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# STUDIES ON THE HAEMATOLOGICAL PARAMETERS OF *ANABAS TESTUDINEUS* (BLOCH, 1792) AFTER LONG TERM EXPOSURE TO METHYL MERCURY

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**Abstract:** Effect of sublethal methylmercury toxicity on the haematological parameters in *Anabas testudineus* was studied during long term (21 days) exposure at a concentration of 0.16 $mgL^{-1}$. Blood samples were collected from 4 fishes each from experiment and control groups. The parameters analyzed were: total red blood cell count (RBC), haemoglobin concentration (Hb), haematocrit (Hct), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC). There was a decrease in the haemoglobin value but the rest of the parameters were comparable to the control at the end of 21 days of exposure.

***Key words:*** Heamatology, mercury, toxicology, Anabas, haemoglobin

The discharge of methylmercury through effluents into the environment has been causing severe damage in various ecosystems. There have been cases like Minamata (Harada, 1995) and Niigata, Japan (Tsubaki, 1979) during the 1950s; Iraq in 1970s (Bakir *et al.*, 1973) as well as in Canada have projected the risk caused by methylmercury pollution to the biosphere. Methylmercury is one of the most toxic chemicals known to man. It is formed by conversion of elemental and inorganic mercury by bacteria present in the water column and sediment of the aquatic ecosystems. Methylmercury has the ability of bioaccumulation upto 100,000 ppm in the body especially in fish (USPHS, 1997) which ends up at the table for human consumption.

Toxicity of various chemicals and pollutants have been studied by using various fish species under laboratory conditions. The mortality, behavioural changes, reproductive stress and changes in histology can be utilized to evaluate the changes caused by the toxicant. Cellular constituents of the blood have been studied and are of importance in the physiological evaluation of animal blood parameters. They are affected due to the presence of chemicals and toxicants in the aquatic environment (Juneja and Mahajan, 1983; Ranzani-Paiva *et al*., 1997). Haematological studies have been used for the detection of changes due to stress conditions like heavy metal presence (Nussey *et al*., 1995). Many species of fishes have been used to decipher the impact of exposure to mercury on the haematolgical parameters of fishes including *Aphanius dispar* (Hilmy *et al.,* 1980), *Tilapia mossambicus* (Menezes and Quasim, 1984), *Pleuronectes platessa* (Fletcher and White, 1986), *Oreochromis aureus* (Allen, 1994), *Ctenopharyngodon idella* (Shakoori *et al.,* 1994), *Acipenser baeri* (Miknakov and Lapirova, 1996), *Hoplias malabaricus* (Oliveira-Ribeiro *et al.,* 2006) and the common indigenous freshwater fish *Channa punctatus* (Sastry and Sharma, 1980; Juneja and Mahajan, 1983; Gupta and Kumar, 2016).

*Anabas testudineus* (Bloch, 1792) is an indigenous air breathing fish belonging to the family Anabantidae commonly found in the freshwater ponds and streams. It is a hardy food fish capable of surviving 6 – 10 hours without water (Hughes and Singh, 1970) and a common occupant of fresh

water ecosystems receiving industrial effluents. The present study was undertaken to understand the impact of long term exposure of methyl mercury on the haematological parameters of *A. testudineus*.

