THE REVOLUTION IN SEEING HOW CELLS WORK

Guest Editorial

Our understanding of how cells work has been dramatically transformed by the ability to see molecules in living cells. Over the last decade the development of green fluorescent protein (GFP) and the array of differently coloured probes have now made it possible to label almost any molecule and directly probe its function in live cells by light microscopy. The ability to visualise the dynamics of proteins in transport vesicles, organelles and cells has provided new insights into how normal cells function and the consequence of mutations resulting in disease. The spatial and temporal dynamics of a variety of cellular processes have been examined, for example, membrane trafficking, mitosis, cell signalling and cytoskeleton dynamics. Cellular imaging is now an essential toolkit of molecular cell biologists. Stunning images and 'art' portfolios help to give our discipline a broad appeal and time-lapse movies have become a common feature of seminars and publications in the field.

The highly dynamic nature of processes such as membrane transport have been very challenging to image. Recent technical advances in confocal microscopy provide enhanced sensitivity and spectral resolution that is ideal for multicolour imaging in space and time (4D imaging) – see the article by Hammond, Stow and Lock. Additional optical techniques like Total Internal Reflection Fluorescent Microscopy and Fluorescence Correlation Spectroscopy now allow biochemistry to be done at the single vesicle and molecule level inside cells. The recent development of spinning disk confocal systems provides further improvements in transmission and speed which is particularly relevant for high quality time-lapse imaging of rapid processes. Quantitation of interactions and dynamics of individual molecules can been obtained through the application of FRET and FRAP, respectively. With further refinements we can look forward to tracking the interactions of molecules as they move in space and time (5D imaging). The development of automated image analysis also has exciting consequences for genome-wide analysis of the subcellular location and function of gene products – see the article by Teasdale and Hamilton.

To date, most of the live imaging has been restricted to the use of cultured cells, however, the most relevant setting is within living organs. Two-photon laser microscopy provides the means to observe the behaviour of cells within the normal physiological environments of tissues and organs at depths of more than 50 μm. For example, imaging of exposed lymph nodes of an anesthetised animal has allowed dynamic events of lymphocyte migration and antigen presentation to be viewed as it occurs within the living animal. As cell biologists increasingly turn their attention to understanding the features associated with specialised cells, two-photon imaging will be adaptable to a broad range of both physiological and pathological processes.

A limitation of light microscopic systems is the absence of fine structural information. The development of techniques to correlate light and electron microscopy is now providing a more detailed cellular landscape to complement the fluorescent images. Cellular tomography is an exciting development that brings ultrastructure back to centre stage – see the article by Marsh. The ambitious aim is to reconstruct and map the components of an entire cell in 3D. With further advances in instrumentation and probes, the next decade will continue to open up a new world where cellular events will be visualised at the molecular level within the context of the fine structure of the cell and tissue. It is bound to be an amazing time ahead.

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Cover Illustration

The Golgi apparatus in NIH3T3 cells.

NIH3T3 cells were labelled with GM130 (a Golgi-specific marker), Alexa-Phalloidin (to detect actin) and DAPI (to show nucleus) and viewed by epifluorescence microscopy. Selected images were then pseudo-coloured to highlight the extent of Golgi-labelling.

Image courtesy of Darren Brown and Jenny Stow, University of Queensland.

Cell Imaging

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In the next issue...

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