(OP 193) Morphometric and Mechanical Characterization, Insulin Loading and *In-Vivo* Biocompatibility of Chitosan Particles Aggregated Scaffolds for Tissue Engineering

P.B. Malafaya[1,2], J.T. Oliveira[1,2], T.C. Santos[1,2], M. van Griensven[3], R.L. Reis[1,2]

[1]3B's Research Group—Biomaterials, Biomimetic and Biodegradables, Department of Polymer Engineering, University of Minho, Campus de Gualtar, 4710-057 Braga, Portugal
[2]IBB—Institute for Biotechnology and Bioengineering, PT Government Associated Laboratory, Braga, Portugal
[3]Ludwig Boltzmann Institute for Experimental and Clinical Traumatology, Austrian Cluster for Tissue Regeneration, Donaueschingenstrasse 13, A-1200 Vienna, Austria

In tissue engineering, scaffolds development presents, among others, 3 key requirements: adequate morphological characteristics, mechanical performance and *in-vivo* biocompatibility. The aim of the present study was to evaluate chitosan-based scaffolds produced by particle aggregation in these key issues. Furthermore, chitosan scaffolds were loaded with insulin to promote chondrogenic differentiation. Micro-Computed Tomography (μ-CT) was carried out for accurate morphometric characterization quantifying porosity, interconnectivity, particles and pores size that shown to be adequate. Dynamical Mechanical Analysis (DMA) showed that scaffolds are mechanically stable in wet state with a storage modulus of 4.21 ± 1.04 MPa at 1 Hz frequency. Insulin-loaded scaffolds were characterized and studied with a pre-chondrogenic cell line (ATDC-5). The *in-vitro* release was carried out mimicking cell culture conditions quantified by micro-BCA. When seeded with ATDC-5, insulin-loaded scaffolds promoted the chondrogenic differentiation as assessed by SEM, DNA and GAG content, histology and real time-PCR. Furthermore, chitosan scaffolds were evaluated *in-vivo* using a rat muscle-pockets defect model for different implantation periods (1, 2 and 12 weeks). The histological and immunohistochemistry results have demonstrated that chitosan scaffolds are biocompatible. In addition, scaffolds interconnectivity shown to be favourable to the connective tissues ingrowth into the scaffolds and to promote the neo-vascularization even in early stages of implantation. It is concluded that chitosan scaffolds produced by particle aggregation could serve as alternative, biocompatible, and safe biodegradable scaffolds for tissue engineering applications.

Acknowledgements: FCT (SFRH/BD/11155/2002) and EU funded projects HIPPOCRATES and EXPERTISSUES.