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Research Article

Histopathological Studies On Sonata (Biofungicide) Induced Stress in *Channa punctatus*

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ABSTRACT

The present study is aimed to assess the histological damage caused to the fish *Channa punctatus* was exposed to lethal concentration (15.39 ppm) to Sonata (Fungicide). Light microscopic studies exhibited severe histopathological changes in the Gill, Liver and Brain. The histopathological changes in the gill of fish include: epithelial lifting, degenerated secondary lamella, curling of secondary filaments and degeneration of epithelial cells. The histopathological changes in the Liver of fish include: blood cells among hepatocytes, appearance of blood streaks among hepatocytes, formation of vacuoles and degenerated hepato pancreatic tissue. The changes in the Brain of fish include: Degenerated dorsal olfactory area, degenerated Ventral olfactory area, blood streaks and degenerated septal area.

1. Introduction

In order to meet the growing population needs and demands, use of agrochemicals is inevitable for enhanced food production. Pesticides are highly effective substances used in control of pest and vectors of human disease. The increasing use of pesticides has caused concerns about their effects on human health and the environment. In spite of potential applications in agriculture, horticulture and other allied fields, they also exert some disadvantages, they include toxicity to animals, plants and human beings. Persistence of some of these chemicals in the environment and their subsequent entry into aquatic systems causes a great havoc. Pesticides and fungicides exert their toxic action on arthropods, mussels, fishes, frogs, turtles, water birds and other wild life too. Excessive use leads to bioaccumulation in farm workers, fruits, vegetables, nuts and food crops, consumers, and it also causes biomagnification at various trophic levels of the food chain. Although Indian average consumption of pesticide is far lower than many other developed economies, the problem of pesticide residue is very high in India [1]. Fungicides also threaten non target aquatic and terrestrial organisms through drift either by consumption or by ground water contamination. They enter water from agriculture and run off. The pollution of normal waters with synthetic chemicals has caused serious problems to the aquatic biota [2, 3,4,5]. Fishes are useful bio indicators and integrators of contaminants. They accumulate in gills, liver, kidney, and fat and induce metabolic changes associated with these organs.

2. Material and Methods

2.1 Animal Collection

The fish *Channa punctatus* specimen samples were collected from the freshwater lake located in Waddepally, Warangal district, Telangana State. Fish measuring 14-15cms in length and weighing 250-300gms specimens were brought to the laboratory immediately and analysed for various biological and nutritive value studies.

The fishes are acclimatized to the laboratory conditions in large plastic tanks with unchlorinated ground water for two weeks at a room temperature of 28±2°C. During the period of acclimatization, the fishes were fed with groundnut oil cake and rice bran. Feeding was stopped one day prior to the experimentation. All the precautions laid by committee on toxicity tests to aquatic organisms [6] were followed. After Acclimatization, Fishes were divided into groups and treated with concentrations of 10 and 20 ppm biofungicide sonata at time points 48, 72 and 96 hrs decide the lethal toxicity (LC50). The LC50 values were calculated the using probits analysis based on finney's (1952) table.

2.2 Tissue Collection

Gill, liver, and brain tissues were isolated from normal (not exposed to the toxicant) and experimental fish. Physiological saline solution (0.75% NaCl) was used to rinse and clean the

tissue. They were fixed in aqueous Bouins solution for 48 hr, processed through graded series of alcohols, cleared in xylene and embedded in paraffin wax. Gills alone were processed by double embedding technique. Sections were cut of 4-6 μ (microns) thickness; stained with Hematoxylin-Eosin (dissolved in 70% alcohol)[7] and were mounted in Canada balsam. Histopathological lesions were examined and photographed with the help of Intel Pentium QX3 computer attached microscope under 400X lens.

3. Results and Discussion

3.1 Gills

No histopathological changes were observed in the gill of the control fish. The structural detail of the gill of control *C. punctatus* is shown in Fig. 1A. The most common changes in 15.39 ppm concentrations of Sonata Fungicide were epithelial lifting, degenerated secondary lamella, curling of secondary filaments and degeneration of epithelial cells. The histological changes noticed in the pesticide exposed and control fishes are shown in fig.1 A (A1&B1).

