Scholars LITERATURE

Journal of Womens Health Care and Management

Research Article

DOI: https://doi.org/10.47275/2692-0948-135
Volume 3 Issue 2

Study on Inflammatory Markers in Premenstrual Syndrome(Emphasis on ESR, TLC and C-Reactive Protein Levels) in South Indian Women

Sai Anirudh A[1]*, Lakshmi PVV[2], Syamala Devi M[3], Nikshiptha J[4] and Sree Gouthami K[5]

[1]Rangaraya Medical College, Andhra Pradesh, India
[2]Department of Physiology, Andhra Medical College, Andhra Pradesh, India
[3]GVP Medical College, Andhra Pradesh, India
[4]Mallareddy Medical College, Telangana, India
[5]Rangaraya Medical College, Andhra Pradesh, India

Abstract

Aim: This study aims to study the role of inflammation as a key contributor to the pathophysiology of PMS, which is still undefined to this day; by studying serum levels of inflammatory markers in women with Premenstrual syndrome (PMS) symptoms and women without PMS symptoms at a different point of their menstrual cycle. This study also sheds light on the significance of correlates such as BMI, onset age of menarche, type of lifestyle etc on the severity of PMS symptoms.

Methods: 100 South Indian women chosen as per the above-mentioned subject inclusion criteria were made to fill out a questionnaire with questions regarding common symptoms of PMS and whether they have experienced those symptoms in the late-luteal phase or not. Women who answered yes to most questions were asked to grade the severity of said symptoms as mild, moderate and severe. Their clinical correlates such as height and weight were measured to calculate BMI. Other factors such as age at menarche, current pulse, blood pressure and type of lifestyle (active, slightly active or sedentary) were also noted. In the last 7-10 days of their menstrual cycle, their samples were collected for analysis of their ESR, TLC and C reactive protein levels. Statistical graphical analysis of the results attained with consideration of age and other correlates was done to gauge the relevancy of the correlation of inflammatory markers in PMS diagnosis, onset, and degree of symptom severity.

Results: There was a significant rise of inflammatory marker levels i.e., ESR ($Z = -8.651$ at $p < 0.001$), TLC ($Z = -6.614$, $p < 0.001$) and CRP ($Z = -7.743$, $p < 0.001$) in PMS subjects over women in the control group, when analyses were done using the Wilcoxon signed ranks test. However, the inflammatory markers studied did not seem to affect symptom severity as the relationship between the degree of severity of symptoms and inflammatory marker levels were not statistically significant. But the correlates BMI ($W = 896.500$, $p = 0.019$) and onset age of menarche ($W = 984.500$, $p = 0.030$) significantly seemed to influence PMS symptom severity when statistical significance was assessed via Wilcoxon-Mann-Whitney U Test, with the median BMI (Kg/m2) and mean onset age of menarche being highest in the severe category of the degree of PMS symptoms experienced.

Conclusions: There is a role played by inflammation in causing PMS or its symptoms (owing to the rise of levels of inflammatory markers such as ESR, TLS and CRP in PMS) but the inflammatory markers studied did not seem to affect symptom severity. However, the correlation between BMI and the onset age of menarche seemed to influence PMS symptom severity. While the type of lifestyle lived by the woman did not directly influence symptom severity, it was found to have a significant influence on BMI and onset age of menarche; thus, indirectly influencing PMS symptom severity. The findings of this study also pave way for further studies to shed light on the role of lectin in influencing PMS symptom severity, as well as the significance of the onset age of menarche in various other studies of female reproductive or menstrual physiology.

Correspondence to: Sai Anirudh Athaluri, Rangaraya Medical College, Kakinada, Andhra Pradesh, India; E-mail: saiathaluri@gmail.com

Citation: Anirudh AS, Lakshmi PVV, Moparthi SD (2022) Study on Inflammatory Markers in Premenstrual Syndrome (Emphasis on ESR, TLC and C-Reactive Protein Levels) in South Indian Women. J Womens Health Care Manage, Volume 3:2. 135. DOI: https://doi.org/10.47275/2692-0948-135

Received: September 15, 2022; **Accepted:** December 23, 2022; **Published:** December 28, 2022

Introduction

Premenstrual syndrome (PMS) is a condition associated with a group of consistently occurring symptoms, some of which include irritability, bloating, oedema, decreased ability to concentrate, depression, headache and constipation; which usually occur during the last 7-10 days of the menstrual cycle, corresponding to the secretory or luteal the phase of the menstrual cycle. These symptoms have conventionally been attributed to salt and water retention.

However, it is rather unlikely that this or any of the other hormonal alterations that occur in the late luteal phase are responsible, because the duration and severity of the symptoms are not modified if the luteal phase is terminated early. Several studies have shown that the effects of antidepressant fluoxetine, which is a serotonin reuptake inhibitor, and the benzodiazepine alprazolam produces symptomatic relief, and so do GnRH-releasing antagonists in doses that suppress the pituitary ovarian axis. Hence, the pathophysiology of PMS remains undefined to this day [1].

Moreover, as of now, there are no unique lab findings that facilitate an easy diagnosis of PMS. Currently, doctors attribute particular symptoms to PMS if it is a part of the predicted menstrual pattern [2]. Henceforth, it is seemingly apparent that PMS isn't always effectively diagnosed.

