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**Toxicity Evaluation and Behavioural Studies of Fresh Water
Fish *Labeo rohita* Exposed To Cypermethrin (Synthetic
Pyrethroid)**

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Abstract

A short term definitive test by the static renewal bioassay method was conducted to determine the acute toxicity (LC50) of technical grade pyrethroid insecticide, cypermethrin (10%EC) on the fresh water teleost, *Labeo rohita*. Fishes were exposed to various concentration of cypermethrin for 96 hours and the percent mortality was recorded. The LC50 value was found to be 0.06 μ l/L and 1/10th of LC50 0.006 μ l/L was selected as sub-lethal concentration (1, 5, 10, 15 days). *L. rohita* in toxic media exhibited erratic and darting movements with imbalanced swimming activity, which might be due to the malfunctioning of neurotransmitters followed by hyper and hypo opercular activity, loss of equilibrium and mucus secretion all over the body were observed.

Key-Words: Toxicity, Behaviour, Cypermethrin, *Labeo rohita*

Introduction

The pollution of rivers and streams with chemical contaminants has become one of the most critical environmental problems of century. As a result of the pollutants transport from industrial and agricultural areas into the environment and their chemical persistence, many fresh water ecosystems are faced with specially or temporally alarming high levels of xenobiotic chemicals (BRACK et al. 2002, DIEZ et al. 2002).

Due to increasing restrictions on organophosphate pesticides, pyrethroids pesticides are used instead of organophosphates in residence and in the field of agriculture.

Cypermethrin is a synthetic pyrethroid insecticide used to control many pests like moth pests of cotton, pests of fruit and vegetable crops, including structural pest control, residential and garden use. This has resulted in its discharge into the aquatic environment and consequently several laboratory studies have been performed, which evidenced that cypermethrin is extremely toxic to fish even at very low concentration and to aquatic invertebrates also.

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Fish is very sensitive to pyrethroid insecticide because it has slow metabolism and elimination of these compounds is through excretion. *Labeo rohita* is one of the prime cultured fresh water fish and having great economic importance.

Hence, the present study was undertaken to evaluate aquatic toxicity of Cypermethrin based pesticides on the fresh water teleost, *Labeo rohita* exposed to lethal and sub lethal concentration of technical grade Cypermethrin (10%EC).

Material and Methods

Collection and Maintenance of experimental animal

Healthy, fresh fingerlings of *L. rohita* (11.0 \pm 1.5cm in total length, 20.0 \pm 2.0gm in weight) were obtained from Bhadaran fish farm, Bhadariana, Borsad, Anand, Gujarat, India. The collected fishes were maintained into glass aquarium containing 50 L dechlorinated tap water for 7 days to acclimatize under laboratory conditions. The aquarium water was aerated continuously and food was provided in the form of dried, small pellets. Water was change at every 24 hour. Experimental water conditions were water temperature 29.5-30.5 $^{\circ}$ C, pH 7.4-7.5, dissolved oxygen 4.4-6.3ppm, free CO₂ 15.5-17.5ppm, bicarbonate alkalinity 25.5-50.5 ppm.

For the present investigation, technical grade cypermethrin (10% EC) manufactured by United

Phosphorous Limited (UPL) purchased from local market of Bhavnagar, Gujarat, India.

Toxicity Assay

Toxicity experiment was performed by the method of Singh and Agarwal. The fingerlings of *L. rohita* were exposed at different concentration of cypermethrin. Five aquaria were set up for each concentration and each aquarium contains 10 fishes in 50 L dechlorinated tap water. Control fishes were kept in same condition without any treatment.

Mortality was recorded at every 6 hour up to 24 hour exposure period. Fishes were considered dead if they fail to respond to stimulus provided with glass rod.

