Leptin Levels in Women with Uterine Leiomyomas

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ABSTRACT:

BACKGROUND:

Uterine leiomyomas are important, common pathological conditions that impose a major healthcare burden. Leptin, a product of the obese (Ob) gene, is produced predominantly in adipose tissue and expressed in several tissues and organs and in myomas and the surrounding myometrium.

OBJECTIVE:

To assess serum leptin levels in women with and without uterine leiomyomas.

METHODS:

This prospective case- control study was carried out on ninety women in their reproductive age. They were divided into two groups; the study group consisted of 45 women with uterine leiomyomas proved by histopathology after operation, and a control group consisting of 45 healthy women without uterine leiomyomas proved by ultrasound examination. Body mass index (BMI) was calculated in all women. Blood samples were collected from all women in both groups in the proliferative phase of their menstrual cycle. Hemoglobin, blood urea nitrogen, serum creatinine, fasting blood sugar and serum leptin levels were examined. Serum leptin levels were determined by Radioimmunoassay and compared in myomatic and normal women.

RESULTS:

The mean serum leptin levels were significantly lower (p=0.0001) in the myomatic women (7.57± 2.88 ng/mL) than in the normal women (9.56± 1.77 ng/mL). The mean ratios of serum leptin levels / body mass index in the myomatic women (0.267± 0.09) were significantly lower (p=0.0001) than in the normal women (0.34±0.05). In addition, a significant correlation was found between serum leptin levels and body mass index in both the myomatic women (r=0.587, p=0.0001) and the normal women (r=0.608, p=0.0001).

CONCLUSION:

In myomatic women compared to normal women, the lower serum leptin levels observed were independent of body mass index with no significant up-regulation of leptin production in response to increased adiposity.

KEY WORDS: leptin levels, uterine leiomyomas

INTRODUCTION:

Uterine leiomyomas is a rather common disease affecting women over 30 years of age and are the most common benign tumours of the female genital system [1]. Their incidence among women is generally cited as 20 to 25 percent, but has been shown to be as high as 70 to 80 percent in studies using histologic or sonographic examination [2-4] and comprised 27 percent of gynecologic admissions [5]. First identified in 1994, leptin is a 167- amino acid protein encoded by the *ob* gene and produced in white adipose tissue [6,7]. Leptin receptors have been identified in the central nervous system and a wide range of peripheral tissues [8,9]. Markowska et al [10] reported expression of the leptin gene both in myomas and in the surrounding myometrium but not in the myometrium of healthy women. Circulating leptin levels are positively correlated with adiposity [11]. Higher body mass index, estrogen, progesterone, dietary fat intake, and lower physical activity levels are known risk factors for uterine myomas [12-14]. These risk factors may also be involved in the regulation of leptin [15]. Chan et al [16] reported lower serum leptin levels in women with myoma

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uteri than those in the normal women. This study aimed to assess whether or not serum leptin levels differ between women with myomas and women without myomas.

MATERIALS AND METHODS:

This prospective case-control study was conducted at the department of Obstetrics and Gynecology at AL-Yarmouk Teaching Hospital for period of one year extending from July 2010 to July 2011. The study was approved by the Ethics Committee of our hospital and an informed verbal consent was obtained from each woman before participation in the study. This study enrolled a total of ninety women in their reproductive age; 45 women with myoma uteri (case group) and 45 normal women (control group) who were recruited from the outpatient gynecological clinic and/or hospital admissions. Myomas were diagnosed according to patients' clinical presentation and confirmed by ultrasonographic findings and pathology after operation (myomectomy or hysterectomy). The control group included 45 healthy participants with a normal, myoma-free uterine structure, based on ultrasonographic examination, matched for their age, parity and BMI, with the case group. Women enrolled in the study were questioned carefully about their medical, surgical, gynecological history and history of hormonal therapy and women with hypertension, diabetes mellitus, chronic renal insufficiency, or chronic drug usage including hormonal therapy and pregnant women were excluded.

