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OBSERVATIONS ON THE FINE STRUCTURE OF *MICROSPORUM AUDOUINI**

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No account of the fine structure of *Microsporum audouini* has been published. No startling differences were found when the fine structure of *M. audouini* was compared with that of closely related forms. Thus, to date, the electron microscope has been of no aid in taxonomic determinations; yet, rather frequently subtle morphologic variations in organoids and inclusions exist. The attempt herein to clarify and restrict the usage of the term "Woronin body" based on differences in fine structure and position is an example of the contribution of EM observation.

METHODS

Microsporum audouini was cultivated in the following medium: phytone 10.0, dextrose 10.0, agar 15.5, cycloheximide 0.4, and chloramphenicol 0.05. The hyphae were fixed in 2 percent unbuffered potassium permanganate. As always, the addition of a detergent was used to facilitate penetration (1).

The hyphae were subsequently rinsed in distilled water, dehydrated in a graded series of ethanol, and embedded in Maraglas (2). Polymerization was carried out at 60° C for 48 hours. Sections were cut with a diamond knife using a Porter-Blum MT-2 microtome. All sections were doubly stained with a 1.5 percent aqueous solution of uranyl acetate for 1 minute, dipped in 3 changes of distilled water, dried, and then stained again with lead citrate for 5 minutes (3). Photographs were taken with a HU-11A Hitachi electron microscope.

OBSERVATIONS

The hyphae of *M. audouini* were identical to all others we have investigated; i.e., filamentously elongate structures. Figure 1 illustrates such morphology and, in addition, dichotomous branching is evident. The cell wall (Fig. 3) was a double structure consisting of an outer, relatively thin but electron-dense layer and an inner, much broader and slightly fibrillar, electron-lucid layer. Internal and contigous to the wall was a thin electron-dense plasma membrane which appeared to be a single entity (Fig. 1); however, in previous work on *Microsporum gypseum,* the plasma membrane was resolved into a trilaminar structure (4). Thus the plasma membrane is not to be construed as a single structure, rather the classical "unit membrane" (5). Again septal walls with pores were seen (Fig. 2). In and at either side of the septal pore, 5 electron dense bodies are discernible. We have mentioned them in two instances other than in this investigation (1, 6). These septal "plugs" always had a single membrane and contents of uniform electron opacity. These bodies showed a distinct predilection for the immediate vicinity of the septal pores. The significance of septal pores and the "plugs" as shown herein and in *M. canis* and *M. gypseum* was covered quite adequately in *Coprinus lagopus* (7). However, we have reported relative to yet another type of spherical electron-dense body in both *Rhizopus rhizopodiformis* and *Microsporum canis* (1, 8). In these two instances, the vesicles (not to be confused with "plugs") were located uniformly just inside of the plasma membrane and showed central areas of high electron density. With light microscopy it would be impossible to detect these morphological differences.

Recently a brief review of septal pores, associated "plugs," and sundry other opaque bodies appeared (9). Some of the bodies were referred to as Woronin bodies—a description of which appeared in the literature about a century ago (10). It is our belief that the term "Woronin body," if it is to be introduced again, should be restricted to the electron dense bodies which are present in the immediate vicinity of the septal pores.

Binucleated and multinucleated hyphae were quite common. The latter can be seen in Fig.

