

Effects of Chromium on Histological Alterations of Gill, Liver and Kidney of Fresh Water Teleost, *Cyprinus carpio* (L.)

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Abstract: Lethal effects of chromium on the histological alterations of fresh water teleost, *Cyprinus carpio* were studied. The fish exposed to lethal concentrations of chromium for 96 h showed clubbing of the secondary lamella in the ends, fusion of adjacent secondary lamella, epithelial lifting, necrosis and curling of secondary lamella. Hepatic cirrhosis, fatty changes, degeneration of parenchyma cells results in atrophy, tissue ischemic and extensive necrosis of liver cells were recorded. Hypertrophy of epithelial cells, contraction of glomerulus, increase of space inside the grouping of tubules, distortion of architecture, glomerular edema were observed when the fish exposed to lethal concentrations of chromium. Bowman's capsule, atrophy and dispersed interrenal cells with pyknosis of some nuclei were observed in kidney of *C. carpio* exposed to lethal concentrations of chromium.

Key words: *C. carpio*, chromium, histology, lethal toxicity, organs, India

INTRODUCTION

Population explosion, process of urbanization and industrialization has no doubt augmented food production and better life amenities but only at the cost of concomitant degradation of the aquatic environment. The disposal of waste leads to the contamination of the rivers, lakes and chronically affects the flora and fauna. Ecological and environmental issues have aggravated over the last two decades, threatening extensive devastation of flora and fauna in many parts of the world. The fish habitats are being contaminated alarmingly through a number of aquatic pollutants (Oner *et al.*, 2008). Among these pollutants, heavy metals are most injurious for fish life. Metals also become increasingly concentrated at higher trophic levels, possibly due to food-chain magnification (Martin and Knauer, 1973).

During all contamination processes in fish, heavy metals cross biological barriers, the gill epithelium and skin for direct route and the wall of digestive tract for indirect route (Lloyd, 1992) and accumulate mainly in metabolically active tissues such as the kidney, liver and gills. Fish are often at the top of the aquatic food chain and may concentrate large amounts of some metals, this metal accumulate in fish organs and made many disturbances in its vital processes (Gomaa *et al.*, 1995).

Histology acts as an integrated parameter, providing a more complete evaluation of the organism's health, effectively mirroring the effects of exposure to environmental pollutants (Teh *et al.*, 1997; Van Der Oost *et al.*, 2003). Chromium compounds are frequently encountered as environmental pollutants and have been known to produce toxic, mutagenic (Kuykendall *et al.*, 2006) and carcinogenic effects (Prabakaran *et al.*, 2007) in biological systems although, Cr is an essential nutrient. Hence, the present study aims to investigate the histological alterations of gill, liver and kidney of *Cyprinus carpio*, exposed to lethal concentrations of chromium.

MATERIALS AND METHODS

The fingerlings of carp, *Cyprinus carpio*. L (20±2 g) were procured from Tamil Nadu Fisheries Development Corporation Ltd. (Fish farm) Mettur, Tamil Nadu, India and were brought to laboratory without any mechanical injury and washed with 0.1% KMnO₄ solution to remove dermal infection if any and were acclimatized to laboratory conditions (24.2°C) for 1 month. Fish were fed *ad libitum* with commercial fish feed once a day. A major portion of the water was changed daily in order to avoid any accumulation of excretory products and unused feed

which might cause further stress to the fish. Preliminary tests were carried out to find the median lethal tolerance limit (LC_{50}) of the fish to the toxicants chromium for 96 h. After determining LC_{50} for 96 h, 500 fish were stocked in a large tank (120×180×90 cm) after it was cleaned and disinfected with potassium permanganate. Fish with an average weight of (20±2 g) were selected for the experiment. The acute toxicity experiment (96 h) was carried out in two circular plastic tubs, filled with 100 L of water. A normal pH (7.1) and chromium concentrations (LC_{50} for 96 h) were maintained throughout the experiment. About 100 fish were introduced into each tub. Control was maintained in two circular plastic tubs with 100 fish per tub. The fish were fed *ad libitum* with commercial feed once in a day. A facility for oxygenation of the test solution was provided.

