Immune Biocompatibility of a Bio-Synthetic Whole Blood Analogue (WBA) for Hemorrhagic Shock Resuscitation

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Background

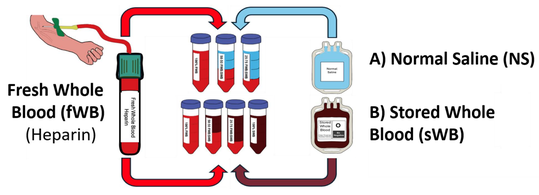
- Hemorrhagic shock remains a principal cause of preventable mortality in battlefield and civilian settings
  - Timely transfusion of fresh whole blood (fWB) or balanced components reduces morbidity and mortality
- Robust efforts are directed at resolving challenges regarding implementation of in-field transfusion
  - Including development of bio-synthetic blood analogues.
- We developed a whole blood analogue (WBA) comprised of:
  - RBC Analogue: ErythroMer (EM)
  - Platelet Mimetic: SynthoPlate (SP)
  - Rabbit Plasma: Freeze-Dried Plasma (FDP)

Goal: WBA to meet progressively restrictive non-inferiority (NI) margins (40% in Y1 to 10% in Y4) compared to stored whole blood (sWB) at 26 days (median in-theater storage duration).

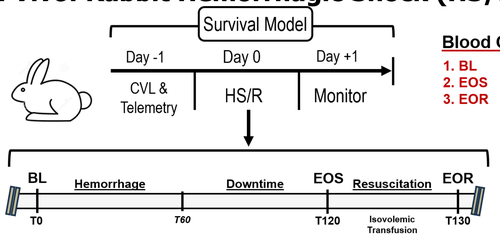
Methods

Objective: Evaluate immune response to WBA via 1) *In Vitro* and 2) *In Vivo* models of massive transfusion and hemorrhagic shock/resuscitation
- Complement Activation (innate, humoral response)
- Cytokine Generation (adaptive cellular response)

1) In Vitro: Human Massive Transfusion (HMT)



2) In Vivo: Rabbit Hemorrhagic Shock (HS/R)



Blood Collection:
1. BL
2. EOS
3. EOR
4. +1hr
5. +3hr
6. +6hr
7. +24hr

We use **Effect Size** to quantify the **degree of superiority** for:
(A) Stored whole blood (sWB): Target Product Profile (TPP)
(B) Whole blood analogue (WBA)
OVER (C) Normal Saline (NS): negative control

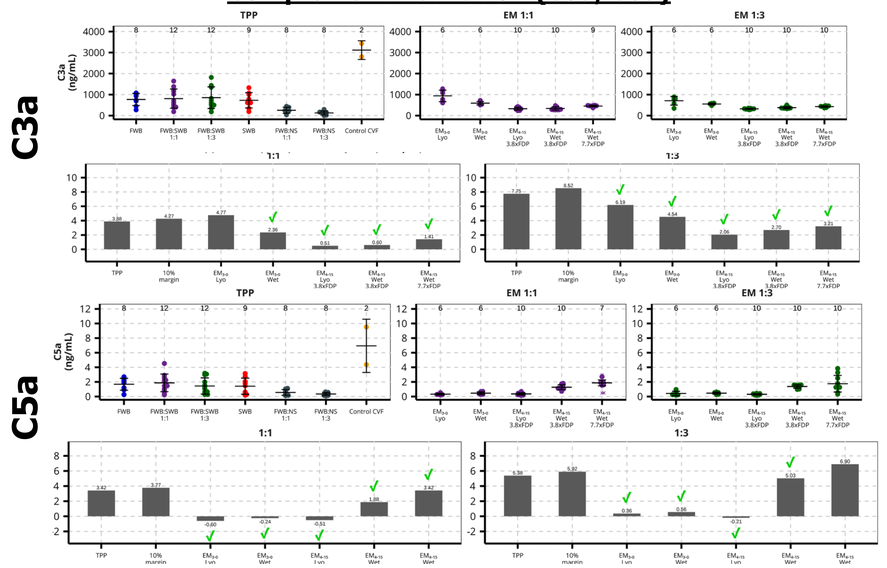
$$\textbf{Effect Size} = \frac{\text{mean(x)} - \text{mean(NS/LR)}}{SD_{NS/LR}}$$

