Time Resolved Raman Microscopy Imaging of Human LnCaP Cells

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Raman images of living LnCaP cancer cells were constructed by plotting the concentration of various cellular components for each position of the exciting beam. Spectra are taken by scanning the sample across the laser beam [1]. Some attractive points of this technique are that it needs no special dyes or specific excitation wavelengths and also that a single scan contains information on all different chemical components. Consecutive scans, performed on the same cell as biochemical changes occur, provide time and space resolved information on cellular processes. To this date an example has been demonstrated where a chemical change signalling cell death was detected in a Raman image at an earlier time than was possible with a fluorescence image [2]. Real time spectroscopic studies of cancer cells yielding detailed chemical information will be invaluable for testing the efficacy of apoptotic or cytotoxic agents.

LnCaP cells grow attached to surfaces, however, to produce Raman images of cells which are naturally in suspension (such as blood cells) some method is needed to immobilise the cells. Optical Trapping (OT) experiments performed on budding S. cerevisiae cells trapped using only one beam showed that after some time the trapped cell can flip by 90 degrees ending up parallel with the trapping beam propagation direction. In this case, because of this movement relative to the excitation beam Raman images cannot be acquired.

Our current system uses a dual beam OT system to manipulate cells and perform spectroscopy [3]. We plan to expand this system to include Holographic Optical Tweezers (HOT) to provide multiple tweezing sites which will trap cells by using a calculated number of beams around the periphery of each cell to immobilise them over the course of the experiment. The HOT system can then be automated to scan the sample across the exciting beam to provide a whole cell image. We plan to use HOT to recover the same level of spatially resolved information from floating cells as from attached cells. Data will be presented for time and space resolved Raman imaging of live cells.

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