A Trilayer Fibrous Composite Scaffold with nHA for Periodontal Regeneration

Vinoy Thomas, Jordan Ball, Marco C. Bottino, Yogesh K. Vohra.
Center for Nanoscale Materials and Biointegration, Department of Physics,
University of Alabama at Birmingham (UAB), Birmingham, AL 35294, U.S.A.

Statement of Purpose: Guided tissue regeneration/guided bone regeneration (GTR/GBR) is a very popular way of treating periodontal defects. Membranes based on bioabsorbable polymers such as polylactide (PLA), polyglycolide (PGA), and/or processed-collagen do not meet the mechanical requirements of their application and also lack the nanoscaled morphology of extracellular matrix proteins, provided by electrospinning [1]. The native ECM in the periodontal region consists of 65-70% inorganic matter, mostly consisting of apatite particles [2]. Because of this, nano-particulate hydroxyapatite (nHA) has often co-spun to form nanofibrous scaffolds with uniformly distributed nHA. But, an interface-implant such as periodontal membrane needs to utilize a graded-structure with compositional gradients and sub-compartments that meets the local functional requirements. Therefore, in this study we reports the processing and characterization of a composite scaffold with bioactive nHA-gradient by concurrent electrospinning and spraying techniques. The rationale of having a periodontal membrane with a graded-structure again relies on the idea that one can tailor the properties of the different layers to design a membrane that will retain its structural, dimensional and mechanical integrity long enough to enhance periodontal regeneration.

Methods: Polydioxanone (PDO) and Polyglyconate (Maxon) were obtained in the form of surgical sutures. nHA with an average particle diameter of 100-150 nm was purchased from Nanocerox (Ann Arbor, MI). PDO and Maxon were mixed at a 1:1 mass ratio and dissolved in HFP (15%, w/v polymer solution) for electrospinning. nHA dispersed in HFP (4% ,w/v) was sonicated for 90 min, 1% (w/v) bovine gelatin (Type B) was added, and sonicated for another 90 min to form a nHA/gelatin suspension for electrospraying. The sequential spinning was in the order; PDO/Maxon solution was spun onto a rotating mandrel (100 rpm) for the first layer (for facing epithelial cells), then, concurrent spinning of PDO/Maxon and spraying of nHA/gelatin on to the first layer, and finally concurrent spinning/spraying from three syringes (one PDO/Maxon spinning and two nHA/gelatin spraying, from opposite directions) on to the second layer to form the nHA-rich layer (interfaces with alveolar bone). The free-standing functionally graded membrane (FGM) was further solvent sintered by exposing to dichloromethane for 10 min in a closed chamber at room temperature. Thereafter, the FGM was pressed at 5 MPa for 2 min to increase the inter-fiber bonding. The FGM was dried in vacuum for 48 h before the compositional, morphological, and mechanical characterizations.

Results: We have designed and fabricated a functionally graded membrane (FGM) with bioactive nHA gradient as a periodontal membrane (~450 µm) to enhance the bone growth while preventing the migration of gingival epithelial cells. Thermogravimetric analysis (TGA) and infrared spectroscopy (IR) confirmed the gradient composition of nHA in the trilayered FGM. IR spectra showed an increase in phosphate peak intensity around 570 cm^{-1} with respect to nHA content.

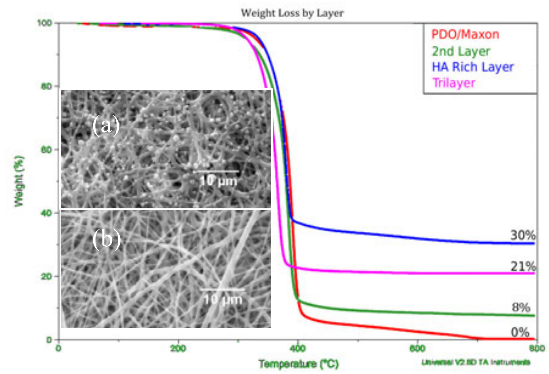

Figure 1. TGA scans of various layers and FGM. Inset shows SEM images of FGM layers with 30% nHA (a) and 0 % nHA (b).

Scanning electron microscopic (SEM) analysis has shown that a highly porous nanofibrous matrix with random fiber fashion was formed. Majority of fibers have diameters in the range of 400-800 nm in all the three layers, close to the upper range of collagen fibers (50-500 nm). nHA spraying has resulted in spherical nHA gels of diameters from 500 nm to 1 µm. The mechanical properties were characterized by un-axial tensile testing. FGM exhibited an average tensile strength of 4.3 MPa and an average modulus of 18 MPa with a failure strain of 235%. Solvent sintering followed by mechanical pressing has resulted in a drastic increase in tensile strength (to 10.4 MPa) and modulus (to 50 MPa) and a decrease in strain value (to 190 %). The sintering /pressing processes have improved the adhesion between the layers, creating a stronger laminar composite.

Conclusions: The simultaneously electrospun/sprayed trilayer membrane with a nHA gradient composition exhibits an ECM-like nano-topography and favorable mechanical properties. The solvent sintering and mechanical pressing have increased the fiber-to-fiber fusion (point-bonding) significantly as evident from the improved mechanical properties. In the future, in vitro degradation and cell interactions using osteoblasts and gingival fibroblasts will be performed.

References:

1. Bottino MC. Acta Biomater 2011; 7: 216-224.
2. Eastoe JE. J Biochem 1954; 57: 453-459.

Acknowledgement: The funding from NSF grant-Research Experiences for Undergraduates (REU)–site under DMR-0646842 is gratefully acknowledged.