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The effect of Cadmium on antioxidant enzymes in the liver of fresh water fish *Cyprius carpio* (Linn).

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ABSTRACT

In the present investigation, the fresh water fish C. carpio and Cadmium were used. Bio assays were conducted to find LC50 96hr value of the metal to the fishes and it was found to be 1.5%. Then groups of fishes were reared in various sublethal concentrations of Cadmium for 30days. The liver from the control and Cadmium –treated fishes was dissected out for the estimation of Superoxide Dimutase (SOD), Catalase (CAT), Glutathione Peroxidase (GPx), and Gulutathione-S-Transferrase (GST). In treated fishes, the liver antioxidant enzymes increased under metal toxicity suggesting the initiation of the production of these enzymes to suppress the action of ROS. The liver might have severely affected by Cadmium as it is the site of multiple oxidative reactions.

Key words: Cyprinus carpio, Superoxide Dimutase (SOD), Catalase (CAT), Glutathione Peroxidase (GPx), and Gulutathione-S-Transferrase (GST).

INTRODUCTION

Water pollution is due to continuous and alarming influx of toxicants into the aquatic environments from both naturally occurring and anthropogenic sources. The polluted water may cause destruction of beneficial species either directly affecting their life on indirectly through breaking biological food chains. Alterations in the chemical composition of natural aquatic environments by the pollutants usually induce changes in the behavior and physiology of the inhabitants particularly the fishes (Saroja et al., 2013). In short, the health of the fish could reflect and give a good indication of the health status of the aquatic ecosystems in which they live.

The heavy metals have long been recognized as serious pollutants of the aquatic environment because they enter water bodies by industrial and consumer processes and even from acidic rain (Obasohan et al., 2008). According to El – Sayed et al., 2013, Cadmium is one of the most toxic contaminant in aquatic environments causing toxicity at each level of the ecological station. Zeynap Abedi et al. (2013) have also noticed that Cadmium even at sublethal concentrations cause cumulative effects resulting in serious physiological disturbances in fishes. Literature are made available by many investigators to show the effect of various heavy metals in Piscean species (Prasath and Arivoli, 2008; Shaheen and Akhtar, 2011; Shwetha et al., 2012; Kabilan et al., 2013; Hog and Hague, 2014).

The fish tissues consistently produce reactive oxygen species (ROS) during environmental stress leading to oxidative stress. The oxidative stress is due to over production of ROS in the tissues of animals and is one of the important mechanism in toxic conditions. In the opinion of Sudha Summarwar and Deepali lall (2013), the oxidative stress affecting the metabolic adaptive strategies of fishes can be assayed in the form of changes in antioxidant enzymes level. Among the main function of vertebrate liver, detoxication is the most prominent one through a number of enzymatic reactions. Since fishes constitute an excellent model to understand the oxidative stress due to contaminations of aquatic ecosystem, the present study has been carried out to evaluate the antioxidant enzymes in the liver of freshwater fish Cyprinus carpio under cadmium toxicity.

MATERIALS AND METHODS

Fishes with weight and length were procured from and Cadmium was from Bioassays were carried out to find LC50 96hr value of cadmium to the fishes by the method of Sprague (1973) and it was found to be 1.5%. Then a group of 10 laboratory acclimatized fishes were exposed to various sublethal concentrations namely 0.05%, 0.10%, 0.15%, 0.20% and 0.25% for 30 days. Appropriate combos were also maintained. The dechlorinated tap water was used as a diluent medium. In the test water, variables such as temperature (30 ± 1°C), pH (7.1 – 7.9), salinity (0.4 – 0.7ppm) and DO (5.5 – 6.2 mg/l) were controlled.

The physicochemical characteristic of the test water were determined by the methods given by APHA (1980). During experimentation, the test solutions were replenished daily and the test fishes were given artificial fish meal. At the end of the experimental period, the liver was dissected out and processed to estimate various antioxidant enzymes namely SOD by the method of Marklund and Marklund (1974), CAT by the method of Sinha (1972), GPx by the method of Rotruck et al (1973) and GST by the method of Habig et al (1974).

