





Master in Photonics – "PHOTONICS BCN" Master ERASMUS Mundus "EuroPhotonics"

MASTER THESIS PROPOSAL

Starting full time from April 2024 Presentation at the end of July or beginning of September 2024

Laboratory: Wavefront Engineering Group&Grup de Biofotònica, Departament de Física Aplicada Institution: Universitat de Barcelona (UB) City, Country: Barcelona, Spain.

Title of the master thesis: STED beam generation: structured illumination via doble pass of single spatial light modulator.

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Keywords: Structured illumination microscopy, depletion (STED and RESOLFT) microscopy, fluorescence microscopy, spatial light modulation, digital holography, beam design, donut beams.

Summary of the subject (maximum 1 page):

New microscopy techniques, such as super-resolution, STED, RESOLFT and others, require specially designed light beams to have a very specific behavior when being highly focused. This is because these techniques seek to exceed the limits imposed by optical diffraction. In the case of STED microscopy, two beams of different wavelengths are combined, one for excitation and one for depletion [1]. The excitation beam aims to generate fluorescence in its zone of action, with a Gaussian profile limited by diffraction, while the depletion beam depresses that fluorescence in an annular region, constraining the excited fluorescence in a much smaller area. The final resolution is determined by the size and depth of the central hole on the depletion beam, Fig. 1(a).

In STED microscopy, depletion beams are generated by using phase masks (binary, segmented into zones or spiral phase plates) or with polarization masks (such as those segmented into angular portions or retarder vortex plates) that allow the generation of azimuthally polarized beams. Other possibilities could be explored in order to achieve donut beams with deeper and sharper central holes [2]. In particular, much progress has been made in the field of beam design in last years, gaining more and more flexibility when generating arbitrary fields. This requires absolute control of the amplitude, phase, and polarization distributions throughout the beam. Using liquid crystal spatial light modulators, together with digital holography techniques, the amplitude and phase distributions of the



field can be controlled. Sculpting arbitrary distributions of polarization requires acting independently on the two transverse components of light: an interferometric setup with one spatial light modulator in each arm is usually used and then, both components are recombined [3]. This approach has some drawbacks, especially on the alignment stability and wavefront distortions. In this master's thesis we propose to design and build a system capable of achieving full control of amplitude, phase, and polarization distributions with a double pass of light onto a single reflective spatial light modulator, Fig. 1(b).



Figure 1. (a) Excitation-depletion of fluorescence in STED microscopy. A Gaussian laser beam (purple) excites fluorescent molecules. A donut (blue) brings back the periphery to the ground state by stimulated emission in a controlled manner. The final fluorescence is emitted from a sub-wavelength region near the optical singularity of the depletion beam. (b) Initial approach of double pass onto a single SLM technique to sculpt arbitrary and non-homogenous amplitude, phase, and polarization distributions on a light beam.

[1] K. Willig et al, "STED microscopy with continuous wave beams." Nat Methods 4, 915-918 (2007).

[2] J. Tang. et al, "Fluorescence imaging with tailored light." Nanophotonics 8, 2111-2128 (2019).

[3] D. Maluenda et al, "Reconfigurable beams with arbitrary polarization and shape distributions at a given plane" Opt. Express **21**, 5432-5439 (2013).

Objectives:

The final goal of this TFM is to mount and align an optical system able to generate arbitrary amplitude, phase, and polarization beam distributions by means of a double pass of light through a single spatial light modulator.

First, different setup approaches will be designed and analyzed in terms of optical efficiency, polarization components requirements, distances, etc. Then, the optimal setup will be implemented on an optical table. On the other hand, different methods of hologram codification will be simulated and compared. Finally, several beams will be generated and fully characterized.

More details will be individually discussed with the candidates.

Additional information:

* Required skills: Disposition for experimental work in a microscopy lab, interested in biophotonics, proficient in a computer language with preference for LabVIEW and Python.

* Miscellaneous: Early incorporation is possible.