Low prevalence of obesity in Behçet’s disease is associated with high obestatin level

Süleyman Serdar Koca1, Murat Kara2, Metin Özgen3, Ramazan Dayanan4, Caner Feyzi Demir5, Kader Aksoy6, Nevin İlhan7, Emir Dönder4, Ahmet Işık1

Abstract

Objective: Chronic inflammatory diseases are associated with altered body composition. Ghrelin has anti-inflammatory effects, and its level is altered in obesity and inflammatory diseases. The aim of the study was to evaluate the prevalence of obesity and ghrelin and obestatin levels in patients with Behçet’s disease (BD).

Material and Methods: One hundred and forty-three (143) patients with BD and 112 healthy controls (HC) were enrolled. Participants were subdivided according to the body mass index (BMI) as lean (<18.5 kg/m²), normal weight (18.5-24.9 kg/m²), overweight (25-29.9 kg/m²) and obese (≥30 kg/m²). In addition to the routine evaluations (fasting blood glucose, lipid profile, and kidney and liver function tests), serum acylated-ghrelin (AG), unacylated-ghrelin (UAG), total ghrelin (TG) and obestatin levels were analyzed. Student’s t-test and chi-square test were used for statistical analysis.

Results: The prevalence of obesity was relatively lower in the BD group than in the HC group (12.6% vs. 20.5%, p=0.089). Serum ghrelin levels were similar in the BD and HC groups (p>0.05 for all) although the obestatin level was higher in the BD group compared to the HC group (p<0.001). Serum UAG, TG and obestatin levels were lower in obese BD patients (n=18) than non-obese BD patients (p=0.027, p=0.014 and p=0.001, respectively).

Conclusion: The obestatin level was high and the prevalence of obesity was low in the BD group. Moreover, obese BD patients had low obestatin levels. These results suggest that obestatin may protect BD patients from obesity.

Keywords: Behçet’s disease, obesity, ghrelin, obestatin

Introduction

Obesity is a major public health threat since it is a well-established risk factor for the development of cardiovascular and metabolic diseases, such as diabetes, hyperlipidemia and insulin resistance, in the general population (1). Ghrelin, a gastric mucosa-derived peptide, is demonstrated to have various metabolic functions including regulation of energy balance, control of appetite, stimulation of gastric acid secretion, and regulation of gastrointestinal motility (2, 3). The administration of ghrelin increases food intake and body weight along with a reduction in fat utilization (2). Negative correlations between circulating ghrelin levels and body mass index (BMI) are found in humans (3, 4). On the other hand, the ghrelin level is reported to be high in patients with anorexia nervosa (5) and subjects with diet-induced weight loss (4). Obestatin is a peptide hormone derived from proghrelin. It also has a role in regulating food intake and energy expenditures (6). In addition to energy homeostasis, some evidence suggests that ghrelin has direct anti-inflammatory effects (7, 8). Therefore, the ghrelin level has been evaluated in a variety of inflammatory diseases with contradictory results (9-11).

Behçet’s disease (BD) is a chronic relapsing inflammatory disease characterized by mucocutaneous lesions, and it can affect ocular, neurological, and gastrointestinal systems (12, 13). BD has been described by a Turkish dermatologist as a triple symptom complex, i.e., oral aphthosis, genital ulcer, and uveitis. BD has a greater incidence and prevalence in the regions along the ancient Silk Road. Although the etiology of BD is not fully known, immune abnormality is thought to be associated with the development and maintenance of BD (12, 13). The prevalence of obesity in rheumatoid arthritis (RA) and systemic lupus erythematosus (SLE) is reported to be higher (14-19). In RA and SLE, the potential association of obesity with the progression of the diseases has been researched (17-19). However, there is an obvious gap in the literature reporting body composition in BD. In contrast to RA and SLE, the knowledge about body composition and the levels of ghrelin and obestatin in BD is limited (20, 21).

