

The effect of short-term royal jelly supplement on testosterone levels in sedentary and healthy individuals

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Summary. This study with a placebo-controlled experimental design intends to investigate the effect of short-term Royal Jelly (RJ) on the testosterone levels in sedentary men at a dose of 1000 mg/day. For this purpose, a total of 20 adult sedentary men aged 21 to 23 were included in this study. The subjects visited the laboratory every day for 15 days between 08:00 and 10:00 to get their portion of royal jelly. The subjects were randomly divided into two groups, namely the experimental group (n = 10 individuals, 1000 mg/day of Royal Jelly) and the placebo group (n = 10 individuals, corn starch mixed with 1000 mg/day of water) and they took royal jelly in glass vials at the same time. Blood samples were taken from both groups of subjects one day before and one day after the study and analyzed to determine their testosterone levels. 2x2 mixed factor ANOVA and LSD tests were used to analyze data obtained from the experimental and the placebo group. A sharp increase in the testosterone levels of the experimental group that took RJ for a short time was found to be statistically significant ($p < 0.05$). The pre- and post-test values of the placebo group were not found to be statistically significant ($p > 0.05$). The study shows that a short-term 1000 mg/day dose of RJ supplements was effective in increasing testosterone levels in sedentary, healthy men.

Key words: Testosterone, royal jelly, supplement

Introduction

Royal jelly is a special nutrient secreted by the hypopharyngeal and mandibular glands of young worker honey bees. The queen bee and the worker bee have the same genetic structure during hatching. However, dietary differences during the larval period leads to the distinction between the queen and the worker bee. All fertilized bee eggs are fed with royal jelly during the first three days of maturation (1-3). From the third day onwards, larvae fed with a mixture of royal jelly, honey and pollen produce worker bees while larvae fed with pure royal jelly that is a homogenous substance rich in water, and include rich water, proteins, carbohydrates, lipids, vitamins and minerals (4-9), produce queen

bees. While the queen bee can produce eggs almost twice its own weight per day with exceptional productivity, worker bees, though female, cannot lay eggs (3). In the literature, the biological and pharmacological effects of royal jelly on humans, animals and culture cells have been studied. Studies that used rats and mice as test subjects to examine the antibacterial, fungal, antiviral and antiparasitic effects of royal jelly have demonstrated estrogenic and gonadotropic effects (10-12), effect on growth and development (13-15), impact on increased life expectancy (16), role in preventing hypoxia and increasing oxygen carrying capacity (17-19), role in increasing fertility in male rabbits, rats and mice (20-23), role in increasing reproductive capacity in sheep and rats (24-27), association with high sperm

quality, increased sperm concentration and motility (28), testicular protective effect (29), role in protecting the autoimmune system (30), preventing inflammation (31, 32), protecting against cancer (33), protecting the cardiovascular system (34, 35), minimizing neural damage, supporting memory (36), as well as antioxidative properties and role in mitigating osteoporosis, protecting the liver and preventing liver damage (18, 37-39). In a similar vein, studies that examined the effects of royal jelly on humans have demonstrated that it reduces fatigue, improves performance (10, 40), has a positive impact on blood parameters with regards to cancer, allergies and wound healing (41-46), causes a decrease in lipid metabolism, prevents cardiovascular occlusion, dilates the veins, regulates the blood pressure (47-50), has an antioxidant effect and protects against radiation (51-54), has positive effects on fertility in both men and women (55). However, there is limited information regarding the effect of short-term using the royal jelly, as 15-day intake, on serum testosterone levels in humans. At this scope, it is considered that the present study has an importance.

It is said that royal jelly has an estrogenic effect and a positive impact on reproduction in humans and animals. We hypothesized that short-term royal jelly intake increases testosterone levels in sedentary and healthy individuals. The purpose of this study was to investigate the effects of royal jelly supplements on the testosterone hormone.

Materials and methods

Participants

The study protocol was approved by the Ethics Committee of Gaziantep University (2017-311) and a voluntary consent form was obtained from all participants before the study. The power analysis was performed for sample size with GPower 3.1. with a priori test protocol.

