Neutrophil-to-lymphocyte ratio and platelet-to-lymphocyte ratio are useful markers of inflammation in polycystic ovary syndrome?

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ABSTRACT

Background: Polycystic ovary syndrome (PCOS) is a heterogeneous metabolic disorder characterized by hyperinsulinemia, abnormal lipid levels, altered hormonal milieu such as elevated androgen levels, and chronic anovulation. PCOS is one of the important reasons for infertility. PCOS is the most common cause of irregular menstruation with diverse clinical manifestations affecting women of reproductive age. **Aims and Objectives:** The present study aimed to examine systemic inflammatory markers that can be detected in complete blood count, such as neutrophil–lymphocyte ratio (NLR) and platelet–lymphocyte ratio (PLR), in patients with PCOS and to compare them with healthy women. **Materials and Methods:** This case–control study was conducted in the Department of Obstetrics and Gynecology, Akash Institute of Medical Sciences and Research Centre, Devanahalli, Bengaluru, Karnataka. After following the inclusion and exclusion criteria, a total of 160 subjects were included. Among them, 80 were PCOS patients as cases and 80 were healthy women as controls. A detailed clinical and gynecological history was obtained from all subjects. Women presenting gynecology OPD in the group of 15–45 years with irregular menstrual cycles were included in the study. The diagnosis of PCOS was done according to the Rotterdam criteria, diagnosis of PCOS requires the presence of at least two of the following three symptoms: (1) oligomenorrhea/polymenorrhea; (2) biochemical hyperandrogenism (Ferriman–Gallwey scores of ≥ 8) or the presence of clinical hyperandrogenism; and (3) detection of polycystic ovaries in an ultrasonographic examination (≥ 12 follicles, 2–10 mm in size). Under aseptic conditions, venous blood samples were collected from all the subjects to assess the hormones such as follicle-stimulating hormone (FSH), luteinizing hormone (LH), inflammatory markers, white blood cells, neutrophils (N), lymphocytes (L), and platelets. NLR and PLR were calculated. Lipid profiles were obtained in the fasting state. **Results:** In the present study, BMI 24.5 ± 4.1 kg/m², total cholesterol 160.1 ± 24.2 mg/dL, triglyceride 99.1 ± 28.1 mg/dL, LDLC 99.1 ± 25.1 mg/dL, LH 8.4 ± 3.8 mIU/mL, LH/FSH ratio 1.2 ± 0.91, lymphocytes 16.30 ± 4.69, NLR 5.73 ± 1.91, and PLR 17.01 ± 7.13 were significantly increased in PCOS cases and HDLC 39.7 ± 6.2 mg/dL and FSH 6.1 ± 1.1 mIU/mL were decreased in PCOS cases compared to healthy controls. **Conclusion:** The study concludes that increased inflammatory markers such as NLR and PLR in PCOS cases compared to the age-matched individuals in the control group, suggesting the presence of chronic low-grade inflammation in PCOS cases of early reproductive age.

Key words: Inflammation; Polycystic ovary syndrome; Neutrophil–lymphocyte ratio; Platelet–lymphocyte ratio

INTRODUCTION

Polycystic ovary syndrome (PCOS) is a heterogeneous metabolic disorder characterized by hyperinsulinemia, abnormal lipid levels, altered hormonal milieu such as elevated androgen levels, and chronic anovulation. PCOS is one of the important reasons for infertility.[1-3] PCOS is the most common cause of irregular menstruation with diverse

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clinical manifestations affecting women of reproductive age. Globally, the incidence of PCOS is 5–20%, and anovulatory infertility cases account for 15%.[4]

In addition, all phenotypes of PCOS have been shown to be associated with metabolic abnormalities.[5] Emerging evidence shows that metabolic changes such as hyperandrogenism, T2DM, insulin resistance (IR), and cardiovascular disease occurring during the course of PCOS in the long term may be linked with low-grade inflammation.[6,7] It has also been reported that subclinical inflammation is one of the components of PCOS with and without obesity. Recently, studies have suggested that inflammatory markers, such as C-reactive protein (CRP), leukocytes/white blood cells (WBCs), interleukin-6, and tumor-necrosis factor-α (TNF-α), levels are elevated in PCOS patients.[8,9] Persistent and untreated subclinical chronic inflammation in PCOS is a risk factor for the development of IR.[10,11]

It has been well known that inflammation involves many complex mechanisms. A few studies have shown that an increase in inflammatory marker levels in the complete blood count (CBC) in PCOS patients may be an indicator of subclinical inflammation.[12,13] The exact mechanism and cause of chronic inflammation in PCOS have not yet been fully established, but it is suggested that the risk of IR, obesity, and cardiovascular diseases is higher in patients with the inflammatory process.

