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STUDIES ON TOXICITY DETERMINATION

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ABSTRACT: -

In nature the overall effect of a pesticide on the biota LS influenced by number of environmental factors which may alter the rate of metabolism of the organism. Alternatively, these factors may get the toxicant increase its adsorption to the particular matter, thereby making it unavailable.

KEYWORDS: *Toxicity Determination , environmental factors , predict toxicity .*

INTRODUCTION

Sigel (1 986) defines the toxicity of a particular chemical in terms of its capacity to reduce the fitness of a population and its ability to disturb the interactions between the population. Though, it is very difficult to predict toxicity of a particular toxicant to fish, nevertheless, the toxicologists have seriously felt the need of knowledge for the determination of an appropriate concentration that may be toxic to a particular animal. For such study a few bioassay methods have .been adopted in practice. In this context Sprague (1973) has modified the previous method especially for fishes. His method is being frequently followed in recent times.

The most important way to evaluate the toxicity is to find out the very minimum concentration of a particular toxicant which kills fish in a given time.

Thus most widely acknowledged methods to determine the toxicity of a chemical is a specific time are the acute toxicity tests. The toxic levels of various chemicals including pesticides to different organism in form of LC50 (concentration lethal to 50% of the test organisms) have been established following acute toxicity tests. The LC_{50} values have been referred to as quick and useful devices in determining the toxicity of a particular pollutant to a particular species under specific *physico-chemical* conditions (Shaw *et al*, 1990). Some of the notable experiments on toxicity to fishes are those of *Nuvacron, Mathur,* 1975, *diazinon* to fat head Minnows, *Alison & Hermanutz,* 1977, *organic biocides to Labeo-rohita, Verma et al*., 1977, *Parathion on Colisa fasciatus, Sastry and Malik,* 1979, *carbaryl to Mystus vittatus, Arunachalam et al* 1989, endosulfan to *Macrognathus aculecitum, Rao et al*., 1981, malathion to *Brachydanio rerio, Kumar and Ansari,* 1984, *Lindon to some air-breathing fishes, Bakhtharathsalam*, 1988, carbofuron to *H. fossilis and C. batrachus, Ghosh*, 1990, Phosphamidon to *Gambusia affinis, Govindum et al*., 1994, Methionyl to *Cyprirtus carpio, Choudhary* (1996) has explained toxicity of fenvalerate to the *fish Channa punctutus. Trivedi and Saksena,* 1999, Metacid 50 to *Channa* punctatus, *Alam,* 2000, *fenvalerate* to Sarotherodon mossambicus (Patnaik et al., 2002), Singh, 2002, reported toxicity of OP & OC to fish H. fossilis. Recently Sonawane & Gathe (2003) studies combined effects of herbicide with pendimethal. Similarly Jha (2006) studies effects of herbicide (Herboclon) on C. batrachus, Rita & Milton (2006) studied effect of carbamate pesticide to fish *Oreochronis mossambicus*. Very recently Rakesh et al., 2007 reported acute toxicity bioassay of dimethoate on fresh water fish *H. fossilis*. Very recently Rani et at. (2008) reported Toxicity of Nuvan on kidney cholesterol on Labeo-rohita. Kumari et at. (2008) has explained

impact of Herbicide (Glyphosate) on the bio–chemical components of the fish catla–catla, while Jha (2009) has explained Nuvan toxicity to the fish *H. fossilis*. Poonam et at. (2010) has explained effects of Fenvalerate on some organs of a freshwater Fish Anabas testudineus (Bloch).

2 .RESULTS :

The physico-chemical characteristics of the test water are enlisted in Table-I. The toxicity of Fenvalerate obtained under static system have been shown in Table–II, Fig (1.5) by regression equation. The 24 hr., 48 hr., 72 hr. and 96 hrs. of LC_{50} values for Fenvalerate by regression equations in Table – II is 0.15 ppm, 0.25 ppm, 0.244 ppm, 0.35 ppm respectively.

SUBLETHAL OR TEST CONCENTRATION :

The formula of Hart et al. (1945) when applied to the LC_{50} values of Fenvalerate, gave the sublethal dose of 0.107 ppm respectively.

Behavioural response :

Fish were exposed to various concentrations of the pyrethroid to determine acute toxicity values for different intervals. The behavioural responses of the fish under the stress of individual synthetic pyrethroidal insecticide were recorded, which are being mentioned below.

