REVEALING STRUCTURAL DYNAMICS OF LIVER SINUSOIDAL ENDOTHELIAL CELLS

Viola Mönkemöller[a], Cristina Øie[b], Peter Heimann[c], Christian Kaltschmidt[c], Barbara Kaltschmidt[c], Peter McCourt[b], Thomas Huser[a], and Wolfgang Hübner[a]

[a] Biomolecular Photonics, Department of Physics, Bielefeld University, Bielefeld, Germany
[b] Faculty of Health Sciences, Department of Medical Biology, Vascular Biology Research Group, University of Tromsø, Norway
[c] Cellbiology and Molecular Neurobiology, Department of Biology, Bielefeld University, Bielefeld, Germany

KEY WORDS: single molecule localization microscopy, nanoscopy, *d*STORM, liver sinusoidal endothelial cells (LSEC), structured illumination microscopy (SIM), dynamics

Liver sinusoidal endothelial cells (LSEC) are a special class of endothelial cells acting as a filter of lipoproteins and small molecules between the liver and blood. A number of clinical conditions, especially metabolic and aging-related disorders, are implicated by improper function of LSECs. The disruption of these cells due to e.g. paracetamol poisoning contribute to acute liver failure and death, thus underlining their biomedical relevance.
Furthermore, LSECs have a unique morphology - containing a large number of transcellular pores, so-called *fenestrations* that are grouped into sieve plates. Until recently, fenestrations could only be resolved by electron microscopy (EM) due to their small size below the optical diffraction limit (50 to 200 nm in diameter). EM methods are not suitable to study the function and dynamics of fenestrations in living LSECs, which is why fenestrations are still poorly understood and thus provide an ideal target for biomedical super-resolution optical microscopy. As "hollow/empty" structures, fenestrations can only be visualized by negative counterstaining, which poses a challenge for imaging by super-resolution techniques.
To date, we have obtained the highest optical resolution of fenestrations [1]. We also compared the higher degree of structural detail that single molecule localization microscopy (e.g. *d*STORM) provides to results obtained by 3D structured illumination microscopy (3D-SIM) on the same microscope setup [2]. A combination of the best of two super-resolution microscopy techniques helps to maximize the information obtained from imaging these physiologically important cells. Here, we present first dynamic imaging data of LSEC fenestration dynamics by 3D structured illumination microscopy (3D-SIM) [3]. By optimizing labeling approaches of the plasma membrane of LSECs for live cell imaging we observed the dynamic formation and closing of fenestrations under different physiological conditions, including drug treatments such as paracetamol and actin disruptors.

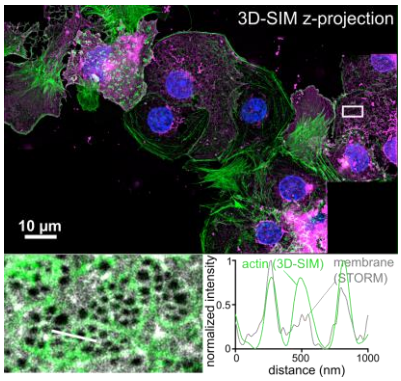

Figure 1 Correlative SIM and *d*STORM of LSECs

References:

[1] Mönkemöller, V. et al. *Imaging fenestrations in liver sinusoidal endothelial cells by optical localization microscopy*. Physical Chemistry Chemical Physics : PCCP 16, 12576-12581 (2014).
[2] Mönkemöller, V., Øie, C., Hübner, W., Huser, T. & McCourt, P. Scientific Reports 5, 16279 (2015).
[3] Müller, M., Mönkemöller, V., Hennig, S., Hübner, W., & Huser, T. (2016). Nature Communications, 7.