

Nervous Evaluation Conducted by the Changes of Ghrelin and Obestatin Executed by Aerobic Exercise

Ramin AGHAJANI¹,
Zahra Hojjati ZIDASHTI²,
Nematollah NEMATI³,
Tahereh BAGHERPOUR⁴

¹Department of English Language
Teaching and Translation, Shahr-e-Qods
Branch, Islamic Azad University, Tehran,
Iran, fakhracelaleh@yahoo.com

²Corresponding Author: Associate
Professor, Physical Education
Department, Rasht Branch, Islamic Azad
University, Rasht, Iran,
z_hoj@yahoo.com

³Associate Professor, Department of
Sport Sciences, Islamic Azad University,
Damghan Branch, Damghan, Iran,
nnemati258@gmail.com

⁴Assistance Professor, Department of
Sport Sciences, Islamic Azad University,
Damghan Branch, Damghan, Iran,
bagherpoor_ta@yahoo.com

Abstract: *Background: Ghrelin and obestatin are some effective peptides for regulating energy balance, which investigating them change caused by physical activity are important. The objective of this research was to evaluate the effect of eight weeks of aerobic exercise on plasma ghrelin and obestatin of obese and overweight women.*

Material and Methods: This quasi-experimental research was conducted in Rasht in 2018. In this research, 24 obese and overweight subjects were selected and randomly divided into two groups of aerobic exercise (N = 12) and control (N = 12). The aerobic exercise group performed an aerobic exercise program for 8 weeks in three sessions of 60 to 70 minutes. Acylated ghrelin, obestatin and body composition of the subjects were measured before and after 8 weeks. Data were analyzed by using SPSS software and T test.

Independent t-test revealed no significant difference between the levels of plasma ghrelin,

Results: The results showed that after an aerobic training period there was no significant difference between the two groups in the levels of ghrelin and obstatin ($p > 0.05$). However, BMI showed a significant decrease in aerobic training group ($p < 0.05$).

Conclusion: In conclusion, this exercise regimen had a positive effect on reducing BMI which is related to Slight changed ghrelin and obstatin responses over time. This finding lends support for a role of exercise in Nervous systems that stimulated some anabolic hormones such as ghrelin and abstatin and it can change relationship between ghrelin, obestatin and some body composition indexes.

Keywords: *Aerobic Exercise; Ghrelin; obestatin; Body Composition.*

How to cite: Faruji, L.F., Kharaghani, N. (2019). Nervous Evaluation Conducted by the Changes of Ghrelin and Obestatin Executed by Aerobic Exercise. *BRAIN. Broad Research in Artificial Intelligence and Neuroscience*, 10(4), 101-114. doi:10.18662/brain/07

Introduction

Obesity is currently considered a serious problem all over the world (Lancha, Frühbeck & Gómez-Ambrosi, 2012). The prevalence of obesity is very worrying, as obesity increases the risk of development of many chronic diseases such as hypertension, cardiovascular disease, diabetes, psychiatric problems, arthritis, gout, gallbladder disease, gastrointestinal diseases and cancer (Timper & Brüning, 2017).). Hence, it is important to investigate the factors associated with obesity (Wang & Nakayama, 2010). The best time for exercise during the day is not known to control weight in obese and overweight patients. Exercise time might influence appetite and food intake. The biological rhythm is related to cyclic changes, which occur at certain periods of time in day and influence the biological processes. In human, two groups of nerve cells called as Suprachiasmatic nucleus (SCN) are located on the base of the brain, known as anterior hypothalamus. Suprachiasmatic is the initiator of the biological rhythm in the body, which coordinates the rhythm of all the body organs during the 24-hour (Timper & Brüning, 2017). Studies have indicated that circadian rhythm controls some metabolic and hormonal indicators affecting the metabolism. The temperature of the body and the level of oxygen consumed in rest time are at the minimum at 4 am and they reach the maximum level at 3 and 4 pm (Azizi & Mohebie, 2013). The pattern of gastrointestinal movements, rate of intestinal absorption, activity of the gastrointestinal enzymes, and secretion of gastric acids also have a circadian rhythm (Wang & Nakayama, 2010; Azizi & Mohebie, 2013).

