
SEM studies on the conidial morphology of some *Pestalotiopsis* species occurring on mangrove plants

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The conidial morphology of 6 species of *Pestalotiopsis*, viz., *P. agallochae* Pal & Purkayastha, *P. caseolaris* Purkayastha & Pal, *P. disseminata* (Thüm.) Stey., *P. mangiferae* (P. Henn.) Stey., *P. palmarum* (Cooke) Stey., *P. versicolor* (Speg.) Stey. have been studied under both light and scanning electron microscope (SEM). The characteristic differences between the conidia of relevant species are described with appropriate electron micrographs.

Key words: *Excoecaria agallocha*, frill, mangroves, Sundarbans, ultra-structure

INTRODUCTION

The conidial morphology is an important character in the taxonomic study of fungi. Sometimes surface ultra-structures of conidia are also considered as diagnostic characters in the identification of a particular taxon. A number of *Pestalotiopsis* species were isolated from several mangrove plants of Sundarbans, the largest mangrove complex of the world. This anamorphic and endophytic genus (Suryanarayanan *et al.*, 1998; Cannon and Simmons, 2002; Kumar and Hyde, 2004; Liu and Gus, 2007; Kamil *et al.*, 2012) is responsible for leaf spot disease of mangrove plants. *Pestalotiopsis* is a complex genus and consists of members difficult to classify at the species level (Kamil *et al.*, 2012). At present, inter-specific delineation of this genus is based on morphology of the conidia (Guba, 1961; Nag Raj, 1993), conidiogenesis (Sutton, 1980) and teleomorph association as well as the size of appendages and their number. In this investigation, their conidial morphology was studied under both light and scan-

ning electron microscopes with a view to determine whether any characteristic surface ultra-structure could be used in the identification of taxa under this investigation where only SEMs have been taken into critical account. The views of modern taxonomists still differ regarding the basic criteria used in delimiting the genera *Pestalotia* / *Monochaetia* / *Sematosporium* (Griffiths and Swart, 1974). It appears from the available literature that no work has been done so far on SEM studies on the conidia of the genus *Pestalotiopsis*. Hence, it was considered worthwhile to study the conidial morphology of six species of the genus *Pestalotiopsis* under SEM, which is expected to provide valuable biological information to those working in the field of fungal taxonomy.

MATERIALS AND METHODS

Six species of *Pestalotiopsis*, viz., *Pestalotiopsis agallochae* Pal & Purkayastha, *P. caseolaris* Purkayastha & Pal, *P. disseminata* (Thüm.) Stey., *P. mangiferae* (P. Henn.) Stey., *P. palmarum* (Cooke) Stey. and *P. versicolor* (Speg.) Stey. were isolated from *Excoecaria agallocha* L., *Sonneratia*

caseolaris (L.) Engl., *Aegiceras corniculatum* (L.) Blanco, *Derris scandens* Benth., *Phoenix paludosa* Roxb. and *Ceriops decandra* (Griff.) Ding Hou respectively. All the host species were found to grow in different regions of the Sundarbans mangrove forest (Eastern India). The cultures were maintained throughout in Potato-dextrose-agar medium. The identity of the cultures was confirmed by the International Mycological Institute (IMI), Kew, Surrey, England.

For SEM studies spores of the *Pestalotiopsis* species were removed from the culture tubes with a small metal spatula and put into a suspension of 95% ethanol. They were then pipette onto clean SEM stubs and left overnight to dry, before coating with platinum for 180 seconds in a BAL-TEC SCD 050 Sputter Coater. They were then examined using a Hitachi S-2400 SEM.

OBSERVATIONS AND DISCUSSION

Light microscopy:

Pestalotiopsis agallochae Pal & Purkayastha (Fig. 1. A); Pal & Purkayastha (1992). IMI 348454

Conidia fusiform, straight or slightly curved, 5-celled, slightly constricted at the septa, 20.4–27.2 x 3.4–6.8 μ m; 3 median cells thick-walled, 12.75–15.3 μ m long, olivaceous brown, the two upper cells faintly darker than lower; apical and basal cells thin-walled, hyaline; apical cell with 2–3 apical appendages, 8.5–17 μ m long with obtuse distal ends; basal cell truncate with a marginal frill and a single centrally inserted endogenous appendage, 3.4–10.2 μ m long.

