Q 32: Biophotonics

Time: Wednesday 11:00–13:00 Location: f342

Q 32.1 Wed 11:00 f342

Label-free imaging of single proteins and viruses ejected from a living cell — •Matthew McDonald, Katharina König, Marek Piliarik, Rui Qi Zhao, and Vahid Sandoghdar — Max Planck Institute for the Science of Light, Erlangen, Germany

A number of important physiological processes–such as cellular signaling and viral invasions–are marked by their secretomic behavior. Real-time, in-vivo sensing of these cellular secretion events commonly relies on labeled proteins and is thus inherently accompanied by the perturbation of the system. Here, we present a novel, label-free method that enables the detection of single protein secretions from living cells via an interferometric scattering technique (iSCAT). Secretomic events from an individual cell are imaged by way of iSCAT wherein single proteins and bioparticles from the kDa to MDa range are synchronously detected in real-time. The developed method has the potential to solve a wide range of problems in cellular physiology, such as intercellular signaling, immunology, and cancer malignancy.

Q 32.2 Wed 11:15 f342

Extending the applicability of Scanning Laser Optical Tomography using antibody staining and nonlinear contrast mechanisms — •Lena Nolte[1], Nadine Tinne[1], Georgios Antonopoulos[1], Marko Heidrich[1], Jennifer Schulze[2], Kristin Schwanke[2], Robert Zweigerdt[2], Athanasia Warnecke[2], Alexander Heisterkamp[3], Tammo Ripken[1], and Heiko Meyer[1] — [1]Laser Zentrum Hannover, Germany — [2]Medizinische Hochschule Hannover, Germany — [3]Leibniz Universität Hannover, Germany

Scanning laser optical tomography (SLOT) enables three dimensional visualization of large samples up to a magnitude of several centimeters using absorption and autofluorescence as intrinsic contrast mechanisms. However, this intrinsic contrast is sometimes not strong enough to image significant details inside the sample. One challenge is the visualization of hair cells and neurofilaments inside the human cochlea. For this reason, we developed a protocol for decalcification, antibody staining and optical clearing to image the cochlea in toto using SLOT.

Optical clearing is an efficient way to look into thick and turbid samples, but also prohibits the application of in vitro studies. Using near-infrared light, the scattering coefficient of the sample is lower and imaging of non-cleared samples can be improved. Therefore, we integrated a fs-pulsed laser source into the SLOT to enable the generation of two-photon fluorescence inside the sample. This way, living cell aggregates, with a diameter up to hundreds of micrometers, can be studied with respect to their three-dimensional structure without optical clearing.

Q 32.3 Wed 11:30 f342

Aberration correction in STED nanoscopy for super-resolution imaging deep inside living tissue — •Jasmin K. Pape, Nicolai T. Urban, Jennifer-M. Masch, and Stefan W. Hell — Max Planck Institute for Biophysical Chemistry, Department of NanoBiophotonics, Göttingen, Germany

Stimulated emission depletion (STED) nanoscopy is a far-field fluorescence imaging technique capable of resolving structures on the nanometer scale. It is remarkably well suited for dynamic imaging of live cells and tissues, especially due to its short acquisition times. When imaging deep inside living tissue, however, two main factors limit the imaging performance: one is the loss of intensity due to absorption and scattering, the other is the distortion of the wavefront shape caused by the inhomogeneous refractive index inside the sample medium.

We address the latter problem by pre-shaping the wavefront of the STED beam using a spatial light modulator, with the aim of recovering a high-quality donut-shaped intensity distribution, which is essential for achieving high spatial resolutions. We determine the optimal correction parameters for compensating the sample-induced aberrations by employing an algorithm which records a series of images and then evaluates different properties of each image. The correction process is improved in a way that reduces the number of acquisitions necessary to find the best correction. The correction capabilities of this method will be compared both in artificial samples and in live cells.

Q 32.4 Wed 11:45 f342

Synchronization-free all-solid-state laser system for stimulated Raman scattering microscopy — •Moritz Floess[1], Tobias Steinle[1], Vikas Kumar[2], Andy Steinmann[1], Marco Marangoni[2], Giulio Cerullo[2], and Harald Giessen[1] — [1]4th Physics Institute and Research Center Scope, University of Stuttgart, D-70569 Stuttgart, Germany — [2]2IFN-CNR, Dipartimento di Fisica, Politecnico di Milano, Piazza Leonardo da Vinci 32, I-20133 Milan, Italy

We demonstrate a simple all-solid-state laser source for stimulated Raman scattering (SRS) microscopy. An 8 W, 450 fs Yb:KGW oscillator with 41 MHz repetition rate pumps an OPA that is seeded by a cw tunable external-cavity diode laser (ECDL). The second-harmonic of the OPA output radiation, generated in a PPLN crystal, acts as the Raman pump beam, tunable between 760 and 820 nm. In contrast to using an OPO as tunable laser source the cw-seeding of the OPA avoids synchronization issues. We demonstrate SRS images with pixel dwell times down to 30 μs with signal-to-noise ratios of up to 50 when investigating polymer beads. Thanks to the favorable noise properties of the solid-state oscillator SNRs of 5 are still possible with 500 ns pixel dwell time.

