

Research Article

**HAEMATOLOGICAL STUDIES OF FRESHWATER CATFISH
MYSTUS VITTATUS EXPOSED TO SODIUM ARSENATE**

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ABSTRACT

Experiments were conducted to study toxic effect of sodium arsenate on haematological parameters of the freshwater catfish *Mystus vittatus*. LC₅₀ value of sodium arsenate was calculated for a period of 96hrs. *M. vittatus* exposed to sub lethal doses such as 0.15, 0.75, 1.5 and 2.0 ppm sodium arsenate. Sodium arsenate toxicity induced a significant decrease in Haemoglobin content, and RBC count and increase in WBC count.

Key words: Sodium arsenate, sublethal toxicity, haematology, *Mystus vittatus*.

INTRODUCTION

Arsenic is the 20th most abundant element in the Earth's crust, and is primarily associated with igneous and sedimentary rocks where it occurs mostly as inorganic forms (Tamaki and Frankenberger, 1992). Arsenic is known to cause adverse effects in aquatic biota and is a major concern to human health (Shaw *et al.*, 2007). Increasing aquatic pollution and other anthropogenic effect on water resources damage the natural fish stocks. The natural aquatic system may extensively be contaminated with heavy metals released from domestic, industrial and other man-made activities (Zaki *et al.*, 2009). Various studies in fishes have shown that heavy metals may alter the physiological activities and biochemical parameters both in tissue and blood (Basa and Rani, 2003).

Under laboratory conditions, toxicity testing procedure (mortality studies, LC₅₀ estimates) provides information regarding the harmfulness

of industrial stress on aquatic animals including fishes (Marier, 1973). Acute the toxicity of contaminants in static bioassays are widely recommended as preliminary step in toxicological studies (McLeay, 1976; Whittle and Flood, 1977; Chapman, 2000; Ali and Sreekrishna, 2001; Parrott *et al.*, 2006).

A number of haematological indices such as haemoglobin (Hb), Red blood cells (RBC) and white blood cells (WBC) count are used to assess the functional status of the oxygen carrying capacity of the blood stream and have been used as indicator of metal pollution in the aquatic environment (Shah and Altindag, 2004). Blood parameters are considered pathophysiological indicators and therefore consider as important in diagnosing structural functional status of a fish exposed to toxicants (Adhikari *et al.*, 2004). The present study was under taken to study the toxic effect of sodium arsenate on blood parameters of freshwater fish *M. vittatus*.

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MATERIALS AND METHODS

Test Animal and Test Chemical

The experimental fish specimens of *M. vittatus* having average length 10 ± 2 cm and average weight, 85 ± 4 g, were collected from Kathirasan Aqua pond Orathanadu and acclimated under laboratory condition for 30 days. The heavy metal Sodium arsenate was obtained from (S.D. Fine-Chem Ltd, Mumbai) and toxic studies were conducted under static bioassay system (Doudoroff, *et al.*, 1957).

Determination of LC₅₀ Value

The 96 hrs LC₅₀ value was determined by adopting Finney's Probit method (Finney, 1964). The four sub lethal concentrations from the LC₅₀ value such as 0.15, 0.75, 1.5 and 2 ppm were selected for further studies. The test media was renewed daily during the experimental period and the fishes were fed *ad libitum*.

Haematological Study

After 30 days, blood from the control and sodium arsenate treated medium fishes were obtained by severance of caudal peduncle and transferred to Eppendorf tubes containing EDTA as anticoagulant (Dacie and Lewis, 1975). These treated and blood samples were used to estimate the haematological parameters.

Total RBC Count

Total count of RBC was made using an improved Neubaur haemocytometer (Shah and Altindag, 2004). Blood was diluted 1:200 with Hayem's fluid (Mishra *et al.*, 1977). Erythrocytes were counted in the loaded haemocytometer chamber and total numbers were reported ($10^6 / \text{mm}^3$) (Wintrobe, 1967).

Total Count of WBC

Total WBC count was done using an improved Nebular haemocytometer (Shah and Altindag, 2005).

