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Investigation of acute toxicity of deltamethrin on guppies (*Poecilia reticulata*)

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Abstract

Deltamethrin, a synthetic pyrethroid pesticide contaminating aquatic ecosystems as a potential toxic pollutant, was investigated in the present study for acute toxicity. Guppy fish (*Poecilia reticulata*) were selected for the bioassay experiments. The 48-h LC_{50} was determined for the guppies. The experiments were repeated three times and a total of 210 guppies were used. The static test method of acute toxicity test was used. In addition, behavioral changes at each deltamethrin concentration were determined for the individuals. Data obtained from the deltamethrin acute toxicity tests were evaluated using the Probit Analysis Statistical Method. The 48-h LC_{50} value for guppy was estimated as 5.13 μg/L.

Keywords: Deltamethrin; Acute toxicity; Guppy; *Poecilia reticulata*; Pyrethroid; Behavioral effects

1. Introduction

Synthetic analogs of the pyrethrins, extracts from the ornamental *Chrysanthemum cineraiaefolium*, have been developed to circumvent the rapid photodegradation problem encountered with the insecticidal natural pyrethrins. The pyrethroids are widely used in field pest control and household use and as veterinary and human pediculicides and are among the most potent insecticides known (Smith and Stratton, 1986). The widespread use of these pesticides consequently leads to the exposure of manufacturing workers, field applicators, the ecosystem, and finally the public to the possible toxic effects of these pesticides.

Pyrethroids have been reported to be extremely toxic to fish and some beneficial aquatic arthropods, for example, lobster and shrimp (Bradbury and Coats, 1989; URL 1; Srivastav et al., 1997). Bradbury and Coats (1989) also reviewed pyrethroid toxicology in mammals, birds, amphibians, and both terrestrial and aquatic invertebrates. Toxicity is highly dependent on stereochemical structure. Most products however, are mixtures of isomers. Several larvicides and adulticides, including resmethrin and permethrin, were evaluated for toxicity to standard test (in-house cultures) and resident organisms to measure effects of mosquito control pesticides to nontarget organisms (Milam et al., 2000). Pyrethroids are especially adventageous for use in northern climate zones, since they exhibit a negative temperature coefficient of toxicity. They are also considered relatively nonpersistent and therefore are not expected to biomagnify through the food chain. Maximum bioconcentration factors ranged from 698X for whole fish (deltamethrin) to 6090X (bifenthrin) (URL 2). Toxic effects of pyrethroids on nontarget organisms have been reviewed and reported to be in the parts per billion value of toxicity (Smith and Stratton, 1986). In both the laboratory and field, adsorption of pyrethroids substantially reduces toxicity. Deltamethrin has been classified "immobile" by the US EPA (URL 2). Therefore, in the field, most of the affected organisms show rapid recovery. The environmental fate and effects of synthetic pyrethroid insecticides have been summarized (Hill, 1989).

Due to their lipophilicity, pyrethroids have a high rate of gill absorption, which in turn would be a contributing factor in the sensitivity of the fish to aqueous pyrethroid exposures. Fish seem to be deficient in the enzyme system that hydrolyzes pyrethroids. The main reaction

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involved in the metabolism of deltamethrin, cypermethrin, or cyhalothrin in mice and rats is ester cleavage mainly due to the action of carboxyesterase. Metabolism in fish is largely oxidative (Demoute, 1989). Fish make intimate contact with the surrounding water through the gills. After short-term deltamethrin exposure, adult *Heteropneustes fossilis* (freshwater catfish) showed hypocalcemia and the researchers attribute this condition to the possible impairment of either net electrolyte influx at the gill or renal function. Deltamethrin exposure also caused hypophosphatemia and was linked to the possible redistribution of electrolytes between intracellular or extracellular compartments and/or impairment of renal function. Deltamethrin may disturb the calcium and phosphate homeostasis and may lead to an effect on the reproductive state of the fish (Srivastav et al., 1997).

Synthetic pyrethroids have been shown to be toxic for fish, aquatic arthropods, and honeybees in laboratory tests. Acute toxicity data for deltamethrin in fish have been summarized in a report of the World Health Organization (WHO, 1990) and classified as highly toxic to fish, being in the $LC_{50} < 1.0$ ppb. The potential hazard to fish is due to its heavy use in many aquatic larvicidal programs. Synergistic interactions between the active ingredient and other components of the formulation should be taken into consideration when evaluating toxicity.

