

**Research Article**

**ACUTE AND SUBCHRONIC EFFECT OF MONOCROTOPHOS ON  
HAEMATOLOGICAL INDICES IN *CATLA CATLA* (HAMILTON)**

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**ABSTRACT**

The present study was aimed to find out the effect of sublethal concentration of monocrotophos on freshwater fish Indian major carp *Catla catla*. The fishes were exposed to sublethal concentration (0.0250mg/l) for 5 days (acute) and 30 days (sub chronic). Haematological parameters such as TEC, PCV, Hb, MCH, MCHC, TLC, MCV and differential blood cell count were carried out to assess the effect of pesticide on fish physiology. Significant changes were observed in the blood cell indices of fishes exposed to pesticide compared with control. A statistically significant decrease in the TEC, Hb, PCV, MCH, MCHC, TLC and increase in MCV were observed in the 5 and 30 days exposure compared with control. The present results indicate that the pesticide monocrotophos induce changes in the blood indices which may be used as indicator for the health status of fish.

**Key words:** Monocrotophos, acute concentration, subchronic concentration, *C. catla*, haematological parameters.

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**INTRODUCTION**

Pesticides are used worldwide in agriculture and aquaculture to control the pest and insects (Enis Yonar *et al.*, 2012). Organophosphate pesticides like ethion, dimethoate, monocrotophos and chlorpyrifos are widely used for paddy crop pests. The widespread use of synthetic organic pesticides over decades has led to their frequent exposure in the environment. Also acute and chronic exposures of humans to pesticides occur during their commercial production and their application. Synthetic pesticides are deliberately sprayed on crops or agricultural land to increase food production but these agrochemicals are not very selective in producing their effects. They are toxic to many non-target species and contaminate the environment (Singh *et al.*, 2006). Usage of pesticides in the ecosystem leads to development of various types of morphological, physiological, biochemical and

behavioral changes in individuals. Potentially hazardous environmental toxicants like pesticides display a broad spectrum of biological effects, being toxic not only to target organisms but also to humans (Jamil *et al.*, 2007).

Aquaculture apart from agriculture is common in India, where fish, the non-target organisms are directly exposed to pesticides used for the control of insects and pests. The pesticides affect the survival, growth rate, fecundity and reproductive activity of fish. Toxic substances even in very low concentration which is sublethal have been reported to interfere with basal metabolism and suppressed reproduction, steroidogenesis, lipid metabolism, degenerative changes in gonadotropin cells and reduction in interstitial cells size, gonadotropin levels act as reproductive biomarkers and also as endocrine disruptors (Singh and Vandana Singh, 2006).

The exposure of fish to several types of chemical agents may induce changes in several haematological parameters, which are frequently used to evaluate fish health. Haematology has been widely used for the detection of physiopathological alterations following different stress conditions. Therefore, haematological techniques are the most common method to determine the sublethal effects of pollutants (Modesto and Martinez, 2010; Kumar *et al.*, 2011). Monocrotophos (MCP) [O, O -dimethyl-O -(2 methyl carboxy-1methyl vinyl) phosphate], an organophosphate ester, is one of the OP insecticides with systemic and contact action. The present study was aimed to study the effect of monocrotophos at acute and subchronic concentration on haematological parameters.

## MATERIALS AND METHODS

The Indian major carp *C. catla* were collected from local commercial fish ponds and acclimatized to laboratory conditions by keeping them in cement tanks. The fish were fed with supplemented commercial feed. For the experimental fishes weighing about 20-25 g were selected. Feeding was stopped one day before commencing of experiments in order to minimize the quantity of excretory products in the test tank. The physico chemical conditions of well water used in the present study has the following characteristics; Dissolved oxygen 7.5 - 8 ppm; Salinity 0.3 - 0.5 ppm; Alkalinity 252 mg/l as CaCO<sub>3</sub>; Hardness 373mg/l; as CaCO<sub>3</sub> PH: 7.4 to 7.7; Temperature : 28 ± 2°C. Based on the percent mortality values the median lethal concentration values of monocrotophos for 6, 12, 24, 48, 72, 96 and 120 hours for *C. catla* were calculated by following the method of Litchfield and Wilcoxon, (1949). The experiments were carried out in sublethal concentration (0.0250mg/l) of monocrotophos for 5 days (acute) and 30 days (sub chronic).

