

## Effects of Monocrotophos 36%EC on Haematological Indices in the Fresh Water Fish *Labeo Rohita* (Hamilton)

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### ABSTRACT

In toxic effects of Monocrotophos 36%EC, at different time intervals (24, 48, 72 & 96 hours) in the haematological indices like Haemoglobin content (Hb), Total Red Blood Corpuscle Count (RBC), Total White Blood Corpuscle (WBC) count and Mean Corpuscle Hemoglobin (MCH) content in the blood of fish *Labeo rohita*, was exposed to sub lethal concentration (0.40 ppm). The total amount of haemoglobin content (8.2, 8.1, 7.9, 7.7 mg.), and red blood corpuscles (2.84, 2.70, 2.60, 2.50 million) were gradually reduced. Meanwhile, White Blood Corpuscles (9780, 9820, 9980, 10110) count were increased. Mean Corpuscle Hemoglobin (MCH) count (28.87, 29.12, 30.82, 30.38 mg.) was gradually increased by hours to hours. In the present investigation it clearly indicates that very adverse effect of Monocrotophos 36% EC on fish.

**Keywords:** Monocrotophos 36%EC, Hematological indices – Hb – RBC – WBC - MCH

### INTRODUCTION

Population explosion, rapid industrialization and consequent anthropogenic stress on the environmental degradation, particularly environment have resulted in alarming levels of pollution and of the aquatic environment. A wide range of toxic chemicals called pesticides are used to control or eradicate the pests of agriculture, horticulture and other species of crops throughout the world.

The uses of pesticides have served to improve our crops from the ravages of insect pests. Control of pests has always been one of the major factors in enhancing the agricultural production (Hague and Freed, 1975).

Commonly using pesticides can be harmful to living organisms, pets and their environment. Part of the problem is the toxicity of some pesticides but even more important is the sheer volume of pesticides. The increasing use of pesticides causes chemical pollution results potential health hazards to live stock, especially to fish, birds, frogs, and mammals (Nagaraju and Venkata Rathnamma, 2013).

Repeated exposure to sub-lethal doses of some pesticides can cause physiological and behavioral changes in fish that reduce populations, such as abandonment of nests and

broods, decreased immunity to disease and increased failure to avoid predators (Veeraiah *et al.*, 2012). Pesticide poisoning is an important cause of the morbidity and mortality in developing countries. Now –a- days, farmers are using verity of pesticide, insecticide, herbicide and etc. for their agricultural field.

The pesticides are mainly two type sorganochlorine and organophosphate. Is ong organophosphate pesticides, Monocrotophos is one of the important pesticideto control insect pest and indiscriminately being used by Indian farmer. The residues of pesticide reaches to the environment by direct application, spray drift, aerial spraying from the atmospheric precipitation and runoff from agricultural lands where they ravage the biotic life (Thangniponet.*al.*,1995). When the pesticides come in contact with internal organs, irreversible changes in metabolic activities, many pesticides have been reported to produce a number of biochemical changes in fish at sub-lethal levels (Nagaraju and Venkata Rathnamma, 2013).

Blood plays an important role in the normal functioning of the body of any organism. Blood is a patho physiological reflector of the whole body and therefore, blood parameters are important in diagnosing the structural and functional status of fish exposed to toxicants

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(Adhikari *et al.*, 2004). Hematological parameters can provide satisfactory information on the physiological response of fish to environmental stressors for two major reasons, namely, the close association of the circulatory system with the external environment and the ease of availability of fish blood (Cazenave *et al.*, 2005; Houston, 1997).

### MATERIALS AND METHODS

#### Selection and Acclimatization of the Experimental Fish

Healthy freshwater fish, *Labeo rohita* of the weight ( $15 \pm 1$ g) and length ( $8.0 \pm 0.5$  cm) were collected from the commercially cultured local pond. Fishes were screened for any pathogenic infections. Glass contamination aquaria were washed with 1% KMnO<sub>4</sub> to avoid fungal contamination and then dried in the sun light. The healthy fishes were transferred to glass aquaria (35 x 20 x 20 cm) containing dechlorinated tap water and water was changed every day. They were maintained in the aquaria at room temperature  $28 \pm 2^\circ\text{C}$ . Fishes were acclimatized to laboratory conditions for 10 to 15 days prior to experimentation. The rate of mortality during acclimatization was less than 10%. They were regularly fed with commercial food. Feeding was stopped two days before the experiment so as to reduce the function of excretory substance in the test tank (Arora, *et al.*, 1972).