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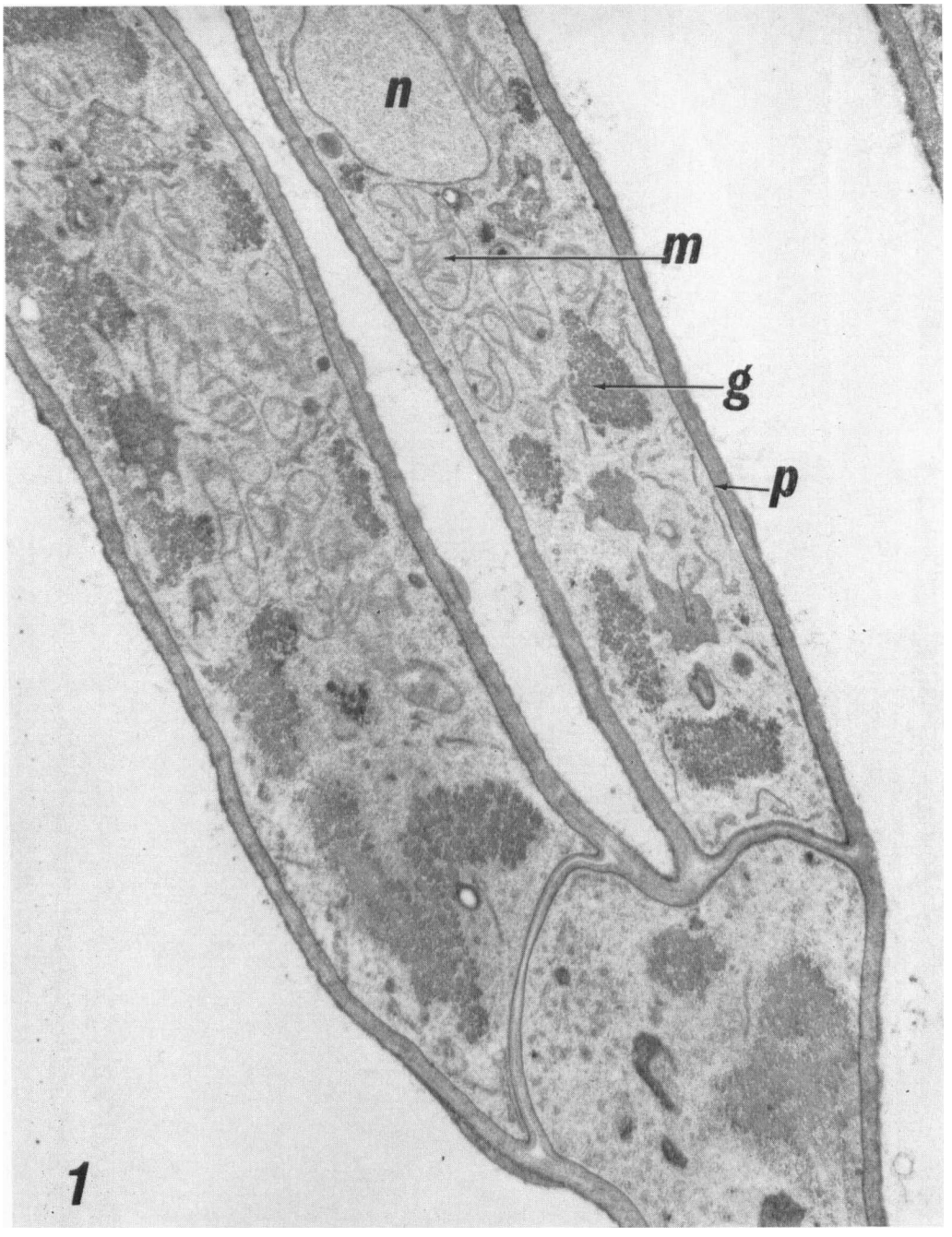
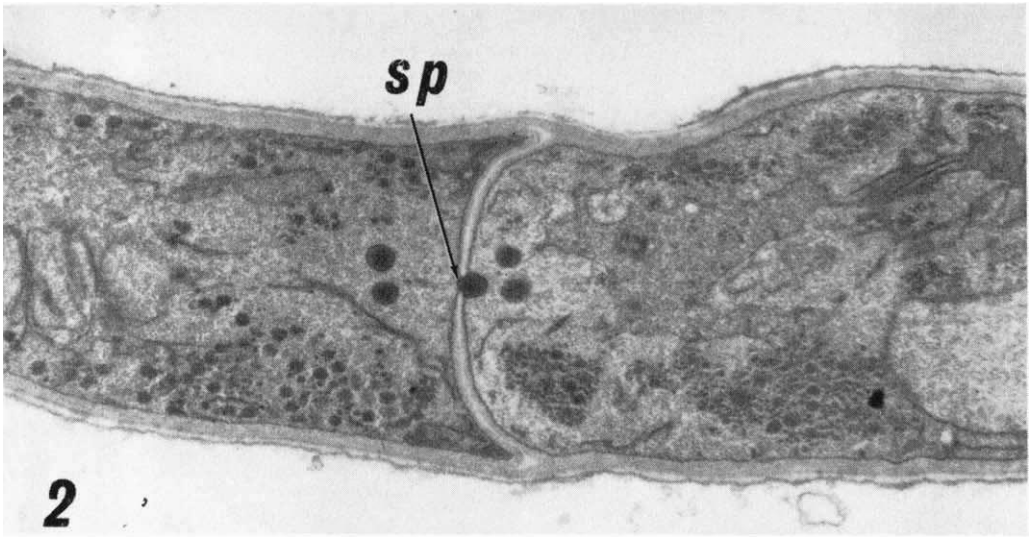


FIG. 1. Longitudinal section of a dichotomous branching hypha showing the plasma membrane (p), glycogen (g), mitochondria (m), and nucleus (n). × 12,500.



