Original Article

Study of relationship between Platelet Volume Indices and Hyperlipidemia

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Abstract

Background: Mean platelet volume (MPV) correlates well with platelet activity as larger platelets tend to be metabolically and enzymatically more active than smaller platelets and secrete more prothrombotic factors. Studies have shown that there is a constant association of increased MPV with atherosclerotic diseases like coronary ischemic disease, myocardial infarction and cerebral infarct as well as with diabetes and hypertension. Hyperlipidemia, being a closely associated condition, is also expected to show changes in platelet size. We try to study the relationship between platelet volume indices and hyperlipidemia.

Methods: 100 patients with deranged lipid profile on recent lab biochemical lab reports in OPD and indoor ward patients of Sassoon general hospital, Pune have been studied. Included cases have been segregated as separate groups for patients with deranged lipid profile along with atherosclerotic disease, diabetes or hypertension. The platelet parameters like platelet count and the platelet volume indices (PVI), including mean platelet volume (MPV), platelet distribution width (PDW), and platelet large cell ratio (P-LCR); were compared with 30 age matched controls.

Results: Mean MPV (9.79 ±1.01), PDW (13.18 ± 2.44) and P-LCR (24.85 ± 7.14) of cases were significantly higher than the controls (mean MPV = 9.22 ± 0.91, mean PDW= 11.64 ± 1.75, mean P-LCR= 20.70 ± 7.07; p-value < 0.05). Also, hyperlipidemic patients having an associated disease had significantly higher PVI than the patients having hyperlipidemia in isolation.

Conclusion: The estimation of these indices can be considered as an early, economical and rapid procedure for identification of complications in hyperlipidemic patients.

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1. Introduction

Hyperlipidemia is emerging as a major risk factor for coronary heart disease, myocardial infarction and stroke.[1] Many studies have concluded that hyperlipidemia has a common association with diabetes mellitus, coronary heart disease, obesity and hypertension.[2,3,4,5] Despite this, hyperlipidemia is a commonly missed condition, firstly due to it being totally asymptomatic most of the times, secondly due to ignorance of the initial early screening for abnormal lipid profile and lastly but importantly, due to cost factor.

Platelets are small anucleate highly complex blood cells that participate in critical reactions central to hemostasis and thrombosis. Larger platelets are considered to be metabolically, enzymatically and functionally more active than the smaller platelets.[6] These produce more thromboxane B_2 than platelets in normal steady state function and are hemostatically more active and hence have more thrombogenic potential.[7,8] Thus, platelet activation is indirectly measured via platelet size or mean platelet volume (MPV).[8]

Automated cell counters have made the platelet count (PC) and the platelet volume indices (PVI) including mean platelet volume (MPV), platelet distribution width (PDW), and platelet large cell ratio (P-LCR); routinely available in most clinical laboratories at no additional cost. Easy availability of direct markers of platelet volume has prompted authors to study the role of platelet activation in the pathogenesis of various diseases. Studies have shown a constant association of increased MPV with various types of diseases like coronary artery disease, myocardial infarction, cerebral infarction and diabetes mellitus.[8,9,10,11]

As all of these are closely associated with deranged lipid profile, a correlation of these platelet volume indices is expected to be observed with hyperlipidemia also. Therefore the present study deals with testing the platelet volume indices in patients of hyperlipidemia solely as well as in association with closely related disorders.

2. Materials and Methods

A total of 130 subjects were studied including 100 cases and 30 control subjects. The study group was further subdivided into four groups – Group A of hyperlipidemia only, Group B of hyperlipidemia with atherosclerotic disease (included patients of coronary heart disease with a previous history of ischemic event and now stable; also those presenting with acute coronary events and acute cerebrovascular events), Group C of hyperlipidemia with diabetes mellitus, Group D of hyperlipidemia with hypertension (above stage 1 hypertension systolic >140 mmHg and/or diastolic >90 mmHg). However, some of the patients had multiple diseases and hence were placed in more than one group. Control group consisted of 30 age and sex matched normal subjects attending the hospital outpatient department for a fitness-check up or some unrelated complaints and whose serum triglyceride and serum cholesterol levels were normal on recent laboratory reports. None of the controls had apparent atherosclerotic disease, diabetes mellitus or hypertension.
The Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III, or ATP III), published in 2002 recommends a complete lipoprotein profile: Total Cholesterol, LDL -Cholesterol, HDL-Cholesterol, and Triglycerides as the initial test for evaluating blood cholesterol. In our study, the selection of hyperlipidemia cases was done on the basis of laboratory reports of either hypercholesterolemia and/or hypertriglyceridemia as per the third report of the NCEP evidence-based guidelines for cholesterol testing and management:

Total Cholesterol: $\geq$ 240 mg/dl
Total Triglyceride: $\geq$ 200mg/dl [12]

The selection of the patients was done solely on the basis of their serum cholesterol and serum triglyceride levels on recent laboratory results regardless of their drug history. It is to be mentioned however, that most of these patients who had hyperlipidemia were on medication for the same.

