

Association Between Glycemic Control and Hematological Indices in Type 2 Diabetes Mellitus: A Cross-sectional Study

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ABSTRACT

Background: Type 2 diabetes mellitus (T2DM) is a major preventable cause of morbidity due to its chronic complications. Emerging evidence suggests that hyperglycemia affects hematological parameters, which may serve as accessible indicators of disease severity and associated risks.

Objective: This study aimed to evaluate the impact of glycemic control on hematological indices among patients with T2DM.

Materials and Methods: A cross-sectional study was conducted involving 300 patients with T2DM. Participants were categorized into two groups based on glycemic control: Group A with controlled diabetes (HbA1c <7%) and Group B with uncontrolled diabetes (HbA1c >7%). Data on clinical history, laboratory findings, and hematological parameters were collected and analyzed.

Results: Of the 300 patients, 115 (38.3%) had controlled diabetes and 185 (61.7%) had uncontrolled diabetes. Patients with poor glycemic control showed significantly higher levels of total leukocyte count (TLC), monocytes, basophils, red cell distribution width (RDW-CV), platelet distribution width (PDW), mean platelet volume (MPV), platelet large cell ratio (PLCR), plateletcrit (PCT), neutrophil-to-lymphocyte ratio (NLR), and platelet-to-lymphocyte ratio (PLR) (p < 0.05). Conversely, red blood cell (RBC) count, hemoglobin, mean corpuscular volume (MCV), and mean corpuscular hemoglobin (MCH) were significantly lower in this group (p < 0.05). HbA1c levels showed positive correlations with TLC, neutrophils, basophils, PDW, MPV, PCT, NLR, and PLR, and negative correlations with RBC count, hemoglobin, hematocrit (HCT), and MCV.

Conclusion: Poor glycemic control in T2DM is associated with increased inflammatory and prothrombotic markers. Routine hematological indices, being cost-effective and readily available, can serve as useful indicators to assess the severity and complications of diabetes..

Keywords: Type 2 Diabetes Mellitus, Glycemic Control, Hematological Parameters, HbA1c, Inflammation, Platelet Indices, Neutrophil-to-Lymphocyte Ratio, Anemia, Chronic Inflammation, Diabetes Complications

1. INTRODUCTION

The term "diabetes mellitus" describes a metabolic ailment of a couple of etiology characterized by persistent hyperglycemia due to defective secretion and/or action of insulin, which leads to an imbalance in carbohydrate, fat, and protein metabolism. Diabetes mellitus is one of the most common chronic illnesses in almost all nations. It is increasing rapidly in every part of the world, to the extent that it has now assumed an epidemic proportion.

It is estimated that 54 crore adults in the age-group of 20–79 years are living with (T2DM). This number is expected to rise to 64.5 crore by 2030 and 78.4 crore by 2045. 1DM caused 6.7 million deaths in 2021. The prevalence of type 2 DM varies significantly from one geographical location to another due to environmental and lifestyle risk factors. Several sedentary lifestyle-related factors are responsible for the increasing prevalence of T2DM in developing countries, including overweight [body mass index (BMI) = 25 kg/m²], obesity (BMI = 30 kg/m²), physical inactivity, and increased caloric intake. These are all shown to be primary risk factors for the onset of T2DM, irrespective of age and sex.³

Dr Nischal Garg,, Dr Tushar Krishnan Saini, Dr Suryavir

Diabetes mellitus is a very common metabolic disease worldwide and, particularly in India, where it has been called "The Diabetes Capital of the World." Diabetes mellitus itself results in a range of complications affecting all major systems of the body.

Diabetes mellitus is associated with macrovascular as well as microvascular complications. Type 2 diabetic patients are at a higher risk of developing cardiovascular diseases (CVDs) and stroke. T2DM is the most common risk factor for the development of coronary artery disease (CAD), along with other risk factors like hypertension, increased low-density lipoprotein cholesterol (LDL), and weight gain.⁴,⁵

Inflammation is closely related to the secretory defect of beta cells as well as insulin resistance. Circulating markers of inflammation can reduce the function of beta cells through secretory dysfunction or uncontrolled apoptosis.⁶ A classical inflammatory marker includes total leukocyte count (TLC); it alters carbohydrate metabolism and

increases insulin resistance, which predisposes individuals to T2DM and various cardiovascular risks.⁷ In addition to atheroma formation, the combination of hypercoagulability, impaired fibrinolysis, and impaired vasodilation may further increase the risk of vascular occlusion and cardiovascular events in diabetes.⁸

