Super resolution far field infrared microscopy

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Abstract. We present and compare the performances of label-free super-resolution infrared microscopy based on non-linear properties intrinsic to all vibrational transition in three configurations such as pump-probe, differential absorption, and attenuated total reflection configurations. We then highlight experimental demonstrations achieved in label-free super-resolution chemical imaging that succeed in exploiting this novel concept.

1. Introduction

The widespread optical techniques used for chemical imaging are coherent anti-Stokes Raman spectroscopy (CARS) [1], sum-frequency generation (SFG) spectroscopy [2], and Fourier-transform infrared absorption microscopy (IRAS). Vibrational microscopy based on IR absorption provides a simple way to perform label-free imaging with chemical selectivity. It has been widely applied to study biological tissues and cells [3-4]. Because of the longer mid-IR wavelengths spanning from 3 μm to 20 μm, the IR microscopy suffers from a low spatial resolution in comparison to microscopy operating in the visible range. [1]. In 1994, Hell demonstrated the possibility of bypassing this Abbe criterion limit in fluorescent microscopy by using stimulated-emission depletion (STED) [5]. These approaches require the use of fluorescent chromophores. Label-free sub-diffraction chemical imaging was first suggested through the theoretical use of CARS by suppressing the vibrational Raman coherence [6]. However, the requirement for vibrational-electronic coupling in the sample means that this can only be applied to specific samples. Super-resolution has also been demonstrated in photoacoustic microscopy [7]. We recently reported that super-resolution can be achieved in optical microscopy, label free, by exploiting a non-linearity intrinsic to any optical transitions [8-10]. Hereafter, we briefly review the proposed concepts and compare, with particular attention to infrared microscopy, the spatial resolutions that can be achieved in the three configurations that is pump-probe, differential absorption, and ATR (Attenuated Total Reflection). We then highlight recent progress achieved in label-free super-resolution chemical imaging that exploits these novel concepts.
2. Model

We evaluate numerically the spatial resolution of the microscope in three configurations. The first configuration implies irradiating the sample using a pump pulse with a doughnut profile intensity profile and a probe beam with Gaussian profile. The doughnut beam profile can be obtained using a vortex phase plate or a spiral phase mirror \[11\]. This last has the advantages of operating over a large spectral range. In the second configuration, the super resolution is achieved by comparing the distortion of the images acquired using saturating pulses with two different intensity profiles, doughnut and Gaussian. The third one implies irradiating the sample using two IR pump beams with same intensity counter-propagating inside a prism and overlap at its base surface with incidence angle larger than the critical angle. The standing evanescent wave bleaches locally the adsorbed layer vibrational transition. The adsorbate absorbance is further probed by a third beam incident perpendicularly on the interface.

We adopt the absorbance proprieties to a layer of CH$_3$-(CH$_2$)$_{17}$-SiCl$_3$ deposited on mica \[12\] and considering an infrared beam tuned in resonance with the dominant CH$_2$ symmetric stretching of the alkane chain, the infrared dipole moment is of 0.12 Debye at 2917 cm$^{-1}$. The infrared pulse duration is set to 1 ps to match the experimental bandwidth of ~10 cm$^{-1}$ and is smaller than typical vibrational relaxation times. The saturation of the optical transition is evaluated using the optical Bloch equations for a two level system \[10\]:

$$\frac{d(\rho_{22}-\rho_{11})}{dt} = -\left(\frac{2B_{12}}{\epsilon_0 \Gamma_\chi} I(t) + \Gamma_\chi\right)(\rho_{22}-\rho_{11}) - \Gamma_\chi$$,

(1)

With

$$B_{12} = \frac{-\frac{\partial \mu_n}{\partial Q_n}}{\epsilon_0 \epsilon \Gamma_\chi h}$$,

where $\epsilon_0$, $c$, and $h$ are the dielectric constant of vacuum, the speed of light and the Planck constant, respectively.