Estimation of Haemoglobin

Haemoglobin (Hb) was estimated using a haemoglobin test kit (DIAGNOVA, Satan India)

by Cyanmethemoglobin method. Values of treated groups were compared statistically with control by student's t-test. Significance was established at $p < 0.05$ using the Microsoft excel 2010 programme. Significance of data further checked with the percent (+ increase and - decrease) in blood parameters of *M. vittaus*

RESULTS

Total RBC Count

The RBC count of healthy controls showed a mean value of 1.55, ($10^6 / \text{mm}^3$). The fish exposed to sub lethal concentration of sodium arsenate showed mean value of RBC as 1.51, 1.47, 1.43 and 1.39 ($10^6 / \text{mm}^3$) for sub lethal concentration 0.15, 0.75, 1.5, and 2ppm treatments respectively. The treatment with sodium arsenate was found to inflict a drastic reduction in the total count of RBC. The reduction was dosage dependent; as concentration of sodium arsenate increased the RBC levels declined (Table 1).

Total WBC Count

The result of the total count of WBC revealed that the blood of the control fish showed a mean value of 6.10 ($10^6 / \text{mm}^3$). The fishes exposed to sub lethal concentration of sodium arsenate showed mean values of WBC as 10.60, 16.50, 22 and 28 ($10^6 / \text{mm}^3$) for 0.15, 0.75, 1.5, and 2ppm of sodium arsenate treatments respectively (Table 1). The values mentioned above showed a significant increase when compared to the control.

Haemoglobin Content

The control fishes showed mean value of 71.00g/dl for haemoglobin. The fishes were exposed to sub lethal concentration of sodium arsenate showed haemoglobin mean values of 60.26, 49.36, 37.20 and 22.68 g/dl haemoglobin at 0.15, 0.75, 1.5 and 2 ppm of Sodium arsenate treatments, respectively (Table 1). The values for treatments showed a significant decrease when compared to the control ($p < 0.05$).

Table 1. Total count of RBC, WBC, and Haemoglobin in the control and sodium arsenate treated *M. vittatus*.

Haematology parameter	Concentration of Sodium arsenate				
	+	0.15 ppm	0.75 ppm	1.5 ppm	2 ppm
Total RBC (x 10 ⁶ / mm ³)	1.55±0.01	1.51±0.02 (-6.12)	1.47±0.01 (-9.86)	1.43±0.02 (-11.78)	1.39±0.01 (-14.65)
Total WBC (x 10 ⁶ / mm ³)	6.10±0.01	10.60±0.45 (63.30)	16.50±0.33 (72.36)	22±0.21 (80.43)	28±0.52 (96.18)
Haemoglobin (g/dl)	71.00±4.00	60.26±0.41 (-9.38)	49.36±0.57 (-30.24)	37.20±0.45 (-34.12)	22.68±0.27 (-42.62)

Each value is mean ±SD 5 observations.

+ indicates increase over the control.

- Figures in brackets indicates decrease or increase over the control indicated as + or -.