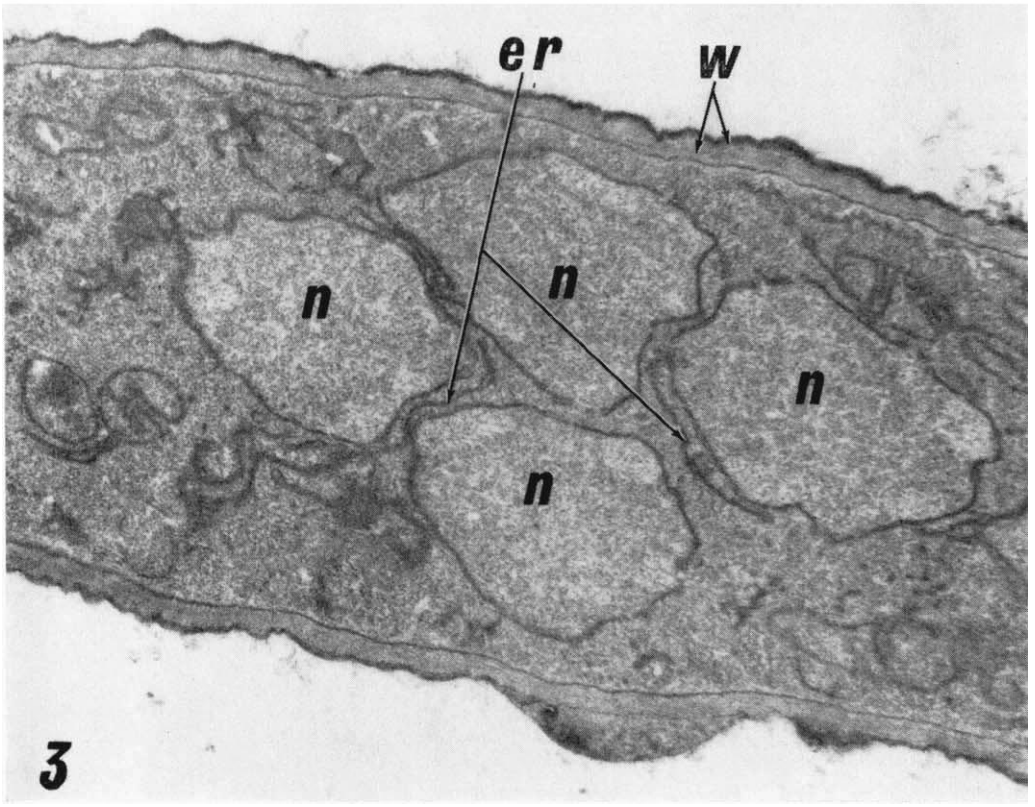


FIG. 2. Longitudinal section showing a septal pore (sp) and 5 electron dense bodies, probably homologous to the Woronin bodies of light microscopy. × 15,000.

FIG. 3. Longitudinal section showing the 2 layers of the cell wall (w), a typical multinucleated condition (n), and the endoplasmic reticulum (er). × 17,500.

3. In Fig. 3 the nuclear membranes are obviously double and porous.

The endoplasmic reticulum was randomly distributed throughout the cytoplasm. This organelle was sparse and most commonly appeared as long, narrow tubules (Fig. 3).

Mitochondria were present abundantly. The usual morphology was evident—double membranes with internal cristae. We noticed that a type of polarity existed in that the long axis of a mitochondrion conformed with the long axis of the cell.

Glycogen (Fig. 1) was invariably present in clumps through the cell.

SUMMARY

A study of the fine structure of *M. audouini* showed that the walls of the hyphae consisted of a thin, outer, electron-dense area and a broad, inner, fibrillar electron-lucid area. A thin electron-dense plasma membrane was contiguous with the inner surface of the cell wall. Electron dense bodies in the region of the septal pores were considered as being homologous to Woronin bodies. Hyphae were characteristically multinucleated. Mitochondria were abundant and the narrow, tubular endoplasmic reticulum was sparse. The inclusion, glycogen, was always present.

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