Original Article

Leptin Levels in Pre and Post Menopausal Pakistani Women

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Abstract

Objective: To evaluate the correlation of fasting serum leptin levels with anthropometric measurements and menopausal status, in women.

Methods: The study comprised of 80 non-obese women who were divided into 2 groups as pre-menopausal (n=46) and post-menopausal (n=34).Anthropometric measurements i.e. height, weight, waist and hip circumference were measured for all the subjects and the Waist-hip Ratio (WHR) and Body Mass Index (BMI) were calculated from these measurements. A fasting venous blood sample was taken from all the subjects and serum leptin concentrations were determined by ELISA.

Results: A comparison of the mean values for BMI and WHR between the two groups showed a non significant difference. Within each group, significant associations were noted between the fasting serum leptin level and values of BMI and WHR. A comparison of the mean serum leptin concentrations between the two groups, showed a highly significant difference ($p<0.001$).

Conclusion: Our results indicate that in the non-obese women of our population, leptin levels associate with BMI and WHR and together with menopausal status seem to be important determinants of serum leptin levels (JPMA 56:3;2006).

Introduction

Bodyweight is regulated by complex mechanisms involving numerous afferent metabolic and hormonal signals informing the brain about the body's energy status. Abnormal production or action of any of the afferent messengers may lead to weight gain.[1]

Leptin (derived from leptos meaning thin), a 16Kd protein synthesized and secreted from adipocytes in both animals and humans, influences multiple neuro endocrine systems and plays a significant role in the regulation of body fat storage, energy expenditure, and body weight changes.[2] The main determinant of plasma leptin concentration in humans appears to be total body fat content.[3,4] After adjustment for body fat, females show higher leptin concentrations than males.[5,6] Normally menstruating women show higher leptin levels than postmenopausal women, thus suggesting a stimulatory effect of oestrogen and/or progesterone and an inhibitory action of androgens on plasma leptin concentrations.[7]

Studies carried out in different parts of the world have suggested that racial differences in body composition and pattern of fat distribution may contribute to variations in serum leptin concentrations. Thus, the aim of the present study was to evaluate the correlation of leptin levels with body mass index, waist hip ratio, and menopausal status in the women.

Subjects and Methods

The present study comprised of eighty women who were selected by convenient sampling method including those who were healthy, sedentary and non-obese. A detailed history was taken from each subject to ascertain age, menopausal status and to exclude any history of diabetes, hypertension, thyroid and other endocrine disorders, use of HRT or oral contraceptives. The subjects were then divided into 2 groups as pre menopausal, (n=46) and post menopausal (n=34) Anthropometric measurements e.g. height, weight, waist and hip circumference were measured for all the subjects. Waist circumference was measured at the level of the umblicus and hip circumference at the widest point around the hips. BMI [wt (kg)/ht. $(m)^2$] and waist to hip circumference ratios were then calculated. After taking all aseptic precautions , a fasting venous blood sample was taken from each subject. Serum leptin concentrations were measured by an enzymatically amplified "two-step" sandwich-type immunoassay, using the commercially available DSL-10-23100 Active™ Human Leptin Enzyme-Linked Immunosorbent (ELISA) kit.

In the assay, standards, controls, and unknown serum and plasma samples are incubated in microtitration wells, which have been coated with anti-human leptin antibody. After incubation and washing, the wells are treated with another anti-human leptin detection antibody, labeled with the enzyme horse radish peroxidase (HRP). After a second incubation and washing step, the wells are incubated with the chromogenic substrate tetramethylbenzidine (TMB). An acidic stopping solution in then added and the degree of enzymatic turnover of the substrate is determined by dual

wavelength absorbance measurements of 450 and 620mm. The absorbance measured is directly proportional to the concentration of human leptin present. A set of human leptin standards is used to plot a standard curve of absorbance versus human leptin concentrations from which the human leptin concentration in the unknown can be calculated.

Results

Fasting serum leptin concentrations, in both groups, ranged from the minimal detectable concentration of 0.7ng/ml to a highest value of 67.2ng/ml in the premenopausal group and 66.5ng/ml in the group of postmenopausal women.

A significant difference in fasting serum leptin levels was noted between the two groups ($P<0.001$), with the premenopausal group having a significantly higher mean serum leptin level of 33.8ng/ml as compared with 24.2ng/ml estimated for women in the post menopausal group. Comparison of the mean values for BMI and WHR between the two groups showed no significant difference. On evaluating the correlation between fasting serum leptin level and BMI in the postmenopausal group, a strongly negative correlation was noted (r=-0.2). Whereas, in the premenopausal group, a much weaker negative correlation was seen (r=-0.09). The correlation between WHR and leptin levels in the two groups of women was found to be negative in the postmenopausal group (r=-0.02) and positive in the premenopausal group of women (r=+0.15).