#### Experimental fish *Labeorohita*

The fish were introduced into 10 liter capacity containers of water containing specific Organophosphate Monocrotophos 36% EC was exposed to sub lethal concentration (0.40 ppm). All such treated fishes were separated from the experimental containers and blood samples were

collected from six experimental individuals in each group at each time with an interval of 24 hours. Fishes were collected and gently wiped with a dry cloth to remove water. Caudal peduncle was cut with a sharp blade and the blood was collected in a watch glass containing 6% EDTA (Ethylene Diamine Tetra Acetic acid), as an anticoagulant. The blood was mixed well with the EDTA solution by using a needle and the samples were used for estimation of Haemoglobin content (Hb), Total Red Blood Corpuscle (RBC) count, Total White Blood Corpuscle (WBC) count and determination of Mean Corpuscle Hemoglobin (MCH).

The Sahli-Hellige method was followed for haemoglobin determination. For Total Red Blood Corpuscle (RBC) count, a method by Christensen *et al.* (1978) was followed. The standard RBC diluting pipette and a 1:200 dilution was used for the RBC count. Yokayama's fluid was used for dilution. For counting the total number of WBC, the pipette with white bead was used. The number of cells present in the four large corner squares marked by capital letter 'L' was counted and multiplied by 103 which give the total number of WBC per cubic millimeter of blood.

The mean corpuscle hemoglobin (MCH) was determined as follows:

$$\text{M.C.H} = \frac{\text{Hemoglobin (g/100ml)} \times 10}{\text{RBCs million's per Cummm}} \times 100$$

And the values are expressed as pictogram (Seiverd, 1964).

### RESULTS

#### Haemoglobin Content

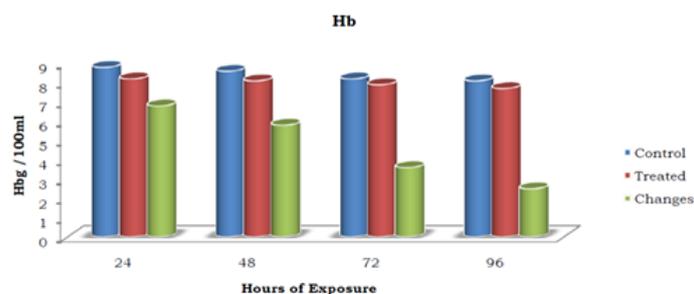
The haemoglobin content in the blood of *Labeo rohita* are shown in the Table.1 and Fig.1. In this study, fish treated with sub lethal concentration (0.40 ppm) of Monocrotophos 36% EC, the hemoglobin content (mg) and hours of exposure were 8.2, 8.1, 7.9, 7.7 and 24, 48, 72, 96 respectively. The total amount of hemoglobin content was gradually reduced by hour to hour.

**Table1.** Total Haemoglobin content in the blood of *L. rohita* control and treated with (0.40 ppm) Monocrotophos 36%EC, at various time intervals. (Values are expressed in mgs.)

Hours of Exposure	Control	Treated	% of Changes
24	$8.8 \pm 0.12$	$8.2 \pm 0.72$	6.8
48	$8.6 \pm 0.12$	$8.1 \pm 0.30$	5.8
72	$8.2 \pm 0.72$	$7.9 \pm 0.26$	3.6
96	$7.9 \pm 0.30$	$7.7 \pm 0.06$	2.5

Values are Mean  $\pm$  SD of six observation; - or + indicate percentage decrease or increase over control.

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**Fig1.** Total Haemoglobin content in the blood of *L. rohita* control and treated (0.40 ppm) with Monocrotophos 36%EC, at various time intervals. (Values are expressed in mgs.)

### Red Blood Corpuscle (RBC) Count

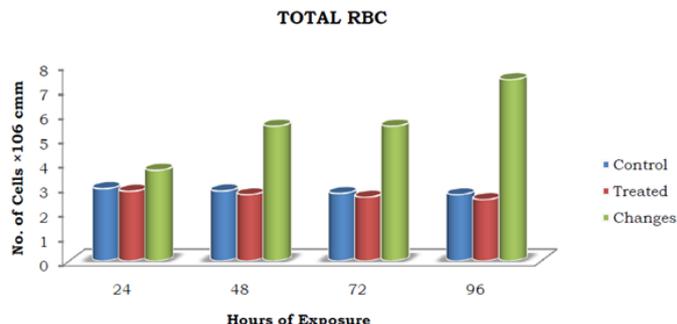
The Red Blood Corpuscle (RBC) count in the blood of *Labeorohita* are shown in the Table.2 and Fig. 2. In this study fish treated with sub lethal concentration (0.40 ppm) of Monocrotophos 36% EC, the red blood

corpuscle count and Hours of exposure were 2.84, 2.70, 2.60, 2.50 million and 24, 48, 72, 96 respectively. The Red Blood Corpuscle was increased by hour to hour.

