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1. REPORT DATE *(DD-MM-YYYY)*	2. REPORT TYPE	3. DATES COVERED *(From - To)*
20/08/2018	Poster	08/20-23/2018

4. TITLE AND SUBTITLE
Expression of Mobility Group Box 1 Protein in a Polytrauma Model Treated with ECLS at Ground Level and High Altitude

5a. CONTRACT NUMBER

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)
Lt Col Sams, Valerie G

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)
59th Clinical Investigations and Research Support
1100 Wilford Hall Loop, Bldg 4430
JBSA – Lackland, TX 78236-9908
210-292-7141

8. PERFORMING ORGANIZATION REPORT NUMBER
18009

9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)
59th Clinical Investigations and Research Support
1100 Wilford Hall Loop, Bldg 4430
JBSA – Lackland, TX 78236-9908
210-292-7141

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION/AVAILABILITY STATEMENT
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13. SUPPLEMENTARY NOTES
MHSRS 2018, Kissimmee, FL, 20-23 August 2018

14. ABSTRACT

15. SUBJECT TERMS

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
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Expression of high mobility group box 1 protein in a polytrauma model treated with ECLS at ground level and high altitude

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Introduction

- Acute respiratory distress syndrome (ARDS) is the most severe form of acute lung injury, characterized by acute onset of hypoxemia, bilateral radiographic pulmonary infiltrates without cardiogenic pulmonary edema, and may lead to sepsis and multi-organ failure.
- Injuries incurred in austere environments, particularly in the combat setting, require immediate evacuation with en-route critical care support.
- Extracorporeal membrane oxygenation (ECMO) may be used to support ARDS patients during transport, including during aeromedical evacuation.
- High mobility group protein box 1 (HMGB1) is an important indicator of damage-associated molecular pattern (DAMP) expression and disease progression in ARDS.
- HMGB1 has been identified as a mediator of ARDS and is expressed in blood following activation of damaged cells.
- Little is known regarding HMGB1 expression in a pulmonary contusion model of ARDS supported by ECLS at ground level..
- Altitude change effect on HMGB1 expression during air transportation is also unknown

Hypothesis

We hypothesized that HMGB1 expression in systemic blood increases following chest contusion and that HMGB1 expression is affected by changes in altitude to a greater extent in injured animals supported by ECLS versus healthy animals on ECLS undergoing the same altitude exposure.

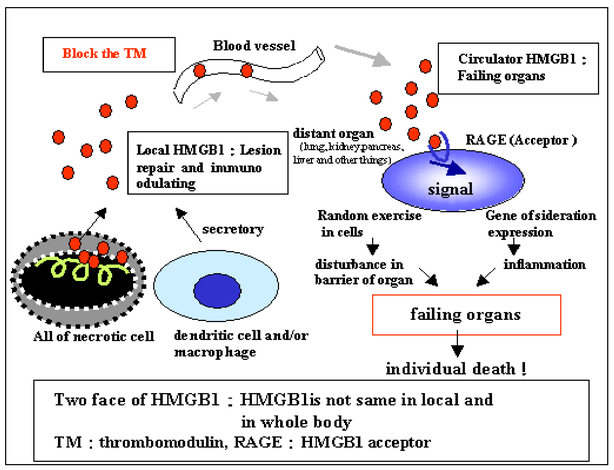

Figure 1. Mechanism of HMGB1 release
Courtesy of Prof. Ikuro Maruyama, Kagoshima University.

Methods

- Female Yorkshire pigs (54.17 ± 1.27 kg) (n=15) were anesthetized and received arterial and venous catheters, followed by tracheostomy.
- Following baseline measurements, animals were cannulated and veno-venous ECMO was initiated (CardioHelp, Maquet Gmbh, Gettinge Group, Rastatt, Germany), via an Avalon 23 Fr. catheter inserted into the right jugular vein.
- Blood flow was 1.2-3 /min and sweep gas flow ranged at 4-8L/min. Continuous heparinization was started at cannulation and titrated to 30-50% higher than baseline ACT levels.
- Animals were then transported via a standard NATO litter fitted with a next-generation medical equipment rail kit (MERK, Smeed Technologies, Cummings, GA) to an adjacent building housing hypobaric chambers.
- The altitude simulation profile consisted of the multiple levels of simulated atmospheric exposure, and is depicted in Figure 2.
- Altitude exposure occurred in healthy state on Day 1, and injured state on Day 2.
- Injury consisted of bilateral pulmonary contusions using a modified captive-bolt stunner (Model ML, Karl Schermer, Packers Engineering, Omaha, NE) and chest tube placement.
- HMGB1 ELISA (IBL international, ST51011, NC, US) was utilized to analyze the level of HMGB1 in the blood at each time-point.
- Plasma free hemoglobin (pfHb) was measured in real time by spectrophotometer method.
- Plasma total protein concentration (PTPC) was measured by Pierce™ BCA protein assay kit (Thermo scientific, Rockford, IL, US)
- Post-mortem lung tissue samples were fixed by 10% normal buffered formalin and paraffin embedded, thickness 4 *um* sliced tissues were stained by Hematoxylin & Eosin or primary antibody immunohistochemistry for HMGB1/TLR 4 (abcam, CA, US).

