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ACUTE TOXICITY OF ORGANOPHOSPHATE PESTICIDES ON JUVENILES OF THE MARINE FISH *(OREOCHROMIS MOSSAMBICUS)*

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ABSTRACT

Pollution of marine ecosystem by various xenobiotic chemicals including pesticides is of great concern, because of their high toxicity and persistence in the environment. In the present study acute toxicity of organophosphates (Malathion, Chlorpyrifos) pesticides on the marine fish *(Oreochromis mossambicus)* was investigated. Acute toxicity of the pesticides on marine organism was estimated by determination of the 24 hour LC50. The toxicity tests were performed separately for each pesticide. Data generated from the acute toxicity tests were evaluated using the probit analysis statistical method. The LC_{50} values obtained at 24 hour show that the fish were sensitive to all the two pesticides tested. The rate of mortality (%) was directly proportional to the concentration of pesticides. In the present study, we noted that Malathion was more toxic than chlorpyrifos. The result of the study suggested that organophosphates pesticides were highly toxic to fish juveniles.

Key words: Pesticides, Malathion, Chlorpyrifos, Fish, *Oreochromis mossambicus*.

INTRODUCTION

Widespread use of agrochemicals in agriculture has contributed significantly to increase in crop yield and improved national earnings of the country. However such hazardous chemicals may result in environmental pollution and toxicity risk to non-target organism (Velisek *et al*., 2007). A wide range of pollutants is continuously discharged into marine environment, which ultimately affect the marine ecosystem. Fish are used as sentinel organisms in ecotoxicological studies as they are important part of the food web, accumulate toxic substances and respond to mutagenic substances (Cavas *et al.*, 2005).Therefore, fish are use as biomarkers for the effects of pollution and early detection of aquatic environmental problems (Lopez-Barea, 1996; Van Der Oost *et al*., 2003).The pollutant accumulates in the muscle and other tissues of fish and hence poses a serious threat to human health (Sekhar *et al*., 2003).

These pesticides are rapidly degradable but their high acute toxicity to non-target species are reported in many studies (Phyu *et al.*, 2005; Sial *et al.*, 2009). Pesticides are acutely toxic to non-target organisms like invertebrates, mammals, birds and fishes, especially those inhabiting the marine environment (Burkepile *et al*., 2000; Bhavan and Geraldine, 2001; Selvakumar *et al*., 2005; Suryavanshi *et al.,* 2009; Shoaib *et al.*,2012).Some of the pesticides have been reported to persist in the environment and tend to bioaccumulate in the organisms (Jayashree and Vasudevan, 2007).Various authors have reported that pesticides can be acutely toxic to fish (Gurusamy and Ramadoss, 2000; Moore and Waring, 2001; Prasad *et al*., 2002; Shrivasatava *et al*., 2002; Eder *et al.,* 2004; Shaikh and Yeragi, 2004 and Visvanthan *et al.*, 2009).

Oreochromis mossambicus is found in many tropical and subtropical habitats around the globe, lives in rivers, lagoons, creeks and streams. *Oreochromis mossambicus* are omnivorous, very hardy, euryhaline fish have a broad salinity and temperature tolerance. The Mozambique tilapia is an invasive species in many parts of the world, having escaped from aquaculture or been deliberately introduced to control mosquitoes (Moyle, 1976).

Acute toxicity studies are generally employed to compare the sensitivities of different species to different potencies of the chemicals. The present study has been carried out to study the lethal concentration 50% (LC_{50}) of pesticides organophosphates (malathion, chlorpyrifos) on the marine fish *(Oreochromis mossambicus)*acting individually.

MATERIAL AND METHODS

Preparation of Chemicals

Pesticides were purchased from the market, organophosphate (chlorpyrifos 40% EC, malathion 57% EC).Stock solution of 100 ppm and appropriate working concentrations were prepared in filtered seawater.

Collection and maintenance of fish

The fishes *Oreochromis mossambicus* (2.7 ± 1cm in length, 5 ± 1 g in weight) were collected from Chilya hatchery Thatta. The fish were transported in clean aerated water to the laboratory ensuring minimum stress. The fishes were allowed to acclimate in the laboratory conditions for one week prior to experiments. The fishes were kept in clean aerated seawater in glass aquaria (92 cm Length x 39cm width x 47 cm height) at temperature (23°C ± 1°C), Salinity 30ppt, pH 7.5 with photoperiod 16 hour of light and 8 hour of dark (16 L: 8 D) cycle. Seawater in each aquarium was replenished every day in order to remove faeces and remaining food and to maintain the water quality and oxygen saturation level above 60%. Fishes were fed ad labium and commercial diet two times a day.

