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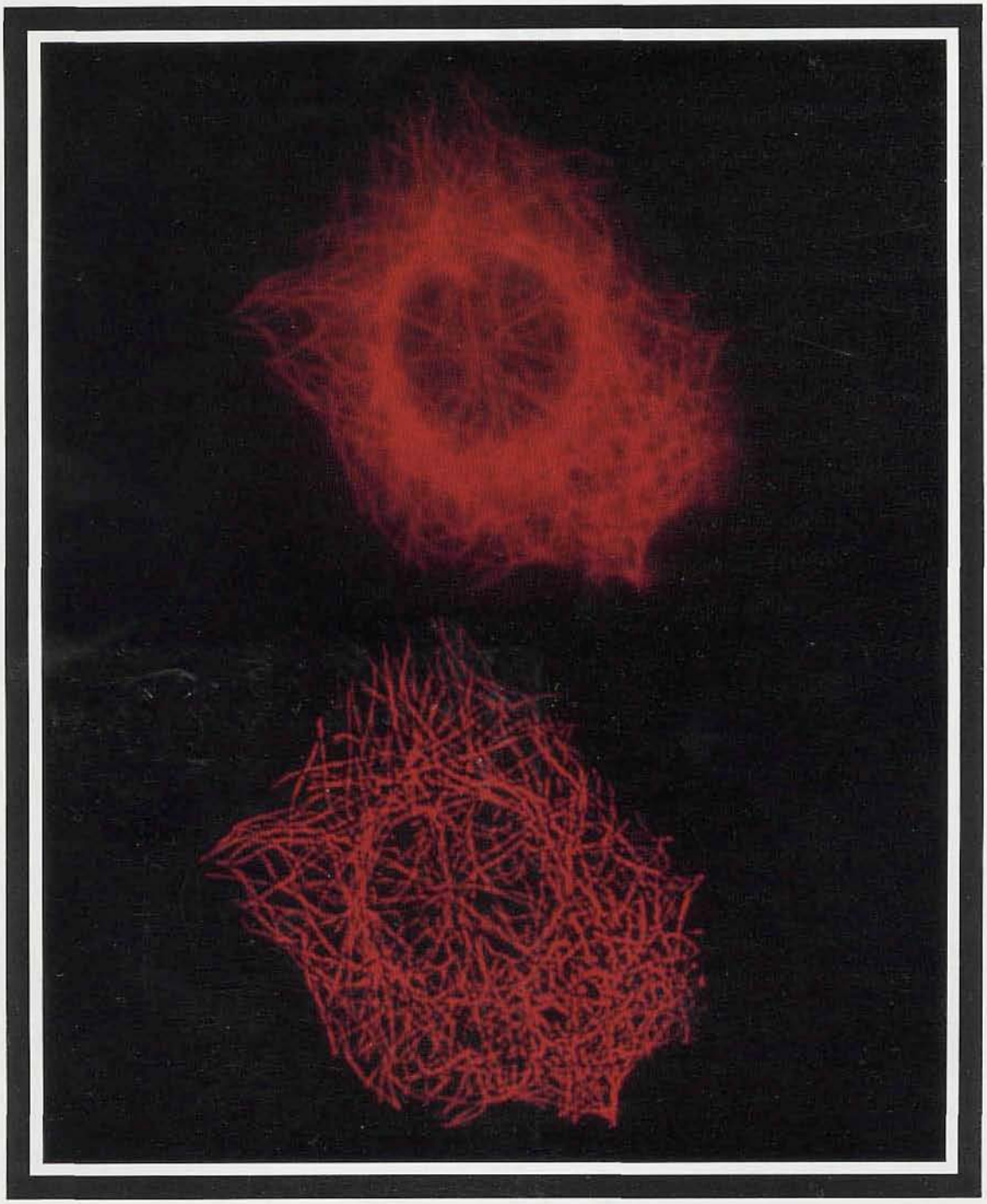
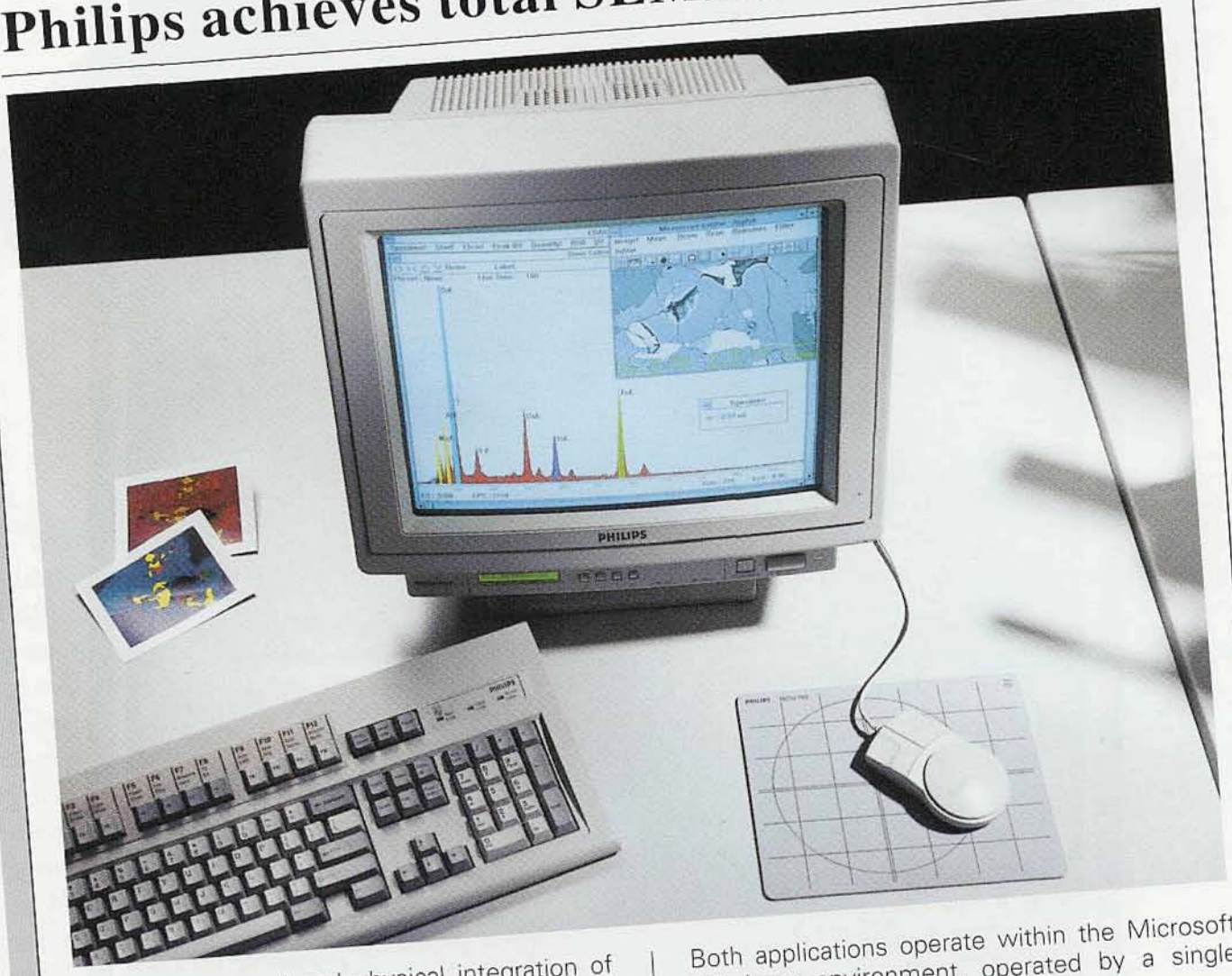