Energy balance is regulated through a complex system, including central and environmental factors. Ghrelin peptide is one of the known environmental factors playing a major role in regulating food intake and body weight. Ghrelin is a 28-amino acid peptide, which is mainly secreted by the stomach fundus cells and poured into the bloodstream (Azizi & Mohebie, 2013; Hedayati, Saghebjo, & Ghanbari-Niaki, 2012). After secretion, ghrelin leaves an effect on the center of satiety and hunger in the hypothalamus and stimulates the intake of food and the weight gain. The results of the researchers suggest that the expression of ghrelin gene in the stomach increases during fasting and decreases in satiety. In fact, the ghrelin plasma level decreases in the positive energy balance conditions and increases under the negative energy balance conditions (Wang & Nakayama, 2010; Hedayati et al., 2012). The ghrelin hormone is also an effective hormone in the process of appetite control. It is circulating in the blood in two active forms of acylated ghrelin and non-acylated ghrelin (Alizadeh,

Mostafaei, Mazaheri & Younespour, 2015). The blood level of this hormone also represents the hunger state and increases the food intake (Tiryaki-Sonmez et al., 2013). An increase in plasma concentration of ghrelin before meals and its reduction after taking food show its role in energy balance (Ebrahimi, Rahmaninia, Damirchi, Mirzaie & Asgharpur, (2013). Weight gain also decreases the circulating ghrelin level. Ghrelin in a negative-feedback loop is also involved in regulating the body weight (Howe et al., 2016).

Scientists have recently found another 23-amino acid peptide known as obestatin. This peptide is derived from the ghrelin-constructing gene, which has undergone different changes after translation. The results of this study showed that the treatment of rodents with obestatin leads to a negative energy balance by reducing food intake and gastric emptying. Obestatin is another peptide synthesized from a common gene with ghrelin identified in recent years with effects opposite to ghrelin in gastrointestinal functioning and homeostasis. In some studies, obestatin, as an inhibitory peptide, has been effective on ghrelin function (Wynne, Stanley, McGowan & Bloom, 2005).). The results show that in simultaneous injection of ghrelin and obestatin, the inhibitory effects of obestatin on the ghrelin function are seen in food intake, intestinal contraction inhibition, slow food intake from the stomach and loss of weight gain. These facts show that there is probably a complex balance between ghrelin and obestatin might be involved in different conditions of energy in regulating body weight (Zhang et al., 2007).

Ebrahimi et al (2013) reported that an exercise program along with diet decreases ghrelin and increases the base metabolism. This hormone is one of the factors disrupting the short and long-term balance of the energy during physical activity. Accordingly, the effect of physical exercises on the changes in this peptide requires more studies. It has been reported that eight weeks of aerobic activity has no significant effect on acylated ghrelin concentration (Howe et al., 2016). The results of Tremblay et al. (2007) show no effect of obestatin on food intake and weight loss. However, Zhao, Furnes, Stenström, Kulseng and Chen (2008) found results similar to Zhang et al. (2007) with the removal of obestatin receptors in mice.

As obesity is one of the greatest problems in modern societies today, recognizing factors and mechanisms for combating obesity would be helping in promoting community health and saving medical costs. Some studies have shown that ghrelin and obestatin have significant roles in energy balance and weight control (Van der Lely, Tschöp, Heiman & Ghigo, 2004; Lagaud et al., 2007). Thus, studying the relationship between changes in levels of these peptides and performing aerobic exercises seems to provide appropriate

strategies for weight control by appropriate exercises. Hedayati et al. (2012) stated that a diet-based exercise program reduces ghrelin and increases base metabolism. Hence, some researchers have concluded that ghrelin and obestatin had opposite effects on weight regulation and the inappropriate functioning of obestatin may be involved in obesity pathophysiology (Hedayati et al., (2012). While most studies have paid attention on the short-term effects of sports activities in the physical activity area, it seems that studying the changes in this peptide as a result of physical exercises is necessary, given the role of ghrelin and obestatin in long-term energy regulation. Thus, this research aims to evaluate the effect of aerobic exercises on variables of ghrelin, obestatin and body composition in obese women.

Methodology

This study was semi-experimental due to the lack of controlling some intervening factors. The population was 120 obese students of Payam-e Noor University of Guilan, who were studying in 2017. Non-random convenient sampling was used to do the research, with the sample size 24. First, coordination was done with Guilan Payam-e Noor University security. After coordination with the university, the obese and overweight people interested in participating in the research were informed and invited. After describing the research objectives and how it is done, health evaluation questionnaire was distributed among them and the health of the subjects was evaluated so that they were no sick. After the above steps and obtaining consent from all participants, 24 female obese women with an average age of 20-35 years with no regular physical activity during the past 6 months were selected to participate in the study. After this stage, the information of height, weight, and BMI of the subjects was collected. After the above steps, 12 subjects were assigned to the exercise group and 12 to the control group according to homogenization based on age and fat percentage. The exclusion and inclusion criteria of the individuals were included as not suffering from diseases like cardiovascular disease, hypertension, renal diseases and non-use of drugs, and lack of regular physical activity in the past six months.

