The effect of intermittent training on ghrelin and leptin in overweight female students

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Abstract

Increasing prevalence of obesity and its related diseases are the proper strategies in weight loss and it needed the recognition of the effective factors in weight gaining. Recently, the researchers have discovered some hormones, including ghrelin and leptin witched active the regulating of the energy balance and finally in regulating the body weight. The present study is to determine the effect of intermittent training on Ghrelin and leptin in overweight female students. Twenty overweight female students subject (BMI ≥ 25) were randomly assigned to two groups (intermittent training and control). The experimental training programs were performed three days a week for 12 weeks at a definite intensity and distance. Before and after 12 weeks intervention, Ghrelin, Leptin, weight and body composition, Vo2max was measured for all subjects. For statistical data analysis, the independent t-test of α=0.05 was applied. The result showed that intermittent training caused a significant increase in Ghrelin and Vo2max, significant decrease in Leptin, weight and body composition of the experimental group in comparison to control group. Apparently, intermittent training not only reduces the fat mass but also impacts the secretion of leptin. However, intermittent training increases ghrelin, followed by reduce in the weight loss.

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Introduction

Obesity is now one of the problems originating numerous diseases such as hypertension, atherosclerosis, type II diabetes, certain types of cancer and respiratory and gastrointestinal disorders (Kopelman, 2000), due to its importance in health, of body weight control lead the researchers to identify interesting hormonal signals associated with the body’s homeostasis and metabolic diseases (Tritos Na et al., 1999). Researchers have recently discovered hormones that regulate energy homeostasis and are involved in the regulation of body weight. Some of these hormones are ghrelin, adiponectin, leptin, obestatin, and resistin (Kojima et al., 2005).

Ghrelin is a peptide of 28 amino acids that is largely isolated from the stomach of humans and animals (Kojima et al., 1999) and is involved in the regulation of food intake (Kojima et al., 2005), energy homeostasis and body weight regulation (Broom et al., 2007; Fujimiya et al., 2006; Tschop et al., 2000). Adipocytokine leptin is secreted by adipose tissue and is effective on energy homeostasis and in controlling body weight by influencing hypothalamus and decreasing appetite and increasing sympathetic nerve activity and lipolysis (Rahmani Nia et al., 2009).

On the one hand, exercise improves health of obese individuals is associated with weight loss and ghrelin and leptin hormones are also related with obesity (Gil-Campos et al., 2006; Imai et al., 2009). Thus, the effect of exercise on these hormones is important in order to better understand the impact of physical activity on improving the health of people. Some studies examined the response of ghrelin to a single session of exercise but the results are contradictory (Cummings et al., 2002; Dall et al., 2002; Ghanbari-Niaki et al., 2006., Hansen et al., 2002., Schmidt et al., 2004). Also the results of previous studies on the effects of exercise training (short or long term) on the response of plasma ghrelin levels in humans and rats are limited and inconsistent (Foster – Schubert et al., 2005; Kraemer et al., 2004; leidy et al., 2004). For example, Leady et al studied plasma ghrelin levels during a period of weight loss with diet and exercise. In this study subjects went through an exercise program with 70 to 80 percent heart rate 5 sessions a week and the results showed that ghrelin concentration decreased significantly in the exercise-induced weight loss group (Kraemer et al., 2004). In contrast Foster - Schubert et al studied the effect of 12 weeks of interval aerobic training on 173 overweight women. Upon completion of the protocol, ghrelin in patients who lost over than 3 kg of weight increased by 18%. (Foster – Schubert et al., 2005).

On the other hand, several studies have examined the changes in leptin in response to exercise, and have reported different results. H. Su et al observed that a month of aerobic exercise in sedentary women reduced body weight and was associated with a significant leptin reduction (Hsu et al., 2007). The results of the study by T. Sao et al indicate a significant reduction in leptin after endurance running exercise at two intensities of 65% and 85% Vo2max and at no difference between the two intensities (Tsao et al., 2009). In studies with durations of less than 12 weeks, however, exercise without weight loss program has no significant impact on leptin concentration whereas in studies with duration of over 12 weeks the results are contradictory (Kraemer et al., 2002). Therefore, considering the conflicting results of these studies, further research in this area is needed to identify the influencing mechanisms of intermittent training on ghrelin and leptin.

