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DIFFERENCES IN SYNCHRONIZATION OF STAGES OF CONIDIAL DEVELOPMENT IN *LEPTOGRAPHIUM* SPECIES

By P. S. VAN WYK

Department of Plant Pathology, University of the Orange Free State, P.O. Box 339, Bloemfontein 9300, South Africa

M. J. WINGFIELD

Plant Protection Research Institute, Private Bag X5017, Stellenbosch 7600, South Africa

AND W. F. O. MARASAS

South African Medical Research Council, P.O. Box 70, Tygerberg 7505, South Africa

Stages involved in conidial and conidiogenous cell development in three *Leptographium* spp. are illustrated by SEM and TEM micrographs. A diagrammatic scheme is presented to illustrate duration of the different stages relative to each other and the effect this has on the morphology of the conidiogenous cell. Differences in the synchronization of the five stages of conidial development can result in conidiogenous cells that appear different but represent identical types of conidial development. Overlapping of the duration of secession and proliferation can create the illusion of sympodial development in species with enteroblastic percurrent proliferation such as *L. truncatum*.

Hughes (1953) recognized annellidic conidial development in *Leptographium* Lagerberg & Melin and established *Verticicladiella* Hughes to accommodate species in which conidia developed sympodially. Kendrick (1961) added *Phialocephala* Kendrick to include those species in which conidia were produced in phialides. These three genera, commonly referred to as the *Leptographium* complex (Kendrick, 1961, 1962, 1963) have thus been distinguished solely on the basis of annellidic (*Leptographium*), sympodial (*Verticicladiella*) and phialidic (*Phialocephala*) conidial development. This basis of differentiation has caused confusion and has been criticized (Kendrick, 1980; De Hoog & Scheffer, 1984; Minter, Kirk & Sutton, 1982; Tsuneda & Hiratsuka, 1984; Wingfield & Marasas, 1983). A detailed scanning electron microscopic (SEM) examination of conidial development in *Leptographium* and *Verticicladiella* revealed that various species in these genera develop both sympodially and percurrently (Wingfield, 1985). For this reason, *Verticicladiella* was reduced to synonymy with *Leptographium* (Wingfield, 1985).

Micrographs (SEM) of conidiogenous cells in some *Leptographium* species (Wingfield, 1985) show collarettes, apparently due to the displacement of outer wall layers. In some cases, this creates an impression of distinct phialides being present. This could cause confusion in distinguishing *Leptographium* from certain *Phialocephala* species. Wingield (1985) showed that *Phialocephala* includes species with apical wall-building conidiogenous cells (Minter *et al.*, 1982) as well as others more like *Chalara* (Corda) Rabenh. with ring wall-building development (Minter *et al.*, 1983). Conidium development in *Phialocephala* and *Leptographium* spp. was examined more closely by Wingfield, Van Wyk & Wingfield (1987), who placed species of *Phialocephala* with ring wall-building in *Sporendocladia* Arnaud ex Nag Raj & Kendrick. Critical examination of transmission electron micrographs (TEM) from the latter study revealed that conidial development in various species of *Leptographium* was different. In this paper, conidial development in three species of *Leptographium* is described, compared and interpreted. In addition, a diagrammatic scheme for the interpretation of conidial development according to the terminology of Minter *et al.* (1982) is introduced.

MATERIALS AND METHODS

Transmission and scanning electron micrographs of *L. terebrantis* Barras & Perry CBS 298.85 (Barras & Perry, 1971), *L. truncatum* (Wingfield & Marasas) Wingfield PREM 45698 = ATCC 58100 (Wingfield & Marasas, 1983; Wingfield, 1985) and *L. procerum* (Kendrick) Wingfield DAOM 33940 were examined. Material was prepared for SEM and TEM as described by Wingfield (1985).

