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Histopathological Impact of Dimethoate on the Liver of Freshwater Fish, *Garra mullya* (Sykes)

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ABSTRACT- The present investigation was carried out to the effect of Dimethoate on histopathological changes in liver of freshwater fish, *Garra mullya* using standard methods. Fish was exposed to sub lethal concentration of Dimethoate (0.0238ppm of 96hrs) for 7, 14, and 21days. Administration of pesticide to determine lesion of liver as indicators of tissue damage. Histopathological changes in liver ranged from vacuolization, necrosis, fibrosis of perivascular region and disposition of yellow brown grains at different time of exposure. Liver histology exhibited various abnormalities, including hyperplasia, nuclear pyknosis, fatty necrosis and degeneration of hepatocytes leading to tumor and syncytium formation, which are the indicative of carcinogenesis. In chronic treatment of dimethoate exposure may pose serious threat to fish health and affect their population.

Key-words- Dimethoate, Histopathology, Liver, *Garra mullya*

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INTRODUCTION

Pesticides are occasionally used indiscriminately in large amounts causing environmental pollution and potential health hazards. Dimethoate is systemic insecticides produced by reacting salts of Dimethyldithio-phosphoric acid with N-methylchloroaecetamide, in aqueous medium in the presence of some organic solvents is widely used against a broad range of insects and mites and is also used for indoor control of houseflies. The extensive use of DM poses a health hazard to animals and humans because of its persistence in soil and crops [1]. One of the major agricultural chemical groups is pesticide which play important role in increasing agricultural productivity through controlling pest. But on the other hand, they cause much damage to the non-target organisms both in terrestrial and aquatic environment. Fish accumulate pollutants directly from contaminated water and indirectly via food chain [2]. The runoff from treated areas enters the river and aquaculture ponds that are supplied by rivers and adversely affect the quality of water surfaces and creates hazards for aquatic life resulting in serious damage to non-target species, including fishes [3].

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Histopathology deals with the study of pathological changes induced in the microscopically structure of body tissue. Any alteration in normal structure of tissue indicates presence of disease or the effect of toxic substances like heavy metal and pesticides. [4] described histopathology as important tool for evaluating the action of any toxicant at tissue level. Histopathology provides data concerning tissue damage. Histopathological alterations can be used as indicators for the effect of various anthropogenic pollutants on organisms and are a reflection of overall health of the entire population in the ecosystem. The present study was under taken to analyze the impact of chronic concentration of dimethoate in liver of fish, *Garra mullya.*

MATERIALS AND METHODS

Healthy adult fish 4-5cm. in length *Garra mullya* were collected from local river Shivan Dist. Nandurbar, India. Fishes were washed with 0.1% of potassium permagnate (Km No 4) solution to avoid dermal infection. They were then rinsed in water and acclimatized to the laboratory conditions in the department of zoology for two weeks in 1000 *l.* capacity glass aquaria. Dead fish were removed immediately, Such as mortality may deplete dissolved oxygen with resultant effect on other fishes. During acclimatization fishes were fed with pieces of live earthworm on alternate days. Water also changed once in every day. The experiment was conducted natural and photoperiod of temperature 25·1±3·2^{0}C. Water quality was measure as per by [5], Conductivity 0.64±0.3, Dissolved O_2^- 6.3±1.1 (ml/L), pH 8.60±0.3, Acidity 2.5±0.1, Alkalini-

ty 44.1 ± 0.5, Total hardness 67.5 ± 0.3. LC50 of dimethoate for 96 hours was determined by probit analysis method [6]. The animals were dissected and liver tissue carefully removed. Tissues were immediately washed in 1% saline solution to remove the adherin mucus and blood and soaked between the blotting papers. The tissue from the control and exposed batches were taken out and preserved in aqueous Bouin's fluid for 24 to 48 hrs. This was followed by successive dehydration in ascending grades of alcohol. Tissues were cleared in xylene and embedded in paraffin wax (at 58˚- 60˚C). The tissue was then processed routinely and prepared into paraffin block cut at 6µm thickness using microtome and stained with Haematoxyline and Eosin [7]. Standard histopathological procedures were followed for histopathological investigations [8]. Observations were taken under light microscope.

