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A Study on the Toxicity of an Organophosphate Pesticide, Phorate on *Oncorhynchus mykiss* (Wal.)

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ABSTRACT

Fish and other organisms are affected by pesticides which pollute the natural water through agricultural runoff. Fishes are common bio-indicators of water pollution. In the present study bioassay of organophosphate pesticide, Phorate was done on *Oncorhynchus mykiss.* Data obtained from the toxicity tests were evaluated using the Probit Analysis Statistical Method. The 96h LC_{50} of phorate was found to be 19.81ppm respectively. The fish exhibited erratic swimming, copious mucus secretion, loss of equilibrium and hitting to the walls of test tank prior to mortality.

Keywords: Phorate, *Oncorhynchus mykiss*, Toxicity and 96h LC_{50}

INTRODUCTION

Increased use of pesticide results in the excess inflow of toxic chemicals, mainly into the aquatic ecosystem (Baskaran *et al*., 1989; Kalavanthy *et al*., 2001). The aquatic environment is currently under threat by the indiscriminate use of synthetic pesticides by the human activities and causing high risk to non-target organisms (Kumar *et. al*., 2010). Among different classes of pesticides, organophosphates are more frequently used, because of their high insecticidal property, low mammalian toxicity, less persistence and rapid biodegradability in the environment (Singh *et al.*, 2010). Phorate [O, O-diethyl-S - [(ethylthio) methyl] phosphorodithioate] is an organophosphate pesticide used to control sucking and chewing pests. It is used in pine forests and on root crops and field crops, including corn, cotton, coffee, and some ornamental plants and bulbs (Gan and Jans, 2007; Wub *et al.,* 2010). In 2001, U.S. annual use of phorate was estimated at 3.2 million pounds. The LD50 of Phorate was reported to be 1.1–2.3 mgL^{-1}, depending on the pathway of intake (Ku and Lin, 2002; Wub *et al.,* 2010). Due to its severe acute risk for human health, the WHO has classified Phorate as a technical product of extreme hazard. During its use as an insecticide, Phorate primarily contaminates the environment through constant leaching and superficial runoff during rainfall.

Phorate is recommended for application as a high or a low volume spray on crops such as paddy, wheat, soyabean, apple, sugarcane, mustard, sunflower and cashew. The Environment Protection Agency (EPA) has classified Phorate as toxicity class 6.1 highly toxic (URL: 1). Several species of fish are susceptible to deleterious effects when exposed to heavy metals, pesticides and other environmental stressors (Khangrat *et al*., 1988; Areechon and Plump, 1990).

To overcome the hazardous effects of these organic pesticides, recent emphasis is on the use of natural pesticides, which are usually of plant origin. Some plants contain compounds of various classes that have insecticidal, piscicidal and molluscicidal properties. However, little work has been done on the toxic effect of insecticides based pesticides on fish. Hence the present study was carried out to evaluate the effect of organophosphate pesticide Phorate to *Oncorhynchus mykiss* (Wal.).

MATERIAL AND METHODS

Healthy and active adult *Oncorhynchus mykiss* were obtained from India fish farm Poondi, Thiruvallur (T.N). They weighed 45g±1g and their length was in the range 10cm±1. They were brought to laboratory carefully in oxygen filled polythene bags in card board boxes to avoid any injury and disinfected by giving a bath for five minutes in $KMno_4$ solution. Thereafter, they were transferred to glass aquariums filled with dechlorinated water. The fishes were acclimated to the laboratory conditions for at least 20 days prior to the experiment. During acclimatization fishes were fed daily with commercial fish food which was given at morning hours. Water was replaced every 24h after feeding in

order to maintain a healthy environment for the fish during acclimation and experimental period.

This ensures sufficient oxygen supply for the fish and the environment is devoid of any accumulated metabolic wastes. Dead fishes when ever located were removed immediately to avoid fouling of the water.

Water quality characteristics were determined and maintained. Phorate manufactured by United Phosphorus Ltd. purchased from local market of Thiruvallur was used for evaluation of their toxicity to fish. For determining LC_{50} concentration different stock solutions were prepared, separate glass aquariums were taken and different concentrations of Phorate were added from the stock solution. Simultaneously a control set was run with the experiment. During assay no food was administered to fishes. The LC_{50} concentration for 96h was calculated by probit analysis method of Finney's (1971). The control and Phorate exposed fish were kept under continuous observation during experimental periods.