Therefore, the aims of the present study were to evaluate the prevalence of obesity and the levels of ghrelin and obestatin in BD.
Table 1. Demographic characteristics and serum ghrelin and obestatin levels

<table>
<thead>
<tr>
<th></th>
<th>BD (n=143)</th>
<th>HC (n=112)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>37.7±10.9</td>
<td>40.1±13.9</td>
<td>0.139</td>
</tr>
<tr>
<td>Sex (female ratio, %)</td>
<td>57.3</td>
<td>57.1</td>
<td>0.974</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>25.3±4.8</td>
<td>26.3±5.5</td>
<td>0.108</td>
</tr>
<tr>
<td>Diabetes mellitus (n)</td>
<td>8 (5.6%)</td>
<td>9 (8.1%)</td>
<td>0.459</td>
</tr>
<tr>
<td>ESR (mm/h)</td>
<td>16.4±8.7</td>
<td>12.1±4.2</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>CRP (mg/dL)</td>
<td>5.9±6.2</td>
<td>3.2±1.7</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Uric acid (mg/dL)</td>
<td>4.2±1.1</td>
<td>4.5±1.8</td>
<td>0.102</td>
</tr>
<tr>
<td>LDL cholesterol (mg/dL)</td>
<td>124.7±21.5</td>
<td>131.2±43.7</td>
<td>0.121</td>
</tr>
<tr>
<td>Triglyceride (mg/dL)</td>
<td>136.4±54.2</td>
<td>128.7±62.4</td>
<td>0.294</td>
</tr>
<tr>
<td>Acylated ghrelin (pg/mL)</td>
<td>22.6±19.2</td>
<td>24.1±13.6</td>
<td>0.587</td>
</tr>
<tr>
<td>Unacylated ghrelin (pg/mL)</td>
<td>100.1±84.3</td>
<td>97.3±58.9</td>
<td>0.801</td>
</tr>
<tr>
<td>Total ghrelin (pg/mL)</td>
<td>194.3±167.1</td>
<td>181.8±104.2</td>
<td>0.593</td>
</tr>
<tr>
<td>Obestatin (pg/mL)</td>
<td>252.5±86.1</td>
<td>174.1±51.1</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Table 2. Ghrelin and obestatin levels in obese and non-obese subgroups

<table>
<thead>
<tr>
<th></th>
<th>BD group</th>
<th>HC group</th>
<th>p</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>BMI&lt;30</td>
<td>BMI≥30</td>
<td>p</td>
</tr>
<tr>
<td>AG (pg/mL)</td>
<td>18.8(5.6-198.2)</td>
<td>15.9(4.9-40.2)</td>
<td>0.346</td>
</tr>
<tr>
<td>UAG (pg/mL)</td>
<td>87.6(32.1-946.1)</td>
<td>69.2(32.6-130.1)</td>
<td>0.027</td>
</tr>
<tr>
<td>TG (pg/mL)</td>
<td>164.3(41.1-1517.5)</td>
<td>118.3(88.4-179.4)</td>
<td>0.014</td>
</tr>
<tr>
<td>Obestatin (pg/mL)</td>
<td>255(89-602)</td>
<td>198(98-268)</td>
<td>0.001</td>
</tr>
</tbody>
</table>

Material and Methods

Participants

This cross-sectional comparison study included 143 patients with BD. Patients were diagnosed according to the established criteria (22), and they were in the age range of 18-72 years. Age-, sex- and origin-matched (Table 1) 112 healthy subjects were recruited from the staff employed in our hospital and faculty to serve as healthy controls (HC). The protocol of this study was approved by the Institutional Ethics Committee of Firat University, and all the participants gave informed consent before enrolling in the study.

Detailed histories of all the participants were obtained and printed on research forms. Their systemic and rheumatologic examinations were performed by one rheumatologist (SSK). Glucocorticoid and disease-modifying anti-rheumatic drugs usages were also recorded. The pathergy test was performed in all the patients with BD, and 24-48 hours later, the patients were evaluated in terms of papulopustular lesions.

