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Research Article

EFFECT OF PROFENOFOS AN ORGANOPHOSPHATE ON PROTEIN LEVELS IN SOME TISSUES OF FRESH WATER FISH *LABEO ROHITA* (HAMILTON)

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ABSTRACT

An insecticide profenofos used on a wide variety of crops to control many pests but mainly Lepidoptera and mites. The objective of present study, an attempt was made to examine the sub lethal toxic effect of profenofos insecticide on protein metabolism in the different tissues of freshwater fish *Labeo rohita.* The LC_{50} values determined for profenofos at 24 hrs were 100µg/l respectively. The $1/10^{th}$ of 96 hrs, LC_{50} value was selected as sub lethal concentrations. The fish were exposed to sub lethal concentrations for 1, 4 and 8 days and the changes in the protein levels of vital organs such as brain, gill, kidney, liver and muscle were studied by Lowry *et al.,* method. The results in sub lethal exposure minimum percentage of protein depletion was (13.84%) in kidney and maximum percentage was (24.21%) in Liver for 1 day , minimum depletion was (9.19%) in gill and maximum(22.38%) for 4 days, minimum percentage (7.5%) in brain and maximum (19.34%) in Liver for 8 days were observed at different days when compared with controls. The changes and decrease in protein level might also be due to inhibition of metabolizing enzymes by administration of toxicants.

Keywords: Profenofos, *Labeo rohita*, Protein, Depletion and liver.

INTRODUCTION

The increasing use of pesticides causes chemical pollution results potential health hazards to live stock, especially to fish, frogs, birds and mammals. Since majority of pesticides are known to bio accumulate in the lipid tissues of fish and other animals, transfer via to human bodies ,the grave risk to the health of the people who consume these fish seems to considerable. When the pesticides come in contact with internal organs, irreversible changes in metabolic activities, many pesticides have been reported to produce a number of biochemical changes in fish at sub-lethal levels. The present study has been undertaken to understand the biochemical alterations induced by (Profex) profenofos 50% EC on exposure to sub-lethal concentrations on fish *Labeo rohita* in different tissues exposed. The calculated values for total proteins and percent changes over control along with standard deviation, in contrast to the control.

MATERIALS AND METHODS

The fish *Labeo rohita* measuring 6 to 8 cm in length and 6.5 to 7.5 gm in weight irrespective of the sex were used in the experiment. Fish were washed with 0.1% $KMnO_4$ solution to avoid dermal infection. All the precautions laid down by APHA *et al.,* (1998) are followed, for maintaining the fish. The fish were exposed to organ phosphorus pesticide Profenofos 50% EC to sub lethal $1/10^{th}$ 96 hrs lc_{50} value, i.e. 100µg/l concentrations for 4 and 8 days. If mortality occurred during the experimental period, dead fish were removed immediately to avoid depletion of dissolved oxygen (DO) level which adversely affects other fish (Schreck and Brouna, 1975). The vital tissues like muscle, brain, liver, gill and kidney of the fish were taken for the estimation of Total proteins.

Estimation of total protein

Total protein content was estimated by the modified method of Lowry *et al.,* (1951). 5% homogenates of gill, muscle and brain and 2% homogenates of liver and kidney were prepared in 5% trichloroacetic acid and centrifuged at 3000 rpm for 10 minutes. The supernatant was discarded. The suspended protein residue was dissolved in 1 ml of 1N NaOH. From this 0.2 ml of the extract was taken into the test tube and 5 ml of alkaline copper solution (50 ml of 2% Na_2CO_3 and 1ml of 0.5% CuSO4. $5H_2O$ in 1% sodium potassium tartrate) was added. The contents were mixed well and allowed to stand for 10 minutes. To this 0.5 ml of 50% folin phenol reagent (diluted with distilled water in 1:1 ratio) was added. After 30 minutes, the optical density was measured at 540 nm in a spectrophotometer (ELICO Model SL171) against a blank. The standard graph was plotted by the method of Lowry *et al.,* (1951) with bovine serum albumin supplied by Sigma chemical Company, U.S.A. The values were expressed as mg/gr wet weight of the tissue.

