High-speed super-resolution structured illumination imaging of intracellular organelles

Dong Li[1,2]

[1]National Laboratory of Biomacromolecules, CAS Center for Excellence in Biomacromolecules, Institute of Biophysics, Chinese Academy of Sciences, Beijing, 100101, China. [2]College of Life Sciences, University of Chinese Academy of Sciences, Beijing, 100049, China.

Email: lidong@ibp.ac.cn

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In eukaryotic cell, various organelles and cytoskeletons are very dynamic, yet highly organized to orchestrate complex cellular functions. Visualizing these interactions requires noninvasive, long duration imaging of the intracellular environment at high spatiotemporal resolution. However, the tradeoffs between spatial and temporal resolution, and low phototoxicity/photobleaching in current super-resolution imaging techniques prevent biologists from accurately characterizing these dynamic processes, and discovering new interaction forms. To achieve these normally opposing goals, in this talk I will discuss our developments in grazing incidence structured illumination microscopy (GI-SIM) [1], and lattice light sheet microscopy (LLSM). GI-SIM improves ~10 fold deeper penetration depth and ~10 fold higher signal strength than TIRF mode. By integrating GI with SIM, GI-SIM practically achieves the imaging performance of 97 nm resolution, 266 Hz frame rate, hundreds to thousands of time points, and multi-color imaging. Moreover, we applied multi-color GI-SIM and LLSM to characterize the dynamic interactions between different organelles. Finally, we are pushing GI-SIM to higher resolution level by using of our previously developed patterned activation nonlinear SIM (PA NL-SIM) method [2].

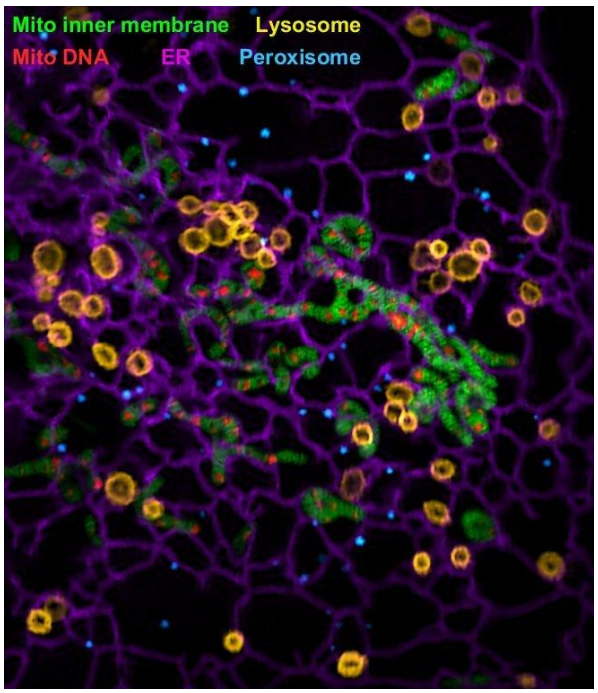

Figure 1. Representative multi-color GI-SIM live cell image of different organelles.

Reference

[1] Y. Guo, *et al.*, "Visualizing intracellular organelle and cytoskeletal interactions at nanoscale resolution on millisecond time scales", Cell. **175**, 1430-1442 (2018).

[2] D. Li, *et al.*, "Extended-resolution structured illumination imaging of endocytic and cytoskeletal dynamics", Science. **349**, aab3500 (2015).