Anthropometric measure of body compositions

In all the participants, the height and weight were measured to determine the BMI. Participants were subdivided according to the body mass index (BMI) as lean (<18.5 kg/m²), normal weight (18.5-24.9 kg/m²), overweight (25-29.9 kg/m²) and obese (≥30 kg/m²) according to the World Health Organization (WHO) guidelines (23).

Determination of disease activities

In the BD group, patients were interpreted as active when they had at least two of the following: genital ulcer, skin lesions, recent eye involvement, recent vascular involvement, recent neurological involvement, active arthritis, positive pathergy test sign in addition to oral ulcer, as well as high erythrocyte sedimentation rate (ESR) and/or C-reactive protein (CRP) levels (10).

Laboratory analysis

Blood samples were drawn from all the participants who had fasted overnight. Subsequently, in addition to the routine laboratory evaluations, the serum samples were stored at -20°C for further measurements of acylated-ghrelin (AG), unacylated-ghrelin (UAG), total ghrelin (TG), and obestatin levels. Serum AG, UAG, TG, and obestatin levels were analyzed using commercially available enzyme-linked immunosorbent assay (ELISA) kits (AG, UAG, and TG kits were purchased from the Bertin Pharma, Paris, France; obestatin kit was purchased from the Wuhan ELIAab Science Co. Ltd; Wuhan, China) by ELISA method. The minimal detectable levels were 5 and 21.5 pg/mL for ghrelin and obestatin, respectively.

Statistical analysis

The Statistical Package for the Social Sciences version 16.0 (SPSS Inc.; Chicago, IL, USA) was used for analysis. Normal distributions were tested with the Kolmogorov-Smirnov test with Lilliefor correction. Quantitative data were presented as mean±standard deviation (S.D.). Parametric data were analyzed using the Student’s t-test. Mann-Whitney U test was performed to compare nonparametric data (obese and non-obese subgroup comparisons) and to compare the skewed data (AG, UAG, TG, and obestatin). Fisher’s exact tests or Pearson’s χ² tests were used to compare categorical variables, and the odds ratio (OR) and the 95% confidence interval (CI) were used for the assessment of risk factors. Correlation analyses were made using the Pearson’s product moment test in both the BD and HC groups. A model of multiple regression analysis (standard linear regression analysis) was constructed with BMI as the dependent variable and the age, usage of any medication, and the levels of AG, UAG, TG and obestatin as the independent variables in the BD group. Moreover, it was analyzed whether the same independent variables predicted the presence of obesity by logistic regression analysis. P values less than 0.05 were considered significant.

Results

The demographics and clinical laboratory data of the BD and HC groups are summarized in Table 1. There was no difference in terms of demographics between the study groups. The mean disease duration was 5.3±6.2 years in the BD group. Mean ages were 37.7±10.9
and 40.1±13.9 years in the BD and HC groups (p=0.139). 57.3% (n=82) of BD patients and 57.1% (n=64) of healthy subjects were female (p=0.974).

The prevalence of obesity was 12.6% in the BD group and 20.5% in the HC group (p=0.089, OR: 0.56, 95% CI: 0.28-1.09, Figure 1).

The serum ghrelin levels were similar in the BD and HC groups (Table 1), although the obestatin level was higher in the BD group compared to the HC group (p<0.001). Serum UAG, TG and obestatin levels were lower in the obese BD patients (n=18) than the non-obese BD patients (Table 2) (p=0.027, p=0.014 and p=0.001, respectively). However, they were not significantly different in the obese and non-obese controls (Table 2).

