Lung Microparticles are an Antimicrobial in Injured Mice

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BACKGROUND

Infection is the leading cause of burn-related deaths[1]. Specifically, burn-injured patients are commonly infected with *Pseudomonas aeruginosa*[2]. In patients with both burn-injury and infection, antibiotic therapy is often ineffective[3]. The mortality rate for patients can be as high as 50%[4]. An alternative to antibiotic therapy is immune modulatory intervention. The inflammatory response to infection, however, is not completely understood. The role of microparticles (MPs) in the immune system is a possible intervention. MPs are small vesicles of disparate lipid and protein composition[5]. During infection, the quantity and type of MPs changes as compared to a healthy state[6].

HYPOTHESIS

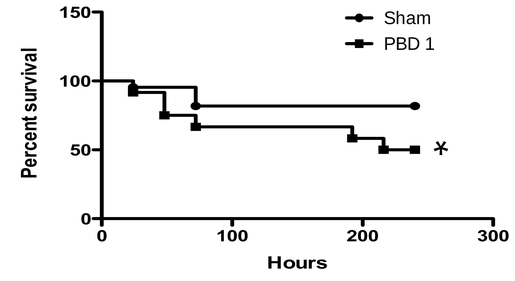
Injection of Bronchoalveolar microparticles (BAL MPs) will increase the survival and bacterial clearance of burn injured mice as compared to mice without the injection of BAL MPs.

METHODS

Mice were subjected to a dorsal 28% total body surface area scald injury and monitored for survival. On Post Burn Day One, MPs were isolated from BAL fluid and analyzed with Nanoparticle Tracking Analysis (NTA). Next, a cohort of mice was injected intranasally with MPs from healthy mice. In each of these cohorts, mice were subjected again to burn injury and subsequently, survival and lung bacterial loads were determined. Afterward, a cohort of mice was injected with MPs and subjected to cecal ligation an puncture. Bacterial load for both cohorts was then determined. Additionally, direct bacterial killing effects of MPs were measured by an *in vitro* assay.

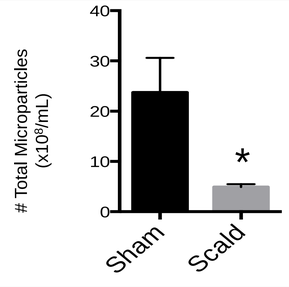
RESULTS

Figure 1. Burn-injured Mice Have Decreased Survival



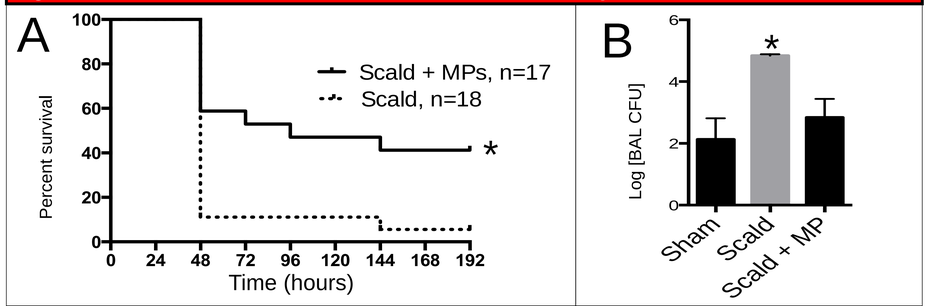
Survival of Post Burn Day One (PBD1) mice compared to survival of sham-injured.

Figure 2. Decreased BAL MPs in Burn-injured Mice



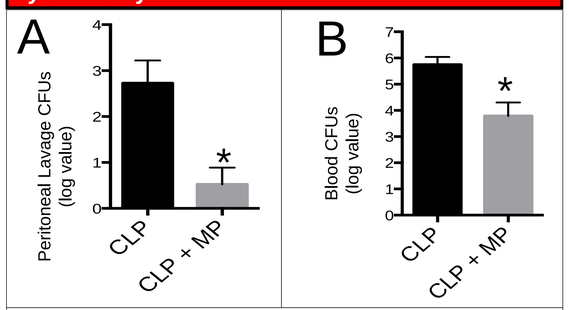
One day after scald-injury, the number of BAL MPs decrease as compared to sham-injured.

Figure 2. Pretreament with BAL MPs Decreases Mortality and Bacterial Load



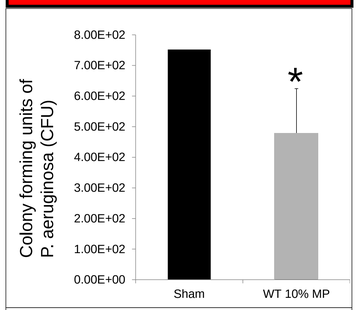
A) Prior treatment with $1x10^9$ BAL MPs, rescued mice from death.
B) Prior treatment with $1x10^9$ BAL MPs, decreased lung bacterial burden.

Figure 3. BAL MPs Decrease Bacterial Load Systemically



A,B) Using another model of infection Cecal ligation and puncture (CLP), BAL MPs reduced both local and systemic bacterial burden.

Figure 4. BAL MPs Directly Kill Bacteria



35% decrease in bacterial load through WT 10% MPs direct in-vitro killing.

CONCLUSIONS

- Burn injured mice, as compared to sham injured mice, have a decreased rate of survival and are observed to also have a reduced number of BAL MPs.
- Intranasal injection of BAL MPs can improve the survival and decrease the bacterial load in the lungs of burn-injured mice.
- BAL MPs decrease the bacterial load and increase survival systemically as well as locally.
- BAL MPs directly kill bacteria in an *in vitro* killing assay.

FUTURE DIRECTIONS

The cellular origin of many BAL MPs is unknown. NTA can currently only account for the cellular origin of 4.220173% of BAL MPs:

- 4.22 % epithelial cells
- 9.00E+05 % platelets
- 3.80E+05 % leukocytes
- 4.50E+05 % red blood cells

Future study will use western blots and NTA to target cell specific markers on BAL MPs to elucidate a larger percent of the cellular origin. First, the cell marker CD 62E will be used to target endothelial cells.

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