Next-Generation Platelet Storage Units for Better and Safer Transfusions

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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

- The hydrophobic surface of the bags activates platelets and plasma proteins[1].
- Storage and activation comprise the quality of the platelet concentrates, known as the Platelet Storage Lesion (PSL)[2].

Bacterial Contamination

- One in 1500-2000 PCs is contaminated[3].
- The bacterial screening period is 24 h., putting great strain on platelet supply and shelf-life.

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

Approach

By developing anti-adhesive coatings with platelet-friendly and antiseptic properties, we may develop self-sterilizing platelet storage units that improve transfusion safety, quality, and supply.

Objectives

1. Develop a storage bag coating which extends the shelf life of platelets.
2. Modify coating components for antimicrobial performance

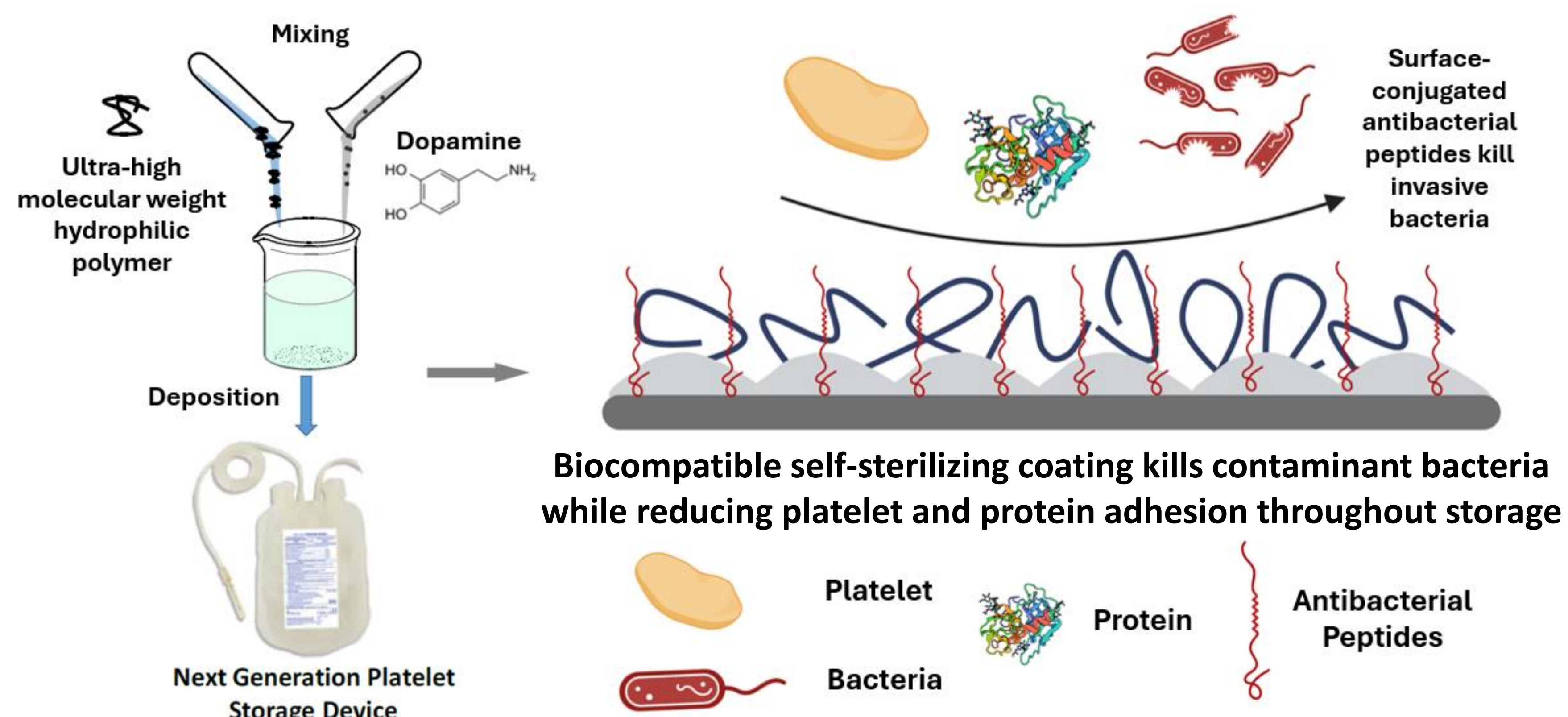## Antifouling coating for platelet storage