RESPONSES UNDER FENVALERATE ATE EXPOSURE :

Fish that were exposed to different concentrations of fenvalerate exhibited exciting and agonistic behaviour. Definite symptoms of restlessness reflected by erratic opercular movements, difficulty in respiration and convulsion were observed. The fish came to the surface of the solution again and again, possibly to gulp the atmospheric air directly and to avoid the toxic environment Air bubbles were noticed coming out of the mouth. These hyperactive response were more marked at higher concentrations and that too in the earlier phase of exposure. However, in the latter stage of exposure, the affected fish showed hypoactivity laying for several hours on their side on the bottom of the aquaria making very little movement. Another characteristic behavioural change was that some individuals frequently dashed against the walls of the experimental aquaria. Such affected fish at times swam with head downwards, jabbing and scraping at the bottom of the aquaria with the snout. This sort of behaviour was apparent during later phase of exposure and continued until the fish became quiescent just before death.

Further, the exposed fish initially revealed an increased rate of opercular movement followed by its sharp fall. In control fish the rate of opercular movement ranged between 48 and 51 per minute, however, as against this under fenvalerate stress the movement increased ranging between 61–73 per minute for first 30 minutes after which it became gradually feeble ranging between 36–40 per minute. In addition the exposed fish secreted excessive mucous from the skin as evident from its deposition over the body and gills.

Further, pale colouration of the body was another change noticed during this study.

3.DISCUSSION :

The present findings are in conformity with those of Steven et al. (1985), Gupta et al. (1985) and Ghosh & Chatterjee (1988). However, Reddy and Bashamohideen (1989) estimated 48 hr LC_{50} value of fenvalerate to Cyprinus carpio as 0.030 ppm and calculated its safe concentration to be 0.0096 ppm. The safe concentration is a function of relative toxicity between 24 and 48 hrs. If the safe concentration is low then the toxicity is high and vice versa. The safe concentration values in the present study was found to be 0.107 ppm for fenvalerate. Yellama et al. (1989) found 24 hr LC¬ of fenvalerate for juvenile and adult Tilapia mossambicus to be 0.3 ppm and 0.1 ppm respectively. Ghosh (1990) evaluated LC_{50} of fenvalerate for 48 hrs. to the fish *C. Carpio* and *O. mossambicus* as 0.048 mg/L and 0.045 mg/L respectively. Ghosh and Chatterjee (1988) found 48 hr LC_{50} of fenvalerate to Anabas testudineus, as 0.0084 mg/L. The present findings are in conformity with Choudhary (1996) has also reported LC_{50} values of Fenvalerate for 24, 48, 72 and 96 hrs to the fish *Channa punctatus* i.e., 0.138 ppm, 0.130 ppm, 0.098 ppm

and 0.078 ppm respectively. Very recently Poonam et al. (2010) has also reported LC_{50} values of fenvalerate for 24, 48, 72 and 96 hrs as 0.128 ppm, 0.120 ppm, 0.088 ppm and 0.068 ppm respectively to the fish *Anabas testudineus* (Bloch).

The comparison of the LU values with the present ones and the subsequent differences therein may be because of various factors influencing the bioassay techniques like, temperature, pH, DO etc. (Kaviraj & Konar, 1982), or the viability in the bioassays techniques or inter and intra species variations due to different body size and weight as also the age of fish. Agrawal (1991).

The bioassay tests in the present study have been carried out at static laboratory conditions. The laboratory bioassay provides the quickest and most reliable information about the toxicity of chemicals, although it differs to a certain extent from the field results (Sandors, 1969). Acute toxicity appears at higher concentration in the field than in the tocicity of the chemicals to the aquatic organisms helps in establishing tolerable and safe limits of chemicals. By knowing the LC_{50} values of a toxicant its discharge into the water resources can be regulated to protect the aquatic life. Calculation of safe concentrations may be meaningful at present, as these may help to a great extent in regulating their discharge in water.