For the disease activity in the BD group, 67 (46.1%) patients were active while 76 (53.1%) were inactive. ESR (23.9±22.7 vs. 12.8±10.1 mm/hour, p<0.001), CRP level (21.7±37.2 vs. 4.1±3.4 mg/L, p<0.001) and white blood cell count (8.4±3.2 vs. 7.2±2.6 /μL, p=0.022) were higher in the active BD patients compared to the inactive ones. While there was no significant difference between the active and inactive BD patients in terms of serum UAG (111.5±113.4 vs. 19.8±9.2 pg/mL, p=0.130) and obestatin (255.6±100.6 vs. 249.8±71.4 pg/mL, p=0.631) levels, serum AG (25.7±25.9 vs. 19.8±9.2 pg/mL, p=0.065) and TG (230.7±236.5 vs. 163.6±54.1 pg/mL, p=0.068) levels were relatively higher in the active subgroups than the inactive subgroup. 8 (12.6%), 23 (34.3%) and 11 (16.4%) of the active BD patients and 6 (7.9%), 35 (46.1%) and 7 (9.2%) of the inactive BD patients were lean, overweight and obese, respectively (p=0.120). The mean BMI was not significantly different in the active and inactive BD subgroups (25.3±5.3 vs. 25.3±4.4 kg/m², p=0.958). There were no obvious effects on the BMI and body compositions in the BD group.

Prediction of obesity
The BMI was correlated with age (r=0.534, p<0.001) in the HC group. The BMI was correlated with age (r=0.471, p<0.001) and serum obestatin level (r=0.267, p=0.001) in the BD group. The BMI was not correlated with the levels of the acute phase reactants. Multiple regression analysis also showed that only the age (β=0.438, p<0.001) and serum obestatin level (β=0.189, p=0.012) were significant predictors of BMI. Similarly, logistic regression analysis also showed that age (p=0.011, OR: 1.12, 95% CI; 1.03-1.23) and serum obestatin levels (p=0.011, OR: 0.98, 95% CI; 0.96-0.99) were predictors of the presence of obesity in BD.

Discussion
The present study demonstrated that the prevalence of obesity was lower in the BD group. In addition, the serum obestatin level was higher in the BD group than in the healthy subjects, while the circulating ghrelin levels were not different.

Behçet’s disease is a multisystemic vasculitis of unknown etiology characterized by mucocutaneous, ocular, arthritic, and vascular manifestations, and it has a high prevalence all along the ancient Silk Road, from Asia to the Mediterranean basin (12, 13). Although the etiopathogenesis of BD remains uncertain, immunological abnormalities including innate and adaptive immunity in humoral and cellular immunity settings are supposed to be the cornerstone of the pathogenesis of BD (13, 24). A variety of cytokines such as IL-6, IL-17, IL-18, and IL-21 are increased in BD; moreover, the numerous polymorphisms of the cytokine gene including TNF-α, IL-1, IL-12, IL-23, and IFNγ are also associated with the disease (13, 24).

The close associations of obesity with inflammatory status and increased atherosclerotic complication lead to a great interest in the body composition in chronic inflammatory diseases. The prevalence of overweight and obesity patients is common in RA and SLE (14-19). However, in our study, the prevalence of obesity was not higher in the BD group than in the HC group and general population (25). The causes of obesity are associated with sedentary lifestyle, physical inactivity, and glucocorticoid usages in RA and SLE (17). BD may have a milder disease course than RA and SLE. A low percentage of BD patients require the use of glucocorticoid usage. Physical inactivity is observed in a low percentage of patients, or it continues a short time. Moreover, oral ulcers reduce food intake in BD patients. Therefore, the prevalence of obesity is not increased in BD in contrast to other chronic inflammatory diseases.

