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# Biochemical Changes in Cirrhinus Mrigala (Hamilton) Exposed to Cypermethrin

Zoology

**KEYWORDS :** freshwater fish Cirrhinus mrigala , lethal ,sublethal ,proteins ,carbohydrates, cypermethrin

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## ABSTRACT

*Many pesticides have been reported that to produce a number of biochemical changes in fish both at lethal and more often at sub lethal levels. Changes accur in ion Concentrations, Organic constituents, enzyme activity, and Endocrinal activity The favorable eco-toxicological properties of cypermethrin are related to their ability to penetrate rapidly and interact with the site of action in fishes. Since synthetic pyrethroid exposure is very common, it is imperative to assess the biochemical changes in the tissues of the organism with reference to the toxicant have been considers by the toxicologists for the chronic effects. Due to chronic effects alterations of energy sources and cycles by the animals is inevitable due to stress and as a whole results change in the system.*

*Hence the present study has been taken to study the effect of cypermethrin 10% E.C. formulation at sub-lethal and lethal concentrations to fish. The gill, Kidney, Liver , Brain and Muscle tissues of the exposed and control fish of cirrhinus mrigala (Hamilton) were selected for Biochemical studies. The results indicate that the liver, a vital organ of carbohydrate metabolism was drastically effected by cypermethrin. In all most all the tissues of the organs i.e. Kidney, Liver, Brain, Muscle and Gill tested at sub lethal concentrations of cypermethrin, a decrease in glycogen value was noticed during the exposure periods. Exposure to sub-lethal doses of cypermethrin 10% E.C, the total protein content was found to decrease in all the tissues. Maximum decrease was noticed in brain and liver at 24 hrs exposure. Same trend was noticed in the same tissues even at 96 hrs exposure. At 48 hrs exposure, maximum decrease was observed in liver and muscle.*

**Introduction:**

Water pollution has become inevitable owing to over industrialization, civilization and green revolution. Among the pollutants the pesticides have been recognised as one of the serious pollutants of the aquatic ecosystems with deleterious effects on the living resourses. Although effects of pesticides on fishes are extensively studied and has also been received by Holden (1973), Mekin etal (1976), Brungs etal (1977), Edwards (1983), Gupta (1986) and Murty (1986) but, yet there is a need for information from the physiological angle. Incidentally a great deal of attention has been paid to evaluate the hazardous effects of Various Pesticides on Physiology of many non-target organisms by Ghosh (1986), rao etal (1986), Sprangce (1971), Sastry and Sidiqqi (1984), Sriniva Murthy etal (1985), Ghosh (1986), Rao etal (1986), Concluded that changes in physiological and behavioural activity may serve as indicators of estimating the level of sub lethal effects of pollution.

Pesticides usage while desirable for the pest control has resulted in unprecedented Chemical pollution. Matsumura (1975) affecting the non - target organisms. Pimevlal (1971) ware (1980). The Bio chemical responses of non - target organisms exposed to different pesticides have been well documented by Coppage and Mathews (1974), Corbelt (1974). Exposure to chemical pollutant elicits many molecular and biochemical changes in fish which precede cellular and Systematic dysfunctions. So that if appropriate parameters are monitored early warning signs of distress may be detected (Palmer 1976). Many pesticides have been reported that to produce a number of biochemical changes in fish both at lethal and more often at sub lethal levels. Changes in ion Concentrations, Organic constituents, enzyme activity, Endocrinal activity and as Chemoregulators in fish have been attributes to pesticides (Murty 1986).

The general classes of toxicants especially pesticides are extensively studied for Biochemical changes. Generally, neurotoxic pesticides of organophosphates and carbamates inhibits the enzyme cholinesterase and chlorinated Hydrocarbons inhibits only the total proteins and carbohydrates observed by (O'Brein 1967, Zitko 1975). In India, recently introduced Photostable synthetic Pyrethroids (Deltamethrin, Cypermethrin, Fenvalerate and permethrin) are used extensively as possible replacements of some of the organophosphate, Carbamate and organochlorine insecticide. Cypermethrin. [( Irs - Cyano 3 - phenoxybenzyl) - 2 -2 - dimethyl cyclopropane carboxylate)] is one of the promising synthetic pyrethroid which is used to control insect pests. The natural and synthetic pyrethroids are found to be more toxic to fish than organophosphates and carbamates (Braudbury etal., 1985). Though the toxic mechanism of pyrethroid is not yet fully elucidated, yet, it is suggested that pyrethroids definitely act on the nervous system (Chatterjee etal, 1986).

