

Effect of Cartap hydrochloride (50% SP) insecticide on gill histology of the fish, *Cirrhinus mrigala* (Hamilton)

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ABSTRACT

The fish, *Cirrhinus mrigala*, Indian major carp was exposed to Cartap hydrochloride and the static LC₅₀ values for 24, 48, 72 and 96 hours were found to be 0.436, 0.419, 0.394 and 0.376 mg⁻¹ and 0.399, 0.371, 0.361 and 0.339 mg⁻¹ in Continuous flow-through system. The gills which are the primary organs to get exposed to foreign contaminants and pesticides were studied for histological changes. The following histological changes like epithelial lifting, degeneration of primary and secondary gill lamellae, curling of secondary gill filaments, atrophy of secondary gill lamellae, congestion of secondary lamellae, fusion of secondary gill filaments were observed. The results obtained were discussed at length with the available literature. By studying the histological deformities, it was concluded that the pesticide caused enough damage to the gills of the fish and can influence on the survival of the fish

Key words : Cartap hydrochloride, *Cirrhinus mrigala*, Gill histopathology

Introduction

Indiscriminate and extensive use of pesticides in modern agricultural practices globally for achieving increased food production is one of the major sources of water pollution. Presence of pesticide in aquatic bodies is largely due to outfall from pesticide manufacturing factories and the runoff from agricultural fields (Ganeshwade, 2012). Pesticides are not highly selective but are generally toxic to macrophytes, non-target organisms such as fish (Ayoola, 2008 and Franklin *et al.*, 2010).

Fish accumulate these pollutants directly or indirectly from polluted waters and food chain (Jabeen *et al.*, 2016). In India, Cartap hydrochloride, a carbamate pesticide is extensively used in rice, sugar-cane cabbage and cauliflower crops to control pests.

In fish, the gill is the major organ for respiration, excretion and osmotic regulation. Gills are appropriate for the assessment of environmental impact as they are considered as a good tissue indicator of the water quality (Fanta, 2003). Histopathological analysis is a very sensitive parameter and is helpful in determining cellular damage that may occur in target organs (Altinok and Capkin, 2007). In this viewpoint an attempt was made to study the effect of sublethal and lethal concentrations of Cartap hydrochloride on gill histopathology of freshwater fish, *Cirrhinus mrigala* exposed for 24 and 96 hours.

Materials and Methods

The fingerlings of the test fish *Cirrhinus mrigala* of size 6-8 ± ½ cm and weight 6-7 ± ½ gm were pro-

cured from local fish hatcheries of Nandivelugu, Tenali mandal, Guntur district, Andhra Pradesh. The fish were acclimated at ($28 \pm 2^\circ\text{C}$) in the laboratory conditions for two weeks. All the precautions laid down on recommendations of the toxicity tests to aquatic organisms were followed (Annon, 1975). Fish were regularly fed with rice bran and one day prior to the experimentation feeding was stopped. Fingerlings were exposed to sub-lethal ($1/10^{\text{th}}$ of 96 h LC_{50} value 0.0376 mg^{-1}) and lethal (96 hours LC_{50} value 0.376 mg^{-1}) concentrations of Cartap hydrochloride for 24 and 96 hrs.

Gills were processed in laboratory for routine histological characterization by the double staining method using Haematoxyline and Eosin. All samples were fixed in 10% phosphate- buffered formalin for about 24 hrs. The specimens were dehydrated in a series of graded ethanol (50%, 70% and 90%) and then after embedded in paraffin. Five-micron sections were cut using an ultramicrotome (Leica, Japan) and deparaffinized by means of xylol. The sections were dehydrated in 90%, 70% and 50% ethanol followed by a 10 min wash in water and further stained with Hematoxylin and Eosin (HE). Sections were observed in digital microscope (Intel Play QX3) at $400 \times$ magnification.

Results

The transverse section of gill tissue of normal fish shows branches from the central axis called the primary gill lamellae. Each of the primary gill lamella further divides into secondary gill lamellae or filaments. Within each division of the gills are the adjacent afferent vessels and efferent vessels with hemocytes. A thin septum separates the primary and secondary gill filaments. The secondary non branching filament lamella possesses epithelial pillar cells separated by large lacunae Fig. 1.

Cartap hydrochloride has induced marked pathological changes in gills of fish *Cirrhinus mrigala*

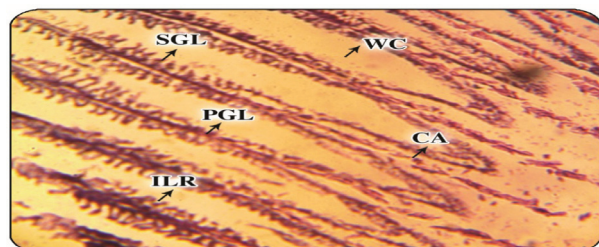


Fig. 1. Normal Gill tissue of *Cirrhinus mrigala*, H.E x 400

mrigala. In the current study, it was observed that with the increase in the time of exposure the histopathological alterations increased in the gills of fish, *Cirrhinus mrigala*.

