Effect of Steroidal Extract for Fenugreek Seeds on ICSH and SSH in Albino Male Mice

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Abstract

Plants are used to treat many conditions of diseases but have many side effects which lead to deleterious effect for several systems in the body. The objectives of this Study are to investigate effective role for steroidal extract of fenugreek on ICSH and SSH in male mice hormones. The steroidal extract of fenugreek was prepared by hydrolyzing the grinded powder of fenugreek seeds with HCl (2N) in water bath, then filtered and air-dried and put in soxhlet apparatus for 16 hrs with addition of chloroform as a solvent; the solution was condensed in the incubator and preserved in refrigerator until use. Thirty (30) adult Albino male mice of 8-10 weeks old, their weight ranged between 25-35 gms. they were randomly divided into 3 equal groups, 10 mice for each group. The steroidal extract of fenugreek seeds administered at dose (100, 200) mg/kg B.Wt. to first 2 groups (G1 and G2) respectively orally for 34 days, while the third group (Control) administered daily ethyl acetate 20% orally. The results of the present study demonstrated significant decrement ($P<0.05$) in both G1 and G2 mice in all parameters measured (ICSH and SSH hormones) as compared with G3 (control group). It was concluded that the steroidal extract of fenugreek seeds has a significant antifertility effects on male reproductive mice and this effect increase with increasing in dose of this extract.

تأثير المستخلص الاستيرويدي لنبات الحلبة على هرموني SSH, ICSH في ذكور الفئران البيضاء

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الخلاصة

النباتات تستعمل لعلاج العديد من الحالات المرضية لكن لها تأثيرات جانبية وتؤدي إلى تأثيرات تثبيطية للعديد من أجهزة الجسم. الهدف من هذه الدراسة هو معرفة تأثير المستخلص الاستيرويدي لنبات الحلبة على هرموني ICSH SSH, في ذكور الفئران البيضاء. حضر المستخلص الاستيرويدي لنبات الحلبة بعد طحن النبات وتحويله إلى بودر ووضع له (2N) HCL ثم وضع في الحمام المائي وبعدها رشح المستخلص وجفف ومن ثم وضع في جهاز السكسوليت لمدة 16 ساعة ووضع له الكلوروفورم كمذيب والمستخلص الأخير وضع في الحاضنة وبعدها في الثلاجة لحين الاستعمال. أستخدم في هذه الدراسة ثلاثون 30 ذكر فار ابيض بالغ بعمر 8–10 أسابيع ومعدل الوزن 25–35 غم قسمت عشوائيا إلى ثلاث مجاميع متساوية في كل مجموعة 10. جرع المستخلص الأخير لنبات الحلبة بجرعة 200,100 ملغم/كغم من وزن الجسم لكل من المجموعة الأولى والثانية (G1,G2) على التوالي عن طريق الفم لمدة 34 يوم وتركت المجموعة الثالثة (G3) كمجموعة سيطرة وجرعة 20% Ethyl acetate عن طريق الفم وبعد انتهاء

فتره التجريع قتلت الحيوانات وتم سحب الدم منها وقياس كلا الهرمونيين ICSH,SSH. أظهرت النتائج تناقص معنوي واضح على مستوى ($P<0.05$) في كلا المجموعتين المعاملة بالمستخلص الاستيرويدي مقارنة مع مجموعة السيطرة. ونستنتج من هذه الدراسة إن المستخلص الاستيرويدي لنبات الحلبة له تأثير سلبي على التكاثر في ذكور الفئران البيضاء ويزداد هذا التأثير بزيادة الجرعة.

Introduction

Fenugreek (Trigonella foenum graecum) is an annual herb which belongs to the family Leguminosea, widely grown in India, Egypt, and Middle Eastern countries (1). Herbal medicine is the oldest form of healthcare known to humankind (2). Fenugreek (Trigonella foenum graecum L.) is an annual plant from the family of Papilionaceae–Leguminosae and is extensively cultivated in India, the Mediterranean region, North Africa and Yemen. Fenugreek seeds are well known for their pungent aromatic properties (3). The medical uses of fenugreek are antidiabetic and hypocholesterolemic in diabetic animals and both insulin-dependent and non-insulin-dependent (Type I and Type II) diabetes mellitus (4). Fenugreek is considered to be a rich source of steroidal sapogenins (5). It is also considered to be hypoglycaemic (6). Saponins have considerable potential as pharmaceutical and/or nutraceutical agents in natural and synthetic form, Saponin from a variety of sources have been shown to have hypocholesterolemic, anti-coagulant, anti-carcinogenic, hepatoprotective, hypoglycemic, immunohodulatory, neuro-protective, anti-inflammatory, anti-fungal, antiviral and antioxidant activities (7,8). According to several reports, Trigonella seed extract contains saprogenic and diosgenin, which are precursor of progesterone and have anti-gonadotropine and anti-androgenic character (9). In light of the fact that previous examination of fenugreek seeds at 30% lowered plasma levels of cholesterol (10), the precursor of steroid hormones, and that fenugreek is considered as a rich source of steroids (5). T. foenum-graecum extract exerts both antifertility and antiandro-genic activities (11). The aim of this study was to investigate the antifertility activity for oral administration steroidal extract of fenugreek seeds on albino male mice.

