Metal-dielectric nanostructures for label-free SERS detection of miRNAs for early cancer diagnostics

Daniel Montesi[1], Alessandro Chiadò[1], Sofia Bertone[1], Chiara Novara[1], Niccolò Paccotti[1], Francesco Geobaldo[1], Gerolama Condorelli[2,3], Silvia Catuogno[3], Carla Lucia Esposito[3],Vittorio De Franciscis[3], Daniela Taverna[4], Paola Rivolo[1] and Fabrizio Giorgis[1]

[1]*Dept. of Applied Science and Technology, Politecnico di Torino, Torino, Italy*

[2]*Dept. of Molecular Medicine and Medical Biotechnology, University of Naples, Italy*

[3]*Institute of Endocrinology and Exp. Oncology of Italian National Research Council, Naples, Italy*

[4]*Dept. of Molecular Biotechnology and Health Sciences and MBC, University of Torino, Torino, Italy*

Corresponding author: daniel.montesi@polito.it

Surface Enhanced Raman Spectroscopy (SERS) is a highly sensitive spectroscopic technique based on Raman signal amplification that has been successfully used to detect biomolecules, cancer cells, pathogens, environmental pollutants and food contaminants. In this work, porous silicon (pSi) membranes decorated with silver nanoparticles (AgNPs) and integrated in a polydimethylsiloxane (PDMS) microfluidic chip were exploited as SERS-active substrates for the detection of microRNAs (miRNAs), short regulatory non-coding RNA sequences typically over- or under- expressed in connection with several diseases including oncogenesis. The SERS efficiency and uniformity of the AgNPs were initially tested using resonant and off-resonant probe molecules, while their near-field plasmonic response was investigated by modeling a simplified representative system (Ag hemispheres dimers on pSi) by 3D Finite Element Method. For the detection of the target miRNA (e.g. miR-222), an innovative two-step hybridization bioassay have been optimized. In the first step, one piece (half1) of a complementary DNA probe is immobilized on the AgNPs through a thiol group to catch the specific miRNA from the sample. In the second step, the captured miRNA is bound by the second piece of the probe (half2) , which is labelled with a Raman reporter (R6G, Cy3 or Cy5), thus enabling the label-free and highly sensitive detection of the target. The sensitivity of the two-step assay was demonstrated by the detection of several miR-222 concentrations in buffer solution down to the sub-nanomolar concentration range, while the selectivity of the bioassay was proved by the detection of miR-222 mixed with different interfering oligos. The influence of the distance of the Raman reporter from the plasmonic surface was also evaluated. Finally, the optimized functionalization protocol was successfully used to detect other miRNAs

involved in lung cancer (i.e. miR-214, miR-146a, miR-148b, miR-320a and miR-20a), even in cancer cell extracts, confirming the potentialities of SERS-based microfluidics for early-cancer diagnosis.