The experimental group performed aerobic exercises at 9 am for 8 weeks in three sessions of 60-70 minutes. The exercise program included warming-up (10 minutes) and running on a smooth surface in sports hall. By increasing the number of exercises, the intensity of activities was also increased. At the end of each exercise session, the cooling-down step was considered for 5 minutes including slow running and stretching movements.

The intensity of exercises was at 55-75% of maximal heart rate during the exercise. Given the low readiness of the subjects, the minimum exercise intensity was considered at 55% of maximal heart rate in the first two weeks. In the next sessions, with observing the principle of overdose, the intensity of exercise increased up to 75% of maximal heart rate (Manshouri, Ghanbari-Niaki, Kraemer and Shemshaki, 2008; Reinehr, De Sousa and Roth, 2008).

Initial blood sampling was performed in 48 hours before the first exercise session in all subjects in the fasting state. 10 cc of blood were taken from the brachial vein of the samples. In addition, 48 hours after the last exercise session, 10cc of blood were taken from brachial vein of subjects in the fasting state in order to eliminate the effect of exercise. Subjects were asked to avoid intake of food for 12 hours up to the blood sampling time. In order that blood samplings in the pretest and post-test to be similar, sampling started at 8am and ended at 9am. The blood samples were collected in EDTA-containing tubes, and then, they were immediately centrifuged and the obtained plasma was stored in separate tubes at -80 ° C for performing next experiments. Body mass index was calculated by dividing body weight (in kg) by height (in m).

The ghrelin measurement kit (Glory Science Co. China) by ELISA method was used to measure ghrelin concentration in terms of ng/ml unit. Ghrelin level of plasma samples were measured by Sandwich ELISA method. The sensitivity of the method was 15.6 Pg/ml. The percentage of intra-test changes coefficient was determined to be 7.4%. Obestatin measuring kit with ELISA method, made in China, Glory Science Co., was used to measure Obestatin concentration in nano-milligrams units. Plasma Obestatin levels were measured by Sandwich ELISA method. The sensitivity of the method was 78 pg / ml. The coefficient of in-test variation was 6.9%.

Descriptive statistics are used to describe the variables and inferential statistics are used to analyze the results in this research. To examine the normality of the variables in each group, the Kolmogorov Smirnov test is used. Correlation t-test was used to evaluate the effect of the exercises and compare the pre-test and post-test in each group and independent t-test was used for inter-group comparisons. The acceptance or rejection of the hypotheses was considered at a significant level of $p < 0.05$.

Results

The mean of the variables in the pre and post-intervention periods is presented in Tables 1 and 2. In-group analysis showed no significant

differences between the BMI of obese women in the exercise group before and after the intervention ($P = 0.091$); Furthermore, no significant differences between the BMI of obese women in the control group before and after intervention ($P = 0.151$) (Table 3). In addition, the results of intra-group study showed a significant difference between the WHR of obese women in the exercise group before and after intervention ($P = 0.003$), while there were no significant differences between WHR of obese women in the control group before and after intervention ($P = 0.81$) (Table 3). However, independent t-test showed a significant difference between BMI and WHR in three groups ($P = 0.035$ and $P = 0.0001$, respectively) (Table 3). Likewise, intra-group analysis showed a significant difference between the fat percentage of obese women in the exercise group before and after the intervention ($P=0.0001$). However, there were no significant differences between the fat percentage of obese women in control group before and after intervention ($P = 0.53$) (Table 3). The results of independent t-test showed a significant difference between the level of body fat percentage in the training group and the control group ($P = 0.0001$).

Intra-group analysis showed no statistically significant differences between obestatin in obese women in the exercise group before and after the intervention ($P = 0.733$). Additionally, there were no significant differences between blood obestatin in control group before and after intervention ($P = 0.91$) (Table 3). Moreover, independent t-test showed no significant differences between obestatin changes in the level in three groups ($P = 0.817$) (Table 3). Using correlation t-test, it was found that there was no statistically significant difference between the ghrelin level in obese women in the aerobic exercises group before intervention and after the intervention ($P = 0.562$). Thus, there is no significant difference between the ghrelin blood levels of obese women in the aerobic exercises group before and after the intervention (Table 3).

Table 1: Description of the variables examined before intervention
(mean $\pm$ standard deviation)

Variable	exercise group	control group
Age (year)	29.9 $\pm$ 7.65	25.3 $\pm$ 7.4
Height (centimeter)	170.12 $\pm$ 5.34	167.97 $\pm$ 32.13
Weight (kg)	76.53 $\pm$ 22.31	80.31 $\pm$ 12.14
BMI(kg/m ²)	31.32 $\pm$ 4.01	31.27 $\pm$ 0.04
WHR	0.78 $\pm$ 0.06	0.82 $\pm$ 0.04
Body fat percentage	47.83 $\pm$ 1.61	44.86 $\pm$ 3.89
Lean Body Mass (kg)	40.02 $\pm$ 8.46	44.6 $\pm$ 4.47

Vo2max (ml/Fg/min)	31.53±6.23	36.6±3.3
Systolic blood pressure (mmHg)	12.0±0.66	12.11±0.6
Diastolic blood pressure (mmHg)	8.0±0.66	8.33±0.5
obestatin (ng/mg)	1.16±0.67	0.9±0.14
Ghrelin (ng/ml)	3.72 ± 1.17	3.8 ± 1.07

The values are presented as mean ± standard deviation.

