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Histopathological Changes in the Liver of *Channa Gachua (Bloch)* Exposed to Biofertilizer

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Abstract: ***The present work deals with histopathological alterations in the liver of an air breathing teleost, Channa gachua (Bloch) after getting an exposure to a biofertilizer (mustard oil cake). The 96hr-LC50 dose of mustard oil cake for fish was found 5065mg/L. In this study, fishes were exposed to 1% and 5% of 96hr-LC50 dose for 96hrs and 45 days to the biofertilizer and their light microscopically studies were made. It showed the oozing of blood from the capillaries, congestion of blood vessels, vacuole formation, vasodilatation, fatty changes and hypertrophy. The result showed that mustard oil cake is slightly toxic to the Channa gachua as it had caused mild degenerative changes in fish organs like liver. The results are significant from environmental pollution and human health point of view.***

Keywords: Histopathology, Liver, *Channa gachua*, Biofertilizer

1. Introduction

The liver of fish performs a vital role in a number of metabolic processes and detoxifies the xenobiotics [1]. The histological changes in the liver over tale of dietary supplementation are important markers for the assessment of the effects of dietary alterations.

Biofertilizers contain microorganisms that are able to convert macronutrients from unavailable to available form during biological processes [2]. Mustard oil cake is one of the randomly used biofertilizers for supplementary feeding in pisciculture in India [3]. Moreover, The Government of India is also trying to promote use of biofertilizers along with fertilizers.

Histolopathogical alterations in response to environmental stress are authenticated in various fishes e.g. carbamate on *Channa marulius*, Malathion on *Channa punctatus* and quinalphos on *Channa gachua* etc [4]-[6]. In this context, study of adverse effect of biofertilizers of fishes seems essential.

Channa gachua (Dwarf Snakehead) is a hardy fish inhabiting in paddy fields and ponds in India and Shahabad region of Bihar. The IUCN status of this fish indicates that its number is decreasing [7].

The aim of this work is to study effect of acute and chronic sub-lethal doses of mustard oil cake on the histology of liver in *Channa gachua*. The work will help in deciding the optimum does of a biofertilizer as well as its safe level and toxicity status for higher yield of this fish.

2. Materials and Methods

Fresh specimens of *Channa gachua* (Hamilton, 1822) (BW: 55-75g and TL: 13-16cm) were obtained from local market of Arrah (Bhojpur) during 2018. They were acclimatized for a fortnight in Departmental Laboratory of VKS University, Arrah.

Fishes were randomly selected from the stock and divided into two groups. One group was taken to control or 'C' and other as treated or 'T'. Powder of mustard (*Brassica napus*) oil cake was selected as a biofertilizer for the experiment.

The 96hr-LC50 dose of mustard oil cake for fish was found 5065mg/L. In this study, fishes were exposed to 1% and 5% of 96hr-LC50 dose for 96hrs and 45 days to the biofertilizer and their light microscopically studies were made.

The control as well as experimental fishes were taken out of aquaria at the end of each exposure period and sacrificed by decapitation. The liver was dissected out and cut into small pieces each about 2-3 mm in size and immediately fixed in 2% calcium acetate in 10% formalin. After fixation the pieces were washed under slow running tap water for equivalent time of fixation. The pieces were dehydrated through alcohol grades for 30 minutes in each grade, cleared in xylene and embedded in paraffin wax. The sections were cut at 6µ thick. The sections were stained with haematoxylin and eosin staining, mounted in DPX and used for observations.

3. Observations and Results

The histological details of liver of controlled *Channa gachua* has been given in Figure 1. The histopathological changes after exposure to sublethal concentrations of biofertilizer in the liver of test fish have been depicted in figures 2 to 5.

Liver is the most important organ as it performs several functions at a time and playing a prominent role in metabolism of carbohydrates, proteins, lipids, storage of glycogen, synthesis of amino acids, secretion of bile and also in detoxification [8]-[9]. Any damage to it may lead to a number of functional disturbances. Liver cord disarray, shrinkage in the liver cells, degenerated nuclei and focal necrosis reported in *Channa punctatus* due to lead intoxication [10].

Liver in *Channa gachua* is bilobed, reddish brown and dense, located in the upper region of the body cavity. Right lobe is large and thick whereas left lobe is further subdivided

into anterior and posterior lobe. The gall bladder is embedded in the right lobe. The vascular system of this organ consists of two afferent blood vessels (hepatic artery and hepatic portal vein) and a single efferent vessel (hepatic vein) located at the hilum region of kidney.