3.2 Liver:

No histopathological changes were observed in the liver of the control fish. The structural details of the liver of control *C. punctatus* are shown in Fig. 2A. In the liver tissues of fish exposed to sonata concentrations of 15.39 ppm, blood cells among hepatocytes, appearance of blood streaks among hepatocytes, formation of vacuoles and degenerated hepato pancreatic tissue. The histological changes noticed in the pesticide exposed and control fishes are shown in Fig.2A (A1&B1).

3.3 Brain:

No histopathological changes were observed in the brain of the control fish. The structural detail of the brain of control *C. punctatus* is shown in Fig. 3A. The most common changes in 15.39 ppm concentrations of Sonata Fungicide were degenerated dorsal olfactory area, degenerated ventral olfactory area, blood streaks and degenerated septal area. The histological changes noticed in the pesticide exposed and control fishes and prawns are shown in fig.3 A (A1&B1).

The gills, which participate in many significant functions in fish, such as respiration, osmoregulation and excretion, remain in close contact with the external environment, and particularly sensitive to changes in the quality of the water, are considered the primary target of the contaminants [7,8,9]. Alterations like epithelial lifting, hyperplasia and hypertrophy of the epithelial cells, besides partial fusion of some secondary lamellae are examples of defence mechanisms, since; in general, these result in the increase of the distance between the external environment and the blood and thus serve as a barrier to the entrance of contaminants [10, 7, 11, 12].

Liver, the first organ to encounter any foreign molecule through portal circulation is subjected to more damage [13]. Liver is an important organ of detoxification which breaks down toxic substances and metabolites of administered substances. This breakdown is carried out by endoplasmic reticulum of hepatocytes. Due to these reasons the hepatic cells are damaged severely.[13] reported that in fish *Tilapia mosambica* exposed to the toxicant resulted in vacuolation and

necrosis in liver.[14] reported that *Channa punctatus* under Malathion toxicity showed the degenerative changes in liver.[15] reported that in teleost fish *Nemacheliusednesoni* (Day) exposure to phosphamidon caused highly vacuolated and cloudy swelling and even the connective tissue was damaged in liver. [16] Reported significant alterations in the hepatic cell count and the nucleocytoplasmic index in the liver of zebra fish *Brachydaniorerio* (cyprinid) exposed to 0.9 mg/l concentration of Malathion.

Like gills and liver in sonata treated *Chana punctatus* fish, Pathological changes were observed in brain samples also. Changes include Degenerated dorsal olfactory area, degenerated Ventral olfactory area, blood streaks and degenerated septal area. Similar changes were observed by [17] reporting swelling of the axon, atrophy, necrosis and pycnosis in the fish *Ctenopharyngodonidellus* under fenvalerate toxicity, and [18] on *Cirrhinusmrigala* exposed to the sublethal and lethal concentrations of technical grade as well as 20% EC of Chlorpyrifos for 8 days and the severity of damage is more in lethal exposures than in sublethal exposures. Quinalphos technical grade caused more degenerative changes in brain than in 25% EC exposures (Plate VI.3, Fig. B, C, D and E). [19] Reported that hexachlorocyclohexane was neurotoxic and induced vacuolation of brain parenchyma and moderate swelling of pyramidal cells of the cerebrum and opined that vacuolation may have been due to glycolysis leading to microsomal and mitochondrial dysfunctions. Loss of Nissle substances and glial cell reaction, with evidence of glial nodule formation in places, were proof of the neurotoxic nature of the chemical.

In the present investigation, gill, liver and brain tissues shows changes in their structures were observed during acute and sublethal sonata exposure which may indicate the different rates of free radical generation and different antioxidant potentials of these tissues. The present study also demonstrated that sonata has a high oxidative-stress-inducing potential in *Channa punctatus* and gill is the most sensitive organ in both acute as well as sub lethal concentration.