Pestalotiopsis caseolaris Purkayastha & Pal (Fig. 1. B); Purkayastha & Pal (1993). IMI 356251

Conidia fusiform, straight, rarely curved, 5-celled, constricted at the septa, 10.2–22.1 x 3.4–6.8 μ m, 3 median cells thick-walled, 3.4–4.25 μ m long, olivaceous brown, in some cases middle cell is darker. Apical and basal cell hyaline; apical cell with 2 apical appendages or apical setulae (rarely 3), 5.1–10.2 μ m long; basal cell with single, hyaline, straight appendage, 1.7–6.8 μ m long.

Pestalotiopsis disseminata (Thüm.) Stey. (Fig. 1. C); Guba (1961). IMI 352585

Conidia fusiform to slightly clavate, straight, rarely

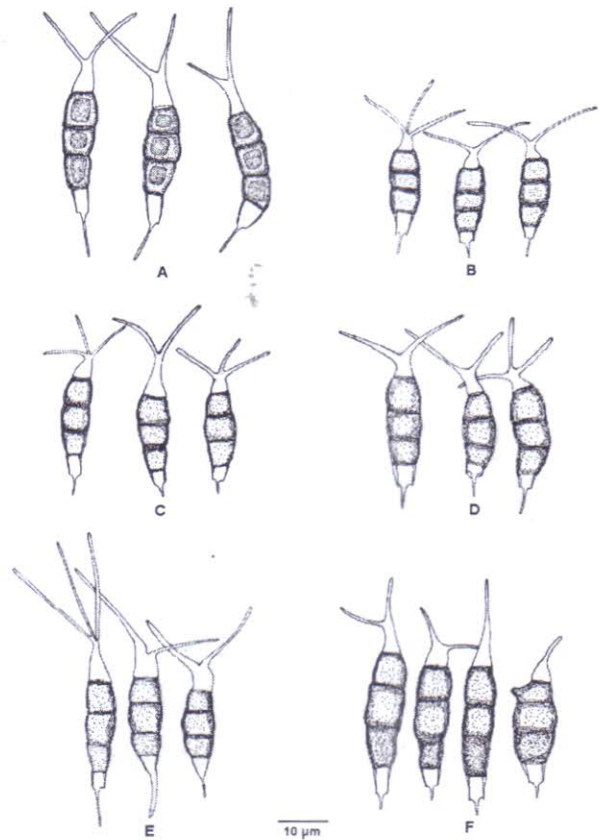


Fig. 1 : Conidia of *Pestalotiopsis* under light microscope.

A. *P. agallochae* B. *P. caseolaris* C. *P. disseminata*
D. *P. mangiferae* E. *P. palmarum* F. *P. versicolor*

curved, 5-celled, hardly constricted at septa, 23.4–35.1 x 5.85–7.8 μ m; 3 median cells 3.4–4.25 μ m long, equally olivaceous or with the two superior median cells or the septum separating them from each other slightly more strongly pigmented. Apical and basal cells hyaline; apical cell with 2–3 hyaline apical appendages, 9.35–13.6 μ m long; basal cell with single straight, hyaline, 2.55–5.1 μ m long appendage.

Pestalotiopsis mangiferae (P. Henn.) Stey. (Fig. 1. D); Mordue (1980). IMI 351338

Conidia fusiform, straight or slightly curved, slightly constricted at septa, 13.6–18.7 x 4.25–5.1 μ m; 3 median cells equidistantly euseptate, thick-walled, 8.5–11.9 μ m long, olivaceous brown. Apical and basal cells thin-walled, hyaline; apical cell with 2–3 apical appendages (sometimes 4), 5.1–15.3 μ m long with obtuse distal ends; basal cell truncate with a single centrally inserted, endogenous appendage, 3.4–8.5 μ m long.

Pestalotiopsis palmarum (Cooke) Stey. (Fig. 1. E);

Mordue & Holliday (1971).

