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Measurement-based estimation of global pupil functions in 3D localization microscopy

- Phase aberrations distort the experimentallymeasured point spread function relative to its theoretical shape, limiting the accuracy and precision of 3D localization procedures.
- We demonstrate an algorithm for improving the PSF model by estimating the aberrations using the Tetrapod engineered PSF.

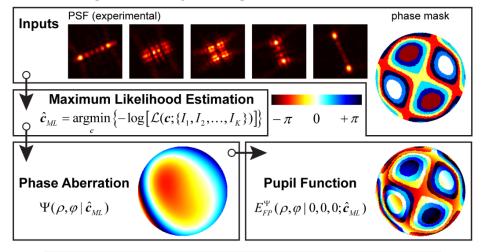
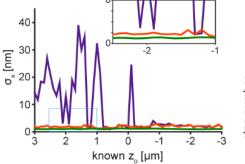
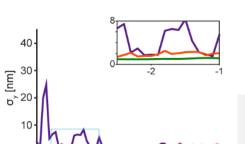
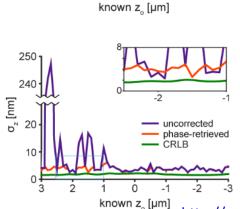


Figure 1: Schematic of the phase retrieval algorithm. A map of the phase mask pattern is used to produce the theoretical model, and a set of experimental images of the (tetrapod) PSF is used to perform the MLE step. The estimation procedure returns a phase aberration term, which is added to the original phase mask pattern to produce the overall corrected pupil function.





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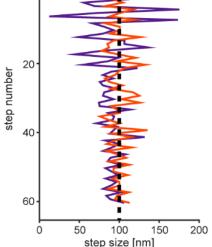


Figure 2: An axial scan of a fluorescent bead in 100 nm steps over 6 μm reveals poor 3D localization performance of the uncorrected PSF model (purple). Estimation of the phase aberration produces a phase-retrieved PSF model (orange), which shows improved accuracy (above) and precision (left), approaching the Cramér-Rao Lower Bound (CRLB).