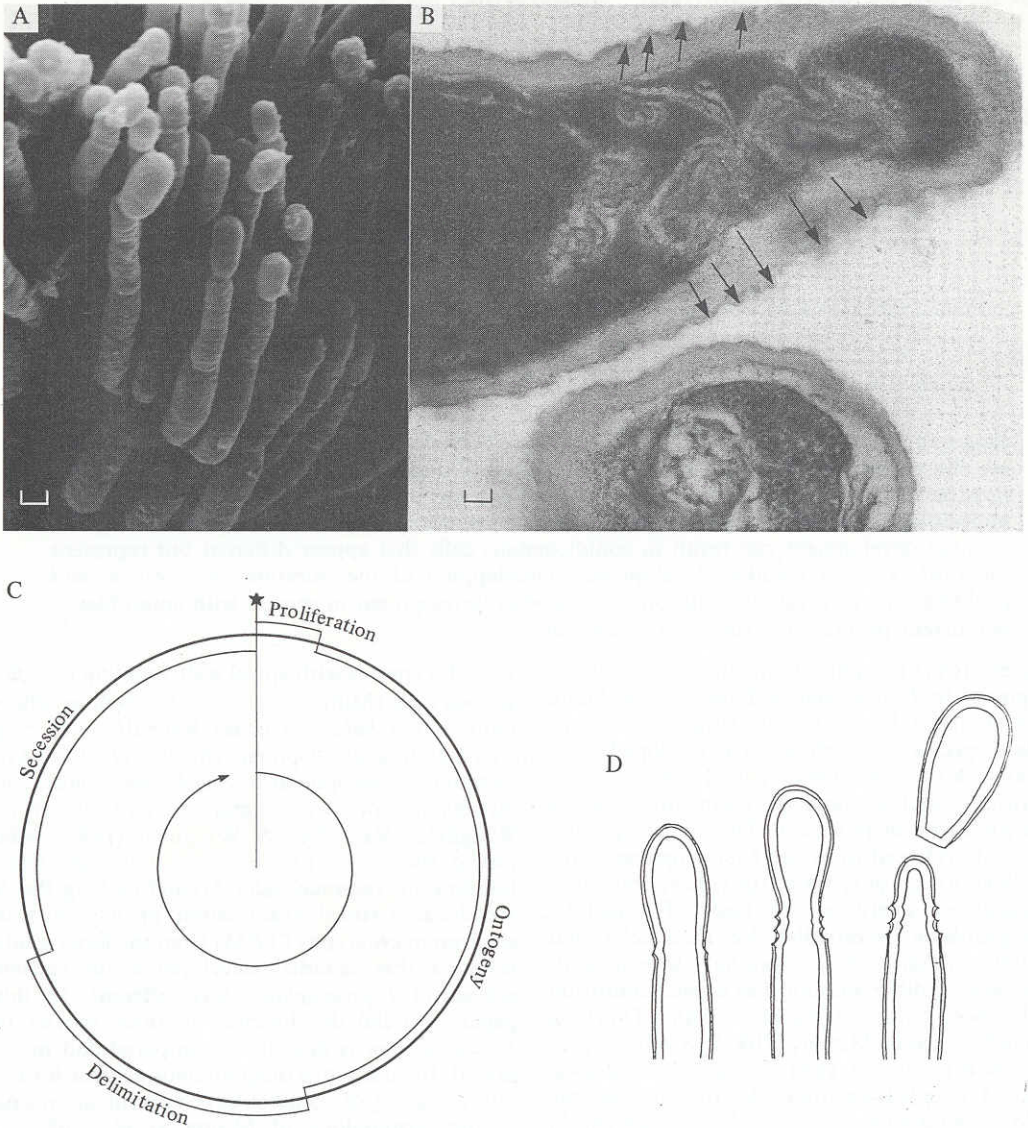
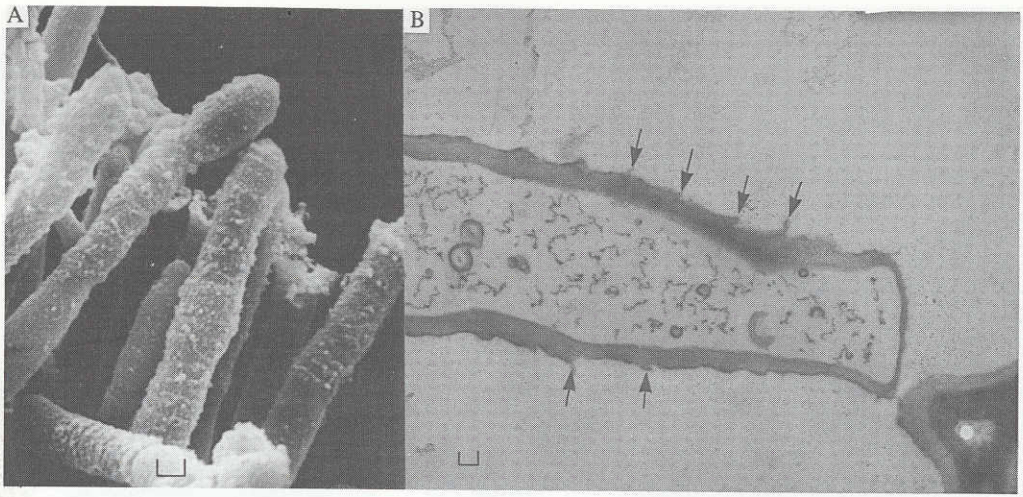


