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## RESEARCH ARTICLE

### EFFECTS OF LAMBDA-CYHALOTHRIN ON FISH FINGERLINGS OF COMMON CARP, *Cyprinus Carpio*

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#### ABSTRACT

The toxicity tests using early life stages of fish are of great importance in assessing risk of growth, reproduction and survival in polluted environments are important tools for good environmental monitoring. In the present study, the toxic effect of Lambda-cyhalothrin to the fingerlings of Common carp *Cyprinus carpio* was evaluated. The aim of the present work was to study the toxic effects of lambda-cyhalothrin on the total protein, total carbohydrates, amount of cholesterol and sterols. The LC<sub>50</sub> of lambda-cyhalothrin were estimated by Static renewal method. The Lambda-cyhalothrin of LC<sub>50</sub> was 0.2µl/l, 0.18µl/l, 0.12µl/l, 0.1µl/l at 24h, 48h, 72h, and 96h respectively. The fish exhibited erratic swimming, loss of equilibrium, copious mucus secretion and hitting to the walls of test tubs prior to mortality. During the present investigation we observed significant alterations in the total protein, total carbohydrates and amount of cholesterol and sterol contents in the tissue of *Cyprinus carpio* exposed to Lambda-cyhalothrin compared to control group. So, that the indiscriminate uses of synthetic pesticide have caused environmental contamination and toxicity to living organisms. The study strongly suggests that the use of eco-friendly products such as bio-pesticide agents which are available abundantly in nature.

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## INTRODUCTION

Pesticides are the chemical products used for plant protection. A pesticide is any substance or mixture of substances used to destroy, suppress or alter the life cycle of any pest (Bhushan *et al.*, 2013). There are 234 pesticides registered in India. A total of 210,600 metric tons of pesticides were consumed during 2005-2010 all over India. The harmful effects of the pesticides are now established worldwide. The harm caused may be acute or chronic in nature. Farmers and agricultural laborers are the direct users of pesticides and are more likely to get affected by the acute toxicity of pesticides. The chronic toxicity affects the whole population. The residues left in the crops, soil and water after use get into the human food chain. Aquatic resource potentially useful to humans for agricultural, industrial, household, recreational, research, navigation etc, about 97.5% of water on the earth is salt water, leaving only 2.5% as fresh water. Now a day, pesticides are being widely used in agricultural fields to get rid of some toxic harmful pests and in turn to increase production. The pesticides which enter the animal system through food chain are known to cause deleterious effects. In India, the use of pesticides has now reached over 1020 tons per annum (Hicks *et al.*, 1998).

Since, the middle of the last century, the world has witnessed a steady growth in the use of synthetic pesticides. These synthetic pesticides are more potent and cheaper than natural products. Majority of the synthetic pesticides that are used in today's agriculture are highly toxic, relative and they are connected with serious health hazards and environmental pollution (Luo *et al.*, 1997). Fish have been used as indicator for contamination of aquatic environment. During environmental catastrophe, fish are unable to escape from the site affected, thus bio-accumulate toxic substance (Andrade *et al.*, 2004). Fish acts as the last link in the food chain in the aquatic environment, they may negatively influence the food safety when they get exposed to contaminated environment. One such dangerous and toxic synthetic pesticide is Lambda cyhalothrin. It is an insecticide registered by the U.S Environmental protection agency (EPA) in 1988. Lambda-cyhalothrin belongs to a group of chemicals called pyrethroids. Pyrethroid including Lambda-cyhalothrin disrupts the normal functioning of the nervous system in an organism. By disrupting the nervous system of animals Lambda-cyhalothrin may cause paralysis or death. An effect of Lambda-cyhalothrin on environment depends on the amount of pesticide present, the length and frequency of exposure.

Scientists have observed that Lambda-cyhalothrin is extensively metabolized in mammalian species. The main routes of metabolism include ester hydrolysis, oxidation and conjugation. However, the metabolism of this compound in fish is oxidative. The present investigations were carried out to obtain further information about disturbances total protein, total carbohydrates and amount of cholesterol and sterol contents in the tissue of *Cyprinu scarpio* due to exposure to Lamda-cyhalothrin.

