TauSTED: The new lifetime-based approach to STED in the STELLARIS confocal platform

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KEYWORDS: STED, TauSTED, TauSense, FLIM, Super-resolution microscopy, Nanoscopy

Super-resolution microscopy techniques have emerged as powerful tools for life science research to extract detailed molecular information in the cellular context beyond the diffraction limit. Among the different technologies, stimulated emission depletion (STED) process induces changes in the spatial fluorescence lifetime distribution, therefore the combination of STED and fluorescence lifetime-derived information can be applied to enhance applications requiring nanoscale resolution in living cells. Pioneering developments using lifetime-derived information gave rise to gated STED (1) and recent implementations based on FLIM phasor analysis (2, 3). The underlying principle is the encoding/decoding of the nanoscale spatial distribution of fluorophores from the fluorescence lifetime information. The original work on this concept with FLIM phasors gave rise to the SPLIT (Separation of Photons by LIfetime Tuning) method, increasing the spatial resolution and eliminating uncorrelated background. Inspired by this approach, we developed a new functionality called TauSTED, that enables to perform STED in gentle conditions keeping the desired resolution, eliminate the uncorrelated background contribution, and perform STED co-localization studies.

In this talk we will show how TauSTED has been implemented on the STELLARIS confocal microscopes as a TauSense tool. We will explain how the TauSTED approach delivers cutting-edge resolution and image quality at low light dose, key to study nanoscale dynamics of cellular processes.

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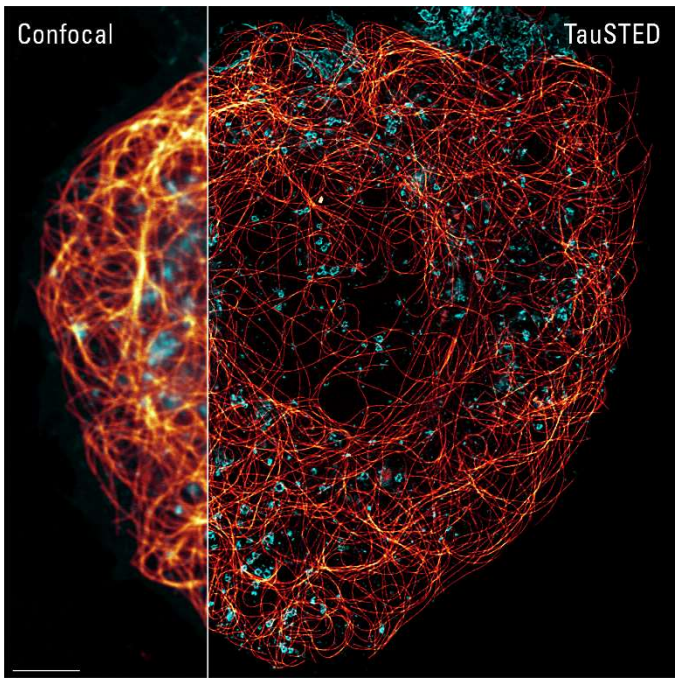

Live-cell TauSTED 775 on cells labeled with SiR-tubulin (glow) and WGA-CF594 (cyan). Scale bar: 5 µm. SiR-tubulin is available from Spirochrome. WGA-CF594 courtesy of Biotium, Inc.