

Effects of eight weeks of aerobic activity with omega-3s on the ghrelin and insulin hormones and body weight in men

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Abstract

Introduction: The normal amount of the hormones ghrelin and insulin play an important role in energy balance, regulation of weight, and prevention of type 2 diabetes. The aim of this study was to investigate the effect of 8 weeks of aerobic activity on hormones ghrelin and insulin and weight in men.

Materials & Methods: In this semi-experimental study, 80 young men with an average age of 24.25 ± 1.73 years, weight 74.23 were divided into four groups, Exercise – supplementation (ES), exercise (E), supplementation (S) and control groups (C). The subjects of the ES and E groups participated in an aerobic exercise program with an intensity of 65-75% of maximum oxygen consumption (V_{O_2} max) for 3 sessions per week and for 8 weeks. Subjects in the ES and S group was given 21 omega-3 capsules (one gram each capsule) each week to take 3 times a day after each main meal for

8 weeks. At baseline and at the end of 8 weeks, the levels of the hormones ghrelin and insulin were measured by blood samples by ELISA method and also weight was measure.

Results: The results of the analysis of variance test showed that after 8 weeks, the total amount of ghrelin and insulin respectively, in the ES groups ($p = 0.001$, $p = 0.001$), E ($p = 0.001$, $p = 0.001$) and S ($p = 0.045$, $p = 0.016$) increased and decreased significantly. There was no change in the levels of ghrelin and insulin in the control group ($p = 0.621$). Also weight in the ES group ($p = 0.005$), E ($p = 0.032$) and S ($p = 0.034$) decreased significantly. No change was observed in C group.

Conclusion: Based on the results, it was found that physical exercise alone and physical exercise along with omega-3 supplementation cause significant changes in the levels of ghrelin and insulin hormones and weight in young men. These changes may help control and prevent diabetes and its complications.

Keywords: ghrelin, insulin, omega-3, physical exercise, young men

1. Introduction

In today's society, overweight, obesity and type 2 diabetes are a health problem facing people and health officials. These problems are usually caused by metabolic disorders and energy imbalances. Energy balance is regulated by a complex system that includes central and environmental factors. Ghrelin is a factor that appears to play an important role in regulating food intake and body weight (1). Studies have shown that approximately 75 to 80 percent of this peptide hormone is derived from 28 amino acids from the stomach, and the rest is often secreted by the small intestine and some other tissues such as the pancreas (2,3). Therefore, most of the ghrelin that enters the blood through the venous system is synthesized by the gastrointestinal tract and it plays an important role in absorbing food and increasing fat mass (4). Researchers have found that the expression of the ghrelin gene in the stomach increases during fasting and decreases during satiety. In fact, the plasma levels of ghrelin decrease under positive energy balance conditions and increase under negative energy balance conditions (5,6).

Ghrelin also has positive cardiovascular effects (7). In addition, ghrelin plasma levels are negatively associated with obesity (8).

Blood ghrelin is effective in anorexia, fuel consumption, weight and body composition, all of which help to regulate the energy balance. Therefore, one of the trends in research on the effects of exercise on ghrelin is due to the effect of exercise on energy balance, because the stimulation of appetite by ghrelin is affected by exercise and the energy balance changes equally (9).

In addition, insulin is a peptide hormone with a composition of 51 amino acids. The hormone is secreted into the blood from the islets of Langerhans, located in the inner part of the pancreas. Effective insulin on liver cells reduces blood sugar by taking sugar from the blood and storing it as glycogen, and increases energy by accumulating glycogen in muscle cells as a source of fuel. It also stops the use of fat as a fuel source by affecting adipose tissue. In the absence or lack of insulin in the blood, the body uses fat as a source of fuel. Insulin acts as the body's control center for metabolism (10). During increased exercise, the concentration of insulin in the blood first decreases, and subsequent changes depend on the carbohydrate consumed and dehydration (11), although the effect of ghrelin on insulin secretion and changes in blood glucose concentration in humans is not yet fully understood. However, clinical studies continue to support the role of this hormone in regulating glucose metabolism and energy balance (12). Some studies also suggest that ghrelin secreted by epithelial pancreatic cells, on the one hand, and ghrelin receptors on beta cells, on the other, regulate insulin secretion from these cells (13). In some studies, injections of ghrelin have led to inhibition and decreased Insulin secretion from pancreatic beta cells (14). These studies suggest that ghrelin has an inhibitory effect on insulin secretion from pancreatic beta cells (15).