It is clear from the present study that pyrethroids undertaken are hazardous to environment as also to the aquatic lives. The presumable safe concentration suggested in the present study may help to minimize water pollution by both the pyrethroids, to determine water quality criteria and in the present study may help to minimize water pollution by both the pyrethroids, to determine water quality criteria and in the setting of water quality standards. Further, it as presumed that even though the pyrethroids have low mammalian toxicity and non-persistent nature due to their biodegradable properties, nevertheless, they are highly toxic to fishes and can in no way be considered as safe substitutes for the organochiorine and organophosplate group of pesticides as far as their relative toxicity to fish is concerned. The results of this study further show that at low concentrations the exposure of fish to the test pyrethroids in the aquatic environment may not cause immediate death, but they certainly would bring about some undesirable behavioural changes which may lead to a reduction in the number of fish in the population which in turn ma disturb the ecobalance of the aquatic system in which these fish live.

FISH BEHAVIOUR :

The test fish exposed to different graded concentrations of synthetic pyrethroid, fenvalerate exhibited more or less similar abnormal behavioural responses. During the exposure time fish initially showed rapid movement, faster opercular activity, surfacing and gulping of air at all concentrations. However, at higher concentrations fish also showed erratic swimming, hyper excitability, difficulty in respiration and convulsion as also tendency to escape from the aquaria. It was observed that with the increasing concentration and exposure Inn these activities were relatively increased initially and subsequently reduced. This reflects the expression of the sign of distress. These behavioural changes were more pronounced under decamethrin exposure, thereby indicating greater toxicity of this chemical to the test fish. Besides, an interesting observation was that the fish hail a visible increase in body dispigmentation along with profuse mucus secretion and its coagulation all over the body and gills. Later on, fish struggled hard for aerial-breathing with their restrictive swimming movements and indicated poor response to external stimulant. This was followed by a loss of equilibrium and fish slowly became lethargic and underwent it hypoactivity. Similar behavioural responses have been reported by earlier workers under synthetic pyrethroidal insecticide exposure and other such related posticides (Ghosh, 1990, Bhatnagar and Tyagi, 1994; Dutta et al, 1994).

Behavioural changes are sensitive indicators of pollutants and the optomotor response is essential for behaviours such as food searching, orientation towards food odour, location of a mate, escaping from a predator and avoidance of an adverse situation. Any alteration in the chemical composition of the natural aquatic environment usually affects a change in the behaviour and health of the fish. Since the behaviour of the animal reflects the physiological status of the body, an alteration in the body metabolism can be visualized through abnormal behaviour. It was reported that for proper ethological responses both nervous and energy system of an animal work in a coordinated manner (Lehninger, 1978). There exists considerable information on the effects of pesticides on ethology of fish (Babu et al., 1986; Ghosh, 1989; Palawski et al, 1983; Radhaiah and Rao, 1988). The synthetic pyretrhoids are found to inhibit acetylecholinesterase and clevate acetyicholine level (Chatterjee et al.,

1986; Hollingworth and Green, 1989). The observed violent movement and loss of equilibrium may be attributed to accumulation of Ach content at nerve ends and neuromast organs, thus disrupting the synaptic transmission of nerve impulses along neurous (Ghosh, 1990). Further, such abnormal swimming behaviour and altered movements as observed during this study have been considered to be the result of excessive elimination of skeletal minerals. The heavy exudation of mucus over the body and body dispigmentation are attributed to the dysfunction of endocrine/pituitary gland under the toxic stress, causing changes in the number and area of mucous glands and chromatophores (Pandey et al., 1990).

The lethargic condition tint ed in this study due to pyrethroidal exposure would affect the fish in several ways. The fish that became lethargic would fall easy prey to predators. Feeding and food capture will be hampered by lethargy and loss of orientation caused by the action of the pollutants. It is presumed that fish living in streams may not be able to maintain their position and may be swept downstream. Bull and Mclnerney (1974) reported that many juvenile coho salmon were unable to maintain position and were swept downstream after being exposed to sumithion, an organophosphorus insecticide, in a stream aquarium.. Burton et al., (1972) demonstrated that acute Zn poisoning in Salmo gairdneri involved a modification of gas exchange process at the gills with subsequent hypoxia at tissue level. Thus, the intense opercular movements exhibited by the fish after lethal exposure to fenvalerate and decamethrin may be attributed to a sort of hypoxic stress accompanied by a sequential inhibitory influence of these toxicants on the respiratory system.

The large amount of mucus secretion observed in the present case may also be associated with the minimization of irritating effects of the pyrethroid and this may be one of the vital factors for the death of the poisoned fish. In this context, Jha and Pandey (1990) have indicated that mucus clogs the gill resulting into the death of the fish.

