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Controlling the Fluctuating Tip-Enhanced Raman Spectra of Chloramben on Silver Nanocubes

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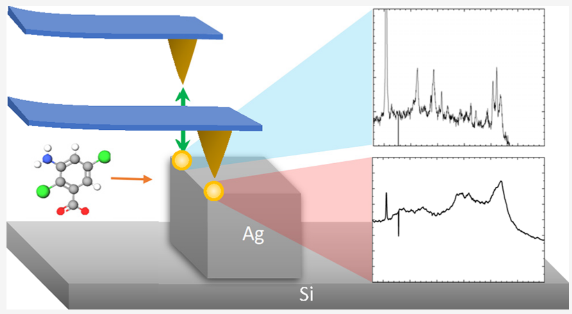
ABSTRACT: Tip-enhanced Raman (TER) scattering from molecules residing at plasmonic junctions can be used to detect, identify, and image single molecules. This is most evident for flat molecules interrogated under conditions of extreme temperatures and pressure. It is also the case for (bio)molecular systems that feature preferred orientations/conformations under ambient laboratory conditions. More complex molecules that can adopt multiple conformations and/or feature different protonation or charge states give rise to complex TER spectra. We illustrate how the latter can be controlled in the case of chloramben molecules coated onto plasmonic silver nanocubes. We show that characteristic molecular Raman spectra cannot be obtained when tunneling plasmons are operative, i.e., when the tip is in direct contact with the chemically functionalized plasmonic nanoparticles. We rationalize these observations and propose an approach to less invasive and hence more analytical TER spectral imaging.



INTRODUCTION

Tip-enhanced Raman (TER) spectral imaging is a powerful approach to chemical and chemical reaction imaging. Its benefits, stemming from the combination of scanning probe and (non)linear optical spectroscopy,[1,2] are perhaps most evident in measurements that take advantage of the stability of ultrahigh vacuum/ultralow temperature setups, where sub-molecular resolution has been demonstrated.[3−5] Even though the information content is blurred as a result of thermal fluctuations and instabilities, performing TER measurements under ambient laboratory conditions is appealing, particularly for applications in catalysis and bioimaging that often require interrogating solid−liquid interfaces.[6−8]

Ambient TER setups are typically built around an atomic force microscope (AFM).[9] Within the realm of AFM-TER measurements, most reported experiments are performed using contact mode feedback.[10] In other words, optical signals are recorded when the tip exerts force on the sample; see Figure 1. More modern approaches to TER spectral imaging take advantage of hybrid AFM feedback modes, wherein sample motion is achieved using tapping (or intermittent contact) feedback, and nano-optical signals are recorded when the tip is in contact with the sample.[11] This mode preserves sample integrity and prolongs the lifetime of the plasmonic probe, all while retaining an optimal optical enhancement.

Besides considerations that have to do with mechanical and first order electrostatic interactions between the tip and the sample, there are additional motivations for avoiding the use of contact mode or even hybrid tapping/contact mode feedback in TER spectral imaging.[11,12] This is particularly the case for substrates consisting of either extended or nanostructured plasmonic metals, where optimal enhancement and spatial resolution have been demonstrated. Indeed, the interplay between classical and quantum plasmons in plasmonic nanogaps[13] complicates the information content in TER, as discussed in recent reports from our group.[12] In our recent study, through TER mapping experiments performed either using tapping mode or contact mode feedback, we showed how molecular charging and optical rectification that are noticeable in the latter can be suppressed when tip−sample interactions are minimized.[12] This was associated with localized vs quantum plasmon driven TER using tapping vs contact mode feedback, as schematically illustrated in Figure 1.

