

EFFECT OF DIMETHOATE ON SOME HISTOARCHITECTURE OF FRESHWATER FISH *OREOCROMIS MOSSAMBICUS* (PETERS, 1852)

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ABSTRACT

The presence of Insecticide in the environment, due to extensive use in agriculture and their low degradation capacity, are of potential toxicological concern for fish. Histological studies have been widely used as biomarkers in the evaluation of the health of fish exposed to the pesticides, both in the laboratory as well as in the field studies. In the present study Adult fish of nearly similar weight ($25 \pm 1.9\text{g}$) and length ($15.5 \pm 1.2\text{cm}$) were exposed to two sub lethal concentrations i.e. $45.0 \mu\text{g/L}$ and $22.5 \mu\text{g/L}$ of dimethoate for 21 days. The treated fish groups were compared with the control group for the histological changes in the selected tissues (Gills, Liver, Kidney, and muscle) and marked changes were observed.

INTRODUCTION

Ecotoxicology is the integration of toxicology and ecology or, as Chapman (2002) suggested, "ecology in the presence of toxicants". Ecotoxicology incorporates aspects of ecology, toxicology, physiology, molecular biology, analytical chemistry and many other disciplines. The evaluation of the ecotoxicological risks caused by pesticides to ecosystems is based on toxicity data and the effects of pesticide preparations on non-target organisms (Maltby and Naylor, 1990). For centuries pesticides have been used in agriculture to enhance the production of food by eradicating unwanted insects and controlling disease vectors (prakasam *et al.*, 2001). But the increasing chemization in all areas of man's activities has off-putting impact also besides its benefits. Though they have contributed considerably to agricultural outcomes, their adverse effects on non-target organisms are significant (John, 2007). The contamination of surface waters by pesticides used in agriculture is a problem of worldwide importance (Sibley and Kaushik, 1991).

Dimethoate (DM) is one of a class of insecticides referred to as organophosphates. DM is an insecticide used to kill mites and insects systemically and on contact. Dimethoate is moderately toxic by ingestion, inhalation and dermal absorption. As with all organophosphates, dimethoate is readily absorbed through the skin. (Secaucus, 1991)

Fish, among the group of non-target aquatic organisms, represent the largest and most diverse group of vertebrates. A number of characteristics make them excellent experimental models for toxicological research, especially for the

contaminants which are likely to exert their impact on aquatic systems (Leblond and Hontela, 1999; Lacroix and Hontela, 2001; Law, 2003 and Shiekh and Lee, 2008)

Histopathological changes have been widely used as biomarkers in the evaluation of the health of fish exposed to contaminants, both in the laboratory (Wester and Canton, 1991; Thophon *et al.*, 2003) and field studies (Hinton *et al.*, 1993; Schwaiger *et al.*, 1997; Teh *et al.*, 1997). One of the great advantages of using histopathological biomarkers in environmental monitoring is that this category of biomarkers allows examining specific target organs, including gills, kidney and liver, that are responsible for vital functions, such as respiration, excretion and accumulation and biotransformation of xenobiotics in the fish (Gernhofer *et al.*, 2001), and serve as warning signs of damage to animal health (Hinton and Lauren, 1993).

Hence, the present study is aimed to look in to the histoarchitectural alterations in DM induced toxicity in some of the vital organs of the teleosts fish *Oreochromis mossambicus* (Peters, 1852), so as to assess the damage and get an insight in its functional consequences.

MATERIALS AND METHODS

The fresh water fish *O. mossambicus* were collected from Sama pond of Baroda city ($22^{\circ}22' 39.03''$ N to $22^{\circ}20' 25.98''$ N and $73^{\circ}12' 99''$ E to $73^{\circ}12' 69''$ E). The fish were stored in a glass aquaria containing 50 L dechlorinated tap water for 10 days for acclimation under laboratory conditions. Water was

changed after every 24 hrs. Commercial fish food was supplied to fish during acclimation period. Adult fish weighing (25 ± 1.9 g) and length (15.5 ± 1.2 cm) were selected for experiments. Acclimated fishes were treated with DMEC 30%. The LC analysis of DM was performed. The fish were treated with two sub lethal concentrations *i.e.* $45.0 \mu\text{g/L}$ and $22.5 \mu\text{g/L}$ for 21 days. Six aquaria were set, two for each concentration and each aquarium containing 10 fish in 10 L dechlorinated tap water. Water temperature was kept at $28 \pm 1.5^\circ\text{C}$

during whole experimental period. Control group were kept in dechlorinated water without any treatment. Fishes from both the group were monitored for their Behavioral changes also. After completion of 21 days, Fish were removed from aquaria and washed with fresh water. Fishes of both treated as well as control groups were killed by decapitation. After decapitation, the blood was allowed to drain and the fishes were dissected open to take out Gills, Muscle, liver and kidney. Fresh tissues were fixed in 4% paraformaldehyde for 24 hrs, dehy-

