Section: Pathology & Orthopaedics

Original Research Article

EVALUATION OF NEUTROPHIL-LYMPHOCYTE RATIO AND PLATELET LYMPHOCYTE RATIO AS A PREDICTOR OF SEVERITY IN KNEE OSTEOARTHRITIS

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ABSTRACT

Background: Two widely recognized indicators of systemic inflammation are the erythrocyte sedimentation rate (ESR) and serum C-reactive protein (CRP) concentrations. These biomarkers serve as valuable tools in identifying inflammatory diseases throughout the body. In recent times, the neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) have gained prominence as a simple and reliable method for assessing inflammation under various inflammatory conditions. The objective of this study was to assess the significance of the neutrophil-to-lymphocyte ratio and platelet-to-lymphocyte ratio as indicators of inflammation in patients with osteoarthritis. Furthermore, the study aimed to evaluate these markers in relation to various grades of the Kellgren-Lawrence classification. **Materials and Methods:** A research project involving 300 patients diagnosed with osteoarthritis was carried out in a specialized healthcare facility in northern India. The study was a collaborative effort between the Pathology and Orthopaedics departments of a tertiary care hospital, focusing on individuals clinically identified as having osteoarthritis. **Result:** Patients with osteoarthritis exhibited notable alterations in their neutrophil-to-lymphocyte ratio (NLR) (2.10±1.10) (p=<0.0001) and platelet-to-lymphocyte ratio (PLR) (139±56.1) (p=0.0052). These modifications in NLR and PLR indicate shifts in the inflammatory condition of individuals suffering from osteoarthritis, potentially signaling the advancement of the disease or the body's reaction to therapeutic interventions. **Conclusion:** The potential of the NLR and PLR is to improve patient outcomes through more precise monitoring and tailored treatment strategies, providing significant advancement in the management of inflammatory diseases, further emphasizing the role of inflammation in the pathogenesis of osteoarthritis.

INTRODUCTION

Among all arthritic conditions in humans, osteoarthritis (OA) is the most common. While this degenerative disease can impact any joint throughout the body, it most frequently targets the knee joint. Cartilage degradation leads to deformities in affected joints resulting in symptoms such as pain, swelling and stiffness.[1] The neutrophil-to-lymphocyte ratio (NLR) and platelet-lymphocyte ratio (PLR) have gained significance as crucial marker for assessing systemic inflammation and disease activity across a spectrum of conditions as a readily available and cost-effective parameter derived from routine blood tests. NLR and PLR offer a convenient and reliable measure of immune response. The ratio of neutrophils to lymphocytes serves as a significant marker for various health conditions, as these white blood cells are essential components in the body's inflammatory and stress responses. As the population ages, identifying reliable biomarkers for early diagnosis, disease monitoring, and treatment assessment becomes crucial. Consequently, NLR and PLR have gained attention for their potential utility in evaluating disease activity, particularly in inflammatory and autoimmune disorders.[2]

Contemporary research has underscored the importance of NLR and PLR across various health conditions, including heart disease, malignancies, and autoimmune disorders. In rheumatologic ailments such as ankylosing spondylitis and

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rheumatoid arthritis, NLR and PLR have demonstrated potential as indicators of disease sensitivity and treatment efficacy. Their straight forward calculation and correlation with established indicators like C-reactive protein and erythrocyte sedimentation rate make them valuable tools for healthcare professionals.[3,4]

The potential utility of NLR and PLR extends beyond rheumatologic disease offering a promising avenue for assessing inflammation and disease activity in osteoarthritis. These ratios could provide clinicians with a cost-effective and readily available tool to monitor disease progression and treatment efficacy in osteoarthritis patients by exploring the relationship between NLR, PLR and osteoarthritis severity.[5] This study aims to contribute to the development of more personalized and targeted treatment approaches for an individual suffering from this degenerative joint condition. These haematological parameters determine the potential of NLR and PLR as diagnostic indicators for osteoarthritis severity and progression.