RESULTS AND DISCUSSION

Histopathological changes have been widely used as biomarkers in the evaluation of the health of fish exposed to contaminants, both in the laboratory [9] and field studies [10]. In normal liver the hepatocytes form a cord-like pattern. These cords are arranged around tributaries of the hepatic vein. The liver cells are large in size, polygonal in shape with homogenous eosinophilic cytoplasm and centrally located nuclei. A large number of blood sinusoids are observed and separates the hepatic cords one from another. Exposure of dimethoate to *G. mullya* induced histopathological changes in the liver. The hepatocytes have lost their normal architecture. The lumen of sinusoid contains mainly erythrocytes and macrophages. Numerous hepatocytes show marked cytoplasmic vacuolization. The liver cells are degenerated with necrosis because of lymphocytic infiltration. The histopathological changes of liver were more pronounced after the exposure period of fenvalerate [11], similar observation by [12]. Fish exposed to sub lethal concentration of dimethoate during 7, 14 and 21 days shown considerable degree of alteration in the liver.

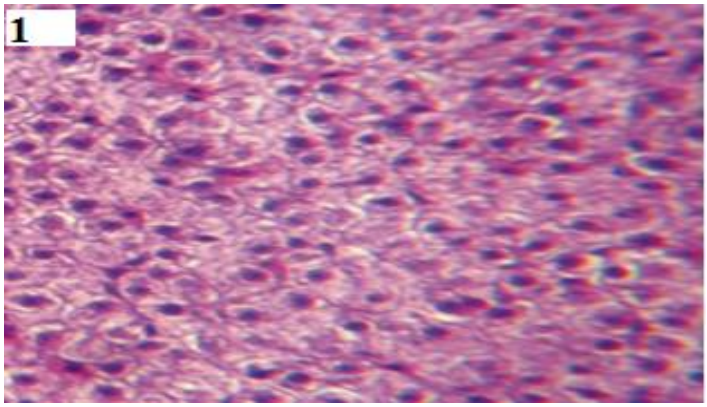

Fig. 1: Normal structure of liver showing blood sinosoids and hepatic cells, centrally placed nucleus containing nuclei (H&E, 450X)

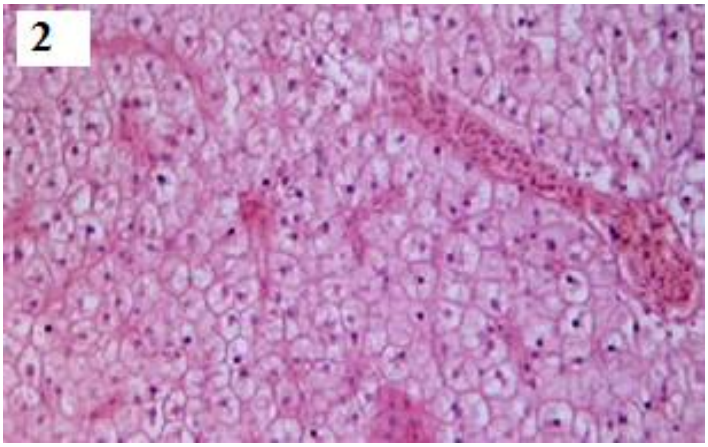

Fig. 2: On Dimethoate treatment after 7 days structure of liver shows hemorrhage at some places vacuolation in the hepatic cells (H&E, 450X)

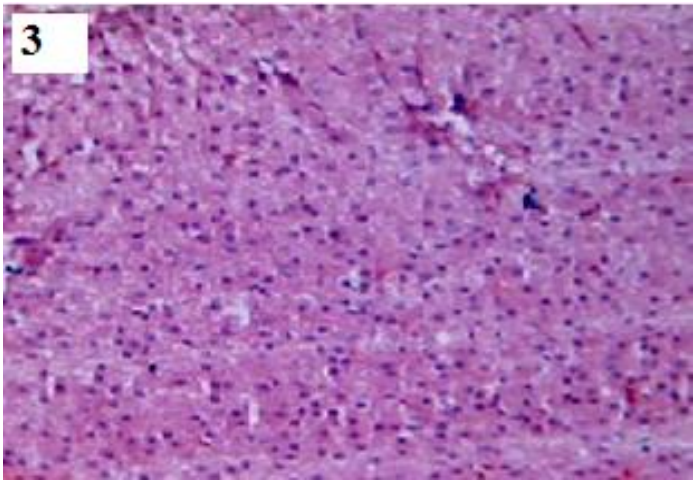

Fig. 3: On Dimethoate treatment after 14 days of liver shows haemorrhage at some places sinusoids are enlarged with vacuolation in the hepatic cells. Extensive degeneration of hepatocytes (H&E, 450X)

