SOCIETY OF
ELECTRON MICROSCOPE
TECHNOLOGY

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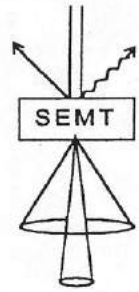

THE FUTURE OF IMAGING

Wednesday 20 May - 2.00 p.m. 1998

LONDON SCHOOL OF PHARMACY
Brunswick Square

2.00 **The multi-imaging centre - present and future**
Pat Echlin (University of Cambridge)

2.30 **Understanding the practical differences between analogue & digital imaging and identifying their application in the microscopy laboratory**
Steve Pierce (Kodak Europaean R & D, Harrow)

3.00 TEA and Trade Exhibition

3.30 **Imaging plates; a digital film replacement ?**
Angus Kirkland (Jeol UK Ltd, Welwyn Garden City)

4.00 **Digital imaging for the TEM using a CCD camera - system overview and application examples**
Neil Wilkinson (Gatan Ltd., Corby, Northants)

4.30 Close

Members and others interested are welcome to attend this meeting
RSVP to the Secretary by Monday 18 May

S.E.M.T. 20 May 1998 The Future of Imaging

The Multi-Imaging Centre - present & future
Pat Echlin
University fo Cambridge

The brain puts images into schemata.
A consensible statement gives an unambiguous message, - perception
A consensual statment gives a universally accepted truth - communication.

How much information is needed? A small number of pixels may be sufficient, e.g. if the information relates to colour. A satellite picture of a delta may resmeble a picture of the retine; interpretation depends on previous information.
Images of fractured surfaces, and scanning images, may give ambiguous up-&-down signals. Escher was a master of these ambiguous images. Drawings can be more representative than photographs, e.g. of snowdrop. Cartoons and diagrams can be used to reduce the information to a few elelments to communicate information.
At low resolution, or if the object is familiar, less information is needed. It must be put in context. We must be flexible with readings and hypotheses.

The Cambridge Multi-Imaging Centre is for use only by biologists; usage is divided into 2-hour segments. 15% of its income is from projects with industry.

e-mail pe13@cus.cam.ac.uk
www: bio.cam.ac.uk/dept/mic

Understanding the practical differences between **Analogue and Digital Imaging** & identifying their application in the microscopy laboratory
Steve Pierce
Kodak European R & D, Harrow

With reference to microscopy and particle analysis.
Method of image capture is electronic vs. film.
Analog structure is 3-D, because the structure of the film gives a number of layers for the different colours; the image therefore contains moee information. Increase in magnification is without grain-size effect.

The initial cost of the analog system is about double; running cost - it eats batteries but does not use film.
Digital system is more difficult at the beginning (so is analog).
The analog system can have the same soft ware for numerous systems. It is much faster, and a print can be obtained 1 scond after taking the picture.
The analog system is easier to manipulate, because it goes through a computer; but MUST MAKE SURE THE RAW DATA IS SAFE before beginning manipulations !!!

Analog systems can be used with Camscan S4; JEOL 100CX TEM; Leica DMRX; & 3 others.

Magnification of a digital image must be done in the microscope, not by the enlarger.
A video camera is not as good as a digital system.
An analog image can be put into other EMs; e.g. to re-run batches of analyses.
The Database structure is: acquisition & storage of data; calibration; storage; reporting. The images are stored on disk - removable disks or hard disk.
It is ESSENTIAL to save TWO images of each, at opposite ends of the site/ institute, in case of fire etc.

Imaging plates: a digital film replacement?
Angus Kirkland
JEOL UK Ltd, Welwyn Garden City

Imaging plates were originally developed for X-rays. A high sensitivity & wide dynamic range are needed; linearity or response for quantitative intensity measurements; high spatial resolution; high pixel resolution; good signal/noise characteristics; large field of view.

The present imaging plates:
re-usable flexible sheets compatible with atandard film cassettes;
digital readout of stored electron intensities;
readout possible many times;
digitisation at 14 Bits
16384 grey levels
large dynamic range.
linear electron response (over 5 orders of magnitude)
high resolution (comparable with that of emulsions)
low noise level
easily handled, & almost insensitive to visible light.

