

## Effect of Garlic Hydro-Alcoholic Extract on Pituitary-Gonad Axis in Female Mice

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### Abstract

Garlic (*Allium sativum*) is belonging to Alleacea family. This plant has anti microbial, anti cancer properties because of its organosulfuric compounds. In this research, effect of hydro-alcoholic extract of garlic was studied on reproduction system of female mice (Balb/C).

At first, samples were divided in five groups with 10 members: one group as control, three groups received treatments, and one group received placebo. Samples were kept in same places. For synchronizing estrous cycles of rats, cloprostenol and progesterone were injected in their peritoneum with a three days interval. Hydro-alcoholic extract of garlic was prepared in three doses (50,100, and 200 mg/kg) and was injected in peritoneum for 20 days. Control group was not injected and placebo group was injected using physiological serum. After 10 injections, sample bloods were analyzed for FSH, LH, estrogen, and progesterone using RIA method. Results showed significant reduction in FSH and LH levels and significant increase in estrogen and progesterone levels. According to results of this study, hydro-alcoholic extract of garlic has a negative effect on female reproduction system in all various doses (50,100, and 200 mg/kg).

**Keywords:** Garlic; Reproductive System; Female Mice

### Introduction

Traditional medicine has a long time history. This division of medicine is a complex of knowledge, experiences, and functions according to theories, believes, indigenous experiences, and various cultures which is used in presentation, diagnosis, and treatment of physical or mental disease. Complementary, replacement, or non-official medicine phrases have been used in some countries in place of “traditional medicine” [1]. Traditional medicine includes using medicinal plants, parts of animal bodies, and minerals. Medicinal plant is a plant with definite effective matter, which is used in prevention or treatment diseases, and is known by one of international valid Pharmacopoeia [2,3]. Effective matters are the same components of medicinal plants with therapy effects. Those pharmaceutical plants which their effective matters are known must be standardized with appropriate analysis methods to have definite amount of effective matters [3]. If it is not possible to recognize effective matters, whole plant can be considered as effective matter.

Among medicinal plants, Garlic has many medicinal effects on various organs of body and many researchers have been conducted on it. It has a lot of antioxidant and anti cancer properties but there are not many researches about its effect on reproduction system, then the effect of garlic on reproduction system was the main goal of our study. Allium is a big genus with many biennial and perennial species. Many of these plants are aromatic and some are not [4]. Garlic (*Allium sativum*) is from tulip family. It is called *Saum* in Arabic, *Theriquedes pourres Allicaulitre* in French, *Knoblouch* in Spanish, and *Aglio* in italic. Garlic bulbs have: allicin, oleanolic acid, adenosine, ferulic acid, endolysin linoleic acid and phytoestrogen [5]. Allicin is the strongest compound of garlic which has the most effect on healthiness. Plus antibiotic specifics, it is an organic anti fungi. One of the most important compounds of garlic which amplifies anti oxidant effect of garlic is *allicin*, the natural compound of garlic [6].

### Material and Methods

Female mice (*Balb/C*) were prepared from animal division of

Isfahan University in range of 25-40 g. Samples were divided to five groups (with 10 rats in each group) including one control group, three treatment groups and placebo group, and were kept in separate cages. All samples were kept in natural radiation, water, and food at 25-30°C for two weeks to environment adaption. This situation was continued in injection period too. Garlic's extract was injected in peritoneum in four levels: group 1: 50 mg/kg of body weight, group 2: 100 mg/kg of body weight, group 3: 200 mg/kg of body weight, placebo group: this group received only 9% salt solution, control group: this group didn't receive any injection. All injections were done for 20 days, every other day from 8 am to 1 pm.

The estrous cycle of all female rats must be synchronized for evaluating drug effect. To this, half micro gram of cloprostenol was injected in peritoneum at first. Three days after, three microgram of progesterone was injected under the skin of all samples and then they became synchronized. One day after progesterone injection, extract injection was started. After the last injection, blood samples were prepared.