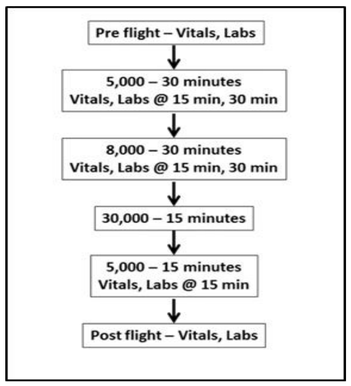

Figure 2 Altitude exposure profile

Results

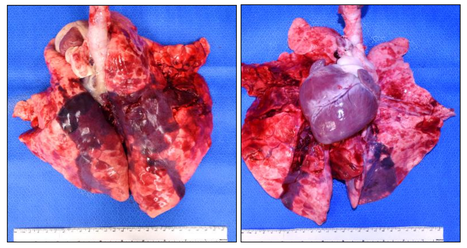

Figure 3. Post-mortem Image of Lungs after Bilateral Pulmonary Contusion Treated with ECMO

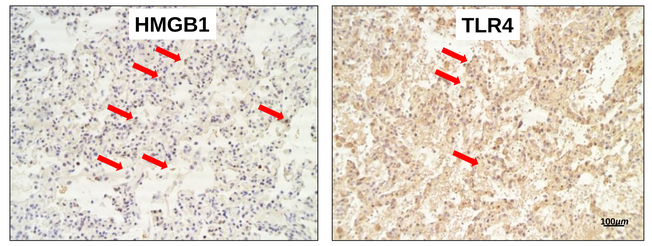

Figure 4. Post-Mortem Expression of hmgb1, TLR4 after Bilateral Pulmonary Contusion Treated with ECMO

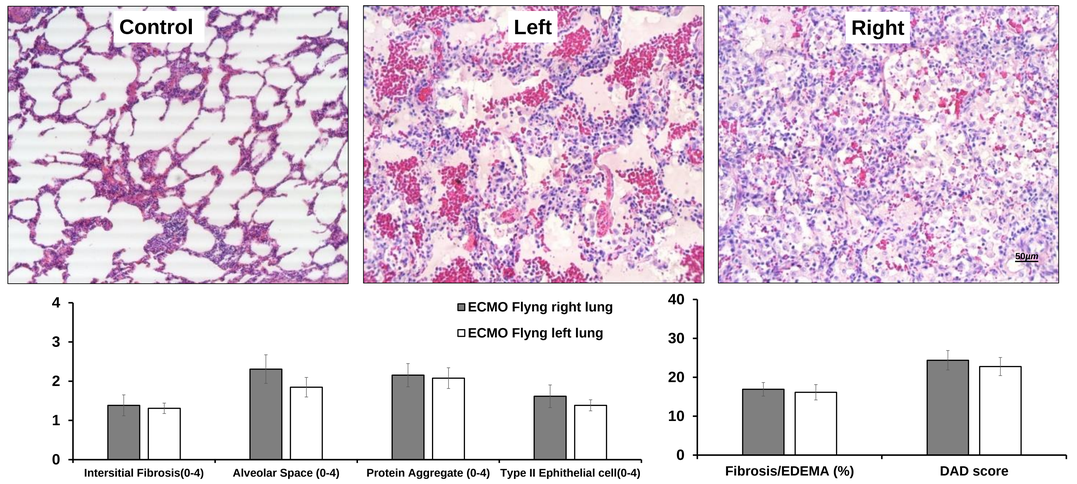

Figure 5. Histology image and diffused alveolar damage score in a bilateral pulmonary contusion

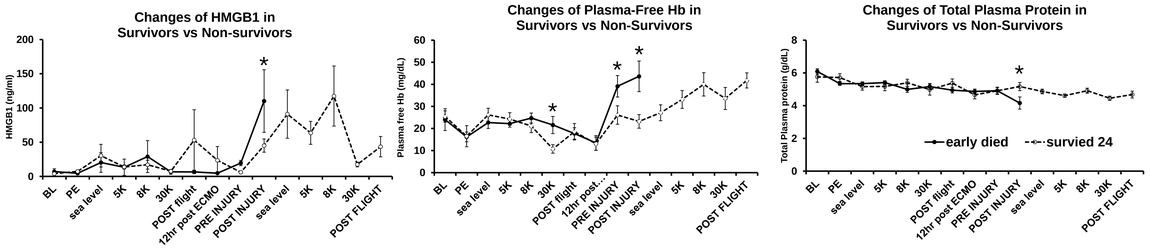

Figure 6. Dramatic change of of HMGB1 protein, pfHb and PTPC in a bilateral pulmonary contusion treated with ECMO at ground level and high altitude during en-route care.

Conclusion

High altitude does not alter HMGB1, pfHb and PTPC to expression in uninjured state on ECLS. Pulmonary contusion causes a transient increase in HMGB1 and pfHb levels. The level of HMGB1 and pfHb of early died animals were significantly higher than survived group. Bedside assessment of HMGB1 and pfHb confirms injury and may provide a useful monitoring capability during en-route care, and should be a part of precision medicine lab-on-a-chip type assays in the future.

Acknowledgements

This study was funded by the United States Air Force, and administered through the 59th MDW via The Geneva Foundation under Contract #FA8650-15-C-6692, PI: Dr. Andriy Batchinsky, MD. The experiments reported herein were conducted according to the principles set forth in the National Institute of Health Publication No. 80-23, Guide for the Care and Use of Laboratory Animals and the Animal Welfare Act of 1966, as amended.