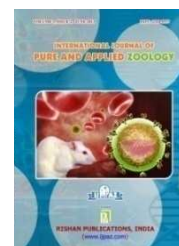




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STUDIES ON THE ACUTE TOXICITY OF PESTICIDES ON THE FRESHWATER FISH *LABEO ROHITA*

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ABSTRACT

The present study was carried out to investigate the LC₅₀ of two different pesticides such as monocrotophos and lambda cyhalothrin on the freshwater fish *Labeo rohita*. Monocrotophos caused 100% mortality of *L. rohita* at 0.0044ppm and 50% mortality (96 hours) at 0.0036 ppm, and for lambda cyhalothrin, the lethal effect was at 0.0029 ppm and LC₅₀ at 0.0021 ppm. The LC₅₀ values obtained at 24, 48, 72 and 96 hours exposures and the 95% confidence limits for the two pesticides revealed that lambda cyhalothrin showed higher toxicity than monocrotophos. The LC₅₀ values of monocrotophos for 24, 48, 72 and 96 hours were 0.0041, 0.0039, 0.0037 and 0.0036 ppm respectively, whereas the LC₅₀ value of lambda cyhalothrin for 24, 48, 72 and 96 hours were 0.0026, 0.0024, 0.0022 and 0.0021 ppm, respectively.

Key words: Monocrotophos, lambda cyhalothrin, mortality, *Labeo rohita*.

INTRODUCTION

Indiscriminate use of different pesticides in agriculture to prevent crop damage from pests has been increasing over two decades especially in developing countries (Santhakumar and Balaji, 2000). These pesticides through surface run off reach unrestricted areas like ponds and rivers and alter the physico-chemical properties of water and consequently affecting aquatic organisms (Kamble and Muley, 2000, Bhachandra *et al.*, 2001; Madhab Prasad *et al.*, 2002 and Sindhe *et al.*, 2007).

In assessing the safety level of any poisonous chemical for higher animals, the first task is to determine the acute toxic LC₅₀ value, a simple expression of the degree of toxicity that can be understood by toxicologists (Doubois and Geiling, 1959). The increasing awareness of aquatic pollution demands toxicity tests to assess the efficacy of the contaminants and to extrapolate their safe levels permissible in the environment. The median tolerance limit of any pollutant is meant as an elementary guide in the field of toxicology (Ward and Parrish, 1982). Without reference to the median tolerance limit, no information on sublethal effects can be

deduced (Patin, 1982). Dermal acute toxicity tests represent an important method for establishing criteria to evaluate water quality and therein to protect the aquatic environments (NAS/NAE, 1972). Acute toxicity studies are generally employed to compare the sensitivities of different species to different potencies of the chemicals and to derive, by using LC_{50} values, environmental concentration of chemicals which could be considered 'safe'.

Toxicity data for a variety of pesticides such as organophosphate, organochlorine, carbamide and pyrethroid pesticides have been reported for number of fish species by various authors (Anees, 1975; Arunachalam and Palanichamy 1982; Arunachalam *et al.*, 1980; Baskaran *et al.*, 1989; Roy and Dutta Munshi, 1988; Singh *et al.*, 1981; Malla Reddy and Basha Mohideen, 1989; Gurusamy and Ramdoss, 2000; Sapna Shrivastava, 2002; Nishar Shailkh and Yeragi, 2004 and Visvanthan *et al.*, 2009). Variations in LC_{50} if any, under different energy balance should reflect on the nutritional status of the animal. This would help to assess the productivity of the aquatic medium concerned and to take appropriate corrective measures. The present work has been carried out to study the lethal concentration 50% (LC_{50}) of pesticides monocrotophos and lambda cyhalothrin on the freshwater fish *Labeo rohita*.