The ultrasonography assessment was performed using Siemens Real Time Scanning ultrasound machine – filtered with a 3.5 MHz convex probe for the abdominal scan. Myomas were defined as hypoechogenic focal solid masses on ultrasound examination.

All participants were scheduled to undergo blood tests in the proliferative phase of the menstrual cycle (2^{nd}-5^{th} day). Blood samples from myomatic women were all collected before operation between (8 - 10 am). Five milliliters of venous blood were collected for the measurement of fasting blood glucose, hemoglobin, blood urea nitrogen (BUN), serum creatinine and serum leptin levels. In both groups, BMI was calculated. For the measurement of leptin level, the collected samples were put in plain tubes without anticoagulant and then centrifuged for 10 minutes and serum was separated and stored at -20°C until analyzed. Serum leptin levels were measured by enzyme-linked immunosorbant assay (ELISA) kit, DRG Instruments GmbH, Germany. The analysis was done in the hormones laboratory of Al-Yarmouk Teaching Hospital.

Statistical analysis:

Analysis of data was performed with SPSS-18 (Statistical Packages for Social Sciences- version 18"PASW" Statistics). Data were presented in simple measures of frequency, percentage, mean, standard deviation, standard error of the mean, and range (minimum-maximum values). The significance of difference of different means (quantitative data) was tested using analysis of variance (ANOVA) for more than two groups and using independent student-t-test for difference between two independent means. Pearson correlation was calculated for the correlation between two quantitative variables with its t-test for testing the significance of correlation to determine the relationship between two quantitative variables, r=correlation coefficient (- inverse, + direct, <0.5 weak, 0.5-<0.7 moderate, =>0.7 strong. Statistical significance was defined as $p<0.05$.

RESULTS:

The study represents a case - control study on a total of ninety Iraqi women, in their reproductive age, 45 women with the diagnosis of uterine leiomyoma (case group), while the other 45 women served as controls. The demographic characteristics for the myomatic women and the control group are shown in table 1. Both groups were demographically comparable regarding their age, parity and body mass index (BMI). The laboratory parameters for the myomatic women versus the control group show no statistically significant difference between the two groups with respect to fasting blood sugar (p= 0.376), blood urea (p= 0.071) and serum creatinine (p= 0.608) levels, however, hemoglobin levels, were significantly lower (p = 0.043) in the myomatic women (10.92± 1.51g/dL) than in the control group (11.42± 0.68 g/dL) with a range of (7.0-13.6) versus (10.2-13.0) respectively as shown in table 2. Serum leptin levels were significantly lower (p = 0.0001) in the myomatic women (7.57± 2.88 ng/mL) than in the normal women (9.56± 1.77 ng/mL) with a range of (0.75-12.91) versus (5.02-12.95) respectively as illustrated in table 2. The correlation between serum leptin levels and the demographic characteristics and laboratory parameters were shown in table 3. We observed a significant correlation between serum leptin levels and the body mass index in both the myomatic women (r =0.58, p=0.0001) and the normal women

(r =0.60, *p*=0.0001) as shown in table 3. No correlation was found between serum leptin levels and age, haemoglobin, fasting blood sugar, blood urea and serum creatinine levels for both myomatic women and control groups. The ratios of serum leptin levels / body mass index for the two groups are illustrated in table 4. The mean ratio in the myomatic women (0.267± 0.09) was significantly lower (*p*=0.0001) than in the normal women (0.34± 0.05). Regarding the correlation between the ratios of serum leptin levels/body mass index and the age, body mass index, haemoglobin, FBS, blood urea and serum creatinine levels, no correlation was observed in both groups as shown in table 5.

Serum leptin levels according to BMI were also compared and the whole study group was divided into non-obese with a BMI<30 and obese with a BMI ≥30. The mean serum leptin levels of the myomatic women were (6.627± 2.421) and (10.166± 2.484) for non-obese and obese participants respectively. The mean serum leptin levels of control group were (8.898± 1.403) and (11.382± 1.375) for non-obese and obese participants respectively. There was statistically significant difference between the two groups (*p*= 0.0001) with a lower leptin levels in the case group as compared with the controls, and leptin levels were significantly higher in patients who had BMI ≥30 (*p*= 0.0001) as illustrated in table 6.

