LETTERS TO THE EDITORS

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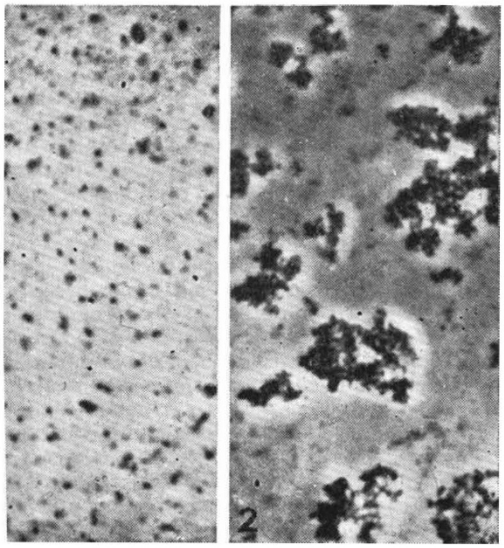
Phase-Contrast Microscopy of Viruses

THOSE who have used phase-contrast microscopy will have been impressed by the apparent high resolving power of the method. It is frequently possible to observe very fine fibrils and particles with a clarity hitherto only obtained by dark-ground illumination. Thus, I have observed living *Leptospira*, said to be only 0·15 μ in width, as well as very small granules inside bacteria.



It was to be expected, therefore, that at least the larger viruses would be visible by phase-contrast illumination, and this has now been verified. Fig. 1 shows a smear of psittacosis virus (× 1600), and Fig. 2 one of vaccinia (× 1600). The preparations were unstained and mounted in water. It is important to distinguish between *visibility* and *resolution* of the particles. The photographs were taken with commercial (Cooke, Troughton and Simms) equipment, using an oil immersion lens, rated N.A. 1·3 with phase-ring diameter approximately two thirds of the aperture. The expected resolution under these conditions, using green light, would be of the order of 0·3 μ. One would thus scarcely expect to obtain a true resolution of virus particles; but that does not preclude the possibility of detecting their presence. Thus, the vaccinia particles seemed spherical, not barrel-shaped, as in the electron microscope. The latter appearance may, of course, be due to a drying artefact.

As a rough approximation, it would be expected that particles below the limit of resolution would appear to remain about the same order of size, but would have less contrast, appearing in shades of grey instead of black. The visibility of small particles in the phase-contrast microscope depends on contrast, which is in turn governed by the refractive index of the particles and by the characteristics of the apparatus. Leaving aside the possibility of improving contrast by variation of the refractive index of the mounting medium, perhaps the most hopeful method of increasing the visibility is by alteration of the phase, and particularly the amplitude, characteristics of the phase plate. Given favourable material, there is probably no reason why much smaller viruses should not be seen, using a strongly absorbing phase plate with an intense light source.

It remains to be seen how useful the method will prove in virus research. It is valuable in that it allows fresh unstained material to be used; but it is, of course, non-specific and there is no simple means of distinguishing viruses from other small particles. An obvious application is to the study of the development of virus bodies in living cells.

I wish to thank Prof. S. P. Bedson for supplying the virus material.

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Experimental Hydronephrosis Studied by the Colchicine Method

THE colchicine method was first used to study compensatory hypertrophy of the kidney by A. P. Dustin and S. Zylberszac[1,2], who noticed that a few days after ligation of the ureter, colchicine induced numerous 'stathmocineses' (arrested mitoses) in the kidney (Fig. 1).

This phenomenon has been reinvestigated in the adult rat, nine hours after injection of 2·0 mgm./kgm. of colchicine, mitosis being counted in the obstructed and the opposite kidneys. The total number of divisions was counted in median sections and, in order to compare the figures more accurately, these were corrected per 100 gm. of animal weight (Fig. 2).

In the obstructed kidney, two days after ligation, numerous stathmocineses are found in the interstitial cells, convoluted tubules, loops of Henle, distal convoluted tubules and collecting tubules. None is seen in the glomeruli. The mitoses remain numerous during the five succeeding days, especially in the proximal convoluted tubules. After one week, mitoses decrease as the kidney undergoes progressive atrophy through dilatation of the pelvis. In the opposite kidney, mitoses in the tubules increase on the

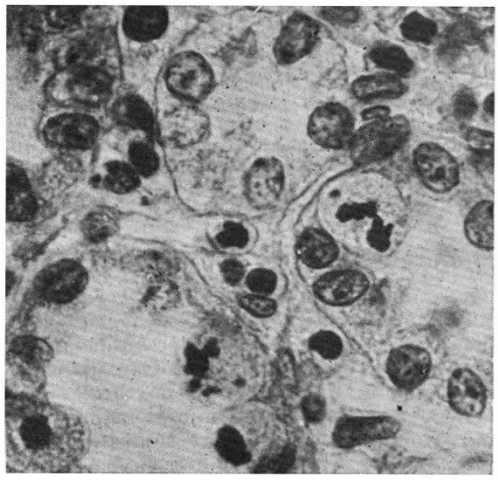

Fig. 1. Stathmocineses in convoluted tubules six days after renal obstruction