A COMPARATIVE STUDY EVALUATING THE ROLE OF UTILITY OF RED BLOOD CELL COUNT, RED CELL DISTRIBUTION WIDTH, AND NEUTROPHIL/LYMPHOCYTE RATIO AS PROGNOSTIC MARKERS IN TYPE 2 DIABETES MELLITUS PATIENTS WITH AND WITHOUT MICROVASCULAR COMPLICATIONS

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ABSTRACT

Aim: The aim of the present study was to evaluate the role of utility of Red Blood Cell count, Red Cell Distribution Width, and Neutrophil/Lymphocyte Ratio as prognostic markers in type 2 Diabetes Mellitus patients with and without microvascular complications.

Methods: The present study is a cross-sectional analytical study. It was undertaken in the Department of Medicine of the parent institute Dr D.Y Patil Medical College, Hospital and Research Centre, Pimpri, Pune, during the period of October 2020 to September 2022 with the aim to evaluate the role of utility of Red Blood Cell count, Red Cell Distribution Width, and Neutrophil/Lymphocyte Ratio as prognostic markers in type 2 Diabetes Mellitus patients with and without microvascular complications.

A total of 300 patients were included in the study- 150 cases, with microvascular complications, and 150 controls, without microvascular complications.

Results: It was evident from the results that majority of patients were in the 51- 60 age group (29%) followed by 61-70years (25%) and 71-80years of age (20%).

The mean RBC in Microvascular complication group was 4.64 and in non-microvascular complication group was 3.17.

The mean RDW in Microvascular complication group was 15.60 and in non-microvascular complication group was 11.26.

The mean NLR in Microvascular complication group was 4.03 and in non-microvascular complication group was 1.37.

The association between these groups is statistically significant (P<0.05).

Conclusion: Thus, we concluded that in this study it is evident that Red Blood Cell count, Red Cell Distribution Width, and Neutrophil/Lymphocyte Ratio can be used as prognostic markers in type 2 Diabetes Mellitus patients with and without microvascular complications.

Keywords: RBC, RDW, NLR, Diabetes mellitus, microvascula

INTRODUCTION

Type 2 diabetes mellitus (DM), a chronic metabolic condition, is characterised by hyperglycemia, insulin resistance and a relative decrease of insulin production. The interaction of behavioural, environmental, and genetic risk factors contributes to type 2 diabetes.[1]

Diabetic complications can be categorised into microvascular and macrovascular complications. Macrovascular problems include cardiovascular disease, stroke, and peripheral artery disease.

Microvascular sequelae include neuropathy, nephropathy, and retinopathy.[2,3,4] The most common and dangerous eye complication of uncontrolled diabetes is diabetic retinopathy. Glaucoma and cataract can also occur in longstanding diabetics.

Early diagnosis and rapid institution of treatment can prevent diabetic retinopathy-related blindness.[5,6] Diabetic nephropathy is considered in the presence of albuminuria and a progressive reduction in glomerular filtration rate. [7,8]

Diabetic neuropathy is considered where there is peripheral nerve involvement following the elimination of alternative causes. It is divided into five categories: focal, diffuse, sensory, motor, and autonomic neuropathy.

In order to provide effective therapy, which improves people's lives, early diagnosis of diabetic retinopathy, nephropathy, and neuropathy using competent screening and diagnostic procedures is essential.[9]

Hematological parameters that have earlier been used as indicators of endothelial dysfunction and inflammation include white blood count (WBC), mean platelet volume (MPV), platelet distribution width (PDW), platelet count, and neutrophil to lymphocyte ratio (NLR). Elevated white blood cell count (WBC) is a classic marker that has been linked to diabetes and a number of risk factors for cardiovascular disease. [10]

Thus, this study was designed to evaluate the role of utility of Red Blood Cell count, Red Cell Distribution Width, and Neutrophil/Lymphocyte Ratio as prognostic markers in type 2 Diabetes Mellitus patients with and without microvascular complications.

MATERIALS AND METHODS

The present study was undertaken in the Department of Medicine of the parent institute Dr. D.Y Patil Medical College, Hospital and Research Centre, Pimpri, Pune, during the period of October 2020 to September 2022 with the aim to evaluate the role of utility of Red Blood Cell count, Red Cell Distribution Width, and Neutrophil/Lymphocyte Ratio as prognostic markers in type 2 Diabetes Mellitus patients with and without microvascular complications.

A total of 300 patients were included in the study- 150 cases, with microvascular complications, and 150 controls, without microvascular complications.