## MATERIAL AND METHODS

**Experimental animal:** *Cyprinus carpio* L. is one of the important Indian major carp (Fig. 1), which together with other major carps contribute to more than 81.53% of the inland aquaculture production in India (FAO, 2003) is being selected as test animal for the present study. It is a bottom dweller and omnivorous and they feed mainly on insects, crustaceans, crawfish and benthic worms.



Fig. 1. Experimental animal *Cyprinus carpio*

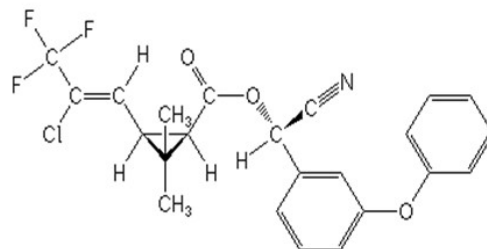
**Procurement and maintenance:** Fingerlings with 8±2cms length and 25±2grams of weight of *C. carpio* were collected from the State Fisheries Department, Bhadra Reservoir Project, Shimoga, Karnataka and were reared in a large tubs and acclimatized in tap water having dissolved oxygen 6.02±0.8 mg/L, P<sup>H</sup> 7.4±0.2, Total hardness of 28±2 mg/L and room temperature 29±1°C for 15 days prior to bringing them to laboratory. Prior to stocking in the large plastics tubs, fishes were washed with Potassium permanganate (1ppm). Fishes were fed with powdered rice bran and groundnut oil cake (3:1) and also a commercially available food then feeding was stopped 24 hours prior to expose to the test medium for acute toxicity tests. The water of the plastic tubs was changed daily once to avoid any fungal and bacterial contamination and 1% Potassium permanganate solution was sprayed to eradicate any bacterial or fungal infection. The temperature of water in the tubs was 27±1°C and the same was the maintained throughout the course of investigation.

**Physico-chemical characterization of water:** The physico-chemical characteristics of fish habitat water were analyzed by following the methods mentioned in APHA, AWWA and WEF (2005) viz., temperature, pH, dissolved oxygen, Carbon dioxide, Total hardness as CaCO<sub>3</sub>/L, Alkalinity and Chlorine. All the water quality parameters were monitored throughout the experimental period except for minimal variation which is tolerated by the fish in the wild.

**Nature of Lambda-cyhalothrin:** Lambda-cyhalothrin is a synthetic pyrethroid insecticide and acaricide used to control a wide range of pests in a variety of applications. Pest includes aphids, colarado beetles and butterfly larvae. It may be used for pest management or in public health applications to control insects such as cockroaches, mosquitoes, ticks and flies which

act as a disease vectors. Lambda-cyhalothrin (Fig. 2) is available as an emulsifiable concentrate, wettable powder or ULV liquid.

