

### Physikalisches Kolloquium Einladung

# Physics Colloquium

# Monday, 02 May 2022

Lecture Hall O25/H2, 16:15

## Using Light to illuminate the Dynamics of Life

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Fluorescence spectroscopy and microscopy offers very sensitive, non-contact approaches to investigate biological systems. Over twenty-five years ago, the ability to detect Förster Resonance Energy Transfer (FRET) on a single molecule was demonstrated. This ability to measure the motion of individual molecules on the nanometer scale has opened a whole new realm for investigating the dynamical function of biological systems. I will demonstrate the use of advanced FRET methods to follow the dynamics of the heat shock protein 70, Ssc1. We first characterized the dynamics of the protein *in vitro* where we have full control overall all the interacting components. Then we moved to investigate the conformation of Ssc1 in their native environment, within mitochondria. We discovered a fine balance in the distribution of Ssc1 that allows the protein system to react quickly to protein folding stress and initiate rescue programs.

In the second part of my talk, I will discuss moving beyond spFRET to three-color FRET. Although the technical challenges of building a third color channel is small, the challenges in extracting the relevant information is more difficult. Hence, we developed a three-color photon-distribution analysis approach to get at the underlying correlations within the data (Barth et al., 2019). We utilized 3-color FRET to investigate the order of folding of a two-domain protein, the Maltose Binding Protein. The two domains are discontinuous and, by using 3c FRET, we could show that the two domains fold jointly.

Lastly, I will briefly mention our investigations of viral assembly and maturation. Here, we used *ex post facto* approaches to follow the assembly of HIV. To follow the last step of assembly, maturation, we introduce a new approach using Fluorescence Lifetime Imaging Microscopy (FLIM) to follow the conformational changes in HIV during maturation (Chen et al. 2021)

Barth, A., Voith von Voithenberg, L., and Lamb, D.C. (2019). Quantitative Single-Molecule Three-Color Forster Resonance Energy Transfer by Photon Distribution Analysis. J Phys Chem B.

Qian, C., A. Flemming, B. Muller, and D. C. Lamb. (2022). Dynamics of HIV-1 Gag Processing as Revealed by Fluorescence Lifetime Imaging Microscopy and Single Virus Tracking. Viruses 14.



Host: Prof. Dr. Jens Michaelis, Institute of Biophysics

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