Aims and objectives
The present study aimed to examine systemic inflammatory markers that can be detected in CBC, such as neutrophil–lymphocyte ratio (NLR) and platelet–lymphocyte ratio (PLR), in patients with PCOS and to compare them with healthy women.

MATERIALS AND METHODS

This case–control study was conducted in the Department of Obstetrics and Gynecology, Akash Institute of Medical Sciences and Research Centre, Devanahalli, Bengaluru, Karnataka, for a duration of 1 year between October 2018 and October 2019. The study has been approved by the Institutional Ethics Committee (AIMSRC/BLR/IEC/15/2018, dated: September 04, 2018). In this study, a total of 160 subjects were included in the study. Among them, 80 were PCOS subjects as cases and 80 were healthy women as controls. A detailed clinical and gynecological history was obtained for all subjects. BMI was calculated for both cases and controls. Women presenting gynecology OPD in the group of 15–45 years with irregular menstrual cycles were included. The diagnosis of PCOS was done according to the Rotterdam criteria, diagnosis of PCOS

requires the presence of at least 2 of the following three symptoms: (1) oligomenorrhea/polymenorrhea; (2) biochemical hyperandrogenism (Ferriman–Gallwey scores of ≥8) or the presence of clinical hyperandrogenism; and (3) detection of polycystic ovaries in an ultrasonographic examination (≥12 follicles, 2–10 mm in size).[14]

Exclusion criteria
Women with pre-existing cardiovascular diseases, other causes for irregular menstruation such as hyperprolactinemia, thyroid dysfunction, pregnancy, Cushing syndrome, acute and chronic infections, general and systemic malignancies were excluded. Gynecological conditions such as acute and chronic PID and fibroid uterus, who are on contraceptives. Anti-androgen therapy (within 6 months), smoking, and alcohol use were excluded from the study.

Under aseptic conditions, venous blood samples were collected from all the subjects to assess the hormones such as follicle-stimulating hormone (FSH), luteinizing hormone (LH), inflammatory markers, WBC, neutrophils (N), lymphocytes (L), and platelets. NLR and PLR were calculated. Lipid profiles are obtained in the fasting state.

Statistical analysis
The data were represented in mean±SD. The Mann–Whitney U-test was used for continuous non-normally distributed variables. Data were analyzed using SPSS version 20. $P<0.05$ was considered statistically significant.

RESULTS

In the present study, BMI 24.5±4.1 kg/m², total cholesterol 160.1±24.2 mg/dL, triglyceride 99.1±28.1 mg/dL, LDLC 99.1±25.1 mg/dL, LH 8.4±3.8 mIU/mL, LH/FSH ratio 1.2±0.91, lymphocytes 16.30±4.69, NLR 5.73±1.91, and PLR 17.01±7.13 were significantly increased in PCOS cases and HDLC 39.7±6.2 mg/dL and FSH 6.1±1.1 mIU/mL were decreased in PCOS cases compared to healthy controls, as shown in Table 1.

DISCUSSION

In the present study, we evaluated inflammatory markers such as NLR and PLR in PCOS patients and healthy women. We observed that these inflammatory markers - NLR and PLR were significantly increased in PCOS patients, suggesting that there is an inflammatory process in PCOS patients.[15,16] In the present study, BMI was significantly increased in PCOS cases. It is well known that IR is associated with the pathophysiology of PCOS and obese patients are more prone to develop abnormalities in lipid metabolism and IR.[17]

Table 1: Comparison of biochemical, hematological, and inflammatory markers in PCOS cases and controls

Parameters	PCOS (n=80) (Mean±SD)	Controls (n=80) (Mean±SD)	P-value
Age (years)	22.2±2.5	21.3±2.4	0.107
BMI (kg/m^2)	24.5±4.1	22.2±3.5	0.001
Biochemical parameters			
Glucose (mg/dL)	91.2±6.8	90.6±8.9	0.454
Total cholesterol (mg/dL)	160.1±24.2	150.2±20.1	0.525
Triglyceride (mg/dL)	99.1±28.1	82.2±20.3	<0.041
HDLC (mg/dL)	39.7±6.2	46.2±12.5	<0.001
LDLC (mg/dL)	99.1±25.1	87.1±22.7	<0.051
VLDLC (mg/dL)	20.1±2.9	16.6±3.1	0.152
FSH (mIU/mL)	6.1±1.1	7.2±1.5	0.013
LH (mIU/mL)	8.4±3.8	5.4±3.4	0.001
LH/FSH ratio	1.2±0.91	0.83±0.52	0.043
Hematological parameters			
Hemoglobin (%)	10.90±2.08	11.35±1.77	0.229
WBC (10^3/μL)	14.15±4.66	12.98±3.58	0.051
Neutrophils (%)	75.56±6.10	74.86±5.85	0.545
Lymphocytes (%)	16.30±4.69	14.00±4.67	0.008
Platelets×(10^9/L)	229.1±85.2	232.24±72.13	0.624
Inflammatory markers			
NLR	5.73±1.91	4.12±1.62	0.008
PLR	17.01±7.13	16.12±7.19	0.042