2 ml blood sample was collected in EDTA bulbs from the antecubital vein by a clean puncture avoiding bubbles and froth. These samples were analyzed by the Sysmex K-4500 (TOA Electronics, Koebe, Japan) automated hematology analyzer for obtaining the platelet parameters – PVI (eg. MPV, PDW, P-LCR) and PC. The samples were run within two to six hours of venepuncture using the analyzer to avoid bias due to excessive platelet swelling.[13]

The statistical tests applied on our samples were mean, standard deviation, independent sample t test, ANOVA test, tuckey's test and Pearson's correlation coefficient (r) test. All these tests were done using the statistical software SPSS-17.0 at 95% confidence interval. The p-value < 0.05 was accepted as significant.

3. Result

A total of 130 subjects were enrolled for the present study. Out of these, in 1 case, the machine was not able to generate the numerical values of platelet volume indices PDW, MPV and P-LCR as the platelet count was too low. This one case was excluded for the calculations of PVI. Out of the total 100 cases of hyperlipidemia, 57 were males and 43 were females. The patients fell into the age group between 12 to 86 years. Maximum number of patients was between 51 to 70 years with a mean age of 52.86 years in study group and 57.10 in control group The study and control groups were found to be age and sex matched statistically. Hence, no bias in the results was observed due to age and sex in the results.

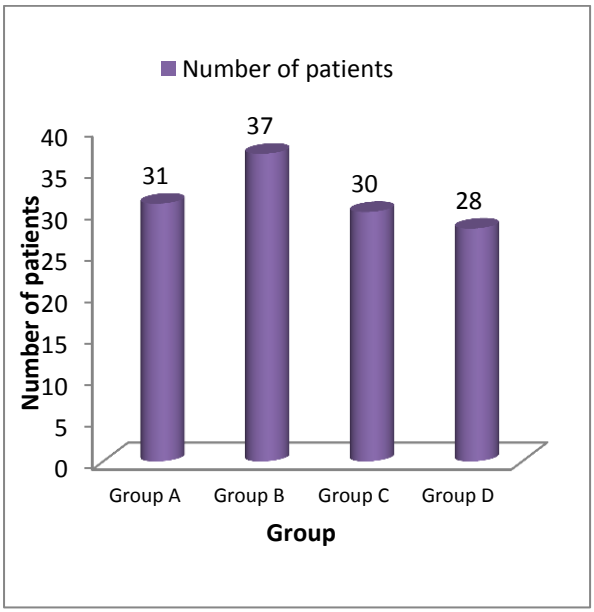

Figure 1 - Distribution of patients in groups

Figure 1 shows the distribution of patients in various groups with the maximum number of patients in group B consisting of patients having hyperlipidemia with atherosclerosis. 23 of the patients had multiple diseases and overlapped in groups B, C and D. The PC and PVI observed were compared between the study and control groups (Table1).

Mean MPV (9.79 ±1.01), PDW (13.18 ± 2.44) and P-LCR (24.85 ± 7.14) of cases were higher than the controls (mean MPV = 9.22 ± 0.91; mean PDW= 11.64 ± 1.75, mean P-LCR = 20.70 ± 7.07) as shown in Table-1. By using 2 independent sample t- test, p-value < 0.05 therefore there is significant difference between study group and control group with respect to MPV, PDW and P-LCR. And p-value > 0.05 for PC, therefore there is no significant difference between the study group and control group with respect to PC. So, all 3 studied PVI were found to be significantly higher in the hyperlipidemic patients than the normolipidemic patients.

On sub-categorizing the study groups and comparison of platelet parameters among various groups and the control group it was found that all three PVI were significantly higher in groups B, C and D than the control group whereas there was no significant difference in these indices in group A and the control (Table 2).

Additionally, patients in groups B, C and D had significantly higher PVI in comparison to the group A patients and the control population i.e.; hyperlipidemic patients having an associated disease had significantly higher PVI than the patients not having an associated disease with hyperlipidemia.

