Editorial

Platelet storage lesion: An update

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Platelets are unique blood cells that play a pivotal role in mediating hemostasis and thrombosis. Over the years, the demand for platelets is steadily rising because of their increased use in patients with thrombocytopenia and platelet function defects. The availability of platelets for transfusion is restricted by the markedly short shelf life of platelets owing to risk of bacterial contamination and storage-related deterioration known as platelet storage lesion (PSL).[1] PSL is best defined as the sum of all the deleterious changes in platelet structure and function that arise from the time the blood is withdrawn from the donor to the time the platelets are transfused to the recipient.[2] The mechanisms responsible for PSL are multifactorial and not clearly understood. Several factors including the methods of collection, processing, storage, and manipulation after collection can result in PSL.[3] These lesions are associated with decreased *in vivo* platelet recovery, survival, and hemostatic activity after transfusion.[4]

Preservation of platelet structure, composition, and function during preparation and storage is one of the primary goals of transfusion medicine practice.[5] Technical advances in the form of introduction of automated cell separators, optimized centrifugation procedures, improved storage containers, leucoreduction, and platelet additive solutions (PAS) have greatly facilitated the collection and storage of platelets. Despite these advances, PSL is still a matter of concern. Platelets get activated following exposure to foreign surfaces, trauma, low pH, agonists (thrombin, ADP), and shear stress. Upon activation, the platelets lose their discoid morphology and become more spherical with multiple pseudopods. Conformational changes in GPIIb/IIIa complex exposes binding sites for adhesive proteins (fibrinogen, vWF) resulting in platelet aggregates. Activation further stimulates release of granular contents and expression of sequestered membrane proteins on outer surface. Release of these granular contents at the site of injury is indispensable for platelet function as the contents not only mediate the recruitment of leucocytes and platelets, promote clotting but also provide immunity against infection and contribute to wound healing.[6] Moreover, the presence of these contents in storage medium can lead to various transfusion reactions.[7] Expression of negatively charged phospholipids (phosphatidyl serine and phosphatidyl ethanolamine) increases on the outer leaflet of activated platelets, providing a surface for the prothrombinase complex (X-Va), thereby contributing to procoagulant activity.

Exposure to shear stress, as in centrifugation, during component separation and agitation while storage not only activates platelets but can also cause their lysis and calpain activation.[5] Platelet lysis leads to discharge and accumulation of cytosolic lactate dehydrogenase (LDH) and granular contents. Calpain (protease) activation results in degradation of cytoskeletal proteins such as actin, talin, and actin binding protein generating platelet microvesicles.[5] Microvesicle formation leads to decrease in mean platelet volume (MPV) and also contributes to procoagulant activity. Adenosine triphosphate (ATP) depletion occurs as ATP is required for each aspect of platelet function and is generated via oxidative and glycolytic pathways. Glycolysis is upregulated under low oxygen conditions. Lactate produced via glycolysis is buffered primarily by plasma bicarbonate. Progressive lactate production and bicarbonate exhaustion during storage results in deleterious decrease in pH augmenting platelet damage. Platelet apoptosis occurring simultaneously during storage further contributes to loss of mitochondrial function, cytoskeletal damage, and surface expression of phosphatidyl serine. In addition, consumption of nutrients by platelets and contaminating leucocytes and accumulation of metabolic end products such as cellular debris, activated clotting factors, proteolytic enzymes, and cytokines in plasma also adversely affect the functional integrity of platelets.[8]

A battery of *in vivo* and *in vitro* tests have been adopted to assess PSL. Corrected count increment after platelet transfusion and bleeding time studies are commonly used for evaluating product efficacy and improved hemostasis, respectively. However, these do not reflect the true picture due to uncontrollable patient variables. Another alternative approach to measure the efficacy of product transfused is to measure the *in vivo* recovery and survival of fresh or stored radiolabelled or biotinylated platelets in healthy volunteers.[9] However, these *in vivo* studies are expensive, time consuming, and complex to perform.

Biochemical tests like decrease in pH, pO_2, LDH accumulation, glucose consumption, and ATP

depletion are employed to assess platelet viability. Aging has also been quantified by *in vitro* tests measuring changes in morphology of platelets. Swirling phenomenon, decrease in MPV, reporting of the percentage of discoid forms, and Kunicki morphological score have been used for measuring the alterations in the discoid morphology. Extent of shape change in response to an agonist and increase in hypotonic shock response provide more objective measure of morphological changes.

