

Stimulated Raman Spectroscopic Imaging by Microsecond Delay-line Tuning

Chien-Sheng Liao¹ and Ji-Xin Cheng^{1,2,3,4,5}

¹Weldon School of Biomedical Engineering, ²Department of Chemistry, ³Purdue University Center for Cancer Research, ⁴Birck Nanotechnology Center, ⁵School of Electrical and Computer Engineering, Purdue University, West Lafayette, IN 47907, USA

Coherent Raman scattering (CRS) imaging techniques, including coherent anti-Stokes Raman scattering (CARS) and SRS, are powerful tools to visualize the spatial *distribution* of molecules in cells or tissues [1]. To resolve overlapped Raman bands in biological samples, there has been a great effort in developing spectroscopic CRS imaging technology. One efficient way to utilize the bandwidth of femtosecond pulses for spectroscopic measurement is to linearly chirp the pump and Stokes pulses and focus their entire bandwidth into a narrow spectral region [2]. Therefore, each temporal delay between the chirped pulses corresponds to a Raman shift. Here we implement a high speed resonant mirror to scan the temporal delay between two chirped pulses for SRS spectroscopic imaging. By synchronization of the resonant mirror with galvo-mirrors used for imaging scan, we collected an SRS spectrum, covering $\sim 200\text{ cm}^{-1}$ determined by the excitation pulse width, at each pixel of an image within $83\ \mu\text{s}$. Owing to the microsecond spectral acquisition speed, we successfully acquired chemical images from highly dynamic organelles in live *C. elegans* (Figure 1).

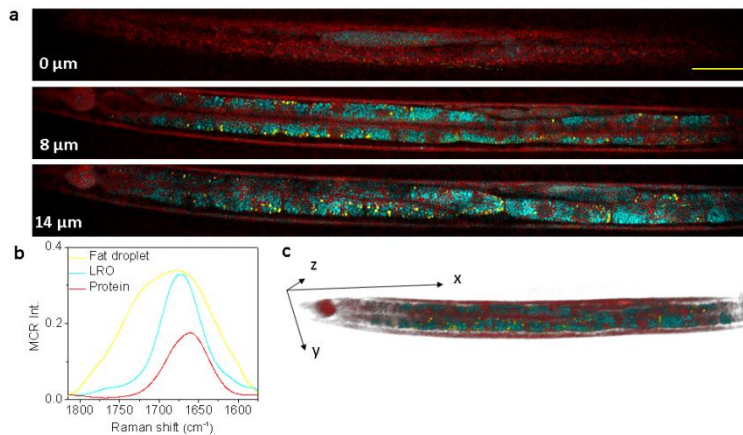


Figure 1. SRS spectroscopic images of *C. elegans* at 0, 8 and 14 μm depths from the surface. (a) MCR output concentration map at the depth of 0, 8 and 14 μm from the surface. (b) MCR output spectra. (c) 3-D reconstructed image of MCR output concentration maps at 12 depths. Scale bar: 30 μm

References

1. A. Zumbusch, G. R. Holtom, and X. S. Xie, Phys. Rev. Lett. 82, 4142-4145 (1999).
2. C. W. Freudiger, W. Min, B. G. Saar, S. Lu, G. R. Holtom, C. He, J. C. Tsai, J. X. Kang, and X. S. Xie, Science 322, 1857-1861 (2008).