Large platelets are more active than smaller platelets and carry more prothrombotic factors, including thromboxane A2, thromboxane B2, platelet factor 4, serotonin, and platelet-derived growth factor. This suggests that changes in platelet count and mean platelet volume (MPV) reflect the state of thrombogenesis. It has been shown that MPV is significantly higher in diabetic populations. Platelet activity and aggregation ability, which are important in atherogenesis and thrombogenesis, can be easily predicted through various hematological indices, such as MPV, plateletcrit (PCT), and platelet count as part of a complete blood count.

Within the closing many years, platelet-to-lymphocyte ratio (PLR) and neutrophil-to-lymphocyte ratio (NLR) were introduced as capability markers to determine inflammation in cardiac and noncardiac issues. 11 Starting from this point, we hypothesized that the elevated hazard of diabetic vascular complications in sufferers with bad glycemic manage can be associated with impaired numerous hematological indices, which can be recovered through tight glycemic control. Consequently, we need to evaluate the association among hematological indices, glycemic control, and microvascular complications in type 2 diabetic sufferers.

2. MATERIALS AND METHODS

Study Design

This cross-sectional, observational, comparative analysis was conducted on a total of 300 patients with T2DM from January 2024 to December 2024, who visited for outpatient department (OPD) or inpatient department (IPD) at Muzaffarnagar Medical College, India. This study was permitted by the Institutional Ethics Committee of this institute. In this study, apparently healthy diabetic sufferers were included. About 115 sufferers with type 2 DM and controlled sugar [hemoglobin A1c (HbA1c) <7%] were considered in group A, and 185 patients with uncontrolled T2DM (HbA1c >7%) were considered Informed consent was received from each subject included in Patients with a history of hematological disease and thrombotic disease, on anticoagulant medications, chronic kidney disease, chronic liver disease, malignancy including leukemia and lymphoma, male patients with Hb <12.5 gm/dL, female patients with Hb <11.5 gm/dL, current or former smokers, pregnancy, and acute infectious disease altering these indices were excluded from this study.

Methods

Patients presenting or diagnosed with T2DM coming to the outpatient or emergency department were identified and enrolled in the study. Any patients with an HbA1c level >6.5 gm% or a history of taking any oral hypoglycemic agents and/or insulin were considered diabetic patients. Data including blood pressure, height and weight measurements, age, gender, accompanying disease history, smoking habits, medication, and medical history were recorded. The Quetelet index formula was used to calculate BMI. Complete blood count from the advanced hematology lab, biochemical profile including blood glucose, creatinine, total lipid profile, HbA1c, and spot urinary albumin creatinine ratio were obtained. NLR and PLR were calculated as the ratio of neutrophils to lymphocytes and platelets to lymphocytes, respectively. Sufferers with DM have been evaluated concerning metabolic regulation and nephropathy on the premise of morning spot urinary albumin/creatinine ratio. All of the data were gathered on a predesigned proforma. Information thus accrued has been entered in the form of a master chart. The information has been categorized and analyzed according to the objectives and goals of the study. Inferences were drawn using suitable tests of significance.

Statistical Analysis

Collected data were tabulated in Microsoft Excel and arranged for analysis. Quantitative data were expressed as mean \pm SD, and qualitative data as percentages. For the comparison of quantitative and qualitative data, the t-test and Chi-squared test were used, respectively. The Pearson correlation coefficient was used to determine correlations between variables. A p-value

of <0.05 was considered significant.

Results

A total of 300 patients enrolled in this study. On the basis of their glycemic control, the study population was divided into two groups, that is, group A with controlled sugar (HbA1c <7%) and group B with uncontrolled sugar (HbA1c <7%). Among the selected study population, 115 patients (38.33%) have controlled blood sugar with HbA1c <7 gm%, and 185 (61.67%) patients have uncontrolled blood sugar with HbA1c >7 gm%.

The mean age was significantly higher in the uncontrolled diabetic group (55.56 ± 15.58 years) as compared to the controlled diabetic group (50.94 ± 11.4 years) with a p-value of 0.0063 (Table 1). Men are able to control their diabetes efficiently as compared to their female counterparts (p < 0.001). The duration of diabetes and systolic blood pressure were found to be significantly higher in the uncontrolled diabetic group as compared to the controlled diabetic group (p < 0.001).

