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The effects of hydro-alcoholic extract of spinach on pituitary-gonadal axis in male mice

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ABSTRACT

Spinach (*Spionacia oleracea L.*) is from *chenopodiaceae* family. This plant has many medicinal effects which are the most important aspects. This plant is really important because of having antioxidants and the highest absorption potential among vegetables for oxygen radicals. 75 male mice were divided randomly in five groups including control, placebo, and three experimental groups (50, 100, and 200mg/kg of the extract). The extract was injected in peritoneum for 20 days. Normal saline was used for placebo group. At the end of period, blood samples were taken and testosterone, LH and FSH amounts were measured. Obtained data were analyzed using SPSS program and one way ANOVA. Results showed that FSH and LH amounts were increased by 200 mg/kg group. Testosterone was increased in 50 and 100mg/kg groups but not in 200 mg/kg group. On the whole, results showed dose dependent effect of spinach extract on pituitary-gonadal axis of male sex which can affect reproduction potential.

Keywords: spinach, LH, FSH, testosterone, mice

INTRODUCTION

Infertility is one of the complicated medicine problems which man has been involved in it from ancient times [1]. About 13% of each community is infertile which in many cases they can be treated [2]. The relationship between nutrition and reproduction is subject that its importance is increased more and more. Primary researches proved that nutrition plays important role in reproduction and in most cases intensive deficits cause reproduction problems [3].

Pharmaceutical plants have been used from ancient times to maintain good health and improve various diseases. Spinach has been produced in traditional medicine as an effective medicinal plant. It seems that this effect is because of its organic and mineral compounds. Spinach is a diploid long day annual plant. Special properties of this plant including few chromosomes, sex chromosomes and shorten the development period made it as a suitable model plant for genetic studies and sex determining molecules. Gene translocation in this plant needs appropriate method of tissue culture [4].

Spinach is an important vegetable because of its minerals, proteins, A, B, C vitamins, antioxidants and the highest absorption potential of oxygen radicals [5]. This plant has been recommended in traditional medicine for people who do intellectual tasks [6]. Spinach also is recommended as an appetizer, anti-cough, and anti-seizure and also for pain relief and cells growth and activity in patients. Researches about anti-stress effects of spinach on Syrian mice confirmed its anti-stress effects in higher doses [2].

Considering the effective compounds of this plant this study was carried out to investigate the effects of its extract on pituitary-gonadal axis of male mice.

MATERIALS AND METHODS

75 mature male mice in weight range of 25-30g were kept for 14 days to adapt to environment. Samples had free access to food and water, 25-30°C temperature and natural light. Mice were divided in five groups randomly with fifteen members in each group. Groups were control, placebo, and three experimental groups (0.5cc of extract in 50, 100, and 200 mg/kg doses every day for twenty days). After twenty days blood samples were taken and LH, FSH, and testosterone hormones were measured using Eliza test.

Obtained data were analyzed using SPSS program and One way analysis of variance ($P < 0.05$).

RESULTS

Mean comparison results of LH and FSH hormones (mIU/ml) showed significant difference between third experimental group (200mg/kg) and control group. Other groups were not different from control group (Figure 1&2).

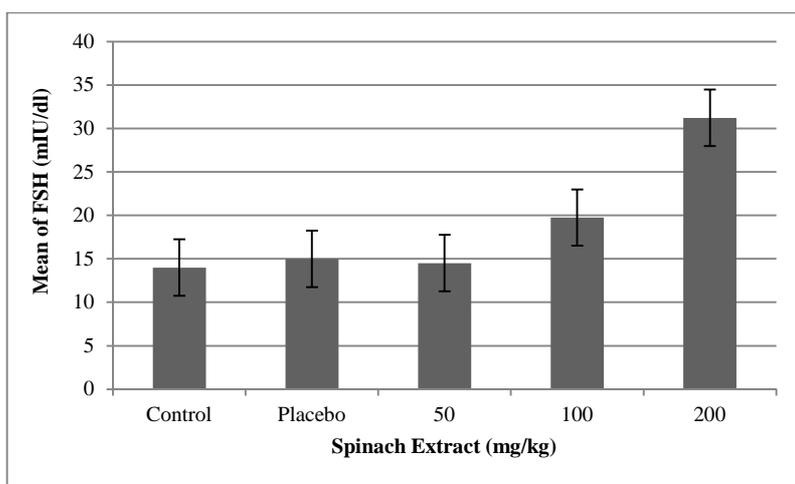


Figure 1. FSH Concentration in all groups

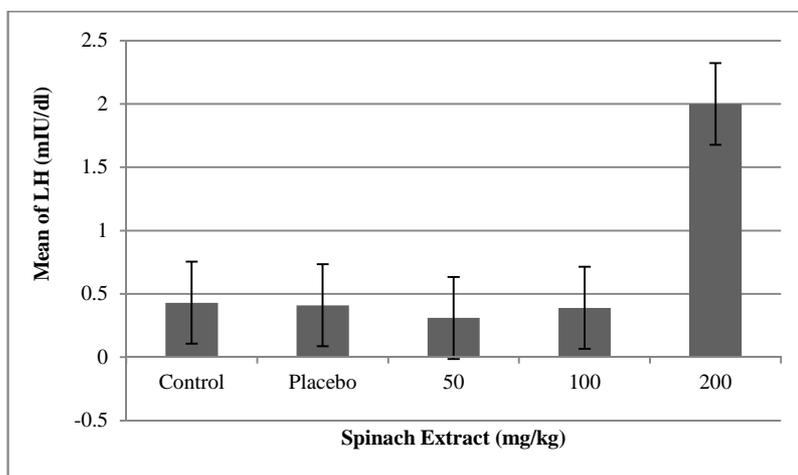


Figure 2. LH Concentration in all groups

Testosterone amount of first and second experimental groups (50 and 100 mg/kg) were different from control group whereas third group did not show significant difference (Figure 3).