Material and Method

The *Trigonella foenum graecum* (fenugreek) was purchased from the local market in Ramadi. The fenugreek seeds were cleaned and grinded by electrical blender. The plant powder was stored at room temperature until extraction. The powder of fenugreek seeds were hydrolyzed with 2N HCl for 4h on water bath cooled and filtered. The material after filter was air dried for 48h and then defatted in a soxhlet apparatus for 16h with chloroform a solvent using double-thickness cellulose extraction thimbles. The extract was dried by incubator (12). Thirty male mice were used in this study, the age of these mice was ranged between 8-10 weeks, and their weight was around 25-35 grams. These animals were kept under suitable environmental conditions of 20-25 ċ in an air conditioned room and photoperiod of 12 hours daily. Thirty (30) male mice were divided into 3 groups (10 animals per each group) and handled as follows the first group was given the orally daily dose 100mg/kg B.wt of the steroidal extract of fenugreek seeds orally, the second group was given 200mg/kg B,wt of the steroidal extract of fenugreek seeds orally, the third group worked as a control group which was given daily ethyl acetate 20% orally. The experiment extends for 34 days and scarified the animals for the following parameters:

1- LH (ICSH)
2- FSH (SSH)

The hormones were measured by Automatic Gamma Counter kit after treating the samples with I^{125} (Labelled testosterone tracer), then by Gamma Counter the connection between I^{125} with hormone were measured in ng/ml unit. The data from both groups were compared using independent student's "t" test. Statistical analysis was done using the SPSS, version 12.0. (ANOOVA and LSD), "P" value <0.05 was considered to indicate statistical significance.

Results

- **ICSH hormone level:** The results of this study demonstrated the effect of steroidal extract of fenugreek seeds oil on ICSH level (Fig.1). There was a significant decrease (P<0.05) in both treated groups at dose 100mg/kg B.wt (1.66±0.19) and at dose 200mg/kg B.wt (1.48±0.28) (G1 and G2) as compared with the control group (1.88±0.30).

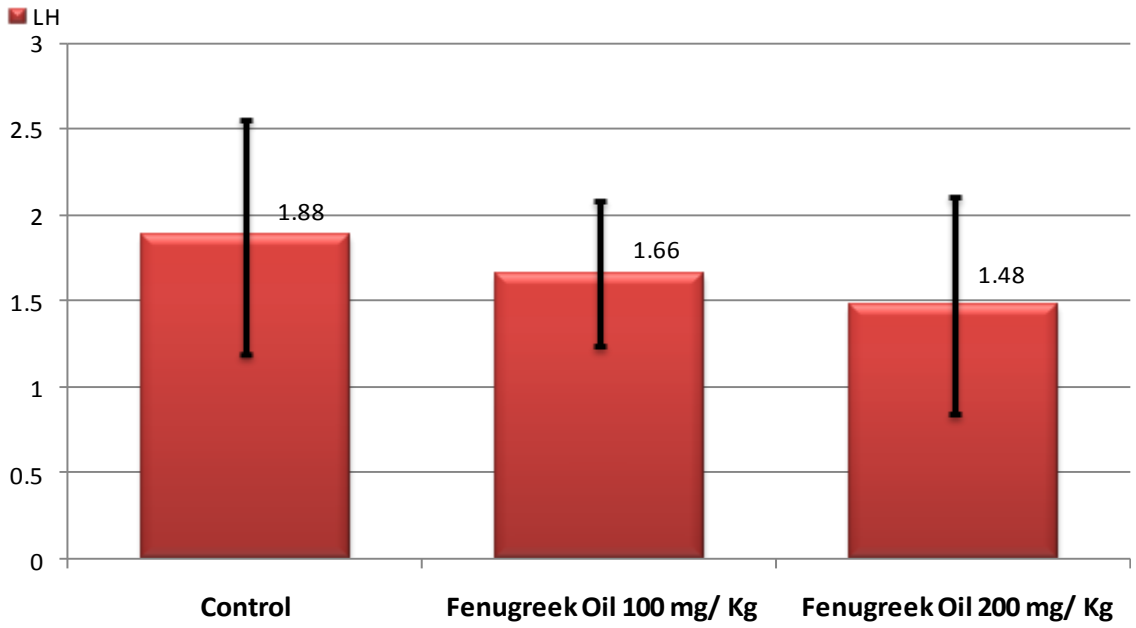

Fig. (1) ICSH hormone level
(Significant difference using t-test at 0.05 level of significance)

