Investigation of LC50, NOEC, and LOEC of Oxadiazon, Deltamethrin, and Malathion on Platy Fish (*Xiphophorus Maculatus*)

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ABSTRACT

Background: The presence of pesticides is very prevalent in surface waters of Iran due to their huge consumption for agricultural purpose. These toxic substances may accumulate in the food chain and cause serious ecological and health problems. Chemical pesticides with persistent molecules (long half-life periods) pose a threat to aquatic life forms and the human population consuming the affected fish.

Methods: Fish samples were exposed to different concentrations of oxadiazon 25% (0, 2, 4, 8, 16 and 32 ppm), deltamethrin 2.5% (0, 0.02, 0.05, 0.10, 0.20, and 0.30 ppm), and malathion 57% (0, 5, 10, 15, 20, and 30 ppm) within a 120 L capacity glass aquaria for 96 h. Their cumulative mortality of platy fish was calculated with a 24-hour interval.

Results: $LC_{50\text{-}96h}$ was 7.59±0.42, 0.11±0.46, and 12.05±0.75 for oxadiazon, deltamethrin, and malathion, respectively. The very low LC_{50} obtained for oxadiazon (7.59±0.42 ppm), deltamethrin (0.11±0.46 ppm), and malathion (12.05±0.75 ppm) indicates that oxadiazon, deltamethrin and malathion are highly toxic to platy fish.

Conclusion: The results of this study demonstrate that deltamethrin and malathion had the lowest and highest rate of mortality on the platy fish, respectively.

Keywords: Deltamethrin, LC_{50}, Malathion, Oxadiazon, Platy Fish.

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INTRODUCTION

There is a growing concern over aquatic pollution because of its detrimental effects on biological life, including human beings. Increased use of pesticides results in contamination of natural ecosystems, especially the aquatic ecosystem [1]. These toxic substances may accumulate in the food chain and cause serious ecological and health problems. Chemical pesticides with persistent molecules (long half-life) pose a threat to aquatic life forms and human populations consuming the affected fish. Presence of pesticides in surface waters has been reported in Europe and North America for nearly 70 years. Since then, many documents have proven the toxic effects of these pollutants on aquatic environments [2, 3].

Deltamethrin is a pyrethroid insecticide. Pyrethroids are synthetic compounds made to mimic the pyrethrins that are isolated from chrysanthemum flowers. Deltamethrin is a broad-spectrum insecticide that works by interfering with a nerve cell's ability to send a normal signal by jamming open tiny gates on the cell that need to open and close rapidly to carry the message. Deltamethrin can be found in a wide variety of products used in farms, gardens, lawns, indoors, and even in those used for pets [4, 5].

Oxadiazon is the active ingredient in the herbicide ronstar, intended for early post-emergence application. When applied to soil during these phases, oxadiazon controls the growth of certain undesirable weeds, such as broadleaves, grasses, sedge, brush vines, and bramble [6]. Acute toxicity of a pesticide refers to its ability to cause damage to an animal from a single exposure, generally of short duration. A number of studies have used acute toxicity tests of pesticides on fish to acquire rapid estimates of the concentrations that cause direct, irreversible harm to the tested organisms [7, 8]. Malathion is an organophosphate parasympathomimetic which binds irreversibly to cholinesterase [9]. It

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is an insecticide of relatively low human toxicity. In the former USSR, malathion was known as carbophos, in New Zealand and Australia it is known as maldison, and in South Africa it is called mercaptothion [10]. Malathion is a pesticide that is widely-used in agriculture, residential landscaping, public recreation areas, and public health pest control programs, such as mosquito eradication. In the US, it is the most commonly used organophosphate insecticide [11, 12]. The present study was performed to determine the acute toxicity of oxadiazon, deltamethrin, and malathion as potential dangerous organic pesticides to assess the mortality induced by these chemicals in the freshwater platy fish.

MATERIALS AND METHODS

In the present study, the selected fish species was platy fish. Test chambers were glass aquaria with 120 L capacity. All fishes were acclimated for a week in these aquaria before assays with continuous aeration; water temperature was regulated at 25^0C by using aquarium heater. The fishes were fed twice per diem with formulated feed and the dead fishes were immediately removed to avoid possible water deterioration [13].

