

Economic and Resource Impact of System of Rice Intensification (SRI): an Empirical Study of Pudukkottai District in Tamil Nadu, India.

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ABSTRACT

Paddy is an important food grain crop, which has been cultivated predominantly throughout Asia and other parts of the world. The cultivation of paddy crop under conventional method has been facing several constraints. Poor remuneration is also posing a serious threat to paddy crop, which is cultivated using conventional method. A new System of Rice Intensification method (SRI) of paddy cultivation introduced relatively recently is proved to increase the yield of paddy significantly with less water, less seed as well as with less chemical inputs than the conventional method of paddy cultivation. The available results on SRI increasingly suggest that the farmers can more than double their paddy yields with lesser of farm inputs and irrigation water. The study is based on the impact of SRI on the pattern as well as the quantity of irrigation water consumption and cost of cultivation under the three different settings. The study has been carried out in Pudukkottai District of Tamil Nadu State, India. A detailed household survey was conducted in study area during the agricultural year 2009-10. In order to capture the impact of SRI on different parameters of paddy cultivation, comparison has been made between the data collected from the different categories of farmers. The study was concluded that the SRI method of cultivation not only increased rice yield but also improves efficiency of water, land and labour use, reduced cost of cultivation and production and increases sustainability.

Key words: System of Rice Intensification, Water consumption, sustainability, Farmyard Manure.

Rajakumar**INTRODUCTION**

Paddy is an important food grain crop, which has been cultivated predominantly throughout Asia and other parts of the world. Rice, which is a processed form of paddy, is a staple food for over half of the world's population. In India, paddy's contribution to the agricultural sector is very significant. Paddy is currently cultivated in over 44 million hectares, which accounts for over 35 per cent of the total food grains area and over 23 per cent of the cropped area. With the current production of over 91 million tonnes, rice accounts for little less than fifty per cent of the total food grains production in India. Besides providing livelihood opportunities to millions of farmers in India, cultivation of paddy crop directly provides employment opportunities to the rural labourers who are relying on agriculture for their livelihood. Unlike the other crops, paddy, which is also paid as wage (in kind) to the agricultural labourers after harvesting in most part of south Asia, also helps to reduce poverty of those families working in the paddy field to a greater extent.

In spite of its significant role in food security as well as its overall contribution to agricultural development, the overall performance of paddy cultivation is not very impressive especially in recent years. While the area under paddy has not declined over the last 50 years, the production of rice has not increased since the beginning of 2000. For instance, the production of rice was at 93.3 million tonnes in 2001-02 but it declined to 90 million tonnes in 2006-07 despite introducing various promotional policies by the State and Central governments for augmenting the production of paddy. One of the important reasons for the reduction of rice production is that the productivity of rice has not increased appreciably or increasing at slower rate in most of the paddy growing states during the last ten years, beginning from the last 1990s. This reduction of rice production has already made some impact on its price, which is causing severely affecting the purchasing power of the poor people.

The cultivation of paddy crop under conventional method (inundation method or flood method) has been facing several constraints. First, since paddy is a water-intensive crop, its cultivation and water availability are closely linked. It is well known that about 70-80 per cent of freshwater withdrawals at global level are used for the agricultural purpose and rice accounts for about 85 percent of it. Paddy being an important water intensive crop (consumes 3000-5000 litre to produce one kg of rice as against the requirement of only 900 litre for wheat), irrigation water supply is essential to increase the productivity. But, the fast decline of irrigation water potential and increased demand for water from various sectors has been reducing the availability of water for agricultural sector, which is causing serious impact on the productivity of paddy in many parts of the country. The inundation method of paddy cultivation (conventional method), which normally requires large amount of chemical inputs, is also no longer effective in increasing the productivity of crop for two reasons: (a) the applied yield increasing inputs do not effectively reach the crop because of leaching and evaporation losses, and (b) the soil fertility has also declined over the years because of intensive use of external chemical inputs.

Poor remuneration is also posing a serious threat to paddy crop, which is cultivated using conventional method. Owing to steep increase in cost of cultivation, farmers are unable to recover even the cost of cultivation that they spent on paddy crop. Evidence available from the data of the Commission for Agricultural Costs and Prices (CACP) clearly reinforces this trend. For instance, an analysis carried out using the data of CACP relating to Andhra Pradesh (an important paddy growing state) suggests that paddy cultivated under inundation method is no longer profitable to farmers. The ratio of value of output to cost of cultivation (C₂) of paddy was 1.006 during 1991-92, but it declined to 0.940 during 2001-2002. Evidence also suggests that this trend seems to have not changed much even today. This low remuneration also discourages the farmers to adopt the required yield-increasing inputs, which is causing some impact on the productivity of paddy.

Paddy crop is also losing out to other crops because of low remuneration. Paddy has been predominantly cultivated under irrigated condition, where the yield is significantly higher than that of the un-irrigated condition. Estimates suggest that irrigated paddy contributes to over 75 percent of rice production in India. But, the irrigated agriculture

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is becoming cost-intensive owing to increased use of groundwater that is capital-intensive source of irrigation. Therefore, farmers of groundwater-irrigated area tend to allocate more of their cultivable lands that have been traditionally used for cultivating paddy crop to more remunerative crops like sugarcane, banana and other similar crops. Apart from increasing the support prices for paddy, new methods of production that can reduce the cost of cultivation as well as increase the productivity need to be introduced so as to change it as remunerative crop.

While the paddy cultivation under conventional method is waning now, the demand for rice has been on the rise because of population pressure and increased income growth. Some estimates suggest that the demand for rice is expected to increase by about 38 percent by 2040. Given the limit for new arable land required for expanding paddy cultivation in India, the improved production of paddy must be achieved largely from higher productivity. The slow growth in productivity of paddy cultivated under the conventional method has forced the researchers and policy makers to find out novel production methods that are cost-effective and also better in terms of land and water productivity.

A new method of paddy cultivation introduced relatively recently is proved to increase the yield of paddy significantly with less water, less seed as well as with less chemical inputs than the conventional method of paddy cultivation. The new method of paddy cultivation is popularly known as the System of Rice Intensification (SRI). SRI is not a new variety or hybrid, but it is only a new method of cultivation, where a set of innovative principles are followed for cultivating paddy. It was first developed in the 1980s by Henri de Laulanie, a French priest and farming practitioner living in Madagascar, and furthered in the 1990s by passionate farmers, scientists and researchers. A note from World Bank (2007) surmises that six key elements distinguish SRI farming practices from traditional rice growing methods. They are: (a) transplanting seedlings much earlier than in conventional methods, (b) planting only one seedling per hill, rather than a handful, (c) spacing plants wider apart than in conventional methods and arranging them in a square pattern, (d) applying water intermittently instead of continuous flood irrigation, (e) using rotary weeding to control weeds and promote soil aeration, and (f) applying organic fertilizers to enhance soil fertility and yield.

Statement of the problem

The available results on SRI increasingly suggest that the farmers can more than double their paddy yields with lesser of farm inputs and irrigation water. Using SRI method of paddy cultivation, countries like India, Indonesia, Cambodia, Vietnam, and the Philippines have recorded increase of rice yield from 60% to over 170%. Studies carried out in different locations in India also suggest that the productivity of paddy cultivated using the method SRI can significantly increase the productivity of paddy. Recognising the importance of cultivating paddy under the method of SRI, both the Central and State governments have introduced various promotional measures including subsidy scheme for popularising this method in a big way. While the area under paddy cultivated using the method of SRI has been increasing in India, comprehensive studies using farm level data covering various socio-economic aspects of it are lacking. Studies available in India are mostly based on experimental data, the results of which are expected to change substantially from the field condition. Along with other benefits, SRI is expected to impact on the cost of cultivation, labour use, water use as well as on the productivity of paddy. However, most of the studies seem to have highlighted the impact of SRI on productivity of paddy, ignoring the other important aspects of SRI. Therefore, there is a need to study the various issues surrounding the adoption of SRI method using farm level survey data. Especially we need to provide solid answers to questions such as: Who are the adopters of SRI? What are the characteristics of the farmers adopting the method of SRI? How much is the real water saving due to the adoption of SRI? What is the impact of SRI on the labour use, both family and hired labour? Whether SRI reduces the cost of cultivation or cost of production? What is the total saving in cost of cultivation due to SRI? Does it capable of increasing the productivity of paddy across all categories of farmers? In view of absence of comprehensive studies covering all the issues cited here, the present study aims to fill this gap using the field data collected from two settings located in Tamil Nadu, a southern State of India.

Rajakumar**Objectives of the Study**

- To study the impact of SRI on the pattern as well as the quantity of irrigation water consumption under three different settings.
- To study the pattern of human labour uses under the method of SRI vis-à-vis the method of conventional paddy cultivation.
- To analyse the pattern of non-labour input use of paddy cultivated under the method of SRI vis-à-vis the conventional paddy cultivation.
- To study the impact of SRI on the cost of cultivation under three different settings.
- To analyse the impact of SRI on the productivity of paddy under three different settings.
- To analyse the efficiency of various inputs used for cultivating paddy under the method of SRI vis-à-vis the conventional paddy cultivation.
- To identify the factors (state's subsidy, etc) that motivate the farmers to adopt the method of SRI.

METHODOLOGY

The study has been carried out in Tamil Nadu State, which is one of the important paddy growing and consuming states of India. As per the recent data (2005-06), paddy is grown in an area of 2.05 million hectares in the State, which is almost five percent of the India's total paddy area. Paddy is cultivated using different sources (tank, canal and groundwater) of irrigation water in the State. Since the impact of cultivating paddy under the method of SRI on different parameters is expected to be varied under different settings, the study has been carried out in three locations (settings), tank irrigated, canal irrigated area and groundwater irrigated area. The major objective of the study is to capture the impact of paddy cultivated under SRI method on various parameters. In order to do this, farmers cultivating paddy using SRI method as well as conventional (inundation) method have been selected for the study.

