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ORIGINAL ARTICLE

Toxicity Test to Furadan Exposure in *Channa punctatus* in Culture Medium under Laboratory Conditions

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ABSTRACT

The present study assesses the acute toxicity and behavioral alterations due to Furadan, an organophosphate pesticide on Channa punctatus. The sub-lethal concentration of Furadan is 910 mg/L. In the present study the alterations in the hematological profiles were investigated in C. punctatus after exposure to lethal and sub-lethal exposures of Furadan. The bioassay test may be determined on the mortality and behavioral changes. Behavioral bioassay has been widely used in toxicity assessment. Behavioral bioassay is more promising alternatives than lethality evaluating bioassay which are currently used for the risk assessment of toxicant. Behavioral changes provide early warning signals about the health of exposed population which other standard tests do not take in to consideration.

Key words: *Toxicity, Furadan Exposure, Channa punctatus, Culture Medium*

INTRODUCTION

The fishes are important source to provide protein-rich diet and also help to tide over malnutrition in human beings. The recent anthropogenic activities to enhance crop production employed various types of insecticides to control over insect infestation in crop fields. There are several reports on the fish toxicity (Gupta 2003; Zahra and Shreshtha 2006), but most of the works have been carried out on technical grade, while pesticides used in agricultural works are in their commercial formulations. The toxicity of a pesticide is altered in commercial form and the fish susceptibility also frequently influenced by environmental factors (Dalela, *et al*, 1981). The knowledge about lethal concentration of any pesticide it is easier to protect its discharge above the limit. Also, toxicity of pesticide may be classified as extreme, high, moderate, slight and relatively harmless chemicals. The furadan is less toxic than organophosphate and organochlorine pesticides and thus widely applied by farmers. Hence, this study is devoted to find out the median lethal value of pesticide to freshwater fish in controlled environment.

METHODS AND MATERIALS

Healthy and living specimen of *Channa punctatus* varying in weight group between 33.94 to 37.10 gm were procured from local market carefully. They were subjected to quarantine and acclimatization. These fishes were treated with Terramycin solution (15 mg/ litre) in ground water for 48 hours, washed with water and then treated by $KMNO_4$ solution (2 mg/ litre) to remove any possible infections. The fishes were provided grind goat liver prior to change of water in culture. The environmental parameters were monitored with procedures and thereafter stock solution preparewd by dissolving a biocide in minimal acetone (Pickering, *et al*, 1962).

The static bio-assay test for acute or LC_{50} value for different exposures of furadan performed following Doudoroff, *et al*, 1951; APHA 1985). Then, the logarithmic series of doses were used to prepare different concentrations to evaluate lethal concentration for different exposures. The experimental fishes were kept without food to 48 hours prior to bio-assay test and also not during the test along with control fishes. The fishes showing no response to prodding with a glass rod and no respiratory movement was considered as dead and removed immediately.

There are several ways to determine LC_{50} value, however, average of three applied methods was confirmed. The values were determined with graphical interpolation, Dragstedt Behren and probit

analysis methods. In graph interpolation of log10 value of the furadan concentration at one axis and mortality plotted on another axis for 50% mortality. Dragstedt Behren method was applied through formula as:

Log LC50=Log A+50-a/b-a*log of concentration factor

Where,
A = pesticide concentration has a percent kill immediately below 50% mortality,
a = percent kill immediately below 50% mortality, and
b = percent kill observed immediately above 50% mortality.

In Probit analysis, percent mortality values are transformed to Probit with use of probit level plotted as ordinate, while concentrations are converted into log values and plotted as axis. The regression line is drawn by least square method and and LC_{50} values are calculated as described by Finney (1981).
The ventilation rate confined by opercular activity of live fish in each concentration as well as control at different exposures were counted by visual observations using a magnifying glass for 5-10 minutes and average per minute were calculated to fishes.

RESULTS AND OBSERVATIONS

The percent mortality of the fish in different concentrations of the furadan were plotted against the log value and LC_{50} values at selected times periods were read directly and by transferring the log value into the normal value with the help of algorithm table were recorded (Table 1, Figure 1).