Laboratory Findings

As per indicators of glycemic control, blood glucose and HbA1c were found to be significantly higher among patients with poorly controlled T2DM as compared to those with well-controlled T2DM (p < 0.001). Diabetic risk factors like total cholesterol, triglycerides, and LDL cholesterol were also found to be significantly higher among patients with poorly controlled T2DM as compared to those with well-controlled T2DM (p < 0.05). Diabetes-related microvascular complications like urine albumin-to-creatinine ratio (UACR) were significantly higher among patients with poorly controlled T2DM as compared to those with well-controlled T2DM (p < 0.001). Among white blood cell indices, inflammatory markers like total WBC count, monocyte count, and basophil count were significantly higher in patients with poorly controlled T2DM as compared to those with well-controlled T2DM (p < 0.05) (Table 1).

Red blood cell indices like RBC count, hemoglobin, mean corpuscular volume (MCV), and mean corpuscular hemoglobin (MCH) were significantly lower among the uncontrolled diabetic group as compared to the controlled diabetic group (p < 0.05), while red cell distribution width (RDW-CV) was found to be significantly higher among the uncontrolled diabetic group ($14.24 \pm 2.87\%$) as compared to the controlled diabetic group ($13.12 \pm 2.56\%$) (p = 0.0007). Other indices like platelet distribution width (PDW), MPV, platelet large cell ratio (P-LCR), PCT, NLR, and PLR were also significantly higher among patients with poorly controlled T2DM as compared to those with well-controlled T2DM (p < 0.001).

Parameters	Group A $HbA1c < 7 gm/dL$ $(n = 115)$	Group B $HbA1c >7 gm/dL$ $(n = 185)$	p-value
Age (year)	50.94 ± 11.4	55.56 ± 15.58	0.0063
Gender			
Male	102 (88.69%)	108 (58.38%)	< 0.001
Female	13 (11.31%)	77 (41.62%)	
Duration of diabetes (years)	4.6 ± 1.4	6.8 ± 2.1	< 0.001
BMI (kg/m²)	28.44 ± 1.29	28.57 ± 1.37	0.4146
Systolic blood pressure (mm Hg)	131.50 ± 5.10	137.29 ± 5.83	< 0.001
Diastolic blood pressure (mm Hg)	88.88 ± 10.56	89.17 ± 6.35	0.7666
Fasting blood glucose (mg/dL)	103.6 ± 8.64	165.4 ± 25.76	< 0.001
Postprandial blood glucose (mg/dL)	153.5 ± 10.65	214 ± 32.44	< 0.001
HbA1c (%)	6.32 ± 0.64	8.94 ± 2.37	< 0.001
UACR (mg/gm)	247 ± 139.06	704.24 ± 231.61	< 0.001
Total cholesterol (mg/dL)	174.94 ± 15.78	183.00 ± 10.83	< 0.001
Triglycerides (mg/dL)	94.55 ± 13.69	98.81 ± 14.28	0.0112
LDL-C (mg/dL)	125.06 ± 19.59	137.18 ± 22.68	< 0.001
HDL-C (mg/dL)	41.88 ± 5.07	41.10 ± 6.36	0.2665
White blood cells indices			
Total WBC count (109/L)	6.60 ± 1.88	7.15 ± 1.91	0.0153
Neutrophil count (109/L)	3.74 ± 1.34	4.12 ± 2.33	0.1124
Lymphocyte count (109/L)	2.20 ± 1.01	2.01 ± 1.29	0.1800
Monocyte count (109/L)	0.42 ± 0.34	0.71 ± 0.51	< 0.001
Eosinophil count (109/L)	0.13 ± 0.21	0.16 ± 0.12	0.1165
Basophil count (109/L)	0.11 ± 0.07	0.15 ± 0.13	0.0026
Red blood cells indices			
RBC count (10 ¹² /L)	4.31 ± 1.12	3.98 ± 1.17	0.0164
Hemoglobin (gm/dL)	13.05 ± 2.33	12.3 ± 2.42	0.0086
HCT (%)	46.14 ± 9.62	44.29 ± 10.31	0.1222
MCV (fL)	89.12 ± 15.32	84.25 ± 22.87	0.0444
MCH (pg)	28.89 ± 3.91	27.98 ± 3.84	0.0484
MCHC (%)	34.50 ± 3.38	34.12 ± 3.53	0.3576
RDW-CV (%)	13.12 ± 2.56	14.24 ± 2.87	0.0007
Platelet indices			
Platelet count (109/L)	246.50 ± 19.52	251.87 ± 26.58	0.0618
PDW (fL)	9.32 ± 1.82	10.11 ± 1.98	0.0006
MPV (fL)	9.12 ± 2.12	9.74 ± 2.54	0.0296
P-LCR (%)	16.94 ± 4.12	19.84 ± 5.24	< 0.001
PCT (%)	0.22 ± 0.04	0.25 ± 0.05	0.0037
NLR	1.70 ± 1.33	2.04 ± 1.50	0.0473
PLR	112.04 ± 19.33	125.30 ± 20.60	< 0.001

