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The Effect of Short-Term Antidepressant Treatment on Serum Levels of Nesfatin-1, Nitric Oxide and Ghrelin in Patients with Major Depressive Disorder*

Objective: Recent studies have shown that major depressive disorders (MDD) have important effects on nesfatin-1, nitric oxide (NO) and ghrelin hormones. To understand the possible roles of nesfatin-1, NO and ghrelin on effectiveness of MDD treatment, the effects of short-term antidepressant venlafaxine treatment on these parameters in patients with MDD were evaluated.

Material and Methods: Sixty subjects (20 MDD patients and 40 healthy controls) enrolled in the study. Venlafaxine (225 mg/day for 3 months) was given to patients. Fasting blood samples were taken at onset of the study and at the end of the 3 months. Biochemical parameters, serum ghrelin, nesfatin-1 and NO were analysed.

Results: After 3 months treatment, the patients prognosis, evaluated by the HAM-D scale, improved from 36.6±1 to 15.9±0.6 (P=0.04). Nesfatin-1 (p=0.001) and ghrelin (P=0.001) levels decreased and NO levels increased significantly (P=0.04) after treatment in MDD patients.

Conclusions: Decreased nesfatin-1, and increased NO levels comparing to their normal levels after venlafaxine application could be an important criteria to evaluate patients' response to treatment and also prognosis in addition to the improvement of HAM-D scales.

Key Words: Nesfatin-1, NO, ghrelin, major depressive disorders, venlafaxin.

Major Depressif Hastalarda Kısa Süreli Depresyon Tedavisinin Serum Nesfatin-1, Nitrik Oksit ve Ghrelin Düzeylerine Etkileri

Amaç: Son zamanlarda yapılan çalışmalar majör depresif bozukluğu (MDB) olan hastalarda nesfatin-1, nitrik oksit (NO) ve ghrelin hormonlarının önemli etkilere sahip olduğunu göstermiştir. MDB tedavisinin etkinliğinde nesfatin-1, nitrik oksit ve ghrelinin olası rollerini anlamak için depresif hastalarda bu parametrelerin kısa ve uzun dönem venlafaksin tedavisindeki etkileri değerlendirilmelidir.

Gereç ve Yöntem: Toplam 60 kişi (20 MDB hastası, 40 sağlıklı kontrol) çalışmaya dahil edildi. Venlafaksin (3 ay boyunca günde 225 mg) hastalara verildi. Açlık kan örnekleri çalışmanın başlangıcında ve 3 ayın sonunda alındı. Biyokimyasal parametreler, serum nesfatin-1, nitrik oksit ve ghrelin düzeyleri analiz edildi.

Bulgular: Üç aylık tedavi sonunda hastaların prognozu HAM-D ölçeği ile değerlendirildi, 36.6±1 den 15.9±0.6 (P=0.04)'e doğru iyileşmeler gözlemlendi. MDB hastalarında tedavi sonrasında nesfatin-1 (P=0.001) ve ghrelin (P=0.001) düzeyleri anlamlı oranda azalırken, nitrik oksit düzeyi (P=0.04) anlamlı oranda artmıştır.

Sonuç: Venlafaksin tedavi sonrasında HAM-D ölçeğindeki ilerlemeye ek olarak nesfatin-1'in azalması nitrik oksitin ise artarak normal değerlere doğru düzelmesi tedavi cevabının değerlendirilmesinde önemli bir kriter olabilir.

Anahtar Kelimeler: Nesfatin-1, NO, ghrelin, majör depresif bozukluk, venlafaksin.

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Introduction

Nesfatin-1 is a newly described peptide hormone that reduced food intake and regulate energy metabolism (1). It has been shown that nesfatin-1 has an affect on hypothalamus via melanocortin 3-4 receptors (1, 2). Importantly, the effects of reduced food intake of nesfatin-1 is related with leptin-independent mechanism (1-3). Since its first description, nesfatin-1 has been studied in many field including, diabetes mellitus (4), obesity (5), cardiovascular system abnormalities (2), puberty (6), epilepsy (7) and even in Major Depressive Disorders (MDD) (8).

