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Histopathological Effects of Cadmium Chloride (CdCl₂) Exposure on the Liver of Freshwater Fish *Heteropneustes fossilis*

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Abstract:

The histopathological effects of CdCl₂ on the liver of *H. fossilis* by acute and chronic exposure of 25 mg/l, 50 mg/l, and 75 mg/l during interval of 30 and 60 days were studied. Similarly, control groups were operated simultaneously with experimental set-up to study alterations in the morphology and architecture of liver in order to understand the injury. The Liver of *H. fossilis* Normally, the liver of *H. fossilis* displayed a bilobed structure consisting of polygonal hepatocytes that were radially disposed around the hepatic veins, which maintained its characteristic architecture with an evident nucleus and interspersed binding connective tissue. Histopathological changes by exposure to CdCl₂ Both acute and chronic exposure were associated with hematogenesis characterized by darker fragile livers with hepatocyte hypertrophy. Hepatocytes lost the polygonal shape and were dispersed; cytoplasmic vacuolization, pyknotic changes, and bile canaliculi dilation. Prolonged exposure worsened these effects, and resulted in marked atrophy accompanied by necrosis, cellular shrinkage and tissue distortion. In addition, higher concentrations of CdCl₂ resulted in enlarged Kupffer cells, ruptured nuclei, cellular necrosis and focal necrosis in liver tissue. Our results suggest that progressive histopathological alterations in the liver of *H. fossilis* due to CdCl₂ exposure, signifying the susceptibility of this organ to heavy metal toxicity and its ecological prospects in aquatic environs.

Key Words: Histopathology. Liver, *H. fossilis*, CdCl₂

Introduction:

In the list of heavy metals such as lead (Pb), mercury (Hg) and cadmium (Cd) are considered to cause public health hazards. Cd is a naturally occurring non-essential heavy metal present at higher concentrations in association with Cd-rich soils, including shales, oceanic and lacustrine sediments, and phosphorites. However, more than 90% of Cd in the surface environment is the result of industrial and agricultural processes (Pan *et al.*, 2010). Cd is primarily stored in the liver and kidneys, which account for half of the body's total stores of Cd, rest in bone, pancreas, adrenals and placenta (Pope

and Rall, 1995). Nowadays, the aquatic life of water resources/bodies is in danger due to continuous addition of industrial pollutants, domestic and agricultural wastes, heavy metals etc.

Material and Methods:

The experimental design was based on Static Renewal Test (SRT), Range Finding and Definitive Test (Acute Toxicity Test) described by Sprague²² and USEPA²³. For each bioassay test, a series of three test concentrations of Cadmium and a control were used.

The tissue samples were taken from the fishes exposed to the first three concentrations only. At the end of the experiment (60 days), live fish samples were collected from the above-mentioned three concentrations, sacrificed and their liver and kidney were excised out, they were subsequently washed in distilled water and processed through graded series of alcohol, cleared in xylene and embedded in paraffin wax. Sections of 10-micron thickness were cut; stained with Harris hematoxylin and eosin and mounted in DPX. Stained sections were examined with light microscope for histopathological changes and also, light photomicrographs were taken. The morphological changes of the liver and kidney sections noted in the experimental fish were compared with those of control fish.

Reagents:

Physiological saline (0.75%NaCl sol.), Neutral buffered formalin, Graded series of alcohol, Xylene, Harris hematoxylin, Eosin, DPX

Results and Discussion:

HISTOPATHOLOGICAL CHANGES

In the present work liver of *H. fossilis* has been obtained for their histopathological investigations. For above observation, fishes were kept in 25 mg/l, 50 mg/l and 75mg/l CdCl₂ mixture for durations of 30 and 60 days, with every aquarium containing experimental fishes, parallel control were set.

Liver of *H. fossilis* is bilobed consisting of two subequal lateral lobes disposed longitudinally, small anterior lobe and larger posterior lobe with the gallbladder between the two. The liver consists of polygonal glandular hepatic cells arranged in groups. The hepatic cells are provided with a centrally placed darkly stained spherical and granular cytoplasm.

In our study liver of control fish *H. fossilis* is a dark brownish red colored, bilobed gland composed of hepatocyte cells which are the parts of mass and forms a typical normal architecture. The hepatocytes

are arranged in a radial manner around hepatic veins to form hepatic cords. The cords are however formed of liver parenchyma which encloses the blood sinusoids. There are lymphatic glands which contain bile pigments and lymphocytes. The liver cells are polygonal in shape and contain a prominent nucleus which possesses densely stained nucleoli. The binding together of liver cells is brought about by the connective tissue (Plate 1).

After acute and chronic exposure to CdCl_2 , liver of *H. fossilis* becomes more fragile and darker in color but no tumor like out growth is seen anywhere in the liver. Histologically, following the exposure to CdCl_2 , hypertrophy of hepatic cells has been observed. Polygonal shape of the hepatic cells is completely lost at various places. Hepatocytes are found scattered in the hepatic tissue, vacuolization and pycnotic changes have been observed. Dilation of extra cellular spaces, bile canaliculi has also been noticed. Histopathological changes were much more remarkable after chronic exposure to CdCl_2 as higher degree of atrophy has been noticed. Principal histological changes are necrosis hypertrophy, shrinkage in various hepatic cells and complete deformation of polygonal shape of the hepatic cells. Progressive splitting in tissues and clumping of cells were important pathological lesions. Nuclei have also been noticed acentric.

In the liver of the fish exposed to CdCl_2 at 25 mg/l, histological changes included ruptured nucleus, increased Kupffer cell, ruptured hepatic tissue, cellular necrosis and increased pycnotic nucleus. Cd at 50 mg/l concentration induced highly distinct changes such as cellular necrosis, ruptured hepatic tissue, ruptured nucleus and focal necrosis in the liver of fish. Changes became more pronounced when the concentration of Cd was 75 mg/l. The changes included focal necrosis, increased pycnotic nucleus, cellular necrosis and ruptured hepatic tissue (PLATE 1).

The toxicity effect of heavy metals on liver has been studied by several workers. The effects of acute Cd on the liver of *H. fossilis* is in conformity to other similar kind of studies (Naigaga, 2002; Nasiruddin et al., 2009; Bais and Lokhande, 2012; Sharma et al., 2013; Selvanathan et al., 2013). Histological alterations like degeneration of hepatocytes, vacuolization, congestion of hepatic tissues, subcapsular vacuolization, necrosis, indistinct cell boundaries and pyknotic nuclei were observed in the liver of the catfish, *Clarias batrachus* exposed to Cd (Selvanathan et al, 2013). Degenerative changes like hepatocellular dissociation, necrosis and hypertrophy were observed in the freshwater fish, *Ophiocephalus striatus* exposed to CdCl_2 (Bais and Lokhande, 2012). Chemicals like DDT can induce notable histological changes in liver, and intestine also described. Elezaby et al., (2001) studied

hemorrhage, necrosis and lipidosis in the liver *Oreochromis niloticus* and *Clarias gariepinus* due to malathion and organophosphorus insecticide (Hostathion) toxicity.



Photomicrograph of section of Liver (control) of *H. fossilis*



Photomicrograph of section of Liver (30 days exposure of CdCl₂) of *H. fossilis*



Photomicrograph of section of Liver (60 days exposure of CdCl₂ control) of *H. fossilis*

Plate-1

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