TPP Effect Size: determined by comparing performance of sWB and (-) NS/LR groups

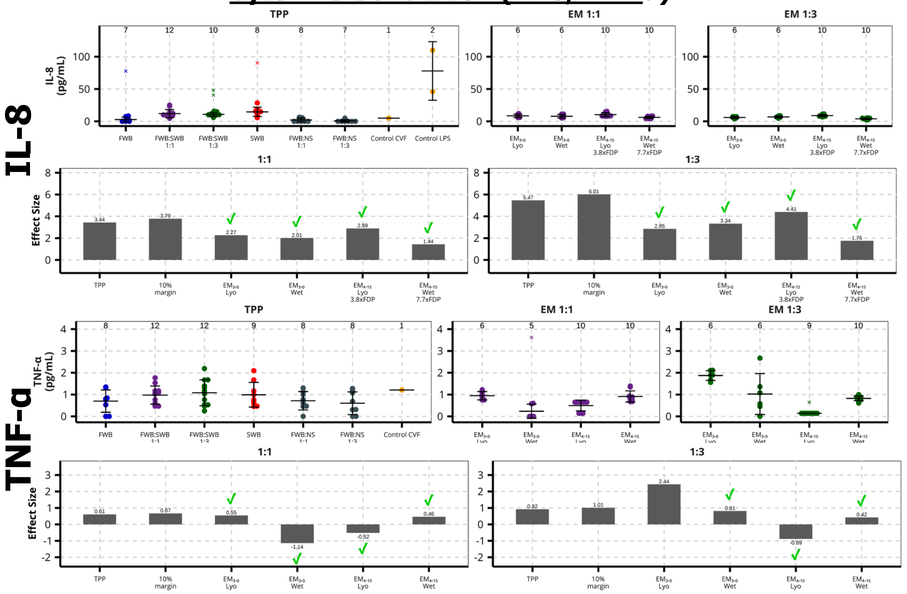
Results

In Vitro: Human Massive Transfusion (HMT)

Complement Activation (C3a, C5a)

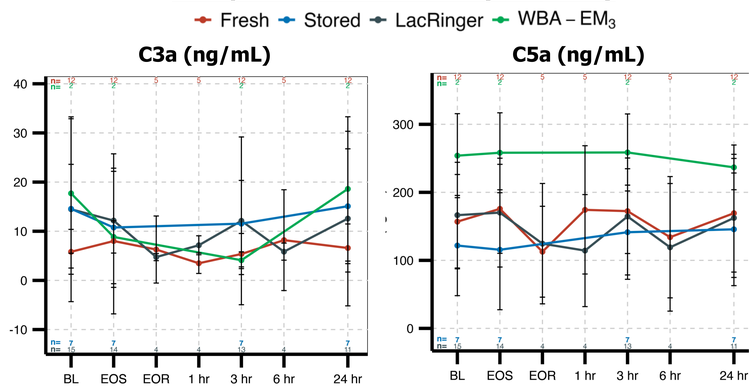


Cytokine Generation (IL-8, TNF-α)

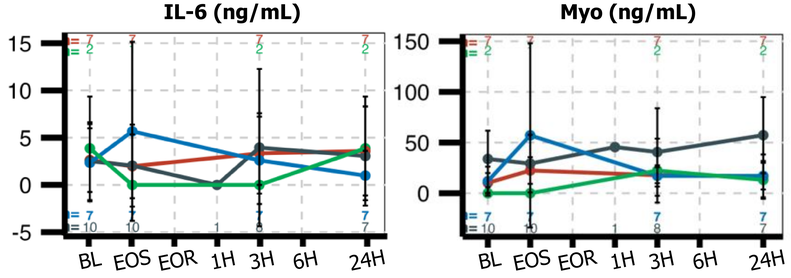


In Vivo: Rabbit HS/R

Complement Activation (C3a, C5a)



Cytokine Generation (IL-6, TF)



Conclusions & Future Directions

- sWB (26d) demonstrates moderate immune activation
- *In Vitro* (HMT), WBA relative to sWB exhibits:
  - No increase in C3a or C5a activation at either BVR level
  - No increase in IL8 or TNFα for WBA at either BVR levels
- *In Vivo* (Rabbit HS/R):
  - Complement: WBA does not exhibit significant increase relative to BL
  - Cytokine: Limited signal detection via rabbit Luminex panel
    - No significant change relative to BL
- Ongoing EM optimization:
  - Shell lipid composition (surface charge) and lyophilization (cryoprotectant & method)

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