Progress in Nutrition 2020; Vol. 22, N. 1: 281-287 DOI: 10.23751/pn.v22i1.8747

Original article

Effect of royal jelly supplementation on aerobic power output and anaerobic power output

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Summary. This study with a placebo-controlled experimental design intends to investigate the effect of Royal Jelly (RJ) on the aerobic and anaerobic power output of sedentary men at a dose of 1000 mg/day. For this purpose, a total of 20 adult sedentary men aged 21 to 23 years 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 Royal Jelly) and the placebo (n = 10 individuals, corn starch mixed with 1000 mg/day water) group and they took royal jelly in glass bottles at the same hour during these visits. In both groups, aerobic and anaerobic power measurements were performed in the laboratory one day before and after the 15-day period. 2×2 mixed factor ANOVA and LSD tests were used to analyze data obtained from the experimental and the placebo group. No significant difference was found in the analysis performed between the pre- and post tests for anaerobic power outputs of the placebo group ($p > 0.05$). Concerning the anaerobic power output of the experimental group, there was a significant difference in the fatigue index value in favor of the post-test ($p < 0.05$). The intergroup analysis of the difference between pre- and post-test in the same parameter showed a significant difference in favor of the experimental group ($p < 0.05$). In the pre-test and post-test analysis of the aerobic power outputs of the placebo group, no significant difference was found ($p > 0.05$). A significant difference was found in the pre- and post-test analysis of the experimental group in favor of the post-test in terms of aerobic power output ($p < 0.05$). The intergroup analysis of the difference between pre- and post-test showed a significant difference in favor of the experimental group in terms of aerobic power outputs ($p< 0.05$). Consequently, it can be argued that royal jelly supplementation taken daily for 15 days at 1000 mg has a positive effect on the aerobic capacity of sedentary men.

Key words: Supplement, performance, power

Introduction

Royal jelly, a sticky, jelly-like matter, is excreted from the glands (hypopharyngeal and mandibular) of bees. This secreted substance is used to feed queen bees and larvae. The color of this nutrient ranges from light cream to dark yellow. Royal jelly is acidic and has a pungent taste (1). Royal jelly impacts the organism in many ways. Studies have shown positive effects on blood parameters (2).

Studies on the physiological effects of royal jelly in rats and mice have shown positive results. Many studies show it helps with suppressing humoral immunity in rats, stimulating proliferation of immune component cells and antibody production in mice (3), raising hematopoietic stem cell production (4) and lowering cholesterol levels (5). It has also been reported that it has a positive effect on skeletal muscle weight, muscle strength and recovery of injured muscle groups (6).

Studies in humans have revealed the positive effects of royal jelly. One study reported that taking 6 g of royal jelly per day for a period of 4 weeks lowered LDL cholesterol levels (7*)*. In addition, royal jelly has been acknowledged for its anti-aging properties (8, 9). It has been shown that it has a positive influence on the endurance of athletes (10), chronic fatigue and vagosympathetic balance of swimmers, regulation of heart rhythm and energy deficiency (11), regulation of the fat-muscle ratio and the body mass index in footballers (12). Another study has shown that royal jelly increases physical performance and dilates blood vessels (13). In a study on elderly people, royal jelly has been shown to have a positive effect on muscle strength and physical performance (6-minute walking test) (14).

Taking into account the contribution of royal jelly to animals and humans, the hypothesis is posited that controlled use of royal jelly supplements can have positive effects on aerobic and anaerobic power. The present study is important since the studies on the effect of royal jelly on athletic performance in general, including aerobic and anaerobic power, are not comprehensive enough in scope. The aim of this study is to investigate the effects of royal jelly supplements on aerobic and anaerobic power outputs in sedentary male subjects, taking into account the positive effects of royal jelly on human health in line with the abovementioned information and studies.

Materials and Methods

Experimental 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 fifteen 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 water in glass vials for 15 days between 08:00 and 10:00 on an empty stomach, while the experimental group (n = 10) took a 1000 mg/day pure Royal Jelly supplement during the same hours. They did not drink anything before royal jelly intake. 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. The groups carried out royal jelly intake only in the morning. At the other time of the day, they carried on a normal diet. Subjects did not receive any energy drink or coffe before measurement. Royal jelly was kept in the freezer and given to the subjects.

