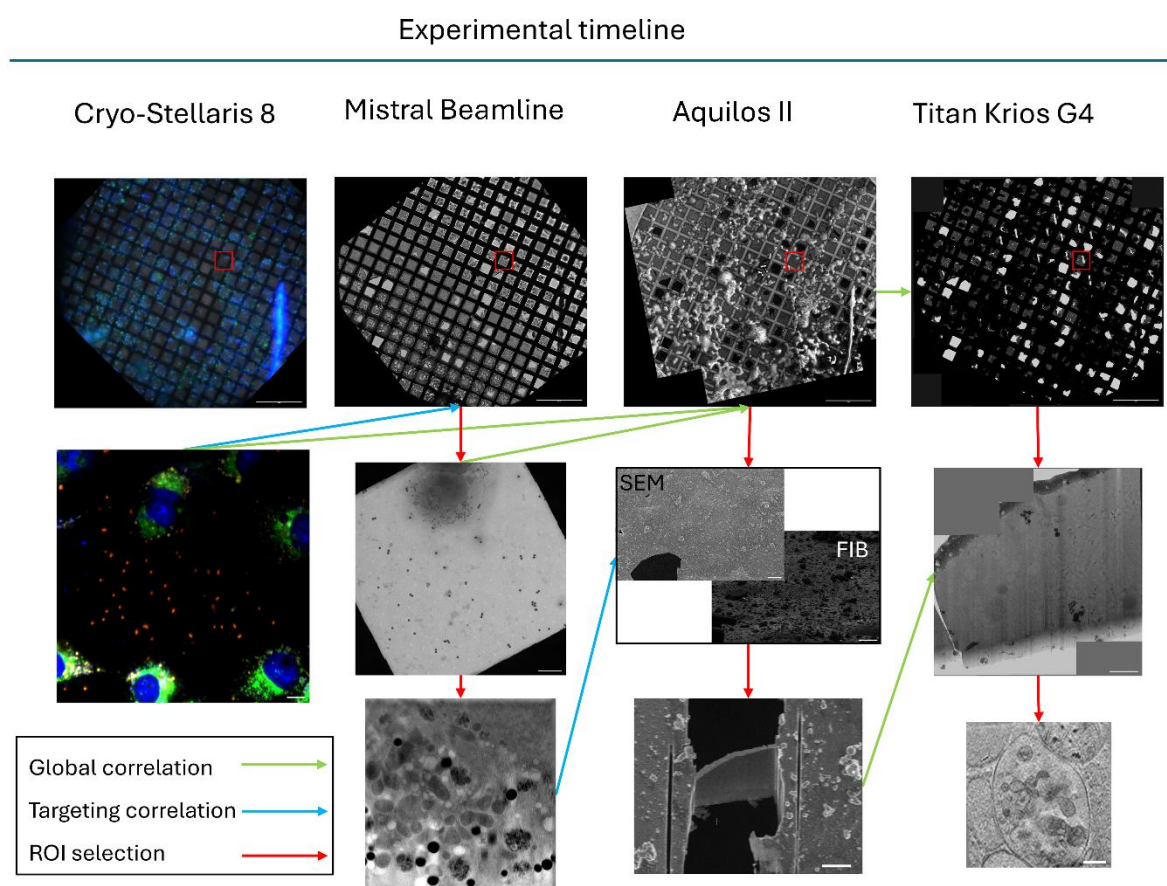
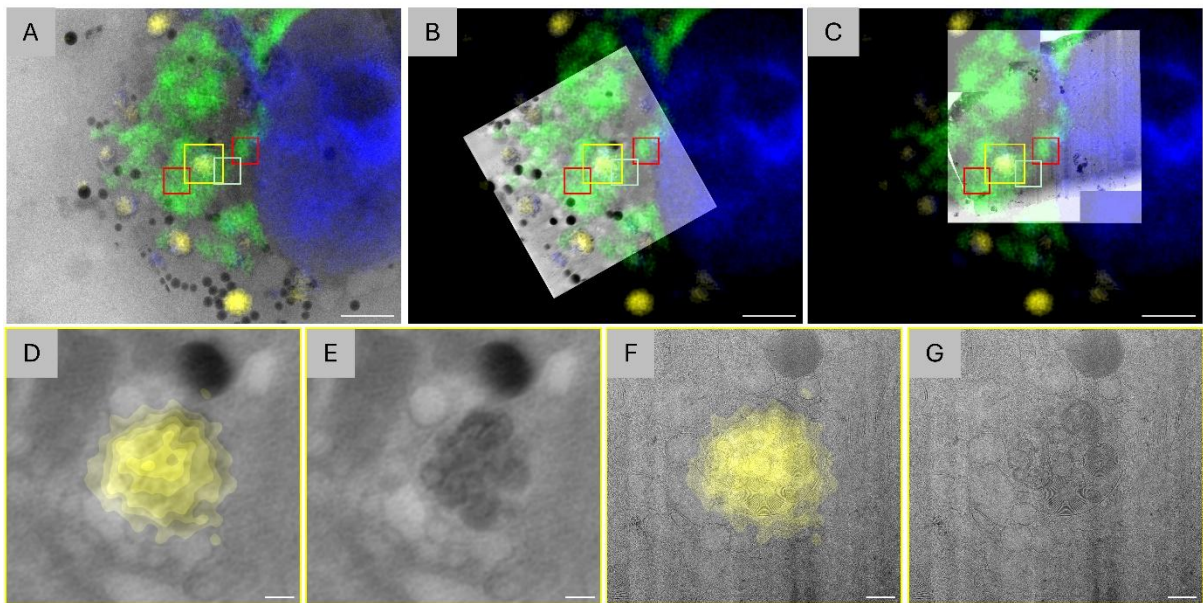