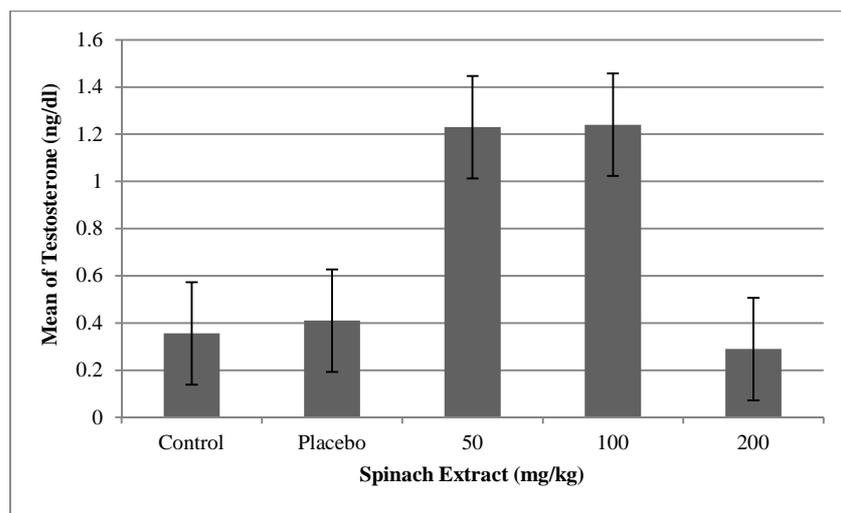


Figure 3. Testosterone Concentration in all groups

DISCUSSION

There was significant difference between third experimental group (200mg/kg) and control group whereas other groups were not different ($p < 0.05$). FSH variation was slower and less than LH hormone because FSH is less related to GnRH hormone from hypothalamus and is changed slowly and over a longer period in response to long time changes of GnRH [7].

On the other hand, connected hydrocarbon strings to TSH hormone are more than LH. Therefore, metabolic clearance of FSH is less than LH and half-life time of FSH is more [8], thus testosterone increment can affect FSH amount in serum. On the other hand, FSH feedbacks are not probably enforced only by testis steroids, but also inhibin, activin and follistatin hormones regulate the FSH concentration by central effect on GnRH [9]. FSH follows GnRH pulsatory changes less than LH [10].

So, disorder in FSH amount leads to spermatogenesis disorder. Also, FSH is essential for connecting and sticking spermatids to Sertoli cells. Studies have shown that LH secretion is affected by factors such as LH-RH, Potassium and serotonin. Direct involvement of serotonin receptors on LH secretion in rats has been approved, in normal situation, increase in testosterone amount activates negative feedback by affecting hypothalamus and front pituitary and reduces LH amount [11].

The amount of testosterone hormone was increased significantly in first (50mg) and second (100mg) groups. Studies have shown that testosterone has negative feedback effects on LH and GnRH secretion so that increases in its amount controls LH secretion which is done via controlling GnRH secretion from hypothalamus and direct effect on pituitary gonadotropins [12].

So that the hormone was increased up to definite dose, and after that was reduced. This can also be a reason for LH increment in third group. Testosterone amount of first and second experimental groups were different from control group whereas third group did not show significant difference. Furthermore, this vitamin can reduce estrogen production. Therefore, testosterone production will be increased [13]. Vitamin A is essential for reproductive organs including testes and many studies showed deficit effects of it on testis tissue [14].

Previous studies have shown that Testosterone is the survival factor of spermatogenesis, especially conversion of round spermatid to long spermatids needs it highly. Furthermore, FSH and testosterone enforce their effects in final spermatogenesis stage by increasing intracellular calcium levels and slit connections between cells and enhance this stage [9].

Spinach has anti-oxidants such as vitamins A, C, E, and polysaccharides. In general, antioxidants, particularly vitamin E found in spinach extract acts as first line of defense against peroxidation of cell membrane phospholipids. E Vitamin has the highest biological antioxidant activity and prohibits reduction of liding and sertoli cells. Increase in the number of liding cells increases produced testosterone. This vitamin also improves testes weight, somniferous tube diameter and the thickness of the germinal epithelium [11].

On the whole, flavonoids have estrogenic- non estrogenic properties at the same time. Phytoestrogens can increase testosterone level by competitive connecting to aromatase enzyme and reducing its gene expression and connecting to estrogen receptors [8].

Mentioned subjects explain increase in testosterone level of first and second experimental groups. Reduction of hormone in 200 mg/kg group can be explained by dose dependent effect of spinach.

CONCLUSION

On the whole, results showed dose dependent effect of spinach extract on pituitary-gonadal axis of male sex which can affect reproduction potential.

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