**Table2.** Total Red Blood Corpuscle (RBC) contents in the Blood of *L.rohita*control and treated (0.40 ppm) with Monocrotophos 36% EC at various time intervals.(Values are expressed in million).

Hours of Exposure	Control	Treated	% of Changes
24	2.95 ± 0.52	2.84 ± 0.23	3.7
48	2.85 ± 0.75	2.70 ± 0.35	5.5
72	2.75 ± 0.75	2.60 ± 0.47	5.5
96	2.70 ± 0.59	2.50 ± 0.75	7.4

Values are Mean ± SD of six observation; – or + indicate percentage decrease or increase over control.



**Fig2.** Total Red Blood Corpuscle (RBC) contents in the Blood of *L. rohita* control and Treated (0.40ppm)with Monocrotophos 36%EC. at various time intervals.(Values are expressed in million).

### White Blood Corpuscle (WBC) count

White Blood Corpuscle (WBC) count in the blood of *Labeo rohita* are shown in the Table.3 and Fig. 3 in this study fish treated with sub lethal concentration (0.40 ppm) of

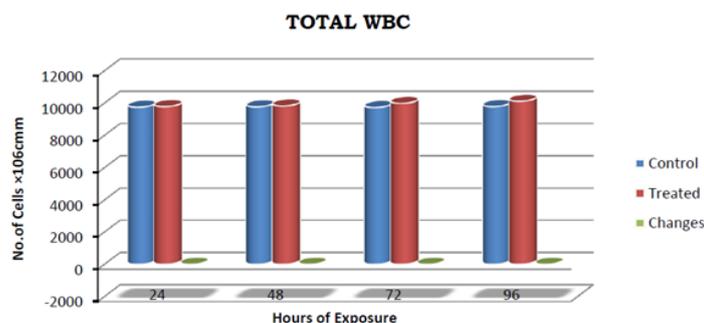
Monocrotophos 36% EC, the White Blood Corpuscle count and hours of exposure were 9780, 9820, 9980, 10110 and 24, 48, 72, 96 respectively. White Blood Corpuscles were gradually increased by hour to hour.

**Table3.** Total White Blood Corpuscle (WBC) content in the Blood of *L.rohita* control and (0.40ppm) treated with Monocrotophos 36%ECat various time intervals.

Hours of Exposure	Control	Treated	% of Changes
24	9740 ± 0.12	9780 ± 0.25	-0.41
48	9765 ± 0.15	9820 ± 0.87	-0.56
72	9770 ± 0.85	9980 ± 0.90	-2.14
96	9780 ± 0.13	10110 ± 0.88	-3.37

Values are Mean ± SD of six observation; – or + indicate percentage decrease or increase over control.

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**Fig3.** Total White Blood Corpuscle (WBC) content in the Blood of *L.rohita* control and (0.40ppm) treated with Monocrotophos 36%EC at various time intervals.

### Mean Corpuscle Hemoglobin

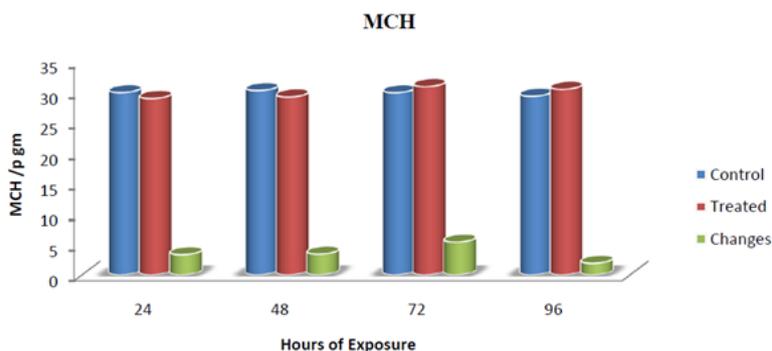
The total Mean Corpuscle Hemoglobin (MCH) count in the blood of *Labeo rohita* is shown in the Table.4 and Fig. 4. In this study fish treated with sub lethal concentration (0.40 ppm) of

Monocrotophos 36% EC, the Mean Corpuscle Hemoglobin count and hours of exposure were 28.87, 29.12, 30.38, 30.82, and 24, 48, 72, 96 respectively amount of Mean Corpuscle Hemoglobin count gradually increased by hour to hour.

**Table4.** Total Mean Corpuscle Hemoglobin (MCH) contents in the Blood of *L.rohita* control and (0.40ppm) sublethal concentration of monocrotophos 36%EC. at various time intervals. (values are expressed in pictogram).

