A Membranous Component of the Cytoplasm in *Streptomyces coelicolor*. BY AUDREY M. GLAUERT* AND D. A. HOPWOOD. (*From the Strangeways Research Laboratory, and the University Botany School, Cambridge, England.*)‡

Most studies of bacterial fine structure in thin sections have shown the cytoplasm to consist mainly of minute granules (1). Occasionally, small regions with a membranous structure have been observed (2, 3, 5, 10–12). In the present work, improved fixation of the actinomycete *Streptomyces coelicolor* by the method of Kellenberger, Ryter, and Séchaud (5), has revealed an unusually complex organisation of the fine structure of a bacterium. The most remarkable feature is an extensive membranous component in the cytoplasm.

Much of the cytoplasm of the hyphae of *Streptomyces coelicolor* is occupied by membranous material (Fig. 1), which is often continuous with the plasma membrane (Figs. 2 and 3). The individual membranes are "double," consisting of two dense layers, each 2 to 3 mμ thick, separated by a lighter region, the whole complex having a thickness of about 9 mμ. In our preparations the plasma membrane also appears to be double (Fig. 3), as described by Tokuyasu and Yamada (12) in *Bacillus subtilis*, and in contrast to the single structure observed by Kellenberger and Ryter (4) in *Escherichia coli*, and it has the same dimensions as the membranes in the cytoplasm. In some regions the membranes in the cytoplasm appear to have a random orientation with respect to each other (Fig. 1, *M*), while in others they are associated in an ordered arrangement to form discrete bodies with oval or circular outlines. In some of these bodies the membranes are closely packed (Fig. 4), while in others they are separated by larger spaces containing cytoplasmic granules (Fig. 5). Thus the membranes have some features in common with those of the endoplasmic reticulum of mammalian cells (9). Some of the membranous bodies are similar to the cytoplasmic inclusions observed by Ryter, Kellenberger, Birch-Andersen, and Maaløe (10) in *Bacillus subtilis* and by Kellenberger, Ryter, and Séchaud (5) in *Escherichia coli*, which these authors term "chondrioids"; they also bear some resemblance to the "mitochondria" observed in *Mycobacterium tuberculosis* by Shinohara, Fukushi, Suzuki, and Sato (11). Sometimes the membranes of the discrete bodies are seen to be continuous with those in the less ordered regions (Fig. 5), and in other areas the arrangement of the membranes shows varying degrees of regularity (Fig. 1). It seems, therefore, that we are dealing with a single system of membranes, and many of the different appearances observed may, in fact, be due to cutting the membranous system in different planes. Membranous bodies are often found in apparent association with cross-walls, and frequently there is one on either side of a wall (Figs. 4 and 6). This suggests that the bodies may be concerned in the formation of the walls.

The continuity of the membrane system with the plasma membrane is of interest in relation to the work of Mitchell and Moyle (6) on gram-positive bacteria. They postulate that the bulk of the cytochrome system and many other enzymes are situated in the plasma membrane, and it is possible that a proliferation of this membrane allows a greater intensity of certain types of enzymic activity. The concentration of the membranous elements into discrete regions may account for the appearance of localised sites of oxidation-reduction in mycobacteria treated with mitochondrial stains (7). Such an association between sites of oxidation-reduction and specialised regions of the cytoplasm in *Escherichia coli* has been suggested by Niklowitz (8).

It appears from this investigation that the cytoplasmic organisation in *Streptomyces coelicolor* is more complex than that described in other bacteria. This is probably due in part to better preservation of the fine structure by improved methods of fixation, and to examination of only the thinnest sections; details of the membranous system are not seen in thicker sections, in which the sole indication of the system is an occasional ill defined membranous body. It must be borne in mind, however, that we are dealing with an actinomycete, a member of one of the most advanced groups of bacteria as judged by external morphology. This organism, therefore, may possess a greater degree of complexity of fine structure than the eubacteria which have been the subject of more numerous investigations.

