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EXPERIMENTS ON THE BIOCHEMICAL CHANGES IN THE FRESH WATER PRAWN *MACROBRACHIUM ROSENBERGII*, EXPOSED TO A SELECTIVE ORGANOPHOSPHATE INSECTICIDE.

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ABSTRACT
Environmental protection has attracted the attention of the wide cross-section of people all over the world, which has now become a global issue among scientists and researchers working in this area (Aruna Khare and Sudha Singh, 2002) pollution of water, is an important dimention of environmental degradation. Prawns are the important part of aquatic ecosystems and adversely affected by these pollutants and they are recognized as an excellent source of high quality protein (Gautam et al., 2002). As the prawns are economically important non-target organisms, they are quite sensitive to a wide variety of pollutants and are used as pollution indicators in water quality management. The prawns could serve as biological indicators to monitor are aquatic pollution. To known the adverse effects of pollutants on prawns, it is necessary to understand the mode of action of pollutants and more knowledge about biochemical changes brought about pesticides. The insecticidal treatment leads to biochemical alterations in the body tissues of prawns. Keeping this in view, in the present study has been designed to understand the alterations in the biochemical components of the tissue of a fresh water prawn, Macrobrachium rosenbergii made after the treatment of Monocrotophos.It was confirmed that biochemical alterations gave two conclusions firstly, prawns could be used as the best bio indicators of aquatic pollution. Secondly prawns have high nutritive values with more biological importance.

KEYWORDS: *Insecticide, Fresh water prawn, Pollution and Monocrotophos.*

INTRODUCTION

Persistent organic pollutants (POPS) are toxic substances released into the environment through a variety of human activities (RashmiSanghi 2001). They are very stable and long –lived chemicals that build up in the food chain and slowly poison animals and humans (Adhya et al.,2000) POPs are lipophilic and tend to accumulate and also magnify in the biochemical constituents of living beings (Nagabushanam et al.,1987). When they enter the living body, they don't leave it and are persistent. They are also semi-volatile which means that they can stay on the ground for a number of years and then be transported hundreds of miles away and be accumulated in another place until they eventually end up in animals and humans. Environmental protection has attracted the attention of the wide cross-section of people all over the world, which has now become a global issue among scientists and researchers working in this area. Pollution of water is an important dimention of environmental degradation. Pesticidal pollution constitutes the most dangerous health hazard apart from creating adverse effects on Prawn production (Holden,1972). They are quite sensitive to a wide variety of pollutants and are used as pollution indicators in water quality management.

Initially pesticides were handled for the total elimination of pest species but instead, the pest species have become resistant to them. The three basic kinds of organic pesticides are organophosphates, cerbamates, and chrorinated hydrocarbons. Pollutants such as pesticides are known to after the physiological and biochemical mechanism. (Mortimer&Cornel 1995; Casselderry et al., 1995). Indiscriminate use of pesticides, careless handling, accidental spoilage and discharge of untreated effluents of the aquatic bodies create serious problems to the aquatic biota such as fishes.Prawns are the important part of aquatic ecosystem and adversely affected by the these pollutants and they are recognized as an excellent source of high quality protein (Gautam et al.,2002).

As the prawns are economically important non-target organisms, they are quite sensitive to a wide variety of pollutants and are used as pollution indicators in water

quality management. The prawns could serve as biological indicators to monitor the aquatic pollution. To known the adverse effects of pollutants on prawns, it is necessary to understand the mode of action of pollutants and more knowledge about biochemical changes brought about pesticides. It is a known fact that biochemical functions get disturbed on exposure to pollutants. The aquatic organisms such as prawns and fishes are susceptible to pollutants. Proteins are important organic substance required by an organism in tissue building and repair. Amino acids help in toxic balance of cellular media and formation of many enzymes under extreme stress conditions proteins have been known to act as the energy suppliers in metabolic pathways and biochemical reactions (Arunakhare and SudhaSingh 2002).The pesticidal treatment leads to biochemical alterations in the body tissues of prawns. Keeping this in view, the present study has been designed to understand the alterations in the biochemical components of the tissue of a fresh water prawn, *Macrobrachium rosenbergii*made after the treatement of monocrotophos.

METHODOLOGY

Test Animals Collection And Maintenance

200 Female freshwater prawns, Macrobrachium rosenbergii were collected from the *kollidam*near *Annaikkarai*. They were brought to the laboratory and reared in glass aquaria, each containing 100 specimens (40x30x60cm). Each aquaria is filled with 5litres of freshly collected river water. Accumulation to the laboratory conditions (Temp. 26+ 10 C; PH6.4+ L.D=16.8). The prawn's water were provided with pieces of boiled fish meat daily and the medium water was renewed daily to avoid faecalmater.

