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### Original article

## Study of Sodium Arsenate Induced Haematological Changes in Catfish, *Clarias batrachus*

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#### ABSTRACT

In the present study haematological parameters of control and sodium arsenate affected *Clarias batrachus* were observed, and provided an experimental view on arsenic contaminant. The action of sodium arsenate on experimental animal is well known and several studies have shown that chronic exposure to low levels of arsenic can also cause anaemia. The result of present study indicate that the RBC, Hb, Platelets, PCV, MCV and MCH value were decreased, fluctuation in differential leucocytes count and increased in the number of WBC due to exposure of sodium arsenate.

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#### INTRODUCTION

Environmentally heavy metals are defined as total condition surrounding an organism or group of organism especially, the combination of external and physical conditions that affect and influence the development, growth and survival of the organisms. Heavy metals are largely found in dispersed form in rock formations. Increasing industrialization and urbanization have the anthropogenic contribution of heavy metals in the biosphere and had the largest availability in soil and aquatic ecosystems and to a relatively smaller proportion in the atmosphere as particulates. Its toxicity in animals varies with animal species, specific metal, concentration, chemical form and pH, as many heavy metals are considered to be essential for animal growth. Heavy metals are significant environmental pollutants; their toxicity is a big problem of increasing significance for evolutionary, ecological, environmental and nutritional reasons [1].

Arsenic is a naturally occurring element found widely in the environment. However, on recent days the level of arsenic (arsenic tri-oxide, arsenic penta-oxide, sodium arsenate, sodium arsenite etc.) in the environment has increased several folds due to its use as pesticides, insecticides, rodenticides, defoliants, electronics, thermal power plants, wood preservatives and metal industries. Arsenic, an important environmental contaminant, arises not only from anthropogenic activities but also from rocks, mountains, possibly due to geothermal activities and leaching. Arsenic has been reported as one of the most alarming chemical in the environment [2], present in different forms and the toxicity depends upon its chemical form and oxidation states.

Arsenic toxicities have been shown in various organisms and the data suggest that the inorganic forms of arsenic showing the highest toxicity level, while organo arsenicals are usually less toxic [3]. The trivalent inorganic arsenic looks to be the form ultimately responsible for most of the toxic effects of the semimetal [4, 5]. Most affected by arsenic, those involved in accumulation, absorption and/or excretion, i.e., the circulatory system, gastrointestinal tract, skin, liver, and kidney. It affects the biochemical, haematological and ion regulatory parameters of organisms and particularly fish in aquatic medium and alterations of these parameters can be beneficial in environmental bio monitoring of arsenic contamination [6]. Fish are usually referred as an organism of choice for evaluating the effects of environmental pollution on aquatic ecosystems [7, 8].

Fishes are excellent bio indicator of heavy metal pollution. Arsenic is a heavy metal which effect on behaviour of *Clarias batrachus*. Behavioural changes are observed for chronic toxicity test. The two test concentrations 1/10 and 1/16 ppm to the Lc50 at 96 hours as sub lethal concentration which are 1.31ppm and 0.74ppm for sodium arsenate and sub-acute concentration of sodium arsenate for 7, 15,30,45 and 60 days[9].

Arsenic enters in the body through skin absorption, ingestion, or inhalation. Most ingested and inhaled arsenic is easily absorbed through the lung and gastrointestinal tract into the blood stream. Gastrointestinal tract are mostly absorbed trivalent arsenic, approximately 95 per cent is absorbed from the gastrointestinal tract. This arsenic is distributed in a large number of organs including the liver, lungs, kidney and skin. After absorption through the gastro-intestinal tract and lungs, 95 to 99 % of the arsenic is placed in erythrocytes, bound to globin of haemoglobin and then transported in different parts of the body [10,11]. In the present study there are some abnormalities in haematological parameters due to exposure of sodium arsenate.

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## MATERIAL AND METHODS

### Test Chemical

The analytical grade sodium arsenate ( $\text{Na}_2\text{HAsO}_4 \cdot 7\text{H}_2\text{O}$ ) (CAS No.: 10048-95-2) (Heptahydrate) was taken from Spectrum chemical mfg. corp., Mumbai, India and used without further purification for the experiment.

