

Selenophoma eucalypti and *Stigmina robbenensis* spp. nov. from *Eucalyptus* leaves on Robben Island

P. W. CROUS¹, C. L. LENNOX¹ AND B. C. SUTTON²

¹ Department of Plant Pathology, University of Stellenbosch, 7600 Stellenbosch, South Africa

² International Mycological Institute, Bakeham Lane, Egham, Surrey TW20 9TY, U.K.

Collections of fungi associated with foliar lesions of *Eucalyptus* on Robben Island yielded two undescribed species, *Selenophoma eucalypti* and *Stigmina robbenensis*. *Selenophoma eucalypti* has also been collected in the forestry regions of the Cape and Transvaal Provinces. *Stigmina robbenensis*, however, is so far only known from the one collection.

Amongst a collection of foliar fungi of *Eucalyptus* made from Robben Island, two undescribed taxa were found. A species of *Selenophoma* Maire was consistently isolated from light brown, amphigenous, irregular leaf spots, which were mainly situated along the leaf margins. Single-conidial isolates produced a yeast-like growth when cultured on 2% malt-extract agar (Biolab) (MEA). When these isolates were placed on carnation-leaf agar (Fisher *et al.*, 1982; Crous, Phillips & Wingfield, 1992), however, they produced pycnidial conidiomata of the *Selenophoma* species which was originally isolated. A closer examination showed this fungus to produce a yeast-like synanamorph in culture. The *Selenophoma* species is also known from collections made in the Transvaal and Cape Provinces of South Africa. Two other fungi known from *Eucalyptus* resemble this species of *Selenophoma*. *Kabatiella dalgeri* M. Morelet (1968) is described with septate conidiophores having 1–4 conidiogenous loci per conidiogenous cell, 9.5–26.5 × 4–7.5 µm. Conidia are curved, cylindrical, 4–12 × 1.5–3 µm. The *Selenophoma* species is distinct in lacking septate conidiophores, having 1–2 loci per conidiogenous cell, and having larger conidia. *Cryptosporium eucalypti* Cooke & Harkn. (1881) is described from *Eucalyptus* twigs with

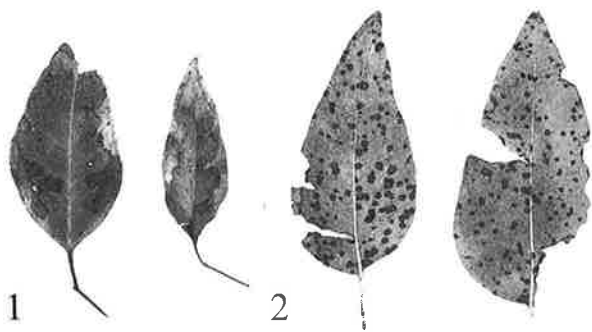
fusoid, abruptly curved conidia 20 × 3.5 µm. An examination of the type (K 1286) revealed dark brown to black acervuli opening by irregular rupture. Conidia are hyaline, fusoid, strongly falcate, 10–20 × 1.5–2 µm in size, forming on inconspicuous conidiogenous cells. The occurrence on twigs, black acervuli, and narrow, strongly falcate conidia clearly distinguish it from the foliicolous *Selenophoma* species. The collection from *Eucalyptus* is furthermore distinguished from the five species presently acknowledged in *Selenophoma* (Sutton, 1980) by having smaller conidia, and is therefore described as new.

Selenophoma eucalypti Crous, C. L. Lennox & B. Sutton sp. nov. (Figs 1, 3, 5–9, 17)

Maculae foliorum amphigenae, pallide brunneae ad brunneae, irregulares demum confluentes, praecipue in marginibus foliorum, 6–30 mm diam. *Mycelium* immersum, ex hyphis septatis olivaceis laevibus ad 4 µm longis compositum. *Conidiomata* pycnidialia, amphigena, praecipue epigena, separata, subepidermalia, ovoidea, cum laesione concolorata, strato basali saepe atriore brunneo, ex *textura angulare* parietum crassorum composita, ad 100 µm lata × 50 µm alta; ruptilia pariete superiore. *Conidiophora* nulla. *Cellulae conidiogena*e phialidicae, locis 1–2, saepe parietibus crassis, ex strato interiore parietis conidiomatis compositae et in parte inferiore tertio conidiomatis dispositae, ampullaceae ad cylindraceae basim tumidae, ad apicem contractae, rectae vel parum flexae, hyalinae, laeves; colla canalisque minuta, periclinale incrassatae, 10–20 × 3.5–5 µm. *Conidia* holoblastica, aseptata, hyalina, laevia, fusiformia, recta ad falcata, ad apicem acuta et basin subtruncata contracta, 8–15 × 2–3.5 µm.