Tissue preparations for histological studies: At the end of acute exposure time (96 h), fish of all the groups were sacrificed by decapitation. Gill, liver and kidney of control and treated fish were dissected out, fixed in 10% buffered formalin and then processed by conventional method, sectioned at 6 μ m and stained with Haematoxylin-eosine (Bancroft *et al.*, 1996). The sections were observed under light microscopy. The histological changes in the tissues were examined in the randomly selected sections from each fish. Histological changes induced by treatments in the tissues were photographed using Nikon photomicroscope.

RESULTS

In fish, gill is the 1st organ to which the pollutant comes into contact. Hence, it is more vulnerable to damage than any other tissue. The gill morphology of untreated fish (control) of common carp remains an ordinary structure in which lamella are lined by squamous epithelium composed of non differentiated cells (Fig. 1). The damage to gill of fish exposed to the higher concentration of Cr were severe. During lethal treatment, Clubbing of the ends of the secondary lamellae, fusion of adjacent secondary gill lamellae and necrosis in the primary lamellae, epithelial lifting, necrosis and curling of secondary lamellae were well marked (Fig. 2).

Hyperplasia and hypertrophy of nuclei were seen. Besides these changes, pyknotic nucleoli, hemorrhage, vacuolization, swelling and degeneration of the epithelial cells and pillar cells were prominent. The morphological section of liver of untreated (control) fish showed the normal parenchyma cells arranged to form lattice net work. The interspaces are the sinusoid of thin strip with sparse connective tissues. Hepatocyte is normal and exhibits a

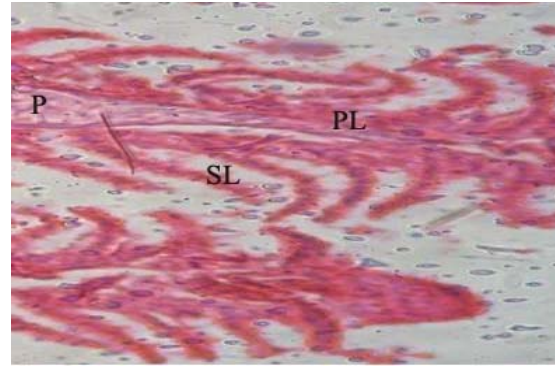


Fig. 1: Histological section of gill of control fish showing normal Primary Lamella (PL), Secondary Lamella (SL) and Pillar Cells (P)

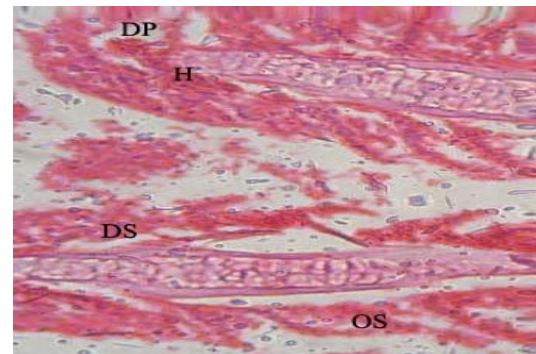


Fig. 2: Histological section of gill of fish exposed to lethal dose of Cr for 96 h showing fusion of gill filaments, over lapping of Secondary lamella (OS), Detachment of Secondary lamella (DS), Degeneration of Primary lamella (DP) and Hemorrhage (H)

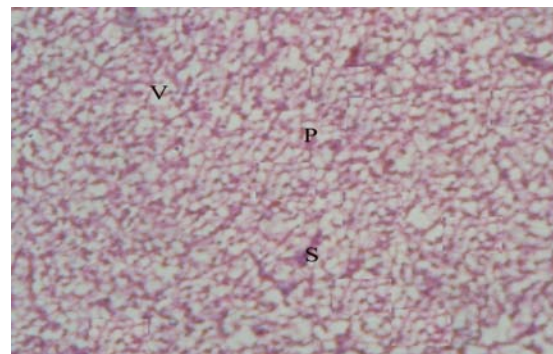


Fig. 3: Histological section of liver of control fish showing the network of Parenchyma cells (P), convergence of Sinusoid (S) into the large central Vein (V)

homogenous cytoplasm around the centrally located spherical nucleus (Fig. 3). Hepatic cirrhosis, fatty

