Meetings

Industrial and Biological Microscopy: New Paths

The various techniques of microscopy have been applied in new ways and to new problems in chemistry, biology, medicine, and the study of fibers and polymers for industrial purposes. Many of these new applications and the results obtained through them were described or demonstrated at the biennial symposium of the New York Microscopical Society held in New York City on 19–20 March.

Among the most outstanding presentations at the symposium were two time-lapse films shown during the sessions on medical and biological microscopy. The first, "The Dynamics of Bone Resorption" by Paul Goldhaber (Harvard School of Dental Medicine), demonstrated the behavior of living cells taking part in this process. The film revealed that osteoclasts played an active role in the destruction of bone, and that in areas of rapid resorption, numerous giant "bubbles," ranging in size up to 50 microns, developed within the osteoclasts. Fusion of these "bubble osteoclasts" and consequent development of multinucleated giant cells were clearly shown. Also demonstrated was the enhancement of bone resorption by oxygen concentration in the gas phase of the system, as well as by the concentration of parathyroid extract, vitamin A, and vitamin D in the medium. Marked enhancement of the effect of suboptimal concentrations of these factors was caused by addition of heparin, which by itself had no effect.

The second film, "Birefringence of the Mitotic Spindle," by Shinya Inoué, Hidemi Sato, and Geoffrey P. Pollitt (Dartmouth Medical School), was also concerned with the behavior of living cells as shown by time-lapse cinematography. It represented a new approach to an understanding of the molecular mechanisms of cell division and movements of chromosomes during mitosis. Birefringence, the property by which a specimen can be seen in the polarizing microscope, reflects the regular molecular orientation of cells. However, this birefringence is weak and is often drowned in a diffuse haze of altered polarized light at the high magnifications required to study cell details. A new microscope, with a polarization rectifier, has been developed by Inoué and has been used in these studies to follow changes of molecular orientation within living cells. The film followed the development of jellyfish eggs, spermatogenesis in a grasshopper, division of the pollen mother cell in Easter lily, and endosperm division of the African blood lily. Another film showed mitosis in endosperm cells of the African blood lily under phase and interference microscopy. The molecular alignment of the spindle fibers in cell division, as revealed by Inoué's microscope, was shown to be controlled by the centrioles at the spindle poles. The degree of alignment was measured by the birefringence. The structure of the spindle fibers could be seen undergoing change when temperature fluctuated and when heavy water or puromycin was added to the system. The existence of spindle fibers, heretofore a controversial point, has been confirmed by the use of the new microscope, thus settling a 50-year controversy.

Following this film, a paper by Joseph Altman (M.I.T.) introduced a new application of autoradiography. With this technique he demonstrated that a postnatal development of microneurons occurs in the brain of the rat. The technique involved injection of a radioactive tracer into newborn rats which were then killed at intervals of 6 hours to 20 days. Thymidine-H^3 was used as the tracer, because, being a specific precursor of chromosomal DNA, it is incorporated into the nuclei of proliferating cells. A time-lapse record was thus made of the behavior and migration of the originally labeled cells. Dilution of the tracer indicated cell multiplication; unlabeled cells were those in existence before the injection, and heavily labeled cells were the first to develop after injection. Autoradiograms showed that migration of undifferentiated cells from a "reservoir" and subsequent development into microneurons occurred after birth. Altman suggested that those microneurons which develop postnatally, as the animal comes to respond to its environment, may be the modulatory and plastic elements of the central nervous system.

New applications of fusion methods in chemical microscopy were presented by Donald Laskowski (Cleveland Metropolitan Hospital) and Francis Jones (U. S. Dept. of Agriculture). Laskowski discussed a procedure for the study of donor-acceptor complexes between carcinogenic hydrocarbons, aromatic amines, and aromatic azo compounds as donors and a variety of quinones as acceptors. These studies are of value both as elucidation of the behavior of some carcinogens and as a more general method for the microscopist to study formation of such complexes. The application of fusion methods, as the name implies, involves melting the prepared samples on microscope slides in contact with each other and studying the zone of mixing for characteristic behavior on heating or cooling. A novel use of a cautery needle under the slide in place of the more elaborate hot stage was described by Jones. This technique, which included an embedded thermocouple, gives all the required information as to the nature of the reaction between two compounds by melting and cooling of a narrow strip of the contact preparation. Color photomicrographs presented by both speakers illustrated the formation of complexes, compounds, solid solutions, eutectics, and so on.