The lipophilicity of pyrethroids indicates that they are absorbed strongly by the gills of the fish even at low concentrations in water (Clark etal., 1985). The favourable ecotoxicological properties of cypermethrin are related to their ability to penetrate rapidly and interact with the site of action in fishes. ( Chatterjee etal., (1986). Since synthetic pyrethroid exposure is very common, it is imperative to assess the extent of biochemical changes caused by cypermethrin in vital organs viz. liver, brain, muscle and kidney of the fish. The biochemical changes in the tissues of the organic with reference to the toxicant have been considers by the toxicologists for the chronic effects. Due to chronic effects alterations of energy sources and cycles by the animals is inevitable due to stress and as a whole results change in the system (Martin etal., 1983).

Hence the present study has been under taken to study the effect of cypermethrin 10% E.C. formulation at sub-lethal and lethal concentrations to fish. The gill, Kidney, Liver, Brain and Muscle tissues of the exposed and control fish of cirrhinus mrigala (Hamilton) were selected for to study the Biochemical studies .

**MATERIALS AND METHODS**

The fish cirrhinus mrigala (Hamilton) were acclimatized at room temperature 28 +/- 2oc in the laboratory condition for 4 days. The acclimatized fish were exposed for 4 days to sub lethal (1/10 of 96 hrs LC50) and lethal concentrations of cypermethrin 10% E.C. formulation. The functionally active tissues such as gill Liver, Kidney, Brain and Muscle were taken out for estimation of glycogen The total glycogen was estimated by the method of Kemp etal (1954) and Total protein content was estimated by the modified method of Lowry et al (1951).

**RESULTS AND DISCUSSION**

The calculated values of glycogen and standard deviation along with percentage changes when exposed to cypermethrin 10%

E.C over control was given.

In the tissue of control fish, glycogen content was in the order of

Liver >Kidney>Brain>Muscle>Gill.

Among the various tissues, higher glycogen level was observed in Liver. Highest glycogen content of liver is acceptable due to its involvement in glycogen synthesis and utilization. Glycogen is the major storage form of carbohydrate in animals which occurs mainly in liver and muscle. Liver glycogen is largely concerned with storage and export of hexose units for maintenance of blood glucose. The function of muscle glycogen is to act as a readily available source of hexose units for glycolysis within the muscle itself (Harpler 1985). Though Brain tissue is metabolically active, lower glycogen contents was observed since it lacks the inherent potential to store glycogen and is dependent on blood glucose for all its metabolic activities (Lehninger 1983).

Exposed to sub lethal cypermethrin 10% E.C, the total glycogen level was found to decrease in all the tissues of Indian major carp cirrhinus mrigala. Maximum decrease was observed at 24 hrs exposure to the toxicant. The percent of decrease was in the order of 24 hrs Kidney > Liver > Brain > Muscle > Gill. Exposed to lethal concentration of cypermethrin 10% E.C, the glycogen value were found to decrease in most of tissues of fish cirrhinus mrigala. The percent of decrease was in the order f

Liver > Kidney > Brain > Muscle > Gill

The results indicate that the liver, a vital organ of carbohydrate metabolism was drastically effected by cypermethrin. In all most all the tissues of the organs i.e. Kidney, Liver, Brain, Muscle and Gill tested at sub lethal concentrations of cypermethrin, a decrease in glycogen value was noticed during the exposure periods. A fall in glycogen levels indicated its rapid Utilization to meet the enhanced energy demands in pesticide treated animals through glycolysis (or) hexose monophosphate path way (cappon and Nicholas, 1975). There is a progressive decrease in carbohydrate level in all the tissues when exposed to the lethal and sub lethal concentrations of Deltamethrin (Venkata Ratnamma, 2002). The decrease in tissue glycogen and protein content closely indicates its rapid utilization by respective tissues as a consequence of chloropyriphos stress (Tilak and Marina Samuel, 2001). The calculated values for total protein and percent change over control along with standard deviation are given in the tissues of control fish cirrhinus mrigala, the protein content was in the order of

Liver > Muscle > Brain > Gill > Kidney

The variation in distribution suggest gradual difference in metabolic calibres of various tissues. Maximum amount of protein was noticed in liver and minimum in kidney. The Liver is much in protein because of metabolic potential being oriented towards it and is seat for the synthesis of various proteins and also regulating centre of metabolism. In sub-lethal and lethal concentrations of cypermethrin, the total protein was found to decrease in most of the tissues. The leotropic series in terms of decrement in protein contents is

24 hrs Gill > Kidney > Muscle > Liver > Brain.

