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Original Research Article

NEUTROPHIL LYMPHOCYTE RATIO AND PLATELET LYMPHOCYTE RATIO AS PREDICTOR OF MICROALBUMINURIA IN TYPE 2 DIABETES PATIENTS IN TERTIARY CARE HOSPITAL- A CROSS SECTIONAL STUDY

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ABSTRACT

Background: Aim: The aim of this study is to evaluate the predictive value of haematological indices for microalbuminuria in type 2 diabetes patients.
Materials and Methods: It was a Cross-sectional observational study conducted at ESIC Medical college & Hospital, Hyderabad, during the period 18 months from August 2022 to February 2024. The study population consisted of 150 patients who had fulfilled the inclusion and exclusion criteria and they are divided into two groups and all of them belong to type 2 diabetes mellitus. Group A diabetic patients with microalbuminuria. Group B diabetic patients without microalbuminuria.
Results: In the present study about 54% had diabetes more than 10 years and 46% had diabetes less than 10 years. Present study revealed that hose who have more NLR have more no cases of microalbuminuria compared to those who have low NLR. The association is statistically significant (P<0.05). Those who have more PLR have more no cases of microalbuminuria compared to those who have low PLR. The association is statistically significant (P<0.05).
Conclusion: This study highlights Neutrophil-to-Lymphocyte Ratio (NLR) and Platelet- to-Lymphocyte Ratio (PLR) as promising biomarkers for predicting microalbuminuria in patients with Type 2 diabetes. The research found that elevated levels of both NLR and PLR were significantly linked to a higher prevalence of microalbuminuria within this patient group. These findings suggest that NLR and PLR could potentially serve as useful tools in clinical settings to assess the risk of diabetic nephropathy. By refining the application of NLR and PLR in diabetes care, healthcare providers could potentially enhance the early detection and management of diabetic nephropathy, thereby improving overall patient care and outcomes.
Keywords: Neutrophil-to-Lymphocyte Ratio (NLR), Platelet- to-Lymphocyte Ratio (PLR), Diabetes Mellitus, Microalbuminuria, Diabetic Nephropathy.

INTRODUCTION

The increasing incidence and prevalence rates of diabetes mellitus (DM) are a serious threat to world health. Theprevalence of diabetes is exceptional and the statistics are astonishing. The burden of the diabetes epidemic becomes evident when one considers the morbidity, mortality, and cost of medical care. According to a 2023 study by the Indian Council of Medical Research-India Diabetes,10.1 crore people have diabetes.[1]
The diabetes epidemic has resulted in Diabetic Nephropathy (DN) becoming the most frequent cause of chronic kidney disease in most countries. Globally, there were 697.5 million cases of chronic kidney disease in 2017, with India ranking second in terms of incidence.[2]

Diabetic Nephropathy is a characterized by pathological levels of proteinuria, diabetic glomerular lesions, and a decline in the glomerular filtration rate (GFR) in individuals with diabetes.[3]
It is a metabolic disorder of high morbidity and mortality. Because of accelerated atherosclerosis, the risk of cardiovascular death from atherosclerosis is 10–20 times higher in people with chronic kidney disease (CKD) than in the general population. Atherosclerosis is known to be primarily caused by an increase in leukocyte count, which sets off a chain reaction of inflammation in the artery wall. In addition to leukocyte count, inflammatory markers like tumour necrosis factor-α, IL-6, IL-8, andinterleukin (IL)-1 have been connected to end organ damage in diabetes; however, their unavailability in routine clinical practice is exacerbated by their high cost and technical difficulties in application
According to Turkmen et al., platelets may contribute to atherosclerosis by binding to endothelial cells and secreting proinflammatory cytokines. Patients with higher platelet counts may have increased inflammation as a result of platelets' production of thromboxane and other mediators.[4]
India, a nation with limited resources and a rising hub for diabetes worldwide, requires affordable and reliable indicators of end organ damage.
White blood cell (WBC) counts, both total and differential and platelet count are a sensitive and straightforward laboratory test for inflammation.

Aims and Objectives

Aim

The aim of this study is to evaluate the predictive value of haematological indices for microalbuminuria in type 2 diabetes patients.

Objective

To compare levels of Neutrophil lymphocyte ratio and Platelet lymphocyte ratio in type 2 diabetes patients with and without microalbuminuria.