Nominal concentrations of active ingredients tested were 0, 2, 4, 8, 16, and 32 ppm of commercial dose (25%) for oxadiazon, 0, 0.02, 0.05, 0.10, 0.20, and 0.30 ppm of commercial dose (2.5%) for Deltamethrin, and 0, 5, 10, 15, 20, and 30 ppm of commercial dose (57%) for malathion. Fifteen groups (five for oxadiazon, five for deltamethrin, and five for malathion) of seven platy fish were exposed for 96h in aerated glass aquaria of the test medium. During the experiment, the water in each aquarium was aerated and the temperature was kept at 25^0C. No food was provided to the specimens during the assay and the test media was not renewed. Mortality rates were recorded during 0, 24, 48, 72, and 96 h of exposure. Acute toxicity tests were carried out according to an earlier study [14]. The nominal concentration of oxadiazon, deltamethrin, and malathion estimated to result in 50% mortality of platy fish within 24 h (24-h LC_{50}), 48 h, 72 h, and 96 h was attained by probit analysis using Finney's method and the maximum-likelihood procedure (SPSS 2002, SPSS Inc., Chicago, Illinois, USA). The LC_{50} value is obtained by fitting a regression equation arithmically and also by graphical interpolation by taking logarithms of the oxadiazon, deltamethrin and malathion concentrations versus the probit value of percentage mortality. After the acute toxicity test, the LOEC (Lowest Observed Effect Concentration) and NOEC (No Observed Effect Concentration) were determined for each measured endpoint [14].

RESULTS

No fish died during the acclimation period before exposure. Moreover, no control fish died during acute toxicity tests. The mortality of platy fish at 0, 2, 4, 8, 16, and 32 ppm doses for oxadiazon; 0, 0.02, 0.05, 0.10, 0.20, and 0.30 ppm doses for deltamethrin; 0, 5, 10, 15, 20, 30 ppm doses for malathion were determined during the exposure times at 24, 48, 72 and 96 h (Tables 1-3). The mortality of platy fish increased significantly with increasing concentrations from 2 ppm to higher concentrations for oxadiazon and 0.02 ppm to higher concentrations for deltamethrin, and 5 ppm to higher concentrations for malathion.

Table 1. Cumulative mortality of platy fish (n=7 each concentration) exposed to oxadiazon acutely.

Concentration (ppm)	No. of mortality			
	24h	48h	72h	96h
Control	0	0	0	0
2	0	0	0	0
4	1	2	3	3
8	2	4	4	5
16	5	6	6	6
32	7	7	7	7

Table 2. Cumulative mortality of platy fish (n=7 each concentration) exposed to deltamethrin acutely.

Concentration (ppm)	No. of mortality			
	24h	48h	72h	96h
Control	0	0	0	0
0.02	0	0	0	0
0.05	1	1	1	2
0.10	2	2	3	3
0.20	3	4	5	6
0.30	7	7	7	7

Table 3. Cumulative mortality of platy fish (n=7 each concentration) exposed to malathion acutely.

Concentration (ppm)	No. of mortality			
	24h	48h	72h	96h
Control	0	0	0	0
5	0	0	0	0
10	2	3	3	3
15	4	4	5	6
20	5	6	6	6
30	7	7	7	7

Median lethal concentrations of 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, and 90% test are presented in Tables 4-6. The mortality (or survival) data are collected for each exposure concentration in a toxicity test at various exposure durations (24, 48, 72, or 96 hours). Therefore, the data can be plotted in other ways; the straight line of best fit is then drawn through the points. These are time–mortality lines. LT_{50} (median lethal survival time) can be estimated for each concentration.

Table 4. Lethal concentrations (LC_{10-99}) of oxadiazon depending on exposure time (24-96h) for platy fish.

Point	Concentration (ppm) (95 % of confidence limits)			
	24h	48h	72h	96h
LC_{10}	4.63±0.53	2.39±0.46	1.25±0.42	0.89±0.42
LC_{20}	7.19±0.53	4.48±0.46	3.65±0.42	3.19±0.42
LC_{30}	9.03±0.53	6.09±0.46	5.38±0.42	4.85±0.42
LC_{40}	10.61±0.53	7.47±0.46	6.86±0.42	6.27±0.42
LC_{50}	**12.08±0.53**	**8.76±0.46**	**8.24±0.42**	**7.59±0.42**
LC_{60}	13.56±0.53	10.05±0.46	9.62±0.42	8.91±0.42
LC_{70}	15.14±0.53	11.43±0.46	11.10±0.42	10.33±0.42
LC_{80}	16.98±0.53	13.05±0.46	12.83±0.42	11.99±0.42
LC_{90}	19.55±0.53	15.29±0.46	15.23±0.42	14.29±0.42
LC_{99}	25.631±0.53	20.61±0.46	20.11±0.42	19.75±0.42

Table 5. Lethal concentrations (LC_{10-99}) of deltamethrin depending on exposure time (24-96h) for platy fish.

