

ACUTE TOXICITY AND BEHAVIORAL RESPONSE OF FRESH WATER FISH *CLARIAS BATRACHUS* WHEN EXPOSED TO MONOCROTOPHOS.

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ABSTRACT

Aquatic environment is influenced by various pollutants like pesticides, heavy metals, detergents, and discharge of effluents from various industries, which induce changes in the behavioral, physiological and biochemical aspects of inhabitants particularly fishes. Aim of present study was to determine acute toxicity of Monocrotophos pesticide to fresh water fish *Clarias batrachus*. The LC₅₀ value for 24, 48, 72 and 96 hrs was found to be 91.39, 83.39, 75.46 and 67.45 respectively. During this acute toxicity study fishes were exposed to different concentrations of Monocrotophos showed behavioral response such as fishes became more active, trying to jump outside aquarium, showed surfacing tendency, some fishes frequently dashed against wall of aquarium, followed jerky movements, erratic swimming, lastly fishes became progressively sluggish and lethargic, loss of sensation of direction, lost their balance and equilibrium, skin discoloration was also observed, excess secretion of mucous was observed all over body, and death of fish occurred and fish sank to bottom of container and turbidity of trough water also increased gradually.

KEYWORDS: Acute toxicity, behavior, *Clarias batrachus*, Monocrotophos

INTRODUCTION

Monocrotophos C₇H₁₄NO₅P is an organophosphate insecticide. It is principally used in agriculture, to control a broad spectrum of pests, including sucking, chewing and boring insects and spider mites on cotton, olives, rice, maize, sorghum, soyabeans and tobacco. The toxicity of this compound is studied by several workers Sekhal *et al.*, 2011; Jeyapriya *et al.*, (2013); Parrish (1995). Acute toxicity of a pesticide refers to the chemicals ability to cause injury to an animal from a single exposure, generally of short duration. The acute toxicity test of a pesticide to fish has been widely used to acquire rapid estimates of the concentrations that cause direct harm to test organisms. Therefore present investigation was designed to study acute toxicity of Monocrotophos to fish *Clarias batrachus* and effect of pesticide Monocrotophos on behavioral response of *Clarias batrachus*.

MATERIALS AND METHODS

Live specimen of *Clarias batrachus* were collected from near- by water bodies of Aurangabad city (Maharashtra) and were brought to laboratory without any mechanical injury. The fishes were treated with 0.1 ppm potassium permanganate solution for any bacterial or viral infection. The fishes were allowed to acclimatize to laboratory condition for a week. During the period of acclimatization the fishes were fed with small pieces of intestine of poultry birds. Aged water was used to maintain fish.

The toxicity tests to calculate LC₅₀ value for 24, 48, 72 and 96 hrs was carried out by desired concentrations of Monocrotophos. The pilot tests were performed for the selection of test concentration. The range of concentration selected ranged 0-100% mortality. Water was changed after every 24 hrs to maintain concentration of pesticide. On the basis of results of pilot tests, series of different concentration grade were prepared. Healthy 10fishes were tested for each concentration for 96 hrs. The bioassays were started in morning and behavioral changes were noted. The mortality and survival was recorded after every 24,48,72 and 96 hrs. The % mortality of each concentration was converted to probit values and plotted against log of concentration Ghosh (1962). The LC₅₀ values were calculated for 24, 48, 72 and 96 hrs by the method of probit analysis (Finney 1971).

Safe concentration for Monocrotophos to fish *Clarias batrachus* was calculated by method described by Hart et al (1945) using the formula.

$$\text{Safe concentration} = \frac{48 \text{ hrs TLM} \times 0.2}{S^2}$$

$$\text{Where } S = \frac{24 \text{ hrs TLM}}{48 \text{ hrs TLM}}$$

Where TLM is median tolerance limit or LC₅₀.

