Effect of aqueous rosemary extract on some sexual hormones in male rats with high thyroxine level

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Abstract

The main purpose of this research is to evaluate the high level of thyroxine affects on each of the luteinizing hormone (LH), testosterone, follicle-stimulating hormone (FSH) of rats and to estimate the potential effect of the administration of aqueous rosemary extract and propylthiouracil against testicular toxicity induced by levothyroxine in male rats.

Introduction

Levothyroxine (LT4), synthetic thyroxine administrated orally, is the favorable treatment for patients with hypothyroidism. Moreover, it could be administered for the repression of pituitary production of thyroid-stimulating hormone in cases of not poisonous multinodular goiter and diverse forms of neoplasia in thyroid (1). However, Levothyroxine has a limit curative index, subjected patients to the risk of iatrogenic hyper- or hypothyroidism at dosage just 25% more than normal, or that are more than the most favorable dose, depending on patients' serum level of TSH (2). Data from many investigations showed that Levothyroxine induce hyperthyroidism “high level of thyroxine” in male rats at 0.5mg/kg i.p or subcutaneously for successive 12 days (3,4).

Testosterone is the major secretive production of the testis, besides the 5a-dihydrotestosterone (DHT), 17-hydroxyprogesterone, progesterone, pregnenolone, androsterone, and androstenedione (5,6). Testosterone is a first-rate endocrine agent, a piece of evidence available as a local organizer of spermatogenesis (7). Also, it is testosterone that determines the former phase of testicular migration and the growth of male exterior genitalia (8). FSH and LH are hormones of glycoprotein secreted from the pituitary gland which regulate growth, maturation, and roles of the gonad (9).

Rosemary (Rosmarinus officinalis L.) (general name, rosemary; family Labiatae), is a common domestic herb grown in abundant parts of the world. It is used for cosmetics, flavouring food, and as a drink. Rosemary is known to be a rich origin of effective metabolites (13) and is used in popular medicines (14). Archaeologists and anthropologists have found proof that herbs of rosemary have been used as a medical herb, culinary in cooking and for beauty in Mesopotamia, ancient Egypt, India and China.
(15). *Rosemary officinalis* enhanced the immunity (16), and considered as an antimutagenic agent (17). Internal use of rosemary for dyspeptic troubles and external use for rheumatic cases and hypotonic-circulatory disorders. As well it is used in conventional medicine for migraine, headache, digestive symptoms, amenorrhea, dysmenorrhea, oligomenorrhea, states of exhaustion, poor memory, and dizziness. Externally, it is used for poorly healing wounds as a poultice, also in eczema, for injuries of the mouth and throat as an analgesic, for myalgias used topically, intercostal sciatica and neuralgia. However, it must not be used throughout the pregnancy period. There were no side effects or health prohibitions are known in synchronism with the suitable administration of fixed curative doses. Allergies have been noticed on occasions. Extremely large amounts of rosemary leave misapplied for the abortion, which could lead to spasm, gastroenteritis, vomiting, kidney irritation, uterine bleeding, deep coma and death in humans (18). This study showed the effect of both and rosemary (*Rosmarinus officinalis*) and Propylthiouracil serum of luteinizing hormone, testosterone, follicle-stimulating hormone levels with experimental hyperthyroidism.

**Materials and methods**

**Chemicals**

L-Thyroxine (T4), propylthiouracil (PTU) taken up from Sigma Chemical Co., USA. The testosterone, FSH, LH and T4 hormones were managed by using commercially available kits supplied by Calbiotech Inc., USA.

**Preparation of extracts**

Dried leaves of Rosemary were purchased from a local supermarket in Kufa, Iraq, the leaves were grind and store in the dark. Briefly, we made the extract by stirring of powder 10 g/100 ml in distilled water for 30 minutes at 50 °C succeeded by fast refinement throughout a rough piece of cloth, then, through the refinery sheet. The definitive nominate was freeze-dried (lyophilized) and thereafter applied in the experiment (19).