The study was conducted in Pudukkottai district. The average rainfall in the district is 918 mm per annum (taken as the average of 50 years). Of the total land area of 4.66 lakh hectares in the district, the net sown area is around 33 per cent and gross sown area is around 34 per cent. Tanks are the major source of irrigation in the district. The total number of tanks available in the district is 5451. All the tanks are rain-fed. Wells are the second major source of irrigation. Although rivers are not the main source of irrigation in the district, the GA Canal irrigation is available only to a small portion of the cropped area. While about 79 per cent of the net sown area is irrigated, 85 per cent of this area is irrigated by tanks and wells. Since tanks and wells are rainfed, a failure of the monsoon has a direct impact on the agricultural activities in the district.

A detailed household survey was conducted in Pudukkottai district in Tamil Nadu during the agricultural year 2009-10. As regards the sample size, a sample of 200 farmers, 100 from SRI method and 100 from conventional method, have been selected. In terms of agro-climatic settings, the sample is distributed as follows: 50 samples from tank irrigated area (25 each from SRI and conventional method), 50 samples from groundwater irrigated area (25 each from SRI and conventional method), and 100 samples from canal irrigated area (50 each from SRI and conventional method). Purposive sampling method has been followed for selecting the sample farmers because the spread of adoption of SRI method is very limited in each village. Farmers who have cultivated paddy using conventional method very close to the field of SRI method have been considered for selecting the non-adopters of SRI.

Variation in the agro-climatic characteristics of the district can be identified in terms of source of irrigation and cropping pattern with the help of secondary data. Though tank is the major source of irrigation in the district, regional variations exist in the use of agricultural water. For instance, tank irrigation dominates in Kulattur taluk, tubewell and well irrigation is the predominant source in Alangudi taluk and canal is the main source of water in

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Avudaiyarkoil taluk. The differences in the mode of irrigation significantly influence the cropping pattern in each taluk. Early communication with the agricultural department and the SRI adoption data show that the adoption of SRI has very much related to the mode of irrigation in each taluk. As the spread of SRI is limited at village level, the study chose to work at development block as unit of study and the samples were chosen from a cluster of villages in the particular block. While Kunnandarkoil block in Kulattur taluk is chosen in the tank irrigated area, Thiruvankulam block in Alangudi taluk and Avudaiyarkoil block in the same taluk have been selected for representing the well irrigated and canal irrigated areas respectively.

RESULTS

In order to capture the impact of SRI on different parameters of paddy cultivation, comparison has been made between the data collected from the two categories of farmers. This section provides the empirical results of the study.

Socio-economic characteristics of SRI and CPC households

The average age and farming experience of farmers in all the three regions together show that relatively young farmers adopt new method of cultivation practices such as SRI. Though it is true to some extent at tank irrigated areas, as the data reveals, the age and experience is higher among SRI farmers than the CM cultivators in areas where canal and borewell irrigation is a predominant source of irrigation. Unlike age and experience, the percentage of households having agriculture as the main occupation is relatively low among SRI farmers as compared to the CM paddy growing counterpart, albeit a slight difference seems to exist in tank irrigated area. Since a considerable percentage of SRI farmers are taken up non-farm employment (private and public sector, business, etc.), the percentage of households having agriculture as the main occupation is relatively low in the SRI farming class as compared to the CM paddy cultivating counterpart.

The average education of the households cultivating SRI paddy is higher than the CM paddy cultivating counterpart in all the three villages. The average education of the households cultivating SRI paddy is 5.55 years, whereas the same come to 4.83 years for the CM paddy cultivating households. The present study reveals that many endogenous and exogenous factors (such as resource position of the farmers, irrigation availability, role of extension network and co-farmers, etc.) have also played an important role in adopting the SRI method, besides the education of farmers. In fact, the study shows that about 80 per cent of the SRI farmers from all the three villages could adopt this novel method using the information provided by agricultural extension officials. The percentage of farmers who have adopted SRI method from SC is absolutely negligible in all the three villages. Moreover, the number of SC households in the sample is also very small as landownership in the community is very less which has reflected in the overall sample. A major proportion of adopters of SRI method belong to other than Schedule Caste households, they belong to Other Backward Community (OBC). Importantly, there is no a single farming household that adopts SRI method in both tank-irrigated borewell-irrigate areas. Observation in the study area indicates that the weaker section farmers, who are mostly small and marginal farmers with poor irrigation facilities, are not going to adopt this technique immediately, due to hesitation and fear about the new method. However, since this method is scale-neutral, the same farmers might adopt it at a later stage after completely getting convinced with the advantage of this technique.

The landholding details of both the SRI and conventional method of paddy cultivators shows that the sample households endowed a considerable amount of land resources. However, the average landholding size of SRI farmers is marginally higher (6.53 acres) than that of the conventional paddy cultivating counterparts (6.23 acres). This variation is only slightly higher (7.40 acres) in areas where tanks provide irrigation water for paddy cultivation. Therefore, the average landholding size of SRI farmers is not much different from the conventional method of paddy growers.

Rajakumar**Agro-economic characteristics of SRI and CPC households**

The cropping intensity of the SRI farmers is higher than that of the conventional method of paddy cultivators only in areas where canal irrigation is the major source of water. It is lower for SRI farmers in both the borewell irrigated and tank irrigated villages. As the study is carried out paddy cultivation under three modes of irrigation conditions, it is observed a considerable difference in the per cent of irrigated area between the SRI and conventional method of paddy growers. The share of irrigated area to gross cropped area (GCA) comes to 66.7 per cent for the SRI farmers, whereas the same comes to only 62.3 per cent for the conventional method of paddy cultivators. Among the three selected villages, the difference in irrigation percentage is found to be higher in two villages irrigated by canal and borewells respectively.

Cropping pattern of SRI and CM farmers

The cropping pattern of the sample farmers shows that there exists only a marginal difference between SRI and conventional method of paddy cultivators. Paddy is the main crop, which account for about 50 per cent of the gross cropped area in both the group of farmers. However, in the two study villages (CIA and BIA), farmers who cultivate paddy under conventional method have allocated relatively higher share of their cropped area (about 54 to 56 per cent) to paddy as compared to the SRI farmers (about 38 to 48 per cent). In tank irrigated village, SRI farmers have allocated relatively higher share of their cropped area (58%) to paddy as compared to the conventional method of paddy growers (54%). The share of cropped area allocated for crops like pulses and oil seeds are relatively higher among the SRI farmers, whereas the relative share of area allocated to crops such as sugarcane, banana and tree crops is marginally higher among the conventional paddy growers.

The study shows that the SRI method of paddy cultivation account for only about 20 per cent in all the three villages together (it is only 16% in TIA). This suggests that the households cultivating paddy under SRI method still allocate a substantial area of their cropped land to conventional method of paddy cultivation. However, the spread of SRI method of paddy cultivation is slowly and steadily increasing in all the three study villages. For instance, the average area allocated for SRI paddy by the sample households in all the three villages together has doubled from 0.8 acres in the agricultural year 2006-07 to 1.8 acres in the year 2009-10. Over all, the cropping pattern of the sample farmers suggests that both the groups of farmers are predominantly paddy cultivators.

Water use pattern

A substantial amount of water (45.3%) has been saved in SRI farms in all the three villages together. The rate of water saving is highest in the BIA area which account for about 50 per cent. Irrigation water has been saved with a considerable reduction in the amount of water used for paddy crops throughout the cultivation period. Two important indicators have been adopted to measure the relative use of water for irrigation in both the SRI and convention method of paddy cultivation. They are: 1) number of irrigation per acre and 2) hours of irrigation per acre. Both these indicators show that a significant difference exists in the application of water in the two farm types. For instance, on average, the number of irrigation used per acre during the cultivation period for SRI paddy is only 10, whereas the same was found to be about 16 for paddy under conventional method. In other words, the number of irrigation water per acre applied by households cultivating SRI paddy seems to have been 37 per cent lower than that of the households practicing conventional method of paddy cultivation. Similarly, the hours of irrigation water application per acre is found to be substantially lower (39 hours) for SRI farms, whereas it is higher (74 hours) among households undertaking paddy cultivation under conventional method. The reduction of water use in terms of hours is 47 per cent in SRI paddy cultivating households when compared to the conventional paddy cultivating counterpart. This is the prime reason for the substantial quantum of water saved in SRI paddy farms. Moreover, a significant variation is evident in the use of irrigation water for paddy among the three study villages. Generally, irrigation water application for both SRI and conventional paddy is lower in areas where surface irrigation (both tank

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and canal) is the main source of irrigation, whereas it is higher in borewell irrigated area (BIA). This variation is a manifestation of the nature of the main source of irrigation in each village.