The liver showed hexagonal parenchymal hepatic cells within the networks of bile canaliculi. Hepatic cells having large quantity of lipid glycogen granules in their cytoplasm and contain centrally placed spherical nucleus. Hepatic cells located around sinusoids forming cord like structure. There was no clear division of hepatic cells in lobules and distribution of bile duct and blood vessels were irregular. Portal vein covered with pancreatic acini and hepatic arteries having narrow lumen covered with thick layer of endothelial cells was noticed. Hepatocytes surrounding the central vessels like rosette pattern and in lumen there were erythrocytes. Von Kupffer cells or satellite cells were occasionally seen in sinusoids as well as glycogen vacuoles were also seen (Figure 1). Liver is mostly associated with detoxification due to its function, position and blood supply and it is most affected organ by toxicants [11].

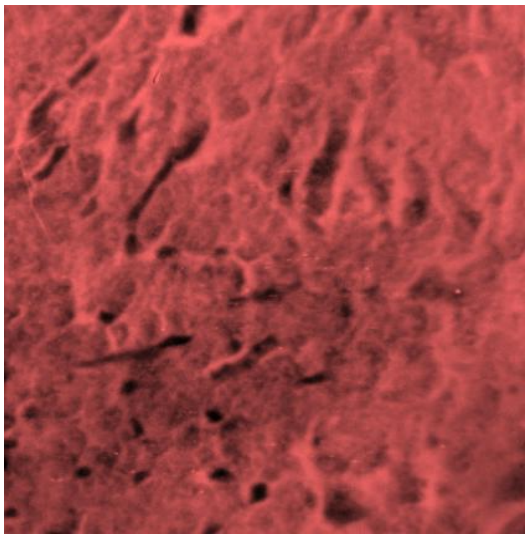

Figure 1: Micro photograph of T.S. of liver of *Channa gachua* under control condition (×400)

Various histopathological alterations are seen in liver depending upon exposure period and concentration of biofertilizer. Acute toxicity studies showed less vacuolization around the hepatocytes, initiation of disorganization of hepatic cords, loss of hepatocytes architecture. Initiation of sinusoidal spaces, less amount of blood vessels rupture, nuclear hypertrophy and degeneration of wall of the central vessel was also evident. Liver of *Channa punctatus* showed an increase in sinusoidal space and lipidosis during early days, followed by a recovery from the stress of mustard oil cake on the 28th day [3]. Considerable degradation in cellular structure viz. vacuolization, lipid infiltration, ruptured blood capillaries and hemorrhage, narrowing and disappeared wall of central vessel were observed. The vacuolization in hepatocytes might indicate imbalance between synthesis of biochemical substances and their release [12]. Severe necrotic patches, lesions, nucleus enlargement, hepatic cords disarray, complete destruction of hepatic cell membrane, cell proliferation, congestion and sinusoidal spaces were observed in liver of fish (Figures 2 and 3).

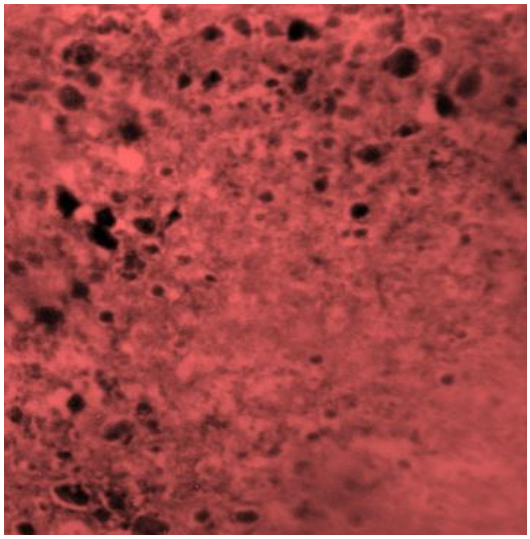

Figure 2: A. Micro photograph of T.S liver of *Channa gachua* after 96 hrs exposure to 50.65mg/L of mustard oil cake (×400)

