Variations of Serum Leptin and Resistin levels in Healthy non-Diabetic women with different degrees of Obesity

Davoud Gholizadeh1, Mohammad Rahmati-Yamchi1,2, Ahmad Poursadegh Zonouzi3, Amir Maleksabet2, Mohammad Pourhassan Moghaddam4, Saeidghorbian4, Nosratallah Zarghami1

1Department of Clinical Biochemistry, Faculty of Medicine, Tabriz University of Medical Sciences, Tabriz, Iran.
2Department of Medical Biotechnology, Faculty of Advanced Medical Sciences, Tabriz University of Medical Sciences, Tabriz, Iran.
3Biotechnology Research Center, Tabriz University of Medical Sciences, Tabriz, Iran
4Young Researchers and Elite Club, Ahar Branch, Islamic Azad University, Ahar, Iran.

ABSTRACT

The role of leptin and resistin serum concentrations in pathogenesis of obesity is considered an emerging topic issue in clinical biochemistry researches. This study was carried out to evaluate the variation of leptin and resistin serum levels in women with different grades of obesity. A total of 149 non-diabetic women were included in this study: 85 women as obese group, 31 women as a overweight group and 33 women as a normal weight group. The serum of blood samples were separated by centrifuge and plasma concentrations of leptin and resistin were measured by ELISA technique. The serum leptin and resistin levels were significantly higher in obese group compared to normal weight group (42.85±9.46 ng/µl vs. 15.46±5.96 ng/µl; p=0.000 and 1.97±0.56 ng/µl vs. 1.48±0.24ng/µl; p=0.000, respectively). We failed to show a statistically significant correlation between serum leptin and resistin levels and anthropometric indices in each groups. In conclusion the results indicate that the serum leptin and resistin levels elevated in obese group, and obesity could be one of the most important factors in promotion of plasma levels of these proteins. However, further studies on large scale populations may be needed, to better understanding the pathobiology of obesity.

Key words: leptin, resistin, obesity, Body Mass Index

INTRODUCTION

Obesity is a major risk factor for insulin resistance, type 2 diabetes, cardiovascular disease, hypertension, and many other diseases [1, 2]. However, the pathophysiology of obesity is not well understood, it has been widely accepted that obesity is a complicated, multifactorial diseases [3]. Results of studies postulate that genetic (more than one hundred gene loci), endocrine, dietary, lifestyle and psychological factors which contribute to regulation of body weight, as etiology of obesity [4]. Adipose tissue is an endocrine organ that secretes a wide variety of proteins which contribute to regulation of body weight [5-7]. These secreted proteins include hormones, cytokines, neuron related proteins and extracellular matrix proteins which have autocrine, paracrine and endocrine effects on metabolism [8]. Among of these proteins, we have focused on leptin and resistin. Human leptin is an adipose-derived protein hormone encoded by the ob gene that is secreted proportionally to the total amount of fat in the body [9]. Although, Leptin is synthesized predominantly by white adipose tissue, small amounts of leptin are also secreted by cells in the epithelium of intestine, placenta, muscles and brain tissues [10]. Leptin receptors are highly expressed in areas of the hypothalamus known to be important in regulating body weight, as well as in T lymphocytes and vascular endothelial cells. Leptin stimulates a subset of neurons in the hypothalamus to produce peptides that decrease feeding and promote increased energy consumption [11]. Moreover, leptin inhibits hypothalamic neurons that produce peptides promoting feeding and decreased energy consumption [12]. Therefore, Serum concentrations of Leptin determined body fat percentage and body mass index, and it rises in parallel with the increased amount of body fat storage [13-16].
Resistin is a secretory cysteine-rich protein that is encoded by the RETN gene in human. Human resistin synthesized by adipose, heart, lung and intestine tissues contains 108 amino acids but circulating resistin in plasma as a dimeric protein consisting of two 92 amino acid polypeptides because its hydrophobic signal peptide is cleaved before it is secreted [17]. Resistin acts as a cytokine and plays many physiological roles, and seems to suppress insulin ability to stimulate glucose uptake into adipose cells [18-20]. Hence, it may be an important link between obesity and insulin resistance. However, the exact role of resistin in obesity, type 2diabetes and insulin resistance in humans is still unclear and controversial [21].