This study was conducted to determine the acute toxicity of deltamethrin, a synthetic pyrethroid pesticide, to the guppy (*Poecilia reticulata*) using the static test system.

2. Materials and methods

Male, adult guppies were obtained from a local breeder in Ankara and brought to the laboratory within 30 min in plastic bags with sufficient air. The plastic bags were placed into the maintenance aquarium for about 30–35 min for acclimatization. Then the bags were cut open and the fish were allowed to swim into the aquarium water. Test chambers were glass aquaria of about a 25-L capacity. Temperature was regulated at $22° \pm 1°C$ by using heaters. Except for dosing instance, all aquaria were aerated.

Test chambers were filled with 20 L of tap water. Some characteristics of this aquarium water were, temperature, $22° \pm 1°C$, dissolved oxygen, 7.2–7.9 mg/L, and conductivity, 0.212–0.260 mS.

Following the preliminary experiment, all determinations were repeated three times. Groups of experimental animals, each consisting of 10 individuals, were selected at random and placed into aerated aquaria. After 48 h of adaptation, the different concentrations of deltamethrin 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 controlled 24, 48, 72, and 96 h after the start of the tests. Dead individuals were removed immediately. Behavioral changes were followed closely.

Technical grade (98%) deltamethrin was from the Insecticide Testing Laboratory of Hacettepe University, Ankara. Deltamethrin stored at $+4°C$ was prepared by weighing a certain amount and diluting it in acetone to give the stock material. Dosing solutions were prepared from this stock by diluting with acetone to give the dosing concentrations of 1.00, 4.00, 6.60, 7.56, 8.64, and 10.8 μg/L. The dosing volume never exceeded 0.2 mL. Control group received acetone at the maximum acetone volume used in the dilution of the dosing concentrations. The bioassay system was as described in standardized methods (APHA, AWWA, WEF, 1998; OECD, 1993) and the national regulation (Turkish Official Gazette, 1991). The LC_{50} and 95% confidence limits were calculated by a computer program (US EPA, 1999).

3. Results

The calculated 48-h acute LC_{50} value (95% confidence limits) of technical deltamethrin, dissolved in acetone, using a static bioassay system to adult, male guppies *P. reticulata* was 5.13 μg/L (3.3263–6.6955). Control mortality was zero. The results show that deltamethrin is highly toxic to fish. The selected species is as recommended by the reference/standard methods (APHA, AWWA, WEF, 1998; OECD, 1993) and Turkish national regulation (Turkish Official Gazette, 1991). Results appear in Table 1.

Observations of the behavioral responses of the guppies were conducted at 1–8 and every 12 h during the acute toxicity tests. The control group showed normal behavior during the test period. The changes in behavioral response started 1 h after dosing in all deltamethrin concentrations tested. In guppies, 1.00 μg/L (lowest) concentration had close to normal behavior. At 4.00 μg/L, less general activity was recorded when compared with the control group and loss of equilibrium was observed.

The responses of loss of equilibrium and hanging vertically in the water were observed at all concentrations above 4.00 μg/L. The highest concentration (10.80 μg/L) showed all responses at high levels and the onset was within the first hour after dosing with deltamethrin. Rapid gill movement, erratic swimming, swimming at the water surface and "gulping for air," and prolonged and motionless laying down on the aquarium bottom were other responses observed at all concentrations above 4.00 μg/L.

Table 1
Acute 48-h toxicity of technical deltamethrin in adult male guppies (*Poecilia reticulata*)

Point	Concentration (μg/L)	95% confidence limits	Slope ± SE	Intercept ± SE
LC 1.00	0.7937	0.0563–1.7053	2.871790 ± 0.822359	2.961881 ± 0.668930
LC 5.00	1.3706	0.1936–2.4455		
LC 10.00	1.8341	0.3726–2.9748		
LC 15.00	2.2326	0.5780–3.4047		
LC 50.00	5.1251	3.3263–6.6955		
LC 85.00	11.7650	8.5308–29.5437		
LC 90.00	14.3211	9.8530–45.4131		
LC 95.00	19.1638	12.0724–86.7609		
LC 99.00	33.0945	17.4151–296.4663		

Note. Control group (theoretical spontaneous response rate) = 0.0000.

