Available online at http://www.journalijdr.com

ISSN: 2230-9926

International Journal of Development Research
Vol.1, Issue, 1, pp.001-004, March, 2011

International Journal of
DEVELOPMENT RESEARCH

Full Length Research Article

EFFECT OF HEAVY METAL NICKEL ON THE LIPID PEROXIDATION AND ANTIOXIDANT PARAMETERS IN THE LIVER TISSUE OF *Cirrhinus mrigala* (HAM.)

Parthiban, P* and Muniyan, M.

Department of Zoology, Annamalai University, Annamalainagar-608002, Tamilnadu, India

ARTICLE INFO

Article History:

Received 2nd January, 2010
Received in revised form
28th February, 2011
Accepted 1st March, 2011
Published online 13th March, 2011

Key words:

Nickel,
Fresh water fish,
LPO, Antioxidants,
Chirrhinus mriaala.

ABSTRACT

Metals are biologically important. If the threshold concentration of the metal increases in the environment, they may interfere with the metabolic activity of organisms. In the presenting study, an attempt was made to investigate the effect on nickel on oxidative stress and antioxidant enzymes in the liver tissue of *Cirrhinus mrigala*. The fish were exposed to sub lethal dose of 96 hr LC_{50} of nickel for 30days and removed the liver tissue as well as from control fish and studied the tissue lipid peroxidation (LPO), Reduced glutathione (GSH), glutathione peroxidase (GPx), Catalase (CAT), and superoxide dismutase (SOD). The present study of metal treated fish shows the increased level of lipid peroxidations and decreased level of reduced glutathione (GSH), glutathione peroxidase (GPx), catalase (CAT), superoxide dismutase (SOD) in the liver tissues of fish, *Cirrhinus mrigala*. These observed mean data were statically significant at $P< 0.05$ student 'T' test. The results are discussed with available literature.

INTRODUCTION

The rapid development of industrialization and anthropogenic activities have leads to contamination of many ecosystems (Gayathri *et al.*, 2008) especially the aquatic ecosystem, which receives a wide range of pollutants. Pollution of aquatic habitats seems to be an inevitable problem of universal nature and the intrusion of various pollutants into the aquatic environment affects the survival growth and reproduction of the biological organisms present in the environment. Heavy metals are extensively used in industries like electroplating, medicine, pesticide and battery manufacturing (Seiler and Siege, 1988), which constitute a serious type of pollution in freshwater and being stable compounds they are not readily removed by oxidation. (Nammalwar,1985). Metals after entering the aquatic environment it may precipitate or adsorb on the surface of solids, remain soluble or suspended in it or may be taken up by fauna and flora. Some of the metals are biologically important. In trace quantities some of the metals such as iron, copper, manganese, magnesium and zinc are essential for the metabolism of organisms (Desilva. 1978). If the threshold concentration of the metals increases in the environment they may interfere with the metabolic activity of organisms. As a fish being exclusively aquatic, a number of potentially hazardous xenobiotics confront the fish life in the sphere of their activities of which a category of special interest in that of heavy metals (Nammalawar, 1985). Nickel is one of these heavy metals. It is used extensively in electroplating as nickel sulphate and nickel hydroxide is used in nickel – cadmium batteries. Nickel induces a morphological transformation and chromosomal aberration in cells (Coen *et al.* 2001). Induces metabolic biomarkers (Vinodhini and Narayanan 2008). Anthropogenic sources of nickel (Ni) in natural ecosystems include mining, smelting, refining, metal reprocessing, fuel combustion and waste incineration (Chau and Kulikovsky-Cordeiro, 1995). Enzymes are necessary for normal cellular metabolism including that of the liver, and the degenerative changes due to the combined metal toxicity exhibited in the liver alter level of a number of its enzymes. For example lipid peroxidation (LPO), Glutothione (GSH), Glutathione peroxidase (GPx) catalase (CAT) and superoxide dismutase (SOD). These enzymes are biomarkers of acute hepatic damage, thus their bioassay can serve as a diagnostic tool for assessing the functions of the liver (Coppo *et al.*, 2002). A limited number of laboratory studies have investigated Ni uptake in fish through aqueous exposures (Tjalve *et al.*, 1988; Sreedevi *et al.*, 1992). However, the biomarkers enzymes assay is still scandy on freshwater fish. Hence, the present investigation has been carried out to investigate. The heavy metal Nickel on the lipid peroxidation and antioxidant level in the liver tissue of *Cirrhinus mrigala.*

***Corresponding author:** drmuniyan@gmail.com*

MATERIALS AND METHODS

Experimental fish

The majar carp, *Cirrhinus mrigala* were collected from the fish farm located in Pinnalur Cuddalore District, 15 Km away from the University campus. The fish were brought to the laboratory and transferred to the rectangular cement tanks (125X100X75cm) of 1000liters capacity containing chlorine free aerated well water and acclimatized to the food and laboratory conditions with 12 hr dark and 12 hr light cycles, pH range of 6.95 to 7.20 and temperature ranging from 16 to 24 °C for 15 days.

