

ORIGINAL ARTICLE

Histopathological fluctuations in the muscle of *Etroplus suratensis* followed by Lambda-cyhalothrin exposure

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ABSTRACT

The current study was done to assess the influence of pesticide lambda-cyhalothrin on muscle tissues in histological aspects of the fish, *Etroplus suratensis*. Lambda-cyhalothrin is a pyrethroid insecticide, commonly utilized in agricultural, homes, public health & gardens. It can affect aquatic organisms since it has high solubility in water & could leach into water sources nearby. Fishes have an important role in assessing possible risk related to pollution of aquatic environment, hence they are desirable bio indicators of environmental contamination. They are directly presented to chemicals because of agricultural production or indirectly by food chain of ecosystem. To visualize the stress-induced structural alterations in cells & tissues, histopathology is considered as a successful apparatus. For this study, to observe the histopathological changes, 10 fishes lacking sex determination were presented to 0.005, 0.008, 0.006, 0.026 & 0.013 ppm (1/20, 1/12, 1/16, 1/4 & 1/8 near-lethal concentration of LC₅₀ value). For histopathological observation, fishes were randomly chosen from treated & control groups by sampling after 60 days of pesticide exposure. From this investigation, *E. suratensis* exposed to lambda-cyhalothrin, shortening of muscle bundles, rupture of muscle bundles, degeneration in muscle bundles, edema between muscle bundles, severe damage of muscle bundles and thickening of muscle bundle, necrosis and disorganization of muscle bundles were noticed. All these alterations show that the fish was under the highly stressful situations. The results of this examination propose that, lambda-cyhalothrin pollution is harmful to aquatic environment & this should be considered when the insecticide is utilized in agriculture & different fields.

Keywords: *Etroplus suratensis*, Hazardous, Histopathology, Lambda-cyhalothrin, Muscle, Stressful.

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INTRODUCTION

The control of insect pests involved in the heavy utilization of artificial insecticides, although the widespread utilization of these compounds has resulted in serious problems including toxic residue on grass & also to non-target organisms like fishes, birds & mammals [1,2]. These substances might intervene with a lot of significant physiological functions & change the levels of different biochemical constituents in fishes [3,4]. The primary sources of possible environmental dangers include herbicides, fungicides & insecticides & they not only affect fish, birds & other organisms but also humans when they get included in the food chain [5]. Numerous abnormalities & decrease in the life span of organisms are caused because of long term exposure to these substances [6,7]. Due to their low toxicity, high efficiency & easy biodegradability, pyrethroids are preferred over carbonates, organophosphates & organochlorines [8,9].

Pollution of water by pesticides, both indirectly or directly, can result in fish lethality, decrease in fish production, or increasing concentrations of harmful chemicals in edible fish tissue that harms humans that consume these fish[10,11]. In food & drinking water, remaining amounts of pesticides & their metabolites are seen, which poses a threat to human health due to introduction to these chemicals[12,13]. At regional, local, global & national levels, pollution of surface waters has been recorded worldwide & is considered as a major issue[14]. The toxicity of these contaminants is evaluated by the degree of histopathological damages produced in the test organism & the extent of cell damage is apparent relative to the concentrations of contaminants employed[15]. Due to the pathological lesions caused by contaminants, mortality of fishes is seen. For the evaluation of stress response to contaminants, histopathological methods are comparatively inexpensive, sensitive, rapid & reliable. So, various near-lethal effects of lambda-cyhalothrin on structure of muscle tissues from *E. suratensis* were recorded[16].

MATERIAL AND METHODS

A group of 10 fishes lacking sex determination were presented to 0.005, 0.008, 0.006, 0.026 & 0.013 ppm (1/20, 1/12, 1/16, 1/4 & 1/8 near-lethal concentration of LC₅₀ value) to observe the histopathological alterations. The pesticide free medium & the test solution were taken, inside which the fishes were kept and was renewed every day. From treated & control groups, fishes were chosen randomly for histopathological observations after 60 days of exposure to pesticide by sampling. The muscle of pesticide & control treated *E. suratensis* were removed & a histological evaluation was done by employing Culling (1974) process.

The dissected tissues were fixed using formalin (25 %), acetic acid (5 %) & Bouin's fixative saturated aqueous solution of picric acid (75 %) for 24 hrs. The fixed tissues were cleaned in running tap water overnight. After that, the tissues were maintained in 50% isopropyl alcohol, and then dehydrated using ascending isopropyl alcohol grades. At last, the tissues were dehydrated totally by two alterations of 100 percent alcohol for 20-30 minutes each. Thus, the tissues were, dealcoholized through alterations of xylol, 20-30 minutes in each alteration. Then it was in filtered with paraffin wax of congealing point 56-58°C. The tissues were kept in molten wax for a minimum of 30 minutes in the embedding bath. Three alterations in paraffin were done for infiltration, so as to remove the entire clearing agent (xylol). Then, after correct orientation of the prepared tissues, they were inserted in labeled paper boats. In a rotary microtome (WEXWAX-SPENCER), the inserted tissues were cut at 7 µm (micrometer) in thickness. On warm water poured which was put on acid-cleaned glass slides & glued with a thin layer of a fixative the ribbon of section was slashed & floated. As fixatives, Hampt's fixative & Mayer's albumen were used.