MATERIALS AND METHODS

Fish Acclimatization

The freshwater healthy fish, *L. rohita* of the weight (10 ± 1 g) and length (8 ± 0.5 cm) were selected for the experiment and were collected from Katherasan Aquafarm near Thanjavur. Fish were screened for any pathogenic infections. Glass aquaria were washed with 1% $KMnO_4$ to avoid fungal contamination and then sun dried. Healthy fishes were then transferred to glass aquaria (35×20×20 cm) containing dechlorinated tap water (Temperature $28 \pm 2^\circ C$; total hardness 518 ± 23 mg/l; dissolved oxygen 5.6 ± 0.2 mg/l; salinity 1.2 ± 0.13 ppt and pH 7.8 ± 0.04). Fish were acclimated to laboratory conditions for 10 to 15 days prior to experimentation. They were regularly fed with commercial food *ad libitum* and the medium (tap water) was changed daily to remove faeces and food remnants.

Acute toxicity test

Toxicity tests were conducted in accordance with standard methods (APHA, 1992). Stock solution of monocrotophos with a concentration of 1 ml per litre (equivalent to 1 ppt) was prepared in distilled water and different dilutions were prepared by adding required amount of distilled water. The stock solution of lambda cyhalothrin with a concentration of 1 ml per litre was also prepared in distilled water and the desired degree of concentrations was prepared. Based on the progressive bisection of intervals on a logarithmic scale, log concentrations were fixed after conducting the range finding test. The fish were starved for 24 hours prior to their use in the experiments as recommended by storage to avoid any interference in the toxicity of pesticides by excretory products. After the addition of the toxicant into the test tank with 10 litres of water having twenty fish, mortality was recorded after 24, 48, 72 and 96 hours. Five replicates were maintained simultaneously.

Per cent mortality was calculated and the values were transferred into probit scale. Probit analysis was carried out as suggested by Finney (1971). Regression lines of probit against logarithmic transformations of concentrations were made. Confidential limits (upper and lower) of the regression line with chi-square test were calculated by a computerized programme for Finney's (1971) probit analysis.

RESULTS AND DISCUSSION

Monocrotophos caused 100% mortality of *L. rohita* at 0.0044 ppm and 50% mortality (96 hours) at 0.0036 ppm, and for lambda cyhalothrin, the lethal effect was at 0.0029 ppm and LC_{50} at 0.0021 ppm. The LC_{50} values obtained at 24, 48, 72 and 96 hours exposures and the 95% confidence limits for the two pesticides revealed that lambda cyhalothrin showed higher toxicity than monocrotophos. The LC_{50} values of monocrotophos for 24, 48, 72 and 96 hours were 0.0041, 0.0039, 0.0037 and 0.0036 ppm respectively (Table 1; Figures 1-4), whereas the LC_{50} value of lambda cyhalothrin for 24, 48, 72 and 96 hours were 0.0026, 0.0024, 0.0022 and 0.0021 ppm, respectively (Table 2 and Figures 5-8).

Table 1. Per cent mortality of *Labeo rohita* exposed to different concentrations of monocrotophos for different periods.

Hours of Exposure	LC ₅₀	L.C.L	U.C.L	Regression Equation	Calculated χ^2 value	Table χ^2 value
24	0.004084	0.004003	0.004167	$Y = 32.08157 + 11.3365 X$	8.2512	12.59
48	0.003870	0.003796	0.003946	$Y = 33.42346 + 11.78303 X$	9.0221	11.07
72	0.003726	0.003662	0.003791	$Y = 37.45153 + 13.36139 X$	3.5032	11.07
96	0.003556	0.003502	0.003611	$Y = 42.9766 + 15.50699 X$	6.7556	11.07