Point	Concentration (ppm) (95 % of confidence limits)			
	24h	48h	72h	96h
LC_{10}	0.06±0.49	0.06±0.49	0.04±0.48	0.03±0.46
LC_{20}	0.10±0.49	0.09±0.49	0.08±0.48	0.06±0.46
LC_{30}	0.12±0.49	0.12±0.49	0.10±0.48	0.08±0.46
LC_{40}	0.15±0.49	0.14±0.49	0.12±0.48	0.10±0.46
LC_{50}	**0.17±0.49**	**0.16±0.49**	**0.13±0.48**	**0.11±0.46**
LC_{60}	0.19±0.49	0.18±0.49	0.15±0.48	0.13±0.46
LC_{70}	0.21±0.49	0.20±0.49	0.17±0.48	0.15±0.46
LC_{80}	0.24±0.49	0.22±0.49	0.19±0.48	0.17±0.46
LC_{90}	0.28±0.49	0.26±0.49	0.23±0.48	0.20±0.46
LC_{99}	0.37±0.49	0.34±0.49	0.30±0.48	0.26±0.46

Table 6. Lethal concentrations (LC_{10-99}) of malathion depending on exposure time (24-96h) for platy fish.

Point	Concentration (ppm) (95 % of confidence limits)			
	24h	48h	72h	96h
LC_{10}	7.36±0.73	6.41±0.73	6.10±0.70	5.85±0.75
LC_{20}	9.97±0.73	8.81±0.73	8.37±0.70	7.98±0.75
LC_{30}	11.85±0.73	10.54±0.73	10.01±0.70	9.51±0.75
LC_{40}	13.45±0.73	12.02±0.73	11.41±0.70	10.82±0.75
LC_{50}	**14.96±0.73**	**13.40±0.73**	**12.72±0.70**	**12.05±0.75**
LC_{60}	16.46±0.73	14.78±0.73	14.03±0.70	13.27±0.75
LC_{70}	18.06±0.73	16.26±0.73	15.43±0.70	14.59±0.75
LC_{80}	19.94±0.73	17.99±0.73	17.09±0.70	16.12±0.75
LC_{90}	22.55±0.73	20.39±0.73	19.34±0.70	18.25±0.75
LC_{99}	28.74±0.73	26.08±0.73	24.74±0.70	23.31±0.75

Toxicity Testing Statistical Endpoints included: a. Hypothesis Testing: Is there a statistically significant difference between the mean response in the treatments and the mean response in control or reference sample? (LOEC: Lowest Observed Effect Concentration; NOEC: No Observed Effect Concentration), and 2. Point Estimates: What toxicant concentration will cause a specific effect on the test population? (LC_{50}: the Median Lethal Concentration). Acute toxicity testing statistical endpoints of oxadiazon, deltamethrin and malathion for platy fish after exposure during different times are shown in Figures 1, 2, and 3, respectively.

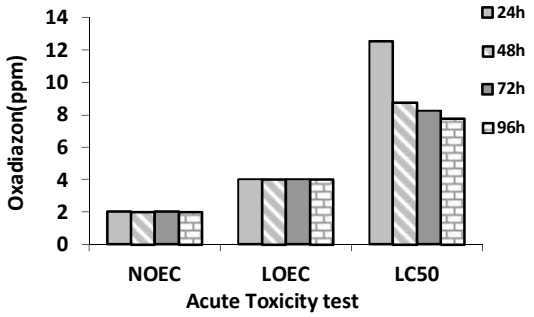

Figure 1. Acute toxicity testing statistical endpoints in platy fish exposed to oxadiazon at different times (24, 48, 72, and 96 h, respectively).

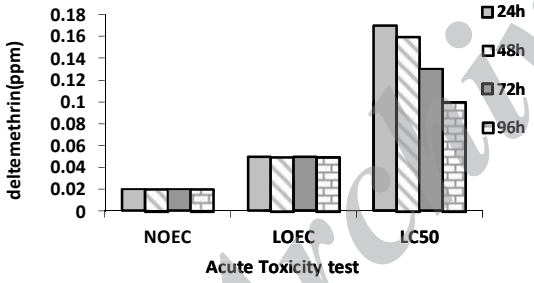

Figure 2. Acute toxicity testing statistical endpoints in platy fish exposed to deltamethrin at different times (24, 48, 72, and 96 h, respectively).

