Progress in Nutrition 2018; Vol. 20, Supplement 2: 137-144 DOI: 10.23751/pn.v20i2-S.5915

Original article

Obestatin and Nerve Growth Factor in Patients with Metabolic Syndrome

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Summary. *Purpose:* Obestatin and nerve growth factor (NGF) have recently been introduced as adipokines. Several evidences have implicated involvement of these two peptides in some metabolic disorders. We aimed to investigate the association between serum obestatin and NGF levels with metabolic syndrome (MetS) parameters in women. *Methods:* Forty three female patients with MetS and 43 BMI-matched healthy women as control group, aged 30- 50 years, participated in the study. Anthropometric parameters, blood pressure, fasting serum obestatin, NGF, TG, TC, HDL-C, LDL-C,FBS, insulin and HOMA-IR were measured. The relationship between serum levels of obestatin and NGF with measured variables was analyzed. *Results:* Women in MetS group had significantly higher values of WHR ($p = 0.01$), systolic ($p = 0.01$) and diastolic blood pressure ($p = 0.01$), TG ($p < 0.001$), FBS ($p < 0.001$), insulin ($p = 0.004$), HOMA-IR ($p < 0.001$) and lower level of HDL-C ($p < 0.001$) as compared with healthy women. Serum levels of obestatin partially ($p = 0.08$) and NGF levels significantly was lower ($p = 0.02$) in MetS group than control. Both serum levels of NGF ($\beta = -0.41$, $p = 0.01$) and obestatin ($\beta = -0.38$, $p = 0.02$) were negatively associated with serum TG levels in MetS group. There was significant and positive association between serum obestatin and NGF levels in MetS group ($\beta = 0.41$, $p = 0.01$). *Conclusion:* The results indicating that obestatin and NGF might be important regulators of TG level and through this involved in the pathogenesis of MetS.

Key words. NGF, obestatin, metabolic syndrome

Introduction

Metabolic syndrome (MetS) is a complicated and multifactorial disease of modern life which has become increasingly important in terms of socio-medical aspects by involving about 56 and 43 percents of women and men population around the world, respectively (1–3). Based on Adult Treatment PanelIII(ATPIII) criteria this syndrome is known by presence of at least three of five traits including central obesity, glucose metabolism impairment, dyslipidemia, high blood pressure and insulin resistance (4, 5).There is a potent correlation between this syndrome and increased risk of atherosclerotic cardiovascular disease and type 2 diabetes mellitus (6). According to reliable evidences insulin resistance and abdominal obesity are the major and pivotal risk factors for MetS (5, 7).Adipose tissueis an active endocrine organ that the function of other organs can be affected by it. This organ is the source of the wide variety of adipokineswhich are peptides and proteins with numerous roles in body's metabolic regulation (8) and their dysregulationis involved in the pathogenesis of the metabolic disorders including insulin resistance and related disorders (8). In recent years, there has been an increasing interest in research about "adipobiology" and deficit of metabotrophic factors in the pathogenesis of some metabolic disorders which are linked with adipose tissue (9). Two peptides, in recent years, have suggested to be placed on the list of adipokines, which include obestatin and nerve growth factor (NGF) (9,10).

Obestatin is a pre-proghrelingenederived peptide that was introduced in 2005 as a hormone in the regulation of food intake and energy homeostasis (11).Its dysregulation can be involved in the pathogenesis of insulin resistance and MetS (12–15). NGF is the first member of the neurotrophin family that was discovered (16). There are some evidences that the systemic and local levels of NGF are subjected to change in some metabolic disorders such as obesity, diabetes mellitus, atherosclerosis and MetS (3, 17). Since there are so limited studies on the association between obestatin and NGF levels with metabolic syndrome components and the relationshipof these two factorshasnot beenstudied yet, we aimed to investigate the association between these two novel adipokineswith each other and with the components of MetS in women.

