Antonie van Leeuwenhoek **51** (1985) 121–128

Ultrastructure of septa in *Blastobotrys* and *Sporothrix*

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SMITH, M. TH. and BATENBURG-VAN DER VEGTE, W. H. 1985. Ultrastructure of septa in *Blastobotrys* and *Sporothrix*. Antonie van Leeuwenhoek **51**: 121–128.

Transmission electron micrographs of septa in *Blastobotrys* species invariably showed central micropores. Septa of species of *Sporothrix,* however, exhibited three types of pores: (a) micropores which were central if single, or scattered; (b) central simple pores with Woronin bodies; (c) dolipores. The results confirm the heterogeneity of the genus *Sporothrix*.

INTRODUCTION

The taxonomic significance of septal ultrastructure is being increasingly recognized (Tu and Kimbrough, 1978). Three basic types of septal pores are known, viz. dolipores, simple pores and micropores (Kreger-Van Rij and Veenhuis, 1973). In the first, septa are characteristically swollen around the central pore canal. In Basidiomycetes the dolipore is provided with a usually well-developed pore cap. The dolipores known in ascomycetous yeast-like fungi have no caps (Kreger-Van Rij and Veenhuis, 1969, 1973).

Simple pores are found in septa of Ascomycetes. The pores are usually single, central pores; one account of septa with several simple pores has been published (Reichle and Alexander, 1965). Woronin bodies are mostly present near simple pores, although absence of these bodies has been reported (Trinci and Collinge, 1973; Mason and Crosse, 1975).

A third type of septal pore is the micropore. The term is generally used to indicate the narrow, randomly distributed perforations of a septum (Hashimoto et al., 1964; Takada et al., 1965; Van der Walt et al., 1983); they may also be arranged in a circle parallel and close to the cell wall (Kreger-Van Rij and Veen-

huis, 1972). In some cases, however, there is only one micropore in the centre of the septum (Kreger-Van Rij and Veenhuis, 1973, 1974).

A marked taxonomic heterogeneity is supposed to occur in the *Blastobotrys/Sporothrix*-complex (De Hoog et al., 1985; Weijman and De Hoog, 1985). Therefore, concurrently with the mentioned studies of morphology, physiology and carbohydrate patterns, the ultrastructure of the septa of species assigned to *Blastobotrys* and *Sporothrix* was examined.

Materials and Methods

The studied specimens are listed in Table 1. Cultures were grown on YPG agar at 25°C except for CBS 140.71, CBS 421.78 and CBS 530.83 A, which were grown at 17°C. After the cells were washed with water, they were fixed with 1.5% aqueous $KMnO_4$ for 30 min at room temperature, washed again with water and centrifuged in Beem capsules. The pellets were dehydrated in an ethanol series, stained with 1.5% uranyl acetate for 2 h at the 50% ethanol step and finally embedded in Spurr resin. Ultrathin sections were cut with a Reichert model OM-04 ultra-microtome, poststained with Reynolds lead citrate for 10 sec and examined with a Philips model EM 201 electron microscope at 60 kV.

Results and Discussion

The types of septal pores found are summarized in Table 1. The hyphal septa of all strains assigned to the genus *Blastobotrys* by De Hoog et al. (1985) are characterized by the presence of one, central micropore (Figs 1–3). The results indicate that the genus is uniform with respect to the ultrastructure of the septa. Similar central micropores were found in *Sp. fungorum,* and in *Stephanoascus farinosus* (Figs 4–5). The micropores showed a random distribution in *Sp. guttuliformis* (Fig. 6) and *St. ciferrii* (Figs 7–8). The location of micropores was attributed taxonomic significance by some authors (Kreger-Van Rij and Veenhuis, 1973, 1974). The value of this feature, however, seems to be limited, since two *Stephanoascus* species with *Sporothrix*-like anamorphs had septa with one central and with several scattered micropores, respectively.

In general the data coincide with those given by Weijman and De Hoog (1985) on the carbohydrate composition of whole cells. They found chromatograms of all *Blastobotrys* species to be characterized by a predominance of mannose and glucose and by the absence of both rhamnose and xylose. The authors suggested that the strains examined form a biological entity, which seems to be supported by the observations of the septal ultrastructure.

