

Bilayered scaffolds colonized with dental pulp stem cells for osteochondral tissue engineering application

Laura Bertoni¹, Manuela Zavatti¹, Elisa Resca², Tullia Maraldi¹, Francesca Beretti¹, Gianluca Carnevale¹, Alessandra Pisciotta¹, Anto De Pol¹

¹Dipartimento chirurgico, medico, odontoiatrico e di scienze morfologiche con interesse trapiantologico, oncologico e di medicina rigenerativa, Università di Modena e Reggio Emilia, Modena, Italy - ²Dipartimento di Scienze Mediche e Chirurgiche Materno-Infantili e dell'Adulto, Università di Modena e Reggio Emilia, Modena, Italy

The development of osteochondral tissue engineered interfaces would be a novel treatment for traumatic injuries and aging associated diseases that affect joints [1]. Dental Pulp Stem Cells (DPSC) are a promising alternative source in cell-based tissue engineering for the easily recruitment, the self-renewal and differentiation potential, being able to differentiate toward chondrogenic and osteogenic lineage [2].

The aim of this study was to develop scaffold that would support an osteochondral interface. DPSCs, isolated from dental pulp by selection with c-Kit-CD34 and STRO1 antibodies, were either cultured on fibroin and collagen scaffolds and differentiated into osteogenic cells or encapsulated in alginate and differentiated into chondrogenic cells. After 3 weeks of culture, the differentiation of the cell-scaffold constructs was analyzed by histology and immunofluorescence. The cell seeded scaffolds were then brought together (fibroin+alginate and collagen+alginate) in order to form a combination that would provide a supportive scaffold for bone formation as well as a soft matrix for cartilage growth. The resulting single constructs were cultured for 2 weeks in a osteochondral media, developed by combining some of the growth factors and nutrients of osteo- and chondrogenic media. The constructs were then characterized for expression of bone and cartilage specific markers to evaluate the capability of the cells to not dedifferentiate in the new cultured conditions.

We demonstrated that both collagen and fibroin appear to be excellent scaffold for cells differentiated into osteogenic lineage, although they have different structural characteristics. Similarly, the alginate has allowed cells to produce ECM-rich in collagen II and aggrecan during chondrogenic differentiation. Moreover DPSC in the bilayered constructs remain viable and differentiated even in the osteochondral medium.

Thus the integrated fibroin-alginate or collagen-alginate proved to be functional *in vitro* and may lead to the development of new strategies for osteochondral repair and regeneration.

References

- [1] Rodrigues MT et al. (2012) Bilayered constructs aimed at osteochondral strategies: the influence of medium supplements in the osteogenic and chondrogenic differentiation of amniotic fluid-derived stem cells. *Acta Biomater.* Jul;8(7):2795-806.
- [2] Maraldi T et al. (2013) Human amniotic fluid-derived and dental pulp-derived stem cells seeded into collagen scaffold repair critical-size bone defects promoting vascularization. *Stem Cell Res Ther.* May 21;4(3):53.

Keywords

DPSC; fibroin; collagen; osteochondral regeneration.