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Investigation of Acute Toxicity of Two Pesticides Diazinon and Deltamethrin, on Blue Gourami, *Trichogaster trichopterus* (Pallus)

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Abstract: There is a growing concern over aquatic pollution because of its detrimental effects on biological life including human beings. The goal of the present study was to determine the acute toxicity of diazinon and deltamethrin as potential dangerous organic pesticides to assess mortality effects of these chemicals to the blue gourami, *Trichogaster trichopterus* in the form of LC50. Fish samples (7 fish in each test group) were exposed to different concentrations of diazinon and deltamethrin (between0-50ppm for diazinon and 0-0.40ppm for deltamethrin) for 96 h in 120 l glass aquaria. The very low LC50s obtained for deltamethrin (0.223 ± 0.07ppm) and diazinon (14.5 ± 0.91ppm) indicate that deltamethrin and diazinon are highly toxic to *Trichogaster trichopterus*. Further researches are recommended to study the processes by which these chemicals affect physiology and histology of fish and their accumulation in fish tissues.

Key words: LC_{50} · Blue gourami · Diazinon · Deltamethrin · Toxicity

INTRODUCTION

There is a growing concern over aquatic pollution because of its detrimental effects on biological life including human beings [1]. Chemical pesticides with persistent molecules (long half-life periods) pose a threat to aquatic life forms and also to the human population consuming the affected fish.

Presence of pesticide in surface waters was reported in Europe and North America since 50 years ago and since then many documents have been proved the toxic effects of these pollutants to aquatic environment [2-5].

Organophosphorus pesticides (OPs) are widely used in agriculture and the aquatic environment near to fields is under influence of OPs such as diazinon [O,O-diethylO-(2-isopropyl-4-methyl-6-pyrimiinyl) phosphorothioate] [4].

Diazinon is an organophosphorus pesticide extensively used in agriculture and possesses moderately persistence constitution [6,7].

The toxicity of diazinon is due to blocking of acetyl cholinesterase (AChE) activity, which causes harmful impacts on non-target aquatic species close to agricultural fields [7].

The pyrethroids including deltamethrin are largely used as pediculicides and are among the most potent insecticides known [8, 9]. Pyrethroids have been proved to be extremely toxic to fish and some aquatic arthropods, such as shrimps [9-11]. The toxicity of Pyrethroids on amphibians, birds and mammals have been reviewed by Bradbury and Coats [10].

Acute toxicity of a pesticide refers to the chemical's ability to cause damage to an animal from a single exposure, generally of short duration. Many workers have been used the acute toxicity tests of pesticides on fish to acquire rapid estimates of the concentrations that cause direct, irreversible harm to test organisms [12,13].

The acute toxicity data provide useful information to identify the mode of action of a substance and also help to comparison of dose response among different chemicals. The 96-h LC50 tests are conducted to assess the vulnerability and survival potential of organisms to particular toxic chemicals. Chemicals with lower LC50 values are more toxic because lower concentrations results 50% of mortality in organisms.

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The present study was performed to determine the acute toxicity of diazinon and deltamethrin as potential dangerous organic pesticides to assess mortality effects of these chemicals to the freshwater fish blue gourami, *Trichogaster trichopterus*.

MATERIALS AND METHODS

The selected fish species for present study was blue gourami, *Trichogaster trichopterus*. Lethal experiments were conducted using 70 young blue gouramies. Test chambers were glass aquaria of 120 l. All fish were acclimated for a week in these aquaria before assays with continuous aeration. Water temperature was regulated at 27°C by using aquarium heater. Fish were feed twice per diem with formulated feed and dead fish were immediately removed to avoid possible water deterioration [14].

Nominal concentrations of active ingredient tested were 0, 5, 15, 30 and 50 ppm of commercial dose(60%) for diazinon and 0, 0.03, 0.04, 0.06, 0.10 and 0.20ppm of commercial dose(2.5%) for deltamethrin were used. 10 groups(5 for diazinon and 5 for deltamethrin) of seven gouramies were exposed for 96h in aerated glass aquaria with 120 l of test medium. During acute toxicity experiment, the water in each aquarium was aerated and the temperature was 27°C. No food was provided to the specimens during the assay and test media was not renewed. Mortality rates were recorded at time 0, 24, 48, 72 and 96 h.Acute toxicity tests carried out according to Hotos and Vlahos [15]. The nominal concentration of diazinon and Deltamethrin estimated to result in 50% mortality of blue gourammi within 24 h (24-h LC_{50}), 48 h, 72 h and 96 h was attained by probit analysis by Finney's method [16] and using 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 diazinon and deltamethrin concentrations versus probit value of percentage mortality.