They are very good low-dose detectors!! They can even image unexposed silver halide crystals.

The CCD is complementary to imaging plates. The imaging plate has a rapid decay of point spread function over 3 pixels; the CCD has a long tail of decay of funciton.
The multiple transfer function with imaging plate is almost invariant with kV; with the CCD, the MTF varies strongly with kV.

The imaging plate is used off-line, and is pseudo-film-based; the CCD is online.
The Imaging plate background is 0.1 - 0.3 counts; the CCD 50 counts.
Imaging plate background noise is 0.09 counts; CCD 1 count.

At low-dose, the iamging plate is better - but few people work under these conditions.
£45-60,000 to buy with all the technology - plates, reader etc.

Digital imaging for the TEM using a CCD camers - system overview & application examples

Neil Wilkinson
Gatan Ltd, Corby, Northants.

Digital images are easily manipulated by computer; image processing can be done; multiple copies easily made; no wet darkroom is necessary.
Image acquisition device, software, & computer are necessary.
Scintillator of phosphor or YAG (yttrium aluminium garnet); fibre-optic coupling.
The bottom-mounted camera is Peltier-cooled at -35°C.
Many users write their own programs.
The camera software automatically adjusts brightness/contrast; it can select the area e.g. if a whole grid square is being examined, or the user can select.
Diffraction patterns & Fourier patterns are very useful, to produce an inverse mask and aid in reconstruction.
Digital montages look very pretty.
Fine tunign of focus is done using the Fourier transform; and also elimination of astigmatism. Autoalignment is possible.

Via the Internet, it is possible to see iamges being generated in another coutnry.
GIF can be used to colour elemental maps, for chemical imaging.

The future of imaging

Abstracts

The interpretation of electron micrographs.

Patrick Echlin (Multi-imaging Centre, School of Biological Sciences, Cambridge)

The visual acuity of the observer is central to the initial stage of the transformation of the two-dimensional image back to the three-dimensional object from which it was obtained. The image archiving processes are secure and reasonably well understood but have a number of failings. The mental processes of image interpretation are less well understood and involve pattern recognition and relating what we think we see to the canonical view of the image. We need to know whether the consensible message fits into the consensual body of knowledge. We have at our disposal a number of objective techniques to aid with the 2-D to 3-D conversion but these must be related to how much information we consider we need to interpret and understand the image. Inconsistencies and illusions confuse interpretation and we should perhaps consider other means of representing images. In the final analysis, experience and familiarity play a pivotal role but even these powerful attributes must be put into a context of aesthetics, flexibility of perception and correlative information.

Understanding the practical differences between analogue and digital imaging and identifying their application within the microscopy laboratory.

Steve Pierce (Kodak Res and Devt., Headstone Drive, Harrow)

The last few years have seen a massive growth in the digital technology for capturing and manipulating images, both in basic photography, and in micrography. This presentation attempts to uncover the mysteries and practicalities of the current imaging technology, as well as emphasising the obvious advantage of such systems for the user. The talk will then cover the practical aspects and advantage of digital and analogue image capture within the microscopy laboratory, taking the current changes in KODAK microscopy as an example. This will incorporate state-of- the- art capabilities which will, hopefully, fuel discussion on the future of image capture for microscopists.

Imaging plates: a digital film replacement ?

Angus Kirkland (Chem Dept.,Univ of Cambridge & Jeol UK Ltd.,)

In recent years there have been considerable advances in the development of digital detectors appropriate for use on transmission electron microscopes. Advances in CCD based cameras coupled via appropriate scintillators have provided approximately real time imaging with good signal/noise characteristics, large dynamic range and high sensitivity. In parallel, electron sensitive imaging plates based on the photo-stimulation of a phosphor have been developed to provide a direct digital film replacement.

