The use of Gaussian and Bessel beams for multimodal light-sheet fluorescence imaging

Omar E. Olarte[1], Jordi Andilla[1], David Artigas[1], Raphael Jorand[2,3], Bernard Ducommun[2,3,4], Corinne Lorenzo[2,3], Pablo Loza-Alvarez[1]

[1]ICFO-The Institute of Photonic Sciences, Av. Carl Friedrich Gauss, 3, Castelldefels (Barcelona), Spain
[2]University of Toulouse, ITAV, F-31106 Toulouse, France
[3]CNRS, ITAV-USR3505, F-31106 Toulouse, France
[4]CHU de Toulouse, F-31059 Toulouse, France

In light sheet fluorescence microscopy (LSFM), a cylindrical lens is used to illuminate a sample. This produces an illumination plane that lies at 90º from the optical propagation axis. By placing a microscope objective orthogonal to this plane, the fluorescence light emerging from it can be collected.
One interesting variant of SPIM is given by the Digitally Scanned Light Sheet Microscopy (DSLM) technique. This is based on focusing the light with a lens to produce a line that is scanned only in one direction, forming a plane similar to that produced in SPIM. This technique has the advantage that it can be employed with nonlinear excitation, as it preserves the required high intensities for two-photon fluorescence excitation (TPEF).
Here I will show a practical multimodal implementation of the LSFM technique in which the two modalities, linear and nonlinear, can be used. Furthermore, the setup also incorporates the option of introducing an axicon for generating Bessel Beams. In this work, I will present the complete setup working with the four different modalities: DSLM in the linear and nonlinear regime and with Gaussian and Bessel beams. I will also describe the complete system characterization and compare the performance of each of the techniques in terms of different imaging parameters such as contrast, light sheet uniformity, field of view etc., for different bio-samples.