International Journal of Applied and Universal Research **E-ISSN No: 2395-0269**
Volume IV, Issue I, Jan-Feb. 2017 Available online at: www.ijaur.com

CHANGES IN HISTOPATHOLOGY OF LIVER IN SNAKE HEADED FISH, *CHANNA PUNCTATUS* (BLOCH) WHEN EXPOSED TO ZNSO4

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ABSTRACT: - The aquatic ecosystem is faced with the threat of biodiversity loss due to indiscriminate use of pesticides. Other than targeted pests, pesticides affect a wide range of non-target organisms, such as invertebrates and fish inhabiting aquatic environment. The present study deals with the impact of zinc sulphate on histopathology of liver of *Channa punctatus* Bloch. In the laboratory condition fishes are divided into control and experimental groups. The Lethal concentration and sub lethal concentration of zinc sulphate for *Channa punctatus* Bloch. is calculated which is (6.6mg/L) and (3.3mg//L) respectively. Fishes are exposed for 7days, 14 days, and 21 days. Fishes showed severe histological changes in liver. The degenerative changes included the hepatocytes having disruption of regular hepatic cordal arrangement and also noticed the prominent shrinkage of hepatocytes. The nuclei of hepatocytes became prominent along with disarray of hepatic cords.

KEYWORDS: *Channa punctatus*, Liver, Zinc Sulphate.

INTRODUCTION

Freshwater is vital for various aquatic animals and plants but the quality of freshwater has been abruptly change due to the introduction of different chemicals including pesticides, industrial discharges, metallic pollutants etc. Aquatic systems are exposed to a number of pollutants that are mainly released from effluents discharged from industries, sewage treatment plants and drainage from urban and agricultural areas. Industrial growth is an important part of the evolution of human civilization and is vital for the development and property of any country. However, industries also often prove hazardous to aquatic life when their toxic effluents are discharged into water, more so when this is done without any pre-treatment. These pollutants cause serious damage to aquatic life (Mishra and Tripati, 2012).

In the recent years, world consumption of fish has increased simultaneously with the growing concern of their nutritional and therapeutic benefits. The American Heart Association recommended eating fish at least twice per week in order to reach the daily intake of omega-3 fatty acids. However, fish are relatively situated at the top of the aquatic food chain; therefore, they normally can accumulate heavy metals from food, water and sediments. The content of toxic heavy metals in fish can counter act their beneficial effects; several adverse effects of heavy metals to human health have been known for long time. This may include serious threats like renal failure, liver damage, cardiovascular diseases and even death.

Therefore, many international monitoring programs have been established in order to assess the quality of fish for human consumption and to monitor the health of the aquatic ecosystem (Moselhy *et al*., 2014).Zinc metal contamination definitely affects the aquatic life of the freshwater fish. In chronically exposed fish internal environment is disturbed gradually at cellular level. This poses the physiological stress inside the various fish tissues due to the progressively Zn metal accumulation in the course of time. (Kumar *et al.* 2015).

Histological study appears to be a very sensitive parameter and is crucial in determining cellular changes that may occur in target organs, such as the liver. Exposure to heavy metals may cause histological changes in the liver. Fish liver histology could therefore serve as a model for studying the interactions between environmental factors and hepatic structures and functions. It has been noted that heavy metals had a negative impact on all relevant parameters and caused histo-pathological changes in fish. Zinc is one of the most important trace elements in the body and participates in the biological function of several proteins and enzymes. Despite being an essential trace element, Zn is toxic to most organisms above certain concentrations. (Ho. 2004).The concentration of metals was found maximum in liver and minimum in muscles. The degree of accumulation among the five tissues differed and it was in the order: gill>liver>kidney>blood>muscle in case of Zn, gills>kidney>blood> liver>muscle. (Shukla *et al.* 2007).

MATERIALS AND METHODS

Fish collection and acclimatization: Live and apparently healthy fish specimens of *Channa punctatus*, available throughout the year in greater part of India, were procured from local fishermen of Adan dam, Taluka-Karanja (Lad) Dist. Washim, (MS). This fish were brought to the laboratory in well oxygenated bag without any injury. The fish were subjected to repeated washing with tap water and 0.1% $KMnO_4$ solution for 5 minutes, to remove dermal infections. Fishes were

allowed to acclimatize to laboratory condition for a period of fort night, in aerated glass aquaria in 10 days aged tap water. Particularly in the morning hours fish fed on small pieces of boiled egg once in a day.