**Experimental animal design**

Twenty-four male albino rats 170-200 gram weight were housed in the animal house under the steady status of temperature 24±2°C for three weeks prior to the beginning of the experiment, the animal house under laboratory conditions 12/12 hours light - dark cycle (20,21) and being preserved on a standard water and diet ad libitum (22,23). The animals were treated according to the specific guidelines of the veterinary medicine faculty and the study was approved by the Animal Ethics Committee of the University of Kufa. The experimental rats were divided as the following (in which every group has six rats). Group 1 animals were received 0.1 ml/day distilled water orally by gavage and considered as negative control group. Group 2 animals were treated subcutaneously for successive 12 days with Levothyroxine 0.5mg/kg. From the 13th to 24th day the animals were received 0.1ml/day distilled water by gavage orally in which considered positive control group. Group 3 animals were treated subcutaneously for successive 12 days with Levothyroxine 0.5 mg/kg. From the 13th to 24th day the animals were received 10 mg/kg propylthiouracil orally by gavage. Group 4 animals were treated subcutaneously for successive 12 days with Levothyroxine 0.5 mg/kg. From the 13th to 24th day the animals were received 10 mg/kg aqueous leaves extract of rosemary (*Rosmarinus officinalis*) orally by gavage.

**Blood collecting**

At the end of the experiment, all rats were subjected to light ether anesthesia, then sacrificed for the collection of blood, then serum separation was performed for thyroxine hormone to confirm the state of hyperthyroidism, serum testosterone level, serum follicle stimulating hormone level, and serum luteinizing hormone level.

**Statistical analysis**

The data were subjected to analysis of variance and the significance differences at P≤0.05 which were determined by analysis of variance (ANOVA), one-way by using the statistical soft ware’s sigma statistical (24).

**Results**

T4 level had significantly increased in positive-control group as compared with the negative-control group as presented in (Table 1). Data in Table 2 showed that levels of serum testosterone, LH and FSH were significantly increased in the positive-control group as compared with the negative-control group. The serum testosterone level of the experimental groups was significantly decreased in PTU treated group while highly significant decreased in aqueous rosemary extract treated group in comparison with the negative control group. Serum levels of LH and FSH in the experimental groups were significantly decreased in PTU treated group while significantly increased in aqueous rosemary extract treated group as compared with the negative-control group, in other words there were insignificance differences between the LH and FSH serum levels in rosemary treated group and positive-control group.

**Table 1: Effect of Levothyroxine on serum thyroxine in male rats**

| Group           | Thyroxine (T4) µg/dl (Mean± SE) |
|-----------------|---------------------------------|
| Negative control | 58.91±2.22B                     |
| Positive control | 76.74±1.93A                     |
| LSD             | 8.279                           |

Capital letters denote differences between groups, P<0.05.
Toxicosis of the free radical scavenging activity which leads to widespread damage to cellular compounds as membrane lipids and highly raised the normal cell viability. Thus, rosemary extract appears as a cytoprotective factor including antioxidants items rosmarinic acid, carnosic acid, carnasol, ursolic acids, butylated hydroxytoluene, and butylated hydroxyanisole, without ally hazard to carcinogenic or the cytotoxicity of the artificial antioxidants (33-35). In the present work, the administration of rosemary leaves aqueous extract after high level of thyroxine induced by levothyroxine had a poor effect on testosterone serum level which is firmly due to the low dose of rosemary extract 10 mg/kg b.w, whereas the favor was to the propylthiouracil.

### Conclusion

Although the poor effect of rosemary leaves extract on serum testosterone, we must as well be mindful of its ameliorative effects if utilized in higher dosage. Thus, further studies must be done in the future.

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### Conflicts of interest

No conflicts.

### References

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تأثير مستخلص إكليل الجبل المائي على بعض الهرمونات الجنسية في ذكور الجرذان ذات المستوى المرتفع للثايروكسين

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الخلاصة

الهدف الرئيسي من هذا البحث هو تقييم تأثير المستوى المرتفع من الهرمون الثايروكسين على هرمون لوتين، هرمون تستوستيرون، والهرمون المنبه للجريب في ذكور الجرذان. وتم استخدام مستخلص إكليل الجبل المائي والبروبيلثيوراسيل ضد سمية الخصية المستحدثة بواسطة الليفوثيروكسين. تم استخدام مجموعة من ثلاث مجموعات جرذان للتجربة. المجموعة الأولى تم معالجتها بالماء المقطر، والمجموعة الثانية والثالثة تم معالجتها بمستخلص إكليل الجبل المائي والبروبيلثيوراسيل. أظهرت النتائج انخفاضًا معيّنًا في مستويات هرمون لوتين، هرمون تستوستيرون، والهرمون المنبه للجريب في مصل الدم. يمكن الاستنتاج أن تأثير المستخلصات على هرمونات الجرذان كان له تأثير محتمل عند إعطاء مستخلص إكليل الجبل المائي والبروبيلثيوراسيل ضد سمية الخصية المستحدثة بواسطة الليفوثيروكسين.