Lal et al. (1984) are also of the view that respiratory distress and increased opercular movement are the signs of oxygen depletion. Difficulty in respiration marked during the present study may be discussed on these lines. Natarajan (1981) suggested that pesticides damage the red blood corpuscles which reduce the efficiency of the fish to trap dissolved oxygen resulting in respiratory trouble. So far as pale colouration of the body noted in this study is concerned, it can be correlated with the destruction of melanophores in the dying cells. The increased irritability of test fish which was reflected in aggressiveness of the fish seems to be the results of the drastic changes in the Na+ and K+ levels in various tissues, since sodium and potassium exert effect on muscle irritability (Swarup et al., 1981).

CONCLUSION

Thus, the present toxico-behavioural studies conclude that the synthetic pyrethroid impair the homeostatic mechanism critical to the survival of the fish.

TABLE – I
Physico–chemical characteristics of test water

Characteristics unit	No.of obs.	Mean	Range
PH	10	7.6	7.2–7.9
Temperature (^{0}C)	10	24.6	2.3–2.6
Total solids mg / l	10	38.5	32.6–38.2
Dissolved solids mg/l	10	12.3	10.5–12.6
Dissolved oxygen mg/l	10	6.5	5.4–7.6
Free CO_2 mg/l	5	2.1	1.3–3.0
Total hardness mg/l as $CaCO_3$	5	180.0	165.0–200.0
Total alkalinity mg/l as $CaCO_3$	5	150.0	136.0–165.0

TABLE – II
LC_{50} values of Fenvalerate with their regression equations
to Anabas testudineus

Pesticide	Time (hr)	LC_{50} (ppm)	Regression equation
Herboclin	24	0.15	–10 + 400 ×
Synthetic	48	0.25	–22 + 360 ×
Pyrethroid	72	0.24	–24 + 360 ×
Fenvalerate	96	0.35	–10 + 200 ×