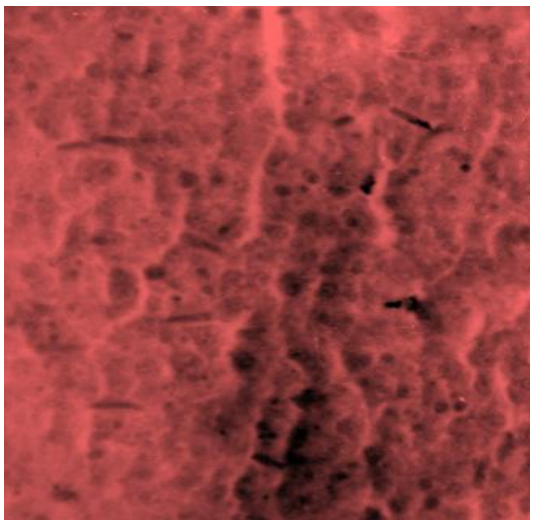

Figure 3: Micro photograph of T.S liver of *Channa gachua* after 96 hrs exposure to 253.25mg/L of mustard oil cake (×400)

When fishes were exposed to chronic toxicity, liver showed karyolysis, necrosis, severe hepatic cords disarray, parenchymal vacuolization, severe irregular shaped cells with irregular nucleus, necrotic patches, ruptured sinusoids and increasing sinusoidal spaces. Hepatocytes are seen in compactly arranged with significant cytoplasmic vacuolization due to swelling of cell and breakdown of cell boundaries. Enlargement of central vessels were observed with degeneration of endothelial lining leading to damage. severe necrosis and vacuolization in centrolobular region where as degenerative changes in hepatocytes began to swell with nuclei, disorganization of parenchymal architecture because of breakdown of hepatic cords, ruptured blood vessels and aggregation of blood cells were seen (Figures 4 and 5). Necrosis is observed in the liver cells in *Channa punctatus* when exposed to Alachlor [13]. Vacuolation, cellular swelling, nuclei pleomorphism, increase in kupffer cells and dilated sinusoids were observed in

Oreochromis mossambicus exposed to effective microorganisms in organically manured aquadams [14].

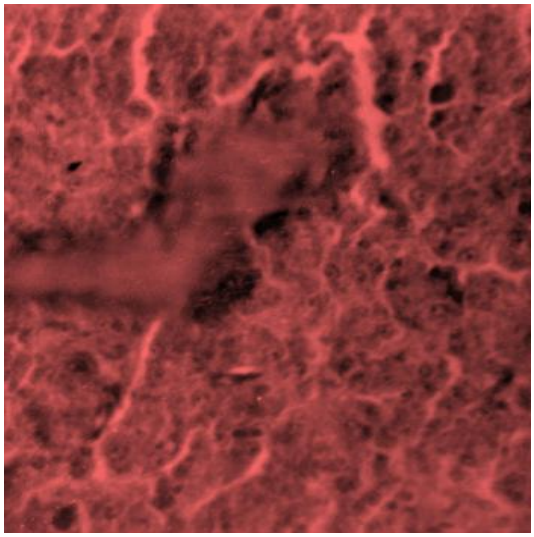

Figure 4: Micro photograph of T.S liver of *Channa gachua* after 45days exposure to 50.65mg/L of mustard oil cake (×400)

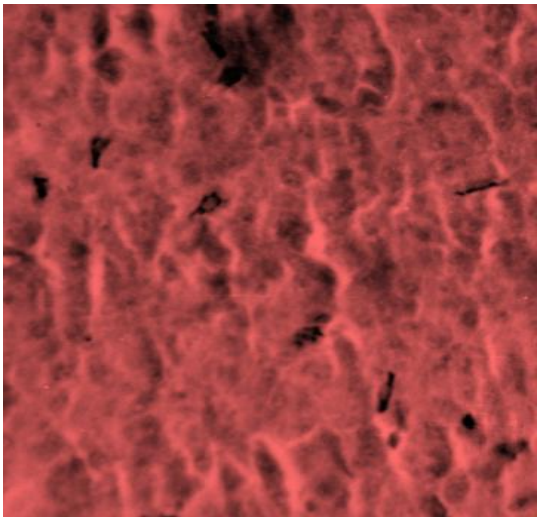

Figure 5: B. Micro photograph of T.S liver of *Channa gachua* after 45days exposure to 253.25mg/L of mustard oil cake (×400)

The findings of present study also match the earlier work who studied the impact of monocrotophos on the liver of *Labeo rohita* [15].

4. Conclusion

It may be concluded that mustard oil cake reaches the liver through the blood vascular system. The disorientation of cells and their functional incapability increases with an increase in exposure duration and toxic concentration. The necrosis, vacuolization and breakage of hepatic chord might be a reason of decreased metabolic activity, activeness of body which ultimately leads to the death of fish. Hence the indiscriminate use of biofertilizers not only harms the aquatic organisms but other organisms of higher trophic level too.

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