Inclusion criteria

Cases: Type II Diabetes mellitus patients between the ages 30-80 years with established:

1)   Diabetic retinopathy or

2)   Diabetic nephropathy or

3)   Diabetic neuropathy

Controls

Type II Diabetes mellitus patients without any established microvascular complications, between the ages of 30-80 years

Exclusion criteria

•   Type I DM patients

•   Type II DM patients with iron deficiency/megaloblastic anaemia/recent blood loss

•   Type II DM patients with any evidence of sepsis/infectious disease in prior 4 weeks

•   Type II DM patients with any form of arthritis on NSAIDs/active GI ulcer

•   Type II DM patients with hypertension

•   Type II DM patients who are pregnant

•   DM patients with known collagen vascular diseases

ETHICS AND CONSENT

The study protocol was approved by the institutional Ethical Committee. All the study participants and/or relatives were also informed about the study procedure and the information required from them for the study. All subjects and/or relatives were explained in detail about the potential benefits and risk of the study. A voluntary informed written consent was obtained from the relatives whose patients were included in the study.

The consent form was available in English, Hindi and Marathi. In the case of illiteracy, the consent of the participants / parents was obtained in front of the witnesses. Third parties not

directly involved in the study were treated as witnesses. At all levels, strict confidentiality was maintained regarding the personal information of participants and the investigation information under the study.

Clinical evaluation: Informed consent was obtained from all the selected participants. Detailed medical history, including personal data, chief complaints, medical history, and personal habits, was obtained.

Lab Investigations:

• Random BSL, BSL F/PP1/PP2

• CBC- Estimation of RDW, RBC count, NLR

• Liver function tests, renal function tests, serum electrolytes

• Fasting lipid profile, HbA1c

• Serology – HIV/HBsAg/HCV

Investigations specific to microvascular complications of diabetes:

• Fundoscopy

• Microalbuminuria estimation, Urine Albumin to Creatinine Ratio

• Clinical examination for diabetic neuropathy–testing of reflexes, monofilament testing, 40-gauge needle test, vibration perception threshold

• Nerve conduction velocity (for patients who exhibit signs and symptoms of diabetic neuropathy)

Statistical Analysis

The data was entered in Microsoft Excel and analyzed using SPSS software. Continuous variables were summarized using mean (SD), and the categorical variables (like sex), were summarized using proportion.

To find a statistically significant association between each of the three parameters (Red Blood Cell count, Red Cell Distribution Width, and Neutrophil/Lymphocyte Ratio) and type 2 Diabetes Mellitus patients with and without microvascular complications t test was applied.

 To study the association between categorical variables, (*Pearson's) Chi square test was applied as a test of significance. P value <0.05 will be considered as statistically significant.

RESULTS

Table 1: Demographic characteristics

Sex	Frequency	Percentages
Female	117	39.00%
Male	183	61.00%
Age groups		
41-50	78	26%
51-60	87	29%
61-70	75	25%
71-80	60	20%
Groups		
Case	150	50.00%
Control	150	50.00%

It was observed that majority of the patients were male with proportion of 61%. It was evident from the above table that majority of patients in this study were in the 51- 60 age group (29%) followed by 61-70 years (25%) and 71-80years of age (20%). The majority of patients in this study were in the 51-60 age groups.

Table 2: Descriptive analysis of fasting blood sugar, postprandial blood sugar, RBC and creatinine in study population

Parameter	Mean ± SD	Median	Minimum	Maximum
Fasting Blood Sugar	194.13 ± 61.05	181.00	120.00	367.00
Postprandial Blood Sugar	267.99 ± 136.06	217.00	111.00	778.00
RBC	4.24 ± 0.64	4.20	2.90	5.50
Creatinine	1 ± 0.33	1.00	0.20	1.90

The mean difference of fasting blood sugar, Postprandial Blood Sugar, RBC levels and Creatinine were 194.13 ± 61.05, 267.99 ± 136.06, 4.24 ± 0.64 and 1 ± 0.33 respectively.

Table 3: Descriptive analysis of HbA1c and Red Cell Distribution Width, NLR (Neutrophil/Lymphocyte Ratio) in study population

Parameter	Mean ± SD	Median	Minimum	Maximum
Hba1C	9.65 ± 2.44	9.15	6.80	17.50
RDW	14.07 ± 6.94	14.00	12.00	132.00
NLR	2.6 ± 1.65	1.81	0.89	8.78

The mean difference of Hba1C, RDW and NLR were 9.65 ± 2.44, 14.07 ± 6.94 and 2.6 ± 1.65 respectively.

Table 4: Comparison of RBC (Red Blood Cell) Count, RDW and neutrophil lymphocyte ratio among study participants

RBC	Mean	SD	P value
Microvascular complication	4.64	0.67	0.04
No microvascular complication	3.17	0.62	
RDW			**P value**
Microvascular complication	15.60	3.21	0.01
No microvascular complication	11.26	1.24	

NLR			P value
Microvascular complication	4.03	0.82	0.01
No microvascular complication	1.37	0.57	

The mean RBC in Microvascular complication group is 4.64 and in non-microvascular complication group is 3.17. Association is statistically significant (P<0.05).

