Bio-hybrid Composite Scaffolds for Bone Tissue Regeneration

Sang Jin Lee, Jang-Won Lee, Grace Lim, Anthony Atala, and James Yoo
Wake Forest Institute for Regenerative Medicine, Wake Forest University Health Sciences, Winston-Salem, NC

Statement of Purpose: Numerous materials have been proposed for bone tissue regeneration. However, none has shown to be entirely satisfactory. The ideal scaffold for bone tissue engineering should be biocompatible and possess mechanical stability, a controlled degradation rate, hydrophilic surface chemistry and an appropriate porosity for cell accommodation.[1,2] Recently, a cell-based approach has been proposed as a new concept in bone tissue regeneration. While many biomaterials serve as a scaffold that augments the body's ability to heal itself, a tissue engineering approach uses cells added to a scaffold to achieve formation of bone tissue.[3] In this study we aimed to develop a composite scaffold for bone regeneration that would meet these criteria by hybridizing BSM as a natural bioactive material[4,5] with synthetic PLGA polymers.

Methods: Poly(D,L-lactide-*co*-glycolide) (PLGA), biodegradable synthetic polymer and a naturally derived collagen matrix derived from porcine bladder submucosa (BSM) were fabricated into a composite scaffolding system using a solvent casting/particulate leaching process. The structural properties were examined by scanning electron microscopy (SEM), mercury intrusion porosimeter, hydrophilicity assay and biomechanical testing. The biological properties of the scaffolds were evaluated by seeding with both human embryonic stem cells and bovine osteocytes. Cell adhesion, survival, proliferation and toxicity were assessed.

Results / Discussion: The BSM-PLGA composite bone scaffolds possessed uniform porous structures with a consistent interconnectivity throughout the entire scaffold. The average pore size of the composite scaffolds was 121.84 ± 23.44 μm and had a porosity of 94.79 ± 10.76% (Figure 1). The surface hydrophilicity of the BSM-PLGA composite scaffolds was significantly enhanced, when compared to the hydrophilicity of each material separately, resulting in uniform cell seeding and distribution. Cells seeded on the composite bone scaffolds readily attached, survived and proliferated (Figure 2), as confirmed by histological examination, cell viability and MTT assays. There was no evidence of toxic effects of the scaffolds.

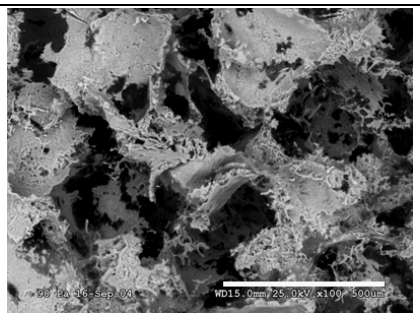

Figure 1. Cross-sectional observation of the BSM-PLGA composite scaffold. Scale bar indicates 500 μm. (SEM, Original magnification: ×100).

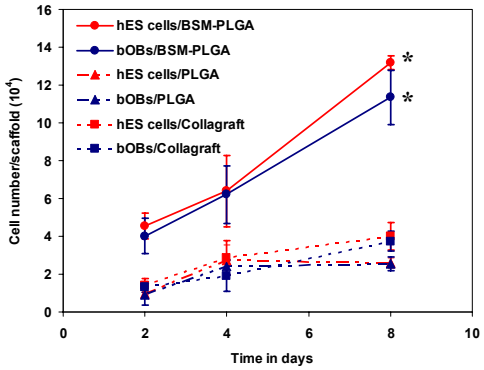

Figure 2. Cell Proliferation of the BSM-PLGA composite scaffold, PLGA scaffold and Collagraft® after 8 days of culture as determined by MTT assay. The cells seeded on BSM-PLGA composite scaffolds show a higher proliferation rate when compared to the other experimental scaffolds (*$P < 0.05$). Cells proliferated gradually in the BSM-PLGA composite scaffold, PLGA scaffold and Collagraft® for 8 days of culture. The initial number of seeded cells was 4×10^4 cells/scaffold.

Conclusions: We successfully fabricated a bio-hybrid composite scaffold composed of BSM and PLGA that possesses the necessary characteristics for bone tissue regeneration. The BSM-PLGA composite scaffolds are non-toxic, easily fabricated and provide structural features, consisting of abundant pores that are homogenously distributed throughout the inner structure allowing cell adhesion and proliferation. The use of the composite scaffolding system with cells may enhance the formation of bone tissue for therapeutic regeneration.

References:

1. Hutmacher DW, Sittinger M, Risbud MV. Scaffold-based tissue engineering: rationale for computer-aided design and solid free-form fabrication systems. Trends Biotechnol 2004;22:354-62.
2. Sachlos E, Czernuszka JT. Making tissue engineering scaffolds work. Review: the application of solid freeform fabrication technology to the production of tissue engineering scaffolds. Eur Cell Mater 2003;5:29-39; discussion 39-40.
3. Langer R, Vacanti JP. Tissue engineering. Science 1993;260:920-6.
4. Chen F, Yoo JJ, Atala A. Acellular collagen matrix as a possible "off the shelf" biomaterial for urethral repair. Urology 1999;54:407-10.
5. De Filippo RE, Yoo JJ, Atala A. Urethral replacement using cell seeded tubularized collagen matrices. J Urol 2002;168:1789-92;discussion 1792-3.