Improving resolution by cryogenic and interferometric imaging

Wei Ji, Xiaojun Xu, Yanhong Xue, Buyun Tian, Lusheng Gu, Yuanyuan Li, Shuwen Zhang, Tao Xu
Institute of Biophysics, Chinese Academy of Sciences
Datun road 15, Chaoyang district, Beijing 100101, China
E-mail: jiwei@ibp.ac.cn

KEY WORDS: Single molecule localization, cryogenic, interferometric, Superresolution

Remarkable progress in Single molecule localization microscopy (SMLM) has been made in the past decade. The localization precision of SMLM is mainly dependent on the number of detected photons, therefore tremendous efforts have been invested to increase photon budget by specially designed fluorophores and anti-bleaching agents. Here we developed cryogenic and interferometric single molecule localization methods which exhibit excellent localization precision performances compared to conventional SMLMs. We built an ultra-stable super-resolution cryo-FM and demonstrated the super-resolution imaging capability of this system. The results suggest that our system is particularly suitable for single molecule localization imaging and cryogenic super-resolution correlative light and electron microscopy (csCLEM)[1,2]. We also introduced an interferometric SMLM named Repetitive Optical Selective Exposure (ROSE), with which a fluorescence molecule is located by the intensities of multiple excitation patterns of an interference fringe, providing around two-fold improvement in the localization precision compared to the conventional centroid fitting method at the same photon budget. We demonstrate this technique by resolving a nanostructure down to 5 nm[3].

Reference

[1] Liu, B., Xue, Y., Zhao, W., Chen, Y., Fan, C., Gu, L., Zhang, Y., Zhang, X., Sun, L., Huang, X., Ding, W., Sun, F., **Ji, W.**, and Xu, T. Three-dimensional super-resolution protein localization correlated with vitrified cellular context. ***Sci Rep*** 513017, 13017 (2015).

[2] Xu, X., Xue, Y., Tian, B., Gu, L., Li, W., **Ji, W**., Xu, T.. Ultra-stable super-resolution fluorescence cryo-microscopy for correlative light and electron cryo-microscopy. ***Science China Life Sciences***. doi: 10.1007/s11427-018-9380-3 (2018).

[3] Gu, L., Li, Y., Zhang, S., Xue, Y., Li, W., Li, D., Xu, T., **Ji, W**. Molecular resolution imaging by repetitive optical selective exposure. ***Nature Methods****, 2019 Nov;16(11):1114-1118.*