Preparation of Experimental Aquarium: To study the effect of zinc sulphate i.e. $ZnSO_4$ on various organ & different biochemical parameters, the experiment were conducted in 2 separate aquariums. The lethal concentration (6.6mg/L) & sub-lethal concentration (3.3mg//L) of the experimental toxicant i.e. $ZnSO_4$ were collected from the literature. First set is the normal or control set while second set is referred as experimental set. In each set total 4 fish is taken for the experiment. Static bioassay was carried out as per standard methods (APHA 1986), aqueous solution of the toxicant was added drop by drop with constant stirring and then acclimatize 4 fishes were transferred to the glass aquarium containing 40 L of toxicant water in set 2. Simultaneously set containing total 4 fishes of normal & control set.

For above studies the acclimatized fishes were divided into 2 set.

Set1: Aquarium containing 4 fish in aged taped water which served as control set.

Set2: Aquarium with 4 fish kept in toxicant water containing 0.280 mg zinc as an experimental set.

The fish were fed on small pieces of boil egg once in a day especially early in the morning hours. Observation was made for 24 hours. The time at which fish losses its sense of balance was noted, keeping these observation in mind for final experiment. Fish also shows erratic movement. The experiment was carried out in sub-lethal concentration of toxicant $ZnSO_4$ for a period of 7, 14 and 21 days. Both the set of normal/control & experimental were run simultaneously in separate aquarium. The total 8 fish including male & females were selected for experimental work maintain in a separate aquarium containing free edge tap water.

HISTOPATHOLOGICAL STUDIES:

- After 7, 14, and 21 days the male & female fish of control as well as experimental set were scarifies immediately by giving blow on the head & were dissecting it.
- The tissue (Liver) were dissected out & rinsed in a saline to remove cell debris & blood stain.
- Then tissue (Liver) was cut into small pieces of desirable size & fix into aqueous Bovines Fluid. All possible precautions were taken to insure proper fixation of tissue.
- After fixation the tissue (Liver) was washed thoroughly under running water for 3 hours. Then it was dehydrated, clean & embedded in paraffin wax as per regular procedure.
- The section of the tissue (liver) was cut at 5µ thickness & was stained with Hematoxilin-Eosin.

Effect of sub-lethal concentration of zinc sulphate on a liver:

- The liver is the main organ for metal regulation in fish. In zinc sulphate exposed fish, the histopathological observe in liver were duration dependent. After 7 days of exposure the hepatocyte showed disruption of regular cordal arrangement and prominent shrinkage of hepatic cells observed in a structure of liver (Fig.2).
- After 14 days exposure to the zinc sulphate solution, in a liver structure shows the nuclei of hepatocytes became prominent along with disarray of hepatic cords (Fig.3).
- After 21 days exposure of fish to the toxicant, zinc sulphate noticed that the hepatocytes became vacuolated and blood coagulation also observed, shrinkage of blood vessels, clump erythrocytes and widely separated bile canaliculi were observed.
- Several other degenerative changes occurred due to acute toxicity of zinc sulphate includes picnotic nuclei and damaged connective tissue, disorganization of hepatic cells and hepatic cords. The necrosis and acute hemorrhage was also prominent, observed in a transverse section of liver (Fig.4).

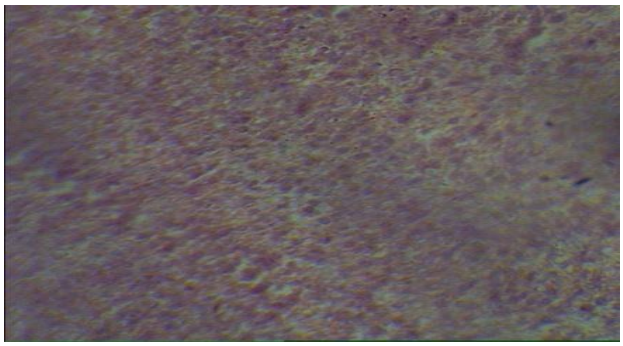

Fig.1.T.S. Through liver of fish *Channa punctatus* (Control) (Hematoxyline-Eosin × 630)