Omega-3 supplements contain essential fatty acids, the benefits of which have been confirmed in several studies, for example, the beneficial effects of omega-3s on heart and vascular function have been accepted (16). However, there have been conflicting reports about the effects of omega-3s on weight loss. Studies in mice have shown that omega-3s reduce fat mass, reduce body weight, and limit hypertrophy of adipose tissue (17).

Considering that obesity, overweight and type 2 diabetes are the health problems of today's societies, and the hormones ghrelin and insulin and weight control are effective in reducing these problems, because the effect of physical exercise on the secretion of these hormones is unclear we have done this study with the aim of affecting 8 weeks of physical exercise with omega-3 supplementation on weight changes, serum levels of ghrelin and insulin hormones in men.

2. Materials & Methods

The present study is a quasi-experimental type of clinical trial of single-blind. The research plan included pre-test and post-test with 4 groups. The statistical population was young men from Shiraz city of Fars province of Iran that 80 of them voluntarily and purposefully selected as a sample and participated in the study based on the information obtained from the health questionnaire and completing the consent form. Subjects were asked to avoid any physical exercise for one week before the program started. The subjects were then divided into 4 groups ES, E, S and C based on their body weight. In this study, variables of weight, insulin and ghrelin were evaluated. Not all participants had a history of physical activity in the past year.

First, the height was measured by a meter that can be mounted on the wall made in china with an accuracy of 0.5 cm and the weight was measured using a digital Sega scale made in Germany with an accuracy of 0.1 kg. The statistical method of this research consists of descriptive and inferential methods. Descriptive statistics were used to draw graphs, determine the mean and the standard deviation. Kolmogorov-Smirnov test was used for statistical analysis and normal data determination. One-way analysis of variance analysis (ANOVA) was used to compare the mean variables before and after training and Pearson correlation coefficient test was used to investigate the relationship between variables. The significance level of the tests was considered to be $P = 0.05$. All data were analyzed using Spss Version 20.

Biochemical measurements

In order to determine the serum levels of the hormones ghrelin and insulin at the beginning of the study and at the end of the eighth week,

blood samples were taken from the participants with similar conditions after 8-12 hours of fasting and before breakfast at the laboratory. The temperature was 20 degrees Celsius. 5 ml of blood was taken from a vein in each person. Blood samples were centrifuged in a laboratory to determine the levels of the hormones ghrelin and insulin, and after isolation, the blood serum was kept at -70. C. ghrelin and insulin serum levels were analyzed by Raybiotech's ghrelin kit and the Mombide Insulin Kit, both made in the United States.

Exercise program

Participants in the ES and E groups performed three sessions of aerobic exercise each week for 60 minutes each session for 8 weeks as follow.

*: The first two week with an intensity of 55 to 60% VO_{2max}

*: The third and fourth week with an intensity of 60-65 percent VO_{2max}

*: Fifth to eighth week with an intensity of 65-75% VO_{2max}

Drug Dosage

The subjects of ES and S groups was given 21 omega-3 capsules (one gram each capsule) each week to take 3 times a day after each main meal (manufactured by Formulated Sciences of the United States) (19).

3. Results

The data in Table 1 shows that the subjects of the four groups were similar in terms of physical and physiological characteristics in the baseline conditions and did not have significant differences with each other.

Table 1. Anthropometric characteristics and maximum heart rate (MHR) of subjects

Groups	Age/ year	Height/ cm	Weight/ kg	MHR/min
ES	24.22	170.32	74.31	152.55
E	23.95	172.61	74.15	148.23
S	24.40	173.90	74.58	147.00
C	24.46	171.66	73.90	149.32
Mean	24.25	172.12	74.23	149.28
Standard deviation	1.80	4.28	5.06	15.14

The results of correlated t showed that serum levels of ghrelin in the ES group ($p = 0.001$) E group were ($p = 0.000$) and S ($p = 0.016$) in the post- test compared to the pre-test have increased significantly. In the C group, there was no significant difference in serum levels of ghrelin in the post-test compared to the pre-test ($p = 0.741$) (Table 2).