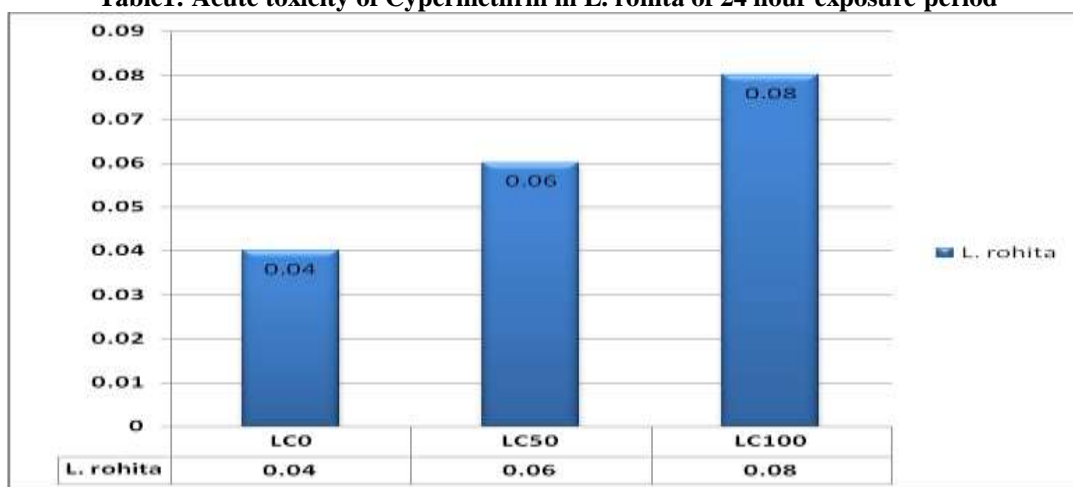
Results and Discussion

Acute toxicity

Acute toxicity test was found in *L. rohita*. In *L. rohita* LC0 was found 0.04µ/L, LC50 was found 0.06µ/L and LC100 was found 0.08µ/L. (Table 1).

Acute toxicity of Cypermethrin in <i>L. rohita</i>			
	LC0	LC50	LC100
<i>L. rohita</i>	0.04	0.06	0.08

Table1: Acute toxicity of Cypermethrin in *L. rohita* of 24 hour exposure period



Graph 1: Acute toxicity test was found in *L. rohita*. In *L. rohita* LC0 was found 0.04µ/L, LC50 was found 0.06µ/L and LC100 was found 0.08µ/L

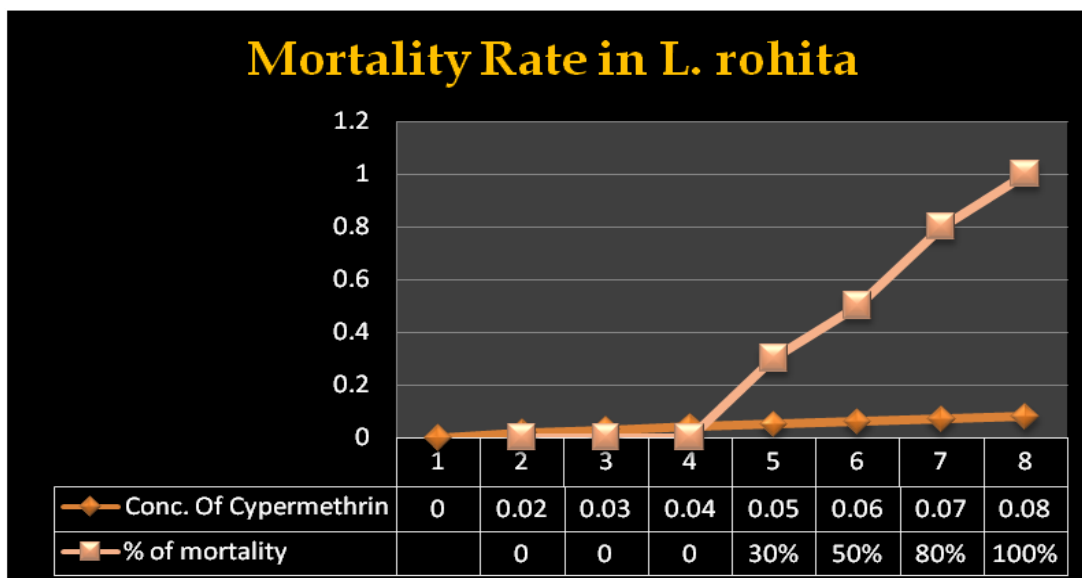
Mortality Rate

Mortality rate in *L. rohita* is shown in below table. Result shows that in *L. rohita* 0% mortality was seen in 0.04µ/L while 50% mortality was found in 0.06µ/L

and 100% mortality found in 0.08µ/L of Cypermethrin.

