MICRO-84

Electron microscopy 50 years on

from Colin J. Humphreys

IN 1934, *Nature* published the first electron micrographs of biological material. Fifty years later, the Royal Microscopical Society, in association with *Nature*, organized MICRO-84, an international microscopy conference and exhibition held in London from 9 to 13 July 1984. The conference was attended by about 800 scientists from all over the world.

One of the highlights of the meeting was a description by Albert Crewe (University of Chicago) of a revolutionary 0.6 Å point resolution scanning transmission electron microscope he has recently started to build. This represents a dramatic advance in resolution over existing electron microscopes, the best of which have a point resolution of about 1.7 Å. Modern electron microscopes have computer control added on, as it were. Crewe proposes to reverse this procedure and build his microscope around a computer system large enough to operate and control the microscope, and process and store the images. His computer system, comprising four computers with 3.6 Giga bytes of storage, has been donated by IBM, which demonstrates the importance that major microelectronics companies attach to high-resolution electron microscopy. If successfully built, Crewe's microscope should have major applications in biology, chemistry, materials science and physics.

Many microelectronic circuits are now so small that they cannot be resolved using an optical microscope, and an electron microscope is necessary simply to see them. D. Cherns (University of Bristol) and W.M. Stobbs and D.J. Smith (University of Cambridge) demonstrated how high-resolution electron microscopy can be used to determine the positions of atoms lying in and near the interfaces of semiconductor devices, so that their electrical properties can be understood.

In recent years, not only the resolution but also the analytical capabilities of electron microscopes have dramatically improved. It is now possible to analyse chemically very small regions of materials using an electron beam less than 10 Å in diameter, by detecting the signals (for example, characteristic X rays) generated when the incident electron beam is inelastically scattered by the specimen. Moreover, it is possible to measure the energy spectrum of the transmitted electrons by a technique called electron energy loss spectroscopy, which is sensitive to all elements and is increasingly being used for the analysis of both biological and non-biological materials.

If the fine structure of an electron energy spectrum is studied, a technique known as electron loss near edge structure (ELNES), results can be obtained equivalent to those achieved on a synchrotron source, but for any atomic number element and with far higher spatial resolution (C. Colliex, University of Paris). ELNES tells us not only what atoms are present in a sample, but also their environment. Mick Brown (University of Cambridge) described some novel applications of electron energy loss spectroscopy which exploit the very high spatial resolution available, for example, the measurement of energy band gaps in insulators.

Topics covered in the wide-ranging programme included microsurgery, forensic science, archaeology and art. Among the relatively new fields, C.F. Quate (Stanford University) gave an outstanding review of the advances in acoustic microscopy from a resolution of 10 μm 10 years ago to one of 300 Å, using helium, today, and G. Schmahl (University of Göttingen) described the latest results from his 500 Å resolution X-ray microscope, which is probably the best in the world. X-ray microscopy offers the prospect of studying living biological material with a resolution far better than that of optical microscopy, and with much less damage than is produced in an electron microscope. To be really useful, however, its resolution will have to be improved still further. Perhaps this will be achieved by the next meeting of MICRO, in 2 years time. □

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Vaccine development

Recombinant vaccinia for prevention of hepatitis B

from John Beale

THE idea of using vaccinia virus as a means of carrying other immunogens for the purpose of vaccination has seemed very attractive since Paoletti and Moss and their colleagues started to investigate the use of vaccinia recombinants in 1982 (Panicali, D. & Paoletti, E. *Proc. natn. Acad. Sci. U.S.A.* **79**, 4927; 1982 and Mackett, M., Smith, G.L. & Moss, B. *op. cit.* **79**, 7415; 1982). The rationale is that the live virus will express a foreign gene inserted into its DNA when it is applied to an animal, which will subsequently produce an immunological response to the foreign gene product. So far, recombinant DNA technology has been used successfully to express various antigens in vaccinia, including hepatitis B virus surface antigen (HBsAg), malaria sporozoite antigen, and influenza and herpes glycoproteins, and the list is growing all the time. On page 67 of this week's *Nature*, Moss and his colleagues take the hepatitis story a stage closer to a practical vaccine by showing that chimpanzees can be protected against hepatitis by vaccination with live recombinant vaccinia virus.

Last year, Smith, Mackett and Moss reported the construction of a fusion gene between DNA encoding HBsAg and an early vaccinia promoter (*UCLA Symp. molec. cell. Biol.*, New Ser. **8**, 543; 1983). By inserting the fusion product into the thymidine kinase-encoding portion of the vaccinia genome such that thymidine kinase activity was no longer expressed, they could select the new recombinant virus by growth in the presence of 5-bromodeoxyuridine. The plaques were tested for HBsAg expression by dot-blot hybridization and for HBsAg gene expression by immunological methods.

What Moss and his colleagues have now done is to vaccinate two chimpanzees with the HBsAg-containing recombinant vaccinia virus and one chimpanzee with wild-type normal vaccinia virus, and then challenge them with living hepatitis B virus. They find that treatment with HBsAg–vaccinia hybrids produces smaller lesions and lower antibody titres to vaccinia than does treatment with vaccinia alone. None of the animals produced antibodies to HBsAg after vaccination. On challenge with hepatitis B virus, however, the chimpanzee given vaccinia alone developed typical signs of hepatitis B virus infection: hepatitis surface antigenaemia, antibodies to the core antigen as well as to the surface antigen of hepatitis B (the production of core antibodies indicates some hepatitis B virus replication had occurred in the primed animal) and biochemical evidence of hepatitis. By contrast, the two HBsAg–vaccinia-treated chimpanzees, although producing no antibodies before challenge, were protected against the challenge, as shown by the lack of biochemical evidence of liver damage and of antigenaemia, and the rapid onset of antibodies to the surface antigen. They had thus been immunologically primed by the expression of HBsAg when the recombinant vaccinia was growing in the skin.

These are impressive results that pose in a sharper form the policy implications of this approach. The attraction is that the accepted and proven instrument of smallpox eradication can be adapted to the formidable task of hepatitis B prevention.