BACKGROUND
- Acute respiratory distress syndrome (ARDS) remains a significant cause of mortality and morbidity in ICU patients. Oxidative stress and inflammation have significant role in pathogenesis of lung injury in ARDS.
- Extracellular superoxide dismutase (EC-SOD) is an important enzymatic defense against superoxide.
- A R213G polymorphism in matrix binding region of EC-SOD results in release of EC-SOD into alveolar fluid without affecting enzyme activity.
- Carriers of R213G SNP have attenuated risk of exacerbations of COPD and allergic airway inflammation in asthma.
- We have demonstrated the protective effects of R213G SNP in bleomycin-induced lung fibrosis and LPS-induced lung injury.
- While the role of R213G SNP has been investigated in COPD and Asthma in human studies, its role in infectious pneumonia and sepsis remains unknown.

HYPOTHESIS
- R213G EC-SOD variant results in redistribution of EC-SOD to the alveoli in response to MRSA pneumonia.
- R213G EC-SOD variant is protective against MRSA-induced acute lung injury and inflammation via attenuated neutrophil recruitment and functionality.

METHODS
- MRSA pneumonia: C57BL/6 (WT) and R213G mice infected IT with 1x10^8 CFUs of methicillin-resistant S. Aureus (MRSA) strain. 24hrs post-inoculation, lungs, spleen and bronchoalveolar fluid (BALF) was collected.
- Protein analysis: EC-SOD protein expression was measured in lung and BALF by Western blot.
- Evaluation of lung injury: Total cell counts and differentials, total protein and albumin, measured in BALF. Agerose inflated lungs were assessed for alveolar overdistention, atelectasis, cellularity, hemorrhage, alveolar wall thickening, perivascular edema and peribronchial edema by two blinded individuals.
- Evaluation of inflammation: IL-1β, IL-6, and TNF-α measured by ELISA in BALF and qPCR in individual.
- Neutrophil studies: CXCL-1 was measured in BALF.
- Statistical analysis: Data were analyzed by unpaired t-test or 2-way ANOVA with Tukey’s post-tests. Significance defined as p<0.05, **p<=0.01, ***p<=0.001, ****p<=0.0001.

RESULTS
- R213G mice release EC-SOD into alveolar fluid in response to MRSA pneumonia.
- R213G mice are protected from MRSA-induced neutrophil influx into alveoli.
- Increased alveolar-capillary barrier permeability and lung injury following MRSA infection is blocked in R213G mice.
- R213G mice have attenuated NETosis in response to MRSA and are protected from extrapulmonary dissemination of bacteria.
- R213G mice have attenuated expression of neutrophil chemoattractant CXCL-1 in BALF.

CONCLUSIONS
- R213G EC-SOD variant results in release of EC-SOD into alveolar lining fluid in response to MRSA pneumonia.
- In response to MRSA pneumonia mice with R213G EC-SOD variant:
  1. Are protected against acute lung injury and inflammation.
  2. Have attenuated neutrophil influx into alveoli.
  3. Have preserved MPO activity but decreased NETosis.
  4. Are protected from spread of MRSA to extrapulmonary sites.
  5. Have decreased expression of neutrophil chemoattractant CXCL-1 in BALF.

FUTURE DIRECTIONS
- Further studies will interrogate the mechanisms driving this protection and therapeutic implications, specifically:
  1. Characterization of alveolar redox environment in WT and R213G mice following MRSA infection.
  2. Determining if administration of catalase and extracellular superoxide dismutase attenuates neutrophil-mediated lung injury in MRSA pneumonia.
  3. Interrogation of how alveolar redox environment affects interaction between ATII cells and recruited neutrophils.
  4. Testing whether R213G mice are also protected from sepsis and end-organ dysfunction as a result of decreased extrapulmonary dissemination of MRSA.

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