Chronic inflammation, however, has been deemed a manifestation of depression and other important psycho-somatic aspects associated with PMS, paving way for research to establish a definite detection of PMS via inflammatory markers [3]. This inflammation occurs when cytokine production remains active in cells. Cytokines are hormonelike molecules that act generally in a paracrine fashion to regulate immune responses, enabling inflammation. They are secreted not only by lymphocytes but also by endothelial and endometrial cells to facilitate vascularisation and tissue repair after periodic endometrial shedding [4]. Related studies done on women in western countries on inflammatory markers in depression to date have only taken subjects with either reference BMI of 20.5 or a cut-off BMI as 25 which is considerably different from the Indian ideal which is 23, as dictated by the American Diabetes Association as the threshold cut-off for Asian ethnicity people [3,5, and 6]. The subjects of said studies have reported being on a protein-rich diet, which is contrary to the current South Indian scenario (carbohydrate-rich diet which is an attribute of rice consumption). Also, the parameters of this particular study haven't been evaluated before collectively. With this sufficient pre-requisite knowledge, we intend to evaluate how inflammatory markers are correlated with PMS and its effective clinical diagnosis.

Review of Literature

Premenstrual syndrome (PMS) and premenstrual dysphoric disorder (PMDD) affect millions of women of reproductive age as the prevalence of PMS is given as 10-98% while PMDD affects 2-8% [7]. PMS and PMDD symptoms compromise the well-being of women and harm their quality of life. Definitive diagnosis is based on prospective self-reporting of the symptoms. Most cases are unrecognized as the presence of the symptoms is not usually questioned during gynecological exams and routine check-ups [7]. PMS, a common cyclic disorder of young and middle-aged women, is characterized by emotional and physical symptoms that consistently occur during the luteal phase of the menstrual cycle. PMS and PMDD are diagnoses of exclusion; therefore, alternative explanations for symptoms must be considered before either diagnosis is made. The disorders can manifest with a wide variety of symptoms, including depression, mood lability, abdominal pain, breast tenderness, headache, and fatigue [8]. Although evidence for a hormonal abnormality has not been established, the symptoms of the premenopausal disorders might be related to the production of progesterone by the ovary. Thus, ovulation suppression and hormonal regulation are areas of focus for diagnostic and treatment options [9]. Women with PMDD experience affective or somatic symptoms that cause severe dysfunction in social or occupational realms. Proposed aetiologies include increased sensitivity to normal cycling levels of estrogen and progesterone, increased aldosterone and plasma renin activity, and neurotransmitter abnormalities, particularly serotonin [10].

Hence, there are various schools of thought offering to explain the pathogenesis of PMS which remains mostly unknown to this day. The perspective upon which this study was devised is the possibility of PMS symptoms occurring in an inflammatory background.

Work by Gold EB, et al. (2016) [12], presented in the Journal of Women's Health contributes importantly to the knowledge of chronic inflammation and PMS. Using data from the large, multi-ethnic Study of Women's Health Across the Nation, the authors assessed the association of elevated CRP levels with premenstrual symptom severity among 2939 premenopausal and early perimenopausal women. At baseline, women reported the presence or absence of each of eight symptoms in the week before menses or during menses, and whether symptoms ended within 1-3 days of menses onset. Factor analysis was used to identify five distinct symptom groups from among reported symptoms. Women provided a fasting blood sample, assayed for hs-CRP; women with levels >3 mg/L were classified as having elevated CRP. After adjustment for important demographic and behavioral factors including body mass index and level of social support, elevated CRP was associated with significant 26%–41% higher odds of four of the five symptom groups evaluated. This is not only the largest but also the most racially and ethnically diverse study of inflammation and premenstrual symptoms conducted to date [11]. In another study wherein, PMS diagnosed women's CRP levels were studied, an hs-CRP level >3 mg/L was significantly positively associated with premenstrual symptoms, paving way for inflammation to be implicated as a backdrop for PMS [12].

(In both the above studies, Elevated CRP and the hs-CRP level were not associated with chronic headaches that some women were already suffering from irrespective of their PMS status). In another study that studied the effect of zinc supplementation on physical and psychological symptoms, biomarkers of inflammation, oxidative stress, and brain-derived neurotrophic factor in young women with PMS, it was found that zinc supplementation for 12 weeks among women with PMS had beneficial effects on physical and psychological symptoms of premenstrual syndrome, total antioxidant capacity, and brain-derived neurotrophic factor. Zinc is known to have multiple beneficial effects especially anti-inflammatory, antioxidant and anti-depressant actions. Its ability to alleviate PMS symptoms again hints at inflammation being a possible cause of PMS [13]. In another study when compared with the controls, women with PMDD had higher BMI, higher leptin concentrations in the EL and LL phase, and leptin concentrations increased from the EL to the LL phase. And this study concluded that young women with PMDD had higher leptin concentrations and BMI in the luteal phase. Though this study hinted at BMI being a possible indicator of PMS along with the fact that Leptin possibly might play a role in the pathophysiology, the role of BMI was not further assessed in this study or other similar studies to our knowledge [14]. Other studies also suggest that leptin could have a role in the pathophysiology of PMS and indicate the degree of severity of PMS [15]. However, future studies on the role of leptin in PMS are needed. ESR and TLC are useful, inexpensive inflammatory markers that are widely used for routine testing of patients in India. But their levels in relation to PMS pathophysiology have not been assessed satisfactorily by other studies to date to our knowledge. Hence through this study, we intend to understand the role of inflammation in PMS, and assess changes in the levels of ESR, TLC and CRP in patients with varying degrees of severity of premenstrual syndrome.

Aims and Objectives

To study the relationship between inflammatory markers and Premenstrual syndrome (PMS).

To establish significant inflammatory markers as reliable detectable indicators for the onset of PMS and its clinical diagnosis.

To understand the significance of correlates like age, BMI etc on PMS symptoms severity.