The mean RDW in Microvascular complication group is 15.60 and in non-microvascular complication group is 11.26. Association is statistically significant (P<0.05).

The mean NLR in Microvascular complication group is 4.03 and in non-microvascular complication group is 1.37. Association is statistically significant (P<0.05).

DISCUSSION

Type 2 diabetes mellitus (T2DM) is an important public health problem in both developed and developing nations at present.[11] Its microvascular complications consist of nephropathy, retinopathy, and neuropathy, besides macrovascular complications (cerebrovascular disease, coronary artery disease and peripheral artery disease).[12]

In T2DM, hyperglycaemia and insulin resistance are related with the stimulation of proinflammatory processes.[13] Pro-inflammatory responses give rise to low-grade inflammation.[14] Neutrophil lymphocyte ratio (NLR) is a significant indicator of systemic inflammation.[15,16]

Various studies have been conducted on the association of haematological parameters with diabetic microvascular complications, which can be compared with the present study.

According to a retrospective research study by Onalan E. et al., conducted between May 2018 to October 2018, comparisons between diabetics and healthy people reveal that specific hematologic parameters may be linked to microvascular complications. This study further suggested that basic complete blood counts, which can measure hematologic parameters including haematocrit, mean platelet volume and Neutrophil Lymphocyte Ratio can be used as inexpensive indicators of diabetic microvascular problems.[17]

Chittawar et al examined the likelihood of NLR as a predictor of CAD and microvascular consequences in type 2 diabetes. Clinical and anthropometric tests were also performed on

consecutive T2DM patients (CAD). The findings demonstrated that patients with higher NLR quartiles had longer-lasting diabetes, lower GFR, and more episodes of nephropathy, albuminuria, retinopathy, and CAD. As a result, the study concluded that two factors that predicted microvascular problems best were NLR and BMI. NLR was discovered to be an inexpensive, simple to use, and accurate predictor of nephropathy, retinopathy, and CAD in Indian T2DM.[18]

In South West Ethiopia, a research study conducted by Asmamaw et al, evaluation of red blood cell characteristics as a biomarker for long-term glycaemic surveillance in T2 DM patients was the main goal. It was a cross sectional study. 60% of T2DM patients had poor glycaemic control. HbA1c level and red cell distribution width had a positive correlation and an inverse relationship, respectively, with red blood cell count.[19]

In 2017, Khandare et al conducted cross-sectional observational research to clarify the relationship between the neutrophil-lymphocyte ratio and diabetic nephropathy. 115 individuals who had been diagnosed were included in the research. The difference between the mean NLR for the normal group and the DN (diabetic nephropathy) group, 2.83 +/- 0.85, was extremely significant (P 0.001). The investigation came to the conclusion that NLR and DN had a meaningful relationship.[20]

Fawwad et al conducted a retrospective study from the years 2005 to 2016, the aim being to determine the relationship between diabetic microvascular problems and the neutrophil-to-lymphocyte ratio in participants with type 2 diabetes. The NLR is a reliable, affordable, and easily accessible marker of inflammation that has been shown in this study to be a significant predictor of the existence of microvascular complications in type 2 diabetic individuals.[21]

Xiong et al in 2017 conducted a study among 809 patients, in order to find out if RDW can be used as a prognostic marker for patients of type 2 diabetes mellitus. According to the results of the study, having a high level of RDW increases the likelihood of developing diabetic nephropathy and predicts a worse prognosis.[22]

Ma et al conducted a retrospective study between the years 2014 to 2019 in Fudan University, China. A study of the relationship between RDW and diabetic retinopathy (DR) was conducted. RDW values were found to be significantly greater in DR patients, and a higher RDW was found to be associated with a higher prevalence of DR in DM patients.[23]

At Safdarjung hospital, New Delhi, Bhattacharya et al conducted a cross-sectional study to investigate the NLR in type 2 diabetes patients and its relationship to microvascular problems in these individuals. The strongest predictor of diabetic nephropathy and diabetic neuropathy was discovered to be NLR. Additionally, NLR was noticeably greater in DM patients with many microvascular complications. NLR may be used as a straightforward metric to predict the occurrence of diabetic microvascular problems, the study concluded.[24]

CONCLUSION

Thus, we concluded that in this study that Red Blood Cell count, Red Cell Distribution Width, and Neutrophil/Lymphocyte Ratio can be used as prognostic markers in type 2 Diabetes Mellitus patients with and without microvascular complications. People with type 2

DM are more prone to a variety of short- and long-term complications, which frequently result in early mortality. They are likelier to have higher morbidity and death rates due to the condition's prevalence, insidious onset, and delayed diagnosis, especially in resource-poor developing nations.

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