The hyphal septa of *Sporothrix* strains exhibited three different types of pores. In *Sp. luteoalba* (Fig. 9) as well as in *Sp. cyanescens* (Fig. 10) the hyphal septum

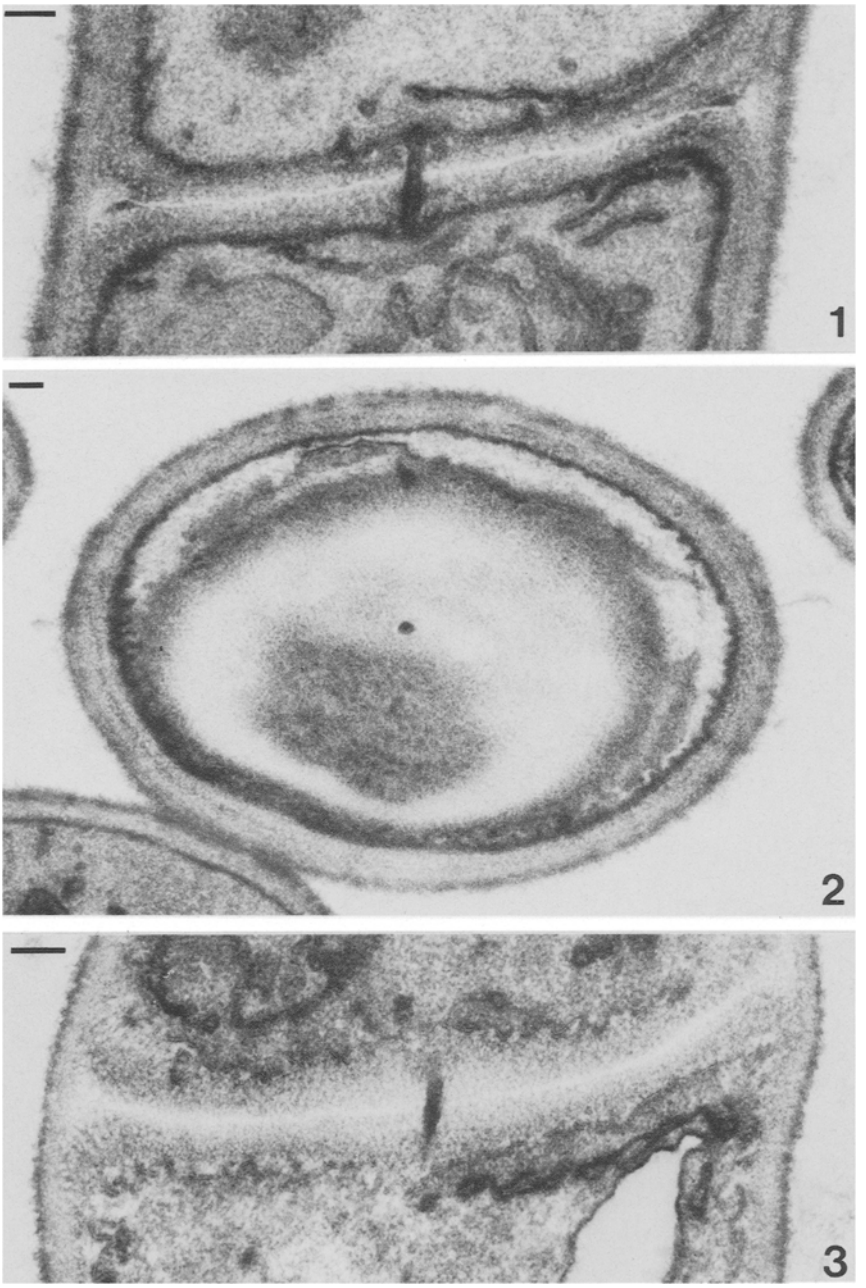

Fig. 1. *Blastobotrys proliferans* CBS 522.75. Transmission electron micrograph of a longitudinal section through a hyphal septum with one central micropore.
Fig. 2. *Blastobotrys proliferans* CBS 522.75. Transmission electron micrograph of a transverse section through a hyphal septum with one central micropore.
Fig. 3. *Blastobotrys* species 4 CBS 181.75. Transmission electron micrograph of a longitudinal section through a hyphal septum with one central micropore.
Bars represent 0,1 μm.

