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RESEARCH ARTICLE FLUBENDIAMIDE A PHTHALIC ACID DIAMIDEEFFECT ON PROTEIN METABOLISM OF FRESHWATER FISH *LABEO ROHITA* (HAMILTON) ¹Nirmalakallagadda And ²Venkata Rathnamma.

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ABSTRACT

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FlubendiamideA New lepidopteron Insecticide is used on to control important lepidopteron pests. The objective of present study, an attempt was made to examine the sub lethal toxic effect of flubendiamide insecticide on protein metabolism in the different tissues of freshwater fish *Labeorohita*. The LC ₅₀ values determined for flubendiamide at 96 hrs were selected. The fish were exposed to sublethal concentrations for 1, 5 and 10th day and the changes in the protein levels of vital organs such as brain,gill, kidney, liver and muscle were studied by lowry*et al.*, (1951)method. The results in sublethel exposure minimum percentage of protein depletion in Kidney (10.46%) and maximum percentagein Liver (25.42%) For 1 day, minimum depletion in kidney (6.53%) and maximum in Liver (13.49%) for 5 days, minimum percentage of Liver (6.93%) For 10 days were observed when compared with controls. The changes and decrease in protein level might also be due to inhibition of metabolizing enzymes by administration of toxicants.

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INTRODUCTION

Rapid industrialization and increase in human population, the pollution of water bodies has become a universal phenomenon in the present day world (Bela, 2008). The important sources of water pollution are industrial effluents, domestic, sewage, drainage and pesticides, which pollute the river and major water sources (Maruthanayagam, 2004). Most of the pesticides find their way into rivers, lakes and ponds through flowing water and have been found to be highly toxic not only to fishes but also to the organisms which contribute to the food chain comprising of fishes (Anees, 1975).

The nutritional value of different species of fish and shellfish depend on their biochemical components such as protein, carbohydrate and lipids. These proximate components could serve as sensitive indicators for detecting potential adverse effects, particularly the early events of pollutant damage because their alterations appear before the clinical symptoms produced by the toxicant(Rao, J.V.,2006).

It is important that potential effects of acute and chronic concentrations of pollutants on proximate composition are determined and interpreted to delineate mechanisms of pollutant action and possibly ways to mitigate adverse effects (Matos, P., 2007). The present investigation was studied on effect of Flubendiamide onsublethal concentrations protein metabolisam of *Labeo rohita* freshwater fish, Flubendiamide is the first member of a new chemical class, it is a phthalic acid diamide, with a novel chemical structure. The structure consists of three parts; a heptafluoroisopropyl group in the anilide moiety, a

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sulfonylalkyl group in the aliphatic amide moiety, and an iodine atom at the 3-position of the phthalic acid moiety.

MATERIALS AND METHODS

The test fish Labeo rohita measuring 6 to 8 cm in length and 10 to 12 gm in weight were collected from the unpolluted ponds. They were acclimatized to the laboratory conditions and water was renewed every day to provide freshwater, rich in oxygen, fed daily with commercial fish pellets having around 50% protein content and chopped sheep liver once in two days, and allowed to acclimate for 15 days at $28+2^{\circ}$ c. Fish were washed with 0.1% KMnO4 solution to avoid dermal infection. All the precautions laid down by APHA et al., (1998) are followed, for maintaining the fish. Such acclimatized fish were exposed for 96 hours to a phthalic acid diamide pesticide flubendiamide 480sc to lethal (LC_{50}) and sub lethal concentrations. If mortality occurred during the experimental period, dead fish were removed immediately to avoid depletion of a vital tissues like muscle, brain, liver, gill and kidney. Total proteins were estimated by the method of Lowry et al., (1951).

 Table1 Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub lethal concentrations of flubendiamide24 h (1 day)

Tissues	Dose (µg /L)	Control	Sublethal	% Change
Brain	11	116.2	99.7±0.057	11.58%
Gill	11	107.3	97.5 ± 0.018	10.46%
Kidney	11	135.2	92.8 ± 0.010	12.54%
Liver	11	175.2	145.1 ± 0.033	25.42%
Muscle	11	143.4	125.6 ± 0.023	18.01%

Values are the means of five observations: (±) indicates the standard deviation

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



Fig. 2: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub lethal concentrations of flubendiamide 5days

Under exposure to sublethal concentrations of flubendiamide the percent depletion of total protein content in the test tissues of the fish *Labeo rohita* is in the order of: Liver > Muscle > Brain >Gill> kidney.