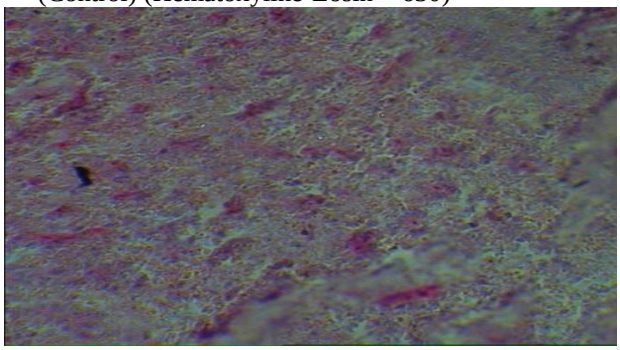

Fig.(2): T.S. Through liver of Fish *Channa punctatus* Exposed to Sub-lethal concentration of $ZnSO_4$ for 7days.

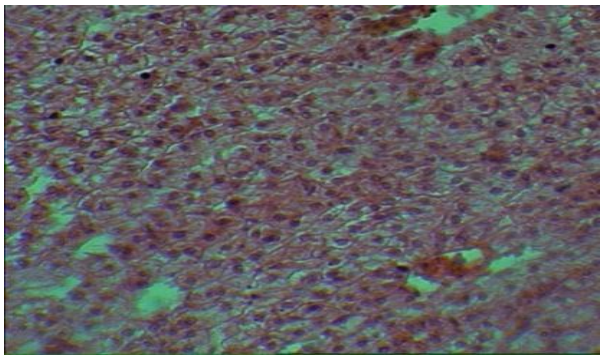

Fig. (3): T.S. Through liver of *Channa pucntatus* Exposed to sub-lethal concentration of $ZnSO_4$ for 14 days. (Hematoxyline-Eosin × 630)

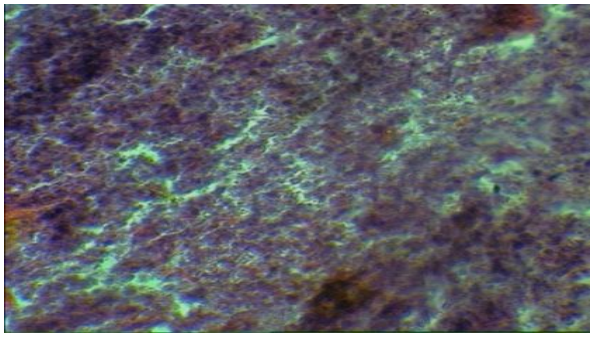

Fig.(4) : T.S. Through liver of *Channa pucntatus* Exposed to sub-lethal concentration of $ZnSO_4$ for 21 days. (Hematoxyline-Eosin × 630)

DISCUSSION:

Avinashe and Patil (2012), have reported that the typical change in the size of hepatocytes and their nuclei were displaced to the periphery and cell membrane of some cells were ruptured resulting into fusion between two or more cells, exhibiting binucleate or multinucleate appearance of cells at several places.

Hoq and Haque (2014), at higher concentration of zinc sulphate it exerts adverse effect by occurring structural damage, which affect the growth, development and survival of fish.

Figueiredo-Fernandes *et al*. (2007), concluded from their study that the changes induced by chromium in the liver hepatocytes such as vacuolization, necrosis and nuclear condensation were also reported for copper exposure.

Bhatkar (2011), the liver of *Labeo rohita*, after exposure of the fish to zinc chloride for ten days revealed swelling of hepatic nuclei, disorganization of hepatic cells with edematous hepatocytes and many cells were devoid of cytoplasmic contents.

Olurin *et al*. (2006), noticed the lesions developed in the liver might be due to the cumulative action of toxicant on blood and ultimately to other cellular structures.

There seems to be a definite correlation between tissue damage and certain physiological alterations.

Ojolo *et al*. (2005) and Saxena *et al*. (2008), liver is the major metabolic center and any damage to this organ would subsequently do, so many physiological disturbances leading to subsequent mortality of fish.

The result of the present investigation is conformities to the results reported by the workers as mention above.

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