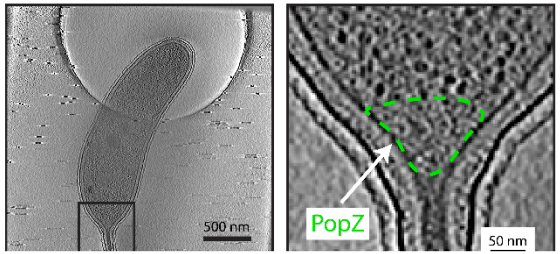
Correlative Cryogenic Super-Resolution Fluorescence and Electron Tomography

Moerner Lab

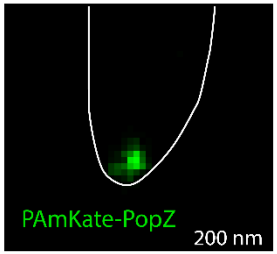
Motivation: Cryogenic electron tomography (CET) and super-resolution fluorescence (SR) are two powerful methods for the observation of subcellular organization, but both methods suffer from unique limitations. Specifically, there are no specific and non-perturbative labelling methods for CET and even high-resolution SR reconstructions lack detailed cellular context.

Result: Here we demonstrate accurate and precise single-molecule fluorescence localizations correlated with CET. This correlation identifies specifically labeled proteins of interest within a high-resolution context.

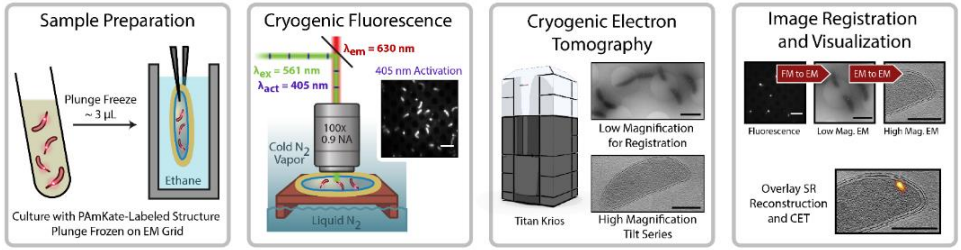
Cryogenic Electron Tomography



Room Temperature SR



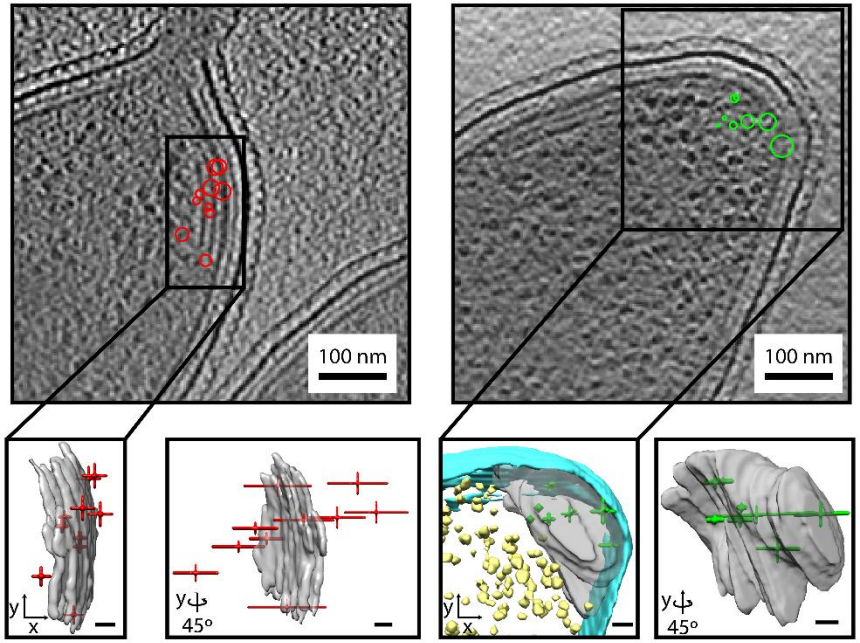
CIASM Workflow



CIASM Results For Two Proteins of Interest

McpA-PAmKate Localizations

PopZ-PAmKate Localizations



Highlights:

- Mean localization precision 9 nm
- Registration error ~30 nm
- Compatible with any protein fused to PAmKate fluorescent protein label
- Workflow “Correlative Imaging by Annotation with Single Molecules” (CIASM)

Dahlberg, P.D., Saurabh, S., Sartor, A.M., Wang, J., Mitchell, P.G., Chiu, W., Shapiro, L. and Moerner, W.E., *PNAS* **117**, 13937 (2020).