Materials and Methods

Participants

This cross sectional study was conducted from January to June,2015;on 86 women volunteers (aged 30–50years). According to the ATP III criteria,43of women were with MetS and another 43 BMI-matched women without MetS and apparently healthy as control group. Women in control group were with noobvious symptoms of any disease at the time of sampling. Consecutive sampling method was used for recruitment of participants from medical weight loss center, prior to treatment. The study was approved by the Ethical Committee of Tabriz University of Medical Sciences, Tabriz, Iran, and a written informed consent was obtained from each subject.

Inclusion criteria for MetS group were having at least 3 of 5 these ATPIII criteria including 1-abdominal obesity, given as waist circumference> 88cm,2-serum triglyceride (TG) ≥ 150 mg/dl,3-high-density lipoprotein cholesterol(HDL-C) ≤ 50 mg/dl,4-blood pressure ≥ 130 / 85 mm Hg,5-fasting blood glucose (FBG) ≥ 100 mg/dl. Exclusion criteria were:1-body mass index (BMI) ≥ 40 kg/m^2,2-havinginfectious and chronic inflammatory diseases, history of psychiatric diseases, thyroid disorders,endocrine diseases,irregular menstrual periods and previous stomach surgery, 3-smoking andexcessive alcohol consumption, 4-beingpregnant,lactating and, menopause and5-receiving anti-obesity, anti-inflammatory, anti-hypertensive, corticosteroid,estrogen and contraceptive drugs, also being on diet 3 months prior to participation in the study.

Anthropometric and blood pressure assessments

Anthropometric variables including height, weight, waist circumference (WC) and hip circumference were measured by an expert individual. Body weight was measured using a digital scale, with the examinee wearing a light gown. Height was measured barefoot using a wall-mounted stadiometer to the nearest 0.5 cm. BMI was calculated as body weight (kg) divided by the square of height (m^2). WC was measured by a tape measure at the midpoint between the lower costal margin and iliac crest to the nearest 0.5 cm. Hip circumference was measured around the widest portion of the buttocks, with the tape parallel to the floor. Waist-to-hip ratio (WHR) was calculated as waist measurement divided by hip measurement. Blood pressure was measured with a calibrated sphygmomanometer, after 5-minute rest in a sitting position. Blood pressure values were estimated as the mean of 2 readings.

Physical activity and stress levels assessment

Physical activity level was determined by International Physical Activity Questionnaire (IPAQ) short form (18)and expressed as a standard Metabolic Energy Turnover in MET-minutes/week. Three levels of physical activity were classified: low (<600 MET-minutes/week), moderate (from 600 to 2999 MET-minutes/week) and high (3000 MET-minutes/week or more). The stress level of participants was assessed by means of the Holmes and Rahe stress scale (19). Stress level was defined as mild (score< 150), moderate (score= 150-300) and severe (score> 300) with face to face interview.

Biochemical assessment

After an overnight fasting, 5 ml venous blood was collected. The serum samples were separated from whole blood by centrifugation at 2000 rpm for10 min at 4°C.Aliquots were stored at -70°C until analysis.

The levels of serum total cholesterol (TC), HDL-C and TG were measured by enzymatic colorimetric methods with a commercially available kit

(Pars Azmone, Tehran, Iran) on an automatic analyzer (Abbott, modelAlcyon 300, USA). Serum LDL-C was calculated by Friedewald equation (20). Fasting blood sugar were determined by the glucose oxidase method. Insulin levels were determined by insulin ELISA kit (MonobindInc, lake forest, CA 92630, USA). To investigate insulin sensitivity the homeostasis model assessment (HOMA-IR) was calculated. Obestatinand NGF levels were measured using a commercially ELISA kits (Hangzhou Eastbiopharm CO.,LTD).

Statistical analyses

Demographic characteristics of the participants were summarized by either mean ± standard deviation (SD) for continuous data, or frequency (percentage %) for proportional data. Differences between two groups were assessed by independent samplet-testfor normally distributed data and Mann-Whitney U test for nonparametric variables and Chi-squared test for qualitative variables. Spearman's coefficients correlations were performed for associations between NGF and obestatin. Univariate and multivariate linear regression were performed to find associations between NGF and obestatin with MetS components. Data analysis was performed by SPSS 22 Software (IBM-SPSS statistics, IL, Chicago, USA) and P- values less than 0.05 were considered statistically significant.