Table 2: Description of the variables examined after intervention
(mean ± standard deviation)

Variable	Morning group	Control
Weight (kg)	75.53±18.41	81.44±8.25
BMI(kg/m2)	30.6±4.43	31.35±2.07
WHR	0.76±0.06	0.82±0.04
Body fat percentage	45.4±2.01	44.9±3.82
Lean Body Mass (kg)	40.7±8.81	44.6±4.15
Vo2max (ml/Fg/min)	37.93±6.57	36.8±3.2
Systolic blood pressure (mmHg)	12.0±0.66	12.1±0.6
Diastolic blood pressure (mmHg)	8.0±0.66	8.33±0.5
obestatin (ng/mg)	1.07±0.63	0.91±0.1
Ghrelin (ng/ml)	3.35 ± 1.66	3.66 ± 0.69

Table 3: Changes in Variables in pre and post intervention among the three study groups

Variable	Group	Pre-intervention	Post-intervention	Statistical estimate	Differences in pre and post-intervention	Statistical estimate
BMI(kg/m ²)	exercise	31.32±4.01	30.6±4.43	t= 1.89 P= 0.091	-0.68±1.13	F= 3.8 P= 0.035**
	control	31.27±0.04	31.35±2.07	t= 1.59 P= 0.151	0.08±0.16	
WHR	exercise	0.78±0.06	0.76±0.06	t= 4.11 P= 0.001*	-0.01±0.01	F= 13.43 P= 0.0001**
	control	0.82±0.04	0.82±0.04	t= 2.00 P= 0.081	0.003±0.005	
Body fat percentage	exercise	47.83±1.61	45.4±2.01	t= 5.73 P= 0.0001*	-2.39±132	F= 23.00 P= 0.0001**
	control	44.86±3.89	44.9±3.82	t= 0.65 P= 0.53	0.12±0.58	
obestatin (ng/mg)	exercise	1.16±0.67	1.07±0.63	t= 0.35 P= 0.733	-0.08±0.79	F= 0.203 P= 0.817

	control	0.9±0.14	0.91±0.1	t= 0.11 P= 0.91	0.007±0.2	
Ghrelin (ng/ml)	exercise	3.35 ± 1.66	3.72 ± 1.17	t= 0.602 P= 0.562	-0.36 ± 1.92	F= 1.69 P= 0.16
	control	3.66 ± 0.69	3.8 ± 1.07	t= 0.585 P= 0.575		

* P < 0.05; Significant difference in compare to Pre values (p≤0.05). ** P≤ 0.05, Significantly different between groups.

Discussion and conclusion

The objective of this research was to evaluate the changes in plasma ghrelin peptide after eight weeks of aerobic exercises in obese and overweight women. No significant change was seen in the levels of acylated ghrelin after 8 weeks of aerobic exercise. The results of this research are not consistent with the results of the research conducted by Ghanbari Niaki, Saghebjo, Rahbarizadeh, Hedayati, and Rajabi (2008), who examined the effect of one-stage multi-intensity on the levels of plasma obestatin levels in young female college students. Manshoury et al (2008) also reported no significant change in plasma obestatin levels after one week of aerobic exercise in female students. In addition, the results of Reinehr, De Sousa and Roth (2008) also showed that long-term aerobic exercise associated with weight loss led to an increase in plasma obestatin levels in children. Some of the possible mechanisms related to ghrelin can contribute in properly understanding of this change in this research. Various studies have shown that exercises lead to an increase in glycogen breakdown and a fraction of energy, and protein and glycogen reconstruction is delayed after exercises (Ghanbari-Niaki, Jafari, Abednazari & Nikbakht, 2008).

