SPEEDYTRACK: MICROSECOND WIDE-FIELD SINGLE-MOLECULE TRACKING

Tech ID: 34012 / UC Case 2025-124-0

PATENT STATUS

Patent Pending

BRIEF DESCRIPTION

Single-particle/single-molecule tracking (SPT) is a key tool for quantifying molecular motion in cells and in vitro. Wide-field SPT, in particular, can yield super-resolution mapping of physicochemical parameters and molecular interactions at the nanoscale, especially when integrated with single-molecule localization microscopy techniques like photoactivation and fluorophore exchange. However, wide-field SPT is often limited to the slow (<10 $\mu m^2/s$) diffusion of molecules bound to membranes, chromosomes, or the small volume of bacteria, in part due to the ~10 ms framerate of common single-molecule cameras like electron-multiplying charge-coupled devices (EM-CCDs); for unbound diffusion in the mammalian cell and in solution, a molecule readily diffuses out of the <1 µm focal range of high-numerical-aperture objective lenses within 10 ms. While recent advances such as ultra-highspeed intensified CMOS cameras, feedback control by locking onto a molecule, trapping, and tandem excitation pulse schemes address the framerate issue, each also introduces drawbacks in light/signal efficiency, speed, uninterrupted diffusion paths, and/or trajectory resolution, e.g., number of time points.

UC Berkeley researchers have overcome these myriad challenges by introducing spatially-encoded dynamics tracking (SpeedyTrack), a strategy to enable direct microsecond wide-field single-molecule tracking/imaging on common microscopy setups. Wide-field tracking is achieved for freely diffusing molecules at down to 50 microsecond temporal resolutions for >30 timepoints, permitting trajectory analysis to quantify diffusion coefficients up to 1,000 um^2/s. Concurrent acquisition of single-molecule diffusion trajectories and Forster resonance energy transfer (FRET) time traces further elucidates conformational dynamics and binding states for diffusing molecules. Moreover, spatial and temporal information is deconvolved to map long, fast single-molecule trajectories at the super-resolution level, thus resolving the diffusion mode of a fluorescent protein in live cells with nanoscale resolution. Already substantially outperforming existing approaches, SpeedyTrack stands out further for its simplicity—directly working off the built-in functionalities of EM-CCDs without the need to modify existing optics or electronics.

SUGGESTED USES

» Facile microsecond tracking/imaging of single molecules and their super-resolution mapping in the wide field

ADVANTAGES

» Wide-field tracking of freely diffusing molecules down to 50 microsecond temporal resolutions and for >30 timepoints

» Resulting trajectory analysis can quantify diffusion coefficients up to 1,000 um^2/s

CONTACT

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INVENTORS

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OTHER INFORMATION

CATEGORIZED AS

» **Optics and Photonics**
» All Optics and Photonics
» **Agriculture & Animal Science**
» Animal Science
» Devices
» Other
» **Biotechnology**
» Other
» **Computer**
» Software
» **Imaging**
» Molecular
» Other
» Software
» **Nanotechnology**
» NanoBio
» **Research Tools**
» Other

RELATED CASES

2025-124-0

» Simply integrates with existing commercial microscopes and detectors

RELATED MATERIALS

» M. A. Steves and K. Xu, "SpeedyTrack: Direct microsecond wide-field single-molecule tracking and super-resolution mapping via CCD vertical shift," bioRxiv, 2025.04.07.647376, 2025.

ADDITIONAL TECHNOLOGIES BY THESE INVENTORS

- Facile, Excitation-Based Spectral Microscopy For Fast Multicolor Imaging And Quantitative Biosensing
- Superresolution Microscopy And Ultrahigh-Throughput Spectroscopy
- Direct Optical Visualization Of Graphene On Transparent Substrates

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