Monocrotophos (MCP) technical grade was obtained from keycer Agrochemicals Ldt. Salem, India. 1% stock solution was prepared by diluting with double distilled water varied **tes t** concentration viz. 5,10,15,20, and 25ppm were prepared according to method described by laughlin et al.,(1983). Acclimated *M. rosenbergii*of uniform body size(14-16mm) and weight (28-32g) were chosen to determine lc50 value. From the mortality values at different concentration of MCP (5 to 25ppm) a concentration rendering 50% of the population dead i.e. lc50/96h was computed, adopting a method densed by Litchfield and Willicoxon (1949). The results were confirmed repeating the expriments thrice.

Biochemical assay

Healthy *M. rosenbergii*of uniform body dimensions were chosen .two groups were formed. One group was considered as control and another group as experimental. The experimental group was exposed to acute (short term) treatments the acute treatment consisted of exposure to 9.5ppm (1/2 of 96h lc50)and chronic treatment consisted of exposure to 1.9 ppm (MCP)twenty test prawns in each set of both control and experimental groups were maintained simultaneously. The medium water and monocrotophos (MCP) Consentration were renewed daily. the prawns were sacrificed at interval of 24h in the acute and at intervals of 7.14 and 21days in the chronic treatment groups for biochemical studies. The total protein content was estimated using Folin-Lowry method (Lowry et al.,1951) and total lipids was estimated by in method den so by Barnes &Biochstoch(1973). All experiments were repeated three times. Mean and standard deviation (+S.D.)Were calculated and the significance of the difference was calculated following students't'. All values are statistically significant at $p<0.05$(Snedecor and Cochran, 1967).

RESULTS

Effect of monocrotophos (MCP) toxicity on protein content in selected body tissues of freshwter prawn, *macrobrachium rorenbergii*(Tables1 and 2).

Table 1: Protein content in selected body tissues of control and acute Monocrotophos exposed fresh water prawns, Macrobrachium rosenbergii

Treatment	Body tissue	Protein content (mg/100mg wet weight)			
		24h	48h	72h	96h
Control	Gills	16.79+0.58	18.85+0.56	19.75+0.32	20.86+0.37
	Hepatopancreas	24.91+0.82	25.01+0.65	23.36+0.74	22.34+0.87
	Muscle	27.71+0.67	33.91+0.65	32.79+0.67	32.12+0.73
Acute exposure MCP	Gills	15.3+0.61(-9.43)	16.5+0.96(13.16)	17.32+0.74(12.96)	18.3+0.54(12.89)
	Hepatopancreas	22.9+0.97(-8.41)	22.6+0.96(10.03)	20.9+0.57(-11.00)	19.9+0.82(-11.43)
	Muscle	23.3+1.02(16.51)	29.4+0.73(-13.70)	25.6+0.85(22.61)	23.5+0.71(-27.69)

Each value is mean +S.D Of three individualobservation
All values are statistically significant at P<0.05
Values in parenthese indicate changes over respective controls
Acute exposure (Conc.in ppm) =9.5MCP

Table 2: Protein content in selected body tissues of control and acute chronic monocrotophos exposed fresh water prawns, Macrobrachium rosenbergii

Treatment	Body tissue	protein content (mg /100mg wet wight)		
		7d	14d	21d
control	Gills	16.58+0.60	18.3+0.90	20.74+0.61
	Hepatopancreas	25.56+0.5	31.51+0.32	33.24+0.63
	Muscle	30.62+0.61	34.10+0.71	36.19+0.48
Acute exposure MCP	Gills	14.1+0.54(15.91)	12.5+0.57(33.56)	11.2+0.93(48.32)
	Hepatopancreas	22.8+0.61(11.23)	23.5+0.74(26.25)	25.54+0.61(23.88)
	Muscle	22.9+0.97(26.06)	23.7+0.61(31.41)	29.4+0.59(-19.29)

Each value is mean + S.D of three individual observation
All values are statistically significant at $p<0.05$
Values in parentheses indicate persent changes over respective controls
chronic exposure (conc.in ppm)=1.9 mcp

The protein content decreased in fresh water gills, hepatopancreas and muscle exposed to sublethalconcentrtion 9.5 ppm (acute) and 1.9 ppm (chronic) of monochrotophos. The results are given in tables 1&2. The hepatopancrease protein content exposed to 9.5ppm nd 1.9ppm were dramatically reduced to about 11.43% and 26.25% respectively. At 9.5ppm and 1.9 ppm exposure concentrations there was a significnt ($p<0.05$) reduction in the mean protein content in gills, hepatopancreas and muscle compared to mean protien estimate from the controls. maximum percent decrease for the gill protein content was 48.32% followed by a maximum decrease, 31.41%in muscles and 26.25 in hepatopancrease tissues of *M.rosenbergii*at chronic exposure i.e over 96h exposure.