RESULTS

The calculated values for total proteins and percent changes over control along with standard deviation are given in Table 1,2 and 3 are graphically represented in Fig 1,2and 3. In the control fish, *Labeo rohita* the total protein content is in the order of: Muscle > Liver > Brain > Gill > Kidney. The variation in distribution suggests differences in metabolic caliber of various tissues. The present trend in the tissues is justifiable in the wake of mechanical tissue of muscle intended for mobility and does not participate in metabolism. The liver is also much in proteins because of metabolic potential being oriented towards it and is the seat for the synthesis of various proteins besides being the regulating center of metabolism. Under exposure to sub lethal concentrations of Profenofos percent depletion of total protein content in the test tissues of the fish *Labeo rohita* is in the order of: Liver > Muscle > Gill > Kidney > Brain .

The results in sub lethal exposure maximum percentage was (24.21%) in Liver and minimum percentage of protein depletion was (13.84%) in kidney and for 1 day, maximum (22.38%) minimum depletion was (9.19%) in gill for 4 days, maximum (19.34%) in Liver minimum percentage (7.5%) in brain and for 8 days were observed at different days when compared with controls.

Table1: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub lethal concentrations of Profenofos for 24 h (1 day)

Tissues	Dose (µg /L)	Control	Sub lethal	% Change
Brain	1 0	118.2± 0.015	96.62± 0.001	18.25
Gill	10	109.3± 0.001	88.34± 0.002	19.17
Kidney	10	136.2± 0.008	117.34± 0.003	13.84
Liver	10	177.26± 0.469	134.33± 0.061	24.21
Muscle	10	143.4 ± 0.005	111.1± 0.005	22.52

Values are the means of five observations: (±) indicates the standard deviation values are significant at* $p < 0.05$.

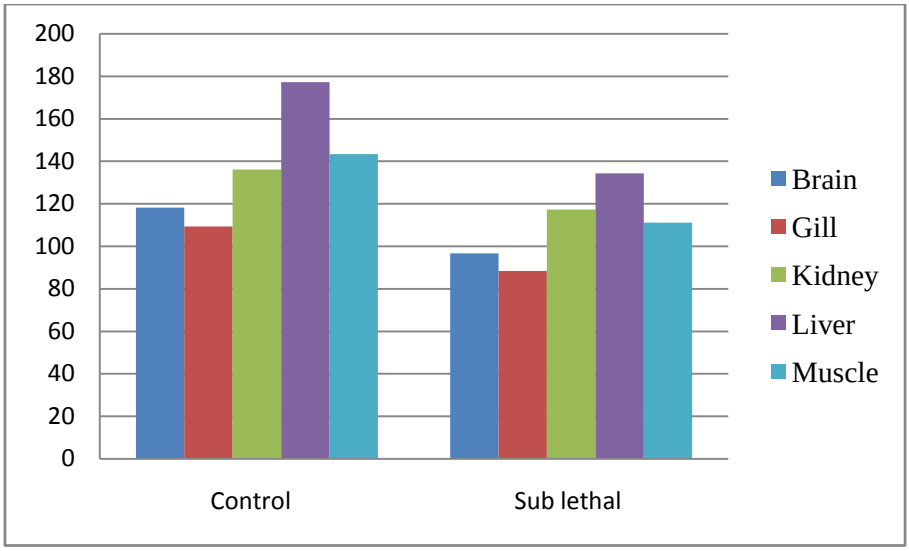

Fig. 1: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub lethal concentrations of Profenofos for 24 h (1day).

Table 2: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub-lethal concentrations of Profenofos for 4 days.

Tissues	Dose (µg /L)	Control	Sub-lethal	% Change
Brain	1 0	96.4± 0.003	86.1± 0.012	10.68
Gill	10	90.3± 0.004	82± 0.003	9.19
Kidney	10	102.3± 0.002	90.2± 0.006	11.82
Liver	10	124.2± 0.0012	96.4±0.0015	22.38
Muscle	10	103.2 ± 0.0015	83.3±0.0014	22.52

Values are the means of five observations: (±) indicates the standard deviation values are significant at* $p < 0.05$

