CHARACTERIZATION OF TWO-PHOTON-SWITCHING CROSS SECTIONS FOR FPALM

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The super-resolution technique Biplane-FPALM \cite{Juette2008} allows to image samples in three dimensions beyond the diffraction limit. This is achieved by sequentially imaging and localizing single photo-switchable molecules in the sample and reconstructing a map of particle positions.

We have replaced the standard violet (405 nm) activation light by two-photon activation using a focused mode-locked laser beam of 700-1000 nm wavelength. This allows limiting of the activation process to a single specimen plane instead of the whole sample thickness \cite{Schneider2005, Ivanchenko2007, Foelling2008} and therefore reduces background and unwanted bleaching. To effectively choose the right fluorescent probes it is important to know the two-photon cross section $\sigma_{2P}$ of the available proteins.

Here we present recent results characterizing $\sigma_{2P}$ as a function of wavelength (Fig. 1) for the widely used photo-activatable/-switchable fluorescent proteins mEos2, PAmCherry, Dendra2 and PATagRFP

![Figure 1: Preliminary two-photon absorption cross section as a function of the wavelength for PAmCherry](image1)

![Figure 2: Activation rate for PAmCherry as a function of the activation power at 700nm wavelength](image2)

\cite{Juette2008, Schneider2005, Ivanchenko2007, Foelling2008}