Fig. 3: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub lethal concentrations of flubendiamide for 10 days

The results in sub lethal exposure in liver maximum percentage of protein depletion was observed (25.42%), and in kidney minimum percentage of protein depletion was (10.46%) for 1 day, observed liver maximum percentage of protein depletion was (13.49%) and in kidney minimum of protein depletion was (6.53%) for 5 days, brain maximum percentage of protein depletion was (6.93%) and minimum percentage of protein depletionin gill (5.22%) for 10th day where as inwhen compared with controls.

Proteins are heterogeneous group of macro molecules having diverse physiological functions (Lehninger, 1999).Hence, protein profiles can be considered as a factor in assessing.

Table2: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub leth
concentrations of flubendiamide for 5 day

Tissues	Dose (µg /L)	Control	Sub lethal	% Change		
Brain	11	95.4	85 ± 0.040	8.10%		
Gill	11	89.3	73.2 ± 0.008	6.53%		
Kidney	11	101.3	71.7 ± 0.008	7.26%		
Liver	11	123.2	109.5 ± 0.024	13.49%		
Muscle	11	102.2	91.1 ± 0.015	9.31%		

RESULTS AND DISCUSSION

The calculated values for total proteins and percent changes over control along with standard deviation were 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: Liver > Muscle > Kidney> Brain > Gill. Variation in distribution suggests differences in metabolic caliber of various tissues of freshwater fish *Labeo rohita*. Liver is also much in protein 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. the physiological status of a tissue or an animal as a whole (Harper, 1992).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. Decrease in protein levels due to severity of pesticides action was also reported by Arsta-Tazeen *et al.*,(1996).

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 Keto acids 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

Table3: Change in the Total protein content (mg/gr wet weight of the tissue) in different tissues exposed to sub lethal

concentrations of flubendiamide for 10 days

Tissues	Dose(µg /L)	Control	Sub lethal	% Change			
Brain	11	81.6	79.7 ± 0.036	6.50%			
Gill	11	79.1	66 ± 0.011	5.22%			
Kidney	11	84.3	66.6 ± 0.012	5.61%			
Liver	11	91.1	76.1 ± 0.015	6.93%			
Muscle	11	83.2	75.3 ± 0.006	6.26%			

the maintenance of osmotic and ionic regulation (Schmidt Nielson, 1975).

Tilak *et al.*, (2003) reported a decreased protein content in *Channa punctatus* exposed to sub lethal concentration of fenvalerate different exposure periods. Similar decreasing trend was observed in total proteins of different tissues like liver, brain and gill tissues of *Catla catla* under sublethal and lethal concentrations of fenvalerate by (Anita Susan *et al.*, 1994).

The biochemical processes represent the most sensitive and relatively early events of pollutant damage. Thus, it is important that pollutant effects be determined and interpreted in biochemical terms, to delineate mechanisms of pollutant action, and possibly ways to mitigate adverse effects.

In connection to this, the phthalic acid diamide significantly altered the level of total protein, in liver and muscle tissues of *Channa punctatus* (Durga and Veeraiah, 2002; Tilak *et al.*, 2005; Ganeshwade *etal.*, 2012; and Nagaraju *et al.*, 2013).The effects of Neem-based pesticides on non-target organisms have been studied in terrestrial ecosystems; however, little attention has been focused on aquatic environments during chronic periods of stress, proteins are a source of energy. The depletion of the protein fraction in gill, liver, brain, muscle and kidney might be due to their degradation and the possible utilization of degraded products for metabolic purposes Singh *et al.* (1996) the intake of insecticide affects the biochemical composition of fishes has been reported previously by many scientists (Kumble and Muley2000; Prasad *et al* 2002).

Schmidt and nielson (1975) stated that the decreased tendency of total protein may also be due to the metabolic utilization of the Keto acids to gluconeogenesis of proteins from the synthesis of glucose or may be due to directing the synthesis of proteins from free amino acidsThe 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. All these investigations support the present study of decreasing trend of proteins in the tissues of the fish *Labeo rohita* exposed to flubendiamide.

CONCLUSION

The present work indicates that flubendiamide caused alterations in the protein metabolism of fish *Labeo rohita*, treated fish tissues showed more decrement in protein levels when compared with controls. 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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