Discussion

Studies have shown that anthropometric measurements such as BMI ,WHR and skin fold thickness (because of a high correlation with body fat) may well predict leptin concentrations. They cannot distinguish between potential hypersecretion of leptin at the site of origin in various adipocyte tissues and the elevated serum leptin concentrations that result from decreased affinity at the receptor site or decreased clearance from the circulating blood.[8] Our study showed a non significant difference in the mean values of BMI estimated for the two groups. However, within the group of postmenopausal women, the fasting serum leptin levels showed a strong negative correlation with BMI. In the premenopausal group, a weaker negative correlation was seen between serum leptin and BMI. Most of the previous studies have shown a strong correlation of serum leptin with BMI. In a study of leptin levels in normal weight and obese individuals, serum leptin levels correlated positively with BMI and were significantly elevated in obese subjects.[4] A population based study in the US also found that BMI could be expected to be highly correlated with leptin concentrations because of its generally high correlation with body fat.[6] However, another study[7] found that leptin was not significantly correlated with BMI.

A primary action of leptin is thought to be its role as an afferent satiety signal in a feedback loop that effects the appetite and satiety centers of the brain. Intake of food triggers insulin and glucocorticoid release, thereby favouring fat accumulatation and the secretion of leptin, which in turn subsequently causes satiation.[9] Our findings for BMI and WHR seem to suggest that in non-obese premenopausal women, as the WHR increases, leptin levels also increase and provide a signal to the brain to suppress body fat by decreasing food intake or increasing energy expenditure. On the other hand, in the non-obese postmenopausal group, the negative correlations suggest that this signal is very weak and because of a lack of a coincident rise in serum leptin concentrations with an increase in BMI and WHR, there is an increased predisposition to fat deposition.

Comparison of fasting serum leptin levels in the two groups showed a highly significant difference. The postmenopausal group having considerably lower leptin levels than the premenopausal group. This finding coincides with that of other workers[7] who found leptin levels to be higher in pre than in the post menopausal women and both groups had

Table 1. Leptin levels in the Premenopausal and Menopausal groups.

Groups	Age (years)	Height (m)	Weight (kg)	BMI (kg/m^2)	W/H Ratio	Leptin Levels (ng/ml)
Premenopausal (n=46)	42.46 ± 7.30	1.56 ± 0.07	66.09 ± 9.10	26.71 ± 3.37	0.84 ± 0.05	33.88 ± 22.43
Menopausal (n=34)	51.15 ± 7.71	1.56 ± 0.05	64.50 ± 9.23	27.05 ± 3.56	0.84 ± 0.06	24.24 ± 20.52

Values given as mean ± standard deviation.

higher levels than males. They suggested that these results could not be accounted for solely by effects of oestrogen and/or progesterone. According to them, it is likely that the suppressive effect of androgens is equally significant. Other workers have shown that oestrogens directly increase in vivo leptin production in rats and human subjects. Therefore, the hypothesis of a stimulatory effect of oestrogen and/or progesterone on plasma leptin concentrations has been formulated.[10] However, a similar comparison done in another study[11] found that menopausal status does not have a significant impact on leptin production.

The results of this study seem to suggest that in the non obese females of our sample, correlation exist between serum leptin levels and anthropometric measurements such as BMI and WHR, but the menopausal status is a more significant determinant of leptin levels. However, larger population studies should be carried out to confirm the same.

References

1. Leibowitz SF. Neurochemical-neuroendocrine systems in the brain controlling macronutrient intake and metabolism. Trends Neurosci 1992;15:491-7.
2. Flier JS. Leptin expression and action: new experimental paradigms. Proceedings of the National Academy of Sciences, USA. 1997;94:4242-5.
3. Dua A, Hennes MI, Hoffmann RG, Maas DL, Krakower GR, Sonnenberg GE, et al. Leptin: a significant indicator of total body fat but not of visceral fat and insulin insensitivity in African-American women. Diabetes 1996;45:1635-7.
4. Considine RV, Sinha MK, Heiman ML, Kriauciunas A, Stephens TW, Nyce MR, et al. Serum immunoreactive-leptin concentrations in normal-weight and obese humans. N Engl J Med 1996;334:292-5.
5. Ostlund RE, Yang JW, Klein S, Gingerich R. Relation between plasma leptin concentration and body fat, gender, diet, age and metabolic covariates. J Clin Endocrino and Metab 1996;81:3909-13.
6. Ruhl CE, Everhart JE. Leptin concentrations in the United States: relations with demographic and anthropometric measures. Am J Clin Nutr 2001;74:295-301.
7. Rosenbaum M, Nicolson M, Hirsch J, Heymsfield SB, Gallagher D, Chu F, et al. Effects of gender, body composition, and menopause on plasma concentrations of leptin. J Clin Endocrinol Metab 1996;81:3424-7.
8. Kuczmarski RJ. Leptin concentrations in U.S. adults. Am J Clin Nutr 2001;74:277-8.
9. Jeanrenaud FR, Jeanrenaud B. Obesity, Leptin and the Brain. NEJM 1996;334:324-5.
10. Shimizu H, Shimomura Y, Nakanishi Y, Futawatari T, Ohtani K, Sato N, et al. Estrogen increases in vivo leptin production in rats and human subjects. J Endocrinol 1997;154:285-92.
11. Douchi T, Iwamoto I, Yoshimitsu N, Kosha S, Nagata Y. Leptin production in pre and postmenopausal women. Maturitas 2002;42:219-23.