

Histopathological Study of Liver And Kidney of The Fish *Ctenopharyngodonidella* Exposed To The Deltamethrin 11% EC, A Synthetic Pyrethroid

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Abstract: Histopathological study of the liver and kidney of the fish *Ctenopharyngodonidella* exposed to Deltamethrin 11% EC for 10 days was studied ($1/10^{\text{th}}$ of 96 h LC_{50} value 0.0172). Due to toxic effect on the 96 h selected organs, alterations are observed as pathological biomarkers indicating the load of pollution of toxic stress. Degeneration of hepatocytes and hepatic cords are observed in the liver of toxicant exposed fish. Similarly in the kidney severe necrosis, cloudy swelling of cytoplasm and a decrease in the epithelial cells of the distal convoluted tubules are also observed. Pathological lesions in liver and kidney culminate the impairment of physiological process of the fish.

Keywords: *Ctenopharyngodonidella*, Liver, Kidney, Deltamethrin 11% EC, Biomarker, Histopathological study.

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I. Introduction

The significance of aquatic environment, the largest subdivision of the planet earth has increased, due to conscious nourishment trends, biological values of aquatic products because of high protein qualities. Water, Water everywhere but is there any water that is not pure, may be impure in the scientific sense that is contaminated. How to detect the situation of that sort that is what in the science of Ecotoxicology which encompasses certain evidences as biomarkers, the biochemical, histopathological, genotoxic and molecular changes.

So contamination by pesticides is viewed as there are four different classes of compounds organochlorines, organophosphates, carbamates and synthetic pyrethroids. Such compounds as the xenobiotic and in fact anthropogenic in an ambition to increase the produce either from agricultural or from aquacultural sources. Being present, in such situation the diversity of organisms are harmed. In doing, research of diverse fauna especially the fish will play a role for determining the effect of chemical substance of pollutant, which is also diverse on aquatic ecosystems (Figen Esin Kayhan et al., 2013).

One such study is histopathology, a pathological condition of different organs, which is made of tissues of course they are made of cells. Marie Francois Xavier Bichat a French anatomist and pathologist and is known as father of histology, a pathobiologist who invented the technique that deals with the study of any disease or abnormalities in the tissues. The pesticides cause certain such changes which as when studied in fishes, will be a tool that guide us by ecotoxicologist to assess the defilement of aquatic environment (Yanchava et al., 2015; Reddy and Kusum, 2013).

Such histopathological studies are mentioned for pesticides in their review articles by Ullah and Jallil (2015); Kaushik Mondal et al., (2015); Shankarmurthy et al., (2013); Prusty et al., (2015); Ahrar Khan et al., 2014, Velisek and Stara (2014) and Velisek et al., (2006, 2007 & 2009) for synthetic pyrethroids to which the present study of the toxicant belongs. Hasibur Rehman et al., (2014) and Bhattarcharjee and Das, (2014) in their reviews on Deltamethrin of the present study toxicant in the above aspect made clear that the studies of fish, organ/tissues i.e., histopathology will be due to toxic action by the toxicant even the concentrations are in sublethal level that are present in waters, inhabited by them.

Velisek and Stara (2014) reported that deltamethrin did not cause histopathological changes in gills, skin, liver, spleen and kidney of rainbow trout and common carp in lethal concentrations. But Srivastava (1997), Cengiz (2006), Cengiz and Unlu (2002, 2003 & 2006) and Staien et al., (2007) reported changes in common carp, mosquito fish *Gambusia affinis* and Nile tilapia *Oreochromis niloticus* respectively. Sayed et al., (2007) too reported on *Carassius auratus gibelio*. But according to Yildirim et al., (2005) who also reported histopathological studies in Nile tilapia (*Oreochromis niloticus* L.) fingerlings in gills, liver, brain and spleen. However, overall paucity of information on the present study of fish and as different opinions are available on

Deltamethrins mentioned in the above as well as in the review articles, the present study is undertaken to study the pathological lesions in the liver and kidney of the fish *Ctenopharyngodonidella*, the exotic carp commonly cultured along with other major carps (may be first time of the toxicant on the grass carp – *Ctenopharyngodonidella*).

II. Materials And Methods

Fresh water fish *Ctenopharyngodonidella* was acclimatized to laboratory conditions for 10 days. 50 Fish are exposed to 11% EC Deltamethrin for 10 days $1/10^{\text{th}}$ of the 96h LC_{50} 0.0172 continuous flow through system (CFTS) value and i.e., for 10 days as per APHA guidelines (1998, 2005, 2012). The toxicant deltamethrin 11% EC is purchased locally, manufactured by GIDC, Industrial Estate Ltd., 629/630, Gujarat, marketed by Sikko Industries Ltd., Ahmedabad. At the end of the exposure, period of 10 days the fish are randomly selected for histopathological examinations and liver and kidney, tissues are isolated and also from fish not exposed to the toxicant which serve as control.