Results

Demographic characteristics

Demographic characteristics of the studied population are presented in Table 1. Average age of MetS and control subjects were 38.77 ± 6.51 and 35.91 ± 5.71 years, respectively (p=0.01).There was no statistical significant difference in levels of physical activity, education, stress and marital status between the two groups (Table 1).

Anthropometric and blood pressure measurement

As shown in Table 2, mean WHR was significantly higher in MetS group as compared with control group (p= 0.01).There were also significant differences in systolic (p= 0.01) and diastolic blood pressure (p=0.01) between the two groups with higher values in MetS group (Table 2).

Lipid profile and metabolic parameters

Regarding lipid profile, as shown in Table 3, subjects with MetS had significantly higher level of TG (p<0.001) and lower level of HDL-C (p<0.001) than control subjects. Total cholesterol and LDL-C levels did not significantly differ between the two groups (p> 0.05) (Table 3).

Higher levels of FBS (p<0.001), insulin (p= 0.004) and HOMA-IR (p<0.001)were observed in subjects with MetS as compared with control group (Table 3).

Table 1. Demographic characteristics of the studied groups

Variables		Patients (n= 43)	Controls (n=43)	p-value
Age a (year)		38.77± 6.51	35.91± 5.71	0.01c
Marital status[b]	Married	93 (41)	79.1 (39)	0.40d
	Single	7 (2)	20.9 (4)	
Literacy level[b]	Under diploma	20 (46.5)	18 (41.9)	0.27d
	Diploma	19 (44.2)	17 (39.5)	
	College	4 (9.3)	8 (18.6)	
Physical activity[b]	Low	12 (27.9)	20 (46.5)	0.08d
	Moderate	31 (72.1)	23 (53.5)	
Stress level[b]	Mild	11 (25.6)	7 (16.3)	0.25d
	Moderate	24 (55.8)	25 (58.1)	
	Severe	8 (18.6)	11 (25.6)	

[a] expressed as mean ± SD, [b] expressed as frequency(percent), [c] p-value was reported based on independent samples t-test, [d] p-value was reported based on Chi-squared test.

Table 2. Anthropometric parameters and blood pressure of the studied groups

Variables	Patients (n= 43)	Controls (n=43)	p-value
Waist circumference (cm)	109.73±10.61	106.00±10.57	0.11
Waist/hip ratio	0.96±0.06	0.93±0.07	0.01
Systole pressure (mmHg)	120.23±20.70	109.30±17.95	0.01
Diastole pressure (mmHg)	79.19±12.30	72.74±10.01	0.01

Data were expressed as mean±SD, p-values were reported based on independent samples t-test.

Serum levels and association of obestatin and NGF

As shown in Table 3, there were no statistically significant difference in serum levels of obestatin between MetS and control groups (4.18±0.33 vs. 3.96 ±0.64ng/ml, p= 0.08). Subjects with MetS had significantly lower level of NGF (292.37±59.16pg/ml) than subjects in control group (327.46±52.33pg/ml) (p=0.02) (Table 3). There was significant and positive association between serum obestatin and NGF levels in MetSgroup (β =0.41, p= 0.01) (Table 4).

Correlation of measured variables with serum levels of NGF

As shown in Table 4, in MetS group, there was significant negative association between serum levels of NGF and serum TG in both univariate and multivariate regression models (β = -0.41, p = 0.01). In control group, there was also significant negative association between serum levels of NGF and serum TG in univariate regression model (β = -0.33, p = 0.04) but In multivariate analysis after adjusting for age, this association was attenuated (β = -0.35, p = 0.05). There was no significant association between serum levels of NGF with other components of lipid profile, FBS, insulin, HOMA-IR, blood pressure, waist circumference and WHR in the two groups (Table 4).

Correlation of measured variables with serum levels of obestatin

In MetS group, there was significant negative association between serum TG and serum obestatin both in univariate and multivariate analysis(β = -0.38, p = 0.02) (Table 5).There was no significant association between serum levels of obestatin with other measured variables in MetS group (Table 5). There was no significant association between serum levels of NGF with components of lipid profile, FBS, insulin, HOMA-IR, blood pressure, waist circumference and WHR in control group (Table 5).

