EXPANSION MICROSCOPY & EXPANSION SEQUENCING

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Ideally one would be able to identify, and localize, biomolecules – such as DNA, RNA, and protein – throughout all the cells throughout a tissue, with nanoscale precision. Such mechanistic maps would reveal how epigenomic configurations, transcriptomic programs, and proteomic cascades are configured to mediate cellular as well as organ-scale emergent functions, and pathologies. They would also provide systematic datasets that could enable generation of unbiased hypotheses that could be tested via causal perturbation, for a wide variety of basic and applied biological questions. However, existing methods of imaging are either too slow, or too expensive, or too low-resolution, to do this.

Recently, we discovered that in contrast to the classical use of lenses to magnify images of biological specimens, that physical magnification of cells and tissue is also possible (Chen, Tillberg, & Boyden, 2015). By synthesizing a swellable polymer throughout a cell or tissue, transferring key biological information from the cell or tissue to the polymer, mechanically homogenizing the original biological structure, and then adding water, we can expand such polymer-tissue composites by manyfold (~4.5x linear expansion, in our original paper). This method, which we call expansion microscopy (ExM), enables nanoprecise imaging over large volumes with conventional dyes, and on conventional microscopy equipment.

The original method of ExM is finding increasingly widespread use, in answering questions ranging from invertebrate development to human cancer pathology. We are optimizing ExM along three axes. First, we are improving the physical magnification factor, aiming for 20x linear expansion and beyond, so that it may be possible to resolve single proteins in dense complexes throughout cells and organs. Second, we are devising chemistries to enable the imaging of many different species of biomolecule, e.g. RNA and DNA as well as proteins, in a multiplexed fashion. Third, we are devising a new method for in situ sequencing of nucleic acids throughout all the cells of an intact tissue, by creating new forms of ExM, as well as fluorescent in situ sequencing (FISSEQ; Lee et al., 2014). Using this new technology, which we call expansion sequencing, users can perform the enzymatic sequencing of RNA directly in expanded cells and tissues, enabling systematic cell type and cell state classification in health and disease.

In this way, we aim to develop a simple toolbox that, ultimately, will enable the identification of essentially unlimited kinds of biomolecules, throughout entire cells and tissues, with nanoprecise spatial resolution.

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