CA-Central Axis, PGL-Primary gill lamellae, SGL-Secondary gill lamellae, ILR-Inter lamellar space, WC-Water channel, FSG-Fusion of secondary lamellae, CSG-Curling of secondary lamellae, DPGL-Degenerated primary gill lamellae, DSGL-Degenerated secondary gill lamellae, BC-Blood congestion, EL-Epithelial lifting, LF-Lamellar fusion, ASL-Atrophy of secondary lamellae.

In the control group, no pathological changes were observed in the gills (Fig.1). After treatment with cartap hydrochloride for 24hrs at sublethal concentration epithelial lifting, curling of secondary gill filaments, fusion of secondary lamellae and degeneration of secondary gill lamellae started to appear (Fig. 2). Degenerative changes like lamellar fusion, epithelial lifting, degenerative secondary gill lamellae and curling of secondary gill filaments were observed after exposure to lethal concentration for 24hrs (Fig. 3). Similar more pronounced degenerative changes like curling of secondary gill filaments, epithelial lifting, degenerative primary and secondary gill lamellae, blood congestion, atrophy of secondary lamellae, mucous secretion were no-

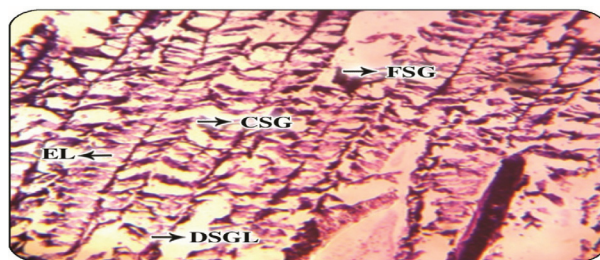


Fig. 2. Gill tissue of *Cirrhinus mrigala* exposed to sublethal concentration of Cartap hydrochloride for 24 hours, H.E x 400

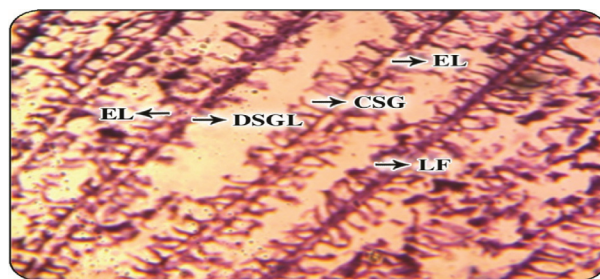


Fig. 3. Gill tissue of *Cirrhinus mrigala* exposed to lethal concentration of Cartap hydrochloride for 24 hours, H.E x 400

ticed in gill exposed to sublethal concentration of cartap hydrochloride for 96 hrs (Fig. 4). Damage in the gills exposed to lethal concentration of cartap hydrochloride for 96 hrs was much extensive with severe degeneration in primary and secondary gill lamellae, hemorrhage between gill filaments, separation of epithelial cells from the basement membrane, collapsed pillar cell, fusion of secondary gill lamellae, lamellar disorganization, atrophy of secondary lamellae, necrosis, blood congestion, mucous secretion, hyperplasia and epithelial lifting (Fig. 5). Thus, the lethal concentration of Cartap hydrochloride has produced significant histopathological changes in comparison to sublethal concentration in *Cirrhinus mrigala*.

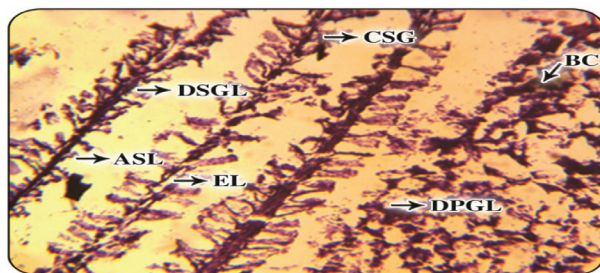


Fig. 4. Gill tissue of *Cirrhinus mrigala* exposed to sublethal concentration of cartap hydrochloride for 96 hours, H.E x 400

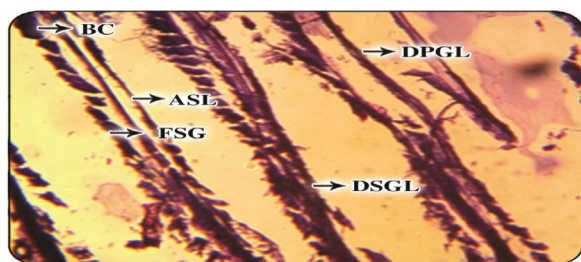


Fig. 5. Gill tissue of *Cirrhinus mrigala* exposed to lethal concentration of cartap hydrochloride for 96 hours, H.E x 400

