TOXICOLOGICAL EFFECTS OF SELENIUM ON SOME PHYSIOLOGICAL PARAMETERS IN MALE RATS (*RUTTUS RUTTUS*)

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ABSTRACT

This study was conducted to detect the effects of Selenium on some physiological and reproductive parameters in rats male, Twenty-four mature male rats were randomly divided into three equal groups; each group consists of eight males. First group was injected intraperitoneally with 0.1 ml of normal saline for 30 consecutive days as control group, while second and third groups were intraprtionally injected with 0.1 ml of sodium Selenite at concentration (2.4 mg/kg body Wight) of selenium for 30 days. The hematological and biochemical parameters including, red blood cells count(R.B.C), white blood cells count(W.B.C), hemoglobin concentration(Hb), package cell volume (P.C.V) , in addition to ALT and AST were performed after animals elimination. The concentration and percentage of normal and abnormal sperms were calculated after epididymis extraction. Statistical analysis reveals that significant differences (p≤0.05) in Red blood cells count, package cell volume, while WBCs count and liver enzymes ALT,AST show increase significant differences (p≤0.05) in comparison with control group. In concern with reproductive parameters, this study indicates there is significant differences in motility and activity of sperms of treated males, in comparison with control group.

INTRODUCTION

Selenium is an essential nutrient for the health of human and animals. Selenium behaves as an essential micronutrient to biological systems at lower concentrations but becomes toxic at more prominent levels (1). Selenium has similar chemical properties to sulphur, Selenium can exist in two forms; as a silvery metal or a red powder. Selenium atomic number is (34) and atomic mass (78.96 amu) and melting point (217.0 C°) and boiling point (684.9 C°), the number of protons/electrons in selenium is (45) and it has a hexagonal crystal structure and the color of selenium is grey, red and black (2). Selenium compounds are used in the glass industry as decolorizing agents and in the rubber industry as vulcanizing agents. Selenium compounds are also found in toning baths used in photography and
xerography, and in insecticides and photoelectric cells. However, selenium is now known to be required by laboratory animals, food animals, and humans for proper growth and immune function (4). Selenium at nutritional levels has been shown to have numerous anticarcinogenic or preventative effects against carcinogen-induced breast, colon, liver and skin cancer in animals (5). The biological role of the element in the active center of glutathione peroxidase, the key enzyme for tissues and body fluids which plays a fundamental defensive role in the antioxidant protection system (6).

Several studies have addressed the toxicity of selenium compounds to animals when administered in either food or drinking water. Groups of 5 male and 5 female Swiss mice were given a daily gavage dose of 0.5 ml containing up to 64 ppm Selenium as sodium selenite for three days, and observed for a further seven days (7). All male mice survived to the end of the study. One female in the 32 ppm group died after the second dose and all females in the 64 ppm group died following the third dose. Pale and slightly necrotic livers were observed at autopsy. Small organs with apparent atrophy accompanied the stunted growth observed in the high dose groups. Mice treated with selenium over 15 days showed significant reduction of R.B.C., W.B.C., Hb. and P.C.V. (8). Reproductive deficits, including teratogenesis and embryonic mortality, occur in avian species (9,10); however, there is no evidence Selenium is teratogenic in mammals (11). (12) reported prooxidant effects of selenium on germ cells after administration of an excess of sodium selenite to the mouse in their feed. (13) Shows the excess of dilatory selenium caused dose-time dependent reduction in body and reproductive organ weights and increase in morphologically abnormal of spermatozoa.

The present investigation aimed to determine the effects of selenium injection on some hematological and reproductive parameters in male rats.

**MATERIALS AND METHODS**

**Experimental Designs:**

The experiment animals was conducted at the animal house of the veterinary medicine college University of Basrah.

Twenty four males rats sexually mature, 12 weeks old, and (200-250 gm) grams of body weigh were used. The experiment conditions were the same for all animals, where the room
temperature was set between 20-25C by the use of an air conditioner, and the daily light period was 12 hours by use of two fluorescent lamp, the rate of humidity was about 50%. All nutritional requirements were provided. The animals were divided into three groups.

Each group contains eight animals. All animals were intrapertoneally injected for 30 days (1 ml injection/ day with volume of 0.1ml/10gm of body weight) as bellow:

1- Group one (control) was treated with physiological saline (0.9% Nacl) and used as a control group.
2- Group tow was treated with 2mg/kg of body weight sodium selenite.
3- Group was treated with 4 mg/kg of body weight sodium selenite.