Physiological saline solutions 0.85% sodium chloride (NaCl) was used to rinse and clean. They were fixed in aqueous Bouin's solution for 48 hr processed through graded series of employing alcohols, cleared xylene and embedded in paraffin wax. Sections of 6 μm thickness were cut stained with Ehrlich-hematoxylin/Eosin dissolved in 70% alcohol and mounted on Canada balsam (Humason, 1972). Sections were observed and photographed with the help of Intel Pentium Q x 3 Computer attached microscope, under 400x lens (made in China).

III. Observations And Discussion

General Histology of Liver (Plate 1 FIG:A)

The surface of the liver is covered with serous membrane and some connective tissue which extends inwards into hepatic cords with hepatic cells and lattice fibers and sinusoids. Hepatic cells are round or polygonal containing clear spherical cells modified cuboid cells rested on cosmically highly secretory on basement membrane. It is the highest secretory gland. Bile canaliculi are centrally located in each hepatic cord and generally obscure. Fairly large quantities of lipid glycogen granules (glycolipids) are located in each cord. The hepatic cell cytoplasm have fairly large quantities in the cytoplasm of the fish liver.

Secretion and storage of bile, rich in vitamin A and K which play an important role in protein and carbohydrate metabolisms and its detoxification, among it the benzopyrin (Lagler, 1962).

Pathology of Liver (Plate 1 FIG:B)

Degeneration and necrosis are prominent. The degeneration of hepatocytes at the polygonal epithelium, atrophy which is prominent, cytoplasm is with vacuoles supposed to have exocytosis, ruptured blood vessels and disposition of hepatic cords. Liver is an organ of portal circulation and any foreign molecule, the antigen can damage the epithelial lining. The toxicity induced the membrane of the largest gland of the body and its detoxification role is very prominent. Due to damage of these metabolism and immunity is effected (Prosser, 1950).

General Histology of Fish Kidney (Plate 2 FIG:A)

Teleostean kidney consists of head and body. Head is the anterior portion of the kidney and consists of lymphoid tissue. Body kidney is composed of many nephrons and interstitial lymphoid tissue. The interstitial tissue is the major hematopoietic tissue in the body. Each nephron consists of two parts, the glomerules and the urinary tubule. The glomerules capsule consists of an inner and outer layer of single flattened epithelia. Renal tubules consist of single layer of epithelial cells. Mesangium fills the space between the loops of glomerular capillaries.

Renal tubules are thin and short in the neck segment. The proximal convoluted segment is divided into two parts i.e., segment I and segment II. The renal tubules are composed of cuboidal epithelial cells with densely arranged microvilli in the tubular lumen. In segment II, renal tubules are composed of cuboidal epithelial cells. Cilia and microvilli are found in the tubular lumen. In the distal convoluted segment, epithelial cells have no microvilli. The cells of this segment are stained with eosin more faintly than those of proximal convoluted segment (Fig. A). Thus, it is easy to distinguish between proximal and distal convoluted segments under light microscopy

Pathology of Kidney tissue under observation (Plate 2 FIG:B)

Highly degenerative changes were observed in haemopoietic tissue which includes severe necrosis, cloudy swelling cytoplasm. The epithelial cells of the distal convoluted tubule decreased in size. The interstitial renal tissue was less affected. Renal interstitial tissue showed formation of vacuoles and cellular contours were not clearly distinguished (Plate 2B).

The waste products collected from the body are eliminated through kidney. The non-detoxified pesticide molecules must be eliminated through the kidney only of the fish and hence, it is susceptible to different chemical substances compounds when exposed to sublethal doses of the toxicant.