Experiments were performed in the Department of Aquatic Biology and Fisheries with a photoperiod of 13L/11D, water temperature of 27 ± 1.7 C and pH was 7.13 ± 0.3. The fish utilized for the exposure were *A. testudineus* (climbing perch) of mean weight 11.024 ± 2.61 gm and mean length of 8.465 ± 0.622 cm and were acquired from a commercial fish farm. The fishes were acclimatized for 20 days in FRP tank and fed commercial feed. The chemical used was Methylmercury iodide from Alfa Aesar, (Japan). A stock solution of 160 $mgL^{-1}$ was prepared with 5 ml absolute ethanol and then made to final volume of 1000 ml with double distilled water. This solution was diluted into 2 aquaria (10 litres) to form a final concentration of 0.16 $mgL^{-1}$. This concentration is 1/100 $LC_{50}$ value as the estimated $LC_{50}$ for the present experiment was 16 $mgL^{-1}$. The number of fish in each experimental tank was 4. Two tanks with 4 fishes each were maintained as the control. The water in the tanks was replaced once in a week. The exposure duration was 21 days and the fishes were sampled at the end of experimental period. The fish were fed once in a week before replacing the experimental water. For haematological analyses, the blood was collected from 4 fishes each, from experiment as well as control groups into heparinized centrifuge tubes by cardiac puncture on the 21st day. The blood samples were used for the determination of total red blood cell count (RBC) using Naubeur's haemocytometer after diluting with RBC diluent. Haematocrit was determined by microhaematocrit method (Blaxhall and Daisley, 1973) whereas haemoglobin was determined by cyanomethaemoglobin method (Drabkin and Austin, 1935).

The Hct, RBC, MCV, MCHC and MCH values after 21 days exposure were comparable with control while Hb was lower than control. This leads to the observation that the amount of Hb in the blood of the fish decreases as a result of long term exposure. Similar findings were observed in *Barbus conchonius* after exposure to toxic mercury for 2 to 3 weeks duration by Gill and Pant (1987). In the study Hb decreased in the first 2 weeks. Significant decrease in Hb was observed in *Cyprinus carpio var. communis* due to the treatment with sublethal dose of mercury for short periods of time (Thangam *et al*., 2016). Statistically significant decrease in Hb were observed in case of *Cyprinus carpio* exposed to 0.30mg/L $HgCl_2$ for 90hrs (Beena and Viswaranjan, 1987), *Aphanius dispar* exposed to $HgCl_2$ (1 mg/L) for 96 hrs and 30 days (Hilmy *et al*., 1980). Similar changes were seen by Fletcher and White (1986) in *Pleuronectes platessa*, plaice, along with splenomegaly (enlargement of spleen) on exposure to a concentration of 0.3 mg/L of $HgCl_2$. In *Clarias batrachus*, on treatment with mercury chloride at different concentrations, the Hb values decreased when compared with control at 35 days of exposure.

The observed decrease in Hb could be due to haemolysis caused by mercury toxicity (O'Connor and Fromm, 1975) and the inhibition of aerobic glycolysis causing reduction in the de novo synthesis of haemoglobin (Matkovics and Wiltas, 1981).

Sublethal concentrations of 0.02 ,0.04, 0.06, 0.08 and 0.10 ppm of $HgCl_2$ caused a decrease in mean Hb values to 67.2, 50, 42.6 and 29.2g/dl from the mean value of 75 g/dl (control) during 96 h exposure in *Channa punctatus* (Gupta and Kumar, 2016). There occurred a reduction of Hb in *Anabas scandens* (Panigrahi and Misra,

**Table 1:** Changes in the haematological parameters of *Anabas testudineus* on exposure to sublethal concentration of methylmercury in experimental tanks

| Parameters | Exposure duration tomethylmercury(Days) | |
|---|---|---|
| | 0 | 21 |
| Haemoglobin (Hb) (g/dl) | 8.0233 ± 1.329 | 5.12 ± 2.24 |

2016) and *Tinca tinca* treated with mercury (0.1 ppm) (Shah and Altindag, 2004). *Channa punctatus* also showed a decrease in Hb on mercury treatment (Sastry and Sharma, 1980). Mc Kim *et al*., 1976 observed that mercury in fish accumulates in the blood. According to Clarkson *et al.,* 1961, half of the mercury in blood is in association with RBC and the remaining half is in the form of a complex with serum albumin combining with the sulphydryl group. There are cases of divergent results and the parameters have been found to be dependent on various factors like size, sex, species, test chemical concentration as well as exposure duration (Fletcher and White, 1986). The present study documented a decrease in Hb on the 21st day indicating marked impact on long term exposure with methylmercury while the other parameters were same as normal values or less than normal values on long term exposure, demanding further studies to understand if there are any impacts on haematological parameters, if any, due to longer term sublethal exposures which are common in natural conditions.

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