The slides were kept all night on a slide warmer whose temperature was controlled at 10-15°C below the melting point of paraffin wax after total air drying for some hours. Then the sections were deparaffinized in xylene and dehydrated in decreasing iso-propyl alcohol grades, 90%, 70% and 50% alcohol series and then in distilled water. Then the sectioned pieces were stained using Ehrlich's haematoxylin for 15 min, and destained in dilute hydrochloric acid. After dehydration in 70%, 50%, 90% iso-propyl alcohol & distilled water, they were counter stained using Eosin. The stained sections were rapidly dehydrated in ascending grades of isopropyl alcohol and counter stained in Eosin. The slides were mounted with DPX mountant & were washed in xylene. The chosen slides were photographed by utilizing computerized Kyowa-Trinocular Microscope with CC TV attached.

RESULTS

In the present study, the results of histological observations of muscle tissues of *E. suratensis* of control and those introduced to sub-lethal concentrations of lambda-cyhalothrin were given in plates 1 to 6. Various structural changes in sections of muscle tissues of fish were taken from treated group and observed under light microscope. The tissues of fishes from control group fishes had different structure from that of lambda cyhalothrin treated groups. The histological study of the muscle tissues of the control fish showed the appearance of normal myotomes having evenly spaced muscle bundles. It indicates the fish in unstressed conditions (Plate 1).

The histopathology of the muscle tissues of the fish *E. suratensis* introduced to lower concentration (0.005 ppm) showed some remarkable changes like, shortening of muscle bundles and rupture of muscle bundles was observed (Plate 2). Fishes introduced to 0.006 ppm of lambda-cyhalothrin showed completely damaged and degenerated muscle bundles in some areas of muscle tissue (Plate 3). Plate 4 revealed vacuolar deterioration in muscle bundles, necrosis, severe damage of muscle bundles and edema between muscle bundles were noticed in 0.008 ppm concentration of lambda-cyhalothrin. The muscle tissue of *E. suratensis* introduced to near-lethal concentration of lambda-cyhalothrin (0.013 ppm) revealed edema between muscle bundles, thickening of muscle bundles, necrosis and disorganization of

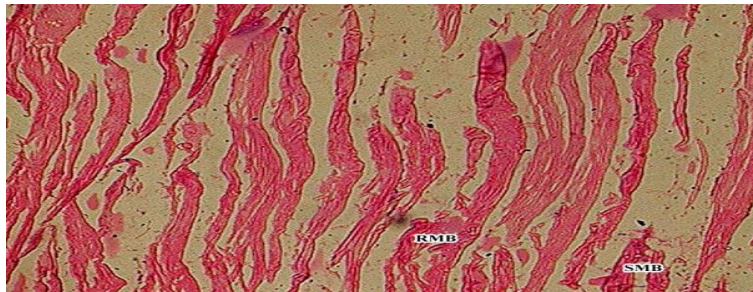
muscle bundles were discovered (Plate 5). In greater concentration (0.026 ppm) of *E. suratensis* considerable changes like disorientation of muscle bundles, condensation of muscle bundles & also severe intra muscular edema were reported in the pathological finding (Plate 6).

Plate 1: The muscle of control *E. suratensis* (200X) can be seen in this section.



MB-Muscle bundles

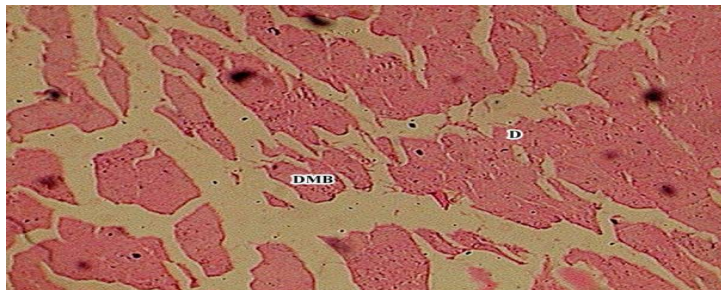
Plate 2: The muscle of *E. suratensis* introduced to 0.005 ppm concentration of lambda-cyhalothrin (200X) can be seen in this section



