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Determination of LC_{50}, NOEC and LOEC for Toxin Butachlor on Gambusia

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Abstract: The aim of the present study was to determine the acute toxicity of butachlor as potential dangerous organic pesticides to assess mortality effects of this chemical to the gambusia in the form of LC_{50}. Gambusia was exposed to the butachlor (0, 5, 15, 25 and 35 ppm). LC_{50} was determined with probite analysis. The 96h toxicity tests showed 100% mortality in 35 ppm and no mortality in 5 ppm. Further researches are recommended to study the processes by which this chemical affect physiology and histology of fish and their accumulation in fish tissues.

Key words: Lc_{50} · Gambusia · Butachlor · Toxicity

INTRODUCTION

Increased using of pesticides resulted 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 periods) pose a threat to fish and also to the human population consuming the affected fish. Acute toxicity data can help identify the mode of toxic action of a substance and may provide information on doses associated with target-organ toxicity and lethality that can be used in setting dose levels for repeated-dose studies. This information may also be extrapolated for use in the diagnosis and treatment of toxic reactions in humans. The results from acute toxicity tests can provide information for comparison of toxicity and dose-response among members of chemical classes and help in the selection of candidate materials for further work [2]. 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 [3-6]. 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 [7, 8].

Butachlor to combat the spread sheet and some broad leaf weeds in rice farming year, before growth May be applied to the plantations. The effect of the toxin into the plant depends on the amount of water available. The 96-h LC_{50} tests are conducted to assess the vulnerability and survival potential of organisms to particular toxic chemicals. Chemicals with lower LC_{50} values are more toxic because lower concentrations results 50% of mortality in organisms.

The present study was performed to determine the acute toxicity of butachlor as potential dangerous organic pesticides to assess mortality effects of these chemicals to the fish gambusia.

MATERIALS AND METHODS

The selected fish species for the present study was gambusia. Lethal experiments were conducted using 105 young gambusia. Test chambers were glass aquaria of 100 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 [9].

Nominal concentrations of active ingredient tested were 0, 5, 15, 25 and 35 ppm of commercial dose (60%) for butachlor was used. During acute toxicity experiment, the water in each aquarium was aerated. No food was

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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 [10]. The nominal concentration of butachlor estimated to result in 50% mortality of gambusia within 24 h (24-h LC_{50}), 48 h, 72 h and 96 h was attained by probit analysis by Finney's method [11] 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 butachlor concentrations versus probit value of percentage mortality. The 95% confidence limits for LC_{50} are estimated by using the formula: LC_{50} (95% CL) = LC_{50}±1.96 [SE (LC_{50})]

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

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 [10]. After the acute toxicity test, the LOEC (Lowest Observed Effect Concentration) and NOEC (No Observed Effect Concentration) were determined for each measured endpoint.

RESULTS

The mortality of the subjected fishes for butachlor doses 0, 5, 15, 25 and 35 ppm were examined during the exposure times at 24, 48, 72 and 96 h (Table 1).

No fish died during the acclimation period before exposure and no control fish died during acute toxicity tests. Fishes exposed during the period 24-96h had significantly increased number of dead individual with increasing concentration. There were significant differences in number of dead fish between the duration 24-96 in each. There was 100% mortality at 35 ppm concentration within the 96 h dose for all fishes. Median lethal concentrations of 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80% and 90% test are presented in Table 2. 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 Fig 1. LOEC (Lowest Observed Effect Concentration) and NOEC (No Observed Effect Concentration) were same for all studied fishes, however LC_{50} (the median Lethal Concentration).

Table 1: Cumulative mortality of gambusia Fish (n=21 each concentration) exposed to acute butachlor

Concentration (ppm)	No. of mortality 24h	48h	72h	96h
Control	0	0	0	0
5	0	0	0	0
15	0	0	6	11
25	9	14	18	20
35	15	18	21	21

Table 2: Lethal Concentrations (LC_{1-99}) of butachlor depending on time (24-96h) for gambusia

Point	Concentration (ppm) (95% of confidence limits) 24h	48h	72h	96h
LC_{1}	10.343	9.557	5.702	3.921
LC_{10}	18.719	16.580	11.506	9.099
LC_{20}	22.246	19.538	13.950	11.279
LC_{30}	24.789	21.670	15.713	12.851
LC_{40}	26.962	23.492	17.218	14.194
LC_{50}	28.993	25.195	18.626	15.450
LC_{60}	31.024	26.898	20.033	16.706
LC_{70}	33.197	28.720	21.539	18.049
LC_{80}	35.740	30.853	23.302	19.621
LC_{90}	39.267	33.810	25.746	21.801
LC_{99}	47.643	40.833	31.550	26.979

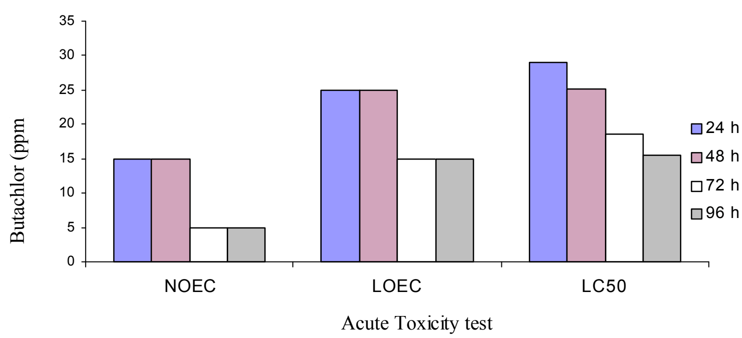

Fig. 1: Acute toxicity testing statistical endpoints in gambusia Fish exposed to crude Butachlor in different times (24h, 48h, 72 h and 96 h respectively).