Effect of monocrophos (MCP) toxicity on lipid content in selected body tissues of freshwater prawns, *Macrobrchiumrosenbergii*(Tables3&4).

Table 3: Lipid content in selected body tissues of control and aqutemonocrotophos exposed fresh water prawns, Macrobrachium rosenbergii

Treatment	Body tissue	lipid content (mg/100mg wet weight)			
		24h	48h	72h	96h
Control	Gills	2.2+0.02	3.3+0.05	2.2+0.02	2.5+0.01
	Hepatopancreas	4.1+0.02	4.5+0.02	3.6+0.01	3.1+0.01
	Muscle	2.8+0.03	2.6+0.01	2.4+0.02	2.5+0.01
Acute exposure MCP	Gills	2.7+0.03(-22.72)	2.5+0.01(-34.78)	2.7+0.01(-22.72)	2.8+0.01(-28.0)
	Hepatopancreas	2.1+0.02(-38.23)	1.8+0.06(-48.57)	1.7+0.06(-52.77)	1.0+0.06(-70.58)
	Muscle	2.0+0.01(+11.12)	1.9+0.04(+18.75)	1.8+0.05(+28.57)	1.6+0.07(+6.67)

Each value is mean + S.D of three individual observation
All values are stastisticaly significant at $p<0.05$
values in parentheses indicate persent changes over respective controls
chronic exposure (conc.in ppm)=9.5 mcp.

Table 4: Lipid content in selected body tissues of control and chronic monocrotophos exposed fresh water prawns, Macrobrachium rosenbergii

Treatment	Body tissue	Total lipid content (mg/100mg wet weight)		
		7d	14d	21d
control	Gills	2.6+0.03	2.5+0.02	2.4+0.03
	Hepatopancreas	3.9+0.01	3.8+0.03	3.7+0.02
	Muscle	1.9+0.01	1.8+0.02	1.7+0.01
Acute exposure to MCP	Gills	1.0+0.05(38.46)	1.9+0.02(-44.0)	1.8+0.01(-25.05)
	Hepatopancreas	2.5+0.01(-35.89)	2.3+0.01(-39.47)	1.7+0.01(-54.05)
	Muscle	2.1+0.03(-10.52)	2.0+0.07(+11.12)	1.9+0.01(+11.76)

Each value is mean + S.D of three individual observation
All values are stastisticaly significant at $p<0.05$
values in parentheses indicate persent changes over respective controls
chronic exposure (conc.in ppm)=1.9 mcp

The protein content decreased in fresh water prawns gills, hepatopancreas and muscle to sublethal concentrations 9.5ppm (acute) and 1.9ppm(chronic) of monochrophos. The results are given in Tables 1&2. The hepatopancrease protein content exposed to 9.5ppm and 1.9ppm were dramatically reduced to about 11.43% and 26.25% respectively. At 9.5ppm and 1.9ppm exposure concentrations there was a significant ($p<0.05$) reduction

in the mean protein content in gills, hepatopancrease and muscle compared to mean protein estimate from the controls. Maximum percent decrease for the gill protein content was 48.32% followed by a maximum decrease, 31.41% in muscles and 26.25in hepatopancreas tissues of *M. rosenbergii*at chronic exposure i.e over 96h exposure. Effect of Monocrotophos (MCP) toxicity on lipid content selected body tissues of freshwater prawns, Macrobrachium rosenbergii (Tables3&4).

In Monocrotophos acute (9.5ppm) exposed *M.rosenbergii*, the lipid content decreased in all tissues except the muscles.