Table 2: Percent mortality rate in *L. rohita* against conc. of Cypermethrin

Mortality rate in <i>L. Rohita</i>				
Conc. Of Cypermethrin (µ/L)	% of Mortality	No. of exposed fish	Live fishes	Died fishes
0.02	0	10	10	0
0.03	0	10	10	0
0.04	0	10	10	0
0.05	30%	10	7	3
0.06	50%	10	5	5
0.07	80%	10	2	8
0.08	100%	10	0	10



Graph: Mortality rate in *L. rohita* is shown in graph. Results show that in *L. rohita* 0% mortality was seen at 0.04µg/L while 50% mortality was found at 0.06µg/L conc. and 100% mortality found in 0.08µg/L conc

Behavioural Observations

The control fish behaved in a natural manner. i.e., they were active with well coordinated movements. They were alert to slightest disturbance, but in the toxic environment fishes exhibited irregular, erratic and darting swimming movements and they loss equilibrium. They slowly became lethargic, hyper excited, restless and they secreted excess mucus all over their bodies. Opercular movements increased initially in all exposure periods but decreased later steadily in the lethal as compared to sub lethal exposure periods. Gulping air at the surface, swimming on the water surface, disrupted shoaling behaviour was seen on the first day itself in the lethal and sub-lethal exposure period and continued throughout the test tenures. Finally the fish settle down to the bottom of aquarium and died with their mouths open.

The acute test for a long time has been a major component in a toxicity testing. In which acute chemical toxicity is determined as a 96 hour LC50 value however the environmental significance of death of individuals after short term exposure to high concentration is questionable. In contrast to this our results shows cypermethrin is very toxic even at lower concentration (0.06µg/L) for 96 hour LC50. Lethality in the present study is compare with the few previously published studies that exist but that LC50s for all species exceeded this concentration. This can be attributed to the inability of the *L. rohita* to withstand and metabolize the cypermethrin intoxication. The acute toxicity treatments showed strong negative effects on survival as pesticide concentration increased.

This suggests that survival is dose-dependent survival and lethality is concentration graded. The varying degree of mortality reported in this study is consistent with the report of David et al. who reported that differences in an organisms biological adjustment and behaviour response to change in water chemistry.

Changed behavioural responses can be taken as index of the stress felt in the fish exposed to cypermethrin by which they try to reduce excess entry of cypermethrin present in the medium or minimize damage to their body tissues. Similar behavioural changes were also observed in guppy fish *Poecilia reticulata*, after exposure to cypermethrin and permethrin (Li et al. 2005, Baser et al. 2003).

In the present study the control fish behaved normal in a natural condition. i.e., they were alert to the slightest disturbance, but in the toxic environment the fish exhibited irregular, erratic and darting swimming movements and loss of equilibrium. This occurs because it inhibits the AchE activity, leading to accumulation of acetylcholine in the cholinergic synapses, leading to hyperstimulation (Mushigeri and David, 2005). They slowly became lethargic, hyper excited, restless and secreted excess mucus all over their bodies. Mucus secretion in fish forms a barrier between body and toxic media thereby probably reduces contact with the toxicant so as to minimize its irritating effect, or to eliminate it through epidermal mucus. Similar observations made by Rao et al. 2003 and Parma De Croux et al. 2002 in *Prochilodus lineatus* under monocrotophos stress. Opercular movements increased initially in all exposure periods

but decreased later steadily in lethal compared to sub lethal exposure periods. The increased opercular gill movements observed initially may possibly compensate for increased physiological activity under stressful conditions. (Shivakumar and David 2004)

In sub lethal exposure fish's body became lean towards the abdomen position compared to control fish and they were found to be under stress, but this was not fatal. Leanness in fish indicates a reduced amount of dietary protein consumed by the fish under pesticide stress which is immediately utilized and not stored as body mass. (Kalavathy et al. 2001).

Conclusion

The analysis of data from the present investigation demonstrated that cypermethrin is extremely toxic and had a profound impact on behaviour in *L. rohita* in both lethal and sub-lethal concentration. Thus it leads to altered fish physiology.

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