



Investigation Of Acute Toxicity Of Synthetic Pyrethroids Fenvalerate In Fish *Oreochromis Mossambicus*

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	Abstract Acute toxicity of synthetic pyrethroids fenvalerate in fish tilapia, <i>Oreochromis mossambicus</i> investigated in the present study. The static renewal test method of acute toxicity was used. The experiments were repeated 3 times and the 96-h LC ₅₀ was determined for the, <i>Oreochromis mossambicus</i> . The data obtained from the acute toxicity test were evaluated using the probit analysis statistical method, developed by USEPA. The 96-h LC ₅₀ value for fenvalerate in fish <i>Oreochromis mossambicus</i> was found to be 30.885 ppb. Behavioural alterations were studied during the exposure period.
CC License CC-BY-NC-SA 4.0	Key Words: Synthetic pyrethroids, Fenvalerate, Tilapia, <i>Oreochromis mossambicus</i> , 96-h LC ₅₀ , Behavioural studies.

INTRODUCTION

Pesticides drained to the aquatic environment are primarily of agricultural origin and which may also stem from effluent from manufacturing plants (Das and Mukherjee, 2000). Synthetic pyrethroids are the newest major class of broad-spectrum organic insecticides, now account for more than 30% of insecticide used worldwide in agriculture, domestic and veterinary applications (Eisler, 1992). Several studies have reported that these compounds are extremely toxic to fish and other aquatic organisms (Bradbury and Coats 1989a; Haya, 1989; Clark 1995). In addition to their acute toxicity, many pyrethroids may have potentially deleterious effects at sublethal levels (Coats et al., 1989; Velmurugan et al., 2006; 2007). Due to their lipophilicity, pyrethroids have a high rate of gill absorption, which in turn would be a contributing factor in the sensitivity of fish to aqueous pyrethroid exposures (Polat et al., 2002).

Fenvalerate, is a relatively recently developed type II synthetic pyrethroid that has replaced other groups of insecticides due to its improved insecticidal potency (WHO, 1990). It combines very high insecticidal activity with moderate to low mammalian toxicity and adequate stability in the field. (Tripathi and Priyanka Verma 2004). Fenvalerate is commonly used for control of biting and sucking insects and mites in cotton, fruit orchards, potatoes, and grain (Leahey, 1985). It is also used in public health sectors and cattle sheds for controlling flies (Battle, 1982; WHO, 1990). Generally, crustaceans, molluscs and fish are more sensitive to fenvalerate followed by amphibians, reptiles, birds and mammals (Mulla et al., 1978; Linden et al., 1979;

Coats and O'Donnell-Jeffrey, 1979; McLeese et al., 1980). Exposure to fenvalerate and tebuconazole exhibits combined acute toxicity in zebrafish and behavioral abnormalities in larvae (Yao et al., 2022).

This study investigates the toxic effects of fenvalerate on tilapia (*Oreochromis mossambicus*), the standard test species pursuant to APHA, AWWA, WEF (1998) and OECD (1993) by the determination of 96-h LC₅₀ values and evaluates behavioral disorders of the fish exposed to different concentrations of the toxicant.

MATERIALS AND METHODS

2.1. Study area and collection of materials

For this experiment, tilapia, *Oreochromis mossambicus*, with an average body weight of $22\text{g} \pm 0.56$ (mean $\pm$ SD) and their length was in the range 13-15 cm, were collected from a freshwater source in the Puzhal lake, Redhills, Chennai. Fish were brought to the laboratory within 30 min in plastic bags with sufficient air. Tilapia, *Oreochromis mossambicus*, was utilized as the model organism in this study, because it is a common fish in the ponds and streams near agriculture areas, most suitable for laboratory studies and is considered one of the most non-target organisms in the ponds and streams surrounded by agriculture areas.