RESULTS AND DISCUSSION

A During environmental stress, the level of ROS increase traumatically suppressing the antioxidant defense system and causing oxidative stress. If the production of ROS is in excess, then the balance between the formation and removal of ROS is destroyed with the resultant oxidative stress (Li et al., 2010). Atli and Canli (2007) have recorded that the antioxidant compounds like antioxidant enzymes, amino acids, peptides, phenolic compounds etc. are found in the tissue of all species of fishes to protect the damage caused by

Concentration of Cadmium (%)	SOD	CAT	GPx	GST
0	29.30 ± 0.27	40.80 ± 0.99	118.29 ± 0.56	20.48 ± 0.26
0.05	29.71 ± 0.31 (+1.40) r = 0.947	41.62 ± 1.33 NS (+2.00) r = 0.992	118.88 ± 0.89 (+0.50) r = 0.932	20.84 ± 0.32 NS (+1.76) r = 0.900
0.10	29.94 ± 0.34 (+2.18) r = 0.940	41.99 ± 1.39 NS (+2.92) r = 0.942	119.44 ± 0.91 (-16.99) r =0.995	21.09 ± 0.51 NS (+2.98) r =0.992
0.15	30.25 ± 0.39 (+3.24) r = 0.990	42.62 ± 1.91 NS (+4.46) r = 0.996	119.84 ± 1.02 (+1.31) r =0.969	21.23 ± 0.65 NS (+3.66) r =0.941
0.20	30.86 ± 0.41 (+5.32) r = 0.996	43.13 ± 2.37 NS (+5.71) r = 0.999	120.28 ± 1.30 (+1.68) r =0.995	21.32 ± 0.77 NS (+4.10) r =0.946
0.25	31.27 ± 0.42 (+6.72)r = 0.999	43.46 ± 2.41 NS (+6.52) r = 0.970	120.91 ± 1.44 (+2.21) r =0.998	21.40 ± 0.84 NS (+4.49) r =0.916

Table.1. The impact of cadmium on oxidative enzymes in the liver of *C. carpio*. (Each value is the mean ± SD of 5 observations). (SOD is expressed as IU /mg protein; CAT as µ moles of H2O2 Utilized/min/mg protein; GPx as n mole of NADPH reduces/min/mg protein).+ indicates percent increase over control; r – Correlation coefficient.

the oxidative stress. It is also reported that the antioxidant defense system that includes several enzymes provides a mean to deal with oxidative stressin organisms.

In the present work, the antioxidant enzymes namely SOD, CAT, GPx and GST are found to increase in the liver of fishes (Table 1.) The percent increase of SOD ranges from 1.40 to 6.72, of CAT from 2.00 to 6.52, of GPx from 0.50 to 2.21 and of GST from 1.76 to 4.49. Though the present increase of the activity levels of antioxidant enzymes are insignificant in the liver, there is positive correlation between the concentrations of cadmium and the level of enzymes. Therefore, it is evident that cadmium would have caused oxidative stress in the liver of C. carpio so that the antioxidant enzymes might have started increased production to enhance the ability of cells to reduce ROS. This is in agreement with the findings of Soorya et al., (2013) and E. Mamidala (2012) who have reported that the induction of antioxidant compounds is a sensitive early warning signal of oxidant stress. In the present experimental fishes, the initiation the production of antioxidant enzymes in the liver in indicative of important adaptational and protective response against toxic stress as also observed by De Lu Zhang et al., (2013) and Vinaya Kumar (2014). It is also found that liver would be heavily affected in organisms by oxidative stress because it is the major site of multiple oxidative reactions and maximum periodical generation (Richardson et al 2010).

In recent times, many environmental pollutants are found to induce oxidative stress in aquatic animals so that the oxidative stress has gained considerable importance in the field of ecotoxicology (Modesto and Martinez 2010; Kim et al., 2012; Venkata Rathnamma and Nagaraj, 2014). In accordance with the findings of Yildirin. et al. (2011), the present study also envisages that the level of antioxidant enzymes can be used as an early warning indicator of environmental pollutants.

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