IMAGE-BASED OPTIMIZATION METRIC ANALYSIS FOR LIGHT-SHEET MICROSCOPY

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ABSTRACT

Lightsheet fluorescence microscopy (LSFM) has inherent depth-sectioning where a sheet-shaped beam is formed at the focal plane of the detection objective lens. LSFM has attracted the modern biologist’s attention for its high spatiotemporal resolution and low toxicity for long-term imaging. However, the ultimate barrier for lightsheet microscopy is the sample-induced optical aberration which destroys the resolution of the microscope. Various clearing methods have given a chance to circumvent the optical aberration. And more recently, LSFM using ‘autopilot’ framework has been developed that realized a physically adapting microscope to fix low-order aberrations caused by transparent samples such as zebrafish or mammalian embryos [1]. However, effective methods for correcting aberrations for highly aberrated samples is still under development. Here, we demonstrate a modular adaptive optics (AO) system which can be incorporated into conventional lightsheet microscopes to achieve fast volumetric imaging with high-resolution.

Wavefront sensorless adaptive optics (WS-AO) is an appropriate choice for lightsheet microscopy since an artificial ‘guide star’ does not exist in natural biological objects [2]. The choice of the optimization metric is primarily critical in WS-AO for estimating the correction for the aberrated wavefront. Here, we investigate various image-based metrics for modal approaches and demonstrate an optimized metric for wide-field imaging. The evaluation was carried out through experiment and simulations, and optimization performance was evaluated based on the robustness to different object structures.

REFERENCE