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The Effect of Aerobic Interval Training on Preptin, Lipid Profile and CRP Levels in Overweight Men

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ABSTRACT

Background and Aim: Preptin is one of the hormones regulating energy consumption. Therefore, in this study the effect of aerobic interval training on serum preptin, lipid profile, and CRP levels was investigated in overweight men.

Methods: 24 volunteer overweight men who were able to participate in regular physical activities were chosen and were randomly divided into experimental (n=12) and control (n=12) groups. For 8 weeks, the experimental group exercised according to the training protocol 3 times per week. Changes in serum preptin, lipid profile, and CRP levels were analyzed before and after the training period using the covariance test at $P < 0.05$.

Results: Intergroup comparison showed that preptin levels had no significant difference (P=0.067); however, CRP levels showed a significant decrease (P=0.048). Intragroup comparisons showed a significant decrease in preptin levels in the experimental group (P=0.042).

Conclusion: It seems that aerobic interval training can play a major role in weight control by reducing serum preptin and CRP levels and lowering some of the harmful indicators in lipid profile.

KEYWORDS

Aerobic Interval Exercises, Preptin, Lipid Profile, Overweight.

Introduction

Advances in technology, changes in lifestyle, poor diets, and reduced physical activity have led to obesity and related diseases in the society. Therefore, it is necessary to investigate weight loss methods and the metabolic disorders caused by weight gain [1]. Energy balance in the body is regulated by a complex system, which is itself affected by internal and external factors [2]. Although many factors contribute to obesity, the imbalance between energy intake and energy consumption is the most important factor [3]. Nutritionists and sports physiologists use exercise and diet to reduce body fat. These two approaches have the greatest impact when used in combination [4].

Preptin is a new hormone that regulates energy consumption [5]. It is a peptide with 34 amino acids and is secreted by pancreatic beta cells along with insulin and amylin. It is involved in glucose homeostasis [6]. Preptin and amylin are peptide hormones that are associated with insulin resistance [7]. Although the main synthesis of preptin is in pancreatic beta cells, it is also produced in several tissues, including the salivary glands, breast tissue, and kidneys. Together with other appetizing peptides, preptin plays an important role in the development of obesity and energy balance regulation by affecting the centers of hunger and satiety in the parenchymal nuclei and arch nuclei [8].

Preliminary information about preptin was obtained from experiments on animals. Some studies have shown that preptin regulates insulin secretion in response to glucose; in rats, intravenous preptin injection reduces blood glucose and insulin secretion during glucose loading [9]. Other researchers have shown that preptin affects protein kinase C and phospholipidase C by activating the insulin-like growth factor 2 (IGF-2R) and stimulating insulin secretion in a calcium-dependent manner in conditions of increased glucose concentration [10]. In the first clinical study of humans, the results showed that levels of preptin in diabetic patients were higher than in normal individuals. Although levels of preptin have a positive relationship with levels of triglycerides, total cholesterol, and insulin resistance, they have no relation with body mass index (BMI) and insulin. Also, the results of high-protein plasma levels were higher in women [11]. However, the results of other studies reported a positive association between serum preptin and BMI levels [8].

Adipose tissue secretes a number of biologically active proteins called adipokines, which play a role in the interplay between energy and inflammation. There is significant evidences that the obesity is associated with the increased plasma levels of inflammatory cytokines in plasma, to the extent that obesity has been described as an inflammatory condition [12]. Adipose tissue appears to be an important source of inflammation in obese people and adipokines play a potential role in this issue. One of the most sensitive cytokines in systemic inflammation, the levels of which are higher in obese people than in normal people, is C-reactive protein (CRP) [13]. This inflammatory index has been introduced as a strong predictor of risk of cardiovascular disease. Serum levels of CRP are also positively associated with overweight, obesity, diabetes, and metabolic syndrome [14].

Overweight and obesity are often associated with lipid metabolism disorders. Also, increased serum levels of fatty acids play a key role in increasing the risk of cardiovascular disease and insulin resistance [15]. Lipid profile includes total cholesterol (TC), triglycerides (TG), high density lipoprotein cholesterol (HDL-C), low density lipoprotein-cholesterol (LDL-C) and very low density lipoprotein-cholesterol (VLDL-C). Lipid profile has a direct and significant correlation with body fat mass, except for the high density lipoprotein-cholesterol. Compared to lean mass, the correlation between fat mass and serum lipids and body mass index is stronger in obese people. There are few studies on the relationship between preptin concentration and lipid profile. However, the results of some studies shows that aerobic exercises and a restricted diet reduce preptin levels and that this decrease had a significant relationship with body composition. Hence, preptin may play a special role in weight control programs [6].

