

## Multi-modality Super-resolution Optical Imaging of Living System

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Various super-resolution (SR) techniques aiming at breaking the diffraction barrier have sprung up in past decades (1), such as stimulated emission depletion microscopy (STED), reversible optically linear fluorescence transitions microscopy (RESOLFT), Structured illumination microscopy (SIM), photoactivated localization microscopy (PALM) and stochastic optical reconstruction microscopy (STORM), Superresolution Optical Fluctuation Imaging (SOFI), etc. Among these SR techniques, switchable fluorophores play a crucial impact on ensuring admirable implementation (2).

Recently, we developed a series of unique beneficial reversibly switchable fluorescent proteins, mGeos2-IES. Taking advantage of their innate optical and biological merits, such as monomeric structure, high brightness, high photostability and excellent fusion for biological imaging in living cells, we successfully achieved SOFI and PALM multi-modality SR images of U2OS living cells expressing actin fused with mGeos2-IES.

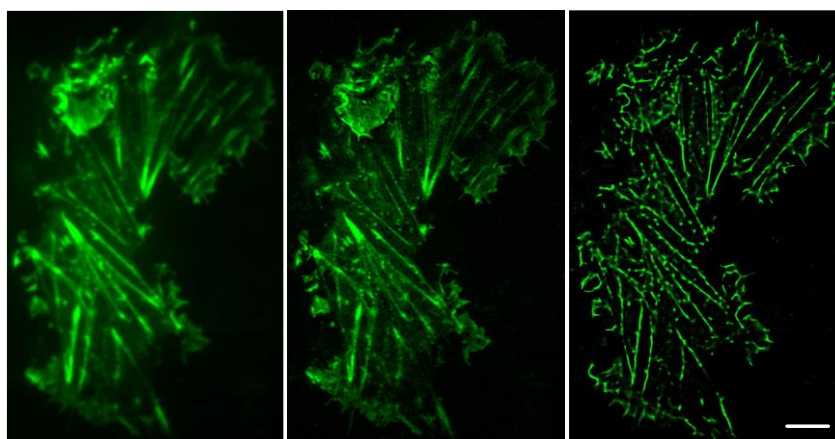


Figure 1. Fluorescence and corresponding 2nd SOFI, PALM images in U2OS living cells expressing actin fused with mGeos2-IES. (a) Average of 1000 original fluorescence frames. (b) 2<sup>nd</sup> order SOFI and (c) PALM image, respectively. (d, e, f) are the corresponding area of (a, b, c) white label region. The SOFI image was obtained by the analysis of the 1000 frames under low 488 nm excitation, then 25000 frames were imaged for PALM under both high 488 nm and low 405 nm excitation, when single molecule can be resolved. Scale bar: 5 $\mu$ m.

### References:

1. Ding Y, Xi P, & Ren Q (2011) Hacking the optical diffraction limit: Review on recent developments of fluorescence nanoscopy. *Chinese Science Bulletin* 56(18):1857-1876.
2. Chang H, *et al.* (2012) A unique series of reversibly switchable fluorescent proteins with beneficial properties for various applications. *Proceedings of the National Academy of Sciences* 109(12):4455-4460.