RESULTS AND DISCUSSION

During this investigation, fishes were exposed to pesticide Monocrotophos and LC₅₀ value for 24, 48, 72 and 96 hrs was determined. The LC₅₀ value for 24, 48, 72 and 96 hrs was found to be 91.39, 83.39, 75.46 and 67.45 respectively (Table 1). In the present study it is observed that LC₅₀ value decreased with increase in exposure period.

Table 1. toxicity of Monocrotophos to fresh water fish *Clarias batrachus* at normal laboratory condition of water.

Time of exposure (hrs)	Regression equation Y= Y+b (X-X̄)	Chi-square	LC ₅₀ Values in ppm	Fiducial limit		Safe conc.
				m1	m2	
24	Y=28.480258X-50.8486561	0.0186286	91.39	1.9542303	1.9774363	13.885 9169
48	Y=25.9340X-44.8223855	0.0219009	83.39	1.9137305	1.9393028	
72	Y=22.964891X-38.122843	0.0047502	75.46	1.8572197	1.8861803	
96	Y=20.50308X-32.5012851	0.0046751	67.45	1.8060308	1.8384692	

Tilak and Kumari (2009) carried out acute toxicity of organophosphate Nuvan to grass carp *Ctenopharyngodon idella* and LC₅₀ values were found to be 13.1, 10.9, 9.8, and 6.5ppm at 24,48,72 and 96 hours respectively. Vasait and Patil (2005) investigated the LC₅₀ values of organophosphate pesticide to *N. botia*. The result show a decrease in LC₅₀ value with increase in exposure period.

Jagtap (2011) exposed fresh water bivalve, *Lamellidens marginalis* to Tributyltin chloride and calculated LC₅₀ value for 24, 48, 72 and 96 hrs and observed that LC₅₀ values decrease with increase in exposure period.

Shejule *et al.*, (2006) reported decrease in LC₅₀ value with increase in exposure period, when freshwater prawn *Macrobrachium Kistnensis* was exposed to tributyltin chloride up to 96 hrs.

During this investigation, the mortality of fish *Clarias batrachus* was observed to increased significantly with increasing the toxicant concentration and time of exposure for pesticide Monocrotophos. Table (2).

Venkateswarlu Naik *et al.*, (2016) observed that mortality rate gradually increased, as the concentration of the toxicant increased in the experiment, when fish *Catla catla* was exposed to organophosphate pesticide Phorate 10 % CG. Tripathi (2015) observed that with increasing concentration of pesticide Phenthoate the mortality rate of *Labeo rohita* fingerlings gradually increased.

Chandrasekara *et al.*, (2016) observed that mortality rate of tilapia fingerlings increased both with the Trebon concentration and the exposure period, and also observed that 96 hrs LC₅₀ value is 8 times smaller than the 24 hrs LC₅₀ value, i.e. increase in exposure period decreases the LC₅₀ value. Ali Rani (2009) showed that the increase in exposure period decreases the LC₅₀ value.

In this study, Safe concentration of Monocrotophos to fish *Clarias batrachus* was found to be 13.8859169.

Water used for toxicity evaluation was with physical parameters such as alkalinity 240 mg/L, hardness 350 mg/L as CaCO₃, pH 7-7.4, Dissolved Oxygen 6.1 to 6.8 ppm. These water parameters were analyzed by using standard methods APHA (2008) (table 3).

Table 2. Summary of toxicity with Monocrotophos in *Clarias batrachus* (at normal laboratory condition of water)

Sr.No	Concentration in ppm	% Survival and Mortality							
		24 hrs		48hrs		72 hrs		96hrs	
		S	M	S	M	S	M	S	M
01	59.5	100	00	100	00	100	00	100	00
02	61.5	100	00	100	00	100	00	80	20
03	63.5	100	00	100	00	100	00	70	30
04	65.5	100	00	100	00	100	00	60	40
05	67.5	100	00	100	00	90	10	50	50
06	69.5	100	00	100	00	80	20	40	60
07	71.5	100	00	100	00	70	30	30	70
08	73.5	100	00	100	00	60	40	20	80
09	75.5	100	00	90	10	50	50	10	90
10	77.5	100	00	80	20	40	60	10	90
11	79.5	100	00	70	30	30	70	00	100
12	81.5	90	10	60	40	20	80	----	----
13	83.3	90	10	50	50	10	90	----	----
14	85.5	80	20	40	60	10	90	----	----
15	87.5	70	30	30	70	00	100	----	----
16	89.5	60	40	20	80	----	----	----	----
17	91.5	50	50	10	90	----	----	----	----
18	93.5	40	60	00	100	----	----	----	----
19	95.5	30	70	----	----	----	----	----	----
20	97.5	20	80	----	----	----	----	----	----
21	99.5	00	100	----	----	----	----	----	----

