THERMAL INJURY PLUS PSEUDOMONAS AERUGINOSA INFECTION INDUCES ACTIVATION OF THE NLRP3 INFLAMMASOME AND ALTERATIONS IN IMMUNE CELLS

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Introduction

Severe burn is a catastrophic event that forces patients to endure a life long battle recovering both physically and psychologically. Thermal injury produces a wide array of stress-associated inflammatory and metabolic changes aimed at restoring systemic homeostasis. Pseudomonas aeruginosa is a gram-negative bacterium that causes serious infections in immune-compromised patients and is particularly common during critical illness and thermal injury. The proliferation of P. aeruginosa infection in the wounds of burn patients in combination with its antibiotic resistant properties contributes towards skin graft failure, increases risk of sepsis, multi-organ dysfunction and mortality. The mechanism for this sequence of events is not well understood; however, recent studies of stress-induced diabetes have brought into question the role of the NLRP3 inflammasome. Using white adipose tissue from burn patients, preliminary data from our lab suggests that there is increased NLRP3 activity in patients after severe burn injury. The specific mechanism of pathogen-induced activation is still under debate. The purpose of this study was to use a two-hit mouse model of infectious thermal injury to determine the role of NLRP3 inflammasome during the development of burn-induced inflammatory response and subsequent infection-induced complications.

Hypothesis

Severe burn with pseudomonas aeruginosa infection induces NLRP3 inflammasome activation in tissue. Collectively, this activation will promote the inflammatory cascade that directly contributes to pathology and mortality

Methods

CS7BL/6 mice (6-8 weeks old) were divided into three groups: sham (n=5), burn (n=6) and burn+PA (n=6) infection. Burn and burn+PA were exposed to a 40% scald injury and the later group received a topical infection of Pseudomonas aeruginosa (PA) 72-hours later. All mice were sacrificed 24-hours after insult.

Flow Cytometry was conducted to characterize the proportions of monocytes, macrophage, neutrophils, B-, T-, NK-cells in the bone marrow, spleen, liver and skin.

A multi-analyte Milliplex (Millipore, MA) platform was used to measure chemokines, pro- and anti-inflammatory cytokines.

Conclusions

Collectively, this suggests that infection causes activation of innate immune cells towards the site of infection suggesting systemic infiltration and migration. This project is the first study to describe the relationship between infectious complications and NLRP3 inflammasome activation after severe burn. These studies will provide insight about the upstream and downstream factors that are inducing inflammasome activation and largely, the degree of diminished metabolic response.