Fig. 1. A–D. Conidium development in *L. procerum*. (A) Conidiogenous cells (bar = 1 μm). (B) TEM section through a conidiogenous cell (bar = 0·1 μm). Compacted annellations as a result of very little proliferation (arrows). (C) Schematic representation of relative duration of four stages of conidial development. One full circle represents the completion of a single conidium. Star (★) indicates onset of regeneration. (D) Illustrative interpretation of conidial development.

RESULTS AND DISCUSSION

Minter *et al.* (1982) proposed that five stages, regeneration, proliferation, ontogeny (wall-building), delimitation and secession are involved in the production of conidia. In addition we suggest that differences in synchronization of these stages or time spent on some of these stages relative to others will markedly affect the morphology of the conidiogenous cell.

In this interpretation of *Leptographium* conidial development, it has been useful to compare these stages in diagrams (Figs 1 C, 2 C, 3 C). In this way



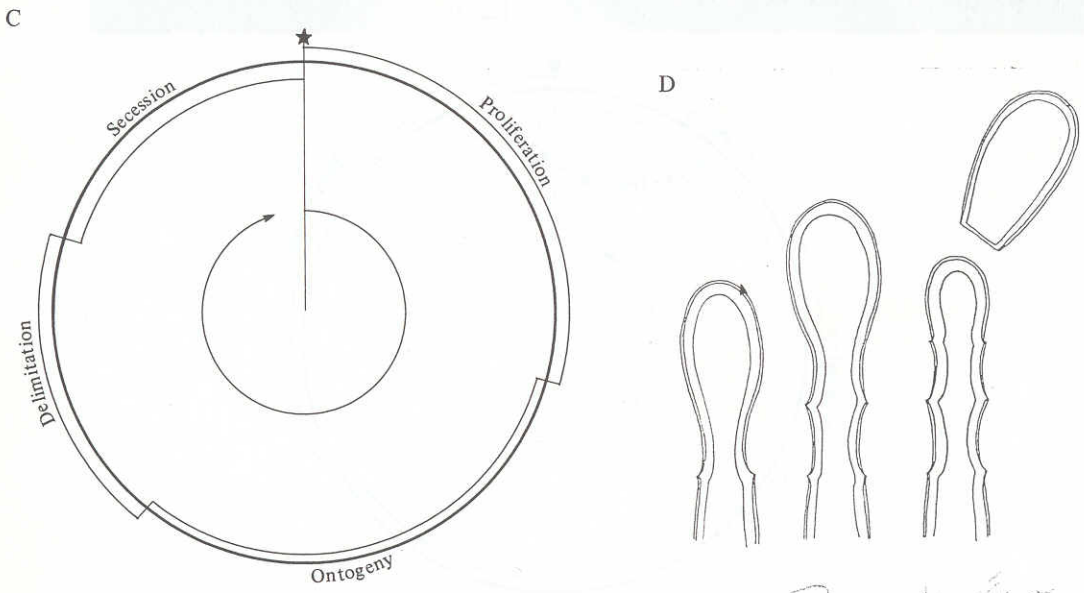


Fig. 2 A–D. Conidium development in *L. terebrantis*. (A) Conidiogenous cells (bar = 1 μm). (B) TEM section through a conidiogenous cell (bar = 0·1 μm). Obvious annellations as a result of considerable proliferation (arrows). (C) Schematic representation of relative duration of four stages of conidial development. One full circle represents the completion of a single conidium. Star (★) indicates onset of regeneration. (D) Illustrative interpretation of conidial development.

the duration of one stage relative to the next and how this affects conidiophore morphology can easily be visualized.

