RECENT DEVELOPMENT IN BLINKING-BASED NANOSCOPY TOWARD HIGH SPATIOTEMPORAL LIVE CELL IMAGING

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Emerging blinking-based super-resolution techniques (Nanoscopy) have been through rapid developments over the past few years. Among them, representative methods that are based on fluorescence fluctuation of probes, such as super-resolution optical fluctuation imaging (SOFI) and Bayesian nanoscopy (3B), have demonstrated several advantages: (i) improved temporal resolution for analysis of high density blinking events; (ii) low phototoxicity for low light intensity; (iii) Economy friendly using lamp-based wide-field microscope, etc.

Here we report our recent development in blinking-based nanoscopy. For SOFI nanoscopy, several studies were carried out: (1) A joint-tagging quantum dots-labeling methods was developed to improve the spatiotemporal resolution and labeling density [1]. (2) Combined with spinning disk confocal microscopy, multi-modality super-resolution microscopy including SOFI and BaLM was achieved for three-dimensional imaging [2]. (3) A new high brightness, high on/off contrast, strong photostability reversibly switchable fluorescent protein, Skylan-S, was developed to achieve high spatiotemporal live-cell SOFI imaging [3]. (4) A qualitative comparison between sCMOS and EMCCD in SOFI imaging was conducted and the study indicates that sCMOS has superior performance in SOFI both in spatial and temporal resolutions [4]. For 3B nanoscopy, we studied the dynamic process of RNA Pol II clusters in live cell nucleus. For the first time, the dynamic processes of individual Pol II clusters, including both cluster assembly and disassembly are directly observed. We revealed the asynchronous nature of Pol II cluster. In addition, our results support the on-demand model for transcription factory formation [5].