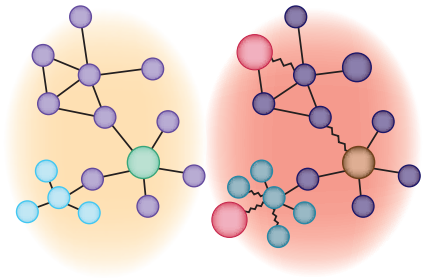
Networking to understand disease

The application of systems approaches to human disease will continue to expand.

The vast majority of human diseases do not result from single genetic changes. Most diseases are the consequence of multiple molecular alterations, genetic and otherwise, with an environmental component frequently also playing a role. What is more, even 'single-gene' diseases are likely to be modulated by alleles at other loci as well as by nongenetic effects. Systems biology—the use of comprehensive large-scale data to understand biology at a more global scale—is therefore widely seen as necessary for a complete understanding of disease.

There are many different ways in which systems approaches are being used for this purpose. In the study of cancer, for instance, several projects have been initiated worldwide to map the genomic, transcriptomic and epigenomic changes that occur in human cancer, and these have already



Disease as a systems-level phenomenon.

yielded maps of glioblastoma, breast and pancreatic cancer genomes, among others. Challenges abound, however, owing in part to the difficulty of distinguishing between 'driver' and 'passenger' events, and to the notorious heterogeneity of cancer, both within a given tumor and between individuals with the disease.

Separately, it has been shown that gene expression profiling data can be combined with information from the human protein-protein interaction network to yield more predictive markers of breast cancer prognosis than is possible with lists of differentially expressed genes alone. It should, however, be recognized that these network-based markers are still far from perfect.

Yet another instance in which systems information may prove useful for understanding the molecular basis of disease is in the more precise identification of causal disease genes, or groups of genes, from candidates suggested by genome-wide association studies. Existing approaches are largely based on bioinformatics analyses, and these are likely to develop further, but experimental strategies using high-throughput imaging to test candidate genes are a potentially exciting complement for the future, in cases for which useful cellular assays can be designed.

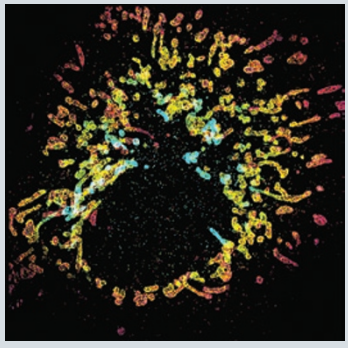
The tremendous interest in the systems biology of disease notwithstanding, both the use of networks as reliable biomarkers of disease and that of integrated 'omics' approaches to profile and describe diseased cells and tissues are still in their early stages. Watch for methods development and much movement in this area in the coming years! **Natalie de Souza**

Fast 3D super-resolution fluorescence microscopy

High-speed fluorescence imaging in all three dimensions at nanometer resolution will resolve, in finer detail, the workings of the living cell.

Light microscopy has given biologists eyes that can peer into small structures in cells and tissues. But owing to diffraction, a wave of light cannot be focused to an arbitrarily small point. Conventional light microscopes therefore have been long thought to be uncapable of resolving two objects closer together than about half the wavelength of light, with an imaging resolution of approximately 200 nanometers laterally and 600 nanometers axially.

Over the last several years, however, super-resolution techniques that allow the acquisition of microscopy images with lateral resolution on the order of tens of nanometers have emerged. Super-resolution fluorescence microscopy has enabled unprecedented resolution for several applications, for instance, live-cell imaging of



A 3D STORM image of the mitochondrial network in a cell (*Nat. Methods* **5**, 1047–1052; 2008).

dynamic cellular structures such as dendritic spines; indeed, it was the *Nature Methods*' choice for Method of the Year 2008.

But biological specimens such as cells are inherently three-dimensional (3D) objects, and early super-resolution methods were limited to improving the resolution either axially or laterally but not along all three axes simultaneously. Not only is it desirable to obtain a super-resolved image along all three axes, but imaging in three dimensions is critical for obtaining dynamic information in living cells. For both 3D and fast,live-cell imaging, current super-resolution techniques often fall short.

In recent years, the axial resolution of super-resolution techniques such as those based on stimulated emission depletion microscopy (STED), stochastic optical reconstruction microscopy (STORM) and photoactivated localization microscopy (PALM) has considerably improved, and in some cases it is now possible to attain sub-20-nanometer resolution in all three dimensions. This additional information comes at the cost of longer imaging times though. Slow acquisition speeds already limit live-cell, two-dimensional super-resolution imaging applications, and improvements are needed so that thick samples with dimensions closer to the full thickness of a cell can be imaged at nanoscale resolution along all three axes and at speeds sufficient to apply these methods to living whole cells.

The eventual arrival of high-speed, 3D super-resolution fluorescence imaging will allow these techniques to be increasingly applied to image the dynamic cellular environment at unprecedented levels of detail, fulfilling their potential as tools of unquestionable value to cell biologists. **Erika Pastrana**

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