Imaging the topology of soft and deformable interfaces by single-molecule localization microscopy

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In the last decade the spatial resolution achieved in fluorescence microscopy has dramatically improved to access length scales as small as tens of nanometers with single molecule localization techniques (SMLM). Their application has unveiled the structure and properties of highly complex systems, such as actin-spectrin periodicity of the cytoskeleton in axons. However, soft and deformable interfaces like emulsions, foams, and ice crystals have scarcely been investigated by SMLM, in part because these cannot be tagged covalently with fluorescent probes. To overcome this challenge, we developed a fluorescence imaging method able to visualize the topology of objects without covalent labeling, irrespective of their surface chemistry. We coin this method interface Point Accumulation for Imaging in Nanoscale Topography (iPAINT) [1].

iPAINT relies on aspecific physical adsorption onto interfaces of polymer chains, tagged with a photo-activatable dye, which are present at micromolar concentrations in the bulk solution. This ensures a constant flux of probes and a continuous replacement of photobleached dyes due to molecular exchange of probes between the solution and the interface. In this contribution we will present the possibilities of this novel approach in SMLM for imaging of soft materials highlighting our recent work on the precise reconstruction of the internal structure of foams, Pickering emulsions and complex coacervate core micelles (C3Ms). Single air nanobubbles were imaged in three dimensions aiming to assess the influence of the nucleation temperature on their morphology. Subsequently, iPAINT was used to visualize colloidal particles of 300 nm in radius at fluid interfaces. The clear reconstruction of both particles and the interface between the two liquids makes iPAINT a true *in-situ* approach to visualize the three-phase contact line at the nanometer length-scale. This enables direct measurements of the contact angle of single particles, which is crucial to understand the behaviour of Pickering emulsions. Finally, we probe the morphology of C3Ms formed upon complexation of oppositely charged block copolymers in aqueous solutions. We find that these undergo a morphological transition from spherical to wormlike structures as the polymer concentration increases.

These three examples demonstrate the applicability of iPAINT in soft matter science as a powerful, complementary tool for in-situ imaging with nanometric resolution of a broad range of soft and deformable interfaces without covalent labelling.

References:

[1] A. Aloi, N. Vilanova, L. Albertazzi, I. Voets, *Nanoscale*, 2016, **8**, 8712-8716, DOI: 10.1039/c6nr00445h.