Original Article

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Platelet Parameters in Controlled and Uncontrolled Type 2 Diabetes Mellitus

*Dutta A[1], Matin MNI[2], Nath MC[3], Das UK[4], Alam MR[5].

Abstract:

Background: Type 2 Diabetes mellitus is one of the most serious public health problems, faced by both developed and developing countries. Poorly controlled diabetes leads to various micro and macro vascular complications. Several studies have shown that glycemic status affects the morphology and functioning of platelets.

Objectives: To see the differences of platelet parameters between controlled and uncontrolled glycemic status in type 2 diabetic patients.

Materials and Methods: This cross-sectional analytical study was conducted in the Department of Physiology, Sylhet M.A.G Osmani Medical College from July 2020 to June 2021. Thirty male patients with controlled type 2 DM (HbA1c ≤7%) and thirtymale patients with uncontrolled type 2 DM (HbA1c >7%) were selected. FPG, PPPG, HbA1c, complete blood count and anthropometric measurement were done in all patients.

Results: The mean [platelet count ($\times 10^3/\mu l$) (269.83 ± 74.67 versus273.97 ± 109.02; p=0.865), mean platelet volume (fl) (11.23 ± 1.01 versus 11.43 ± 1.23; p=0.494) and platelet distribution width (fl) (13.83 ± 2.35 versus 14.12 ± 2.73; p=0.661) did not differ significantly between controlled and uncontrolled type 2 DM.

Conclusion: Platelet indices such as platelet count, mean platelet volume and platelet distribution widthdid not differ significantly between controlled and uncontrolled type-2 Diabetes mellitus. However, large scaled study is warranted.

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Key words: Type 2 Diabetes mellitus, HbA1c, platelet count, mean platelet volume and platelet distribution width.

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Introduction:

Diabetes Mellitus (DM) is a metabolic disorder characterized by chronic hyperglycemia affecting metabolism of carbohydrate, lipid and protein caused by either lack of insulin secretion, impaired insulin action, or both.[1]

1. Dr. Anup Dutta, Assistant Professor, Department of Physiology, BGC Trust Medical College, Chattogram.
2. Dr. Md. Nazrul Islam Matin, Assistant Professor, Department of Physiology, Sylhet MAG Osmani Medical College, Sylhet.
3. Dr. Manik Chandra Nath, Associate Professor, Department of Physiology, BGC Trust Medical College, Chattogram.
4. Dr. Uzzal Kanti Das, Assistant Professor, Department of Medicine, BGC Trust Medical College Hospital, Chattogram.
5. Dr. Muhammad RabiulAlam, Assistant Professor, Department of Anatomy, BGC Trust Medical College, Chattogram.

Corresponding author: Dr. Anup Dutta,
Assistant Professor, Department of Physiology, BGC Trust Medical College, Chattogram.
Email- anupduttarony@gmail.com

According to international diabetes federation (IDF), it is estimated that globally 415 million people had diabetes in 2015 and type 2 diabetes mellitus constituted up about 90% of these cases.[2,3] The prevalence rate of Diabetes Mellitus is 7.4% of adult population in Bangladesh.[3]

Person with diabetes, especially in those having type 2 diabetes, express increased platelet reactivity. Chronic hyperglycemia results in increased platelet reactivity through its direct effects or by increasing glycation of the surface protein of the platelet. Both relative and absolute deficiency and insulin resistance promotes platelet reactivity. Insulin inhibits platelets activation. Mean platelet volume (MPV) and

Platelet Distribution Width (PDW) are display the average size of the platelets and platelets activity. It is reported that these are often elevated in diabetes mellitus. A higher MPV can indicate larger and more reactive platelets and is thought to be an emerging risk factor for cardiovascular disease. Similarly PDW is an exhibitor of changes in platelet size and a higher PDW may suggest a sign of active platelet production.[4]

Mean platelet volume (MPV) and platelet distribution width (PDW) are easily measured platelet indices, which increase during platelet activation. In order to obtain a larger surface platelets change in shape during activation. Their shape changes from discoid to spherical. Pseudopodia are formed as well.[5] Larger platelets are younger more reactive and agreeable than small platelets. These larger platelets show more dense granules producing more pro-coagulant factors such as serotonin, β-thromboglobulin and thromboxane A_2. Hence the larger platelets play an essential role in thrombus formation and vascular thrombotic complications. This suggests an association between the platelet size and function specially MPV and vascular complications in diabetes mellitus. Therefore, higher MPV indicates a state of thrombogenesis. Hence individuals with higher MPV are appear as an emerging risk factor for the vascular complications associated with DM of which major role is driven by atherothrombosis.[6]

