1558
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Hybrid Open-Top Light-Sheet Microscopy for Multi-Scale 3D Imaging of Cleared and Expanded Tissues

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Recent advances in tissue-clearing protocols greatly reduce optical scattering, aberrations, and background fluorescence, enabling deep-tissue imaging with high resolution and contrast. These approaches have yielded new insights in many fields, including neuroscience, developmental biology, and anatomic pathology [1, 2]. Light-sheet microscopy has emerged as a preferred means for high-resolution volumetric imaging of cleared tissues due to its unrivaled speed and low photobleaching [3, 4]. Many variants of light-sheet microscopes have been developed in recent years by academic researchers and commercial entities to tackle a diverse range of imaging applications. Whereas individual light-sheet systems are well-suited for a subset of cleared-tissue applications, trade-offs are inevitable. In particular, no current light-sheet microscope can satisfy all of the following requirements: (1) user-friendly mounting of multiple specimens with standard holders, (2) compatibility with all current clearing protocols, (3) no fundamental limits on lateral specimen size, (4) a large imaging depth to accommodate intact mouse organs and thick tissue slabs, and (5) broad 'multi-scale' imaging capabilities for time/data-efficient workflows. There would be great value in being able to rapidly screen large (cm-scale) tissue volumes at several-micrometer resolution (i.e., mesoscopic imaging) along with the ability to quantify fine structures within localized regions of interest (mm-scale regions) at sub-micrometer resolution. To address the varied requirements of cleared-tissue light-sheet microscopists, we have designed a open-top light-sheet microscopes [5]. In particular, we have recently designed a 'hybrid' open-top light-sheet microscope that leverages the strengths and overcomes the limitations of our and others' previous systems. These unique capabilities open the door for new light-sheet microscopy applications, including efficient multi-scale imaging workflows in which one or more large specimens must be rapidly screened at low resolution to identify localized regions of interest for quantitative interrogation at the sub-micrometer scale.

References:

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