Table3. Serum metabolic parameters of the studied groups

Variables	Patients (n = 43)	Controls (n = 43)	p-value
NGF (pg/ml)	292.37±59.16	327.46±52.33	0.02
Obestatin(ng/ml)	3.96±0.64	4.18±0.33	0.08
TC (mg/dl)	200.74±40.30	189.21±32.04	0.15
TG (mg/dl)	163.15±63.46	101.91±39.69	<0.001
LDL-C (mg/dl)	127.40±30.35	117.98±27.14	0.13
HDL-C (mg/dl)	41.51±8.88	52.65±8.96	<0.001
FBS (mg/dl)	106.31±9.02	95.60±6.18	<0.001
Insulin (mU/L)	26.24±6.09	22.52±5.50	0.004
HOMA-IR	6.92±1.68	5.29±1.37	<0.001

Data were expressed as mean±SD,p-values are based on independent samples t-test.

Table 4 Association of measured variables with serum levels of NGF based on univariate and multivariate linear regression

	Patients				Controls			
	Unadjusted		Adjusted		Unadjusted		Adjusted	
Variables	**Beta**	**p-value**	**Beta**	**p-value[a]**	**Beta**	**p-value**	**Beta**	**p-value[a]**
Obestatin	0.42	0.01	0.41	0.01	0.30	0.06	0.29	0.08
WC	-0.01	0.96	0.05	0.79	0.25	0.13	0.26	0.11
WHR	0.05	0.75	-0.19	0.44	0.37	0.18	-0.16	0.31
SP	0.05	0.75	0.11	0.53	-0.18	0.28	-0.16	0.36
DP	0.04	0.83	0.05	0.75	-0.05	0.76	-0.04	0.81
TC	-0.19	0.23	-0.17	0.25	-0.23	0.16	-0.22	0.18
TG	-0.40	0.01	-0.41	0.01	-0.33	0.04	-0.35	0.05
LDL-C	-0.05	0.76	0.04	0.82	-0.16	0.31	-0.17	0.30
HDL-C	-0.20	0.19	-0.19	0.28	-0.14	0.38	-0.12	0.49
FBS	0.16	0.32	0.19	0.25	0.19	0.26	0.20	0.23
Insulin	0.04	0.82	0.04	0.82	-0.21	0.19	-0.23	0.17
HOMA-IR	0.09	0.59	0.10	0.53	-0.16	0.34	-0.16	0.32

[a] *Data were adjusted for age.*

Table 5 Association of measured variables with serum levels of obestatin based on univariate and multivariate linear regression

	Patients				Controls			
	Unadjusted		Adjusted		Unadjusted		Adjusted	
Variables	Beta	p-value	Beta	p-value a	Beta	p-value	Beta	p-value a
WC	0.08	0.61	0.12	0.48	-0.02	0.89	-0.01	0.95
WHR	-0.02	0.93	0.02	0.93	-0.04	0.79	-0.03	0.88
SP	0.10	0.52	0.14	0.41	-0.17	0.30	-0.13	0.46
DP	0.05	0.76	0.06	0.73	-0.25	0.13	-0.20	0.24
TC	-0.19	0.22	-0.19	0.25	-0.07	0.66	-0.06	0.73
TG	-0.38	0.02	-0.38	0.02	-0.09	0.57	-0.04	0.83
LDL-C	-0.09	0.57	-0.09	0.59	-0.001	0.99	-0.01	0.94
HDL-C	0.02	0.93	0.05	0.79	-0.29	0.07	-0.26	0.14
FBS	0.004	0.98	0.02	0.89	-0.11	0.50	-0.09	0.58
Insulin	-0.04	0.79	-0.05	0.78	-0.90	0.57	-0.11	0.51
HOMA-IR	-0.01	0.97	0.001	0.99	0.04	0.46	-0.13	0.42

a Data were adjusted for age.