Materials and methods

Subjects

First of all call notices were posted in Azad University Shahre Qods Campus in which the researcher invited to identify overweight and obese individuals who were willing to run exercise for weight adjustment and improvement of their physiological conditions. In the next stage the candidates were invited for the purpose of the Initial assessments and from among them, at least 30 individuals with BMI ≥ 25 whose being overweight was not associated with thyroid under-activity and did not have a history of exercise or caloric restriction diet were selected. The subjects
were then divided randomly into two groups; intermittent training and control. The study was approved by the Ethics Committees of Islamic Azad University, Iran. Written consent was obtained from each subject after the experimental procedures and possible risks and benefits were clearly explained.

**Anthropometric measurements**

All anthropometric measurements were made by the same trained general physician and under the supervision of the same pediatrician following standard protocols. Anthropometric measurements of participants were performed while they stood in light clothing without shoes. Height was measured to the nearest millimeter, body weight was measured in duplicate in the morning following a 12-h fast. Body fat percentage was determined using body composition monitor. Body mass index was calculated as weight (kg) divided by squared height (m).

<p>| Table 1. Intermittent training programs. |
|---|---|---|---|---|---|---|---|---|---|---|---|---|</p>
<table>
<thead>
<tr>
<th>Week</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>10</th>
<th>11</th>
<th>12</th>
</tr>
</thead>
<tbody>
<tr>
<td>Target heartbeat (percentage)</td>
<td>70-75%</td>
<td>70-75%</td>
<td>70-75%</td>
<td>75-80%</td>
<td>75-80%</td>
<td>75-80%</td>
<td>80-85%</td>
<td>80-85%</td>
<td>80-85%</td>
<td>80-85%</td>
<td>80-85%</td>
<td></td>
</tr>
<tr>
<td>Distance (meter)</td>
<td>8×200</td>
<td>8×200</td>
<td>9×200</td>
<td>9×200</td>
<td>12×200</td>
<td>200</td>
<td>200</td>
<td>200</td>
<td>200</td>
<td>200</td>
<td>200</td>
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</tbody>
</table>

<p>| Table 2. Pre- and post-test physical, physiological and biochemical variables and t test in the two groups. |
|---|---|---|---|---|---|---|---|---|---|</p>
<table>
<thead>
<tr>
<th>Group</th>
<th>Intermittent</th>
<th>Control</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (year)</td>
<td>22.2 ± 1.68</td>
<td>22.77 ± 1.63</td>
<td>-</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>159 ± 1.88</td>
<td>158.80 ± 3.99</td>
<td>-</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>75.21 ± 2.86</td>
<td>72.92 ± 2.44</td>
<td>0.02</td>
</tr>
<tr>
<td>Body mass index(kg/m^2)</td>
<td>29.8 ± 1.31</td>
<td>28.89 ± 1.22</td>
<td>0.03</td>
</tr>
<tr>
<td>Fat percentage (%)</td>
<td>30.92 ± 1.48</td>
<td>28.06 ± 0.84</td>
<td>0.006</td>
</tr>
<tr>
<td>Vo2max (ml/kg/min)</td>
<td>23.64 ± 1.24</td>
<td>29.83 ± 1.28</td>
<td>0.000</td>
</tr>
<tr>
<td>Ghrelin (pg/ml)</td>
<td>4.42 ± 0.21</td>
<td>6.48 ± 0.96</td>
<td>0.000</td>
</tr>
<tr>
<td>Leptin (ng/ml)</td>
<td>11.70 ± 0.27</td>
<td>9.31 ± 0.45</td>
<td>0.000</td>
</tr>
</tbody>
</table>

Data are expressed as mean and standard deviation

**Blood measurement and Exercise protocol**

After physical examination, all subjects in the experimental and control groups were asked to attend Hematology Lab following 12 hours of overnight fasting, between the hours of 8 to 9 am for blood sampling in order to measuring plasma ghrelin and serum leptin. The subjects were advised to avoid any heavy physical activity 48 hours before the blood sampling. Then the subjects of exercise group were completed an Intermittent training program (12 weeks/3 times per week). A session of training program in intermittent group included a ten-minute warm-up with and stretching exercises. The subjects then continued with running a distance of 1600 to 3200 meters with the intensity of 80 to 95% of their maximum heart rate reserve with the work to rest ratio of one to three (Table 1). They cooled off for five minutes. All anthropometrical and biochemical measurements were repeated 48 after last exercise session in two groups.