NLR: Neutrophil–lymphocyte ratio, PLR: Platelet–lymphocyte ratio, WBC: White blood cells, FSH: Follicle-stimulating hormone, LH: Luteinizing hormone

IR in PCOS may lead to elevation of LDL and reduction in HDLC, which may result in atherosclerosis and hypertension.[18,19] In addition to this, obese PCOS patients may also secrete many adipokines such as adiponectin, TNF-α, and monocyte chemoattractant protein 1. These factors activate nuclear transcription factor-κB and ultimately, promote the inflammatory response.[20]

Abnormalities in the hypothalamic–pituitary–ovarian or adrenal axis are also one of the pathophysiological factors of PCOS. The disorder of gonadotropin-releasing hormone secretion mode leads to a relative increase in the release of sex hormones.[21] Ovarian estrogen is the cause of abnormal feedback mechanism, which leads to an elevation in the secretion of LH and FSH in a downward trend.[22]

In the literature, studies have reported that increased levels of proinflammatory cytokines such as IL-1α, IL-1β, TNF-α, IL-6, IL-18, and CRP in the systemic circulation. These findings suggesting evidence of chronic inflammation in PCOS.[23,24] However, in view of the expensive nature of the above inflammatory markers, there is a need to identify inexpensive and easily accessible new markers to detect chronic low-grade inflammation in PCOS, so the identification of the subset of PCOS patients who are at risk of developing complications such as IR and cardiovascular problems due to altered lipid profiles still a matter of interest for many researchers.

CBC, frequently used laboratory test in clinical practice, gives us information in terms of inflammatory markers. Inflammatory marker levels derived from CBC tests and the ratios of such marker levels to each other are evaluated as inexpensive, easily accessible, and to be used in the diagnosis

as well as follow-up of the disease course. Being a parameter obtained from CBC tests as the ratio of the neutrophil count to the lymphocyte count; NLR is correlated with the activation of diseases accompanied by chronic systemic inflammation including systemic hypertension, atherosclerosis, and chronic obstructive pulmonary disease.[25-27]

There are a few studies, which emphasizing that the CBC parameters can be used as inflammatory markers in PCOS, in the pathophysiology of which low-grade chronic inflammation is thought to have a role similar to the abovementioned diseases.[8,28]

Neutrophils are one of the important leukocyte subsets involved in controlling the inflammatory response. Their level of increase is often related to the progress and severity of inflammatory response.[29] Leukocytes are an important part of the body's immune function.

At the same time, the stress of catecholamine and cortisol hormone in the body is increased, which can lead to further reduction of lymphocytes.[30] Therefore, NLR is an inflammatory index reflecting these two important systems at the same time and has good clinical value. In this context, neutrophils, lymphocytes, NLR, and PLR are used as valuable markers of inflammation.

Kurt et al., compared the NLR, CRP, neutrophil levels, and leukocyte levels of the patients with PCOS and control groups and found that all these parameters were higher in the PCOS group. According to the recommendation of Kurt et al., this significant increase in inflammation markers is independent of obesity.[31]

In another study, Yılmaz et al., found that neutrophil and basophil counts, NLR, and mean platelet volumes as the inflammatory markers were higher in the PCOS group compared to the control group.[32] In their study, Pergialiotis et al., found out that both the PLR and NLR were correlated with some hormonal and metabolic indicators in PCOS cases.[33]

Similarly, Arpaci conducted a study on PCOS cases and BMI-matched healthy controls. Reported elevated PLR in PCOS cases than healthy controls, but was not statistically significant.[34] Yet, another study by AL-Dahhan et al., also reported increased NLR and PLR in PCOS cases compared to healthy controls. Furthermore, reported significant elevation of TNF-α, IL-6, IL-8, IL-10, and CRP in PCOS patients, suggested NLR and PLR may be good markers of inflammation.[35] Studies have reported that the endocrine, blood glucose, and lipid metabolism disorders in PCOS patients will have severe outcomes than those in the normal control group.[36]

Limitations of the study

The present study has some limitations. Less sample size and other inflammatory markers were not assessed.

CONCLUSION

The present study results may indicate increased inflammatory markers such as NLR and PLR in PCOS cases compared to the age-matched individuals in the control group. This finding confirms the presence of chronic low-grade inflammation in PCOS cases of early reproductive age.

ACKNOWLEDGMENT

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