All types of nutrients taken by the subjects along with their names and the quantities consumed were recorded during one-on-one interview for 7 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. One day before and one day after the study, the subjects underwent measurements in a lab environment to determine the aerobic and anaerobic powers of the experimental group and the placebo group with everyone participation in the groups.

Subjects

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, patients with chronic disease, who smoke and train regularly were not included in the study. 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.

Determination of aerobic power outputs in an exercise with increased load

The aerobic power outputs of the subjects were measured directly with an ergoline bicycle (Sana Bike 450F, Ergosana GMBH, Bitz, Germany) and an ergospirometer (Figure 14, Ergo100 PFT Systems, Medical Electronic Construction R&D, Brussel, Belgium).

The measurement was carried out by using the "breath by breath" method on ergospirometer during

Table 1. Descriptive parameters of the study subjects

Variables	Experimental Group (n=10)		Placebo Group (n=10)	
	M	SD	M	SD
Age	21.70	1.16	23.00	1.16
Height	177.60	6.13	174.30	6.53
Weight	71.53	6.42	70.06	8,88
BMI	22.69	1.81	23.08	2.58

exercise with increased load to determine the amount of O_2-CO_2 in the expiratory air. Heart rate, O_2 saturation and blood pressure data were recorded during exercise by making subjects wear an O_2 saturation probe and an arm manometer before the test. At the beginning of the test, the bicycle pedal load was set to 50 watts and the test was continued with an increase of 25 watts per minute. During the test, the subject had a bike screen as reference and tried to pedal at 60 rpm. When the subject realized that he could no longer continue, the exercise was stopped after checking the Respiratory Exchange Rate (RER) value (15). Peak oxygen consumption (VO_{2PEAK}), relative peak oxygen consumption (rVO_{2PEAK}), workload VO_2 ratio ($\Delta VO2/\Delta WR$) minute ventilation (VE_{PEAK}), respiratory rate (RR_{PEAK}), peak carbon dioxide release (VCO_{2PEAK}) and the respiratory exchange rate (RER_{PEAK}) was measured as aerobic power output parameters.

Determination of the anaerobic power outputs with the Wingate anaerobic power test

The Wingate test protocol with a bicycle ergometer with scale (894E Peak Bike, Monark Exercise AB, Vansbro, Sweden) was used to determine the anaerobic power outputs. Subjects did warm-up exercises for ten minutes before starting the test. Weights corresponding to 7.5% of the subject's body weight were put on the scales of the bicycle. The subject was told he could start the test at any time by pressing the button controlling the scale. When the subject felt ready, he pressed the button that controlled the scales to lower the weight on the scales to put more weight on the pedals. From that moment on, he cycled with maximum effort for 30 seconds. The test was ended when the time was out (16). Anaerobic power output parameters were measured as in peak power (PP), relative peak power (rPP), average power (AP), relative average power (rAP), minimum power (MP), relative minimum power (rMP), fatigue index (FI) and time to peak (TTP).

Royal Jelly

Freshness has been attributed a great importance for RJ quality. Royal jelly can be spoiled easily if not properly stored. Immediately after harvest it should be placed in dark vessel and stored 0 - 5°C. Stored under these conditions its quality remains OK for half an year. Deterioration of royal jelly can be prevented by storing RJ in Argon after harvesting (19). After longer storage it will turn rancid. Frozen royal jelly can be lyophilised as it can be transported more easily in the dry state. If frozen, it can be stored for 2-3 years without loosing of its quality. Chauvin states that the physical properties of RJ change after 20 hours after harvest, if left at ambient temperature (29).

Statistical Analysis

SPSS 20 package program was used to analyze the data obtained from the study (SPSS Inc., Chicago, IL, USA). Data were presented as mean, standard deviation values. The Shapiro-Wilk test was used for the normality test. 2×2 mixed factor ANOVA and LSD tests were used to analyze data obtained from the groups. The level of significance was accepted as $p < 0.05$.