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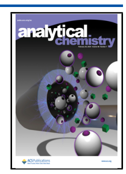

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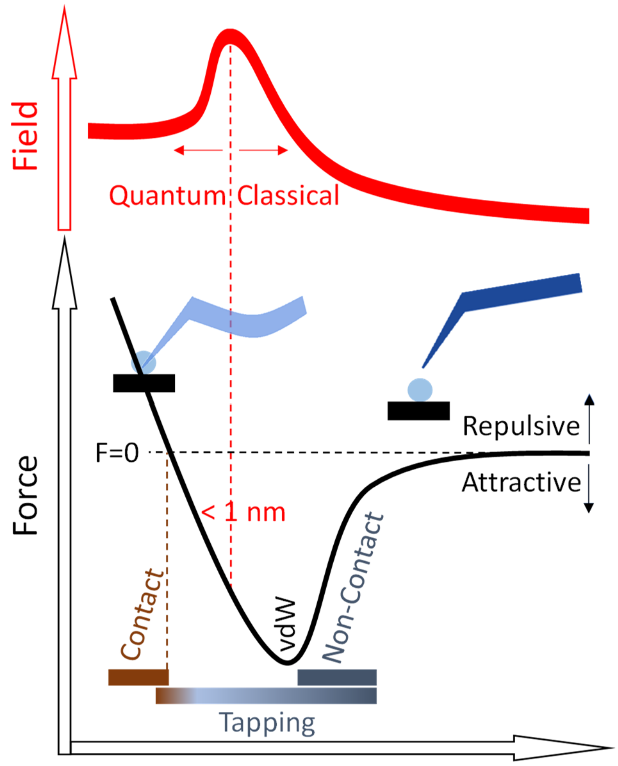

Figure 1. Schematic illustration of a typical AFM force−distance curve (lower black trace) and the expected local optical field enhancement as a function of tip−sample distance (upper red curve). The two curves are annotated to highlight the zero force line ($F = 0$), van der Waals (vdW) contact, (non)contact and tapping mode regimes, and approximate position of the transition from classical to quantum plasmons. Note that the circular blue dot between the tip and sample in the inset designates molecules (e.g., water) that lead to contact and that offset vDW and conductive (<1 nm) contact points.

Note that as TER signal magnitudes are sensitive not only to local fields but also to molecular scattering cross sections, the field picture is modified when molecular loads are taken into account. The most mundane consideration in this context is Raman scattering from neutral vs anionic species in the two plasmonic regimes depicted in the upper panel of Figure 1, where distinct molecular resonances may be operative.[14]

In this study, we initially set out to detect trace quantities of a herbicide, chloramben, via TER spectroscopy and imaging. The detection of herbicide residues (as well as pesticide residues and other metabolites) has traditionally involved the use of chromatography−mass spectrometry[15] and high-performance liquid chromatography.[16] Near-field spectroscopy draws interest as a plausible alternative. Besides general interest in the ultrasensitive detection and identification of pesticides and herbicides,[17−21] chloramben features an interesting chemical structure from fundamental and applied surface science/spectroscopy perspectives; see Figure 2. Namely, this molecule has two possibilities of binding to a plasmonic metal, herein a silver nanocube. The first possibility is binding through the NH_2 moiety, and the second involves the CO_2H group. These structural and binding motifs are common to many small biomolecules, including amino acids. Besides distinct binding geometries, different protonation and charge states are also possible for silver-bound chloramben molecules under ambient laboratory conditions. Modified TER surface selection rules, the possibility of multipolar TER scattering,

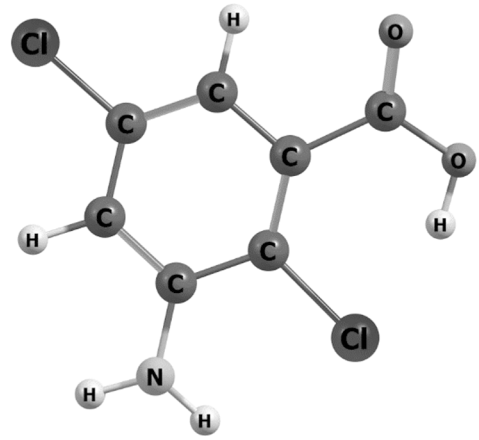

Figure 2. Molecular structure of chloramben (3-amino-2,5-dichlorobenzoic acid).

and optical rectification add further complexity to the recorded spectral patterns.[11] In the following, we show how all of these effects together lead to extreme spectral heterogeneity: the molecule is not recognizable from our recorded contact mode TER spectra.

