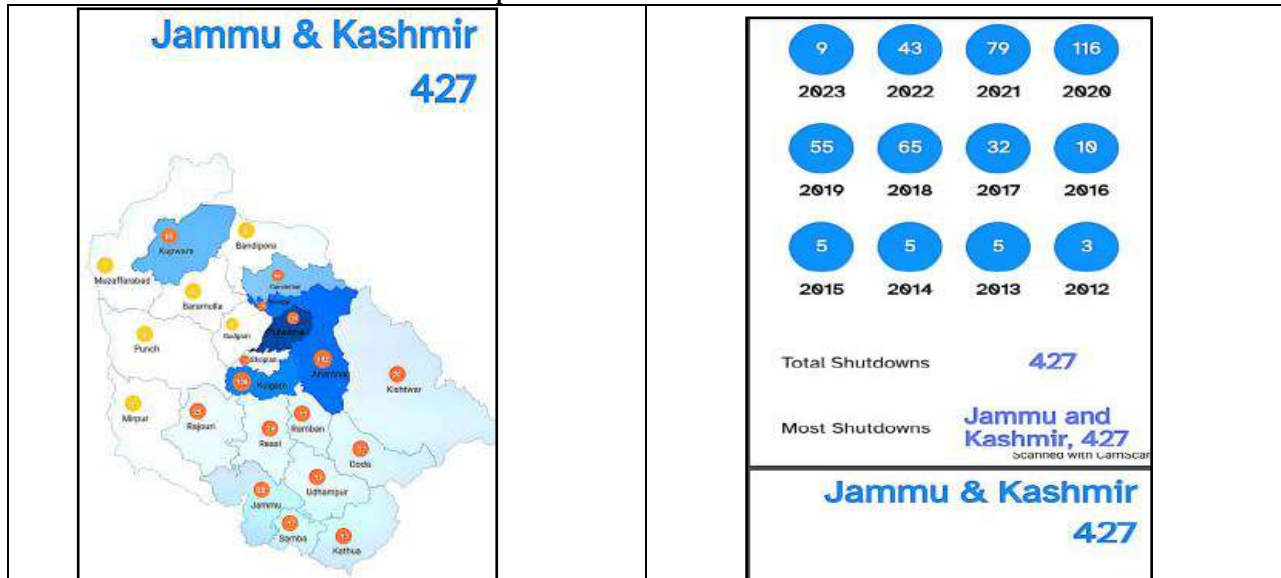


Figure.1 : Year wise report of Internet Shut downs in Kashmir
Source: Government of Jammu and Kashmir Home department order no home- 41 TSTS of 2022 retrieved from <https://internetshutdowns.in/Governmentstatic-page/jammu-kashmir>

