Laboratoire Lasers, Plasmas et Procédés Photoniques
UMR 7341 CNRS - Aix-Marseille Université - Campus de Luminy C917 - 13288 Marseille Cedex 09, France

PH'D THESIS PROPOSAL 2020

Laboratory :

LP3, Laser Plasma and Photonic Processes, www.lp3.univ-mrs.fr

Advisors :

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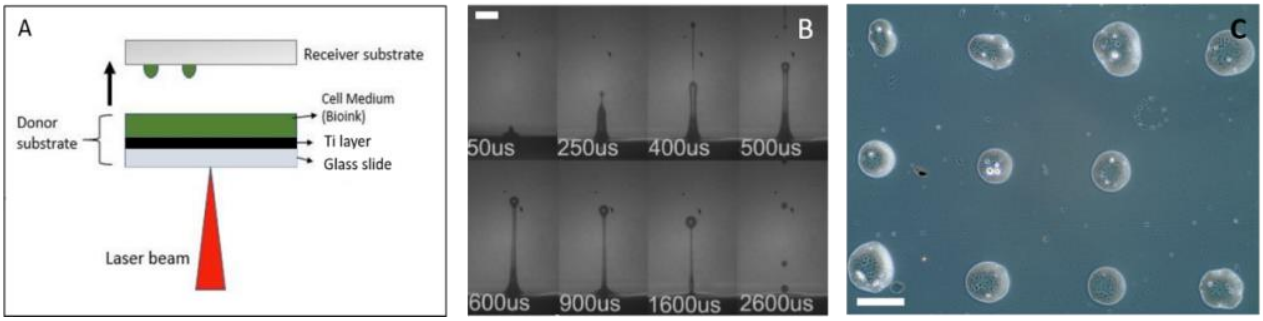
Title : Laser-induced printing of stem cells for the creation of ordered neuromuscular junctions

Printing techniques applied to biology have begun to develop since the 2000s and they undergo a high development since a few years. They are based on **interdisciplinary approaches** and use a combination of cells, chemistry, engineering and sophisticated protocols to create artificial tissues. Their applications range from tissue engineering for organ creation to regenerative medicine and new drug discovery. The main goal of these techniques is to create a 2D/3D microenvironment that more likely reflects the in vivo complexity and architecture of tissues consisting of the extracellular matrix (ECM), biomolecules and cells.

In this context, at LP3, we plan to combine cellular biology based on stem cells research and two laser techniques (laser-assisted printing, laser surface texturing) to investigate different 2D/3D bio-patterns for a better understanding of the growing environment impact (from a topological point of view to chemical one) on the cell behavior to optimize muscle differentiation in mature myotube populations and to finally create active neuromuscular junctions in-vitro.

To do so, the Ph'D student could take advantage of the LP3 experience on the laser-induced forward transfer (LIFT) technique[1], firstly developed for several years for applications in microelectronics, to master the printing of stem cells in ordered patterns.



A) Sketch of LIFT process, B) Example of bioink ejection and C) Droplets with stem cells printed by LIFT

The idea is to focus a short pulse laser beam through a transparent substrate (donor) coated with two layers of material: the first one, very thin, aiming at absorbing the radiation, and the second one, thicker, composed of the targeted biomaterial (typically a hydrogel with embedded cells) (**Figure 1**.A). Under appropriate conditions, the laser beam induces the formation of a liquid jet (**Erreur ! Source du renvoi introuvable.**B) propagating perpendicularly from a donor substrate to a receiver. The biomaterial is then deposited as a droplet on a collector substrate (**Erreur ! Source du renvoi introuvable.**A).

The LIFT process has been successfully applied in laboratory-scale trials for the deposition of organic and inorganic compounds including biomaterials and cells[2,3].

The project implies, firstly, a thorough study of the transfer mechanisms and the jet formation according to laser parameters (energy, wavelength, pulse duration, laser-absorbing layer, irradiation ….) and bioink properties (thickness, composition, rheological properties, cells content….). Then, we aim at printing 2D/3D patterns of biomaterials containing stem cells with a high degree of control in term of volume and location.

In addition, we aim at using the ability of lasers to structure surfaces at the micro and nanoscales and studying its impact on the cells behavior. Indeed, ripples, spikes or groves can be formed with a very good control of size and shape on any kind of materials. These treatments allow tuning some surface properties like hydrophobicity, adhesion or light absorption and could impact proliferation, adhesion, migration, and even higher-order functions such as differentiation[4,5].

Experiments will be conducted using different lasers sources available in LP3. They will be associated with real time visualization of liquid ejection (**Figure 1**.B).

For the biological aspects of this study the applicant will benefit of the collaborations between the LP3 and biology laboratories particularly interested in these studies such as Marseille Medical Genetic (MMG).

Bibliography:

1. *Delaporte et al., Laser-Induced Forward Transfer: A high resolution additive manufacturing technology, Invited paper, J. of Optics & Laser Technology 78,33–41 (2016)*
2. *Zhang et al. Biomicrofluidics 1, 034120 (2017)*
3. *Koch L et al., International Journal of Bioprinting (2017)–Volume 3, Issue 1*
4. *Fadeeva, E., Deiwick, A., Chichkov, B. & Schlie-Wolter, S. Impact of laser-structured biomaterial interfaces on guided cell responses. Interface Focus 4, 20130048 (2014)*
5. *Dinca, V. et al. Bio-Interfaces Engineering Using Laser-Based Methods for Controlled Regulation of Mesenchymal Stem Cell Response In Vitro. in Recent Advances in Biopolymers (ed. Parveen, F. K.) (InTech, 2016)*

To apply: Please provide CV (including notes/marks or anything that could reinforce your application) and letter of motivation to Anne Patricia ALLONCLE patricia.alloncle@univ-amu.fr and Adrien CASANOVA adrien.casanova@univ-amu.fr