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Studies on toxicity of endosulfan on edible fresh water fish *Nemacheilus botia*

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Abstract

The present paper deals with acute toxicity test carried out for pesticide endosulfan. The LC_{50} value, standard error (for accuracy of variance) and maximum and minimum fiducial limit were calculated. The safe concentration for endosulfan was found 2.038 ml µg/lit. The biochemical component glycogen was studied in the control and polluted water.The glycogen content of liver decreased after acute treatment by endosulfan at 2.024 and 3.039 milli µg/lit conc.

Keywords: *Nemacheilus botia, LC_{50}, Glycogen, Liver, Endosulfan*

Introduction

The marked increase in total pesticides usage and rapid proliferation of synthetic organic compounds has deteriorated the water quality which enters through the agriculture discharge, chiefly the pesticides may reach fresh water bodies by runoff or by accident and ultimately enters the fauna residing there. Today over 1000 chemicals are used against about 2000 pest species.

Fishes are the most widely used organisms to determine the toxicity of water and other pollutant. The wide use of fishes is probably due to their adaptability to laboratory conditions as well as their availability and their varying degree of sensitivity to the toxic substance (Verma *et al.* 1980). Undesirable effects caused by pesticide to the aquatic organisms and their hazard are elegantly reviewed by many workers (Sanger, 1964). Brown (1976) listed 12 basic types of investigations of toxicity; these are for preliminary screening of chemicals, for monitoring influence to determine the extent of risk to aquatic organisms and to determine the component causing death. Determination of acute toxicity is essential for determining the sensitivity of the animals to the toxicants and is also useful for evaluating the degree of damage to the target organs and consequent physiological and behavioral disorders. Toxicity tests are useful for suitability of environmental conditions for aquatic life, favorable and unfavorable environmental factors, such as DO, pH, temperature, salinity or turbidity and effect of environmental factors on waste toxicity. The present study was carried on *Nemacheilus botia* which is a fresh water fish. An attempt has been made to study the changes in biochemical composition in tissues of organs like liver of fish, when it is exposed to different concentrations of endosulfan. Glycogen and proteins have main role in the energy metabolism. Therefore the change in glycogen level was studied in the present investigation.

Materials and Method

Nemacheilus botia were collected regularly in live conditions from River Godavari at the place Nandur Madhmeshwar, a famous bird sanctuary in Nashik district. These fishes were kept in the aquarium containing tap water. Fishes are acclimatized to laboratory conditions. The important parameters like pH, temperature, dissolved oxygen, total hardness were determined for the water as per standard methods (APHA, 1981). Endosulfan is a broad spectrum, extremely toxic organochlorine pesticide that is widely used in India. The effect of 35% endosulfan on biochemical constituents of *Nemacheilus botia*,

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were evaluated by exposing fishes to 1/2, 1/3 … $1/10^{th}$ to sublethal concentrations of endosulfan at 24, 48, 72 and 96 hours. The controlled and treated fishes were used for analysis of biochemical constituent glycogen in liver. The fresh isolated tissues were used for estimation of glycogen according to (Dezwann and Zandee, 1972) and protein according to (Lowry *et al.* , 1951).

The results were statistically analyzed by applying standard deviation and chi-square test. Control animals were treated exactly in the same way as the experimental animals but without toxicants.

Results and Discussion

Pesticides have enormous value in agricultural field to control agricultural pest and diseases. The use of chemicals in the field increases along with overgrowing populations because of which pesticides have become environmental contaminants. The toxicity of particular pollutant depends upon many factors such as animal weight, time of exposure, temperature, pH and hardness of water. The evaluation of LC_{50} concentration of pollutant is an important step, before carrying further studies on physiological changes in animals. In present investigation endosulfan showed LC_{50} gradually reducing from 5.76 to 3.35 milli µg/liter as time duration increased from 24 to 96 hrs, whereas safe concentration was found to be 2.038 milli µg/liter. During the course of study it was noticed that the toxicity increases as the exposure time as well as concentration of pollutant increased. The pesticide endosulfan might affect the nervous tissues by blocking the passage of impure across the synaptic junctions and inhibition of acetyl cholinesterase activity because the mode of action of organochlorine pesticide is indicated through the impairment of nerve tissue (Murthy and Devi, 1982).

Table 1 : Relative toxicity of pesticide Endosulfan when fish *N. botia* were exposed to 24 hrs. to 96 hrs.