Table 2. Serum ghrelin and insulin concentrations in pre and post test

Statistic Variables	Groups	Mean		Standard Deviation		P
		Pre-test	Post-test	Pre-test	Post-test	
Ghrelin	ES	14.09	19.94	8.44	10.72	0.001*
	E	14.25	19.85	6.68	7.46	0.001*
	S	13.46	17.66	4.92	5.66	0.016*
	C	14.52	14.95	4.46	4	0.741
Insulin	ES	14.62	12.60	7.23	6.69	0.001*
	E	14.49	13.32	7.25	7.24	0.001*
	S	15.91	14.14	6.42	7.39	0.045*
	C	13.76	14.56	7.47	7.09	0.107

* Significant differences ($P < 0.05$)

Also, in the post-test phase, compared to baseline, a significant decrease in serum insulin levels was observed in the ES group ($p = 0.001$), E alone group ($p = 0.001$) and S group ($p = 0.045$). In C group insulin increased but was not significant ($p = 0.107$). In order to compare the changes in ghrelin and insulin in four groups' variable one-way analysis of variance was used.

The results of this test for Ghrelin are as follows:

- *: There is no difference between groups ES and E in pre and posttest ($p = 0.902$).
- *: There is no difference between groups C and S before the test, but there is a significant difference in posttest ($p = 0.015$).
- *: There is no difference between groups C and ES in pretest, but there is a significant difference in posttest (0.001).
- *: There is no difference between groups C and E in pretest, but there is a significant difference in posttest (0.001).
- *: There is no difference between groups ES and S in pretest, but there is a significant difference after the test for ES (0.047)

*: There is no difference between groups S and E in pre and posttest (0.052) (Table 3).

Table 3. Comparison of ghrelin levels between groups

Comparison of ghrelin between groups	pre- test sig	Post- test-sig
between ES and E	0.0531	0.912
between S and C	0.383	0.015*
between ES and C	0.546	0.001*
between E and C	0.893	0.001*
between ES and S	0.811	0.047*
between E and S	0.302	0.059

* Significant differences ($P < 0.05$)

The results of this test for insulin are as follows:

*: There is no difference between groups ES and E in pre and post - test (0.756).

*: There is no difference between groups C and S before the test, but there is a significant difference in post - test (0.006).

*: There is no difference between groups C and ES in pre - test, but there is a significant difference in post - test (0.003).

*: There is no difference between groups C and E in pre - test, but there is a significant difference in post - test (0.001).

*: There is no difference between groups ES and S in pre - test, but there is a significant difference in post - test for ES (0.657).

*: There is no difference between groups S and E in pre and post - test (0.713) (Table4).

Table 4. Comparison of insulin levels between groups

Comparison of insulin between groups	Pre-test sig	Post- test-sig
between ES and E	0.842	0.776
between S and C	0.963	0.006*
insulin comparison between ES and C	0.884	0.003*
insulin comparison between E and C	0.841	0.001*
insulin comparison between ES and S	0.893	0.657*
insulin comparison E and S	0.764	0.723

* Significant differences ($P < 0.05$)

The findings indicate a significant reduction in the weight of ES, E and S groups, but no change was seen in C group (Table 5).

Table 5: Comparison of subjects' weight in pre and post- test

Groups	Pre-Test	Post -Test	P
ES	74.31	72.41	0.005*
E	74.15	72.31	0.032**
S	74.58	73.66	0.034**
C	73.90	74.12	0.621

4. Discussion

In the present study, the effect of physical exercise along with omega-3 supplementation on serum levels of insulin and ghrelin hormones was investigated. The results showed that serum levels of ghrelin and insulin significantly increased and decreased at the end of the eighth week compared to baseline in the ES group, E group and S group. The results showed weight loss in the three groups mentioned above. Studies have shown that long-term exercise leading to weight loss significantly increases plasma ghrelin levels. On the other hand, studies show that plasma ghrelin levels can be regulated by hormones such as insulin and metabolites such as glucose. In fact, it has been observed that lowering insulin in the fasting state increases the secretion of ghrelin, and immediately after eating, the secreted insulin stop the secretion of ghrelin (6,18). Broglio et al. (2003) found that glucose and insulin reduced and inhibited ghrelin, therefore, reducing messages due to long-term training and starvation removes their inhibitory effect and provides the environment to increase the level of ghrelin. There was a significant inverse relationship between insulin and plasma ghrelin in the study (19).