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Explanation of Plates

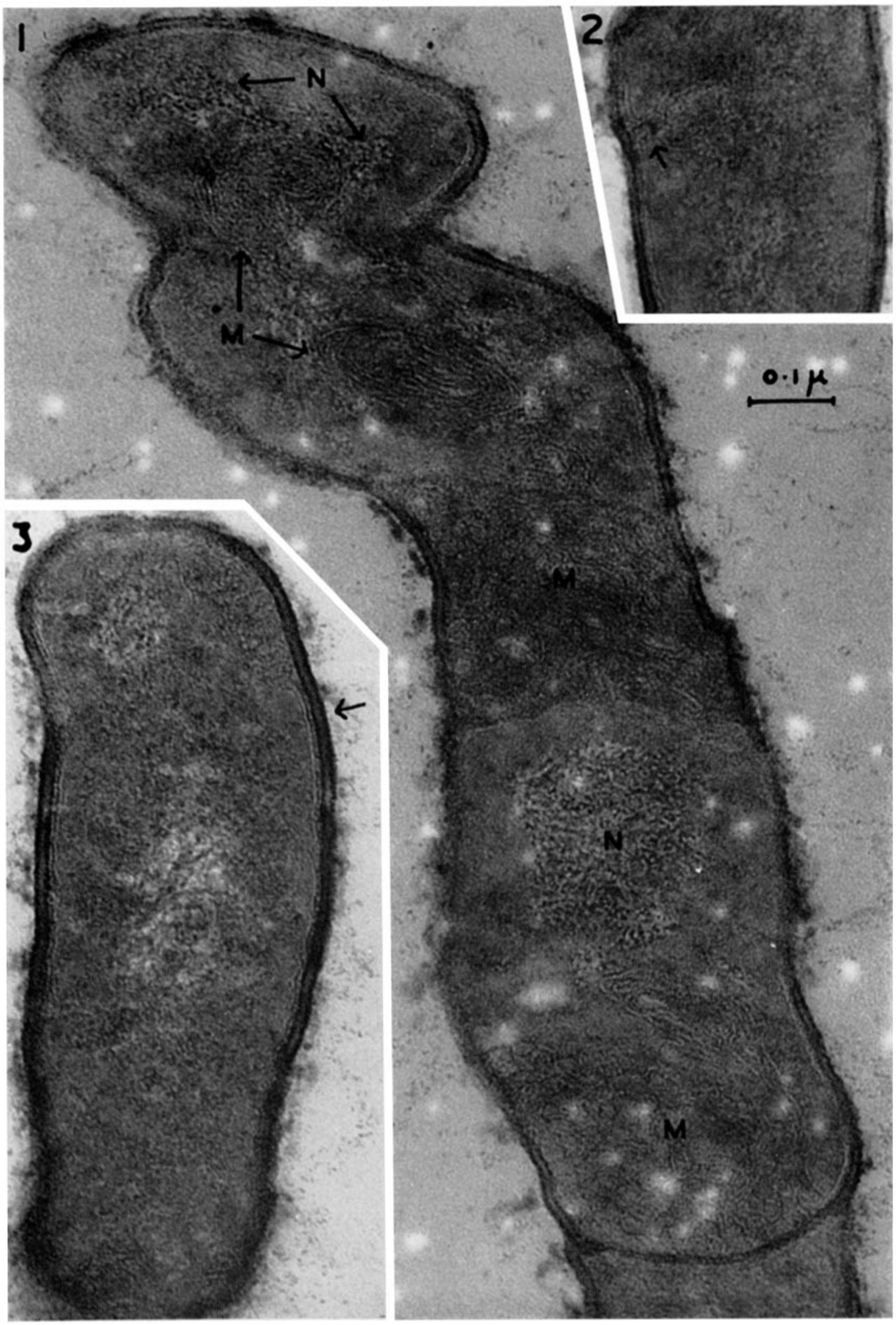
Plate 249

Electron micrographs of thin sections of hyphae of *Streptomyces coelicolor*. × 140,000.

Fig. 1. The membranous component of the cytoplasm (*M*) occupies a large part of the hypha. In places it is in close association with the fibrous nuclear material (*N*). In some regions the membranes have an apparently random orientation with respect to each other; in others they appear more ordered. Above the cross-wall at the bottom of the picture the membranous component is in continuity with the plasma membrane. (The scattered light spots are due to small holes in the carbon support film.)

Fig. 2. The membranous component of the cytoplasm in continuity with the plasma membrane (arrow).

Fig. 3. The double plasma membrane is clearly visible (arrow) underlying the cell wall.