Holotypus: South Africa, Transvaal, Pretoria, Fort Klapperkop, *Eucalyptus* sp., P. W. Crous, Apr. 1990, PREM 51727, IMI 341609 (isotypus).

Leaf spots amphigenous, light brown to brown, irregular, becoming confluent, mainly situated along the leaf margins, 6–30 mm in diam. *Mycelium* immersed, consisting of septate, olivaceous, smooth hyphae up to 4 µm wide. *Conidiomata* pycnidial, amphigenous, predominantly epigenous, separate, subepidermal, ovoid, concolorous with the lesion, basal layer



Figs 1, 2. Leaf symptoms. **Fig. 1.** Irregular lesions of *Selenophoma eucalypti*. **Fig. 2.** Distinct, circular lesions associated with *Stigmina robbenensis*.

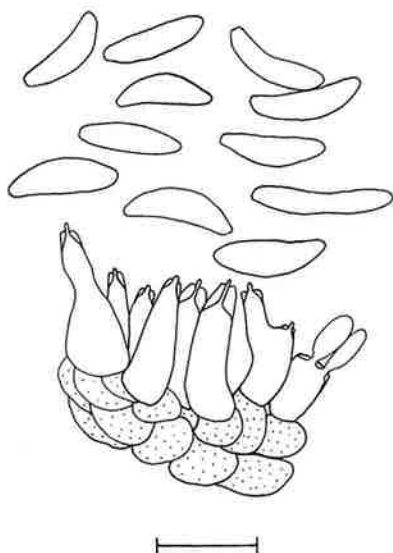


Fig. 3. Conidia and conidiogenous cells of *Selenophoma eucalypti* (bar = 10 μ m).

often darker brown, consisting of 2–3 layers of thin-walled *textura angularis*, up to 100 μ m wide \times 50 μ m high; dehiscence by irregular rupture of the upper wall. *Conidiophores* absent. *Conidiogenous* cells phialidic (*sensu* Sutton, 1980), with 1–2 loci, often thick-walled, formed from the inner layer of the conidiomatal wall and situated in the lower third of the conidioma, ampulliform to cylindrical with a swollen base tapered towards the apex, straight or slightly bent, hyaline, smooth; collarettes and channel minute, periclinal thickening present, 10–20 \times 3.5–5 μ m. *Conidia* holoblastic, aseptate, hyaline, smooth, fusiform, straight to falcate, tapering to an acutely rounded apex and subtruncate base, 8–15 \times 2–3.5 μ m.

Specimens examined: South Africa: Transvaal, Pretoria, Fort Klapperkop, *Eucalyptus* sp., P. W. Crous, Apr. 1990, PREM 51727

(holotype), IMI 341609 (isotype), IMI 359556 (ex type culture); Cape Province, Stilbay, *Eucalyptus* sp., P. W. Crous, Aug. 1993, PREM 51728; Robben Island, *Eucalyptus* sp., C. L. Lennox, Sept. 1993, PREM 51729.

Cultures are effuse, slimy, without aerial mycelium, and 19" d Buff (Rayner, 1970) on MEA. Single-conidial isolates have a minimum temperature requirement 5–10 $^{\circ}$ C, a maximum 30–35 $^{\circ}$, and an optimum growth at 25 $^{\circ}$, with colonies reaching a radial growth of 9 mm after 10 d at 25 $^{\circ}$ in the dark. Single-conidial isolates form a yeast-like synanamorph, with older conidia giving rise to new conidia, the loci being phialidic, with clearly visible collarettes. Conidia of the yeast state are falcate to fusiform, or more ellipsoid with bluntly rounded ends, 8–15 \times 2–7 μ m. Several mature conidia were also observed to become pigmented brown, verruculose and thick-walled, resembling chlamydospores. A similar phenomenon was also reported in the genus by Sutton (1980). The occurrence of yeast synanamorphs in mitosporic fungi is well known (Wang, 1979; Sutton, 1980), and the phenomenon of phialidic conidiogenous cells not unusual (von Arx, 1979).

