Effect of alpha Lipoic Acid on Some Reproductive Hormones in Adult Male Wister Rats Exposed to Hydrogen Peroxide

Mohammed Salih Alwan1 and Baraa Najim Al-Okialy2

1Department of Physiology and Pharmacology, College of Veterinary Medicine/ University of Wasit, Iraq.
2Department of Physiology and Pharmacology, College of Veterinary Medicine/ University of Baghdad, Iraq.

E-mail: dr.Mohammedalali1966@gmail.com

Received date: 6 May 2019 Accepted: (449) 12 Juny 2019 page (79-87) Published: 30 Juny 2019

Abstract

The present study was aimed to investigate the role of alpha lipoic acid (ALA) on reproductive hormonal profile in adult male Wister rats treated with hydrogen Peroxide. Forty adult male rats were divided randomly into four equal groups (10 rats /group) and were handled daily as follows for 56 days: Control group (C) were intubated distilled water and received tap water ; group T1 were intubated 60mg/kg B.W of ALA and received tap water; group T2 administered hydrogen peroxide H2O2 in tap water at concentration of 0.05%, while group T3 were intubated 60mg/kg B.W of ALA and received ordinary tap water containing 0.05% H2O2. Blood samples were collected at 0, 28 and 56 days of the experimental periods for measurement of serum follicular stimulation hormone (FSH), lutelizing hormone (LH) and testosterone (T) concentrations. The results revealed that group T2 showed a significant decrease in FSH and testosterone concentrations. While there were significant increase in previous parameters in T3 group compared to T2 group. Whereas, rats in group T3 shows significant improvement in above serum hormonal profile by repairing role of alpha lipoic acid against hydrogen peroxide in (group T2). In conclusion, alpha lipoic acid mitigated the deleterious effect of that-induced H2O2 induced pituitary-testicular dysfunction through antioxidant mechanism by free radical scavenging properties.

Key Words: Alpha lipoic acid, hydrogen peroxide, LH, FSH and testosterone.
Hydrogen peroxide (H$_2$O$_2$) is non-radical oxidant produced in vivo by many reactions via a wide variety of enzymes, it is a common reactive oxygen species (ROS), which is highly reactive and can interact with nearby molecules, inducing oxidative stress (OS) (4). Many studies showed that H$_2$O$_2$ is one of the most toxic reactive oxygen species in the mammalian spermatozoa (5 & 6). A considerable amount of literature has been published about the role of OS and ROS in the etiology and/or progression of a number of diseases (7).

Alpha-Lipoic acid is a naturally occurring nutraceutical, whose therapeutic effect has been related to its antioxidant activity and its ability to ameliorating oxidative damage (8 & 9). ALA works on the cellular level to help produce energy in the body, as a part as, a coenzyme in the citric acid cycle by preparing the fuel for the mitochondria, and plays a vital role in mitochondrial electron transport reactions required for cellular energy production (10). It has been reported that ALA have powerful antioxidant abilities, equal to that of coenzyme Q 10, vitamin C, and vitamin E (11). Unlike other antioxidants, ALA alpha lipoic acid has ability to neutralize free radicals in intracellular and extracellular environments (12). Therefore, the current study was designated to explore the protective role of ALA to ameliorating the deleterious effect of H$_2$O$_2$ on testicular function.

Materials and Methods
Forty adult male rats were randomly divided into four equal groups (10 rats/group) and were handled daily as follows for 56 days: Group C, rats in this group were intubated distal water plus received ordinary tap water and served as control; Group T1, rats in this group were intubated 60mg/kg B.W. of ALA (24) as well as received ordinary tap water; Group T2, rats were administered H$_2$O$_2$ in tap water at concentration of 0.05% (25) and Group
T3, rats in this group were intubated 60mg/kg B.W. of alpha lipoic acid and received ordinary tap water containing 0.05% \( \text{H}_2\text{O}_2 \). Blood samples were collected and transferred to a gel tube without anticoagulant at zero, 28 and 56 days of experiment. Sera were isolated estimate follicular stimulating hormone (FSH), Luteinizing hormone (LH) and Testosterone hormone (T) concentrations. Assessment of serum Hormonal using ELISA Technique for FSH, LH and T hormones and optical density (OD value) of each well at once, used a micro-plate reader set at 450 nm. Calculation of results of the ELISA results were calculated depend on the optical density reading for each standard and samples optical density. Then the standard curve was plotted by the mean OD value for each standard on the Y-axis against the concentration on the X-axis and draw a best fit curve through the points on the graph.

**Statistics**
Statistical analysis of data was done on the basis of tow – way analysis of variance using a significant levels of (p<0.05), furthermore LSD test was used to determine specific differences (13).