Exercise is an effective treatment for weight loss and obesity and its related risk factors. Intense interval training can be a good alternative to other training methods, including traditional aerobic exercise. The physiological and functional changes caused by intense interval training are similar to or even greater than other methods [16].

Recent evidence suggests that high-intensity activities may alter appetite by altering appetite-regulating hormones and can be considered as causes of the anorexia resulting from exercise. It has also been shown that high-intensity interval training can significantly reduce fat mass due to the involvement of several muscles during training [17].

However, current knowledge of preptin and its association with lipid profile and CRP is limited to biological systems. Due to the higher impact of high-intensity interval training on various health-related factors including body composition, lipid profile, blood pressure, various metabolic variables, and insulin resistance [18] compared to continuous and even periodic exercise with low intensity, we focused on high-intensity interval training in the present study to investigate its potential effects on serum levels of preptin, lipid profile, and CRP in overweight men.

Materials and Methods

The present study is a quasi-experimental study, with a pre-test and post-test design and a control group. The statistical population of this study included non-athletic students aged 18 to 27 years old who were purposefully selected based on the inclusion criteria. Objectives of the program and conditions of the study were explained to the candidates and if they agreed, they would complete a consent form. Finally, the candidates visited a specialist to obtain their research license. Of the 50 participants who volunteered, 28 were eligible to participate in regular training programs; hence, they were chosen and after homogenization based on weight and body mass index, were randomly assigned to experimental (n=14) and control (n=14) groups. Sample size was calculated based on Fleiss's formula taking into account a power of 0.8, alpha of 0.05, and average changes of 5. It should be noted that during the research period, two individuals from each experimental and control groups withdrew their cooperation, causing the numbers in each group to drop to 12.

The inclusion criteria were: being obese with a body mass index (BMI) greater than 25 kg/m^2, not being an athlete, being healthy, no history of drug use affecting lipid profile, no history of smoking, and no history of sleep disorders, cardiovascular disease, or liver, kidney or mental illness. The participants were not given any specific medication or diets during the treatment stage and there was no significant difference in diet or energy intake between them. These criteria were assessed through the personal information questionnaire of PAR-Q, The health and physical activity questionnaire, and the medical history questionnaire. All coordinations in terms of medical ethics and supervision were done in coordination with the specialist physician and with the patients' consent.

The Training Protocol

One week before the start of the training protocol, the participants came into the laboratory to learn how to implement the protocol. After dividing the participants into experimental and control groups, height, weight (using a stadiometer and a scale, respectively), and body mass index (weight in kg dividing by height squared in m) were

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measured. Peak oxygen consumption (VO2peak) was measured using the Stover-Davis test on a Monark Bike Ergometer (model E839 made in Sweden). Measurements were done pre-test and post-test. The Stover-Davis test was also used to determine the maximum power of the participants [20].

For 8 weeks, the experimental group exercised according to the training protocol 3 times per week. The interval training program included cycling on the Monark Bike Ergometer three times per week (one day between sessions) for 8 weeks. Each session included warm-up, main workout, and cooling down. Cycling on the Monark Bike Ergometer during the warm-up stage, the participants reached their maximum leg power for 5 minutes with an intensity of 30-40%. Workload increased during the main practice from the first week to the eighth week; so that it increased from 6 alternations in the first week to 12 alternations in the eighth week. The intensity of activity was considered as a percentage of the maximum power of the individual. In the first week, the participant pedaled with 65% of his maximum power in the activity stage and with 30-40% of his maximum power in the resting stage. The power exerted in the activity stage reached 80% of maximum power in the eighth week. In addition, the duration of the activity stage increased from 30 seconds in the first week to 60 seconds in the eighth week. At the end of each session, each participant performed cooling for approximately 5 minutes at a maximum of 30-40% of their power. In order to apply the workload at the desired intensity, at the end of the fourth week maximum power of the leg was re-measured with the Stover-Davis test. Based on the results of this test, the intensity of work in the activity stage was adjusted for the fifth to eight weeks [21].

