1250
doi:10.1017/S1431927622005189
Microsc. Microanal. 28 (Suppl 1), 2022

A Next Generation Cryo-FIB Microscope for High-Throughput Cryo-Electron Tomography

Alexander Rigort[1*], Abhay Kotecha[2], Steve Reyntjens[2] and John Mitchels[1]

[1.] Thermo Fisher Scientific s.r.o., Brno, Czech Republic
[2.] Thermo Fisher Scientific, Eindhoven, The Netherlands
* Corresponding author: alexander.rigort@thermofisher.com

Dual-beam cryo-FIB/SEM microscopes have become increasingly indispensable tools for cryo-electron tomography over the past decade. Thermo Fisher Scientific has played a leading role in the development together with pioneering scientific groups in the field, setting standards for this still young class of cryo-dedicated instruments. Microscopes such as the Aquilos 2 are now in their second generation and, with new automation routines, along with integrated cryo-lift-out and fluorescence microscopy options, have steadily expanded the application range of the cryo-FIB and are already routinely used in many laboratories for cryo-lamella preparation. With the advent of cryo-FIB instrumentation, as well as improved cryo-TEM data collection schemes utilizing faster direct electron detectors and modern energy filters, completely new possibilities have opened for cryo-electron tomography [1], as evidenced by the steady increase in the number of publications in which new cell and structural biology questions are elucidated using this method [2].

In this contribution, we will present a further step in cryo-electron microscope technology development. Analogous to the development of the automated Krios cryo-TEM approximately 15 years ago, we will discuss a new next generation cryo-FIB system. The new instrument is dedicated to production of cryo-lamellae for the cryo-electron tomography workflow, and features a state-of-the-art plasma ion source, an automated sample loading system, integrated fluorescence microscopy, and the lowest contamination rates, allowing preparation of cryo-lamellae at a higher level of productivity than ever before and thus producing the best possible samples for tomography.

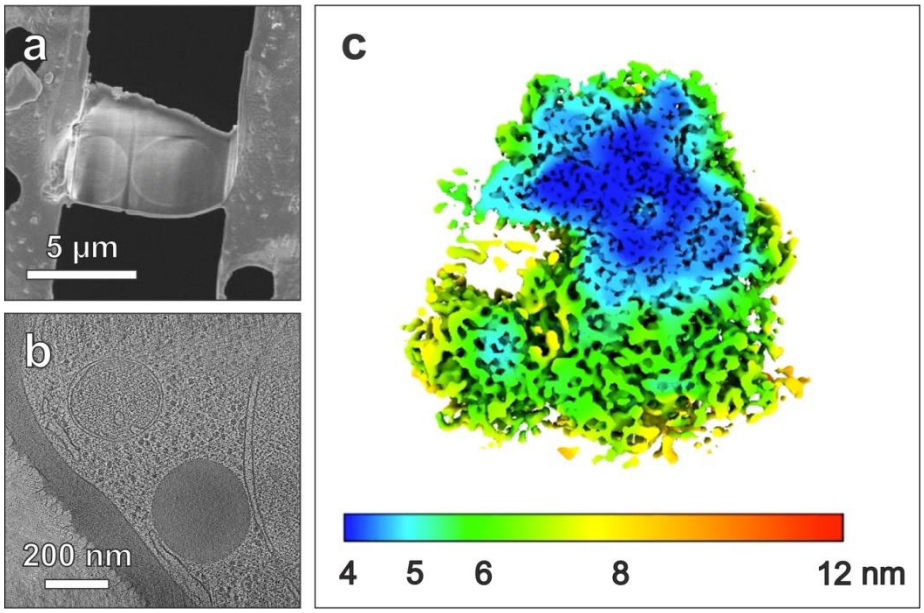

Figure 1. (a) *S. cerevisiae* cryo-lamella produced by Xenon ion milling in the next generation cryo-Plasma-FIB system. (b) Slice from a tomographic reconstruction. (c) Corresponding sub-tomogram averaging yielding the 80S ribosome at a global resolution of 6.5Å from 7000 particles. The 60S large subunit reaches 4.5 Å local resolution.

References:

[1] D Kwon, Nature 598, 558-560 (2021). doi.org/10.1038/d41586-021-02904-w
[2] Z Wang et al, Cell 184, 2135–2150.e1–e5, April 15, (2021). doi.org/10.1016/j.cell.2021.02.047

https://doi.org/10.1017/S1431927622005189 Published online by Cambridge University Press