

Abstract Submitted
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4Pi

Spectral

Self-interference

Fluorescence Microscopy¹ MEHMET DOGAN, Department of Physics, Boston University, ANNA K. SWAN, Department of ECE, Boston University, M. SELIM UNLU, Department of ECE, Boston University, BENNETT B. GOLDBERG, Department of Physics, Boston University — 4Pi fluorescence confocal microscopy [1] improves axial resolution, and Spectral Self-Interference Fluorescence Microscopy (SSFM) [2] provides sub-nanometer localization of fluorescent emitters in biological structures. Here we report on the construction and evaluation of a 4Pi fluorescence confocal microscope and discuss the efforts to combine the high resolution 4Pi technique with SSFM. In the 4Pi microscope, the back focal planes of two opposing high numerical aperture objectives are filled with coherent laser illumination and counter propagating spherical wave fronts form constructive interference at the common focus of two objectives, resulting in an improvement of the axial point spread function (PSF). We characterized the 3-D PSF of the microscope using fluorescent polystyrene beads and fluorescent monolayers. We measured a factor of 3 improvement of the axial PSF compared to a confocal microscope. [1] S. W. Hell et al. *J.Opt.Soc.Am. A* Vol.9, No.12, pp.2159 (1992) [2] A. K. Swan et al. *IEEE JSTQE* Vol. 9, No. 2, pp. 294 (2003)

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