Operation-wise employment (days per acre)

There exists a significant difference in the average number of labour days employed in the SRI farms and conventional farms. On average, the number of person-days employed per acre in the SRI farms comes only to 93.37, whereas it is 134.35 days for conventional farms. Employment of labour days in SRI farms is significantly lower as compared to conventional farms. In other words, about 41 person-days per acre or 31 per cent of labour have been displaced by the SRI method of paddy cultivation in all the three villages. It is a striking result. There is also a striking result regarding gender aspect of labour use in both the farm groups. It is clear from the data presented in the Table 4.7 that a substantial part of work is done by female labour force in the case of conventional farms. For instance, 86 out of 134 person-days are entirely employed by female labour in one acre of conventional paddy land. In other words, female labour forms nearly about two third of the entire workforce employed in the conventional farms. Whereas, it is only 47 days are employed by female labour out of 93 person-days in an acre of SRI field, which illustrate the fact that a sizable number of female labour force is displaced in SRI farms. The number of employment days lost by female labourers in SRI farms is found to be 39 days per acre or 46 per cent. This feature of labour is evident in all the three villages. This observation suggests that the SRI method is not gender neutral. In CM farms hired labour (81.60 days) exceeds family labour (52.76). In SRI reverse is the case- family labour (52.34) is higher than hired labour (41.06).

Input use pattern

There are differences in the use of various inputs between both the farms. Except farm yard manure (FYM), all other inputs used by the SRI paddy cultivators are considerably lower than the conventional paddy farmers. A substantial reduction is registered in the use of quantity of seed and pesticides in the SRI farms. A low intake of chemical fertilizers, particularly potash and urea, is also found in the SRI farms than in the conventional farms. The trend is uniform in all the three regions. For instance, while the average use of seed is only 3 kg/acre among the SRI farmers, the same is about 30 kg/acre among the conventional paddy cultivators, indicating a difference of over 90 per cent. Similarly, the difference in the use of pesticides between the SRI and conventional farms comes to nearly 45 per cent. In the case of fertilizers, the average use of Urea, DAP and Potash in an acre of land is 96 kg, 51 kg and 41 kg respectively in the SRI farms, whereas it is 119 kg, 53 kg and 53 kg in the conventional farms. The use of FYM is higher among the SRI farms than the conventional farms, and the difference comes to 10 per cent. As will be seen shortly, the reduction in input use does not lead to any reduction in yield, but in contrast, SRI farms register a substantial gain in yield. While the reduction of input use among the SRI farms resulted in lower cost of cultivation, it also protects the living organisms in the production environments by way of lower use of chemical fertilizers and pesticides.

Cost of Cultivation

The cost of cultivation of SRI fields are substantially lower than that of the conventional paddy farms in all the three zones. The average cost of cultivation of the three zones comes to Rs. 7203 per acre for SRI farms and Rs. 10984 for conventional paddy farms, a difference of about 33 per cent. There are several reasons for the lower cost of cultivation among the SRI farmers. First, the average seed cost of the SRI farmers is Rs. 90 per acre, which is much lower than the seed cost of the conventional paddy cultivators (Rs. 850). Moreover, there is no cost at all for the preparation of nursery among the SRI farms where family labour is mainly used. Second, the expenditure incurred on fertilisers and pesticides are substantially lower among the SRI farmers: about 34 per cent lower than the conventional paddy farmers. Third, the cost over plugging of seedlings and transplantation is also substantially lower among the SRI farms than the conventional paddy fields, a difference over 75 per cent. Fourth, the cost of

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weeding is found to be much lower among the SRI farmers due to mechanical weeding operations mainly done by family labour. Fifth, SRI farmers do not employ wage labour but use family labour for certain field operations like nursery preparation, application of fertilisers and some farm management works which ultimately reduce the real cost of cultivation. In sum, the survey results clearly indicate that the cost of cultivation is substantially lower in SRI farms than that of the conventional paddy farms.

Productivity

Productivity of SRI farms is significantly higher than the conventional paddy farms. The average productivity of SRI farms in all the three zones comes to 2.77 tonnes per acre, but the same is only about 1.61 tonnes for conventional paddy farms, indicating a difference of about 72 per cent over the conventional paddy farms. Similar trend is observed in all the three zones. While the productivity difference comes to about 71 per cent in the tank irrigated area (TIA), the same comes to about 69 per cent and 75 per cent in canal irrigated area (CIA) and borewell irrigated area (BIA) respectively. A remarkable feature of the results is the consistency of the responses- in all the three areas SRI farms give the better yield than the conventional farms. The reason underlying this consistent response is that farmers paid more care and attention to the fields in which the SRI methods were adopted.

Profitability and economic viability of paddy cultivation

Profit realised from SRI is substantially higher than the conventional paddy cultivation in all the three zones. While the average profit comes to about Rs. 20188 per acre for SRI farms, it is only about Rs. 6930/acre for the conventional paddy farms, indicating a difference of about Rs. 13258. The profit of the SRI farmers is nearly 190 per cent over the profit of the conventional paddy cultivators.

Level of adoption of SRI methods and difficulties

It is found that a mixed pattern of adoption of recommended practices prevails among the SRI farmers. Out of eight recommended practices referred to them, only three were completely adopted in practice. Farmers find no difficulties in adopting seed rate at recommended quantity, transplantation of the seedling in prescribed time and required space. Weeding is done by mechanical weeders which are felt difficult to operate in low lands. Farmers are not satisfied with the application of recommended chemical inputs as they fear about yield risk. Irrigation water control at field level determines the application of water at required time. Farmers located in tank irrigated areas find difficulties in the application of water in time and required level as water storage in tanks is uncertain over the cultivation season.

Adoption and yield

The mixed pattern of adoption practices resulted in differential yield: higher the level of adoption larger will be the yield per acre. Better yield is achieved in farms where SRI methods are intensively followed. Lower level of yield is associated with farms where adoption rate is found to be poor. Out of 100 farmers who are SRI adopters, only 40 per cent has followed the recommended practices completely and the remaining 60 per cent followed partially in all the three areas. While the better adopters get yield rate of 29.83 quintals per acre, the poor adopters have only 22.74 quintals, indicating a difference of 30 per cent over the poor adopters of the SRI methods.

CONCLUSION

It is evidenced from the study that the SRI method of cultivation not only increased rice yield but also improves efficiency of water, land and labour use, reduced cost of cultivation and production and increases sustainability. Thus, SRI can play a major role in such a water scarce situation. The current crisis in agricultural sector should serve

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as a timely wakeup call for both the central and the state governments to refocus on agriculture. However, there is no need to invest in a second Green Revolution to feed the country in the face of a growing population and shrinking land base for agriculture. Promoting SRI through a sustained campaign must be the most desirable option available now.

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A Case Control Study of Type II Diabetes Mellitus on Prediction of Cardio Vascular Risk Factors

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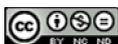
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ABSTRACT

Diabetes Mellitus is one of the leading causes of death & disability in the world. It is a group of metabolic diseases characterized by hyperglycaemia, resulting from defects in insulin secretion, insulin action or both. In patients with type II diabetes mellitus, the risk of CVD events is two to four folds higher than in the non diabetic population. Patients with diabetes mellitus are prone to cardio vascular disease & risk factors presumably unrelated to diabetes such as Hcy, ApoB, Folic acid, GlyHb which are involved in the atherothrombotic process are noted in this work. This study was conducted on a group of 50 male patients with the age group of 45-60 years. The above parameters including anthropometric measurements were analyzed. Results for these parameters were found to be significantly different in diabetic patients than the normal subjects. From the data it seems reasonable to assume that moderately elevated Hcy is an independent risk factor for CVD in the diabetic subjects with the highly declined level of folic acid. Studies have also shown that moderately raised concentrations of Hcy are prevalent in diabetic subjects & that an inverse relation exists between Hcy & folic acid. In conclusion it can say that Hcy hit hard in diabetes. Therefore it has been suggested that lowering serum homocysteine level by increase the level of folic acid, probably the most effective means of decreasing CVD.

Key words: Atherothrombosis, Homocysteine, Apolipoprotein B, Folic acid, GlycosylatedHb.

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INTRODUCTION

Health is not a static phenomenon but a dynamic life process, which begins at birth and is governed by the genetic, nutritional and environmental factors through out life [1]. During the past two decades, great progress has been made forward securing the hypothesized relationship between diabetes mellitus and coronary artery disease and in defining the role of several parameters [9]. Diabetes mellitus is a chronic condition associated with abnormally high levels of glucose in the blood [6]. People with diabetes either do not produce enough insulin- a hormone that is needed to convert sugar, starch and other food into energy needed for daily or cannot use the insulin that their bodies produce. As a result, glucose builds up in the blood stream. If let untreated, diabetes can lead to blindness, kidney disease, nerve disease, heart disease and stroke etc. Patients with diabetes mellitus are prone to cardio vascular disease & risk factors presumably unrelated to diabetes, such as hyperhomocysteinaemia may be involved in the atherothrombotic process in these subjects [12]. Cardiovascular Disease (CVD) is the leading cause of death in individuals with type II diabetes, which affects some 15 million peoples. Among diabetic individuals, CVD accounts for > 50% of all deaths and stroke accounts for an additional 15% [11]. This review mainly features the level of Apo B, homocysteine & folic acid in patients with diabetes mellitus and potential role of them in the development of CVD in diabetic patients. The metabolism of folate and homocysteine are interrelated and increasing foliate intake augments remethylation of homocysteine, leading to a reduction of up to 25% in plasma concentration [4]. This effect occurs despite normal plasma foliate and can be achieved by increased folic acid doses. This has led to the proposal that folic acid treatment may reduce the cardiovascular risk by reducing Hcy. The present study features especially the levels of hemoglobin, glycosylated Hb, homocysteine, folic acid & apolipoprotein B in diabetic patients with the risk of CVD.

MATERIALS AND METHODS

Participants: The selected subjects were known diabetic of 50 male patients with the age group of 45-60 years were included in this study. An equal number of age & sex matched subjects were taken as controls. Data collected included height, weight, body mass index, blood pressure. The study sample (blood and urine) were collected from each subject at Rontgen Diagnostic & Research Centre (RDRC), Thanjavur, Tamilnadu, who volunteered to participate in this study. The protocol was approved by the ethical committee of Bharathidasan University, Trichy. Subjects were clearly informed, in advance about the purpose and a written consent was obtained from them.

