

A comparison of the fungal genera *Phaeophleospora* and *Kirramyces* (coelomycetes)

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Phaeophleospora eugeniae Rangel, the type species of *Phaeophleospora* Rangel, has recently been collected and cultured from leaf spots of *Eugenia uniflora* L. in Brazil. Based on its brown, verruculose, percurrently proliferating conidiogenous cells, conidiomatal anatomy and conidial morphology, *Phaeophleospora* is resurrected as the earlier name for the recently established coelomycete genus, *Kirramyces* J. Walker, B. Sutton & I. Pascoe. New combinations are proposed for the seven former species of *Kirramyces*, namely *P. destructans* (W. J. Wingf. & Crous) Crous, F.A. Ferreira & B. Sutton, *P. epicoccooides* (Cooke & Massee) Crous, F.A. Ferreira & B. Sutton, *P. eucalypti* (Cooke & Massee) Crous, F.A. Ferreira & B. Sutton, *P. hebes* (W. Wu, B. Sutton & A.C. Gange) Crous, F.A. Ferreira & B. Sutton, *P. lillianiae* (J. Walker, B. Sutton & I. Pascoe) Crous, F.A. Ferreira & B. Sutton, *P. proteae* (B. Sutton) Crous, F.A. Ferreira & B. Sutton and *P. phormii* (Naito) Crous, F.A. Ferreira & B. Sutton comb. nov.

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Introduction

This study was initiated when the senior author had an opportunity while recently visiting the Department of Plant Pathology at the University of Viçosa, Brazil, to examine fungi collected by students which is a requirement of their mycology course. Some of the collections represented a coelomycete causing a leaf spot on *Eugenia uniflora* L. The latter fungus, identified as *Phaeophleospora eugeniae* Rangel, occurs commonly on trees around the campus, and is apparently an annual favourite of students for this course. This identification was intriguing, as it is well-known that most taxonomists dealing with coelomycetes regard the monotypic genus *Phaeophleospora* Rangel as a *nomen dubium sensu* Sutton (1977). Several collections of this fungus were available for examination in the department's herbarium, as this pathogen is apparently well-known on *E. uniflora*, a host which occurs throughout Brazil. *E. uniflora* is commonly called 'pitanga', which is the Indian word relating to its edible red fruit. This host is also sometimes planted to form hedges (Braga 1976).

In his examination of the type specimens to determine which generic names are acceptable for coelomycete genera, Sutton (1977) failed to locate the type specimen of *Phaeophleospora* Rangel. Because of the incomplete description by Rangel (1916), and the fact that genera in this complex are separated on a combination of conidiomata, conidia and conidiogenesis, this genus was referred to the *nomina dubia*. Although the holotype specimen has not been relocated for the present study, several new collections clearly representing the description and illustration provided by Rangel (1916) were available for study. *P. eugeniae* was originally described by Rangel as follows: 'Maculis amphigenis, sparsis, gregariis vel confluentibus, orbicularibus vel sub-orbicularibus, 1–3 mm diam, obscure-brunneis dein medio pallescentibus; pycnidiis paucis, epiphyllis, irnmersis, epiderride vestitis dein vix erumpentibus, ovoideis vel subovatis, imperfecte evolutis, late apertis (ca. 40 µm), olivaceis, 100–160 µm diam. Sporulis vermiformibus vel clavato-elongatis, apice rostratis, deorsum obtusis, multiseptatis, haud constrictis, filligi-

