Biological imaging using super resolution stimulated Raman scattering microscopy

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Far-field super resolution imaging has allowed for a better understanding of the complex machinery of biological systems. Recently, a growing interest has emerged to use label-free Raman-based techniques exploiting the intrinsic nonlinear vibrational response of molecules [1]. Nevertheless, proposed super-resolution techniques in Raman processes either require high laser powers that may be phototoxic for biological specimens, or reduce the excitation volume resulting in lower signal levels [2]. Moreover, as light goes deeper inside tissues, wavefront distortion arises inside a scattering medium generating a speckle pattern, hence precluding most super resolution techniques to be used at depth.

Here, we present a single-pixel blind structured illumination microscopy technique (Blind S⁳) that is able to go beyond the diffraction limit of the conventional stimulated Raman scattering (SRS) microscope. The technique uses speckles as structured illumination patterns and a second beam to generate a stimulated Raman scattering process. After various images taken, we reconstruct a super resolved image without knowing the exact speckle illumination used to illuminate the sample [3]. The aforementioned super resolution technique has been demonstrated on polystyrene beads and is compatible with biological specimens imaging (see Fig. 1).

Fig. 1 HeLa cell lipid droplet imaging. (a) Large field of view. (b) Two zoomed-in region of interests imaged with conventional SRS (left) and blind S³ techniques. (c) Line profiles corresponding to the dashed lines of (b).

References