Discussion

In the present study, serum obestatin level was non-significantly lower inMetSgroup as compared to control group.The evidence in this area is remarkably scarce. In a sole previous related study, lower levels of obestatin has been reported inMetS individuals than controls (21). Reduced obestatin levels have also been reported in patients suffering fromsome othermetabolic disorders including obesity and diabetes (14, 22, 23).

In the current study, lower levels of NGF were observed in subjects with MetS compared to control subjects. Hyponeurotrophinemia have already been defined as a characteristics of patients with the MetS (24,25).In contrary,there are some other evidences which indicating higher levels of the neurotrophin in patients with MetS (26,27). Recently, a neurotrophic hypothesis suggests that neurotrophins have a different role in the early or late stage of MetS,whereas a hyponeurotrophinemia appears during the developed stage (28).Hristova*et al.* suggested that neurotrophin levels are high in early stage of MetS in order to compensate and attenuate emerging inflammatory events, but when MetS's criteria are arisen, concentration of neurotrophins begin to reduce because of proinflammatory cytokines effects on the neurotrophins (3).

A novel finding of the current study is determination of a marked positive association betweenserum levels of NGF and obestatin.There is no report to address the relationship between the two peptides. The exact mechanisms by which the two peptidesinteract to each other could not be explained by limited data of the current study. However, comparable functions have been suggested for them in different investigations, separately.For instance, both of the peptides exhibit ananorexigenic effect and treatment of animals with NGF or obestatin leads to reduction in food intake (29,30).Therefore, it appears that they perform in same biologic and regulatory pathway and production of one of them induces the production of the other one. More studies need to elucidate the association between the two peptides.

We found a significant negative correlation between serum NGF and obestatin with TG level. Very few reports exist in the scientific database about NGF effect on lipid profile and mostly have focused onthe involvement of NGF on lipid metabolism.Ng *et al.* in a study on rodents has reported that NGF displays antilipolytic and lipogenic activities in isolated rat adipocytes (31).Colitti*et al.* have also demonstrated the localization of NGF and BDNF along the plasma membrane and cytoplasm of adipocytes and described that these neurotrophins could be related to lipogenesis and lipolysis (32). But about obestatin, the finding is in agreement with previous reports. Agnew *et al.*in a study on animalsobserved a 40% reduction in plasma triglyceridesfollowing a 14-day infusion of rats with 50 nmol/kg/day of obestatin or itsanalog (33).Ghada*et al.*in a study on rats, again, demonstrated increased levels of TG and decreased levels of obestatinin obese or diabetic rats compared with controls.Theyrevealed that plasma obestatin was negatively correlated with TG level (34). Nagaraj*et al.* reported that in adult male mice 8-day obestatintreatment led to 22% decreased TGlevels (35). A later study by the same groupreporteda 32% reduction in plasma TG following 8-day-obestatintreatmentinrodent (36).The mechanisms by which obestatin might exert the TG lowering action is not clearly understoodyet. But, there is substantive evidence supporting obestatin influences TG storage, metabolism and regulates lipolysis in mature adipocytes in rodents (37,38).Wojciechowicz*et al.*evaluatedthe effect of obestatin on lipid accumulation, pre-adipocyte differentiation, lipolysis in rat primary pre-adipocytes. They showed that obestatin enhances lipid accumulation and increases the expression of surrogate markers of pre-adipocyte differentiation. They also revealed that at the early stage of differentiation, obestatin suppressesbut at the late stage of adipogenesisit stimulates lipolysis (37).Taken together, we are thinking, again, that NGF and obestatinact in a same functional pathway and modulate circulating TG level byaffecting adipose tissue metabolism.

In conclusion, NGF and obestatin (partially) levels were lower in MetS individuals. Both the peptides had inverse association with circulating TG and directly correlated together. The results suggest that they might execute in same biologic and regulatory pathway and modulate TG level in serum.