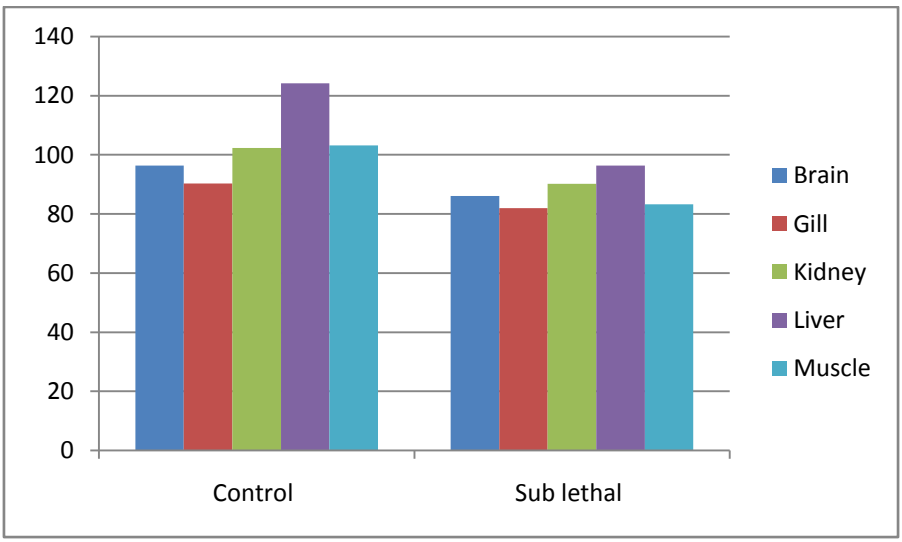

Fig. 2: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub lethal concentrations of Profenofos for 4 days.

Table 3: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub-lethal concentrations of Profenofos for 8 days

Tissues	Dose (µg /L)	Control	sub-lethal	% Change
Brain	1 0	82.6± 0.002	76.4± 0.002	7.5
Gill	10	80.1±0.02	73.2±0.02	8.61
Kidney	10	85.3±0.01	74.3±0.01	12.89
Liver	10	92±0.01	74.2±0.03	19.34
Muscle	10	84.2 ± 0.03	70.2±0.002	16.62

Values are the means of five observations: (±) indicates the standard deviation values are significant at *$p < 0.05$

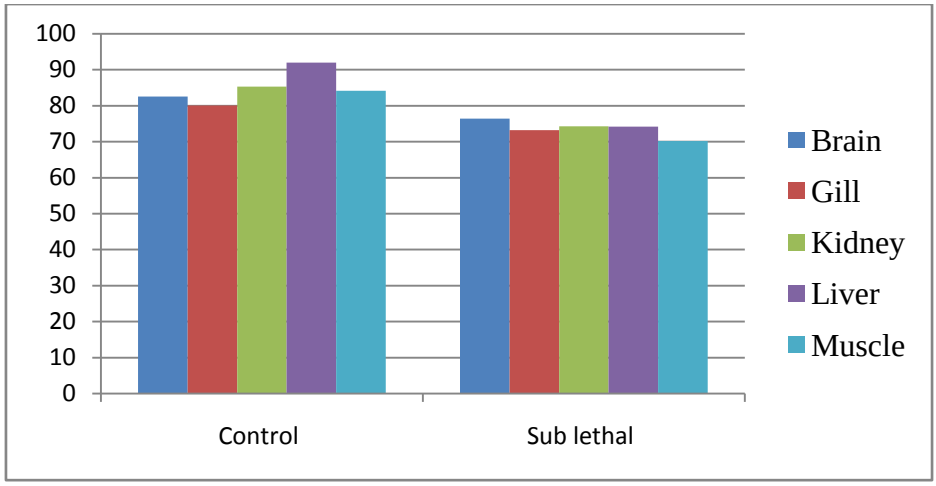

Fig. 3: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub lethal concentrations of Profenofos for 8 days

DISCUSSION

Proteins are indeed of primary and paramount importance in the living world not only because of their peculiars but also because of the fact that they appear to confer their biological specificity among various type of cells (Bhushan *et al*., 2002). According to Venkataramana *et al.,* (2006) *Glossogobius giuris* when exposed to sub lethal concentrations of (0.05, 0.25 and 0.5 mg L^{-1}) Malathion for short duration of 24 to 96 hr. The cardiac muscles showed significant decrease in levels of proteins after treatment with 0.5 mg L-1concentration. The decreased trend of the protein content as observed in the present study in most of the fish tissues may be due to metabolic utilization of the ketoacids to gluconeogenesis pathway for the synthesis of glucose; or due to the directing of free amino acids for the synthesis of necessary proteins, or, for the maintenance of osmotic and ionic regulation (Schmidt Nielson, 1975).