BMI, body mass index; HCT, hematocrit; Hgb, hemoglobin; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; MCV, mean corpuscular volume; MPV, mean platelet volume; NLR, neutrophil to lymphocyte ratio; PDW, platelet distribution width; PLC-R, platelet large cell ratio; PLR, Platelet to lymphocyte ratio; RBC, red blood cells; RDW-CV, red cell distribution width coefficient of variation; RDW-SD, red cell distribution width standard deviation; SD, standard deviation; UACR, urine albumin creatinine ratio; WBC, white blood cells; p-value < 0.05, statistically significant

Association of Hematological Indices with Parameters of Glycemic Index

Fasting blood glucose has a significant positive correlation coefficient with TLC, neutrophil count, monocyte count, basophil count, RDW-CV, RDW-SD, PDW, MPV, P-LCR, PCT, PLR, and NLR, while RBC count, hemoglobin, hematocrit (HCT), MCV, and MCH showed significant negative correlation coefficients (p < 0.05). HbA1c showed a significant positive correlation coefficient with TLC, neutrophil count, basophil count, PDW, MPV, PCT, PLR, and NLR, while RBC count, hemoglobin, HCT, and MCV showed significant negative correlation coefficient with TLC, neutrophil count, monocyte count, basophil count, PCT, and PLR, while RBC count, hemoglobin, HCT, MCV, and MCH showed significant negative correlation coefficients (p < 0.05) (Table 2).

	Table 2: Pearson correlation coe	fficient "r" (p-value	e) of hematological indice	s with diabetic parameters
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Hematological indices	FBG (mg/dL)	PPG (mg/dL)	HbA1c (gm/dL)	BMI (Kg/m²)
White blood cells indices	465-06-100-06-0	DBP		
Total WBC count (10 ⁹ /L)	0.21 (0.0006)	-0.013 (0.8091)	0.272 (<0.001)	0.289 (<0.001)
Neutrophil count (109/L)	0.198 (0.0005)	0.022 (0.6915)	0.280 (<0.001)	0.311 (<0.001)
Lymphocyte count (109/L)	-0.044 (0.4686)	-0.109 (0.0547)	0.035 (0.5459)	-0.042 (0.4686)
Monocyte count (109/L)	0.29 (<0.001)	-0.013 (0.7825)	0.091 (0.1157)	0.130 (0.0243)
Eosinophil count (109/L)	0.110 (0.0617)	-0.038 (0.5009)	0.095 (0.1005)	0.111 (0.05479)
Basophil count (109/L)	0.432 (<0.001)	0.101 (0.0837)	0.314 (<0.001)	0.329 (<0.001)
Red blood cells indices				
RBC count (10 ¹² /L)	-0.325 (<0.001)	-0.135 (0.193)	-0.131 (0.0232)	-0.181 (0.0016)
Hemoglobin (gm/dL)	-0.438 (<0.001)	-0.175 (0.0023)	-0.185 (0.0012)	-0.279 (<0.001)
Hematocrit (%)	-0.401 (<0.001)	-0.172 (0.0028)	-0.160 (0.0054)	-0.230 (<0.001)
MCV (fL)	-0.396 (<0.001)	0.121 (0.0361)	-0.54 (<0.001)	-0.218 (<0.001)
MCH (pg)	-0.337 (<0.001)	-0.090 (0.1198)	-0.070 (0.2267)	-0241 (<0.001)
MCHC (gm/dL)	0.068 (0.2403)	0.25 (<0.001)	0.001 (0.9862)	-0.087 (0.1327)
RDW-CV (%)	0.138 (0.0167)	0.0075 (0.897)	0.020 (0.7300)	0.151 (0.0088)
RDW-SD (fL)	0.162 (0.0049)	0.281 (<0.001)	0.086 (0.1372)	0.105 (0.0693)
Platelet indices				
Platelet count (109/L)	-0.28 (<0.001)	0.161 (0.0053)	0.062 (0.2844)	0.078 (0.1778)
PDW (fL)	0.271 (<0.001)	0.076 (0.1892)	0.136 (0.0184)	0.085 (0.1418)
MPV (fL)	0.295 (<0.001)	0.002 (0.9724)	0.101 (0.0807)	0.030 (0.6047)
PLC-R (%)	0.177 (0.0020)	-0.031 (0.5927)	0.071 (0.2201)	-0.035 (0.5459)
PCT (%)	0.151 (0.0088)	0.101 (0.0807)	0.102 (0.0777)	0.120 (0.0377)
NLR	0.125 (0.0304)	0.106 (0.0667)	0.156 (0.0067)	0.095 (0.1005)
PLR	0.187 (0.0011)	0.115 (0.0465)	0.123 (0.0332)	0.224 (<0.001)