Clinical Examination: Volunteers were subjected to medical examination including anthropometric by well-trained staff. Glucose level was estimated both in fasting and postprandial condition. The blood was pooled in tubes for separation of serum was taken for the further investigations. The other relevant parameters like Hemoglobin (Hb), Glycosylated Hemoglobin (HBA_{1c}), Apolipoprotein B (ApoB), Homocysteine (Hcy) and folic acid. Serum concentrations of fasting and postprandial glucose and HBA_{1c} estimation were performed by a fully automated A15 Biosystem analyzer. Serum ApoB, Hcy and folic acid were estimated by using ELISA Reader. Hemoglobin level was measured by haematometer. The results were fed into the main frame computer and using the statistical analysis system, the data analyses were conducted separately for the diabetic and the non diabetic controls. The significance of the difference in the results was determined using t-test. We used SPSS 12.0 Software for all the statistical analyses.

RESULTS

Table 1 represents the case study of the subjects taken for study. The controls were also in the same range as the diabetics, had no family history of diabetic mellitus. The systolic & diastolic blood pressure was found to be not significant in diabetic patients and normal subjects. The body mass index was found to be significantly increased in normal subjects when compared to diabetic patients.

Amargeetha et al.**Table-1. Anthropometric measurements of normal and diabetic subjects**

General characteristics	Normal	Diabetic patients
Tot no of subjects	50	50
Age (yrs)	45-60	45-60
Sex	MALE	MALE
Height (m)	164 ± 10.7	165 ± 8.7
Weight (kg)	71 ± 12.3	65 ± 10
Body mass index (kg/m ²)	26 ± 5.0	24.2 ± 5.4*
Systolic blood pressure (mm/hg)	132 ± 10.8	132 ± 11.9
Diastolic blood pressure (mm/hg)	86.3 ± 7.3	86 ± 9.0

Statistical analysis - Paired Student t test *p = 0.0133

Table-2. Biochemical indices of control & diabetic patients

S.No	Parameter	Normal	Diabetic patients	Sig.(2-tailed)
1.	Haemoglobin %	13.3 ± 1.35	11.0 ± 0.8	p<0.0001
2.	HBA _{1c} %	6.57 ± 1.09	8.0 ± 1.0	p<0.0001
3.	Blood glucose (F) mg/dl	89 ± 14	142 ± 32	p<0.0001
4.	Blood glucose (PP) mg/dl	117 ± 22	220 ± 68	p<0.0001
5.	Apolipoprotein B [g/l]	0.84 ± 0.28	2.63 ± 0.57	p<0.0001
6.	Homocysteine [μmol/l]	3.63 ± 0.89	7.66 ± 1.24	p<0.0001
7.	Folic acid [μg/ml]	4.72 ± 0.78	1.96 ± 0.59	p<0.0001

Statistical analysis - Paired Student t test

The Table 2 predicts that the level of hemoglobin was decreased while the level of HBA_{1c} was significantly increased in the diabetic subjects. The levels of blood glucose fasting and post prandial condition were significantly elevated in diabetic subjects. The level of Apolipoprotein B and homocysteine were found to be elevated when compare to that of normal patients but folic acid level are highly declined in the diabetic subjects.

DISCUSSION

More than 17 million people currently have diabetes and this number is rapidly increasing [5]. Affected individuals are at high risk of premature death due to cardiovascular & other chronic diseases. Thus the annual estimated costs of diabetes increase of greater than 30 % since 1998 [7]. This social burden & evidence that the diabetes epidemic is being fueled by our current life style [10] means that diabetes is now an urgent public health problem. Blood glucose levels in fasting & post prandial conditions are seems to increase in diabetic subjects than the normal. Due to defects in insulin secretions, insulin action, or both (Alberti & Zimmet, 1998; 1999), glucon levels are elevated and resulting in hyperglycemic condition [13].

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The glycosylated Hemoglobin was found to be more than the normal subjects. The level of HBA_{1c} is strongly linked risk for incident nerve disease and CVD in people with type 1 and type 2 diabetes mellitus. Moreover, randomized trials have clearly shown that decreasing the glycosylated Hb level reduces these risks [3]. First this reports clearly prone that the glycosylated Hemoglobin level is an independent progressive risk factor for incident cardiovascular events, regardless of diabetes status. Every 1-percentage point absolute increase above a clearly normal glycaemic level predicts a 20 % relative increase in the incidence of CVD. These report show that the HBA_{1c} level can now be added to the list of other clearly established indicators of cardiovascular risk, such ad blood pressure and cholesterol level. This dysglycemia epidemic may therefore be the harbinger of a future epidemic of CVD [14].

Apo B is the marker of all potentially atherogenic lipoproteins, as predictor of CVD in patients with type 2 diabetes mellitus. First, diabetes is often concomitant with many lipid & lipoproteins abnormalities characterized mainly by elevated Apo B, which is potentially atherogenic consistent with the results of non- diabetic subjects, the level of Apo B was normal and not having the risk of CVD. From the study it can hypothesize that high level of Apo B was responsible for the development of CVD in type 2 diabetic mellitus subjects.

Plasma homocysteine levels are elevated in patients with non insulin dependent diabetes mellitus (NIIDDM) [15]. From the data, it seems reasonable to assume that moderately elevated homocysteine is independent risk factors for CVD subjects. The data suggest that hyperhomocysteinaemia can enhance atherogenic and thrombogenic pathways common to classic risk factors such as smoking and DM because NIIDDM is associated with the high risk of CVD, interaction with hyperhomocysteinemia may have important implication with regard to risk of CVD conferred by hyperhomocysteinaemia is higher in subjects with diabetes [2]. The data reveals that there was no significant change in serum folic acid levels in normal subjects, but highly declined in diabetic subjects [8]. Studies have also shown that moderately raised concentrations of homocysteine are prevalent in diabetic subjects and that an inverse relation exists between homocysteine concentrations and concentration of folic acid.

Finally there is an increasing evidence that hyperhomocysteinaemia is common in the elderly population especially diabetic subjects. A large part of the prevalence of hyperhomocysteinaemia in the diabetic subject is due to the level of folic acid. Therefore it has been suggested that lowering serum homocysteine levels by increase the level of folic acid, probably the most effective means of decreasing CVD.

CONCLUSION

Finally this work reveals that homocysteine hit hard in diabetes. Given the high cardiovascular risk in diabetes & the fact that hyperhomocysteinemia can easily and safely be improved by folic acid, because folic acid has an inverse relationship with homocysteine.

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Effect of Coirpith based Cyanobacterial Biofertilizers, Jiwamrita and their Combination on Morphometric Parameters of *Oryza sativa* L.

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ABSTRACT

Cyanospray and cyanopith are the novel extracts of coir pith degraded by cyanobacterium, *Oscillatoria annae*. They were compared with other fertilizers to evaluate the efficacy of growth promotion of *Oryza sativa* (Paddy) in pot experiment. In the present investigation, optimization and morphometric analyses were carried out for Jiwamrita, Cyanospray and cyanopith treated *Oryza sativa* L (Paddy). Furthermore, the treatment was conducted individually such as chemical fertilizer of NPK (farmers recommended dose), Cyanospray, Jiwamrita combination of Cyanospray+Cyanopith and Jiwamrita + Cyanospray + cyanopith. The morphometric analyses in combined application of Jiwamrita+Cyanospray+cyanopith were significantly higher when compared to control, chemical fertilizer, Jiwamrita, Cyanospray and cyanopith. The results of this study indicated that the combined application of Jiwamrita+Cyanospray+cyanopith was potentially enhanced the growth of *O. sativa* L.

Key words: Cyanobacteria, Coir pith, Cyanospray, Cyanopith, Jiwamrita, *Oryza sativa*. L and *Oscillatoria annae*.

INTRODUCTION

Paddy (*O. sativa* L.) is plausibly the most important cereal in the world and serves as staple food for about 50% of the world's population [1]. Tamil Nadu is one of the leading rice growing states in India, has been cultivating rice from time immemorial as this State is endowed in all favourable climatic conditions suitable for rice growing. Cyanobacteria play a wide spectrum in the field of biofertilizer, energy production, human food, animal feed and biomedical

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applications etc. Cyanobacterium (*Oscillatoria annae*) is capable of abating various kinds of pollutants and serves as potential biodegradative organism [2]. Coir pith is generally mixture of dust eliminated during extraction which creates disposal related problems and it drives air pollution. *O. annae* grow luxuriant along with coir pith in field condition and during degradation period the color of medium changes from colorless to brown called Cyanospray which indicate the release of phenolic compounds into medium [3]. Combined effect of basal and foliar application of coir pith based cyanobacterial biofertilizer rendered positive impact on the growth of *Basella rubra* L. [4]. The application of coir pith based cyanobacterial biofertilizer would increase the growth and yield of *O. sativa* L. was already reported in our laboratory [5].

In India, organic farming was well developed and systematized agricultural practice during the past and this 'Ancient Wisdom' incurred through Indian knowledge system to specify the use of Jiwamrita (Nectar for life) is one of the traditional biofertilizers. It contains full of microbial inoculum to promote immense biological activity in the soil and makes the nutrients available to the crop. Jiwamrita is not to be considered as a nutrient for the crop but only a catalytic agent to promote biological activity in the soil. To increase the yield, fertility of the soil and population of microbes and earthworms should be enhanced [6, 7]. Hence, investigation was focused on to analyze the combined effect of Jiwamrita, Cyanospray and Cyanopith biofertilizers, control (without treatment) and Chemical fertilizers NPK (farmers recommended dose) on paddy and its growth was ascertained with different morphometric parameters.