Bioassay

The 24hours acute toxicity bioassay was carried out as described by Shoaib *et al*. (2012). All the glassware was acid washed prior to the tests and natural seawater was used throughout the experiments. Fish were exposed to different concentrations of selected pesticides. The different concentrations of pesticides ranged between 20-80ppb.Experiments were carried out in glass aquarium (30.5 cm Length x 30.5cm width x 30.5 cm height).In each aquarium ten experimental fishes were used. All pesticide concentrations were prepared with filtered seawater. The experiment was performed in triplicate. Three controls were also set up for each experiment. The control has only seawater. The experiment was performed at temperature (23°C ± 1°C), Salinity 30ppt, pH 7.5, photoperiod 16 hour light and 8 hour dark. Acute toxicity measured as mortality of organisms exposed to pesticides, acting individually was estimated by determination of the 24 hour LC_{50} (the concentration of the pesticides which kills 50% of the test animals after 24 hour exposure). Organisms were considered dead if they did not exhibit any movement and it lay immobile. The LC_{50} values were determined by using Computer program, Biostat 2009 based on Finney Method 1952 (Probit analysis).

RESULTS

In the present study organophosphates (malathion, chlorpyrifos) pesticides tested on the marine fish (*Oreochromis mossambicus*) show that the fish were sensitive to all the two pesticides tested. The rate of mortality (%) was directly proportional to the concentration of pesticides (Fig. 1). The variability in the degree of sensitivity is reflected by the lethal concentration values of pesticides, at which 50% mortality occurs (LC_{50}), shown in Table 1. In the present study we noted that Malathion is the most toxic pesticide having lowLC_{50}than chlorpyrifos.

Table 1.Toxicity of organophosphate pesticides on marine fish (*Oreochromis mossambicus*) after 24 h of treatment showing LC_{50}.

Pesticides	No. of Fishes	Concentration tested (ppb)	LC50 ppb	Intercept	χ square	p-level
Chlorpyrifos	150	20-80	63	6.06	0.007	0.93
Malathion	150	20-80	28	9.29	0.881	0.64

DISCUSSION

The result of the foregoing study suggests that organophosphates are highly toxic to fish and have low LC_{50} values. These results are in agreement with previous reports (DOSE, 1997; Chindah, 2004; Shrivasatava *et al.,* 2010; Shoaib *et al.,* 2012; Shoaib *et al.,* 2013). The increase in concentration of pesticides increases the mortality rate of fish (Shoaib *et al.,* 2012; Shoaib *et al.,* 2013). Organophosphates are non-persistent in nature and rapidly degrade, therefore acute toxicity test of 24h LC_{50} were considered in the present study. Despite the fact that organophosphates are short-lived, these pesticides are highly toxic compared to a number of other pesticides for example organophosphate pesticide is found to be more toxic to fish than organochlorine (Veeraiah,2001; Deb and Das 2013; Tilak, 2001).Therefore, organophosphates can affect organisms exposed even for a very short period. In the present study we noted that Malathion is the most toxic pesticide to fish having LC_{50}28 ppb as compared to chloropyrifos. In the present study *Oreochromis mossambicus* responded differently, to the organophosphates tested, indicating variability in sensitivity of test organisms. The toxicity of pesticides to marine organisms is affected by age, size and health of the species (Abdul-Farah *et al.,* 2004). Physiological parameters like temperature, pH, dissolved oxygen and turbidity of water, concentration and formulation of chemical, and its exposure influence toxicological studies (Gupta *et al.,* 1981; Young, 2000).

