

## TOXIC EFFECTS OF ARSENIC ON PROTEIN CONTENT IN THE FISH, *LABEO ROHITA* (HAMILTON)

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### ABSTRACT

The toxic effects of two different sublethal concentrations (one-third and one-tenth of the 96 hrs LC<sub>50</sub> value) of arsenic on fresh water fish, *Labeo rohita* have revealed significant changes in the tissues like liver and muscle of the fish. The fish exposed to the lower and higher sublethal concentration of arsenic for 7, 14, 21 and 28 days has showed reduction in the protein content in liver and muscle tissues.

### INTRODUCTION

In recent years, the high rate of increase in human population and rapid pace of industrialization have created problem of disposal of waste waters. The domestic wastes and untreated or partially treated industrial effluents, supplemented with pollutants like heavy metals, pesticides and many organic compounds, have greatly contributed to massive fish death of aquatic ecosystems. These toxic chemicals and metals have changed the quality of waters that affect the fish and other aquatic organisms (Dhasarathan et al. 2000).

Almost all the metals are toxic at higher concentrations and some are lethal even at a very low concentration. The priority list of pollutants compiled by the Environmental Protection Agency (EPA) of the United States contains eight widespread heavy metals, arsenic, cadmium, chromium, copper, lead, mercury, nickel and zinc (Moore & Ramamoorthy 1994). Among these, arsenic is more significant from environmental and toxicological point of view (Newland 1982). Proteins play a vital role in the physiology of living organisms. Proteins are highly sensitive to heavy metals and are one of the earliest indicators of heavy metal poisoning (Jacobs et al. 1977). Proteins are important substances required by organisms in tissue building and play an important role in energy metabolism (Yeragi et al. 2003). A reduction in protein content has been noticed when the fish *Channa punctatus* was exposed to heavy metals such as mercury, arsenic and lead (Jana & Bondyopadhyay 1981).

Heavy metal toxicity induced changes in protein profiles in different tissues of fish have been reported in *Cirrhina mrigala* exposed to lead acetate (Ramalingam et al. 2000), *Catla catla* exposed to chromium (Vincent et al. 1995) and *Channa punctatus* exposed to phenyl mercury acetate (Karuppasamy 2000). However the studies on the effects of arsenic on the liver and muscle tissues of *Labeo rohita* are meagre. Hence, in the present study, the sublethal toxicity of arsenic on protein content of a freshwater fish *Labeo rohita* was investigated.

### MATERIALS AND METHODS

Healthy *Labeo rohita* fish ranging from 10-12 cm in length and weighing 9-14 g were collected from fish farm located in Puthur and acclimatized under laboratory conditions ( $29 \pm 1^\circ\text{C}$ ). The fish were fed daily on oil-less groundnut cake. The unused food was renewed after 2 hours and water was changed daily. Prior to experimentation the fish were acclimatized to experimental tanks for at least

one week. The  $LC_{50}$  for arsenic trioxide for 96 hours was found using probit method (Finney 1971) and  $1/10^{\text{th}}$  of the  $LC_{50}$  value (0.27ppm) and  $1/3^{\text{rd}}$  of the  $LC_{50}$  value (0.91ppm) were taken as lower and higher sublethal concentrations respectively. The fish were exposed to these sublethal concentrations for treated and control period of 7, 14, 21 and 28 days. A control group was maintained in an identical environment. The toxicant and normal waters were renewed every day. The fish were sacrificed from both experimental and control groups on 7<sup>th</sup>, 14<sup>th</sup>, 21<sup>st</sup> and 28<sup>th</sup> days of exposure periods and subjected to analysis for the biochemical changes. The protein was estimated by the method of Lowry et al. (1951).

## RESULTS

The protein content in the different tissues of control and arsenic treated fish, *Labeo rohita*, are presented in Table 1.

**Liver:** In the control fish, the mean protein content in the liver tissues was 98.32, 98.20, 98.51 and 98.34 mg/g wet wt. of tissue on 7, 14, 21 and 28 days of experimental periods respectively. In the fish treated with 0.27 ppm of arsenic, mean liver protein was 85.39, 80.11, 72.33 and 66.53 mg/g wet wt. of tissue and at 0.91 ppm, it was 81.00, 75.17, 68.44 and 51.00 mg/g wet wt. of tissue on 7, 14, 21 and 28 day of the exposure period respectively. The percent decrease over the control was 13.15, 18.42, 26.57 and 32.35 in lower sublethal concentration and 17.61, 23.45, 30.52 and 48.14 in higher sublethal concentration for 7, 14, 21 and 28 days of exposure period respectively. The decrease in mean protein levels was statistically significant ( $p < 0.05$ ) in both sublethal concentrations at all the exposure periods (Table 1).

**Muscle:** When *Labeo rohita* was intoxicated with lower and higher sublethal concentration of arsenic, the protein content in the muscle decreased at all periods from the mean control level. The maximum decrease was observed at 28<sup>th</sup> day of higher sublethal exposure of arsenic. In the control fish, the mean protein content in the muscle tissues was 84.38, 84.27, 84.42 and 84.38 mg/g wet wt. of tissues for 7, 14, 21 and 28 days of exposure periods respectively. In the fish treated with lower sublethal concentration of arsenic, mean muscle protein content was 75.82, 71.17, 67.51 and 62.07 mg/g wet wt. of tissue, and in higher sublethal concentration, 72.25, 65.02, 61.09 and 53.29 mg/g wet wt of tissue for 7, 14, 21 and 28 days exposure periods respectively. The percent decrease over the control was 10.14, 15.54, 20.03 and 26.44 in lower sublethal concentration and 14.38, 22.04, 27.63 and 36.84 in higher sublethal concentration for 7, 14, 21 and 28 days of exposure period respectively. The decreased level of protein in both sublethal exposure periods was statistically significant (Table 1).

