

## LETTERS TO THE EDITORS

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## A Flying-spot Microscope

SURPRISINGLY little of the information obtained with microscopes has been quantitative; most observers are content to sit at the microscope and regard the image, or to photograph it. Theoretically, it is possible to scan the image or its photograph mechanically; but this has seldom been done in practice. The whole method of obtaining resolution by lenses involves so much loss of light, lack of control of contrast, and other difficulties, that it is difficult to provide a good display or method of scanning.

Some of these difficulties can be avoided by using a wholly different means of obtaining resolution and amplification. The essence of the problem of resolution is to separate in some way the light passing through very close regions of an object. The conventional microscope does this by using refraction by lenses to separate the light from neighbouring regions. An alternative method is to use the lens system the other way round, namely, to produce a minute spot of light. Discrimination between neighbouring points is then produced by passing the light through them at different times by making the spot scan it. After passing through the preparation, the spot is made to fall on a photocell, with subsequent amplification and display as required. Such a flying-spot microscope depends on scanning different parts at different times, and will only give accurate information about objects that are stationary or moving only at a rate of a different order from that of the spot.

The method is based on the well-known technique of flying-spot scanning by two cathode-ray tube rasters locked together. In front of the eyepiece of a microscope is placed a tube providing a televizing raster of high brilliance and very short time-constant. The microscope objective thus produces a minute spot of light that scans the preparation to be examined. The amount of light transmitted is determined by the density of the specimen and is picked up by a multiplier photocell. The output of this is used for modulating the raster on a second cathode-ray tube, of projection type.

This combination of optical and electronic techniques provides all the facilities of conventional microscopy, with important additional advantages, among which are the following. (1) A projection display some three foot square and of brightness several foot-lamberts can be obtained. This method of display has substantial advantages, not only for discussion and teaching, but also for measurement and indeed for ordinary surveying of preparations. (2) Since the size of scanning raster can be continuously varied, there is less need for changing of objectives. (3) The brightness of the image is readily controlled. (4) Since the contrast is determined by the amplifier gain as well as by the density distribution, contrast can be made greater, or less, than in the original specimen, with obvious advantages in studying unstained, or vitally stained, or over-stained specimens. (5) It should be possible to produce visible displays, with incident radiation of a wave-length of at least from 2,000 Å. to 10,000 Å.

(6) The quantum efficiency of the photocell is greater than that of the photographic plate by at least 100:1 at 2,650 Å., making it proportionally easier to avoid producing alterations in living specimens. (7) Resolution in the ordinary microscope is affected by the halation produced by interference between neighbouring particles. Since only one resolvable region is illuminated at a time, resolution should be substantially greater with the flying-spot than with the conventional microscope. This possibility has not yet been systematically explored experimentally. (8) Perhaps the most important of all advantages is the possibility of quantitative analysis. The output of the photocell has a voltage-time distribution known as a time series, and can therefore be analysed by well-known techniques to give histograms and spectrograms of the specimens. Particles could probably be counted and sized at a rate of a million per second.

The first model that we have built suggests that these advantages can be realized, and further development is being undertaken.

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## Extra-Corticle Membranes and Layers of the Wool Fibre

It has been shown<sup>1</sup> that peracetic acid renders wool freely soluble in dilute ammonia, except for a small fraction which appears as a tubular membrane. By prolonging both these treatments, it has been possible to obtain further evidence in support of the theory concerning the structure of the outer layers of the wool fibre put forward by Reumuth<sup>2</sup>, Lehmann<sup>3</sup>, Swerdlow and Seeman<sup>4</sup> earlier, Lindberg, Philips and Gralén<sup>5</sup>, and Zahn and Haselmann<sup>6</sup> more recently.

Extracted wool fibres were fixed by means of a solution of methyl methacrylate to an open frame which would accommodate a microscope slide. They were then suspended in peracetic acid for five days. After washing they were immersed in ammonia for a further seven days. The remains of the fibres had an appearance rather like transparent soft elastic and were quite fragile.

Microscopic examination showed them to be membranes similar to those illustrated by Alexander and Earland<sup>1</sup>. In view of the strong evidence supporting the existence of an epicuticle membrane<sup>6-8</sup>, I attempted to break this major membrane with micro-manipulators, hoping that if I could dislodge such an epicuticle I would be able to see some trace of it. This actually happened. The operation of sliding this very transparent sheath off the comparatively bulky major membrane was a hazardous one, and it was only in three attempts out of several on each of a hundred fibres that success was achieved. Although it was invisible in a bright field, by using the phase-contrast microscope and the very minimum of illumination, the first example was seen. The micrograph of this involved a 30-min. exposure with a 23 Scheiner orthochromatic plate (Fig. 1). Later, a better result was obtained with a 30 Scheiner plate. In both these cases a thin membrane was removed from the part of the major membrane shown in the micrograph.