Original Article

Levels of Immature Platelet Fraction in Patients of Acute Coronary Syndrome

Sadia Ijaz, Israrul Haque, Muhammad Muzammil Bajwa, Junaid Mushtaq, Ghias un Nabi Tayyab, Mujtaba Hassan Siddiqui

ABSTRACT

Objective: To measure the level of immature platelet fraction in patients with the acute coronary syndrome.

Methodology: It was a cross-sectional comparative study conducted at the Department of Pathology, Postgraduate Medical Institute, Lahore from November 2017 to November 2018. One hundred and seventy respondents were included and divided into two groups, a case group, and a control group. In the case group, 85 patients of acute coronary syndrome and in the control group, 85 age-matched normal healthy individuals were included using a non-probability convenient sampling technique. The immature platelet fraction (IPF) was measured in both groups using Sysmex haematology analyser (XN-1000). An independent sample t-test was applied to compare the mean of normally distributed data in both groups.

Results: The ages of the patients in case group ranged between 18-86 years and in control group 17-80 years. No statistical significant difference was found when the age of the patients and the controls were compared (p-value >0.05). In the case group, there were 45(26.47%) males and 40(23.53%) females whereas in the control group there were 47(27.65%) male and 38(22.35%) female cases. The mean immature platelet fraction in the case group and control group was 8.716±6.2834% and 3.83±1.63%, respectively. A significant difference was found in the comparison of mean IPF in the case & control groups (p-value <0.001).

Conclusion: The levels of immature platelet fraction were raised among patients with the acute coronary syndrome. So, increased levels of IPF can lead to other thrombotic events or unfavorable prognosis in patients with acute coronary syndrome (ACS).

Keywords: *Immature platelet fraction. Mean platelet volume. Acute coronary syndrome. Cardiovascular disease.*

INTRODUCTION

Cardiovascular diseases are the major cause of death around the world.[1] Among cardiovascular diseases, acute coronary syndrome is the leading cause of morbidity and mortality. Acute coronary syndrome accounts for the first presentation of coronary vessel diseases and is a broader term used for describing signs and symptoms of cardiac ischemia.[2] On the basis of signs and symptoms, findings on ECG and certain laboratory parameters ACS can be sub-grouped as ST-segment & non-ST-segment elevation myocardial infarction, and unstable angina. The acute coronary syndrome can cause fatal events like cardiogenic shock causing the sudden arrest of cardiac activity further leading to death.[3] Amongst today's challenges, the major goal is to design such strategies which can prevent the occurrence of adverse coronary events and identify the individuals who are at greater risk of occurrence of ACS.[4]

Cardiac troponins and iso-enzymes of creatine kinase are the routine investigations that are being used for the diagnosis of ACS. Troponin is the most sensitive, specific, and reliable cardiac marker and is being currently used as a gold standard investigation for the diagnosis and risk stratification in ACS patients. However, it remains undetectable in 40-60% of the patients who are suffering from ACS. Platelet indices are a more reliable and accurate tool, as a new cardiac biomarker, of cardiovascular events and can be used potentially for the risk stratification of cardiovascular diseases.[5] Platelets are now being considered as the source of release of inflammatory mediators. These mediators further lead to platelet adhesion. The thrombotic potential of platelets causes the release of further mediators which leads to the activation of the inflammatory process and the propagation of the coronary vessel thrombosis predisposing to the thrombotic events.[6]

Platelets vary in properties like size, density, and activity. These variations can be associated with the initial triggering factor of ACS. Platelets that are large in size have stronger adhesion potential and aggregating effect than the platelets with smaller size. An increase in the volume of platelets is associated with the increased prothrombotic potential of atherosclerotic plaque in ACS and is the risk factor for thrombus formation in coronary vessels in patients of acute myocardial infarction (AMI).[7]

Mean platelet volume (MPV) is a component of complete blood count (CBC) and is the commonly used most reliable index for platelet size identification and the status of its activation. An increase in MPV is associated with various cardiovascular risk factors and

Sharif Medical & Dental College, Sharif Medical City. Sharif Medical City Road, Off Raiwind Road, Jati Umra, Lahore 54000, Pakistan.