### Results

Comparing FSH hormone (mlu/ml) of blood serum using Duncan test ( $P<0.05$ ) showed significant reduction in experimental groups, in proportion to control group. Figure 1 shows these differences.

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**Received** November 19, 2012; **Accepted** December 31, 2012; **Published** January 03, 2013

**Citation:** Modaresi M, Taromsari S (2013) Effect of Garlic Hydro-Alcoholic Extract on Pituitary-Gonad Axis in Female Mice. J Veterinar Sci Technol S11: 001. doi:10.4172/2157-7579.S11-001

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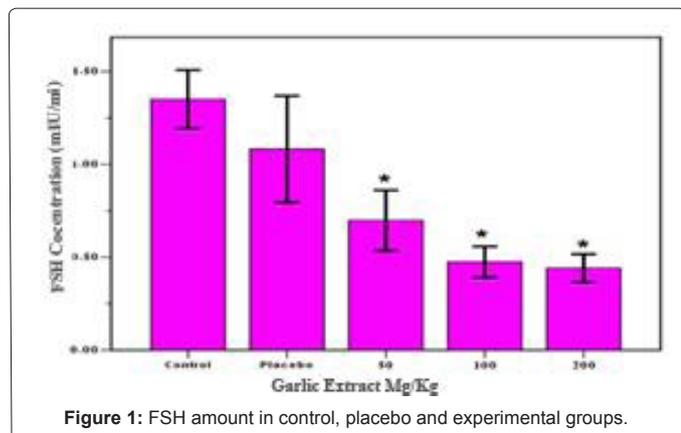


Figure 1: FSH amount in control, placebo and experimental groups.

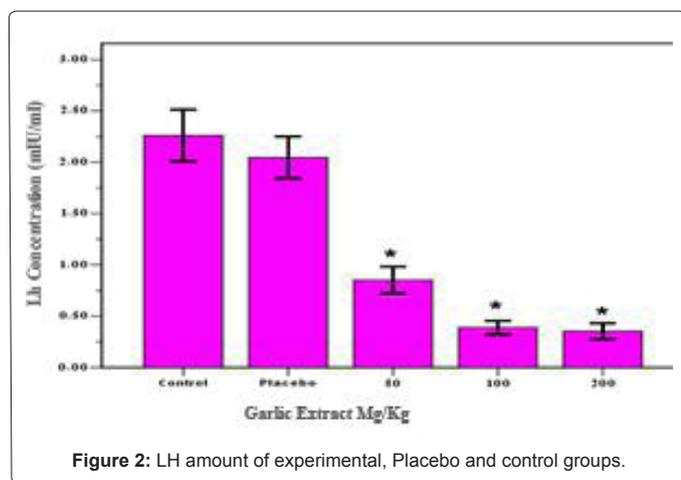


Figure 2: LH amount of experimental, Placebo and control groups.

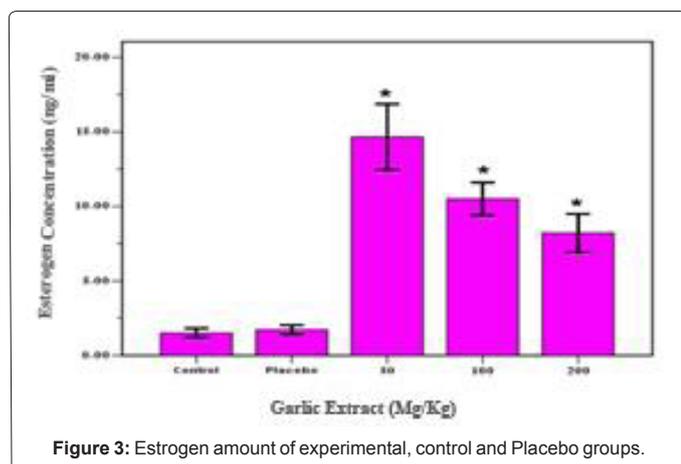


Figure 3: Estrogen amount of experimental, control and Placebo groups.