To pave way for further understanding of the pathophysiology of PMS with this newly established relation with inflammatory markers and the bio-chemical pathway of their action, as the pathophysiology remains undefined to this day.

Materials and Methods

Type of Study

A cross-sectional study involving statistical analysis and clinical biochemical investigations to evaluate the ESR, TLC and C reactive protein levels in seemingly healthy South Indian women aged 18 to 30 years, during the last 7-10 days of their menstrual cycle to establish a correlation with PMS diagnosis.

Sample Population

South Indian women visiting the government general hospital of respective college for clinical tests under study, aged 18-30 years, who consume a quintessential Indian carbohydrate-rich diet (rice in this context) rather than a western protein-rich diet.

Criteria for choosing subjects: Subjects were chosen such that they were aged between 18-30 and not afflicted with amenorrhea, and without any familial history of high BP or increased cholesterol levels, hepatic or renal afflictions, or any inflammatory autoimmune diseases like AIDS or rheumatoid arthritis. Subjects who are on medications were not chosen.

Sample size: A total of 100 subjects who met the above-mentioned criteria were chosen.

Duration of Study: Two months.

Study Design

Categorisation of subjects for study:

Subject category 1: Women aged between 18-30 with normal BMI.

Subject category 2: Women aged between 18-30 with higher BMI indicating overweight condition.

Subject category 3: Women aged between 18-30 with higher BMI indicating obese condition.

Method

Women chosen as per the above-mentioned criteria were first informed regarding the study via an informed consent form. Only after receiving their signed approval for participation in the study, the subjects matching the selection criteria were taken into consideration. The subjects were made to fill out a questionnaire with questions regarding common symptoms of PMS and whether they had experienced those symptoms in the late-luteal phase or not. Women who answered no to most questions were not taken into the study as their parameters proved to be difficult to correlate with the study being done, as it indicated that they were not suffering from PMS at the time. On the other hand, women who answered yes to most questions were made to grade the severity of said symptoms as mild, moderate and severe. (However, during data collection none of the subjects experienced mild PMS symptoms; they only reported that they were suffering from moderate or severe PMS symptoms, or they were not suffering from PMS at all at the time. Hence the three categories of mild, moderate and severe were inevitably reduced to two: moderate and severe categories during statistical analysis).

Their clinical correlates such as height and weight were measured to calculate BMI. Other factors such as age at menarche, pulse rate, blood pressure and type of lifestyle (active, slightly active or sedentary) were also noted.

In the last 7-10 days of their menstrual cycle, their samples were collected for analysis of their ESR, TLC and C reactive protein levels.

ESR, TLC and CRP levels of women diagnosed with PMS were compared with marker levels of women in the control group (same women who were matching the subject selection criteria but were not suffering from PMS symptoms when their blood sample collection was done at a subsequent visit during a different phase of their menstrual cycle other than the late-luteal PMS phase).

Statistical analysis of the results attained with consideration of age and other correlates was done to gauge the relevancy of the correlation of inflammatory markers in PMS diagnosis, onset and severity.

Statistical Analysis

First, to assess the role of inflammation and the rise of inflammatory marker levels in the pathophysiology of PMS (as hypothesized), ESR, TLC and CRP levels of women diagnosed with PMS was compared with marker levels of women in the control group (same women who were matching the subject selection criteria but were not suffering from PMS symptoms when their blood sample the collection was done at a subsequent visit during a different phase of their menstrual cycle). Before comparison, however, the normality of data distribution of each marker was assessed, and it was found that all the data were not normally distributed (as per Shapiro-Wilk Test). So, a non-parametric test (Wilcoxon-Signed-Ranks Test) was used to assess the rise in marker levels in the PMS group compared to the control group instead of parametric tests (such as paired sample t-tests). After establishing a rise in marker levels as significant in PMS occurrence, the effects of correlates and inflammatory markers on PMS severity (moderate and severe) were assessed. Upon analysis, it was evident that the variables BMI (Kg/m^2) and onset age of menarche has significant effects on PMS symptom severity. To assess the magnitude of their influence on PMS symptom severity, a non-parametric test (Wilcoxon-Mann-Whitney U Test) was used to make group comparisons. (Non-parametric test was used as the data distribution was not normal)..

Observation and Results

ESR Levels in PMS

Table 1: Distribution of the participants in terms of ESR (mm/Hr) (n = 100).

ESR (mm/Hr)	
Mean (SD)	45.25 (22.87)
Median (IQR)	42.5 (28-60)
Range	8 - 100

The variable ESR in PMS diagnosed women (mm/Hr) was not normally distributed (Shapiro-Wilk Test: p = 0.002).

The mean (SD) of ESR in the PMS diagnosed group (in mm/Hr) was 45.25 (22.87). The median (IQR) of ESR (mm/Hr) was 42.50 (28-

60). The ESR (mm/Hr) ranged from 8-100.

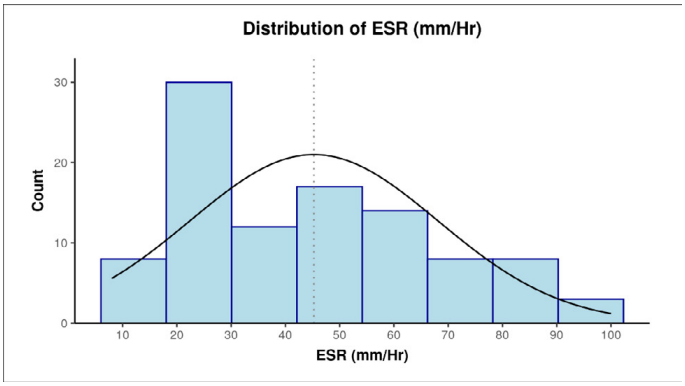

Figure 1: Distribution of ESR (mm/Hr).

