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Optical beam shaping and microscopy methods for biological and biomedical imaging applications[1]

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While observing live biological specimens under the fluorescence microscope, scientists encounter a critical barrier defined as phototoxicity, which is the degeneration of a specimen under the light exposure. In this presentation, we will review how changing the geometry of excitation and collection of the light paths in a fluorescent microscope, by generating a sheet of excitation light, can overcome limitations given by phototoxicity. This new geometry allows improving the spatial resolution of the microscope to obtain isotropic resolution, 3D fast imaging and low phototoxicity, and obtain previously inaccessible dynamic information about biological processes, for example during cell proliferation and cell migration. For imaging single cells with sufficient spatial resolution and field of view, a Bessel beam light-sheet microscope was developed, in which a Bessel beam was generated using a fixed amplitude mask and then used for the excitation light instead of Gaussian beam. The separate geometry of excitation and detection also provides freedom to act on each of them separately with adaptive optics or beam shaping techniques, to improve imaging when aberrations are present or to lower phototoxicity by generating a thinner sheet of excitation light and a more versatile instrument

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