Universal Modular Subsystem for Multi-modal Multi-color Extended-depth 3D Imaging & Tracking using Engineered Point Spread Functions

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Conventional 3D light microscopy has allowed for keen insights into biological questions, but is often limited in resolution, imaging depth, or speed. Here, we describe Double Helix (DH) Light Engineering™, which offers a library of engineered point-spread-functions (PSFs) for a variety of 3D imaging applications to overcome many of the limitations of conventional imaging. The DH phase masks (DH-PSF) have been used to extend the imaging capabilities of microscopes for nanometer-scale single molecule imaging [1], light sheet [2], 3D particle tracking [3], and more.

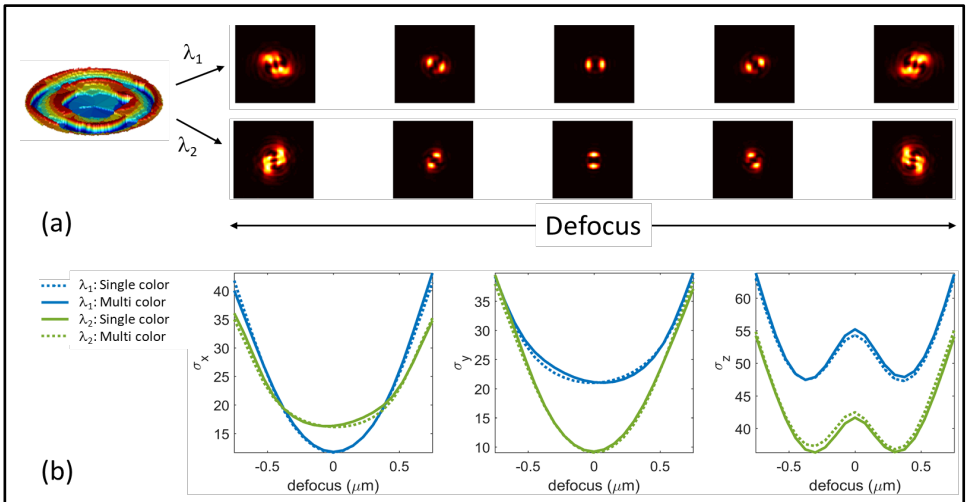

Fig. 1 (a) PSF of the multi-color phase mask for the two design wavelengths. (b) Cramer-Rao Lower Bound (CRLB) comparison of the multi-color mask to the original single-color designs.

Here we describe a recent addition to our library, namely multicolor phase masks (MC-PM) [4] for simultaneous imaging of two or more colors in the same camera sensor area by encoding the spectral information, and thus 3D position, into distinct PSFs for each color. In order to demonstrate this, we designed an MC-PM that encodes DH-PSFs at perpendicular orientations for two selected wavelengths. The resulting PSFs are shown in Fig 1(a). We also compared the Cramér–Rao lower bound (CRLB) of the original DH-PSFs at their respective wavelengths to the CRLB of the PSFs resulting from the MC-PM as shown in Fig 1(b).

References

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