Vishal Tiwari (2004) exposed *Cirrhina mrigala* sub lethal concentration of 2 mg L^{-1} of malathion for 7, 14 and 21 days and observed decrease in total, structural and soluble proteins and an increase in free amino acids and protease activity levels in contrast to protein decrement noticed in 7 and 14 days of exposure. But on 21 days of exposure all the values came to normally. The restoration of different protein fraction to normalcy indicates that after 14 days of exposure there seems to exist an oscillatory phase in protein turnover towards a more synthetic phase leading to the establishment of recovery and adaptation phenomena. Aruna Khare *et al.,* (2000) observed that the sub lethal concentrations of Malathion showed a significant increase in total protein content in kidney of exposed fish, *Clarias batrachus* during the first week and there after a gradual decrease in protein content was observed in the later periods of exposure.

Jha and Verma (2002) studied the impact of the pesticidal mixture (Endosulfan; Malathion and Agrafun 1:1:1) on total protein content in the stomach, intestine and ovary of the fish *Clarias batrachus* acute (96 hr) sub chronic (7 & 14 days) and chronic (21 days) exposures and found that reduced protein profiles in the exposed fish were dose duration dependent.

Tilak *et al*., (2003) reported a decrease in protein content in *Channa punctata* exposed to sub lethal concentration of fenvalerate. The similar decreasing trend in total proteins was also reported in the liver, brain and gill tissues of *Catla catla* under sub lethal and lethal concentrations of fenvalerate by Anita Susan *et al*., (1999). A significant decrease was reported in the protein content in almost all tissues in *Ctenopharyngodon idella* by Tilak and Yacob, (2002). Tilak *et al*., (2001d) reported that when the freshwater fish, *Labeo rohita* was exposed to sub lethal concentrations of pesticide mixture of monocrotophos and fenvalerate, the protein content was decreased. K.Suneetha *et al.*, reported Effects of Endosulfan and fenvalerate on carbohydrate metabolism of the freshwater fish, *Labeo rohita* (Hamilton) by Int. J. Pharm. Sci, 2011.

Decline in muscle protein profile in early period of suggests stress in metabolic process and impairment of protein synthesis machinery in fish; the catabolic process was initiated by increased proteolysis that led to rapid decline in protein concentration to meet the energy demand in extremely stressful environment Baruah *et al.,* (2004). Hypoprotenemia was observed in the selected tissues of fish exposed to organophophorus pesticides by many investigators, thus supporting the findings of the present study (Ramalingam, 1985 and Deva Prakash Raju, 2000).

The decrease in protein content of quinalphos intoxicated fish in the present study also indicates the physiological adaptability of the fish to compensate for pesticide stress. To overcome the stress the animals require high energy. This energy demand might have led to the stimulation of protein catabolism. The present analysis also coincides with the findings of Sastry and Siddiqui (1984) who reported that the protein content was decreased in liver, muscle, kidney, intestine, brain and gill when *C. punctatus* treated with quinalphos. Similar reports of Dura raj and Selvarajan (1992), Anusha Amali *et al.,* (1996); Yeragi *et al*., (2000) and Tilak *et al*., (2005) support the present data. The changes and decrease in protein level might also be due to inhibition of metabolizing enzymes by administration of toxicants. Several other investigations also revealed a decrease in protein profiles with organophosphate compounds. All these investigations support the present study of decreasing trend of proteins in the tissues of the fish *Labeo rohita* exposed to Profenofos.

CONCLUSION

The present work indicates that profenofos caused alterations in the protein metabolism of fish *Labeo rohita*, treated fish tissues showed more decrement in protein levels this may be due to more pesticidal stress. The altered mobility and low content of proteins reflects a change in the rate of synthesis and degradation of protein, lowered working capacity under the impact of accumulation of pollutants leading to an alteration in function indicating the vulnerability of the organ.

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