TOXIC EFFECT OF COPPER CHLORIDE ON THE AIR BREATHING FISH, *HETEROPNEUSTES FOSSILIS* (BLOCH, 1794)

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Abstract

Environmental contamination from heavy metals is a serious problem of global dimension. Discharge of heavy metals into aquatic environment from various sources even below permissible levels creates health hazards in aquatic organism like fish. The harmful effect of any toxicant which was moved to water bodies can be assessed by investigating health of fish fauna. In the present study an attempt has been made to examine the sub lethal concentration (0.005mg/L) of copper chloride on fresh water fish Heteropneustes fossilis for a period of 7, 14, 21 and 28 days, which were duration dependent. All these changes in liver histology can be considered to monitor the water contaminated with heavy metals.

Key words: *copper chloride, aerial breathing, aquaria, sub lethal.*

Introduction

In the recent years industrial development and agricultural processes have resulted in increased levels of toxic metal in the environment, although relatively high concentration can also occur naturally. The contamination of fresh water with a wide range pollutant has become matter of concern over the last few decades. The natural aquatic system may extensively be contaminated with heavy metal released from domestic, industrial and man-made activities. Heavy metal contaminations have devastating effects on the ecological balance of the recipient environment and diversity of aquatic organism (Vutukura, 2005; Voegborlo *et al.,* 1999). Heavy metal contamination severely interfere with ecological balances of an ecosystem and produces devastating effect on environment quality anthropogenic inputs like waste disposal directly adds to the burden of environmental degradation (Farombi *et al.*,2007).

Untreated community wastes uses of fertilizers and pesticides as well as dumping of organic and inorganic waste from industries are increasing environmental pollution to a great extent. Heavy metals have been recognized as strong biological poison because of their persistent nature, toxicity tendency to accumulate in organism and undergo food chain amplification they also damage the aquatic fauna including fish (Dinodia *et al.,* 2002). Copper is a very important element which could influence the body metabolism and it is also a nutritional element for living being. But if the intake is to much will cause toxicity of pollutant is either acute or chronic (Nekoubin *et al.,* 2012). Majority of the studies concerning the effects of heavy metals on fish have been confined to the acute toxicity test with the death of fish. Hence in the present study an attempt has been made to assess the toxic effect of copper chloride ($CuCl_2$) on fish *Heteropneustes fossilis*.

Materials and Methods

In the present work, the fresh water fish *Heteropneustes fosillis* were selected. The fish were obtained for the

experimental purpose from Adan dam, Taluka –Karanja (Lad) Dist – Washim.M.S. India. They were washing with 1% of $KMNO_4$ solution for five minute for dermal disinfection. The fish are allowed to acclimatize to the laboratory condition for a period of fortnight before conducting the experiment. Particularly in the morning hours, fish fed on small pieces of boiled eggs once in a day. The 16 fish including male and females weighing between 40 to 50 gm and length between 22-24 cm. were selected for experimental work. They were maintained in separate aquaria containing aged tap water. The physicochemical parameters of used tap water as per standard method were determined periodically (APHA 1998). To study the toxic effect of $CuCl_2$ on fish *Heterpneustes fossils*, 96h Lc_{50} and sub lethal concentration of $Cucl_2$ for the fish was taken from literature. This was 0.005 mg/lit for 96h Lc_{50} value, approximately 5 times less concentration (O.001mg/l) used to expose the fish for 28 days. Parallel set of control fish were run simultaneously in separate aquaria.

After 7,14,21 and 28 days parallel set of control as well as experimental fish groups were weighted and sacrificed immediately by giving blow on head and dissected. The liver tissue from both the control and experimental groups were removed and rinse in saline water to removed cell debris. Then all tissues were embedded in paraffin wax using routine technique and section were cut 5µ thickness and stained with hematoxylene-eosin staining method. Histopathological observations were made to assess the effect of toxicant copper chloride on fish *Heropneustes fossilis*.

Results and Discussion

Experimental fish struggled hard for aerial breathing with their restricted swimming movement and poorer response to external stimuli. Fish lose equilibrium and slowly moved upward in a vertical direction. Thereafter fish become progressively lethargic.

Tissues like liver of the fish *Heteropneustes fossilis* showed varied degenerative changes observed after7, 14, 21 and 28 days exposure to copper chloride. Histological studies revealed that liver section from control fish show normal histoarchitecture by polygonal shaped hepatocyte with granular cytoplasm and centrally placed round nuclei. Hepatocyte arranged in well-organized hepatic cords and separated by narrow blood sinusoids.

In copper chloride toxicant exposed fish, the histopathalogical changes observed in liver were duration dependent. After 07 days copper chloride exposure, the hepatocyte showed disruption of regular cordial arrangement and prominent shrinkage in hepatic cells. The similar observations were recorded by Avinashe and Patil (2010) while studying the effect of selenium dioxide on the histopathology of the liver *Channa punctatus*.

After 14 days of exposure to the toxicant the nuclei of hepatocyte observed become prominent along with disarray of hepatic cords. Radhakrishanan and Hemlatha (2010) noted that hepatocyte with marked cytoplasmic vacuolization, sinusoids in most areas distended and central veins appeared severely damaged due to marked swelling and degeneration of the endothelial lining cells. Cope *et al.*, (1969) also reported swollen hepatocyte, disorientation of

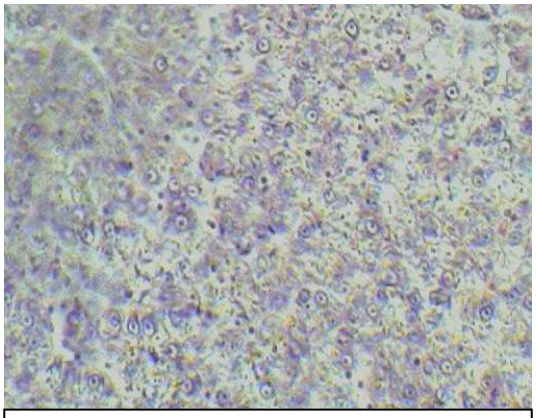

Fig. 2 : T. S. of liver of the fish *Heteropneustes fissilis* showing histomorphological changes after exposure to $CuCl_2$ for 7 days.