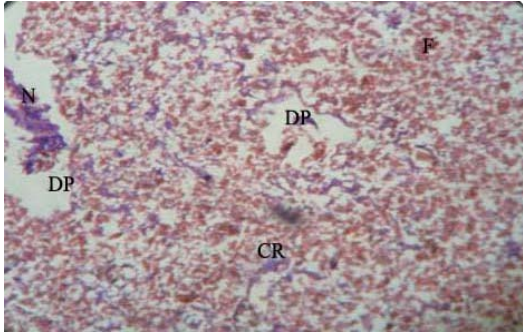


Fig. 4: Histological section of liver fish exposed to lethal dose of Cr for 96 h showing Cirrhosis (CR), Fatty changes (F), Degeneration of Parenchyma cells (DP) and Necrosis (N)

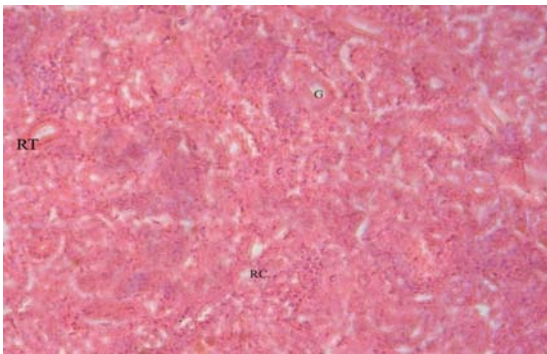


Fig. 5: Histological section of kidney of control fish showing normal arenal Tubules (RT) and Glomerulus (G)

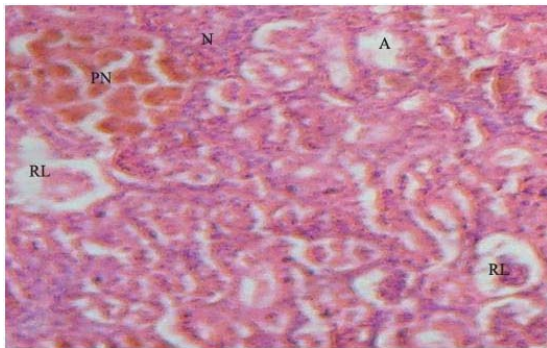


Fig. 6: Histological section of kidney of fish exposed to lethal dose of Cr for 96 h showing Reduction of Lumen (RL), Atrophy (A), Necrosis (N) and Pyknotic Nuclei (PN)

changes, degeneration of parenchyma cells results in atrophy, tissue ischemic and extensive necrosis of liver cells were recorded when the *C. carpio* exposed to lethal concentration of chromium for 96 h (Fig. 4). The section of control kidney tissues exhibits an ordinary pattern with no

abnormal changes in the renal cells (Fig. 5). Hypertrophy of epithelial cells, contraction of glomerulus, increase of space inside the grouping of tubules, distortion of architecture, glomerular edema and inflammatory cell infiltration were observed in the kidney when the fish exposed to lethal concentrations of chromium. Bowman's capsule, atrophy and dispersed interrenal cells with pyknosis of some nuclei were observed in kidney of *C. carpio* exposed to lethal concentrations of chromium (Fig. 6).

