Modified Platelet Storage Device to Improve Quality During Storage

Nicolas Pereyra[1], Helen Chen[2], Kai Yu[2], Jayachandran Kizhakkedathu[2], and Dana Devine[1]

Department of Biomedical and Molecular Sciences[1] and Chemistry[2], University of British Columbia, Vancouver, BC, Canada

Abstract

Platelet transfusion is a lifesaving therapy for trauma and various disorders. Current platelet storage devices limit their shelf life to 5-7 days, which applies pressure on supply of these cells. The bio-incompatible bag materials activate platelets and lead to a decreased product quality in a process known as the Platelet Storage Lesion (PSL). Platelet concentrates are also prone to bacterial contamination which results in adverse transfusion reactions and sepsis. Pathogen Inactivation Technologies (PITs), a host of techniques which destroy nucleic acids, have recently been brought to market. These technologies can damage platelets and exacerbate the PSL. This project aims to develop a platelet bag coating formulation which extends the storage life of platelets and has antimicrobial properties. The Kizhakkedathu lab has developed a universal anti-biofilm coating based on polydopamine co-assembly with an assortment of ultra-large hydrophilic polymers. This coating displays antibacterial properties and can decrease adverse platelet-bag interactions. Various polymer coating formulations will be screened to determine which have the best biocompatible and antibacterial properties for both PIT-treated and untreated platelets. Platelet activation and apoptosis during storage will be measured through surface expression of P-selectin and phosphatidylserine. Platelet activation and metabolism will also be tracked using a blood gas analyzer for the measure of pH, glucose, lactate, pO_2, and pCO_2. Platelet function will be assessed by rotational thromboelastometry (ROTEM) and aggregometry. These results will guide the development of an optimized device. This research will lead to better platelet storage bags which alleviate supply strain and improve transfusion outcome.

Background

Storage conditions

- Platelets are stored on 22 °C shakers for 5-7 days
- Bags are made from polyvinyl chloride (PVC) plasticized with di(2-ethylhexyl) phthalate (DEHP) (Figure 1.).

Platelet Storage Lesion

- Platelets are activated by the hydrophobic surface of the PVC bags[1].
- Storage and activation comprise the quality of the platelet concentrates, known as the Platelet Storage Lesion (PSL)[2].

Bacterial Contamination

- Pathogen contamination of platelets is a problem for both recipient safety and blood product supply.
- One in 1500-2000 PCs is contaminated[3].
- The bacterial screening period is 24 h., putting further strain on platelet supply and shelf-life.

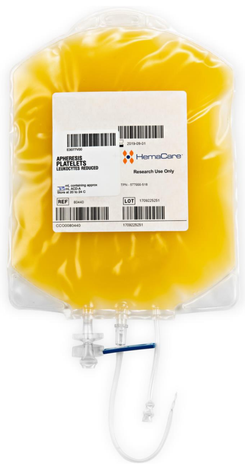

Figure 1. Apheresis platelet storage bag. Approximately 350 mL of platelet-rich plasma is stored in PVC-DEHP bags.

Pathogen Inactivation Technology

- Pathogen Inactivation Technologies (PITs) destroy nucleic acids.
- PITs also damage platelets, leading to faster clearance and poorer transfusion outcome[4].
- Intercept™ Blood System (Cerus Corporation) uses a photoactivated psoralen to crosslink pyrimidines.
- Mirasol™ (Terumo BCT, Inc.) uses photoactivated riboflavin (vitamin B_2) to generate reactive oxygen species which oxidize guanines.

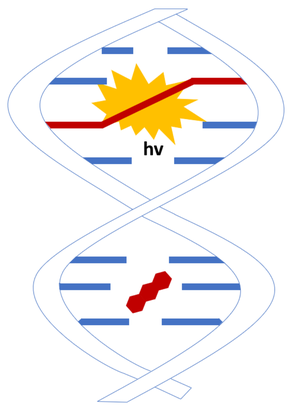

Figure 2. Diagram of the Intercept™ blood system mechanism. Amotosalen (red hexagons) is activated by UV light between 320-400 nm (hv). Amotosalen then cross-links pyrimidines (red line).

