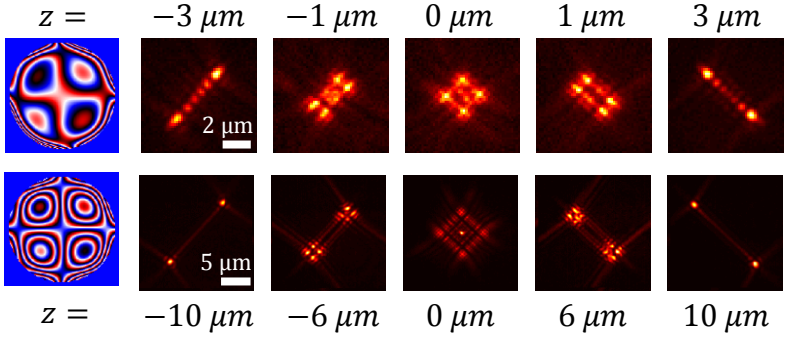
Precise Three-Dimensional Scan-Free Multiple-Particle Tracking over Large Axial Ranges with Tetrapod Point Spread Functions

A major limitation of current 3D super localization methods is limited depth of field, or applicable axial (*z*) range (~3 µm). As a result, **such methods are inapplicable for directly imaging or tracking in thick samples**, such as mammalian cells.

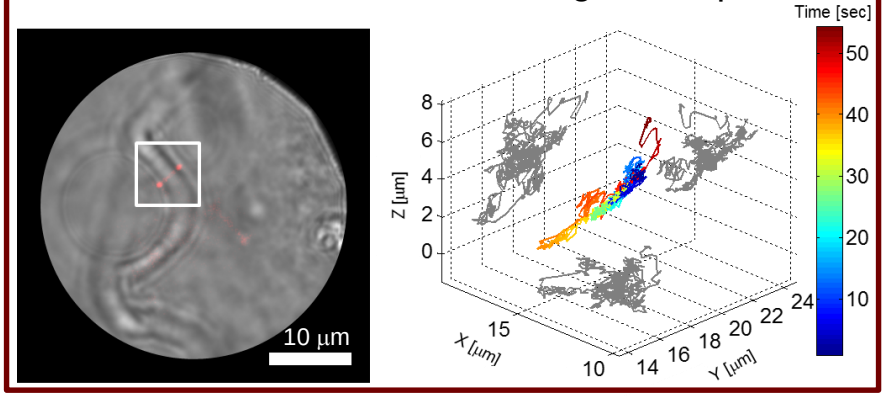
Using our recently developed PSF optimization framework, we are able to algorithmically design **information optimal PSFs with unprecedented applicable *z* ranges of up to 20 µm.**

Tetrapod PSFs with a 6um range (top) and 20um range (bottom)

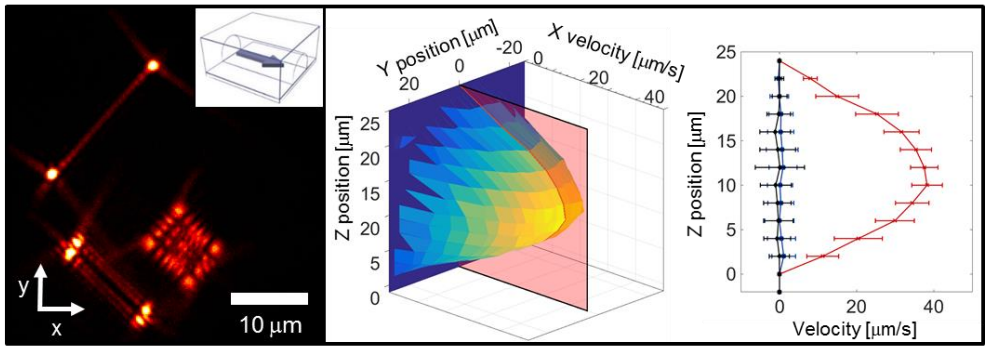


New possibilities enabled by such PSFs are demonstrated in example applications:

Tracking a QD diffusing on the membrane of a live mammalian cell over a z-range of ~6 µm



Flow profiling in a 20 µm thick micro-fluidic channel



Shechtman, Y., Weiss, L.E., Backer, A.S., Sahl, S.J. & Moerner, W. E. *Nano. Lett.* **15** (6), 4194-4199 (2015)