native approaches were offered by T. Nei (University of Sapporo, Japan), H. Skaer (University of Cambridge) and A. Walter (Clinical Research Centre, Harrow) who advocated the use of penetrating and non-penetrating cryoprotectants. Nei showed images of remarkable preservation in red blood cells frozen and thawed in the presence of 20% glycerol, and Skaer gave details of some high molecular weight polymers which although they do not enter cells, limit the size of intracellular ice crystals to nanometre dimensions even in pieces of tissue 1 mm in diameter. Such polymers have important implications in the preparation of tissues for freeze fracture and X-ray microanalytical studies which are best carried out on chemically unperturbed tissues.

There was extensive discussion on the process of sectioning and freeze fracturing and the nature and extent of the artefacts associated with these techniques. A. Saubermann (Harvard Medical School) demonstrated that the process of cutting frozen material is a mixture of sectioning and multiple fracturing depending on temperature, cutting speed and knife angle, and U. Slevtr (Vienna) and T. Robards (University of York) showed that plastic deformation can occur af temperatures well below the glass transformation point of ice. Sufficient energy is released during cleavage to deform biological material and great care must be taken when interpreting freeze fracture images. The same point was taken up by D. Branton (Harvard University) and S. Bullivant (University of Aukland) who showed that some of the artefacts associated with membrane structure are produced either during specimen pretreatment and freezing or simply by the collapse of the lipid layers after membrane cleavage.

Low temperature techniques are clearly no longer the preserve of the morphologists and a number of papers were given showing how both frozenhydrated and frozen-dried tissues and sections could be used for the analysis of diffusible ions and electrolytes. K. Zierold (Max Planck Institute, Dortmund) and M. Sjostrom (University of Umea, Sweden) gave details of low temperature X-ray analytical studies on mammalian muscle. T. Barnard (Wenner-Gren Institute, Stockholm) related the diffusion of electrolytes in frozen sections of adipose tissue to changes in the electron density of the sections. One of the main problems of low temperature microscopy is to ensure that the biological material stays cold enough during preparation and examination. P. Echlin (University of Cambridge) considered these problems in relation to cryofractured frozen bulk samples and sections examined at

low temperatures in the scanning electron microscope and suggested a number of criteria which could be used to judge the hydration state of material in the microscope. Such problems are less exacting for the smaller samples examined by K. Taylor (MRC Laboratory of Molecular Biology, Cambridge) in the transmission microscope who used the disappearance of the ice diffraction pattern as an indicator of specimen hydration in catalase crystals and bacterial cell walls. There were close similarities between the negative stain images of catalase and the hydrated crystals, and it looked as if the ice was acting rather like a negative stain. The earlier hopes that low temperatures might dramatically diminish radiation damage in high resolution images were dashed when R. Glaeser (University of California, Berkeley) showed that depending on the specimen, the damage is only lessened by a factor of two when operating at liquid nitrogen temperatures and a factor of five when operating only a few degrees above absolute zero

There was considerable discussion on the interpretation of morphological data obtained from frozen sections and fractures. T. Gulik-Krzywicki (CNRS Gif-sur-Yvette) showed that it is possible to correlate low temperature Xray diffraction patterns with images obtained by freeze fracture electron microscopy, and H. Chanzy (CNRS Grenoble) described a technique for low temperature electron diffraction. Whereas these specialised techniques and the use of image processing can help with the interpretation of the inert freeze fracture images, such methods are of little use for unstained frozen sections which give images containing few of the subcellular features recognisable from conventional microscopy. Cryohistologists are going to have to re-educate themselves to recognise images in material produced and examined at low temperatures. П

Head-on proton collisions

from Peter Landshoff

Most of the information on the structure of protons has been obtained by bombarding them with very high energy electrons, and these are the experiments that have provided strong, though indirect, evidence that protons contain quarks. The quarks apparently

Peter Landshoff is Reader in Mathematical Physics at Cambridge University.



A hundred years ago

THE Superintendent's Report on the Botanic Garden and Public Plantations for 1875-76 has recently been officially published in Jamaica. It deals almost entirely with plants of economic value, foremost of which is the coffee, the ordinary kind (Coffea arabica), apparently giving way to its formidable rival Coffea liberica, which was introduced to Jamaica in 1874, and is now thriving, especially in some districts. In one situation, at a height of about 1,000 feet above the sea, a plant that had only been planted out a little over a year has already fruited. This seems to indicate that in the course of a few years the new coffee may be widely cultivated in Jamaica from plants raised from seeds ripened in the island.

From Nature 16, 24 May, 73; 1877.

have very small diameters, or are maybe even pointlike, although the proton as a whole has a more diffuse structure in which the quarks are embedded. The nature of the remaining constituents is not yet clear. Certainly they include quark-antiquark pairs, some of which may be bound tightly together to form mesons. Very possibly they include also the 'gluons' that bind quarks and antiquarks together in the quantum field theories that mathematical physicists are constructing in attempts to unify the various fundamental forces of nature.

The evidence that there are quarks inside the proton is indirect, because no free quarks have yet been seen. Indeed, many physicists believe that it may not be possible for quarks to be knocked out of their parent protons. (There have been various attempts to construct such theories of 'quark confinement'.) For this reason, it is desirable to verify the quark structure in other types of experiment. Beams of high-energy muons and neutrinos have also been used to probe the proton, and the information that they give, while again indirect, seems to confirm and complement that which comes from electron scattering. A quark structure in the proton seems rather sure.

Electrons, muons and neutrinos are particularly useful as probes because their own structure is especially simple and well understood. However, additional information can be obtained by making pairs of protons collide at very high energy.