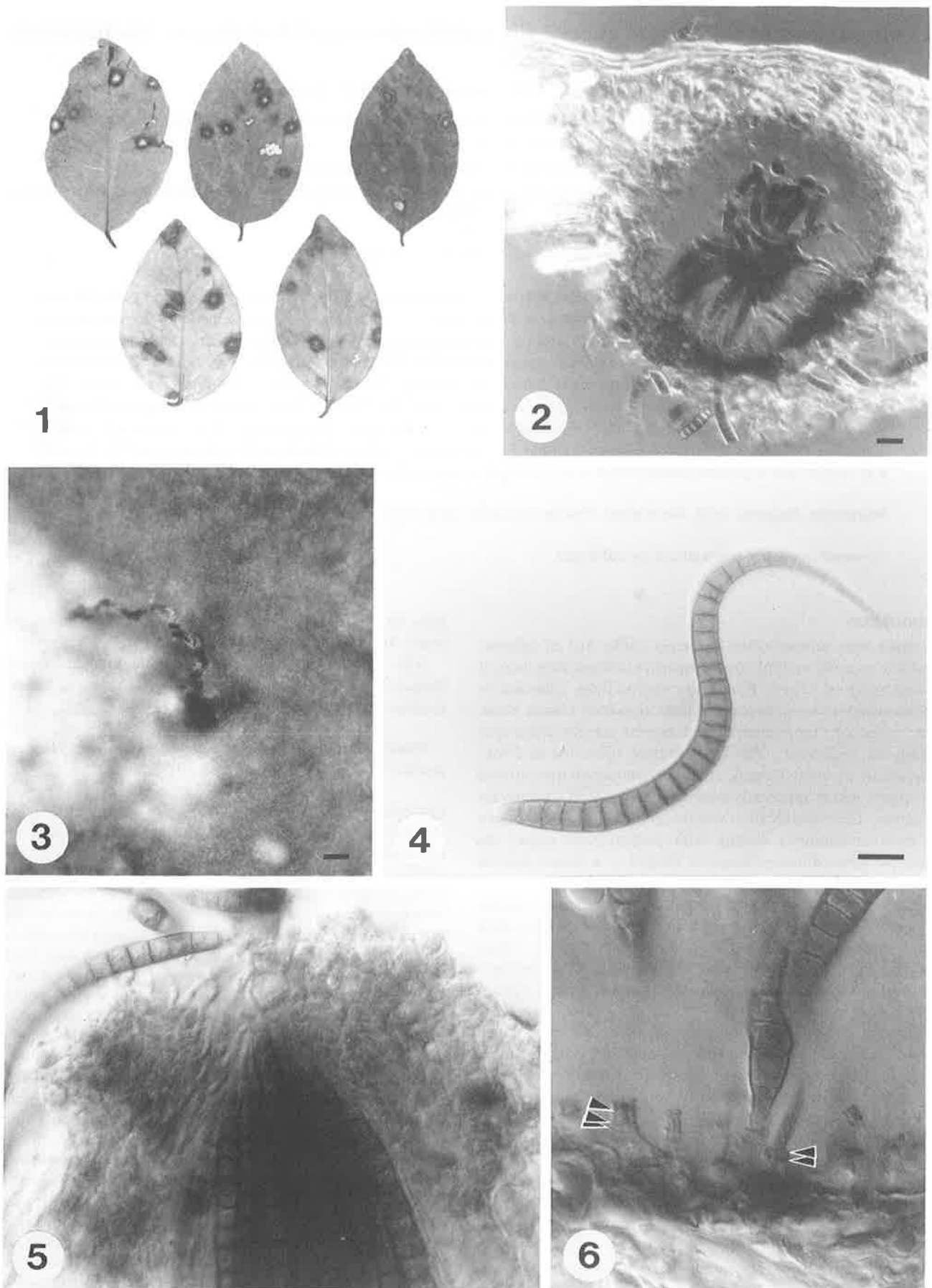
neis, 60–90 × 3–5 µm; basidiis filiformibus, simplicibus, brevissimis, hyalinis ad basim dispositis.'

After an examination of several specimens and cultures of the fungus, the following revised description is provided based on a neotype designated below.

Phaeophleospora eugeniae Rangel, Arch. Mus. Nac. Rio de Janeiro 18: 162 (1916). Figures 1–10.

