STUDIES ON THE INFLUENCE OF HEAVY METALS ON ANTIOXIDANT SYSTEM IN THE TISSUES OF FRESHWATER FISH *Cyprinus carpio*

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ABSTRACT:

After the determination of LC_{50} 96 hr values of zinc, cadmium and lead *to C.carpio* (1.0%, 1.5% and 1.5% respectively). Group of ten fishes were reared in sublethal concentrations of indidual metals (0.5%, 0.1%, 0.15%, 0.20% and 0.25%) for 30 days. Tissues like liver, kidney and muscle were used to estimate the antioxidant enzymes (SOD, CAT, GPx and GST) from the metal treated fishes. The heavy metals were found to cause oxidative stress so that the defensive enzymes were begin to rise the activity levels of the enzymes to neutralize the reactive oxygen species (ROS) under metal toxicity in the fishes.

Key words: Antioxidant system, heavy metals and *Cyprinus carpio.*

Introduction:

The contamination of aquatic environment is of great concern throughout the world because a number of chemicals from multifarious sources cause deleterious effect on aquatic organisms. Among all types of pollution, the aquatic contamination especially by heavy metals is deteriorating the ecological balance of aquatic environment. Due to rapid industrialization and urbanization, a large quantity of heavy metal and there compound are continuously being released into the aquatic bodies. In the opinion of Malik *et al* (2010), the heavy metal pollution in aquatic system is growing at an alrming rate and has become an important worldwide problem.

The fishes are one of the major components of aquatic ecosystem as well as food. Shukla and Upadhyam (1998) have reported that fishes are being used as protein diet to meet the deficiency in India. While the fish population provides protein – rich food to human beings, they are very sensitive to a wide variety of toxicants in water. They are often at the top of aquatic food chain and can observe heavy metals through skin, gills and gastro intestinal tract (Jovanovic *et al* 2011). As a result the fishes are the best biosensors of aquatic pollution.

Therefore the present work has been planned to study the effect of heavy metals on antioxidant enzyme in various tissues of *Cyprinus carpio.*

Materials and Methods:

The common fresh water fish *C.carpio* procured from local reservoir, Nerunjipet were used and heavy metals (zinc, cadmium and lead) from Gharda chemicals, Mumbai, India) were used in the present study. The concentrations of metals (0.5%, 1.0%, 1.5%, 2.0% and

2.5%) were prepared indually for each metal using dechlorinated tap water. Static bioassays were carried out to find out LC_{50} 96 hr value for zinc was 1.0% for cadmium and lead were 1.5%. Then a group of 10 fishes were exposed to various sublethal concentrations of indidual metals (0.5%, 0.10%, 1.5%, 0.20% and 0.25%) for 30 days. Various tissues such as liver, kidney and muscle were selected from the treated fishes and processed to estimate various antioxidant enzymes namely superoxide dismutase (SOD) by using the method of Marklund and Marklund (1974), Catalase (CAT) by the method of Sinha (1972), Glutathione peroxide (GPx) and Glutathione- S- Transferase (GST) by method given by Habig *et al* (1974).

The SOD activity was expressed in terms of units/ min/mg protein,GPx activity as µmoles of glutathione oxidized min/mg protein and CAT activity as µmoles of H_2O_2 utilized/min/mg protein, GST activity as µmoles of CDNB formed/min/mg protein.

Results and Discussion:

In the present investigation, the antioxidant enzymes such as SOD, CAT, GPx and GST are found to increase in tissues like muscle, liver and kidney of *C.carpio* in dose – dependent manner as shown in tables 1-3.

The overproduction of ROS (reactive oxygen species) such as hydrogen peroxide (H_2O_2), superoxide anion (O_2) and Hydroxyl radical (OH) in the tissues of organisms cause oxidative damage in organisms. Many pollutants are shown to induce the production of ROS in aquatic organisms (Kurutas *et al* 2009; Ahmed *et al* 2010; Seveikova *et al* 2011). One of the main consequences of oxidative stress is lipid peroxidation (LPO) in which the ROS reactive with membrane lipids. Thus the antioxidant responses such as oxidative stress and the activity levels of antioxidant enzymes would be reliable biochemical indicators to assess the environmental risk.