RMB-Rupture of muscle bundles

SMB-Shortening of muscle bundles

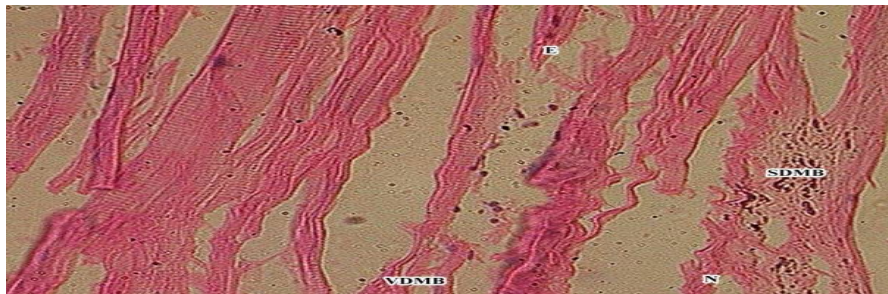
Plate 3: The muscle of *E. suratensis* exposed to 0.006 ppm concentration of lambda-cyhalothrin (400X) can be seen in this section



D-Damaged muscle bundles

DMB-Degeneration in muscle bundles

Plate 4: The muscle of *E. suratensis* introduced to 0.008 ppm concentration of lambda-cyhalothrin (400X) can be seen in this section

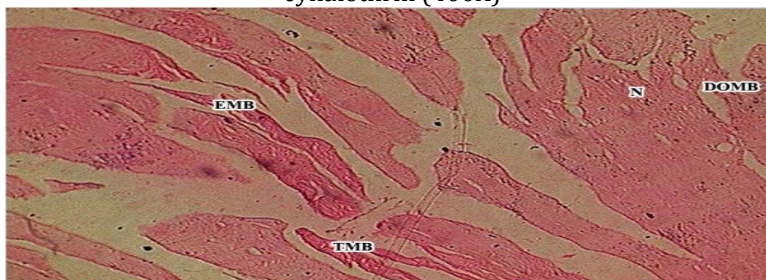


VDMB-Vacuolar degeneration in muscle bundles N-Necrosis

SDMB-Severe damage of muscle bundles

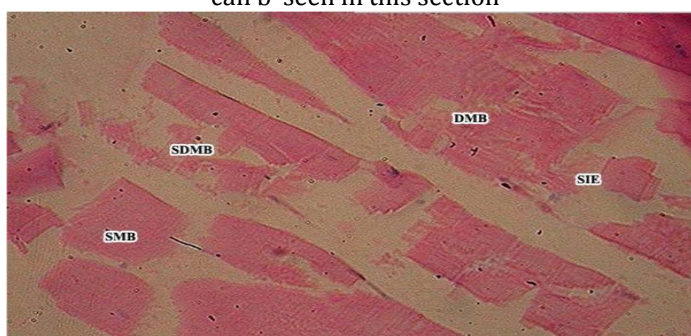
E-Edema between muscle bundles

Plate 5: Section showing the muscle of *E. suratensis* exposed to 0.013 ppm concentration of lambda-cyhalothrin (400X)



EMB-Edema between muscle bundles
TMB-Thickening of muscle bundle N-Necrosis
DOMB-Disorganization of muscle bundles

Plate 6: The muscle of *E. suratensis* exposed to 0.026 ppm concentration of lambda-cyhalothrin (400X) can be seen in this section



DMB – Disorganization of muscle bundles
SDMB – Severe damage of muscle bundles
SMB – Shortening of muscle bundles
SIE – Severe intra muscular edema

DISCUSSION

In the present study, *E. suratensis* exposed to lambda-cyhalothrin, shortening of muscle bundles, rupture of muscle bundles, deterioration in muscle bundles, edema between muscle bundles, severe damage of muscle bundles, thickening and disorganization of muscle bundles were noticed. All these alterations indicate the fish under the highly stressful conditions. These findings are in dependable with the works of Dhevkrishnan and Hussain [7] on *L. rohita* exposed to Cauvery river pollutants. The histopathology of the muscle tissues of the freshwater fish *L. rohita* collected from less polluted region showed some remarkable changes like, shortening of muscle bundles, thickening of muscle bundles and necrosis of muscle bundles were observed. Similar alterations have also been recorded by [10, 6, 11, 3, 12].

The histopathological changes were more evident in *E. suratensis* revealed to lambda-cyhalothrin and were not found in the control fish. The histological investigation findings showed that destructive effect was caused in the muscle of *E. suratensis* due to exposure to lambda-cyhalothrin utilized for controlling. Therefore, considering the muscle injuries of the organism exposed to lambda-cyhalothrin, it has been shown that the conditions of this environment were not satisfactory for the development and survival of the fishes and other organisms including human beings. Biological methods could be used for controlling insect pests in agriculture, public health, homes and gardens instead of lambda-cyhalothrin in order to protect the natural environment.

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