Table 1: The demographic characteristics for women with uterine leiomyoma and normal women.

Characteristics	Cases (n=45)		Controls (n=45)		P value
	Mean±SD	Range	Mean±SD	Range	
Age (years)	38.56±4.92	30-49	38.56±4.92	30-49	-
Parity	1.31±1.73	0-7	1.42±1.71	0-6	0.760
BMI (Kg/m2)	28.02±3.51	19.9-36.0	28.15±3.50	19.8-36.2	0.864
*Significant using Students-t-test for difference between two independent means at 0.05 level of significance. BMI=body mass index					

Table 2: Serum leptin levels and laboratory parameters for women with uterine leiomyoma and normal women.

	Cases (n=45)		Controls (n=45)		P value
	Mean±SD	Range	Mean±SD	Range	
Serum leptin (ng/mL)	7.57± 2.88	0.75-12.91	9.56± 1.77	5.02-12.95	0.0001*
Haemoglobin (g/dL)	10.92±1.51	7.0-13.6	11.42±0.68	10.2-13.0	0.043*
FBS (mg/dL)	86.79±5.43	78.0-98.5	87.78±5.09	75.0-98.0	0.376
Blood urea (mg/dL)	27.39±2.86	22.0-38.0	26.57±2.71	20.5-30.1	0.071
Serum creatinine (mg/dL)	0.79±0.08	0.7-0.9	0.80±0.09	0.6-0.9	0.608
*Significant using Students-t-test for difference between two independent means at 0.05 level of significance					

Table 3: The correlation between serum leptin levels and the demographic characteristics and laboratory parameters.

		Leptin (ng/mL)	
Age (years)	r	0.150	0.197
	P	0.324	0.193
BMI (Kg/m²)	r	0.587**	0.608**
	P	0.0001	0.0001
Haemoglobin (g/dL)	r	-0.118	-0.097
	P	0.440	0.526
FBS (mg/dL)	r	0.289	0.048
	P	0.081	0.755
Blood urea (mg/dL)	r	0.196	0.101
	P	0.197	0.511
Serum creatinine (mg/dL)	r	0.125	0.053
	P	0.413	0.731
*Correlation is significant at the 0.05 level. **Correlation is significant at the 0.01 level.			

Table 4: The ratios of serum leptin levels / body mass index (BMI) in the myomatic and normal women.

	Leptin/BMI ratio	
	Cases	Controls
Number	45	45
Mean	0.267	0.340
Std. Deviation	0.091	0.053
Std. Error of Mean	0.014	0.008
Minimum	0.038	0.198
Maximum	0.414	0.459
Mode	0.038	0.198
Percentiles 3rd	0.038	0.205
5th	0.062	0.225
25th	0.216	0.315
50th (Median)	0.279	0.341
75th	0.335	0.376
95th	0.399	0.443
97th	0.411	0.457
P value	0.0001*	
*Significant using Student-t-test for two independent means at 0.05 level of significance.		

Table 5: The correlation between serum leptin levels/Body mass index ratio and the demographic characteristics and laboratory parameters for myomatic women and controls

		Leptin/BMI ratio	
Age (years)	r	0.044	0.006
	P	0.773	0.969
BMI (Kg/m²)	r	0.285	0.125
	P	0.058	0.414
Haemoglobin (g/dL)	r	-0.001	-0.025
	P	0.995	0.869
FBS (mg/dL)	r	0.309	-0.014
	P	0.055	0.928
Blood urea (mg/dL)	r	0.215	0.063
	P	0.156	0.681
Serum creatinine (mg/dL)	r	0.216	0.013
	P	0.153	0.934
*Correlation is significant at the 0.05 level. **Correlation is significant at the 0.01 level.			