Results

Table 2 illustrates the analysis of participants' aerobic measurements between pre-test and post-test. As a result of the analysis, no significant difference was found between the pre- and post-test values of the placebo group ($p > 0.05$). In the analysis for the

Table 2. Analysis of participants' aerobic measurements between pre-test and post-test

M		Experimental Group (n = 10)		Placebo Group (n = 10)	
		SD	M	SD	
VO_{2PEAK} (L/min)	Pre-test	2.46	1.35	1.74	0.64
	Post-test	2.96[a]	0.64	1.61	0.51
	Difference	0.49[b]	1.13	-0.13	0.49
rVO_{2PEAK} (ml/kg/min)	Pre-test	34.85	19.77	25.17	10.77
	Post-test	41.40[a]	7.52	22.69	6.16
	Difference	6.55[b]	15.80	-2.48	7.93
$\Delta VO_2/\Delta WR$ ($mlO_2.min^{-1}.W^{-1}$)	Pre-test	11.50	6.08	10.62	4.17
	Post-test	11.30	2.88	9.78	3.49
	Difference	-0.20	5.60	-0.84	3.16
VE_{PEAK} (L/min)	Pre-test	47.19	28.30	35.29	10.71
	Post-test	81.71[a]	27.04	32.97	7.86
	Difference	34.52[b]	21.55	-2.32	8.73
RR_{PEAK} (breath/min)	Pre-test	36.40	8.98	38.30	12.55
	Post-test	42.30[a]	6.09	34.00	10.84
	Difference	5.90[b]	8.44	-4.30	10.81
VCO_{2PEAK} (L/min)	Pre-test	2.39	1.35	1.70	0.57
	Post-test	3.31[a]	0.87	1.55	0.44
	Difference	0.91[b]	1.05	-0.15	0.54
RER_{PEAK} (VCO_2/VO_2)	Pre-test	1.05	0.12	1.09	0.09
	Post-test	1.16[a]	0.09	1.04	0.07
	Difference	0.113[b]	0.14	-0.05	0.11

SD-standard deviation, VO_{2PEAK}-peak oxygen uptake, rVO_{2PEAK}-relative peak oxygen uptake, $\Delta VO2/\Delta WR$-oxygen uptake to work rate slope, VE_{PEAK}-peak minute ventilation, RR_{PEAK}-peak respiratory rate, VCO_{2PEAK}-peak carbon dioxide output, RER_{PEAK}-peak respiratory exchange ratio. a-significant difference between pre- and post-tests of group, b-significant difference between the experimental and placebo groups.

experimental group between pre-test and post-test values in aerobic measurements, there was a meaningful difference in favor of the post-test concerning the VO_{2PEAK}, rVO_{2PEAK}, VE_{PEAK}, RR_{PEAK}, VCO_{2PEAK} and RER_{PEAK} values ($p < 0.05$). Concerning the VO_{2PEAK}, rVO_{2PEAK}, VE_{PEAK}, RR_{PEAK}, VCO_{2PEAK} and RER_{PEAK} values, a significant difference was found in favor of the experimental group in the intergroup comparison of the differences between the pre-test and post-test ($p < 0.05$).

Table **3** illustrates the analysis of participants' anaerobic measurements between pre-test and post-test. As a result of the analysis, no significant difference was found between the pre- and post-test values of the placebo group concerning anaerobic measurement results ($p > 0.05$). In the analysis of the anaerobic measurements of the experimental group, a significant difference was found concerning the FI value in favor of the final test ($p < 0.05$). In the intergroup analysis of the difference between pre-test and post-test of the measurements, a significant difference was found in favor of the experimental group concerning the FI value ($p < 0.05$).

Discussion and Conclusion

This study investigated the effect of a daily dose of 1000 mg royal jelly on the aerobic and anaerobic power of sedentary men. Immediately after the preliminary measurements before starting the royal jelly supplements, the 15-day long process for royal jelly supplements was initiated. At the end of the process, the final measurements were taken to evaluate the aerobic and anaerobic performance of the individuals. Our study

Table 3. Analysis of participants' anaerobic measurements between pre-test and post-test