MATERIALS AND METHODS

Study Site: ESIC Medical College and Hospital, Hyderabad
Study Population: Patient attending General Medicine outpatient department
Study Design: Cross-sectional observational study
Sample Size: 150
Study Period: 18 months from August 2022 to February 2024

Inclusion Criteria

1. Patient who had type 2 diabetes for at least 4 years
2. Patient above 18 years

Exclusion Criteria

1. Patient with type 1 diabetes
2. Patient with acute and chronic infection or inflammation, malignancy
3. Medications affecting on the number of leucocytes like steroids, anti-inflammatory drugs
4. Knowncase of hypertension, heart disease, renal disease, blood diseases, autoimmune disorder, systemic disorder
5. Patient with condition affecting urinary protein excretion- dehydration
6. Patient who are chronic alcoholic

Ethical consideration

The study was approved by the institutional review board prior to commencement of data collection. All the patients/ legal guardians were explained about the study; the investigations, expected results and the benefits of the study. If they agreed, then the patients were enrolled in the study. It did not cause economic burden to the patients. Informed consent was taken from each patient/guardian. Data were collected by approved data collection form.

Study Procedure

Age, gender, weight, height, duration of diabetes, haemoglobin A1c (HbA1c), blood pressure, serum urea, serum creatinine, serum albumin, UACR, glomerular filtration rate (GFR), C- reactive protein (CRP), and complete blood count (CBC), which includes total white blood cell count (WBC), neutrophil count, lymphocyte count, neutrophil/lymphocyte ratio (NLR), total red blood cell count (RBC), haemoglobin (Hb), platelet count (PLT), and platelet/lymphocyte ratio (PLR), were among the demographic, clinical, and laboratory data obtained from patient file records. Weight/(height)2 was used to compute the body mass index
(BMI) (kg/m2). The 2021 CKD-EPI Creatinine was used to compute the predicted GFR. Calculating NLR involves dividing the total neutrophil count by the total lymphocyte count, while calculating PLR involves dividing the total platelet count by the total lymphocyte count. Spot urine sample collected, Urinary Microalbumin calculated using the method Immunoturbidimetry, Urine Creatinine calculated using Jaffe's method.
Microalbumin to creatinine ratio = Microalbumin in mg/L/Urinary creatinine in mg/dl X 1000
Eligible patients were classified into two groups based on the microalbuminuria, Group A Diabetes with microalbuminuria and Group B Diabetes without microalbuminuria

Statistical Analysis

Statistical Package for Social Science (SPSS for Windows, version 20.0; SPSS, Inc., Chicago, IL, USA) was used for statistical calculations. In this study, various statistical methods were used to analyse the data collected from patients with type 2 diabetes. The primary goal was to determine the relationship between inflammatory markers (neutrophil-to-lymphocyte ratio [NLR] and platelet-to-lymphocyte ratio [PLR]) and microalbuminuria.

RESULTS

The study population consisted of 150 patients who had fulfilled the inclusion and exclusion criteria and

they are divided into two groups and all of them belong to type 2 diabetes mellitus.
Group A diabetic patients with microalbuminuria.
Group B diabetic patients without microalbuminuria.
The baseline characteristics of all groups were analysed and are shown in the following tables.

The mean value for each parameter like Age, Sex, Duration of DM, Total count, Neutrophils, Lymphocytes, Platelet, NLR, PLR were calculated. NLR, PLR, HBA1C AND CRP values compared between groups and co related with microalbuminuria to find out the significance.

Table 1: Age wise Distribution of study participants

Age in years	Frequency	Percentage	Mean±S.D
21-30	20	13.33	38.87±9.21
31-40	61	40.66	
41 -50	53	35.34	
51-60	16	10.67	
TOTAL	150	100	

In the present study majority of the study participants (41%) were in the age group of 31 to 40 years. About 35% were in the age of 41 to 50 years. About13% were in the age group of 21 to 30 years. Only 10% were in the age of 51 to 60 years. Mean age is 38.87 and standard deviation is 9.21.

Table 2: Sex wise distribution of study participants

Sex	Frequency	Percentage
Male	94	62.67
Female	56	37.33
Total	150	100

About 63% were males and 37% were females.

Table 3: Body mass index among study participants

BMI	Frequency	Percentage	Mean ±S.D
18.5-22.99	11	7.33	24.38±5.48
23.0-24.99	102	68	
≥25.0	37	24.67	
Total	150	100	

In the present study about 68% were overweight,25% were obese and 7% were in the normal BMI range. Mean BMI is 24.38 and standard deviation is 5.48

Table 4: Distribution of duration of diabetes mellitus among study participants

Duration of DM in years	Frequency	Percentage
≤10 years	69	46
>10 years	81	54
Total	150	100

In the present study about 54% had diabetes more than 10 years and 46% had diabetes less than 10 years.

