Seeing the unseen with 'super-resolution' fluorescence microscopy

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Thanks to a new "super-resolution" fluorescence microscopy technique, Harvard University researchers have succeeded in resolving the features of cells as miniscule as 20-30 nanometers (nm), an order of magnitude smaller than conventional fluorescence light microscopy images, according to a presentation at the American Society for Cell Biology (ASCB) 48th Annual Meeting, Dec. 13-17, 2008, in San Francisco.

"Super resolution" microscopy techniques enable scientists to visualize cells laterally below 200-300 nm, which is the length scale of most intracellular structures and the level at which the cell gets most of its work done.

Harvard's "super-resolution" technique, developed by Bo Huang, Xioawei Zhuang and colleagues at the university, is called Stochastic Optical Reconstruction Microscopy (STORM).

It is one of several higher-resolution fluorescence microscopy techniques that fundamentally surpass the diffraction "blind spot" of conventional light microscopes.

Because conventional light microscopes cannot resolve two objects closer than half the wavelength of the light, they produce images that appear blurry and overlap no matter how high the magnification.

According to the Harvard researchers, STORM can record light emitted from a single molecule in the sample.

Using probe molecules that can be "photoswitched" between a visible and an invisible state, STORM can determine the position of every molecule of interest and can then compile all the molecules' positions to define a structure.

Huang and colleagues have adapted STORM to study three-dimensional structures and can now visualize a whole cell with an axial resolution of 50-60 nm.

Multicolor imaging also has been achieved by using photoswitchable fluorophores made of combinatorial pairs of various activator dyes and reporter dyes. Multicolor, 3-D STORM is able to visualize detailed interactions between cell organelles and the cytoskeleton.

In brain tissue, the researchers used STORM to reveal the fine details in the synaptic structure of the olfactory system.

Source: American Society for Cell Biology