

Platelet Volume Indices in Peripheral Artery Disease

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Abstract: This study aimed to assess the relationship between platelet volume indices (PVI) and peripheral artery disease (PAD). Platelet count (PLT), platelet distribution width (PDW), platelet large cell ratio (P-LCR), and mean platelet volume (MPV) were studied in adult patients with PAD. Two hundred ninety two patients were divided into two groups according to Fontaine classification. Group-1 contained 183 patients with stage I and II with mild symptoms, and the remaining 109 patients referred to the group-2 with severe symptoms. Blood samples were analyzed in a Sysmex SF-3000 for platelet indices. There were no significant differences between patients in group-1 and group-2 in terms of PVI ($P > 0.05$). No significant correlation was identified between severity of peripheral vascular disease and PVI in patients with PAD. PVI seem not to associate with severity of PAD.

Keywords: Peripheral artery disease, Platelet volume indices, Risk factor, Severity.

INTRODUCTION

The pathomechanism of atherosclerosis and PAD is a multifactorial phenomenon. Atherosclerosis is an inflammatory disorder and platelets play a key role in the pathogenesis of vascular diseases. Platelets activation is triggered by their interaction with the vessel wall. Platelets cover the exposed sub-endothelial tissue and cause further platelet and leukocyte recruitment at the site of vascular injury [1]. Platelet volume has been demonstrated to correlate with its function. Larger platelets are believed to be enzymatically and metabolically more reactive. The frequently reported inverse correlation between platelet count and MPV reflects the need to preserve a constant platelet mass [2]. However, platelet mass changes as both platelet count and MPV increase during stimulated thrombopoiesis [3]. It has been reported that platelet heterogeneity is not related to ageing and changes in platelet volume determines by megakaryocyte [4]. PVI could therefore be a novel marker or a predictor of PAD.

PVI are widely available by hematologic autoanalysers, and it makes them accessible and inexpensive for routine medical practice. Previous studies had shown that MPV was elevated in acute myocardial infarction (AMI), coronary artery disease (CAD), and stroke [5, 6]. The clinical usefulness of MPV, PDW, P-LCR, and platelet mass (MPV \times platelet count) studied in different pathological conditions [7-9]. There are also conflicting results in the literature [10]. We sought to investigate the relation between PVI and severity of peripheral vascular disease.

PATIENTS AND METHODS

Two hundred ninety two PAD patient subjects (107 females, 185 males, mean age 66 years) were enrolled in the study at the outpatient clinic of the Department of Medicine during the period of 2005-2011. Then, patients were divided into two groups according to Fontaine classification. Asymptomatic patients and patients with mild claudication comprised Group-1 (Fontaine I and II) and patients with rest pain and necrosis and/or gangrene included in group-2 (Fontaine III and IV). The study was approved by Debrecen University Ethics Committee. Signed informed consent was obtained from all participants.

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BIOCHEMICAL ANALYSES

Peripheral blood was collected after eight hours fasting at venipuncture in Vacutainer tubes (Dade Behring, Marburg, Germany) for measurement of total cholesterol, triglyceride, creatinine (Cobas Integra 700, Roche Diagnostics, Mannheim, Germany), and high-sensitivity CRP (hs-CRP) (Cobas Integra 400, Roche Diagnostics, Mannheim, Germany). EDTA anticoagulated whole blood samples were obtained and analyzed by the Sysmex SF-3000 analyzer (Sysmex Corporation, Kobe, Japan). The samples were processed within two hours after venipuncture. PVI were derived from the platelet size distribution curve [11].

Hyperlipidemia defined as cholesterol more than 5.2 mmol/l and/or triglyceride more than 1.7 mmol/l or administration of lipid-lowering agents. Hypertension was defined as blood pressure exceeding systolic 140 mmHg and/or diastolic 90 mmHg, and/or receiving antihypertensive treatment. Diabetes mellitus was defined as a fasting glucose ≥ 7 mmol/L, a non-fasting glucose ≥ 11.1 mmol/L, or treatment for diabetes. Cigarette smoking history was self-reported. Height and body weight were measured with participants standing without shoes and other heavy garments. Body mass index was calculated as weight (kg)/height²(m). All laboratory and vascular measurements were carried out by technicians unaware of the subjects' history of PAD.

DIAGNOSIS OF PERIPHERAL ARTERIAL DISEASE

The diagnosis of PAD was based on the ankle brachial pressure index [12], which has been previously validated [13], and test results have been shown to have high reproducibility when performed by trained personnel [14]. Ankle/brachial ratio was considered to be indicative of PAD if less than or equal to 0.9 [15].