### Collection of blood sample

Blood was collected from control and monocrotophos treated groups by cardiac puncture. Plastic disposable syringe fitted with 26 gauge needle which was already moistured with heparin was used. The collected blood was expelled into separate heparinised plastic vials

and kept immediately on ice. The whole blood was used for the estimation of haematological analysis.

### Haematological analysis

Total erythrocyte count (TEC) and Total leukocyte count (TLC) were counted by haemocytometer method (Rusia and Sood, 1992). Hb concentrations were estimated by Cyanmethaemoglobin method (Drabkin, 1946) and Packed Cell Volume (PCV) was determined by the microhematocrit method (Nelson and Morris, 1989). Erythrocyte indices like Mean Corpuscular Value (MCV), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Haemoglobin Concentration (MCHC) were also calculated according to standard formulas (Lee *et al.*, 1998).

### Statistical analysis

The data was statistically analyzed by statistical package SPSS version 16, in which data was subjected to one way ANOVA. Comparisons were made at the 5% and 1% probability level.

## RESULTS AND DISCUSSION

In the present study, *C. catla* exposed to sublethal concentration of monocrotophos (0.0250mg/l) for 5 days (Acute) and 30 days (sub chronic). The haematological parameters such as TEC, PCV, Hb, MCH, MCHC, TLC and MCV were estimated and compared with the control the results were represented in the Fig (1-8). The total erythrocyte count (TEC) were significantly (P<0.05) decreased with 5 days and 30 days exposure compared with control. The percent of change over control was -7.8 and -19.5 for 5 and 30 days. Likewise the packed cell volume (PCV), haemoglobin level (Hb), Mean corpuscular haemoglobin (MCH), Mean corpuscular haemoglobin concentration (MCHC), Total leucocytes (TLC) were significantly (P<0.05) differs from the control. The percent of change over control were -9.5 -16.7 for PCV; -15.4, -23.6 for Hb; -7.6, -8.7 for MCH; -7.1, -10.4 for MCHC; -0.7, -14.0 for TLC respectively. In contradictory to the above the MCV significantly increased in the fishes exposed to pesticide; the percentage change over control was +0.59, +3.5 for 5 and 30 days. The neutrophil increased

significantly from control; 59.9%, 65% and 70% for control, 5 and 30 days. The eosinophil level was decreased in exposures, 24.9%, 20%, 18% for control, 5 and 30 days respectively. No significant alteration was found in the monocytes. Basophil percentage was not differ from the control in 5 days exposures while in 30 days a significant decreases was noticed 9.9%, 10% and 7% for control, 5 and 30 days respectively. The lymphocytes level was significantly increased in both 5 days and 30 days exposures. The mean percentage was 64.9%, 73% and 80% for control, 5 and 30 days respectively.

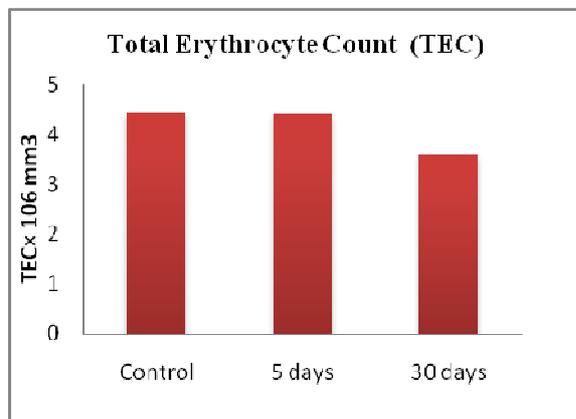
In the present study, decreased in TEC, Hb PCV, MCH, MCHC, TLC were observed in *C. catla* exposed to sublethal concentration of monocrotophos for 5 and 30 days. Similar reports were reported on the fishes exposed to different type of pesticides. Tyagi *et al.*, (1989) found decreases in erythrocytes. Hb, MCH and MCHC and increases in WBC, PCV, MCV in *Ophiocephalus punctatus* exposed to endosulfan. *Cyprinus carpio* exposed to atrazine decrease in ertherocytes, Hb and an increase in WBC (Ramesh *et al.*, 2009), consistent report were also reported by various authors *Cirrhinus mrigala*, to Neem extract; (Saravanan *et al.*, 2011); *C. catla* and *Labeo rohita* fingerlings to cypermethrin (Vani *et al.*, 2012 ; Das and Mukherjee, 2003) *Saccobranchus fossilis* exposed to chlordane (Verma *et al.*, 1979); *Clarias gariepinus* to diazinon (Adedeji *et al.*, 2009); *Clarias gariepinus* and *Cyprinus carpio*; to chlorpyrifos, (Ogueji Okechukwu *et al.*, 2007 Yonar *et al.*,

2012); *Oreochromis mossambicus* to endosulfan (Kumar *et al.*, 2011); *Cyprinus carpo* to lindane (Saravanan *et al.*, 2011).

