Hematological alterations induced by Sodium arsenate toxicity in Albino mice
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Abstract
Arsenic is one of the most dangerous occupational and environmental toxins. Arsenic (As) is a toxic and carcinogenic metalloid. For the general population, arsenic in drinking water is the main exposure source, and more harmful than arsenic in food, because the bioavailability (actual amount absorbed into the bloodstream) of arsenic from water is greater than that from grains or vegetable. Arsenic exists in the trivalent and pentavalent forms and is widely distributed in nature. The drastic adverse changes in the various hematological parameters were observed in treated mice when compared control animals. Haematological parameters are used as sensitive indicators of toxicity due to different pollutant. First group exposed to tap water maintained as control where as second and third groups were treated with different doses of Sodium arsenate. Blood samples were collected after 7 days and 14 days. The lymphocytes were severely damaged by arsenic toxicity. White Blood Cells (WBCs), Red Blood Cells (RBCs) and hemoglobin level in control groups were in normal range whereas level were significantly decreased with the increase dose of arsenic in the respective treatmental groups. The data was analyzed statistically and was found that significant was found among the group (p < 0.05).

Key-Words: Sodium arsenate, Albino mice, Hematology

Introduction
Arsenic is a ubiquitous metalloid present at low concentrations in rocks, soil and natural water. Agricultural soils and drinking water are exposed to geogenic arsenic as this element naturally occurs in the earth’s crust and is a constituent of more than 200 mineral species. Human exposure to arsenic can occur via different routes. Although dermal and inhalation exposure is possible, food and drinking water are the principal routes of exposure to arsenic (1, 2). Arsenic and inorganic arsenic compounds can be emitted into air and then deposited into water and soil during industrial operations such as ore mining and smelting, and during volcanic eruptions and forest fires (3). The maternal circulation levels of arsenicals are influenced by absorption rates. In oral exposure, arsenic is absorbed into the blood from the intestines. It is then transported to the liver and may undergo first-pass metabolism prior to being delivered to the uterus (4, 5).

Prolonged ingestion of As leads to its accumulation in the liver, kidneys, heart and the lungs and in smaller amounts in the muscles, nervous system, gastrointestinal tract and the spleen (6). Blood plays a decisive role in the regulation of life processes to make them function properly. An organism must be able to keep its blood composition relatively constant under normal conditions and must also have the ability to change it under extreme conditions such as stress situations. Hematological parameters after As exposure showed marked decrease in hemoglobin, packed cell volume, erythrocytic count and total leukocytic counts, heterophils and lymphocytes (7). The decrease in erythrocyte indices could be due to acute hemorrhages or hemolysis (8), showing rapid decrease in all hematological parameters leading to anemia.

Material and Methods
Healthy albino mice of same age group 65±5 days and weight (35±5) were taken from veterinary college, Bangalore and maintained a colony. They were kept in well cleaned and sterilized cages. The laboratory conditions were maintained i.e. 26±2°C; 12hr light and 12hr darkness.

Experimental design
The animals were divided into three groups each group having ten animals. Toxicity of sodium arsenate was evaluated according to Finney and found LD_{50} as 45
mg/kg body weight. Tenfold lower concentration of LD_{50} i.e. 4.5mg/kg body weight was taken as sub lethal dose. The first group of animals fed with tap water and intraperitoneal administration of As was done in the second group of animals with 7 days treatment (alternate days, 1, 3, 5 and 7) and third group of animals with 14 days treatment (alternate days). The blood was collected from the orbital venous plexus by puncturing with the tip of Pasteur pipette under diethyl ether anesthesia and the blood was allowed to fall drop by drop in to a graduated centrifuge tubes containing EDTA (Ethylene Diamine Tetra Acetic Acid), anticoagulant to the required quantity for hematological work. The bleeding was arrested by gently pressing the eyeball with the help of dry cotton.

Results and Discussion

In the present study the toxic effect of sodium arsenate on the hematological parameters is determined in albino mice. The alterations in blood of albino mice due to sodium arsenate are indicated (Table: 1) after 7 days and 14 days experimental and control animals. Repeated intraperitoneal administration of sodium arsenate produced gradual decrease in red blood cells (RBC), Hemoglobin (Hb), Packed Cell Volume (PCV), Mean Corpuscular Hemoglobin (MCH), Mean Corpuscular Volume (MCV), and Mean Corpuscular Hemoglobin Concentration (MCHC).

A significant decrease is observed in RBC count in all doses of sodium arsenate injected albino mice. The decrease in RBC count in seven days of sodium arsenate injected animals -20.92%, and -45.59% decreases was recorded in fourteen days treated sodium arsenate Mice. Thus the 14 days treated animal’s shows highly significant change in RBC count. In the case of Hb, the levels also showed decrease in experimental animals when compared to control animals. Thus - 26.128% in 7 days treated animals, and -32.84% in 14 days treated animals decrement was recorded. The packed cell volume (PCV) has shown -24.68%, and 31.39% decrease in 7 days and 14 days treated animals respectively.

The mean corpuscular hemoglobin (MCH), mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) were determined in control and experimental animals. In all the cases there is an increase in MCV and decrease in MCH and MCHC of experimental animals when compared to control group (Table: 1). The WBC count was increased in the experimental animals. The increment was 13.87% in 7 days treated animals and 40.08% in 14 days treated animals respectively.