2.2. Experimental design

The fish were acclimated to the laboratory conditions for at least 20 days prior to the experiment in a glass aquarium (150 l) filled with dechlorinated water. Water quality characteristics were determined. The mean values for test water qualities were as follows: temperature $27.5 \pm 1.5^\circ\text{C}$, pH 7.5 ± 0.03 , dissolved oxygen 6.4 ± 0.2 mg/l, alkalinity 250 ± 2.8 mg/L as CaCO₃, total hardness 456 ± 3.5 mg/L. The fish were fed daily with commercially balanced fish food sticks. The fishes were maintained on a photoperiod period with 12 h light/12 h dark.

Commercial grade Fenvalerate (20% EC) (Rallis India Ltd., Mumbai, India.) was used in this study. Fenvalerate is a synthetic pyrethroid insecticide with the Cas, chemical name ((RS)- α -cyano-3-phenoxybenzyl (RS)-2-(4-chloro-phenyl)-3-methyl-butyrate), CAS registry no. 51630-58-1; chemical formula—C₂₅H₂₂ClNO₃

Acute toxicity of fenvalerate to *Oreochromis mossambicus* is determined using a standard static-renewal technique (APHA-AWWA-WPCF, 1998). Test chambers were filled with 20 l of tap water. Groups of experimental animals, each consisting of 10 individuals, were selected at random and placed into aerated aquaria. After 24 h of adaptation, the different concentrations of fenvalerate in distilled water were added to the experimental aquaria. During the last 24 h of adaptation, and throughout the duration of the experiment, animals were not fed. Mortality was assessed at 24, 48, 72 and 96 h after the start of the tests. Dead individuals were removed immediately. The toxicant in the test chambers was renewed completely with fresh solution of same concentration every 24 h. Following the preliminary experiment, all determinations were repeated three times. Behavioral changes were followed closely.

The toxicant used in static bioassays was fenvalerate in tap water. The fish were randomly distributed in different concentrations of fenvalerate. For the acute bioassay tests, 10 fish were used per concentration per replicate. A total of three replicates were carried out for each dose and the control group. The aquaria were not aerated at the time of dosing with fenvalerate. Fenvalerate concentrations administered in the 20 l capacity test tanks were 26, 30, 34, 38, 40, 50 ppb/L. LC₅₀ and 95% confidence limits were calculated by LC₅₀ software program, version 1.00 computer program developed by EPA (US EPA, 1999).