Specimens collection:
After the end of excrement the rats were anaesthetized, blood samples were collected directly from the heart by the use a disposable syringes. The blood samples were a liquated into two parts, one for hematological study and, the other for biochemical study.

Blood Parameter:

A- Red blood corpuscles count (RBC):
The red blood corpuscles count was obtained by use of hematocytometer (Neubaure improved double) and (Haymes solution) and a special pipette for dilution (14).

B- White blood cells count (WBC):
The white blood cell was obtained by the method of (15).

C- Packed cells volume (PCV):
The Packed cells volume was measured according to the method of (16).

Biochemical parameters:
Serum aspartate transaminase (AST) and alanine transaminase (ALT) activities were measured according to the method of (17)

Reproductive Parameters:
By the end of the injection period, all animals have been killed and the sperms were extracted further reproductive analysis. Massive motility are measured depending upon the gradation basis suggested by (18). The individual motility is done according to (19).
Statistical analysis:

In this study, (ANOVA) analysis and L.S.D. tests are used according to (SPSS version) programmer at the (P<0.05) to find the means for all treatments (SPSS,1989),(20).

RESULTS

The result in the table (1) show significantly at (P<0.05) decrease of R.B.C, Hb and P.C.V. in treated group with sodium selenite where compared with control while the same table appears a significant (P<0.05) increase in W.B.C,ALT and AST when compared with control group.

Table (1). Effects of selenium on some hematological and biochemical parameters on male rats.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>R.B.C (\times10^6) (Cell/mm(^3))</th>
<th>W.B.C (\times10^9) (Cell/mm(^3))</th>
<th>Hb g/100ml</th>
<th>P.C.V %</th>
<th>AST IU</th>
<th>ALT IU</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Control group</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>7.03 ± 474</td>
<td>5.9 ± 153.88</td>
<td>12.56 ± 0.14</td>
<td>38.12 ± 0.29</td>
<td>155 ± 1.62</td>
<td>136.37 ± 1.01</td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>A</td>
<td>A</td>
<td>A</td>
<td>C</td>
<td>C</td>
</tr>
<tr>
<td><strong>T1</strong></td>
<td>4.94 ±134.82</td>
<td>6.59 ± 99</td>
<td>11.05 ± 0.26</td>
<td>33.62 ± 0.65</td>
<td>181.5 ± 1.65</td>
<td>156.25 ± 2.08</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>B</td>
<td>B</td>
<td>B</td>
<td>B</td>
<td>B</td>
</tr>
<tr>
<td><strong>T2</strong></td>
<td>4.10 ± 81.73</td>
<td>6.6 ± 104.84</td>
<td>9.65 ± 0.29</td>
<td>29.12 ± 0.63</td>
<td>194.5 ± 1.83</td>
<td>162.125 ±2.19</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>A</td>
<td>C</td>
<td>C</td>
<td>A</td>
<td>A</td>
</tr>
<tr>
<td><strong>LSD</strong></td>
<td>0.26</td>
<td>0.35</td>
<td>0.72</td>
<td>1.63</td>
<td>5.02</td>
<td>5.42</td>
</tr>
</tbody>
</table>

The different litters refer to significant difference among group (P<0.05) n=8

Sperm viability:

In concerning with seminal fluid analysis table (2) show the effect of selenium on sperm viability. It is obvious that the injected of selenium lead to significant decrease (p<0.05) in all sperm viability with an increase in sperms abnormalities and dead sperms compared with control group and between T1 and T2 groups.
Table(2) effect of selenium on semen quality of rats:

<table>
<thead>
<tr>
<th>Groups</th>
<th>Massive motility (%)</th>
<th>Individual motility (%)</th>
<th>Sperms count ($\times 10^6$/ml)</th>
<th>Abnormal sperms (%)</th>
<th>Dead sperms (%)</th>
<th>Alive sperms (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>83.0 ± 0.88 A</td>
<td>81.37 ± 0.94 A</td>
<td>174.62 ± 3.01 A</td>
<td>10.12 ± 0.63 B</td>
<td>17.75 ± 1.55 C</td>
<td>72.12 ± 1.50 A</td>
</tr>
<tr>
<td>T1</td>
<td>40.00 ± 2.67 B</td>
<td>41.25 ± 3.50 B</td>
<td>140.62 ± 4.20 B</td>
<td>13.25 ± 1.71 B</td>
<td>42.12 ± 1.74 A</td>
<td>45.37 ± 2.55 B</td>
</tr>
<tr>
<td>T2</td>
<td>17.50 ± 2.50 C</td>
<td>25.00 ± 3.27 C</td>
<td>127.62 ± 1.92 C</td>
<td>21.50 ± 2.16 A</td>
<td>39.62 ± 2.12 A</td>
<td>38.87 ± 1.30 C</td>
</tr>
<tr>
<td>LSD</td>
<td>6.39</td>
<td>8.3</td>
<td>9.37</td>
<td>4.82</td>
<td>5.37</td>
<td>5.5</td>
</tr>
</tbody>
</table>