Studying the LH amount (mIU/ml) in blood serum of experimental, control rats and comparing them at 5% level using Duncan test, showed significant reduction in experimental groups, in proportion to control group. The results are shown in figure 2.

Studying the estrogen amount in blood serum of experimental, control groups and comparing them at 5% probability level, showed significant increase in experimental groups, in proportion to the control group. These results are shown in figure 3.

Studying the amount of progesterone hormone (mg/ml) in blood

serum of experimental and control groups and comparing them at 5% probability level, using Duncan test showed significant increase in group one (50 mg/kg), group two (100 mg/kg), and group three (200 mg/kg) in proportion to control group. These results are presented in figure 4.

## Discussion

Garlic extract did not destroy tissues in this study. This is in agreement with Omotoso study which testis tissue of rats was not destroyed by using garlic aquatic extract [7].

FSH reduction increases the level of IGFBP 4.5 (which is an ovarian growth factor) and protease function will be prevented, then FSH antagonists will be increased and follicles will be involved with atresia.

Then, FSH level inside follicle plays an important role in selection phenomenon or atresia. Also, GnRH stimulates production of IGFBP 4.5 in granulosa cells in follicle, and meanwhile reduces IGFBP proteases then causes follicle atresia. So, GnRH can control granulosa cells and cause pyknosis appearance. Also, some other factors are effective in apoptosis process of granulosa cells (EGF, IGF, and BGTF), then we can say that regulating follicles atresia is affected by growth factors and some other factors like FSH and steroids [8].

The amount of LH hormone was reduced significantly in all three experimental groups. LH is regulating by negative feedback mechanism and controlling secreting sexual hormones via Pituitary-gonad system. Low amounts of estrogen controls LH production highly. Also, when progesterone is extant, control effect of estrogen will be increased several times. This will reduce secreting LH from front Pituitary and LH will be reduced [9]. Results show that FSH was reduced in all three groups. Increase in estrogen secreting from dominant follicle at half follicular phase reduces FSH secreting via negative feedback. Also, high progesterone is one of the other reasons of FSH reduction which prevents FSH secreting and prevents new ovulation. Estrogen increasing was occurred more in group one (dose 50 mg/kg) which has been because of more graffian follicles in this dose. Granulosa cells of follicle are estrogen producers in ovary [9].

## Conclusion

Considering the obtained results, it seem that garlic reduced graffian follicle of every three doses and stopped folliculogenesis via affecting ovary, and increase in corpus luteum is because of previous yellow bodied. Folliculogenesis has been stopped after injecting garlic extract and then increase in corpus luteum has been stopped

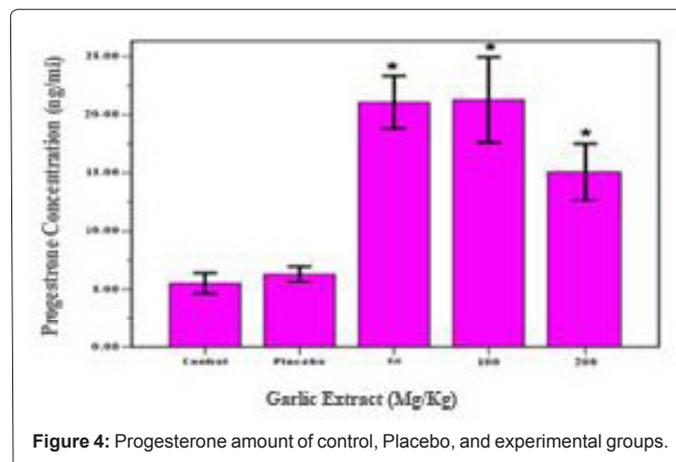


Figure 4: Progesterone amount of control, Placebo, and experimental groups.

consequently. Also, this extract causes estrogen secreting which cause itself negative feedback on pituitary-gonad system, and reduced FSH, and LH. Considering obtained results, garlic plant affects secondary sexual characteristics and increases them, but it has anti fertility effect because of folliculogenesis reduction. According to results, 50 mg/kg can be the most effective dose.

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