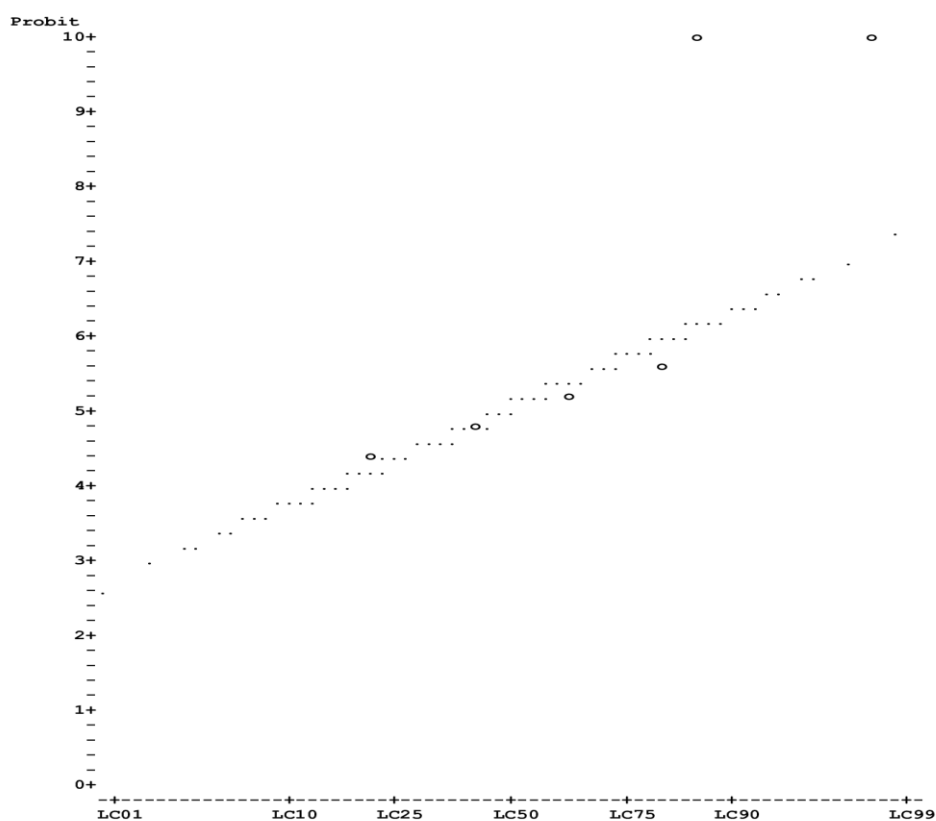
3. RESULTS AND DISCUSSION

Table 1 shows the relation between the fenvalerate concentration and the mortality rate of *Cirrhinus mrigala* according to Finney's Probit Analysis using EPA Computer Program. The calculated 96-h acute LC₅₀ value (95% confidence limits) of commercial grade fenvalerate, using a static bioassay system to *Oreochromis mossambicus* was 30.885 ppb (27.045 - 33.605). Control mortality was zero. The results show that fenvalerate is highly toxic to fish. The estimated LC₅₀ values and confidence limits are listed in Table 1 and Fig 1.

Table 1Acute 96-h toxicity of commercial fenvalerate in *Oreochromis mossambicus*

Point	Concentration (ppb/l)	95% confidence limits	Slope $\pm$ SE	Intercept $\pm$ SE
LC 1.00	18.415	10.056 - 22.686	10.36 $\pm$ 2.65	-10.43 $\pm$ 4.04
LC 5.00	21.426	13.575 - 25.192		
LC 10.00	23.229	15.912 - 26.672		
LC 15.00	24.530	17.696 - 27.744		
LC 50.00	30.885	27.045 - 33.605		
LC 85.00	38.886	35.512 - 47.376		
LC 90.00	41.065	37.113 - 52.441		
LC 95.00	44.518	39.450 - 61.222		
LC 99.00	51.799	43.954 - 82.374		

Note: Control group (theoretical spontaneous response rate) = 0:0000.

FIG 1. PLOT OF ADJUSTED PROBITS AND PREDICTED REGRESSION LINE OF *Oreochromis mossambicus* EXPOSED TO FENVALERATE

Observations of the behavioral responses of the *Oreochromis mossambicus* were the loss of equilibrium, rapid gill movement, erratic swimming, swimming at the water surface and “gulping for air,” hanging and swimming vertically in water, convulsions and prolonged and motionless laying down on the aquarium bottom, abnormal swimming behavior increased and the fish were observed to hit the aquarium walls. Additionally, increased mucus secretion and paleness of the fish colour was obvious in all these concentrations. The behavioural results of the present study agree with similar observations made for some pyrethroid compounds by other workers (US EPA, 1999; Edwards et al. 1986; Polat et al. 2002; Baser et al. 2003; Amit Kumar et al., 2007).

The 96-h LC₅₀ value of fenvalerate in *Oreochromis mossambicus* was found as 30.885 ppb in the present work and here we report fenvalerate to be potentially toxic to fish. Fenvalerate is widely produced and used in agriculture, particularly in developing countries. Over the last 20 years, large amounts of formulated fenvalerate, approximately 1000 metric tons per year, have been used as an agricultural insecticide (WHO, 1990). In runoff studies monitoring the pyrethroid insecticide fenvalerate, values between 0.5 and 39.7 mg/L (total concentration of water/sediment mixture) were measured (Smith *et al.* 1983). River sediments have been shown to contain fenvalerate concentrations between 0.6 and 3.6 mg/kg (House *et al.*, 1991), while estuary sediments contained levels up to 100 mg/kg (Chandler, 1990).