Table 1. Training protocol

Week	frequency	Exercise/rest (Second)	Intensity Training (Maximum power %)	Rpm
First	6	30/180	65	45-55
Second	7	30/180	70	45-55
Third	8	40/180	70	45-55
Fourth	9	40/180	75	45-55
Fifth	10	50/180	75	45-55
Sixth	11	50/180	80	45-55
Seventh	12	60/180	80	45-55
Eighth	12	60/180	80	45-55

Biochemical Analysis

In the laboratory, blood samples were taken 48 hours before the start of the protocol and 48 hours after the last training session. 10-ml blood samples were taken from both groups by a specialist in accordance with standard precautions. Samples were poured into special serum tubes and frozen at -20 °C after centrifugation and separation of serum, and were later used to measure the study variables. The enzyme-linked immunosorbent assay method (ELISA) and a preptin kit with a sensitivity of 5 ng/l (Zell Bio Company, Germany) were used to measure the concentration of preptin. The level of CRP was measured using the hs-CRP very sensitive ELISA method and a kit made by the Pars Azmoun Company with a sensitivity of 0.1 mg/l. Lipid profile indices were measured using a kit made by the Pars Azmoun Company.

Statistical Analysis

The Shapiro-Wilk test was used to determine whether or not the data distribution was normal, and the Levon test was used to investigate the equivalence of variance. After assuring that the data were normal, the statistical test of covariance (ANCOVA) and t-pair were used at $P<0.05$ level. Data analysis was performed via SPSS software version 23.

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Results

Physical characteristics and body composition measurements of participants at baseline and post-intervention are included in Table 2. particpants in the experimental and the control groups were of a similar age and height. After 8 weeks of aerobic interval training, a significant decrease in the weight index (P=0.001) and BMI (P=0.001), and a significant increase in peak oxygen consumption (P=0.007) were observed.

Table 2. General characteristics of the subjects

Variables	Group	Pre-test Mean and SD	Post-test Mean and SD	In-group t P	Intergroup F P
Weight (kg)	experimental	87.2±13.4	83.5±11.2	3.63 *0.004	14.4 ¥0.001
	control	89.1±12.5	88.5±12.8	1.61 0.14	
BMI (kg/m^2)	experimental	26.2±1.62	25.1±1.17	3.81 *0.003	19.8 ¥0.001
	control	26.9±1.38	26.7±1.32	1.66 0.12	
VO_2max (ml.kg/min)	experimental	33.4±8.8	35.6±9.6	3.76 *0.003	8.97 ¥0.007
	control	31.9±8.8	32.1±9.6	1.15 0.28	

*In-group Statistical significance; ¥ intergroup Statistical significance

Statistical analysis of the data showed that there was no significant difference in rest preptin levels ($F_{1,21}$=3.72, P=0.067) after 8 weeks of aerobic interval training (Figure 1). The intragroup changes showed that there was a significant difference between the mean before and after the training period in the experimental group (P=0.042). The results of CRP data also showed that there was a significant reduction after the exercise intervention ($F_{1,21}$=4.4, P=0.048) (Figure 2). Examination of intragroup changes of the CRP index showed that there was no significant difference in any of the groups (P> 0.05).

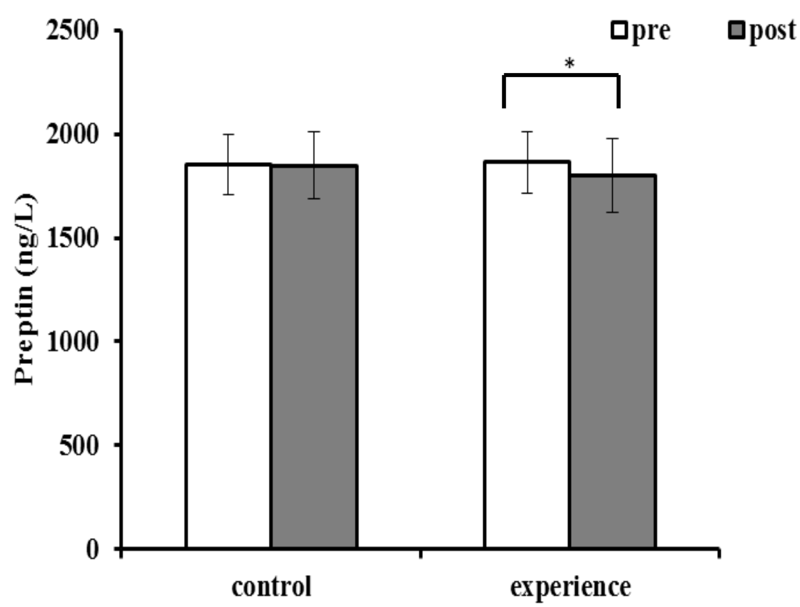

Figure 1. Mean (±Standard error) of Preptin before and after training in groups. *In-group Statistical significance