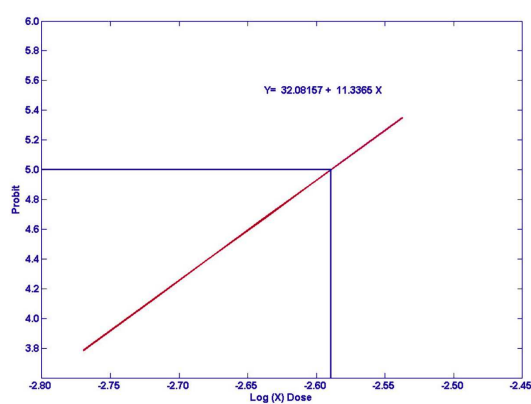
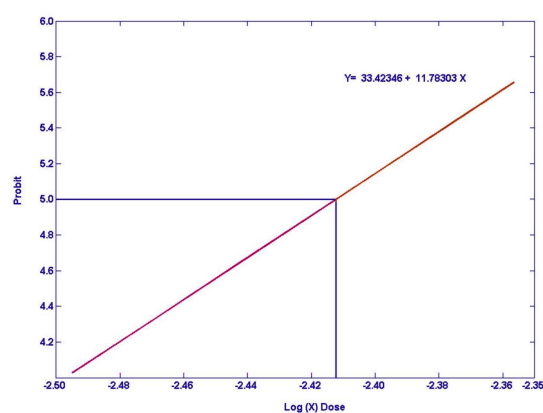
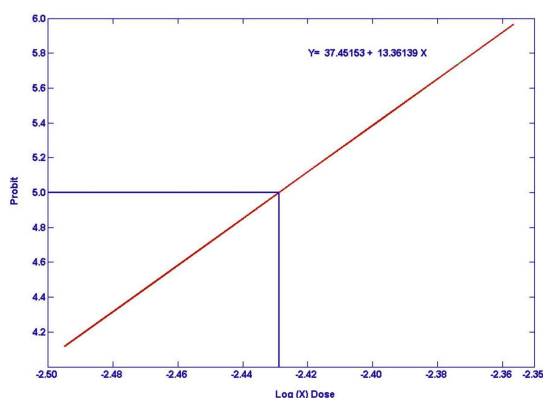
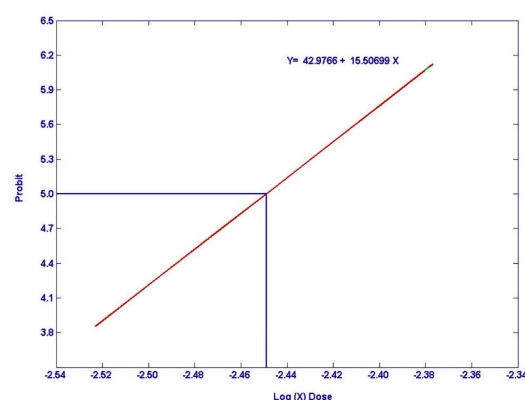
**Figure 1.** Regression line (based on Probit analysis) of log concentration of pesticide monocrotophos Vs Per cent mortality of *L. rohita* for 24 hours.**Figure 2.** Regression line (based on Probit analysis) of log concentration of pesticide monocrotophos Vs Per cent mortality of *L. rohita* for 48 Hours.**Figure 3.** Regression line (based on Probit analysis) of log concentration of pesticide monocrotophos Vs Per cent mortality of *L. rohita* for 72 hours.**Figure 4.** Regression line (based on Probit analysis) of log concentration of pesticide monocrotophos Vs Per cent mortality of *L. rohita* for 96 hours.

Table 2. Per cent mortality of *Labeo rohita* exposed to different concentrations of lambda cyhalothrin for different periods.