Platelet activation markers such as release of specific granular contents into plasma (β thromboglobulin, platelet factor 4, and RANTES), changes in GP expression on platelet surface (GPIb, GPIIb, and GPIIIa), expression of sequestered granular membrane proteins (P-selectin - CD62P, CD63, and CD40L), and phosphatidyl serine (determined by Annexin V binding) on surface occurring during processing and storage have been reported.[9] Tormey and Stack observed diminution of secretory capacity of platelets during processing and storage using cytokine (RANTES) release assay.[6] Wenzel *et al*., used soluble CD40 ligand (sCD40L) to study the effect of storage on platelet viability by monitoring sCD40L levels in storage medium. They noticed an increase in sCD40L concentration and decrease in platelet release capacity with an increase in duration of storage.[7]

Jain *et al*., used these *in vitro* tests to observe correlation of morphological changes with platelet activation markers in platelet concentrates (PCs) prepared either from whole blood using platelet-rich plasma (PRP) method or buffy coat reduced method (BC) and apheresis platelets (AP-PC). Morphology score (MS), decrease in pH, glucose levels, lactate concentrations, soluble P-selectin (sP-selectin) levels were determined in PRP-PCs, BC-PCs and AP-PCs from day of collection to the last day of storage. A negative correlation was observed between MS and sP-selectin and a positive correlation was found between MS and pH. AP-PCs were found to be superior to BC-PC and PRP-PC with respect to quality control parameters, morphological changes, and platelet activation markers.[10]

Though *in vitro* tests are more economical and rapid, but none of these tests can accurately predict *in vivo* recovery of transfused platelets and correlation has been documented only in extreme values.[11] Moreover, majority of these tests are usually applied for research purposes and only a few tests like pH, platelet volume, platelet count, and leucocyte content have been used for daily quality control monitoring.[8]

Efforts till date at optimizing and improving the viability of stored platelets have largely focused on maintaining pH by buffering lactate production and enhancing gaseous exchange. Gaseous exchange in turn is dependent upon platelet content, gas diffusion through the plastic container, and agitation. Improved second generation containers currently in use for storing platelets (manufactured from polyolefin or polyvinyl chloride (PVC) plasticized with compounds such as triethylhexyltrimellitate and butyryltrihexyl citrate) have twice the oxygen permeability compared with first generation bags (PVC with diethylhexyl phthalate).

Growing use of leucoreduced PCs has further contributed towards enhancing platelet safety as contaminating leukocytes not only compete with platelets for nutrients but the cytokines (IL-6, TNF-α, and IL-β) generated following their fragmentation and activation have also been associated with various transfusion reactions such as febrile nonhemolytic transfusion reactions, transfusion-related acute lung injury (TRALI).[12] Leucoreduction also decreases alloimmunization and transmission of leucotropic viruses. Kaur *et al*., studied the levels of cytokines generated during storage and their correlation with leucocyte content. Significant rise in cytokine levels were observed in non-leucofiltered group compared with leucofiltered group.[13]

Development of synthetic storage solutions also known as PAS to store platelets is a step further in ameliorating PSL and improving platelet quality. Removal of plasma results in removal of enzymes reducing their proteolytic effect on platelet membrane. In addition, the use of PAS minimizes the adverse effects mediated by plasma like allergic reactions and TRALI, allows the use of photochemical pathogen reduction techniques, and potential improvement in platelet storage by engineered manipulation of storage medium.[14] Reduction in adverse reactions has been reported when comparing PAS preserved platelets to plasma suspended units.[15] Platelet morphology and function was found to be better preserved in additive solution on day seven as compared to platelets stored without additive solution by Chandra *et al*.[16]

Platelet demand has increased over the past decade as they are not only being used to control or prevent bleeding but also being increasingly used as a source of growth factors in tissue repair, wound healing, and skin rejuvenation. Studies have shown promising results of use of topical platelet therapy in various clinical conditions.[17-20] Therefore, retention of platelet functionality as much as possible is of prime importance as this may help in extension of shelf-life, and thus better inventory management.