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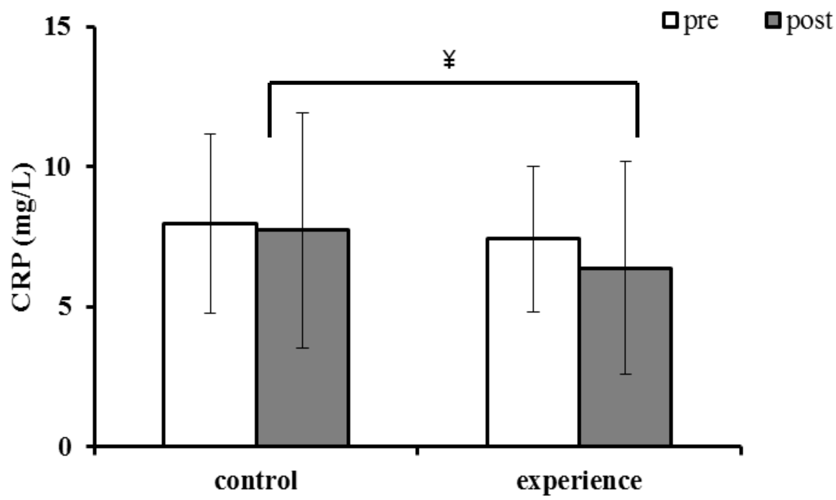

Figure 2. Mean (±Standard error) of CRP before and after training in groups. ¥ intergroup Statistical significance

In terms of TC and HDL indices, there was no significant difference between changes in the two groups after the exercise intervention TC ($F_{1,21}$=3.19, P=0.088), HDL ($F_{1,21}$=3.36, P=0.038). However, there was a significant difference in the TG ($F_{1,21}$=4.87, P=0.039) and LDL ($F_{1,21}$=4.41, P=0.048) indices between the two groups. The intragroup changes observed in the experimental group showed that there was a significant difference between the mean before and after the training period in terms of the TC (P=0.037), TG (P=0.031), HDL (P=0.004) and LDL (P=0.015) indices (Table 3).

Table 3. Investigation of inter- and intra-group changes in variables in two groups

Variables	Group	Pre-test Mean and SD	Post-test Mean and SD	In-group t P	Intergroup F P
Preptin (ng/L)	experimental	1863±161	1798±177	2.3 *0.042	3.72 0.067
	control	1852±142	1846±150	0.7 0.49	
CRP (mg/L)	experimental	7.43±4.2	6.38±3.8	2.13 0.057	4.4 ¥0.048
	control	7.95±3.2	7.73±2.6	0.84 0.42	
TC (mg/dl)	experimental	167±29.1	161.3±26.3	2.37 *0.037	3.19 0.088
	control	179.1±36.5	177.3±29.8	0.49 0.63	
TG (mg/dl)	experimental	138.3±18.9	130.5±13.2	2.48 *0.031	4.87 ¥0.039
	control	137.5±13.8	137±10.8	0.178 0.86	
HDL (mg/dl)	experimental	37.8±7.6	43.2±9.1	3.63 *0.004	3.36 0.081
	control	39.9±8.3	40.5±9.8	0.3 0.77	
LDL (mg/dl)	experimental	76.7±18.2	69.3±19.6	2.86 *0.015	4.41 ¥0.048
	control	76±17.6	77.1±18.5	0.36 0.73	

*In-group Statistical significance; ¥ intergroup Statistical significance

Discussion and Conclusion

Overweight and obesity are pervasive metabolic complications that have affected both developed and developing countries [21]. Physical activity plays a very important role in the prevention and control of overweight and obesity, and high-intensity aerobic interval training is one of the best exercise regimens for obese people.