Experimental design

Fish were selected for the experiment from the stock irrespective of the sex. The size selected for the experiments were 80-100mm length and 5-10g of weight fish were divided into two equal groups each comprising of 20 fishes. Each group was kept in separate plastic trough. The first group was kept as control and were maintained in normal water without any treatment. The second group was exposed to a sub-lethal concentration of 96hrs LC_{50} of nickel (3.75ppm) for 30 days. Solution was renewed once in 24hrs exposure period. The fish from the respective experimental as well as control groups were sacrificed and liver tissue was isolated from the fish and used for the estimation lipid peroxidation and antioxidant parameters.

Estimation of lipid peroxidation and antioxidants in liver tissue

The isolated liver tissue of the control and experimental fish was used for the level of lipid peroxidation in liver tissue by the method of Nichans and Samuelson (1968), reduced glutathione was determined by the method of Beutler and Kellay, (1963), glutathione peroxidase activity was determined by the method of Rotruck *et al.* (1973), Catalase was determined by the method of Sinha (1972) and Superoxide dismutase activity was assayed by the method of Kakkar *et al.* (1984). The Statistical significance of control and experimental means were analysed by student 'T' test.

RESULTS

Level of Lipid peroxidation (LPO) and antioxidant parameters

The Table-1 shows the lipid peroxidation and antioxidants level in the liver tissue of *Cirrhinus mirgala* exposed to nickel as well as control. The lipid peroxidation, reduced glutathione and glutathione peroxidase of the liver tissue of control fish were 1.17 ± 0.058; 5.11 ± 0.204 and 1.89 ± 0.076 µmole/mg of protein/hr. where as in the sublethal concentration of nickel treated liver tissue were, 2.09 ± 0.084, 3.62 ± 0.181 and 1.41 ± 0.042 µ mole/mg of protein/hr. The catalase activity of the control liver tissues were 2.82 ± 0.113 and 1.36 ± 0.048 unit/mg of protein where as in sublethal concentration of nickel treated fish were 1.36 ± 0.063 and 0.78 ± 0.039 unit/mg of protein. If compared the control with experimental group, the lipid peroxidation has increased in the experimental group then the contral group. The per centage increased was 78.63. The reduced glutathione, glutathione peroxidase, catalase and superoxide dismutase has decreased in the experimental group then the control group. The per centage decreased were -29.51; -25.39. -51.77; and -42.65. The mean values of lipid peroxidation and antioxidant values of control and Nickle treated group was compared for their statistical significance by student 'T' test. The 'T' values were 8.8975; 5.4658; 5.5363; 11.2915 and 2.9682, These 'T' values are statistically by significant at $P<0.05$.

DISCUSSION

Heavy metal promotes the formation of reactive oxygen species (ROS) such as hydrogen peroxides. The ROS enhances the peroxides and reactive hydroxyl radicals (Miller *et al*, 1991; Hussan *et al*, 1999). These lipid peroxides and hydroxyl radicals may cause cell membrane damage and thus destroy the cell. Heavy metal increases the rate of formation of reactive oxygen species including superoxide anion radical O_2^- and hydroxyl radical (OH) through a chain reaction (Yamanaka *et al*., 1991). In the present study, the free radical scavenger enzymes GPx, CAT, and SOD were reduced in the sublethal concentration of nickel toxicity in *Cirrhinus mrigala*.