Correspondence: Dr. Sadia Ijaz
Demonstrator Department of Pathology
Postgraduate Medical Institute, Lahore
E-mail: drsadia.amir87@gmail.com

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comorbidities like diabetes mellitus, hypertension, hypercholesterolemia, and obesity.[8]

Amongst the platelets circulating, the youngest form is the reticulated platelets. They are bigger than the senescent platelets and they have the residual RNA that gives the reticulated appearance and are hyperactive because they express more GPIb and GPIIb/IIIa receptors. Now a days, a fast fully automated method is available for the quantification of reticulated platelets via a parameter called IPF, which is a ratio of reticulated platelets and a total number of platelets. Immature platelet fraction may be a more sensitive and specific indicator as compared to MPV for the measurement of platelet reactivity.[9] It is noticed that in subjects with coronary artery disease, IPF is increased as compared to normal healthy subjects. The formation of atherosclerotic plaque results in complications like myocardial infarction and stroke due to the occlusion of thrombotic vessels. Vascular damage repair and the maintenance of narrow capillaries to remain patent is a complex, mechanism and platelets act as a key for the regulation of the process.[10]

Platelets contribute to both the dysfunction of endothelium and the rupture of plaque in the process of atherosclerosis. The platelet interaction with endothelium lining vessels causes excessive activation of platelets which reduces the half-life and increases the turnover of platelets, hence influence the MPV and IPF.[11] Activation of platelets causes the release of specific proteins. β-TGF is the first platelet-specific protein that is released during the aggregation of platelets resulting in inflammatory and thrombotic processes that both play part in the pathogenesis of the development of ACS.[12]

Platelet activation parameters can be beneficial tools for coronary disease progression before the occurrence of cardiac cell necrosis.[13] Hence the goal of this study was to evaluate platelet morphological index in patients of ACS as compared to controls without ACS.

METHODOLOGY

It was a cross-sectional comparative study and carried out at the Department of Pathology, Postgraduate Medical Institute, Lahore. The study was approved by the ethical committee of the institution (ethical approval # UHS/Education/126-17/3368, 01-08-2017). The study patients were selected from the Punjab Institute of Cardiology Lahore, employing a non-probability convenient sampling technique. The duration of the study was from November 2017 to November 2018. A total of 170 subjects were included and divided into two groups, the case group, and the control group. In the case group, 85 patients of acute coronary syndrome, and in the control group, 85 age-matched normal healthy individuals were included. Both the study groups included individuals greater than 18 years of age. Patients included in the case group had raised levels of cardiac troponin I >0.04 ng/ml and at least one of the following was a must to be present for the inclusion in the current study: clinical symptoms of ischemia/significant ST-segment & T wave changes/ development of pathological Q waves.

Patients with acute inflammation, chronic circulatory insufficiency, cancer, renal failure, diabetes mellitus, history of acute coronary syndrome, patients on anticoagulant treatment (acenocoumarol, warfarin, dabigatran, rivaroxaban), and patients taking antiplatelet drugs (acetylsalicylic acid, clopidogrel, ticagrelor) were excluded from the study.

A questionnaire was filled for each patient, samples were collected and tests were performed to collect the required data. After obtaining the informed consent of the patients, the personal information of patients was recorded on the proforma designed for the study. The blood pressure of all the patients and controls was measured. Under aseptic measures, venous blood was collected in EDTA & gel vacutainers. Random blood sugar and serum creatinine were measured in both groups. Complete blood count was performed within 3-4 hours of sample collection using Sysmex automated hematology analyzer (XN-1000) and immature platelet fraction was checked.

STATISTICAL ANALYSIS

Data was entered and analyzed using Statistical Package for the Social Sciences (SPSS) version 24. Mean±standard deviation (SD) was used for quantitative data like age, blood pressure, random blood glucose, serum creatinine, and immature platelet fraction. An independent sample t-test was applied to compare the mean values of study variables of both groups. A p-value ≤0.05 was considered as significant.

RESULTS

The ages of the patients in case group ranged between 18-86 years and in control group 17-80 years. No statistical significant difference was found when the age of the patients and the controls were compared (p-value >0.05).

In the case group, there were 45(26.47%) males and 40(23.53%) females whereas in the control group there were 47(27.65%) male and 38(22.35%) female cases. Regarding gender, no significant results were seen when both groups were compared (p-value=0.758). Figure 1 shows gender distribution in both groups.

The mean systolic blood pressure (BP) in patients and controls was 137.68±15.621mmHg and 128.89±18.813 mmHg, respectively, while the mean diastolic BP in patients and controls was 86.36±9.716 mmHg and 81.39±11.536 mmHg, respectively. The

mean systolic and diastolic BP was statistically higher in the case group as compared to controls (p-value ≤0.05). The mean blood glucose levels in the case group were 132.21±33.63 mg/dL and in controls was 132.84±32.04 mg/dL. The mean glucose level was not statistically different in both groups (p-value=0.984).

The mean serum creatinine in ACS patients was 1.01±0.32 mg/dL and in controls was 0.913±0.26 mg/dL with statistically higher levels in ACS patients (p-value=0.02).