Innovation in Integration

Philips achieves total SEM/EDX integration



Absolute functional and physical integration of scanning electron microscopy and energy dispersive microanalysis have been realized in the Philips XL Series SEMs.

Both applications operate within the Microsoft Windows environment, operated by a single mouse and single keyboard.

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The Amazing ■ **XL** Series from Philips

PHILIPS

Comments on Electronic Libraries at NESEM/CMS meeting

Sterling Newberry

Each year The New England Electron Microscope Society and the Connecticut Microscope Society hold a joint Spring conference at Woods Hole which is usually reported briefly in The MSA Journal. The purpose of this note is to report on a discussion which is of great importance to our Microscope Society but which was not a formal part of the meeting and very unlikely to be reported else where, namely a discussion of the strengths and weaknesses of electronic access to library information.

The discussion arose when one of our MSA Tour Speakers, past president Mike Isaacson, stated that he is some what of a heretic about how to use libraries in that he believes that ability to physically browse the stacks is an essential function by which he has often discovered reports needed in his work. This brought a lively discussion from the audience which showed a quite different assessment of electronic library services than is gained by reading the current literature and public press on the subject.

Without belaboring the point, it is my hope that our readers will be encouraged to do two things. First to report on problems, proposed solutions to such problems and/or useful aids in library searches which they have found. The second thing which I wish to encourage is the use of this magazine for repeating a condensed version of good papers which are given at your Local Section meetings. Here the Program Chairman could suggest that the author let them submit his abstract to *Microscopy Today*. Perhaps the author would like to prepare a more extended summary or at least add more references and include a picture or diagram. The local committee might be able to assist the author with the chores of submitting this summary to our editor. We are a very good forum for extending the audience, which learns of an author's work, without prejudicing prior or future publications. It is good to note that our picture reproductions, B&W and color, are of excellent quality. Condensed republication of good work is a win situation for all concerned, authors, readers, local societies, *Microscopy Today* and the advertisers who make it possible. ■

Front Cover Image
3-Dimensional Volume Views (displayed as maximum-intensity projection views) of a Normal Rat Kidney Epithelial (NRK) cell showing rhodamine-labeled tubulin

The upper cover image shows the data as acquired using a standard epifluorescence microscope, namely a Zeiss IM-35 Microscope, equipped with a Nikon 60X 1.4 NA oil-immersion objective. 20 images were acquired at successive optical depths through the specimen, with a z-spacing of 0.25 µm between sequential focal planes. The lower image shows the data after restoration using Exhaustive Photon Reassignment™ (EPR™) processing in the Scanalytics CELLscan™ system (technology described as follows). Images were acquired and processed by Douglas Bowman of the Biomedical Imaging Center at the University of Massachusetts Medical School in Worcester, MA.

Fluorescence microscopy is an increasingly popular technique in the field of light microscopy. However, when one uses an objective lens of high magnification and numerical aperture, out-of-focus fluorescent objects in planes above and below the plane of focus contribute light haze to this plane, compromising the visual quality of the specimen image. The upper, unprocessed image clearly demonstrates this problem. This haze also renders it impossible to accurately quantify light intensities of fluorescent objects present in this plane.

Dr. Fredric S. Fay and members of the Biomedical Imaging Group at the University of Massachusetts Medical School in Worcester, MA have spent the last sixteen years developing and perfecting a digital imaging system that helps scientists overcome these problems, while avoiding many of the drawbacks associated with confocal microscopes. This system uses a method in which the distortion and dept-of-field properties of an individual wide-field microscope are characterized and mathematically *reversed*, yielding quantifiable, high-resolution images of the true object being visualized.

In brief: Images are obtained using a standard (wide-field) epifluorescence microscope and high dynamic range cooled CCD camera, resulting in fast, gentle image acquisition (perfect for live cells) and the ability to use any available fluorochrome. The optical system is characterized by acquiring an empirical Point Spread Function (PSF) using a sub-resolution-sized fluorescent bead. This knowledge about how the image of a point source appears when the source is in focus or at various distances out of focus is utilized to *reassign* out-of-focus photons in the 3D specimen image stack back to their points of origin with the stack. Since all available out-of-focus photons are reassigned rather than removed as is common with subtractive methods or filters, and since this procedure is performed utilizing a true, i.e., empirical PSF, the result of the process is a three-dimensional image in which out-of-focus light haze is reassigned to its proper locations, one in which fluorescent objects may be accurately quantitated and one where fine or faint structures are clearly visualized. The approach is thus based on *retention of all relevant signal rather than rejection of out-of-focus signal. The lower image demonstrates an example of results obtained with this technology.*

Research performed utilizing this approach has been regularly published in international journals and the technology is thus well characterized, and widely known and accepted. However, obtaining blur-free images using this process was computationally intensive and therefore not easily implemented. Advances in computer technology have now made it possible to perform this technique using standard desktop PCs. Scanalytics (a division of CSPI) is the exclusive worldwide licensee of this patented technology called Exhaustive Photon Reassignment or EPR. Scanalytics may be contracted at 40 Linnel Circle, Billerica, MA 01821. Telephone: (800)882-6247.

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