■ RESULTS AND DISCUSSION

Simultaneously recorded AFM and TER maps of the edge of a chloramben-coated silver nanocube are shown in Figure 3. These images were recorded using a hybrid feedback mode: sample motion is accomplished using tapping mode feedback, whereas TER spectra are recorded when the tip is in contact with the substrate, i.e., above the zero force ($F = 0$) line in Figure 1. The AFM (Figure 3a) and TER (Figure 3b) images are both indicative of a complex tip morphology. Indeed, deviations from the expected field profile can be associated with artifacts both in AFM as well as TER imaging.[22] The more important observation is the extreme spectral heterogeneity across the map, which can be seen in Figure 3c. The recorded single-pixel spectra are highly congested, consisting of numerous sharp molecular lines that span the entire 500−1700 cm^{-1} spectral region. Note that these observations (Figure 3c) are quite general: extreme spectral heterogeneity was observed in the TER spectral images of various chloramben-coated surfaces and nanostructures; see Figure S1. In other words, these observations are not a function of any specific tip geometry or nanoparticle shape/makeup. None of the spectra we recorded can be reliably used to identify the chloramben molecules that we deposited onto the nanocube. It is interesting to stress that this is not the case for aromatic thiol derivatives interrogated under nearly identical conditions.[11] If one assumes that the operative local optical fields are similar in this study vs prior work irrespective of the molecular load, the observation of extreme spectral heterogeneity must have to do with our current choice of molecule. So, beyond the realm of functionalized aromatic thiols, a few dye molecules, and porphyrins immobilized in UHV chambers, how can one use TER as a noninvasive analytical chemical imaging and identification technique without having to overengineer the sample or tip?

Before attempting to offer one answer to the question raised above, we inspect the spatiotemporally averaged TER response in Figure 3c. This response is similar to traces often seen in diffraction-limited surface enhanced Raman scattering meas-

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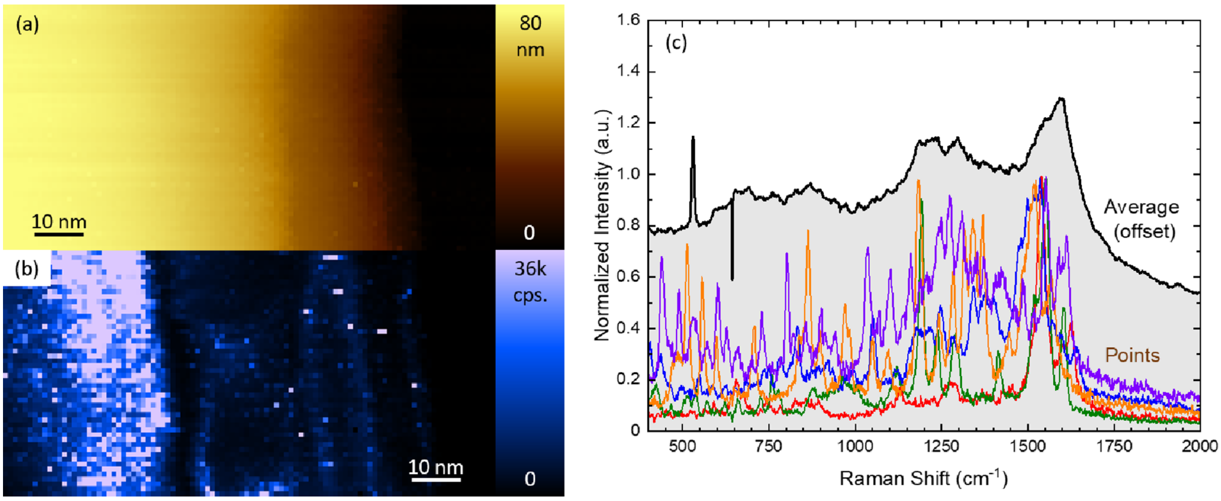

Figure 3. Simultaneously recorded AFM (a) and contact mode TER (b, binned in the 1500−1700 cm^{-1} spectral region) images of the edge of a chloramben-coated plasmonic silver nanocube. Selected single pixel TER spectra are shown in (c) along with the optical response that we obtained by spatial averaging over the entire area in (b). Parameters: 100 $\mu W/\mu m^2$ at the sample position, 1 nm lateral/vertical steps, 0.25 s/pixel time integration.

