Electrospraying of primary chondrocytes for cartilage repair

Ângela Semitela [(a)], Andreia Pereira [(a)], Cátia Sousa [(b)], Alexandrina Mendes [(b)], Paula A.A.P. Marques [(a)] and António Completo [(a)]

(a) TEMA, Department of Mechanical Engineering, University of Aveiro, Portugal.

(b) CNC, Centre for Neuroscience and Cell Biology, University of Coimbra, Portugal

(a) Aveiro, Portugal; (b) – Coimbra, Portugal

***Abstract*— Electrospun scaffolds have long been used for cartilage repair, due to the topographic similarity between the electrospun fibers and the collagen fibers of the extracellular matrix (ECM) in the native cartilage. Still, while their nanotophography can be beneficial for the cell proliferative and spreading behavior, it greatly reduces the inter-fiber pore size, hindering cell migration and relegating tissue formation to the surface of the scaffold [1]. A possible solution for this structural limitation would be the direct incorporation of cells into the fibers during electrospinning of the fibrous scaffold, overcoming the challenges of cell infiltration into small pore sizes by literally surrounding cells with the fiber matrix as it is produced [1]. This can be achieved using cell electrospraying, a concept first introduced in 2005 by Jayasinghe, enables the deposition of living cells onto specific targets by exposing the cell suspension to an external high intensity electric field [2]. Cell exposure to the electric field, as well as the shear stress of passing through the cell electrospraying apparatus may affect cell viability and function, so several types of cells have been electrosprayed, and no significant influence was observed on a genetic, genomic and physiological level [4]. In fact, our previous work has demonstrated this inertness from a chondrocyte cell line (C28-I2) [5]. Still, these immortalized cells are genetically modified, and might not not accurately replicate the physiological conditions. Primary chondrocytes possess little proliferative ability, showing considerable dedifferentiation from a chondrocyte-like to a more fibroblast-like phenotype over time, particularly if growth factors are not used [5].**

In this regard, electrospraying experiments were performed with primary chondrocytes to assess the process influence on chondrocyte viability. After 24 hour-incubation, chondrocyte metabolic activity was measured, and these electrosprayed (E) cells were then slip and cultured in well plates and in three-dimensional anisotropic fibrous/porous scaffolds under static and perfused conditions. Non-electrosprayed (NE) cells were considered for comparison.

The obtained results confirmed that the behaviour of primary chondrocytes upon electric field exposure was significantly different from that obtained for the chondrocyte cell line, which can be attributed to the lower recovery ability of these cells. Nonetheless, an increasing proliferation rate was observed over time. The proliferation performance of NE and E primary chondrocytes on 3D environment followed a similar trend, with E primary chondrocytes possessing a significantly lower viability than the NE primary chondrocytes. The application of perfused conditions to the E chondrocyte-seeded scaffolds greatly increased the chondrocyte viability to values similar to the ones obtained for NE chondrocyte-seeded scaffolds. Even though the electrosprayed primary chondrocytes suffered a substantial proliferative delay, they were able to recover, particularly under perfused conditions, suggesting that these conditions should be implemented after the electrospraying process, so that this technology might become an effective approach to uniformly incorporate primary chondrocytes into electrospun scaffolds.

Keywords*— *cartilage tissue engineering, chondrocyte, electrospraying

ACKNOWLEGEMENTS

This work was supported by the Portuguese funding of Program COMPETE-FEDER, Programa Operacional Competitividade e Internacionalização through the projects POCI-01-0145-FEDER-028424, and CENTRO-01-0145-FEDER-022083. Also, by Fundação para a Ciência e Tecnologia I.P. (FCT, IP) through the projects PTDC/EME-SIS/28424/2017 and UID/EMS/00481/2019. The authors thank to FCT for the PhD grant SFRH/BD/133129/2017.

TOPIC

2) a.: Multiscale technologies and devices for medicine, environment and energy

REFERENCES

[1] J. J. Stankus *et al*, Fabrication of cell microintegrated blood vessel constructs through electrohydrodynamic atomization, Biomaterials. 28 (2007) 2738–46.

[2] S. N. Jayasinghe, A. Townsend-Nicholson. “Stable Electric-Field Driven Cone-Jetting of Concentrated Biosuspensions”. Lab Chip 2006, 6 (8), 1086–90.

[3] S. N. Jayasinghe. “Cell Electrospinning: A Novel Tool for Functionalising Fibres, Scaffolds and Membranes with Living Cells and Other Advanced Materials for Regenerative Biology and Medicine”. Analyst 2013, 138 (8), 2215.

[4] Semitela et al, “Chondrocyte electrospraying for cartilage tissue engineering”, unpublished.

[5] E. M. Darling, K. A. Athanasiou. “Growth Factor Impact on Articular Cartilage Subpopulations”. Cell Tissue Res. 2005, 322 (3), 463–473.