Improving the Imaging Quality and Depth of Biological Objects Using Phase-Only Wavefront Modulation

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Light scattering is the main cause of the impaired image quality and limited probing depth within biological objects when using optical imaging techniques. Object-related wavefront distortions, resulting from light scattering, substantially reduce image quality — these distortions are common to all biological objects and are inherently similar. Increasing imaging depth and image quality is a key challenge in bioimaging to improve diagnostic efficacy and treatment monitoring. Currently, the methods employed individually or in combination to reduce light scattering in biological objects are limited mainly to: 1. optical clearing (reduces the mismatch of refractive indices, low-invasive method); 2. use of the orbital angular momentum of light (scatters minimally in tissue, non-invasive method); 3. use of excitation light sources in the infrared wavelength range (scatters minimally compared to other wavelengths, non-invasive method). So far, the enhancement of image quality is mainly limited to the application of these methods, and other non-invasive approaches are still under development.

The impact of object-related aberrations on image quality can be significantly diminished by using adaptive optical elements, such as spatial light modulators and wavefront sensors, integrated into the optical scheme of microscopes. This method is completely non-invasive and therefore highly promising for measuring and correcting wavefront distortions caused by the sample in real-time. Adaptive optical elements control the wavefront profile of the excitation and/or detection paths and quickly correct it when object-related conditions change, *e.g.* by shifting the focus deeper, where scattering is enhanced.

Here we demonstrate the concept of adaptive optics in microscopy to enhance the image quality and imaging depth in biological objects, utilizing a novel piston micromirror-based spatial light modulator for fast and highly precise phase-only modulation. To further reduce scattering and maximize image quality and imaging depth of biological objects, combining different methods is advantageous.