Table 1. The level of lipid peroxidation and antioxidants in liver tissue of *Cirrhinus mrigala* exposed to sublethal concentraction of Nickel

Parameters	Control	Exposure of 30 days	% change	'T' Value
Lipid peroxidation (LPO) (µmole/mg.of protein)	1.17 ±0.058	2.09 ± 0.084	78.63	8.8975*
Glutathione (GSH) (µmole /mg.of protein)	5.11± 0.204	3.62 ±0.181	-29.15	5.4658*
Glutathione peroxidase (GPx) (µmoles/mg protein)	1.89±0.076	1.41 ± 0.042	-25.39	5.5363*
Catalse (CAT) (Unit/mg.of protein	2.82 ± 0.113	1.36 ± 0.063	-51.77	11.2915*
Superoxide dismutase(SOD)(Unit/mg.of protein	1.36 ± 0.048	0.78 ± 0.039	-42.65	2.9682*

The values are Mean ± S.E of six individual observations, *Significance ($p<0.05$) of student 't' test

The inhibition of these fress radical scavenger enzymes might be due to interaction of sublethal concentration of nickel directly with metal ion, which is dependent on subcellular origin. It is possible that nickel in the tissue interacts with the metal moiety and produces inhibition of enzyme activity (Chandravathy and Reddy, 1999). Similarly, Rana *et al*., (1996) and Romeo *et al.*, (2000) reported that cadmium increases the formation of lipid peroxidation in rats and fishes. Several studies have been shown that there is increase in the formation of oxygen free radicals or reactive oxygen species (Stohs and Bagchi, 1995). The present results showed the decrease in GSH levels, which can explain the increased ROS concentration and of LPO levels in nickel exposed fish. The similar results have been observed in the Indian catfish (*C. batrachus*) after exposure to low concentration of Arsenic (Battacharya and Battacharya, 2007). The decrease in the activities of these two enzymes can inhibit the citric acid cycle and thereby decrease the generation of reducing equivalents such NADH and NADPH, which impairing ATP production (Ramanathan *et al.,* 2003) and oxygen reduction to form water. Moreover, Metal can affect NADH dehydrogenase and cytochrome oxidase. The significant decline in the activity of these two enzymes would result in the inhibition of electron flow from NADPH to oxygen, augmenting the chance of ROS generation and lowering oxygen consumption. (Rmanathan *et al*., 2003; Battacharya and Battacharaya, 2007)

Soundararajan *et al.,* (2009) reported that the alterations of GSH, SOD and Catalase in liver tissues of *Tilapia mossambica* treated with Arsenic and suggested that liver is an active site for synthesis of these antioxidant enzymes. Basha and Rani (2003) also noted significant elevations of SOD and catalase activities in liver and kidney from day 7 onward, and these activities were maintained until day 15 and then decreased slightly on day 30 of exposure. Basha and Rani (2003) suggested that upregulation of enzyme production might be a defense mechanism, providing first line of defense against metal toxicity before the induction of metallothionein synthesis. Fattorini and Regoli (2004) observed remarkable accumulation of Arsenic in the branchial crown of *Sabella spallanzanii* with dimethylarsinate (DMA) as the main Arsenic metabolite, while in another polychaete species, *Arenicola marina*, Arsenic is accumulated mostly in the inorganic forms (Geiszinger *et al.,* 2002). It can be stated that nickel affect the antioxidant responses in *Cirrhinus mrigala* in terms of increased lipid peroxidation which could impair ATP production, and triggering oxidative damage.

Acknowledgement

The authors are thankful to the Professor and Head, Department of Zoology, Annamalai University for providing necessary laboratory facilities to carry out the work successfully.

REFERENCES

Basha, P.S. and Rani, A.U. 2003. Cadmium-induced antioxidant defense mechanism in freshwater teleost *Oreochromis mossambicus* (tilapia). *Ecotoxicol Environ Saf* ., 56 (2), pp. 218–221.

Battacharya, A. and Battacharya, S. 2007. Induction of oxidative stress by Arsenic in *Clarias batrachus*: involvement of peroxissomes. *Ecotoxicol. Environ. Saf*., 66: 178–187.

Beutler, E. and Kelley, B. M. 1963. The effect of disodium nitrate on RBC glutathione. *Experientia.,* 29: 97.

Chandravathy, V.M. and Reddy, S.L.N. 1999. Effect of lead on antioxidant enzymes activities and Lipid peroxidation in old male mice. *Mus musculus. J. Environ. Biol.* 20: 103-106.

Chau, Y.K., Kulikovsky-Cordeiro, O.T.R., 1995. Occurrence of nickel in the Canadian environment. *Environ. Rev.*, 3:95–120.

Dallinger, R., Kautzky, H., 1985. The importance of contaminated food for the uptake of heavy metals by rainbow trout (*Salmo gairdneri*): a field study. *Oecologia*, 67: 82– 89.

Duncan, B.D. 1957. Duncan's multiple range tests for correlated and heteroscedastic mean. *Biometrics.* 13: 359-364.