Percurrent proliferation of conidiogenous cells was observed in all species of *Leptographium* examined here and elsewhere (Wingfield, 1985; Wingfield *et al.*, 1987). However, the extent of the proliferation appears to vary markedly between species. In conidiogenous cells of *L. procerum*, annellations indicative of percurrent proliferation were not obvious in SEM micrographs (Fig. 1 A). Although highly compressed, they were, however, obvious in TEM micrographs (Fig. 1 B). In *L. procerum* very little proliferation occurs (Fig. 1 C);

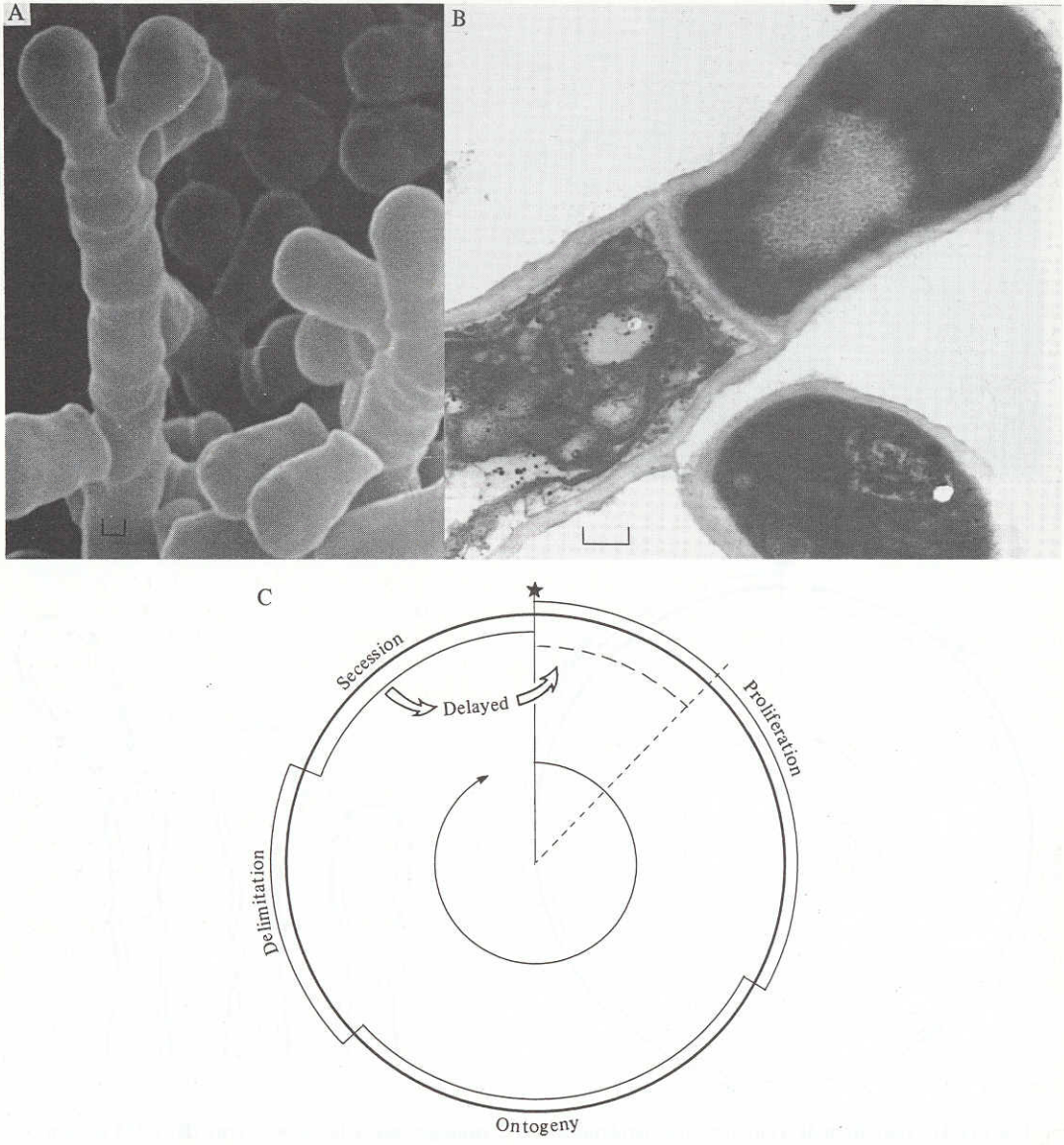


Fig. 3 A–C. Conidium development in *L. truncatum*. (A) Conidiogenous cells (bar = 1 μm). (B) TEM section through a conidiogenous cell (bar = 0·1 μm). (C) Schematic representation of relative duration of four stages of conidial development. One full circle represents the completion of a single conidium. Star (★) indicates onset of regeneration.

this results in compacted annellations (Fig. 1 D) that are not obvious except at very high magnification.

