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## Abstract: 97O

### 3D super-resolution imaging of whole nuclear lamina

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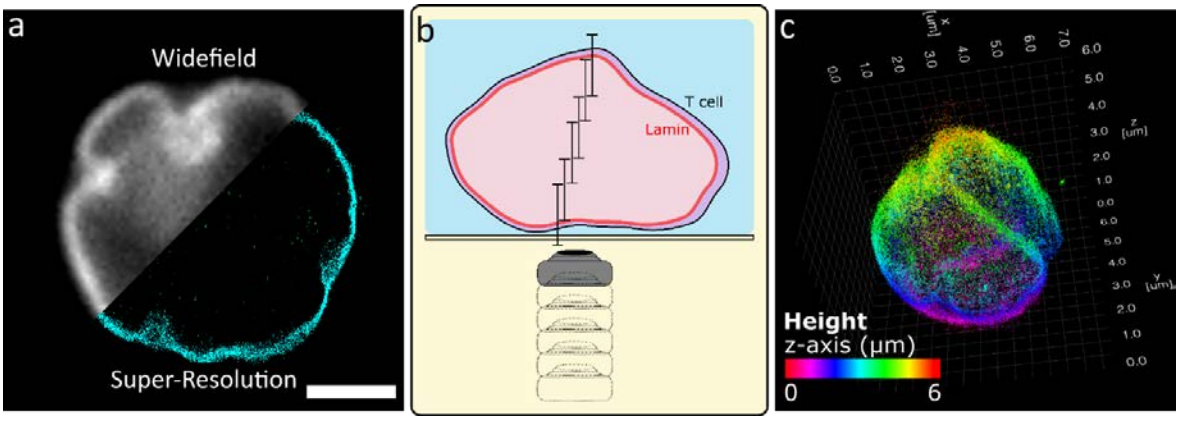
Fluorescence microscopy in biology provides specificity to observe cellular components and interactions *in situ*. The spatial resolutions, however, are limited to ~200 nm due to the diffraction of light, perturbing visualization of biomolecules at the nanoscale. Single molecule localization microscopy (SMLM) achieves imaging resolutions as good as 20 nm laterally and 50 nm axially[1]. A conventional method for 3D SMLM is to induce astigmatism such that single molecule emissions become laterally elongated based on their axial position[2]. However, this approach only provides an axial range of ~1 µm which is unsuited to capture cellular structures and protein distributions that span the entire cellular volume, typically several microns wide in each dimension. As such, 3D visualization of whole cells in SMLM resolution remains challenging.

We have applied single molecule astigmatism with multiplane imaging to visualize whole nuclear lamina; the protein network adjacent to the inner nuclear membrane[3] (Figure 1). We demonstrated 3D SMLM for an axial range up to 8 µm to image nuclear lamina in COS-7 cells and T cells, and quantified nuclear surface area and volume using 3D convex hull fitting. The super-resolution detail revealed membrane features such as folds, blebs and invaginations within the context of the whole nucleus 3D image. For T cells, the nuclear lamina can be used as a reference structure to quantify the spatial distribution of the chromatin landscape in response to cell differentiation.

**Figure 1.** (a) SMLM enables super-resolved detail of nuclear lamina in 2D. Scale bar = 2 µm. (b)




Schematic demonstrating multiplane imaging with each step affording 1 µm of *z* observation. (c) Combined 3D coordinates to reveal whole nuclear lamina in super-resolution, colour coded for height.

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