Q 32.5 Wed 12:00 f342

Microcavity based detection of single ions interacting with plasmonic nanorods — •Martin D. Baaske and Frank Vollmer — Max-Planck-Institut für die Physik des Lichts, Erlangen, Germany

Whispering gallery mode based microresonators provide powerful tools for the all optical detection of nanoscopic objects such as nanoparticles and viruses[1,2]. Their sensitivity can further be boosted by modification of the microcavities with resonant plasmonic nanoparticles allowing for the detection of single molecules and their interactions[3].

Here we present experimental data on the microcavity based observation of single ions interacting with immobilized plasmonic nanorods in aqueous environment. We show that different types of interactions can be identified by their transient behavior. Furthermore we discuss the influence of the solution's ionic strength on the type of interaction observed for bivalent ions of two different elements.

[1] L. He et al., *Nature Nanotech.*, **6**, 428 (2011)
[2] F. Vollmer et al., *PNAS*, **105**, 20701 (2008)
[3] M.D. Baaske et al., *Nature Nanotech.*, **9**, 933 (2014)

Q 32.6 Wed 12:15 f342

Optimizing thiol DNA-gold reaction using whispering gallery microcavities — •Eugene Kim, Martin D. Baaske, and Frank Vollmer — Max-Planck-Institute for the science of light

In this work, the transient kinetics of thiol DNA-gold interactions in aqueous environment are studied at the single-molecule level utilizing high Q plasmonic-photonic whispering gallery mode microcavities. Three different regimes of thiol-gold interactions were found depending on the environmental conditions. Statistical analysis of the detection frequency, dwell-/binding-time, and size distribution of transient interactions with respect to their dependence on pH and electrolyte concentration allow us for the optimization of thiol-gold bonding.

Q 32.7 Wed 12:30 f342

Detection and simulation of optoacoustic signals generated in layered tissues — •O. Melchert, E. Blumenroether, M. Wollweber, M. Rahlves, and B. Roth — Hannover Centre for Optical Technologies, Leibniz Universität Hannover, Hannover, Germany

The absorption of electromagnetic waves by media induces a spatial pressure distribution, proportional to the density of the deposited energy, followed by thermoelastic expansion and emission of acoustic waves. While the deposition of energy is assumed to be instantaneous, the propagation of acoustic waves is determined by the sound velocity of the material. From a theoretical point of view, the resulting mathematical problem is governed by an inhomogeneous wave equation featuring an optoacoustic source term.

Here, we consider optically inhomogeneous, i.e. layered media, where scattering is effectively negligible and the absorbed energy density follows Beer-Lambert's law, i.e. is characterized by an exponential decay within the layers and discontinuities at interfaces. We complement test experiments on samples where the material properties are known *a priori*, with numerical simulations based on solving the optoacoustic wave

equation, tailored to suit our experimental setup. Experimentally we characterize the acoustic signal observed by a piezoelectric detector in the acoustic far-field in backward mode and we discuss the implication of acoustic diffraction on our measurements as well as possibilities to retrieve the absorption coefficient from measurements in the forward mode.

Q 32.8 Wed 12:45 f342

Simulation of the OCT-depth signal of homogeneous turbid media via an extended Monte-Carlo model — •Arthur Varkentin, Maya Otte, Merve Wollweber, Maik Rahlves, and Bernhard Roth — Hannoversches Zentrum für Optische Technologien - HOT, Leibniz Universität Hannover, Germany

Optical coherence tomography (OCT) is widely used for imaging of biological tissue. In most cases the result is a 2D or 3D tomogram showing scattering structures of the studied sample. This qualitative information indicates the morphology of the tissue. The extraction of quantitative information such as the scattering coefficient μ_s is straight forward only for weakly scattering media where ballistic photon scattering can be assumed. For highly scattering media, however, additional phenomena have to be taken into account. For example, multiple scattering has to be considered, where photons that are scattered more than once, but are still within the coherence length of the OCT also contribute to the signal. A cluster of equal scatterers can appear as one single scatterer with different optical properties. These effects lead to concentration dependent scattering which shows nonlinear behavior. We present a simple model to simulate OCT-depth signals in weakly and strongly scattering media. Multiple scattering is implemented and, in addition, a weighting function rescales the photon signal according to the number of undergone scattering events. Based on a parameter study of this weighting function we are able to implicitly predict the influence of dependent scattering without modeling the process explicitly. In future, our quantitative approach could improve biological imaging.