In addition, our current understanding of OA development extends beyond cartilage degradation. It now encompasses bone remodelling beneath the cartilage, abnormal bone formation, joint capsule enlargement, and synovial membrane inflammation. This indicates that OA affects the entire joint, with inflammatory processes driving numerous pathological changes. The inflammation observed in OA differs from that seen in rheumatoid arthritis and other autoimmune conditions. It is characterized by its chronic nature, relatively mild intensity, and primary mediation through the innate immune system.[4]

The objective of this research was to explore the potential of the neutrophil-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) as markers for evaluating disease activity in individuals with osteoarthritis. Furthermore, the investigation aimed to analyze the relationship between these ratios and the progression of the disorder correlates with the level of inflammation. By comparing NLR and PLR with clinical findings, we aimed to highlight their effectiveness as reliable markers of disease progression, which could enhance treatment planning and lead to better outcomes for patients.[5]

MATERIALS AND METHODS

This cross-sectional study was implemented in a hospital environment, specifically within the Orthopaedics and Pathology Departments of a tertiary healthcare facility located in Lucknow, India. The duration of the study spanned from October 2022 to March 2023. The research focused on patients with clinically diagnosed osteoarthritis from the Department of Orthopaedics during the aforementioned period. Samples from clinically diagnosed and confirmed cases of osteoarthritis were collected from the Department of Orthopaedics in a tertiary care hospital for complete blood count (CBC) analysis in an ethylene diamine tetraacetic acid (EDTA) tube. CBC was analyzed using a five-part analyzer in the central laboratory. The samples were processed in the Department of Pathology, according to standard protocols. The total sample size was 300.[6] Approval for this investigation was granted by the Institutional Ethics Committee (IEC/IIMS&R/2022/09). Before being enrolled in the study, all participants provided their informed consent. The research was limited by its small sample size and cross-sectional design, which restricts the broad applicability of its conclusions. To confirm these findings, further research involving larger populations and multiple centres is necessary.

In standard haematological procedures, blood samples were collected using tubes containing a set amount of EDTA for complete blood count analysis. Samples were processed within two hours of collection. A 5-part ERBA analyzer, employing laser flow cytometry, was used in the central laboratory for complete blood count analysis. The analyzer underwent daily quality control checks and periodic calibration. Patient blood count data were recorded in a digital database. NLR above 3 and PLR above 150 in males and 173 in females were deemed high. The study data were collected from patients diagnosed with osteoarthritis at a tertiary care hospital's Orthopaedics OPD, based on physical examination and the Kellgren-Lawrence grading system.

The research eliminated participants who had previously been diagnosed with rheumatoid arthritis, other inflammatory joint conditions, or autoimmune disorders. Additionally, those with ongoing long-term infections, heart problems, chronic obstructive pulmonary disease, recent injuries (within two months), musculoskeletal surgeries, pregnancy, thrombosis, cancer, or chronic kidney insufficiency were not included in the study.

Statistical analysis was performed using SPSS 16.0 (SPSS Inc., Chicago, IL, USA). Distributions of variables were evaluated using the Shapiro-Wilk test. Data are presented as mean ± standard deviation. Student's t-test and Mann-Whitney U test were used to compare continuous variables. Correlations between variable pairs were analyzed using Spearman's correlation test. Differences were considered statistically significant at $p < 0.05$.

RESULTS

The study revealed that females were more commonly affected than males. The average age of individuals affected by OA was 50. Most patients were between the age group of 70-79 ($p=<0.001$) years. Grade I was the most common form of osteoarthritis. There was a significant correlation between NLR and PLR with different grades of

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osteoarthritis, proving it to be a good indicator of the progression of osteoarthritis.