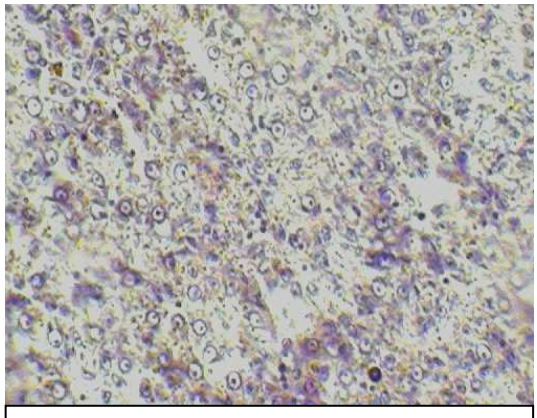

Fig.3 : T. S. of liver of the fish *Heteropneustes fissilis* showing histomorphological changes after exposure to $CuCl_2$ for 14 days.

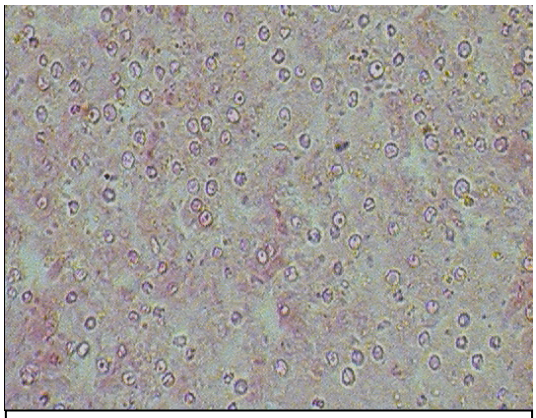

Fig. 1 : T. S. of liver of *Heteropneustes fissilis* showing control structure.

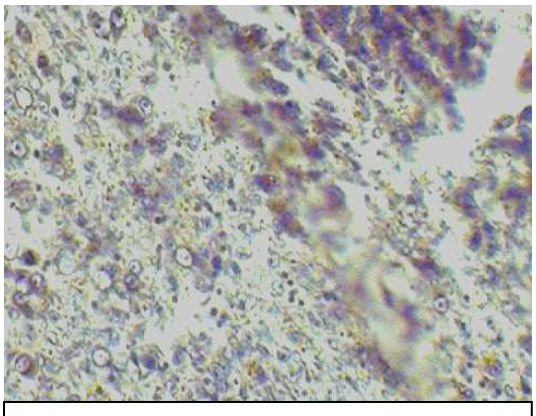

Fig.4 : T. S. of liver of the fish *Heteropneustes fissilis* showing histomorphological changes after exposure to $CuCl_2$ for 21 days.

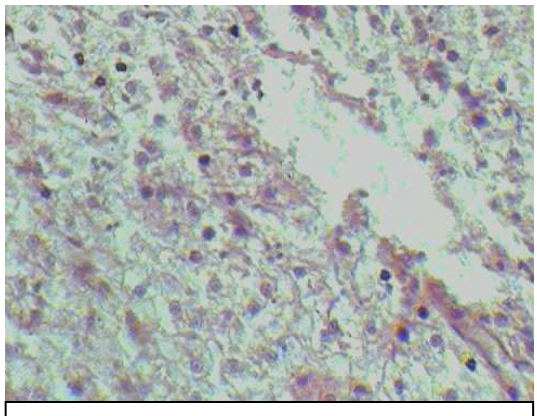

Fig.5 : T. S. of liver of the fish *Heteropneustes fissilis* showing histomorphological changes after exposure to $CuCl_2$ for 28 days.

liver cords, cytoplasmic vacuolization and karyolysis of hepatocytes of blue gills after exposure of dichlorobennil.

After 21 days of exposure, the hepatocyte becomes vacuolated and blood coagulation was also seen, shrinkage of blood vessels and clumped erythrocyte widely separated bile canalliculi were also noticed. Severe bile secretion and dilation of sinusoids were reported in the liver of *Hoplobatrachus occcipitalis* by Ikechukwu and Ajeh (2011) when exposed to sub lethal concentrations of cadmium. Rani and Ramamurthi (1989) and Dangre *et al.*, (2010) noticed pathological changes in liver tissues including engorgement of blood vessels, congestion, vacuolar degeneration of hepatocyte, necrosis of pancreatic cells.

After 28 days of exposure of $CuCl_2$ the high degree of atrophy was notice in the liver. There was typical change in the size of hepatocyte and their nuclei to periphery. The cell membrane ruptured.The vacuolization and clumping of hepatocyte was prominent. Other degenerative changes like increase in connective tissue damage that occur in hepatic cells of fish liver which was exposed to copper chloride toxicant. Ahmad *et al.,* (2011) when exposed the fish *Clarias batrachus* to 4 ppm and 8 ppm cadmium chloride for 60 days resulted in severe loosening and necrosis of hepatic tissue, hepatic cells lost their original shape, enucleation takes place in majority of cells and the centrally situated nuclei shifted to the periphery of the cells. Mohammad *et al.,* (2013) also reported the histopathological changes in the liver of *Clarias gariepinus* including loss of cellular architecture of liver, vacuolar degereation, pycnotic nuclei and focal area of necrosis of the hepatocyte.

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