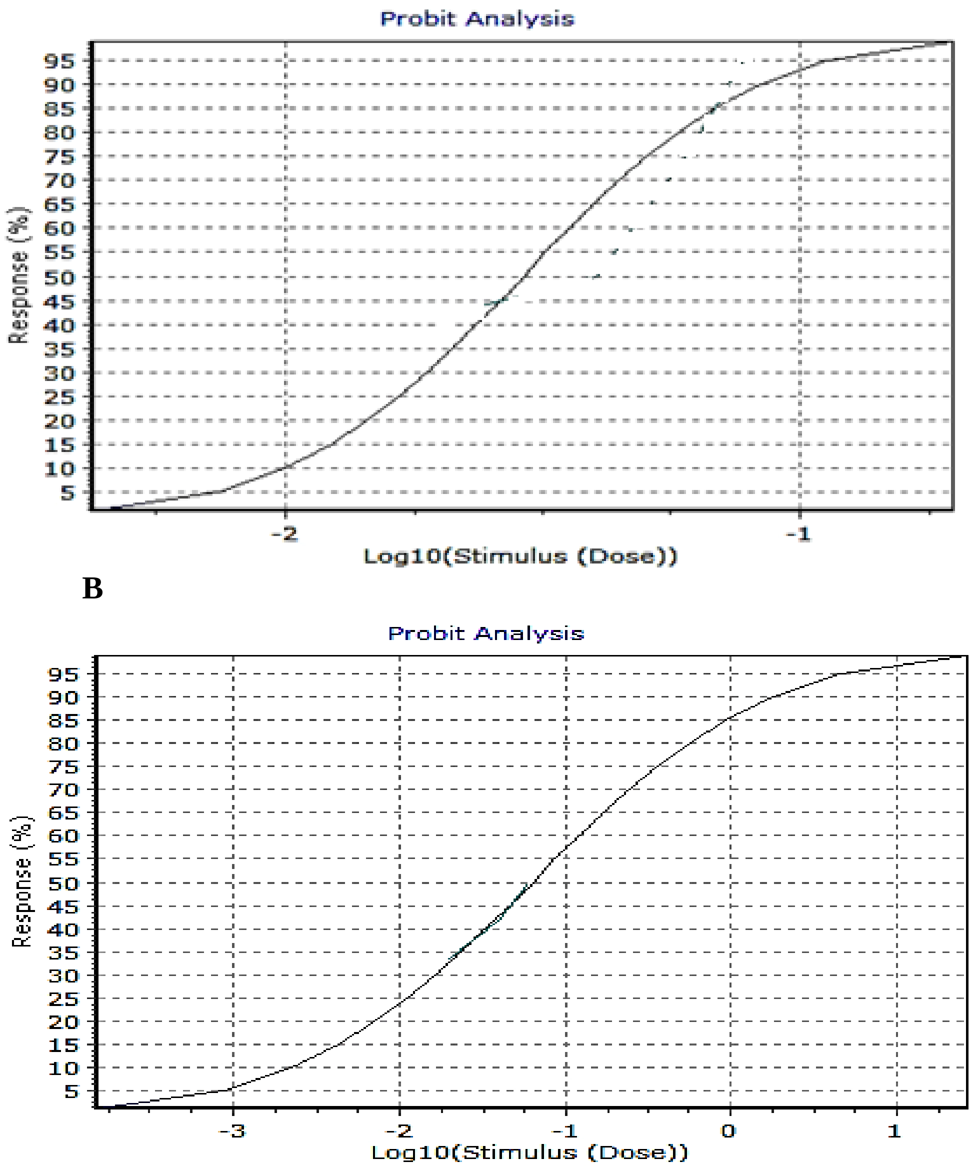

Fig. 1.Probit analysis curve showing response of marine fish (*Oreochromis mossambicus*) after exposed to different concentrations (dose) of: A, Malathion pesticide; B, chlorpyrifos pesticide.

In the present study we use juvenile of fish to study effects of pesticides as in bioassay generally juveniles are employed for eco toxicological test to predict environmental risk. Juveniles are more sensitive to environmental impacts than the adult (Kefford *et al.,* 2004).The fish juveniles appeared to be more susceptible to pesticides, for example, as reported for *Heteropneustes fossilis* (Dutta, 1995) and *Micropterus salmoides* (Pan and Dutta, 1998) and *Aphanius dispar* (Shoaib *et al.,* 2012*;* Shoaib *et al.,* 2013).

According to PEPA, the National Environmental Quality Standards (NEQS) relating to municipal and liquid industrial effluents for pesticide is 150 ppb (The Gazette of Pakistan, 1993). However, in our result the value of LC_{50} when exposed to two pesticides is recorded as 28 ppb and 63 ppb for fish, which is quite low. Many investigators have reported that various kind of pesticides are responsible to cause severe impairment in physiological set up of fish (Begum, 2004; Monteiro *et al*., 2006; Siang *et al*., 2007; Banaee *et al*., 2009). Physiological and biological process are disturbed due to the effect of pesticides include neurological disorder and disruption of nerve functions, respiratory dysfunction and suffocation that can lead to death of fishes (Banaee *et al*., 2011). In short, pesticides have lethal effect on organisms and exposure to these chemicals may disturb the marine food web through bioaccumulation (Abedi *et al*., 2013). Hence continuous use of organophosphates pesticides may harm fish juveniles in mangrove area, may attribute to decline of fisheries and marine resources resulting in loss of revenue generated by fishery industry.

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