4. Discussion

The 48-h LC_{50} value of deltamethrin in guppies was found as 5.13 μg/L in the present work and here we report deltamethrin to be highly toxic to fish. The USDA National Agricultural Pesticide Impact Assessment Program's EXTOXNET document (URL 1) reports deltamethrin acute toxicity to fish in laboratory tests to be in the average range LC_{50} value of 1–10 μg/L. Mittal et al. (1994) reported deltamethrin toxicity to *P. reticulata* as the most toxic of the pyrethroids studied: $LC_{50} = 0.016$ ppm. Our results are in good agreement with the reports of other investigators using fish species (Smith and Stratton, 1986; Bradbury and Coats, 1989; WHO, 1990; Mestres and Mestres, 1992; Golow and Godzi, 1994; URL 1).

Mestres and Mestres (1992) report 96-h fish LC_{50} values as follows: *Salmo gairdneri*, 0.39 μg/L; *Cyprinus carpio*, 1.84 μg/L; and *Sarotherodon mossambica*, 3.50 μg/L. Golow and Godzi (1994) report 96-h fish LC_{50} value for *Oreochromis niloticus* (L., 1757) fingerlings as 14.50 μg/L. They concluded that deltamethrin was two times more toxic to the fish species than dieldrin. Bradbury and Coats (1989) have reviewed the toxicology of pyrethroids in mammals, birds, fish, amphibians, and invertebrates (terrestrial and aquatic) and cited deltamethrin toxicity to Atlantic salmon (*Salmo salar*), mosquitofish (*Gambusia affinis*), and rainbow trout as 96-h LC_{50} values of between 0.50 and 1.97 μg/L. The US EPA states deltamethrin's bioconcentration factor as 698X for whole fish (URL 2). Although under field conditions deltamethrin is considered to pose less risk due to its high adsorption to soil, these data should be considered when assessing possible/potential ecosystem risks.

The effect of deltamethrin on the sensitive early life stages of *Brachydanio rerio* (Hamilton, 1822), zebrafish, were examined by Görge and Nagel (1990). The development of larvae was influenced by deltamethrin. Ontogenesis was also impaired. Hatchability of embryos was reduced in a dramatic way at 0.80 μg/L and higher. The calculated LC_{50} values for deltamethrin at 35 days (95% confidence limits) was 0.52 (0.46–0.58) μg/L.

Deltamethrin was inhibitory to the monooxygenase system of carp liver (*Cyprinus carpio* L.) at the higher concentration of 2 μg/L. The low concentration of 0.2 μg/L resulted in faster metabolism of deltamethrin and was evaluated as induction of hepatic microsomal cytochrome P450-dependent monooxygenases (Deér et al., 1996).

It is interesting to note that only a few studies on the acute toxicity of one of the most toxic pyrethroids, namely deltamethrin, to fish exist in the open literature.

5. Conclusion

Deltamethrin is a highly toxic synthetic pyrethroid pesticide widely used in agriculture. Here special attention is drawn to its heavy use in mosquito control programs, which necessitates in-depth subchronic and chronic toxicity tests to fish species and to nontarget species to be undertaken. In addition, potential risk from deltamethrin metabolites should be investigated to get a more complete picture in terms of toxicity. The low toxicity of deltamethrin to mammals may be misleading at this point in terms of ecotoxicology and lead to extrapolation problems to aquatic species. Delistraty (2000), in the study of examining relationships among physicochemical properties and acute toxicity endpoints of 231 chemicals in rats and trout, concluded that trout aquatic LC_{50} was predicted from rat inhalation LC_{50} with moderate success. Therefore, such data are useful in ecological risk assessment but there are limitations and uncertainties. Further work with toxicity testing methods directly on fish will be very useful in assessing possible ecological risk assessment of these pesticides. To overcome discrepancies and potential synergistic effects from the components of the pyrethroid formulations, toxicity tests with formulations must be included together with active ingredient tests. Using only the pyrethroid active ingredient in the tests is insufficient.

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