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Direct DNA quantification using gold nanoparticles as label free biosensor

Prayanka Rajendran, Marcy Zenobi-Wong and Janos Vörös
ETH Zurich, Switzerland

Single-cell mRNA quantification holds great promise to understand diversity among a homogeneous cell population, but the methodology to process these samples in a reproducible, quantitative and parallel fashion remains challenging. The techniques being developed are aimed to form the foundation for highly parallel single-cell mRNA level studies. Here, we utilize microwell arrays to hold sub nanolitre solution, equivalent to single cell volume, together with sequence specific oligonucleotides tagged to reporter probe (gold nanoparticle, fluorophore etc.) and other accessory components such as salt, for probe hybridization. The ultimate aim is to demonstrate single-cell lysis for mRNA isolation and quantification in a single pot. Direct oligonucleotide quantification assays can be attractive detection systems since they offer the potential to assess single cells without the use of reverse transcription and amplification steps as required for Reverse Transcription- Polymerase Chain Reaction (RT-PCR). Even though PCR and microarrays have already become standard methods for the analysis of nucleic acids, still more direct and less cumbersome assays are needed to make a commercial breakthrough for oligonucleotide detection technology.

The aim of this project is to develop a scattering spectra - based detection system to quantify the oligonucleotides of interest using metal nanoparticles. Gold nanoparticles will be used as nanosensors. The assay results in gold nanoparticles forming pairs via side-by-side hybridization events between the oligonucleotides and their complementary mRNA target. The pair formation leads to concomitant changes in the scattering spectra of the particles which can be utilized for the detection and quantification of the target. The binding of the analyte (target DNA/RNA molecule) to the oligonucleotide-modified gold nanoparticles during the assay brings the plasmonic nanoparticles in close vicinity to each other (<5 nm). Due to this proximity effect they become optically coupled, creating a more enhanced field and giving a strong resonance depending on the coupling strength or interparticle distance. As a result there is a second scattering peak towards the red region.

A custom made dark field spectro-microscope by combining a spectrometer and an inverted optical microscope with halogen lamp illumination is used as read out system.

Biography

Prayanka Rajendran has completed her M.Sc. in Photonics in École Normale Supérieure de Cachan and is currently pursuing her Ph.D. in Laboratory for Biosensors and Bioelectronics, ETH Zurich.

rajendran@biomed.ee.ethz.ch