DISCUSSION

Heavy metals caused the serious impairments in the metabolic, physiological and structural system which is present in high concentration in the milieu (De Smet and Blust, 2001; Kuykendall *et al.*, 2006). Among them chromium is the most harmful pollutant for fish (Al-Akel and Shamsi, 1996). The severity of damage depends on the toxic potentiality of a particular compound or toxicant accumulated in the tissue and therefore, exposure to this heavy metal may adversely affect various organs/systems in fish which ultimately could lead to overall toxic impact on organs like gill, kidney and liver may seriously affect the metabolic as well as physiologic activities and could impair the growth and behavior of fish. The chromium exposure exhibited marked degenerative changes in the histology of liver and gill tissues. Similar kind of toxicity affects effects were noticed in various fish exposed to other heavy metals (Karlsson-Norggren *et al.*, 1986; Randi *et al.*, 1996; Arellano *et al.*, 1999; Ortiz *et al.*, 2003; Olojo *et al.*, 2005; Figueiredo-Fernandes *et al.*, 2007).

The gills serve as a respiratory organ in fishes that has direct contact with water. Gill is the principle tissue site for concentrating trace metals and it has different binding affinity for metals. The damage to gills of fish exposed to the higher concentration of Cr were severe than the fish exposed to the sublethal concentrations of the toxicant. During lethal treatment, Clubbing of the ends of the secondary lamellae, fusion of adjacent secondary gill lamellae and necrosis in the primary lamellae, epithelial lifting, necrosis and curling of secondary lamellae were well marked (Fig. 2).

Hyperplasia and hypertrophy of nuclei were seen. Besides these changes, pyknotic nucleoli, hemorrhage, vacuolization, swelling and degeneration of the epithelial cells and pillar cells were prominent. Mishra and Mohanty (2008) also reported the similar findings that epithelial hyperplasia, lamellar fusion, epithelial lifting, necrosis and desquamation and aneurism and curling of secondary lamellae. The gills of heavy metal exposed fish showed

epithelial lesions, the edema in the filament region with intense lamellar vasodilatation and numerous space are found in the metal treated *C. carpio* fish. Similar results were reported by Vinodhini and Narayanan (2009).

Hepatic tissues of the fish treated with sublethal concentrations chromium showed hepatic cirrhosis patchy degeneration around the paranchymal cells, more congestion and acute and extensive necrosis of liver cells have been recorded at the end of the periods (Fig. 4). The liver is the vital organ of detoxification. The exposure alterations in the liver due to toxicity effect are associated with degenerative changes (Arellano *et al.*, 1999; Olojo *et al.*, 2005; Figueiredo-Fernandes *et al.*, 2007). The changes caused by chromium in the liver such as vacuolization, necrosis were reported for copper exposure (Figueiredo-Fernandes *et al.*, 2007).

In the liver, chromium caused a loss of normal architecture, fatty changes, extensive vacuolization in hepatocytes, eccentric nuclei and Kupffer cell hypertrophy (Das and Mukherjee, 2000). Similar vacuolation in the hepatocytes were reported by Bruno and Ellis (1988) in *Salmo solar* exposed to tributyltin In the present study also recorded similar hypertrophy of epithelial cells, contraction of glomerulus, glomerular edema and inflammatory cell infiltrations. Pyknosis nuclei have also been observed in the chromium treated *C. carpio* fish (Fig. 6).

During the sublethal treatment, hypertrophy in the epithelial cells of the renal tubules, glomerulus contraction and increase in the space inside the bowmans capsule and atrophy have also been observed in the toxicant exposed fish (Fig. 6). Kirubakaran and Joy (1988) reported the similar results that hypertrophy of epithelial cells, degenerations of renal cells, reduced lumen in the mercurial compounds exposed fish *C. batrachus*. In the present study also heavy metal chromium caused severe alterations in the gills, liver and kidney of fish exposed to lethal concentrations of chromium.

CONCLUSION

Histological alterations in carp, under the influence of chromium can be used as a sensitive model to monitor the aquatic pollution. The current result indicates that the heavy metal contamination definitely affect the gills, liver and kidney of the fish exposed to chromium toxicity. Hence, a scientific method of detoxification is essential to improve the health of these economic fish. The present research served as experimental tools for the evaluation of environmental pollution.

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