Selenium Nanoparticles And Silymarin To prevent Lead Acetate-Induced Toxicity On Reproductive Performance Of Male Rats

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Abstract

The current study was carried out to study the effect of selenium nanomaterials and the extract of the silymarin plant on male rats poisoned by lead phastates and to study their effect on male genital. The study was conducted in the eighth month of last year and for 40 days, Where 50 males were taken from rats and were divided equally into 5 groups, which are as follows: Control group 10 animals were gavage with distilled water for 40 days. The lead acetate group was administered in single dose and left for the duration of the experiment. The third group was administered with a single dose of lead acetates and then gavage with selenium nanoparticles for 40 days. The fourth group administered lead acetate with a single dose then gavage with extract of silymarin for 40 days. And the fifth and final group was gavage with lead acetate with a single dose and then administered combination of selenium nanoparticle and extract of silymarin for 40 days. The results of the study exposed that selenium nanotubes and salimarin extract had a positive effect(P < 0.05) on all the parameters measured in this study. In conclusion, ScNPs and extract of silymarin in the supranutritional dose has a positive effect on the reproductive function of male.

Keywords: Selenium nanoparticles, Silymarin, Reproduction, Glutathione, MDA, LH, FSH and Testeostrone.

Introduction

Lead forms everywhere environmental heavy and polluting professional pollution. The US Centers for Disease Control (Pb) has been identified as the most widespread environmental toxin that stimulates most common diseases in children [1]. Continuous exposure to the public to this metal leads to many harmful health problems, including blood, nervous, gastrointestinal and renal diseases [2]. Thus,
reduced activity of these enzymes, including the antioxidant enzyme, glutathione and MDA may rise tissue exposure to oxidative damage [3]. Nanotechnology enables researchers to design nanoparticles with larger spaces, allowing for faster interactions with biological targets. These new properties of nanoparticles contribute to its promising future application in manufacturing, energy and medical fields [4]. It is clear that nanoparticles (NPS) are used to enhance the antioxidant potential. For example, Nano-Se has been shown to enhance the activity of vital proteins effectively with pathological bioavailability and minimal toxicity. Vehicles extracted from natural plants have effective preventive and preventive roles against stressful and pathological conditions. Silymarin is a flavonoid with an effective anti-oxidant activity has been isolated from the seeds *Siebum marianum* milk. Anti-inflammatory action is one of the roles of protection against silymarin, where silymarin protection has been reported from hepatotoxicity caused by many different factors. Furthermore, Silymarin has been reported to have anti-reproductive, antiviral, antiviral and immunosuppressive properties [5].

**Materials and methods**

**Experimental design:-**

Male rats were assigned to 5 equal groups (10 each) and treated as follow:

1. C:- 10 male rats were drenched with drinking water daily for 40 days.
2. T1:- 10 male rats were drenched with Lead acetate (0.5 mg/ kg b.w) daily for 40 days.
3. T2:- 10 male rats were Lead acetate induce toxicity and drenched with Selenium nano with (0.8 mg/ kg b.w) daily for 40 days.
4. T3:- 10 male rats were Lead acetate induce and drenched extract of silymarin (200 mg/ kg b.w) daily for 40 days.
5. T4:- 10 male rats were Lead acetate induce and drenched with silymarin suspension (200 mg/ kg b.w) daily and in same time drenched Selenium nano with (0.8 mg/ kg b.w) for 40 days.

Male rats were monitored throughout experimental periods. At the end of each treatment and control group, male rats were anesthetized (by injecting ketamine 0.3 ml + 0.1 ml of zylazine / kg bw. ip), the blood samples obtained from the ventral vein were dissected in non-heparinized tubes. Serum samples were separated (centrifuged at 3000 rpm for 5 minutes) and kept at -20 °C until the evaluation of GSH and MDA concentrations. Testicular samples were obtained from each male and kept directly at -70 °C to assess the levels of GAPDH expression as homogenous, gene, hydrogen and testosterone genes using green-pigmented qRT-PCR.
Laboratory measurements: Blood samples were obtained after fasting for at least 10 hours. Blood serum was obtained and analyzed for GSH and MDA. The Elisa test was used to determine FSH, LH and Testosterone.

Molecular analysis: Total RNA was isolated from blood samples that give the procedure formulated by the TRIzol® inhibitor (Promega co. USA). After the separation, the quantity (ng / μL) in addition to total RNA differentiation was critical using the UV / VIS Nanodrop spectrometer (OPTIZEN POP.MECASYS, Korea). The single stranded cDNA was regenerated to the second strand cDNA which was used as a re-copy prototype. QRTTPCR has been achieved using the AccuPower® Greenstar™ qPCR PreMix (Bioneer, Korea) and Exicycler™ 96 Block Quantum Real Time Block (Bioneer, Korea). After completion of response, data analysis has been achieved, where the housework gene (GapdH) was epitomized as a regulate gene that can be charity for intention of the virtual gene expression or fold variation in mark gene (LH, FSH and Testestrone gene).