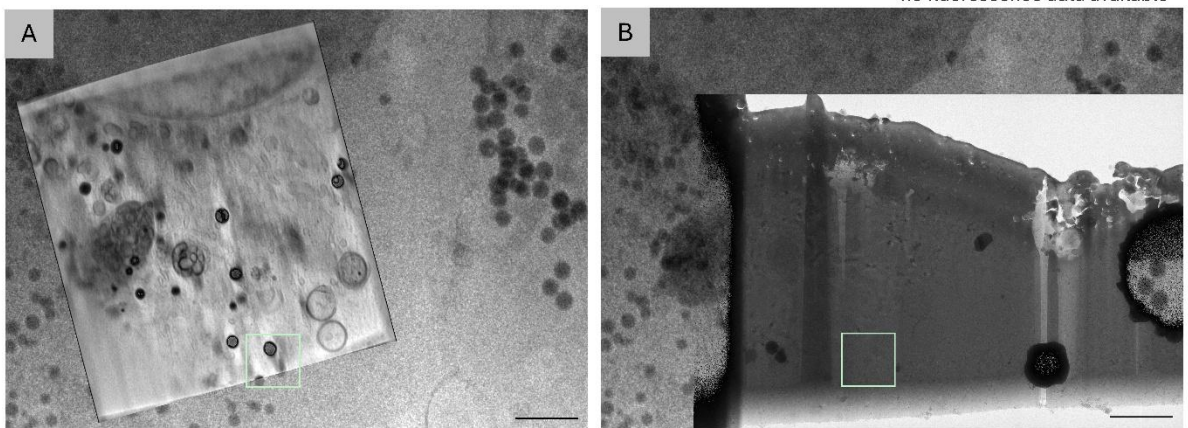


Supplementary Figure 1: Experimental timeline and correlations that have been performed between the different imaging modalities - cryo-confocal (Leica Cryo-Stellaris 8), cryo-SXT (Mistral Beamline), cryo-FIB-SEM (Thermo Fisher Aquilos 2) and cryo-TEM (Titan Krios G4). Green arrows highlight global correlations, used mostly for navigation purposes. Blue arrows show the targeting correlations that needed to be accurate for targeting the ROI. Red arrows show the manual ROI selection process, guided by the global correlations. Scale bars : top row : 50 μm ; middle row, cryo-stellaris and Mistral : 10 μm , Aquilos : 20 μm , Titan : 2 μm ; bottom row, Mistral : 2 μm , Aquilos 5 μm , Titan : 200 nm.