Table 2: Distribution of the Participants in Terms of ESR (n = 100).

ESR	Frequency	Percentage
≤20 mm/Hr	13	13.0%
>20 mm/Hr	87	87.0%
Total	100	100.0%

13.0% of the participants had ESR: ≤20 mm/Hr. 87.0% of the participants had ESR: >20 mm/Hr.

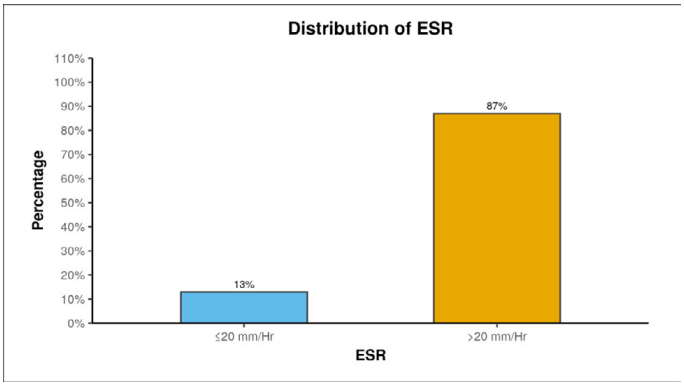

Figure 2: Distribution of participants in terms of ESR.

As the Shapiro-Wilk test indicated that the data was not normally distributed, Wilcoxon signed ranks test (a non-parametric test) was used to compare and contrast the alterations in ESR levels in control group (women whose blood samples were taken at a subsequent visit during their menstrual cycle when they weren't suffering from PMS) and in the PMS diagnosed group.

Table 3: Descriptive statistics.

	N	Mean	Std. Deviation	Minimum	Maximum
ESR in control group women	100	9.71	5.368	1	19
ESR in PMS diagnosed women	100	45.25	22.867	8	100

The test indicated that ESR levels in women diagnosed with PMS were significantly higher when compared to women in the control the group who weren't experiencing symptoms of PMS during the sample collection (Z= -8.651, p <0.001).

TLC Levels in PMS

The variable TLC (/cu.mm) was not normally distributed (Shapiro-Wilk Test: p = 0.034).

Table 4: Ranks (Wilcoxon Signed Ranks Test).

		N	Mean Rank	Sum of Ranks
ESRpms - ESRcontrol	Negative Ranks	2a	4.50	9.00
	Positive Ranks	98b	51.44	5041.00
	Ties	0c		
	Total	100		
a. ESRpms < ESRcontrol				
b. ESRpms > ESRcontrol				
c. ESRpms = ESRcontrol				

Table 5: Test statistics-a.

	ESRpms - ESRcontrol
Z	-8.651b
Asymp. Sig. (2-tailed)	<.001
a. Wilcoxon Signed Ranks Test	
b. Based on negative ranks.	

Table 6: Distribution of the PMS diagnosed group in terms of TLC (/cu.mm) (n = 100).

TLC (/cu.mm)	
Mean (SD)	10473.00 (2614.25)
Median (IQR)	10300 (9200-12000)
Range	3300 - 19100

The mean (SD) of TLC (/cu.mm) was 10473.00 (2614.25). The median (IQR) of TLC (/cu.mm) was 10300.00 (9200-12000). The TLC (/cu.mm) ranged from 3300-19100.

Table 7: Distribution of the PMS diagnosed group in terms of TLC (n = 100).

TLC	Frequency	Percentage
≤11000 /cu.mm	61	61.0%
>11000 /cu.mm	39	39.0%
Total	100	100.0%

61.0% of the participants had TLC: ≤11000 /cu.mm. 39.0% of the participants had TLC: >11000 /cu.mm.

As the Shapiro-Wilk test indicated that the data was not normally distributed, Wilcoxon signed ranks test (a non-parametric test) was used to compare and contrast the alterations in TLC levels in control group (same women whose blood samples were taken at a subsequent visit during their menstrual cycle when they weren't suffering from PMS) and in the PMS-diagnosed group.

Table 8: Descriptive statistics.

	N	Mean	Std. Deviation	Minimum	Maximum
TLCcontrol	100	7690.00	1947.104	4500	10900
TLCpms	100	10473.00	2614.252	3300	19100

Table 9: Ranks (Wilcoxon Signed Ranks Test).

		N	Mean Rank	Sum of Ranks
ESRpms - ESRcontrol	**Negative Ranks**	23a	26.15	601.50
	Positive Ranks	77b	57.77	4448.50
	Ties	0c		
	Total	100		
a. TLCpms < TLCcontrol				
b. TLCpms > TLCcontrol				
c. TLCpms = TLCcontrol				

Table 10: Test statistics-a.

	TLCpms - TLCcontrol
Z	-6.614b
Asymp. Sig. (2-tailed)	<.001
a. Wilcoxon Signed Ranks Test	
b. Based on negative ranks.	

The test indicated that TLC levels in women diagnosed with PMS were significantly higher when compared to women in the control a group who weren't experiencing symptoms of PMS during the sample collection (Z= -6.614, p <0.001).

CRP Levels in PMS

Table 11: Distribution of the participants in terms of CRP (mg/L) (n = 100).

CRP (mg/L)	
Mean (SD)	9.23
Median (IQR)	7.00
Range	2 - 79

The variable CRP (mg/L) was not normally distributed (Shapiro-Wilk Test: p = <0.001).

The mean (SD) of CRP (mg/L) was 9.23. The median (IQR) of CRP (mg/L) was 7.00. The CRP (mg/L) ranged from 2-79.

