

## Volume Imaging: From HeLa Cells to the Human Nervous System

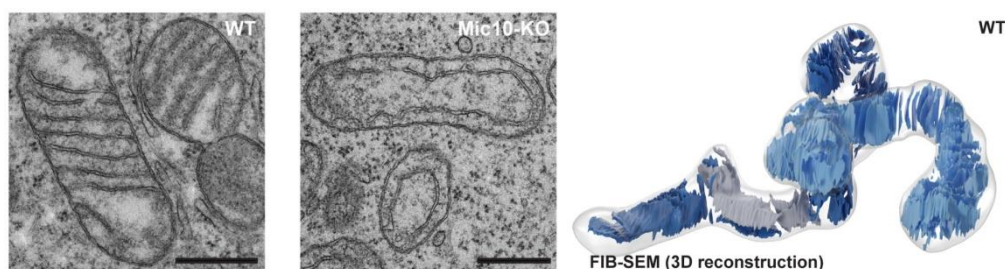
Anna M. Steyer<sup>1\*</sup>

<sup>1</sup>European Molecular Biology Laboratory, Imaging Centre, Heidelberg, Germany.

\* Corresponding author: [steyer@embl.de](mailto:steyer@embl.de)

Focused ion beam scanning electron microscopy (FIB-SEM) has emerged as a flexible method that enables semi-automated volume acquisition at the ultrastructural level. New questions can be approached and research questions that were so far only addressed in 2D can now be revisited at nanometer resolution. Very important steps besides the actual data acquisition include a suitable sample preparation and the right set of tools to do data processing and model building afterwards. More recently volume imaging under native conditions at cryogenic temperatures has been gaining importance.

Here I would like to show examples of volume electron microscopy using the focused ion beam scanning electron microscope at room temperature from mitochondrial structures in single cells to sperm cells in *C. elegans* as well as mouse nervous tissue and even human nerves. Different projects and workflows will be discussed including various sample preparation strategies (chemical fixation as well as high-pressure freezing) and image analysis pipelines including different segmentation methods.



**Figure 1.** Adapted from Stephan *et al.* 2020 [1]. Mitochondria in wildtype and knockout cells (Mic 10 KO) in transmission electron microscopy image (left and middle). 3D reconstruction of cristae morphology and rendering (right). Scale bars 500 nm.



**Figure 2.** Semi-automated segmentation of axons in the optic nerve of a wildtype mouse based on a focused ion beam scanning electron microscopic dataset (left and middle, scale bar 500 nm). 3D rendering of the 50 segmented axons (right, scale bar 4  $\mu$ m).

### References:

- [1] T Stephan *et al.*, the EMBO Journal **39** (2020), doi/full/10.15252/embo.2019104105