M		Experimental Group (n = 10)		Placebo Group (n = 10)	
		SD	M	SD	
PP (W)	Pre-test	727.97	139.67	695.99	123.56
	Post-test	730.05	132.77	728.52	125.70
	Difference	2.078	52.54	32.53	79.44
rPP (W/kg)	Pre-test	10.14	1.65	9.84	1.65
	Post-test	10.25	1.52	10.30	1.64
	Difference	0.12	0.78	0.46	1.11
AP (W)	Pre-test	507.58	77.48	471.50	69.95
	Post-test	509.29	71.56	488.96	78.39
	Difference	1.71	31.67	17.46	22.31
rAP (W/kg)	Pre-test	7.07	0.80	6.65	0.86
	Post-test	7.16	0.71	6.89	0.80
	Difference	0.09	0.40	.239	0.28
MP (W)	Pre-test	269.75	33.61	247.04	43.07
	Post-test	293.81	46.46	240.97	94.82
	Difference	24.05	50.60	-6.07	83.31
rMP (W/kg)	Pre-test	3.78	0.52	3.48	0.55
	Post-test	4.15	0.68	3.39	1.27
	Difference	0.37	0.74	-0.09	1.18
FI (%)	Pre-test	62.24	5.43	64.04	5.73
	Post-test	59.23[a]	6.39	66.85	11.63
	Difference	-3.01[b]	4.94	2.816	8.84
TTP (msec)	Pre-test	3.01	1.39	2.90	1.56
	Post-test	2.66	1.59	3.07	1.29
	Difference	-0.353	1.45	0.17	1.67

SD-standard deviation, PP-peak power, rPP-relative peak power, AP-average power, rAP-relative average power, MP-minimum power, rMP-relative minimum power, FI-fatigue index, TTP-time to peak. a-significant difference between pre- and post-tests of group, b-significant difference between the experimental and placebo groups.

came up with two major findings. Firstly, Royal Jelly was positively affect the aerobic power], and secondly anaerobic power values. In the analysis of the data of the experimental group, a statistically significant difference was found in the parameters VO_2, relative VO_2, VE, RR, VCO_2 and RER in favor of the final test in the analysis of the difference between the pre- and post-test in aerobic measurements. In the anaerobic measurements of the experimental group, a significant difference was found concerning the FI value in favor of the final test. According to the results obtained by this study, using royal jelly supplements for 15 days can have a positive effect on aerobic and anaerobic power in sedentary men.

Previous studies have shown that royal jelly has beneficial effects on animals and humans. Due to its easy applicability and the convenience it affords with regards to monitoring of the results, the positive effect of royal jelly on animals in a clinical setting has been demonstrated in almost all studies. It is assumed that the positive results from animal experiments may also apply to the human organism. Research on humans has shown that royal jelly intake has contributed positively to physical performance **(in elders),** a strong memory (13,17), overall health, insomnia, vigor, heart health and vascular ailments (18), renewal of energy spent, faster recovery from fatigue (19), physical endurance and energy efficiency **(in athletes)** (11), reducing high choles-

terol levels (7), and posture and body mass index (12). Researchers also found that royal jelly has a positive effect on the body of **footballers**; and contributes to the growth and development of the body (20, 21).

Studies have shown that royal jelly has positive effects on physical performance (14), muscle strength and the fat-muscle ratio (12). Aerobic power is adversely affected by the increase in lipid hydroperoxides in the blood (22). As an antioxidant, royal jelly can inhibit lipid peroxidation (23). A previously conducted study revealed that royal jelly supplementation stimulates lipid peroxidation and inhibits its increase. After the supplementation, a significant difference was obtained by ensuring a decrease in the amount of lipid peroxidation (10). Based on the results of this study, it can be assumed that in our study, a certain part of the increase in aerobic power may be due to this factor.

Anaerobic power is affected by a number of hormonal changes in the organism. It has been shown that individuals with high values of total testosterone and androstenedione have low fat in their bodies and a high bone density and that such individuals have high performance values and a maximum oxygen consumption capacity (24). Testosterone can directly stimulate glycogen synthesis, and a previous study has shown that circulating testosterone levels increase during short-term intensive exercise (25). Since royal jelly increases testosterone production (26, 27, 28), in our study, the effect of royal jelly on testosterone can be put forward as the physiological basis of the increase in the fatigue index value that occurs in anaerobic power.

It can thus be argued that royal jelly taken at a dose of 1000 mg for 15 days has positive effects on aerobic and anaerobic capacity in sedentary healthy male subjects. On the basis of the research results, it can be argued that the increase in the aerobic and anaerobic values of the subjects is due to the positive effects of royal jelly on physical performance, overall health and fatigue, and to the presence of highly nutritious components in royal jelly.

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