Methyl Parathion Induced Haematology Profile Alteration Of Climbing Perch, *Anabas testudineus* (Bloch.)

ABSTRACT

Water pollution from agricultural wastes sources such as pesticides is now considered to be a major problem in worldwide. The present study includes the alterations induced by chronic (21 days) exposure of the fish Anabas testudineus to a sublethal concentrations (0.047 ppm conc.) of methyl parathion on the haematology profile. The induced fish shows significant alteration in the haematological parameters such as decreasing value in Hb, RBC, WBC while increasing value observed in DLC, Neutrophil, Monocytes and Eosinophil. The methyl parathion induced fishes may have various haematological diseases: Erythropoesis, anaemia, Leucocytopaemia, Neutropaemia, Lymphopaemia, Eosinophilia and Erythropaemia. Therefore, for the optimum growth performance of fish cultivation in contaminated water with pesticide, methyl parathion concentration should not be more than 0.047 ppm.

Key words: *Anabas testudineus*, DLC (Differential leucocytes count), PCV (packed cell volume), haematology profile, methyl parathion and pesticides.

INTRODUCTION :

The use of chemicals in field is viewed as a panacea to improve the productivity of crops. As a result, the application of chemical pesticides to improve crop health and yield has increased worldwide. Pesticides are used to control pests in field for the crop production and vector control for the public health. It has been observed that not all the pesticides applied will reach the targeted organisms. It has been estimated that only approximately 0.3 % of pesticides applied reach the target organisms, whereas 99.7 % contaminates the surrounding environmental, such as air, soil, and water, through runoff, spray drift and leaching. Methyl parathion is an organochlorine pesticide have high insecticidal property and low cost production make them worldwide popular. But due to its high persistent in nature and toxicity on non target organisms it is now banned.

Anabas testudineus (Bloch.), locally known as “kawai” is belonging to order Perciformes and family Anabantidae. This is an important fish of paddy field culture in wetland region of this subcontinent. This is also subjected to severe effect of pesticides on fishes when huge application of pesticides in fields.

Therefore, in current paper an effort has been made to illustrate the methyl parathion induced haematological profile alterations of air breathing climbing perch, *Anabas testudineus.*

MATERIALS AND METHODS :

The climbing perch, *Anabas testudineus*, live fish were procured from the local fish market, Darbhanga and brought to lab in open container. The healthy fish were measured 10±2 cm and weight 32±2 g, washed with 0.1% $KMnO_4$ to remove dermal infection if any. Acclimatization for 15 days before experiment started. Fish was fed with commercial feed (28% crude protein) through the experiment period at the rate 3% of body weight. No aeration was done and follows the methods of APHA (1985).

The LC_{50} values of methyl parathion were determined for 24, 48, 72 and 96 hours following the static bioassay methods of APHA, AWWA & WPCF (1985). The resulted LC_{50} values for given period were 0.35 ppm, 0.25 ppm, 0.13 ppm and 0.095 ppm respectively. The sub-lethal concentration 0.045 ppm was determined (Hart *et al.* 1945). Ten fish were treated with concentration 0.047 ppm of methyl parathion and along with ten fish were taken as control for 21 days. On 21st day the fish were anaesthetized with 1:4000 MS 222 (tricane, methane, sulfonate, sandoz) for two minutes and blood samples were extracted at the site of caudal dorsal of the test fish. The haematological parameters were estimated haemoglobin, RBC, WBC, Lymphocytes, neutrophil, monocytes, basophil, eosinophil and determination of PCV (packed cell volume) as follow the methods (Akela *et al.* 1996; Shrivastav, 1979).

