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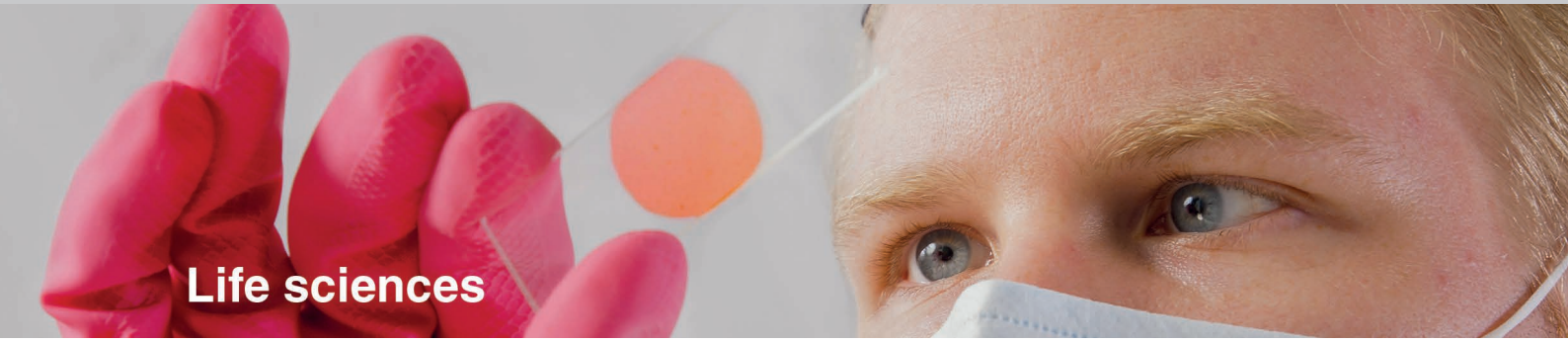
Redox biology with the inVia confocal Raman microscope

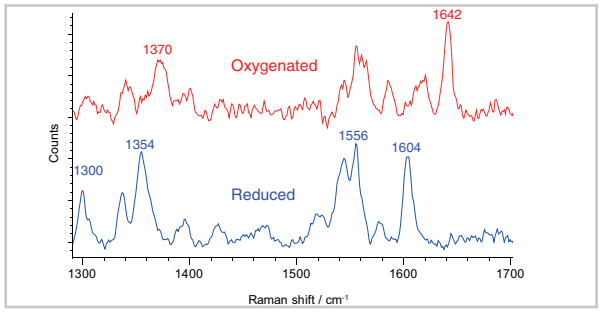


Raman spectroscopy is sensitive to the presence of haem proteins and is ideal for studying their redox biology, without the need for isolation or staining. The redox of haem proteins is closely linked to their protein functions – oxygen transport and storage, electron transport, and scavenging of free radicals. By using Raman spectroscopy to elucidate redox states within biological systems, researchers can study redox dynamics and its effects on health regulation and diseases.

Images generated from Raman spectra can be used to visualise the spatial relationships, distribution, redox and spin states of haem proteins within biological systems. You can process the data to reveal a multitude of information.

Identify haem protein type and their electronic states

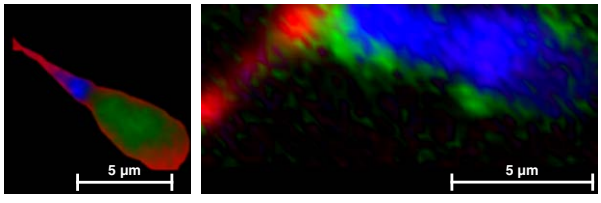
- Identify haem proteins by their Raman spectra
- Determine their redox and spin states by Raman band positions
- Resonance Raman spectroscopy with visible wavelengths can specifically enhance the signal intensities of haem protein bands, offering high sensitivity
- Isolation of protein not required
- Analyse haem proteins within cells (live/fixed) and tissues (whole organ/section)



Clearly define redox state according to the Raman features – oxygenated and reduced myoglobin.