PLATE I

Fig. A

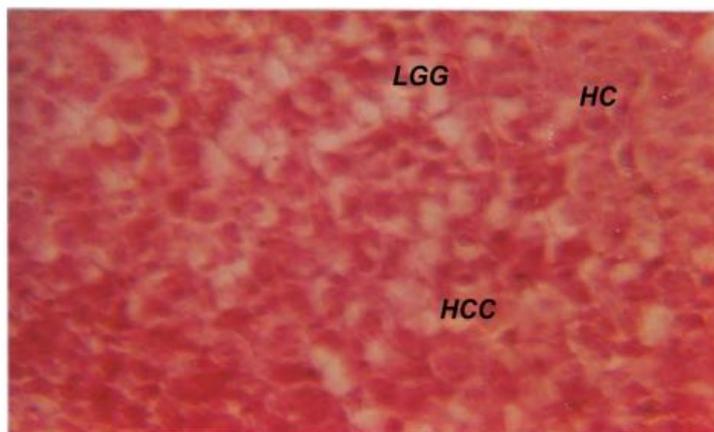


Fig. B

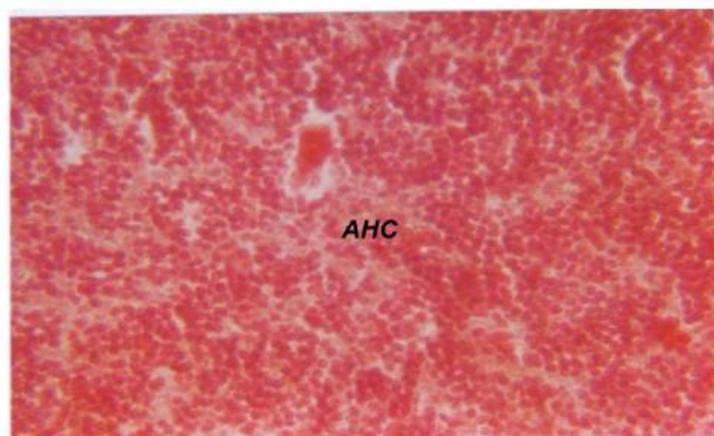


Plate 1: Fig. A. Normal Structure of Liver in Ctenopharyngodonidella Haematoxylin/Eosin Stain (HE), x400 HC: Hepatic Cells; HCC: Hepatic Cells Cords; LGG: Lipid and Glycogen Granules.

Fig. B. Liver of Ctenopharyngodonidella exposed for 10 days to sublethal concentration of Deltamethrin 11% EC Haematoxylin/Eosin Stain (HE), x400 AHC: Atrophy in Hepatic Cells.

PLATE 2

Fig. A

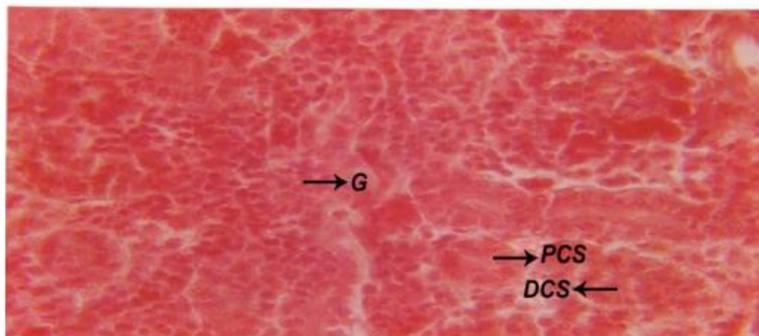


Fig. B

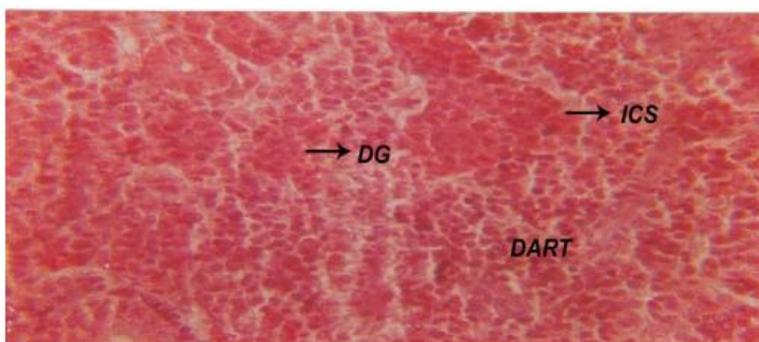


Fig. A. Normal Kidney Structure in *Ctenopharyngodonidella* Haematoxylin/Eosin Stain (HE); x400
PCS: Proximal Convoluted Segment; DCS: Distal Convoluted Segment; G: Glomerulus.

Fig. B. Kidney of *Ctenopharyngodonidella* exposed for 10 days to sublethal concentration of Deltamethrin 11% EC Haematoxylin/Eosin Stain (HE), x400 DART: Degeneration and Atrophy in Renal Tubules; DG: Degeneration in Glomerulus; ICS: Intercellular Spaces formation giving mesh like appearance.

Suvetha *et al.*, (2015) reported on the intoxication of deltamethrin in an Indian major carp *Labeo rohita* due to hormonal and enzymatic homeostatic responses. They reported significant alterations in the enzymes of both liver and kidney of the fish. These biochemical changes will be due to the tissue damages of the respective organs of the fish, one is metabolically active whereas the other one is for excretion.

Cengiz (2006) reported kidney histopathology in the freshwater fish *Cyprinus carpio* after acute exposure to deltamethrin. The study revealed appearance of lesions in the kidney tissues of the fish and is characterized by degeneration of the epithelial cells of renal tubule, pyconotic nuclei in the hematopoietic tissue, dilatation of glomerular capillaries and degeneration of glomerulus, intracytoplasmic vacuoles in the epithelial cells of renal tubule with hypertrophied cells and narrowing of the tubular lumen. The freshwater fish needs to maintain osmoregulation and will be definitely a problem if kidney is effected as in the reported fish and also in the present studied fish.