The 95% confidence limits for LC50 are estimated by using the formula:

$$LC50\ (95\%\ CL) = LC_{50} \pm 1.96\ [SE\ (LC_{50}\ )]$$

The SE of LC_{50} is calculated from the formula: $SE(LC50) = 1/b\sqrt{pnw}$

Where: b=the slope of the chemical/probit response (regression) line; p=the number of chemical used, n = the number of animals in each group, w = the average weight of the observations [15]. After the acute toxicity test, the LOEC (Lowest Observed Effect Concentration) and NOEC (No Observed Effect Concentration) were determined for each measured endpoint.

RESULTS

No fish died during the acclimation period before exposure and no control fish died during acute toxicity tests. The mortality of blue gouramies for diazinon doses 0, 5, 15, 30 and 50 ppm and 0, 0.03, 0.04, 0.06, 0.10 and 0.20 ppm for deltamethrin were examined during the exposure times at 24, 48, 72 and 96 h (Table 1,2). The mortality of blue gouamies was increased significantly with increasing concentrations from 15 ppm to higher concentrations for diazinon and 0.20 ppm to higher concentrations for deltamethrin.

Median lethal concentrations of 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80% and 90% test are presented in Table 3 and 4. Because mortality (or survival) data are collected for each exposure concentration in a toxicity test at various exposure durations (24, 48, 72, or 96 hours), data can be plotted in other ways; the straight line of best fit is then drawn through the points. These are time-mortality lines. The LT_{50} (median lethal survival time) can be estimated for each concentration.

Toxicity Testing Statistical Endpoints are in tow part: 1- Hypothesis Testing: is there a statistically significant difference between the mean response in the treatments and mean response in control or reference sample? LOEC: Lowest Observed Effect Concentration; NOEC: No Observed Effect Concentration. 2- Point Estimates: what toxicant concentration will cause a specific effect on the test population? LC50: the median Lethal Concentration. Our result for Toxicity Testing Statistical Endpoints is shown in Fig 2

DISCUSSION

The results of the present study indicate that both chemicals diazinon and deltamethrin varied in their acute toxicity to blue gourami, *Trichogaster trichopterus*.The toxicity of deltamethrin and diazinon on blue gourami increased with increasing concentration and exposure time.

Table 1: Cumulative mortality of blue gourami Fish (n=7 each concentration) exposed to acute diazinon.

Concentration (ppm)	No. of mortality 24h	48h	72h	96h
0.00	0	0	0	0
5	0	0	0	0
15	0	0	3	4
30	0	4	6	7
50	7	7	7	7

Table 2: Cumulative mortality of blue gourami Fish (n=7, each concentration) exposed to acute deltamethrin.

Concentration (ppm)	No. of mortality 24h	48h	72h	96h
0.00	0	0	0	0
0.03	0	0	0	0
0.04	0	0	0	0
0.06	0	0	0	0
0.10	0	0	0	0
0.20	0	0	2	3
0.30	4	5	6	6
0.40	7	7	7	7

Table 3: Lethal Concentrations ($LC_{1\text{-}99}$) of diazinon (mean ± Standard Error) depending on time (24-96h) for blue gourami.

Point	Concentration (ppm) (95 % of confidence limits) 24h	48h	72h	96h
LC_1	28.9 ± 3.83	19.3 ± 11.0	2.06 ± 0.72	8.50 ± 0.91
LC_{10}	33.6 ± 3.83	23.7 ± 11.0	8.69 ± 0.72	11.2 ± 0.91
LC_{20}	35.6 ± 3.83	25.6 ± 11.0	12.3 ± 0.72	12.3 ± 0.91
LC_{30}	37.0 ± 3.83	27.5 ± 11.0	14.9 ± 0.72	13.1 ± 0.91
LC_{40}	38.2 ± 3.83	28.0 ± 11.0	17.1 ± 0.72	13.8 ± 0.91
LC_{50}	39.3 ± 3.83	29.1 ± 11.0	19.2 ± 0.72	14.5 ± 0.91
LC_{60}	40.5 ± 3.83	30.2 ± 11.0	21.3 ± 0.72	15.1 ± 0.91
LC_{70}	41.7 ± 3.83	31.3 ± 11.0	23.6 ± 0.72	15.8 ± 0.91
LC_{80}	43.1 ± 3.83	32.7 ± 11.0	26.2 ± 0.72	16.7 ± 0.91
LC_{90}	45.1 ± 3.83	34.5 ± 11.0	29.8 ± 0.72	17.8 ± 0.91
LC_{99}	49.8 ± 3.83	38.9 ± 11.0	38.4 ± 0.72	20.5 ± 0.91

Table 4: Lethal Concentrations ($LC_{1\text{-}99}$) of deltamethrin (mean ± Standard Error) depending on time (24-96h) for blue gourami.