The Einstein coefficient of equation (1) can be evaluated directly from the adsorbate vibration absorbance ($Abs$) and oscillator density according to

$$\frac{B_{12}}{\epsilon} = \frac{Abs}{D_v \hbar}$$

(2)

From the IRRAS data of octadecyltrichlorosilane adsorbed on a mica-substrate \[12\], we can deduce that $D_v = 6.3 \times 10^7 \mu$m$^{-2}$ corresponds to 17 CH$_2$ groups per molecule and per 27 A$^2$ and Abs $\sim$0.5 %, leading to $B_{12}/\epsilon = 1400 \mu$m$^2$ kHz$^{-1}$.

3. Results and discussions

Figures 1 show the CH$_2$ vibration excitation response to the saturating pulse fluence. The near-horizontal asymptote of the curve in Figure (1) at the right, observed for fluences >0.7 nJ/µm$^2$, corresponds to the transition bleaching that is exploited to improve the microscope spatial resolution.

Figure 2 at the left shows the PSF (point spread function)-FWHM as a function of the pulse energy for the microscope NA equal 0.85 while Figure 2 at the right shows an example of the microscope PSF for a saturating pulse energy of 40 nJ. For these calculations, the spiral phase mirror (12 mm in diameter with a central hole of 2 mm in diameter) and the optics are irradiated homogenously. The beam incidence on the spiral phase mirror is set to 15° which does not induce significant asymmetry in the doughnut beam profile. Therefore, in the absence of a saturating pump, the microscope PSF is an Airy function with a first minimum radius of 0.61×λ/NA = 2.5 µm and an FWHM of 2.16 µm. The need to discern the probe pulse from the saturating pump pulse at the detector is an experimental difficulty which can be circumvented by a second configuration called differential infrared absorption microscopy. It relies on...
the observation of the distortions of the microscope image when using laser pulse fluencies in the bleaching regime but with different intensity profiles [10], because these distortions contain the super-resolution information.

Figure 1. At the left. Population inversion to the optical transition according to equation (1). At the right. Absorbance saturation for the probe pulse emitted 1 ps after the saturating pump pulse.

Figure 2. At the left. PSF-FWHM of the microscope for pump-probe, differential and ATR configurations. The PSF at the red cross, which corresponds to the limit in spatial resolution gain of the differential configuration, are given in Fig. 2 at the right for pulse energy of 40 nJ.

Figure 3 (a) illustrates the experimental arrangement of an infrared microscope operating in ATR. Figure 3 (b) shows the PSF of the microscope using the two beam profiles (doughnut and Airy) obtained using the same experimental configuration. In this configuration, the ripples in the beam profile will severely limit the spatial resolution. These ripples are intrinsic to the limited diameter of the optics and are present in both the Airy and doughnut profiles. Therefore, in differential infrared absorption microscopy, improvement in the spatial resolution is practically limited to a factor of four to five-compared to the Abbe criterion. This poor gain results from the ratio of the Airy function first ring intensity (1/57) compared to the Airy function maximum and the ring intensity allowed in the PSF (1/10). It should be noted that the ripples are more important in the present calculations than in the original article [10], where the microscope PSF was approximated by a Gaussian profile. Finally, the best spatial resolution can be achieved by combining these super-resolution effects with the ATR configuration. In this later scheme, the spatial resolution is improved by both of the refractive index of the ATR prism and the saturation of the vibration. For numerical analysis, we assumed a hemisphere of ZnSe with a refractive index of 3.4. The ATR condition is fulfilled if the incidence angle is larger than...
17°. Differential infrared microscopy is precluded in the ATR condition because the larger hole in the spiral phase mirror further enhances the ripples in the PSF which further limits the possible resolution gain. The obtained FWHM PSF in the ATR configuration is about 3.4 times smaller than that achieved with the pump-probe scheme.

**Figure 3.** At the left. Experimental arrangement for a super-resolution infrared microscope operating in ATR. The probe beam propagates through the spiral phase mirror center. At the right. PSF of the microscope for a peak intensity of 57 nJ.µm^-2.

**Figure 4.** Nano-structure defects in a single layer of graphene as measured by absorption microscopy. A: Absorbance of graphene at 1020 nm acquired using a pump-probe scheme without super-resolution. B: The same image with improved spatial resolution using a saturating doughnut beam to bleach the absorption (Ref. [16]) C: shows the plot profiles of the line between the two arrows drawn in the images A and B.

