

# Quo vadis?

## - Recent Developments in Microscopy -

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An overview some recent advances in fluorescence microscopy will be given with a special focus on enhancement of optical resolution.

In **structured illumination** the sample is illuminated with a number of different patterns of light. In our case this is a series of sinusoidal grids at different grid positions and orientations generated by a programmable spatial light modulator. Experimental datasets acquired under these conditions and reconstructed results from these data, demonstrating a resolution improvement of up to a factor of two over standard widefield microscopy are presented.

The non-linear approach of **saturating optical transitions** (for structured illumination as well as beam-scanning approaches) has a great potential especially in combination with photo-switchable dyes such as the recently released DRONPA protein by Atsushi Miyawaki's group or the Cy3-Alexa647 system used in Xiaowei Zhuang's group. An interesting approach is to push molecules into dark states in a patterned way shortly before imaging and exploiting the saturation of this transition.

A further approach to high resolution imaging is based on the **localization** of multiple particles in an image. This approach was named Pointillism. Experimental data with particle separation based on independent component analysis will be presented.

Finally a method will be presented in which the emitted fluorescence of a confocal microscope passes through two separate paths. These paths are **interferometrically recombined** in such a way that the images undergo a mutual rotation of 180 degrees. The self-interference of the fluorescent light is then only constructive if it originated from the optical axis of the scanning laser beam, thus leading to an efficient detection of a high resolution fluorescence image.