A total of 20 healthy, sedentary men (Table 1) aged 21 to 23 participated voluntarily in the study. Healthy individuals without a chronic disease who do not smoke and who do not train regularly were included in the study. As per the exclusion criteria, individuals with a chronic disease, who smoke and train regularly were

not included in the study. Besides, the reason for our age limitation was for reach to individuals who were in their first adulthood after puberty.

Study Design

This is a study with a placebo-controlled experimental design. A total of 20 male subjects were randomized with the stratified randomization method and divided into two equal groups. The subjects visited the laboratory every day for 15 days between 08:00 and 10:00 to get their portion of royal jelly. The placebo group ($n = 10$) took corn starch mixed with 1000 mg/day of water in glass vials for 15 days between 08:00 and 10:00, while the experimental group ($n = 10$) took a 1000 mg/day of pure royal jelly supplement during the same hours. Royal Jelly (Civan, Bee Farm, Bursa) was prepared in 1000 mg glass vials according to cold chain criteria and stored in a refrigerator. The subjects were instructed not to engage in any physical exercise or strenuous physical activity for 15 days.

All types of nutrients taken by the subjects along with their names and the quantities consumed were recorded during one-on-one interviews for seven days. The results of these records were calculated as daily average nutritional values. Energy levels, macronutrients (carbohydrates, fat, protein), micronutrients (vitamins, minerals) and fluid intake of the subjects who were asked to maintain their dietary habits throughout the experiment were analyzed with certain nutrients ruining the dietary balance of the group removed from the program to better monitor their diets. Testosterone hormone levels were measured in 5 ml of blood taken

Table 1. Descriptive parameters of the study subjects

		Mean	SD
Experimental Group ($n=10$)	Age	21.70	1.16
	Height	177.60	6.13
	Weight	71.53	6.42
	BMI	22.69	1.81
Placebo Group ($n=10$)	Age	23.00	1.16
	Height	174.30	6.53
	Weight	70.06	8.88
	BMI	23.08	2.58

SD. standard deviation; BMI. body mass index. There is no significant difference between groups in descriptive parameters ($p>0.05$)

Table 2. Statistical analysis of testosterone levels in the experimental and the placebo group

	Experimental Group (n=10)		Placebo Group (n=10)	
	Mean	SD	Mean	SD
Pre-test	453.74	81.21	459.93	107.45
Post-test	510.25 ^a	70.50	466.03	108.09
Testosterone (ng/dL) Difference	56.51 ^b	34.59	6.10	45.53
p (between pre-post tests)	0.003		0.546	
p (between groups)			0.001	

a. significant difference between the pre-post tests ($p<0.05$); b. significant difference between the groups ($p<0.05$); SD. standard deviation

before and after the study.

Measurement of serum testosterone level

One day before and after taking royal jelly supplements, venous blood samples of 5 ml were taken from the right arms of fasting participants between 09:00 and 10:30 in the central laboratory of the Medical Faculty Hospital of Gaziantep University. The samples were placed in tubes with yellow covers. The blood samples collected were centrifuged for seven minutes at 4000 rpm in a Nüve-NF800 device to separate the serum. Serum testosterone levels were measured by electrochemiluminescence immunoassay (Hitachi Cobas 6000) (56).

Anthropometric Measurements

Weight has been measured to the nearest 0.1 kilogram using a digital weight scale. Height has been measured with a wall-mounted digital stadiometer. BMI was calculated as weight in kilograms divided by height in meters squared (57).

Statistical Analysis

SPSS 22.0 was used for statistical analysis. Data were provided in mean and standard deviation. Significance was defined as $p \leq 0.05$. 2x2 mixed factor ANOVA and LSD tests were used to analyze data obtained from the experimental and the placebo group.

Results

The effects of taking royal jelly at a dose of 1000 mg/day for a period of 15 days on testosterone levels are illustrated below in a table and diagram, for both the ex-

perimental and the placebo group. The groups consisted 20 subjects and there was no significant difference between groups in terms of descriptive parameters.

