

NONLINEAR MODULATION LIGHT SHEET MICROSCOPY WITH ISOTROPIC SPATIAL RESOLUTION

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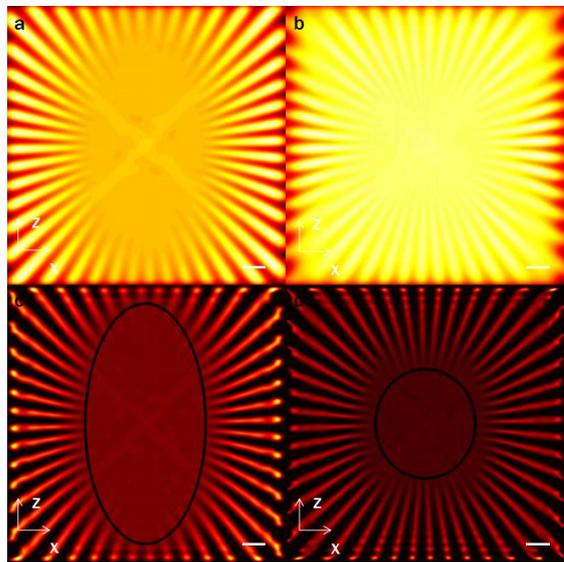
KEY WORDS: nonlinear modulation, multiview fusion algorithm, light sheet fluorescence microscopy, isotropic spatial resolution.

1. PRINCIPLE

Light sheet microscopy is becoming a popular tool in imaging biological process because of its major advantages of high temporal-spatial resolution, low photobleaching[1]. However, the resolution anisotropy is hard to achieve in traditional two-objective light sheet microscope because of the lower axial resolution.

We proposed a new strategy of solving the problem to obtain an isotropic spatial resolution. Firstly, we use the traditional 2D gaussian light sheet generated by a cylinder lens to illuminate the sample and get a 3D imaging result. Secondly, we generate a saturated central zero-intensity plane light sheet using a spatial light modulator and illuminate the sample to get a 3D result. Finally, the two obtained 3D results are fused with a 3D multiview algorithm or a frequency domain stitching algorithm[2].

2. EXPERIMENT RESULTS



We make the stimulation and experiments using this method. The stimulation result is shown in Fig. 1. The xz section image of the normal light sheet[Fig. 1(a)], the saturated central zero-intensity light sheet[Fig. 1(b)], the deconvolved result of the normal light sheet[Fig. 1(c)] and the fusion result[Fig. 1(d)]. In experiments of fluorescence beads, we have obtained isotropic resolution around 200nm. The further experiments have also been made to prove the availability of this method in live-cell biological imaging.

Figure 1: XZ section of 3D stimulation results. (a) Normal light sheet image. (b) Saturated central zero-intensity light sheet image. (c) Deconvolved normal light sheet image. (d) Fusion result. Scale bar 1 μm .

REFERENCE :

1. Huisken, J., Swoger, J., Del Bene, F., Wittbrodt, J. & Stelzer, E.H. Optical sectioning deep inside live embryos by selective plane illumination microscopy. *Science* 305, 1007–1009 (2004).
2. Zhao G, Zheng C, Kuang C, et al. Nonlinear Focal Modulation Microscopy[J]. *Physical Review Letters*, 2018, 120(19):193901.