université PARIS-SACLAY
FACULTÉ DE PHARMACIE

Graduate School: Health and Drug Research
Master 2 Pharmaceutical Technology and Biopharmacy

Company/laboratory/public institution: Laboratory for Vascular Translational Science (LVTS U1148 INSERM)

Address: 46 rue Henri Huchard, 75018 PARIS

Supervision of trainee:

Name of tutor: Teresa Simon-Yarza & Elliot Lopez

Position: TSY: PharmPhD Chargé de recherche Inserm

EL: PhD student

E-mail: teresa.simon-yarza@inserm.fr & elliot.lopez@inserm.fr

Internship period: 20 January - 18 July 2025

Title of the project: Role of cancer-associated fibroblasts in an in vitro vascularized tumor model

Three-dimensional culture models are considered as a promising tool to study physiologically relevant *in vitro* human models. They represent a compromise between simplistic monolayer models and compelling animal models. Over the past decade, cell-biomaterial and cell-cell interactions have been proved highly valuable for the constitution of a relevant model, and further studies have been conducted on the mechanobiology of the matrices[1], as well as on the synergistic effects of coculture both in physio- and pathological cases for example[2–5]. It is now well established that supportive cells such as fibroblasts, pericytes or mesenchymal stromal cells play a role during angiogenesis, as well as during tumor development. Besides, the need for a vascular compartment, and the reciprocal influence of endothelial and cancer cells are widely studied as well[6,7]. Yet, few quantitative studies have been conducted on the interactions of supportive, endothelial and cancer cells regarding development and perfusion of the endothelial network, secretions by any of these cell types, and possible migration of cancer cells, among others. It is therefore challenging to understand what is the influence of each cell type over the others.

This internship aims at adapting a coculture system already studied in the lab to a triculture setup with fibroblasts, endothelial and cancer cells, using hydrogels of pullulan and dextran[4,8,9]. The mechanical and biological properties of the matrix have been studied already[10], and can be tuned to convenience to help the cells adhesion, migration, and communication. Then, the influence of each cell type will be studied by investigating the three two-cell-type-system independently by proteomics analysis, evaluation of the permeability under perfusion, and immunostaining and 3D imaging. In particular, the work will involve:

- Hydrogel synthesis and possible protocol optimizations.
- Cell culture with human umbilical vein endothelial cells (HUVECs), breast cancer cells and breast-cancer-derived fibroblasts.
- Protein quantification assays and permeability testing under flow.
- Immunostaining and confocal microscopy to assess the integrity of the vascularization.

The intern is expected to be interested in cell culture, in biomaterials, and in engineering and microfluidics. The intern is highly encouraged to take initiatives and to propose ideas. This internship will take place in our laboratory in Bichat Hospital, 18e, Paris, within a team and a lab constituted by more than 200 people, including researchers, clinicians, engineers, postdocs, PhD students, and interns. The team is specialized in biomaterials and hydrogel production for tissue engineering, and therefore owns the equipment necessary to accomplish this project, including the process of fabrication and characterization of the materials, as well as 3D cell dynamic culture and biomolecular analysis. The lab is young and dynamic, and the student will benefit from many soft-skills and communication trainings such as journal club sessions, weekly team meetings, or a "3-min-M2" contest. We are also committed to facilitate student integration by sharing time together, such as meals and social activities.

1. Kretschmer, M., Mamistvalov, R., Sprinzak, D., Vollmar, A. M. & Zahler, S. Matrix stiffness regulates Notch signaling activity in endothelial cells. J Cell Sci 136, (2023).
2. Nguyen, M. et al. Dissecting Effects of Anti-cancer Drugs and Cancer-Associated Fibroblasts by On-Chip Reconstitution of Immunocompetent Tumor Microenvironments. Cell Rep 25, 3884-3893.e3 (2018).
3. Song, H. H. G. et al. Transient Support from Fibroblasts is Sufficient to Drive Functional Vascularization in Engineered Tissues. Adv Funct Mater 30, (2020).
4. Dellaquila, A. et al. Fibroblasts mediate endothelium response to angiogenic cues in a newly developed 3D stroma engineered model. Biomaterials Advances 154, 213636 (2023).
5. Kenny, H. A., Krausz, T., Yamada, S. D. & Lengyel, E. Use of a novel 3D culture model to elucidate the role of mesothelial cells, fibroblasts and extra-cellular matrices on adhesion and invasion of ovarian cancer cells to the omentum. Int J Cancer 121, 1463–1472 (2007).
6. Lopez-Vince, E., Wilhelm, C. & Simon-Yarza, T. Vascularized tumor models for the evaluation of drug delivery systems: a paradigm shift. Drug Delivery and Translational Research 2024 1–26 (2024) doi:10.1007/S13346-024-01580-3.
7. Dellaquila, A., Le Bao, C., Letourneur, D. & Simon-Yarza, T. In Vitro Strategies to Vascularize 3D Physiologically Relevant Models. Advanced Science 8, (2021).
8. Simon-Yarza, T., Labour, M. N., Aid, R. & Letourneur, D. Channeled polysaccharide-based hydrogel reveals influence of curvature to guide endothelial cell arrangement in vessel-like structures. Materials Science and Engineering C 118, (2021).
9. Le Bao, C. et al. Spatial-Controlled Coating of Pro-Angiogenic Proteins on 3D Porous Hydrogels Guides Endothelial Cell Behavior. Int J Mol Sci 23, 14604 (2022).
10. Grenier, J. et al. Mechanisms of pore formation in hydrogel scaffolds textured by freeze-drying. Acta Biomater 94, 195–203 (2019).