MDD is a severe psychiatric disorder that characterized by impaired mood and reduced of interest or pleasure in daily activities, and accompanied by weight change, sleep disturbance, fatigue, reduced physical capacity, developing countries,

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MDD prevalence rates are high and increasing continuously. The interesting point is the relationships between nesfatin-1 and MDD. The impaired food intake and energy regulation changes are commonly seen problems in patient with MDD (9). Barim et al (10) have studied the change in ghrelin level, which stimulate food intake in contrast to nesfatin-1, in response to the citolopram treatment. They observed an increase in body weight and decrease ghrelin level following depression treatments.

The nitric oxide (NO) level in MD D patients and its response to treatment is other important point to be clarified in this study. This is because, NO may have an important role in pathophysiology of depression. In addition, it has also been suggested that NO could be a criteria in treatment and following prognosis in depressive patients (11). It should be emphasize that effects of treatment on nesfatin-1 and NO levels has not been studied in patients with MDD. There is one study, performed to show the nesfatin-1 level in patient with MDD (8). However, this study contains only basal high nesfatin-1 values in MDD patients. In this study we concerned the relation between nesfatin-1, NO, ghrelin levels and MDD.

We were purposed to examine the effects of three months of venlafaxine treatment on nesfatin-1, NO and ghrelin levels in patient with MDD to find a criteria used to evaluate the effectiveness of MDD treatment.

Materials and Methods

All procedures were in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans. The study protocol was approved by the local ethics committee, and informed written consent was obtained from each subject at the start of the study. Before the study, each participant underwent medical screening to rule out abnormalities, including medical history, physical examination, hormonal analyses, and cardiovascular risk assessment, which included electrocardiography. It is known that different comorbidity as a chronic inflammation, gastrointestinal or nutritional disorders can significantly affect serum levels of nesfatin-1, ghrelin as well as NO production.

The sample size was determined by being used power analysis. A total of 60 subjects were participated in this study: 20 MDD patients (12 female and 8 male) and 40 control subjects (22 female and 18 male). For the first time MDD diagnosed patients were participated to this study. The MDD patients were chosen using diagnostic and statistical manual of mental disorder-fourth edition (DSM-IV) criteria (12). The patients were participated to Hamilton Depression Rating Scale (HAM-D) before and after the treatment to evaluate the patients' response to applied treatment (13).

They were randomly assigned to receive oral treatment with Venlafaxine (Efexor, Wyeth) 225 mg/day, the dosage that has been shown to be the most effective in resistive MDD treatment (14). The body mass index

and age were similar in control group and MDD group. The control group was chosen from healthy subjects with taking no alcohol or smoking or taking any medication or severe illness including metabolic and psychiatric. Blood samples were drawn at study entry and after 3 months of therapy. After an overnight fast, a 5 ml venous blood sample was obtained between 8:00 AM and 10:00 AM, always at approximately the same time in the morning to avoid reductions in serum hormones levels over time. Blood samples were taken into the tube contaminated with the aprotinin to prevent desaturation of proteins. The samples were separated by centrifugation (4500 rpm for 5 minutes at 4°C) and stored at -20°C and analyzed immediately at the end of study (Hettich, Zentrifugen Universal 32 R, Germany).

The biochemical parameters (including cholesterol, triglyceride, HDL, LDL) were analyzed using OLYMPUS AU400 (Japan). Acylated ghrelin (SPI BIO CATT NO: A05119), des-acylated ghrelin (SPI BIO CATT NO: A05119) (SPI BIO, Bertin Pharma Biotech, Montignyle Brettonneux, France) and nesfatin-1 (RAY BIO CATT NO: EIA-NES-1) were analyzed using ELISA method. Total ghrelin was obtained from the sum of acylated ghrelin and des-acylated ghrelin. All analysis performed in the same experimental set in a blind fashion. NO determination was made using spectrophotometric Griess reaction. Intra- and interassay coefficients of variation were 8.1% and 8.3% for acylated ghrelin and 3.2% and 3.8% for desacylated ghrelin, respectively. Care was taken to perform the measurements with kits from the same company, as different commercial ghrelin kits measure the same biological sample differently. Intra- and interassay coefficients of variation were below %10 and below 15% for nesfatin-1, respectively. Nesfatin-1 values were linear between 1-100 ng/mL.

Values are expressed as means±SEM. The Kolmogorov-Smirnov Z test showed that the data were normally distributed Therefore. within-group comparisons of data at baseline and study end were assessed using the Paired t test. Unpaired t test used to assess between-group data and P<0.05 was considered significant.