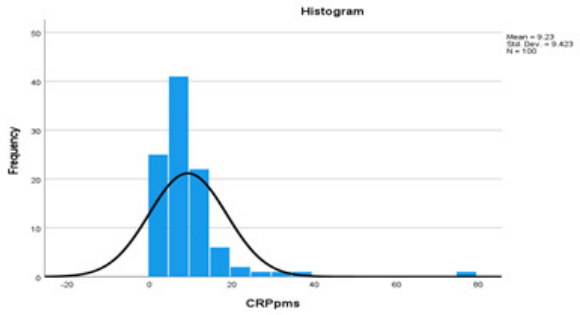

Figure 3: Histogram representation of the participants in terms of CRP (mg/L) (n = 100).

Table 12: Distribution of the PMS diagnosed group in terms of CRP (n = 100).

CRP	Frequency	Percentage
≤5 mg/L	35	35.0%
>5 mg/L	65	65.0%
Total	100	100.0%

35.0% of the participants had CRP: ≤5 mg/L. 65.0% of the participants had CRP: >5 mg/L.

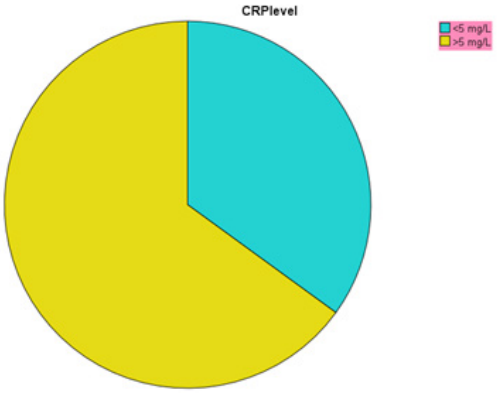

Figure 4: Distribution of the PMS diagnosed group in terms of CRP (n = 100).

As the Shapiro-Wilk test indicated that the data was not normally distributed, Wilcoxon signed ranks test (a non-parametric test) was used to compare and contrast the alterations in CRP levels in control group (same women whose blood samples were taken at a subsequent visit during their menstrual cycle when they weren't suffering from PMS) and in the PMS-diagnosed group.

Table 13: Descriptive statistics.

	N	Mean	Std. Deviation	Minimum	Maximum
CRPcontrol	100	2.97	1.460	1	5
CRPpms	100	9.23	9.423	2	79

Table 14: Ranks (Wilcoxon Signed Ranks Test).

		N	Mean Rank	Sum of Ranks
CRPpms - CRPcontrol	**Negative Ranks**	10a	18.30	183.00
	Positive Ranks	84b	50.98	4282.00
	Ties	6c		
	Total	100		
a. CRPpms < CRPcontrol				
b. CRPpms > CRPcontrol				
c. CRPpms = CRPcontrol				

Table 15: Test statistics-a.

	CRPpms – CRPcontrol
Z	-7.743b
Asymp. Sig. (2-tailed)	<.001
a. Wilcoxon Signed Ranks Test	
b. Based on negative ranks.	

The test indicated that CRP levels in women diagnosed with PMS were significantly higher when compared to women in the control the group who weren't experiencing symptoms of PMS during the sample collection (Z= -7.743, p <0.001). From the above results, it is clear that there is a significant rise of inflammatory marker levels (ESR, TLC and CRP) in women suffering from PMS, suggesting the possible role of inflammation in PMS. With the above positive correlation, further statistical analysis was done to assess the influence of the same inflammatory markers and other correlates on PMS symptom severity and the results are published in table 16.

Table 16: Association between degree of symptoms and parameters.

Parameters	Degree of Symptoms		p value
	Moderate (n = 44)	Severe (n = 56)	
Age (Years)	25.43 ± 2.68	25.34 ± 2.88	0.8991
BMI (Kg/m²)***	27.02 ± 2.57	28.39 ± 3.63	0.0191
BMI			0.0602
<18.5 Kg/m²	0 (0.0%)	1 (1.8%)	
18.5-22.9 Kg/m²	1 (2.3%)	1 (1.8%)	
23.0-24.9 Kg/m²	3 (6.8%)	3 (5.4%)	
25.0-29.9 Kg/m²	34 (77.3%)	30 (53.6%)	
30.0-34.9 Kg/m²	6 (13.6%)	19 (33.9%)	
35.0-39.9 Kg/m²	0 (0.0%)	2 (3.6%)	
Lifestyle			0.0623
Active	16 (36.4%)	11 (19.6%)	
Sedentary	28 (63.6%)	45 (80.4%)	
Onset Age of Menarche***	12.14 ± 0.63	12.57 ± 1.02	0.0301
TLC (/cu.mm)	10840.91 ± 3136.16	10183.93 ± 2102.10	0.2364
TLC			0.9473
≤11000 /cu.mm	27 (61.4%)	34 (60.7%)	
>11000 /cu.mm	17 (38.6%)	22 (39.3%)	
ESR (mm/Hr)	47.14 ± 24.05	43.77 ± 22.00	0.4731
ESR			0.6663
≤20 mm/Hr	5 (11.4%)	8 (14.3%)	
>20 mm/Hr	39 (88.6%)	48 (85.7%)	
CRP (mg/L)	9.91 ± 12.203	8.70 ± 6.536	0.9531
CRP			0.8663
≤5 mg/L	15 (34.09%)	20 (35.71%)	
>5 mg/L	29 (65.90%)	36 (64.28%)	

***Significant at p<0.05, 1: Wilcoxon-Mann-Whitney U Test, 2: Fisher's Exact Test, 3: Chi-Squared Test, 4: t-test

From table 16, it is evident that the variables BMI (Kg/m^2) and onset age of menarche has significant effects on PMS symptom severity. To assess the magnitude of their influence on PMS symptom severity, a non-parametric test (Wilcoxon-Mann-Whitney U Test) was used to make group comparisons. (Non-parametric test was used as the data distribution was not normal).

