# The effect of inspiratory muscle training and royal jelly supplement on lipid metabolism in smoking addicts

Serkan Pancar<sup>1</sup>, Zarife Pancar<sup>2</sup>

<sup>1</sup> Yeniceabat Anatolian High School, Ministry of Education, Bursa, Turkey

<sup>2</sup> Sport Science Faculty, Gaziantep University, Gaziantep, Turkey

**Abstract.** *Study Objective:* This study aimed to investigate the effects of inspiratory muscle training and royal jelly supplementation on lipid metabolism in smoking addicts. *Methods:* A total of fourty males aged between 20-22 years were included in the study. Before the study, the groups who were addicted to smoking were divided into four groups as the control group (n: 10), royal jelly supplement group (n: 10), IMT group (n: 10), and royal jelly + IMT group (n: 10). Blood samples were taken from all groups for analysis of cholesterol, triglyceride, HDL, LDL levels. Groups were made to perform respiratory muscle training through an inspiratory muscle training device with 40% of their MIP values. Groups supplemented with royal jelly were provided with 1000 mg/day royal jelly supplied in glass vials. The training sessions were carried out at the same time every day for four weeks / five days. *Results:* A statistically significant difference was found in favor of the final test in royal jelly, inspiratory muscle exercise, royal jelly + IMT groups (p <0.05). *Conclusion:* It can be said that royal jelly and inspiratory muscle exercises affect blood lipids.

Keywords: Respiration, supplement, smoking, exercise

#### Introduction

The importance of supplements has increased with the spread of epidemics. One of themost important of these is royal jelly which is natural food. In the treatment of many diseases, the use of royal jelly is increasing day by day as a supplement to the medical treatment process(1). Royal jelly is a dense milk product in which young worker bees secrete from mandibular and hypopharyngeal glands and is used to feed their larvae (2). Queen bees are fed with royal jelly starting from the larvae period and royal jelly directly affects the life of the bees, allowing them to live up to five years by giving eggs as heavy as their weights each day (3). It is recommended to minimize the damage caused by chemicals taken to the liver and kidneys and to protect these organs, especially in patients who use intensive antibiotics and receive radiotherapy and chemotherapy (4). Due to these superior features, the use of royal jelly as human food and its importance for human life and health are increasing more and more (5).

The strength or weakness of respiratory muscles in comparison to other skeletal muscles due to their more specialized structures is considered as an indicator or cause of a disease. The most important of these diseases are specified as asthma, cystic fibrosis, neuromuscular diseases, and chronic obstructive pulmonary disease (COPD) (6). Respiratory muscle training has a rehabilitative effect as well. It is one of the primary methods used in pulmonary rehabilitation (7). Due to the strength-enhancing effect of respiratory muscle training on inspiratory muscle, it decreases the perception of dyspnea caused by decreased inspiratory muscle strength in COPD patients and provides an increase in exercise capacity (8). When studies conducted are examined, it can be seen that the positive effects of inspiratory muscle training have been determined on healthy individuals (9), individuals with lung disease (10), healthy athletes (11), obese individuals (12), hypertension patients (13).

When the studies are examined, there are no studies on royal jelly supplements and inspiratory muscle training in cigarette addicts. This study was thought to contribute to science in terms of its method and findings. In this study, it was thought that royal jelly supplementation together with inspiratory muscle training would affect lipid metabolism in smokers.

#### Material and Method

## Participants

A total of 40 healthy male subjects volunteered to participate in this study (Table 1). The aim of the study was explained to all subjects and written informed consent was obtained from all subjects at the familiarization session.

1st Group: The Control (natural Sedentary, n:10) Group,

2nd Group: The Royal Jelly supplement group which takes only Royal Jelly (RJ, n:10),

3rd Group: Training Group which is inspiratory muscle training (IMT, n:10),

4th Group: The Inspiratory muscle training which is supplied with royal jelly (RJ+IMT, n:10).

Before the study, the groups who were addicted to smoking were divided into four groups. A Voluntary consent form was obtained from all participants in the study. In addition, permission to do the study was obtained from the Gaziantep University Clinical Research Ethics Committee.

## Experimental design

This study is a randomized, experimental study with a control group. The subjects visited the lab four times. During their first visit, all subjects were informed about the study, and their descriptive information was recorded. First, by applying the Fagerström Test for Nicotine Dependence (14), those with advanced degrees of dependence (6-7 points) were divided into groups. During their second visit, blood samples were taken from all subjects before the study. During their third visit, the MIP and MEP values of individuals were determined and a nutrition program was given to all groups to establish a standard innutrition. On their fourth visit, subjects performed the warm-up procedure with 40% of their MIP values for respiratory muscle training. The group was given royal jelly supplement (1000 mg/day) and the groups performing IMT were invited to the laboratory at the same time (between 09:00 and 11:00) 5 days a week. No groups were allowed to do exercise and high intensity physical activity.