The mean immature platelet fraction in ACS patients and controls was 8.716±6.2834% and 3.83±1.63%, respectively (Table 1). Mean IPF was statistically high in the case group as compared to controls (p-value=0.001). Figure 2 shows immature platelet fraction (%) in both study groups.

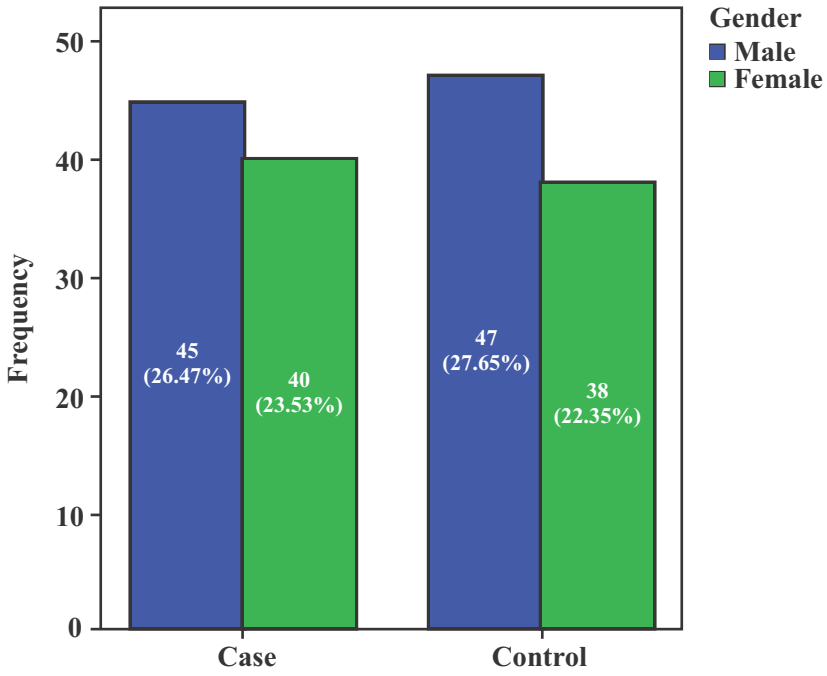

Figure 1: Gender Distribution in Both Groups

Table 1: Study Variables of Both Study Groups

Study Variables	Study Patients	Mean±SD	Minimum	Maximum	p-value
BP Systolic (mmHg)	Case	137.68±15.621	110	160	0.001
	Control	128.89±18.813	112	155	
BP Diastolic (mmHg)	Case	86.36±9.716	70	100	0.003
	Control	81.39±11.536	70	100	
Blood glucose levels (mg/dL)	Case	132.21±33.63	183	60	0.984
	Control	132.84±32.04	183	60	
Serum creatinine (mg/dL)	Case	1.01±1.10	0.3	1.7	0.02*
	Control	0.913±1.10	0.6	1.5	
Immature Platelet Fraction (%)	Case	8.716±6.2834	31.6	6.8	<0.001*
	Control	3.83±1.63	6.8	2.8	

**Significant p-value*

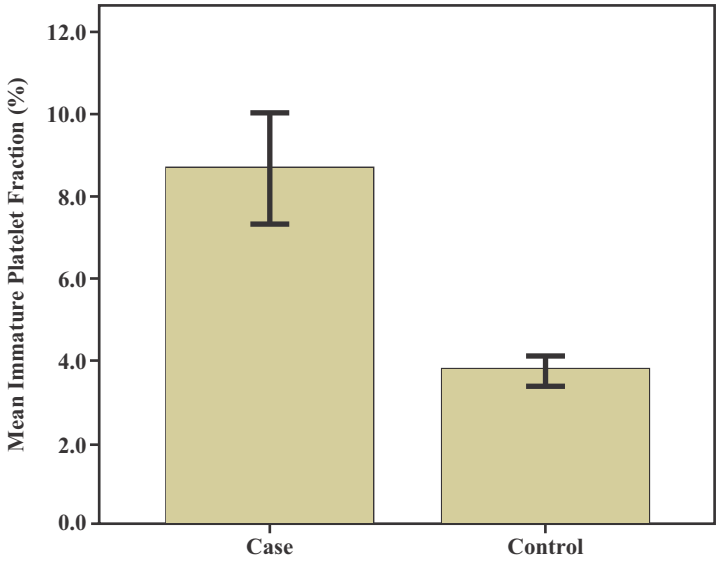

Figure 2: Immature Platelet Fraction (%) in Both Study Groups

DISCUSSION

Among the cardiovascular diseases acute coronary syndrome is considered to be one of the most prevalent and a major cause of mortality worldwide.[3] Raised levels of immature platelet fraction are observed among patients with acute coronary syndrome when compared with normal healthy individuals. Age is considered the most important factor that plays a significant role in the progression of numerous diseases because most of the diseases are prevalent among elderly people with increasing age.[1]