MCV (fl) = [PCV (%) x 10] / [RBC count in millions/mm^3]

MCH (pg) = [HB (g/dl) x 10] / [RBC count in millions/mm^3]

MCHC (g/dl) = [HB (g/dl) x 100] / [PCV (%)]

Results and Discussion:

The current study undertaken was the alteration in haematological profile of the fish, *Anabas testudineus* induced to (0.047 mg/l) sublethal concentration of methyl parathion for 21 days. The result in Table -1 showed a highly significant ($P<0.001$) decreases was observed in haemoglobin (Hb) of induced fish 5.78±0.10 gm/dl than control 8.89 ±0.05 gm/dl. The present study revealed the decreases in haemoglobin (Hb) (5.78±0.10 gm/dl) of methyl parathion induced fish that was conformity with the works of Raizada and Gupta, (1982) in their study found a decrease of haemoglobin in the fungicide RH-216 induced fish, *Trichogaster fasciatus*. Similar alteration in haematological parameters in *Channa striata* was also studied by Sasikala *et al*. (2011). Arjun *et al.* (2009) have

observed highly significant (P<0.001) as a decreased level of haemoglobin in the chromium exposed fish, *Clarias batrachus*. Similar haematological alterations results were observed by earlier workers with various toxicants treated fish; Hb decline was reported by Revathi *et al.* (2003), Shipra *et al.* (2005), Bruska *et al.* (2005), Anwar and Choudhary (2009). Roy and Nath, (2011) also reported similar haematological changes in case of Thiamethoxam treated *Oreochromis niloticus.*

The study revealed that RBC count in control fish was 5.35±0.05 x 10^6µl while in treated fish 4.11±0.05 x 10^6µl. The result showed decreasing a significant value (P<0.001) of RBC count in treated fish. Verma *et al.* (1982) also reported the alteration in RBCs count and haemoglobin concentration in *Mystus vittatus* induced by pesticides and infection of parasites. That physiological change was caused of deleterious effect of toxicants on the erythropoietic tissue of fish. *Heteropneustes fossilis* induced by pesticide, malathion showed decrease in RBC count from 6,400,000 to 3,460,000/cm in LC_{50} 96 hr at 7.6 ppm reported by Mishra and Srivastava, (1983). Muthalagi (2006) has found similar nature of decrease RBC count under sewage treatment to the fish *C. mrigala.* The present study showed also conformity with Arjun *et al.* (2009) observed a decline in RBC under the treatment of chromium to the fish *C. batrachus.* Recently Pratibha and Kumar (2013) have observed the *H. fossilis* (Bloch) induced to mercury chloride showed similar decline nature of RBC.

The Table -1 showed in treated fish the values of Neutrophil, Monocytes and Eosinophil were increasing such as 13.12±0.05, 7.0±0.05 and 3.2±0.05 in compare to control fish value such as 5.45±2.05, 4.0±0.05, 2.2±0.05 respectively. The value of Neutrophil was highly significant (P< 0.001), while Eosinophils showed significant value (P < 0.01). Whereas Basophil has decreasing value 1.4 ±0.02 in treated fish than 1.8 ±0.02 control. That has been found non significant (P < 0.05).

The Table-1 revealed that in treated group the DLC (Differential leucocytes count) of Lymphocytes showed value was decreases 32.0±0.02 from control 52.13±2.40. The Lymphocytes value showed significant (P < 0.01).

The Table-1 showed that the PCV (Packed Cell Volume) value was decreases in treated fish group 12.85±0.03 while in control fish group 34.91±0.06. The PCV value showed significant (P < 0.01).

During present study the WBC decreases are close conformity with various workers, under the treatment of fertilizers, pesticides, alkaloids to fishes or mammals. In fishes Muthalagi (2006) has been reported similar decrease of WBC under domestic sewage to the fish *C. mrigala.* Recently Arjun (2010) has explained similar decrease of WBC under chromium exposure to *Clarias batrachus.* The present findings are conformity with various mammals, such as rat, rabbit etc. under the exposure of metals, pesticides, alkaloids etc. The present findings are conformity with Revathi *et al.* (2003), Shipra *et al.* (2005), Anwar and Choudhary (2009). Pratibha and Kumar (2013) have explained exposure of mercury chloride to the fish *H. fossilis.* On the basis of above facts it is quite clear that WBC plays a very important role in the defense mechanism of body. A decrease in WBC count in exposed fish is termed as leucopaemia. Another observation support the present work, Vasait and Patil (2005) found decreasing lymphocyte count in *Nemacheilus botia* fish induced to organophosphorous insecticide.