Hours of Exposure	LC ₅₀	L.C.L	U.C.L	Regression Equation	Calculated χ^2 value	Table χ^2 value
24	0.002575	0.002474	0.002679	Y= 22.44465 + 6.737374 X	5.6685	11.07
48	0.002370	0.002286	0.002457	Y= 21.49032 + 6.281464 X	8.8858	12.59
72	0.002156	0.002023	0.002298	Y= 24.59749 + 7.350035 X	17.6868	12.59
96	0.002030	0.001967	0.002095	Y= 23.85884 + 7.004488 X	8.9140	12.59

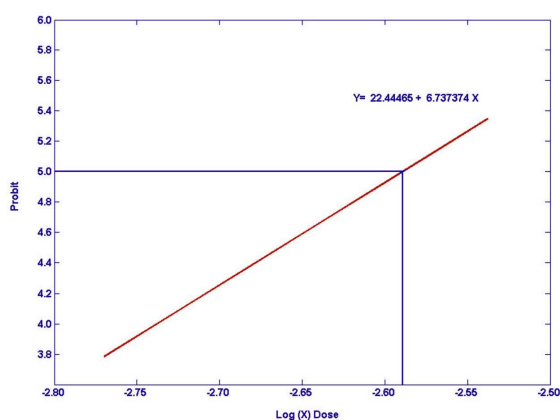


Figure 5. Regression line (based on Probit analysis) of log concentration of pesticide Lambda cyhalothrin Vs Per cent mortality of *L. rohita* for 24 hours

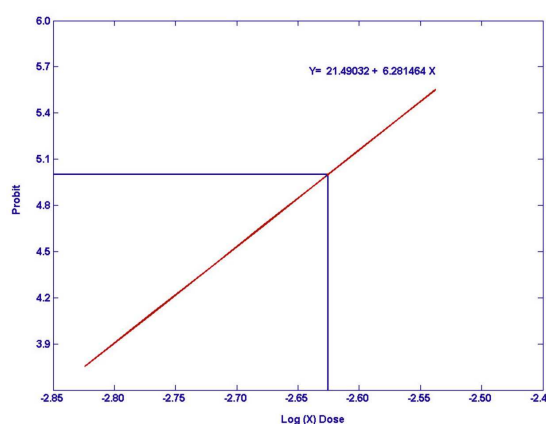


Figure 6. Regression line (based on Probit analysis) of log concentration of pesticide Lambda cyhalothrin Vs Per cent mortality of *L. rohita* for 48 hours

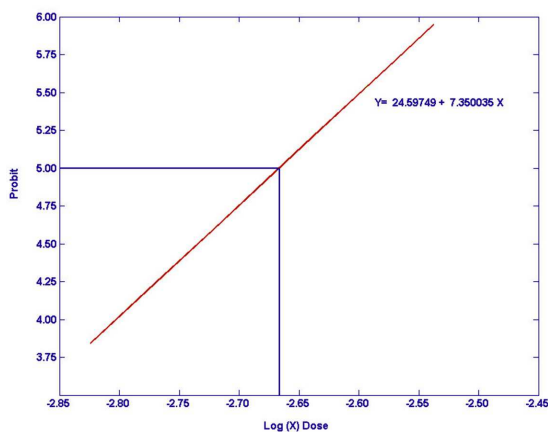


Figure 7. Regression line (based on Probit analysis) of log concentration of pesticide Lambda cyhalothrin Vs Per cent mortality of *L. rohita* for 72 hours

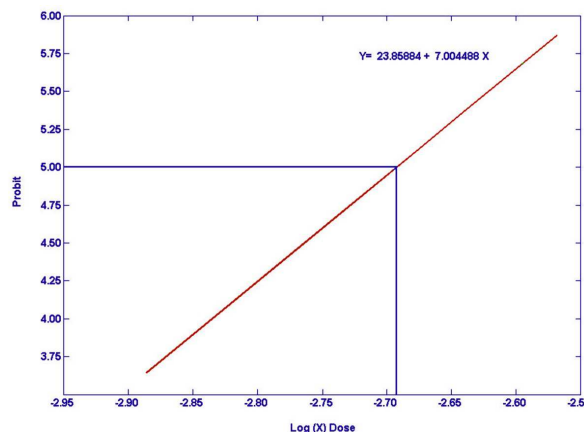


Figure 8. Regression line (based on Probit analysis) of log concentration of pesticide Lambda cyhalothrin Vs Per cent mortality of *L. rohita* for 96 hours

L. rohita was silvery white in body in the control group throughout the experiment. The body colour changed from original silvery white

to dark colour in pesticide treated fish. The fish maintained in freshwater behaved normal as usual. But when the fish was exposed to

pesticides monocrotophos and lambda cyhalothrin, erratic swimming, abnormal posture, disbalance, sluggishness, imbalance in posture, increase in surface activity, opercular movement, gradual loss of equilibrium and spreading of excess of mucus all over the surface of the body were observed.

