

Research Seminar

2pm Friday 4th December 2015

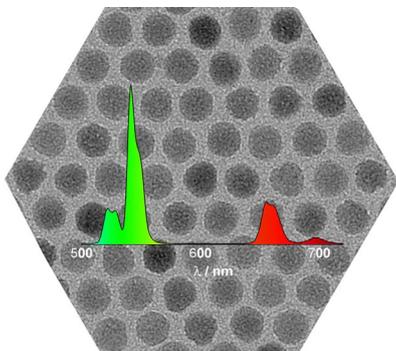
Room: JA5.04

DEPARTMENT OF PHYSICS



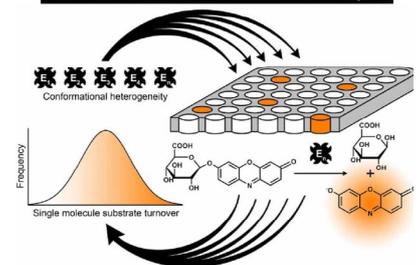
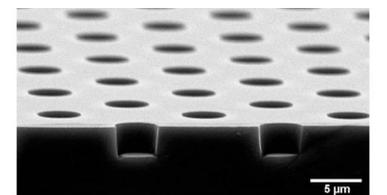
Photon-upconverting nanoparticles and femtoliter arrays for background-free imaging and ultrasensitive biomedical applications

Fluorescence microscopy is one of the most sensitive and versatile detection methods, which can reach the single molecule level. Autofluorescence and light scattering of the surrounding medium, however, leads to strong background interference in imaging and biomedical applications. Hence, we follow two strategies to eliminate the high background fluorescence.



1) Photon-upconverting nanoparticles (UCNPs) emit short-wavelength light under near-infrared excitation (anti-Stokes emission), which strongly reduces autofluorescence and light scattering. As bioanalytical applications require UCNPs that are well dispersible in aqueous solutions and do not aggregate, we have developed new strategies to characterize and purify UCNPs by gel electrophoresis. UCNPs have provided new opportunities for the design of highly sensitive immunoassays as well as optical encoding schemes that enable the multiplexed detection of diagnostic markers. Recently, we have developed a new upconversion laser scanner for the ultrasensitive readout of UCNPs on a macroscopic scale. I will also show an upconversion microscope setup for imaging single UCNPs against zero background.

2) Large arrays of 62,500 femtolitre-sized chambers are etched into the surface of fused silica slides to isolate hundreds of individual enzyme molecules and observe their substrate turnover in parallel. If a fluorogenic reaction takes place in a femtolitre volume, a single enzyme molecule generates a product concentration high enough for detection by wide-field fluorescence microscopy. I will demonstrate that individual molecules of β -galactosidase, β -glucuronidase, and horseradish peroxidase display broadly distributed catalytic activities as a consequence of different protein conformations. This single molecule approach also provides new insights into molecular evolution and enzyme inhibition. These femtolitre arrays lay the foundation for developing single molecule immunoassays.



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