The differential count (DC) of leukocytes was found a reliable haematological index to investigate the environmental contamination by various pollutants (Goger and Sawant, 1989). Sharma and Gupta (1982) found considerable lymphocytosis; i.e., within 6 days, lymphocytes increased from 33 to 72% when fish exposed with CCl_4 at concentration 0.03 and 0.06 ml/100 g body weight at intervals of 3 days. The methyl parathion induced fish were showed that DLC- Neutrophil, Monocytes and Esnophil increase while Lymphocytes, Basophil decreases in present study has close conformity with earlier works like under exposure of sewage (Muthalagi, 2006), chromium exposed to fish (Arjun, 2010) and mercury chloride induced to the fishes (Pratibha and Kumar, 2011). Pratibha (2013) reported that induced *Heteropneustus fossilis* to murcury chloride showed haematological alteration. Recently, the induced European whitefish to propofol showed the significant reduction in the counts of lymphocytes, neutrophils and monocytes in haematology profile reported by Gomulka *et al.* (2014). Neutropaemia might be under exposure of methyl parathion under 21 days increased secretion of Adraline (Table:-1).

The haematological profile parameters like Hb, RBC, WBC were found decreases while in DLC, Neutrophil, Monocytes and Eosinophil values increases. Further Lymphocytes and PCV values decrease. There were three ways as significant, highly significant or non-significant resulted by analysis of obtained haematological data. The methyl parathion induced fishes showed various physiological disorder in form of Erythropoesis, anaemia, Leucocytopaemia, Neutropaemia, Lymphopaemia, Eosinophilia and Erythropaemia.

Muthalagi (2006), Arjun (2010) and Pratibha (2013) found haematological changes in fishes under exposure of sewage, chromium as well as cadmium chloride that results conformity of present work. Fish induced to pesticide, methyl parathion showed increase in MCH and MCHC levels which may be due to increased haemolysis of RBCs and the reduction in the Hb concentration resulted by a decrease in cellular blood iron. The present work also conformity of work of Revathi *et al.* (2003), they observed similar in tannery effluent induced fish a decrease in PC, MCV, MCH and MCHC. The tannery effluent concentration influences the haematology profile. Similarly Arjun (2010) observed a significant decrease in PCV, MCV, MCH and MCHC of *Clarias batrachus* induced to chromium. Under the Cadmium chloride exposure to the fish, *H. fossilis* (Bloch) same decrease of PCV, MCV, MCH and MCHC found reported by Pratibha and Kumar (2013). In contrary found an increase in MCV, MCH and MCHC levels of *C. gariepinus* exposed by manganese (Olojo and Ladeji 2012). No significant effect on monocytes and basophils in *A. testudineus* under exposure of sub-lethal concentrations of methyl parathion used in present study. The reduction in basophils and lymphocytes and an increase in monocytes and neutrophils counts were found in present work. Whereas an increase in lymphocyte, eosinophils and monocytes with a decrease in neutrophils and basophils concentration in *Garra gotyla gotyla* exposed to various concentrations of manganese was reported by Sharma and Langer (2014).

TABLE-1

Haematological profile changes in methyl parathion induced *Anabas testudineus*

Variable		Methyl parathion (96 hrs) exposure
Parameter	**Control**	**20 mg/l**
Blood Hb (gm/dl)	8.89 ±0.05	5.78±0.10 ***
TEC(RBC) (10^6 cell/mm^3)	5.35±0.05	4.11±0.05 ***
WBC (10^4 cell/mm^3)	4.92±0.05	2.25±0.05 ***
Neutrophil (10^4 cell/mm^3)	5.45±2.05	13.12±0.05 ***
Lymphocytes (10^4 cell/mm^3)	52.13±2.40	32.0±0.02 **
Monocytes (10^4 cell/mm^3)	4.0±0.05	7.0±0.05 *
Eosinophil (10^4 cell/mm^3)	2.2±0.05	3.2±0.05 **
Basophil (10^4 cell/mm^3)	1.8 ±0.02	1.4 ±0.02 *
PVC (%values)	34.91±0.06	12.85±0.03 **

MCV(fl/cell)	150.25±0.86	156.00±1.35
MCH(pg)	42.53±1.02	47.20±1.43
MCHC(g/dl)	24.05±1.05	29.38±1.05