A survey of LC₅₀ values of different pesticides to the fish for different periods of exposure reveals the occurrence of a wide differences between duration of exposure and types of fishes (Macek and Mc Allister, 1970; Holden, 1972; Carter and Graves, 1973; Bakthavathasalam 1980; Koundinya and Ramamurthi, 1980; Padmini, 1980; Rani *et al.*, 1990; Dhanalakshmi, 1991; Sadhu, 1993; Pickering and Henderson, 1966; Santhakumar and Balaji, 2000; Mathivanan, 2004 and Ramasamy *et al.*, 2007). Changes in body colour have been reported in *Anabas testudineus* after exposure to monocrotophos (Santhakumar and Balaji, 2000), *C. punctatus* to organophosphorus (Sandhu, 1993) and *Cyprinus carpio* to ammonia stress (Israeli-weinstein and Kimmel, 1998). The behavioural changes are considered directly related to complex physiological responses and have often been used as a sensitive indicator of stress (Little and Finger, 1990).

Fish exposed to sub lethal concentrations of pesticides irregular, erratic and darting movements with imbalanced swimming activity and attempt to jump out of the toxic medium were observed. Similar behaviour patterns were observed in fish, trout and *L. rohita* exposed to fenvelrate (Murthy, 1987). Increased opercular movements, loss of equilibrium, erratic swimming and jerky movement and mucous secretion all over the body were observed in *Heteropneustes fossilis* after exposure to rogor and endosulphan pesticides (Borah and Yadav, 1995). Erratic swimming, imbalance in posture, increased surfacing activity with gradual decrease in opercular movement, loss in equilibrium, excess of mucus all over the body surface followed by sluggishness and death of *A. testudineus* after exposure to monocrotophos was reported by Santhakumar and Balaji (2000).

Many workers have observed erratic swimming, equilibrium loss and surfacing phenomenon in the fish following pesticide exposure. Surfacing phenomenon shown by the fish might be to gulp maximum possible air to ease the tension. Rao and Rao (1987) also observed this phenomenon in the fish, *Channa punctatus* exposed to two different pesticides viz., carbaryl and phenthoate. In relation to this they also reported that the surfacing phenomenon was due to hypoxic condition of the fish. Increased opercular movements were seen in the fish, *L. rohita* exposed to pesticides, which was in accordance to the report put forth by Amita kiran and Jha (2009) in *Clarias batrachus* exposed to herbicide, herboclin. The rapid opercular movements may be due to accumulation of mucous over gill due to the toxicant (Sadhu, 1993; Sabita and Yadav, 1995 and Jagadeesan and Vijayalakshimi, 1999). Similar findings were observed by Prasanth *et al.* (2005), when freshwater fish *C. mrigala* exposed to cypermenthrin. The fish *L. rohita* exhibited irregular, erratic darting movements with imbalanced swimming activity. Occasionally the fish tried to jump out of the toxic medium, which shows the avoidance behaviour of the fish to the toxicant. Similar behavioural patterns were observed in *L. rohita* exposed to endosulfan (Shivakumar and David, 2004).

The change of body colour, behavioural changes such as irregular swimming movements, loss of equilibrium, restlessness and excess secretion of mucous suggest that *L. rohita* has undergone chemical stress when exposed to pesticide and the present study could be taken as an indicator of aquatic pollution.

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