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Effect of Olea europaea Extract on Male Rats' Reproductive Parameters

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Abstract

Objective: Phytoestrogens plant compounds with biologic-estrogenic activity, structurally similar to 17-β estradiol, are first converted to heterocyclic compounds similar to estrogens in structure and then conjugated in the liver. Olive (Olea europaea), from the oleaseae family, is known as a phytoestrogen plant compound since it contains lignans and phenolic compounds. The aim of the present study was to evaluate the effect of O. europaea extract on male rats' reproductive parameters.

Materials and Methods: In this experimental study, 40 male Wistar rats with the average weight of 200-250 g and age of 8-10 weeks, divided into 5 groups. Group 1, which called control group, received no treatment. Group 2 received normal saline and Groups 3-5 received extract at a dose of 50, 100 and 200 mg/kg/day of the olive leaves extract (1 cc), respectively. Before the administration of the first gavage and 24 h after that of the last one (i.e., in the 49th day), all the rats were weighed, and blood samples were taken from their tail vein. The blood samples were then centrifuged at 1500 rpm for 20 min, then serum was separated and stored at −80 °C for the measurement of estradiol and testosterone, using immunoassay technique.

Results: The results show a significant decrease in testosterone and estradiol level among the five groups, which is dependent on the concentration of the extract; the decrease in testosterone and estradiol is positively correlated to the concentration of the extract.

Conclusion: In conclusion, olive leaves extract significantly decreased fertility parameters in the male adult rat dose-dependently.

Keywords: Estradiol, Olive Leaves Extract, Rats, Testosterone

Introduction

Phytoestrogens plant compounds with biologic-estrogenic activity, structurally similar to 17-β estradiol, are first converted to heterocyclic compounds similar to estrogens in structure and then conjugated in the liver (1). Phytoestrogens are categorized into three major classes: Isoflavones, lignans, and coumestans (2). These plants are vastly available in food sources such as soybean, flax seed, fennel and Actinidia chinensis (3). Epidemiological studies show that food sources containing phytoestrogens cause lower risk of cardiovascular diseases and also prostate and breast cancers (4).

Australian pastures developed a widespread infertility in the 1940s. A particular type of clover (Trifolium species), rich in formononetin, is included in the sheep grazing, which in the rumen during the process of fermentation will be changed to daidzein (5). Other studies claim that the phytoestrogens present in a type of summer

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grass reduced the reproduction rate of sparrows and deer in California; these studies also report that young mice fed by their mothers suffered from infertility problems because they were exposed to high amounts of phytoestrogens (6). It was also observed that soy bean caused infertility in Cincinnati's panthers, a problem solved by eliminating soy bean from the food supply (7).

Olive (Olea europaea), from the oleaseae family, is known as a phytoestrogen plant compound since it contains lignans and phenolic compounds (8). Olive contains stilbenoids, phenolic acid and flavonoids, and, due to the presence of oleuropein, has antioxidant, anti hyperlipidemic and anti-ischemic effects (9). It is also useful in curing gastrointestinal problems since it has laxative effects (10). What's more, olive is employed in treating dermatological diseases like psoriasis and atopic dermatitis (11).

In addition, the plant has antimicrobial, antivirus and anti-fungus attributes (12). It should be mentioned that olive reduces osteoporosis in Menopausal women (13). Therefore, with regard to the phytoestrogenic effects of this plant, the present study investigated the effects of olive extract on the fertility reduction of male rats.

Materials and Methods

Olive leaves was collected in summer from Rodbar jungles, Iran, and the class was specified by an expert to be O. europaea L (14). Then, the leaves supply was dried in exposure to air and away from sun beam, and after being crushed, was taken to the percolator where it was percolated by means of ethanol 70% (4 times/day, 20 cc solvent each time, for 25 days). The resultant ethanol extract was preserved in closed and dark containers in refrigerator until the time of experiment.

In this experimental study, 40 male Wistar rats with the average weight of 200-250 g and age of 8-10 weeks, divided into 5 groups. Group 1, which called control group, received no treatment. Group 2 received normal saline and Groups 3-5 received extract at a dose of 50, 100 and 200 mg/kg/day of the olive leaves extract (1 cc), respectively. They were kept at the Animal Center of the Tabriz research center at a temperature of 23 ± 2°C, a cycle of 12 h/12 h light/dark. They had access to food and water ad libitum for 49 days. The study adheres to the principles of laboratory care established by Ethics Committee of Tabriz research center.