Veliseket *et al.*, (2007) reported the effects of deltamethrin on rainbow trout *Onchorynchus mykiss*. Being strongly toxic to fish the fish showed alterations both biochemically and haematologically due to pathological conditions of the damage of the largest gland of the body and also ammonotelic excretory organ.

Yeldrimet *et al.*, (2005) reported on histopathological effects of deltamethrin on tissues (gills, liver, brain, spleen, kidney, muscle and skin) of Nile tilapia (*Oreochromis niloticus* L.) fingerlings. Hydrophobic degenerations in liver were reported. The fish not only have alterations but also exhibited certain behavioural changes too due to severity of the organs damage.

Cengiz and Unlu (2006) reported the sublethal effects of commercial deltamethrin on the structure of the gill, liver and gut tissues of mosquito fish, *Gambusia affinis*. The significant pathological alterations reported in liver are, hypertrophy of hepatocytes, significant increase of Kupffer cells, circulatory

disturbances focal necrosis fatty degeneration, nuclear pycnosis and narrowing of sinusoids. Some of these change are also observed in the present study of the fish.

Dobsikova *et al.*, (2006) reported histopathological parameters of common carp *Cyprinus carpio* after exposure to Cypermethrin the other one of type II synthetic pyrethroid. The study revealed vacuolization of pancreas particularly the exocrine cells. The importance of the pancreas associated with liver which play a significant role in digestion.

Velumurugan (2009 & 2007a) reported histopathological changes in the liver of the fresh water fish *Cirrhinus mrigala* exposed to Dichlorovos an organophosphate. Hepatic lesions in the liver tissues of fishes exposed to dichlorovos were characterized by cloudy swelling hepatocytes congestion vacuolar degeneration karyolysis karyotaxial dilation of sinusoids and nuclear hypertrophy.

Veliseket *et al.*, (2009) reported the histopathological effects of bifenthrin, a synthetic pyrethroid in the rainbow trout (*Ochrochinchus mykiss*). Degeneration of liver hepatocytes were reported as a histological biomarker.

Velumurugan (2007b) reported the effects of fenvalerate which is also type II synthetic pyrethroid on different tissues of fresh water fish *Cirrhinus mrigala*. The study on liver and kidney of the fish showed significant changes such as necrosis of tubular epithelium pycnotic nuclei in the hematopoietic tissue hypertrophied epithelial cells of renal tubules, narrowing of the tubular lumen expansion of space inside the Bowman's capsule and contractions of the glomerulus were observed in the kidney tissues of the fish. Hepatic lesions in the liver of the fish exposed to the toxicant which were characterized by congestion and cloudy swelling of hepatocytes and focal necrosis.

Veliseket *et al.*, (2006) reported the effects of Cypermethrin on rainbow trout (*Oncorhynchus mykiss*). Being highly toxic to fish, the liver showed severe pathological condition resulting significant changes in the enzymes of the respective organs.

Neelima *et al.*, (2015) reported histopathological alterations in the gill, liver and kidney of *Cyprinus carpio* exposed to cypermethrin (25% EC). Hepatocytes damage and glomerulus damage respectively in liver and kidney are reported.

Hasan *et al.*, (2014) reported histomorphometric profile of grass carp *Ctenopharyngodonidella* during acute Endosulfan toxicity. The study reported degenerative changes including vacuolization pyknosis etc.

Andemet *et al.*, (2017) reported histopathological changes of synthetic pyrethroid pesticide cypermethrin exposed to African clariid mud catfish (*Clarias gariepinus*) fingerlings. The term hyperplastic hepatocytes and necrosis is reported.

Mohammed *et al.*, (2016) too reported the subchronic toxicity of Deltamethrin and tetramethrin in the Nile tilapia fish. The report mentioned about the necrosis of spleen.

Thus not only synthetic pyrethroids any pesticides preclude damage of the important organs and using histopathology of fish as a protocol serve as a biomarker in the assessment of aquatic pollution. The liver is known for the metabolic process such as glycogenesis, glyconeogenesis and glycolysis. If they are impaired the growth of the fish especially in aquaculture the venture is at loss. Similarly the kidney, the primary organ of excretion ammonotelic in nature is disturbed if the organ is pathologically damaged.

IV. Conclusion

The xenobiotic compounds affect the fish organs like liver and kidney and the susceptibility of the fish in the context of environmental monitoring and the biomarker approach. Metabolically active organs the liver and kidney when damaged the very survival of the fish, the non-target organisms is questionable. Due emphasis must be on sublethal concentrations and formulations have to be viewed seriously.

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