

# METHOD OF IMPROVING THE AXIAL RESOLUTION AND IMAGE CONTRAST IN A SLIT SCANNING CONFOCAL MICROSCOPE

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Line scanning confocal microscopes can provide much faster image acquisition than their point scanning counterparts, as only one-dimensional scanning is required. However they do not block out-of-focus light travelling along the slit direction, resulting in a full width half maximum (FWHM) sectioning strength approximately 1.5 times larger than that for a point scanning confocal microscope, and much slower attenuation of signal away from the focal plane [1].

We present a simple implementation of a slit scanning confocal microscope that can attain an axial resolution better than that of a point scanning confocal microscope, and with faster suppression of signal with defocus. Under line illumination, images of a fluorescent object are captured using an array detector instead of a line detector. Out-of-focus light is recorded in virtual slits alongside the normal confocal virtual slit and is then subtracted from the confocal image to produce the improved sectioned image.

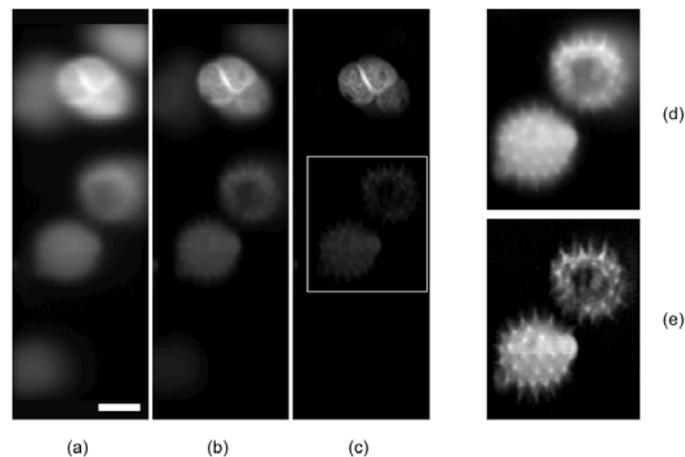


Fig. 1 60X images of a stained pollen grain cluster. Scale bar is 6 microns. (a) Reconstructed widefield image. (b) Conventional slit confocal image. (c) Improved slit confocal image. (d)-(e) Auto-scaled zoom of the highlighted pollen grain cluster in images (b) and (c), respectively.

Optical sectioning theory for the new microscope is described that shows a 10% sectioning improvement over a point scanning confocal system for a fluorescent sheet object. It also predicts an approximate  $u^{-2.9}$  out-of-focus signal suppression compared to  $u^{-1}$  and  $u^{-2}$  for conventional line and point scanning confocal systems respectively. Because the detection slit and line detector are replaced by an area CCD camera in a conventional line scanning system, this simple microscope modification could potentially be very easily implemented on existing commercial systems.