Table 5: Association between NLR and microalbuminuria

NLR	Microalbuminuria present (n=80)	Microalbuminuria absent (n=70)	P value
≥ 1.89	71	22	0.01
<1.89	9	48	
Total	80	70	

Those who have more NLR have more no cases of microalbuminuria compared to those who have low NLR. The association is statistically significant (P<0.05)

Table 6: Association between PLR and microalbuminuria

PLR	Microalbuminuria present (n=80)	Microalbuminuria absent (n=70)	P value
≥ 122	68	22	0.01
<122	12	48	
Total	80	70	

Those who have more PLR have more no cases of microalbuminuria compared to those who have low PLR. The association is statistically significant (P<0.05)

Table 7: Comparison of variables among patients with and without microalbuminuria

Variables	Microalbuminuria present		Microalbuminuria absent	
	Mean	SD	Mean	SD
Age	31.66	10.23	33.66	13.55
BMI	24.21	10.21	23.68	15.56
HbA1C	7.86	1.23	7.12	0.97
S.urea	28.26	6.7	26.12	4.5
Creatinine	1.12	0.12	0.98	0.13

GFR	86.08	10.5	106.5	10.75
UACR	112	12.32	9.90	2.98
CRP	2.21	0.24	0.84	0.36

DISCUSSION

This study investigates the predictive value of Neutrophil Lymphocyte Ratio (NLR) and Platelet to Lymphocyte Ratio (PLR) for microalbuminuria in patients diagnosed with Type 2 diabetes at a tertiary care hospital. Microalbuminuria is recognized as an early indicator of diabetic nephropathy, underscoring the critical need for effective biomarkers to aid in risk assessment and early intervention. NLR and PLR, reflecting systemic inflammation and thrombosis respectively, have shown promise as potential indicators in various disease states, including complications arising from diabetes.[5]

The study enrolled 150 participants with Type 2 diabetes, predominantly aged between 31 to 50 years (86.67%), with a higher proportion of males (62.67%). This was consistent with the study conducted by Sahu et.al. who found that mean age of the diabetic individuals in their study was 51 years. Sahuet.al. similarly found that nearly 60% of the participants were males.[6]

Agarwal et.al. concluded that men are more likely than women to have diabetes because of a confluence of biological, behavioural, and healthcare-use factors. Men tend to have lower visceral fat distribution and larger muscle mass biologically, which might result in decreasedinsulin sensitivity. Diabetes risk is greatly elevated by behavioural factors, including greater rates of smoking, excessive alcohol use, and poorer nutritional practices. There may also be a function for genetic predispositions influencing insulin regulation. Men's delayed diagnosis and inadequate treatment are also influenced by differences in healthcare access and adherence to medical guidance.[7]

Studies have indicated that male gender and poor glycaemic control during an extended period of diabetes are important risk factors for microalbuminuria.[8] Asghar et al found that the mean duration of diabetes among patients with microalbuminuria was 10.1 years with SD of 6.2 years.[9] The mean BMI was 24.38 ± 5.48, and diabetes duration was evenly distributed, with 54% having diabetes for more than 10 years. Martin et al found that nearly one-fourth of diabetic individuals with microalbuminuria were overweight and obese with respect to the BMI.(44) Systolic blood pressure averages 112.36 mmHg (SD 9.82), and diastolic blood pressure averages 89.22 mmHg (SD 4.12), both within normal ranges. This was in line with the study conducted by Asghar et al. (9)

We found that HbA1C is 7.86% (SD 2.39), indicating fair to poor diabetic control. Fasting blood sugar (FBS) averages 122.64 mg/dL (SD 26.60), suggesting moderately elevated levels. The current body of research indicates that albuminuria is influenced by HbA1c levels. HbA1c mayhave an impact on the development of albuminuria and renal disease in individuals with diabetes, according to findings from the DMIDS project and the FinnDiane study. Concurrently, the AdDIT study—which concentrated on teenagers—came to the same conclusion.[10-12] Serum creatinine averages 1.12 mg/dL (SD 0.16), and GFR averages 69.21 ml/min (SD 12.86), indicating normal kidney function. Spot urine ACR averages 1.76 (SD 0.28), suggesting increased risk of kidney damage. Asadujjaman et al in their stud to find out the prevalence of microalbuminuria and overt proteinuria in diabetes mellitus and their association with renal function had similar results.[13] Table 6 revealed a significant association between NLR and microalbuminuria ($P < 0.05$). Participants with an NLR ≥ 1.89 exhibited a higher prevalence of microalbuminuria (71 out of 80 cases) compared to those with an NLR <1.89 (9 out of 70 cases). This finding suggests that elevated NLR levels may serve as an indicator of systemic inflammation contributing to renal microvascular complications in Type 2 diabetes.