We correlated PC and PVI with the severity of hyperlipidemia and found a positive correlation between PVI and severity of hypercholesterolemia. MPV, PDW and P-LCR showed a poorly positive correlation with serum cholesterol levels (Correlation coefficient (r) and p-value were 0.300 and 0.003 respectively for MPV, 0.396 and < 0.001 respectively for PDW and 0.272 and 0.007 respectively for P-LCR) but not with triglyceride levels (p-value > 0.05).

4. Discussion

Hyperlipidemia, often an asymptomatic hidden state of blood might be the root behind a large number of complications including the life-threatening thromboembolic events. Role of platelets in such thromboembolic events is well-known and various platelet volume indices have been largely studied in these conditions. Present study was a humble attempt to study the changes in platelet volume indices in cases of hyperlipidemia.

The collection of samples was done in EDTA bulbs and the samples were run between 2 – 6 hours

Table1: Mean PC, MPV, PDW and P-LCR of cases against controls

Parameters	Group	No. of patients	Mean ± SD	p-value
PC	Study	100	247.41 ± 72.80	0.957
	Control	30	246.43 ± 89.68	
MPV	Study	99	9.79 ± 1.01	0.005
	Control	30	9.22 ± 0.91	
PDW	Study	99	13.18 ± 2.44	< 0.001
	Control	30	11.64 ± 1.75	
P-LCR	Study	99	24.85 ± 7.14	0.007
	Control	30	20.70 ± 7.07	

Table2: MPV, PDW, & P-LCR in groups A – D against that of the controls

GROUP	No. of patients	MPV		PDW		P-LCR	
		(Mean ± SD)	p-value	(Mean ± SD)	p-value	(Mean ± SD)	p-value
Control	30	9.22 ±0.91		11.64 ±1.75		20.70 ± 7.07	
A	31	9.12 ±0.87	0.660	11.69 ±2.13	0.920	20.35 ± 6.97	0.847
B	37	10.34±0.92	< 0.001	14.26 ±2.18	< 0.001	28.22 ± 6.03	< 0.001
C	29	9.90 ±0.87	0.005	13.49 ±2.47	0.002	25.73 ± 6.58	0.006
D	28	10.39 ±0.93	< 0.001	14.51 ±2.07	< 0.001	29.26 ± 6.19	< 0.001

after venepuncture to avoid bias due to swelling of platelets in EDTA in the light of the finding by Thompson et al that MPV increased for up to first 2 hours and then remained stable for eight hours in EDTA samples.[13]

We have used Sysmex K-4500 hematology analyzer for obtaining the various PVI and PC like some other recent studies.[8, 14,15,16] The chief advantage of using this analyzer is that it provides a relatively newer platelet parameter P-LCR which has been studied much lesser with respect to the other platelet parameters.

Studies have shown that platelet count and size might be gender and age dependent. [17,18] Hence we have conducted this case control study with both age and sex matched controls to avoid any such bias in our results.

We found that all three PVI – MPV, PDW & P-LCR were significantly higher (p-value <0.05) in the study group than the controls, i.e. hyperlipidemic patients had significantly higher MPV, PDW and P-LCR than the normolipidemic patients. We know that larger platelets are considered to be metabolically, enzymatically and functionally more active than the smaller platelets.[6] They contain more dense granules and hence are more potent and thrombogenic and this might be a cause for hyperlipidemia being a pre-thrombotic state.[15]

It is observed that hyperlipidemia is widely associated with many diseases so we decided to divide our study group of hyperlipidemia into further sub-groups- groups A – D based on the presence of the associated diseases.

Group A (n = 31; 23.85%) comprising of hyperlipidemic patients without any other associated disease, had no significant difference ($p > 0.05$) in their platelet count, MPV, PDW and P-LCR (p-value – 0.429, 0.66, 0.92 and 0.847 respectively) against the controls. A similar study using the same platelet volume indices in dyslipidemic patients done by Grotto et al found that MPV, PDW and P-LCR were significantly higher in dyslipidemic patients than in controls ($P < 0.00001$).[16] This is in discordance with our study if we take into account the results of group-A patients having hyperlipidemia only. However, an important point to be considered here is that Grotto et al did not mention whether their patients had associated diseases along with hyperlipidemia or not.[16] Similar to our study, Dogru et al found no association between MPV and abnormal lipid profile.[19] However, Pathansali R et al found an increased MPV in hypercholesterolaemic subjects and observed no changes in platelet count in hypercholesterolaemia.[20] Ravindran et al studied PC, PDW and plateletcrit in hypercholesterolemic patients and found that there was no significant difference in platelet counts between the healthy controls and the hyperlipidemic patients and an increase in PDW only in patients who had hyperlipidemia associated with CAD, similar to our study. They emphasized that existence of more than one risk factor is found to have a significant effect on the platelet hyper-responsiveness.[21] Similarly, we also found in our study that existence of an associated disease with hyperlipidemia alters the platelet volume indices significantly as compared to the presence of hyperlipidemia alone. Fuchs J et al found an increased percentage of big platelets in hyperlipidemic patients which persisted even after treatment despite the improvement in lipoprotein profile. So they thought that the relation of big platelets and lipid abnormalities is questionable.[22] We also could not establish the relationship between mere hyperlipidemia and platelet size. However, in contrast to their study, mere hyperlipidemia group of our study did not have big platelets.

