SOME CYTOPLASMIC DETAILS OF YEAST REVEALED BY THE ELECTRON MICROSCOPE

It was reported recently\(^1,2\) that even in ultra-thin sections of yeast fixed in osmium tetroxide structural details could not be made out owing to the cytoplasm being impenetrable to the electron beam. Bartholomew and Mittwer\(^3\) used ultra-violet photolytic methods to obtain electron micrographs of the internal structure.

In bacteria the cytoplasm has a strong affinity for basic dyes resulting in difficulties in demonstration of the chromatinic structures. This practical difficulty was overcome by mild hydrolysis.\(^4\) Hydrolysis of yeast cells was therefore carried out to discover whether such a treatment would make the cytoplasm transparent to an electron beam.

The strain of yeast used was the riboflavin excreting mutant, \(\text{BY2.}^5\) Young cells from wort cultures at 28° C. fixed in 40% neutral formaldehyde for 1 hour and stored in 5% formaldehyde for 3 days were washed for 3 hours in repeated changes of distilled-water and hydrolysed in \(\text{NHCl}\) for 12-15 minutes. Small droplets of distilled-water suspension of cells were transferred to supporting membranes of Formvar on specimen carriers, dried and examined with a Philips three-stage electron microscope operating at 60 KV. The cells were viewed on the fluorescent screen at 10,000 diameters and photographed at a quarter of that magnifi-
cation on Kodak 35 mm safety positive film. The negatives were enlarged six times.

The progressive removal of opacity could be seen in Figs. 1, 2, 3 & 4. The removal is not uniform but in patches. Attention is invited to the spherical electron opaque body in Fig. 3 which reminds one of the nucleus described from stained preparations. While the impenetrability of the cytoplasm to the electron beam makes a study of all the cell organelles difficult, the fact that hydrolysis removes the opacity offers the hope that investigation of the nucleus and its behaviour during cell division may be possible.

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5. Royan, S., Ibid., 1953, 19, 267.