Glycated hemoglobin (HbA1c) is a form of hemoglobin that is quantified mainly to ascertain the average plasma glucose over extended period of time.[7] The patients of type 2 DM can be divided into two categories based on their HbA1c level in blood. Patients with HbA1c ≤ 7% are considered as having controlled diabetes whereas those with HbA1c > 7% are categorized as having uncontrolled diabetes.[1] Diabetes, especially when poorly controlled, leads to complications such as nephropathy, retinopathy and neuropathy as well as several disordered metabolic processes including oxidative stress which causes oxidative damage to tissue and cells.[8]

Several studies demonstrated that platelet indices were significantly higher in uncontrolled DM compared to controlled type 2 DM.[9,10] But other study stated that platelet indices did not differ significantly between controlled type 2 DM and uncontrolled type 2 DM.So this study aiming to evaluate platelet parameters (platelet count, MPV, PDW) in type 2 DM with glycemic status.

Materials and Methods

This cross-sectional analytical study was conducted in the Department of Physiology, Sylhet M.A.G Osmani Medical College during the period from July 2020 to June 2021. Sixty male type 2 DM, duration of diabetes 6 months or more and aged 20-70 years attending the Outpatient or inpatient Department of Endocrinology and Department of Medicine, Sylhet M.A.G Osmani Medical College Hospital of which 30 controlled (HbA1c ≤ 7%) and 30 uncontrolled (HbA1c > 7%) type 2 DM were selected. Exclusion criteria were any acute or chronic illnessrelated to hematological abnormalities and use of drug affecting platelet function.

All the study subjects, who fulfilled the selection criteria, were briefed about the objective, procedure of study and regarding questionnaire. Then informed written consent was taken. A brief history was taken regarding disease and clinical examination was done accordingly.

Height and weight were measured with the help of stadiometer and weight machine respectively. Body weight was measured in light clothing and without shoes. Weight was recorded in kilogram and height was recorded in meter. Body mass index (BMI) was calculated using following formula (BMI=Weight in Kilogram/Height in meters^2).

All patients were advised to take 10–12 hours of fasting with the exception of water and medication.On the next morning after confirmation of fasting status of patient, 10 ml venous blood was drawn from antecubital vein by using disposable syringe under aseptic precaution. Blood sample was transferred to 2 test tubes; one tube with anticoagulant EDTA K2 for estimation of CBC (MPV and PDW are

part of complete blood count) and HbA1c and second tube with Sodium fluoride and EDTA K2 for estimation of plasma glucose.
Another blood sample was taken at 2 hour after breakfast and was transferred in a test tube containing Sodium fluoride and EDTA K2 for estimation of Post Prandial plasma glucose.

Collected blood samples were kept in room temperature for one hour. After clot retraction centrifugation of blood was done at 3000 rpm for 30 minutes. Afterward, serum was removed by disposable pasture pipettes and transferred into air tight tube.

All biochemical analysis of blood was performed at biochemistry laboratory in Sylhet MAG Osmani Medical College by researcher himself with technical assistance of department stuff.

Fasting Plasma glucose and Post Prandial Plasma glucose were measured by *Ortho-Clinical Diagnostics VITROS 350 System,* automated biochemistry analyzar (India) with calibration. Whole blood for HbA1c was measured by GH 900 automachine (Lifotronic Technology Co Ltd, China) by ion exchange chromatography, to separate HbA1c directly. Platelet count was estimated by Haemocytometer. Some parameters (MPV and PDW) were measured by hematology analyzer, Sysmex 500i (China).

Statistical analysis:
All the collected data were compiled and analyzed using the Statistical Package for Social Science (SPSS) Version 25.0. Quantitative data were analyzed by mean and standard deviation and comparison was done using unpaired t test. Qualitative data were presented as frequency and percentage and comparison was done using Chi-Square (χ^2) test or Fisher's Exact test. In all statistical tests $p< 0.05$ was considered as significant.

Ethical Consideration:
Informed written consent was taken from each patient after explaining all procedures. The consent form was clearly described the purpose and method of the study, confidentiality of the interview, risk and benefits of participating in this study, their right to participate voluntarily and refuse at any point of time. Prior to the commencement of the study, the research protocol was submitted for approval by the ethical committee of Sylhet M.A.G Osmani medical college, Sylhet and approval was obtained.

Results:
The mean age was 55.50 ± 8.00 years and 58.03 ± 4.47 years in controlled and uncontrolled type-2 DM respectively; difference was not considered significant (t=-1.514; p=0.135). The age group of the participants between controlled type-2 DM and uncontrolled type-2 DM was no also significant (χ^2=3.915; p=0.141) (Table- I).