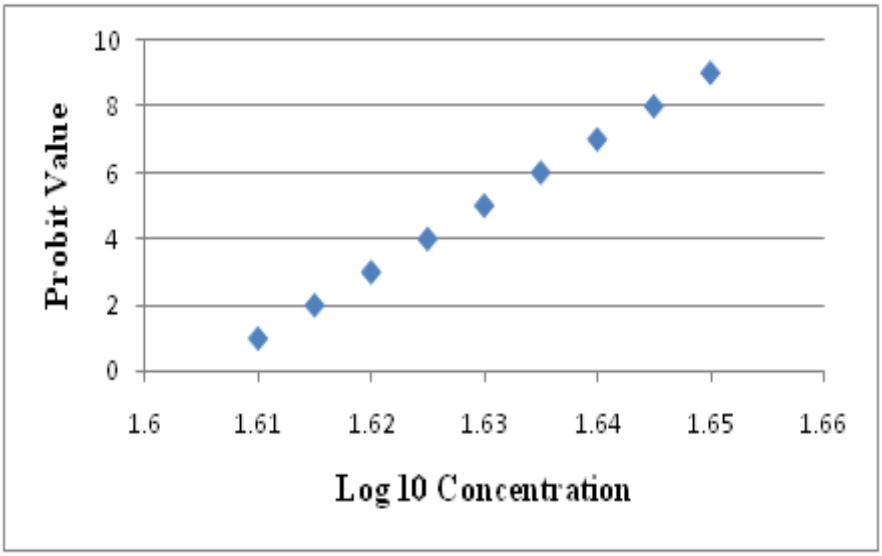
RESULTS

The 96h LC_{50} value of Phorate was found to be 19.81ppm respectively. The LC_{50} concentration for 96h was calculated by probit analysis method of Finney's (1971). Table 1 shows the relation between the Phorate concentration and the mortality rate of *Oncorhynchus mykiss* and the graph 1 below show the plot of Finney's probits against log10 conc. for calculating LC_{50} value of the pesticide.

Table 1: For Phorate exposed to *Oncorhynchus mykiss*

S. No	Conc. Mg/L	Log_{10} Conc.	Total No.	No. Dead	% Mortality	Probit
1.	12	1.1461	10	0	0%	-
2.	13	1.1671	10	16	10%	3.72
3.	14	1.3041	10	3	30%	4.48
4.	15	1.3304	10	6	60%	5.25
5.	16	1.3753	10	8	80%	5.84
6.	17	2.3988	10	10	100%	-

After exposure of the pesticides, the *Oncorhynchus mykiss* showed behavioral changes, they aggregated at one corner of aquarium, irregular, erratic and darting swimming movements and loss of equilibrium. They slowly became lethargic, hyper excited, restless and secreted excess mucus all over their bodies. The fish exhibited peculiar behavior of trying to leap out from the pesticide medium which can be viewed as an escaping phenomenon. They often spiral rolled at intervals and finally the fishes sank to bottom with their least operculum movements and died with their mouth opened. However, the behavioral changes were more prominent for the organophosphate pesticide Phorate.



Graph 1: For Phorate exposed to *Oncorhynchus mykiss*

DISCUSSION

Newer biological pesticides are developed to replace deleterious chemical pesticides. Even though chemical pesticides are target specific and effective, their impact on the environment is mostly deleterious. In the present study, the pesticide was high toxic to fish compared to Phorate. The 96h LC_{50} of Phorate is 19.81 ppm. Cagauan *et al*. (2004) showed that the lethal concentration of neem to Nile tilapia *Oreochromis niloticus* L. was 12.4 ml/L and mosquito fish *Gambusia affinis* Baird and Girard was 8.31 ml/L and the corresponding 96 h LC50 values were 2.57 and 3.0 ml/L). Hassanein *et al.,* 2007 reported the 96h LC_{50} value of a neem biopesticide (Triology) on the grass carp fish, *Ctenopharyngodon idella* and was found to be 112ppm. The LC_{50} values of Phorate has been reported by various workers as in *Cyprinus carpio* 6gm it was 0.34ppm for 96h (Verma *et al.,* 1981) in *Cirrhinus mrigala* it was 9.1ppm for 96h (Velmurugan *et al.,* 2009) and in *Ctenopharyngodon idella* it was 13.1ppm for 24h (Tilak and Swarna Kumari 2009).

The LC_{50} values clearly indicate that the chemical based pesticide is highly toxic. To reduce the chemical load on the environment, it is suggested that use of plant based pesticides should be encouraged (Schmutterer, 1990). However, care should be taken to use even the plant based pesticide at moderate levels. Furthermore, plant based pesticides disintegrate easily into constituent elements without leaving any indelible impression in different regions of the environment (Khan and Ahmed, 2000). It is advocated that more and more plant products should be developed with proper and targeted action and this eventually helps in keeping the environment free from hazardous chemicals. From the present study, it could be concluded that Phorate contamination is dangerous to aquatic ecosystems, and this fact should be taken into consideration when this insecticide is used in agriculture or in the control of mosquito populations. It can be also concluded that although bio pesticides are considered as less toxic and environmental friendly, but precautions must be taken when it is used in fish inhabiting areas since the excess application can affect the life of organisms. This type of study can also be useful to compare the sensitivity of the various species of aquatic animals and potency of chemicals using LC_{50} values and to derive safe environmental concentration by which there is no lethality and stress to the animals.

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