Several of the leaves infected by *Selenophoma eucalypti* also had prominent lesions associated with a species of *Stigmina* Sacc. Presently there are four species of *Stigmina* known from *Eucalyptus* (Sutton & Pascoe, 1989). This collection has ellipsoid to cylindrical conidia which are 1–9-distoseptate, 10–30 \times 6–8 μ m. It can easily be distinguished from *S. hansfordii* B. Sutton & Pascoe which has ellipsoid to fusiform, 1-distoseptate conidia, 12–17.5 \times 6.5–8 μ m. The leaf symptoms of this collection resemble those of *S. eucalypticola* B. Sutton & Pascoe (1989). However, *S. eucalypticola* has ellipsoid to fusiform conidia which are 1–3 transversely distoseptate, and occasionally have a single longitudinal distoseptum. Furthermore, the conidiogenous cells of *S. eucalypticola* are ampulliform to doliiform or cylindrical, and form up to four

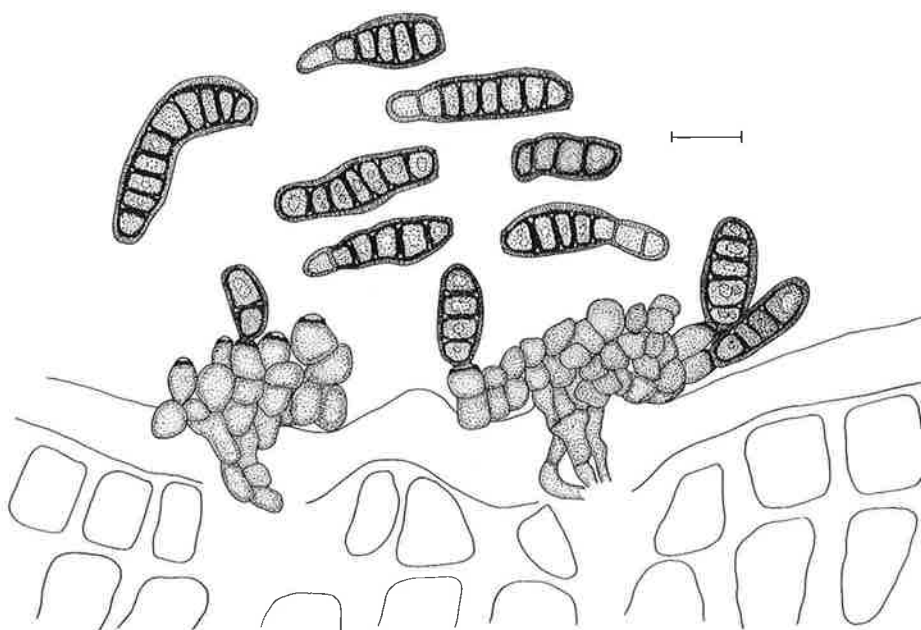
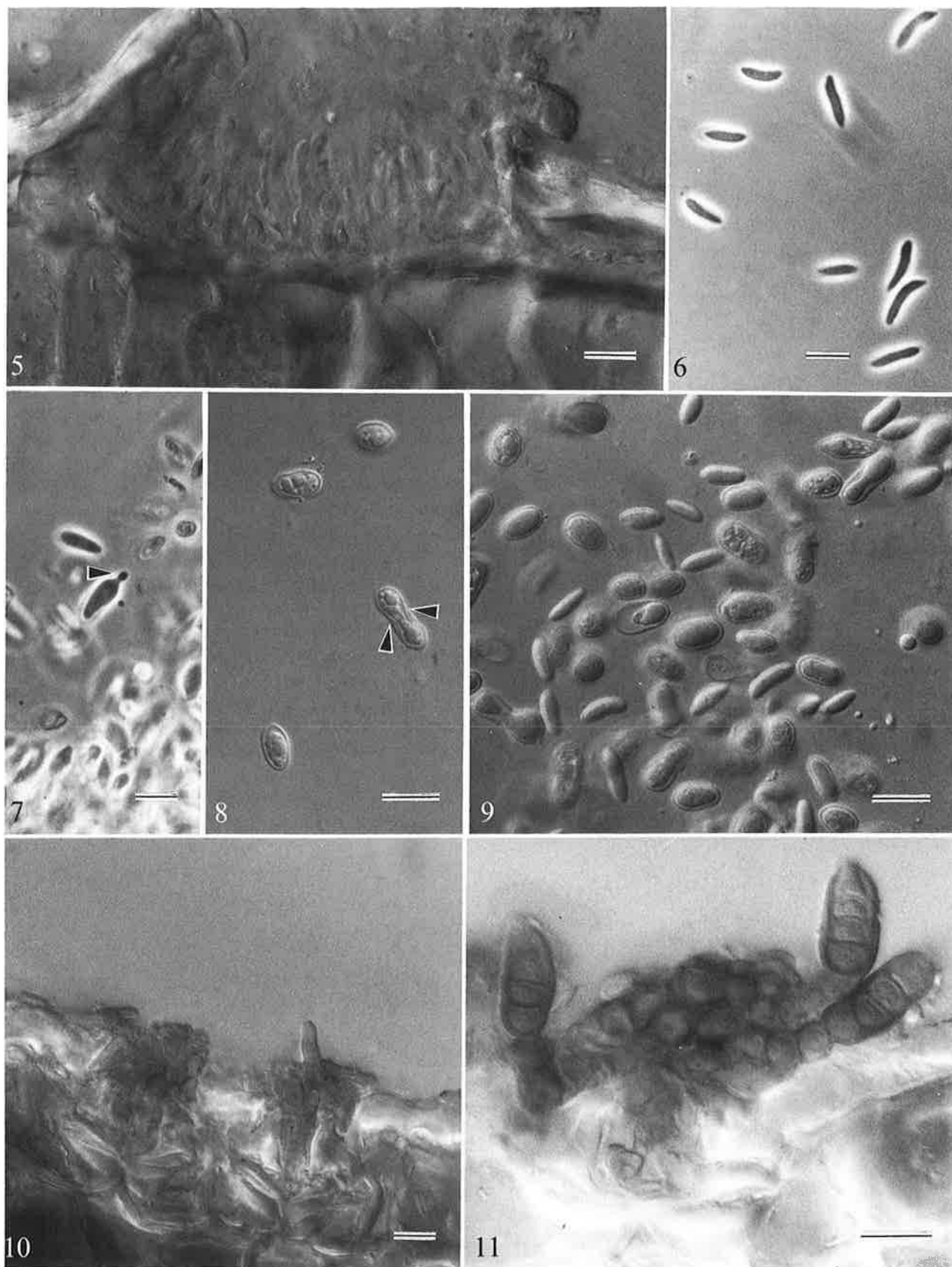
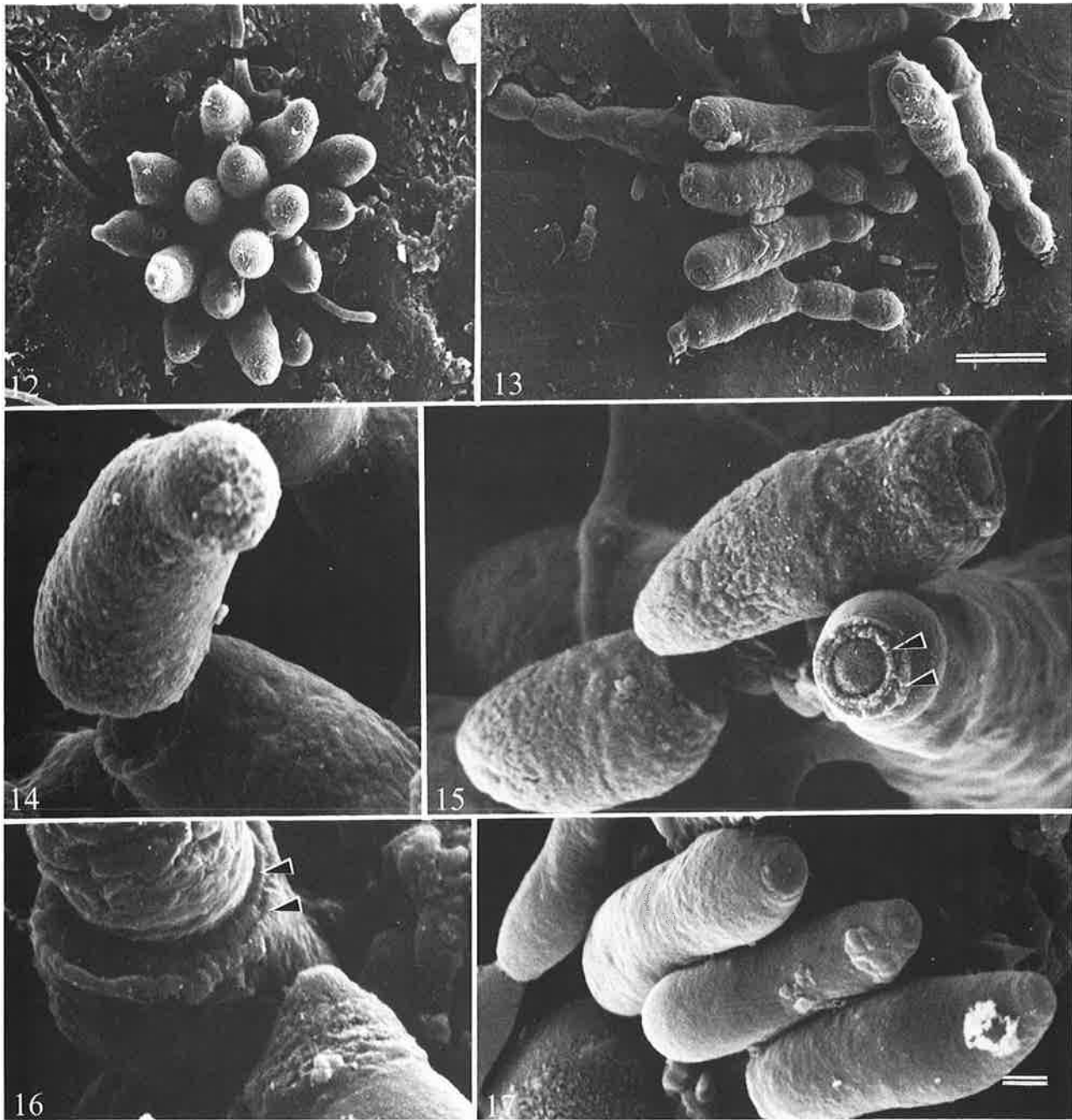