### Determination of Lethal Concentration of Sodium Arsenate

To determine the lethal concentration (LC50) of sodium arsenate, catfish (*Clarias batrachus*) were selected from the stock and exposed to different concentrations of sodium arsenate in different tanks. Ten fish were kept in each tank and water was replaced daily with fresh sodium arsenate mixed water to maintain a constant level of sodium arsenate during the exposure period. The mortality or survival of fish was observed at the end of 24 hours and the concentration at which 50% mortality of fish occurred was taken as the lethal concentration (LC50)[12].

### Experimental Animal

The healthy *Clarias batrachus* were used as an experimental animal and it was collected from local fish market of Indore and acclimatized in the laboratory for 7 days.

### Experimental Design and Duration

In the present investigation experimental fishes were divided into two groups. Ten (10) fishes were kept in control group and exposed to normal water and in experimental group forty (40) fishes were exposed to concentration of sodium arsenate at 24 hour's intervals. In both control and experimental group fishes were exposed to maximum 96 hours.

### Haematological Analysis

The blood collected from cardiac puncture of catfish and kept in sterilized appropriate vials then processed for various haematological analyses [13]. The RBC, WBC, platelets, PCV, MCV and MCH were counted by Haemocytometer method [14]. Hb concentrations were estimated by Sahil's method [15], differential leucocyte count by Leishmann method[16].

## RESULTS

In the present investigation the 96 hours LC50 value of sodium arsenate to *Clarias batrachus* was estimated and found to be 42 mg/l. On the other hand, in the experiment haematological estimation of control and sodium arsenate treated fishes were done. The haematological parameters were RBC, total WBC, DLC (Neutrophils, Eosinophils, Lymphocytes, Basophils and Monocytes), Haemoglobin, Platelets, PCV, MCV and MCH.

In control group haematological values were, RBC (3.53 million/cmm), total WBC (106.80 x103/cmm), Hb (10.64 g/dl), Neutrophils (2.18%), Eosinophils (5.32%), Lymphocytes (93.12%), Basophils (0.82%), Monocytes (6.00%), Platelets (162 cells/cmm), PCV (33.36%), MCV (126.20 fl) and MCH (38.64 pg).

Haematological values were at the 24 hrs. in experimental group haematological values were RBC (3.34 million/cmm), total WBC (121.72 x103/cmm), Hb (10.24 g/dl), Neutrophils (2.72%), Eosinophils (4.84%), Lymphocytes (94.78%), Basophils (0.76%), Monocytes (6.94%), Platelets (158.60 cells/cmm), PCV (31.58%), MCV (123.80 fl) and MCH (36.22 pg).

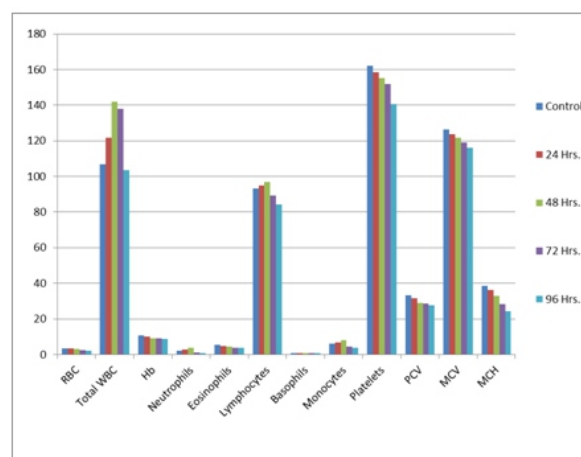
Moreover, in the present investigation at the 48 hrs. haematological values were RBC (3.24 million/cmm), total WBC (142.00 x103/cmm), Hb (9.08 g/dl), Neutrophils (3.66%), Eosinophils (4.44%), Lymphocytes (96.92%), Basophils (0.76%), Monocytes (8.14%), Platelets (155.20 cells/cmm), PCV (28.94%), MCV (121.60 fl) and MCH (32.94 pg).

At the 72 hrs. haematological values were RBC (2.60 million/cmm), total WBC (137.90 x103/cmm), Hb (9.00 g/dl), Neutrophils (1.04%), Eosinophils (3.84%), Lymphocytes (89.34%), Basophils (0.72%), Monocytes (4.62%), Platelets (152.00 cells/cmm), PCV (28.64%), MCV (19.20 fl) and MCH (28.28 pg).