The results of the present study showed that 8 weeks of aerobic interval training did not lead to a significant changes in the preptin, TC, and HDL indices in participants, while the concentrations of CRP, TG, and LDL experienced significant changes. Examination of the data obtained from measurement of preptin levels showed that intergroup changes of preptin were not significant despite the reduction. The intragroup data showed a significant reduction in preptin levels in the experimental group when comparing pre- and post-training levels. Analyzing these results, it can be said that changes in the experimental group could possibly reflect the changes resulting from weight loss and a decrease in BMI, since previous studies have reported a positive relationship between preptin, body mass index, and obesity.

There have been limited studies on the effect of physical activity on preptin. In the study by Ramezan Khani et al. (2015), in line with the results of the present study, a decrease in preptin was reported after 16 weeks of exercise [6]. Nazarali et al. (2019) examined the effect of high-intensity circular exercises on preptin and the insulin resistance index and concluded that six weeks of intense circular exercises was effective on serum levels of preptin in women [2]. Based on this, high-intensity exercises improve metabolism in skeletal muscles and increase anaerobic capacity, muscle glycogen content, and glucose transporter type 4 (GLUT4). Part of the adaptation of skeletal muscles is due to the intensity of the exercises. Because the intensity of physical activity is the main factor in increasing PGC1-α[1], the main enzyme of mitochondrial biogenesis. On the other hand, hormones produced by the adipose tissue play a vital role in energy regulation and consumption and metabolism of fats and carbohydrates through the production and secretion of adipokines. Preptin, as one of these peptides, appears to play a compensatory role in the process of blood glucose homeostasis along with insulin; therefore, as the capacity and sensitivity of muscle cells to insulin increase due to the high-intensity of exercises, this compensatory need decreases and preptin production and secretion decline [22]. Studies by Ozkan et al. (2013) and Yang et al. (2009) demonstrated the positive and significant relationship of serum levels of preptin with BMI and the insulin resistance index [8,11]. The present study is consistent with these studies.

The results of the present study showed that CRP level decreased significantly after the exercise intervention. Previous studies have shown that CRP level is higher in people with diabetes than in healthy people [23]. Some studies have reported a decrease in CRP levels after aerobic exercises [14,24]. Two main mechanisms can explain the decrease in the level of this inflammatory index. Many studies report that weight loss due to exercise is a major factor in decreasing CRP levels, and other physiological effects that may follow exercise may not play a role [25]. As the results of this study show, weight loss in the experimental group was significant after the exercise intervention and weight loss was probably the reason for decreased CRP in the participants. Other studies suggest that decreased CRP concentration is achieved only by high physical fitness, the physiological effects of exercise, and the anti-inflammatory properties of exercise [6]. In this study, peak oxygen consumption also increased significantly, which seems to hint that both mechanisms had significantly reduced CRP concentration in the experimental group.

The most common lipid disorders in overweight and obese people are increased TG and decreased HDL. These parameters are risk factors for cardiovascular diseases. An increase in TG is associated with an increase in LDL, which is known as an atherogenic factor. There is credible evidence that high-intensity physical activities have significant positive effects on plasma levels of lipids. The intergroup results of the present study showed that the exercise intervention had significantly reduced TG and LDL; however, exercise did not make a significant difference in TC and HDL levels. At the same time, the intragroup results showed that TC, TG, and LDL levels decreased significantly and HDL levels increased significantly in the experimental group. There is no clear mechanism for the effect of exercise on lipoproteins. It seems that regular aerobic physical activities with relatively high intensity can significantly improve lipoprotein indices [27]. Studies have shown that the liver lipase enzyme is reduced and inhibited after Regular exercises; therefore, the production of LDL triglycerides is reduced. Exercise duration seems to impact triglyceride reduction [28]. Researchers believe that HDL and LDL are hardly affected by exercise. The

1. Peroxisome Proliferator-Activated Receptor Gamma Coactivator 1-Alpha

mechanism of HDL changes following complex training. Enzymes such as lipoprotein lipase (LPL) play an important role in HDL concentration. This enzyme is the most important factor in changing HDL concentration through the hydrolysis of plasma triglycerides [29].

In general, the results of this study showed that aerobic interval training had a positive effect on preptin levels, CRP, and some lipid profile indicators. The findings showed that interval training had an important role in regulating weight in overweight men. However, it is not possible to comment conclusively on the matter due to the limited number of studies on this topic, and thus more studies are needed.

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