48 hrs Brain > Kidney > Gill > Muscle > Liver.

96 hrs Kidney > Muscle > Gill > Liver > Brain.

Exposure to sub-lethal doses of cypermethrin 10% E.C, the total protein content was found to decrease in all the tissues tested. Maximum decrease was noticed in brain and liver at 24 hrs exposure. Same trend was noticed in the same tissues even at 96 hrs exposure. At 48 hrs exposure, maximum decrease was observed in liver and muscle.

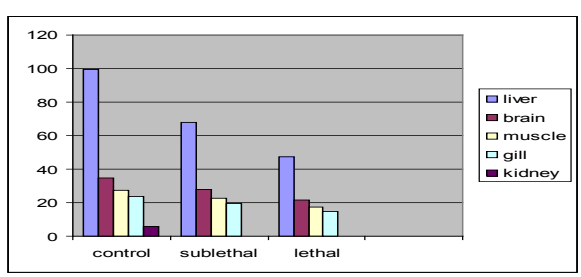
In the present study, decrease was more apparent in sub-lethal concentration than in lethal concentration. It may be due to detoxification of enzymes. The decrease tendency of total protein may also be due to the metabolic utilization of the Keto acids to gluconeogenesis path way for the synthesis of glucose (or) may be due to directing the synthesis of proteins from free amino acids (Schmidt and Nielson, 1975). The decrease in protein content was more due to break down rather than retarded synthesis which is supported by the findings of Radhaiah (1988). The protein break down and synthesis proceed simultaneously in all the tissues. Where synthesis is more, growth results. But during the pesticide stress, break down of protein occurs and acts as an alternative source of energy. Exposure of fish to toxic stress will stimulate protein metabolism (Kabeer et al, 1981). During protein metabolism, the removal of amino group from different amino acids was observed supporting the elevated levels of amino acids in the fish exposed to pesticide (Campbell and Davies, 1975). All these investigations support the present study of decreasing trend of proteins in the fish cirrhinus mrigala exposed to lethal and sub-lethal concentrations of cypermethrin 10% E.C.

**Table 4.1**
**ESTIAMATION OF GLYCOGEN**
Changes in the total glycogen contents and percent change over the (mg/g wet weight tissue) control in the different tissue of the fresh water fish cirhinus mrigala. Exposed to sub-lethal and lethal concentrations of cypermithrin ( 10% E.C.) for 24 hrs:

24 hrs;

| TISSUE | CONTROL | SUBLEATHAL | %CHANGE | LEATHAL | %CHANGE |
|---|---|---|---|---|---|
| LIVER | 99.6 +/-1.140 | 67.8 +/-0.84 | 31.9% | 47.6 +/-1.14 | 52% |
| BRAIN | 35 +/-1.140 | 27.8 +/-1.140 | 20.5% | 21.6 +/-1.14 | 38% |
| MUSCLE | 27.6 +/-0.836 | 22.4 +/-1.140 | 18.8% | 17.6 +/-0.836 | 36% |
| GILL | 23.8 +/-0.84 | 19.6 +/-1.140 | 17.6% | 15 +/-1.000 | 37% |
| KIDNEY | 5 .7 +/-0.052 | | | | |

.Resuls are the mean ;value of the five observations. Standard deviation is indicated (+/are significant of p<0.05



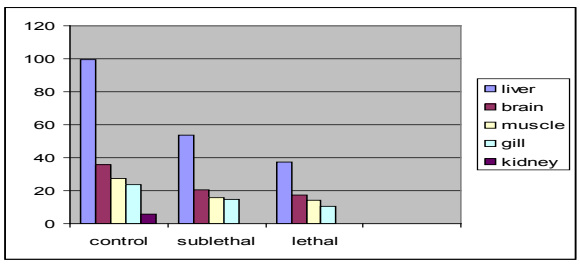

Changes in the total glycogen contents and percent change over the (mg/g wet weight tissue) control in the different tissue of the fresh water fish cirhinus mrigala. Exposed to sub-lethal and lethal concentrations of cypermithrin ( 10% E.C.) for 48hrs:

48hrs;

| TISSUE | CONTROL | SUBLEATHAL | %CHANGE | LEATHAL | %CHANGE |
|---|---|---|---|---|---|
| LIVER | 99.6 +/-1.143 | 59.6 +/-1.140 | 40% | 42 +/-0.707 | 51.8% |
| BRAIN | 35.6 +/-1.140 | 24 +/-0.7071 | 33% | 20.4 +/-1.140 | 43% |

| | | | | | |
|---|---|---|---|---|---|
| MUSCLE | 27.6 +/-0.836 | 17.6 +/-1.140 | 36.2% | 16.4 +/-1.140 | 41% |
| GILL | 23.8 +/-0.836 | 17.4 +/-1.140 | 27% | 13 +/-1.000 | 45% |
| KIDNEY | 5 .7 +/-0.023 | | | | |

.Results were the mean ;value of the five observations. Standard deviation is indicated (+/are significant of p<0.05

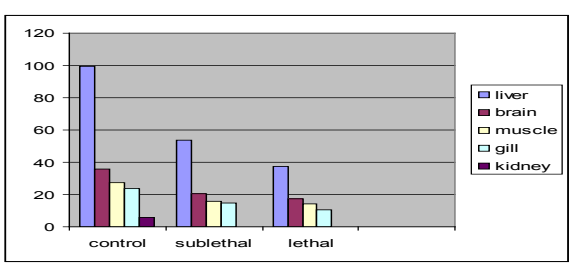



Changes in the total glycogen contents and percent change over the (mg/g wet weight tissue) control in the different tissue of the fresh water fish cirhinus mrigala. Exposed to sub-lethal and lethal concentrations of cypermithrin ( 10% E.C.) for 96hrs:

96hrs;

| TISSUE | CONTROL | SUBLEATHAL | %CHANGE | LEATHAL | %CHANGE |
|---|---|---|---|---|---|
| LIVER | 99.6 +/-1.143 | 53.6 +/-1.140 | 46% | 37.6 +/-1.140 | 62% |
| BRAIN | 35.6 +/-1.140 | 20.4 +/-1.141 | 43% | 17.6 +/-1.140 | 51% |
| MUSCLE | 27.6 +/-0.836 | 15.6 +/-1.140 | 43.4% | 14.2 +/-0.836 | 49% |
| GILL | 23.8 +/-0.836 | 14.8 +/-0.836 | 38% | 10.4 +/-0.84 | 56% |
| KIDNEY | 5 .7 +/-0.029 | | | | |

.Resuls are the mean ;value of the five observations. Standard deviation is indicated (+/are significant of p<0.05




**BIO CHAMICAL CHANGES IN PROTEIN ESTIMATION**

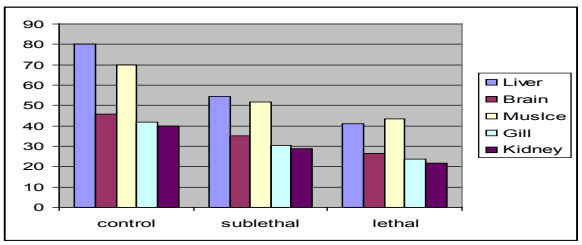
Changes in the total protein Contents (mg/gr wet weight of the tissue) and percent change over the control in different tissues of the fresh water fish cirrhinus mrigala.

Exposed to lethal and sub-lethal concentrations of cypermethrin (10%EC) for

24hrs.

| TISSUE | CONTROL | SUBLETHAL | %CHANGE | LETHAL | %CHANGE |
|---|---|---|---|---|---|
| LIVER | 80 +/-1.58 | 65.6 +/-1.140 | 18% | 47.6 +/-1.140 | 40.5% |
| BRAIN | 45.6 +/-1.120 | 3706 +/-1.140 | 17.5% | 28.6 +/-1.140 | 37.2% |
| MUSCLE | 70 +/-1.581 | 55.6 +/-1.140 | 20.5% | 48.4 +/-1.140 | 30.8% |
| GILL | 42 +/-1.582 | 31.6 +/-1.140 | 24.7% | 25.6 +/-1.140 | 39% |
| KIDNEY | 40 +/-1.5811 | 30.6 +/-1.140 | 23.5% | 22.4 +/-1.14 | 44% |

Resuls are the mean ;value of the five observations. Standard deviation is indieated as (+/-) value are significant of p<0.05.