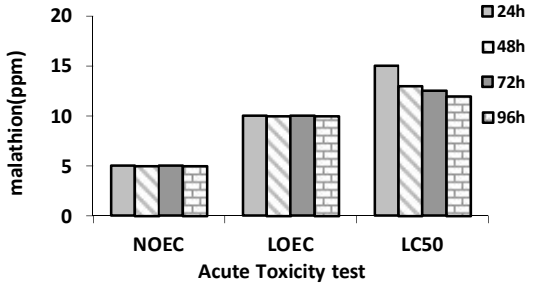

Figure 3. Acute toxicity testing statistical endpoints in platy fish exposed to malathion at different times (24, 48, 72, and 96 h, respectively).

DISCUSSION

Even though chemical pesticides are target specific and effective, their impact on the environment is mostly deleterious. Thus, new pesticides are developed to replace deleterious chemical pesticides. Plant-based pesticides contain active agents with short half-life period and their effects on the environment are not very detrimental [1]. Increased use of chemical pesticides results in the excess inflow of toxic chemicals, mainly into the aquatic ecosystem. The aquatic flora and fauna are affected by the toxic substances which eventually enter into their systems or bring about external damages [11]. Several species of fish are susceptible to deleterious effects when exposed to pesticides and other environmental stressors [3].

The results of the present study indicate that oxadiazon, deltamethrin, and malathion varied in their acute toxicity to platy fish. The toxicity of oxadiazon, deltamethrin, and malathion in platy fish increased with increases in concentration and exposure time.

Presence of pesticides at high concentrations in agricultural wastewaters and their toxicity to aquatic organisms, especially fish species, have been reported by many researchers [15]. Contamination of aquatic environments with pesticides via rainfall runoff is highly possible [16]. Fishes are sensitive to aquatic contamination and serious concerns remain due to their potential adverse effects on human and wildlife populations. In addition, in this study, oxadiazon, deltamethrin and malathion were lethal substrates to platy fish.

Previous studies indicate the high toxicity of deltamethrin to fish species. The results of this study are in good agreement with these reports. Boateng and colleagues reported that young fish were more susceptible and different species respond differently to concentrations of chemicals [17]. Mittal et al. estimated deltamethrin toxicity to *P. reticulate* to be LC_{50}=0.016 ppm [18]. Larkin and colleagues reported the LC_{50} value of deltamethrin in *Clarias gariepinus* to be 5.13 mg/L [6]. Mestres *et al.* found 96-h fish LC_{50} values as follows: *Salmo gairdneri*, 0.39 mg/L; *Cyprinus carpio*, 1.84 mg/L; *Sarotherodon mossambica*, 3.50 mg/L [19]. LC_{50} value of deltamethrin in Tilapia, *Oreochromis niloticus*, was reported to be 15.47 µg/l by Boateng and colleagues [17].

Although deltamethrin is thought to be less toxic in field conditions due to its adsorption in sediments, these data are useful in potential ecosystem risk assessment [6]. Fishes are sensitive to aquatic contamination and serious concerns remain due to their potential to cause adverse effects on human and wildlife populations.

The toxicity of malathion on platy fish (*Xiphophorus maculatus*) increased with increasing concentration and exposure time. Using a variety of methods to detect the acute and chronic toxicity of malathion by preparing various water makes comparisons between fish species difficult. For example, 96 h LC_{50} values were reported to be 6.84 ppm for the *Abramis brama* [10], 10.20 ppm for *Acipenser persicus* [11], and 8.41 ppm for the *Cyprinus carpio* [12].

The 96 h LC_{50} values of oxadiazon for different fishes are reported from tenths to several tens of mg l^{-1} [20]. Values of oxadiazon 96h LC_{50} were 6.42 mg l^{-1} for *Acipenser nudiventris* and 8.85 mg l^{-1} for *Acipenser persicus* [21].

CONCLUSION

The results of this study demonstrated that deltamethrin and malathion had the lowest and highest rates of mortality in platy fish, respectively. However, the rate of mortality increased with increases in the concentration of toxins and exposure time.

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