FIGURE – 1
LC_{50} VALUES OF FENEVALERATE FOR 24 HRS.
TO FISH *A. TESTUDINEUS*

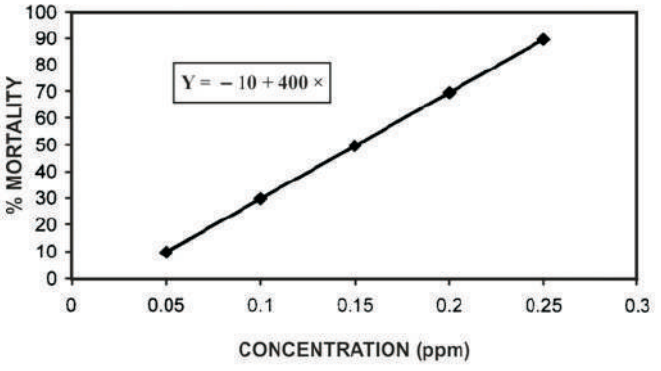


FIGURE – 2
LC_{50} VALUES OF FENEVALERATE FOR 48 HRS.
TO FISH *A. TESTUDINEUS*

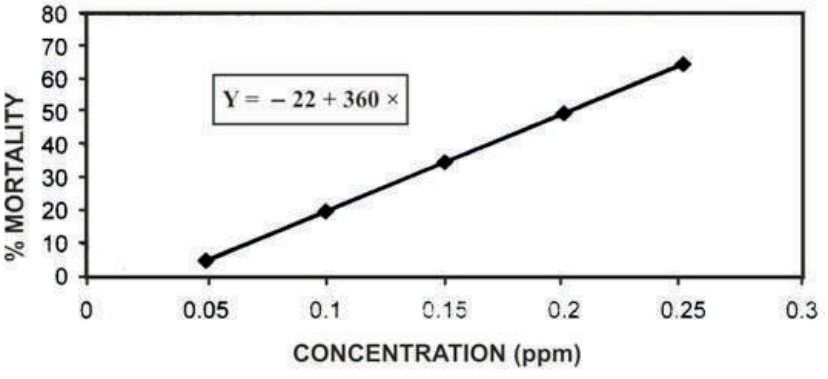


FIGURE – 3
LC_{50} VALUES OF FENEVALERATE FOR 72 HRS.
TO FISH *A. TESTUDINEUS*

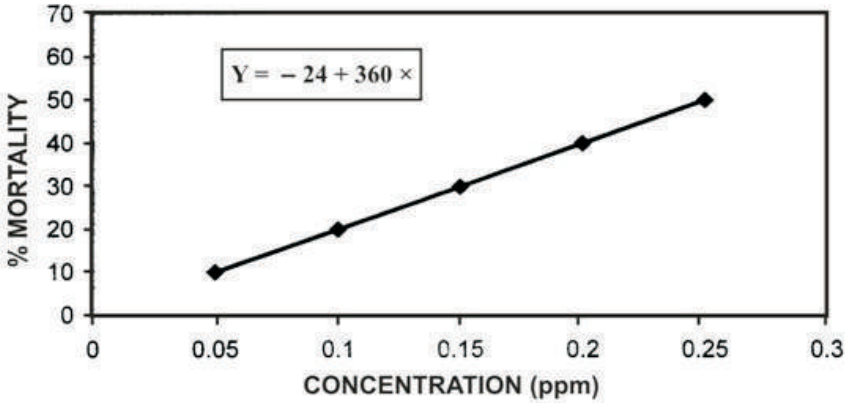


FIGURE – 4
LC_{50} VALUES OF FENEVALERATE FOR 72 HRS.
TO FISH *A. TESTUDINEUS*

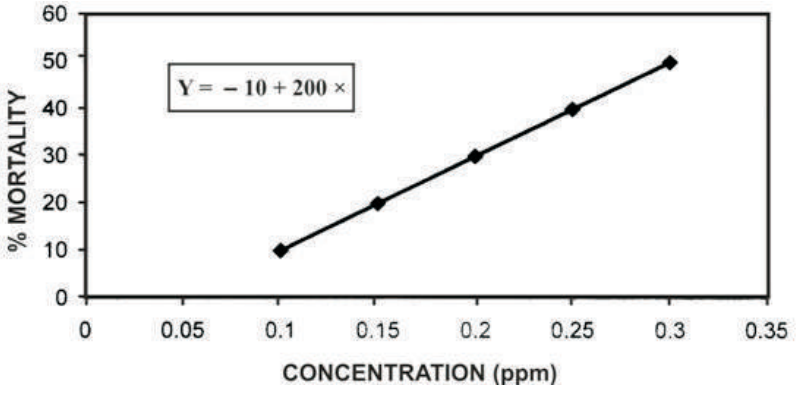
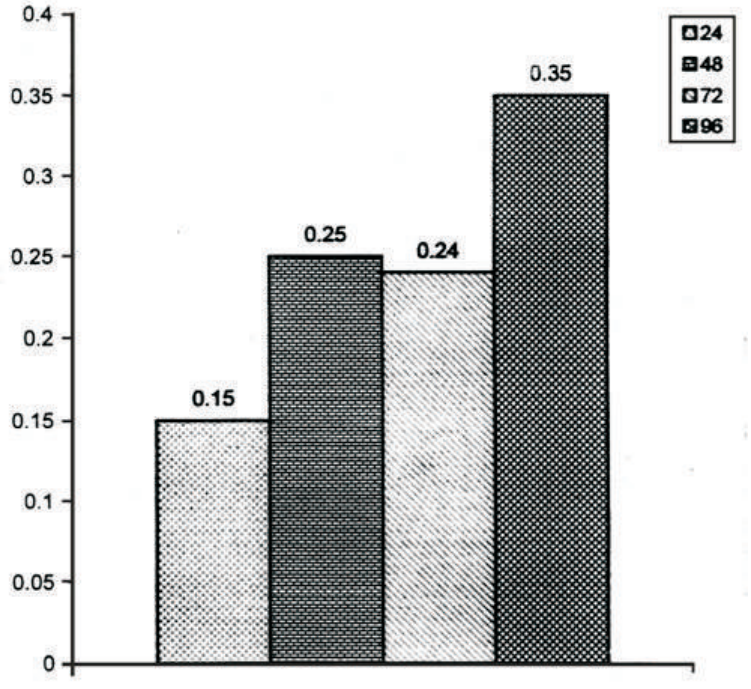


FIGURE – 5
LC_{50} values (ppm) of Fenvalerate for different time intervals obtained by regression equations to *Anabas testudineus*



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