## Collection of Data

#### MIP and MEP measurement

Electronic respiratory pressure meter was used to calculate MIP and MEP (Pocket Spiro MPM-100, Medical Electronic Construction R&D, Brussels, Belgium) according to the 2002 guidelines of the American Thoracic Society and European Respiratory Society (15). Measurements were made using the nasal plug in a sitting position. For MIP, the individual was asked to perform maximum expiration and was requested to perform maximum inspiration against the closed respiratory tract and maintain it for 1-3 seconds. For MEP, the individual was asked to do maximum inspiration and was requested to perform maximum expiration against the closed respiratory tract and maintain it for 1-3 seconds. The measurements were repeated between the 2 best findings until there was a 5% difference, and the average was recorded in cm  $H_2O(16)$ .

## Inspiratory Muscle Training Procedure

A specific inspiratory training device (POWER®Breathe Classic, IMT Technologies Ltd., Birmingham; UK) was used for IMT. Training group subjects performed the IMT procedure at 40% of MIP (with +10% load increase each week and MIP test repeated on the first training day of every week). The IMT procedure included 30×2 dynamic inspiratory efforts (with 1 min interval) daily for 4 weeks (17). A separate inspiratory muscle training device was used for each subject.

#### Royal Jelly Supplement

Royal jelly is seen as a beneficial natural food source for human metabolism and systems due to valuable substances contained in royal jelly content produced in the hypopharyngeal and mandibular glands of worker bees for the feeding of queen bees (18). Royal jelly (Civan, Bee Farm, Bursa) was obtained in 1000 mg glass vials and was kept ready in the refrigerator. Groups supplemented with royal jelly received 1000 mg/day royal jelly in glass vials between 08.00 and 10.00 in the morning for four weeks.

## Blood Test Procedure

Venous blood samples were collected from the right arm of the participants into 5ml purple capped tubes at the central laboratory of Gaziantep University Faculty of Medicine between the hours of 09:00 and 10:00 in the morning a day before the study and the day after the study. At the end of the study, to analyze cholesterol, triglyceride, HDL, LDL levels in the blood samples collected, serum samples obtained by centrifuging for 5 minutes at 4000 rpm in Nuve trademark centrifuge device were studied in Beckman Coulter brand autoanalyser and the results were recorded.

#### Statistical Analysis

The SPSS version 22.0 (SPSS Inc., Chicago, IL) program was used for statistical analyzes. Values were represented as mean and standard deviation, and significance was set at 0.05. Kolmogorov-Smirnov test was performed to assess normality, and 2x4 mixed-factor analysis of variance and least significant difference tests were performed to analyze intra- and intergroup differences.

## Results

The analysis of the obtained data is shown in Table 1. In the findings obtained in the cholesterol levels, Royal Jelly (147.83 ± 23.04); In the IMT (160.75 ± 36.29) and RJ + IMT (147.57 ± 27.52) groups, statistically significant was found in favor of the post-test (p <0.05). At triglyceride levels, Royal Jelly (89.67 ± 25.62); A statistically significant difference was found in the IMT (105.75 ± 51.57) and RJ + IMT (109.71  $\pm$  45.72) groups in favor of the post-test (p <0.05). At high density lipoprotein (HDL) levels, Royal Jelly (48.17 ± 5.42); In the IMT (50.88 ± 11.34) and RJ + IMT (46.86 ± 8.13) groups, statistically significant was found in favor of the post-test (p <0.05). Considering the difference between the groups, a statistically significant difference was found in the cholesterol, triglyceride, HDL and LDL levels in the supplement group, exercise group, and supplement + exercise group compared to the control group (p < 0.05).

#### Discussion

This study aimed to examine the effects of inspiratory muscle exercise and royal jelly supplementation on blood lipid levels in smokers. Two major findings were identified in this study. The first finding was that royal jelly supplements and IMT affected blood lipids, resulting in changes in their levels. The second finding is that royal jelly supplements and IMT changed HDL levels, which is the healthy cholesterol.

Many studies have shown that respiratory muscle training has significant effects on respiratory muscles. It has been reported in several studies that the respiratory muscles will be stronger in a few days with respiratory muscle exercise, that the frequency of respiration decreases within three weeks, and that the performance increases as a result of the four-week respiratory muscle exercise (19-21). However, smoking causes functional disorders in the respiratory muscles by affecting the respiratory functions. The leading cause of the harmful effects of smoking on the respiratory system is the deterioration of the oxidant / antioxidant balance in favor of oxidants (22).