a. (S)-alcohol (Z)-(IR)-cis-acid



b. (R)-alcohol (Z)-(1S)-cis-acid

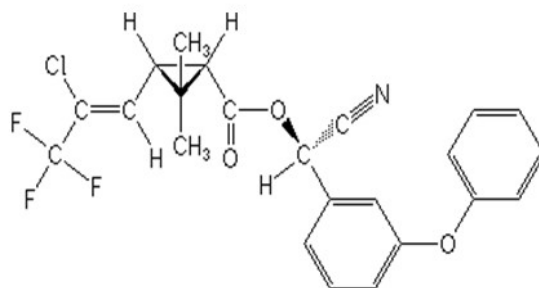


Fig. 2. The structure of two isomers of Lambda-cyhalothrin

**Toxicity test:** The LC<sub>50</sub> value of Lambda-cyhalothrin was determined by treating the fishes by Static renewal bioassay method (Benoit *et al.*, 1993). In aquatic organisms the dermal route of administration is the natural way of exposure to pollutants and since the route of administration in evaluation of toxicity is usually the route through which the organism is naturally exposed to toxicants. The test animal i.e., fish were kept in large plastic tubs, capacity of containing 20 liter of water. The plastic tubs, which contained the test solutions of different concentrations of insecticides. The fingerlings size (8±2cms length and 25±2grams of weight) of common carp fishes were introduced into each plastic tubs and chronic test was conducted. In each tubs, 10 fishes of the same species (*Cyprinus carpio*) were introduced containing known concentration of test solution for the toxicity test, during the test period, they were not fed to avoid variation in feeding habits. The mortality was noted down for every 24hours intervals till 96hours duration. The dead animals were removed immediately and the test solution was renewed for every 24hours till 96hours along this, controls were also maintained without test chemicals. The LC<sub>50</sub> were calculated by plotting percent mortality v/s toxicants concentration in a double logarithmic grid. Straight lines were fitted and concentration of toxicants at which there was 50% mortality was noted to represent LC<sub>50</sub> for a given duration of exposure.

**Analysis of Biochemical Parameters:** The muscle tissue was extracted by dissecting the fishes from both control and treated fishes and homogenized the 100mg of tissue using 10ml of distilled water separately and estimated total protein, total carbohydrates and Cholesterol by Lowry's Method (Lowry *et al.*, 1951), Hedge and Hofreiter Method (Hedge and Hofreiter,

1962) and Zak's Method (Zak, 1957) respectively. The estimation of Sterols is by Liebermann-Burchard Method (Liebermann, 1885) taking 100mg of tissue from same fishes and homogenized in 10ml of glacial acetic acid.

**Statistical Analysis:** The statistical analysis was done for Biochemical parameters by using SPSS software. Mean  $\pm$  Standard Deviation (SD) was calculated.

## RESULTS AND DISCUSSION

The physico-chemical characteristics of water sample were analyzed. temperature  $26\pm 1^\circ\text{C}$ , pH  $7.4\pm 0.2$  at  $29^\circ\text{C}$ , dissolved oxygen  $6.02\pm 0.8\text{mg/L}$ , Carbon dioxide  $3.8\pm 1.0\text{mg/L}$ , Total hardness  $28\pm 2\text{mg}$  as  $\text{CaCO}_3/\text{L}$ , Alkalinity  $40\pm 10\text{mg/L}$ , Chlorine  $12.78\pm 1.0\text{mg/L}$ .  $\text{LC}_{50}$  values were decreased with the increase in chemical exposure duration and Lambda-cyhalothrin was more toxic to fingerlings. The present study showed that during the exposure of Lambda-cyhalothrin, the 24 h  $\text{LC}_{50}$  was  $0.2\mu\text{l/l}$  while for 48 and 72h it is declined to 0.18 and  $0.12\mu\text{l/l}$  respectively and  $0.1\mu\text{l/l}$  after 96h of exposure (Table 1; Fig 3). Result will indicate that the synthetic pyrethroids are much toxic for the early life stage of Common carp. Toxicity test with fingerlings is valuable for assessing potential impacts on growth, reproduction and survival of fish in polluted environment and are important tools for good environmental monitoring. The fingerlings exhibited normal swimming behavior in the control condition while erratic and jerky swimming was observed among in the treated media. They became restless, aggregated at one corner of the tub. These changes may be related to the consequent alteration in the physiological process due to chemical exposure. At 96h exposure the pectoral and pelvic fins were expanded and they rolled vertically prior to death. In some cases the fingerlings exhibited inconsistent jumping, frequent surfacing, secreted mucous from whole body, loss of equilibrium and decrease in opercula beat. The fish exhibited peculiar behavior of trying to leap out from pesticide medium which can be viewed as an escaping phenomenon. They often spiral rolled at intervals and finally the fishes sank to bottom with their least operculum movements and died with their mouth opened. During the present investigation we observed significant alterations in the total protein, total carbohydrates, and amount of cholesterol and sterol contents in the tissue of *Cyprinus carpio* exposed to Lambda-cyhalothrin. The amount of protein estimated in control fish was  $7.293\pm 0.267\text{mg/g}$  whereas in Lambda-cyhalothrin  $1.37\pm 0.252\text{mg/g}$  (Table 2). The treated fishes showed significantly lower protein levels when compared to untreated control group (Fig 4). The amount of carbohydrate estimated in control and Lambda-cyhalothrin treated were  $7.183\pm 0.300\text{mg/g}$  and  $2.097\pm 0.108\text{mg/g}$  respectively (Table 2). The highest reduction of carbohydrate was noticed after 96hrs of exposure (Fig 5). The total cholesterol estimated in the control group was  $0.413\pm 0.015\text{mg/g}$  whereas Lambda-cyhalothrin treated tissue was  $0.073\pm 0.006\text{mg/g}$  (Table 2). The exposure of Lambda-cyhalothrin caused significant decrease in total sterol in the tissues. The amount of total sterol was  $0.917\pm 0.006\text{mg/g}$  and  $0.14\pm 0.01\text{mg/g}$  in control and Lambda-cyhalothrin treated fishes respectively (Table 2). Lambda-cyhalothrin exposure caused a significant decrease in the level of cholesterol and sterol contents in the tissues compared to control group (Fig 6&7).