The present study indicated that the majority of the patients in both groups were more than 50 years old. Similar results were found in another study conducted by Pervin and teammates, who reported that most of the patients were more than 40 years old while some of them were up to 40 years old.[14] Our study indicated that the mean age of the cases was 57.04±11.62 years and the mean age of controls was 54.14±9.84 years. The results of a recent study performed by Khalifa and coworkers highlighted that the mean age of the cases was 61.32±7.20 years while the mean age of the controls was 57.40±7.89 years.[15] Another study indicated that the mean age of the cases was 50.87±11.02 years while among controls mean age was 47.31±12.09 years.[16]

High blood pressure and cholesterol are considerable risk factors associated with acute coronary syndrome.[4] It was found during the study that both systolic and diastolic blood pressure was raised in ACS patients as compared to the controls. The mean systolic BP in ACS patients and controls was 137.68±15.621 mmHg and 128.89±18.813 mmHg, while the mean diastolic in patients and controls was 86.36±9.716 mmHg and 81.39±11.536 mmHg, respectively. The findings of a study undertaken by Abdallah et al., showed results similar to our results indicating that mean systolic blood pressure among cases was 132±30 mmHg and 136±30 mmHg among controls while mean diastolic blood pressure was 80±17 mmHg and 79±15 mmHg in cases and controls, respectively.[17]

Our study showed that among patients with ACS the immature platelet fraction levels are raised as compared to normal healthy individuals. During this study when the levels of IPF were evaluated, the study demonstrated that among the majority of the controls, the levels of immature platelet fraction were found normal while in a major proportion of ACS patients, raised levels of immature platelet fraction were observed. The study pointed out that the mean immature platelet fraction in patients was 8.72±6.28% and among controls was 3.83±1.63% with statistically significant results showing raised levels in ACS patients. The findings of a study performed by Berny-Lang and fellows indicated that mean immature platelet fraction among patients suffering from ACS was 5.0±2.8% and in controls, it was 4.6±2.7% showing statistically insignificant results.[18] These findings are in contrast to the present study results.

The results of our study were supported by a similar study conducted in 2020 concluding that IPF is a novel biomarker for the prediction of major adverse cardiovascular outcomes in patients of ACS.[19]

Immature platelet fraction is a biomarker of ACS. A study conducted by Huang et al. concluded that MPV and IPF were significantly raised in patients of ACS. Mean platelet volume represents the volume of platelets and remains constant within 4 months after measurement whereas IPF represents the measure of platelet producing activity by the marrow. Hence MPV can be used for predicting the onset of ACS, whereas

peripheral immature platelets are degraded within 24 hours. Thus, a sudden increase of IPF may predict the prognosis of patients with ACS.[20]

Another study was conducted to evaluate platelet count, IPF, and MPV in patients undergoing coronary artery bypass grafting. Daily changes in platelet count, IPF, and MPV were observed along with the daily prognostic subjective improvements of the patients. Of the three markers used, immature platelet count was the most surrogate index for the production of platelets.[21]

The acute coronary syndrome includes ST elevation myocardial infarction (STEMI), non-ST elevation myocardial infarction (NSTEMI), and unstable angina (UA). A study was aimed to analyze the differences of platelet indices among subtypes of ACS in Dr. Soetomo hospital in 2019. The results of the study concluded that IPF values were significantly higher in STEMI patients than NSTEMI and UA and the values of NSTEMI patients were higher than the values of UA patients. The platelet count, MPV, platelet distribution width, and plateletcrit were higher in both STEMI and NSTEMI compared to UA. The median of IPF values of STEMI, NSTEMI, and UA patients showed a gradual decrease of 3.8%, 2.7%, and 2.0%, respectively. The IPF values were significantly different among each type of ACS patient giving an opportunity to use this marker to differentiate the ACS types.[22]

Several studies were conducted in regard to the immature platelets showing increased levels of reticulated platelets in patients with ACS. In patients of ACS, an evaluation showed that patients with an increase in reticulated platelet count came up with the increase in the incidence of major ischemic cardiovascular events, which again signifies the importance of IPF in the diagnosis of ACS.[23]

CONCLUSION

The levels of immature platelet fraction were raised among patients with the acute coronary syndrome. So, increased levels of IPF can lead to other thrombotic events or unfavorable prognosis in patients with ACS.

LIMITATIONS & RECOMMENDATIONS

- Immature platelet fraction should be used for screening of patients at risk of a thrombotic event.
- Further diagnostic studies should be carried out on a large scale in patients of ACS categorizing them in different types of ACS and with at least three months follow up for the evaluation of the extent of adverse outcomes.

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