In the presents study the heavy metals are found to cause oxidative stress in the tissues of fishes as the antioxidant system has been estimated to increase the production of various defensive enzymes. This observation falls in line with the findings of Aina *et al* (2012) who have shown that the oxidative stress would increase the levels of defensive enzymes under the toxicity of pollutants. Saliu and Bawa Allah (2012) have found that the increased quantity of antioxidant enzymes would help the organisms to overcome to oxidative stress by regulative bioaccumulation of the metals to levels the body can tolerate. According the Begum and Sengupta (2014) increased activity levels of antioxidant enzyme is indicate the adaptational and protective response of animals under pollutant toxicity.

The present findings in freshwater fishes clearly indicate that the entire antioxidant system has been stimulated to start increased production of antioxidant enzymes to detoxify the free radicals and to remove from the cells to overcome the oxidative stress caused by heavy metals.

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Table 1. Percent changes in the concentrations of various antioxidant enzymes in the tissues of *C. carpio.* under zinc exposure.

Concentration (%)	Muscle				Liver				Kidney			
	SOD	CAT	GPx	GST	SOD	CAT	GPx	GST	SOD	CAT	GPx	GST
0.05	+0.74	+3.50	+0.11	+2.23	+0.40	+0.55	+0.19	+1.08	+0.61	+0.48	+0.75	+2.68
0.10	+0.54	+9.13	+0.21	+4.02	+0.92	+1.43	+0.46	+2.36	+1.93	+1.92	+2.06	+4.46
0.15	+2.79	+12.82	+0.39	+6.70	+1.84	+1.88	+0.73	+3.69	+3.07	+2.13	+3.09	+8.93
0.20	+3.79	+18.06	+0.48	+8.04	+2.38	+2.95	+1.07	+4.23	+4.77	+2.41	+4.50	+11.61
0.25	+4.45	+25.44	+0.49	+12.05	+2.23	+3.48	+1.26	+5.46	+6.37	+4.20	+6.13	+13.39

+ indicates percent increase over control

Table 2. Percent changes in the concentrations of various antioxidant enzymes in the tissues of *C. carpio.* under cadmium exposure.

Concentration (%)	Muscle				Liver				Kidney			
	SOD	CAT	GPx	GST	SOD	CAT	GPx	GST	SOD	CAT	GPx	GST
0.05	+0.35	+8.16	+0.66	+1.78	+1.40	+2.00	+0.50	+1.76	+0.76	+0.58	+1.03	+5.45
0.10	+0.82	+12.90	+0.95	+3.56	+2.18	+2.92	+0.97	+2.98	+3.64	+1.54	+2.17	+10.00
0.15	+3.54	+20.87	+1.27	+6.22	+3.24	+4.46	+1.31	+3.66	+5.15	+2.71	+4.07	+13.64
0.20	+4.43	+29.22	+1.62	+8.44	+5.32	+5.71	+1.68	+4.10	+7.71	+3.74	+5.41	+15.45
0.25	+5.40	+35.67	+2.03	+16.00	+6.72	+6.52	+2.21	+4.49	+9.31	+4.63	+6.43	+20.00

+ indicates percent increase over control

Table 3. Percent changes in the concentrations of various antioxidant enzymes in the tissues of *C. carpio.* under lead exposure.

Concentration (%)	Muscle				Liver				Kidney			
	SOD	CAT	GPx	GST	SOD	CAT	GPx	GST	SOD	CAT	GPx	GST
0.05	+0.88	+4.86	+0.50	+1.76	+0.48	+0.66	+0.26	+1.13	+1.57	+0.58	+0.83	+2.63
0.10	+2.07	+9.91	+0.69	+2.64	+1.44	+1.09	+0.42	+2.94	+3.23	+1.33	+1.84	+6.14
0.15	+3.11	+14.21	+1.10	+4.85	+2.30	+1.87	+0.70	+3.82	+4.33	+2.25	+2.85	+8.77
0.20	+3.49	+18.88	+1.69	+7.49	+3.53	+2.80	+1.00	+4.26	+6.13	+2.86	+3.53	+11.40
0.25	+4.15	+25.05	+2.01	+11.01	+5.36	+3.35	+1.40	+4.31	+7.27	+3.85	+4.39	+13.16

+ indicates percent increase over control