Rapid Coherent Raman Spectroscopic Fingerprint Imaging

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Abstract

Raman spectroscopy is a powerful technique for label-free identification of chemical species within biological samples. Spontaneous Raman scattering has been extensively applied to a variety of tissues types and pathologies, but with typical spectral acquisition times of (0.1 to 10) s, it is not a practical method for imaging.

Broadband coherent anti-Stokes Raman scattering (BCARS) provides signal over the full Raman spectrum of interest, including the typically weak Raman “fingerprint” region, which is important for chemical discrimination, and has done so at speeds 50X faster than spontaneous Raman, making imaging practical. We have recently built a new BCARS instrument that exhibits an unprecedented combination of speed, sensitivity, and chemical selectivity. The system obtains Raman ~20X faster than previous BCARS instruments, (> 1000 times faster than spontaneous Raman methods), with very good spectral quality. (The images below were obtained in 2 minutes.) I will discuss innovations that have facilitated recent progress, and some applications.

(left) BCARS image from healthy murine liver. Collagen, DNA, and general protein are shown, but elastin, and other structural proteins can also be highlighted based on the Raman spectra.
(right) Same sample, intrinsic 2-photon fluorescence image and second harmonic generation image, highlighting elastin and collagen, respectively.

The BCARS image above of glioblastoma in a mouse, required no staining, but provides chemical sensitivity sufficient to identify accumulations of DNA, red blood cells, structural proteins of various types, accumulated metabolites, and much more.