Results

The anthropometric and biochemical characteristics of the patients and the control subjects are given in Table. Nesfatin-1 (before and after treatment) and control values are shown in Figure. After 3 month treatment, HAM-D scale values decreased from 36.6±1 to 15.9±0.6 (P=0.04). Before treatment, nesfatin-1 level was found to be significantly higher (P=0.001) in patients compared to the control group: 82.2±2.1 ng/mL and 11.4±4.7 ng/mL, respectively (Figure). After 3 months treatment, nesfatin-1 decreased to 34.8±6.5 ng/mL (Figure 1) (P=0.001). Despite the significant decrease in nesfatin-1 level, it was still significantly higher than the control level (P=0.006) (Figure 1).

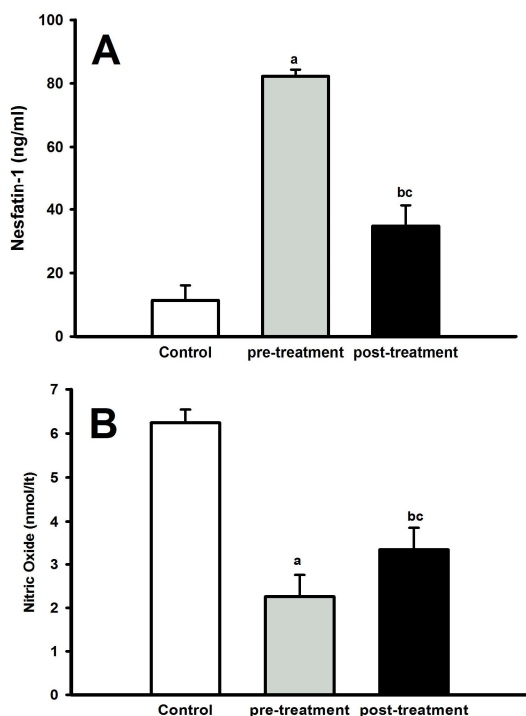


Figure 1. Nesfatin-1 (A) and nitric oxide (B) values (mean±SEM) for control group, for MDD group pre-treatment and post-treatment (a: significance between control and pre-treatment group. b: significance between pre and post treatment groups. c: significance between post-treatment and control groups).

Table 1. The anthropometric and biochemical characteristics of the subjects (age, weight and body mass index, BMI), total ghrelin and biochemical parameters in patients with MDD before and after treatment and control subjects

Parameters	Control (n=40)	Patients (n=20)	
		Pre-treatment	Post-treatment
Age (yr)	39.8±2.0	38.6±3.3	-
Weight (kg)	71.7±2.5	69.0±2.9	72.4±2.9 ^b
BMI (kg/m ²)	27.0±1.1	26.3±0.9	27.6±0.8 ^b
Total Ghrelin (pg/mL)	316.9±49	40.6±7.7 ^a	31.8±4.1 ^{bc}
HDL (mg/dL)	64.2±3.4	55±2.4	60±2.8 ^b
LDL (mg/dL)	120.6±4.7	127±8.2	148±11 ^{bc}
Triglyceride (mg/dL)	123.4±10.3	98.3±9	150.5±13.3 ^b
Cholesterol (mg/dL)	189±5.6	204.1±10.7	228.2±13.6 ^{bc}

Values are mean±SEM

^a: Significance between control and pre-treatment group (P<0.05)

^b: Significance between pre and post-treatment groups (p<0.05)

^c: Significance between post-treatment and control groups (P<0.05)

NO for the control, before and after treatment are shown in Figure. The NO level was markedly low (P=0.001) in MDD patients group before treatment compared to control values: 2.2±0.5 nmol/L and 6.2±0.3 nmol/L, respectively (Figure 1). Importantly, after treatment, NO level increased to 3.3±0.5 nmol/L (P=0.04) (Figure). However, NO was still significantly lower than the control group level (P=0.001) (Figure)

In MDD patients group, before treatment, total ghrelin levels was markedly lower than the control group: 40.6±7.7 pg/ml vs 316.9±49.7 pg/ml, respectively (P=0.001) and it decreased significantly to 31.8±4.1 pg/ml, after treatment (P=0.001) (Table 1).