## DISCUSSION

Proteins play a vital role in the biological functions and serve as building blocks for cellular mass. The depletion of protein in tissues after treatment with sublethal concentrations of arsenic might be due to reduction in rate of protein synthesis and/or excessive proteolysis during stress condition.

In the present investigation, fish exposed to both lower and higher sublethal concentrations of arsenic have showed decrease in protein content in liver and muscle at all periods. A significant decrease in protein content of gill, brain, intestine, liver, kidney and muscle has been observed in the fish exposed to nickel (Joseph Thatheyus et al. 1992). The reduction in the protein level of liver has also been observed in the fish *Labeo rohita* exposed to pesticides due to intensive proteolysis during stress conditions (Rajamannar & Manohar 1998). A significant decline of protein content in liver of fish *Cyprinus carpio* when exposed to the pesticide monocrotophos suggests an increase in the pro-

Table 1: Effects of lower and higher sublethal concentrations of arsenic on protein levels in liver and muscle tissues of *Labeo rohita* at different periods of exposure.

Tissues	Group	Exposure period in days				F Value
		7	14	21	28	
Liver	C	98.32±0.474	98.20±0.664	98.51±0.545	98.34±0.821	0.050 <sup>NS</sup>
	LC	85.39±0.411 (-13.15)	80.11±0.492 (-18.42)	72.33±0.536 (-26.57)	66.53±0.098 (-32.35)	391.49*
	HC	81.00±0.541 (-17.61)	75.17±0.366 (-23.45)	68.44±0.067 (-30.52)	51.00±0.281 (-48.14)	659.78*
Muscle	C	84.38±0.488	84.27±0.338	84.42±0.627	84.38±0.348	0.017 <sup>NS</sup>
	LC	75.82±0.398 (-10.14)	71.17±0.314 (-15.54)	67.51±0.210 (-20.03)	62.07±0.211 (-26.44)	314.16*
	HC	72.25±0.228 (-14.38)	65.02±0.447 (-22.84)	61.09±0.268 (-27.63)	53.29±0.174 (-36.84)	814.07*

C - Control; LC - Lower sublethal concentration; HC - Higher sublethal concentration

Mean ± S.E. - Indicates the mean of six individual observations; (+/-) Indicates the percent increase/decrease over the control; Values are expressed in mg/g wet wt. of tissue; NS - Non significant; \*indicates significant at 5% level of F test.

teolytic activity caused by exposure to the pesticide (Maruthanayagam & Sharmila 2004).

Depletion of protein as a result of toxicant stress has also been reported by number of workers. Jana & Bondyopadhyay (1981) have reported such a reduction in protein content when the fish *Channa punctatus* was exposed to heavy metals such as mercury, arsenic and lead. Muniyan (1999) has found the depletion of protein content in the liver, gill, kidney and muscle of *Oreochromis mossambicus* when exposed to ethofenprox. Jeyachandran & Chockalingam (1987) have also observed a similar response in liver, muscle, gill and kidney of *Channa punctatus* after being exposed to tannery effluent. Moreover, they have reported that depletion in the protein content in intoxicated fish may be due to intense proteolysis.

Appreciable decrease in protein level of liver, muscle, intestine, gill and blood of *Heteropneustes fossilis* was noticed after the exposure of fish to nickel for 30, 60 and 90 days (Prasanta Nanda et al. 2000). Jha & Jha (1995) have observed protein depletion in liver, muscle and gonads of *Anabus testudineus* under the stress of nickel chloride. Decrease in the liver and muscle protein level has been reported in the fish *Channa punctatus* exposed to phenyl mercuric acetate (Karuppasamy 2000) and also *Channa punctatus* exposed to oleandrin (Tiwari & Singh 2004). A similar decreased liver protein level has also been found in *Mystus vittatus* exposed to nuwan (Tazeen Arasta et al. 1996), *Lepidocephalicthus thermails* exposed to copper (Geetha et al. 1996), *Channa puntatus* exposed to arsenic (Jatyajit Hota 1996), *Channa straiatus* exposed to mercury, cadmium and lead (Palanichamy & Baskaran 1995), *Cirrhina mrigala* exposed to lead acetate (Ramalingam et al. 2000), *Cyprinus carpio* exposed to endosulfan (Jenkins et al. 2003), *Labeo rohita* exposed to cypermethrin (Veeraiyah et al. 1998) and *Heteropneustes fossilis* exposed to petroleum oil (Sabita Borah 2005). A reduction in the protein content in the present investigation in *Labeo rohita* suggests that the tissue protein might have undergone proteolysis, during the stressful situation in the intoxicated fishes.

#### ACKNOWLEDGEMENT

The authors are thankful to Prof. Dr. L.S. Renganathan, Head of the Department of Zoology, Annamalai University, Annamalai Nagar for providing laboratory facilities. I exuberantly acknowledged Dr. M. Vasanthi, Prof. and Head, P.G. Deptt. of Environmental Sciences, Govt. Arts College Ariyalur, for

the encouragement.

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