Chromium Toxicity Induced Histopathological Alterations in The Liver of Fresh Water Carp Fish, Catla Catla

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Abstract: *Catla catla*, the fresh water carp fish, fingerlings when exposed to different sublethal concentrations of chromium for a different periods 1, 8, 16, 32 days of time brought changes in the structure and morphology of the nonosmatic organs such as liver. In trivalent chromium exposed fish, in the liver a few pathological changes were seen during the initial days of exposure and then structural reorganization observed in later days of exposure. The degenerative changes observed in the liver of the fish exposed to the sublethal concentration of hexavalent chromium were the disarray of liver cords, dilation of sinusoids, vacuolization in liver tissue, etc., all these changes confirmed structural disruption by hexavalent chromium ions on prolonged exposure.

I. INTRODUCTION

Freshwaters are highly vulnerable to pollution since they act as immediate sinks for the consequences of human activity always associated with the danger of accidental discharges or criminal negligence (Vutukuru, S., 2003). The contamination of freshwater with a broad spectrum of pollutants has become a matter of concern all over the world (Voegborlo et al., 1999; Vutukuru, 2005; Rauf et al., 2009). The concentration of heavy metals is especially high in water bodies that deteriorate the life sustaining quality of water and damage both flora and fauna (Kasherwani, D., et al., 2009; Zyadah, M.A et al., 2000; Lliopoulou - Georguadaki, J.et al., 2001; Verma, et al., 2005; Sharma, et al., 2005).

Heavy metals toxicity received considerable attention in aquatic organisms especially in fish (Javed, M, 2002). The metals enter fish bodies through body surface, gill or the digestive tract (Vincent, S., 2002). The alteration in biochemical contents in different tissues of fish due to toxic effects of different heavy metals and pesticides have been reported by number of workers Verma et al., (1983); Gupta et al., (1987); Khan et al., (1992); James & Sampath (1995); Das et al, (1999); Khare and Singh (2002); Desai et al, (2002); Remia et al, (2008); Hadi et al, (2009) and Ganeshwade, (2011).

The higher levels of trace elements such as lead and chromium in liver relative to other tissues may be attributed to the affinity or strong coordination of metallothionein protein with these elements (Ikem et al., 2003). Toxicological histopathology gives useful data concerning the changes induced by toxicants at cellular levels. All the tissues and organs in the body of an animal may be potential targets for the toxic effects of any chemical or heavy metal. The finer cytoarchitectural changes produced during chemical intoxication can be traced by microscopic examinations of the tissues; such studies may explain to certain extent the tissue specificity of the drug action. Their field of study is called histotechnology (Merck Source, 2002 and Stedman’s medical dictionaries, 2005).

II. MATERIALS AND METHODS

Catla catla (Hamilton, 1822), the Indian major carp is an economically important edible fish, having a great commercial value, occurs abundantly in fresh water tanks and ponds, collected from the department of fisheries, Anantapur, Andra Pradesh, and were immediately transported in big fish containers in the laboratory. The fish were fed with commercial fish pellets having around 40% protein content, and allowed to acclimatize for 15 days.

Then the fish were isolated into batches having weight of 10±2gms were maintained in static water without any flow. Water was renewed every day to provide fresh water rich in oxygen. The quality of dechlorinated tap water used for the experiment was analysed and various parameters such as dissolved oxygen - 6.8mg/l, alkalinity-130mg/l, hardness-125mg/l and pH-7.3 were measured and maintained. Water temperature was maintained between 22 ± 3oC as recommended by APHA during experiment. During experimentation water was aerated once a day to prevent hypoxic conditions. As the level of toxicity reported to vary with the interference of extrinsic and intrinsic factors like temperature, salinity, PH, hardness of water, exposure period, density of the animals, size, sex etc., (Sivaramakrishna et al., 1991), and precautions were taken throughout this investigation.
Lethal concentration (LC50) of chromium chloride (trivalent and hexavalent) to fish Catla catla was determined by “Probit method” of Finney (1971). Based on the fact that the effect of a metal on fish becomes consistent within 96 hour of exposure (Eisler, 1977), LC50S/96 hours of trivalent and hexavalent chromium are considered as lethal concentrations. So, about 1/10 th of the 96 h LC50 lethal concentration was taken as sublethal concentration i.e., 59.68mg/l, 100 mg/l(Cr as 35.40mg/lit) were the lethal concentrations, 5.96 mg / l of trivalent chromium and 10 mg /l(Cr as 3.54 mg/lit) of hexavalent chromium respectively was taken as the sublethal concentration for further studies.

