IAP Seminar

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Tuesday, 9th November 2021, 16:00 s.t.
TU Wien, Institut für Angewandte Physik, E134
1040 Wien, Wiedner Hauptstraße 8-10
Yellow Tower „B“, 5th floor, SEM.R. DB gelb 05 B

Verifying biomolecular nanoclustering in single molecule localization microscopy

Single molecule localization microscopy (SMLM) circumvents the diffraction limit of light by separating the signals from individual fluorophores in time. While the main focus of SMLM analysis up to now has been the precise localization of the fluorophores, the natural next step concerns qualitative and quantitative interpretation of the localization maps and their biological relevance. Such an interpretation is challenging, as fluorophore blinking leads to repeated detections of the same molecule. The resulting apparent localization clusters can be easily mistaken for biomolecular nanoclustering.
We developed two different methods for distinguishing true biomolecular clustering from overcounting artifacts. First, a comprehensive characterization of fluorophore blinking behavior combined with Monte Carlo simulations allows for robust evaluation of localization maps with respect to true molecular clustering. Second, an assessment of molecular clustering completely independent of fluorophore blinking can be achieved by targeting the same molecule of interest with two different labels competitively achieves. Thus, biomolecular clustering can be reliably detected with high sensitivity down to the level of dimers.

All interested colleagues are welcome to this seminar lecture
(30 min. presentation followed by discussion).

Friedrich Aumayr
(LVA-Leiter)

Gerhard Schütz
(Seminar Chair)