References

1. Resnick HE, Jones K, Ruotolo G, Jain AK, Henderson J, Lu W, et al. Insulin resistance, the metabolic syndrome,

and risk of incident cardiovascular disease in nondiabetic American Indians the Strong Heart Study. Diabetes Care 2003;26:861–7.
2. Gupta A, Gupta R, Sarna M, Rastogi S, Gupta VP, Kothari K. Prevalence of diabetes, impaired fasting glucose and insulin resistance syndrome in an urban Indian population. Diabetes Res Clin Pract 2003;61:69–76.
3. Hristova MG. Metabolic syndrome–From the neurotrophic hypothesis to a theory. Med Hypotheses 2013;81:627–34.
4. Grundy SM. Metabolic syndrome scientific statement by the american heart association and the national heart, lung, and blood institute. Arterioscler Thromb Vasc Biol 2005;25:2243–4.
5. Grundy SM, Cleeman JI, Daniels SR, Donato KA, Eckel RH, Franklin BA, et al. Diagnosis and management of the metabolic syndrome an American Heart Association/National Heart, Lung, and Blood Institute scientific statement. Circulation 2005;112:2735–52.
6. Kerala HIN. ORIGINAL ARTICLE 2014;3:5534–40. doi:10.14260/jemds/2014/2625.
7. Grundy SM, Brewer HB, Cleeman JI, Smith SC, Lenfant C. Definition of metabolic syndrome report of the National Heart, Lung, and Blood Institute/American Heart Association Conference on scientific issues related to definition. Circulation 2004;109:433–8.
8. Gnacinska M, Malgorzewicz S, Stojek M, Lysiak-Szydlowska W, Sworczak K. Role of adipokines in complications related to obesity. A review. Adv Med Sci 2009;54:150.
9. Tore F, Tonchev AB, Fiore M, Tunçel N, Atanassova P, Aloe L, et al. From adipose tissue protein secretion to adipopharmacology of disease. Immunol Endocr Metab Agents Med Chem (Formerly Curr Med Chem Endocr Metab Agents) 2007;7:149–55.
10. Granata R, Gallo D, Luque RM, Baragli A, Scarlatti F, Grande C, et al. Obestatin regulates adipocyte function and protects against diet-induced insulin resistance and inflammation. FASEB J 2012;26:3393–411.
11. Li J-B, Asakawa A, Cheng K, Li Y, Chaolu H, Tsai M, et al. Biological effects of obestatin. Endocrine 2011;39:205–11.
12. Seim I, Walpole C, Amorim L, Josh P, Herington A, Chopin L. The expanding roles of the ghrelin-gene derived peptide obestatin in health and disease. Mol Cell Endocrinol 2011;340:111–7.
13. Zamrazilová H, Hainer V, Sedlácková D, Papezova H, Kunesova M, Bellisle F, et al. Plasma obestatin levels in normal weight, obese and anorectic women. Physiol Res 2008;57:S49.
14. Nakahara T, Harada T, Yasuhara D, Shimada N, Amitani H, Sakoguchi T, et al. Plasma obestatin concentrations are negatively correlated with body mass index, insulin resistance index, and plasma leptin concentrations in obesity and anorexia nervosa. Biol Psychiatry 2008;64:252–5.
15. Qi X, Li L, Yang G, Liu J, Li K, Tang Y, et al. Circulating obestatin levels in normal subjects and in patients with impaired glucose regulation and type 2 diabetes mellitus. Clin Endocrinol (Oxf) 2007;66:593–7.
16. Sofroniew M V, Howe CL, Mobley WC. Nerve growth factor signaling, neuroprotection, and neural repair. Annu Rev Neurosci 2001;24:1217–81.
17. Chaldakov GN, Fiore M, Hristova MG, Aloe L. Metabotrophic potential of neurotrophins: implication in obesity and related diseases? Med Sci Monit 2003;9:HY19–21.
18. Silva-Batista C, Urso RP, Silva AEL, Bertuzzi R. Associations Between Fitness Tests and the International Physical Activity Questionnaire—Short Form in Healthy Men. J Strength Cond Res 2013;27:3481–7.
19. Holmes TH, Rahe RH. The social readjustment rating scale. J Psychosom Res 1967;11:213–8.
20. Friedwald WT, Leve RI, Fredrichson DS. Estimation of concentration of low density lipoproteins separated by three different methods. Clin Chem 1972;18:499–502.
21. Cui A, Gai N, Zhang X, Jia K, Yang Y, Song Z. Decreased serum obestatin consequent upon TRIB3 Q84R polymorphism exacerbates carotid atherosclerosis in subjects with metabolic syndrome. Diabetol Metab Syndr 2012;4:1.
22. Anderwald-Stadler M, Krebs M, Promintzer M, Mandl M, Bischof MG, Nowotny P, et al. Plasma obestatin is lower at fasting and not suppressed by insulin in insulin-resistant humans. Am J Physiol Metab 2007;293:E1393–8.
23. Lippl F, Erdmann J, Lichter N, Tholl S, Wagenpfeil S, Adam O, et al. Relation of plasma obestatin levels to bmi, gender, age and insulin. Horm Metab Res Horm Stoffwechselforschung= Horm Metab 2008;40:806–12.
24. Chaldakov GN, Fiore M, Stankulov IS, Manni L, Hristova MG, Antonelli A, et al. Neurotrophin presence in human coronary atherosclerosis and metabolic syndrome: a role for NGF and BDNF in cardiovascular disease? Prog Brain Res 2004;146:279–89.
25. Manni L, Nikolova V, Vyagova D, Chaldakov GN, Aloe L. Reduced plasma levels of NGF and BDNF in patients with acute coronary syndromes. Int J Cardiol 2005;102:169–71.
26. Bulló M, Peeraully MR, Trayhurn P, Folch J, Salas-Salvadó J. Circulating nerve growth factor levels in relation to obesity and the metabolic syndrome in women. Eur J Endocrinol 2007;157:303–10.
27. Atanassova P, Hrischev P, Orbetzova M, Nikolov P, Nikolova J, Georgieva E. Expression of Leptin, NGF and Adiponectin in Metabolic Syndrome. Folia Biol (Praha) 2014;62:301–6.
28. Hristova M, Aloe L. Metabolic syndrome–neurotrophic hypothesis. Med Hypotheses 2006;66:545–9.
29. Brunetti L, Di Nisio C, Recinella L, Orlando G, Ferrante C, Chiavaroli A, et al. Obestatin inhibits dopamine release in rat hypothalamus. Eur J Pharmacol 2010;641:142–7.
30. Williams LR. Hypophagia is induced by intracerebroventricular administration of nerve growth factor. Exp Neurol 1991;113:31–7.
31. Ng TB, Wong CM. Epidermal and nerve growth factors manifest antilipolytic and lipogenic activities in isolated rat adipocytes. Comp Biochem Physiol Part B Comp Biochem 1985;81:687–9.
32. Colitti M, Loor JJ, Stefanon B. Expression of NGF, BDNF

