Enhanced FIB-SEM Technology: Large Volume Imaging at Fine Resolutions

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Focused Ion Beam Scanning Electron Microscopy (FIB-SEM) offers unique benefits for volume imaging, such as isotropic high-resolution (< 10 nm) and robust image alignment. However, deficiencies of conventional systems in imaging speed and duration cap the maximum possible image volume. We transformed FIB-SEM from a lab tool lacking long term reliability to a robust imaging platform with 100% effective reliability: capable of years of continuous imaging without defects in the final image stack. As a result, we have expanded the imageable volume by more than four orders of magnitude from $10^3 \, \mu m^3$ to $3 \times 10^7 \, \mu m^3$ while maintaining isotropic 8-nm voxels. Moreover, by trading off against imaging speed, the system can achieve even higher resolutions at 4-nm voxels [1, 2].

The expanded volumes open up a new regime in scientific learning, where nano-scale resolution coupled with meso and even macro scale volumes enable fruitful discoveries. The largest connectome to date (https://neuprint.janelia.org) has been generated at 8-nm voxels [3]. A reference library of whole cells and tissues imaged at 4-nm voxels has also been established: the higher resolution further improves the interpretation of otherwise ambiguous details, and the open access (https://openorganelle.janelia.org) inspires wider research communities to explore comprehensive cellular architecture [4].

In this presentation, I will discuss the technological advances of enhanced FIB-SEM, followed by a variety of biological examples including Drosophila brain, mouse liver, and mammalian cells to illustrate the power of fine isotropic resolution coupled with large imaging volume.

References