The lipid content in the hepatopancreas declined from 3.4to 2.1; 3.5to 1.8; 3.6 to 1.7 and 3.4to 1.0 at 24, 48, 72, and 96h monocrotophos acute exposure respectively. A maximum depletion in percentage of lipid content was 70.58 in hepatopancreas, followed by 34.78% in gills. There was an increase in the lipid content in the muscle from 2.0 to 1.8; 1.9to 1.6; 1.8to 1.4 and 1.6to 1.5 at 24, 48,72 and 96h acute exposures. The average lipid content in gills and hepatopancreas decreased from 2.31.7($p<0.05$) and from 3.5 to 1.7 at acute (9.5ppm)treatement. In Monocrotophos chronic treated (1.9ppm) fresh water prawns, a fall in lipid content in the hepatopancreas and gills with greater decrease (54.05%) in the former 3.9 to 2.5 ($p<0.05$);3.8 to 2.3 ($p<0.05$) and 3.7to 1.7($p<0.05$) at 7,14 and 21 days exposure they was a slight increase 11.76%in lipid content in muscle tissue as in acute exposed *M.rosenbergii*.

DISCUSSION

In recent years, the aquatic bodies such as rivers, seas, estuaries get contaimined by a variety of chemicals such as heavy metals, pesticides hydro carbons etc., Among the pollutants pollution due to pesticides considerable attention because of harmful effects on beneficial organisms (Raja et al., 2001). Therefore the present study was undertaken to investigate the biochemical mechanism underlying the chronic effect of MCP in freshwater prawns, Macrobrachium rosenbergii. It has a good consumer preference because of its high nutritive value i,e., protein and lipids consititude the cheif biochemical constituents in its body tissues various account for 25% of total global protein supply.

The present study revealed that MCP showed differential toxicity in different tissues viz. gills, hepatopancreas and muscles of *M. rosenbergii*both at acute and chronic treatments. MCP produced a time - does dependent decrease in the biochemical substances namely total proteins and total lipids, Similar observation have been reported by Karuppasamy et al.,(2001)in their studies. The observed depletion protein content in tissues was recorded in this order viz Gills >Hepatopancreas> Muscle and the maximum depletion in muscle both at acute and chronic treatments indicated that muscles contained more proteins than the other tissues. The increased protease activity levels in the tissues of *M.rosenbergii*during pesticides exposures revealed that the protein utilisation was very high in proteolytic pathway. The might have been fed into TCA cycle throuth aminotransferase system to cope with excess demand of energy caused by MCP stress as suggested by Jha (1991).

There was a possibility for the decreased protein contents in the tissues may also be due to tissues description or necrosis or disrturbance of cellular function and consiequent impairment in protein synthetic machinery. From the results of present study, it was concluded that the hepatopancrease was the main target tissues and severely affected both at acute and chronic exposures of MCP. A maximum depletion of 25.43% and 45.63% was noticed in treated prawns and values were statistically significant ($P<0.05$) over the control tissue. Hepatopancrease in the cheif metabolic organ for both protein and lipid macromolecules and could be compaered functionally to the liver of higher animal. During the exposure periods of MCP, proteins from hepatopancrease was *utillition*in greater quantities to provide energy and therefore maximum decreases in protein content in hepatopancreas was evident in the present study (Table 1 and 2).

Another significant result obtained in the present study was depletion in the lipid content of tissues in *M. Rosenbergii*both at short - term and long -term exposer (Chronic). The biochemiccal of vital tissues is assuming an important role as a biochemical tool for understanding environmental toxicity, as the toxicity induced biochemical alteration precede the onset of morphological and pathological manifestation of the toxicityThe lipid content is an important organic consituent of the animal tissues and plays an important role in energy metabolism. Lipids are also important in cellular and sub cellular membranes (Sheela & Muniandy 1992) The fall in lipid content levels during MCP toxic exposure periods (acute and chronic) may be due to increased catabolic and decreased anabolism of lipids.

The reducing trend in all tissues except in muscles of *M. rosenbergii* may be attributed to metabolic utillization of fatty acids for the synthesis of sterols of the main enhance of osmo and ionic regulation. When the animal, *M. rosenbergii*is under stress due to MCP toxicity, the energy rich compounds such as lipids stored in vital organats namely gills and hepatopancreas get oxidized rapidly by lipases and therefore, there is a decrease in the lipid content of gills and hepatopancreas alone due to the increased lipase activity. In time dependent experiments (Acute and chronic), the MCP exposure altered the protein and lipid contents either by interfering directly their storage or by an increased expenditure.

CONCLUSION

The biochemical alteration caused by MCP in freshwater prawns may be used as valuable index for determining

the environmental pollution by pesticides. Furthermore, in the protein and lipid due to MCP give the nutritive index of prawns.

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