Fabrication of Electrospun 3D Scaffolds That Mimic Cortical Bone

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Statement of Purpose: Bone grafts are used in the orthopaedic reconstructive procedures to provide mechanical support and promote bone regeneration. Bone is organized into to two different organizational structures: trabecular and cortical bone. Cortical bone is composed of tightly packed units, called osteons, oriented parallel along to the axis of the bone. The high organization and compact nature provide excellent micro-crack propagation prevention and high tensile and compressive mechanical properties.

In this study we utilized electrospinning techniques to create scaffolds that will mimic structural organization of osteons. Multiple osteon scaffolds are then sintered together to create 3D scaffolds that mimic the organization of cortical bone.

Methods:

Electrospinning: Poly (ethylene oxide) (PEO) was electrospun onto rotating mandrel and cut into 4 mm strips and rolled into fibers. PEO fibers were then placed into the rotating stage build, shown in Figure 1, and placed in front of negative target. Poly (L-lactide) (PLLA) was dissolved in 75% dichloromethane (DMC) and 25% DMF to 7 wt% solution, and solution of gelatin was added to create 10% w/w of gelatin with respect to PLLA. Volume of 1 ml of 10% gel/PLLA mixture was electrospun onto rotating PEO fibers to create osteon-like scaffolds. Gelatin was cross-linked in vapor of 2.5 % v/v of glutaraldehyde for 2 hours (CL).

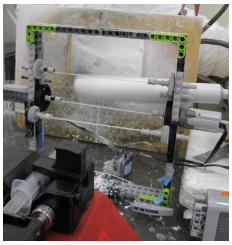

Figure 1. Fiber rotating stage

Heat sintering: Osteon-like scaffolds were cut into 1cm pieces, which were then stacked and wrapped with 1cm wide strip of electrospun 10%gel/PLLA sheets into cylinders with 5mm diameter. The scaffolds were placed into mold and heat sintered at 54°C for 45 min.

Mineralization: Scaffolds were mineralized by incubation in 10X simulated body fluid (SBF). Scaffolds were incubated in vacuum, and SBF was changed every 2h. Mineralization of the individual scaffolds and sheets prior to sintering was evaluated as treatment (premin1h) for increasing mineralization and mechanical properties. Two mineralization times, 6h (M6h) and 24h (M24h) were evaluated. Mineral distribution was imaged using alizarin red stain.

Compression testing: Scaffolds were tested using Instron 5869 in pH7.4 PBS at 37°C. Scaffolds were tested at 10% strain rate until failure.

Results: Scaffolds were successfully fabricated into 5mm diameter and 10mm height cylinders. Cross-sections stains reveal the structures of the scaffolds that consisted of packed concentric layers of electrospun polymer nanofibres with channels running along the length, mimicking the structure of cortical bone (Figure2). Alizarin red stains revealed presence of the mineral lining the channels and surface. Scaffolds that were cross-linked, then premineralzied for 1h, and heat sintered and mineralized for 6h (CL_premin1h_M6) had the most mineral.

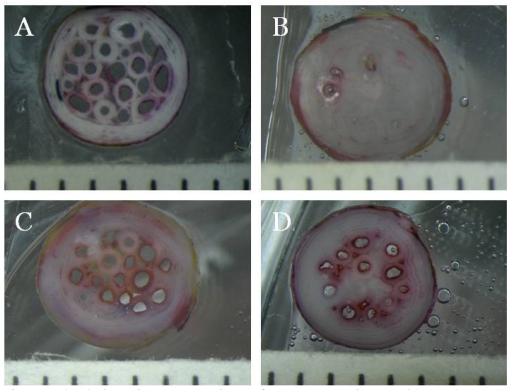

Figure 2. Alizarin red stains of cross-sections: A) M6h , B) premin1h_M6h, C) CL_M6h, and D) CL_premin1h_M6h

Pre-mineralization for 1h and cross-linking resulted in a significant increase in elastic moduli for scaffolds mineralized for 24h.

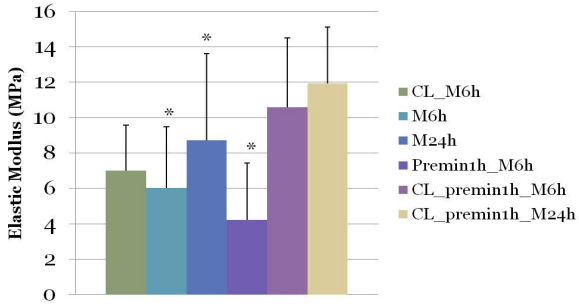

Figure 3. Compressive elastic moduli (MPa). * significantly different from CL_premin1h_M24h ($p<0.05$)

Conclusion:

We were able to successfully fabricated 3D electrospun scaffolds that mimic the organization of cortical bone. Cross-linking the gelatin and mineralizing the individual scaffold's components resulted in significant increases in mechanical properties and mineral deposition on the scaffolds. Future studies will focus on increasing strength and cell behavior on the scaffolds.