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		Control	Koyal Jelly		KJ+IMI Maara SD
		Wieali+5D	Wiean+SD	Wiean+SD	Wiean+5D
	Pre-test	162.00±29.74	165.67±27.76	166.63±24.88	159.29±35.16
Colest.	Post-test	163.14±21.84	147.83±23.04 <sup>A</sup>	160.75±36.29 <sup>A</sup>	147.57±27.52 <sup>A</sup>
	Difference	1.14±20.40	-17.83±17.83 <sup>в</sup>	-15.88±31.47 <sup>в</sup>	-11.71±19.37 <sup>в</sup>
Trigl.	Pre-test	138.14±52.02	131.83±67.87	143.00±52.14	161.43±46.17
	Post-test	135.29±24.23	89.67±25.62 <sup>A</sup>	105.75±51.57 <sup>A</sup>	109.71±45.72 <sup>A</sup>
	Difference	-2.86±43.26	-42.17±57.31 <sup>B</sup>	-37.25±28.67 <sup>B</sup>	-51.71±41.86 <sup>в</sup>
HDL	Pre-test	45.43±10.63	46.50±4.81	48.50±12.21	44.86±6.31
	Post-test	45.71±6.58	48.17±5.42 <sup>A</sup>	50.88±11.34 <sup>A</sup>	46.86±8.13 <sup>A</sup>
	Difference	0.29±6.55	1.67±4.63 <sup>B</sup>	2.38±14.20 <sup>B</sup>	2.00±6.03 <sup>B</sup>
LDL	Pre-test	89.14±30.48	112.83±22.98	130.25±37.47	101.57±25.57
	Post-test	87.71±24.05	82.67±22.98 <sup>A</sup>	94.50±27.25 <sup>A</sup>	70.43±22.16 <sup>A</sup>
	Difference	-1.43±21.27	-30.17±23.84 <sup>B</sup>	-35.75±20.44 <sup>B</sup>	-31.14±21.61 <sup>в</sup>

Table 1. Statistical analysis of the values of the groups

**S-CG:** smoker control group, **S-RJG:** smoker royal jelly group, **S-IMTG:** smoker inspiratory muscle training group, **S-IMT+RJG:** smoker inspiratory muscle training with royal jelly group, **A:** significant difference between pre- and post-tests, **B:** a significant difference from CG.

Royal jelly is used effectively against mental and physical fatigue and wrinkles and acne on the skin. Royal jelly lowers blood cholesterol, total lipid, phospholipid, triglyceride, β-lipoprotein levels, has blood pressure-lowering, and vasodilating activity, has hypoglycemic and immunological effects due to its containing insulin-like peptides (23, 24). In addition to these, it has therapeutic, hormonal regulatory, sexual functions regulating effects in skin and hair diseases, and has cell repairing and rejuvenating effects (25, 26). In our study, it was observed that the levels of cholesterol derived lipids decreased and the levels of positive healthy cholesterol increased. Royal jelly has been used by people for many purposes including cancer as well as its aphrodisiac effect from past to present. It is known that physical activity and physical fitness increase lipoprotein metabolism and reduce the risk of coronary artery disease. HDL, rich in cholesterol but poor in triglycerides, carries cholesterol from the periphery to the liver, the liver converts this cholesterol into bile directly as cholesterol or into bile salts (27). Therefore, it is considered as an effective factor in protection from atherosclerosis. Factors such as obesity, smoking, diabetes, renal failure decrease HDL level,

while exercise increases HDL level. It is necessary for the normal structure and function of cells as well as for the repair of the cell membrane (28). It has been observed that the royal jelly supplement used increases the height and muscle component of athletes and causes a decrease in the fat component compared to pre-exercise (29). It has also been reported that royal jelly improves performance (30), is used as a fatigue reliever (31), and meets the energy needs of athletes (23).

Harmful substances such as carbon monoxide, nitrogen oxide, and hydrogen cyanide, nicotine, and tar in cigarette smoke increase the production of free oxygen radicals such as singlet oxygen, superoxide, hydrogen peroxide, and hydroxyl (32). Active or passive inhalation of cigarette smoke, exercise, stress, and increased body temperature increase free oxygen radicals in the body, and these increase free oxygen radicals in the body, and these increased radicals increase the body's need for vitamins (33). Cigarette smoke increases lipid peroxidation in the respiratory epithelium (34), increased free radicals resulting from cigarette smoking cause decreased lung volumes and capacities in the long run (35), respiratory and circulatory system diseases (36) and more importantly, lung cancer (37). It is known that respiratory exercises have positive effects on the lung system (38). The strengthening of the lung parameters depends on the performance of the respiratory muscles (39).

As a result, it can be said that royal jelly supplement and respiratory muscle exercises applied for four weeks positively affect lipid metabolism.

## **Conflicts of Interest**

The authors declare that there is no conflict of interest in this manuscript.

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Correspondence: Serkan PANCAR

Yeniceabat Anatolian High School, Ministry of Education, Bursa, Turkey

E-mail: sdpancar@gmail.com