Figure 1. Simple one-step preparation of self-sterilizing novel platelet storage bags. A protein and platelet adhesion resistant, and platelet friendly coating is prepared inside of the platelet by using a one-step deposition of polydopamine and hydrophilic high molecular polymer (uHMWPs) poly (N, N-dimethylacrylamide) (PDMA) in aqueous solution (left side). Antibiofilm peptides (ABPs) will be covalently conjugated to coating to inhibit biofilm formation and kill bacteria. Through this novel coating technology, which maintains the platelets throughout storage, the bag is simultaneously sterilized. Coating chemistry is simple enough to adapted in a manufacturing process.
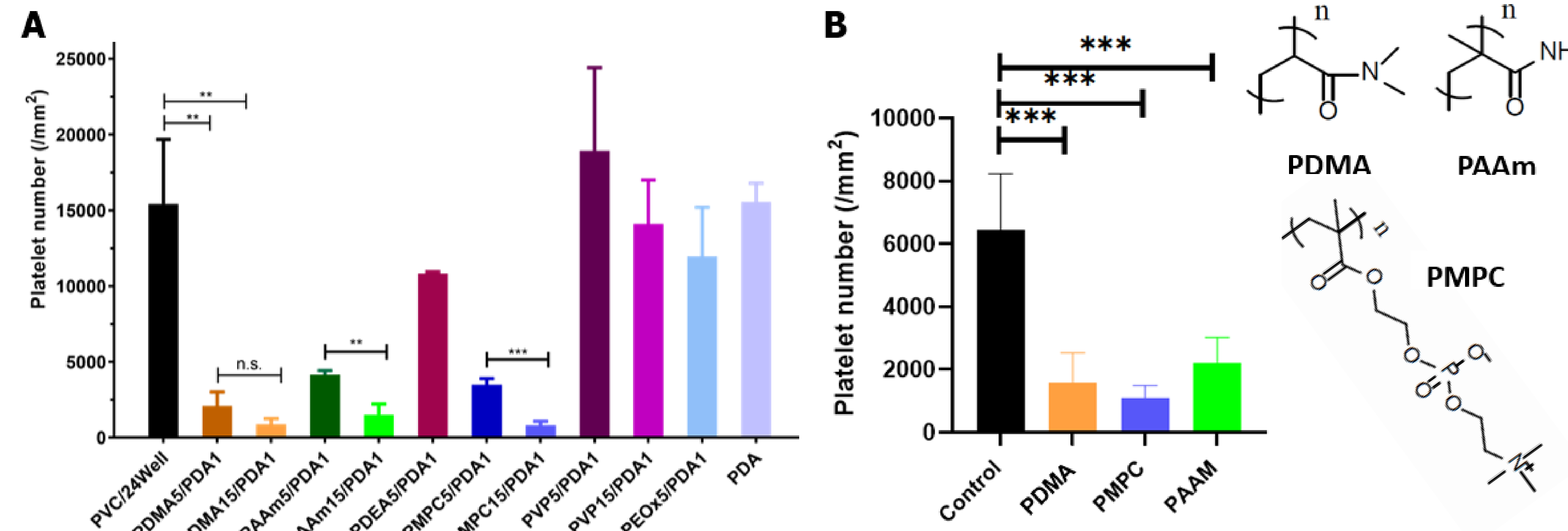
Storage Quality

Figure 4. Selection of low platelet adhesive-coating formulations. (A) Different hydrophilic polymer-based coatings were investigated in a **screening study using 24-well plates**. The wells were filled with platelet-rich plasma and incubated for 4 h, before platelet adhesion was quantified by LDH assay. Polymers that reduced platelet adhesion >90% were selected for a 7-day storage study (PDMA (polydimethyl acrylamide), PAAm (polyacrylamide), and PMPC (poly (methacryloyl phosphatidylcholine)). It was also found that 15:1 ratio of polymer:PDA was superior in reducing platelet adhesion than 5:1. **(B)** PVC mini-bags coated with 15:1 PDMA, PAAm, and PMPC, or remained uncoated as control. Platelet rich plasma was then stored within for 7 days, and platelet adhesion was quantified using the LDH assay, demonstrating that each coating had effective anti-fouling properties. **(C)** Chemical structure of the 3 top polymer compositions used in this study. N=3 (minimum) different donors. Results are mean ± SD.