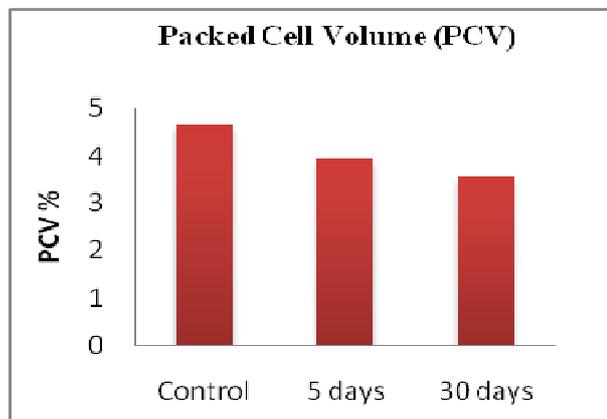
In the present study MCV level was increased in the 5 and 30 days exposure when compared with the control. Zhang *et al.*, (2007) found no change in the MCV, MCH and MCHC in *Carassius auratus* injected with microcystins.

In *Saccobranchus fossilis* exposed to chlordane shows no change in the MCHC level (Verma *et al.*, 1979). Adedeji *et al.* (2009) also support the above findings in *Clarias gariepinus* exposed to diazinon. The changes in erythrocytes counts may be due to damage to RBC by the monocrotophos toxicity.

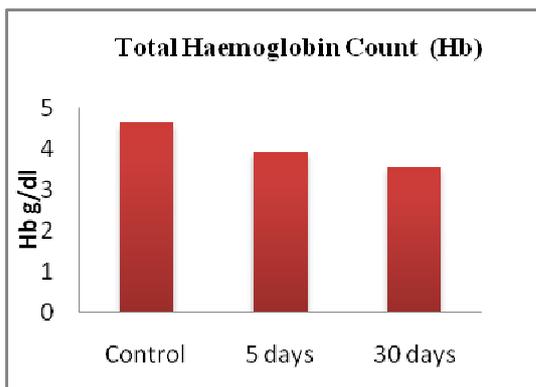
In the present study, the TLC decreased in 5 and 30 days compared with control. Compared with control the total leucocytes counts in 5 days exposure was not distinct where as 30 days a significant different exists. Consistent supports also made for other fishes to different pesticides (Verma *et al.*, 1979; Adedeji *et al.*, 2009; Neeraj kumar *et al.*, 2011). Contradictory to the above the several results show a significant increase in the TLC and differential counts in the polymorphonuclear leucocytes (Enis yonar *et al.*, 2012; Saravanan *et al.*, 2011; Das and Mukherjee, 2003). Increase in TLC may be due to lymphopoiesis or enhance release by lymphocytes from lymphomyeloid tissues. There are various factors to cause anemia and other change in animals such as hemorrhage, hemolysis and renal diseases (Zhang *et al.*, 2007).



