Label-free monitoring of microcapsule-enabled intracellular release using gold-nanoparticle coated microchips

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By using gold and silver nanostructures for intracellular SERS, several applications of highly sensitive and selective labelfree single cell analysis have been demonstrated[1]. While colloidal nanoparticles have been shown to be minimally invasive to cells, these experiments suffer from a poor reproducibility due to the unpredictible behaviour of the nanoparticles. On the other hand, Tip-Enhanced RS probes contain a fixed metal nanopattern, but intracellular applications with these probes require an incision of the cell membrane during the measurement. In order to enable intracellular SERS detection with a fixed metal pattern, but without an incision of the cell wall during measurements, we fabricated micron-sized silicon-nitride chips using UV contact lithography. These planar structures are entirely incorporated by the cell and offer a high surface/volume ratio, which maximizes the probe area and is expected to limit cytotoxicity. Equally important, our fabrication scheme allows to use the wide variety of nano-antenna fabrication techniques developed for reproducible SERS subsrates for extracellular applications. We demonstrate this by coating the SiN chips both with adsorbed gold nanoparticles (fig 1, left) and with a predefined gold nanopattern using nanosphere lithography (fig 1, right). In a proof-of-concept application, we use our chips for a labelfree monitoring of the intracellular delivery of dextrane-RhodamineB (D-RhoB). These fluorescent molecules allow for correlative fluorescence and Raman Spectroscopy. Prior to delivery, a high concentration (1mg/ml) of D-RhoB is loaded into a polymeric capsule[2]. These capsules and the gold-coated SiN chips are delivered into the cytoplasm of a cell via electroporation (fig 2,left). Next, a specific capsule is opened through laser-triggered heating. Immediately afterwards, SERS spectra are collected on top of a SiN chip in the same cell. Despite the complex intracellular environment, specific SERS peaks originating from the released D-RhoB are observed at low laser power (1-2mW) and short integration time (500msec) (fig2, right). This experiment indicates the potential of our approach for monitoring (in-vitro) drug delivery. Furthermore the planar SiN chips enable us to design more complex photonic chips and gold coatings in the near future.

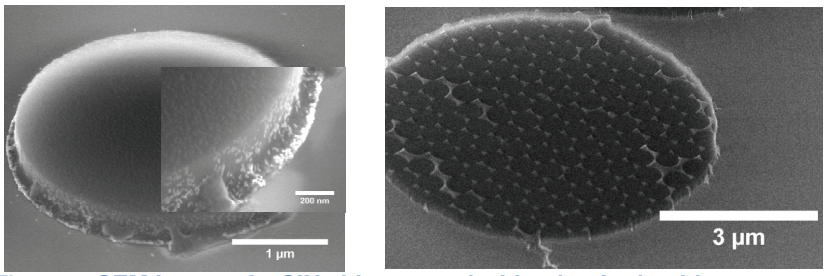

Figure 1: **SEM image of a SiN chips coated with adsorbed gold nanoparticles (left) and nanosphere-lithography fabricated gold triangles (right)**

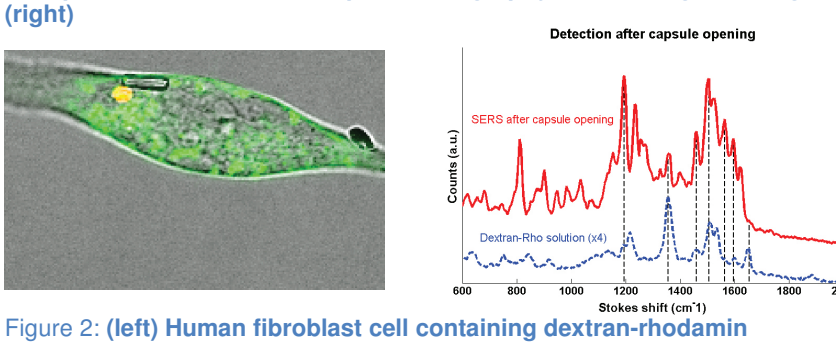

Figure 2: **(left) Human fibroblast cell containing dextran-rhodamin loaded capsules (yellow) next to a rectangular gold SiN chip with adsorbed gold nanoparticles (right) SERS spectra measured on top of the SiN chips after capsule release (red) and reference spectrum of dextran-rhodamin**

[1] E. Vitol et al., *J Raman Spectrosc.*., 2012, 43-7, 817-27, [2] A. Skirtach et al., *Nano Letters*, 2005, 5-7 1371-77