In *L. terebrantis*, the extent of percurrent proliferation that occurs between the ontogeny of each successive conidium (Figs 2 A, B) is greater (Fig. 2 C) than that in *L. procerum*. Consequently, annellations are more obvious (Fig. 2D). Conidial secession in *L. terebrantis* often leaves a greater portion of the outer wall layer on the conidiogenous cell (Fig. 2 B) than is observed in species such as *L. procerum*. These wall remnants on the conidio-

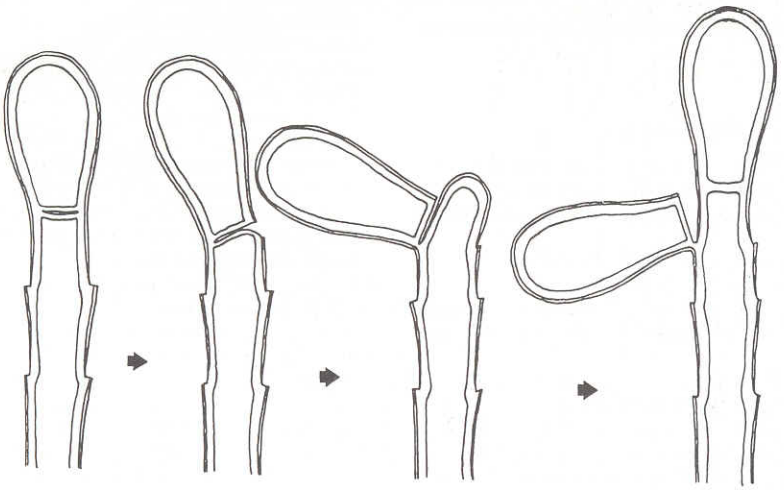


Fig. 4. Illustrative interpretation of conidial development in *L. truncatum*.

genous cell outer wall are fundamentally similar to those that make up collarettes of what have been termed phialides (Subramanian, 1971; Cole & Samson, 1979) in apical wall-building Hyphomycetes. However, extended proliferation leading to a distinctly annellate conidiogenous cell in *L. terebrantis* and other *Leptographium* spp. clearly differentiates between these and phialidic apical wall-building Hyphomycetes such as *Fusarium* sp. (Van Wyk *et al.*, 1987) and *Verticillium* sp. (Subramanian, 1971). In the latter fungi, all proliferation occurs below the base of the first collarette (Minter *et al.*, 1982, 1983). In *L. truncatum*, the five stages of conidial development are apparently not synchronized as they are in *L. procerum* and *L. terebrantis*. Extended proliferation, similar to that in *L. terebrantis* and unlike *L. procerum*, occurs (Figs 3A, B, 4) and distinct annellations are present. Conidial secession in *L. truncatum* is apparently delayed, and overlaps the onset of regeneration and proliferation (Fig. 3C). The result is that conidia are attached at either side of the conidiogenous cells. This creates an illusion of sympodial development in conidiogenous cells that proliferate percurrently (Fig. 4).

The illusionary sympodial development in *L. truncatum* and other *Leptographium* spp. (Wingfield, 1985) calls into question understanding of sympodial development as a whole. Cole & Samson (1979) describe sympodial development as 'a mechanism of conidiogenous cell proliferation in which each new growing point appears just behind and to one side of the previous apex, often resulting in a geniculate cell configuration'. Definitions of this process (Hughes, 1953; Ellis, 1971; Sutton, 1980; Cole & Samson, 1979) suggest that this type of development refers to the way in which the conidiogenous cell is rearranged to commence further conidium development after production of conidia has terminated. Using the terminology of Minter *et al.* (1982), the conidiogenous cell becomes non-functional after a conidium is produced, and the process that leads to production of the subsequent conidium is termed regeneration. This regeneration of the conidiogenous locus in a typical sympodial species such as *Tritirachium oryzae* (Vincens) de Hoog (Cole & Samson, 1979; Minter *et al.*, 1982) would be followed by holoblastic ontogeny. In contrast, *Leptographium* species that appear to have sympodial conidiogenous cells have enteroblastic percurrent proliferation followed by holoblastic ontogeny.

Results of this study suggest that differences in synchronization of the five stages of conidial development introduced by Minter *et al.* (1982) can result in conidiogenous cells that appear to be distinctly different, despite the fact that the processes involved in their conidial development are the same. Accurate interpretation of conidial development is significantly aided by consideration of the five developmental stages separately as well as in relation to each other. Sympodial development should refer to cases where conidiogenous cell proliferation is holoblastic or enteroblastic revitalization of the conidiogenous locus. This should not be confused with enteroblastic percurrent proliferation in *Leptographium*, where the sympodial arrangement of conidia apparently results from delayed conidial secession.

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