3. DISCUSSION

Diabetes mellitus is a persistent condition that could result in several complications over the years. The diabetes-related microvascular complications are highly prevalent in individuals with poor glycemic control, longer duration of T2DM, associated high blood pressure, and obesity. This leads to elevated morbidities and mortalities in T2DM.¹² Persistent hyperglycemia results in a series of interrelated changes that could cause evident endothelial dysfunction and diabetes-related vascular complications. The formation of advanced glycation end products, protein kinase C activation, and polyol pathway defects are the possible mechanisms through which elevated sugar levels produce vascular complications.¹²

Sufferers with T2DM have an elevated risk of coagulation defects and thromboembolic events. The activation, increased adhesion, and aggregation of platelets due to metabolic disturbances, insulin resistance, high blood sugar, and elevated cholesterol levels have been observed in T2DM.¹² Various factors, including systemic inflammation, impaired calcium metabolism, reduced nitric oxide, and increased phosphorylation and glycosylation of cell proteins, are responsible for prolonged platelet activation and increased release of pro-thrombotic factors in T2DM.

Increased TLC is a standard marker of inflammation, and it recruits inflammation with hyperglycemia and insulin resistance. In recent years, PLR and NLR have been identified as novel markers of inflammation in various metabolic disorders. Moreover, increased levels of PLR and NLR have been reported in T2DM and diabetes-related nephropathy. Besides thromboembolic issues, associations of platelet indices with inflammation and disordered activity of anti-inflammatory factors have also been demonstrated.

Dr Nischal Garg,, Dr Tushar Krishnan Saini, Dr Suryavir

In the present study, a total of 300 patients with T2DM were enrolled. Elderly people and female patients had poorly controlled diabetes, and the difference was significant.

In our study, the mean BMI among both groups did not differ significantly. This suggests that cases with poorly controlled diabetes are more obese than those in the well-controlled diabetic group, but the difference is statistically nonsignificant (p = 0.4146).

In our study, uncontrolled diabetics had a longer history of diabetes compared to the controlled diabetic group. Indices of glycemic control, including duration of diabetes, blood sugar level, HbA1c, and systolic blood pressure, were significantly higher among those with uncontrolled diabetes compared to the controlled diabetic group. In our study, the mean SBP of the good glycemic control group (HbA1c <7%) was 131.50 ± 5.10 mm Hg, and that of the poor glycemic control group (HbA1c <7%) was 137.29 ± 5.83 mm Hg. A significant difference was observed (<0.001) between the groups.

Diabetes-related microvascular complications, like UACR, were significantly higher in patients with poorly controlled T2DM compared to those with good control (p < 0.001). Thus, patients with poorly controlled diabetes had significantly more albuminuria than those with good control, further increasing the risk of nephropathy.

In this study, the mean values of total cholesterol, triglycerides, and LDL levels were significantly higher in patients with poorly controlled T2DM compared to those with good control (p < 0.001). Therefore, patients with poorly controlled diabetes had a significantly more deranged lipid profile than those with good control, further increasing the risk of atherosclerotic burden. In this study, we found that among white blood cell indices, inflammatory markers such as total WBC count, monocyte count, and basophil count were significantly higher in patients with poorly controlled T2DM compared to those with good control (p < 0.05).