This was corroborated by Li et al who showed that the frequency of albuminuria increased 1.01 times and 4.75 times for every unit increase in NLR and MLR, respectively (NLR: OR = 2.01; 95% CI: 1.10, 3.68;

MLR: OR = 5.75; 95% CI: 1.76, 18.76). People with T2DM who have higher NLR levels should be aware of kidney function. This may be dueto the fact that the readily available and reasonably priced NLR,[15] reflects both innate immunity, which is mediated by neutrophils, and adaptive immunity, which is mediated by lymphocytes. Furthermore, NLR stability is better and less impacted by pathological and physiological conditions.[14]

Similarly, Kahraman et al in their study to find the relationship between neutrophil-to-lymphocyte ratio and albuminuria in type 2 diabetic patients, found that for the microalbuminuria group, the neutrophil/lymphocyte ratio was observed to be 2.6 ± 1.0, indicating a significant increase compared to the normoalbuminuric group ($p = 0.003$). This suggests a potential association between microalbuminuria and elevated neutrophil/lymphocyte ratios, highlighting inflammatory processes in this subgroup.[15]

Similarly, Table 7 demonstrated a significant association between PLR and microalbuminuria ($P < 0.05$). Individuals with PLR ≥ 122 had a higher prevalence of microalbuminuria (68 out of 80 cases) compared to those with PLR < 122 (12 out of 70 cases). Elevated PLR levels, reflecting increased thrombotic and inflammatory processes, appear to correlate with the presence of microalbuminuria, suggesting a potential role in diabetic kidney disease progression. Similarly, Li L etal found that PLR levels in patients with type 2 diabetes were linked

with renal function indicators and increased with increasing albuminuria.[16]

In addition, Duan etal found that PLR was revealed to be an independent risk factor for renal development and was connected with the prognosis of Diabetic Kidney Disease (DKD) in a follow-up analysis of 167 individuals with biopsy-proven DKD.[17] Milasetal highlighted that multiple inflammatory pathways are responsible for the pathological alterations in kidney structure caused by the NLRP3 inflammasome and other inflammatory cytokines, such as interleukins and tumor necrosis factors. They worsen renal fibrosis, tubular damage, and glomerular sclerosis. They also raise the excretion of albumin in the urine.[18-19]

The findings emphasize the clinical relevance of NLR and PLR as accessible biomarkers for identifying Type 2 diabetes patients at risk of developing microalbuminuria. Integration of these ratios into routine clinical practice could potentially enable early detection and intervention, thereby potentially slowing the progression of diabetic complications. Future research directions should focus on longitudinal studies to validate these findings across diverse patient populations and elucidate the underlying mechanisms through which NLR and PLR exert their predictive value.

CONCLUSION

This study highlights Neutrophil-to-Lymphocyte Ratio (NLR) and Platelet- to-Lymphocyte Ratio (PLR) as promising biomarkers for predicting microalbuminuria in patients with Type 2 diabetes. The research found that elevated levels of both NLR and PLR were significantly linked to a higher prevalence of microalbuminuria within this patient group. These findings suggest that NLR and PLR could potentially serve as useful tools in clinical settings to assess the risk of diabetic nephropathy.

The study implies that monitoring NLR and PLR levels could help clinicians identify Type 2 diabetes patients who are at greater risk of developing microalbuminuria. This early detection could enable timely interventions aimed at managing kidney complications and improving patient outcomes. Nevertheless, further research is needed to fully understand the underlying mechanisms that connect NLR, PLR, and microalbuminuria. Additionally, larger studies are necessary to validate these biomarkers' predictive capabilities across different patient populations and clinical settings. By refining the application of NLR and PLR in diabetes care, healthcare providers could potentially enhance the early detection and management of diabetic nephropathy, thereby improving overall patient care and outcomes.

Limitation of Study

limitations include the cross-sectional design, which precludes establishing causality, and the single-centre setting, which may limit the generalizability of findings to broader populations.

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