Table 3. Physical and chemical parameters of water used for toxicity test.

Sr. No.	Parameters	
1	Temperature	24°C (22-26°C)
2	Alkalinity (Bromocresol)	240 mg/L
3	Total Hardness as CaCO ₃ (EDTA)	350 mg/L
4	Dissolved Oxygen	6.1 – 6.8 ppm
5	pH	7 – 7.4 ppm
6	Weight of fish	260 – 300 gm
7	Length of fish	280 – 320 cm

During this study of acute toxicity, behavioural response was studied when exposed to Monocrotophos, the fishes became more active initially, trying to jump outside container, showed surfacing tendency, some fishes frequently dashed against wall of container, followed by jerky movements, erratic swimming, lastly fish activity became progressively sluggish and lethargic, loss of sensation of direction, lost their balance and equilibrium, Skin

discolouration was also observed, excess rapid gill movements, vertical hanging, fading of body colour, increased opercular movements, excess secretion of mucous all over body, restlessness, finally fish became motionless with widely open mouth prior to death at the bottom of the container (Table 4).

Table 4. Physical reaction of *Clarias batrachus* to pesticide Monocrotophos during acute toxicity study.

Sr. No.	Behaviour in control water.	Behavioral changes in treated fish.
1	Fishes were neither trying to jump out, nor showed surfacing tendency.	trying to jump outside container, showed surfacing tendency.
2	No jerky movements, nor erratic swimming, fish activities were normal.	Jerky movements, erratic swimming, lastly fish activity became progressively sluggish and lethargic.
3	No loss of sensation, no loss of balance and equilibrium.	loss of sensation of direction, lost of balance and equilibrium.
4	No skin discoloration, gill movements were normal.	Skin discoloration, rapid gill movements, vertical hanging.
5	Fish activities were normal, no secretion of mucous all over body, no restlessness.	excess secretion of mucous all over body, restlessness, finally fish became motionless with widely open mouth prior to death.

Venkateshwarlu Naik *et al.*, (2016) observed fishes were hitting against container wall, irregular swimming, loss of balance, restlessness, body of fish became pale in colour and surfacing activity, secretion of mucous was observed all over the body and fish death occurred and fish sank to bottom of container, and turbidity of trough water also increased gradually by fish *Catla catla* when exposed to organophosphate pesticide Phorate 10% CG.

Similar observations such as fish irritation, very fast opercular movement, violent action of pelvic fins and spreading fins, excess mucus secretion, loss of equilibrium were observed by Ganeshwade R.M. *et al.* (2012) when fresh water fish *Channa striatus* was exposed to Endosulfan.

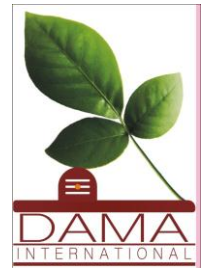
Christopher Didigwu Nwani *et al.*, (2013) observed abnormal behavior, trying to avoid test water by swimming fast, jumping, displaying erratic and vigorous jerky movements, faster opercular movement, hyper excitation, surfacing and gulping of air by fresh water African catfish *Clarias gariepinus* when exposed to Organophosphorus pesticide, Termiphos. Tripathi *et al.*, (2015) observed fishes frequently coming to surface water, trying to jump out of water, loss of equilibrium erratic and darting swimming movements.

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