Results of previous literatures have been indicated that expression of ob and RETN genes in adipose tissue in obese individual higher than healthy individual [22-25]. Therefore, seems there are strong positive correlation of serum leptin and resistin concentrations with obesity. On this basis, the current study was designed to evaluate of leptin and resistin serum level, and also its relationship with body mass index and other anthropometric indices in women with different grades of obesity.

MATERIALS AND METHODS
Subjects
We enrolled 149 non-diabetic women from many various regions within the Northwest of Iran that were all referred by Nutritionists which were divided into normal weight (33 women, aged 24.63±7.21 years), overweight (31 women, aged 27.32±7.86 years) and obese groups (85 women, aged 38.34±42.7 years) according to WHO classification of body mass index (BMI) (Table 1,2). Informed consent was obtained from all participants before enrolment.

Anthropometric indices assessment
Anthropometric parameters (weight, height, Body Mass Index (BMI), hip and waist circumferences and Waist-Hip Ratio (WHR)) were measured for normal weight, overweight and obese groups. To anthropometric indices measurement individuals were in the standing position and wearing light clothing without shoes. Body weight and height were measured in kilograms and in centimeters, respectively. The BMI was calculated as weight in kilograms divided by the square of height (in meters). The hip circumference was taken at the widest area of the hips at the greatest protuberance of the buttocks; the waist circumference was measured at the narrowest part of the waist, between the lowest rib and iliac crest. WHR was also calculated from the ratio of waist circumference in centimeters to hip circumference in centimeters as waist circumference divided by hip circumference.

Analysis of leptin and resistin serum concentrations
Fasting venous blood sample (5 cc from each) was taken from all subjects. The serum of blood samples were separated by centrifuging at 3000 RPM for 10 minutes and stored at -70°C until further analysis. Measurement of fasting plasma glucose concentration was carried out on all the samples by the glucose oxidase method to make sure that the participating subjects were none-diabetic. Quantitative analysis of leptin and resistin serum levels were measured by enzyme-linked immunosorbent assay (ELISA) with a commercially available kit (mediagnost, Germany) according to the manufacturer’s instruction after the serum samples were thawed at room temperature. The sensitivity was 0.2μIU/ml for leptin and 0.012ng/ml for resistin. Intra- and inter-assay coefficients of variation were 10% in both cases for leptin and (5.9% and 7.6%) for resistin in both cases, respectively.

Statistical analysis
All statistical analysis was performed by using the SPSS software version 16 and all continuous variables were expressed as Mean ± SD. One way ANOVA test was used to compare means. Determination of the correlation between obesity parameters (BMI and WHR) with leptin and resistin levels were performed by Pearson’s correlation coefficient. The data with less than 0.05 probabilities regarded as statistical significant.

RESULTS
The comparison means of anthropometric and hormonal parameters of the all studied groups are shown in Table 2. Comparison of the means for serum leptin and resistin concentrations and anthropometric indices (weight, BMI, waist-to-height rate (WHR), waist and hip circumferences) between the all study subjects showed statistically significant difference (P < 0.05). However, comparing total groups regarding height no difference was detected (Table 2).

Our results show that the serum leptin level were statistically significant higher in obese group compared to both groups normal weight and overweight (42.85±9.46 ng/µl vs. 15.46±5.96 ng/µl; p = 0.000 and 42.85±9.46 ng/µl vs. 32.78±17.7; p=0.000, respectively) (Table 2, 3). Resistin level was significantly higher in obese group compared to normal weight group (1.97±0.56ng/µl vs. 1.48±0.24ng/µl; p=0.000) (Table 2, 3).
Resistin concentration was compared between normal weight and overweight groups and no meaningful differences was detected (P < 0.05) (Table 2, 3). Obese group had a statistically significantly higher mean BMI value when compared normal weight and overweight groups (37.10±4.47kg/m² vs. 21.99±2.33kg/m²; p=0.000 and 37.10±4.47kg/m² vs.27.62±1.37kg/m²; p=0.000, respectively) and they also had a significantly higher mean WHR value when compare do normal weight (0.92±0.08 vs. 0.82±0.10; p=0.000) (Table 2, 3). The mean of the WHR was higher in the overweight compared to normal weight, but did not show a statistically significant difference (P < 0.05) (Table 3).

