Imaging of microspheres for optimisation and adaptations of DNA-PAINT

Tobias Lutz[1], Alexander H. Clowsley[1], Ruisheng Lin[1], Lorenzo Di Michele[2], Christian Soeller[1]
[1]Living Systems Institute & Biomedical Physics, University of Exeter, United Kingdom
[2]Cavendish Laboratory, University of Cambridge, United Kingdom
E-mail: c.soeller@exeter.ac.uk

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DNA-PAINT (Point Accumulation Imaging in Nanoscale Topography) is a super-resolution imaging technique which allows imaging at sub-5 nm resolution. It is largely independent of dye photophysics, multiple options for multiplexed imaging exist and quantitative imaging is possible in synthetic samples, fixed cells or tissue sections[1,2]. Being based on well-studied DNA nanotechnology, DNA-PAINT allows a high degree of flexibility and opportunity for adaptations and improvements, e.g. optimisation of background fluorescence, signal duration, and protocol simplification. Evaluation of such adaptations is often carried out using DNA origami, however, these nanostructures can be technically challenging, time consuming and expensive to prepare. We demonstrate the alternative use of streptavidin-coated microspheres as straightforward and cost-effective DNA-PAINT targets[3]. The coating allows quick and high-affinity binding of biotinylated oligonucleotide structures, and the regular shape of the particles provides a positive control for imaging, allowing to optimise imaging parameters, e.g. buffer conditions, or to test new imaging approaches, e.g. 3D or multiplexed imaging as well as other DNA binding schemes. We demonstrate a variety of practical assays and their use in optimising DNA-PAINT for high-quality super-resolution imaging.

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