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Correlative synchrotron infrared spectroscopy and super-resolution fluorescence microscopy for the detection of cellular DNA damage

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Single molecule localization microscopy (SMLM) and Synchrotron Fourier transform infrared (S-FTIR) spectroscopy are two techniques capable of elucidating unique and valuable detail and are especially suited for interrogation of biological samples. SMLM provides images of the structures and distributions of targeted biomolecules at spatial resolutions up to an order of magnitude better than the diffraction limit, whereas S-FTIR spectroscopy objectively measures the holistic biochemistry of an entire sample thereby revealing any variations in overall composition.

Previously, we have correlated these two techniques to probe the biochemical changes that common cell fixation and labelling methods cause, characterizing crosslinking and dehydration-induced losses of biomolecular composition and perturbation to cellular ultrastructure. More recently, we have expanded these studies to investigate the effects of drugs that cause DNA replicative and transcriptional stress.

In this study we acquired S-FTIR spectra of single drugged live cells alongside SMLM super resolution images of these cells after fixation, visualizing the DNA damage sites and their associated repair proteins. The complementary nature of these techniques allowed us to detect subtle changes to the cellular metabolism as well as the chromatin structure. Remarkably, we were even able to differentiate undamaged cells from those treated with low drug dosages that cause damage usually undetectable by conventional methods. These studies strikingly demonstrate the potential sensitivity of these combined techniques for the correlated detection of biochemical changes while also highlighting the types of structural and compositional changes that could only be revealed by one of the two techniques.

Primary author(s) : Dr WHELAN, Donna (La Trobe University); Dr BELL, Toby (Monash University); Mr ROZARIO, Ashley (Monash University); Ms MIRIKLIS, Esther (Monash University)

Presenter(s) : Dr WHELAN, Donna (La Trobe University)

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