MATERIALS AND METHODS

Organisms and Culture Condition

Axenic fresh water cyanobacteria, *Oscillatoria annae* was obtained from the germplasm of the National Facility for Marine Cyanobacteria (NFMC), Bharathidasan University, Tiruchirappalli, Tamil Nadu, India. The culture was maintained in BG11 medium [8] under 1500 lux at 25±2 °C with 10/12 hrs light /dark cycle.

Lignocellulosic Material

Coir pith was collected from coir pith industry, near Srirangam, Tiruchirappalli, Tamil Nadu, India.

Experimental Layout Preparation of Cyanospray (Foliar Spray) and Cynopith solution

A shallow pit was prepared 7 feet length, 3 feet width, 2 feet depth and a rexin sheet was stretched over the pit. 100 liters of water was added to the pit with 100g of neem extract. The pit was inoculated with 1 kg of *O. annae*. After 3 days the coir pith and cyanobacterium (*O.annae*) was added to the ratio of 1:10. Hence, the cyanobacteria were allowed to act on the coir pith for 30 days for degradation. The uprising solution when coir pith degraded by Cyanobacterium was filtered and used as a foliar spray (Cyanospray) in different concentrations.

Preparation of Jiwamrita Solution [7]

Local cow dung of 10Kg, local cow urine 10L, jaggery 2kg, dicot flour (Bengal gram) 2g and fertile soil 1kg were mixed in a plastic container or tank and made up to 200L of water. Then the plastic container or tank was incubated under shadow for 24 hours then the mixture was stirred clockwise direction twice in a day.

Pot Experiments

This study was accomplished at National Facility for Marine Cyanobacteria (NFMC), Tiruchirappalli, Tamil Nadu, India. The paddy (*O.sativa* L.) of Andhra ponni variety BPT5204 seeds were obtained from Tamil Nadu Rice Research

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Institute (TNRRI), Aduthurai, Thanjavur District, Tamil Nadu. Seventy (70) cement pots with diameter of 30 cm and a height of 30 cm were used. The holes log at the bottom of each pot was carefully sealed before filling with 5 kg of soil. Seedling were transplanted in a rate of one seedling per hill to a depth of 3.4 cm. Totally, 7 experiments were carried out by treating with biofertilizers like JK (70ml/pot), CYS (60ml/pot), CYP (25g/pot), combined application of CYS+CYP and JK+CYS+CYP in 10 replicates and are compared with control (without treatment) and chemical treatment (farmers recommended dose). Throughout the experiment, water level was maintained in 2.5cm depth.

Measurement of Plant Growth

The morphological parameters such as number of leaves, leaf length, leaf width, tiller length, number of tillers, culm length, primary internode length, secondary internode length, plant height, seminal root length number of crown roots, shoot fresh weight, shoot dry weight, root fresh weight, root dry weight and total plant height (15, 30, 45 and 60th day) were recorded.

Determination of shoot fresh and dry weight, root fresh and dry weight [9].

The harvested plants were washed in tap water to remove the debris and weighed the fresh weight. Then the plants were kept in paper bags and dried at 60°C in an oven until reaches constant weight and this was defined immediately after harvesting by using an electronic weighing balance

Statistical analysis

The Statistical analysis was performed using SPSS 11.5 software (Statistical program for social sciences). All probabilities were 100 tailed. Differences between the means were evaluated for significance by using least significant difference (LSD) and Duncan's multiple range test (DMRT) values ($P < 0.001$).

RESULTS AND DISCUSSION

Morphological parameters of *O. sativa* L. including number of leaves, leaf length, leaf width, tiller length, number of tillers, culm length, primary and secondary internode length, plant height, seminal root length number of crown roots, shoot fresh weight, shoot dry weight, root fresh weight, root dry weight and total plant height on 15, 30, 45 and 60th days were significantly higher in combination of biofertilizers (Jiwamrita + cyanospary + cyanopith) than combination of cyanospray+cyanopith treatment and individual treatments like Jiwamrita, cyanospray, cyanopith, chemical and control ($P < 0.001$) (Table. 1,2,3 and 4) and (Fig. 1). This is due to the presence of nutrients and plant growth regulators in the organic biofertilizers. Some of earlier research on cyanobacterial based biofertilizers for its growth promoting abilities also supports the present investigation. For instance, Viswajith 2008 [10] tested the culture filtrate of lignocellulosic waste degraded by *Osillatoria annae* for its plant growth promoting ability as foliar spray on *Tagetes erecta*. The results showed that *Tagetes erecta* sprayed with *O. annae* degraded lignocellulosics (*O. annae* + coir pith; *O. annae* + *Prosopis juliflora*; *O. annae* + *Lantana camara*) showed better growth and flowering than the control and their individual treatment.

Moreover, Cyanospray fertilizer at 0.4% concentration was optimally found to increase the morphometric characteristics and yield of *Aloe barbadensis* [11]. Subramanian and Malliga, 2011 [12] demonstrated that the combined application of Cyanopith 25g + Cyanospray 0.4% biofertilizers potentially increased the morphological, biochemical parameters and yield in *Zea mays* (Corn). Various morphological growth parameters of *Manihot esculenta* such as shoot length and width, numbers, length and width of leaves, number of branches showed effective variations and better results by addition of Cyanopith biofertilizers when compared with inorganic (NPK) and combined fertilizers (NPK and organic manure) [13].

Durairaju and Malliga**CONCLUSION**

The present research work concluded that the pot experiment of *Oryza sativa* L. was incredibly higher in the combination of Cyanospray+Jiwamrita+Cyanopith than all other treatments. This method of farming system with the use of organic fertilizers Cyanospray, Jiwamrita and cyanopith is successful in releasing nutrients to crop for increased sustainable production in an ecofriendly manner and rendering pollution free environment with less input to lead a healthy life.

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Table 1: Effect of Coirpith based Cyanobacterial Biofertilizers, Jiwamrita and their Combination on Morphometric Parameters of *Oryza sativa* L. (15th Day).

S. No	Parameters	CON	CHE	JK	CYS	CYP	CYS+CYP	JYK+CY S+CYP
1	Tiller length (cm)	9.63±.13 ^b	9.62±.14 ^a	9.69±.18 ^e	9.63±.04 ^b	9.67±.14 ^d	9.63±.14 ^b	9.65±.16 ^c
2	Number of leaves (numbers)	2.7±.48 ^c	2.5±.52 ^a	2.7±.48 ^c	2.7±.48 ^c	2.7±.48 ^c	2.7±.48 ^c	2.6±.51 ^b
3	Primary leaf length (cm)	2.55±.07 ^a	2.64±.17 ^e	2.58±.18 ^b	2.66±.17 ^f	2.62±.12 ^c	2.62±.16 ^c	2.63±.12 ^d
4	Secondary leaf length (cm)	5.62±.16 ^a	5.63±.11 ^b	5.65±.15 ^c	5.62±.13 ^a	5.78±.09 ^e	5.75±.09 ^d	5.62±.17 ^a
5	Culm length (cm)	3.42±.13 ^a	3.50±.12 ^c	3.47±.09 ^b	3.53±.08 ^d	3.57±.09 ^f	3.56±.12 ^e	3.61±.07 ^g

CON- Control, CHE- Chemical, JK- Jiwamrita, CYS- Cyanospray, CYP- Cyanopith

The mean significant difference at p<0.001 (99.9%) level of probability using Duncan's Multiple Range Test.

Table 2: Effect of Coirpith based Cyanobacterial Biofertilizers, Jiwamrita and their Combination on Morphometric Parameters of *Oryza sativa* L. (30th Day).

S.No.	Parameters	Control	Chemical	Jiwamrita	Cyanospray	Cyanopith	Cyanospray + Cyanopith	Jiwamrita + Cyanospray + Cyanopith
1	TL	18.5±.20 ^a	19.4±.19 ^b	26.5±.17 ^e	21.4±.19 ^c	22.4±.22 ^d	28.5±.19 ^f	36.7±1.4 ^g
2	NOT	1.0 ±.00 ^a	1.0 ±.00 ^a	1.4±.51 ^c	1.0 ±.00 ^a	1.3±.48 ^b	1.5±.52 ^d	2.2±.42 ^e
3	NOL	4.6±.51 ^a	5.2±.42 ^c	6.8±.42 ^f	4.8±.42 ^b	5.5±.52 ^d	6.1±.56 ^e	13.1±1.2 ^g
4	LL	16.6±.84 ^a	16.9±.68 ^b	19.5±.69 ^e	17.8±.66 ^c	18.6±.82 ^d	19.5±.63 ^e	24.3±1.01 ^f
5	LW	0.11±.03 ^a	0.13±.04 ^b	0.42±.06 ^f	0.18±.04 ^c	0.29±.05 ^d	0.39±.05 ^e	.60±.04 ^g
6	CL	3.6±.02 ^a	3.7±.22 ^b	4.3±.15 ^e	3.8±.07 ^c	3.9±.06 ^d	4.4±.13 ^f	4.8±.07 ^g
7	PIL	3.3±.11 ^a	3.5±.07 ^b	4.7±.09 ^f	3.8±.10 ^c	3.9±.06 ^d	4.0±.10 ^e	5.02±.25 ^g
8	SIL	2.7±.07 ^a	2.9±.08 ^b	5.8±.09 ^e	4.4±.07 ^c	3.6±.08 ^e	4.8±.06 ^e	6.2±.08 ^f

TL - Tiller Length

NOT - Number of Tillers

NOL - Number of Leaves

LL - Leaf Length

LW - Leaf Width

CL - Culm Length

PIL - Primary Internodes Length

SIL - Secondary Internodes Length

The mean significant difference at p<0.001 (99.9%) level of probability using duncan's multiple range test.

