Multicolor bio-imaging at molecular scales with 2D and 3D MINFLUX nanoscopy

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Imaging protein complexes at the molecular scale has been on the wish list of researchers in the life sciences for decades. With the advent of MINFLUX nanoscopy [1], this demand can now be met.

MINFLUX is capable of resolving individually switchable fluorophores at distances as small as 1 - 2 nm. This is possible by localizing single fluorophores with an excitation pattern featuring a spatially well-controlled intensity zero. In our case, we use a donut-shaped excitation beam whose central zero is targeted to pre-defined positions close to the fluorophore. The fluorophore position can thus be determined with a minimal number of photons and consequently within a spatio-temporal regime that exceeds alternative techniques [1, 2]. Here, we report on a MINFLUX microscope that is based on a common fluorescence microscope stand **[3]**. This novel MINFLUX microscope combines high localization precisions with standard workflows, therefore allowing even non-experts to apply this technique.

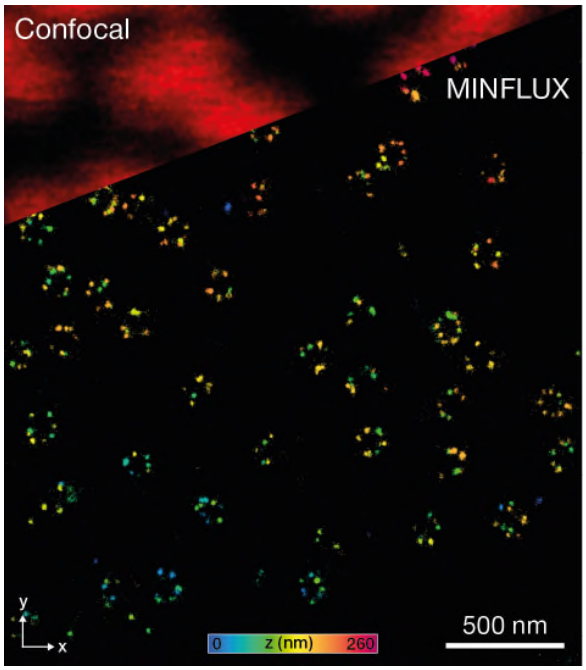

Figure 1. Confocal and 3D MINFLUX images of a nuclear pore complex sample.

To demonstrate the capabilities of our system, we present MINFLUX imaging of Nuclear Pore Complex samples in 2D mode, with localization precisions below 2 nm. In 3D MINFLUX imaging mode, isotropic localization precisions below 2.5 nm (Fig. 1) were achieved.

Images from additional samples from the life sciences will also be presented, highlighting the option for multi-color imaging in the nanometer range. We amply demonstrate that MINFLUX will allow researchers to address numerous biomedical and biophysical questions on the molecular scale.

References

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