

Fabrication and Optical Characterization of Nanoimprinted Plasmonic Constructs for Biosensor Applications



Isaias Tesfaslassie

Applied Physics, Appalachian State University

NNIN REU Site: Lurie Nanofabrication Facility, University of Michigan, Ann Arbor, MI

NNIN REU Principal Investigator(s): L. Jay Guo, Department of Electrical Engineering and Computer Science, University of Michigan, Ann Arbor, MI

NNIN REU Mentor(s): Brandon D. Lucas, Applied Physics, University of Michigan, Ann Arbor, MI

Contact: tesfaslassieim@appstate.edu, guo@eecs.umich.edu, bdlucas@umich.edu

Abstract:

This work focuses on the fabrication of Au nanoparticle arrays using nanoimprint lithography (NIL), and their application as biosensor transduction elements. Noble metal nanoparticles such as gold (Au) are unique in that upon illumination by light of certain frequency, collective oscillations of the free electrons can be produced. This characteristic response is known as the localized surface plasmon resonance (LSPR). Optical characterization of the nanoparticle arrays show that the LSPR is not only sensitive to nanoparticle shape and size but it is also sensitive to the surrounding dielectric medium. Therefore, a biomolecular binding event on the particle surface causes the LSPR spectral line to shift. Specifically in this project, the LSPR of Au nanoparticles is used to transduce the interaction between two non-structural proteins responsible for viral genome replication, NS3 and NS5. This goal was accomplished by implementing an immobilization strategy for purified His-tagged NS3 proteins to the nanoparticle surface using a nickel-nitrilotriacetic acid (Ni-NTA) surface chemistry.

Introduction:

Surface plasmons are collective electron-density oscillations generated on the surface of metallic films. As the resonant excitation of these plasmons is highly dependent on the dielectric function of the material on the metal surface, monitoring the resonance condition serves as a suitable method to transduce dielectric changes in the near-field of the metallic surface. Based on this principle, surface plasmon resonance (SPR) biosensors have been utilized extensively over the last decade to characterize various biomolecular systems. Most significant among the benefits of SPR is the ability to monitor biomolecular interactions in real-time without the need for various labels that can generate less robust signals and putative effects on the interaction of interest. An extension of this technique, known as localized surface plasmon resonance (LSPR), uses noble metal nanoparticles. LSPR is gaining interest in part due to the availability of more reliable fabrication methods, adaptability for use in commercially available spectroscopic systems and enhanced sensitivity. In this work, nanoimprint lithography (NIL) is utilized to produce plasmonic constructs that are optically characterized using microextinction spectroscopy. We subsequently utilize a surface-modified Au nanoparticle array to detect interactions between two non-structural proteins NS3 and NS5 which are found in a *Flavivirus replicase* complex.

Fabrication Process:

Nanoimprint lithography (NIL), a mold-based fabrication

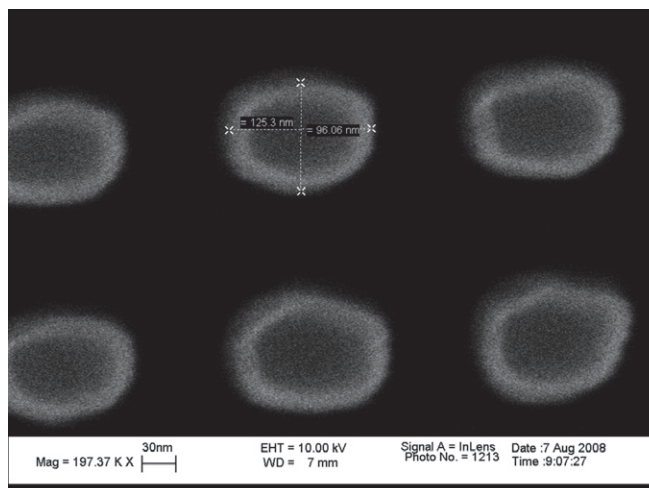


Figure 1: SEM of the nanoimprint mold.

method, was used to fabricate nanoscale patterns on glass. Glass substrates were cleaned in a 1:1 piranha solution of $\text{H}_2\text{O}_2:\text{H}_2\text{SO}_4$ for 20 minutes followed by a rinse in deionized water solution and dried using N_2 . Nanoimprint resist (MRi-8030) was spin-coated on the glass substrates to the appropriate thickness and then baked on a hot plate at 140°C for 5 minutes to remove residual solvent. A nanoimprinter (Nanonex, NJ) was used to imprint with a mold possessing an array of pillars with oval cross-sections (~ 130 nm by 100 nm) and gaps of ~ 90 nm (Figure 1) directly onto the prepared glass substrates.