Discussion

Several authors observed similar degenerative changes in gill when exposed to carbamate pesticides. Vivek *et al.*, (2016) observed fusion of primary and secondary lamellae, epithelial hyperplasia, curling of secondary lamellae, degeneration of lamellae in gills of *Labeo rohita* exposed to sublethal concentrations of cartap hydrochloride for 24,48,72 and 96hrs. Mariya dasu (2014) observed bulging of tips of primary gill filaments, curling of secondary gill filaments, necrosis, fusion of secondary gill lamel-

lae, hyperplasia, hypertrophy of nuclei, pyknotic nuclei, lifting of epithelium in *Labeo rohita* exposed to Thiocarb, a thiocarbamate pesticide in both sublethal and lethal concentrations. Anitha (2015) observed necrosis, curling and fusion of secondary gill filaments, atrophy, degeneration of primary and secondary gill lamella, blood conjunction, epithelial hyperplasia, lamellar telangiectasis, in gills of *Labeo rohita* exposed to sublethal concentrations of Pyraclostrobin for 24 hrs, 5 days and 10 days and to lethal concentration for 24 hrs. Ali Taheri Mirghaed *et al.*, (2018) noticed edema, lamellar curling, hyperplasia, lamellar fusion in the gill of *Cyprinus carpio* treated with 0.75 mg/L, 1.5 mg/L, and 3 mg/L of indoxacarb for 7, 14 and 21 days. The histopathological changes observed in the present study were in accordance with the above studies.

Similar histopathological changes in gills were reported in *Channa punctatus* exposed to Imidacloprid (Pawara *et al.*, 2019), in *Labeo rohita* exposed to phenol (Butchiram *et al.*, 2013), in *Labeo rohita* exposed to cadmium (Saravanan *et al.*, 2019), in *Oreochromis niloticus* exposed to heavy metals (Shameem Rani, 2018), in *Labeo rohita* exposed to tannery industrial effluent (Diana Handa and Gurinder Kaur Walia, 2019), in *Puntius sophore* treated with Acephate (Gavit and Patil, 2019), in *Cirrhinus mrigala* exposed to cement factory effluent (Juginu and Sujila, 2019) in *Cirrhinus mrigala* exposed to lead arsenate (Vanitha *et al.*, 2017), in *Labeo rohita* exposed to dimethoate (Dey and Saha, 2014), In *Cirrhinus mrigala* exposed to cypermethrin (Sree Veni and Veeraiah, 2014).

In fish, gill is the major organ for respiration, excretion and osmotic regulation. Gills are appropriate for the assessment of environmental impact as they are considered as a good tissue indicator of the water quality (Fanta, 2003). The lifting of lamellar epithelium in the present study may be induced by the incidence of severe Oedema (Pane *et al.*, 2004). Swelling in gill epithelium leads to decreased efficiency of gases exchange and oxygen consumption. (Sudhasaravanan and Binukumari, 2015).

The first line of defense to metal/ toxicant exposure in the gills is mucus secretion and by fusion of lamellae it can temporarily protect the underlying epithelium from injury (Handy and Maunder, 2009). Pronounced secretion of mucus layer over the gill lamellae curtails the diffusion of oxygen (David *et al.*, 2002 and Rudragouda Marigoudar *et al.*, 2009) which may ultimately reduce the oxygen consump-

tion by the animal (Kalavathy *et al.*, 2001). All metabolic pathways depend upon the efficiency of the gills for their energy supply as gills are the major respiratory organs and damage to these vital organs lead to respiratory distress. The present histological studies clearly indicate that the damage in gills of *Cirrhinus mrigala* decreases the uptake of oxygen which results in histotoxic anoxia. Due to this the gill tissue suffers from oxygen debt and losses the capacity to remove CO₂ from blood which is in accordance with (Sathivel *et al.*, 1991).

In the present study the observed alterations such as partial fusion of secondary lamellae, lifting of epithelial cells and proliferation of the epithelial cells, act as defense mechanisms towards toxicants. These alterations increase the distance between the external environment and the blood in the gills which serves as a barrier for the entry of contaminants. (Fernandes and Mazon, 2003 and Mallatt, 1985). When any type of toxicants meets gills, lamellar fusion in gills occur which is a protective measure as it diminishes the amount of vulnerable gill surface area in fish (Mallatt, 1985). when gills come in contact with any types of toxicants oedema appears to be a common feature of the gill pathology. Due to disturbance in branchial Na⁺, K⁺-ATPase pump by toxicants, solute accumulation in the epithelial cells occur and disturb the osmotic influx of water. This exchange protects the lamellar epithelial cells and prevents the entry of waterborne pollutants into the bloodstream (Arellano *et al.*, 1999).

Conclusion

The results of the present study suggest that the changes in gill histomorphology of *Cirrhinus mrigala* may serve as a rapid biological monitor to assess the impact of Cartap hydrochloride on other biotic communities in the water body. The results of the present study also stress on the diligent usage of the pesticide product to prevent the environmental pollution.

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