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THE IMPACTS OF ZINC ON LIVER AND SERUM PROTEINS AND CHOLESTEROL LEVELS OF FISH (*CIRRHINUS MRIGALA*)

ANAND KUMAR[1] AND TEJPAL DAHIYA*
[1]Department of Zoology and Aquaculture, Chaudhary Charan Singh Haryana,
Agricultural University, Hisar - 125 004 (Haryana), INDIA
*Department of Zoology, Government Postgraduate College, Mahendergarh - 123 029 (Haryana), INDIA
e-mail: dahiyatejpal@yahoo.co.in

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***Corresponding author**

ABSTRACT
The present investigation was aimed to observe the impacts of Zinc (Zn) on the protein levels in liver, serum and cholesterol of *Cirrhinus mrigala* fingerlings and experiments were designed having control without Zn and three treatments T1, T2 and T3 containing Zn at concentrations 0.01, 0.02, 0.04 ppm, respectively in triplicates. The decreasing trends were observed in the protein levels of liver tissue in *C. mrigala* with an increase in the concentrations of Zn from T1 then T2 to T3 *i.e.* 16.40% (56.00±0.45 µg/g), 32.83% (44.26±0.52 µg/g) and 47.76% (34.86±0.81µg/g), respectively w. r. t. control (T0) 67.10±0.45 µg/g for 45 days. The decline in liver proteins was found to be maximally *i.e.* up to 47.76% in liver proteins. The declines in serum protein level were observed in T1, T2 and T3 *i.e.* 4.76% (8. 00 ± 2.54 µg/g), 11.90% (7.33 ± 4.34 µg/g) and 24.06% (6.34±0.72 µg/g) at concentrations 0.01, 0.02, 0.04 ppm of Zn, respectively w. r. t. control (T0) 8.40±0.58(µg/g). After 45 days, the levels of cholesterol were increased with an increase in the concentrations of Zn in treatments T1, T2 and T3 and it was increased up to 69 % at 0.04ppm (T3). In conclusion, the levels of liver and serum proteins decrease while cholesterol increases when *C. mrigala* is subjected to Zn metal toxicity.

INTRODUCTION

The natural water quality is deteriorating by dumping waste products from industries synthesizing textiles, agro-chemicals, insecticides, paints, dye and chloroalkali metals etc. (Dahiya *et al*., 2008, 2009) and making it devastating to people, animals, fish and birds unsuitable for drinking, agriculture as well as industry itself. The heavy metals water soluble, non-degradable, vigorously oxidizing agents and strongly bind to many biochemical units and hence entered in food chains. Chromium, zinc and lead at sub lethal concentrations have been found to increase the oxygen consumption, locomotion of fishes and swimming pattern (Brafield and Mathiessen, 1996). James *et al*. (1992) also reported that the presence of sublethal concentration of mercury reduced the rates of feeding, absorption, conversion and metabolism in *Heteropneustes fossilis*. Maniora (2005) observed the differences in correlations of lipid components in the brain of carp (*C. carpio*) under the influence of heavy metals. There were increased amounts of total lipids and the relative content of phospholipids, free fatty acids, free cholesterol, choline, and phosphatidyl ethanolamine under the intoxication of Pb^{2+}, Cu^{2+} and Zn^{2+}. So, by keeping above facts in mind, the present investigation was proposed to observe the impact of Zn on the liver and serum proteins as well as cholesterol levels *C. mrigala*.

MATERIALS AND METHOD

In the present study, 6-8 inch long fingerlings of *C. mrigala* were collected from the local fish farm and acclimatized for two weeks in large tanks already filled dechlorinated tap water with proper aeration under laboratory conditions. These were exposed to the sublethal concentrations (<0.1ppm) of Zn for the period of 45 days; in small plastic tanks of 40 liter capacity in triplicates with 10 fishes each replicate; and were fed with normal standard diet on alternate days at the rate of 2 per cent of total fish body weight. The treatments were T1, T2 and T3 having concentrations of 0.01, 0.02 and 0.04ppm of zinc; while T0 was control having dechlorinated water only. The three fishes were taken and sacrificed from each replicate at the end of 45 days and analyzed for the estimation of liver tissue protein and serum by following Lowry's *et al*. (1951) , cholesterol by Zlatkis *et al*. (1953). For the estimations the blood was removed from the fish in Eppendorf tubes and left for about 30 minutes at room temperature. Serum was separated and kept at 4ºC for 30 minutes and then centrifuged at 3000 rpm for 10 minutes. The supernatant was removed and used as stock solution for protein estimation following Lowry's *et al*. (1951) method. The obtained results were analyzed by statistically (Snedecor and Cochran 1994).

RESULTS

Impacts of zinc on protein

The proteins are most diverse bio-molecule as these are of prime importance in biochemical reactions and cellular structures. In the present investigations; decreased trends were

observed in the liver proteins *i.e.* 16.4 % at 0.01ppm, 32.83 % at 0.02ppm and 47.76 % at 0.04ppm concentrations of Zn after 45 days. It is concluded that there were decline in tissue protein percentage in fish body when exposed to three concentrations of Zn (Table 1 and Fig. 1).