The impact of body composition on the severity and activity of chronic inflammatory diseases is also ascertained due to the inflammatory nature of obesity, including increased CRP and TNF-α levels. Obese RA patients have been documented to have higher disease activity and worse quality of life than non-obese ones (17). Obesity has also been reported to be associated with increased inflammatory markers and impaired functional capacity in SLE (18). Conversely, Chaiamnuay et al. (19) demonstrated that obesity is associated with fibromyalgia but not with disease activity indices in SLE. In spite of these controversial results in the li-
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Indeed, in our study, while the circu-
lar effect (6, 34). Treatment of rats with
has orexigenic properties, obestatin may have
that is derived from the posttranslational cleav-
age of preproghrelin and released from the
lating obestatin level was high in BD, obese
BD patients had lower serum obestatin levels
compared to the non-obese ones. These re-
results may suggest that high obestatin may pro-
tect the BD patients from obesity.

Obestatin exists in plasma, saliva, and semen
(reviewed in 37). Moreover, it is detected in
various tissues including adipose tissue, gas-
trointestinal tract, muscle, lung, and liver (37).
Obestatin leads to anti-inflammatory and
anti-oxidant actions, in addition to metabolic
effects (38, 39). It has been reported that obestatin decreases IL-1β, TNF-α, and nucle-
ar factor-κB expressions and the activities of malondialdehyde and myeloperoxidase (38, 39).
In hemodialysis patients, Beberashvili et al.
(40) have shown that low obestatin levels are
predictors of mortality, especially due to car-
diovascular diseases. In our study, the obestatin
levels are low in the BD patients with obesity, which is related to accelerated atherosclerosis.

We realize that the present preliminary study
has some limitations. First, the assessment of
obesity was performed using BMI according to
the WHO definition in the present study. BMI
is widely used in clinical practice, and it is easy
perform whereas the evaluation of body fat
may be required for more sophisticated meth-
ods. Moreover, the WHO definition is valid in
the general population. However, BMI may be
inaccurate to adjust body composition, espe-
cially in the situation of cachexia since body
fat can be different even at the same BMI level in
cachexia. It can be a limitation of the study that a more accurate method of detecting the
body fat percentage could be used instead of
BMI. On the other hand, this study is an ob-
servational case-control study. This study design
has advantages in addition to the disadvantag-
es. For instance, a longitudinal follow-up could
improve our results.

In contrast to ghrelin leading to hyperphagia
and obesity, obestatin is known as an anorec-
tic hormone. In BD, the prevalence of obesity
was lower than in the general populations, and
the serum obestatin levels were higher than in
healthy subjects. Moreover, when compared to
the non-obese BD patients, serum obestatin
levels decreased in obese BD patients. These
results suggest that the prevalence of obesity
is not increased in BD and that obestatin may
protect the BD patients from obesity. However,
it has to be tested further in prospective stud-
ies.

Ethics Committee Approval: Ethics committee approv-
al was received for this study from the ethics com-
mitee of Firat University School of Medicine.

Informed Consent: Written informed consent was ob-
tained from patients who participated in this study.

Conflict of Interest: No conflict of interest was de-
cared by the authors.

Financial Disclosure: The authors declared that this
study has received no financial support.

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Obesity is a well-established risk factor for
atherosclerotic cardiovascular diseases in the
general population (1). It has been reported
that weight loss ameliorates several metabo-
lic complications associated with atheroscle-
rotic diseases and improves cardiac functions
(30, 31). Similarly, decreased ghrelin levels are
associated with a variety of metabolic abnor-
malities and atherosclerotic cardiovascular dis-
ases (32). RA and SLE are associated with an
increased prevalence of obesity and decreased
ghrelin levels, and they lead to accelerated ath-
erosclerosis. In contrast to the other chronic
inflammatory diseases, BD does not lead to ac-
celerated atherosclerosis (33). Therefore, it may
be suggested that the decreased prevalence of
obesity and unaltered ghrelin levels may also
protect the BD patients from accelerated ath-
erosclerosis.

In our study, the obestatin levels were high, in
contrast to the ghrelin levels, in BD patients.
Obestatin is a 23-amino acid peptide hormone
that is derived from the posttranslational cleav-
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obestatin suppresses food intake, inhibits je-
junal contraction, and decreases weight gain
(34-36). Indeed, in our study, while the circu-
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