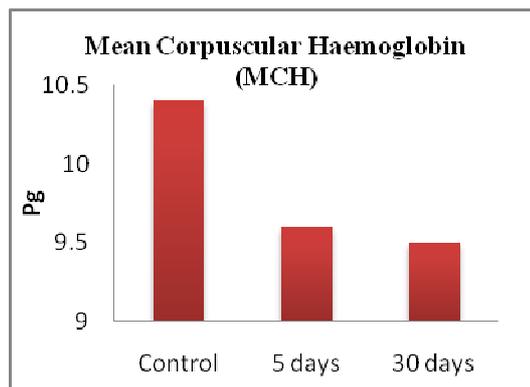
**Figure 1**



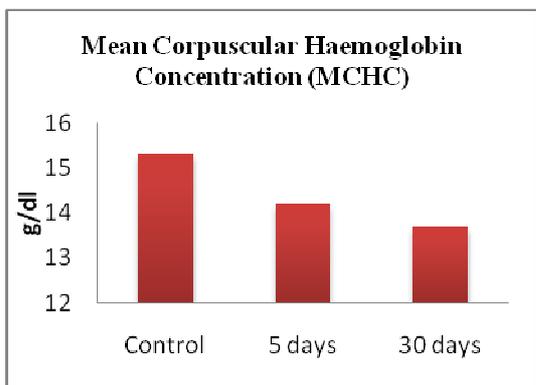
**Figure 2**



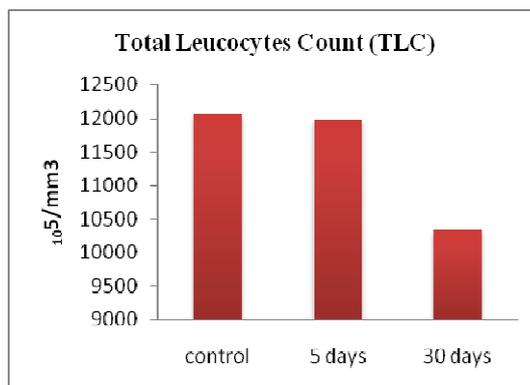
**Figure 3**



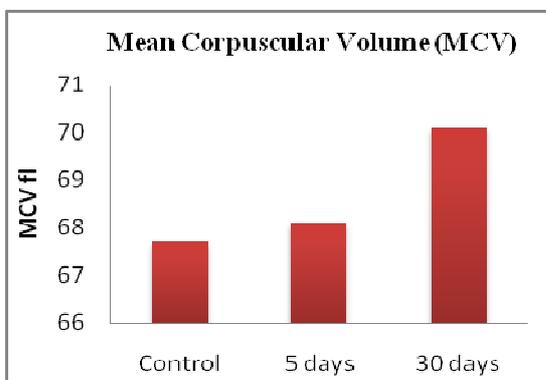
**Figure 4**



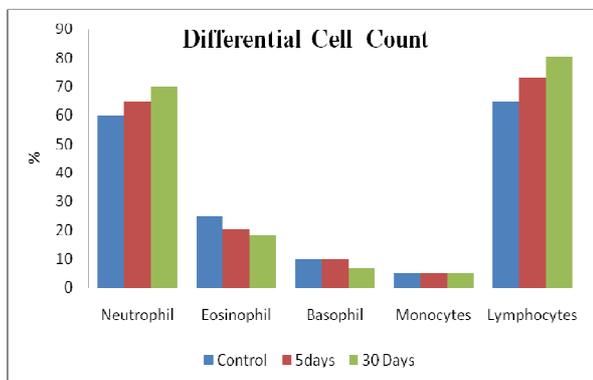
**Figure 5**



**Figure 6**



**Figure 7**



**Figure 8**

**CONCLUSION**

In the present study, monocrotophos induced the changes in the blood indices of *C. catla*, which may be used as indicator of pesticide pollution in the aquatic environment.

**CONFLICT OF INTERESTS**

The author declares that there is no conflict of interests associated with this article.