In this talk the technology behind imaging plates will be outlined with reference to the requirements of an effective film replacement. In particular, the effective resolution, detective quantum efficiency, dynamic range, linearity and sensitivity will be addressed. Examples drawn from a range of disciplines illustrating the use of imaging plates will be presented. Finally the practical requirements for the successful integration of an imaging plate system into a modern EM laboratoy will be discussed.

Digital imaging for the TEM using a CCD camera: system overview and application examples

Neil Wilkinson (Gatan Ltd., Corby, Northants.)

CCD (Charge Coupled Device) cameras fitted to TEM's are proving invaluable in the on-line capture of high-quality digital images, on-line measurement, image analysis and microscope Autotuning procedures. Improvements in computer and related technologies make these tasks increasingly speedy and easy.

With purpose designed cameras utilizing fibre optic and CCD technologies derived from astronomy, their sensitivity is unsurpassed, allowing single fast electrons to be captured. At more normal beam intensities, images with a large dynamic range, good linearity and low geometric distortion can more than replace photographic film. These qualities can expand the scope of normal electron microscopy into areas such as on-line measurement, automated image analysis, automated focussing and astigmatism correction, automated low dose imaging and digital montage.

With a range of cameras to suit varied applications, fibre-optically coupled CCD cameras can replace the photographic recording of images and save large amounts of time. Images can then be transferred into third party applications (reports, image analysis etc.), saved and archived using a variety of storage media and transmitted via networks and the internet.

The CCD camera is suitable for all TEM applications from low-dose/cryo in Life Science to high-resolution lattice imaging in Materials Science. Using automation software, images from the CCD camera can be analysed and the TEM settings remotely adjusted to perform such functions as focus, astigmatism correction and Coma-free alignment. Specialised cameras can be used in conjunction with post column image filters to produce elemental images of high resolution and in real time. In biological sections, marked improvements in image contrast can also be achieved using image filtering.

There seems little doubt that photographic image recording in the TEM will be replaced by CCD image capture, in all but a very few specialised applications.

SEMT Meeting 20 May 1998

List of Registrants

Chris Andrews	School of Pharmacy
Andrea Boyd	Oral Med and Pathol., Guy's Hospital
John Bredl	Royal Veterinary College
Jackie Brown	Biol Dept., Open University
Terry Cooper	Taab Laboratories, Aldermaston, Berks.
Heather Davies	EMU, Biol. Sci., Open University, Milton Keynes
Sheila Davis	CAMR, Porton Down
Paul Davis	Jeol UK, Welwyn Garden City
Anne Drewe	Microbiology, Charing Cross & Westminster Med School
Barry Dowsett	CAMR, Porton Down, Wilts.
Patrick Echlin	Multi-Imaging Centre, Cambridge
Sara Fletcher	Royal Vet College
Alan Gray	Dental Inst., London Hosp Med Coll
Gisele Hodges	Queens University, Belfast
Joanne Hunt	Res and Devt., Kodak, Harrow
Stan Jones	Microlab Scientific Systems
Mike Kelly	Dental Inst., London Hosp Med Coll.
Claire Kendal	Biology Dept., Open University, Milton Keynes
Angus Kirkland	Cambridge Univ & Jeol UK
Gill Lewis	EMU, Eastman Dental Inst
John Manston	EMU, Queen Mary & Westfield Coll.
David McCarthy	EMU, School of Pharmacy
Hilary McPhail	Dept Physiol. St Mary's Hospital Med Sch.
David Michell	Edge Scientific Instrument Co., Milton Keynes
Nicky Mordan	EMU, Eastman Dental Hospital
Phil Salmon	Royal Veterinary College, London
Padmini Sarathchandra	EM, Surgical Research, NPIMR, Harrow
Wendy Tynan	Cortecs Research Lab., School of Pharmacy
Bob Whitenstall	Dept of Materials, Queen Mary & Westfield College
Neil Wilkinson	Gatan Ltd, Corby, Northants.
Amanda Wilson	EM, St George's Hospital Medical School, Tooting
Graham White	Cellular Pathol.,Southmead Hospital, Bristol
Steve Pierce	Kodak