Point	Concentration (ppm) (95 % of confidence limits) 24h	48h	72h	96h
LC_1	0.207 ± 0.15	0.193 ± 0.02	0.107 ± 0.10	0.084 ± 0.07
LC_{10}	0.246 ± 0.15	0.232 ± 0.02	0.165 ± 0.10	0.147 ± 0.07
LC_{20}	0.262 ± 0.15	0.249 ± 0.02	0.189 ± 0.10	0.173 ± 0.07
LC_{30}	0.274 ± 0.15	0.260 ± 0.02	0.207 ± 0.10	0.192 ± 0.07
LC_{40}	0.284 ± 0.15	0.270 ± 0.02	0.222 ± 0.10	0.208 ± 0.07
LC_{50}	0.293 ± 0.15	0.280 ± 0.02	0.236 ± 0.10	0.223 ± 0.07
LC_{60}	0.303 ± 0.15	0.289 ± 0.02	0.250 ± 0.10	0.239 ± 0.07
LC_{70}	0.313 ± 0.15	0.299 ± 0.02	0.265 ± 0.10	0.255 ± 0.07
LC_{80}	0.325 ± 0.15	0.311 ± 0.02	0.282 ± 0.10	0.274 ± 0.07
LC_{90}	0.341 ± 0.15	0.327 ± 0.02	0.307 ± 0.10	0.300 ± 0.07
LC_{99}	0.380 ± 0.15	0.366 ± 0.02	0.364 ± 0.10	0.362 ± 0.07

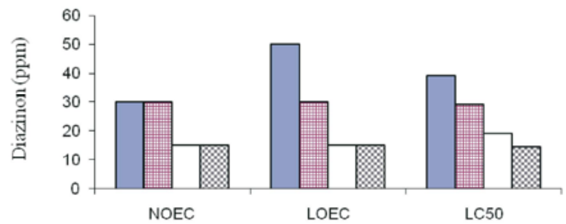

Fig 1: Acute toxicity testing statistical endpoints in blue gourami Fish exposed to crude Diazinon in different times (24h, 48h, 72 h and 96 h respectively).

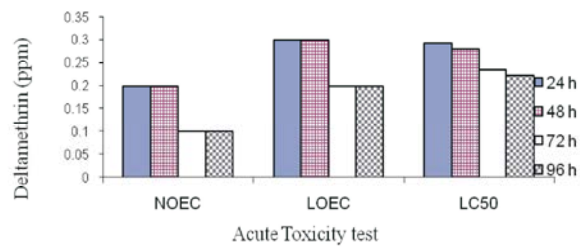

Fig 2: Acute toxicity testing statistical endpoints in blue gourami Fish exposed to deltamethrin in different times (24h, 48h, 72 h and 96 h respectively).

Occurrence of pesticides in high concentrations in agricultural wastewaters and their toxicity to aquatic organisms especially fish species have been reported by many researchers [3,5,7]. Contamination of aquatic environment with pesticides via rainfall runoff is very possible [17]. Fishes are sensitive to aquatic contamination and serious concerns remains due to their potential to cause adverse effects on human and wildlife populations. In addition we found that both diazinon and deltamethrin are lethal substrates to blue gourami.The 96h LC_{50} was calculated to be 14.5 ± 0.91ppm for diazinon and 0.223 ± 0.07ppm for deltamethrin and here we report deltamethrin to be highly toxic to fish.

The 96hLC50 values of diazinon on different fishes reported from tenths to several tens of mg l^{-1} [18,19]. Value of diazinon 96h LC50 was 0.8 mg l^{-1} for guppy (*Poecilia reticulata*) and for zebra fish (*Brachydanio rerio)* was 8 mg l^{-1}[19,20]. Different factors have been suggested to cause selective toxicity of diazinon on different fishes: different detoxification, absorption and different inhibition of acetylcholinesterase [19,21].

Previous studies, indicate the high toxicity of deltamethrin to fish species and our results are in good agreement with these reports. Boateng *et al.*[22] reported that young fish are more susceptible and different species respond unlike to concentrations of chemicals : Mittal *et al.* [23] estimated deltamethrin toxicity to *P. reticulate* to be LC50=0.016 ppm[23].Viran *et al.* reported LC50 value of deltamethrin in guppies as 5.13 mg/L [9]. Mestres and Mestres [24] found 96-h fish LC50 values as follows: *Salmo gairdneri*, 0.39 mg/L; *Cyprinus carpio*, 1.84 mg/L; and *Sarotherodon mossambica*, 3.50 mg/L [24]. LC50 value of deltamethrin in Tilapia, *Oreochromis niloticus* as15.47 µg/l was reported by Boateng *et al.* [22]. Although deltamethrin is thought to be less toxic in field conditions due to its adsorption to sediments, these data are useful to potential ecosystem risk assessment [9].

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