



Microscopy equipment on-site demonstrations this month

Applied Precision - <http://www.api.com/index.html>

Applied Precision will be bringing their mobile Cellular Imaging lab to the UA campus. The lab includes demonstration systems of their two deconvolution microscopes. The mobile lab will be on the Main campus on Feb 14-15 and then it will be parked outside of the Pharmacy/MRB buildings on Friday, Feb 16. If you are interested in a demonstration of this system, contact Doug Cromey. To check for available time on the two systems, see the calendar at: <http://nanometer.cba.arizona.edu/DVdemo/demo>

There is an Applied Precision Deltavision RT system already available for use in the MRB building (contact - Dr. David Elliott, 626-7870 or <Elliott@arizona.edu>). This system was given a significant upgrade in October, so it is state-of-the-art. Deconvolution microscopes like this one are particularly well suited to imaging cells with fluorescence that may not be bright enough for confocal or live cells that respond poorly to high light levels. This system is nicely configured for time lapse studies of living cells.

Nikon high speed confocal microscope -

http://www.nikonconfocal.com/the_right_tool_for_you/products/live_scan.php

Nikon is tentatively scheduled to demonstrate their high speed confocal microscope during the week of Feb 26 - Mar 2. An email will be sent out as more details become available.

Doug Cromey has been collaborating with Dr. David Elliott of the new College of Medicine Research Microscopy Service Core to evaluate high-speed confocal microscopes from several vendors. The currently available campus confocal microscopes do not acquire images rapidly enough to image cellular events that occur in less than about 0.5 seconds. The College of Medicine is strongly considering a purchase of this type of instrument as part of its recruitment of new faculty for the MRB building.

Deconvolution vs. Confocal microscopy, what's the difference?

One of the frustrations with looking at samples in a wide-field fluorescence microscope is the out-of-focus blur that contributes to the image. This can be a particular problem with thicker samples. The excitation light from the lamp causes all the fluorescent molecules to light up, even the ones that are not in the plane of focus. Deconvolution and confocal microscopes take two very different paths to end up with crisp images of different focal planes in a sample.

Deconvolution uses a well calibrated wide-field microscope and a CCD camera. The technique assumes that a fluorescent sample is created by a collection of sub-resolution point sources of light (e.g., the dye molecules). The light from these points is affected (convolved) by the optics of the microscope in known ways. By taking a series of different focal plane images with the microscope and feeding the stack of images into a powerful computer, the blurred (out-of-focus) light can be moved back to its point of origin in the stack of images (de-convolved).

Confocal microscopes use an optical technique that discards light that does not originate from the plane of focus in the sample. The pinhole in the confocal does not allow out-of-focus light to reach the detector.

Which technique is better? - It depends on the needs of your experiment. The images will have very comparable optical resolution and clarity using either technique. Deconvolution can be done with lower light intensities at the sample, a clear advantage with live cell samples. A single focal plane image with a deconvolution microscope requires stack of focal plane images and time for the computer to deconvolve the stack. A single focal plane image with a confocal takes less than 10 seconds. Deconvolution works very well with samples that do not have bright fluorescence. Confocal images of samples with dim fluorescence tend to be electronically noisy.

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