

Human tissue culture of osteochondral defect: An advanced *in vitro* model

Melania Maglio,¹ Matilde Tschon,¹ Stefania Pagani,¹ Leonardo Vivarelli,² Marco Govoni,² Martina Rocchi,² Alessandro Mazzotta,² Lucia Martini,¹ Dante Dallari,² Milena Fini¹

¹IRCCS Istituto Ortopedico Rizzoli, Complex Structure Surgical Sciences and Technologies, Bologna; ²IRCCS Istituto Ortopedico Rizzoli, Reconstructive Orthopaedic Surgery and Innovative Techniques - Musculoskeletal Tissue Bank, Bologna, Italy

Abstract

The optimization of advanced *in vitro* models is essential for the development of alternative methods. The *in vitro* study of osteochondral regeneration still has numerous limitations and wide scope for exploration.

Introduction

The ageing population has led to a higher incidence of degenerative diseases affecting the joints, with pain and limitations in the quality of life of patients. Osteochondral lesions can be treated with autologous/allogenic grafts or with synthetic materials. The evaluation of these treatments today requires mainly the use of *in vivo* models. However, the development of advanced *in vitro* tissue models is an area of growing interest in the field of tissue engineering and regenerative medicine, in order to reduce the use of animal models, thus recreating a reliable microenvironment *in vitro* for preliminary investigations.

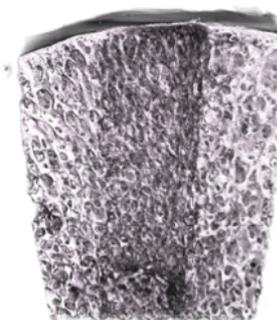


Figure 1. 3D microtomographic image of the defect created in the osteochondral tissue sample.

In this field, the use of human biological materials from surgical procedures or from tissue banks represents a great opportunity for the development of valid alternative models.

To date, as regards the study of osteochondral regeneration, the models present in literature are few, variable and difficult to compare with each other, creating an important gap in this research field.¹

Aim of the present study is to set up an *in vitro* 3D tissue models simulating osteochondral defects using human osteochondral tissue, in order to assess the best protocol to perform a long tissue culture for possible evaluation of the regenerative potential of biomaterials/bank products, cells, growth factors or combination of them.

Materials and Methods

Osteochondral tissue samples cylindrically shaped were carved from the femoral head of cadaver donors. From each sample, a smaller inner portion was extracted (Figure 1); to fully characterize the model, autograft (inner core of the same donor), allograft (inner core from different donor) and empty tissue explants (without inner core) were cultured for 8 weeks in dynamic culture condition. Tissue viability was assessed once a week. At the end of the experimental time, microtomographic and histologic evaluations were performed to assess bone and cartilage status and to study the interface between grafts and tissue. Gene expression analyses were performed on tissue samples at the time of harvesting and at the end of culture to evaluate the possible activation of genes involved in bone and cartilage metabolism.

Results

Viability of cultured osteochondral tissue samples remained stable during all culture time, without significant fluctuations and regardless of the experimental group. Microtomographic images showed signs of initial integration of the graft, mainly along the lateral contact surfaces between graft and defect. The histological observation highlights matrix deposition and bone and cartilage cells activities. Analyses of gene expression profiles are in progress.

Discussion and Conclusions

The development of advanced *in vitro*

Correspondence: Melania Maglio, IRCCS Istituto Ortopedico Rizzoli, Complex Structure Surgical Sciences and Technologies, Bologna, Italy.

E-mail: melania.maglio@ior.it

Key words: 3D *in vitro* models; human tissue; osteochondral defect.

Acknowledgments: The work is supported by Ministry of Health- 5x1000 project "Medicina rigenerativa e riparativa personalizzata per le patologie dei tessuti muscolo-scheletrici e la chirurgia ricostruttiva ortopedica" (year 2019)

Disclosures: The authors declare no conflict of interest.

Conference presentation: This paper was presented at the Third Centro 3R Annual Meeting - L'era delle 3R: modelli *in silico*, *in vitro* e *in vivo* per promuovere la ricerca traslazionale - 30 September - 1 October 2021, Evento online organizzato dal Politecnico di Torino.

Received for publication: 9 July 2021.

Accepted for publication: 7 September 2021.

This work is licensed under a Creative Commons Attribution NonCommercial 4.0 License (CC BY-NC 4.0).

©Copyright: the Author(s), 2021

Licensee PAGEPress, Italy

Biomedical Science and Engineering 2021; 4(s1):177

doi:10.4081/bse.2021.177

tissue models is essential to achieve the goal of reducing and refining and, if possible, replacing preclinical *in vivo* models, but for this purpose it is necessary to validate reliable, reproducible experimental setups that allow complete evaluations. The use of human biological material represents an additional advantage to make such models valid and appealing. Data obtained so far from this 3D human tissue culture suggest that the proposed set up allow to perform a reliable and replicable model, able to resemble an osteochondral defect condition for the preliminary evaluation of treatments and therapies like new biomaterials/scaffolds.

References

1. Maglio M, Tschon M, Sicuro L, et al. Osteochondral tissue cultures: Between limits and sparks, the next step for advanced *in vitro* models. *J Cell Physiol* 2019;234:5420-35.