ORIGINAL ARTICLE

THE STUDY OF PLATELET VOLUME INDICES IN PLATELET APHAERESIS PROCEDURE: AN EXPERIENCE OF 271 PLATELET APHAERESIS PROCEDURES

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ABSTRACT

Background: Platelet activity can be assessed by platelet volume indices like MPV, PDW & P-LCR.

Aim: To develop an approach in blood bank professional, a habit of looking at platelet indices in hematology analyzer report of aphaeresis donors & QC samples of platelet aphaeresis products.

Methods and materials: A retrospective data analysis was done for 271 platelet aphaeresis procedures conducted on CS3000 plus with AMS cell separator, Fenwal, USA& COM.TEC, Fresenius Kabi, Germany. Samples of the donors were collected before aphaeresis & 1 to 2 ml sample from each bag was collected in the satellite pouch attached to bag & analysis was done on day 0 & day 7. Platelet parameters were measured on automated hematology analyzers SYSMEX KX-21 & Horiba Micros 60.Statistical analysis: Statistical analysis was done by calculating 'r value' & a paired t test at 95 % confidence interval. A *P* value of <0.05 was taken as significant.

Results: The mean platelet yield was 3.39±0.88 x 10^{11}/unit. The platelet yield correlated negatively with MPV, PDW and PLCR (r value -0.224, -0.045 & -0.159 respectively for correlation between MPV, PDW & PLCR with the yield, *P* <0.0001).The mean values of PVI of SDP were significantly smaller than that of donor pre-donation samples (paired t test *P* value < 0.05).The size of stored single donor platelet on day 7 were significantly larger than that of day 0 (*P* value < 0.05).

Conclusion: The platelet indices are useful to study - selectively smaller platelet separation by automated cell separators, storage lesions & yield prediction.

Key words: Plateletphaeresis, MPV, yield, storage lesion, Platelet derived micro particles.

INTRODUCTION

Platelet size correlates with platelet activity and can be assessed by platelet volume indices (PVI). Mean Platelet Volume (MPV) is a machine-calculated measurement of the average size of platelets found in blood and is typically included in blood tests as part of the Complete Blood Count. The distribution width at the level of 20% was defined as platelet distribution width (PDW), and the percentage of the platelets with a size of more than 12 fL was defined as platelet large cell ratio (P-LCR).[1](figure-1)

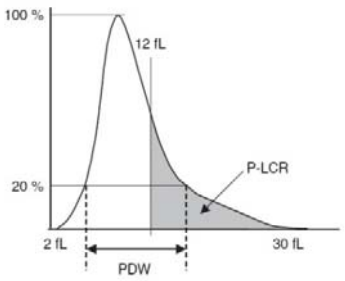

Figure-1: Histogram of platelet size distribution and the definition of platelet size deviation width (PDW) and platelet-large cell ratio (P-LCR)

Platelet recovery in the patient is influenced by the transfused dose of platelets, which in turn is dependent on the quality of Single Donor Platelet (SDP) in terms of platelet yield.[2] Donor platelet count has a direct impact on the yield of platelets.[3] In addition to few other variables, platelet indices of aphaeresis donors have also influences on yield of platelet product.

Platelet Concentrates (PCs) undergo alterations during collection, processing, and storage that adversely affect their structure and function. These changes commonly referred to as the platelet storage defect (PSD) or platelet storage lesion (PSL), are important because they are associated with decreased post transfusion in vivosurvival.[4]Various laboratory tests have been recommended to study PSL ranging from most simple test such as *p*H to more complex tests of platelet function.[5] Platelet count, MPV, PDW, and P-LCR have been also used as markers for the quality control of PCs, as these reflect storage induces shape changes in platelets.[6]

Platelet-derived micro particles (PMPs), small particles derived from the membranes of intact platelets, are present in PCs. PMPs are strongly pro-coagulant and there is evidence that they retain many of the biologic properties of intact platelets. Various mechanisms may contribute to PMPs formation in PCs, including direct mechanical injury and exposure to stresses during component preparation. In addition, PMPs are formed as a result of the inevitable degrees of platelet activation that occur during platelet processing and storage activation that partially depends on interactions between platelets and the plastic storage container.[4]

In present study, the platelet volume indices were studied for its influence on the yield of SDP, PMPs formation during the aphaeresis procedure and PSL in aphaeresis product.