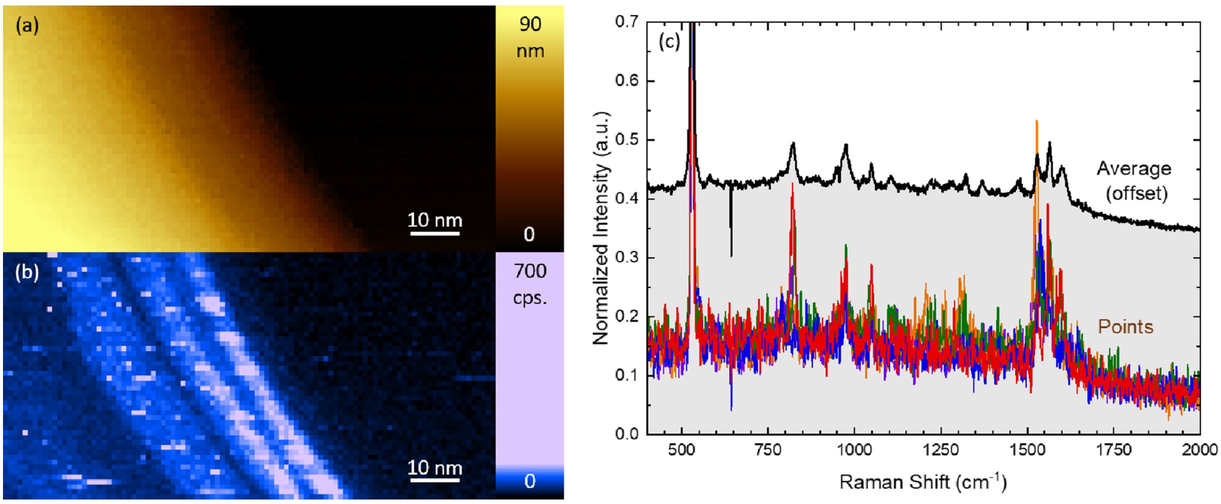

Figure 4. Simultaneously recorded AFM (a) and tapping mode TER (b, binned in the 1500−1700 cm^{-1} spectral region) images of the edge of a chloramben-coated plasmonic silver nanocube. Selected single pixel TER spectra are shown in (c) along with the optical response that we obtained by spatial averaging over the entire area in (b). Parameters: 100 $\mu W/\mu m^2$ at the sample position, 1 nm lateral/vertical steps, 0.5 s/pixel time integration.

urements. There, similar spectra are often associated with amorphous carbon that forms as a result of sample (photo)-damage.[23,24] In other instances, observations of broad and featureless spectra are assigned to flat binding orientations,[25] a classic example of which is enhanced Raman scattering from benzene molecules physiosorbed onto plasmonic metals.[26] Our present results clearly establish that extreme spectral heterogeneity can also lead to broadened background-like spectral profiles upon spatial and temporal averaging. This effect is often underappreciated in the plasmon-enhanced (surface-enhanced in particular) Raman community but should be considered. In the same vein, the analysis of surface-enhanced and TER backgrounds must also account for molecular processes that lead to broad background-like features.

Inspired by recent observations[12] that were highlighted in the introductory comments, we repeated the measurements above using the same tip and nanoparticle, but now using tapping mode feedback; see Figure 4. The stark contrast between the well-defined spectra in Figure 4c and the disparate spectra in Figure 3c is immediately evident. The spectra we observe in Figure 4c can be assigned to chloramben on the basis of standard gas phase density functional theory simulations (Figure S2). The high spatial resolution observed in Figure 4b (∼2 nm) suggests that this approach may be used to readily detect and identify yoctomolar concentrations of chloramben molecules under ambient laboratory conditions. The sample preparation that consists of simply drop-casting solvated chloramben onto a substrate is an added advantage.