Fig. 4. Suprastomatal conidiomata giving rise to conidiogenous cells and dark brown distoseptate conidia of *Stigmina robbenensis* (bar = 10 μ m).



Figs 5–9. *Selenophoma eucalypti* (bars = 10 μ m). **Fig. 5.** Vertical section through a conidioma. **Fig. 6.** Falcate to fusiform conidia. **Fig. 7.** Budding conidium on MEA. **Fig. 8.** Primary conidium forming a secondary conidium enteroblastically, with clearly visible collarette. **Fig. 9.** Conidia of *S. eucalypti* and its yeast synanamorph on MEA. **Figs 10, 11.** *Stigmina robbenensis*. **Fig. 10.** Hyphae emerging through stomata, giving rise to conidiomata. **Fig. 11.** Vertical section through a conidioma with conidiogenous cells and conidia.



Figs 12–16. *Stigmina robbenensis*. **Fig. 12.** Conidiomata viewed from above. **Fig. 13.** Conidia on the leaf surface showing constrictions at some of their apical septa (bar = 10 μ m). **Fig. 14.** Conidium dehiscence. **Fig. 15.** Verruculose conidia with basal, marginal frills, lying next to a conidiogenous cell with percurrent proliferations (arrowed). **Fig. 16.** Verruculose conidiogenous cell with percurrent proliferations (arrowed). **Fig. 17.** Falcate conidia of *Selenophoma eucalypti* with flattened bases (bar = 1 μ m).

percurrent proliferations, whereas those of this collection are doliiform to cylindrical, and only have up to two percurrent proliferations. The more cylindrical conidia of this species resemble those of *Stigmina eucalypti* Alcorn, but can be distinguished by being smaller, and having more septa than those of *S. eucalypti*, which are markedly verrucose and mainly 3-septate, 20–36 \times 7.5–12 μ m (Sutton & Pascoe, 1989). It is similar to *Stigmina inconspicua* B. Sutton & Pascoe (1989) in having conidia of comparable size but differs in being only 1–3 septate. The *Stigmina* species occurring on eucalypts on Robben Island is therefore described as new.