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Table 3: Effect of Coirpith based Cyanobacterial Biofertilizers, Jiwamrita and their Combination on Morphometric Parameters of *Oryza sativa* L. (45th Day).

S.No.	Parameters	Control	Chemical	Jiwamrita	Cyanospray	Cyanopith	Cyanospray + Cyanopith	Jiwamrita + Cyanospray + Cyanopith
1	TL	24.1±.03 ^a	23.7±.07 ^a	37.1±.39 ^d	27.6±.21 ^b	28.3±.47 ^c	37.4±.59 ^d	48.8±.19 ^e
2	NOT	1.7±.48 ^a	1.9±.56 ^{ab}	3.4±.69 ^c	1.6±.51 ^a	2.8±.63 ^{Bc}	3.2±.78 ^c	5.6±.96 ^d
3	NOL	6.9±.87 ^a	7.1±.99 ^a	11.1±.73 ^{bc}	10.1±.56 ^b	10.2±.91 ^b	11.6±.84 ^c	15.9±.99 ^d
4	LW	0.17±.04 ^a	0.25±.05 ^a	0.67±.08 ^{cd}	0.50±.06 ^b	0.57±.10 ^{Bc}	0.76±.05 ^d	1.0±.11 ^e
5	LL	19.9±.16 ^a	20.2±.35 ^b	24.0±1.2 ^c	22.6±.57 ^b	22.5±.37 ^b	24.6±.17 ^c	34.4±.18 ^d
6	CL	3.69±.14 ^a	4.0±.25 ^b	4.7±.09 ^c	4.1±.25 ^b	4.5±.12 ^c	4.7±.11 ^c	5.5±.13 ^d
7	PIL	3.5±.08 ^a	3.7±.09 ^{ab}	4.7±.06 ^d	3.9±.23 ^b	4.2±.12 ^c	4.6±.14 ^d	5.5±.33 ^e
8	SIL	3.0±.13 ^a	3.3±.07 ^b	6.2±.10 ^d	3.7±.11 ^c	5.9±.25 ^c	6.2±.13 ^d	8.51±.12 ^e

TL - Tiller Length, NOT - Number of Tillers, NOL - Number of Leaves, LL – Leaf Length, LW - Leaf Width
 CL - Culm Length , PIL - Primary Internodes Length, PL- Plant length, SIL - Secondary Internodes Length
 The mean significant difference at p<0.001 (99.9%) level of probability using Duncan’s Multiple Range Test.

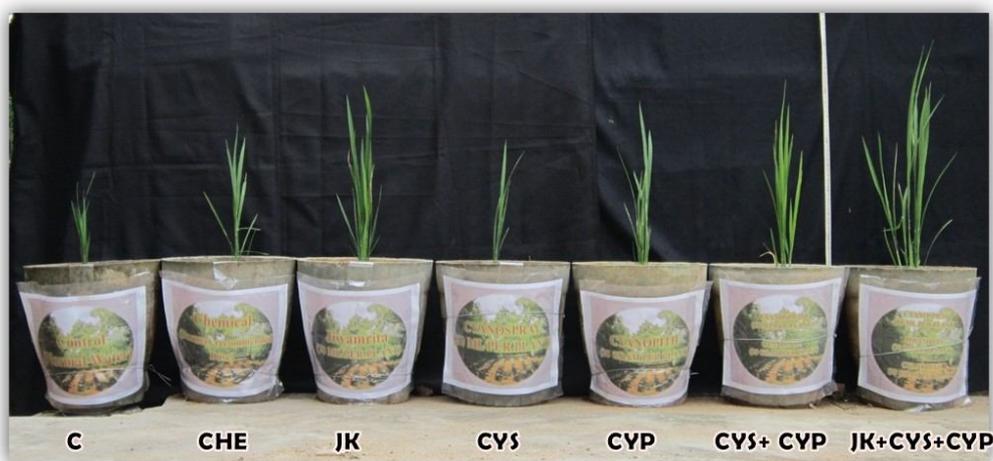


Fig.1: Effect of Coirpith based Cyanobacterial Biofertilizers, Jiwamrita and their Combination on Morphometric Parameters of *Oryza sativa* L. (60th Day).

C - Control, CHE- Chemical, JK- Jiwamrita, CYS- Cyanospray, CYP- Cyanopith

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Table 4: Effect of Coirpith based Cyanobacterial Biofertilizers, Jiwamrita and their Combination on Morphometric Parameters of *Oryza sativa* L. (60th Day).

S.No.	Parameters	Control	Chemical	Jiwamrita	Cyanospray	Cyanopith	Cyanospray + Cyanopith	Jiwamrita + Cyanospray + Cyanopith
1	TL (cm)	29.1±.03 ^a	31.7±.07 ^c	42.1±.39 ^e	29.6±.21 ^b	32.3±.47 ^d	43.4±.59 ^f	58.8±.19 ^g
2	NOT (numbers)	1.7±.48 ^b	3.9±.56 ^d	6.4±.69 ^f	1.6±.51 ^a	2.8±.63 ^{Bc}	6.2±.78 ^e	8.6±.96 ^g
3	NOL (numbers)	11.9±.87 ^a	17.1±.99 ^c	28.1±.73 ^f	15.1±.56 ^b	21.2±.91 ^d	25.6±.84 ^e	35.9±.99 ^g
4	LW (cm)	0.47±.04 ^a	0.61±.05 ^c	0.87±.08 ^f	0.50±.06 ^b	0.63±.10 ^{Bd}	0.86±.05 ^e	2.0±.11 ^g
5	LL (cm)	23.9±.16 ^a	34.2±.35 ^c	46.0±1.2 ^e	32.6±.57 ^b	42.5±.37 ^d	49.6±.17 ^f	57.4±.18 ^g
6	CL (cm)	4.69±.14 ^b	5.1±.25 ^c	6.7±.09 ^e	4.1±.25 ^a	6.5±.12 ^d	7.7±.11 ^c	8.5±.13 ^f
7	PIL (cm)	3.5±.08 ^a	3.7±.09 ^{ab}	4.7±.06 ^f	3.9±.23 ^c	4.2±.12 ^d	4.6±.14 ^e	5.5±.33 ^g
8	SIL (cm)	4.0±.13 ^a	5.3±.07 ^c	8.2±.10 ^f	4.7±.11 ^b	6.9±.25 ^d	7.2±.13 ^e	9.51±.12 ^g
9	TPH (cm)	37.0±10.6 ^a	56.2±15.3 ^b	71.3±5.7 ^c	41.7±11.4 ^a	59.8±5.0 ^{bc}	64.8±9.7 ^{bc}	85.3±5.3 ^d
10	NCR (numbers)	40.0±8.6 ^a	57.4±15.0 ^{ab}	91.1±23.0 ^c	53.3±10.4 ^{ab}	69.3±10.4 ^b	91.9±7.3 ^c	156.8±17.2 ^d
11	SRL (cm)	11.5±4.7 ^a	12.6±4.4 ^a	19.2±3.4 ^b	12.6±3.1 ^a	18.9±4.9 ^b	17.0±2.9 ^{ab}	21.7±3.7 ^c
12	SFW (g)	2.96±0.93 ^a	5.07±0.87 ^b	7.7±0.4 ^c	2.2±0.7 ^a	4.8±0.5 ^b	3.5±0.2 ^c	17.2±1.7 ^d
13	RFW (g)	0.3±0.1 ^a	1.6±4.4 ^{ab}	19.2±3.4 ^c	12.6±3.1 ^{ab}	18.9±4.9 ^{bc}	17.0±2.9 ^{bc}	21.7±3.7 ^d
14	SDW (g)	2.12±0.76 ^a	3.72±0.63 ^{bc}	4.66±1.54 ^c	1.34±.62 ^a	2.83±0.49 ^{ab}	4.12±0.69 ^{bc}	11.14±1.43 ^d
15	RDW (g)	0.25±0.08 ^a	1.56±0.13 ^c	1.85±0.47 ^c	0.48±0.15 ^{ab}	0.85±0.09 ^b	1.80±0.07 ^c	3.07±0.58 ^d

SL – Shoot Length, TL - Tiller Length, NOT - Number of Tillers, NOL - Number of Leaves

LL – Leaf Length, LW - Leaf Width CL - Colum Length, PIL - Primary Internodes Length,

SIL - Secondary Internode Length, PL- Plant length,TPH – Total Plant Height NCR – Number of

Crown Roots,SRL – Seminal Root Length, SFW – Shoot Fresh Weight RFW – Root Fresh Weight

SDW – Shoot Dry Weight, RDW – Root Dry Weight

The mean significant different at p<0.001 (99.9%) level of probability using Duncan's Multiple Range Test.

Studies on the effect of Pesticide Dimethoate 30%EC on the Biochemical Composition of the freshwater fish, *Labeo rohita* F. Hamilton.

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ABSTRACT

The aquatic ecosystem is contaminated by indiscriminate and wide spread of pesticide and other metallic pollutants in controlling the agricultural pests. The non-target organisms like fishes, bivalves, prawns and crabs etc. of the freshwater ecosystem are adversely affected. Pesticides are injurious to non-target organisms like fish. Fishes are very sensitive to a wide variety of toxicants in water; various species of fish show uptake and accumulation of many contaminants or toxicants such as pesticides. Pesticides are one of the most potentially harmful chemicals introduced into the environment. Though they have contributed considerably to human welfare, their adverse effects on non-target organisms are significant. The nature of toxic effect of the pesticide Dimethoate 30% EC on the biochemical aspects of aquatic organisms. Fishes belonging to species *Labeo rohita* were exposed to concentration of 0.398 ppm for 24, 48 and 72 respectively.