Table 17: Comparison of the 2 subgroups of the variable degree of symptoms in terms of BMI (Kg/m^2) (n = 100).

BMI (Kg/m^2)	Degree of Symptoms		Wilcoxon-Mann-Whitney U Test	
	Moderate	Severe	W	p value
Mean (SD)	27.02 (2.57)	28.39 (3.63)	896.500	0.019
Median (IQR)	26.5 (25-29)	28 (26-30.25)		
Range	22 - 34	18 - 35		

The variable BMI (Kg/m^2) was not normally distributed in the 2 subgroups of the variable Degree of symptoms. Thus, non-parametric tests (Wilcoxon-Mann-Whitney U Test) were used to make group comparisons.

The mean (SD) of BMI (Kg/m^2) in the Degree of symptoms: Moderate group was 27.02 (2.57). The mean (SD) of BMI (Kg/m^2) in the Degree of symptoms: Severe group was 28.39 (3.63). The median (IQR) of BMI (Kg/m^2) in the Degree of symptoms: Moderate group was 26.5 (25-29). The median (IQR) of BMI (Kg/m^2) in the Degree of symptoms: Severe group was 28 (26-30.25). The BMI (Kg/m^2) in the Degree of symptoms: Moderate ranged from 22-34. The BMI (Kg/m^2) in the Degree of symptoms: Severe ranged from 18-35.

There was a significant difference between the 2 groups in terms of BMI (Kg/m^2) (W = 896.500, p = 0.019), with the median BMI (Kg/m^2) being highest in the Degree of symptoms: Severe group.

The Box-and-Whisker plot below depicts the distribution of BMI (Kg/m^2) in the 2 groups. The middle horizontal line represents the median BMI (Kg/m^2), the upper and lower bounds of the box represent the 75th and the 25th centile of BMI (Kg/m^2) respectively, and the upper and lower extent of the whiskers represent the Tukey limits for BMI (Kg/m^2) in each of the groups.

The bar graph below depicts the means of BMI (Kg/m^2) in the 2 different groups.

Table 18: Comparison of the 2 subgroups of the variable degree of symptoms in terms of onset age of menarche (n = 100).

Onset age of menarche	Degree of symptoms		Wilcoxon-Mann-Whitney U Test	
	Moderate	Severe	W	p value
Mean (SD)	12.14 (0.63)	12.57 (1.02)	984.500	0.030
Median (IQR)	12 (12-12)	12 (12-13)		
Range	11 - 15	11 - 16		

The variable onset age of menarche was not normally distributed in the 2 subgroups of the variable degree of symptoms. Thus, non-parametric tests (Wilcoxon-Mann-Whitney U Test) were used to make group comparisons.

The mean (SD) of onset age of menarche in the degree of symptoms: Moderate group was 12.14 (0.63). The mean (SD) of onset age of menarche in the degree of symptoms: Severe group was 12.57 (1.02). The median (IQR) of onset age of menarche in the degree of symptoms: Moderate group was 12 (12-12). The median (IQR) of onset age of menarche in the degree of symptoms: Severe group was 12 (12-13). The onset age of menarche in the degree of symptoms: Moderate ranged from 11-15. The onset age of menarche in the degree of symptoms: Severe ranged from 11-16.

There was a significant difference between the 2 groups in terms of the onset age of menarche (W=984.500, p= 0.030), with the mean onset age of menarche being highest in the degree of symptoms: Severe group.

The Box-and-Whisker plot below depicts the distribution of the onset age of menarche in the 2 groups. The middle horizontal line represents the median onset age of menarche, the upper and lower bounds of the box represent the 75th and the 25th centile of onset age of menarche respectively, and the upper and lower extent of the whiskers represent the Tukey limits for onset age of menarche in each of the groups.

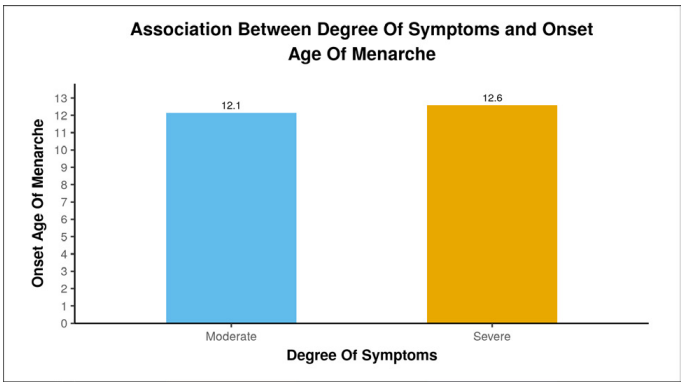

Figure 5: Means of onset age of menarche in the moderate and severe symptom groups.

Table 19: Association between lifestyle and parameters.