Table 1: Demographic profile of patients with clinically diagnosed osteoarthritis

Demographic profile of patients			
Sex	**Osteoarthritis patients (n)**	**Test Statistic (Chi-Square)**	**P-value**
MALE	142(47.3%)	0.853	0.355
FEMALE	158(52.6%)		
AGE (Years)			
50-59	40	39.25	<0.001
60-69	61		
70-79	112		
>80	87		
Kellgren-lawrence grade (clinical characteristics)			
1	110	34.54	<0.001
2(Mild)	98		
3(Moderate)	62		
4(Severe)	30		
BMI	29±4.2	0.52	0.5

Out of 300 patients, males were 142 (47.3%), and 158 (52.6%) were females. The most commonly affected age group was between 70-79 (p=<0.001) years. The most common grade in Kellgren-Lawrence grade was grade I which was 110 (p=<0.001) as shown in [Table 1].

Table 2: Comparative analysis of inflammatory markers in knee joint osteoarthritis

Parameter	**Patients (Mean ±SD)**	**Normal range**	**One sample t-test, P-Value**
Neutrophil count	1.5±3.2	2.0-7.5x103 /µL	-7.67, <0.0001
Lymphocyte count	1.9±2.1	1.5-4.5x10/µL	-1.23, <0.222
Platelet count(103/µL)	2.83±7.2	150-400x103/µL	-324.79, <0.0001
NLR (Neutrophil/lymphocyte)	2.20±1.10	1.30-3.0	9.50, <0.0001
PLR(Platelet/lymphocyte)	139±56.1	100-300	1.97, 0.052

There was an increase in NLR (2.20±1.10) (<0.0001) and PLR (139±56.1) (0.052) correlation was observed in both NLR and PLR values of patients with clinically diagnosed OA as shown in [Table 2].

[Figure 1] NLR showed changes in different grades of osteoarthritis based on the Kellgren Lawerance grading system, grade 1 (2.4±0.6), grade 2 (3.1±0.7), grade 3 (3.4±0.9) and significant increase was observed in grade 4 (3.9±1.1) osteoarthritis as shown in [Figure 2]. The Kellgren-Lawrence (KL) grading system assesses osteoarthritis severity: Grade 0 indicates no OA radiological features; Grade 1 shows possible osteophytic lipping and uncertain joint space narrowing; Grade 2 exhibits clear osteophytes and potential joint space narrowing; Grade 3 demonstrates multiple moderate osteophytes, definite joint space narrowing, some sclerosis, and possible bone end deformity; Grade 4 presents large osteophytes, significant joint space narrowing, severe sclerosis, and definite bone end deformity.[7] NLR represents the neutrophil-to-lymphocyte ratio, PLR denotes the platelet-lymphocyte ratio, SD stands for standard deviation, and P-value indicates the statistical significance of group differences.

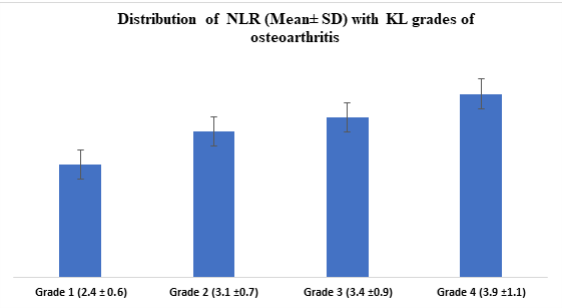

Figure 1: Distribution of NLR (Mean± SD) with KL grades of osteoarthritis

Table 3: Comparing KL grades of osteoarthritis with NLR

KL Grade Multiple Comparisons with NLR	**T-test**	**P-Value**
Grade 1 vs. Grade 2	-3.51	< 0.001
Grade 1 vs. Grade 3	-5.13	< 0.001
Grade 1 vs. Grade 4	-7.31	< 0.001
Grade 2 vs. Grade 3	-1.63	0.103
Grade 2 vs. Grade 4	-3.91	< 0.001
Grade 3 vs. Grade 4	-2.53	0.012

NLR ratios consistently increased with the progression and severity of osteoarthritis. There was a strong correlation between these markers and grades of osteoarthritis as shown in [Table 3].