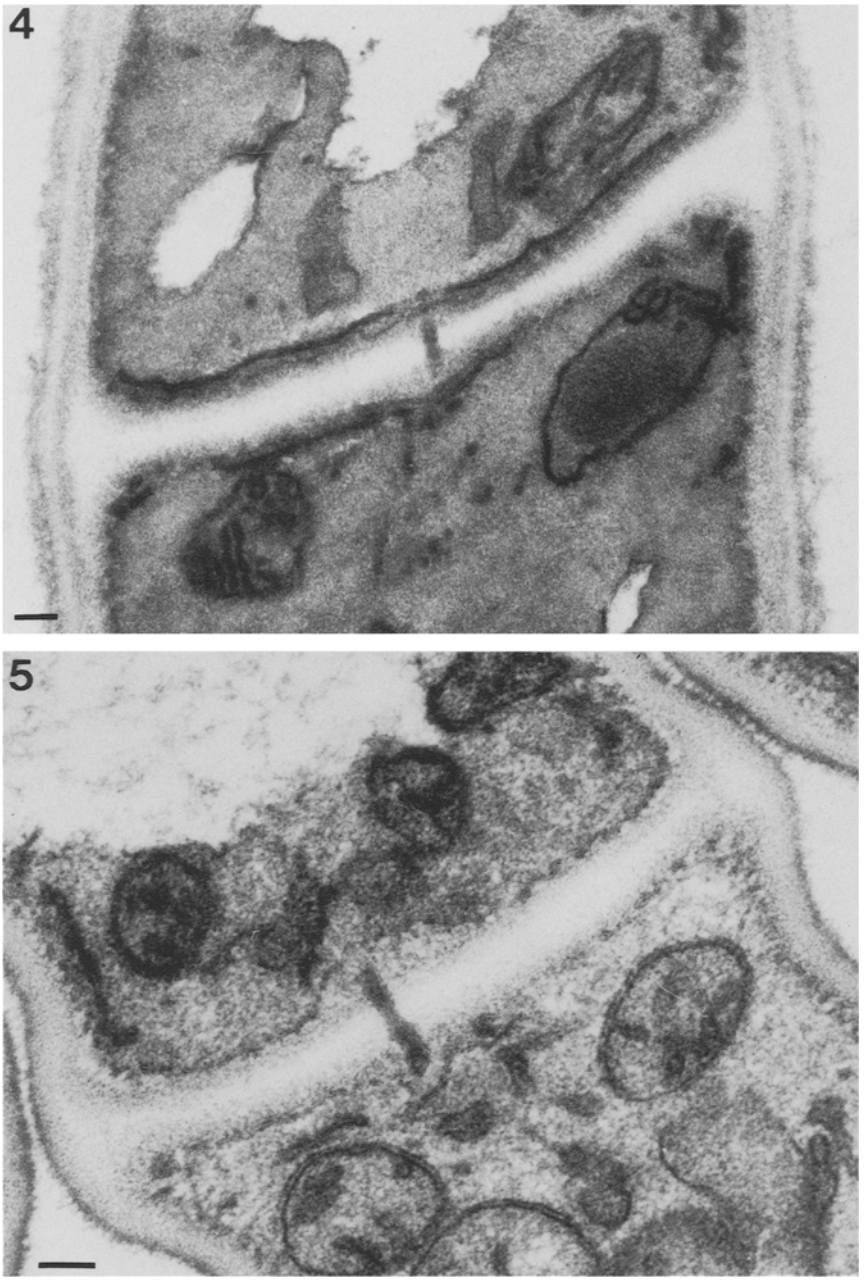

Fig. 4. *Sporothrix fungorum* CBS 259.70. Transmission electron micrograph of a longitudinal section through a hyphal septum with one central micropore.
Fig. 5. *Stephanoascus farinosus* CBS 140.71. Transmission electron micrograph of a longitudinal section through a hyphal septum with one central micropore.
Bars represent 0.1μm.