Objectives

1. Develop a storage bag coating which extends the shelf life of platelets.
2. Optimize the coating for Pathogen Inactivation Technologies.

Hypothesis

I hypothesize that the novel platelet storage device will increase the viable storage period of platelets treated with PITs.

Anti-Adhesive Platelet Bag

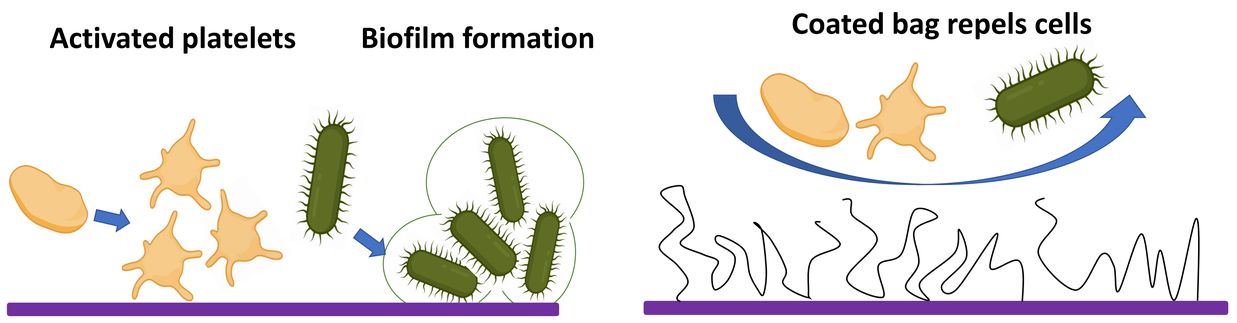

Figure 3. Graphic of the platelet storage bag coating. The figure on the left is a schematic of the uncoated bag. Without a protective layer, platelets (orange cells) are activated (orange cells with filopodia) by the bag's hydrophobic surface (purple), and bacteria (green cells) are free to form biofilms (green outline). The bottom figure shows the schematic of the bag surface with the proposed polymer coating. The polymers (black lines) protect platelets from being activated by the bag. This coating also prevents bacterial adhesion, inhibiting the formation of biofilms. Figure produced using Biorender.com.