Unlike most mammals where pyrethroids have a short life and are readily metabolized, fish are reported to be deficient in the enzymes that hydrolyze pyrethroids (Haya, 1989). Additionally, comparative *in vivo* and *in vitro* metabolic studies have shown that fish have a lower capacity to metabolize and eliminate pyrethroid insecticides (Glickman and Lech, 1981; Glickman *et al.*, 1982).

The toxic effects of fenvalerate have been extensively studied with osmoregulation, (Sivaram Prasad *et al.*, 1995); invertebrates, (Woin, 1998); chromosome aberrations, (Arokia Rita and Selvanayagam 1998, Giri *et al.*, 2002); hematology, (Seth and Saxena 2003); toxicity and residual effects, (Tilak *et al.*, 2003); biochemistry, (Tripathi and Priyanka Verma 2004); acetylcholinesterase activity, (Mushigeri and David 2005), oxidative damages (Prasanthi *et al.*, 2005).

The LC₅₀ values reported for fenvalerate are 3.8 ppb, 7.6 ppb for 24 hours for *Salmo gairdneri* (Mulla *et al* 1978; Coats & O'Donnell-Jeffery 1979), 3 ppb for 48 hours in *Salmo gairdneri*, 35 and 24 ppb for 24 hours and 48 hours in *Cyprinodon macularis* (Mulla *et al* 1978), 0.14, 0.77, 0.68 mg/h for 24, 48 and 72 hours in *Pimephales promelas* (Bradbury *et al* 1985). The 48 h LC₅₀ values of fenvalerate to *Cyprinus carpio* was 0.03 ppm (Malla Reddy and Basha Mohideen 1988). The LC₅₀ value for fenvalerate was estimated as 19 ppb (Tripathi and Priyanka Verma, 2004). The LC₅₀ of 15 ppb for commercial grade fenvalerate is in agreement with reports of Worthing and Walker (1987), Bradbury *et al.* (1987a), Radaiah *et al.* (1989), and Radaiah and Rao (1990). LC₅₀ of fenvalerate in fish has been reported to vary from 0.69 to 5.4 ppb (Holcombe *et al.*, 1982; Bradbury *et al.*, 1985). These differences in LC₅₀ values may be mainly due to differing species, exposure time and grade of pesticide used (Tripathi and Priyanka Verma, 2004). The 96 h LC₅₀ value of fenvalerate for the fish, *C. mrigala* was found to be 6 µg/l (Mushigeri and David. 2005). The 96-h LC₅₀ value for fenvalerate in fish *Cyprinus carpio* was found to be 3.059 ppb Raja *et al.*, 2010). Adult fish, the acute toxicity of fenvalerate to zebrafish larvae is higher than tebuconazole, significant differences in the 96 h-LC₅₀ values between two life stages of zebrafish were discovered, with adults being more sensitive (Yao *et al.*, 2022). In the present study the

96-h LC₅₀ value of fenvalerate in *Oreochromis mossambicus* was found as 30.885 ppb. Fish are sensitive to fenvalerate with 96-hour LC₅₀'s of 0.64 ppb for bluegill, 0.81 ppb for channel catfish and 6.2 ppb for rainbow trout. The sheepshead minnow is somewhat more resistant with a 96-hour LC₅₀ of 430 ppb. Pink shrimp and fiddler crabs have LC₅₀'s of 1.4 ppb and 53.0 ppb, respectively (URL: 1).

Fenvalerate is a highly toxic synthetic pyrethroid widely used in agriculture. Synergistic interactions between the active ingredient and other components of the formulation should be taken into consideration when evaluating toxicity. In spite of these potential risks studies on fenvalerate in aquatic ecosystems are highly limited. The result obtained in this study clearly reveals the fact that it is necessary to carry out more toxicity studies.

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