Dependence of Osteogenic Cell Proliferation on the Morphology of Starch Based Scaffolds

Sangwon Chung[1,2], Vitor M. Correlo[1,2], Nuno M. Neves[1,2], Ricardo Picciochi[1,2], João F. Mano[1,2], Rui A. Sousa[1,2], Rui L. Reis[1,2]
[1]3B's Research Group-Biomaterials, Biodegradables and Biomimetics, University of Minho, Braga, Portugal
[2]Department of Polymer Engineering, University of Minho, Campus de Azurém, Guimarães, Portugal

Statement of Purpose: One of the critical components in tissue engineering is the design and fabrication of tailored scaffolds aimed at promoting specific cellular functions, phenotype expression and extracellular matrix deposition. Although it is well known that ideal scaffolds should exhibit a porous and interconnected morphology coupled with adequate mechanical properties, the influence of their morphological and physical properties on the specific cell response has not been yet thoroughly explored. The scaffold morphology is important as it controls cell functioning by means of nutrient and waste products diffusion kinetics, as well as cell–cell interactions within the scaffold [1]. Furthermore, mechanical performance also plays an important role, as forces experienced by cells are likely to be influenced by the mechanical behavior of the scaffold construct itself and the microenvironment provided by it [2]. In this study, fiber mesh scaffolds based on a blend of starch with polycaprolactone (SPCL) were produced under different processing conditions in order to systematically assess the role of scaffold morphology on osteogenic cell proliferation.

Materials and Methods: A blend of starch and polycaprolactone (SPCL, 30/70 wt/wt) was melt spun into fibers with an average diameter of 210 µm. The melt spun fibers were used for the production of 3D fiber mesh scaffolds using a thermal based fiber bonding methodology. A specially designed compression mold was used to fabricate the scaffolds. Different scaffold morphologies were obtained by systematically varying fiber mass, fiber length and compression ratio parameters according to a L8 experimental design. The scaffolds obtained presented 0.6 cm in diameter, and thicknesses ranging between 1.5 mm and 4 mm. In order to assess the influence of scaffold morphology on proliferation of human osteoblastic cells, *in vitro* tests were performed using an osteoblastic cell line (SaOS-2) for 3, 7 and 14 days under static and dynamic culturing conditions. Morphological characterization of the constructs was conducted, together with quantification of DNA and Alkaline phosphatase (ALP) activity. The scaffolds were additionally characterized by scanning electron microscopy (SEM). 3D virtual modeling and 2D analysis was also conducted by micro-Computed Tomography (µ-CT). Mechanical properties were determined using dynamic mechanical analysis (DMA) between 0 to 15 Hz. Each sample was tested three times in order to assess eventual creep behavior under cyclic stress conditions.

Results and Discussion: The systematic variation of processing parameters enabled the production of random fiber mesh scaffolds with diverse range of porosities and average pore size dimensions. The typical morphology of the developed scaffolds is presented in Figure 1. The scaffolds possessed a high degree of interconnectivity and a range of porosity between 50% and 75%.

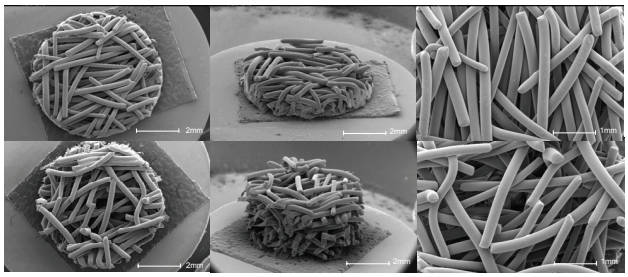

Figure 1-Morphology of SPCL fibrous scaffolds under SEM

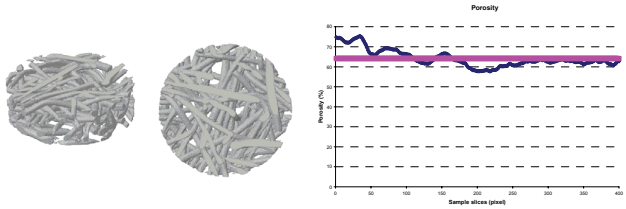

Figure 2- Example of 3D virtual modeling and 2D analysis for porosity measurement by micro-CT (average porosity 65%)

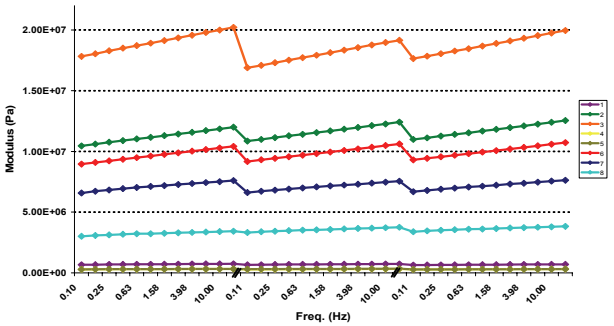

Figure 3-Compression modulus of SPCL scaffolds by DMA

Compression performance was assessed for 8 different morphologies, indicating a stiffness range between 0.3 and 19 MPa. There was a minor creep behavior among the samples as illustrated in Figure 3. Porosity and stiffness have shown poor direct correlation which emphasizes importance of other morphological parameters (fiber length, orientation and average contact points between fibers) on modulation of mechanical performance. Preliminary data showed good cell viability on the scaffolds. Cell proliferation and ALP activities of osteoblasts are currently being assessed for both static and dynamic culturing conditions.

Conclusions: It was possible to systematically assess the influence of morphology on mechanical performance of SPCL fiber mesh scaffolds. The on-going *in vitro* studies may shed extra light on the role of morphological and mechanical parameters on osteogenic proliferation under both static and dynamic conditions.

Acknowledgements: "Alea Jact Est" Marie Currie Early Stage Training (EST) Network and European NoE EXPERTISSUES.

References:

1. Miot S et al. Biomaterials 2005;26:2479-2489.
2. Agrawal CM, et al. J Biomed Mater Res 2001;55(2):141-150.