The BMI was 24.57 ± 1.80 (Kg/M^2) and 25.34 ± 1.87 (Kg/M^2) in controlled and uncontrolled type 2 DM respectively; difference was not significant (t=-1.641; p=0.106). The body mass index did not differ significantly between the patients of controlled and uncontrolled type 2 DM when categorized in different segments (p=0.662) (Table-I).

The duration of diabetes was 6.28± 4.04 years in controlled type 2 DM and 6.77± 3.65 years in uncontrolled type 2 DM; duration of diabetes did not differ significantly between the patients of controlled and uncontrolled type 2 DM (t=-0.486; p=0.629). The duration of diabetes did not differ significantly between the patients of controlled and uncontrolled type 2 DM when categorized in different segments (χ^2=0.325; p=0.850) (Table-II).
The mean fasting plasma glucose was 98.33 ± 20.21 mg/dl in controlled type 2 DM and 182.60 ± 89.90 mg/dl in uncontrolled type 2 DM; the difference was significant (t=-5.009; p<0.001) (Table- III).
The mean postprandial plasma glucose was 164.70 ± 32.12mg/dl in controlled type 2 DM and 270.77 ± 90.79mg/dl in uncontrolled type 2 DM; difference was significant (t=-6.032; p<0.001) (Table- III).
The mean HbA_{1C} (percent) was 6.69 ± 0.25in controlled type 2 DM and 11.08 ± 2.62 in uncontrolled type 2 DM; difference was significant (t=-9.133; p<0.001)(Table- III).

The mean Platelet count ($\times 10^3/\mu l$) was 269.83 ± 74.67 in controlled type 2 DM and count was 273.97 ± 109.02 in uncontrolled type 2 DM; difference was not significant (t=-0.171; p=0.865) (Table- IV).

In controlled type 2 DM the mean MPV (fl) was 11.23 ± 1.01 and in uncontrolled type 2 DM the mean MPV (fl) was 11.43 ± 1.23; the difference was not significant (t=-0.689; P=0.494) (Table-IV).

The mean PDW (fl) was 13.83 ± 2.35 in controlled type 2 DM and 14.12 ± 2.73 in uncontrolled type 2 DM; difference was not significant (t=-0.441; p=0.661) (Table- IV).

Table-I Characteristics of the patients

Characteristics of the patients	Study subjects		p-value
	Controlled DM (n=30)	Uncontrolled DM (n=30)	
Age (years)			
41 to 50	9 (30%)	3 (10.0%)	
51 to 60	12 (40.0%)	17 (56.7%)	*p=0.141
61 to 70	9 (30.0%)	10 (33.3%)	
Mean ± SD	55.50 ± 8.00	58.03 ± 4.47	†p=0.135
BMI (Kg/M^2)			
Normal (18.5-22.9)	4 (13.3%)	3 (10.0%)	‡p=0.662
Overweight (23.0-24.9)	14 (46.7%)	11 (36.7%)	
Obese (25-29.9)	12 (40.0%)	16 (53.3%)	
Mean ± SD	24.57 ± 1.80	25.34 ± 1.87	†p=0.106

*Chi-Square test, unpaired †t test and ‡Fisher's Exact test were used to analyse the data.

Table-II Comparison of duration of diabetes between study groups

Duration of diabetes	Study groups		p-value
	Controlled DM (n=30)	Uncontrolled DM (n=30)	
1 to 5 years	15 (50.0%)	13 (43.3%)	*p=0.850
6 to 10 years	10 (33.3%)	12 (40.0%)	
11 to 15 years	5 (16.7%)	5 (16.7%)	
Mean ± SD years	6.28 ± 4.04	6.77 ± 3.65	†p=0.629

*Chi-Square test and unpaired †t test were used to analyse the data.

Table-III Comparison of glycaemic parameters between study groups

Glycaemic parameters	Study groups		*p-value
	Controlled DM (n=30)	Uncontrolled DM (n=30)	
Fasting plasma glucose (mg/dl)	98.33 ± 20.21	182.60 ± 89.90	p<0.001
PPPG (mg/dl)	164.70 ± 32.12	270.77 ± 90.79	p<0.001
$HbA1_C$ (percent)	6.69 ± 0.25	11.08 ± 2.62	p<0.001

*unpaired t test were used to analyse the data. PPPG: post prandial plasma glucose

Table-IV Comparison of Platelet parameters between study groups

Platelet parameters	Study groups		*p-value
	Controlled DM (n=30)	Uncontrolled DM (n=30)	
Platelet count ($\times 10^3/\mu l$)	269.83 ± 74.67	273.97 ± 109.02	p=0.865
MPV (fl)	11.23 ± 1.01	11.43 ± 1.23	p=0.494
PDW (fl)	13.83 ± 2.35	14.12 ± 2.73	p=0.661