**Statistical analysis**

All values are represented as mean ± SD. As to the inferential statistics, first the Kolmogorov–Smirnov test was used for normal distribution, Leven test was used for data homogeneity. Then independent student t test was used for testing significance between groups. All the statistical operations were performed by spss software version 15.0. A p-value of less than 0.05 was considered to be statistically significant.
Results
The descriptive profile of the groups in variables of age, height, weight, body mass index, body fat percentage, ghrelin and leptin as well as the independent t-test are presented in the table 2. After 12 weeks of Intermittent training ghrelin level (p= 0.000) (Table 2 and Diagram 1) showed a significant increase. Leptin level (p= 0.000) (Table 2 and Diagram 2) showed a significant decrease. Also the difference of measurements of variables of the two groups including Body weight (p= 0.02), Body mass index (p= 0.03), Body fat percentage (p= 0.006), Maximum oxygen consumption (p= 0.000) was significant (P ≤ 0.05) (Table 2).

Discussion
In this study the effect of 12 weeks of intermittent training on plasma acyl ghrelin and serum leptin in overweight female college students was studied. The results of this study reveal that after 12 weeks of intermittent training plasma ghrelin levels increase and serum leptin decrease significantly. A review of the researches on physical activity on plasma ghrelin levels suggests that the results are contradictory; while some are consistent with those of this study others contradict them.

Most previous studies have evaluated ghrelin response in a single session of exercise (Ghanbari-Niaki A., 2006; Schmidt et al., 2004). Also little information is available on the effects of training (short or long term) on the response of plasma ghrelin levels (Foster – Schubert et al., 2005; leidy et al., 2004). Leady et al studied plasma ghrelin levels during a weight-loss diet and exercise program. The results indicate a significant increase in plasma ghrelin in subjects who had weight loss while ghrelin levels were constant in other subjects who had no weight loss (leidy et al., 2004). Also, Foster - Schubert et al found that ghrelin level increases as a result of exercise-induced weight loss without any decrease in the eaten food. Plasma ghrelin levels have been observed to vary with the increase or decrease in BMI (leidy et al., 2004). Karen et al proposed a hypothesis in their research that ghrelin is involved in regulating a negative feedback loop which regulates body weight. The hypothesis maintains that weight loss is a cause of increase in blood ghrelin level which is in fact known as part of adaptation to energy deficiency. In the study of Foster and Schubert upon completion of a one-year exercise protocol ghrelin showed an 18% increase in subjects who had lost more than 3 kg of their weight (Foster – Schubert et al., 2005). Therefore, in this study weight, BMI and body fat percentage showed significant decrease which, consistent with the abovementioned studies, may be a reason for the rise in plasma ghrelin. However, the results of some other studies are contrary to those of this research. Roberts et al showed in a study that a 93-day period of regular exercise in identical twins did not cause significant changes in ghrelin levels (Robert et al., 2007). Also in a study Mirzaei et al found that 8 weeks of aerobic exercise training in obese women increase levels of desacyl ghrelin, while no change are observed in acyl ghrelin levels (Mirzaei et al., 2009). In another study Heywon et al found that after 12 weeks of combined training in 11-year-old boys, desacyl ghrelin concentrations increased, whereas acyl ghrelin

Fig. 1. The pattern of changes in Ghrelin levels before and after 12 weeks exercise in two groups.

Fig. 2. The pattern of changes in Leptin levels before and after 12 weeks exercise in two groups.
concentrations did not change significantly (Kim et al., 2008). The results of few studies also indicate a slight decrease in plasma ghrelin after exercise. In this regard, a study by Maslow et al can be cited. The researchers found in their study that conditions of negative energy balance, such as physical activity can reduce the levels of ghrelin and its activity, reduce the amount of eating in obese people and in this way it can help them with weight loss (Marzullo et al., 2004). Similarly Wang et al also found that after 8 weeks of aerobic exercise, there will be different changes in plasma and hypothalamic ghrelin levels. Their findings showed that appetite and body weight in obese mice reduce by exercise and by methods of decreasing ghrelin levels in the hypothalamus (Wang et al., 2008). With a comprehensive look at the previous findings as well as this study it may be concluded that factors such as subjects’ gender, being overweight or underweight, duration and type of exercise protocol may be effective on changes in ghrelin levels following a period of training. Most of these studies, however, have focused on total ghrelin while Acyl ghrelin plays a role in the energy balance, which was assessed in the present study. Thus, the evident findings in previous research on the effects of exercise training on the levels of total ghrelin cannot be generalized to acyl ghrelin.