Table 1: Definitive test for direct interpolation method

Conc. of toxicant (ml/l)	No. of fishes	24hrs		48hrs		72hrs		96hrs	
		M	% M	M	% M	M	% M	M	% M
0.00010	10	0	0	0	0	0	0	2	20
0.00025	10	0	0	0	0	2	20	3	50
0.00040	10	1	10	2	30	3	60	1	70
0.00055	10	2	20	4	60	2	80	1	90
0.00070	10	7	70	1	80	1	90	1	100
0.00085	10	8	80	1	90	1	100		
0.001	10	8	80	2	100				

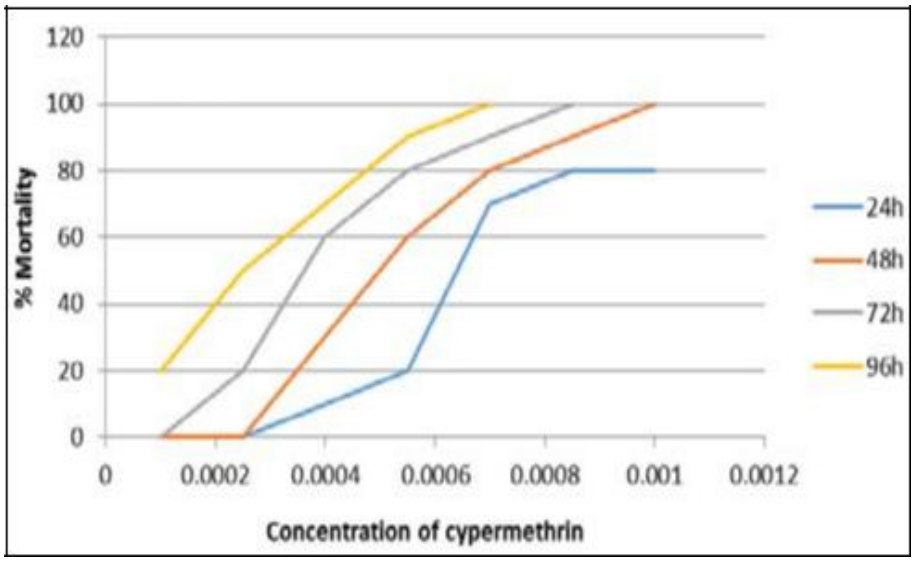

Fig 1: Estimation of LC_{50} at different exposure period

In Dragstedt Behren method, cumulative frequency and percent kill were first determined and then LC_{50} values at selected time intervals were calculated using the formula (Table 2).

The percent mortality were transferred to probit and plotted against log concentration of the biocide, which showed a straight line (Figure 3). Accordingly, the LC_{50} values with their standard error, slope value and regression equations were calculated (Table 3).

The values obtained through different methods showed similarity, however, probit is more appropriate than other methods applied in the study to determine lethality. Accordingly, the median lethal concentration values for 24, 48, 72 and 96 hours obtained as 13.80-21.88, 10.23-16.79, 7.24-13.65 and 5.01-10.96 mg/liter for furadan.

Table 2: Log conc and probit to 24 and 48 hour furadan exposure

S. No.	Conc. 24h	Log Conc.	No. of Fishes	24h			48h		
				% dead	Correct %	Probit	% dead	Correct %	Probit
1	0.00010	-4	10	0	2.5	3.04	0	2.5	3.04
2	0.00025	-3.60206	10	0	2.5	3.04	0	2.5	3.04
3	0.00040	-3.39794	10	10	10	3.72	30	30	4.48
4	0.00055	-3.25964	10	20	20	4.16	60	60	5.25
5	0.00070	-3.1549	10	70	70	5.52	80	80	5.84
6	0.00085	-3.07058	10	80	80	5.84	90	90	6.28
7	0.001	-3	10	80	80	5.84	100	97.5	6.96