At the last at 96 hrs. haematological values were RBC (1.98 million/cmm), total WBC (103.34 x103/cmm), Hb (8.82 g/dl), Neutrophils (0.96%), Eosinophils (3.66%), Lymphocytes (84.20%), Basophils (0.70%), Monocytes (3.68%), Platelets (140.60 cells/cmm), PCV (27.54%), MCV (116.20 fl) and MCH (24.38 pg).

In the present experimental investigation due to sodium arsenate RBC, Hb, Eosinophils, Platelets, PCV, MCV and MCH value were decreased as compared to control value at 24, 48, 72 and 96 hours. Basophils value was same at 24 and 48 hours then decreased at 72 and 96 hours as compared to control value. Total WBC value was also increased at 24, 48 and 72 hours and decreased at 96 hours as compared to control value. Neutrophils, Lymphocytes and Monocytes values were increased at 24 and 48 hours and then decreased at 72 and 96 hours as compared to control value. The impact and variation on haematological values of control and sodium arsenate treated fish were represented by graph.

**Graph 1: Showing haematological changes in *Clarias batrachus* due to sodium arsenate**



## FINDING AND DISCUSSION

The leucocytes and agranulocytes were increased by the arsenic group, in contrast to decreased levels of RBC count, granulocytes, haemoglobin, MCV, MCH, PCV and platelets. These values represent a marker of anaemia (microcytic, hypochromic anemia) with subsequent result of suppression of erythropoiesis in the haemopoietic system. Arsenic compound such as sodium arsenate may cause oxidative stress in the liver of fish and bring fluctuation in haematological profile. Arsenic exposure decreases in the counting of red blood cells [17]. The haematological parameters are also referred to notice the indicators of stress and physiological status of lives [18]. In the present study granulocytes, agranulocytes, MCV, MCH and PCV were also decreased due to effect of sodium arsenate.

The decrease in haemoglobin was due to decrease of RBC count, which might be due to sodium arsenate effect on erythropoietic process which lead into inhibited erythropoiesis and thus poor haemoglobin synthesis. Anaemic condition can also be generating by other pathological conditions such as lysis of RBC, erythropenia or haemodilution [19]. The above discussion led us to conclude that sodium arsenate even in sub-lethal concentration alters the normal hematological value.

The suppression of erythropoiesis due to toxicants of arsenic by its action on membrane or may another possible reason [20,21]. The decrease level of haemoglobin and packed cell volume in *Clarias batrachus* exposed to waterborne arsenic because of lysis of erythrocytes reduced the haemoglobin and packed cell volume [22]. The decreased number of Haemoglobin, RBC, granulocytes and platelets in fish may due to sodium arsenate. Decrease in RBC in the present investigation resulted from suppression of production of RBC or Haemoglobin synthesis by the sodium arsenate or the accumulation of arsenic contaminants in the gill region might have affected the structure of gill leads to haemolysis.

In the present investigation the LC50 value of sodium arsenate to the *Clarias batrachus* was found to be 42 mg/l indicating that the sodium arsenate is toxic for fish. RBC, Hb, Platelets, PCV, MCV and MCH value were decreased, fluctuation in Differential leucocytes count and increased in the number of WBC due to effect of sodium arsenate.

## CONCLUSION

The present investigation confirmed that the haematological parameters are very sensitive parameters for monitoring toxic responses of the *Clarias batrachus*. Sodium arsenate toxicity reveals the anaemic condition in catfish. Increased WBC indicates the stimulation of defence system due to arsenic toxicity. Other haematological profile also could be effectively used as potential biomarkers of sodium arsenate toxicity.