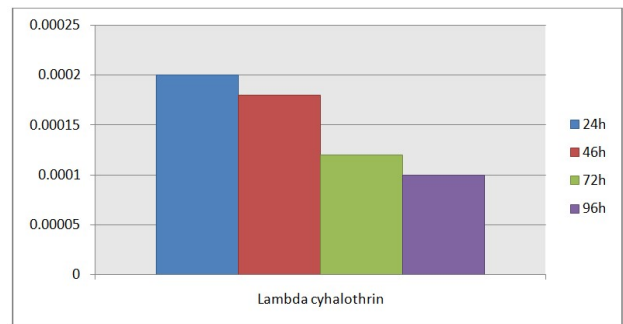


Fig 3.  $\text{LC}_{50}$  values of Lambda-cyhalothrin for *C. carpio* from 24h to 96h time intervals

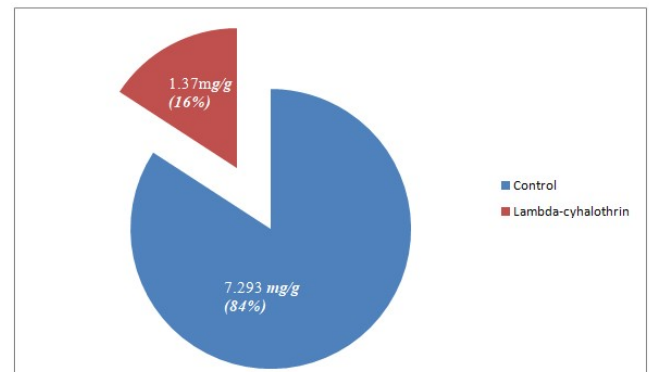


Fig. 4. Total protein in the muscle tissue of experimental animal (in mg/g)

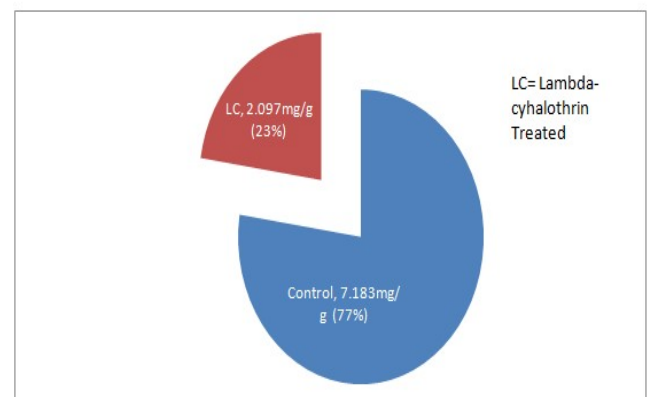


Fig 5. Total carbohydrates in the muscle tissue of experimental animal (in mg/g)