Our group-B patients (n = 37; 28.46%) had atherosclerotic diseases and/or acute thromboembolic events in conjunction with hyperlipidemia and all the PVI (MPV, PDW, P-LCR) ($p<0.001$) were significantly higher in this group than the normal controls. This was comparable to the results obtained by many earlier studies.[8, 14, 23] However, in our study; PC was not significantly different from the normal controls ($p = 0.997$). This was similar to the Pizulli et al study but in contrast Khandekar et al and Ranjith et al found a decreased platelet count in AMI patients as compared to the controls.[8, 14, 23]

Group-C patients (n = 29; 29%) in our study had diabetes mellitus along with hyperlipidemia and it was found that these patients also had significantly higher MPV, PDW and P-LCR (p-value is 0.005, 0.002 and 0.006 respectively) than the control group. Here also we didn't get any significant difference for PC between the group-C and the controls. Our results were similar to a very recent study done by Jindal et al who also found all these three platelet volume indices higher in the diabetic patients compared to the controls and comparable platelet counts in the diabetics and the controls.[15] This strengthens the hypothesis that hyperglycemia contributes to heightened platelet reactivity directly as well as through glycation of platelet proteins and hence results in an increase in platelet volume indices.

Group-D patients (n = 28; 21.54%) in our study had hyperlipidemia in association with hypertension. We found that all three PVI; MPV, PDW and P-LCR were significantly higher in this group as compared to the controls (Table- 2; p value <0.001) while there was no significant difference in the PC between the two groups.

These findings concluded that the patients who were having hyperlipidemia associated with other diseases had significantly higher PVI i.e. larger platelets than the normolipidemic patients. However, the patients who had hyperlipidemia alone without any other disease condition did not have any significant difference in their platelet size against normolipidemic patients. This led us to analyze whether there was any difference in these indices in hyperlipidemic patients themselves when patients had hyperlipidemia with and without complications. It was noticed that hyperlipidemia when associated with atherosclerosis (group B), diabetes mellitus

(group C) or hypertension (group D) had higher PVI than when it was present in isolation. However, again no significant difference in platelet count was noticed in different groups. It leads us to a new insight that hyperlipidemic patients who have larger platelets are more likely to have an associated disease condition and the platelet volume indices may form a basis for the prediction of these diseases in hyperlipidemic patients.

An attempt was also made to correlate these platelet volume indices with the severity of hyperlipidemia. For this, PC and PVI values were correlated with the increasing values of total cholesterol and triglyceride levels. It was found that all 3 PVI had a poor positive correlation with increasing total cholesterol; however no correlation was seen with increasing triglyceride. This means that platelet size was directly proportional to the severity of hyperlipidemia (hypercholesterolemia). We suggest that this might be possibly due to the more likelihood of occurrence of hyperlipidemia associated complications like thrombotic diseases with increase in its severity. Another hypothesis can be that it might be possible that hyperlipidemia alone may play some role in the activation of the platelets causing raised PVI when it increases in severity even in the absence of other diseases.

Categorical division of the hyperlipidemic patients into different groups and further group-wise analysis was the main strength of this study. A previous study which found these PVI to be higher in dyslipidemic patients missed to take this point into consideration and did not mention the status of other diseases in their cases of dyslipidemia which might have resulted into a different result from us.[16] We had certain limitations and constraints in this study. Although we took a sample size of 100 patients in this study, the division of them into subgroups gave us a very small sample size for each sub- group. So, further such studies with larger sub- groups may throw some more light in this matter.

5. Conclusion

The present study highlights the platelet volume indices in hyperlipidemic patients compared to control subjects. Categorical analysis, which is the strength of this study, showed that it is the presence of other diseases in hyperlipidemic patients that brings the platelet in pre-thrombotic active state as reflected by the changes in its PVI. We recommend further studies to emphasize this hypothesis with larger sample sizes for each category with similar categorical analysis. The estimation of these PVI can be considered as an early, economical and rapid procedure for identification of complications in hyperlipidemic patients.

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Competing Interests

None declared

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