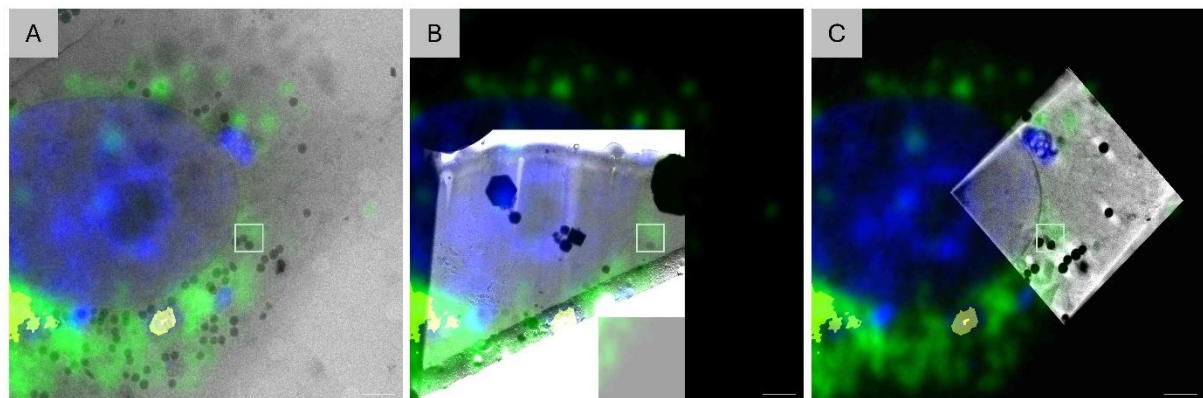
P1



P2*

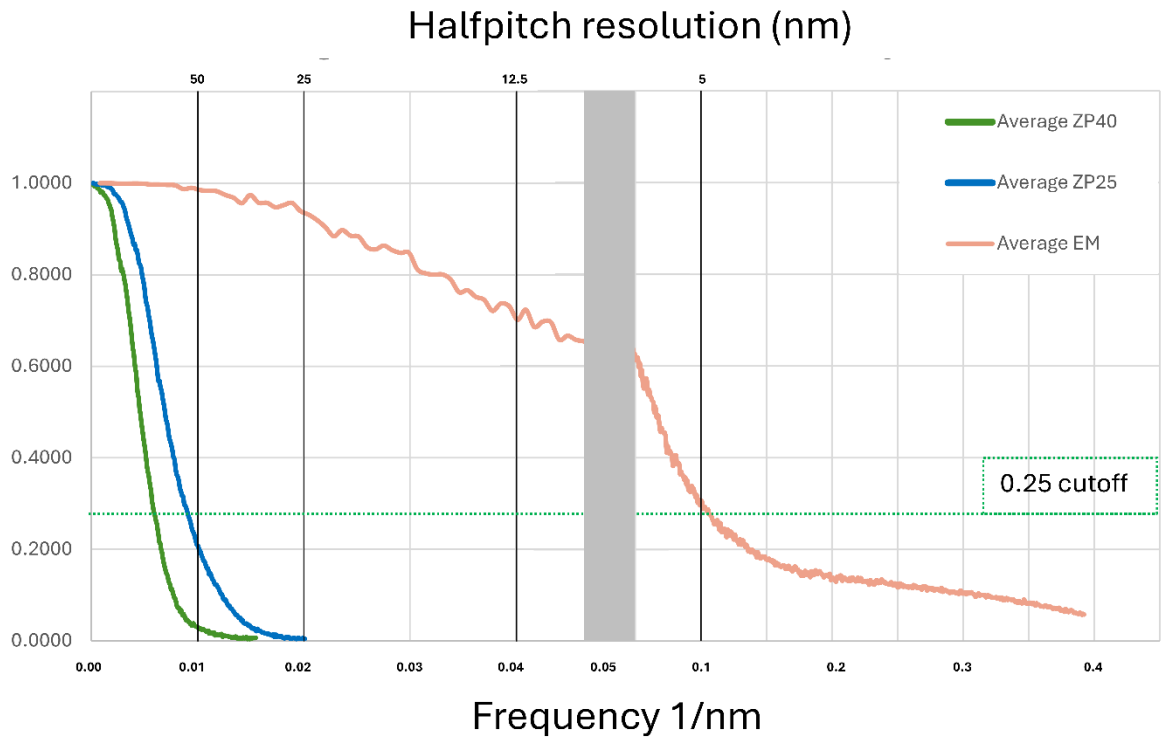


P3

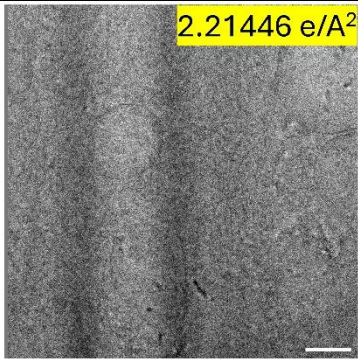
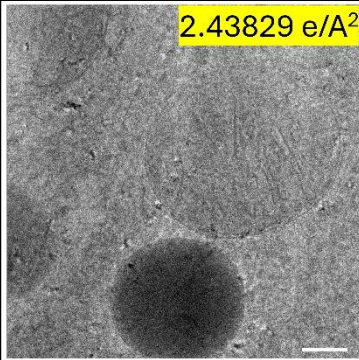
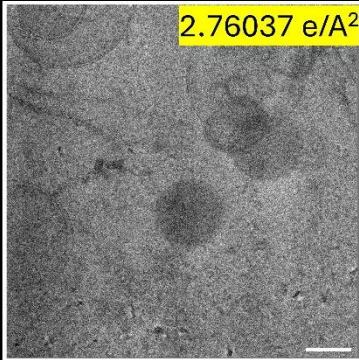
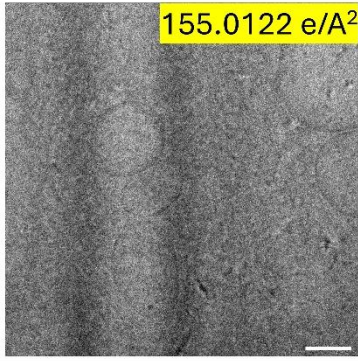
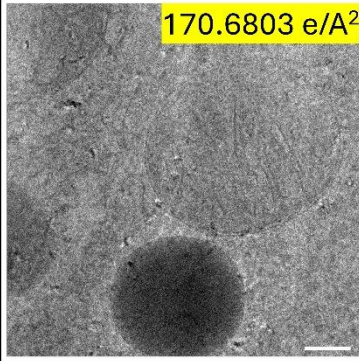
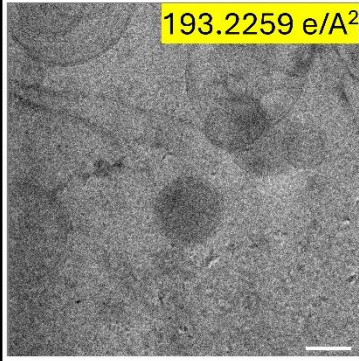


Supplementary Figure 2: Additional examples of areas with partial overlap of the cryo-SXT tomogram and a lamella. P1 corresponds to the correlation shown in Figure 2, highlighting the 3-modalities-correlation that shows an acidic compartment (stained by lysotracker) that can be identified both in the cryo-SXT tomogram (panel D and E) and in the projection image from the cryo-EM lamella (panel F and G). P2 and P3 show areas from two other grids with partially overlapping cryo-

SXT/cryo-EM areas, where tomograms were collected outside the overlapping regions. The red squares in P1 indicate the tiltseries areas shown in Figure 2. The yellow square in P1 marks the acidic organelle location, seen in panels D-G. The light green squares in P1, P2 and P3 indicate the areas used for the no-tilt acquisition to measure the allowed exposure dose show in the supplementary figure 4. Scale bars : P1-ABC, P2-AB, P3-ABC : 2 μm ; P1-D-G : 200 nm.



Supplementary Figure 3: Averaged Fourier Shell Correlation (FSC) curves. The curves show the achieved resolution for the cryo-ET and the cryo-SXT data, the latter acquired with either the 40 or 25 nm Fresnel Zone Plate (ZP). Measured resolutions are: ZP 40 nm ~80 nm ; ZP 25 nm ~53 nm ; EM ~5 nm. The raw data can be found in the supplementary data 1 excel file.

	P1	P2	P3
Image 1	 2.21446 e/A ²	 2.43829 e/A ²	 2.76037 e/A ²
Image 70	 155.0122 e/A ²	 170.6803 e/A ²	 193.2259 e/A ²

Supplementary Figure 4: Exposure Dose test. First and last images from a 70-image stack acquired from 3 areas that were previously imaged by cryo-SXT, highlighting the lack of visible radiation damage. P1, P2 and P3 correspond to the same lamellae of supplementary figure 2, each coming from a different grid. Scalebars: 200 nm.