The damage threshold of the substrate and/or of the adsorbate will limit the performance of the super-resolution microscope. For CaF_2 exposed to picosecond mid-IR pulses, the damage threshold is 6.5 J/cm^2 = 65 nJ/µm^2 [13]. In the case of biological tissues, thermal stress is considered acceptable if the beam intensity is below 0.05 mW/µm^2 [14]. Using a 1-kHz rate laser would limit the pump laser peak intensity to the range of 50 nJ/µm^2.

The first two experimental cases described in Refs [15, 16] adopt the first pump-probe scheme for imaging graphene nano-structures. Wang [15] chose pump and probe frequencies of 1064 nm and 830 nm, respectively, and demonstrated an improvement of the spatial resolution to 225 nm, in comparison to 385 nm when no super-resolution is applied. Bianchini [16] recently improved the super-resolution on graphene nano-structures to 237 nm by using pump and probe frequencies of 800 nm and 1020 nm, respectively, as illustrated on the Figure 4. Liu [17] was the first to adopt the pump-probe configuration with all beams generated at the same frequency. The microscope was operated at 680 nm.
corresponding to the CdSe interband resonant transition. Using 2-ps pulses with energies of < 0.1 nJ, Liu demonstrated a super-resolution of the order of 150 nm.

4. Conclusion
The performances of a label free super-resolution infrared microscope were evaluated and compared on the model system constituted by methylene vibration in three different schemes such as pump-probe, differential absorption and ATR. The spatial resolution obtained in the ATR configuration is about 3.4 times smaller than that achieved in the pump-probe scheme. A super resolution in the pump probe scheme is demonstrated experimentally [15-17].

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References
[1] Evans C L and Xie X S 2008 Annu. Rev. Anal. Chem. 1 883–909
[2] Guyot-Sionnest P, Hunt J H and Shen Y R 1987 Phys. Rev. Lett. 59 1597–1600
[3] Holman H Y N, Bechtel H A, Hao Z, and Martin M C 2010 Anal. Chem. 82 8757–65
[4] Holman H Y, Miles R, Hao Z, Woezi E, Anderson L M and Yang H 2009 Anal. Chem. 81 8564–70
[5] Hell S W and Wichmann J 1994 Opt. Lett. 19 780–82
[6] Beeker W P, Groß P, Lee C J, Cleff C, Offerhaus H L, Fallnich C, Herek J L and Boller K J 2009 Opt. Exp. 17 22632–38
[7] Yao J, Wang L, Li C, Zhang C and Wang L V 2014 Phys. Rev. Lett. 112 014302
[8] Silien C, Liu N, Hendaoui N, Tofail S A M and Peremans A 2012 Opt. Exp. 20 29694–704
[9] Pita I, Hendaoui N, Liu N, Kumbham M, Tofail S A M, Peremans A, and Silien C 2013 Opt. Exp. 21 25632–42
[10] Hendaoui N, Mani A, Liu N, Tofail S M, Silien C and Peremans A 2017 Opt. Com. 382 574–579
[11] Ghai D P 2011 Appl. Opt. 50 (10) 1374–81
[12] Guzonas D A, Hair M L, and Tripp C P 1990 Appl. Spect. 44 290–293
[13] Lee H 2007 J. Mech. Sci. Technol. 21 1077–82
[14] Holman H YN, Martin M C and McKinney W R 2003 J. Biol. Phys. 29 275–286
[15] Wang P, Slipchenko M N, Mitchell J, Yang C, Potma E O, Xu X and Cheng J X 2013 Nat. Photonics 7 449–453
[16] Bianchini P, Korobchevskaya K, Zanini G and Diaspro A 2018 20th International Conference on Transparent Optical Networks (ICTON) 1–4
[17] Liu N, Kumbham M, Pita I, Guo Y, Bianchini P, Diaspro A, Tofail S A M, Peremans A and Silien C 2016 ACS Photonics 3 478–485