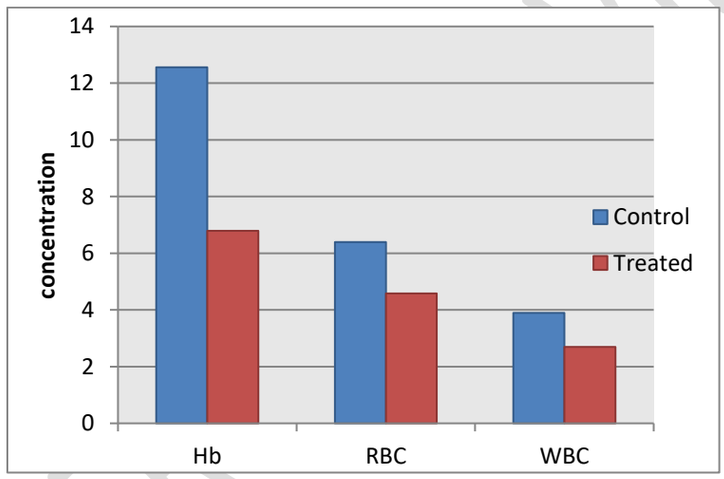
Values are mean ± SE of 5 individual observations:-

* *P<0.5 Non Significant,*

** *P<0.01 Significant,*

*** *P<0.001 Highly Significant*

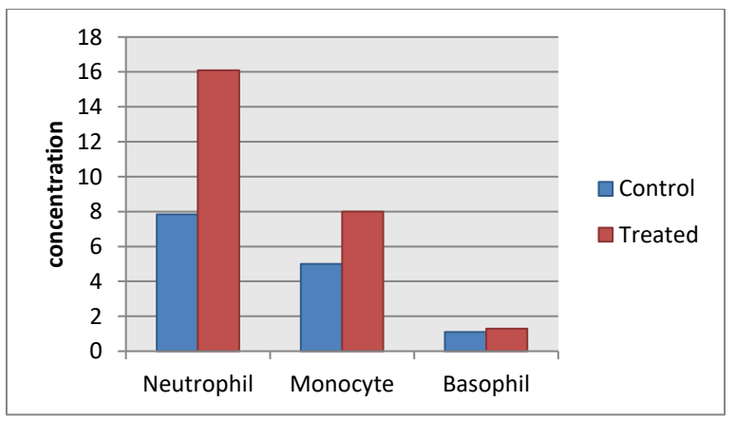
Showing the effect of Methyl parathion on Hb, RBC, WBC in *Anabas testudineus* (96 hrs) ***P < 0.001.



PARAMETERS

Figure-2

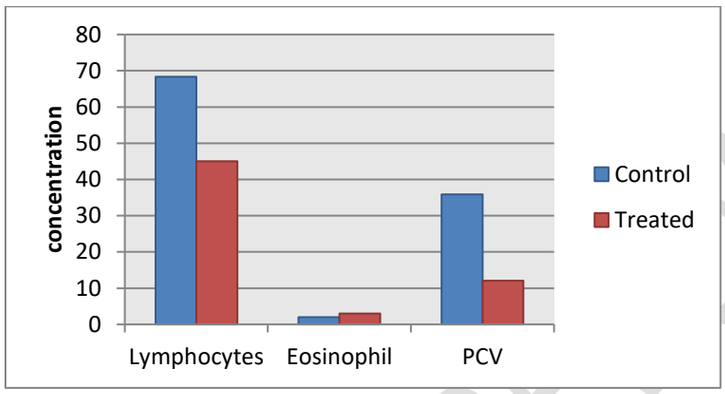
Showing the effect of Methyl parathion on Neutrophil, Monocytes, Basophil in *Anabas testudineus* (96 hrs) *P<0.05, *** P<0.001.



PARAMETERS

Figure:-3

Showing the effect of Methyl parathion on Lymphocytes, Eosinophil, PCV, in *Anabas testudineus* (96 hrs) ** P<0.01.



PARAMETERS

CONCLUSION:

In present study we found that methyl parathion pesticide is a toxic for aquatic organisms and *Anabas testudineus* (average weight 30.0± 4.0 g) culture at water contaminated with this at the concentration of < 0.047 mg/l may be suitable for optimum growth performance and survival rate than other water conditions.

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