Statistical Analysis: The results were articulated as SEM. A assessment was made between groups using a t-test for students. The variances were careful important at P <0.05. Arithmetical exploration was achieved using SPSS (SPSS,2010, USA).

Results

Chang in characteristics of sperm: The results exposed in Table (1) exhibited a important variance between the control group and the study groups in (P <0.05) . The results indicated that there was a reduction in mean of the sperm count for both T1 group treated with Lead acetate for 40 days, compared to control group and other groups. The results exposed that there was a reduction in the mean of sperm motility for both T1 group treated with lead acetate for 40 days compared to the control group and other groups. The results exposed an important rise in the mean of motility of sperm, compared to other groups. The results indicated a reduction in mean of the sperm viability of T1 group treated with lead acetate for 40 days, compared to control group. The results exposed an important rise in mean of the viability of the sperm in others groups, compared to other groups. while the results exposed an important reduction in the morphological of the sperm in T1 group treated with lead acetate for 40 days, compared to control group.

| Groups | Count | Motility | Viability | Morphology |
|--------|-------|----------|-----------|------------|

Table (1): Shows the Chang in sperm characteristics of male rats treated with selenium nanoparticles and silymarin.
The values are expressed as M±SD, The stars signify important difference (p<0.05) between studied groups. Similar letter denote to non-important and difference letter denote important.

**Antioxidant Assay**

The results shown in Table (2) exposed a important rise (P> 0.05) in the level of MDA in the group of animals treated with the Lead acetate for 40 days compared to the control group as well as the groups (T2, T3 and T4), important reduction to nearly of control group. The results exposed a important reduction of P (<0.05) in the level of glutathione in the animal's groups treated with the Gemzer drug for 40 days, compared to the control group, as well as the groups (T2, T3 and T4), The results exposed a important rise in the rate The level of glutathione.

**Table (2):** Shows the Antioxidant levels of male rats treated with selenium nanoparticle and extract of silymarin with lead acetate

| Groups | MDA       | GSH       |
|--------|-----------|-----------|
| C.     | 1.69 ± 0.05 | 2.43± 0.25 |
| T1     | 2.95 ± 0.06 | 1.79 ± 0.05 |
| T2     | 1.70 ± 0.12 | 2.31 ± 0.15 |
| T3     | 1.74 ± 0.025 | 2.45 ± 0.03 |
| T4     | 1.67 ± 0.03 | 2.35 ± 0.05 |
| T5     | 1.73 ± 0.01 | 2.52 ± 0.06 |
| L.S.D  | 0.080 | 0.016 |

The values are expressed as M±SD, The stars signify important difference (p<0.05) between studied groups. Similar letter denote to non-important and difference letter denote important.
Genes expression

Relative quantification of LH genes expression: In the current study, a important reduction (P <0.05) of the level of blood gene expression LH (fold changes) was reported in T1 lead acetate compared control group and other groups, T2 treated with selenium nanoparticle, T3 treated with extract of silymarin and T4 combination group. (Figure 1).

![Figure 1: Gene expression levels (fold changes) of LH gene in Male Rats. The values are expressed as M±SD, The stars signify important difference (p<0.05) between studied groups.](image1)

Relative quantification of FSH genes expression: In the current study, a important reduction (P <0.05) of the level of blood gene expression FSH (fold changes) was reported in T1 lead acetate compared control group and other groups, T2 treated with selenium nanoparticle, T3 treated with extract of silymarin and T4 combination group. (Figure 2).

![Figure 2: Gene expression levels (fold changes) of FSH gene in Male Rats. The values are expressed as M±SD, The stars signify important difference (p<0.05) between studied groups.](image2)
Relative quantification of Testosterone genes expression: In the current study, a important reduction (P <0.05) of the level of blood gene expression testestrone (fold changes) was reported in T1 lead acetate compared control group and other groups, T2 treated with selenium nanoparticle, T3 treated with extract of silymarin and T4 combination group. (Figure 3).

![Testosterone expression levels](image.png)

**Figure (3):** Gene expression levels (fold changes) of Testosterone gene in Male Rats. The values are expressed as M±SD, The stars signify important difference (p<0.05) between studied groups.