Parameters	Lifestyle		p value
	Active (n = 27)	Sedentary (n = 73)	
Age (Years)	25.33 ± 2.35	25.40 ± 2.94	0.7211
BMI (Kg/m^2)***	26.04 ± 3.01	28.44 ± 3.13	0.0071
BMI***			0.0042
<18.5 Kg/m^2	1 (3.7%)	0 (0.0%)	
18.5-22.9 Kg/m^2	2 (7.4%)	0 (0.0%)	
23.0-24.9 Kg/m^2	3 (11.1%)	3 (4.1%)	
25.0-29.9 Kg/m^2	19 (70.4%)	45 (61.6%)	
30.0-34.9 Kg/m^2	2 (7.4%)	23 (31.5%)	
35.0-39.9 Kg/m^2	0 (0.0%)	2 (2.7%)	
Degree of symptoms			0.0623
Moderate	16 (59.3%)	28 (38.4%)	
Severe	11 (40.7%)	45 (61.6%)	
Onset age of menarche***	12.07 ± 0.62	12.49 ± 0.96	0.0421
TLC (/cu.mm)	11285.19 ± 2948.62	10172.60 ± 2432.83	0.0874
TLC			0.1093
≤11000 /cu.mm	13 (48.1%)	48 (65.8%)	
>11000 /cu.mm	14 (51.9%)	25 (34.2%)	
ESR (mm/Hr)	50.37 ± 20.46	43.36 ± 23.54	0.1311
ESR			0.5052
≤20 mm/Hr	2 (7.4%)	11 (15.1%)	
>20 mm/Hr	25 (92.6%)	62 (84.9%)	
CRP (mg/L)	12.33 ± 15.07	8.92 ± 5.20	0.5561
CRP			0.7793
≤10 mg/L	20 (74.1%)	52 (71.2%)	
>10 mg/L	7 (25.9%)	21 (28.8%)	

***Significant at p<0.05, 1: Wilcoxon-Mann-Whitney U Test, 2: Fisher's Exact Test, 3: Chi-Squared Test, 4: t-test.

The bar graph below depicts the means of onset age of menarche in the 2 different groups.

After the effects of BMI and onset age of menarche on PMS symptom severity was studied, further correlates were also assessed; and the effects of type of lifestyle on other correlates found to significantly influence PMS severity (active/sedentary) was studied. It was then found that while the type of lifestyle (sedentary or active) did not influence the severity of PMS symptoms, it did however significantly influence BMI ($p = 0.004$ via the Wilcoxon-Mann-Whitney U Test) and onset age of menarche ($p = 0.042$ via the Wilcoxon-Mann- Whitney U Test), two factors were found to significantly affect the degree of symptoms experienced. From the above analyses, it was noted that there is a role of inflammation in causing PMS or its symptoms (owing to the rise of levels of inflammatory markers in PMS) but the inflammatory markers studied did not seem to affect symptom severity. However, the correlates BMI and onset age of menarche seemed to influence PMS symptom severity. While the type of lifestyle lived by the woman did not directly influence symptom severity, it was found to have a significant influence on BMI and onset age of menarche; thus, indirectly influencing PMS symptom severity.

Discussion

From the above analyses, it can be noted that there is a role played by inflammation in causing PMS or its symptoms (owing to the rise of levels of inflammatory markers in PMS). The findings of this study are in concert with similar studies that assessed the role of inflammation and CRP levels in premenstrual syndrome. They all show a statistically significant elevation in CRP levels in women with PMS [11]. This study also demonstrates a statistically significant rise in TLC and ESR during premenstrual syndrome. The sedimentation rate of erythrocytes in plasma are known to be influenced by the level of acute phase proteins, and thus ESR has historically been used as a laboratory test for assessment of acute phase response to inflammation [16]. Since the elevation of ESR results from RBC rouleaux formation, for which fibrinogen, the main inductor, is known to be a slow-reacting positive acute phase reactant, the role of ESR is rather definitive to imply the possibility of inflammatory processes occurring in the body [17]. Elevation of TLC is also suggestive of inflammatory processes in the body, as neutrophilia (which in turn leads to an elevated TLC), is a the popular pathological hallmark of acute inflammation. However; ESR, TLC and CRP levels did not seem to affect the severity of PMS symptoms. This could be because of the inflammatory processes causing PMS cannot be definitively categorized as acute or chronic. This is because elevated TLC (especially neutrophilia), ESR and TLC represent acute inflammation, as they are acute phase reactants or by-products of acute phase reactants. But PMS is something women continue experiencing for a major portion of their lifespan from menarche to menopause, hence the inflammation associated with it is likely chronic in nature. Hence, inflammation might play a definitive role in causing PMS but not in the degree of symptoms, as acute and chronic inflammation have varying symptomatic and pathophysiological manifestations. But the correlated BMI and onset age of menarche seemed to influence PMS symptom severity. BMI is directly correlated with leptin levels in the body by several studies [18]. We postulate that because a higher BMI is associated with increased leptin levels, it can influence PMS symptom severity as Leptin can influence PMS by directly acting on the hypothalamic-pituitary-gonadal axis.

Western countries have witnessed a downward secular trend in age at menarche, as corroborated in a Brazilian study [19], and this change has been interpreted as a consequence of environmental, socioeconomic, nutritional, and cultural factors. Thus, an association between obesity and younger age at menarche has also been observed more recently [20], while on the other hand there are reports of later menarche in athletes and girls living in rural areas [21].

According to this trend, there is a biological plausibility in the relationship between age at menarche and the manifestation of premenstrual symptoms and PMS. The results of the current study are in agreement when they show a higher PMS symptom severity among women with a lower onset age of menarche.

While the type of lifestyle lived by the woman did not directly influence symptom severity, it was found to have a significant influence on BMI and onset age of menarche, especially sedentary lifestyle contributing to decreased physical activity, disordered eating and hence obesity and an increase in BMI; thus, indirectly influencing PMS symptom severity.

All the objectives hypothesized in the beginning of this study have been adequately addressed owing to an appropriate study methodology and suitable testing methods, on a randomly selected sample population based on well-defined, narrow inclusion criteria. The findings of this study pave way for further studies to shed light on the role of lectin in influencing PMS symptom severity, as well as the significance of the onset age of menarche in various studies of female reproductive or menstrual physiology.