and their receptors in subcutaneous adipose tissue of lactating cows. Res Vet Sci 2015;102:196–9.
33. Agnew A, Calderwood D, Chevallier OP, Greer B, Grieve DJ, Green BD. Chronic treatment with a stable obestatin analog significantly alters plasma triglyceride levels but fails to influence food intake; fluid intake; body weight; or body composition in rats. Peptides 2011;32:755–62.
34. Fard GMA, Madi NM, Zade AAA. Circulating obestatin level in diabetic and obese rats. Tanta Med J 2014;42:1.
35. Nagaraj S, Peddha MS, Manjappara U V. Fragment analogs as better mimics of obestatin. Regul Pept 2009;158:143–8.
36. Nagaraj S, Peddha MS, Manjappara U V. Fragments of obestatin as modulators of feed intake, circulating lipids, and stored fat. Biochem Biophys Res Commun 2008;366:731–7.
37. Wojciechowicz T, Skrzypski M, Kołodziejski PA, Szczepankiewicz D, Pruszyńska-Oszmałek E, Kaczmarek P, et al. Obestatin stimulates differentiation and regulates lipolysis and leptin secretion in rat preadipocytes. Mol Med Rep 2015;12:8169–75.
38. Miegueu P, St Pierre D, Broglio F, Cianflone K. Effect of desacyl ghrelin, obestatin and related peptides on triglyceride storage, metabolism and GHSR signaling in 3T3β-L1 adipocytes. J Cell Biochem 2011;112:704–14.

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