Stigmina robbenensis Crous, C. L. Lennox & B. Sutton sp. nov. (Figs 2, 4, 10–16)

Foliorum maculae praecipue hypogaeae, saepe non per lamina extensae, atrobrunneae, centro albo et margine chlorotico, irregulariter circulares, separatae, 2–5 mm diam. *Mycelium* internum et externum, hypharum septatarum brunnearum, glabrarum ubi internarum, subtiliter scabrarum ubi externarum, ad 4 μ m latarum, stroma suprastomatatae producens 20–40 μ m latum et 15–20 μ m altum, cellularum atrobrunnearum pseudoparenchymatarum compositum. *Conidiomata* supra stomatibus, amphigena, praecipue hypogaea, sporodochialia, brunnea, 40–70 μ m lata, 20–30 μ m alta. *Conidiophora* nulla vel cellulae sustentis composita. *Conidiogaeae cellulae* sub-

stromata exorientes, doliiformes ad cylindratae, discretae, verruculosae, brunneae, proliferationibus ad duobus enteroblasticis, percurrentibus verrucosis, $5-10 \times 5-7 \mu\text{m}$. *Conidia* holoblastica, apicalia, solitaria, brunnea, $10-30 \times 6-8 \mu\text{m}$, verruculosa, 1-9 transverse distoseptata, ellipsoidea ad cylindratae, apice obtuso et base truncata margine undulato.

Holotypus: South Africa, Robben Island, *Eucalyptus* sp., C. L. Lennox, Sept. 1993, PREM 51730, IMI 359478 (isotypus).

Leaf spots predominantly hypogenous, frequently not extending through the lamina, dark brown, with a whitish centre and chlorotic margin, irregularly circular, separate, 2-5 mm diam. *Mycelium* internal and external, consisting of brown, septate hyphae that are smooth when internal, and finely roughened when external, up to $4 \mu\text{m}$ wide, giving rise to a suprastomatal stroma 20-40 μm wide and 15-20 μm high, composed of dark brown pseudoparenchymous cells. *Conidiomata* situated over stomata, amphigenous, predominantly hypogenous, sporodochial, brown, 40-70 μm wide, 20-30 μm high. *Conidiophores* absent or consisting of a supporting cell. *Conidiogenous cells* arising from the stromata, doliiform to cylindrical, discrete, verruculose, brown, with up to two enteroblastic, percurrent verrucose proliferations, $5-10 \times 5-7 \mu\text{m}$. *Conidia* holoblastic, apical, solitary, brown, $10-30 \times 6-8 \mu\text{m}$, verruculose, 1-9 transversely distoseptate, ellipsoid to cylindrical, with an obtuse apex and truncate base with a marginal frill.

(Accepted 5 September 1994)

It is interesting to note that *S. robbenensis* has thus far only been found on eucalypts occurring on Robben Island, and has not been observed on any eucalypts near the coast in the Cape Province, or elsewhere in the commercial forestry plantations of South Africa. Although Koch's postulates could be confirmed for *Selenophoma eucalypti* on 2-month-old *E. grandis* Hill ex Maid. seedlings, this could not be repeated for *S. robbenensis* as no cultures were obtained of the latter.

REFERENCES

- von Arx, J. A. (1979). Propagation in the yeasts and yeast-like fungi. In *The Whole Fungus*, vol. 2 (ed. B. Kendrick), pp. 555-571. National Museums of Canada: Ottawa, Canada.
- Crous, P. W., Phillips, A. J. L. & Wingfield, M. J. (1992). Effects of cultural conditions on vesicle and conidium morphology in species of *Cylindrocladium* and *Cylindrocladiella*. *Mycologia* **84**, 497-504.
- Fisher, N. L., Burgess, L. W., Toussoun, T. A. & Nelson, P. E. (1982). Carnation leaves as a substrate and for preserving cultures of *Fusarium* species. *Phytopathology* **72**, 151-153.
- Rayner, R. W. (1970). *A Mycological Colour Chart*. CMI and British Mycological Society: Kew, Surrey, England.
- Sutton, B. C. (1980). *The Coelomycetes*. CMI: Kew, Surrey, England.
- Sutton, B. C. & Pascoe, I. G. (1989). Reassessment of *Peltosoma*, *Stigmina* and *Batcheloromyces* and description of *Hyphothyrium* gen. nov. *Mycological Research* **92**, 210-222.
- Wang, C. J. K. (1979). Pleomorphic fungi imperfecti. In *The Whole Fungus*, vol. 1 (ed. B. Kendrick), pp. 81-91. National Museums of Canada: Ottawa, Canada.