Impacts of zinc on serum protein

Serum proteins have immunological properties in fishes and other animals as well as in human. Likewise liver proteins; a similar decreasing trend was also observed in serum proteins *i.e.* 4.76 %, 11.90 % and 24.06% of the serum protein decreased at 0.01, 0.02 and 0.04ppm concentrations of Zn, respectively (Table 1and Fig.1).

Impacts of zinc on cholesterol

Polluted water having Zn has a large impact on the concentration of cholesterol in the fish body. From the present study it was observed that after treatments of Zn the cholesterol level increased as compared to the control by 30.72 % at 0.01ppm, 53.87 % at 0.02ppm and 69.44 % at 0.04 ppm, thereby indicating a high physiological disturbance in fish when exposed to Zn for 45 days (Table 2 and Fig.1).

Table 1: Effects of zinc on total protein (μg/g) in *C. mrigala* after 45 days treatment

Treatment	Zn concentration (ppm)	Total tissue protein (μg/g)	Total serum protein (μg/g)
T0	0.00	67.10 ± 0.45	8.40 ± 0.58
T1	0.01	56.00 ± 0.45 (16)	8. 00 ± 2.54 (4.76)
T2	0.02	44.26 ± 0.52 (32)	7. 33 ± 4.34 (11.90)
T3	0.04	34.86 ± 0.81 (47.76)	6. 34 ± 0.72 (24.66)
	SE(m)	0.79	0.01
	CD at 5%	1.88	0.76

Figures in parentheses are the percentage change over control ($p < 0.05$).

Table 2: Effects of zinc on total cholesterol (μg/g) in *C. mrigala* after 45 days treatment

Treatment	Zn concentration (ppm)	Total tissue cholesterol (μg/g)
T0	0.00	26.13 ± 2.41
T1	0.01	34.16 ± 1.25 (30)
T2	0.02	40.06 ± 2.75 (53)
T3	0.04	44.76 ± 0.84 (69)
	SE(m)	0.148
	CD at 5%	0.490

Figures in parentheses are the percentage change over control ($p < 0.05$)

DISCUSSIONS

The protein levels in liver tissue of *C. mrigala* were found to be declined with the increase in the concentrations of heavy metal Zn *i.e.,* from T1 to T3 and it declined maximally up to 47.76% at 0.04ppm (T3) of Zn. Serum protein also showed same declining trend just like liver proteins *i.e.* serum proteins level showed a maximum decline up to 24.66% at 0.04ppm (T3) concentration of zinc. Similar declining trends in total protein and glycogen contents were also observed by Kumar *et al.* (2005) and Maruthappan *et al.* (2005) in freshwater fish *C. mrigala* when exposed to Lead (Pb) and also reported significant hematological and biochemical alterations *i.e.* reduced hemoglobin content, RBC and WBC counts and found

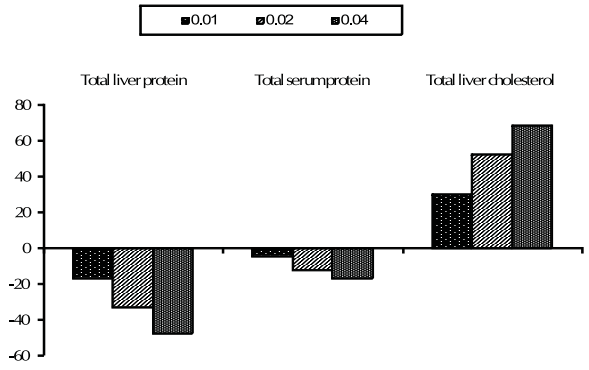

Figure 1: The effect of zinc on total protein (μg/g) and total cholesterol (mg/g) in *C. mrigala* after 45 days treatment

that the adverse effects of lead were higher after a long term exposure when compared to the short term exposure of the lead. While Athikesavan *et al.* (2006) observed histopathological changes in freshwater fish *Heteropneustes malitrix* when exposed for 10, 20 and 30 days in a sub-lethal concentration of nickel 5.7 mg/L. Kaur and Kaur (2006) used increased cadmium concentrations and observed similar results in *Oreochromis mossambicus*. The results observed by Mittal (2004) also found similar with our results in *C. mrigala* but used toxic heavy metals As, Hg, Co and Ni alone and in combination (As + Hg, As + Co, As + Ni).

In the present investigation cholesterol level increased significantly ($(P < 0.05)$) as compared to control by 30.72 % at 0.01ppm (T1), to 69.44 % at 0.04 ppm (T3) in *C. mrigala* when exposed to Zn for 45 days; similar trend was observed by Desai *et al.* (2002) when freshwater fish *Channa punctatus* exposed by nickel. In an another similar study Jain (1999) observed a decrease in the soluble protein, RNA and glycogen contents in the liver and body weight but increased the cholesterol contents when teleost fish *H. fossilis* exposed to sublethal concentrations of lead nitrate in water solution for short (35 days) and long (120 days) period. In conclusion, heavy metal Zn can change the body metabolism of the fish by decreasing liver and serum proteins and increasing cholesterol in polluted water bodies.

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