POLARIZED STRUCTURED ILLUMINATION MICROSCOPY

Karl Zhanghao, Xingye Chen, Qionghai Dai, Peng Xi
Department of Biomedical Engineering, College of Engineering
Peking University, Beijing, China
E-mail: karl.hao.zhang@pku.edu.cn

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Fluorescent dipoles reflect the spatial orientation of the fluorophores, which indicates structural information of the targeted proteins. Imaging of fluorescent dipoles has been widely applied in structural research of cell membranes, biological filaments and macromolecules [1]. However, it suffers from the Abbe’s diffraction limit, which deteriorates the imaging accuracy of both position and orientation of the fluorescent dipoles. Various super-resolution techniques have therefore been developed to provide resolution beyond the diffraction barrier, either by single molecular imaging [2] or polarization demodulation [3]. Unfortunately, the single molecular method takes tens of minutes for acquisition and couldn’t be applied to live cell imaging, while the polarization demodulation method has limited resolution of ~150 nm.

Structured illumination microscopy (SIM) has been demonstrated to be very compatible with live-cell imaging due to fast imaging speed and low photo-toxicity. Here we report polarized SIM (pSIM) [4] which could be directly performed on commercial SIM systems. When the fluorescent anisotropic specimen is illuminated by a linearly polarized light, structured illumination happens in the dimension of dipole orientation, with polarization modulation varying the phases. To generate high-contrast interferometric fringes, SIM rotates excitation polarization as the pattern direction changes, which expands both the spatial and orientational observable region in reciprocal space. With a novel algorithm, super-resolution imaging of fluorescent dipoles could be obtained by pSIM, which maintains all the advantages of SIM, accompanied by easy access to commercial equipment. We believe pSIM would become a widely applied tool in studying the molecular dynamics of biological structures.