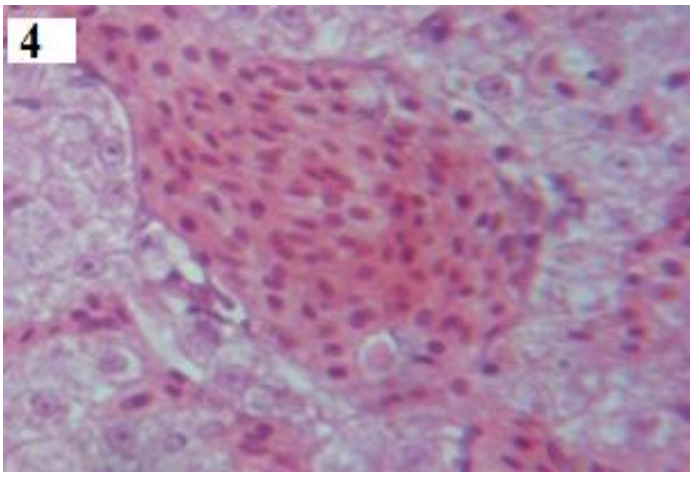

Fig. 4: On Dimethoate treatment after 21 days of liver shows extensive hemorrhage, necrotic hepatocytes, cloudy swelling and cytoplasmic vacuolization of hepatocytes (H&E, 450X)

Liver being the main organ of various key metabolic pathways, the effects of a chemical usually appear primarily on liver. This in turn, provides toxicologists, a significant site for investigation. In the experimental studies, liver histology exhibited various abnormalities, including hyperplasia, nuclear pyknosis, fatty necrosis and degeneration of hepatocytes leading to tumor and syncytium formation, which are the indicative of carcinogenesis.

The histopathological changes were more evident in specimens exposed to dimetoate and were not observed in the control fish. The liver cells in *G.mullya* are polygonal containing spherical central nucleus (Fig.1). After 7 days of exposure the hepatocytes became irregular and loose their polygonal shape. Some cells exhibited cloudy swelling, their contour becoming indistinguishable. There were many regions in the liver where cells were highly vacuolated. Many cells had exhibited pycnosis (Fig. 2 and 3). At the end of 21 days treatment, pronounced structural changes in hepatocytes such as focal necrosis, pycnosis, cloudy swelling and darkly stained specks of necrotic nuclei were observed (Fig. 4). The majority of insecticides are bio-transformed in metabolites by liver through various enzyme systems and as a consequence of this process, liver undergoes different levels of damages. Alteration like irregular shaped hepatocytes, cytoplasmic vacuolation and laterally placed nuclei were also observed in the siluriform *Corydoras paleatus* after exposure to organophosphate pesticide for 96 hrs observed by [13]. [14] also observed lipid vacuoles, hepatocytes swelling and pyknotic nuclei in *Oreochromic nilotitus* exposure to alachlor for 96 hrs. [15] also observed that dimethoate is strongly hepatotoxic and severely affect histology, carbohydrate and protein metabolism on liver of fish, *Cyprinus carpio*. [16] reported histopathological lesions induced in the hepatopancreas of *Channa punctatus* and *Clarius batrachus* exposed to industrial pollutants.

CONCLUSIONS

The present investigation shows After 7, 14 and 21 days of exposure of Dimethoate the hepatocytes became irregular, cloudy swelling and loose their polygonal shape. Liver cells were highly vacuolated, pycnosis and necrotic nuclei were observed in the histological structure of liver in *Garra mullya*. The changes in liver are biomarker in the evaluation of health of fish. The metabolic activities of the fish are affected which in turn become lethal to the fish. Dimethoate used to protect many field crops against disease, hence farmer come direct contact it and may affect their health.

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