Development of a device for the maturation of alveolar cells at the air-liquid interface

Idiopathic pulmonary fibrosis (IPF) is a severe, progressive and unpredictable lung disease that currently has no cure and is often fatal. It is characterized by diffuse interstitial infiltrates, excessive extracellular matrix (ECM) deposition leading to stiffening of the pulmonary parenchyma, and destruction of functional lung tissue composed of alveolar epithelial cells, fibroblasts, endothelial cells and alveolar macrophages.

Many studies have enlightened the role of the mechanical environment in orienting cell fate [1,2]. As it evolves during IPF progression, it has become apparent that it may contribute to the evolution of the disease. However, other factors are also involved in IPF progression, such as secreted factors. Previous in vitro studies on the role of the mechanical environment in IPF onset and progression have used oversimplified systems [3], often neglecting intercellular communication between the different resident cell types. And indeed, the impact of matrix stiffness, mimicking different stages of IPF, on various cell types found in IPF tissues remains unknown. We aim here **to engineer a new culture set up that allows addressing the following questions: Does matrix rigidity contribute to the emergence of altered and undefined cell populations in the alveolar capillary barrier that drive the fibrosing process?**

The objective of the internship is to develop a novel 2D, soft culture support system that allows different cell types to grow in separated compartments while enabling the exchange of soluble factors secreted by the cells. The culture support must be compatible with air/liquid interface culture conditions, so that epithelial cells can differentiate into an alveolar barrier. This development implements on previous studies, in which a first prototype has been designed. The internship will address several steps:

- the implementation of a controlled protocol for surface functionalization of the culture support (click-chemistry, micro BCA assays, immunofluorescence, confocal microscopy)
- toxicity assay to evaluate the inertness of the materials used (propidium iodide, CCK8 or other; fluorescence microscopy, absorbance measurements; IA driven image analysis)
- the analysis of epithelial cell growth on the designed culture system, focusing on cell morphology, proliferation and phenotypic drifts (phase contrast videomicroscopy, western blots).
- measure of their secretion after TGFb stimulation, and the way these secretions diffuse to the neighboring chambers (micro BCA

The internship will be conducted at LTM. Fabrication steps will be performed in a microfabrication lab equipped for sterile manipulations, while cell experiments will be performed in a L2 laboratory. This work will be conducted in close collaboration with E. Boncoeur, expert in physiopathology of lung tissues (CEA/BGE/Edyp), and Cell&Soft, a spin-off of LTM expert in the design of soft culture supports for in vitro assays. This work is conducted in the collaborative context of a joint laboratory between the 3 teams (LabCom SoftLab).

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