A positive correlation but not statistically significant between lepitin level and BMI (R = 0.060; p = 0.585) as well as with WHR (R = 0.089; p = 0.420) were detected in obese group (Table 4). We observed a positive correlation between resistin level and BMI in obese group (R = 0.160; p = 0.143) (Table 4). Negative correlations between resistin level and WHR (R = -0.101; p = 0.358) was observed in obese group (Table 4). We did not observe any statistically meaningful correlation between compared parameters in within each of groups (Table 4).

In the total study subjects, bivariate correlation analysis indicated highest positive correlation of serum leptin with weight, BMI, waist, hip, WHR and resistin (Table 5). Moreover, a positive correlation of serum resistin with weight, BMI, waist and hip were detected. On evaluating the correlation between serum resistin level and WHR in the all subjects, a positive correlation but not statistically significant was observed (r = 0.148, P = 0.072). The correlation between leptin and resistin level to height was negative in the all subjects (Table 5).

**DISCUSSION**

In current study, we evaluated the leptin and resistin serum levels and their relationship with obesity in non-diabetic women with different degrees of obesity. The results of our study indicate the important role of leptin and resistin in the pathogenesis of obesity.

Our results showed the mean of serum leptin level were up to approximately three times higher in obese subjects than in non-obese individuals (Table 2). Our data was concordant with some other reports in this field [26, 27]. Our results were confirmed by Considine et al, who reported the mean serum leptin concentrations as 31.3ng/ml and 7.5ng/ml for obese and normal weight individuals (P< 0.001) [26]. Moreover, in a study by Considine et al, a strong positive correlation between serum leptin concentrations and the percentage of body fat was detected (r=0.85, P< 0.001) [26]. Matsubara et al evaluated serum leptin concentrations in 353 non-diabetic women and confirmed our data [27]. A study performed by Ho et al clarified that there is evidence to support a relationship of serum leptin concentration with obesity [28]. These result led to the view that the high serum leptin concentration in obese individuals might be associated with degrees of obesity and body fat storage. In mice, leptin decreases appetite and elevates energy expenditure, resulting in weight loss [29, 30]. If the actions of leptin in humans are similar, appetite should decrease and energy consumption elevate in obese individuals. It seems that elevating of serum leptin level in obese subjects due to decreased sensitivity or resistance to leptin. However, body weight is regulated by complicated mechanisms involving multitudinous afferent metabolic and hormonal signals informing the brain about the body’s energy status [31]. Abnormal production or action of any of the afferent messengers may lead to weight gain.

Many parameters related with increasing of the serum leptin concentration in obese individuals, including body fat mass, total percentage of body fat, body mass index and body fat distribution [32]. Our results show that among of anthropometric parameters weight, Body Mass Index (BMI), hip and waist circumferences and Waist-Hip Ratio associated with serum leptin concentrations in obese subjects and the mean of these anthropometric indices increased in parallel with rising serum leptin level (Table 2, 3). However, within the group of obese women, the serum leptin levels showed a weaker positive correlation with BMI, hip circumference, waist circumference and WHR (Table 4). Most of the earlier publications have shown a strong correlation of serum leptin with BMI. Serum leptin levels correlated positively with BMI and were significantly elevated in obese individuals [26]. Ruhl and Everhart have also confirmed these data [33]. However, Rosenbaum et al. found that leptin was not significantly correlated with BMI [34]. Peltz et al reported that the serum leptin concentration determined by percent body fat in Mexican-American women. Moreover, the hip circumference and waist circumference are associated with serum leptin concentration [35]. In a study by Al-Daghi et al, an association between serum leptin concentration and hip circumference was observed [36]. We found that serum resistin level was statistically significantly increased in obese subjects compared to normal weight and overweight groups (Table 3). Therefore; we postulate that the highest resistin concentration in obese women might be associated with degrees of obesity and rises in parallel with the increased amount of body fat storage. There are other studies that confirm our data [37, 38].
Gholizadeh et al