IMI 351333

Conidia fusiform, straight, rarely curved, 5-celled, slightly constricted at septa, 13.6–20.4 x 4.76–6.8 µm; 3 median cells 11.05–11.9µm long, olivaceous with the two superior median cells or single me

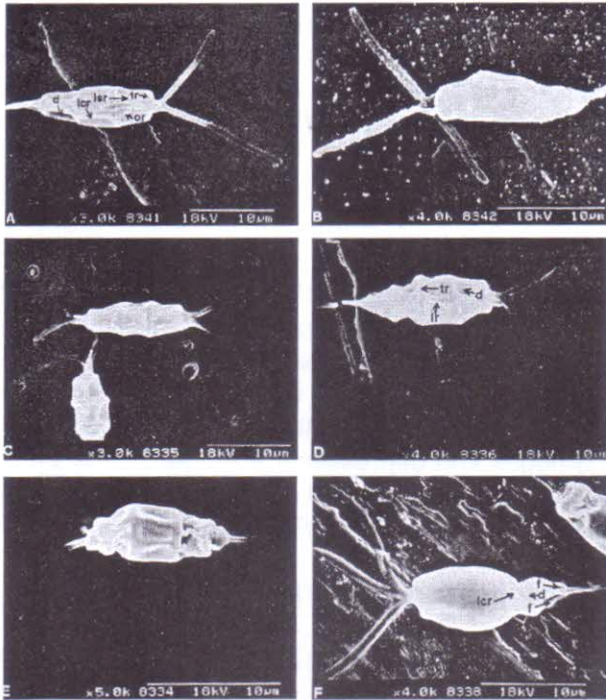


Fig. 2 : Scanning Electron Micrographs of conidia of *Pestalotiopsis* spp. A – B. *P. agallochae* C – E. *P. disseminata* F. *P. caseolaris*

dian cell darker than the inferior one. Apical and basal cells hyaline; apical cell with 3 apical appendages (rarely 2), hyaline, with obtuse apices, 5.1–13.6 µm long; basal cell with a hyaline, straight, 3.4–8.5 µm long appendage.

Pestalotiopsis versicolor (Speg.) Stey. (Fig. 1. F); Guba (1961). IMI 359184

Conidia fusiform, 5-celled, 25.5–37.4 x 6.8–8.5 µm, slightly constricted at the septa; 3 intermediate cells versicolourous (the two upper ones very dark). Apical and basal cells thin-walled, hyaline; apical cell with 2–3 apical appendages or apical setulae, 4.25–12.75 µm long; basal cell with a single endogenous hyaline, straight, basal appendage 1.7–3.4 µm long.

Scanning Electron Microscopy:

P. agallochae Pal & Purkayastha (Fig. 2. A & B)

Conidia fusiform, straight or slightly curved, 5-celled, more or less same as *P. palmarum*; diameter of the central body, however, appears to be smaller. Both the cells adjacent to the central body appears to have distinct depressions. Six longitudinal, straight or curved ridges and one each of transverse and oblique ridges along with distinct depressions were present on the spore wall. Apical cell bearing 2–3 long, smooth apical appendages (8.5 – 17 µm long), with obtuse single distal ends; basal cell with short, single centrally inserted, smooth basal appendage (3.4 – 10.2 µm long).

P. caseolaris Purkayastha & Pal (Fig. 2. F)

Conidial shape variable (fusiform, straight or rarely curved), 5-celled, central cylindrical body is devoid of any depression but the adjacent cell associated with the basal cell has got distinct depressions, unlike *P. disseminata*. Distinct longitudinal curved ridges were present on the surface of basal

