

Increased levels of plasma leptin in relation to etiology of premenstrual syndrome

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Abstract: The objective of the study was to investigate the relationship of leptin with body mass index based categories in different phases of menstrual cycles of both normal and premenstrual syndrome (PMS) symptoms. Female subjects (no=112; age:20±1.94,19-21years), were included to obtain information about their anthropometric profiles (subjects were categorized on the basis of their BMI) and PMS symptoms on a specified prescribed questionnaire and fasting blood samples were collected during specific days of follicular and luteal phases. The presumptive day of ovulation to identify the phases was detected by using biphasic BBT patterns. Plasma leptin levels were determined by EIA using commercially available Kits. Circulating leptin concentrations varied during the different phases of normal menstrual cycle; slightly higher but significant values during the luteal phase compared to the follicular phase only in subjects with less than 26.8 BMI. Further, PMS subjects depicted significantly higher plasma leptin levels ($p<0.001$) in both follicular and luteal phases compared to their corresponding controls of all the categories of BMI. The increased level of leptin in PMS subjects; suggest it could have an important role in the pathophysiology of PMS.

Keywords: Leptin, premenstrual syndrome, BMI.

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INTRODUCTION

Premenstrual syndrome (PMS), has been defined as a combination of distressing and menstrually related dysfunctions involving physical, psychological and or behavioral changes of sufficient severity causing ill-effect on interpersonal relationship or interference with normal activity¹. Its clinical features, screening, diagnosis, epidemiology, etiology, and treatment strategies reviewed recently highlighted its importance in women's reproductive health².

The data on PMS symptomatology has been reported in different populations; however, information about its etiology, with respect to proper diagnosis and treatment is needed³. There occurs overlap between PMS and atypical symptoms and variability in methodology produced further problems in understanding this syndrome^{4,5}. Attempts in recent years have been made to explore the etiology of PMS through association with relevant hormones like ovarian steroids and leptin⁶. Leptin, a tropic hormone for reproductive system, has been suggested to have a dynamic regulatory role in HPO axis participating in a variety of reproductive functions including those of ovary and its related syndrome⁷. Although relationship between leptin and oestradiol or progesterone was not found in a UK based study but leptin was suggested to have a role in PMS pathophysiology requiring further evaluation⁸. On the other hand, an interventional study with Gn-RH analogues failed to find any such role⁹. In this connection, no hormonal data is available on non western PMS subjects. An

association of obesity and energy metabolism with leptin and possibly with menstrual cycle or its dysfunctions/PMS has been reported^{10,11}. Different socio-cultural factors in Pakistan as compared to western countries may influence PMS as indicated by a recent study on Chinese women who showed overlapping and distinct features when compared with pattern in west⁴.

The aim of the present study was to explore the relationship of leptin with BMI based categories in different phases of menstrual cycle of both normal and PMS subjects

SUBJECTS AND METHODS

The study was conducted at random on adult female students of University of Karachi. The subjects with history of hypertension, metabolic or endocrine disease or taking hormonal therapy were excluded. The female subjects belonged to an average socio- economics status. They were provided with PMS charts and relevant instructions with monthly follow up to obtain the necessary data to help determine the duration and regularity of bleeding, pattern and severity of premenstrual and menstrual symptomatology, and the degree of life-style disruption precipitated by these symptoms¹. The completed detailed record, the PRISM calendar (Prospective Record of the Impact and Severity of Menstrual Symptoms, with 24 common behavioral and physical symptoms) for over three consecutive months were analyzed under ACOG guidelines (severity of some of these symptoms during luteal phase)³. Thus, the subjects were distinguished as

DISCUSSION

PMS subjects (with PMS symptoms; N=68) and controls (symptom free; N=44); study size much higher than a similar previous UK study conducted on 32 women with PMS and 28 women with asymptomatic menstrual cycles. This cross sectional study carried out during 2009, was approved by the Board of Advanced Studies and Research, University of Karachi and informed consent was obtained from all subjects.

The weight (Kg) of the subject using simple weighing machine and height (m) measured with height scale, were used to calculate BMI (Kg/m^2) according to the formula given as under ¹².

$$\text{BMI} = \text{weight (Kg)} / \text{height (m)}^2$$

The subjects categorized on the basis of their body weight.

Under weight	BMI < 22.4 kg/m^2
Normal	BMI 22.4 -26.8 kg/m^2
Overweight	BMI > 26.8 kg/m^2

Venous blood collected, centrifuged immediately and plasma stored at -70°C until assayed for leptin. Plasma leptin concentrations were measured in duplicates during the mid-follicular (days 5–9) and mid-luteal phases (days 19–23) of the menstrual cycles in women with PMS and with symptom-free cycles (controls), by EIA (ELISA Kit Cat#20-DR-2395 supplied by Alpco, USA¹³. PMS & control groups were matched for age, BMI, and duration of menstrual cycle. Statistical significance of plasma leptin values of different groups between PMS and control subjects were analyzed using paired *t*-test. Statistical analysis was performed using Statistica 5.0 version. $P < 0.05$ was considered statistically significant.

RESULTS

The present study measured plasma leptin concentrations of underweight, normal and overweight subjects with PMS symptoms and compared with their age and BMI matched controls. The data is shown in Figure 1. It appears that leptin levels in luteal phase as compared to the follicular phase in the controls as well as in the PMS subjects were higher ($p < 0.001$) in all BMI categories of subjects except those with BMI > 26.8 kg/m^2 in the controls. Further, it was also observed that the leptin levels were much higher ($p < 0.001$) both in follicular and luteal phases in PMS subjects of all the categories of BMI as compared to their corresponding controls (Figure 1).