STATISTICAL ANALYSIS

Values are expressed as mean \pm standard deviation (SD), or median (25%-75% confidence interval). Descriptive statistics were calculated to summarize the clinical features of PAD patients. SPSS software (version 18; SPSS Inc., Chicago, IL, USA) was used to perform all analyses. All tests were two-tailed comparisons and P less than 0.05 was considered as statistically significant. The normality of distribution of continuous variables was assessed using the Kolmogorov-Smirnov test. Comparisons between

groups were performed with the χ^2 -test for binary and categorical data and the Student's t-test or Mann-Whitney U test for continuous variables.

RESULTS

Table 1. summarises the clinical and demographic aspects of patients. There were a similar proportion of diabetes mellitus, smokers, and hypertensive individuals between group-1 and group-2. The degree of hyperlipidemia, hs-CRP and creatinine in the two patient groups was not significantly different. Platelet count, MPV, PDW, and P-LCR did not differ between two groups of patients. These findings are summarized in Table 2. MPV was significantly and inversely correlated to platelet count in all patients ($r = -0.421$) and two groups (group-1: $r = -0.403$; group-2: $r = -0.46$) separately. Estimating the Receiver operating characteristic (ROC) curve area under the curve (AUC) showed that PVI has no discriminating power for predicting severity of PAD, separately or combined (Figures 1 and 2).

Table 1: Demographic Features and Baseline Clinical Characteristics of Study Population with Peripheral Artery Disease, in Group-1 and 2

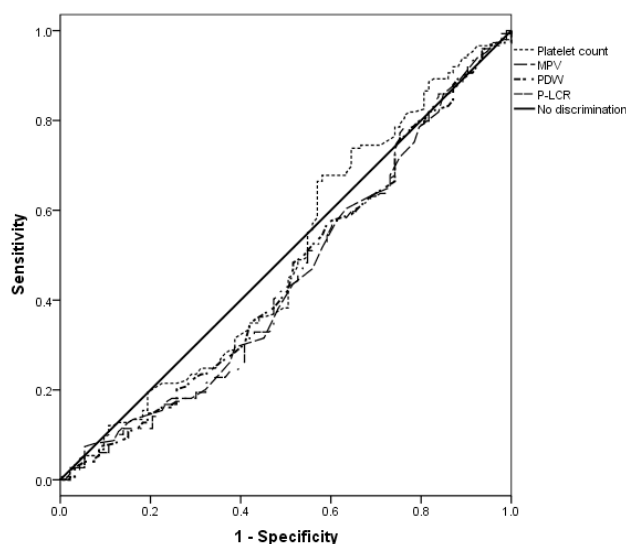
	Patients (n=292)	Group-1 (n=183)	Group-2 (n=109)
Male, n (%)	185 (63)	113 (63.1)	68 (63.5)
Age (years)	66 \pm 12	66 \pm 13	67 \pm 12
Hyperlipidemia, n (%)	219 (75)	126 (70.4)	85 (79.4)
Smoking, n (%)	105 (36)	58 (32.4)	44 (41.1)
Hypertension, n (%)	175 (60)	112 (62.6)	61 (57)
Diabetes mellitus, n (%)	72 (25)	42 (23.5)	29 (27.1)
BMI (kg/m ²)	24.7 \pm 2.8	24.7 \pm 2.7	24.7 \pm 3.0
hs-CRP (mg/l)	3.0 (1.9-5.2)	2.5 (1.9-4.6)	3.7 (2.4-6.0)
WBC count ($\times 10^9/l$)	7.2 \pm 2.0	7.1 \pm 1.9	7.3 \pm 2.1
Creatinine (μ mol/l)	82 (76-90)	82 (76-93)	81.5 (76-88)
Triglyceride (mmol/l)	1.5 (1.2-2.0)	1.5 (1.2-2.0)	1.5 (1.3-2.0)
Cholesterol (mmol/l)	5.7 \pm 1.2	5.5 \pm 1.2	5.9 \pm 1.2
LDL-cholesterol (mmol/l)	3.5 \pm 1.1	3.3 \pm 1.1	3.5 \pm 1.0
HDL-cholesterol (mmol/l)	1.3 (1.2-1.5)	1.4 \pm 0.4	1.4 \pm 0.5
ApoA-I (g/l)	1.5 \pm 0.3	1.5 \pm 0.3	1.5 \pm 0.3
ApoB-100 (g/l)	1.1 \pm 0.3	1.1 \pm 0.3	1.1 \pm 0.3
Lp(a) (mg/l)	144 (75-277)	143 (75-281)	162 (76-267)

Data are expressed as median (inter-quartile range) or mean \pm SD; BMI, body mass index; WBC, White blood cell; hs-CRP, high sensitive C-reactive protein.