Keywords: Ecosystem, *Labeo rohita*, Pesticides, Toxicants, non-target.

INTRODUCTION

Carbohydrates, proteins and lipids play a major role as energy precursors for fishes under stress conditions (Idler and Clemes, 1959 and Umminger, 1970). The carbohydrates levels become altered by (Manoharan and Subbaiah, 1982), (Ayyapasekar, 1982) and (Palanisamy *et al.*, 1986). The decreased levels of carbohydrate *Channa striatus* and *Catla catla* exposed to phosalone have reported by Seenivasan (1986). Changes in normal carbohydrate and muscle of *Sarotherodon mossambicus* exposed to DDT have been studied by Ramalingam, (1988). Effects of oxydemeton-methyl on carbohydrate metabolism of freshwater air breathing *Channa striatus* was analyzed by Natarajan (1989). The

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neurotoxic effects of Hexachlorocyclohexane on glycogen metabolism of teleost fish *Channa punctatus* was noticed by Srinivas Reddy *et al* (1993). The toxicity of titanium dioxide industrial effluent on the biochemical compositions of the muscle and liver of the cichlid freshwater fishes *Oreochromis mosambicus* and *Etroplus maculatus* were studied by Vijayamohan and Achuthan Nair, (2000). Toxic effect of carbaryl on glucose level in the muscles of heteropneustes fossils was analysed by Sapna Shrivastava and Sudha Singh, (2003). Effect of sugar factory effluent on glycogen, protein and free amino acid content in tissues of the *Lepidocephalus thermalis* was studied by Sonawane *et al.* (2004). A study on the effect of heavy metal chromium sulphate on the biochemistry of the fish *Labeo rohita* has been noted by Vasanthi (2007).

Cholesterol is an important and prominent lipid present in all living systems and plays an important role in body metabolism (Sudish Chandra, 1985). Amudhavalli *et al* (1988) have reported the decreased lipid content of muscle and liver of *Tilapia mossambicus* when exposed to the sub lethal concentration of zinc. Changes in the protein and lipid contents of intestine, liver and gonads in the lead exposed freshwater mussel *Channa punctatus* was observed by Jha. (1991). Effects of pesticide carbaryl and phorate on serum cholesterol level in fish, *Clarias batracus* was analysed by Jyothi and Narayanan, (1997). The effect of phosalone on total lipid and triglycerides in freshwater fish *Tilapia mossambicus* were analyzed by Leela Siva Parvathi *et al.* (2000).

India is surrounded by agriculture fields or their source of freshwater is continuously in contact with the agriculture farms in which lot of pesticides, weedicides, insecticides are used. Keeping in view that the present work is planned to study the effect of the pesticide "Dimethoate 30% EC" on the biochemical constituents namely Carbohydrate, Protein, Cholesterol in the tissues namely muscles, gills, liver, kidney of the fish *Labeo rohita*.

MATERIALS AND METHODS

Test animal- *Labeo rohita*

In the present study, fishes were exposed to different concentration of a pesticide namely Dimethoate 30% EC and its effect on biochemical constituents of the fish, *Labeo rohita* was studied.

Test Toxicant

Name: Dimethoate 30% EC

Dimethoate 30% EC is one of the organophosphorus insecticides widely used against vegetables and fruit sucking aphids, mites, saw flies and boring insects on cereals, cotton, chilly, tobacco and oil seeds. During rainy season along with running water, Dimethoate 30% EC insecticides enter the fresh water resources and results into aquatic pollution. Pesticides are well known example for causing more toxic effects in teleost (Scott., 1967; Jackson., 1968).

Collection and maintenance of fish

Bulk of sample of fishes (*Labeo rohita*) ranging in weight from 4-5gms and measuring 4-6cm in length were procured from Tamil Nadu Fisheries Department, Aliyar, Tamilnadu. Fishes were acclimatized to the laboratory conditions for one month in large cement tank. The tank washed using 1% KMnO₄ to prevent fungal infection prior to stocking. The fishes were fed regularly with conventional diet rice bran and oil cake 1:1 ratio. Feeding was stopped one day prior to the start of the experiment. Fishes about the same size irrespective of sexes were selected for the experiment (Plate 2).

RESULTS AND DISCUSSION

The effect of pesticide Dimethoate 30% EC on biochemical constituents in the tissues of the fish, *Labeo rohita* showed a significant change in the present study. The mortality of the fish *Labeo rohita* exposed to different concentration of

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Dimethoate 30% EC was observed and it shown that the Dimethoate 30% EC is very toxic to fish even at very low concentration.

Total protein

Environmental stress invokes compensatory metabolic activity in the organs of an animal through modification and modulation of the quantity and quality of proteins. The result of the present study showed significant decrease in protein content in the tissues studied. The percentage decrease of protein is greater in gill. It is maximum in 72 hours. The percentage of decrease is 67.52. Gill is an important organ because of its direct contact with water, which allows the pesticides to enter through it and get accumulated in the fish body. It has been suggested that water pollutants damage the fish gill, presumably by causing breakdown of the gas exchange mechanism with consequent tissue hypotoxic conditions (Khare and Sing, 2002).

Borah and Yadav, (1995) have reported gradual decrease in protein and glycogen of gill in *Heteropneustes fossilis* under dimethoate toxicity. Susan *et al.* (1999) observed significant decrease in protein content in gills of *Catla catla* under sublethal concentration of pyrethroid fenvalerate. Rao and Ramaneshwari, (2000) observed decrease in protein and carbohydrate content in the gill of *Labeo rohita*, *Mystus vittatus* and *C. punctata* under endosulfan and monocrotophos toxicity.

The alteration in protein value may also be related to some structural changes in the liver, the arrangement of hepatic cords leading to the alteration of liver metabolism. The decrease in liver protein is also attributed to the inhibition of protein synthesis (Ganeshwade, 2011). The decrease in protein content suggests an increase in proteolytic activity and possible utilization of its products for metabolic purpose. The fall in protein level during exposure may be due to increased catabolism and decreased anabolism of proteins. (Ganeshwade, 2012). A significant reduction in the levels of proteins and glycogen (Sreekala *et al.*, 2013). Present findings are in good agreement with the above findings.

Carbohydrate

Carbohydrates form an important organic constituent of animal tissues. It is one of the important macromolecule, which comes first to reduce fish from enduring stresses caused by any xenobiotic by providing energy. The results of the present finding showed a decrease of carbohydrate level in kidney and high in muscle. The percentage decrease of carbohydrate is greater in muscle. It is maximum in 72 hours. The percentage of decrease is 41.84.

The function of muscle glycogen is to act as a readily available source of hexose units for glycolysis within the muscle itself (Harper, 1985). Umminger, (1970) reported that the principle and immediate energy source in carbohydrate when exposed to stress condition. The level of carbohydrate in tissues is decreased when it treated with herbicides. The decrease in carbohydrate indicated the altered carbohydrate metabolism which might have resulted from the enhanced breakdown through glycogenolysis to meet the high energy demand, due to mental stress. A similar trend of decrease in carbohydrate was reported by many workers. Jayashree & Gill, (1987) have reported in *Barbus conchoryis* on the toxicity of aldicars, decrease in carbohydrate probably due to glycogenolysis and utilization of glucose to meet increased metabolic cost as suggested by Viswarajan *et al.* (1998).

Ramalingam, (1998) studied on the toxic affect of DDT, Malathion and mercury on the tissues of fish, *Sarotherodon mossambicus*. Geetha Bhaskar, (1997) in *Anabas testudineus* due to sublethal doses of Fenvalerate; Kumar and Saradamani, (2004) in *Cirrihinus mrigala* on the effect of insecticide; Venkataramana *et al.* (2006) in *Alossogobius giuris* on the impact of malathion. All these workers reported the significant decrease of carbohydrate. The decrease in glycogen content may also be due to inhibition of the enzyme glycogen synthetase or hormones which mediate glycogen synthesis (Stamp and Lesker, 1967; Edwards, 1973). Glycogen content in the muscle of *C.mrigala* under sublethal and lethal concentrations of fenvalerate showed depletion, in *L. rohita*, the glycogen content of the muscle remained similar to that of control fish (Susan *et al.*, 2010).

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A fall in glycogen level clearly indicates its rapid utilization to meet the enhanced energy demands in fish exposed to toxicants through glycolysis or hexose monophosphate pathway; it is assumed that decrease in glycogen content may be due to the inhibition of hormones which contribute to glycogen synthesis (Ganeshwade, 2011). Enhanced utilization of glycogen and its consequent depletion in tissues may be attributed to hypoxia since it increases carbohydrate consumption (Ganeshwade, 2012). A significant reduction in the levels of proteins and glycogen (Sreekala *et al.*, 2013) here the glycogen level is decreased in kidney.

Carbohydrate represents the principle and immediate energy precursor for fish exposed to stress conditions. Hence, the decrease in carbohydrate in kidney of the fish, *Labeo rohita* envisage for excess energy needed to alleviate for excess energy needed to alleviate the toxic stress. Toxic stress imposes an increased energy requirement from the animal adopt to the changed metabolic condition and this achieved through utilization of reserve stores of carbohydrate in fish tissues under toxic stress is due to increased glycogenolysis.