Platelet Activation

Clotting Analysis

Metabolic Health

Coating A   **Coating B**   **Coating C**   **Uncoated**

Figure 5. Evaluation of platelet quality parameters throughout storage in a modified mini platelet bag. Platelet bags were coated with a library of anti-adhesive platelet-friendly coating formulations identified as described in **Fig. 4**. **(A)** Base-level platelet activation was measured by surface-display of P-selectin. **(B)** Platelet responsiveness was assessed by treatment with ADP. **(C)** Platelet apoptosis was measured using annexin V to probe for phosphatidylserine surface-display. **(D)** Maximum Clot Firmness reached from extrinsic coagulation initiation (EXTEM, recombinant tissue factor). Reactions were performed in autologous plasma, and samples were treated with START-TEM before reaction initiation. **(E-F)** The pH and O_2 contents of the plasma. Results are expressed as mean ± SD. N=4 for each test except phosphatidylserine, where N=3. No significant differences were observed across all samples tested, indicating the biocompatibility of the 3 coating compositions when compared to CBS-standard units.

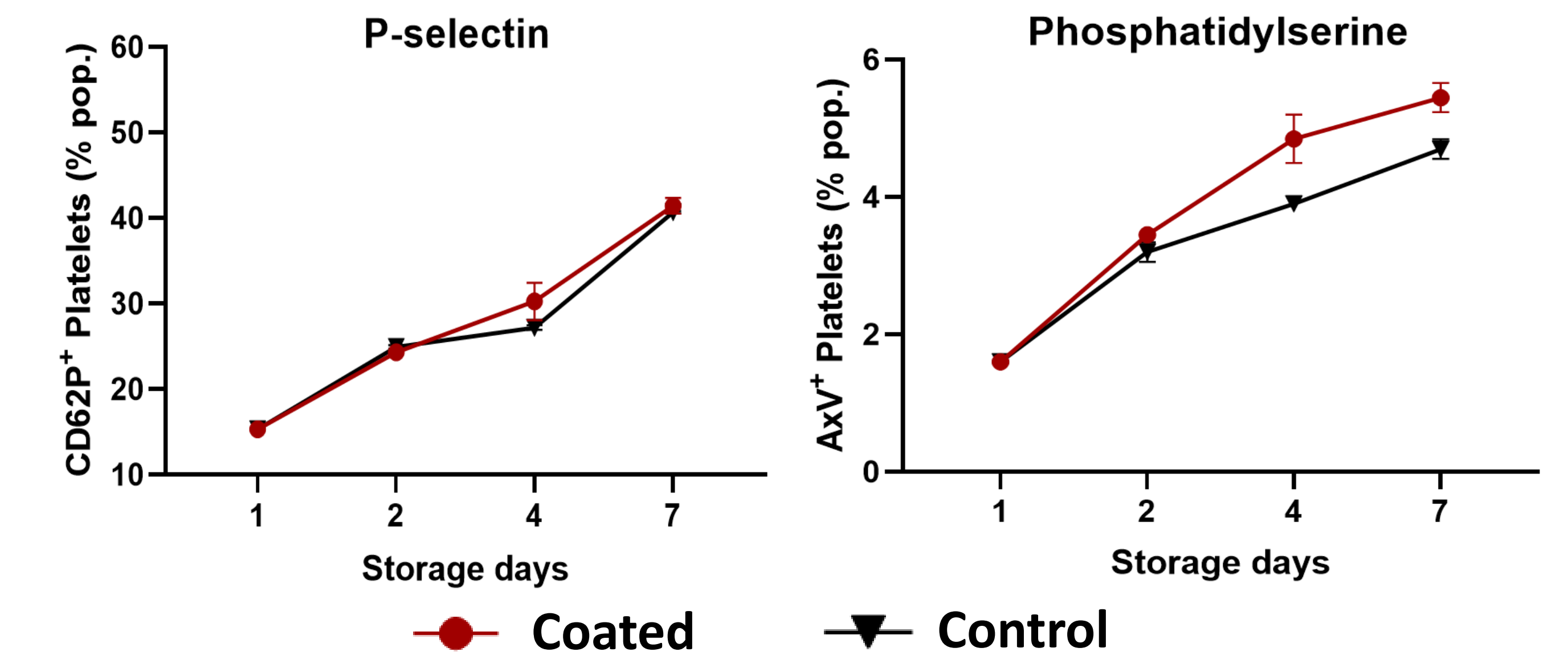## Antimicrobial Peptide Coupling

Coated   **Control**