**Discussion**

The primary objective of this work was to investigate the effective of SeNPs on testicular and squamous rats in male rats, asserting that supranutritional SeNPs were better used to develop the male reproductive system. Sperm analysis is routinely used to evaluate male reproductive performance and to describe the reproductive toxicity of environmental or therapeutic factors [6]. Sperm concentration, morphology, vitality and movement parameters are closely related to sperm quality, which can be regarded as a marker for clinical evaluation of male reproductive performance. SeNPs can be supplemented with supranutritional sperm concentration, vitality, and motion factors. SeNPs improved at the level of these supranutritional traits compared with control. The processes of sperm formation and the formation of steroids (testosterone) are two major physiological functions of the test. It is called the process of sperm formation
where sperm (spermatogonia, primary sperm, secondary sperm, sperm and sperm). Germ cells include tubular tubes and Leydig cells. The seminal tubes consist mainly of sperm cells and supporting cells. Testosterone and androgens are necessary for the development of natural sperm, which activate genes in the cytotolll cells to promote differentiation of sperm [7]. Clear pathological changes and lower concentrations of testosterone weakened sperm formation in a higher dose group than SeNPs. These results are largely supported by increased activity of enzyme markers released as soon as tissues are damaged. Spermatozoa is highly susceptible to attack by the reactive oxygen metabolites by virtue of the high concentrations of polyunsaturated fatty acids in the sperm plasma membrane [8]. If the sperm are attacked by ROS to induce the MDA product, then the lipid peroxide fraction that results in the loss of unsaturated fatty acids from the plasma membrane and a similar decrease in the fertilizing potential of these cells [9]. Selenium functions its biological functions as selenocysteine residues specifically incorporated into polypeptide chains of individual proteins(selenoprotein) [10, 11]. While excess selenium, which is not used in this way, is due to the toxicity of selenium. Glutathione, a representative of selenoprotein, is a family of enzymes with antioxidant functions [12]. As situational evidence indicates that there is no proliferation without selenoprotein, GSH, GSH, traditional cell neurotransmitter, GSH is used exclusively as a substrate for reducing H2O and a limited number of organic hydroperoxides including hydroperoxide cumene and tert-butyl hydroperoxide. GSH activity in the testis, however, is low[13]. Testicular pattern is an unusual pattern with some hegemony in the macrophages [14]. The health effects of nanoparticle exposure were the main focus of nanomolecular studies [15]. Non-specific oxidative stress is a major mechanism of toxicity caused by nanoparticles [16]. The surveys stimulated this higher level of MDA. This result indicated that silymarin suspension suspension at the given dose (200 mg / kg bw) and the duration of certain experiments did not have any side effects on body functions in normal mice, but instead had a positive positive effect in improving the antioxidant status For oxidation. In this study, we tried to find out how SELIMARINE can improve drug interaction in male inhibitory and immunosuppressive rats. Current clinical results revealed that silymarin administration importantly improved antioxidant activity even in a healthy animal model, not only in immunosuppressed animals, researchers reported, [17] It has been reported that high levels of MDA in the blood, as a vital indicator or indicator of the degree of oxidative stress due to its ability to interact with lipoprotein, Usually accompanied by hyperlipidemia that promotes lipid peroxidation and / or oxidative damage, the protection provided by silymarin appears to be due to an increased anti-free radical mechanism by increased scanning mechanism, where the silymarin actions can be performed through its activity against lipid peroxidation, Normal metabolism and oxidative stress There are large amounts of metabolites and free radicals in the production of mitochondria, as a result of energy production, but the available or effective method of oxidation is the important variable between the two states [18]. One of the most important efficacy of silymarin, as demonstrated by previous researchers [19] is by increasing serum GSH levels responsible for the production of glutathione [18].
There have been growing concerns focusing on the beneficial effects of many antioxidants and naturally produced substances against toxin-induced reproductive toxicity, which can be caused by free radical scavenging and by restoring oxidative/antioxidant balance. [19] In this study, giving silymarin to male rats improved all the negative changes observed in the production of reproductive hormones, highlighting their role in combating oxidative damage caused by free radicals. This study revealed that silymarin treatment resulted in no change in serum LH. [20] The lowest dose of cholesterol (phytoestrogen) was found to suppress basal LH levels, whereas high doses did not have a detectable effect. Silymarin therapy which insisted on the prevention of pregnancy and resembles the values of FSH Values of older mice and postmenopausal women [21] and [22]. In male rats, it is an important increase in levels of testosterone in the blood and levels of the hormone lutein through treatment with silymarin in one month. [23] confirmed that estrogen is required for the normal function of the certified channel and is necessary for long-term fertility in male rodents.

Conclusion

SeNPs and extract of Silymarin in the supranutritional dose has a positive effect on the reproductive function of male rats. More research is needed to discern the mechanism by which SeNPs improve reproductive function in rats.

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