New wavelength-tuneable birefringent easySTED phase plate

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Stimulated emission depletion microscopy (STED) is one of the most prominent fluorescence-based far-field tools for imaging intact biological cells with a resolution far beyond the diffraction limit [1, 2]. It usually employs a scanning excitation beam which is superimposed by a donut-shaped STED beam for switching off fluorescence at the periphery of the excitation spot.

Recently, a segmented, chromatic, birefringent wave plate was introduced that generates a donut-shaped depletion pattern while leaving the excitation spot intact [3]. When mounted directly in front of the objective lens and in the co-aligned beam path of both the excitation and STED lasers, this wave plate converts a conventional beam scanning microscope into an easy-to-use and rugged STED microscope (easySTED). So far however, a chromatic phase plate of that type can be used for only one specific pair of excitation and STED wavelengths.

Here we introduce a new, birefringent phase plate for STED which extends the concept of easySTED to include wavelength tuneability. Technically, this phase plate is a combination of a segmented, chromatic element and an achromatic element and allows to easily tune the depletion pattern and the excitation spot according to the applied laser wavelengths. Any pairs of STED and excitation wavelengths now require only a single birefringent beam shaping device.