Single-protein detection in crowded molecular environments in cryo-EM images

Title	Single-protein detection in crowded molecular environments in cryo-EM images
Publication Type	Journal Article
Year of Publication	2017
Authors	Rickgauer, JP, Grigorieff N, Denk W
Refereed Designation	Refereed
Journal	eLife
Volume	6
Issue	e25648
Pagination	1-22
Abstract	We present an approach to study macromolecular assemblies by detecting component proteins' characteristic high-resolution projection patterns, calculated from their known 3D structures, in single electron cryo-micrographs. Our method detects single apoferritin molecules in vitreous ice with high specificity and determines their orientation and location precisely. Simulations show that high spatial-frequency information and – in the presence of protein background – pre-whitening are essential for optimal detection, in particular for images taken far from focus. Experimentally, we could detect small viral RNA polymerase molecules, distributed randomly among binding locations, inside rotavirus particles. Based on the currently attainable image quality, we estimate a threshold for detection that is 150 kDa in ice and of 300 kDa in 100 nm tissue sections.
DOI	10.7554/eLife.25648

Rickgauer_eLife2017.pdf 3.75 MB

Source URL: http://grigoriefflab.janelia.org/node/5618