A side-to-side comparison between the tapping mode (Figure 4) and contact mode (Figure 3) spectra allows us to draw a few key conclusions. Molecular aspects alone, such as the possibility of multiple orientations/conformations, cannot explain the observed spectral heterogeneity in contact mode vs uniform spectral patterns in tapping mode TER spectral imaging. The same can be said of mechanical deformations,

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i.e., molecular flattening, when the tip exerts force on the silver-bound molecules. Namely, what we observe are sharp molecular lines that undergo extreme spectral diffusion. A plausible explanation for our observations requires invoking the distinct (non)local fields that drive the optical processes in tapping vs contact mode measurements. As schematically illustrated in Figure 1 and discussed in prior reports,[12] tunneling plasmons drive TER when conductive contact is established between the probe and the surface. The onset of tunneling is accompanied by optical rectification and molecular charging. Whereas the first is not expected to significantly alter the spectra barring Stark reporters (e.g., nitrile moieties),[27] the second can have more significant effects on the observables. For instance, the possibility of resonance enhancement upon molecular charging has been recently invoked in the context of plasmon-enhanced Raman scattering.[14] Similarly, we propose herein that molecular charging shifts molecular resonances into the spectral window that we access with our excitation wavelength. We propose that this is at the core of the observed spectral variations. Distinct conformations/orientations/binding motifs that may alter the static/dynamic polarizabilities of silver-bound molecules also contribute to our observation of extreme spectral diffusion in contact mode TER experiments. The suppression of molecular charging in tapping mode TER measurements on the other hand leads to recognizable and assignable molecular line spectra that can be understood on the basis of simple simulations, as shown in Figure S2.

■ CONCLUSIONS

In conclusion, tapping mode TER spectral imaging is the recommended approach for the detection and identification of molecules that do not adopt well-defined binding geometries or conformations. Recent observations of brighter signals in tapping mode vs contact mode TER experiments[28] provide additional motivation for adopting the former in Raman nanoscopy. A study that described how to optimize signal levels in tapping mode nano-optical measurements[10] is equally important in this context. Alternatively, it would be attractive to develop modified contact mode feedback methods, wherein the force exerted on the sample is below the zero force line. Here, the objective would be to maintain 1−2 nm separation between the tip and substrate throughout the measurements using a (negative or attractive) force for feedback. Novel scalable approaches to functionalizing metal tips and substrates with ultrathin (1−2 nm) insulator layers are routinely employed in UHV-TER systems. It would be interesting to explore the same tactic for measurements performed under ambient laboratory conditions.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acs.analchem.3c05542.

> Experimental details, additional TER spectra recorded using different tips and plasmonic constructs, and a comparison between computed gas phase density functional theory and experimental Raman spectra (PDF)

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Notes

The authors declare the following competing financial interest(s): Y.G. has equity interest in Klar Scientific.

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■ REFERENCES

(1) Stöckle, R. M.; Suh, Y. D.; Deckert, V.; Zenobi, R. *Chem. Phys. Lett.* **2000**, *318* (1), 131−136.

(2) Kurouski, D.; Dazzi, A.; Zenobi, R.; Centrone, A. *Chem. Soc. Rev.* **2020**, *49* (11), 3315−3347.

(3) Zhang, R.; Zhang, Y.; Dong, Z. C.; Jiang, S.; Zhang, C.; Chen, L. G.; Zhang, L.; Liao, Y.; Aizpurua, J.; Luo, Y.; Yang, J. L.; Hou, J. G. *Nature* **2013**, *498* (7452), 82−86.

(4) Lee, J.; Crampton, K. T.; Tallarida, N.; Apkarian, V. A. *Nature* **2019**, *568* (7750), 78−82.

(5) Schultz, J. F.; Mahapatra, S.; Li, L.; Jiang, N. *Appl. Spectrosc.* **2020**, *74* (11), 1313−1340.

(6) Britz-Grell, A. B.; Saumer, M.; Tarasov, A. *J. Phys. Chem. C* **2021**, *125* (39), 21321−21340.

(7) Wang, Y.-Z.; Wang, J.; Wang, X.; Ren, B. *Curr. Opin. Electrochem.* **2023**, *42*, 101385.