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Table 4. Comparing KL grades of osteoarthritis with PLR

KL Grade Multiple Comparisons with NLR	T-test	P-Value
Grade 1 vs. Grade 2	3.33	0.001
Grade 1 vs. Grade 3	-1.51	0.132
Grade 1 vs. Grade 4	-3.03	0.003
Grade 2 vs. Grade 3	-3.15	0.002
Grade 2 vs. Grade 4	-4.63	< 0.001
Grade 3 vs. Grade 4	-1.51	0.132

PLR ratios consistently increased with the progression and severity of osteoarthritis. There was a strong correlation between these markers and grades of osteoarthritis as shown in [Table 4].

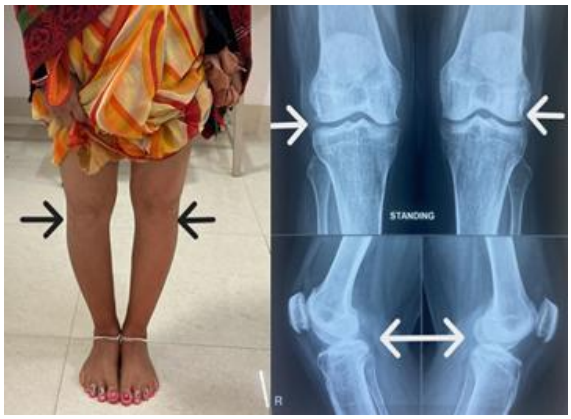

Figure 2: Patient with clinically diagnosed osteoarthritis & varus deformity of bilateral knee (black arrows) and X-rays of bilateral knees anterior-posterior (standing) and lateral views showing moderate multiple osteophytes, definite narrowing of joint space, small pseudo cystic areas with sclerosis (white arrows) and possible deformity of bone contour (Kellgren -Lawrence grade 3).

DISCUSSION

Osteoarthritis is a degenerative bone disease that leads to joint destruction. It can affect various joints but the knee joint is the most commonly impacted. It is clinically assessed by the presence of stiffness and limited joint movements.[7-9] In their studies Loeser et al,[10] and Nelson et al,[11] demonstrated that osteoarthritis affects both males and females with a higher prevalence in females. This increased susceptibility in females is attributed to hormonal changes and lower bone density. This was similar to the findings in our study, affected females were 158 (52.6%) and males were 142 (47.3%) [Table 1].

Jadhao et al,[12] in their study stated global statistics indicate that symptomatic osteoarthritis [Figure2] affects 9.6% of males and 18.0% of females aged 60 and above. This condition impairs everyday physical functions such as stair climbing and squatting, with pain being the primary symptom. They conducted a study determining the prevalence of knee osteoarthritis (OA) among rural adults over 40 years and examined the relationship between various risk factors. In our study also average age for osteoarthritis was 50 years, and the maximum cases were between the age group of 70-79 years (p=<0.001) [Table 1].

Swadi et al,[13] reported that premenopausal women with OA had a mean BMI of 29.56 ± 0.64 kg/m2. This aligns with the general finding that OA patients tend to have higher BMI [Table 1]. Vennu et al,[14]reported that severe knee OA was associated with worse physical function in adults with high BMI (>25 kg/m2) [Table 1]. In the present study also BMI was 29±4.2. The recognition of weight control [Table 1] as a key element in slowing osteoarthritis progression underscores the significance of lifestyle modifications in managing the disease. This insight can enable healthcare professionals to create more holistic treatment strategies that combine both medication-based and non-medication approaches, potentially improving patient's long-term health outcomes. The study by Deepti et al[15] revealed that osteoarthritis symptoms, including pain, transient morning stiffness, and impaired physical function in everyday tasks, can have wide-ranging effects on health, ultimately impacting an individual's quality of life.