Conclusion

There is a role played by inflammation in causing PMS or its symptoms (owing to the rise of levels of inflammatory markers such as ESR, TLS and CRP in PMS) but the inflammatory markers studied did not seem to affect symptom severity.

However, the correlates BMI and onset age of menarche seemed to influence PMS symptom severity.

While the type of lifestyle lived by the woman did not directly influence symptom severity, it was found to have a significant influence on BMI and onset age of menarche; thus, indirectly influencing PMS symptom severity.

References

1. Barrett KE, Boitano S, Barman SM, Brooks HL (2019) Ganong's review of medical physiology twenty. (26th Edtn.), McGraw Hill, India.
2. Mayo Clinic (2021) Premenstrual syndrome (PMS) - Diagnosis and treatment. Mayo Clinic, United States.
3. Bertone-Johnson ER, Ronnenberg AG, Houghton SC, Nobles C, Zagarins SE, et al. (2014) Association of inflammation markers with menstrual symptom severity and premenstrual syndrome in young women. Hum Reprod 29: 1987-1994. https://doi.org/10.1093/humrep/deu170
4. Miller AH, Maletic V, Raison CL (2009) Inflammation and its discontents: the role of cytokines in the pathophysiology of major depression. Biol Psychiatry 65: 732-741. https://doi.org/10.1016/j.biopsych.2008.11.029
5. Evans J, Salamonsen LA (2012) Inflammation, leukocytes and menstruation. Rev Endocr Metab Disord. 13: 277-288. https://doi.org/10.1007/s11154-012-9223-7
6. Hsu WC, Araneta MRG, Kanaya AM, Chiang JL, Fujimoto W (2015) BMI cut points to identify at-risk Asian Americans for type 2 diabetes screening. Dia Care. 38: 150-158. https://doi.org/10.2337/dc14-2391
7. Dilbaz B, Aksan A (2021) Premenstrual syndrome, a common but underrated entity: review of the clinical literature. J Turk Ger Gynecol Assoc 22: 139-148. https://doi.org/10.4274/jtgga.galenos.2021.2020.0133

8. Dickerson LM, Mazyck PJ, Hunter MH (2003) Premenstrual syndrome. Am Fam Physician. 67: 1743-1752.

9. Imai A, Ichigo S, Matsunami K, Takagi H (2015) Premenstrual syndrome: management and pathophysiology. Clin Exp Obstet Gynecol 42: 123-128. https://doi.org/10.12891/ceog1770.2015

10. Biggs WS, Demuth RH (2011) Premenstrual syndrome and premenstrual dysphoric disorder. Am Fam Physician 84: 918-924.

11. Bertone-Johnson ER (2016) Chronic inflammation and premenstrual syndrome: a missing link found. J Womens Health (Larchmt) 25: 857-858. https://doi.org/10.1089/jwh.2016.5937

12. Gold EB, Wells C, Rasor MO (2016) The association of inflammation with premenstrual symptoms. J Womens Health (Larchmt) 25: 865-874. https://doi.org/10.1089/jwh.2015.5529

13. Jafari F, Amani R, Tarrahi MJ (2020) Effect of zinc supplementation on physical and psychological symptoms, biomarkers of inflammation, oxidative stress, and brain-derived neurotrophic factor in young women with premenstrual syndrome: a randomized, double-blind, placebo-controlled trial. Biol Trace Elem Res 194: 89-95. https://doi.org/10.1007/s12011-019-01757-9

14. Yen JY, Lin HC, Lin PC, Liu TL, Long CY, et al. (2020) Leptin and ghrelin concentrations and eating behaviors during the early and late luteal phase in women with premenstrual dysphoric disorder. Psychoneuroendocrinology 118: 104713. https://doi.org/10.1016/j.psyneuen.2020.104713

15. Nicolau ZFM, Bezerra AG, Polesel DN, Andersen ML, Tufik S, et al. (2017) Plasma leptin and premenstrual syndrome: a review. Obstet Gynecol Surv 72: 659-662. https://doi.org/10.1097/OGX.0000000000000502

16. Brigden ML (1999) Clinical utility of the erythrocyte sedimentation rate. Am Fam Physician 60: 1443-1450.

17. Lapić I, Padoan A, Bozzato D, Plebani M (2020) Erythrocyte sedimentation rate and C-reactive protein in acute inflammation. Am J Clin Pathol 153: 14-29. https://doi.org/10.1093/ajcp/aqz142

18. Monti V, Carlson JJ, Hunt SC, Adams TD (2006) Relationship of ghrelin and leptin hormones with body mass index and waist circumference in a random sample of adults. J Am Diet Assoc 106: 822-830. https://doi.org/10.1016/j.jada.2006.03.015

19. Kac G, Velásquez-Melendez G, Valente JG (2003) Menarche, early pregnancy and obesity in Brazilian women selected at a Health Center in Belo Horizonte, Minas Gerais, Brazil. Cad Saúde Pública 19: S111-S118. https://doi.org/10.1590/S0102-311X2003000700012

20. Biro FM, Khoury P, Morrison JA (2006) Influence of obesity on timing of puberty. Int J Androl 29: 272-277. https://doi.org/10.1111/j.1365-2605.2005.00602.x

21. Tavares CHF, Haeffner LSB, Barbieri MA, Bettiol H, Barbieri MR, et al. (2000) Age at menarche in schoolchildren from a rural community in Southeast Brazil. Cad Saúde Pública 16: 709-715.