**Results**
In this study all treatment groups showed non-significant (p>0.05) difference in serum FSH concentration compared to control at zero time Figure 1-A. Statistical analysis of the data indicate that the mean value of serum FSH concentration of T2 group (81.29 ±1.8) and (76.28±1.56) was significantly (p<0.05) decrease after 28 and 56 days of treatment periods while in combination ALA with \( \text{H}_2\text{O}_2 \) in drinking water (group T3) showed a significant (p<0.05) increase in serum FSH level at two treatment periods as compared to group T2. Highest significant (p<0.05) decrease in this parameter was observed in T2 and T3 groups after 28 and 56 days of the treatment as compared with Zero time. While there are no significant (p>0.05) differences in the mean values of serum LH level between experimental groups during pretreatment period (Zero time), a significant (p<0.05) increase in this parameter was observed at days 28 and 56 of the experiment in group T2 with mean value of (46.29±1.41) and (44.85±1.24) respectively compared with T1, T3 and control groups Figure 1-B. Combination of ALA and \( \text{H}_2\text{O}_2 \) caused non-significant (p>0.05) differences in serum LH concentration at the end of the experiment comparing to control group. Within the time, non-significant (p>0.05) differences were observed in serum LH level at 28 and 56 days of the experiment in control, T1 and T3 groups comparing with pretreated period. Figure 1-C pointed to the mean values of serum testosterone concentration of control and three treated groups. The results showed a significant (p<0.05) decrease in serum testosterone concentration in groups T2 and T3 as compared with control and T1 treated groups. The figure also shows non-significant (p>0.05) differences in serum testosterone concentration at two treatment periods between control and T1 groups. With exception to control and T1 groups, within the time significant (p<0.05) decrease in T2 and T3 treated groups comparing to the pretreated period.

**Discussion**
The results that rats received 0.05% \( \text{H}_2\text{O}_2 \) (group T2) cause significant decrease in testosterone concentration as well as FSH and LH as compared to other treated groups. Similar results were obtained by other investigators (14, 15 &16). Study the hormonal profile in adult male is critical point to overview the rate of reproductive performance. Steroidogenic of the testes characterized by an intense synthesis and secretion of testosterone. Furthermore, FSH and LH as well as other biochemical activity play an important role in testicular tissue functions (17). It is well
known that pituitary gland is highly sensitive to oxidative stress (18).

Results of the present study also confirmed that the detrimental effect of H$_2$O$_2$ on the pituitary hormones may be through suppressed serum prolactin, LH, FSH, and GH concentrations. This was agreed by other studies (19), (14) and (16). Moreover, any chemical agent suppressing the secretion of pituitary gonadotropins has produce antispermatogenic and antifertility effects, impaired secretion of LH results in deficient androgen secretion by the testes, inhibition of spermatogenesis and loss of libido (20). It is often well known that small amounts of ROS are produced in the Steroidogenic pathway and spermatozoa are necessary for fertilizing capabilities (21), apoptosis and spermatogenic failure (22).

Hydrogen peroxide toxicity may be affected pituitary gland causing a reduction in gonadotropins secretion leading to inhibition of steroid biosynthesis by Leydig cells and then decrease in testosterone concentration (23). A reduction of testosterone concentration in H$_2$O$_2$ treated group was documented in the present study by histological changes of the testis. As shown in the results significant improvement of testosterone, FSH and LH concentrations were observed in rats treated with ALA+ H$_2$O$_2$ (group T3) as compared to group T2. So, it could be concluded that that alpha lipoic acid reduced testicular damage that caused by H$_2$O$_2$, as evidenced by the significant increase of serum hormonal profile and due to ALA is a multifunctional antioxidant.

**Conclusions**

From the results and discussion of the current study, it can be concluded that oral intubation of 0.05% H$_2$O$_2$ to adult male rats caused significantly decrease in hormonal profile including Testosterone, FSH and LH. On the other hand, intubation of alpha lipoic acid (ALA) possessed antioxidants activity with improvement male reproductive performance against oxidative stressed rats induced experimentally by H$_2$O$_2$. 


Figure 1 (A) Presents effect of alpha lipoic acid (ALA) for 28 and 56 days on serum FSH hormone concentration compared to the zero time of experiment of hydrogen peroxide (H₂O₂) treated male rats.

Values are expressed as means ± SE n = 7/group.

C: control received tap water.

T₁: gavages alpha lipoic acid (ALA) (60 mg/ kg B.W).

T₂: received 0.05% H₂O₂ in drinking tap water.

T₃: received 0.05% H₂O₂ in drinking tap water plus 60 mg / kg B.W of ALA.

Means with different small letters denote significant differences (p<0.05) between groups.

Means with different capital letters denote significant differences (p<0.05) within groups.
Figure.1 (B) Presents effect of alpha lipoic acid (ALA) for 28 and 56 days on serum LH hormone concentration compared to the zero time of experiment of hydrogen peroxide (H$_2$O$_2$) treated male rats
Values are expressed as means ± SE n = 7/group
C: control received tap water.
T$_1$: gavages alpha lipoic acid (ALA) (60 mg/kg B.W).
T$_2$: received 0.05% H$_2$O$_2$ in drinking tap water.
T$_3$: received 0.05% H$_2$O$_2$ in drinking tap water plus 60 mg/kg B.W of ALA
Means with different small letters denote significant differences (p<0.05) between groups.
Means with different capital letters denote significant differences (p<0.05) within groups.
Figure 1 (C) Presents effect of alpha lipoic acid (ALA) for 28 and 56 days on serum testosterone hormone concentration compared to the zero time of experiment of hydrogen peroxide (H₂O₂) treated male rats.

Values are expressed as means ± SE n = 7/group

C: control received tap water.

T₁: gavages alpha lipoic acid (ALA) (60 mg/kg B.W).

T₂: received 0.05% H₂O₂ in drinking tap water.

T₃: received 0.05% H₂O₂ in drinking tap water plus 60 mg/kg B.W of ALA.

Means with different small letters denote significant differences (p<0.05) between groups.

Means with different capital letters denote significant differences (p<0.05) within groups.
References