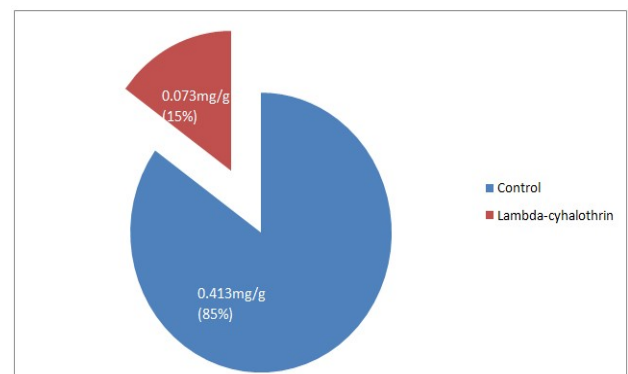


Fig 6. Total Cholesterol in the muscle tissue of experimental animal (in mg/g)

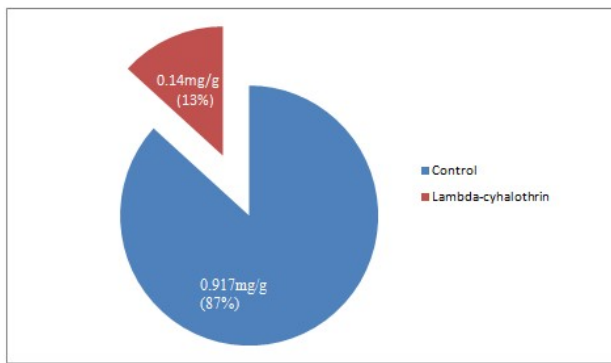


Fig 7. Total sterols in the muscle tissue of experimental animal (in mg/g)

Table 1. LC<sub>50</sub> values of Lambda-cyhalothrin on C. carpio from 24h to 96h time intervals (in ml)

Pesticides	24h	46h	72h	96h
Lambda-cyhalothrin	0.00020	0.00018	0.00012	0.00010

Table 2. Effect of Lambda-cyhalothrin on biochemical parameters in fingerlings of C. carpio (Average±SD in mg/g)

Parameters	Protein	Carbohydrates	Cholesterol	Sterol
Control	7.293±0.267	7.183±0.300	0.413±0.015	0.917±0.006
Lambda-cyhalothrin	1.37±0.252*	2.097±0.108*	0.073±0.006*	0.14±0.010*

\*significant at P <0.0001

## SUMMARY AND CONCLUSION

The aim of the present work was to study the effect of Lambda-cyhalothrin on the total protein, total carbohydrate and amount of cholesterol and sterol contents in tissues of common carp, *Cyprinus carpio* (*Cyprinidae*), after 96h exposure to sub lethal concentration of Lambda-cyhalothrin. During the present study we concluded that the fingerlings of common carp are very sensitive to low level of Lambda-cyhalothrin because the reduction in all the macromolecules was significantly reduced in aquatic environment and significantly affect its population. Because Lambda-cyhalothrin is commonly applied to rice fields to control insects, potential water and sediment contamination may lead to toxicity in aquatic organisms such as mosquito, fish, shrimps, crabs and clams. The widespread use of Lambda-cyhalothrin has resulted in residues in sediment, which have been found to be toxic to aquatic organisms including fish and amphipods. Therefore, pesticides should be used with great caution and in a sustainable way so that it may not be hazardous to aquatic environment and human beings. Moreover, extensive investigations should be done for their safe use.

Consequently, it is important to identify the impact of products have on certain biochemical parameters. Water pollution biomarkers are early diagnostic tools for measuring biological effects and assessing environmental quality. The indiscriminate uses of synthetic pesticide have caused environmental contamination and toxicity to living organisms. The present study strongly suggests that the use of eco-friendly products should be encouraged. Eco-friendly bio-pesticide agents are available abundantly in nature.

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