Papers presented by George Cocks (Cornell University) and Walter McCrone (McCrone Associates) suggested enlargement of the chemical microscopist's arsenal of tools to include the electron microscope, x-ray, and microprobe analysis, as well as ultraviolet and infrared absorption. Cocks discussed the applications of the electron microscope to the problems of the chemist. Both he and McCrone

stressed the unique qualifications of the light microscopist in the interpretation of problems, since, as McCrone stated, "the microscope is the only research tool not adaptable to routine operation by a technician, and the experience of the microscopist is a necessity in sample preparation and interpretation of results." He also proposed a new definition of the term microscopy—as the "study of the composition and behavior of matter on a small scale"—to cover the broad application of the essential tools of the skilled microscopist.

In the session on fibers and polymers, three of the speakers discussed various approaches to the study of the internal structure of fibers. The nature of voids and variations in fiber density was demonstrated by Frederick Morehead (American Viscose) using a double sectioning and embedding technique, involving longitudinal sectioning of viscose rayon. Robert Scott (DuPont), after a brief review of the fundamentals of interferometry, described the combination of this technique with x-ray diffraction and density studies to obtain information about the nature of microvoids and the uniformity of density. Both speakers emphasized the value of birefringence measurements, made with the polarizing microscope, in understanding orientation and density of fibers. Marion Rhodes (University of Massachusetts) described a method of obtaining structural information by mathematical evaluation of low-angle light-scattering patterns of polymers. A polarizing microscope equipped with a Bertrand lens was found most convenient when correlating such patterns with the morphology of the polymers. The size of the lobes of the scattering patterns was related to the average dimension of the spherulites, and this average value gave a more accurate representation of the polymer morphology than could be obtained by direct measurement, which would have been hampered by the presence of background spherulites as well as intergrown material which would be difficult or impossible to measure directly.

Illustrations were presented of these applications and of the changes occurring in polymers during thermal treatment. The surface topography of fibers was explored by Joseph Bloxsom, who described how the Tolansky light-profile technique could be used to make quantitative measurements of height-depth variations. This method involves

the projection of a reticle in the object plane of a microscope. Quantitative measurements are made by observing the displacement of the ghost reticle in a previously calibrated system. The technique may be used on solid or liquid surfaces and does not require that they be highly reflective. It may be used to determine thickness and refractive index of transparent films and, being nondestructive, is of value in studying time-dependent behavior.

The proceedings of this meeting will be published and are expected to be available next fall at $5 per copy through the New York Microscopical Society, located at the Museum of Natural History, 79th Street and Central Park West, New York, N.Y.

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The Electron Microscope and Its Future Development

The first successful operation of an electron microscope in North America was announced in 1939 (*1*). Many of the formidable technical obstacles that lay in the way of obtaining electron optical images at high magnification were overcome in the encouraging atmosphere of E. F. Burton's laboratory at the University of Toronto. Accordingly, to commemorate the anniversary of the announcement, the Burton Society of Electron Microscopists held a special meeting at the university on 16 January 1965. The work of Burton's talented group of students (which included Cecil Hall, J. Hillier, W. A. Ladd, and A. F. Prebus) was described by Hillier (now at RCA, Princeton). The achievement of Hillier and co-workers in rapidly making a modified version of the Toronto microscope commercially available to biologists and physicists considerably influenced the pace of development of ultrastructural investigations.

J. H. Reisner (RCA, Camden) discussed the possibilities of viewing the electron optical image by an image intensifier–television system. It was pointed out that image intensifiers allow electron microscope images to be seen comfortably in a bright room, while the illuminating beam is actually reduced below normal operating intensity. Methods of recording images on video tape and of analyzing the image were discussed. A method of