*unpaired t test were used to analyse the data. MVP: platelet volume, PDW: platelet distribution width

Discussion:

HbA1c level has an important role for clinical diagnosis of diabetes.[11] It has been used to evaluate the level of metabolic control and to determine the developing of diabetic complications, as well as measuring the quality of diabetes care.[12] The persistent increase in glycosylated hemoglobin as a result of poor glycemic management is associated with the structural and functional changes in hemoglobin (Hb) molecule, the osmotic disturbance and the cytoplasm viscosity within each cell.[13]

This study showed that the mean age was 55.50± 8.00 years in controlled type-2 DM and was 58.03 ± 4.47 years in uncontrolled type-2 DM. The difference was not statistically significant (p=0.135). This result correlated with several studies.[2,9]

In this study the body mass index (BMI) was 24.57 ± 1.80 (Kg/M^2) in controlled type 2 DM and 25.34 ± 1.87 (Kg/M^2) in uncontrolled type 2 DM; body mass index did not differ significantly between the patients of controlled and uncontrolled type 2 DM (p=0.106). Several studies supported this result.[9,13]

In the present study the duration of diabetes was 6.28 ± 4.04 years in controlled type 2 DM and 6.77 ± 3.65 years in uncontrolled type 2 DM; duration of diabetes did not differ significantly between the patients of controlled and uncontrolled type 2 DM (p=0.629). Other studies disagreed with the finding that duration of diabetes was significantly higher in those with uncontrolled type 2 DM compared to controlled type 2 DM (p<0.001).[9,14]Difference may be due to sample size, genetic and environmental factors in the present study.

This study revealed that the mean fasting plasma glucose was 98.33 ± 20.21mg/dl in controlled type 2 DM and 182.60 ± 89.90mg/dl in uncontrolled type 2 DM; difference was significant (p<0.001). This finding was almost similar to the studies of others.[9,14] But Hasan et al.[15] found that fasting glucose level did not differ significantly between controlled and uncontrolled DM (p=0.441).

In this study the mean postprandial plasma glucose was 164.70 ± 32.12mg/dl in controlled type 2 DM and 270.77 ± 90.79mg/dl in uncontrolled type 2 DM; difference was significant (p<0.001).Several studies supported this result.[14,15]

In the current study the mean HbA_{1C} (percent) was 6.69 ± 0.25in controlled type 2 DM and 11.08 ± 2.62 in uncontrolled type 2 DM; difference was significant (p<0.001). Several studies were in line with the present study.[9,15]
This study expressed that the mean platelet count ($\times 10^3/\mu l$) was 269.83± 74.67 in controlled type 2 DM and 273.97 ± 109.02 in uncontrolled type 2 DM; difference was not significant (p=0.865). This result was consistent with several studies.[2,9,13] But Jiskani and Singh,[10] reported Platelet Count was significantly higher in uncontrolled DM compared to controlled type 2 DM (p=0.02).

In this study the mean MPV (fl) was 11.23 ± 1.01 in controlled type 2 DM and 11.43 ± 1.23 in uncontrolled type 2 DM; difference was not significant (p=0.494). Other studies also supported this result.[2,13,17] But Demirtas et al.[9] reported significantly lower MPV (fL) in controlled type 2 DM (8.9 ± 0.8) compared to uncontrolled type 2 DM (9.4±0.9) (p<0.001).

The mean PDW (fl) in the present study was 13.83 ± 2.35 in controlled type 2 DM and 14.12 ± 2.73 in uncontrolled type 2 DM; difference was not significant (p=0.661). Samiduraiet al.[13]supported this result. But Demirtas et al.[9] reported significantly lower PDW (fL) in controlled type 2 DM (median 16.0) compared to uncontrolled type 2 DM (median 16.6) (p=0.002).
Limitations were faced during this study: (1) First, since it involves one single institution, it may not represent the general population, (2) Second, more number of study subject needed to be included in this observational study, which could not be done due to limited time and Covid 19 pandemic situation and (3) Third, we cannot determine a cause and effect relationship due to the cross-sectional nature of our study.

Conclusion:
Based on findings in the present study it may be concluded that platelet indices such as total platelet count, mean platelet volume and platelet distribution width are not differ significantly between controlled and uncontrolled type-2 Diabetes mellitus. However, large scaled study involving multicentre should be conducted in order to evaluate platelet indices between controlled and uncontrolled type-2 Diabetes mellitus should be conducted to reach a justifiable conclusion.

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