DISCUSSION

In determining the toxicity of a new chemical to fish, an acute toxicity test is first conducted to estimate the median lethal concentration (LC_{50}) of the chemical in water to which organisms are exposed [12]. The relationship between the degree of response of tested organisms and the quantity of exposure to the chemical almost always assumes a concentration–response form. As in our results the *y*-axis represents percentage mortality and the *x* -axis represents concentration of butachlor. Both variables increased with distance from origin. The cumulative responses to butachlor concentrations yield the sigmoid (S-shaped) curve [12]. The results of the present study indicate that both chemicals butachlor varied in their acute toxicity to gambusia. The toxicity of butachlor on gambusia increased with increasing concentration and exposure time.

Occurrence of pesticides in high concentrations in agricultural wastewaters and their toxicity to aquatic organisms especially fish species have been reported by many researchers [4, 6, 13]. Contamination of aquatic environment with pesticides via rainfall runoff is very possible [14].

Fishes are sensitive to aquatic contamination and serious concerns remains due to their potential to cause adverse effects on human and wildlife populations. Previous studies have used a variety of methods to detect the acute and chronic toxicity of butachlor by preparing various water. This makes comparisons between fish species difficult. For example, it was reported 96 h LC_{50} values for the *Abramis brama* 1.21 ppm [15] and for *Leptodactylus magna astacus* 0.0019 ppm [16] and 48 h LC_{50} values for the *Hipophthalmichthys molitrix* 0.37 ppm [17].

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REFERENCES

1. Stalin, S.I., S. Kiruba and S.S. Manohar Das, 2008. A Comparative Study on the Toxicity of a Synthetic Pyrethroid, deltamethrin and a Neem Based Pesticide, Azadirachtin to *Poecilia reticulata* Peters 1859 (Cyprinodontiformes: Poeciliidae) Turkish Journal of Fisheries and Aquatic Sciences, 8: 1-5.
2. Hedayati, A., A. Safahieh, A. Savar and J. Ghofleh Marammazi, 2010. Detection of mercury chloride acute toxicity in Yellowfin sea bream. World Journal of Fish and Marine Science, 2(1): 86-92.
3. Miller, G.G., L.I. Sweet, J.V. Adams, G.M. Omann, D.R. Passino-Reader and P.G. Meier, 2002. *In vitro* toxicity and interactions of environmental contaminants (Arochlor 1254 and mercury) and immunomodulatory agents (lipopolysaccharide and cortisol) on thymocytes from lake trout (*Salvelinus namaycush*). Fish Shellfish Immunol., 13: 11-26.
4. Galloway, T. and R. Handy, 2003. Immunotoxicity of organophosphorous pesticides. Ecotoxicology, 12: 345-63.
5. Tinoco-Ojanguren, R. and D.C. Halperin, 1998. Poverty, production and health: inhibition of erythrocyte cholinesterase via occupational exposure to organophosphate insecticides in Chiapas, Mexico. Arch. Environ. Health., 3: 29-35.
6. Capel, P.D., S.J. Larson and T.A. Winterstein, 2001. The behavior of thirty-nine pesticides in surface waters as a function of scale. Hydrol. Process., 15: 1251-1269.
7. Parrish, P.R., Acute toxicity tests, 1995. In Fundamentals of Aquatic Toxicology: Effects, Environmental Fate and Risk Assessment, 2nd, ed. G. M. Rand. pp: 947±973. Taylor & Francis, Washington DC.
8. Pandey, S., R. Kumar, S. Sharma, N.S. Nagpure, S.K. Srivastava and M.S. Verma, 2005. Acute toxicity bioassays of mercuric chloride and malathion on air-breathing fish *Channa punctatus* (Bloch). Ecotoxicology and Environmental Safety, 61: 114-120.
9. Gooley, G.J., F.M. Gavine, W. Dalton, S.S. De Silva, M. Bretherton and M. Samblebe, 2000. Feasibility of aquaculture in dairy manufacturing wastewater to enhance environmental performance and offset costs. Final Report DRDC Project No. MAF001. Marine and Freshwater Resources Institute, Snobs Creek, pp: 84.
10. Hotos, G.N. and N. Vlahos, 1998. Salinity tolerance of *Mugil cephalus* and *Chelon labrosus*, Pisces: Mugilidae/fry in experimental conditions. Aquaculture, 167: 329-338.
11. Finney, D.J., 1971. Probit Analysis. Univ. Press, Cambridge, pp: 333.
12. Di Giulio, R.T. and D.E. Hinton, 2008. The Toxicology of Fishes. Taylor & Francis, pp: 319-884.
13. Eisler and G.R. Gardener, 1993. Acute toxicology to an estuarine teleost of mixtures of cadmium, copper and zinc salts. J. Fish. Biol., 5: 131-142.

14. Larkin, D.J. and R.S. Tjeerdema, 2000. Fate and effects of diazinon. Rev. Environ. Contam. Toxicol., 166: 49-82.
15. Willis, G.H. and L.L. McDowell, 1982. Review: Pesticides in agricultural runoff and their effects on downstream water quality. Environ. Toxicol. Chem., 1: 267-279.
16. Jazeb Nikoo, A., 1996. Effects of pesticides and 60% butachlor 57% of malathion on the mortality of *Abramis brama*. Dissertation Master of Fisheries, 65: 20-25.
17. Gholami, N., 2002. Effect of herbicides on pesticide deaths (Machty) and insecticides (malathion and diazinon) on mortality miniatures freshwater crayfish (*Leptodactylus magna astacus*). Thesis Fisheries, Islamic Azad University Lahijan. 130: 14-41.