Cholesterol

Cholesterol is an important normal body constituent used in the structure of cell membranes, synthesis of bile acid and synthesis of steroid hormone moreover, lipid in fish also contribute to the buoyancy mechanism. Thus any change in lipid metabolism effect the ability of fish to store energy obtains nutrients and in long term the stability to survive.

The results of the present study showed high cholesterol content in the tissue muscle. The percentage decrease in 24,48 and 72 hours recorded were ranged from 23.93 to 53.84 in liver, 10.06 to 54.71 in kidney, 2.201 to 13.58 in muscle and 14.01 to 67.38 in gill. The percentage decrease is high in 72 hours for muscle. Maximum decrease of cholesterol level is found in gill.

Decrease in muscle lipid indicates that lipid hydrolysis might be accelerated to derive energy to overcome pesticidal stress (Rao *et al.*, 1985). The disturbance of fat metabolism is an indication of impaired pancreatic functions. Decrease in amount of lipid may possibly due to the utilization of lipid to meet additional energy requirement under stress. Similar reductions of lipids on various tissues were studied by various authors. A decreased lipid level on the liver of *Channa punctatus* exposed to emisan was observed by Ram & Sathyanesan, (1987). Srinivas *et al.* (1991) have reported decreased lipid content in *Tilapia mossambica* exposed to Atrazine. Gradual depletion of lipid content in gills of Catla catla exposed to Malathion was analysed by Mishra *et al.* (2004). Arockiya Reta and John Milton, (2006) have showed declining trend of lipid content of the brain, gill, kidney, liver and muscle on exposure to carbamate in the first *Oreochromis mossambicus*.

Cholesterol content was decreased in liver during exposure period. It might be possible that dimethoate causes general damage and structural changes in liver and it leads to effect on capacity of liver to store cholesterol (Ganeshwade, 2011). The decrease in cholesterol content was observed; it might be possible that dimethoate causes general damage, blockage of enzyme system for steroidogenesis (Ganeshwade, 2012). Cholesterol levels showed a marked increase in the tissues of test fish sampled from lake Yellamallappa chetty lake compared to control fish from Hebbal fish form (Sreekala *et al.*, 2013).

The above study reveal that the changes of proteins, carbohydrate and cholesterol in the Dimethoate 30% EC treated fishes will naturally effect the nutritive value of these animals and all the metabolites studied are found to be sensitive changes in the normal indicators which reflect changes in the normal activities of various functional systems.

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CONCLUSION

To get an idea about the nature of toxic effect of the pesticide Dimethoate 30% EC on the biochemical aspects of aquatic organisms. Fishes belonging to species *Labeo rohita* were exposed to concentration of 0.398 ppm for 24, 48 and 72 respectively. Biochemical characterization like total protein, carbohydrate and cholesterol were estimated in liver, kidney and gill. Protein level is high in gill. It is reduced with increasing hours of exposure. Maximum reduction is found in 72 hours exposure. Protein level is less in kidney. Here also maximum reduction is found in 72 hours. In kidney, carbohydrate showed maximum reduction of 5.8 mg/g after 72 hours exposure. Carbohydrate level is high in muscle and low in kidney. Cholesterol level is high in muscle as 31.1mg/g. It is increased with decreasing hours of exposure. It is found to be high at 72 hours as 36.83mg/g. Cholesterol level is low in gill. Maximum reduction is found in 72 hours.

From the present study it is concluded that the above biochemical parameters could be used as a non specific biomarkers with regard to the effects of toxicants on organisms. It is also suggested that the random use of fertilizers and pesticides must be avoided for preserving our aquatic resources.

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Fig.1: Test animal- *Labeo rohita*



Fig.2: Acclimatization of fish

Table 1: Mortality of *Labeo rohita* in different concentration of Dimethoate 30% EC

Concentration ppm	No. of fishes	No. of fishes dead	Mortality %	Probit kill
0.3	10	0	0	0
0.32	10	0	0	0
0.34	10	0	0	0
0.36	10	1	10	3.72
0.38	10	3	30	4.48
0.39	10	4	40	4.75
0.398	10	5	50	5.00
0.40	10	6	60	5.52
0.42	10	9	90	6.28
0.46	10	10	100	8.09

LC 50 = 0.398 ppm , Lower : 3.72, Upper : 8.09, Mean : 3.78, SD: 2.86

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Table 2 : Changes in protein content (mg/g) in the liver of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	Tissues	C	E	%	C	E	%	C	E
Liver	2.10±0.07	1.70±0.06	19.04	2.10±0.07	0.18±0.03	80.47	2.10±0.07	0.18±0.04	91.42
SE	0.024			0.012			0.028		
't' test	9.98**			49.75**			52.95**		

Table 3 : Changes in protein content (mg/g) in the kidney of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	Tissues	C	E	%	C	E	%	C	E
kidney	1.47±0.03	0.98±0.01	33.33	1.47±0.03	0.71±0.04	51.70	1.47±0.03	0.3±0.04	79.59
SE	0.004			0.016			0.016		
't' test	29.28**			32.58**			49.44**		

Table4: Changes in protein content (mg/g) in the muscle of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	Tissues	C	E	%	C	E	%	C	E
Muscle	1.89±0.03	1.12±0.04	40.74	1.89±0.03	0.7783±0.04	59.25	0.3833±0.21	1.89±0.03	79.89
SE	0.016			0.016			0.085		
't' test	34.03**			53.28**			16.18**		

Table 5: Changes in protein content (mg/g) in the gills of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods

Exposure	24 Hours			48 Hours			72 Hours		
	Tissues	C	E	%	C	E	%	C	E
Gills	3.11±0.04	2.64±0.03	15.11	3.11±0.04	1.74±0.03	44.05	3.11±0.04	1.01±0.03	67.52
SE	0.012			0.012			0.012		
't' test	20.15**			65.31**			93.54**		

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Table 6: Changes in carbohydrate content (mg/g) in the liver of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	C	E	%	C	E	%	C	E	%
Liver	17.40±0.39	11.83±0.44	32.01	17.40±0.39	7.60±0.51	56.32	17.40±0.39	6.42±0.37	63.10
SE	0.179			0.208			0.151		
't' test	21.14**			34.14**			45.63**		

Table 7: Changes in carbohydrate content (mg/g) in the kidney of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	C	E	%	C	E	%	C	E	%
kidney	8.4±0.26	10.7±0.39	21.49	8.4±0.26	6.4±0.26	23.80	8.4±0.26	5.8±0.42	30.9
SE	0.159			0.106			0.171		
't' test	10.96**			12.13**			11.67**		

Table 8: Changes in carbohydrate content (mg/g) in the muscle of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	C	E	%	C	E	%	C	E	%
muscle	46.6±0.35	44.8±0.26	3.862	46.6±0.35	30.9±0.36	33.69	46.6±0.35	27.1±0.26	41.84
SE	0.106			0.146			0.106		
't' test	9.283**			70.5**			100.6**		

Table 9: Changes in carbohydrate content (mg/g) in the gills of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	C	E	%	C	E	%	C	E	%
carbohydrate	30.9±0.39	18.8±0.30	39.15	30.9±0.39	15.6±0.35	49.51	30.9±0.39	11.6±0.51	62.45
SE	0.122			0.142			0.208		
't' test	54.77**			65.6**			67.23**		

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Table10: Changes in lipid content (mg/g) in the liver of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours			
	Tissues	C	E	%	C	E	%	C	E	%
liver	23.40±0.35	17.80±0.28	23.93	23.40±0.35	17.61±0.14	24.78	23.40±0.35	10.80±0.52	63.10	53.84
SE	0.114			0.057			0.212			
't' test	28**			34.79**			45**			

Table 11: Changes in lipid content (mg/g) in the kidney of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	Tissues	C	E	%	C	E	%	C	E
kidney	31.8±0.35	28.6±0.32	10.06	31.8±0.35	17.4±0.30	45.28	31.8±0.35	14.4±0.26	54.71
SE	0.130			0.122			0.106		
't' test	15.26**			69.93**			89.73**		

Table12:Changes in lipid content (mg/g) in the muscle of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	Tissues	C	E	%	C	E	%	C	E
Liver	31.8±0.30	31.1±0.26	2.201	31.8±0.30	28.2±0.32	11.32	31.8±0.30	36.8333±44.71	13.58
SE	0.106			0.130			18.256		
't' test	2.913*			18.37**			74.35**		

Table 13: Changes in lipid content (mg/g) in the gills of *Labeo rohita* exposed to pesticide Dimethoate 30% EC for different periods.

Exposure	24 Hours			48 Hours			72 Hours		
	Tissues	C	E	%	C	E	%	C	E
Liver	21.4±0.50	18.4±0.35	14.01	21.4±0.50	10.5±0.39	50.93	21.4±0.50	6.98±0.08	67.38
SE	0.142			0.159			0.032		
't' test	11.06**			0.159**			63.98**		

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2. Volume with supplement: Shen HM, Zhang QF. Risk assessment of nickel carcinogenicity and occupational lung cancer. Environ Health Perspect 1994; 102 Suppl 1:275-82.
3. Issue with supplement: Payne DK, Sullivan MD, Massie MJ. Women's psychological reactions to breast cancer. Semin Oncol 1996;23(1, Suppl 2):89-97.

Books and other Monographs

4. Personal author(s): Ringsven MK, Bond D. Gerontology and leadership skills for nurses. 2nd ed. Albany (NY): Delmar Publishers; 1996.
5. Editor(s), compiler(s) as author: Norman IJ, Redfern SJ, editors. Mental health care for elderly people. New York: Churchill Livingstone; 1996.
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