

Introduction

For emission microscopy work, debate continues over the best imaging solution. This application note addresses two imaging sources, the Image Intensifier and CCD cameras.

How does an image intensifier work?

Photons strike a photocathode which converts the photons to electrons. The electrons are accelerated along a microchannel intensifier resulting in an increased number of electrons which strike a phosphor screen at the other end of the tube. The resolution is typically low (300x300) as it is defined by the number of available microchannels in the tube as well as electron deflection issues. Consider the Gen III GaAs indium phosphate system. The quantum efficiency of these systems is approximately 40% in the best case. Since signal amplification can also amplify the background noise, background averaging is required to get a clean image.

Sensitivity is limited mainly by the photocathode of the intensifier. An intensifier requires a sufficient packet of photons impinging on the photocathode to generate 1 electron for intensification. The quantum efficiency dictates the size of the required photon packet to release an electron from the photocathode.

Special interlocks are required to protect the intensifier. A burst of light to the intensifier can burn the phosphors off the screen of the intensifier. Replacement cost is quite high.

How does a CCD work?

The Charge Coupled Device was originally developed to be a memory element, but is now the major player for optical electronic imaging. Photons which strike a cell change the charge level of the cell (hole-electron effect). The number of electrons collected at each cell is

linearly quantifiable with time and photon intensity. Sensitivity derives from integration time and quantum efficiency. CCD sensors have quantum efficiencies typically in the 70% range at present. Resolution is limited by array size (512x512 to 4096x4096 are typical ranges). Sensitivity is mainly determined by integration time, so the longer the integration the higher the sensitivity. With long integration times CCD sensitivity can extend beyond that of an intensified system (10 minute integration is typical to reach the maximum sensitivity attained by an intensifier.) Cooling the CCD chip to -40°C allows up to 2 hours integration. Thermal noise (dark current) is reduced by lower temperature, so background subtraction is not required for most analyses. Modifications to the camera electronics also make such high sensitivity, long integrations possible.

Numeric aperture and sensitivity

Numeric aperture is an important part of the overall equation for an efficient emission microscope. However, higher magnification lenses will be less effective with an area emission involving depth such as forward biased junctions or saturated devices due to the limited depth of field. Point source emissions are much more easily detected at these powers since the emission is restricted to the focal plane. On the other hand, wide angle lenses and macro lenses perform best with area emissions but are effective to a lesser degree with point source emissions.

An important point is that corrective optics which match wide angle/macro lenses to the microscope (which is built for objectives) also scatter light. Overall transmission of the optical system is extremely important to the system's performance. Therefore, the more the system is cluttered with intervening optics, the worse the system's performance. The more direct the optical path, the less loss of emission signal.

Swapping out the intensifier for overlaid images

With intensified systems, the emission image (low resolution) taken with the intensifier is superimposed on the device image taken with the CCD camera. This technique becomes a serious limitation if spatial resolution of the emission image and registration accuracy is important.

Mechanical swapping can cause vibration problems and registration error between the image and the emission spot. The size of the emission spot will be in error with the number of pixels from the CCD image, as image intensifiers are inherently lower resolution. The emission resolution is equally important as the device image.

With a single cooled CCD camera directly producing both the emission and device images, the above problems are not introduced, and again, the less cluttered the optical path, the better the results.



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