Hours of Exposure	Control	Treated	% of Changes
24	29.83 ± 0.1	28.87 ± 0.1	3.3
48	30.17 ± 0.1	29.12 ± 0.3	3.4
72	29.81 ± 0.7	30.38 ± 0.2	1.9
96	29.25 ± 0.7	30.82 ± 0.0	5.3

Value are Mean ± SD of six observation – or + indicate percentage decrease or increase over control



**Fig4.** Total Mean Corpuscle Hemoglobin (MCH) contents in the Blood of *L.rohita* control and (0.40ppm) treated of monocrotophos 36%EC. at various time intervals. (Values are expressed in pictogram).

### DISCUSSION

The hematology of fish has gained recognition as an applied science. Hematology tests have become important diagnostic tools. Recent studies have shown that the hematological indices may be equally valuable, in indicating the disease of the stress in the fish. The composition of blood of fish varies with the changing conditions of the environment and response immediately to any changes in water quality because of intimate contact. Out of varied hematological indices differential red blood corpuscle counts are of immense physio

pathological importance. In the present investigation, an attempt has been made to elucidate the effect of pesticide with different hours on certain physiological properties of the blood of fish *Labeo rohita*.

In the present study, the hemoglobin content were gradually decreased at 96 hours administration of monocrotophos 36%EC, because the hemoglobin content directly relationship for RBC content it indicate count leading to anemia as a result of inhibition of erthropoiesis, haemosynthesis and increase in the rate of erythrocyte destruction in

haemopoietic organs. Similar observations were reported for juvenile *C. gariiepinus* separately treated with Lambdacyhalothrin, Cypermethrin and Deltamethrin pesticides (Yekeen, 2009). Similar report have been reported by Houston (1997), Cazenave *et al.*, (2005) and Fukushima *et al.*, (2012).

The *Labeo rohita* exposed to sub lethal concentration (0.40 ppm) of monocrotophos 36% EC resulted in a significant decreased in RBC's count in the 96 hours treated fish blood, leading to anemia as a result of inhibition of erthropoiesis, haemosynthesis and increase in the rate of erythrocyte destruction in haemopoietic organs. Similar report have been reduced the red blood cell count and hemoglobin content. This indicates the high doses of pesticides produced anaerobic condition and limit the oxygen carrying capacity and there by decrease the mobility. Similar reduction in RBC was reported for Cypermethrin treated *Labeo rohita* (Das and Mukherjee, 2003), fresh water common carp (*Cyprinus carpio* L.) treated with diazinone (Svoboda *et al.*, 2001) and African cat fish (*C. gariiepinus*) treated with diazinone (Adedeji *et al.*, 2009). Other toxicants (effluent and heavy metals) had also been reported to have similar reduction effects on RBC of fishes (Goel *et al.*, 1981; Kumari and Banerjee, 1993; Deoi *et al.*, 2004). Reduction in Hb content of treated *C. gariiepinus* may be an indication of decline in hemoglobin synthesis as well as reduction in oxygen carrying capacity which May perhaps is as a result of interference of endosulfan with hemoglobin synthesis pathway. Significant reduction in Hb content and erthyrocyte count in the blood of a freshwater fish, *Sarotherodon mossambicus*, on exposure to an organophosphate (dimecron) and carbamate (cumin L.) pesticides had been reported (Ramaswam *et al.*, 1996). The reduction in values obtained for hematological parameters of treated fish in this study showed that the physiological activities of the treated fish were affected.

At 96 hours treated fish, the total white blood corpuscle count has remarkably an increased in their number in the different hours at sub lethal concentration. Same results noticed by Venkataramana *et al.*, (2006) the white blood corpuscle count were in ceased to toxicity of malathion on the gobid fish, *Glossogobius giuris* (Ham) and Sudha Summarwar (2012), hematological investigation on *Labeo rohita*

following chronic exposure to Zn & Cu. Also studied by Patil and David (2007). Hepatotoxic potential of malathion in the freshwater teleost, *Labeo rohita*.

A linear relationship was established with respect to pesticide monocrotophos and total white blood corpuscle. The constant increasing in the differential count clearly indicates that the pesticide stress certainly stimulate the white blood cells to produce more at all time of exposure. It has been suggested that the enumeration of differential cell ratio counts provides of useful diagnostic procedure to assess the physiological stress in the fish.

The amount of mean corpuscle hemoglobin (MCH) in the blood of *Labeo rohita* exposed to monocrotophos found to increased at different study periods. Same findings given by Binukumari and Vasanthi (2013), on the effect of pesticide dimethoate 30% EC, in the blood of fresh water fish, *Labeo rohita*.

## CONCLUSION

It can be concluded from this study that monocrotophos 36% EC has the potential to impair the physiological activities of the organism which led to changes observed in hematological indices. Persistent exposure to monocrotophos 36% EC may thus lead to mortality. Monocrotophos 36% EC usage in India should therefore be controlled in order to reduce its potential risk to human health.

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