MATERIAL & METHODS

A retrospective data analysis was conducted for platelet aphaeresis donors and procedures conducted during August 2006 to August 2014 at Blood Bank of government tertiary level hospital. During this period total number of 422 platelet aphaeresis procedures were done on CS 3000 plus with AMS cell separator, Fenwal, USA (329 procedures) & COM.TEC, Fresenius Kabi, Germany, Software version 4.03.xx (93 procedures) using closed & open system aphaeresis kits as per the equipment operating manuals & studied various parameters. Out of these 422 procedures, data of 271 procedures were included in the present study. During this period, all donors were met the donor eligibility criteria as laid down by the Food & Drug Administration Act, India. All data was collected from the aphaeresis records maintained in the blood bank, hence ethical committee clearance is not necessary.

Platelet counts & Platelet volume indices (PVI) were measured on automated hematology analyzer SYSMEX KX-21 (Sysmex Corporation Kobe, Japan) & Horiba Micros 60, ABX Diagnostics, France as per operating manual of the hematology analyzer. To exclude the theoretical risk of counting outside the counter linearity, the samples of the SDPs were diluted 1:2 with saline and then the platelets were counted. EDTA samples of the donors were collected before aphaeresis donation and approximately 1 to 2 ml sample from each bag was collected in the satellite pouch attached to bag to ensure representative product of the platelet bag & analysis done on day 0 & day 7.

Statistics*:* Influences of donor pre donation PVI on the yield of SDP were studied by calculating 'r value' (Pearson Correlation) at 95 % confidence interval using MS Excel Software (MS Office 2010). A paired t test was used to find out difference in PVI is significant or not. All statistical tests were two-sided and performed at a significance level of $\alpha = 0.05$. A *P* value of < 0.05 was taken as significant.

RESULTS

A total of 422 platelet aphaeresis procedures performed on automated cell separators. The mean platelet yield of all procedures was $3.39 \pm 0.88 \times 10^{11}$. The mean value of donor pre-donation platelet indices MPV, PDW and P-LCR were 8.66 ± 1.26, 11.61 ± 2.37 and 19.43 ± 7.85 respectively. The mean values of platelet indices of SDP on day 0 & day 7 are given intable-1. A negative correlation was observer between donor's pre-donation MPV, PDW & P-LCR with the yield of SDP. The Pearson value r were -0.224, -0.045 & -0.159 respectively for correlation between MPV, PDW & PLCR with the yield of SDP ($P< 0.0001$). The mean values of PVI of SDP were significantly smaller than that of donor predonation samples (Two sided paired t test *P* value< 0.05 for MPV, PDW & PLCR). The sizes of stored

SDP on day 7 were significantly larger than that of day 0 (Two sided paired t test *P* value< 0.05 for MPV, PDW & PLCR)

Table 1: Platelet volume indices of donor and SDP

Platelet volume indices	Donor (n=271)	SDP-Day 0 (n=213)	SDP-Day 7 (N=36)
MPV	8.66 ± 1.26	7.46 ± 1.16	9.66 ± 1.15
PDW	11.61 ±2.37	10.25 ± 2.42	16.82 ± 3.18
P-LCR	19.43 ± 7.85	11.81 ± 6.38	26.69 ± 9.20

Value are mentioned in mean ± SD

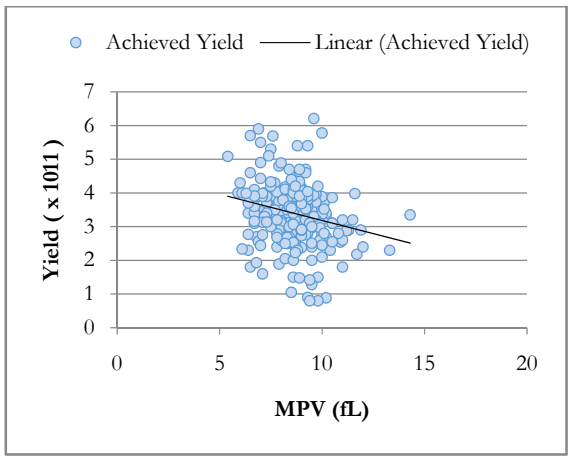

Figure-2.2: PVI (MPV and P-LCR) versus Yield

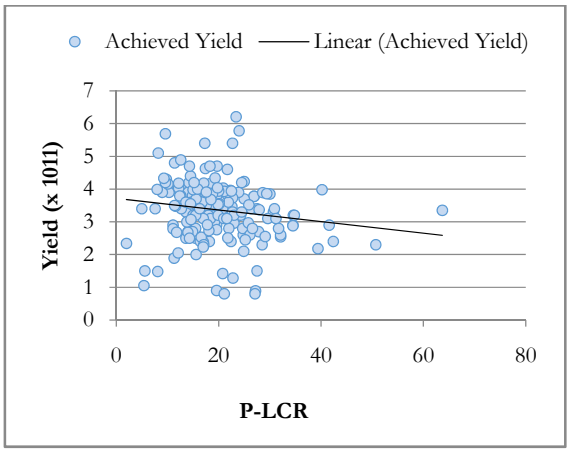

Figure-2.2: PVI (MPV and P-LCR) versus Yield

DISCUSSION

The main aim of this study is to develop an approach in blood bank professional, a habit of looking at hematology analyzer report of aphaeresis donor & QC samples of platelet aphaeresis products.