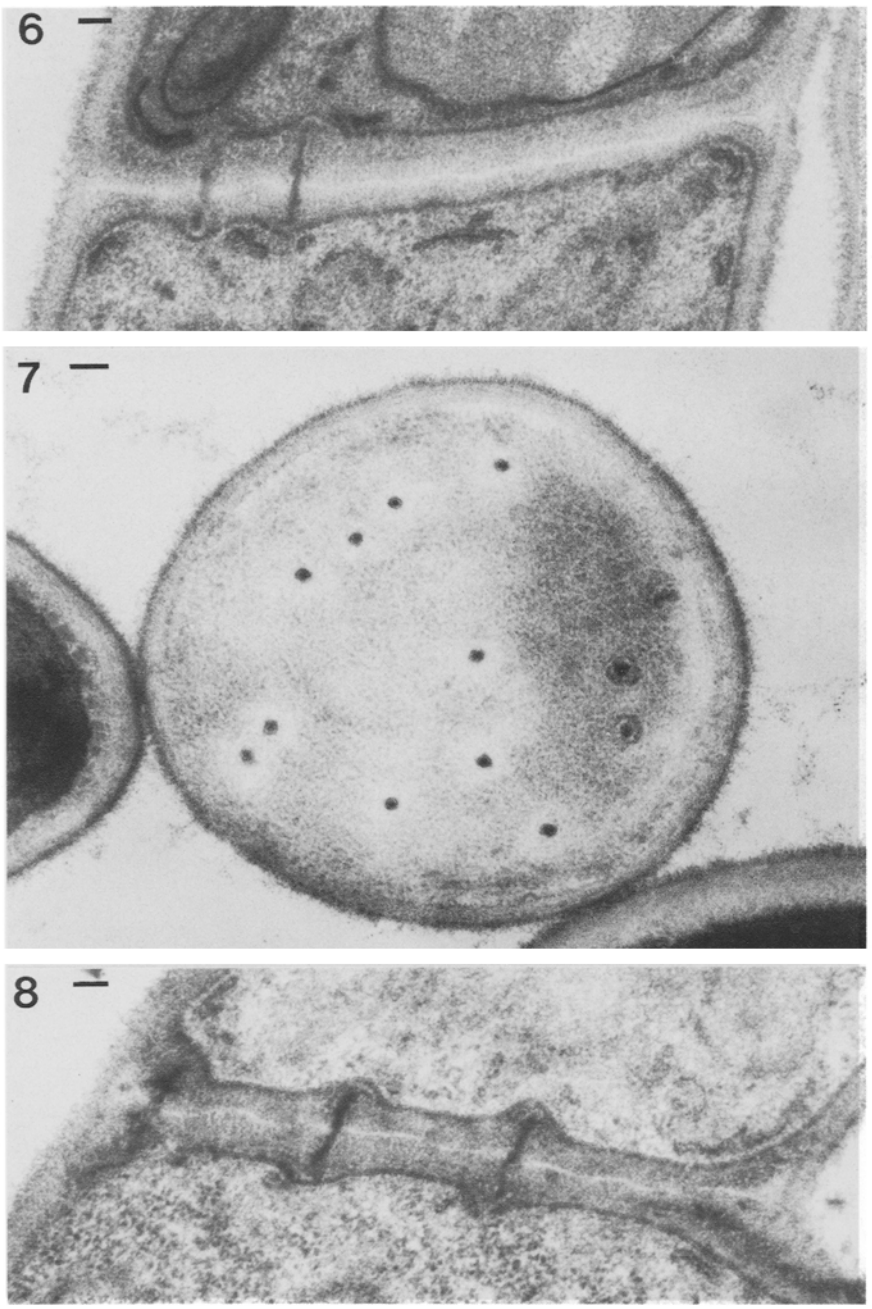

Fig. 6. *Sporothrix guttuliformis* CBS 437.76. Transmission electron micrograph of a longitudinal section through a hyphal septum with randomly distributed micropores.
Fig. 7. *Stephanoascus ciferrii* CBS 6699. Transmission electron micrograph of a transverse section through a hyphal septum with randomly distributed micropores.
Fig. 8. *Stephanoascus ciferrii* CBS 6699. Transmission electron micrograph of a longitudinal section through a hyphal septum with randomly distributed micropores.
Bars represent 0.1 μm.