FBG has a direct significant positive correlation coefficient with TLC (r = +0.21, p = 0.0006), neutrophil count (r = +0.198, p = 0.0005), monocyte count (r = +0.29, p < 0.001), and basophil count (r = +0.432, p < 0.001). HbA1c has a direct significant positive correlation coefficient with WBC count (r = +0.272, p < 0.001), neutrophil count (r = +0.280, p < 0.001), and basophil count (r = +0.314, p < 0.001). BMI has a direct significant positive correlation coefficient with WBC count (r = +0.289, p < 0.001), neutrophil count (r = +0.311, p < 0.001), monocyte count (r = +0.130, p = 0.0243), and basophil count (r = +0.329, p < 0.001). The differential WBC counts are probably deranged due to high blood sugar levels in patients with T2DM.

Red blood cell indices, like RBC count, hemoglobin, MCV, and MCH, were significantly lower in the uncontrolled diabetic group compared to the controlled diabetic group (p < 0.05), while RDW-CV was found to be significantly higher in the uncontrolled diabetic group (14.24 \pm 2.87%) compared to the controlled diabetic group (13.12 \pm 2.56%) (p = 0.0007). FBG has a direct significant negative correlation coefficient with RBC count (r = -0.325, p < 0.001), hemoglobin (r = -0.438, p < 0.001), HCT (r = -0.401, p < 0.001), MCV (r = -0.396, p < 0.001), and MCH (r = -0.337, p < 0.001), while a positive correlation coefficient is found with RDW-CV (r = 0.396, p = 0.0167) and RDW-SD (r = 0.162, p = 0.0049). HbA1c has a direct significant negative correlation coefficient with RBC count (r = -0.131, p = 0.0232), hemoglobin (r = -0.185, p = 0.0012), HCT (r = -0.160, p = 0.0054), and MCV (r = -0.54, p < 0.001). BMI, a most important risk factor of diabetes, has a direct significant negative correlation coefficient with RBC count (r = -0.181, p = 0.0016), hemoglobin (r = -0.279, p < 0.001), HCT (r = -0.230, p < 0.001), MCV (r = -0.218, p < 0.001), and MCH (r = -0.241, p < 0.001), while a positive correlation coefficient is found with RDW-CV (r = 0.151, p = 0.0088).

Every other study has found that diabetics are at risk of anemia because of decreased kidney capabilities and reduced production of erythropoietin hormone, which ultimately results in reduced RBC count within the body. The possible explanation for this difference is probably that chronic hyperglycemia causes nonenzymatic glycosylation of RBC membrane proteins, leading to increased aging of RBCs. Enormous elevations of HCT and MCV are likely due to the type of morphological changes exhibited by RBCs and compositional changes in plasma associated with T2DM.²⁸ In diabetic patients, a number of risk factors, including hyperglycemia, hyperosmolarity, oxidative stress, inflammation, and lipid metabolic disease, may affect RBC metabolism as they increase aggregation, reduce cell deformability, and decrease membrane fluidity.

Indices like PDW, MPV, P-LCR, PCT, NLR, and PLR were observed to be significantly higher in the uncontrolled diabetic group compared to the controlled diabetic group (p < 0.001). FBG has a direct significant negative correlation coefficient with platelet count (r = -0.28, p < 0.001), while a positive correlation coefficient is found with PDW, MPV, PLCR, PCT, NLR, and PLR. HbA1c has a direct significant positive correlation coefficient with PDW, NLR, and PLR.

We examine exhibits that inflammation, tendency to coagulation, and thrombosis may be detected with those smooth-on-hand and cheaper hematological indices. Furthermore, a number of these parameters can also assist to conscious clinicians about impaired glucose regulation and vascular diabetic complications. These assessments are easy, cheaper, and carried out routinely. They may be an alternative to other more expensive inflammatory markers, such as ILs, TNF, and cytokines. Improved glycemic control decreases these indices and can prevent microvascular complications of diabetes.

4. LIMITATIONS

The sample size was limited, and for confirmation of results, we need a larger sample size. It was a single-center, observational, cross-sectional study, and if it had been multicentric, we might have had different results due to a different study population

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