- **SSH hormone level:** The results of this study demonstrated the effect of steroidal extract of fenugreek seeds oil on SSH level (Fig. 2). There was a significant decrease (P<0.05) in both treated groups at dose 100mg/kg B.wt (28.60±8.42) and at dose 200mg/kg B.wt (14.16±2.45) (G1 and G2) as compared with the control group (29.00±3.41).

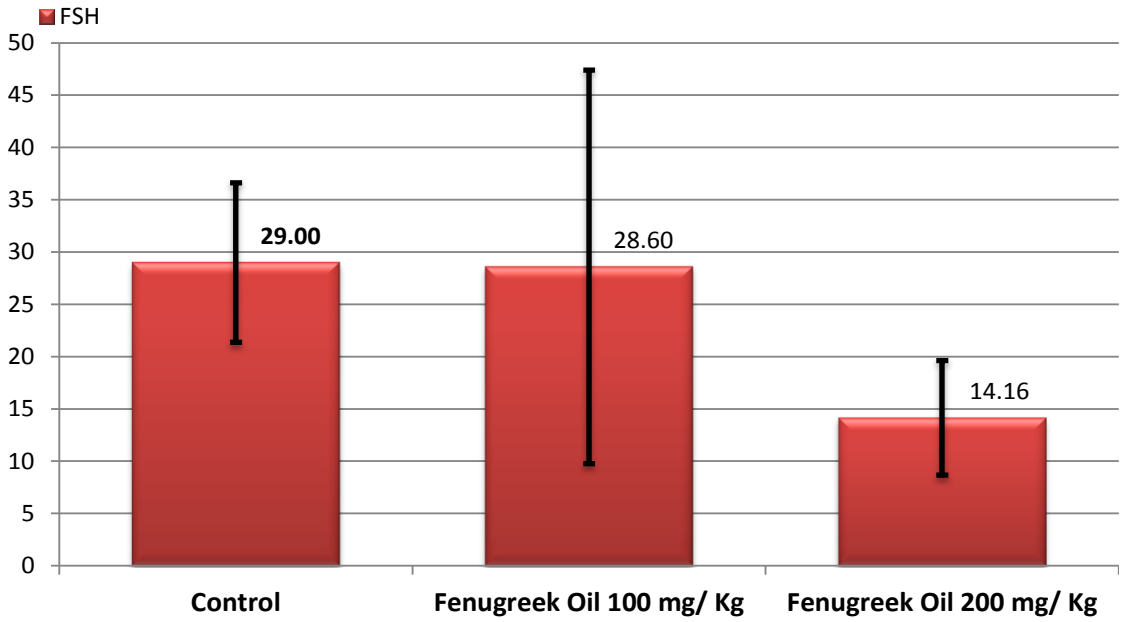

Fig. (2) SSH hormone level
(Significant difference using t-test at 0.05 level of significance)

Discussion

Hormonal level in all groups showed decrease in the ICSH and SSH levels after fenugreek administration of fenugreek seed. This might be attributed to the work on male mice treated with fenugreek that fed on fenugreek seeds at 30% to male rabbits lowered testosterone to 65.8% (13). The negative impact of fenugreek seeds on the male structural and functional integrity of testicular tissues, The secretion of testosterone was under the control of ICSH (11). Therefore, the decreased testosterone concentrations produced by the action of saponin could be explained either by inhibition of the enzymatic pathways of its synthesis in the testes or the adrenal cortex, or may be due to interfering with ICSH release, all of that leading to decrease testicular testosterone synthesis and release (14). Fenugreek has antiandrogen activities, due to beta-sitosterol, palmitic-acid and stearic-acid, and also has the ability to lower total cholesterol, LDL, VLDL cholesterol and triglycerides significantly (15,16). According to several reports, Trigonella seed extract contains saprogenic and diosgenin, which are precursor of progesterone and have anti-gonadotropine and anti-androgenic character (9). T. foenum-graecum extract exerts both antifertility and antiandro-genic activities (11). It was concluded from this study the steroidal extract of fenugreek seeds has significant antifertility effects on albino male mice and this effect increase with increasing in dose of this extract.

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