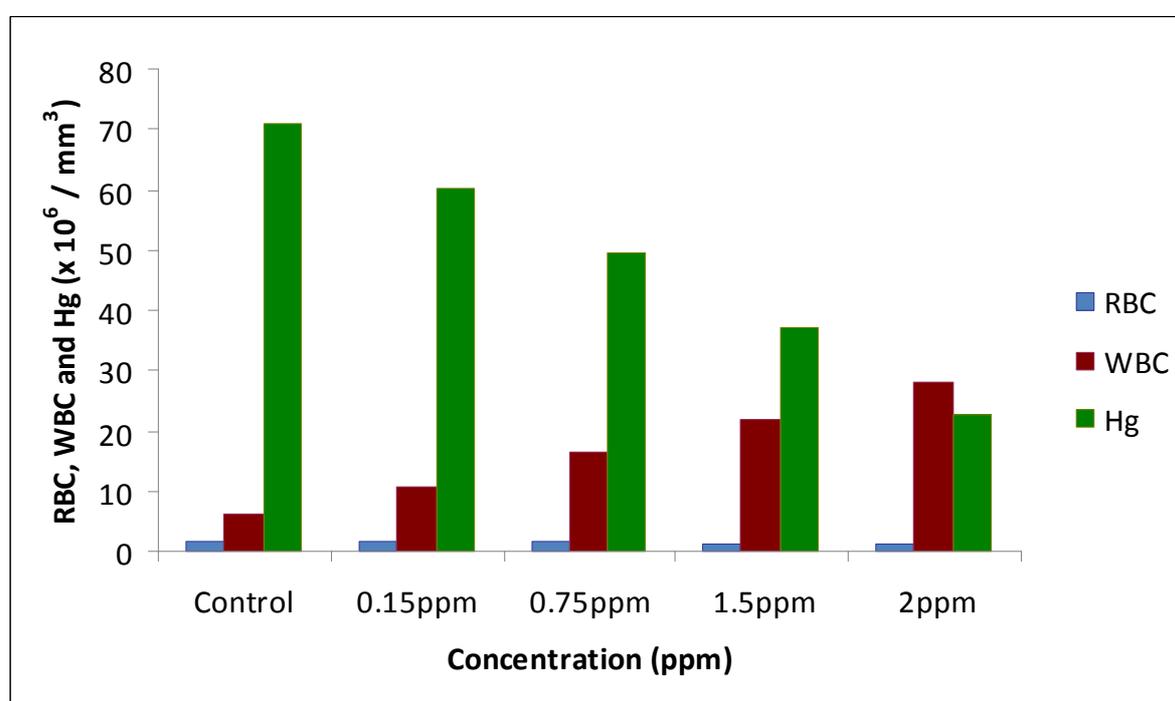


Figure 1. Total RBC, WBC and Hg changes in *M. vittatus*, exposed to different concentrations of Sodium arsenate.

DISCUSSION

Toxic Sodium arsenate harms fish and aquatic animals largely as a function of its toxicity, to the exposure time, dose and persistence in the environment. Toxicity of the sodium arsenate refers to how poisonous it is some heavy metals are extremely toxic.

In recent years haematological parameters have been used more to assess the effect of sub lethal concentrations of pollutants (Webemeyer and Yasutake, 1977). The results of the present investigation show that the sodium arsenic

treatment inflicted a drastic reduction in the total count of the RBC which shows a dosage dependent effect (Panigrahi and Misra, 1978). Reduction in haemoglobin percentage and RBC count of the fish *Anabas scandens* treated with mercury is also reported earlier. Decrease level in RBC count and Hg content was observed in fish *Tincatinca* exposed to mercury, cadmium and lead (Shah and Altindag, 2004). In the present investigation, leucocyte concentration showed greater and quite different pattern of change with the effect of sodium arsenate compared with the erythrocyte levels of the

control groups. An increase in leucocytes number may be the compensatory response of lymphoid tissues to the destruction of circulating the kinds of behavioural faults in orientation and Lymphocytes (Shah and Altindag, 2004). Allen (1994) observed increased WBC counts in *Oreochromis aureus* after mercury exposure. Chandanshive *et al.* (2012) also reported the decrease level of RBC in the fresh water fish *Labeo rohita* after exposure to mixture of heavy metals. All these reports are in agree with the present study. The reduction in total RBC count and Hb have suggested that heavy metal exposure decreases the total RBC count, and Hb content due to impaired intestinal absorption of iron (Joshi *et al.*, 2002).

Increase in WBC content observed in the present study could be attributed by stimulation of the immune system in response to tissue damage caused by sodium arsenate (Gill and Pant, 1985; Dhanekar and Srivastava, 1985)

CONCLUSION

Reduction of RBC and Hb may be suggestive of an appreciate decline in the hematopoiesis leading to anaemia. Increase in total WBC is recorded probably due to thrombocytosis, lymphocytosis or leucopoiesis and or enhanced release of lymphocytes from the lymphoid tissue to mitigate the metal toxicity.

CONFLICTS OF INTEREST

The authors declare that there are no conflicts of interest associated with this article.

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