Dual Lattice Lightsheet and its Application in Large Tissue Imaging

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Physical magnification of the specimen itself becomes the norm since the dawn of expansion microscopy (ExM) in 2015 [1]. ExM retains phantoms of fluorescent tags and expands their relative spatial distributions through a polymer system. Low tags density causes fluorescent specimens to bleach easily, forcing microscopists to seek optics designs that can better utilize its limited photon budget.

Combine forces of expansion microscopy (ExM) and lattice lightsheet microscopy (LLSM) [2] in 2019 has proven to be a valuable strategy for millimeter-scale Drosophila whole-brain imaging [3]. With the recent progression in ExM, tissue specimens push the size boundary further from millimeters to centimeters while maintaining near isotropic cellular resolution. However, typical objective working distance has a hard time reaching sub-centimeter distance; those that have extended coverage are limited in their spatial dimension and optical design, causing difficulties to adapt them in an LLSM setup.

Thus, we propose a dual lightsheet solution to mitigate this challenge and demonstrate its feasibility using renal cortex slices. By generating two lattice lightsheet (LLS) from opposing sides, and scan them transversely in the opposite direction, one may span across a 7 mm column tile by acquiring data from both sides (Figure 1).

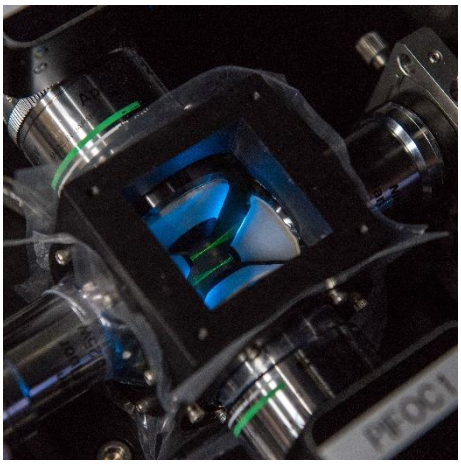

Figure 1. Two LLS illuminated dye in the chamber.

REFERENCES:

1. Chen, F., P.W. Tillberg, and E.S. Boyden, *Expansion microscopy.* Science, 2015. **347**(6221): p. 543.
2. Chen, B.-C., et al., *Lattice light-sheet microscopy: Imaging molecules to embryos at high spatiotemporal resolution.* Science, 2014. **346**(6208): p. 1257998.
3. Gao, R., et al., *Cortical column and whole-brain imaging with molecular contrast and nanoscale resolution.* Science, 2019. **363**(6424): p. eaau8302.