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ORIGINAL ARTICLE

Effect of Zinc on Serum Testosterone Level in Albino Rats

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ABSTRACT

Objective: To study the effect of zinc on serum testosterone levels in Albino rats.

Material and Methods: This study was conducted in the Department of Pharmacology and Therapeutics BMSI JPMC Karachi. In this study 60 albino rats were divided into four groups, 15 rats in each group. Group one was control group (normal diet was given to this group), group two was given indomethacin, group three was given zinc and group four was given combination of Zinc and indomethacin. All the drugs were given for 12 weeks. Serum testosterone level was checked at the end of study and finally data was analyzed statistically using SPSS version 18.

Results: In group 2, serum testosterone level (3.12±0.63) was low as compared with control (6.26±0.15). In group 3, mean testosterone was high (6.97±0.63) when it was compared with control (6.26±0.15). In group 4, mean testosterone was low (5.15±0.73) but not significant when compared with control (6.26±0.15).

Conclusion: Zinc has a protective role on testes and it increases testosterone and fertility.

Key words: Zinc, serum testosterone level, Indomethacin, fertility.

Author`s Contribution
[1] Conception, synthesis, planning of research and manuscript writing Interpretation and discussion
[2,3] Data analysis, interpretation and manuscript writing, [4] Active participation in data collection.

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Introduction

Zinc is an essential mineral and essential trace element present in more than 200 metallo enzymes.[1] Zinc deficiency leads to gonadal dysfunction, decreases testicular weight and causes shrinkage of seminiferous tubules.[2] Previously it was known as "white vitriol". [3] Zinc is available in the form of zinc oxide, zinc acetate, and zinc gluconate. [4] It is a constituent of enzyme superoxide dismutase, which protects cells from damage caused by free radicals. Zinc reduces oxidative stress in developing sperm and improves sperm quality. [5] It possesses antioxidant properties [6] and is present in rich quantity in semen.[7] Testosterone is a steroidal androgenic hormone, it is the main male sex hormone and an anabolic steroid. [8] It is necessary for normal sperm development, it activates genes in sertoli cells and promotes differentiation of spermatogonia.[9] By restricting zinc in diet, significant decrease in serum testosterone concentrations will occur, on the other hand zinc supplementation increases serum testosterone levels.[10] Indomethacin is a non-steroidal anti-inflammatory drug that is a nonselective inhibitor of cyclooxygenase (COX) 1 and 2. Indomethacin produces toxic effect to testes. Oligospermia and azoospermia have been reported after the use of indomethacin. [11] Zinc is the second most abundant element in human and plays multifactorial role in sperm function,[12] Adult human body contains approximately 1-3 g of zinc and recommended daily allowance (RDA) is 11-8 mg/day for men and

women respectively.[13] Zinc has many biologically significant interactions with hormones and has special role in production, storage and secretion of hormones. Zinc deficiency has a major impact on testosterone, insulin and adrenal corticosteroids. [14]

Material and Methods

This experimental study was conducted in the Department of Pharmacology and Therapeutics BMSI, JPMC Karachi from 30th June 2013 to 30th December 2013. Sixty male albino rats, 2.5 to 3.5-month old weighing 200 to 300 grams were taken from animal house. Sample size of 60 rats was calculated by using computer program open Epi version 2 with power of 80 of the test and 95% confidence interval (www.openepi.com/samplesize/sspropor.htm). Zinc was obtained in the syrup form available as 20 mg /5ml. Total 5 mg/kg was given orally for 12 weeks. Indomethacin powder dissolved in distilled water was given orally with a dose of 10mg /kg to selected animals for 12 weeks. Selected sixty albino male rats were divided into four equal groups, 15 animals in each group.

Group I (control group): The 15 albino rates of this group were kept on normal diet.

Group II (Indomethacin group): In this group 15 Albino rats were treated with indomethacin.

Group III (Zinc group): The 15 albino rats in this group were given zinc.

Group IV (Combination group): Combination of zinc and indomethacin in their respective dose was given for 12 weeks.

Before starting the research, animals were kept in room temperature for one week for acclimatization. All the animals were observed daily for their physical activity and weighed weekly. Medication was given to each rat in their proper doses for three months. At the end of study period, weight of albino rats was observed and then they were scarified. A mid line incision was made up to scrotum and extended upwards to the thoracic region. Blood sample was taken from each animal by intra-cardiac puncture with the help of disposable syringe, for the detection of serum testosterone levels. Blood was taken from rat heart and was shifted to plain glass tube and retained in standing position for one hour for clot formation. After that, the centrifugation was done. The separated serum was then shifted to small sterile plastic cups and was stored in laboratory refrigerator at -80°C. Before estimation, the samples were drawn from refrigerator and were kept at room temperature for two hours. Afterwards the serum testosterone levels were estimated. All data was analyzed with SPSS version 18. Quantitative variables were expressed as mean±SD. Statistical comparison between groups was done by analysis of variance (ANOVA). In order to compare means, student t test was applied. p value ≤0.05 was considered as statistically significant.

Results

Albino rats which were given Indomethacin 10mg/kg for 90 days, revealed an overall decrease in mean serum testosterone level as compared to control. Decrease in mean serum testosterone level was highly significant (p-Value=0.0001) on day 90 as compared to control, with overall reduction of 34% (Table 1).

