Systemic and Microcirculatory Effects of Severe Hemorrhage and Resuscitation Using a PEGylated Carboxyhemoglobin (PEG-COHb)

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ABSTRACT

Hemorrhage and its complications are the leading cause of preventable death in combat casualties. Survival of hemorrhage is dependent on prompt hemorrhage control, sufficient - but not excessive - restoration of volume, and mitigation of ischemia/reperfusion injury by careful reintroduction of oxygen carrying capacity.

Oxygen therapeutics are often categorized into hemoglobin-based oxygen carriers (HBOCs) and perfluorocarbons (PFCs). HBOCs are derived from hemoglobin, while PFCs are made from modified carbon chains. This study focuses on the former. SanguinateTM is a PEGylated carboxyhemoglobin (PEG-COHb), which has been engineered with increased particle size to prevent extravasation and attenuate immunomodulatory responses, and affixed with a CO molecule to regulate oxygen delivery.

RESULTS

Figure 1: (A) Vascular cannulations and airway intubation. (B) Animal mounted to thermosplint platform. (C) Spinotrapezius muscle mounted to thermostable pedestal ready for intravital microscopy. (D) Impact of hemorrhagic shock on arteriole/venule pair. Baseline (left), Post-hemorrhage (HS, right).

Figure 2: Survival times were measured from the point of injury (onset of hemorrhage) for Sham and HextendTM and SanguinateTM resuscitated animals and are expressed as mean ± SD. Arteriolar diameters were measured discontinuously at the indicated time points and are expressed as mean ± SD.

Figure 3: Tissue Oxygenation. $P_O_2$ was measured in the spinotrapezius muscle for animals resuscitated with either HextendTM or SanguinateTM. Sham animals (omitted for clarity) showed a truncated profile similar to HextendTM where $P_O_2$ was undetectable following hemorrhage. Data are reported as mean ± SD.

Figure 4: Inflammatory markers. TOP: mRNA was isolated from whole blood collected prior to hemorrhagic shock and at the end of study from untreated, HextendTM or SanguinateTM treated animals. CT values, generated by qPCR, for indicated RNA were normalized to GAPDH and are expressed as fold change in relative RNA levels at end of study compared to baseline. BOTTOM: Average RNA values for IL-6, VEGF, CRP, IL-8, and CYGB ± SD, x, y pairs at end-of-study were calculated and normalized to baseline values. Interaction of dotted lines and arrows represent the baseline and change in the magnitude of RNA, respectively.