Development of a Rat Model for Post-Traumatic Heterotopic Ossification

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Disclosures:

Introduction: Heterotopic ossification (HO) is the ectopic formation of mature lamellar bone in nonosseous tissue. HO occurs following combat injuries, including traumatic amputations, in nearly two-thirds of patients with up to 41% requiring surgical intervention. Though means of primary prophylaxis currently exist, they are impractical or unproven in combat casualty care. As such, effort is directed towards developing novel means of primary prophylaxis and/or treatment. However, there is no animal model for the development of combat-related HO that recapitulates the systemic and local inflammatory response observed in combat trauma, which we believe are critical to HO formation. Using techniques that could be reproduced in any translational laboratory, we sought to create a preclinical animal model for HO that reproduces a severe limb injury in the setting of the prolonged systemic inflammatory response observed in combat casualties.

Methods: Male Sprague Dawley rats underwent the following:(1) systemic inflammation induced by blast overpressure (BOP), (2) severe extremity trauma by creating a thigh crush injury with femur fracture, and (3) limb salvage or amputation. Specifically, BOP were delivered at 120 +/- 7 kPa, open femur fractures were created from a 500g drop weight apparatus, and crush injuries were from compression clamps at 20psi for 1 minute over the fracture site. After the injury, the rats were transferred to the operating room to undergo either transfemoral amputation or fracture fixation using a 0.045 kirschner wire. Radiographs (XR) were obtained weekly for four weeks then monthly to six months. Rats were euthanized once mature HO was seen or at six-months post-operatively in the persistent absence of HO.

Results: We began with 74 rats; 10 were euthanized early for various complications. The 64 surviving rats at eight weeks comprised three groups: blast control (n=9), surgical control (n=18) and study cohort (n=37). HO developed in zero blast controls, eight (44%) surgical controls and 31 (84%) of the study group. Serum analysis demonstrated a pronounced inflammatory state with a prolonged, increased concentrations of tumor necrosis factor a (TNF-a), interleukin (IL)-1b, and IL-6 and lower concentrations of RANTES.

Discussion: We successfully developed a model for post-traumatic heterotopic ossification in rats that reproduces the systemic inflammation seen in human combat casualties. Blast overpressure in the presence of extremity trauma produced radiographically-evident heterotopic ossification in the majority of rats, with acceptable mortality. We plan to use this model to determine the effect(s), if any, of bioburden using clinically-relevant bacterial isolates, in an effort to further evaluate cellular and molecular mechanistic pathways, and to test novel means of systemic and local prophylaxis currently in development.

Significance: We developed a model of post-traumatic HO that recapitulates the systemic inflammation seen in combat casualties. This model will be used for further evaluation of the molecular pathways that lead to the development of heterotopic ossification as well as a means for testing novel prophylaxis measures.

Acknowledgments:
References:

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