Immunostimulatory Peptide Biomaterials as Scaffolds for Defect Repair

Yalini Vigneswaran, Huifang Han, Jacqueline Handley, Tao Sun, Joel H. Collier
The University of Chicago, Department of Surgery, Chicago, IL, USA 60637.

Statement of Purpose: Peptide nanofibers have received significant interest as scaffolds for tissue repair, wound healing, and regeneration. These self-assembling peptide systems have been exploited for their modularity, ease of synthesis, ability to incorporate multiple signaling components, and structural similarity to native extracellular matrix. Recently, we have observed that these materials can be formulated to raise strong immune responses by incorporating specific T cell and B cell epitopes, yet they elicit no detectable inflammation at sites of delivery. We sought to determine whether this nanofiber immunogenicity precluded the use of the materials in wound healing contexts, hypothesizing that some antibody responses may be consistent with good *in vivo* performance of the materials in tissue defects.

Methods: In mice, an excisional dermal wound healing model was used to study the effects of active immune responses at the site of healing. Our work used the self-assembling peptide Q11 (QQKFQFQFEQQ) and the OT-II antigen from the protein ovalbumin (containing a T cell and B cell epitope). Mice were either immunized with dilute solutions of the epitope-bearing nanofibers or not immunized. After vigorous anti-peptide immune responses were established, as measured by serum antibody titers, an 8mm full-thickness dermal wound was created on the dorsum of each mouse. Immunized mice received the epitope-bearing nanofiber scaffold intradermally at the wound edge (n=6), whereas non-immunized mice received bare scaffolds (n=6). Mice were sacrificed 7 and 16 days later. Wounds were harvested, fixed, paraffin embedded, and examined histologically.

Results: Immunized mice raised and maintained high antibody titers against the epitope-bearing peptide scaffold, but not the bare scaffold, throughout the experiment. Interestingly, wound healing did not differ significantly between immunogenic or bare scaffolds at day 7 or at day 16. On day 16 immunized mice had more granulation tissue but this was not statistically significant. Immunohistochemistry showed CD3+ leukocytes in the defect for both groups, but they were more organized at the interface between the dermis and granulation tissue for the immunogenic peptide group (Fig 1).

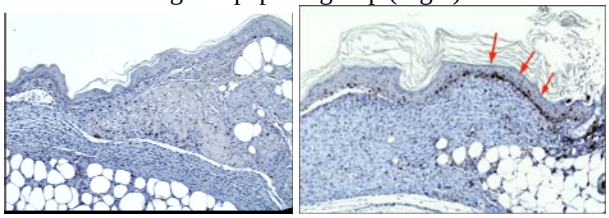

Figure 1. Immunohistochemistry. CD3+ cells were distributed throughout the dermis and granulation tissue in wounds filled with bare scaffolds, but they were localized to the dermal/granulation tissue interface for wounds filled with immunogenic scaffolds (red arrows).

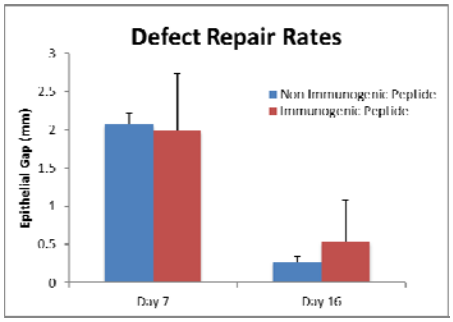

Figure 2. Epithelial gap measured at day 7 and day 16 was not significantly different for mice immunized against the scaffold (immunogenic peptide, n=3 per time point) and mice receiving non-immunogenic peptide scaffolds (non-immunogenic peptide, n=3 per time point), p >0.05 by Student's t-test for testing the two groups.

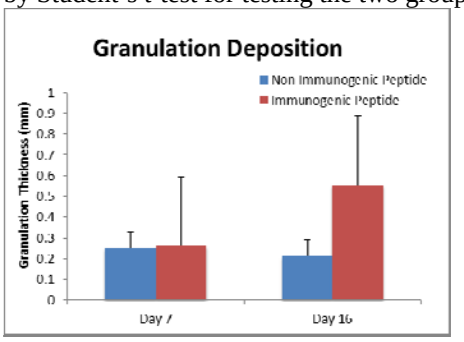

Figure 3. Granulation tissue thickness measured at day 7 and day 16 was not significantly different for mice immunized against the scaffold (immunogenic peptide, n=3 per time point) and mice receiving non-immunogenic peptide scaffolds (non-immunogenic peptide, n=3 per time point). p >0.05 by Student's t-test for testing the two groups.

Conclusions: We conclude that the phenotype of the immune response raised by peptide nanofibers may be compatible with using such materials within healing defects. This finding is consistent with previous studies from our group showing that even vigorous antibody responses raised by the materials are not associated with any measurable inflammation at their sites of delivery, but it runs counter to the generally held view to avoid anti-biomaterial immune responses in tissue engineering applications. We are conducting higher-powered studies to invest this true absence of hindrance to wound healing in the presence of a vigorous antibody response and to better characterize the organized immune response seen at the wound bed.

References:

1. Chen, J. Biomaterials 2013, 34 (34), 8776-85.