Fattorini, D. and Regoli, F. 2004. As speciation in tissues of the Mediterranean polychaete *Sabella spalanzanii*, *Environ. Toxicol. Chem*., 23: 1881–1887.

Gayathri R. Nambiar, K. Raveendran, Changxing Zhao, C.A. Jaleel, 2008. A glimpse of lignicolous marine fungi occurring in coastal water bodies of Tamil Nadu (India), Comptes *Rendus Biologies.*, 331 (6) : 475–480.

Geiszinger, A.E., W. Goessler and K.A. Francesconi, 2002. The marine polychaete *Arenicola marina*: its unusual As compound pattern and its uptake of arsenate from seawater. *Mar. Environ. Res.*,53: 37–50.

Hussain, S., Atkinson, A., Thompson, S.J. and Khan, A.T. 1999. Accumulation of mercury and its effect on antioxidant enzymes in brain, liver and kidney of mice. *J. Environ. Health Sci.*, 34: 645-660.

Kakkar, P., Das, B. and Visvanathan, P.N. 1984. A modified spectrophotometric assay of superoxidase dismutase. Ind. *J. Biochem. Biophys*., 211: 131-132.

Miller, O.M., Lund, B.O. and Woods, J.S. 1991. Reactivity of Hg (II) with superoxide : evidence for the catalytic dismutation of superoxide by Hg (II). *J. Biochem. Toxicol.*, 6: 293-298.

Nammaluvar, P. 1985. Heavy metal pollution in Adyar Eastraury, India Proc symp Assxes *Environ. Poll*. 235-338.

Nichens, W.G and Samuelson, B. 1968. Formulation of malondialdehyde from phospholipid arachidonate during microsomal lipid peroxidation. *Eur. J. Biochem*., 6: 126-130.

Palaniappan, PL. RM. and Karthikeyan, S. 2009. Bioaccumulation and depuration of chromium in the selected organs and whole body tissues of freshwater fish *Cirrhinus mrigala* individually and in binary solutions with nickel.*J.Environ.Sci., 21*:229-236.

Ramanathan, K., Shila, S., Kumaran, S. and Panneerselvan, C. 2003. Ascorbic acid and α-tocopherol as potent modulators on as induced toxicity in mitochondria, *J. Nutr. Biochem.* 14: 416–420.

Rana, S.V.S., Rekja, S. and Seema, V. 1996. Protective effects of few antioxidants in liver function in rats treated with cadmium and mercury. *.Ind.J.Exp. Biol*, 34: 177-179.

Romeo, M., Bennan, N., Gnassia – Barell: M., lafaurime, M., and Grnad, J.P. 2000. cadmium and copper display

different responses towards oxidative stress into kidney of the sea bass. *Aquat. Toxicol*., 48. 185-194.

Rotruck, J.T., Pope, A.L. Ganther, H.E., Swanson, A.B., Hateman, D.G and Hoekstra, W.G. 1973. Selenium biochemical role as a component of glutathione peroxidase. *Science*, 179: 588-590.

Sinha, K.A. 1972. Colorimetric assay of catalase. *Analytical Biochemsitry*, 47: 389-394.

Soundararajan, M., Veeraiyan, G., and Sankar Samipillai, S. 2009. Arsenic-induced oxidative stress in fresh water catfish *Tilapia Mossambica. J.Phy.,1:267-276.*

Sreedevi, P., Suresh, A., Sivaramakrishna, B., Prabhavathi, B., Radhakrishnaiah, K., 1992. Bioaccumulation of nickel in the organs of the freshwater fish, *Cyprinus carpio*, and the freshwater mussel, *Lamellidens marginalis*, under lethal and sublethal nickel stress. *Biosphere*, 24, 29–36.

Stohs, S.J. and Bagchi, D. 1995. Oxidative mechanism in the toxicity of metal ions. *Free. Radic. Boil. Med.* 18: 321-336.

Tjalve, H., Gottofrey, J., Borg, K., 1988. Bioaccumulation, distribution and retention of 63Ni2+ in the brown trout (*Salmo trutta*). *Wat. Res*., 22, 1129–1136.

Yamanaka, K., Hasegawa, A., Sawamura, R. and Okada, R. 1991. Cellular respond to oxidative damage in lung induced by the administration of dimethyl arsinic acid a major metabolite of inorganic arsenics in mice. *Toxicol. Appl. Pharmacol*., 108 : 205-213.

****