Discussion

We examined the effects of three months venlafaxine treatment on nesfatin-1 level in MDD patients for the first time. Venlafaxine is more effective antidepressive drug especially in resistive depressive patient's treatment (14). As shown in a previous study, nesfatin-1 level was found to be markedly high in MDD patients (8). In this study, we clearly showed that nesfatin-1 decreased significantly after treatment. However, it should be strictly emphasizes that, this post-treatment nesfatin-1 value is still higher than the normal subjects' levels (Figure). This may be related with the duration of the MDD treatment. To clarify this topic, further studies, concerning the effects of long treatment period on nesfatin-1 level in MDD patients, are necessary. It is also important to emphasize that after three months treatment the patients depression status as classified by HAM-D scale improved but not to normal levels (8). Because, one may think that the prognosis of the patients could be related the level of nesfatin-1 (4). In addition, high nesfatin-1 level has been reported in generalized epilepsy (7) and in panic disorders compared to healthy subject (15).

The close relationship between nesfatin-1 and central nervous system hormones has been reported (16). Importantly, the interaction of nesfatin-1 and central nervous system hormones has an important role in etiology of depression (8). A markedly high nesfatin-1 level found in raphe nucleus neurons and locus coeruleus neurons of stressed rat compared to non-stressed rat (17). This is important that locus coeruleus neurons have a powerful regulatory role on many mental functions known to be dysregulated in MDD (18). In addition, raphe nucleus, which is the main source of serotonergic innervations in the brain, is the primary neurotransmitter accused for some metabolic and psychiatric changes including, appetite, energy, sleep, mood, libido and cognitive function changes in MDD (19).

It is suggested that an increase in nesfatin-1 level may result an increase in anxiety via affecting melanocortin system (20). In clinical medicine, nesfatin-1 level could be important criteria to describe the patient prognoses and effectiveness of treatments. In previous study performed in epileptic patients, significant decreases in nesfatin-1 level reported following anti antiepileptic treatment (7).

It has been suggested that ghrelin and nesfatin-1 could be closely related in ethiopathologies of psychiatric diseases in addition to their effects on appetite and food intake regulation (7, 10). It is well known that ghrelin and nesfatin-1 is adversely affecting hormones (21). The anorectic effect of nesfatin-1 has been shown after directly administering it into the brain of rodents (1). In literature, it is well known that ghrelin increases food intake in normal condition (3, 10).

The results of a study performed in rodent suggested that ghrelin has antidepressive and depressionogenic effects (22). In this study, basal ghrelin level of MDD patients was markedly lower than the control group, which could be the results of the depression (7, 10). The lower ghrelin level before and after treatment could be related with the low antioxidant capacity, which is known to be reduced in depressive patients. Importantly, it has been shown that ghrelin has important effects on neuroprotection (23).

Despite the decrease in ghrelin level after treatment, we have observed increase in body weight (Table 1). It might be expected decrease ghrelin level results decrease in body weight rather than increases. The increased body weight during treatment period may be related with the reduced nesfatin-1 level or direct effects of drug affecting metabolic rate (24). In addition, it is also emphasized that NO is an important regulatory factor in food intake (25). It is known that basal metabolic rate is one of the important factors in body weight regulation. The main reasons for weight gain in MDD patients after treatment are increased caloric intake, reduced energy expenditure or a combination of both. Venlafaxine may

have important effects on basal metabolic rate and physical activity (26).

In this study, serum NO levels, before and after treatment, were measured and it was found to be markedly low in depressive patients. Importantly, NO level increased significantly after treatment, but it was still lower than the control levels. In depressive patients, end-production of NO metabolism has been shown to be high in some studies (27). In contrast, low NO levels in depressive patients compared to normal subjects has been reported (28). Interestingly, increased NO levels results a suicide attempt especially in mood disorders and schizophrenia patients (29). Thus, evaluation of NO level could be important criteria in depressive patients prognosis (27). However, the clear and satisfactory explanation in NO level and depressive patients has not been documented yet.

In this study, three months of venlafaxine treatment associated with significant improvements in nesfatin-1 and NO levels compared to their basal values over a three months period. In this regard, nesfatin-1 and NO could be useful markers to evaluate the effectiveness of treatment and prognosis of MDD in addition to the other parameters. However, further studies with a large number of sample size and with a longer treatment duration are required to understand the factors affecting nesfatin-1, ghrelin and NO metabolism and their roles in the etiology and also in prognosis of MDD.

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