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## REFERENCES

- Asati A, Pichhode M, Nikhil K. Effect of heavy metals on plants: an overview. *Int. J. Appl. Innov. Eng. Manage.* 2016;5:2319-4847.
- ATSDR. Toxicological profile for Arsenic. Agency for Toxic substances and Disease registry. SUDHHS PHS Washington DC. 2002.
- Duker AA, Carranza E, Hale M. Arsenic geochemistry and health. *Environment international.* 2005;31(5):631-41.
- Rasmussen RE, Menzel DB. Variation in arsenic-induced sister chromatid exchange in human lymphocytes and lymphoblastoid cell lines. *Mutation Research/Reviews in Mutation Research.* 1997;386(3):299-306.
- Kovendan K, Vincent S, Janarthanan S, Saravanan M. Expression of metallothionein in liver and kidney of freshwater fish *Cyprinus carpio* var. *communis* (Linn) exposed to arsenic trioxide. *Am J Sci Ind Res.* 2013;4(1):1-0.
- Lavanya S, Ramesh M, Kavitha C, Malarvizhi A. Hematological, biochemical and ionoregulatory responses of Indian major carp *Catla catla* during chronic sublethal exposure to inorganic arsenic. *Chemosphere.* 2011;82(7):977-85.
- Gernhöfer M, Pawert M, Schramm M, Müller E, Triebkorn R. Ultrastructural biomarkers as tools to characterize the health status of fish in contaminated streams. *Journal of Aquatic Ecosystem Stress and Recovery.* 2001;8(3-4):241-60.
- Gaim K, Gebru G, Abba S. The effect of arsenic on liver tissue of experimental animals (fishes and mice)—a review article. *International Journal of Scientific and Research Publications.* 2015;5(5):1-9.
- Mahurpawar, M. Toxic effect of sodium arsenate on *Clarias batrachus*. *International Research Journal of Biological Sciences.* 2017;6(1):15-18.
- Hunter, F. T., Kip, A. F. and Irvine, J. W. Radioactive tracer studies on arsenic injected as potassium arsenite I. Excretion and localization in tissues. *Journal of Pharmacology and Experimental Therapeutics.* 1942;76(2):207-220.
- U.S. Public Health Service. Toxicological profile for arsenic. USEPA, Washington, DC. 1989.
- Kumari B, Kumar V, Sinha AK, Ahsan J, Ghosh AK, Wang H, DeBoeck G. Toxicology of arsenic in fish and aquatic systems. *Environmental chemistry letters.* 2017;15(1):43-64.
- Dacie V and Lewis SM. In: *Practical Haematology*, ELBS, Longman, Singapore, 1975.
- Sharma IJ and Singh HS. *Students Laboratory Manuals of Veterinary Physiology*, 1st edn. Kalyani Publisher, New Delhi, India. 2000.
- Ratan V. Hb% estimation by Sahil's Method. *Handbook of Human physiology*, 3rd edn. 1978.
- Rumke CL. Variability of result in differential cell count on blood smear. *Triangular.* 1960;44:154-157.
- Kavitha C, Malarvizhi A, Kumaran SS, Ramesh M. Toxicological effects of arsenate exposure on hematological, biochemical and liver transaminases activity in an Indian major carp, *Catla catla*. *Food and Chemical Toxicology.* 2010;48(10):2848-54.
- Lal Shah S. Hematological changes in *Tinca tinca* after exposure to lethal and sublethal doses of Mercury, Cadmium and Lead. *Iranian Journal of Fisheries Sciences.* 2010;9(3):434-43.
- Srivastava G, Srivastava AK. Toxicological effects of selenium on the haematological parameters of a freshwater catfish, *Heteropneustes fossilis* (Bloch). *Indian Journal of Scientific Research.* 2011;109-15.

20. Allin CJ, Wilson RW. Effects of pre-acclimation to aluminium on the physiology and swimming behaviour of juvenile rainbow trout (*Oncorhynchus mykiss*) during a pulsed exposure. *Aquatic Toxicology*. 2000;51(2):213-24.
21. Chowdhury MJ, Pane EF, Wood CM. Physiological effects of dietary cadmium acclimation and waterborne cadmium challenge in rainbow trout: respiratory, ionoregulatory, and stress parameters. *Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology*. 2004;139(1-3):163-73.
22. Tripathi S, Sahu DB, Kumar R, Kumar A. Effect of acute exposure of sodium arsenite ( $\text{Na}_3\text{AsO}_3$ ) on some haematological parameters of *Clarias batrachus* (common Indian cat fish) in vivo. *Indian journal of environmental health*. 2003;45(3):183-8.

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