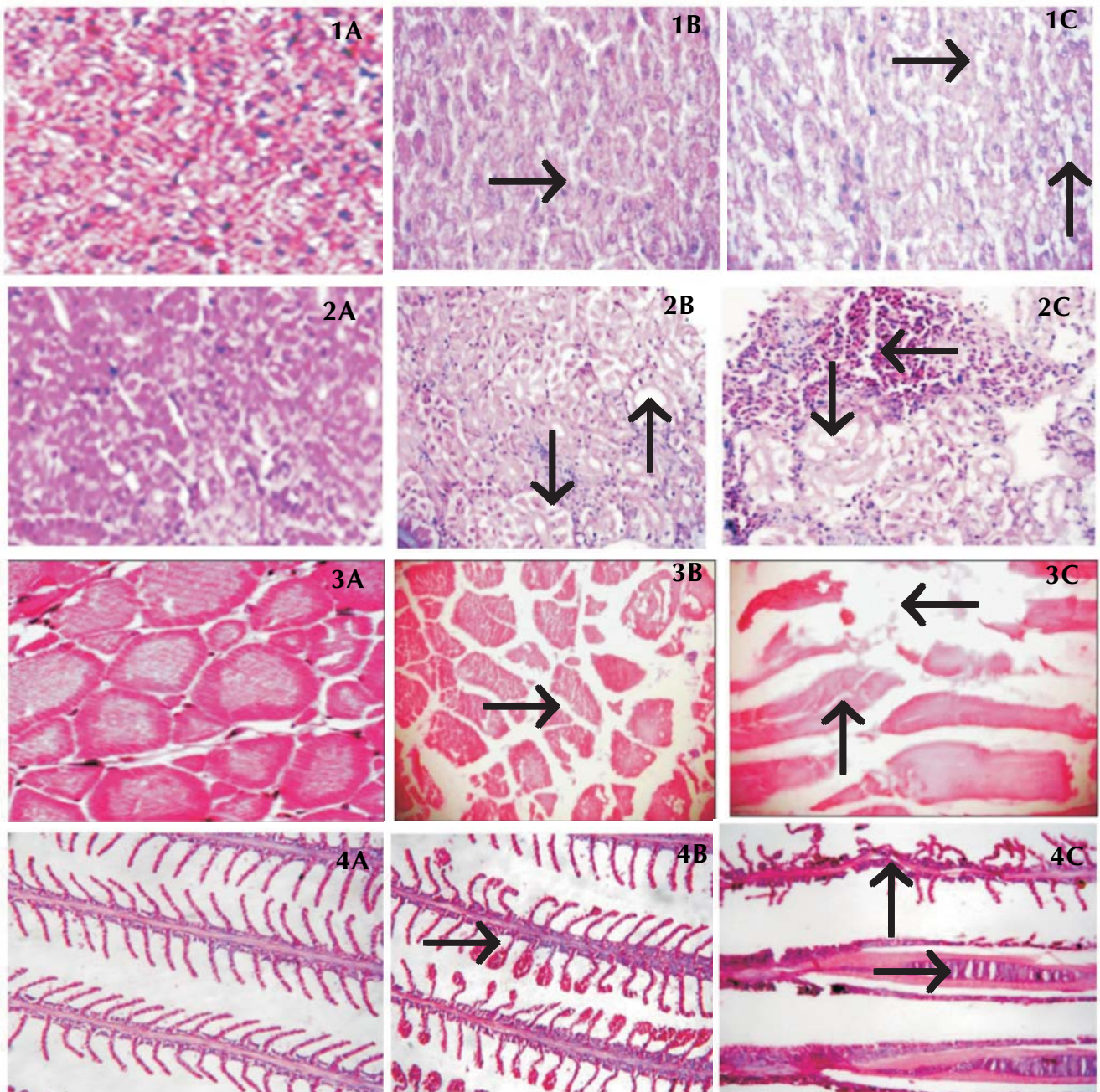


Figure 1, 2, 3 and 4: The normal structure of liver, kidney, muscle and gills shown in the Figure 1A, 2A, 3A and 4A respectively. Marked swelling, Vascular degeneration of hepatocytes (↑) (Fig. 1B) focal necrosis and fibrosis (↑) (Fig.1C); Glomeruli enlargement, edema in bowman's capsules (↑) (Fig. 2B), vacuolar degeneration with aggregate inflammatory cells (↑) (Fig.2C); Degeneration, necrosis and atrophy of muscle bundles (↑) (Fig. 3B) splitting of muscle fibers (↑) (Fig. 3C); Curling and clubbing of secondary lamellae, distortion and of primary lamellae (↑) (Fig. 4B) and enlargement of primary lamellae and loss of secondary lamellae (↑) (Fig. 4C). {HE 400x}

drated, embedded in paraffin wax and sectioned at 10-12 μ m then stained with heamatoxylin and eosin and examined microscopically and photographed using digital camera (400x).