The results of this study show that serum leptin levels in the experimental group decreased significantly after 12 weeks of interval training (Fatouros et al; 2005., Gomez-Merino et al., 2002; Unal et al., 2005a; Unal et al., 2005b). Regardless of the mechanisms of this reductive change, these findings are consistent with the results of certain previous studies which also report decreased leptin level, although they are in contradiction with some others suggesting leptin to remain unchanged (Gomez-Merino et al., 2002; Fatouros et al., 2005; Unal et al., 2005a; Unal et al., 2005b). Indeed, previous findings on the effects of exercise on circulating leptin levels are not consistent (Koutsari et al., 2003; Karen et al., 2004; Aminian-Razavi et al., 2007; Benatti et al., 2007., Hamedinia et al., 2008., Haghighi et al., 2008). Ning et al report that regular physical activity and leptin concentrations are independently and inversely correlated. Put more clearly, in unison with some earlier researchers they found that plasma leptin concentrations would decrease in men and women as a result of regular physical activity (Ning et al., 2005). In another study, Bonati and Junior show that aerobic exercise can lead to a decrease in plasma leptin independent of changes in body fat and insulin (Benatti et al., 2007). Hayas et al also showed that 10 weeks of swimming aerobic exercise leads to a decrease in plasma leptin concentrations (Hayase et al., 2002). The effect of type of exercise on plasma leptin concentration is better revealed by studying the effect of interval training and comparing the same with continuous endurance training. Aminian, Razavi et al (2007) showed that 16 weeks of intermittent training has no effect on leptin concentration, but continuous exercise decreases it significantly. However, in this study where the type of exercise is interval exercise the serum leptin level decreases. It is worthwhile that the subjects in the study are all women while in the study of Aminian et al the subjects were men and it is possible that like some previous study gender is one influential factor (Hayase et al., 2002).

A review of other studies also shows that most of them report lowered leptin levels after exercise training. It should be noted, however, that almost all these researches study the effects of aerobic exercise in obese subjects. The said researchers mention different reasons for the decrease in leptin levels. Hamedinia et al (2008) suggest that this effect could be due to a significant reduction in body fat and cortisol levels and increased lean body mass in obese adolescents prompted by weight elementary swimming training as well as insignificant increase of insulin as a result of these exercises. Haghighi and Hamedinia (2008) report a positive and significant correlation leptin level with insulin and body fat percentage as well as a negative and significant correlation of leptin with free serum fatty acid at baseline level in obese men. They maintain that leptin is compatible with aerobic exercise and argue that
evident decrease in leptin levels in their study is more than the change attributed to fat tissue mass. Body fat percentage and insulin concentrations are variables to which most previous researchers have attributed the changes in leptin concentrations (Aminian-Razavi et al., 2007, Benatti et al., 2007, Hamedinia et al., 2008; Haghighi et al., 2008). They either report simultaneous reduction of body fat percentage and insulin concentrations with decreased leptin concentration as the cause of changes, or base their argument on the correlation between leptin concentrations, on the one hand, and body fat percentage or insulin concentration on the other hand. Of course, some other researchers assume leptin concentrations to be independent of changes in body fat percentage and insulin concentrations (Houmard et al., 2000; Benatti et al., 2007).

Conclusion
The findings of this research show that interval training leads to increased cardiovascular fitness, weight loss, reduced body fat and increased ghrelin and decreased leptin at the same time. Interval training seems to affect leptin secretion by decreasing fat mass. Exercise, however, probably brings about negative energy balance in the bodies of non-athletic obese women and ghrelin is secreted in response to this energy deficiency in order to stimulate food intake behavior to make up for the lost energy and re-establish the energy balance. This increase in ghrelin could stop the post-exercise catabolic processes and may result in glycogen supercompensation. The results in total support depletion of energy stores theory and increased ghrelin.

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