The effects of sublethal concentrations of trivalent and hexavalent chromium on the fish were studied at different periods of exposure in order to understand the influence of time over toxicity. Thus 1, 8, 16 and 32 days were chosen to observe the short term and long term effects of trivalent and hexavalent chromium on the fish Catla catla. After the completion of stipulated exposure period, the fish were sacrificed and isolated tissues such as liver under laboratory conditions for biochemical analysis and histopathological studies. These tissues were removed and washed with saline then fixed in buffer formalin (10%) processed for sectioning (5-6um) and staining with haematoxyline and eosin. The histological sections of the liver were taken by adopting the procedure as described by Humason (1972). Photographs were taken.

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**Fig: III** Fish Control Liver showing continuous mass of large hexagonal cells laminae, granular cytoplasm distinct nuclei, hepatic labarynth of laminae hepatocytes (HC), blood sinusoids (S), with lower magnification (10X); and Higher magnification (40X).
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Fig: IIIa. Fish exposed to sublethal conc. of trivalent chromium at day 1, large hexagonal cells, granular cytoplasm, distinct nuclei, hepatic labarynth hepatocytes (HC) blood sinusoids (S), with lower magnification (10X); and Higher magnification (40X).

Fig: IIIb. Fish exposed to day 8, vacuolization in hepatic cells (HC), dilation of blood sinusoids (S), Liver cords disarranged, atrophic changes in hepatocytes (HC), with lower magnification (10X); and Higher magnification (40X).

Fig: IIIc. Fish exposed to day 16, hepatocytes (HC) with vacuoles, globular bodies, polygonal hepatocytes hypertrophied, cell membranes thickened necrosis in pancreatic acini, with lower magnification (10X); and Higher magnification (40X).

Fig: IIId. Fish exposed to day 32, sinusoids degenerated, intercellular gaps (IG), pancreatic acinar cells (PAC) and degeneration of hepatic cells (HC), with lower magnification (10X); and Higher magnification (40X).

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Fig: IIIe. Fish exposed to sublethal conc. of hexavalent chromium for day 1, near to normal liver hexagonal cells, granular cytoplasm distinct nuclei some blood sinusoids (S), with lower magnification (10X); and Higher magnification (40X).

Fig: IIIf. Fish exposed to day 8, dilation of blood sinusoids (S), hypertrophic nuclei, atrophic changes, vacuolization in liver tissue, granular degeneration and lamellar structure (LS) dissolution, with lower magnification (10X); and Higher magnification (40X).

Fig: IIIf. Fish exposed to day 16, vacuolization in hepatocytes (HC), loss of polygonal shape hypertrophy, cell membranes thickened, pancreatic acini around blood capillaries necrosed, with lower magnification (10X); and Higher magnification (40X).

Fig: IIIG. Fish exposed to day 32, atrophy of liver cords, shrinkage of hepatocytes (HC), granular degeneration, cytoplasmic disintegration hepatocytes (HC) and the sinusoids (S) highly dilated, with lower magnification (10X); and Higher magnification (40X).

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Fig: IIIH. Fish exposed to day 32, atrophy of liver cords, shrinkage of hepatocytes (HC), granular degeneration, cytoplasmic disintegration hepatocytes (HC) and the sinusoids (S) highly dilated, with lower magnification (10X); and Higher magnification (40X).

III. RESULTS AND DISCUSSION

The structure of the normal liver of the fish consists of a continuous mass of large hexagonal cells forming laminae. These cells contained granular cytoplasm with distinct nuclei either eccentric or slightly centrally placed. Suspended in the hepatic labarynth were seen some blood sinusoids. Each lamina of the liver was separated by the thick wall of the peripheral cells (Fig III).

IV. CONCLUSIONS

The degenerative changes observed in the liver of the fish exposed to the sublethal concentration of hexavalent chromium were supported by the metabolic disorders observed in it. The disarrayed structure of liver cords, hepatic cells vacuolization sinusoids dilation, severe degree of nuclear atrophy, coagulation of blood cells followed by the hepatocyte shrinkage and dissolution of laminar structure suggest that the depletion in glycogen reserves. All these biochemical disorders observed in the liver of the fish exposed to the sublethal concentration of hexavalent chromium could be due to its gradual structural disorganization. Bengeri and Patil (1986) stated that the concentration of a metal is more.
important in bringing the histological changes in the liver of the fish; hence these changes could be used as a tool for assessing the toxic effects of heavy metals in aquatic environment.

REFERENCES
