Plasma FIB Applications in Life Science: From Large Volumes to Cryo-lamella Preparation

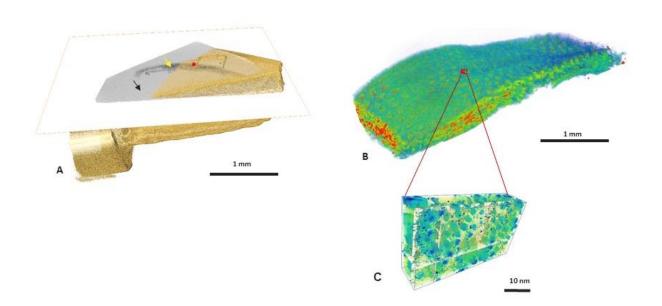
Daniela Slamková^{1*}

The Thermo ScientificTM Helios HydraTM Plasma FIB-SEM with the unique capability to deliver four ion species (Xe, Ar, O, or N) as the ion beam source is a novel technology in the life sciences space and can be a powerful method for multiscale analysis of biological samples.

The study of complex cellular processes often requires multiple levels of structural details and possibilitis for volume imaging or lamella preparation.

The ability to choose between different ions makes it possible to obtain curtain-free surfaces for a wide variety of resin-embedded biological samples as well as optimize and develop new strategies for cryolammela preparation and 3D volume-contextual imaging of bulk hydrated samples.

The Helios Hydra™ offers a new level of application flexibility for addressing scientific questions. It can bridge multiscale 3D to 3D correlation for precise targeting of the region of interest from uCT data to 3D Plasma FIB-SEM tomography (Figure 1). Its Spin Mill capability offers a unique alternative approach to 3D analysis, by planar milling of large-areas (up to 1 mm in diameter) (Figure 2), especially beneficial for accessing and investigating sparse regions of interest The multi-ion Plasma FIB is a powerful tool for analysis of cryo-immobilised, hydrated samples where different ions can be used for automated cryo-lamella preparation (Figure 3).





¹ Thermo Fisher Scientific, Vlastimila Pecha 1282/12, Brno, Czech republic,

^{*} Corresponding author: daniela.slamkova@thermofisher.com

Figure 1. A) 3D uCT data of a marine sponge bulk sample with dimensions 1.2 mm x 1.2 mm x 3.3 mm. The sponge tissue (yellow arrow) is embedded in the resin (black arrow). The red dot indicates the targeted area of interest. B) uCT data used for 3D to 3D correlation was collected on HeliScanTM MicroCT. Voxel size is 0.42 μm. C) Auto Slice & View data of the targeted area was acquired on Helios Hydra. Voxel size is 8 x 8 x 10 nm. Total 3D volume size is 55 x 31 x 10 μm³.

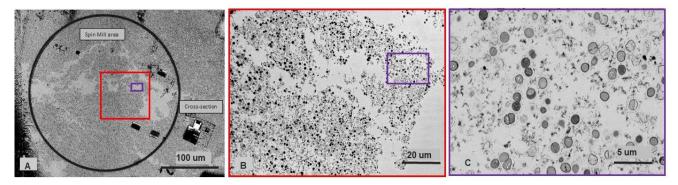


Figure 2. Accessing large areas with Spin Mill. *Staphylococcus* infected by phage sample, embedded in Hard plus resin. A) Large area milling by Spin Mill is applied to the area defined by the circular marker (diameter 450 μ m). B) First area of interest, field of view 100 μ m, pixel size 16 nm. C) Second area of interest, field of view 20 μ m, pixel size 4 nm.

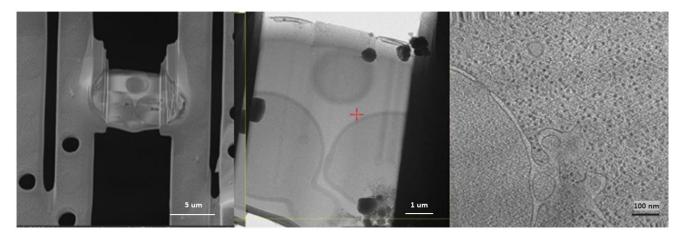


Figure 3. Lamella of Yeast plunge frozen on a grid prepared with Hydra, using Xe as a primary ion species. AutoTEM Cryo 2.4 was used for automated lamellae preparation. Thickness of lamella 150 nm. Krios G4 was used for TEM evaluation, using 300 kV, Selectris energy filter and Falcon 4 direct electron detector.