Reveal haem protein information within biological systems *in situ*

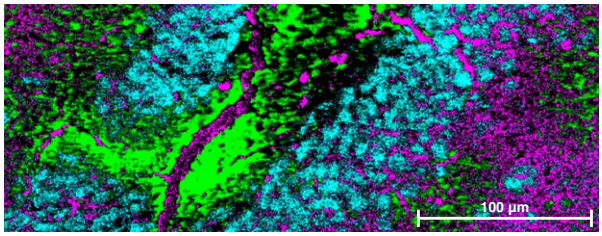
- Raman imaging provides detailed (sub-micrometre) spatial information
- Clearly show the location of haem proteins within the sample
- Provide information on the relationship between their distribution, protein, organelle and cell functions



In cells: analyse haem proteins' distribution, levels and redox state in situ. Reduced cytochrome signals are detected in blue and red domains in normal and abnormal spermatozoa respectively

Shed light on the protein, organelle and cell functions and dynamics through redox biology, such as:

- Correlate the cytochromes' redox state with mitochondrial intermembrane potential and energy production
- Better understand the link between mitochondrial dysfunction and neurodegenerative diseases or infertility
- Relate haemoglobin distribution in erythrocytes to their oxygen transport functions / abnormalities
- Study the oxidoreductase function of man-made redox proteins in synthetic biology
- Assess the oxygenation function of myoglobin in cancer progression and in tissue engineering



In tissue: analyse haem proteins' distribution, level and redox state in situ – neurons (cyan) and glial cells (green and magenta) in the rat brain section displayed varied levels and redox states of cytochromes.

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Raman imaging; the ideal technique for biological research

- Compare the haem proteins' physical and electronic properties between samples
- Simultaneously provide chemical information on other biomolecules, e.g. lipids, proteins, nucleic acids, carbohydrates, minerals
- Gain valuable insights of the effect of redox regulation on biological systems

Renishaw inVia; ideal for studying redox biology with Raman spectroscopy

- Research grade confocal Raman microscope
- StreamLine™ imaging technology for high speed mapping of haem proteins without causing cell/tissue damage
- StreamLine imaging with Slalom for a quick overview of tissue samples
- High confocality StreamHR™ imaging to scrutinise small details

The Renishaw inVia confocal Raman microscope

Relevant reading:

i. Brazhe *et al*, 2012, Mapping of Redox State of Mitochondrial Cytochromes in Live Cardiomyocytes Using Raman Microspectroscopy, PLoS ONE 7(9): e41990
ii. Ramser *et al*, 2012, Resonance Micro-Raman Investigations of the Rat Medial Preoptic Nucleus: Effects of a Low-Iron Diet on the Neuroglobin Content, Applied Spectrosc 12:1454-60
iii. Brazhe *et al*, 2013, In Situ Raman Study of Redox State Changes of Mitochondrial Cytochromes in a Perfused Rat Heart, PLoS ONE 8(8): e70488
iv. Parshina *et al*, 2013, Combined Raman and atomic force microscopy study of haemoglobin distribution inside erythrocytes and nanoparticle localization on the erythrocyte surface, Laser Phys. Lett. 075607
v. Anderson *et al*, 2014, Constructing a man-made c-type cytochrome maquette in vivo: electron transfer, oxygen transport and conversion to a photoactive light harvesting maquette, Chem. Sci. 5: 507-514
vi. Synytsya *et al*, 2014, Raman spectroscopy at different excitation wavelengths (1064, 785 and 532nm) as a tool for diagnosis of colon cancer, J. Raman Spectrosc. 45(10): 903-911

A range of related Renishaw literature is available. Please ask your local Renishaw representative for more information.

Renishaw. The Raman innovators

Renishaw manufactures a wide range of high performance optical spectroscopy products, including confocal Raman microscopes with high speed chemical imaging technology, compact process monitoring Raman spectrometers, structural and chemical analysers for scanning electron microscopes, solid state lasers for spectroscopy and state-of-the-art cooled CCD detectors, for both end-user and OEM applications.

Offering the highest levels of flexibility, sensitivity and reliability, across a diverse range of fields and applications, the instruments can be tailored to your needs, so you can tackle even the most challenging analytical problems with confidence.

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