The pre-test value of the experimental group was 453.74 ng/dL and the post-test value was 510.25 ng/dL. According to the statistical analysis, the difference between the pre-and post-test values was 56.51 ng/dL. The pre-test value of the placebo group was 459.93 ng/dL and the post-test value was 466.03 ng/dL (Figure 1). According to the statistical analysis, the difference between the pre-and post-test values for the placebo group was 6.10 ng/dL (Table 2). Statistically, the increase between the pre- and post-test for the experimental group was found to be significant at $p<0.05$ level, whereas the values of the placebo group were not found to be statistically significant ($p>0.05$). Also a statistical significance was found between the groups in favor of the experimental group ($p<0.05$).

Discussion

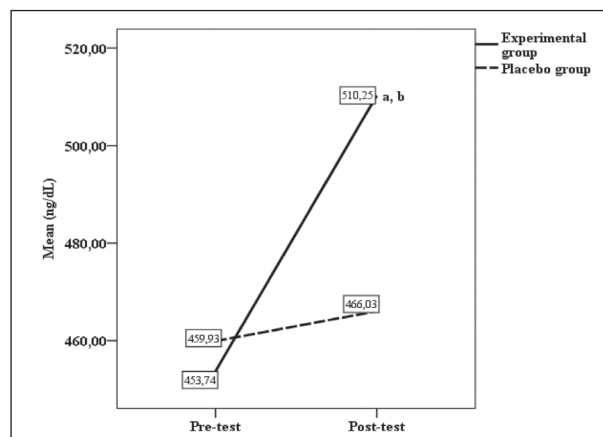


Figure 1. Variation in testosterone levels in the experimental and the placebo group. a, significant difference between the pre-post tests ($p<0.05$); b, significant difference between the groups ($p<0.05$).

The purpose of this study was to determine the effects of short-term royal jelly supplements on testosterone levels in sedentary and healthy men. The strength of the study has come up with two major findings: [1] The royal jelly supplement taken by the experimental group increased testosterone levels in favor of the post-test ($p < 0.05$), and [2] the difference between the pre-post tests was higher in the experimental group compared to the placebo group ($p < 0.05$). The results of this study show that royal jelly supplement increases testosterone levels in sedentary and healthy men.

Our study showed an increase in testosterone levels of sedentary men in the experimental group taking royal jelly supplements for 15 days. The literature shows that royal jelly plays a role in increasing sperm count in infertility due to asthenozoospermia (58). Furthermore, royal jelly has been shown to have a positive effect on testosterone levels, live sperm count, ejaculation volume, sperm motility and fructose rate in seminal plasma in male rats (22).

Elnagar used royal jelly to prevent infertility in male rabbits due to temperature related stress. The study concluded that sperm motility, ejaculation volume, sperm concentration, seminal plasma's fructose rate and testosterone levels increased in groups of rabbits given royal jelly at different doses (200, 400, 800 mg/kg) compared to the control group (21). Another study found similarly that giving 100 mg/kg of royal jelly to diabetic rats over a period of six weeks increased testicular weight, sperm count and motility, sperm viability and testosterone levels (59). For women, taking royal jelly regularly has been shown to increase fertility. It was pointed out that this was due to the fact that royal jelly has an estrogenic effect. This estrogenic effect is mainly due to fatty acids in royal jelly. Royal jelly interacts with estrogen receptors via these fatty acids, leading to altered gene expression and cell proliferation (11). It is also argued that royal jelly accelerates oocyte maturation, and increases the fertilization, cleavage and blastocyst rate (60). All studies have confirmed the positive effects of royal jelly on the male reproductive system through experimental studies in animals and humans and through biochemical and histological findings.

In conclusion, intake of short-term royal jelly,

which is used in many areas, is effective in increasing testosterone levels in sedentary healthy men. Royal jelly supports the development of bee larval cells and maintains the ovulation capacity, and this increases its possibility of having an important role in fertility. It is assumed that the possible physiological mechanism responsible for this effect works through polyunsaturated fatty acids and phospholipids contained in royal jelly protecting the sperm cell membrane from oxidative damage, interacting with testosterone receptors, causing cell proliferation and increasing testosterone levels.

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There is no conflict of interest between the authors.

Limitations

The present study constructed on short-term intake of royal jelly. In order to obtain detailed data on this short-term intake, the limitation of our study is that three or more repeated measurements of the testosterone level were not taken. It could be suggested that daily or every two days measurements of serum testosterone level in order to sensitive determination for further research.

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