Leaf spots amphigenous, circular, 1–3 mm diam., centres grey, surrounded by a medium to dark brown outer region; borders slightly raised, dark red-brown, margins surrounding the borders are diffuse, reddish-purple; on the lower leaf surface lesions appear as described on the upper surface, or are uniformly medium brown with a diffuse brown margin. Mycelium immersed, consisting of brown, septate, branched, finely verruculose hyphae. Conidiomata pycnidial primarily hypogenous, exuding conidial masses in long dark brown to black cirrhi, sub-epidermal, appearing slightly erumpent due to a furfuraceous margin of light brown cells that form around the ostiolar region, often absent at maturity, leaving a broad ostiole; globose to subglobose, unilocular, 150–180 µm diam; ostiolar region composed of several layers of light brown to hyaline cells, wall up to 40 µm thick at maturity; basal and lateral walls composed of 3–5 layers of medium brown cells of *textura epidermoidea*, wall 8–15 µm thick at maturity. Conidiophores absent. Conidiogenous cells discrete, light brown, cylindrical to ampulliform, verruculose, 5–15 × 3.5–6 µm, with 1–8 smooth or slightly irregular percurrent proliferations; formed from the inner layer of the conidiomatal wall. Conidia holoblastic, solitary, exuding in dark brown to black cirrhi, finely verruculose, vermiform, long obclavate, tapering to sub obtuse apices, bases truncate, with minute marginal frills, widest in the middle region, euseptate, with (16–)24–27(–30) septas, generally not constricted at septa when mounted in lactophenol but constricted when mounted in water, and more so in older material, medium brown, basal cell light brown, apical 2–4 cells pale brown, (70–)110–120(–130) × (6–)7–8 µm in lactophenol, 8–10 µm wide in water.

Cultural characteristics. Colonies reaching 11–14 mm in diam. on 2% malt extract agar (MEA) (Oxoid) after 1 month in the dark



Figures 1–6 *Phaeophleospora eugeniae*. 1. Leaf spots on the upper (top row) and lower (bottom row) leaf surfaces of *Eugenia uniflora*. 2. Vertical section through a subepidermal pycnidium. 3. Conidia exuding from a pycnidium in a brown cirrus. 4. Sigmoid conidium. 5. Erumpent, ostiolar region of a sporulating pycnidium. 6. Brown, verruculose conidiogenous cells with percurrent proliferations (arrows). (Scale bars; Figures 2, 3 & 4–6 = 20, 25 & 10 μm).

at 25°C, margins are smooth, lobed to even, and the surface is white, erumpent, irregular; aerial mycelium is moderate, interspersed with black conidial masses; reverse is honey 21"b coloured (Rayner 1970). Cardinal temperature requirements for growth in 5° intervals are above 10°C (min.), 20–25°C (opt.), and below 30°C (max.).

Neotype: Brazil Minas Gerais, Viçosa University campus, living leaves of *Eugenia uniflora*, F.A. Ferreira, 8 Jul. 1996, IMI 372655, a portion of the neotype is also lodged as PREM 55275, cultures ex-type STE-U 1453, 1454.

Additional specimens examined: Brazil, Minas Gerais, Viçosa University campus, living leaves of *E. uniflora*, F.A. Ferreira, 15 Jun. 1990, PREM 55276. Brazil, Minas Gerais, Viçosa University campus, living leaves of *E. uniflora*, F.A. Ferreira, 20 Jun. 1989, PREM 55277.

Generic considerations

The genus *Phaeophleospora* was introduced as the dark form of '*Phleospora*' (*Phloeospora* Wallr.) by Rangel (1916). In contrast to *Phloeospora*, conidiomata were described as pycnidial, and no data were given regarding its mode of conidiogenesis. Several genera can be compared with *Phaeophleospora*, namely *Phaeoseptoria* Speg., *Scoleciasis* Roum. & Fautrey, *Hendersonia* Berk., *Stagonospora* (Sacc.) Sacc., *Sonderhenia* H.J. Swart & J. Walker and *Kirramyces* J. Walker, B. Sutton & Pascoe. According to Walker *et al.* (1992), uncertainty remains regarding *Scoleciasis*, which was described as having hyaline to pale brown conidia with three to several transverse septa, 120–160 × 4 µm, borne on long, thin conidiogenous cells, 8–24 × 2 µm. Sutton (unpublished) examined exsiccata of the type species, *S. aquatica* Fautrey & Roumeguère, but only found ascomata and ascospores of a *Leptosphaeria* Ces. & De Not. sp., so the placement in the coelomycetes is questionable. *Hendersonia* is a *nomen rejiciendum* in favour of *Stagonospora*, which has hyaline spores and conidiogenous cells. *Sonderhenia* has verruculose, brown conidia produced on brown, percurrently proliferating conidiogenous cells, but can be separated from *Phaeophleospora* by its distoseptate conidia. The genus *Phaeoseptoria* is characterised by having filiform, smooth-walled, euseptate, brown conidia borne on hyaline, smooth-walled and apparently holoblastic conidiogenous cells (Morgan-Jones 1974; Walker *et al.* 1992), and is therefore distinct from *Phaeophleospora*.