Before the administration of the first gavage and 24 h after that of the last one (i.e., in the 49th day), all the rats were weighed, and blood samples were taken from their tail vein. The blood samples were then centrifuged at 1500 rpm for 20 min, then serum was separated and stored at −80°C for the measurement of estradiol and testosterone, using immunoassay technique. Spectra testosterone, and estradiol kits were used according to their manufacturer's instruction (Merck, Germany).

Quantitative data are presented as mean ± standard deviation. Sperm count and motility, of control and experimental groups are compared using one-way analysis of variance and Tukey test is use to find the statistical differences among their means. $P < 0.05$ is considered to be statistically significant.

Results

Oral administration of various concentrations of olive leaves extract resulted in significant decrease of male genital system hormones (Table 1).

As shown in table 1, it has been revealed that olive leaves extract caused significant decrease in serum levels of estradiol and testosterone dose dependently. It means that, extract at a dose of 200 mg/kg caused more decrease in serum levels of these hormones.

Discussion

Phytoestrogens are plant compounds with structures and functions similar to those of 17-β estradiol, which produce effects like those by estrogen. The olive, as it contains phenol compounds, is one of the natural plants rich in phytoestrogens, and belongs among the Lignans (15). The plant can highly decrease menopausal syndrome in women (16). It also decreases the occurrence of colorectal, prostate, and breast cancers (4). The findings of the present study show that olive decreases the levels of reproductive indicators such as, testosterone and estradiol in male rats.

The results also show a significant decrease in testosterone and estradiol level among the five groups, which is dependent on the concentration of the extract; the decrease in testosterone and estradiol is positively correlated to the concentration of the extract. Studies by Weber et al. (17) and Roberts et al. (18) on the effects of phytoestrogens on testosterone support these results. McGarvey et al. found that the LH level in rats decreases as a result of exposure to genistein (19). According to their study, it is possible that phytoestrogen has an inhibitory effect on the enzyme 17 β-hydroxy steroid hydrogenase human type 5; therefore, the synthesis of testosterone in adrenal cortex reduced.

Table 1. The effect of olive leaves extract on estradiol and testosterone serum levels

Parameter group	Estradiol (pg/ml)	Testosterone (ng/ml)
Control	12.03 ± 1.02	2.68 ± 0.14
Positive control	11.82 ± 0.96	2.60 ± 0.10
Extract 50 mg/kg	9.63 ± 0.67	1.89 ± 0.06
Extract 100 mg/kg	8.57 ± 0.70	1.53 ± 0.11
Extract 200 mg/kg	7.48 ± 0.86	1.07 ± 0.09

Studies by Weber et al. (17) and Glazier and Boman (20) have also shown that phytoestrogens produce no significant decrease in estradiol levels, and a study by Dehghani and Panahi about the effect of A. chinensis on male rats' spermatogenesis showed an increase in estradiol (21).

It should also be noted that A. chinensis belongs to genisteins while olive is from lignan group, which can justify the discrepancy of the results of different experiments as a result of the different types of phytoestrogen under study and the differences in the concentrations employed.

With regard to the explained results, there is this possibility that the different effects of phytoestrogens on the male productive system is due to estrogenic and anti-estrogenic effects, as phytoestrogens function through estrogen receptors, which have both agonistic and antagonistic properties. Depending on the type of phytoestrogen and the location, the effects can differ. For example, Isoflavones are very weak agonists which bind to estrogen receptors less than estradiol does (22).

When estradiol levels are low in the body and binding is therefore less competitive, isoflavones show stronger agonistic effects. On the other hand, the anti-estrogenic effects of isoflavones are co-dependent on relative concentrations of endogenous phytoestrogens and estrogens, and it is quite possible that when estrogen is high, phytoestrogens make estradiol receptors unavailable to estradiol.

Phytoestrogens produce various physiological effects in both the human body and animal models. Their effects on the male reproductive system depend on the type of the phytoestrogen, concentration and the model under study (11).

Conclusion

Olive leaves extract significantly decreased fertility parameters in the male adult rat dose-dependently. However, it is needed more study about the mechanism by which olive leaves extract create its anti-fertility effects on human being which are still unknown. Nevertheless, considering our findings in this animal model, it is recommended that the olive leaves extract maybe used in the future as a contraceptive in males.

Ethical issues

The local ethics committee approved the study.

Conflict of interests

We declare that we have no conflict of interests.

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