Figure 2. Storage profile and antimicrobial properties of AMP-coupled coating. The antimicrobial peptide (AMP) E6 was conjugated to polymer composition A and deposited on PRP storage bags. PRP was inoculated with *S. epidermis* 10003 then stored in these bags for 7 days. Platelet phosphatidylserine and P-selectin were measured over 7 days. Planktonic and surface-adhered bacteria were also quantified upon inoculation and on days 2 and 4 of storage.
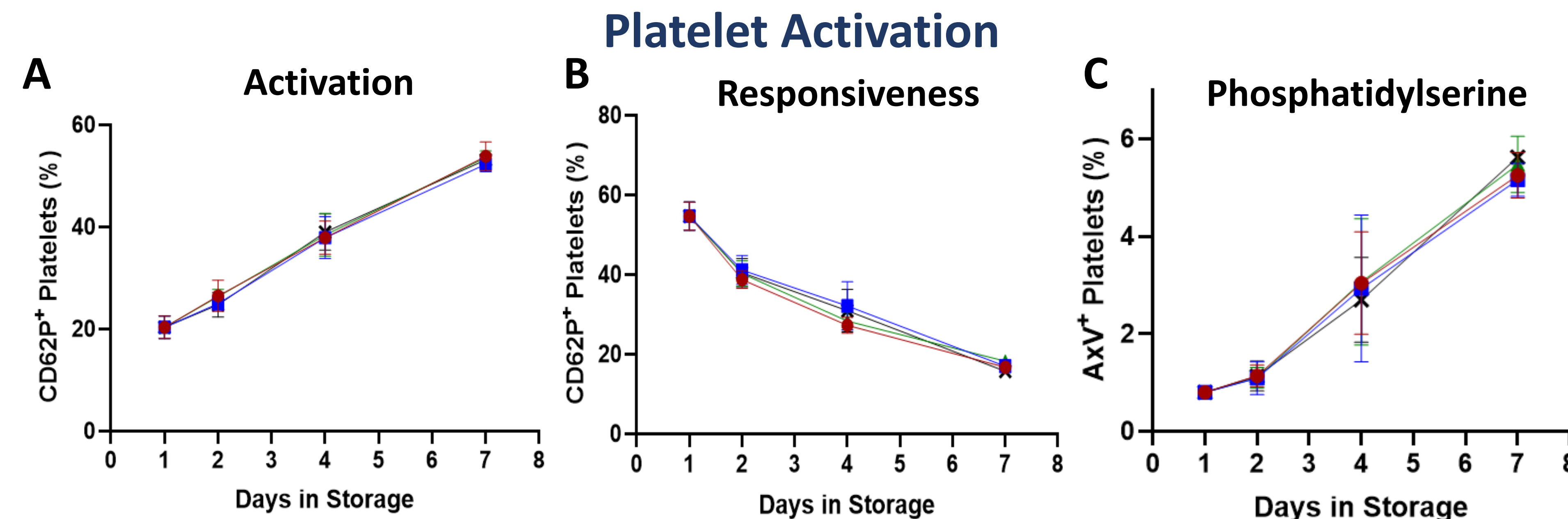
Conclusion & Future Directions

Conclusions

- Preliminary results show the coating does not significantly alter the quality of the stored platelets, indicating it is well-tolerated by the cells. AMP conjugation successfully attenuates bacterial proliferation without destroying platelets.

Future Directions

- Assess other polymers for the platelet-friendly coating.
- Optimize composition for pathogen-reduced platelets.
- Apply coating to other blood-exposed materials, such as implants, tubing, and catheters.

References:

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4. Kaiser-Guignard, J., Canellini, G., Lion, N., Abonnenc, M., Osselaer, J.-C., and Tissot, J.-D. (2014) The clinical and biological impact of new pathogen inactivation technologies on platelet concentrates. *Blood Reviews*. **28**, 235–241