Biocompatible and Antimicrobial Coating

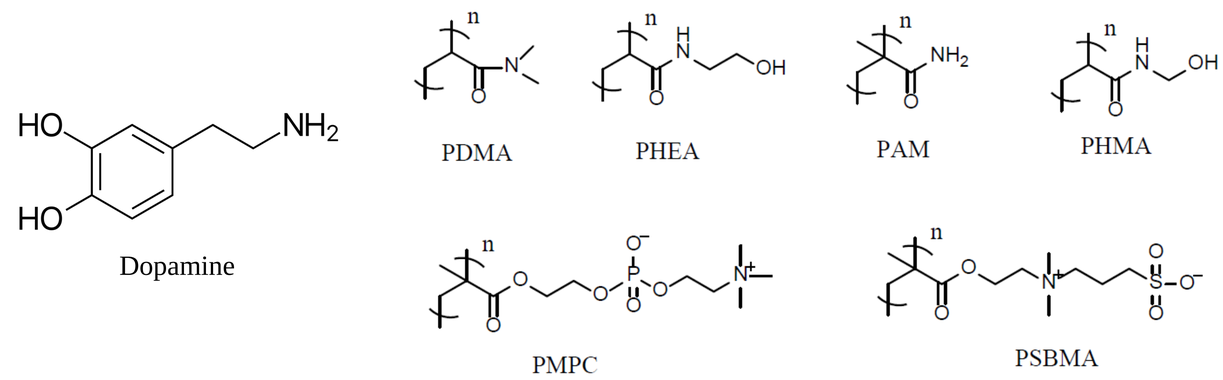

Figure 4. Library of candidate polymers for the platelet storage device coating. A list of ultra-high molecular weight polymers (uHMWP) will be used to determine the optimal coating for the interior of the platelet bag. The names of the polymers are Poly(*N,N*-dimethylacrylamide) (PDMA), Polyacrylamide (PAM), Poly(*N*-hydroxymethyl acrylamide) (PHMA), Poly(*N*-hydroxyethyl acrylamide) (PHEA), Poly(*N*-(tris(hydroxymethyl)methyl)acrylamide) (PTHMAM), Poly(2-methacryloyloxyethyl phosphorylcholine) (PMPC), and polysulfobetaine (PSBMA). UHMWPs with a molecular weight between 800 k and 1 million Da with a polydispersity of < 1.5 will be used. The uHMWPs are anchored to the PVC surface through dopamine polymers (polydopamine) which bind to the bag through the molecule's catechol groups.

Screening Process

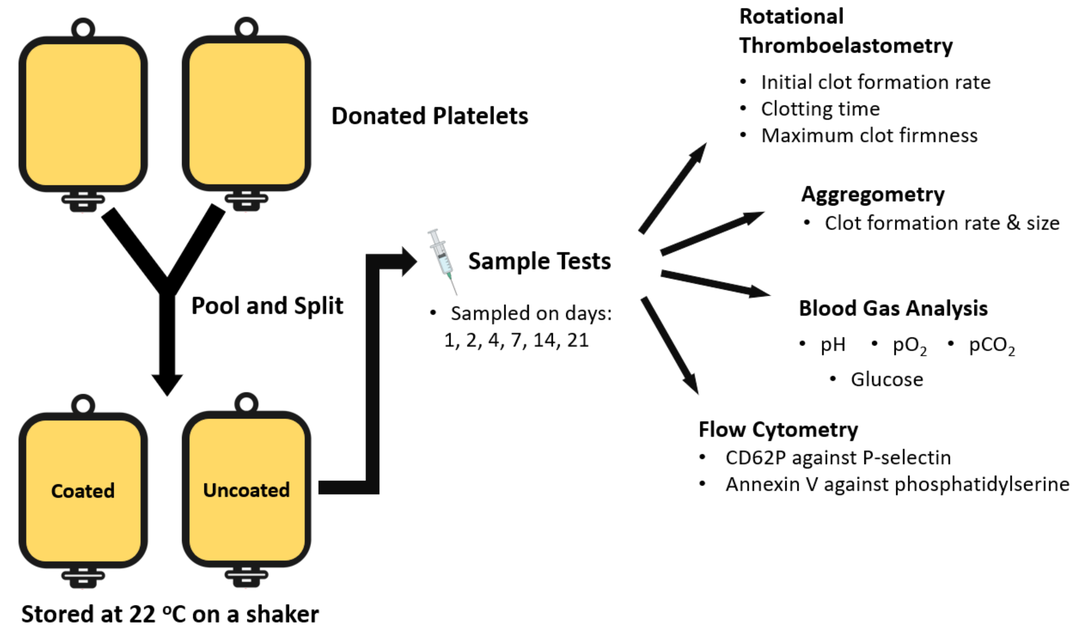

Figure 5. Flowchart of the platelet characterization experiments after storage in the coated platelet bags. Two apheresis platelet concentrates are pooled and mixed together before being split into one coated and one uncoated bag. Platelets are then stored as per Canadian Blood Services standard procedure (22 °C on a shaker). The platelets are sampled on days 1, 2, 4, 7, 14, and 21. On these days, the cells are subject to a host of biochemical and cell physiology assays. Platelet activation and apoptosis are measured by immunofluorescent staining of P-selectin and phosphatidylserine, respectively. Platelet activity and metabolism is tracked through blood gas analysis of pH, pO_2, pCO_2, and glucose. Platelet function is assessed by rotational thromboelastometry and aggregometry. Figure produced using Biorender.com.

Acknowledgements

I would like to acknowledge the blood donors, the Network Center for Applied Development (NetCAD), and Canadian Blood Services. This project has been graciously supported by the Canadian Institute of Health Research funding granted to DVD and Jay Kizhakkedathu.

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