In the present study, 162 (60%) donors had MPV up to 9 fl and the mean platelet yield in these donors was 3.52 ± 0.85 x 10^{11}/unit, 96 (35%) donors had MPV in the range of 9-11fl and the mean platelet yield obtained in this range was 3.25 ± 0.92 x 10^{11}/unit. 13 (5%) donors had MPV greater than 11fl and the mean yield obtained in this group was 2.84 ± 0.58 x 10^{11}/unit. This observation stated that higher MPV values corresponded to lower platelet yield. There was a direct negative correlation between the donor pre-donation MPV & P-LCR with the yield of SDP (r value -0.223 & -0.159 respectively, *P*< 0.05, figure-2). The reason behind this can be attributed to the separation mechanism of the automated cell separators used in this study. The separation of platelets by automated cell separatorsis based on cell size, as the size of platelets increases, these large platelets are excluded from collection mimicking as red cells. The *P* value of two sided paired t test was less than 0.05 which was also favouring the observation of selective separation of smaller platelets during the procedure of platelet aphaeresis. This observation is similar to the study performed by P Sachdeva et al in which the author concluded direct negative correlation between the mean MPV of the donor and the platelet yield obtained (r = - 0.447, *P*< 0.001).[7] According to study performed by Brozovic et al, the mean MPV for PC collected with Spectra and CS-3000 cell separators was 8.48 ± 0.52 fl (n = 20) and 8.94 ± 0.60 fl (n = 12), respectively, and was significantly higher (*P*< 0.01) than that determined in venous blood samples of donors taken before plateletpheresis (7.76 ± 0.74 fl and 8.12 ± 0.62 fl respectively).[8] Thus study performed by Brozovic et al indicates preferential separation of large platelets during aphaeresis, which was not as per the present study.

The observations of the study done by P Sachdeva et al & the present study lead to possibility of either selective separation of smaller platelets or formation of PMPs formation during aphaeresis procedure. To rule out possibility of sheer stress induced PMPs formation during the procedure of aphaeresis, we need to study & quantify by flow cytometric methods based on light-scattering properties and surface expression of GPIb-IX or GPIIb-IIIa.[4]

The PDW as analyzed on standard hematology cell counters is an indicator of size dispersion in the

platelet population. Platelet concentrates prepared for transfusion shows an increase in PDW over storage. This increase correlates strongly with in vitro indicators of platelet viability (pH and response to osmotic stress). Hematology cell counter also measures the largest platelets in the platelet population as a large cell ratio (P-LCR).[9] Table-1 shows that the mean values of MPV, PDW & P-LCR in present study on day 7 was significantly higher than the same values on day 0 of SDPs (*P* value <0.0001). Study performed by Albert Farrugia et al also correlated with the same above findings.[9] Harprit Singh et al had found that mean MPV on days 0 and 7 were 9.65 and 9.61 fl respectively & stated that MPV alone did not change significantly over 7 days of storage.[10] Thus the present study did not correlate with the findings of Harprit Singh et al & there was increase in mean MPV value from day 0 to day 7 during storage indicating morphological changes that had occurred during storage period because of platelets viability & metabolically activity during storage. The studies performed by S Basir et al & T Chandra et al are also in agreement with present study as they concluded that the values of MPV & PDW were significantly higher (*P*< 0.0001) than the values of day 0 for stored platelet products.[11, 12]

CONCLUSION

PVI mainly MPV has negative correlation with yield of aphaeresis products in other words the smaller platelets are collected more efficiently by the automated cell separators and hence yielding a better product. PVI can be used to study platelet derived micro particle formation, platelet activation due to shear stress during aphaeresis procedure, however other tests such as Flow Cytometry is required in addition to PVI study. PVI can be used for quality monitoring of stored single donor platelets and hence one can predict usefulness of SDP in terms by transfusing viable platelets.

The present study was not complete in terms of studying PSL or PMPs formations. The present study was data analysis to represent role of PVI as yield predictors of SDP, role in studying PMPs formation due to sheer and stress & in PSL studies.

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