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Implementation of a commercial Lattice Light Sheet Microscope (LLSM) in an Imaging Facility (PICT-IBiSA)

Lattice Light Sheet Microscope (LLSM) represents the novel generation of 3D fluorescence microscopes dedicated to live single-cell analysis. LLSM[1] uses ultrathin light sheets derived from 2D optical lattices. These are scanned plane-by-plane through the specimen to generate a 3D image. The thinness of the sheet leads to high axial resolution and negligible photobleaching and background outside of the focal plane. By dithering the lattice to create a uniform light sheet, single cells can be imaged in 3D, often at sub-second intervals, from hundreds to thousands of time points at the diffraction limit (300 x 300 x 700 nm, 40 to 100 planes per second per cell). Photo-bleaching and photo-toxicity are typically reduced by one order of magnitude relative to that seen with a 1D scanned Bessel beam or spinning disk confocal microscopy. This allows 3D images to be captured over longer periods of time, and enables the study of signaling, transport, and stochastic self-assembly in complex environments with single molecule sensitivity. However, facing the amount of information provided by LLSM, cutting-edge image processing algorithms need to be investigated, at a time regime compatible with live cell imaging.
LLSM is commercialized by Intelligent Imaging Innovations (3i). However, as it is, the commercial version of LLSM, shows a number of drawbacks deserving a strict metrology control and alignment protocols. As early adopters of the 3i LLSM project, this is one of our aim to determine how this sophisticated system could be useful at the level of a large Imaging facility. Another goal of our project consists to optimize the LLSM for different biological live samples in 3D. In a second part of the project, it is planned to develop different software to help the reconstruction, visualization and analysis of data produced in the microscope.
[1] B.-C. Chen et al., Science 346, 1257998 (2014).

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Terms and Conditions

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