Different studies have reported that eccentric activities lead in muscle damage and defect in re-synthesis of glycogen after activity. Investigations on human and animal samples also revealed that insulin-induced increase in glucose uptake and transfer by the muscle were positively correlated with content of glucose transmitter protein 4 (GLUT-4). In fact, it was found that the content of muscle GLUT-4 had a direct association with muscle glycogen levels. Thus, the defect in the re-synthesis of muscle glycogen after periodic contractions might be involved due to the reduction in the level of GLUT-4 protein caused by muscle damages (Ghanbari-Niaki et al., 2008) because the content of the GLUT-4 protein is one of the determinants of muscle uptake by glucose. Thus, damage in muscle fibers following high intensity exercise may delay the reconstruction of muscle glycogen. Considering that the severe muscle contractions can

damage muscle fibers and reduce the content of glucose transmitters GLUT-4 of muscle cells, it can cause delay in the reconstruction of muscle glycogen stores (Reinehr et al., 2008).

Some studies have also reported that defect in re-synthesis of glycogen is probably due to the accumulation of inflammatory cells and, consequently, the competition between inflammatory cells and muscle fibers to achieve plasma glucose following the physical activities in the human subjects (Manshouri et al., 2008). Hence, it could be stated that exercise with an intensity of 55-75% of the maximal heart rate is the response of these hormones to this energy shortage. It could be in fact stated that breakdown of ghrelin precursor has led to an increase in production of ghrelin. As a result, obestatin secretion and the ratio of ghrelin to obestatin are influenced to affect the behavior of food intake, the lost energy sources and finally the body energy balance. It was also observed that ghrelin expression in aerobic and resistance exercise groups was not associated with significant changes in plasma ghrelin levels (Manshouri et al., 2008; Ghanbari-Niaki et al., 2008). Studies have shown that ghrelin and its receptor are expressed in peripheral blood mononuclear cells in subjects with metabolic syndrome. It has been also shown that ghrelin expression has a positive correlation with TNF- and IL-1 expression. Researchers concluded that ghrelin expression might play a role in the autoimmune immune system (Ghanbari-Niaki et al., 2008). In fact, various studies have indicated that ghrelin might moderate the proliferation of immune cells and activation and secretion of pre-inflammatory cytokines (Reinehr et al., 2008).

In general, it is not still known that how exercise lead to changes in ghrelin and further studies are required in this regard. Several studies have been conducted on the effect of aerobic exercise on plasma ghrelin levels in human and mouse subjects so far, most of which have led to a significant increase in plasma ghrelin (Lagaud et al., 2007; Manshouri et al., 2008). The results of the previous studies have shown that long-term exercise interventions led to exercise-induced weight loss significantly increased plasma ghrelin levels.

The results showed that increased plasma ghrelin was observed only in subjects who lost weight during the exercise program, and tendency to increase plasma ghrelin was more in subjects who lost weight less than 5% (Lagaud et al., 2007). After one year of aerobic activity with moderate intensity in postmenopausal obese women, the levels of the total blood ghrelin increased proportional to the weight loss, and the levels of blood ghrelin remained fixed in subjects whose weight remained unchanged (Reinehr et al., 2008). It seems that increased ghrelin is a compensatory

response to weight loss. In other words, the increase in ghrelin probably acts as a compensatory mechanism for restoring body weight to a regulated point (Ghanbari-Niaki et al., 2008). In general, most studies have reported that long-term activities would result in increased levels of ghrelin plasma, if it is associated with weight loss (Reinehr et al., 2008; Ghanbari-Niaki et al., 2008). As no significant increase was seen in plasma ghrelin in the current research, it can be justified by the lack of weight loss in subjects during the exercise program. Thus, one of the factors which might be effective in affecting the level of ghrelin changes is the duration of the exercise program. Thus, the duration of the exercise program in this research is probably one of the factors that did not lead into significant changes in plasma ghrelin.

After 8 weeks of aerobic exercise program, there was no significant change in the concentrations of obestatin in any of the groups in the study. Moreover, there were no significant changes in the concentration of obestatin in any of the groups. Studies regarding physical activity and obestatin are very few and the interpretation of the results is very difficult due to little background. Rinher et al. (2008) in a study on obese children, showed the concentration of base obestatin higher. In a study by Ganbari-Niaki et al. (2008) on rats, there were no significant changes in the plasma concentrations of obestatin after 6 weeks of training on the rotating band at a speed of 25 m / min for 5 days a week. Nevertheless, there was significant decrease in obestatin of gastric and intestinal tissue. Several hormones including insulin, somatostatin and glucagon regulate Ghrelin and obestatin in plasma and stomach. In a recent study by Ganbari-Niaki et al. (2008), only the growth hormone had been measured, where the results showed a negative correlation between GH and obestatin. However, information about the effect of physical activity on growth hormone and ghrelin shows a negative feedback loop between increasing CH and the decrease in ghrelin plasma and stomach, but information about obestatin is not available yet. Dall et al. (2002) stated that by GH infusion to subjects, ghrelin concentration decreased after physical activity, but in the absence of GH, ghrelin concentration did not change significantly. Wang and Nakayama (2010) examined the effect of 1 session and 8 weeks of endurance training on a belt rig at a speed of 22 m / min for 5 days a week on the levels of obestatin, ghrelin plasma and hypothalamus in rats. In both acute and chronic conditions, the concentrations of obestatin and ghrelin plasma did not change significantly, but the activity of ghrelin and obestatin in the hypothalamus decreased after 8 weeks. The results of the present study, regarding the lack of changes in plasma obestatin, are in line with the three above-mentioned studies, showing that plasma obestatin alone does not