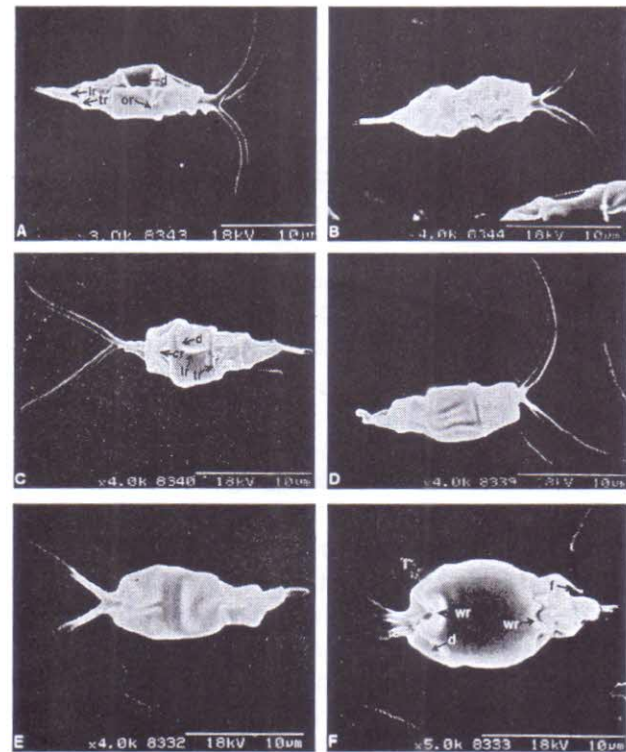


Fig. 3 : SEM of conidia of *Pestalotiopsis* spp. A – B. *P. mangiferae* C – D. *P. palmarum* E – F. *P. versicolor* lcr = longitudinal curved ridge; lsr = longitudinal straight ridge; lr = longitudinal ridge; tr = transverse ridge; or = oblique ridge; wr = wavy ridge; cr = curved ridge; d = depression f = frill.

cell adjacent to central cell. No such ridges were observed on central cell or apical cell, basal cell with two distinct marginal frills. Apical cell with 3

long, smooth, apical appendages or apical setulae (5.1 – 10.2 μm long), cylindrical to the obtuse distal ends; basal cell with single, straight, smooth, centrally inserted, long basal appendage (1.7 – 6.8 μm long).

P. disseminata (Thüm.) Stey. (Fig. 2. C-E)

Conidia fusiform to slightly clavate, straight, rarely curved, 5-celled, main body cylindrical. Central cells with distinct depressions, basal and apical cells adjacent to central cell showed transverse and longitudinal ridges on the surface. Apical and basal cells are smaller and tapering. Apical cells are provided with 2–3 apical appendages (rarely one) like protozoan flagella; apical appendage long (9.35 – 13.6 μm), smooth and cylindrical to the obtuse distal ends; basal cell with single straight, smooth, centrally inserted long appendage (2.55 – 5.1 μm), surface ornamentation of conidia psilate i.e. without having ornamentation.

P. mangiferae (P. Henn.) Stey. (Fig. 3. A & B)

Conidia fusiform, straight or slightly curved; central cylindrical body has irregular depression. Both transverse, longitudinal and oblique ridges present on the surfaces of central, apical and basal cells; irregular depressions were also noticed on the cell surfaces. Both the cells adjacent to the apical and basal cell appears to have distinct depressions; apical cell with 2–3 long, smooth, apical appendage having obtuse distal ends (5.1 – 15.3 μm), basal cell with short straight to slightly curved basal appendage (3.4 – 8.5 μm).

P. palmarum (Cooke) Stey. (Fig. 3. C & D)

Conidia in this case are distinctly fusiform, broad at apical part, gradually tapering towards the basal part; 5-celled. Both longitudinal and transverse ridges were observed on the spore wall; apical cell adjacent to central cell showed a few curved ridges; depressions were also visible in some places on the wall. Apical cell bearing 2–3 long, smooth apical appendages with obtuse distal ends (5.1 – 13.6 μm); basal cell with very short, single, straight to slightly curved basal appendage (3.4 – 8.5 μm).

P. versicolor (Speg.) Stey. (Fig. 3. E & F)

Conidia fusiform, 5-celled, short, curved or wavy ridges and depressions were discerned on the

surfaces of apical and basal cells; 2 frills present in the basal cell, the central cylindrical body devoid of depressions, but the cells adjacent to apical and basal cells are provided with depressions. Apical cell with 2–3 apical setulae; apical setulae (4.25 – 12.75 μm), smooth, with obtuse distal ends and shorter than *P. disseminata* and *P. caseolaris*. In such cases where there are 2 setulae it appears that it is dichotomously branched and having a common base, the basal cell however, are like that of *P. disseminata*; basal cell with short basal appendage (1.7 – 3.4 μm).

SEM studies revealed three distinct group of conidia, viz., (1) ridges and depressions present throughout the surface of conidium, frills absent in the basal cell (e.g. *P. agallochae*, *P. disseminata*, *P. mangiferae*, *P. palmarum*), (2) ridges and depressions restricted to the basal cell only, frills present in the basal cell (e.g. *P. caseolaris*) and (3) ridges and depressions are restricted to apical and basal cells or spread over throughout the spore wall, frills present (e.g. *P. versicolor*).

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