Biomineralized Highly Aligned Nanofiber Array Based Building Unit for Bone Regeneration

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Statement of Purpose: Bone is composed of both organic and inorganic components, with the inorganic hydroxyapatite (HAp) forming between and elongating on the organic collagen fibers. This structure allows for enhanced mechanical properties that give bone its necessary high strength and fracture toughness, which maintain its load-bearing function, as well as elasticity, which allows it to absorb shock.[1] Current treatment methods for critical sized defects in bone do not suffice. The field of bone tissue engineering focuses on mimicking the structural and mechanical properties of bone by creating a scaffold that closely resembles bone's microstructure. In the present study, highly aligned biomineralized polymer nanofibrous scaffolds were created as the fundamental building unit for bone regeneration. By combining the use of our novel electrospinning apparatus, which creates highly aligned electrospun fibers, along with SBF immersion, which provides biomimetic ceramic coating on the fibers, we were able to create a scaffold which mimics the microstructure of the natural extracellular matrix (ECM) of bone, as well as possesses enhanced mechanical properties, which are necessary for this application.

Methods: Highly aligned polycaprolactone (PCL) nanofibrous scaffolds were created by an electrospinning device designed in our lab (Figure 1A). The surface of these scaffolds were then functionalized and mineralized using a biomimetic technique in order to create an even coating of hydroxyapatite nanoparticles on the surface. The parameters of this mineralization process (surface hydrolysis time and 5X simulated body fluid (SBF) immersion time) were altered in order to produce an optimal coating which demonstrated similar chemical and structural properties to that of natural bone. The mechanical properties of these optimized scaffolds were then compared to the mechanical properties of unmineralized scaffolds in order to test their ability to serve as a bone tissue engineering scaffold. Cell studies were also conducted in order to determine if human embryonic palatal mesenchymal (HEPM) stem cells attached and aligned as well on the mineralized scaffolds as compared to the unmineralized controls.

Results: It was determined that scaffolds hydrolyzed for 30 minutes and soaked in 5X SBF for 24 hours created an optimal coating on the scaffolds (Figure 2). Initial mechanical testing data indicates significantly higher ultimate tensile strength, elastic modulus, and toughness of mineralized scaffolds (PCL-HAp) as compared to the umineralized controls (PCL). Further, the ultimate strain of the samples was comparable for both groups, indicating that the mineralized samples exhibited both strength and elasticity, which is very important for bone scaffolds (Figure 3). Preliminary cell studies indicate similar cell attachment and alignment for both mineralized and unmineralized samples (Figure 4).

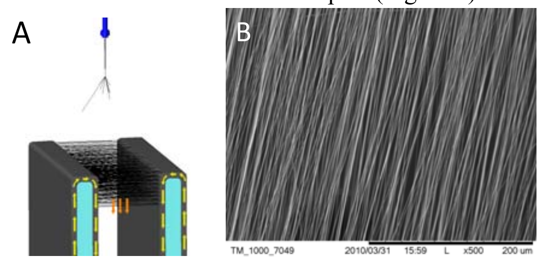

Fig. 1: Novel electrospinning device (A) used to create highly aligned fibers (B)

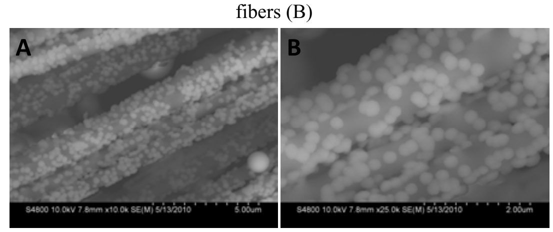

Fig. 2: Biomimetic HAp coating on aligned nanofibers: 10,000X (A) and 25,000X (B)

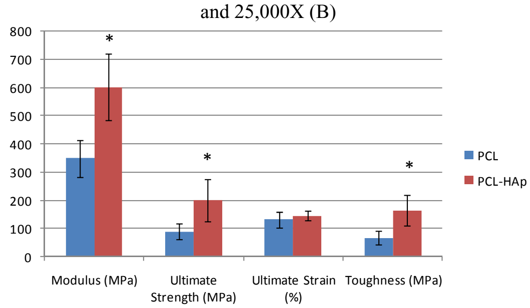

Fig. 3: Mechanical properties of PCL and PCL-HAp scaffolds

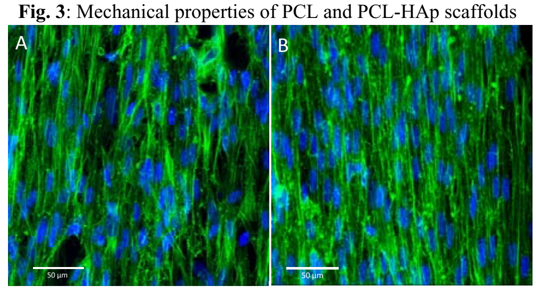

Fig. 4: Cell alignment on PCL-HAp (A) and PCL (B) scaffolds

Conclusions: This study has been successful in determining the optimal parameters that created a bioceramic coating on polymer nanofibers, both in composition and structure. Further, this coating created a scaffold that has significantly enhanced mechanical properties, which is very important for bone tissue engineering. HEPM cells exhibited no cytotoxicity and demonstrated excellent cell attachment, alignment, and proliferation on these scaffolds.

References: 1. Barrere, F. et al. Int J Nanomedicine. 2006; 1:317-32

Acknowledgement: This work is supported by NIH P20RR021949