The glass substrates were heated above the glass transition temperature of the nanoimprint resist (180°C) and pressure (670 psi) is used to create a conformal contact with the mold. After imprinting, a thin layer (~ 5 nm) of Ni was deposited using a shadow evaporation technique to promote undercutting and enhance the subsequent lift-off process. Residual layer removal was accomplished using a suitable O₂ plasma dry etching process (20 sccm; 50W; 20 mTorr). Following the etching, Au was deposited to the desired thickness on the glass substrates using an electron beam evaporator. Lift-off was performed by immersing the glass substrates in a beaker of acetone, and sonication when necessary. The finished samples were rinsed with methanol and IPA and dried with N₂. The resulting Au NP arrays were surface-modified with a Ni-nitrilotriacetic acid (Ni-NTA) surface chemistry in order to immobilize the purified His-tagged NS3 proteins onto the Au NPs. The surface modification technique was accomplished by the incubation of the Au NP substrates in solutions of cystamine, glutaraldehyde, N α ,N α -bis(carboxymethyl)-L-lysine hydrate (NTA), and nickel sulfate respectively.

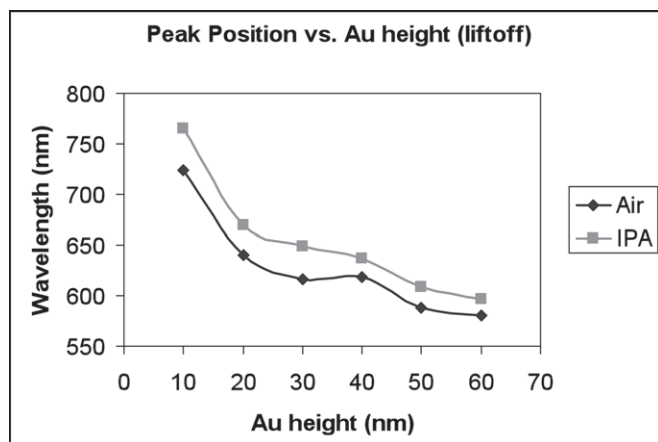


Figure 2: LSPR response of fabricated particle array (in air & IPA).

Experimental / Results:

The optical properties of the nanoparticles arrays were determined by an optical extinction measurement. These measurements were conducted using a Nikon TE300 Eclipse inverted microscope (40x objective) with transmitted broadband light focused into an optical fiber which was coupled to spectrometer (Ocean Optics HR4000). The SpectraSuite software package was used for all data acquisition and analysis. The first test was conducted on a variety of samples that were differentiated only by the height (10-60 nm) of the Au NPs. As shown in Figure 2, with increasing nanoparticle height there is a consistent resonance peak shift in the LSPR to shorter wavelengths (blue-shift). In addition, dielectric sensitivity was measured on the different heights by monitoring the LSPR response of the nanoparticles as

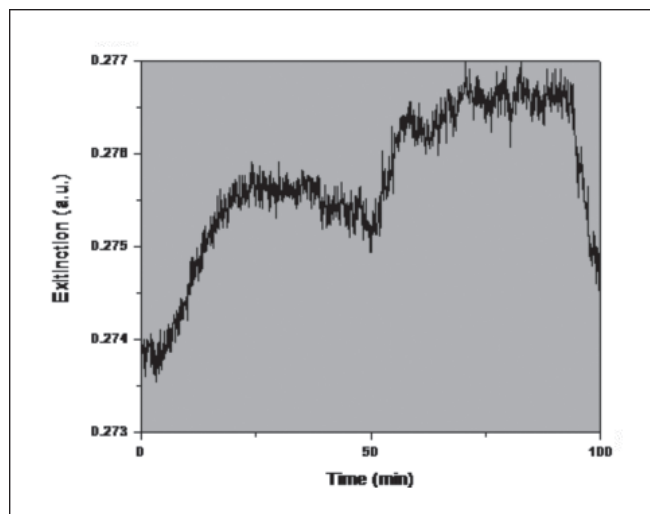


Figure 3: LSPR measurement of NS3 and NS5 interaction.

they interact with ambient air and isopropanol (IPA). Overall behavior shows an increase in wavelength (red-shift) with increasing refractive index. We observed that there is an optimal particle thickness (30 nm) that produces the highest environmental sensitivity.

Lastly, we tested the interaction of NS3 and NS5. A flow cell setup was used to flow NS3, NS3 buffer, NS5, and NS5 buffer respectively. As seen in Figure 3, the interactions of NS3 with the nanoparticle surface and with NS5 are clearly distinguishable based on the extinction vs. time graph (NS3 at ~ 0.0 min, NS3 buffer at ~ 25 min, NS5 at ~ 50 min, and NS5 buffer at ~ 75 min).

Conclusions / Future Work:

We successfully fabricated Au nanoparticles using NIL and have characterized their plasmonic properties. We applied our nanoparticles to detect interactions between NS3 and NS5. Optimization of the surface chemistry used to bind His-tagged proteins is being addressed and should lead to improved biosensor performance.

Acknowledgements:

I would like to thank Professor L. Jay Guo, Brandon Lucas, Dr. Sandrine Martin, Guo Nanogroup, Lurie Nanofabrication Facility, the National Science Foundation, Intel Foundation for funding, and the National Nanotechnology Infrastructure Network Research Experience for Undergraduates (NNIN REU) Program for all their help and support.