Yamauchi et al reported that the mean serum resistin level was 5.3±0.4 ng/µl and 3.6±0.4 ng/µl for obese and normal weight subjects (P< 0.001) [37]. In a study by Savage et al, resistin mRNA levels in whole adipose tissue were elevated in obese subjects when compared with lean controls [38]. Conversely, Silha et al were unable to observe that the mean resistin level increased in obese subjects [39]. Lee et al reported that the circulating resistin levels are not related with obesity [40]. In sum, it appears that the role and regulation of resistin may be different in normal physiology when compared with disease states such as obesity.

<table>
<thead>
<tr>
<th>Table 1. Classification of individuals according to BMI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Classification</td>
</tr>
<tr>
<td>Normal weight</td>
</tr>
<tr>
<td>Overweight</td>
</tr>
<tr>
<td>Obese</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Table 2: The mean of anthropometric indices and serum leptin and resistin levels in different groups of women *</th>
</tr>
</thead>
<tbody>
<tr>
<td>Parameter</td>
</tr>
<tr>
<td>Age (year)</td>
</tr>
<tr>
<td>Weight (kg)</td>
</tr>
<tr>
<td>Height (cm)</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
</tr>
<tr>
<td>Waist (cm)</td>
</tr>
<tr>
<td>Hip (cm)</td>
</tr>
<tr>
<td>WHR</td>
</tr>
<tr>
<td>Leptin (ng/µl)</td>
</tr>
<tr>
<td>Resistin (ng/µl)</td>
</tr>
</tbody>
</table>

Abbreviations: BMI, Body Mass Index. WHR, Waist Hip Ratio.

a Data are means ± SD
b Evaluated by One way ANOVA test and P ≤ 0.05 is considered significant

<table>
<thead>
<tr>
<th>Table 3: Comparison of the mean anthropometric indices and serum leptin and resistin levels between groups</th>
</tr>
</thead>
<tbody>
<tr>
<td>Parameter</td>
</tr>
<tr>
<td>Age (year)</td>
</tr>
<tr>
<td>Weight (kg)</td>
</tr>
<tr>
<td>Height (cm)</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
</tr>
<tr>
<td>Waist (cm)</td>
</tr>
<tr>
<td>Hip (cm)</td>
</tr>
<tr>
<td>WHR</td>
</tr>
<tr>
<td>Leptin (ng/µl)</td>
</tr>
<tr>
<td>Resistin (ng/µl)</td>
</tr>
</tbody>
</table>

Abbreviations: BMI, Body Mass Index. WHR, Waist Hip Ratio.

a to show groups that compared with each other
b P ≤ 0.05 is considered significant
Within the group of obese women, the serum resistin level showed a weaker positive correlation with BMI, hip circumference and waist circumference (Table 4). A negative correlation was detected between resistin and WHR in obese group (Table 4). In agreement with our data, the weaker positive correlation serum resistin and BMI was previously reported by Yannakoulia et al [41]. However, Savage et al did not find any correlation of resistin level and BMI [38]. Our data were confirmed by Yannakoulia et al, who reported that the serum resistin level was negatively correlated to waist to hip ratio [41].

In conclusion, our results show that the serum leptin and resistin levels were significantly higher in obese compared to normal weight subjects. These results suggest that leptin and resistin may be having a critical role in the pathogenesis of obesity.

ACKNOWLEDGEMENTS

Authors would like to express our sincerest appreciation to Dr. Morteza Ghojazadeh and Sina Raiisi for their great help in this project.

REFERENCES


60. Lee JH, Chan JL, Yannakouris N. (2003). Circulating resistin levels are not associated with obesity or insulin resistance inhumans and are not regulated by fasting leptin administration: cross-sectional and interventional study in normal, insulin resistant, and diabetic subjects. J Clin Endocrinol Metab. 88: 4848-4856.


CITATION OF THIS ARTICLE