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**REFERENCES**

Adedeji, O.B, Adeyemo, O.K and Agbede, S.A., 2009. Effects of diazinon on blood

- parameters in the African catfish *Clarias gariepinus*. *African J. Biotech.*, 8(16): 3940-3946.
- Das, B.K. and Subhas Chandra Mukherjee, 2003. Toxicity of cypermethrin in *Lebeo rohita* fingerlings biochemical enzymatic and haematological consequences. *Comp. Biochem. Physiol.*, 134: 109-121.
- Drabkin. D.L., 1946. The crystallographic and optical properties of the hemoglobin of man in comparison with those of other species. *J. Biol. Chem.*, 64:703-723.
- Enis Yonar, M., Serpil Mise Yonar, Mevlit Sener, Ural Sibel Silici and Mustafa dusukcan, 2012. Protective role of propolis in chlorpyrifos-induced changes in the haematological parameters and oxidative antioxidative status of *Cyprinus carpio*. *Food Chem. toxicol.*, 50: 2703-2708.
- Jamil. K., Prabhavathy Das, G., Shaik A.P., Dharmi, S.S. and Sudha Murthy, 2007. Epidemiological studies of pesticide exposed individuals and their clinical implications. *Curr. Sci.*, 92(3): 340-345.
- Kumar, N., Prabhu, P.A.J., Pal, A.K., Remya, S., Aklakur, M., Rana, R.S., Gupta, S., Raman, R.P. and Jadhao, S.B., 2011. Anti-oxidative and immuno-hematological status of tilapia *Oreochromis mossambicus* during acute toxicity test of endosulfan. *Pestic. Biochem. Physiol.*, 99: 45-52.
- Lee, R.G., Foerster, J., Jukens, J., Paraskevas, F., Greer, J.P., and Rodgers, G.M., 1998. *Wintrobe's-Clinical Hematology*, 10<sup>th</sup> En. Lippincott Williams and Wilkins, New York, USA.
- Litchfield, J.T. and Wilcoxon, F., 1949. A simplified method of evaluating dose effect experiments, *J. Pharmacol. Exp. Ther.*, 96: 99-113.
- Modesto, K.A., and Martinez C.B.R., 2010. Effects of Roundup Transorb on fish: hematology, antioxidant defenses and acetylcholinesterase activity. *Chemosphere*, 81: 781-787.
- Neeraj Kumar, P. Antony Jessu Prahbu, Pal, A.K., Remya. S., Md. Aklakur, R.S. Rana, Subodh Gupta, Raman, R.P. and Jadhao, S.B., 2011. Anti-oxidative and immune-hematological status of *Tilapia (Oreochromis mossambicus)* during acute toxicity test of endosulfan. *Pestic. Biochem. Physiol.*, 99: 45-52.
- Nelson, D.A and Morris M.W., 1989. Basic Methodology. Chap. 27. Hematology and Coagulation, part IV, In: *Clinical Diagnosis and Management by Laboratory Methods*. Nelson, D.A., Henry, J.B. seventieth ed. Henry, J.B, W.B. Saunder Company., Philadelphia, U.S.A. pp: 578-625.
- Ogueji Okechukwu, E., Auta, J. and Balogun, J.K., 2007. Effects of acute nominal doses of chlorpyrifos-ethyl on some haematological indices of African catfish *Clarias gariepinus*- Teugles. *J. Fisheries Int.*, 2(2): 190-194.
- Ramesh, M., Srinivasan, R. and Saravanan, M., 2009. Effect of atrazine (Herbicide) on blood parameters of common carp *Cyprinus carpio* (Actinopterygii: Cypriniformes). *Afr. J. Environ. Sci. Technol.*, 3(12): 453-458.
- Rusia, V. and Sood S.K., 1992. Routine haematological tests, in: *Medical laboratory technology*, Kanai, L. Mukerjee, (Edn.), I. Fifth reprint, Tata McGraw Hill Publishing Company Limited., New Delhi, 252-258.
- Saravanan, M., Prabu Kumar, K. and Ramesh, M., 2011. Haematological and biochemical response of freshwater teleost fish *Cyprinus carpio* (Actinopterygii: Cypriniformes) during acute and chronic sublethal exposure to lindane. *Pestic. Biochem. Physiol.*, 100: 206-211.
- Singh, P.B. and Vandana Singh, 2006. Impact of endosulfan on the profiles of phospholipids at sublethal concentration in the male *Heteropneustes fossilis* (Bloch) *J. Environ. Boil.*, 27(3): 509-514.
- Singh, M, Sandhir, R. and Kiran R., 2006. Erythrocyte antioxidant enzymes in toxicological evaluation of commonly used organophosphate pesticide. *Indian J. Exp. Boil.*, 44: 580-583.
- Tyagi, M. Agrwal V.P. and Nandini, 1989. Haematological abnormalities induced in *Ophiocephalus punctatus* by endosulfan. *Recend Trends Toxicol.*, 81-84.
- Vani, T. Saharan, T. N. Roy Ritesh Ranjan, S. D., Pal A. K. and Siddaiah Rajesh Kumar, G.M., 2012. Alteration in haematological and biochemical parameters of *Catla catla*

- exposed to sub-lethal concentration of cypermethrin *Fish Physiol. Biochem.*, DOI 10.1007/s10695-012-9650-0.
- Verma, S.R, Bansal, S.K, Gupta, A.K and Dalela R.C., 1979. Pesticide induced haematological alterations in fresh water fish *Saccorbranchus fossilis*. *Bull. Environm. Contam. Toxicol.*, 22: 467-474.
- Zhang, X., Ping Xie, Dapeng Li, and Zechao Shi, 2007. Hematological and plasma biochemical response of crucian carp *Carassius auratus* to intraperitoneal injection of extracted microcystins with the possible mechanisms of anemia. *Toxicol.*, 49: 1150-1157.