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The mesoSPIM initiative – open-source light-sheet microscopes for imaging in cleared tissue

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Tissue clearing methods have recently seen a renaissance with a wide variety of clearing approaches now available. In neuroscience, the combination of tissue clearing with light-sheet microscopy is ideal to bridge scales from the µm to cm-level, thus providing a link on the mesoscale for detailed 3D anatomical investigations. To optimally image cleared samples, we set out to design a modular light-sheet microscope that combines extremely simple sample mounting and exchange with large field-of-views (FOV) of 2-22 mm to provide users with overview datasets within minutes. Especially for such large FOVs, common light-sheet microscopes suffer from non-uniform axial resolution due to the varying thickness of the light-sheet. To circumvent this problem, we are using tuneable lenses to shift the excitation beam waist through the sample in synchrony with the rolling shutter of the camera. For whole mouse brains, typical datasets are isotropic (5 µm sampling), small (12-16 GB), and generated quickly (7-8 minutes). Together with standardized quick-exchange sample holders, these features allow fast screening of samples for clearing, imaging, and labelling quality and thus speed up data acquisition considerably.
After creating overview datasets, users can zoom in and acquire high-resolution data.
The microscope has been tested and validated in combination with common clearing methods ranging from hydrogel-based techniques such as CLARITY to organic solvent approaches such as iDISCO – by using a modular design of the imaging chambers, switching between different imaging media can be done in less than a minute. Recently, we have realized four such microscopes at various institutions across Switzerland as part of the mesoSPIM initiative (mesospim.org) – a project aimed at creating a community to accelerate the exchange of tissue clearing and mesoscale imaging expertise. Microscope hard- and software are open-source and we welcome suggestions for improvements.

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