Zeptomol Level Vibrational Spectroscopy of Proteins in Lithographically Engineered Plasmonic Nano-antenna Arrays

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Introduction

Infrared absorption spectroscopy enables access to detailed bond specific information of bio-molecules by probing absorption bands in the mid-infrared spectral region (3-20 μm). While absorption cross-sections are nearly 10 orders of magnitude larger than corresponding Raman cross-sections, they are still small in comparison with those of fluorescent labels. Sensitivity improvements are therefore necessary for the method to be applicable to single molecular layer studies. One promising method, surface enhanced infrared absorption (SEIRA) spectroscopy, leverages the enhanced near-fields that accompany the plasmonic excitations of nanoscale metal particles **Error! Reference source not found.**. The bulk of previous SEIRA experiments, however, have obtained enhancements via chemically prepared or roughened metal surfaces. The stochastic nature of these substrates limits enhancement due to the poor spectral overlap of the plasmonic resonances with the vibrational modes of interest [1]. The result is weaker absorption signals and a lack of reproducibility in measurements. In contrast, engineered plasmonic antennas support well defined resonances that can be tuned throughout the mid-infrared in order to maximize field enhancement at the frequency corresponding to a given vibrational mode [2,3]. In addition, periodic lattices of antennas offer a means towards further near-field enhancements and can easily fabricated with most top down methods [3].

In this work, we demonstrate a collectively enhanced infrared absorption (CEIRA) spectroscopy technique using collective excitations of nano-antenna arrays. We show 10^4-10^5 fold enhancements of the amide-I and II backbone signatures of proteins and obtain absorption signals from zeptomole quantities of protein molecules [3].

Results and discussion

Collective excitations in well engineered nanoanntenna arrays can lead to near-field enhancements well beyond those of an individual antenna. This can be seen in Fig.1b, which is the near field distributions obtained by FDTD simulations. These resonances depend strongly on the phase delays experienced by light propagating in between particles in the array and therefore the array periodicity, *d*. The associated suppression of radiation damping results in a characteristic narrowing of the far-field response. For experimental demonstration, we fabricated periodic arrays of engineered antennas by electron beam lithography as shown in Fig. 1a. Reflectance spectra of the periodic antenna arrays (square lattice) are shown along with the spectra from a random arrangement (black curve), Fig. 1c. The line-width of antenna resonance for the d = 1.6 μm array is greatly reduced, in agreement with the enhancement predicted by the FDTD simulation.

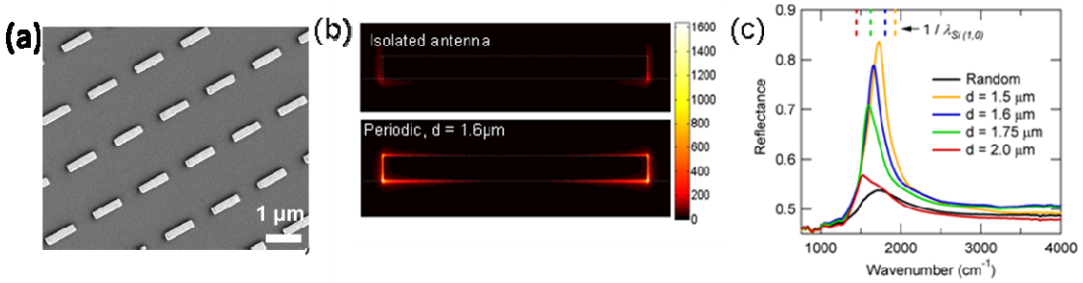

Fig. 1 *(a) SEM image of engineered nano-rod shaped antenna arrays. (b) FDTD simulated near-field intensity enhancement for 1100 nm long rods on Si substrates. (c) Reflectance spectra for 1100 nm long rods in arrays of varying periodicity.*

To observe the effect of the collective array resonances on the CEIRA enhancement, we coated the antenna arrays with a 2 nm thick film of the protein silk fibroin **Error! Reference source not found.**. The reflectance spectra of the antennas following coating with the protein show clear absorption features, as can be seen for the 1.6 μm periodic array in Fig. 2 (a). Subtracting the response of the antenna array alone from that of the protein coated array yields difference spectra closely resembling the protein absorption spectra (Fig. 2 (b)). The strong dependence of the signal strength on the array arrangement is evident. The magnitude of the absorption feature at 1660 cm^{-1} can be seen to correlate well with the narrowing of the far field response, with the 1.6 μm periodic array resulting in signal strength of 6.8 %, nearly an order of magnitude greater than that of the random array, 0.9%. By comparing this signal with that expected from a bare silk film and accounting for the fact that the enhanced signal results from a small region around the tip ends of the rods we have estimated the signal enhancement to be within the range of 10^4 – 10^5 [3].

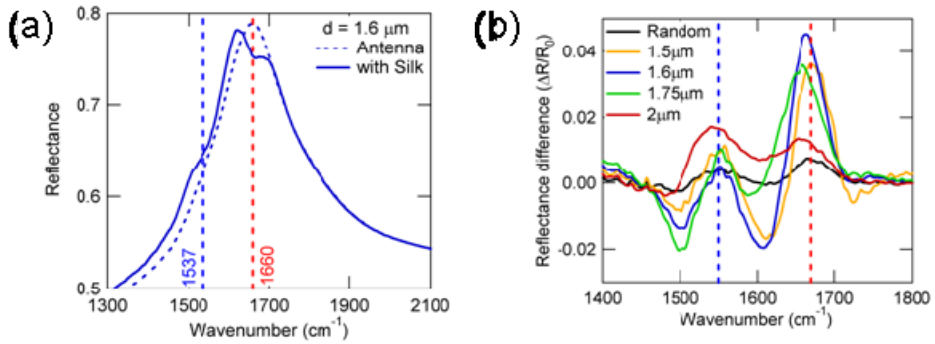

Fig. 2 *(a) Reflectance spectra for the d = 1.6 μm periodic antenna array and (b) difference spectra for randomly arranged and periodic antenna arrays.*

3. Conclusions

In conclusion, we have demonstrated the 10^4-10^5 fold enhancement of the backbone resonances of the protein silk fibroin through the use of periodic arrangements of infrared antennas. The detection volume at the tip ends of the rods can be calculated to contain ~ 300 zeptomoles for the entire array, corresponding to 145 molecules per antenna [3]. Given the large signal-to-noise ratios in our measurements, the observation of signatures from tens of zeptomole quantities of proteins, should be possible.

4. References

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