RESULTS AND DISCUSSION

The treated group showed an insignificant reduction in the body weight. Abnormal behavior such as restlessness, sudden quick and jerky movements, were observed in the fishes exposed to low dose, whereas, increased opercular movements accompanied with surface to bottom movements and loss of equilibrium was observed in the fishes exposed to high dose. Similar observations have been reported by Hinton *et al.*, (1993) and Raheman *et al.*, (2002) with various organophosphate insecticides. The histological changes observed in all the tissues of *O. mossambicus* in the present study indicate that sub lethal concentrations caused moderate to severe alteration in Gill, liver, kidney as well as muscle architecture, which are an important organs performing vital function like detoxification, respiration, osmoregulation, acid-base balance, excretion etc. The organ most associated with the detoxification and biomarker process is liver and due to its function, position and blood supply, it is also one of the organs most affected by contaminants in the water (Camargo and Martinez, 2007). The liver of the fish exposed to both low as well as high dose showed vacuolar degeneration, swelling in the hepatocytes with indistinguishable cellular outline. (Fig. 1 A, B, C) These changes may be attributed to direct toxic effects of pollutants on hepatocytes, since the liver is the site of detoxification of all type of toxins and chemicals. It seems that there is a temporal sequence of the events that starts with vacuolization, swelling and necrosis. Rodrigues and Fanta (1998); Camargo and Martinez, (2007); and Mohamed, (2009) have also reported parallel observations with pesticides in various fishes.

The kidney is a vital organ of body and proper kidney function is to maintain the homeostasis. It is not only involved in removal of wastes from blood but it is also responsible for sensitive reabsorption, which helps in maintaining volume and pH of blood and body fluids and erythropoiesis (Iqbal, *et al.*, 2004). The alterations found in the kidney of fish at low dose were glomeruli enlargement and edema in bowman's capsules; at high dose the kidney exhibited vacuolar degeneration accompanied with hemolysis (Fig. 2, A, B, C). With severe intoxicated conditions, the degenerative process leads to tissue necrosis (Yokote, 1982). The necrosis of the tubules will affect the metabolic activities and promotes metabolic abnormalities in fish. The present results are in agreement with those observed in *C. carpio* exposed to sewage (Kakuta and Murachi, (1997), Veiga *et al.*, (2002) and Thophon *et al.*, (2003).

Separation and degeneration of muscles, atrophy of muscle bundles and focal area necrosis were an interesting observation in muscle tissue at low dose and it was leading to Vacuolar degeneration and splitting of muscle fiber were seen as high dose (Fig. 3 A, B, C). The histopathological alterations in the fish muscle of both the doses are in agreement with those observation by many investigators who have studied the effect of different pollutants on fish muscle (Sakr and Gabr, 1991; Nour and Amer, 1995 and Elnemaki and Abuzinadah, 2003).

The gills, which participate in many important functions in the fish, such as respiration, osmoregulation and excretion, remain in close contact with external environment and particularly sensitive to changes in the quality of the water are considered the primary target on contaminants (Camargo and Martinez, 2007; Fernandes and Mazon, 2003). The gills of fish exposed to low dose showed Curling and Clubbing of secondary lamellae (telangiectasis) Enlargement of primary lamellae, loss of secondary lamellae was seen at high dose. (Fig. 4, A, B, C) These pathological changes may be a reaction to toxicants intake or an adaptive response to prevent the entry of the pollutants thorough the gill surface and probably due to increased capillary permeability (Olurin *et al.*, 2006). The present results are in agreement with those observed in other fish species under the influence of different pollutants (Kakuta and Murachi, 1997; Olurin *et al.*, 2006).

CONCLUSIONS

Finally, the present study proves the toxic potential of the DM pesticide and shows moderate to severe alterations in gills, muscles, liver and kidney which can lead to metabolic changes in the fish. Furthermore, the present study also adds to the concepts that Histopathological studies are one of the effective tools for ecotoxicology and risk assessments.

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