Hrs. of Exposure	Regression Equation	LC_{50} ± SE	Variance	Fiducial Limit		Lethal dose ml µg/Lit	Safe Concentration
				M_1	M_2		
24	$Y = 1.14 + 4.9\ \bar{}$	5.76 ±1.32	1.767	-1.840	3.360	114.240	2.038 milli µg/Lit
48	$Y = 0.71 + 6.09\ \bar{}$	4.88 ±0.98	0.967	-1.240	2.618	234.330	
72	$Y = 2.31 + 4.4\ \bar{}$	4.47 ± 0.90	0.898	-1.140	1.590	322.488	
96	$Y = 2.62 + 4.5\ \bar{}$	3.35 ± 1.10	1.212	-1.390	2.790	430.080	

The LC_{50} value of various fish species varies with pesticide to pesticide which is highly useful in the final evaluation of extent of pollution of aquatic environment by agricultural chemicals. Vasait and Patil (2005) investigated the LC_{50} values of organochlorine pesticide and calculated its effect for 7 and 14 days exposure period. The result indicates decrease in LC_{50} concentration with increase in concentration and duration of exposure. Similar results was shown by Joshi (2001) Many chemicals induce similar precipitation of mucous which fills the space between filaments and gill lamellae ultimately affecting the gaseous exchange leading to stasis of blood and death of the fishes. Many researchers suggested that cytological damage to gills, rather than mucous accumulation results in death by asphyxia. Chindah *et al.* (2001) noted that aquatic organisms (shell and for fishes) in direct contact with the medium in addition to breathing and feeding is vulnerable to respiratory tract damage and other organs of the body.

The acute toxicity effects are generally evolved due to action of the pesticides on the target organs.

The results of the acute toxicity test was observed to be for 24, 48, 72 and 96 hours. The LC_{50} values were calculated for 24, 48, 72 and 96 hours by method described by Finney (1971).

The obtained regression equation to pesticide endosulfan for 24, 48, 72 and 96 hours are listed in Table-1. LC_{50} values for 24, 48, 72 and 96 hours exposures to mutation were found as 5.76, 4.88, 4.47 and 3.35 milli μg/lit respectively. The calculated accuracy for log LC_{50} values are summarized in under column variance, which are 1.767, 0.967, 0.898, 1.212 for 24, 42, 72, 96 hours respectively. The standard error for (accuracy of variance) 24, 48, 72 and 96 hours are 1.32, 0.98, 0.90, and 1.10 respectively. The fiducial limits for log LC_{50} value are summarized in Table 1 under the column fiducial limit M_1 and M_2. The 95% confidence of LC_{50} values are (fiducial limit) to pesticide are M_1 (Minimum limit) and M_2 (Maximum limit). The maximum and minimum fiducial limit for 24, 48, 72, and 96 hours log LC_{50} value of endosulfan are -1.840 to 3.360, -1.240 to 2.618, -1.140 to 1.590, and -1.390 to 2.790 respectively.

Table-2: Glycogen content in milli μg/gm of wet liver tissue of *N. botia* in control and endosulfan exposed

Concentration of Endosulfan	24 hours	48 hours	72 hours	96 hours
Control	7.000(± 0.121)	7.000(± 0.121)	6.833(± 0.171)	6.666(± 0.0906)
Endosulfan Conc. 2.024 milli μg/lit.	6.666(± 0.182)	6.331(± 0.207)	6.166(± 0.1314)	5.833(± 0.1052)
Endosulfan Conc. 3.039 milli μg/lit.	6.333(± 0.186)	6.166(± 0.122)	5.833(± 0.171)	5.666(± 0.0192) 3.833

The standard error for (accuracy of variance) 24, 48, 72 and 96 hours are 1.32, 0.98, 0.90 and 1.10 respectively. The fiducial limits for log LC_{50} value are summarized in Table 1 under the column fiducial limit M_1 and M_2. The 95% confidence of LC_{50} values are (fiducial limit) to pesticide are M1 (Minimum limit) and M_2 (Maximum limit). The maximum and minimum fiducial limit for 24, 48, 72 and 96 hours log LC_{50} value of endosulfan are -1.840 to 3.360, -1.240 to 2.618, -1.140 to 1.590, and -1.390 to 2.790 respectively.The safe concentration for endosulfan is 2.038 ml μg/lit. Lethal dose for pesticide are entered in column 'lethal dose', for immediate 100% mortality of fish, the lethal dose was calculated. The lethal doses for pesticide endosulfan at 24, 48, 72, 96 hours exposure are 114.240, 234.330, 322.488 and 430.080 ml μg/lit. respectively. For 100% immediate mortality the fish require highest lethal dose of pesticide endosulfan. The glycogen content of liver decreased after acute treatment by endosulfan at 2.024 and 3.039 milli μg/lit conc.

The glycogen content in liver decreased from 7.000 to 6.666, 7.000 to 6.330, 6.830 to 6.166, 6.666 to 5.833 mg/gm of wet tissue in 24, 48, 72 and 96 hours respectively at 2.024 milli μg/lit. From the present investigation it is quite clear that glycogen content of fish *N. botia* after endosulfan treatment was altered indicating the effect of tested endosulfan. The average glycogen content after acute treatments were decreased. Whatever may be reasons, the decrease in the level of glycogen contents will adversely affect the growth, development and reproduction of organisms, which in turn disrupt the effectiveness of aquatic organisms in biological control programmes.

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