(Glauert and Hopwood: Membranous component of cytoplasm)

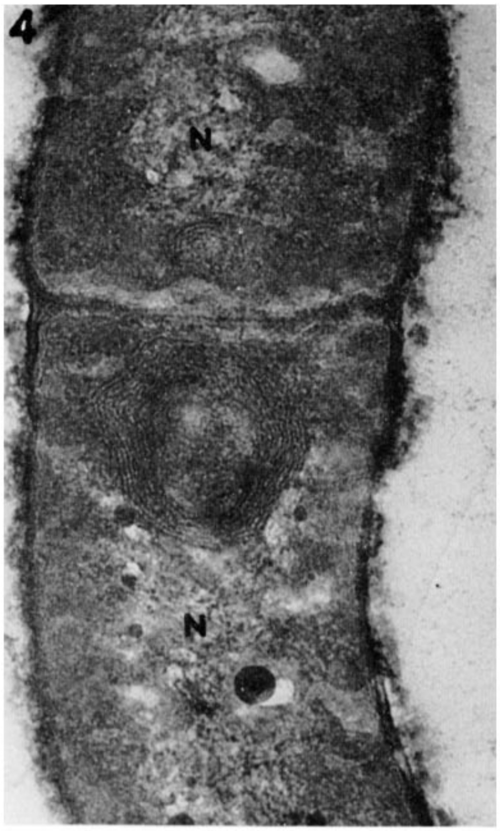
PLATE 250

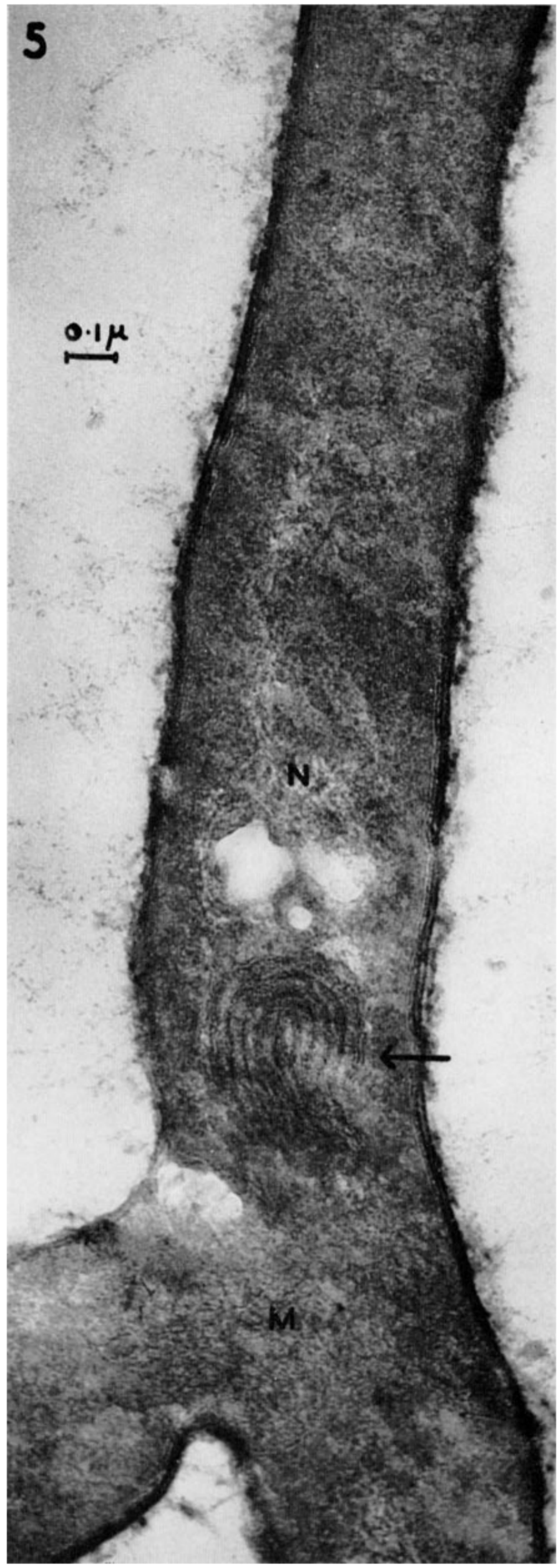
Electron micrographs of thin sections of hyphae of *Streptomyces coelicolor*. N = nuclear material. × 70,000.

FIG. 4. A body with closely packed membranes is visible below the cross-wall, and a smaller one above it.

FIG. 5. A body (arrow) in which the double membranes are separated by spaces containing cytoplasmic granules is in continuity with the membranes of a less ordered region (M).

FIG. 6. Membranous bodies (M) are visible on either side of a cross-wall.





(Glauert and Hopwood: Membranous component of cytoplasm)