The research emphasizes the utility of neutrophil-lymphocyte and platelet-lymphocyte ratios as effective markers for gauging osteoarthritis progression. Results demonstrate that elevated levels of these ratios exhibit a strong association with more advanced stages of osteoarthritis in affected individuals [Table 2,3]. Buyukaavci et al,[3] and Kushwaha et al[9] demonstrated that grade III and grade IV osteoarthritis exhibited significant increases in these ratios. Our study also found that neutrophil-lymphocyte ratio (2.20±1.10) [Table 2,3][Figure1] and platelet-lymphocyte ratio (139±56.1) [Table 4] increased with the progression and severity of osteoarthritis being more pronounced in grade III and grade IV osteoarthritis. Additionally, the elevated neutrophil-lymphocyte ratio and platelet-lymphocyte ratio levels in advanced osteoarthritis suggest that these ratios could be valuable indicators for monitoring drug response and guiding treatment strategies for this disease, which are consistent with those of previous studies,[16] that have elucidated the role of the NLR and PLR in various inflammatory diseases.

The simplicity and cost-effectiveness of calculating the NLR and PLR from routine blood tests render it an advantageous option for clinical practice in monitoring disease progression and further substantiating its inflammatory pathogenesis [Table 2]. Unlike CRP and ESR, which are influenced by various external factors NLR and PLR offer stable and reliable measures of the body's

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inflammatory state. The presence of dependable inflammatory indicators such as NLR and PLR [Table 2-4] could enhance diagnostic accuracy and speed, enabling earlier treatment and potentially slowing the advancement of diseases. This could be especially advantageous in general practice settings, where sophisticated diagnostic equipment might not be readily accessible.[17,18]

Salmmana F et al,[19] and Shi J et al,[20] demonstrated that platelet-to-lymphocyte ratio (PLR) is significantly associated with the radiographic grades of knee osteoarthritis [Table 4]. The PLR reflects the inflammatory response. Both PLR and NLR [Table 2-4] are potential markers for assessing the severity of knee osteoarthritis. These markers were significantly correlated with radiographic findings [Figure2] and were markedly elevated in grade III and grade IV [Table 3,4] osteoarthritis, which was in line with the findings of our study [Table 3,4]. These markers were also elevated with the progression of osteoarthritis, suggesting that NLR and PLR [Table 3,4] could serve as effective and easily accessible markers for monitoring therapeutic responses. These markers show changes in disease progression and severity, thereby offering clinicians a valuable tool for ongoing patient management.[21]

The Correlations observed between neutrophil-lymphocyte ratio and platelet-lymphocyte ratio with different grades of osteoarthritis in this study further validate their usefulness, as the neutrophil-lymphocyte ratio could serve as a complementary marker to CRP and ESR, thereby enhancing a clinician's ability to assess and monitor disease activity more comprehensively. This finding is especially relevant in a context where quick and cost-effective assessment tools are needed.[22-24] The results underscore the importance of ongoing inflammation in osteoarthritis progression and indicate that these markers of inflammation may be useful as early indicators of OA, potentially facilitating timely diagnosis and treatment. NLR and PLR [Table 3,4] show promise as biomarkers not only for tracking osteoarthritis progression but also for anticipating treatment efficacy and tailoring individualized therapeutic approaches. These indicators could prove especially beneficial in settings with limited resources or for regular check-ups, where more sophisticated or costly diagnostic methods may be scarce. Moreover, incorporating NLR and PLR into clinical protocols might facilitate the creation of more thorough osteoarthritis risk evaluation models, potentially enhancing early identification and intervention techniques.[25]

CONCLUSION

This research presents an innovative method for using the neutrophil-lymphocyte ratio (NLR) and platelet-lymphocyte ratio (PLR) as practical and dependable indicators of disease activity in osteoarthritis patients. Although existing studies have extensively explored the application of conventional inflammatory markers like erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP), the inclusion of cost-effective NLR and PLR tests provides a new angle in the clinical evaluation of disease activity for these conditions. In conclusion, neutrophil-lymphocyte ratio (NLR) and platelet-lymphocyte ratio (PLR) are promising biomarkers for disease activity in patients with osteoarthritis. Their significant correlation with established inflammatory markers and disease activity supports their potential use in clinical practice. Incorporating the NLR and PLR into the routine assessment of patients with osteoarthritis could enhance disease monitoring and improve patient management.

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