4. Conclusion

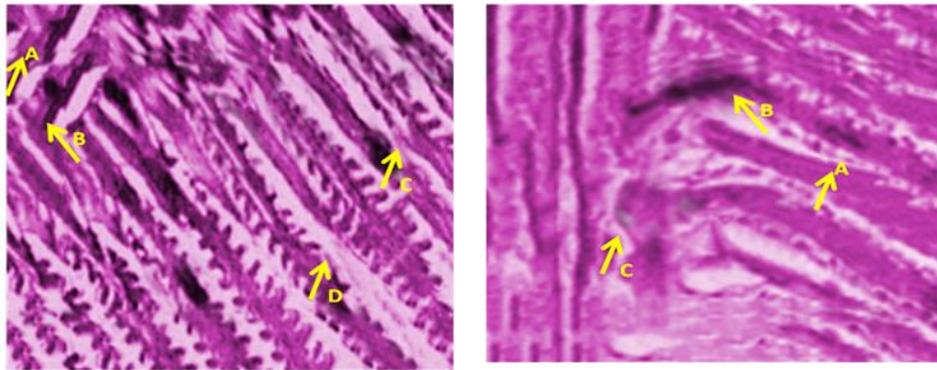
All the histopathological observation indicated that exposure to lethal concentrations of sonata caused destructive effect in the gill, liver and brain tissues of *C. punctatus*. Gill, liver and brain histopathological alterations, such as those observed in this study and findings from previous studies could result in severe physiological problems, ultimately leading to the death of fish. As a conclusion, the findings of the present histological investigations demonstrated a direct correlation between pesticide exposure and histopathological disorders observed in several tissues.

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Conflicting Interests

The authors have declared that no conflicting interests exist.

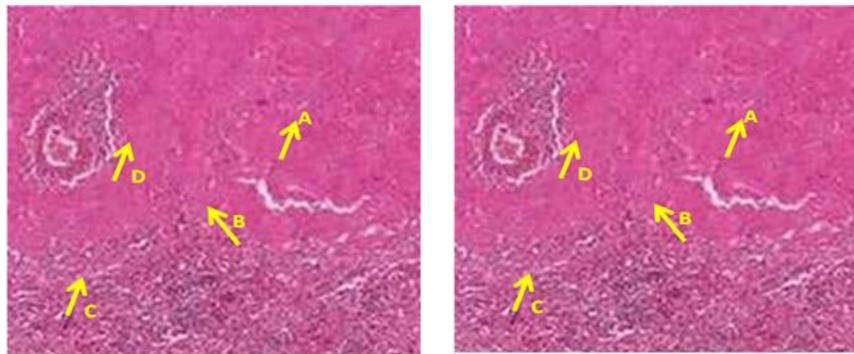


A1

B1

Figure-1: Histopathology Studies of *Chana Punctatus* in Gill samples treated at 96 hrs (A&B);

A1: Control : B1 Treated : In Control, A. Central Axis. B. Erythrocyte . C. Primary Gill Lamella. D. Secondary gill lamella : In Treated A. Epithelial Lifting. B. Curling of secondary gill filaments C. Degenerated secondary lamella

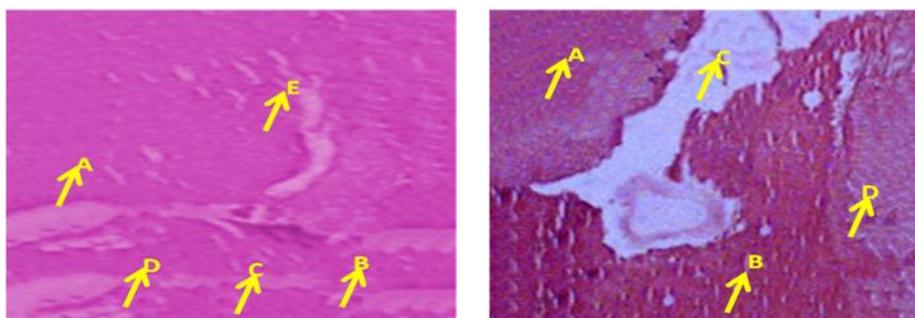


A1

B1

Figure-2: Histopathology Studies of *Channa Punctatus* in Liver samples treated at 96 hrs

(A1 & B1). A1 Control: B1 Treated. In Control, A. Hepatic cell. B. Nucleus. C. Lipid and glycogen granules: In Treated A. Degenerated hepato pancreatic tissue. B. Blood cells among hepatocytes C. Appearance of blood streaks among hepatocytes. D. Formation of vacuoles.



A1

B1

Figure3 A: Histopathology Studies of *Channa Punctatus* in Brain samples treated at 96 hrs (A1 & B1);

A1 Control: B1 Treated: In Control, A Dorsal olfactory area. B. Ventral olfactory area. C. Septal area. D. Tractus olfactorios medialis. E Tractus olfactorios lateralis: In Treated A. Degenerated dorsal olfactory area B. Degenerated Ventral olfactory area C. Blood streaks. D. Degenerated septal area.

Conflicting Interests

The authors have declared that no conflicting interests exist.

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