**BIO CHAMICAL CHANGES IN PROTEIN ESTIMATION**

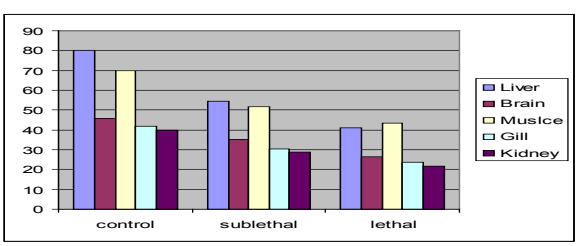
Changes in the total protein Contents (mg/gr wet weight of the tissue) and percent change over the control in different tissues of the fresh water fish cirrhinus mrigala.

Exposed to lethal and sub-lethal concentrations of cypermethrin (10%EC) for

48 hrs;

| TISSUE | CONTROL | SUBLEATHAL | %CHANGE | LEATHAL | %CHANGE |
|---|---|---|---|---|---|
| LIVER | 80 +/-1.58 | 61.6 +/-1.140 | 23% | 41. +/-1.140 | 48% |
| BRAIN | 45.6 +/-1.140 | 35 +/-1.581 | 64% | 26.4 +/-1.1401 | 23.2% |
| MUSCLE | 70 +/-1.58 | 51.8 +/-0.836 | 26% | 43.6 +/-1.140 | 37.7% |
| GILL | 42 +/-1.58 | 30.2 +/-0.835 | 28% | 23.8 +/-0.836 | 43.3% |
| KIDNEY | 40 +/-1.158 | 28.8 +/-0.836 | 28% | 21.6 +/-1.516 | 46% |

Resuls are the mean ;value of the five observations. Standard deviation is indieated as (+/-) value are significant of p<0.05.




**BIO CHAMICAL CHANGES IN PROTEIN ESTIMATION**

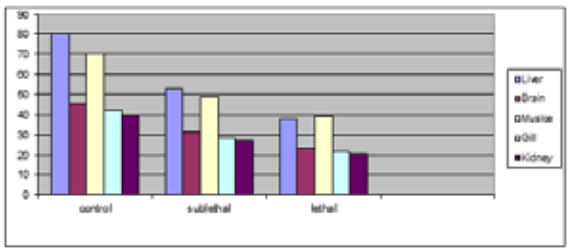
Changes in the total protein Contents (mg/gr wet weight of the tissue) and percent change over the control in different tissues of the fresh water fish cirrhinus mrigala.

Exposed to lethal and sub-lethal concentrations of cypermethrin (10%EC) for

96 hrs;

| TISSUE | CONTROL | SUBLEATHAL | %CHANGE | LEATHAL | %CHANGE |
|---|---|---|---|---|---|
| LIVER | 80 +/-1.58 | 57.6 +/-1.140 | 28% | 37.6 +/-1.140 | 53% |
| BRAIN | 45.6 +/-1.140 | 31.8 +/-0.836 | 30.2% | 23.4 +/-1.140 | 48.6% |
| MUSCLE | 70 +/-1.58 | 48.8 +/-0.836 | 32.2% | 39.4 +/-1.140 | 43.7% |
| GILL | 42 +/-1.58 | 28.2 +/-0.836 | 32.8% | 21.6 +/-1.140 | 48.5% |
| KIDNEY | 40 +/-1.158 | 24.4 +/-1.140 | 39% | 20.4 +/-1.140 | 49% |

Resuls are the mean ;value of the five observations. Standard deviation is indieated as (+/-) value are significant of $p<0.05$.




**Conclusion :**

The glycogen which is the major storage form of carbohydrates has reduced in all the tissues,but the decrease is highly significant in liver. Glycogen depliction is more prevalent under hypoxic conditions and situation similar to hypoxia or anoxia might be occurring in the tissues of fish. The glycogen level is attributed to the convertion of carbohydrates into amino acids. The total protein content was found to be decreased in all the tissues tested. Similar trend was observed in the same tissues even at 96 hrs of exposure. The apparent decrease of total protein content observed, under sub lethal exposure cypermethrin, it may be due to the detoxification of enzymes. The deceased tendency of total protein may also be due to metabolic utilization of the keto acids in gluconeogenisis path way of the synthesis of glucose or may be due to directing the synthesis of protein from free amino acids.

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