Table 3: Log conc and probit to 72 and 96 hour furadan exposure

S. No.	Conc. 24h	Log Conc.	No. of Fishes	72h			96h		
				% dead	Correct %	Probit	% dead	Correct %	Probit
1	0.0001	-4	10	0	2.5	3.04	20	20	4.16
2	0.00025	-3.60206	10	20	20	4.16	50	50	5
3	0.0004	-3.39794	10	60	60	5.25	70	70	5.52
4	0.00055	-3.25964	10	80	80	5.84	90	90	6.28
5	0.0007	-3.1549	10	90	90	6.28	100	97.5	6.96
6	0.00085	-3.07058	10	100	97.5	6.96	100	97.5	6.96
7	0.001	-3	10	100	97.5	6.96	100	97.5	6.96

The response of the pesticide on the fish behavior was depends upon furadan concentration and exposure times. The fishes showed hyperactivity and fast movement initially as evidenced by increased operculum movement and frequent surfacing followed by erratic locomotion, muscular convulsions ultimately resulted into paralysis and bottom surfacing with cutaneous mucus secretion. The frequency of surface breaking also increased 3-4 times than control fishes. The gill ventilation in fishes exposed to different concentrations showed a maximum 11.9 percent increase in lowest concentration and 38.09 percent increase in higher concentration in early exposure, while 10 to 50 percent decrease in later stage of exposure.

DISCUSSION

Furadan continuously pollute the water by its toxic effect on aquatic organisms. The present study was carried out to determine the toxic effect of furadan on *Channa punctatus* that was assessed by the LC_{50} values calculated at different exposure period. The fishes showed mortality at low concentration and with decrease of duration of exposure, the fishes exhibit mortality at higher concentration. It has been reported earlier that pesticide, chemicals and xenobiotic accumulated in natural waters, which results in toxicity to aquatic organisms (Zahra and Vishwakarma, 2010). The values of LC_{50} of furadan obtained by other workers were found to be different for pyrethroid insecticides on different species of fishes (Saha and Kaviraj, 2003). According to Finney's probit

analysis, the LC_{50} values in *Channa punctatus* were found to be 0.00066, 0.00044, 0.00033, and 0.00022 in this investigation. This shows that furadan is highly toxic to fishes. Tiwari *et al* (2012) reported the toxicity of furadan in very low concentration in fingerlings of *Labeo rohita* (Manjulasri and Viraiah, 2014). The result obtained from acute toxicity of furadan on *Tilapia* at 96h LC_{50} value was 5.99 g/L (Sarikaya, 2009). The acute toxicity of fresh water fish *Cirrhinus mrigala* when exposed to 10% furadan was found to be in the range of 2.69– 2.28 ppb after 24 to 96 h exposure (Ayula and Ajni, 2008).

Furadan intoxicated behavioural alternations were reported in several fishes (Manjulasri and Viraiah, 2014). Yaji *et al.* (2011) studied behavioural alternations in *Oreochromis niloticus* juveniles exposed to furadan at different concentrations 0.0007, 0.0008, 0.0009, 0.010 and 0.11 mg/L for 96h (Zahra and Shreshth, 2006). Initially exposed fishes came to surface to engulf air frequently. Hyper excitability was noticed by the jerky and random movement of fishes just after the addition of toxicant. Ojutiku *et al*., (2014) also reported restlessness, loss of balance, excessive accumulation of mucus on skin and jumping. The experimental fishes in this study showed similar observations even at lower concentration of furadan. The changes induced by furadan can be attributed to an increase in physiological stress due to neuronal excitation, low rate of oxygen consumption and histological changes.

Once a pesticide introduced into the water through any sources might affects on trophic structure in more or less extent variable with pesticide concentration and exposure periods. It is well known that water quality have a definite role in heavy metal toxicity (Khangarot and Roy, 1987), but their role for pesticides was yet not be studied. Lloyed and Herbert (1962) have been studied more pronounced toxicity at higher temperature and alkalinity and considered both as important controlling factor.

CONCLUSION

The present study was an attempt to find the toxicity of Furadan on *C. punctatus* and the results conclusively showed that the furadan is highly toxic to fishes even at very low concentration. The study on fishes will be very useful to provide a future understanding of ecological impact.

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