have a significant role in regulating energy balance. Zhao et al. (2009) argued that in obese children and control group with normal weight, after 3 months of diet and free aerobic exercise (basketball, swimming, running and cycling), performed at two turns in the morning and afternoon, plasma obestatin and ghrelin concentration increased significantly. According to them, the levels of ghrelin and obestatin are associated with obesity in children. Reinehr et al. (2008) also found significant increases in obestatin levels in their study on obese children after one year of combined dietary diet and body weight gain, whereas ghrelin showed no significant changes. Following the changing lifestyle in the long run, obestatin has been shown to play a role in regulating energy balance in children. It is probable that the change in lifestyle for a long time, therefore, the change in energy balance will affect its plasma concentration. Food intake, glucose uptake, high-carbohydrate diet and negative energy balance decrease gastric and metastatic gastric abscess in some animal and human specimens. Increased levels of liver and muscle glycogen in trained rats can be related to lower levels of obestatin in the stomach tissue. This is because the studies have shown that endurance training increase muscle and liver glycogen (with or without carbohydrate loading) in human and animal specimens (Ghanbari-Niaki et al., 2008). Andersson et al. (2005) reported that one running session on treadmill could reduce glycogen in liver and muscle. Following this evacuation with high-carbohydrate intake, liver glycogen storage increased and plasma ghrelin levels decreased. In a study by Ganbari Niaki et al. (2008), ghrelin level was not measured in tissue or plasma, but their results showed that obestatin does not have a significant role in regulating energy balance, which is in line with the results of this study. For the first time, Zhang et al. (2007) reported that obestatin has a role in controlling energy balance. Sibilia et al. (2006) found different factors related to energy metabolism like consumed food, body weight, body composition, energy consumption, locomotor activity, respiratory rate, and hypothalamic neuropeptides affecting energy balance regulation in response to long-term injection of obestatin in rats, but found no significant effects. Furthermore, Nogueiras et al. (2007) found the effect of obestatin on the regulation of energy balance with similar results and found it ineffective in regulating energy balance. Zhao et al. (2006) examined the obese children, who had 3 hours aerobic activity per day, performing in three fields of basketball, swimming and Ping-Pong in the morning and afternoon. The diet was performed for a month. The results showed that the concentration of ghrelin and plasma obestatin presents a significant increase after significant weight loss. These researchers believe that ghrelin and obestatin are connected with childhood obesity. In women with severe

obesity, who lost 62% of their extra weight using gastric bypass surgery, ghrelin levels decreased significantly, whereas obestatin concentrations showed no significant changes. Opposite to previous results, weight loss was associated with decrease in ghrelin levels, and obestatin concentrations showed no changes.

After all, in aggressive studies involving stomach surgery, a part of the stomach where the ghrelin producing cells are is eliminated while other areas had more ghrelin secretion to make compensations (Ghanbari-Niaki et al., 2008). Vicennati et al. (2002) reported that obese women had higher concentrations of obestatin and lower ghrelin and lower ghrelin to obestatin ratio compared to the control group. These researchers argue that the lack of balance between ghrelin and obestatin can play a significant role in the pathophysiology of obesity. The role and importance of obestatin in activity as an anti-appetite peptide involved in energy balance to prevent weight gain is still ambiguous due to little understanding of it. Probably, the level of obestatin is affected by exercise and may result in changes in appetite and weight. Due to the recent discovery of this peptide, only a few studies have examined the effect of exercise on this peptide. Due to the effect of obestatin on energy balance, the level of this peptide may be affected by physical activity leading to changes in appetite and weight (Ghanbari-Niaki et al., 2008). As exercise can cause a negative energy balance, it is likely that the level of obestatin is affected by exercise as well. In the few studies conducted so far, different results have been reported about the effect of physical activity on the level of plasma obestatin and various tissues. It seems that the type, duration and intensity of exercise are likely to affect the level of obestatin. Moreover, different tissues may show different responses to exercise. Increased obestatin following weight loss may be a necessary mechanism to maintain weight loss. Factors such as the fasting or non-fasting of the subjects during the study, the body weight, BMI, the type of exercise program used, and even the time of post-activity sampling have changed the results and have had a role in the results. There has been a significant correlation between changes in BMI and changes in plasma obestatin in obese subjects. Moreover, a significant negative correlation was found between the level of growth hormone and the level of obestatin fundus and small intestine. Overall, it seems that the increase in the level of obestatin in the weight loss process is critical to maintaining and preventing weight gain, because obestatin peptide is an anti-appetite and has a function opposite to that of ghrelin (Dall et al., 2002; Andersson et al., 2005).