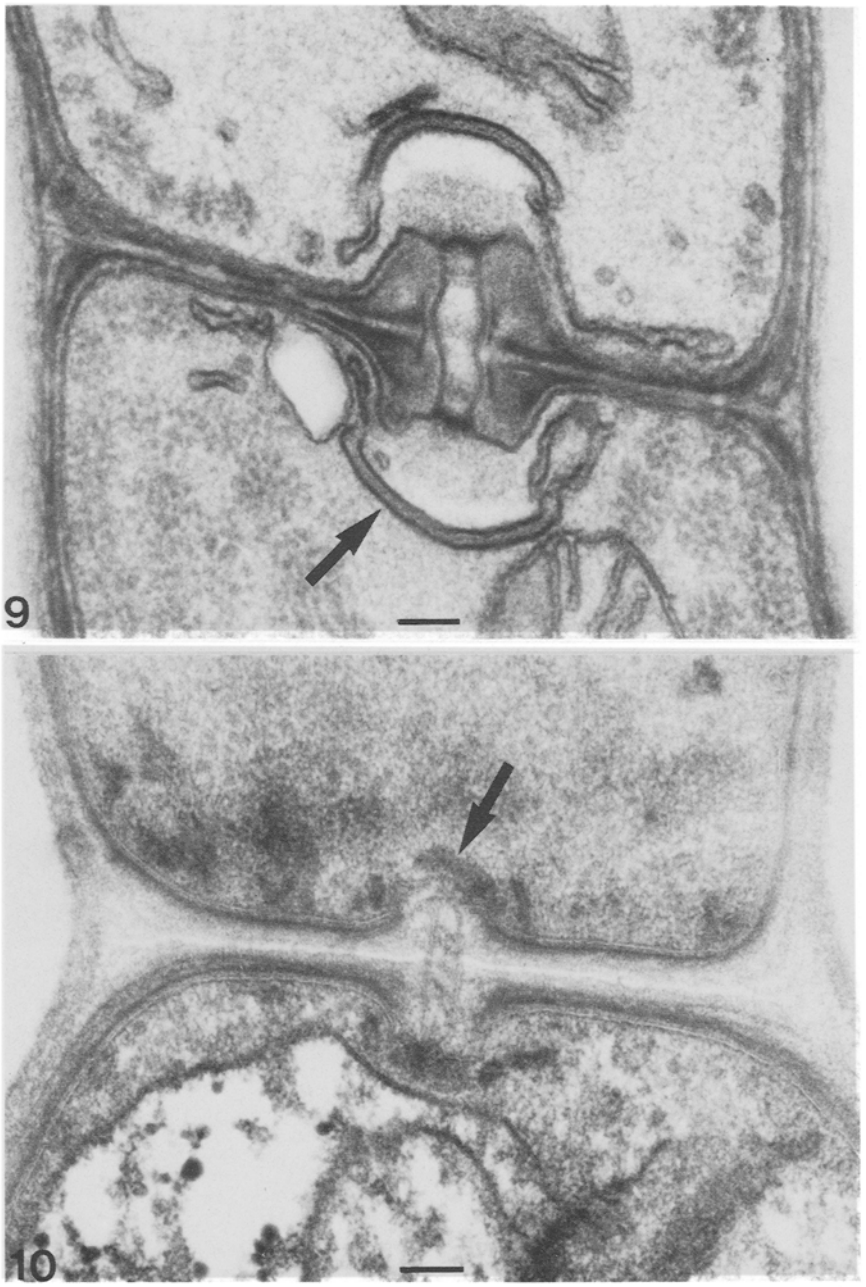

Fig. 9. *Sporothrix luteoalba* CBS 209.48. Transmission electron micrograph of a longitudinal section through a hyphal septum with a dolipore. A pore cap is present (arrow).
Fig. 10. *Sporothrix cyanescens* CBS 357.73. Transmission electron micrograph of a longitudinal section through a hyphal septum with a dolipore. Electron-dense material (arrow) adjacent to the pore.
Bars represent 0.1 μm.

