

MAKING COLOURFUL SENSE OF RAMAN IMAGES OF SINGLE CELLS

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Raman mapping is a powerful technique for single cell analysis, providing vast amounts of spatially resolved biochemical data from which cellular composition and sub-cellular components can be identified. The importance of single cell analysis for many areas of biomedical research, including cell biology, neurobiology, pharmacology and developmental biology means that the label-free, non-destructive technique of Raman imaging, with the potential of live cell imaging, has a pivotal role in the future of single cell imaging. The label-free, non-destructive technique provides a unique opportunity to monitor protein, DNA, RNA and lipid content as well as spatial distribution in live cells as they grow and migrate across, into and through 3D scaffolds.

However, in the field of Raman spectroscopy we are at risk of undermining the routine use of Raman mapping due to a lack of consistency and transparency in the way false-shaded Raman images are constructed. The successful construction of a Raman map requires the generation of a false-colour image in which each pixel is shaded according to the relative intensities of the defined Raman spectral regions or peaks, or specific components such as principal component scores. Yet standards for the way shading is applied are presently lacking and this lack of objectivity means there is very real potential that if false-colouring is not carefully carried out data may be over-interpreted or important details may be inadvertently missed. Only by developing and carefully applying robust protocols will the power of Raman imaging for live cell analysis be truly appreciated.