The results of this research revealed that 8 weeks of aerobic exercise did not significantly decrease BMI levels in the experimental group. The

results of this research were not in line with those of studies which showed a significant change in BMI (Hovanloo, Arefirad & Ahmadizad, 2013) and in line with results of the studies which reported lack of change in BMI (Marandi et al., 2014). Marandi et al (2014) reported that a short period of physical activity intervention had a positive impact on body fat percentage and cardiovascular fitness of the overweight and obese subjects. However, Hovanloo et al. (2013) concluded that there is no significant difference between weight changes and body mass index following a speed periodic exercise and an increasing endurance exercise. Inconsistency and lack of significant changes in waist to hip ratio were probably due to the low intensity of exercise, the duration of exercise and the non-control of subjects' diet in this research. In short, the results of this research suggest that the concentration of acylated ghrelin in obese subjects did not change significantly after eight weeks of aerobic exercise with moderate intensity. It seems that the intensity and duration of exercises were not adequate to cause changes in these variables and exercises with high intensity and longer duration are needed. In addition, the fasting state of subjects might be effective on ghrelin concentration, which should be considered in further studies.

References

- Alizadeh, Z., Mostafaei, M., Mazaheri, R., & Younespour, S. H. (2015). Acute effect of morning and afternoon aerobic exercise on appetite of overweight women. *Asian J Sports Med.* 6(2), 24222
- Andersson, U., Treebak, J. T., Nielsen, J. N., Smith, K. L., Abbott, C. R., Small, C. J., Carling, D., & Richter, E. A. (2005). Exercise in rats does not alter hypothalamic AMP-activated protein kinase activity. *Biochem Biophys Res Commun.* 329(2), 719-25. doi:10.1016/j.bbrc.2005.02.027
- Azizi, M., & Mohebie, H. (2013). The effect of exercise in the morning and evening on the maximum oxidation fat in obese and normal-weight men. *Research in practical exercise physiology*, 10(19), 31-42.
- Dall, R., Kanaley, J., Hansen, T. K., Møller, N., Christiansen, J. S., Hosoda, H., Kangawa, K., & Jørgensen, J. O. (2002). Plasma ghrelin levels during exercise in healthy subjects and in growth hormone-deficient patients. *Eur J Endocrinol.* 147(1), 65-70. doi:10.1530/eje.0.1470065
- Ebrahimi, M., Rahmaninia, F., Damirchi, A., Mirzaie, B., & Asgharpur, S. (2013). Effects of short-term exercises on appetite, energy intake and energy-regulating hormones. *Iran J Basic Med Sci*, 16(7), 829–834.

