# Fibroblast-seeded lung extracellular matrix (ecm)-derived hydrogels as an in vitro model for stromal bed in idiopathic pulmonary fibrosis (ipf)

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## Abstract 452

### DECELLULARIZED EXTRACELLULAR MATRIX FROM HUMAN BONE AS VERSATILE PLATFORM TO STUDY THE COMPLEXITY OF MSC – MATRIX INTERACTIONS IN VITRO

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Despite the considerable volume of research into the biology and regenerative potential of MSC, the lack of suitable models that recapitulate the in vivo situation has led to poor clinical translation1. Decellularized constructs have the ability to present tissue-specific ECM components in their native organization.

In this work, three different constructs, i.e. 2D matrix, 3D scaffold and hydrogel, were optimized as meaningful in vitro models that allow studying the influence of microenvironmental cues on MSC. A 2D decellularized cell-derived matrix was obtained by 10 or 21 days cultivation of human MSC followed by incubation with high-pH detergent solution and DNase. A 3D bioscaffold was produced by EDTA-based decellularization and decalcification of human femoral heads2. A translucent 3D hydrogel was obtained by pepsin digestion of decellularized bone powder followed by neutralization at 37°C for 1h.

Removal of DNA was confirmed by nuclei absence observed on histological sections. Preservation of specific bone ECM-related proteins was analyzed by SDS-Page and immunostaining (particularly strong presence of preserved collagen-I was observed for all models). The ultrastructure of models was investigated by SEM and/or microCT, confirming preservation of the native ECM architecture. Resazurin assay and FACS analysis of recovered cells showed sustained MSC viability. Osteogenic differentiation seemed to be promoted by the models, as observed by alizarin-red staining and gene expression analysis. Future studies will investigate how the different complexity of models affects stemness and mechanosensing in MSC, providing insights in the influence of these microenvironmental cues on the regenerative potential of MSC.

### References

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## Abstract 453

### FIBROBLAST-SEEDED LUNG EXTRACELLULAR MATRIX (ECM)-DERIVED HYDROGELS AS AN IN VITRO MODEL FOR STROMAL BED IN IDIOPATHIC PULMONARY FIBROSIS (IPF)

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Introduction: Idiopathic Pulmonary Fibrosis (IPF) is characterized by aberrant extracellular matrix (ECM) deposition and remodeling, which orchestrates cellular responses to the fibrotic microenvironment[1]. Decellularized lung ECM-derived hydrogels resemble the mechanical properties[2] of native decellularized tissues, potentially providing a 3D model mimicking native cell-ECM interactions. We aimed to characterize this 3D human lung microenvironment model, with respect to stiffness and viscoelastic properties, in the presence and absence of primary human lung fibroblasts.

Materials & Methods: Lyophilized powders of decellularized IPF and control lung matrices (pool of 6 patients) were pepsin digested, and formed to hydrogels seeded with control primary lung fibroblasts ($n=4$ donors), and cultured for 14 days. Stiffness and viscoelastic relaxation were measured by Low-Load Compression Testing[2] (20% strain).

Results: IPF hydrogels were stiffer than controls ($1.84\pm0.33$ kPA vs $1.37\pm0.35$ kPA), and became even more stiff when cell-seeded ($1.91\pm0.37$ kPA) in contrast to controls which became softer ($1.09\pm0.27$ kPA). Time to reach 100% viscoelastic relaxation was shorter in cell-seeded compared to native hydrogels for both IPF ($19.14\pm3.17$ vs $41.6\pm37.66$ seconds) and control ($11.44\pm6.55$ vs $22.21\pm19.59$ seconds).

Conclusion: The mechanical properties of the ECM hydrogels were modified by fibroblasts, while in turn the ECM microenvironment altered cellular responses. These data suggest that higher stiffnesses and altered relaxation patterns of the ECM could contribute to the fibrotic response in IPF by instructing the cells. Fibroblast-seeded ECM-derived hydrogels can provide more insight on cell-ECM interactions in IPF.

### References

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## Abstract 454

### USE OF TRANSCRIPTOMIC DATA FROM NON-WOUND HEALING PATIENTS TO PURPOSE IDENTIFIED GENES INVOLVED IN THE DISEASE.

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The molecular mechanisms for tissue regeneration and wound healing are not yet well understood. Limited success of clinical trials indicates that a crucial aspect of the growth factor wound healing strategy is the effective delivery of these polypeptides to the wound site. A meta-analysis approach in which genetically clear the role of each gene and protein or signal pathway can help to overcome the limitations associated with the application of recombination growth factor proteins. Studies point to the synthesis of factors within the chronic wound environment that could have deleterious effects on the repair process. So, the understanding of how damage or loss of tissue can be reconstructed is one of the biggest challenges in biomedical research and facilitating reconstruction. In this paper, we use a bioinformatics approach and network theory for more accurately and better understanding the genetic underpinnings of wound healing