Innovative 3D Collagen Microsphere Scaffold (MSS) Promotes Robust Cellular Invasion

OA Asanbe, RC Hooper, T Elshazly, HL Osoria, A Jacoby, R Walters, JL Bernstein, BM Vishnevsky, J Morgan, J Joyce, RH Weinreb, AD Stroock, JA Spector

Introduction: Contemporary dermal substitutes are avascular and prone to high failure rates when used in complex settings, such as irradiated wounds or those with exposed hardware or bone. In order to overcome these shortcomings, we designed a scaffold to guide and hasten cellular invasion and neovascularization, via the use of regularly spaced interfaces of differing collagen densities.

Methods: Utilizing Kepler's conjecture of sphere packing, which states that the arrangement of spheres in a 3D space has a density of 74%, we fabricated 7 mm microsphere scaffolds (MSS) with a regular arrangement of density gradients. Type I collagen microspheres (1%, 0.6% or 0.4%), ranging 50 to 150 µm in diameter, were manufactured via an oil emulsion technique. MSS were fabricated by encasing microspheres of varying collagen density into collagen bulk of varying density (0.3%, 0.2% or 0.6%), so that 74% of the scaffold's volume was comprised of microspheres and 26% of bulk collagen. MSS underwent thermal gelation at 37°C for 1 hour. Non-microsphere-containing 1% or 0.3% collagen scaffolds were fabricated as controls. Scaffolds were implanted subcutaneously in the dorsa of 8 week old wild-type mice and harvested for histological analysis after 7 or 14 days.

Results: After 7 and 14 days, fluorescent microscopy revealed MSS with robust cellular invasion spanning the scaffold depth. The optimal collagen density gradient was observed in MSS with 0.6% microspheres embedded in 0.3% bulk. Comparatively, cells sporadically invaded 0.3% collagen scaffolds and failed to invade 1% collagen scaffolds, remaining confined to the periphery. Immunohistochemical analysis identified CD31 expression in all MSS after 7 and 14 days of implantation, indicative of invading endothelial precursors.

Conclusions: By enhancing mechanical and spatial cues of MSS through the regular arrangement of collagen density gradients, we significantly accelerated cellular invasion and neovascularization. In addition to optimizing structural cues sensed by cells, collagen microspheres may be impregnated with growth factors and medications to further hasten wound healing. Our innovative MSS hold tremendous promise for creating the next generation dermal replacement product.

Disclosure/Financial Support: None of the authors has a financial interest in any of the products mentioned in this manuscript.