- Ghanbari-Niaki, A., Jafari, A., Abednazari, H., & Nikbakht, H. (2008). Treadmill exercise reduces obestatin concentrations in rat fundus and small intestine. *Biochem Biophys Res Commun*, 372(4), 741-5. doi:10.1016/j.bbrc.2008.05.097
- Ghanbari-Niaki, A., Saghebjo, M., Rahbarizadeh, F., Hedayati, M., & Rajabi, H. (2008). A single circuit resistance exercise has no effect on plasma obestatin levels in female college students. *Peptides*, 29(3), 487-90. doi:10.1016/j.peptides.2007.11.002
- Hedayati, M., Saghebjo, M., & Ghanbari-Niaki, A. (2012). Effects of circuit resistance training intensity on the plasma ghrelin to obestatin ratios in healthy young women. *Int J Endocrinol Metab*, 10(2), 475-479. doi:10.5812/ijem.2459
- Hovanloo, F., Arefirad, T., & Ahmadizad, S. (2013). Effects of sprint interval and continuous endurance training on serum levels of inflammatory biomarkers. *J Diabetes Metab Disord*, 12(1), 22. doi:10.1186/2251-6581-12-22
- Howe, S. M., Hand, T. M., Larson-Meyer, D. E., Austin, K. J., Alexander, B. M., & Manore, M. M. (2016). No effect of exercise intensity on appetite in highly-trained endurance women. *Nutrients*, 8(4), 223. doi:10.3390/nu8040223
- Lagaud, G. J., Young, A., Acena, A., Morton, M. F., Barrett, T. D., & Shankley, N. P. (2007). Obestatin reduces food intake and suppresses body weight gain in rodents. *Biochem Biophys Res Commun*, 357(1), 264-9. doi:10.1016/j.bbrc.2007.03.138
- Lancha, A., Frühbeck, G., & Gómez-Ambrosi, J. (2012). Peripheral signaling involved in energy homeostasis control. *Nutr Res Rev*, 25(2), 223-48. doi:10.1017/S0954422412000145
- Manshouri, M., Ghanbari-Niaki, A., Kraemer, R. R., & Shemshaki, A. (2008). Time course alterations on plasma obestatin and growth hormone levels in response to short-term anaerobic exercise training in college woman. *Appl Physiol Nutr Metab*, 33(6), 1246-9. doi:10.1139/H08-098
- Marandi, S. M., Minasian, V., Kelishadi, R., Khalighinejad, P., Borojeni, M. M., & Borghi, S. H. (2014). Short-term effects of a physical activity intervention on obesity and cardiovascular fitness of 12-14-year-old boy students. *Int J Prev Med*, 5(Suppl 2), S114-9. doi:10.4103/2008-7802.157668
- Nogueiras, R., Pfluger, P., Tovar, S., Arnold, M., Mitchell, S., Morris, A., Perez-Tilve, D., Vázquez, M. J., Wiedmer, P., Castañeda, T. R., DiMarchi, R., Tschöp, M., Schurmann, A., Joost, H. G., Williams, L. M., Langhans, W., & Diéguez, C. (2007). Effects of obestatin on energy balance and growth hormone secretion in rodents. *Endocrinology*, 148(1), 21-6. doi:10.1210/en.2006-0915
- Reinehr, T., De Sousa, G., & Roth, C. L. (2008). Obestatin and ghrelin levels in obese children and adolescents before and after reduction of overweight. *Clin Endocrinol (Oxf)*, 68(2), 304-10. doi:10.1111/j.1365-2265.2007.03042.x

- Sibilia, V., Bresciani, E., Lattuada, N., Rapetti, D., Locatelli, V., De Luca, V., Donà, F., Netti, C., Torsello, A., & Guidobono, F. (2006). Intracerebroventricular acute and chronic administration of obestatin minimally affect food intake but not weight gain in the rat. *J Endocrinol Invest.* 29(11), RC31-4. doi:10.1007/BF03349204
- Timper, K., & Brüning, J. C. (2017). Hypothalamic circuits regulating appetite and energy homeostasis: pathways to obesity. *Dis Model Mech.* 10(6), 679-689. doi:10.1242/dmm.026609
- Tiryaki-Sonmez, G., Ozen, S., Bugdayci, G., Karli, U., Ozen, G., Cogalgil, S., Schoenfeld, B., Sozbir, K., & Aydin, K. (2013). Effect of exercise on appetite-regulating hormones in overweight women. *Biol Sport*, 30(2), 75-80. doi:10.5604/20831862.1044220
- Tremblay, F., Perreault, M., Klamann, L. D., Tobin, J. F., Smith, E., & Gimeno, R. E. (2007). Normal food intake and body weight in mice lacking the G protein-coupled receptor GPE39. *Endocrinology*, 148(2), 501-6.
- Van der Lely, A. J., Tschöp, M., Heiman, M. L., & Ghigo, E. (2004). Biological, physiological, pathophysiological, and pharmacological aspects of ghrelin. *Endocr Rev.* 25(3), 426-57. doi:10.1210/er.2002-0029
- Vicennati, V., Genghini, S., De Iasio, R., Pasqui, F., Pagotto, U., & Pasquali, R. (2007). Circulating obestatin levels and the ghrelin/obestatin ratio in obese women. *Eur J Endocrinol.* 157(3), 295-301. doi:10.1530/EJE-07-0059
- Wang, Z., & Nakayama, T. (2010). Inflammation, a link between obesity and cardiovascular disease. *Mediators Inflamm*, 2010,535918. doi:10.1155/2010/535918
- Wynne, K., Stanley, S., McGowan, B., & Bloom, S. (2005). Appetite control. *J Endocrinol*, 184 (2), 291-318. doi:10.1677/joe.1.05866
- Zhang, J.V., Klein, C., Ren, P.G., Kass, S., Donck, L.V., Moechars, D., & Hsueh, A. J. W. (2007). Response to comment on obestatin, a peptide encoded by the ghrelin gene opposes ghrelin's affects on food intake. *Scienc*, 315 (5813), 766.
- Zhao, C. M., Furnes, M. W., Stenström, B., Kulseng, B., & Chen, D. (2008). Characterization of obestatin and ghrelin producing cells in the gastrointestinal tract and pancreas of rats: An immunohistochemical and electron-microscopic study. *Cell Tissue Res.* 331(3), 575-87. doi:10.1007/s00441-007-0514-3