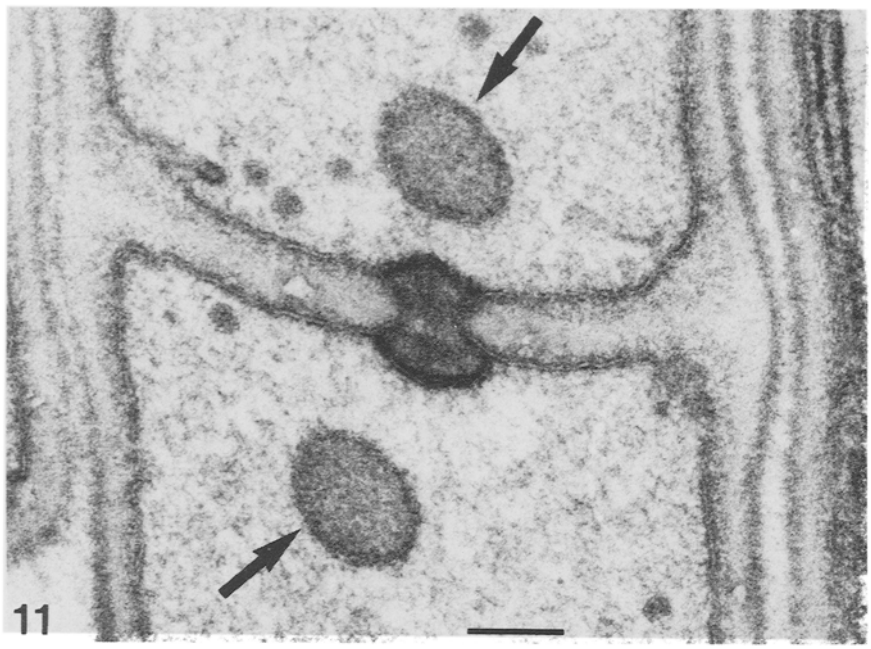

Fig. 11. *Sporothrix schenckii* CBS 292.55. Transmission electron micrograph of a longitudinal section through a hyphal septum with a simple pore. Woronin bodies are present (arrows). Bar represents 0.1 μm.

Table 1. Septal pores in *Blastobotrys* and *Sporothrix* species

Species		CBS number	Pore type	Number	Location
Blastobotrys	*arbuscula*	227.83	micropore	1	central
	aristata	521.75	micropore	1	central
	capitulata	287.82	micropore	1	central
	elegans	530.83A	micropore	1	central
	gigas	421.78	micropore	1	central
	nivea	163.67	micropore	1	central
	proliferans	522.75	micropore	1	central
	species 4	181.75	micropore	1	central
	species 9	151.83	micropore	1	central
Sporothrix	*cyanescens*	357.73	dolipore	1	central
	fungorum	259.70	micropore	1	central
	guttuliformis	437.76	micropore	several	random
	luteoalba	209.48	dolipore with pore cap	1	central
	schenckii	359.36	simple pore with Woronin bodies	1	central
	schenckii	292.55	simple pore with Woronin bodies	1	central
Stephanoascus	*ciferrii*	6699	micropore	several	random
	farinosus	140.71	micropore	1	central

showed a central dolipore with an imperforate pore cap in the former and with electron-dense material adjacent to the pore in the latter.

The hyphal septa of *Sp. schenckii* had only one simple, central pore (Fig. 11). In the vicinity of these, Woronin bodies were present. This type of ultrastructure indicates an affinity with higher, hyphal Ascomycetes (Curry and Kimbrough, 1983). The observation of the presence of basidiomycetous dolipores, ascomycetous simple pores and micropores in *Sporothrix* indicate that this genus is heterogeneous. These results concur with the data of Weijman and De Hoog (1985), who subdivided the genus into three sections. Ultrastructurally these sections can be characterized as follows: *Sporothrix* sect. *Luteoalba* Weijman & De Hoog has septa with dolipores; *Sporothrix* sect. *Sporothrix* has hyphal septa with simple pores and Woronin bodies; *Sporothrix* sect. *Farinosa* Weijman & de Hoog has septa with either one central micropore or several scattered micropores.

Received 3 May 1984

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