Some studies found that subjects with a normal weight who lost more than 3 kg through a combination of physical activity and dietary intervention experienced a significant increase in plasma ghrelin levels, and in subjects who did not lose weight, the level of ghrelin remained constant. Therefore, ghrelin appears to be very sensitive to changes in body weight, and an increase in ghrelin is a compensatory behavior in response to weight loss. In other words, increasing ghrelin may act as a

compensatory mechanism for restoring body weight to a regulated point (20). In general, most studies have suggested that long-term exercise increases plasma ghrelin levels if it is associated with weight loss (21).

Insulin appears to inhibit the secretion of ghrelin at high concentrations in humans, but the physiological relationship of this finding is unclear. Insulin may be required to control serum ghrelin levels after a meal, which has been suggested by some researchers, but not all. Also, in the presence of diseases associated with weight loss and malnutrition such as anorexia nervosa (22) and some cancers (23), plasma ghrelin levels increase up to three times that of normal levels. Confirmation of the present findings was intravenous injection of ghrelin into a group of healthy young men, which led to an increase in the significant level of fasting glucose levels following insulin levels 15 to 30 minutes after ghrelin injection, indicating the inhibitory effect of ghrelin on insulin secretion (24).

It was reported the depletion of energy reserves as one of the mechanisms involved in the increase in ghrelin. In the present study, although the depletion of energy reserves has not been studied, it seems that part of the increase in ghrelin is justified by this mechanism (25).

Based on the findings, it can be concluded that a decrease in insulin in subjects can increase the expression of ghrelin and increase the serum level of ghrelin. Insulin, as an anabolic hormone, plays an important role in the entry of nutrients into cells, so that the transfer of glucose and fatty acids into muscle cells is not possible without it. On the other hand, our findings show that there is a significant inverse relationship between serum levels of insulin and also the function of beta cells. Based on this finding, it can be concluded that increasing ghrelin reduces the function of pancreatic beta cells and leads to a decrease in insulin secretion from these cells. Therefore, ghrelin indirectly increases blood glucose levels by disrupting the function of beta cells. In this regard, a study showed that ghrelin reduces the function of beta cells (26).

The present study is inconsistent with studies that have reported a significant reduction in the amount of ghrelin after exercise. Kim et al. (2008) in a 12-week study of aerobic and resistance training with an intensity of 55 to 75% of maximum heart rate, did not see a change in

the amount of fasting acylated ghrelin. The researchers initially observed an increase in total ghrelin with weight loss, but did not change in the last weeks when weight loss was stabilized, and attributed the lack of change in acylated ghrelin to stabilizing weight loss (27). Some researchers observed no change in acylated ghrelin fasting time after 8 weeks of aerobic exercise with an intensity of 60 to 85% of the maximum heart rate. In their view, the duration of the exercise program and the amount of weight loss were not sufficient for a significant change in ghrelin levels (28,29).

Differences in the type and intensity of exercise and negative energy balance during exercise, as well as differences in the age and sex of the subjects compared to current study can be considered as possible reasons for non- direction. Regardless of the limitations of the present study, such as low sample size, lack of diet control, limitation in controlling the physical exercise of the subjects outside the exercise program, lack of stress control, anxiety and excitement. The results of the present study indicate that aerobic physical exercise causes energy balance in the body due to increased metabolism and omega 3 due to fat fuel and in response to lack of energy, ghrelin is secreted to stimulate the need to receive food, provide lost energy sources, and restore energy balance. Insulin, on the other hand, is reduced by the effect of epinephrine on the pancreas to maintain blood glucose levels, so it is less likely to store fat, therefore omega-3 can make exercise more effective. Although, according to the findings, the present study of aerobic exercise combined with omega-3 fatty acid supplementation for 8 weeks can affect serum insulin and ghrelin levels as well as the weight of young men. However, more studies with more samples, diet control, and longer training are needed to confirm or reject the results of the present study.

5. Conclusion

Based on the above theoretical principles, it can be said that exercise (due to increased metabolism) and omega-3 (due to fat metabolism) probably causes a negative energy balance in the body, and in response to energy deficiency, ghrelin is secreted to stimulate food intake behavior, provide lost energy sources, and restore energy balance. On the other hand, insulin secretion is reduced by the effect of epinephrine on

the pancreas to maintain blood glucose levels, so the body is less likely to store fat. As a result, omega-3 can increase and improve the effects of exercise.

Conflict of interests: The authors have no conflicts of interest to declare.

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