References

1. Thon JN, Schubert P, Devine DV. Platelet storage lesion: A new understanding from a proteomic perspective. Transfus Med Rev 2008;22:268-79.
2. Thon NJ, Devine DV. Translation of glycoprotein IIIa in stored blood platelets. Transfusion 2007;47:2260-70.
3. Seghatchian J, Krailadsiri P. The platelet storage lesion. Transfus Med Rev 1997;11:130-44.
4. Holme S, Moroff G, Murphy S. A multi-laboratory evaluation of *in vitro* platelet assays: The tests for extent of shape change and response to hypotonic shock. Transfusion 1998;38:31-40.
5. Fisk JM, Pisciotto PT, Snyder EL, Perrotta PL. Platelet and related products. In: Hillyer CD, Silberstein LE, Ness PM, Anderson KC, Roback JD, editors. Blood Banking and Transfusion Medicine. 2nd ed. Philadelphia: Churchill Livingstone; 2007. p. 308-41.
6. Tormey CA, Stack G. Use of a cytokine-release assay to demonstrate loss of platelet secretory capacity during blood bank processing and storage. Arch Pathol Lab Med 2014;138:1481-7.
7. Wenzel F, Gunther W, Baertl A, Lasshofer R, Rox J, Fischer JC, *et al.* Comparison of soluble CD40L concentrations and release capacities in apheresis and prestorage pooled platelet concentrates. Clin Hemorheol Microcirc 2011;47:269-78.
8. Shrivastava M. The platelet storage lesion. Transfus Apher Sci 2009;41:105-13.
9. Vassallo RR. Preparation, preservation and storage of platelet concentrates. In: Simon TL, Snyder EL, Solheim BG, Stowell CP,

Strauss RG, Petrides M, editors. Rossi's Principles of Transfusion Medicine. 4th ed. West Sussex: Wiley-Blackwell; 2009. p. 187-98.
10. Jain A, Marwaha N, Sharma RR, Kaur J, Thakur M, Dhawan HK. Serial changes in morphology and biochemical markers in platelet preparations with storage. Asian J Transfus Sci 2015;9:36-42.
11. Murphy S. Utility of *in vitro* tests in predicting the *in vivo* viability of stored PLTs. Transfusion 2004;44:618-9.
12. Heddle NM, Klama L, Singer J, Richards C, Fedak P, Walker I, *et al.* The role of the plasma from platelet concentrates in transfusion reactions. N Eng J Med 1994;331:625-8.
13. Kaur D, Sharma RR, Marwaha N. Defining an appropriate leucoreduction strategy by serial assessment of cytokine levels in platelet concentrates prepared by different methods. Asian J Transfus Sci 2015;9:26-30.
14. Andreu G, Vasse J, Herve F, Tardivel R, Semana G. Introduction of platelet additive solutions in transfusion practice. Advantages, disadvantages and benefit for patients. Transfus Clin Biol 2007;14:100-6.
15. Cohn CS, Stubbs J, Schwartz J, Francis R, Goss C, Cushing M, *et al.* A comparison of adverse reaction rates for PAS C versus plasma platelet units. Transfusion 2014;54:1927-34.
16. Chandra T, Gupta A, Kumar A, Afreen S. Morphological and functional changes in random donor platelets stored for seven days in platelet additive solution. Int J Blood Transfus Immunohematol 2011;1:20-5.
17. Zhao JG, Wang ZL, Zhao L. Platelet rich plasma in arthroscopic rotator cuff repair. Arthroscopy 2015;31:9-10.
18. Bashir J, Panero AJ, Sherman AL. The emerging use of platelet rich plasma in musculoskeletal medicine. J Am Osteopath Assoc 2015;115:24-31.
19. San Sebastian KM, Lobato I, Hernandez I, Burgos-Alonso N, Gomez-Fernandez M, Lopez J, *et al.* Efficacy and safety of autologous platelet rich plasma for the treatment of vascular ulcers in primary care: Phase III study. BMC Fam Pract 2014;15:1129.
20. Godse K. Platelet rich plasma in androgenic alopecia: Where do we stand? J Cutan Aesthet Surg 2014;7:110-1.

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