(8) Wang, X.; Huang, S.-C.; Huang, T.-X.; Su, H.-S.; Zhong, J.-H.; Zeng, Z.-C.; Li, M.-H.; Ren, B. *Chem. Soc. Rev.* **2017**, *46* (13), 4020−4041.

(9) Verma, P. *Chem. Rev.* **2017**, *117* (9), 6447−6466.

(10) Umakoshi, T.; Kawashima, K.; Moriyama, T.; Kato, R.; Verma, P. *Sci. Rep.* **2022**, *12* (1), 12776.

(11) El-Khoury, P. Z. *Acc. Chem. Res.* **2021**, *54* (24), 4576−4583.

(12) Wang, C.-F.; O'Callahan, B. T.; Kurouski, D.; Krayev, A.; Schultz, Z. D.; El-Khoury, P. Z. *J. Phys. Chem. Lett.* **2020**, *11* (15), 5890−5895.

(13) Zhu, W.; Esteban, R.; Borisov, A. G.; Baumberg, J. J.; Nordlander, P.; Lezec, H. J.; Aizpurua, J.; Crozier, K. B. *Nat. Commun.* **2016**, *7* (1), 11495.
(14) Sloan-Dennison, S.; Zoltowski, C. M.; El-Khoury, P. Z.; Schultz, Z. D. *J. Phys. Chem. C* **2020**, *124* (17), 9548−9558.
(15) Alder, L.; Greulich, K.; Kempe, G.; Vieth, B. *Mass Spectrom. Rev.* **2006**, *25* (6), 838−865.
(16) Michel, M.; Buszewski, B. *J. Liq. Chromatogr. Relat. Technol.* **2002**, *25* (13−15), 2293−2306.
(17) Dong, X.; Tang, Z.; Zhang, H.; Hu, Y.; Yao, Z.; Huang, R.; Bai, J.; Yang, Y.; Hong, W. *Anal. Chem.* **2023**, *95* (26), 9831−9838.
(18) Li, B.; Wang, Z. *ACS Appl. Mater. Interfaces* **2023**, *15* (26), 31824−31835.
(19) Sheng, E.; Lu, Y.; Xiao, Y.; Li, Z.; Wang, H.; Dai, Z. *Biosens. Bioelectron.* **2021**, *181*, 113149.
(20) Lee, D. K.; Kim, G.; Kim, C.; Jhon, Y. M.; Kim, J. H.; Lee, T.; Son, J. H.; Seo, M. *IEEE Trans. Terahertz Sci. Technol.* **2016**, *6* (3), 389−395.
(21) Ashrafi Tafreshi, F.; Fatahi, Z.; Ghasemi, S. F.; Taherian, A.; Esfandiari, N. *PLoS One* **2020**, *15* (3), No. e0230646.
(22) Wang, C.-F.; O'Callahan, B. T.; Arey, B. W.; Kurouski, D.; El-Khoury, P. Z. *J. Phys. Chem. C* **2022**, *126* (8), 4089−4094.
(23) Heck, C.; Kanehira, Y.; Kneipp, J.; Bald, I. *Molecules* **2019**, *24* (12), 2324.
(24) Zong, C.; Xu, M.; Xu, L.-J.; Wei, T.; Ma, X.; Zheng, X.-S.; Hu, R.; Ren, B. *Chem. Rev.* **2018**, *118* (10), 4946−4980.
(25) El-Khoury, P. Z.; Peppernick, S. J.; Hu, D.; Joly, A. G.; Hess, W. P. *J. Phys. Chem. C* **2013**, *117* (14), 7260−7268.
(26) Gao, X.; Davies, J. P.; Weaver, M. J. *J. Phys. Chem.* **1990**, *94* (17), 6858−6864.
(27) Wang, H.; Yao, K.; Parkhill, J. A.; Schultz, Z. D. *Phys. Chem. Chem. Phys.* **2017**, *19* (8), 5786−5796.
(28) Bartolomeo, G. L.; Zhang, Y.; Kumar, N.; Zenobi, R. *Anal. Chem.* **2021**, *93* (46), 15358−15364.