Table 6: Leptin levels according to body mass index (BMI)

		Leptin (ng/mL) (Mean±SD)		P value
		Cases	Controls	
BMI (Kg/m2)	Non-obese (<30.0)	6.627±2.421	8.898±1.403	0.0001*
	Obese (≥30.0)	10.166±2.484	11.382±1.375	0.0001*
P value		0.0001*	0.0001*	
*Significant using ANOVA test of t-test for two independent means at 0.05 level of significance.				

DISCUSSION:

Uterine leiomyomas are the most common pelvic tumors leading to hysterectomy [17]. Leptin is a protein encoded by the *ob gene* and is produced mainly by adipose tissue and appears to play an important role in reproductive system [6]. Risk factors for uterine leiomyoma (nutritional and hormonal) may be involved in the regulation of leptin production and secretion [15,18].

In the current study, serum leptin levels were significantly lower in women with myomas than in those without myomas. Our findings were in agreement with those reported by Chan et al [16], but disagree with Dingiloglu et al [19] who observed higher serum leptin levels in the myomatic women than in the normal women with no statistically significant difference.

Women with myomas often suffer from hypermenorrhea and iron deficiency anemia, which was also present in this study. Togo et al [20] reported that serum leptin levels were negatively correlated to hemoglobin levels. In our study, hemoglobin levels were significantly lower in the myomatic women than in normal women. Similar to our findings, Chan et al [16] reported a

significantly lower hemoglobin levels in the myomatic women than in normal women. Dingiloglu et al [19] also reported lower hemoglobin levels in the myomatic group, but there was no statistically significant difference when hemoglobin and leptin levels were compared. The low hemoglobin levels found would not likely explain the decreased plasma leptin levels in myomatic women. Factors which can affect serum leptin levels such as age, BMI, blood glucose levels and renal function tests, were similar between the study and control groups in this study, so they do not explain the situation. Chiaffarino et al [13] found that, a high fat diet was associated with myomas, while a high carbohydrate diet has a protective effect on myomas. Havel et al [18] observed that consumption of high fat/ low carbohydrate meals resulted in lowered 24-h circulating leptin concentrations. These studies, together with observations obtained in this study suggest that lower leptin levels are possibly involved in the pathogenesis of myomas.

In this study, it was observed that serum leptin levels were positively correlated to body mass index in both the myomatic (r=0.587, p=0.0001) and normal women (r=0.608, p=0.0001). However, for a given body mass index, serum leptin level was likely to be lower in women with myomas than in those without. These findings agree with those reported by Chan et al [16]. In addition, ratios of serum leptin levels/body mass index were significantly lower (p=0.0001) in myomatic women than in normal women. Similar results were also observed by Chan et al [16] as ratios in myomatic women were significantly lower, suggesting that the decreased serum leptin levels in myomatic women were independent of body mass index. However, Dingiloglu et al [19] disagree with these findings; in their study, higher ratios of serum leptin levels/body mass index were observed in myomatic women as compared with normal women, still the difference was not statistically significant.

BMI and body fat are the major determinants of plasma leptin, especially total body fat mass [21]. In this study, a significantly higher (p=0.0001) leptin levels were observed in women with high BMI (BMI>30) as compared to those with BMI<30. These findings are consistent with Dingiloglu et al [19], Rosenbaum et al [22] and Ambrosius et al [21] observations. Also Havel et al [23] reported that leptin levels were higher (per unit of adiposity) in obese subjects than in those with normal weight; this finding also supports results of this study.

In this study, no correlation was observed between ratios of serum leptin levels/ body mass index and body mass index in both myomatic and normal women. This partially agree with those reported by Chan et al [16]; in their study, a positive correlation was found between ratios of serum leptin levels/ body mass index and body mass index in normal women but not in the myomatic women. The possible explanation is that, in myomatic women, the normal up- regulation of leptin production in response to increased adiposity is blunted. Further studies are required to prove or disprove this possibility.

CONCLUSION:

Unlike normal women, the lower serum leptin levels observed in the women with myomas were independent of body mass index with no significant up-regulation of leptin production in response to increased adiposity.

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