Table 2: Platelet Volume Indices in Study Population with Peripheral Artery Disease, in Groups 1 and 2*

	Patients (n=292)	Group-1 (n=183)	Group-2 (n=109)
Platelet count ($\times 10^9/l$)	196 (170-220)	204 \pm 72	206 \pm 66
Mean platelet volume (fl)	10.3 (9.8-10.7)	10.3 \pm 1.0	10.4 \pm 1.1
Platelet distribution width (fl)	12.5 (11.5-13.2)	12.3 (11.3-13.1)	12.8 (11.6-13.6)
Platelet large cell ratio (%)	28.7 (24.9-31.8)	28.4 \pm 7.8	30.1 \pm 9.2
Platelet mass	2076 \pm 628	2059 \pm 649	2014 \pm 595

Data are expressed as median (inter-quartile range) or mean \pm SD. *P-value for all comparisons between group-1 and group-2 were >0.05

**Figure 1:** Receiver operating characteristic curve analysis for prediction of peripheral artery disease by platelet volume indices.

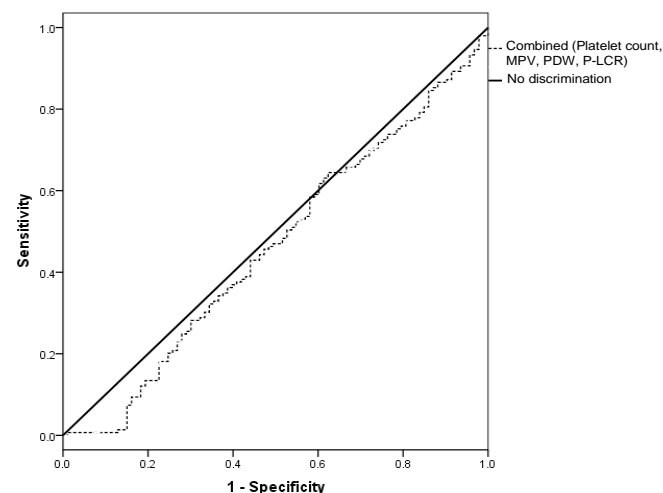
The area under the curve (95% confidence interval; P) is: platelet count 0.493 (0.416-0.57; 0.852), MPV 0.450 (0.374-0.526; 0.191), PDW 0.457 (0.381-0.532; 0.255), P-LCR 0.449 (0.373-0.525; 0.18).

CONCLUSIONS

We investigated the relationship between PVI and PAD. PVI are routinely measured by automated hematology analyzers. Platelets are implicated in the pathogenesis of vascular disorders, including atherosclerosis and its complications, such as AMI, and stroke. Abnormal platelet volume may indicate altered platelet reactivity and potential vascular risk [16].

Platelet volume may increase due to the accelerated platelet turnover with higher level of reticulated platelets from bone marrow [17]. PVI have

been investigated extensively in different pathological conditions.

**Figure 2:** Receiver operating characteristic curve analysis for prediction of peripheral artery disease by combined effect of platelet volume indices.

The area under the curve 0.461 (95% confidence interval 0.385-0.536; P=0.304).

Several reports have demonstrated that changes in MPV and PDW have been linked to increased risk of CAD, AMI, stroke and the level of circulating auto antibodies and inflammatory agents [18-20]. Whereas other demonstrate that MPV and PDW are not associated with increased risk of cardiovascular diseases [21, 22].

Khandekar and coworkers found significantly higher P-LCR in patients with AMI and unstable angina compare to control individuals. On the other hand, De Luca *et al.* failed to demonstrate an association between P-LCR and CAD [23, 24]. There was an inverse relation between platelet count and MPV. Preserved constant platelet mass reflects the tendency to maintain hemostasis in different conditions [2]. The platelet mass alteration was not significantly different between stroke patients and control individuals [25]. PVI do not retain clinical relevance as confirmed by the results of the ROC curve analysis. In a literature search of PAD, we found few reports about PVI and PAD. Berger *et al.* has demonstrated increased MPV in patients with PAD [26]. A positive correlation between MPV and arterial stiffness was shown in healthy individuals by Wang *et al* [27]. In conclusion, we found that PVI are not associated with the extent of PAD.

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