The present study confirms the previous findings conducted on UK subjects⁸, that leptin may be increased in subjects showing suggestive PMS symptoms in comparison to the controls, thus further supporting the probable role of leptin in female reproductive physiology⁷. This study is the first description of a possible association of leptin with PMS in Pakistani women diagnosed with PMS symptoms. The previous studies conducted on Asian women primarily dealt with either prevalence or characterization of PMS symptoms using variety of diagnostic charts; for example, studies on Pakistani, Iranian and Indian populations¹⁴⁻¹⁶.

BMI categorization of the subjects and their relationship to leptin has not so far been reported in any of the previous study although data on American women provided evidence that obesity strongly is associated with PMS¹⁰. The present study demonstrates the raised level of leptin in the PMS subjects in luteal phase of all the three categories of BMI thereby suggesting that obesity may be one of the risk factors for PMS but not the only one.

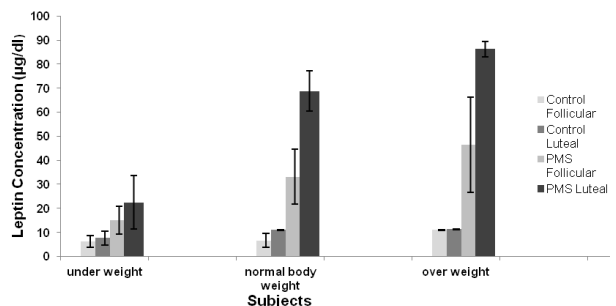


Figure 1: Plasma leptin concentrations ($\mu\text{g/dl}$) in PMS subjects and controls during their menstrual cycle phases. The subjects were categorized according to their BMI (<22.4, underweight; 22.4-26.8, normal body weight; >26.8, overweight)¹².

The etiology of PMS remains mostly unknown^{17,5}. Currently, the diagnosis is done on the record of the menstrual cycle (timing and symptom pattern) obtained on the prescribed daily symptoms chart - a recommended questionnaire. The present study has employed one of such recommended questionnaire¹. There have been a few hypotheses regarding the etiology of this syndrome. For example, it may have genetic basis¹⁷ or may be due to abnormal response by CNS to endogenous steroids or their metabolites either directly or mediated by certain specific neurotransmitters¹⁸. However, certain studies failed to find effective role of progesterone/estrogen in the etiology of PMS⁶.

Very few studies have been attempted to find basis for possible involvement of hormones like leptin in this complex mechanism. Although our data does not indicate precise cause-effect relationship of leptin with PMS, however, it does documents that leptin may indeed play an important role in triggering the onset of PMS symptoms, in agreement with the finding on UK population⁸. Accordingly, the possible causes of a raised level of leptin in PMS patients may include:

A direct action on hypothalamic neuronal receptors are well known for their participation through limbic system in a variety of neuroendocrinological mechanisms including those for food intake, energy expenditure and behavioral physiology¹⁹, well supported by localization and expression of functional leptin receptors OB-Rb in specific hypothalamic nuclei having important link with phenomenon of satiety and reproduction⁸.

Previously, no change in the pattern of leptin was found between controls and those with PMS symptoms²⁰. However, raised levels of leptin during follicular phase of PMS group in comparison to corresponding controls as well as the different levels among two phases in subjects showed that the PMS symptoms in our study may suggest that alteration can take place even in the follicular phase of the menstrual cycle. It may be that higher levels have the ability to prime CNS in women predisposed to PMS, in line with the previous findings that PMS symptoms can appear very early in the luteal phase⁸. Perhaps, measurements of daily pattern of leptin secretion during a complete menstrual cycle may suggest the precise day of leptin rise.

Secondly, leptin may participate in central response to neurosteroids and their metabolites¹⁸. Thus, it may have a role in PMS etiology via its effect on NP-Y transmission pathway. Leptin via L-arginine-nitric oxide pathway induces brain serotonin metabolism²¹. Increased metabolism or uptake of serotonin can reduce its concentrations – suggested to be another mechanism leptin may have for development of PMS²¹.

Leptin is also shown to vary in women with eating disorders. The relationship of increased leptin concentration with food cravings observed in women with PMS remains unexplained as it is well known that leptin stimulates satiety center²¹. However, it may be due to condition of leptin resistance as suggested in obesity²² although women with obesity or insulin resistance may not show PMS symptoms. In this study an association of increased leptin levels is observed in women with high BMI but we have no data in women with

insulin resistance. Moreover, involvement of nitric oxide (NO) has also been postulated to have link with etiology of PMS but awaits confirmation²³.

Briefly, physiological importance of leptin increase during luteal phase is not clear, but does document that leptin, may have a role in the regulation of cyclic female reproductive function. Such a rise as well as the difference of leptin levels between control subjects and PMS patients may be due to highly complex interactions between ovarian hormones and the neuroendocrine mechanisms due to neuropeptides and neurotransmitters responsible for food intake, energy expenditure and adiposity during the luteal phase of the menstrual cycle. Thus, the present study indicates the multiple disturbances regarding neuroendocrine systems in this syndrome. It may be suggested that the variability in the leptin levels of menstrual cycle phases of both groups falling under different categories of BMI, may depend on the ethnicity as well as on different socio-economic status of the subject and is well supported by recent reports^{24,25}. It is apparent that further studies on a larger population are needed to arrive at a conclusion. However, the alterations in leptin concentration during luteal phase in PMS subjects document a true physiological difference which may suggest its clinical importance; perhaps as a biomarker for PMS diagnosis and management.

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