Table 1: Mean serum testosterone levels within different groups in albino rats (n=60)

Groups	Serum testosterone level observed(ng/dl) mean±SD
Control group	6.26±0.15
Indomethacin group	3.12±0.63**
Zinc group	6.97±0.63
Zinc + Indomethacin group	5.15±0.73*

** very low levels * slightly low levels

Albino rats given zinc 10 mg/kg revealed increase in mean serum testosterone level as compared to control. The increase in serum testosterone level was highly significant (p-Value 0.001) on day 90 as compared to control, with overall increase of 6% (Table 2; Figure 1). Albino rats were given combination of zinc and Indomethacin 10mg/kg for 90 days with slight decrease in mean serum testosterone level as compared to control. The decrease in serum Testosterone level was not significant on day 90 i.e.5.15±0.73 as compared to control 6.26±0.15, with overall decrease of 2%. Albino rats given zinc 10mg/kg revealed increase in mean serum testosterone level as compared to indomethacin. The increase in serum testosterone level was highly significant (p-Value 0.0001) on day 90 as compared to indomethacin 3.12±0.6, with overall increase of 38%. Albino rats given

Table2: Comparative effects of different drugs on mean serum testosterone level (ng/dl) in albino rats (n=60)			
Groups	**Animals (n)**	**Serum testosterone at day 90**	**p-value**
Control Vs Indomethacin	15	6.26±0.15 vs 3.12±0.63	0.0001**
Control Vs Zinc	15	6.26±0.15 vs 6.97±0.63	0.0016**
Control Vs Indomethacin +Zinc	15	6.26±0.15 vs 5.15±0.73	0.119
Indomethacin Vs Zinc	15	3.12±0.63 vs 6.97±0.63	0.0001**
Indomethacin Vs Indomethacin +Zinc	15	3.12±0.63 vs 5.15±0.73	0.0001**
Zinc Vs Indomethacin +Zinc	15	6.97±0.63 vs 5.15±0.73	0.001**

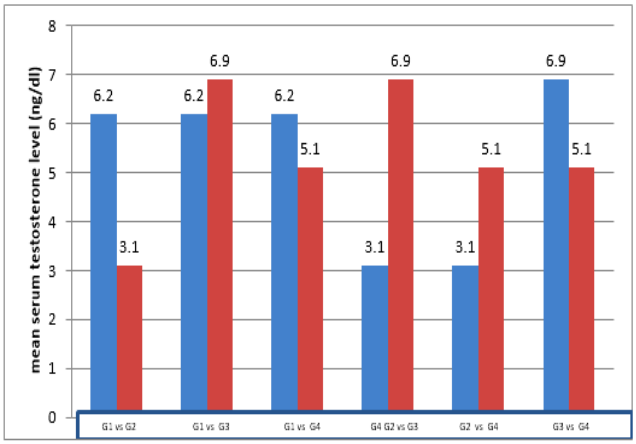

Figure 1: Comparing mean serum testosterone level in different groups G1= Group 1 (control), G2 =Group 2 (Indomethacin), G3= Group 3 (Zinc), G4= Group 4 (Indomethacin+Zinc)

indomethacin + zinc 10mg/kg showed increase in mean serum testosterone level as compared to indomethacin. The increase in serum Testosterone level was highly significant (p-Value 0.0001) on day 90 as compared to indomethacin 3.12±0.6, with overall increase of 32%. Albino rats given indomethacin + zinc for 90 days revealed slight decrease in mean serum testosterone level as compared to zinc. The decrease in serum Testosterone level was highly significant (p-Value 0.0001) on day 90 i.e. as compared to zinc, with overall decrease of 16%. (Table 2; Figure 1).

Discussion

Zinc is essential for spermatogenesis. It causes induction of metallothionein which protect cells from toxicity caused by free radicles.[13] Zinc deficiency in rats causes changes in seminiferous tubules and in serum levels of testosterone.[14] The results of present research indicate that administration of indomethacin in a dose of 10mg/kg suppresses synthesis of testosterone due to toxic effects of indomethacin on seminiferous tubules in testes. Similar finding was reported previously by Jahangir et al in 2009.[15] Seminiferous tubule atrophy was observed by administration of propoxur (PPX) by Oyewopo et al in 2010. [16] The results of combination therapy showed the protective effects of zinc in presence of indomethacin. When combination therapy was compared with control, there was no significant difference. Study conducted by Falana et al observed similar findings, in which rats were divided in four groups and were treated for 30 days, group 1 was control group, group 2 received lead, group 3 received zinc and selenium, group 4 received lead with zinc and selenium. [17] After completion of study period, blood was taken, mean serum testosterone levels were significantly decreased in lead treated group compared with control. The administration of lead with selenium and zinc increased the serum levels of testosterone. The zinc and selenium treated group had significant increase ($p<0.05$) in mean serum level of testosterone when value was compared with control. This is compatible with our study in which group 3 (zinc) showed increased mean serum testosterone (6.97±0.63). When it was compared with control (6.26±0.15) the result was highly significant ($p<0.001$). Similarly, in group 4 mean serum testosterone level was low but when compared with control it was not significant.

Conclusion

Synthesis of testosterone is inhibited by some drugs/ chemicals due to its toxic effects on seminiferous tubules. Zinc plays protective a role on testes and increases production of testosterone to normal range.

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