The toxic effect of sodium arsenate on hierogram is determined in mammalian modal albino mice in the present study. Often the sodium arsenate treated Albino mice gradually has got anaemic condition. The present experiment has proved the significant decrease in red blood cells (RBC) count, hemoglobin (Hb) and packed cell volume (PCV) levels when compared to control mice with sodium arsenate treated animals. In the present study, levels of leucocytes and MCV in the arsenic group increased, MCH and MCHC in the arsenic group decreased, in contrast decreased levels of erythrocyte count, hemoglobin and hematocrit. Furthermore, the reason of increase in the MCV values may be macroglobin type anemia. Singh, 1995 and Griffin, 1999 documented that RBC count, Hb content and PVC values were significantly decreased in Channa punctatus on exposure to copper sulphate. The study shows hematological changes after arsenate exposure as white blood cells (WBC) level was increased and denotes the necrosis activities in the cells. Whereas the decrease in the red blood cells (RBC) and hemoglobin levels denotes inhibition of hem-synthesis path way. Similarly the other hematological parameters also show decreased levels as these are dependent on the hemoglobin and RBC levels. Arsenic is known to cause a decrease of white blood cell during the heavy metal antagonism in male mice (11). This could support the implication of arsenic exposure (in drinking water) as a possible contributory factor in Mycobacterium ulcerans infection in Buruli Ulcer endemic areas as reported by Gyasi et al., (12). Reduction of Hb and RBC accompanied by a compensatory response (increased hematopoietic rate) in lead-intoxicated rainbow trout was reported (13). The increased rate of hemolysis, and particularly high rate of nuclear anomalies in lead treated common carp, was also observed in earlier studies (14). Mercury and the combination of high concentrations of cadmium and mercury could inhibit heme synthesis of red blood cells and cause anemia signs described by (15). Hemoysis occurs in response to toxicity that leads to alteration in the selective permeability of the membrane (16). Chandanshive (17) also reported decrease in RBC of fish Labeo rohita after exposure to mixture of heavy metals. Zinc caused a significant decline in Hb, RBC, and copper caused significant decrease in Hb, MCHC and an elevation of MCV was observed (18) in fish. The MCHC is a good indicator of red blood cell swelling and/or to a decrease in haemoglobin synthesis (19&20, 21). Progressive increased levels of total WBC count have also been reported in C. punctatus exposed to lead (22), Clarias batrachus exposed to mercuric chloride (23)
and Clarias gariepinus to metal finishing company effluents (24). The haematopietic system is also affected by both short-and long-term arsenic exposures. Anemia and leukopenia are common effects of poisoning and have been reported as resulting from acute intermediate, (25) and chronic oral exposures (26). These effects may be due to a direct haemolytic or cytotoxic effect on the blood cells (27) and a suppression of erythropoiesis. Peroxidation of membrane phospholipids in one of them. It is well established that As induces lipid peroxidation and perturbation of antioxidant systems in rat erythrocytes (28).

The elevation of TBARS (Thiobarbituric Acid Reactive Substances) has been associated with reduction of membrane fluidity (29) and the enhanced hemolysis could be a consequence of oxidative stress and lipid peroxidation in the circulating erythrocytes (30). The hematological parameters showed a significant change under the impact of sodium arsenate (30).

In the present investigation the changes are highly significant in 14 days treated animals than the 7 days treated animals of arsenate administered Mice.

References


Table 1: Heamogram of control and sodium arsenate treated albino mice values are mean ± SD of eight individual observations

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>7 days</th>
<th>14 days</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RBC</td>
<td>8.161±0.36</td>
<td>6.453±0.41</td>
<td>4.44±0.38</td>
</tr>
<tr>
<td></td>
<td>(20.92)</td>
<td>(-26.12)</td>
<td>(-32.84)</td>
</tr>
<tr>
<td>Hb</td>
<td>10.211±0.41</td>
<td>7.543±0.45</td>
<td>6.857±0.48</td>
</tr>
<tr>
<td></td>
<td>(13.87)</td>
<td>(-24.68)</td>
<td>(-31.39)</td>
</tr>
<tr>
<td>WBC</td>
<td>5333.733±275.513</td>
<td>6073.633±116.329</td>
<td>7471.817±484.29</td>
</tr>
<tr>
<td></td>
<td>(13.87)</td>
<td>(13.54)</td>
<td>(25.48)</td>
</tr>
<tr>
<td>PCV</td>
<td>22.499±0.42</td>
<td>16.944±0.34</td>
<td>15.435±0.45</td>
</tr>
<tr>
<td></td>
<td>(24.88)</td>
<td>(-24.68)</td>
<td>(-31.39)</td>
</tr>
<tr>
<td>MCV</td>
<td>26.149±0.81</td>
<td>29.69±0.51</td>
<td>32.813±0.37</td>
</tr>
<tr>
<td></td>
<td>(13.54)</td>
<td>(25.48)</td>
<td></td>
</tr>
<tr>
<td>MCH</td>
<td>15.122±0.81</td>
<td>13.459±0.51</td>
<td>14.148±0.37</td>
</tr>
<tr>
<td></td>
<td>(10.99)</td>
<td>(-6.44)</td>
<td></td>
</tr>
<tr>
<td>MCHC</td>
<td>49.242±0.88</td>
<td>43.359±0.62</td>
<td>43.218±0.84</td>
</tr>
<tr>
<td></td>
<td>(-11.94)</td>
<td>(-12.23)</td>
<td></td>
</tr>
</tbody>
</table>

Values in parentheses indicate percent change over control
Values are mean ± SD of eight individual observations
*Significant from control at p<0.05
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