The different litters refer to significant difference among group (P ≤ 0.05)

n=8

Discussion:

Selenium is an essential micronutrient in animals (21). Its three levels of biological activities have been shown in animals: 1- trace concentrations are required for normal growth and development; 2- moderate concentrations can be stored and homeostatic functions are maintained; and 3- elevated concentrations can result in toxic effects (22).

Effect of selenium on some hematological parameters:

The present study showed that administration of Selenium in the different doses caused a decrease in RBC, PCV, Hb level and increase in WBC. selenium in erythrocytes is distributed between glutathione peroxidase and haemoglobin (23) and selenium is taken up by red blood cells within several minutes, reduced to selenium by glutathione, and transferred to the liver (24) and Chronic exposure to selenium has been reported to create a hypochromic anemia in Women (25) and a decrease in blood Hb level may be due to Se supplementation in an inappropriate dose could affect protein synthesis in addition to its ability to bind with protein to form Se binding proteins (26). Thus, the reduction of blood Hb level in Se supplemented rats could be attributed to the toxic effect of this element. this result
is agreement with (8, 27).

The WBCs are inextricably involved in the regulation of immunological function (28) and a prolonged exposure to a metal may inflict immunological deficiencies where the toxicant may work as an antigen. The rise of total WBC counts at different concentrations of selenium may be due to malfunctioning of haematopoietic system caused by selenium intoxication.

The increase of WBC may be due to the biological importance of Selenium at least 3-folds: First, it forms the prosthetic group of some critical selenocysteine-containing enzymes, such as glutathione peroxidase, iodothyronine 5-deiodinase, and thioredoxin reductase (29). Second, sodium selenite is protective against a number of toxicants. Third, selenium excessive intake causes toxic potential (30).

**Effects of selenium on liver enzymes:**

The obtained data revealed that administration of Selenium caused significant increase in ALT, AST. Aspartate transaminase (AST) is widely used to assess the liver function. ALT is a cytoplasmic enzyme while AST is found in both cytoplasmic and mitochondria. ALT is increased in acute hepatitis (viral or toxic), Jaundice, liver cirrhosis. AST is increased in myocardial infarction, liver diseases, liver cancer and liver cirrhosis (31). The biochemical mechanism for the toxicity of Se and related compound is the mediation of oxidative stress mechanisms (3). It has been found that excess Se could inhibit dehydrogenase enzymes, and remove the sulfhydryl groups essential to cellular oxidative processes (32).

**Effects of selenium on some reproductive parameters:**

The injection of selenium caused decrease in the motile, concentration and alive sperms and increase in the dead and abnormal sperms. This results is agreement with (33, 13, 34).

The toxic effect of Selenium might be attributed to destructive impact of high microelement concentration on the spermatozoa insert, regarded as an energetic area of male gamete and with the impairment of processes, including physiological oxidation or cell respiration, occurring in mitochondria (35). Studies have shown that Selenium is localized in the outer membrane of sperm mitochondria implies that Selenium plays the main role in maintaining a proper composition of this structure (36).

These changes might be potentially caused (in view of the short duration of treatment) by...
effects of free radicals in addition with direct effects of stress hormones to the structure and functions of testis. motility are closely related to sperm mitochondrial section, site of energy production (37) reported morphological damage of mitochondrial section of spermatozoa after the peroral feeding of 4 ppm sodium selenite to the rats which supports our findings of reduced parameters of motility. These data suggest that the mitochondrial section might be directly or indirectly the target site of spermatozoon damage caused by selenium. Selenium in excess causes oxidative stress in testicular germ cells, damage of RNA and DNA, reduction of the expression of genes required for synthesis of proteins essential for spermatogenesis (12).

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