Walker *et al.* (1992) introduced the generic name *Kirramyces* for foliicolous pathogenic coelomycetes formerly placed in *Phaeoseptoria*, which has unilocular, pycnidial conidiomata, discrete, ampulliform, lageniform or short cylindrical, brown, verruculose conidiogenous cells with several proliferations, and euseptate, cylindrical to obclavate, brown, verruculose conidia with obtuse apices and truncate bases with marginal frills. A feature typical of *Kirramyces* spp. is that their conidiomata are usually wide open at maturity (Walker *et al.* 1992; Palm 1996). Palm (1996) observed hyaline cells at the apices of conidiomata of *K. phormii* (Naito) M.E. Palm to be pushed outwards due to the enlarging conidial mass, which results in the overlying part of the leaf cuticle and epidermis being absent at the apices of mature conidiomata. The same was also noted for other species of *Kirramyces* (Walker *et al.* 1992), and is also characteristic of *P. eugeniae* (Figure 5). Walker *et al.* (1992) recognised two series of species in *Kirramyces*, namely those with brown, verruculose conidia and conidiogenous cells, and conidiomata of *textura angularis*, and those with paler, finely verruculose conidia and conidiogenous cells, and conidiomata with walls of *textura epidermoidea*. With the description of *K. proteae* B. Sutton (Sutton 1993), the first species with characteristics of both series was described, and Palm (1996) speculated that with the description

of additional species, this separation may even become less distinct.

Phaeophleospora eugeniae corresponds closely with species of *Kirramyces* in having sub-epidermal, dark-walled conidiomata of *textura epidermoidea* that are open and cup-shaped at maturity. Under conditions of high humidity, these conidiomata exude brown to black cirrhi of medium brown, euseptate, sub-cylindrical to obclavate, tapering, finely verruculose, multiseptate scoleospores. These conidia are also frequently visible on the leaf surface, though not as abundant as that observed for *K. epicoccoides* (Cooke & Masee) J. Walker *et al.* Conidia are formed on verruculose, brown, doliiform to cylindrical or ampulliform, percurrently proliferating conidiogenous cells that line the inner layer of the conidiomatal cavity. When mounted in water, conidia of *P. eugeniae* appear constricted at their septa, and when mounted in lactophenol cotton blue, the upper 4–5 cells and basal conidial cells stain more prominently than the rest of the conidium body. Conidia cultured *in vitro* on MEA, however, appear more uniform in colour than those formed *in vivo*. When conidia collected *in vivo* are mounted in H₂O, the apices appear subhyaline, and the basal cell light brown, in contrast to the rest of the conidium body which is medium brown. This difference in pigmentation, and the multiseptate conidia (up to 30-septate), are therefore the only features distinguishing *Phaeophleospora* from *Kirramyces*. Presently no descriptions for species of *Kirramyces* record more than 7 septa. Major differences occur in pigmentation, with conidia of *K. epicoccoides* being medium brown, becoming lighter brown near the apex, and conidia of *K. eucalypti* (Cooke & Masee) J. Walker *et al.* being finely verruculose and pale brown throughout.

Given all the similarities discussed above, we do not consider the number of conidial septa a morphological distinction sufficient to differentiate the two genera, and therefore propose that the older name, *Phaeophleospora* be designated for these species presently placed in *Kirramyces*, and the latter name reduced to synonymy.

New combinations in *Phaeophleospora* are therefore proposed as follows:

Phaeophleospora Rangel, Arch. Mus. Nac Rio de Janeiro 18: 162 (1916).

Kirramyces J. Walker, B. Sutton & I. Pascoe, Mycol. Res. 96: 919 (1992).

Phaeophleospora destructans (M. J. Wingf. & Crous) Crous, F.A. Ferreira & B. Sutton, comb. nov.

Basionym: *Kirramyces destructans* M.J. Wingf. & Crous, S. Afr. J. Bot. 62: 325 (1996).

Phaeophleospora epicoccoides (Cooke & Masee) Crous, F.A. Ferreira & B. Sutton, comb. nov.

Basionym: *Cercospora epicoccoides* Cooke & Masee apud Cooke, Grevillea 19: 91 (1891).

Additional synonyms listed in Walker *et al.* (1992).

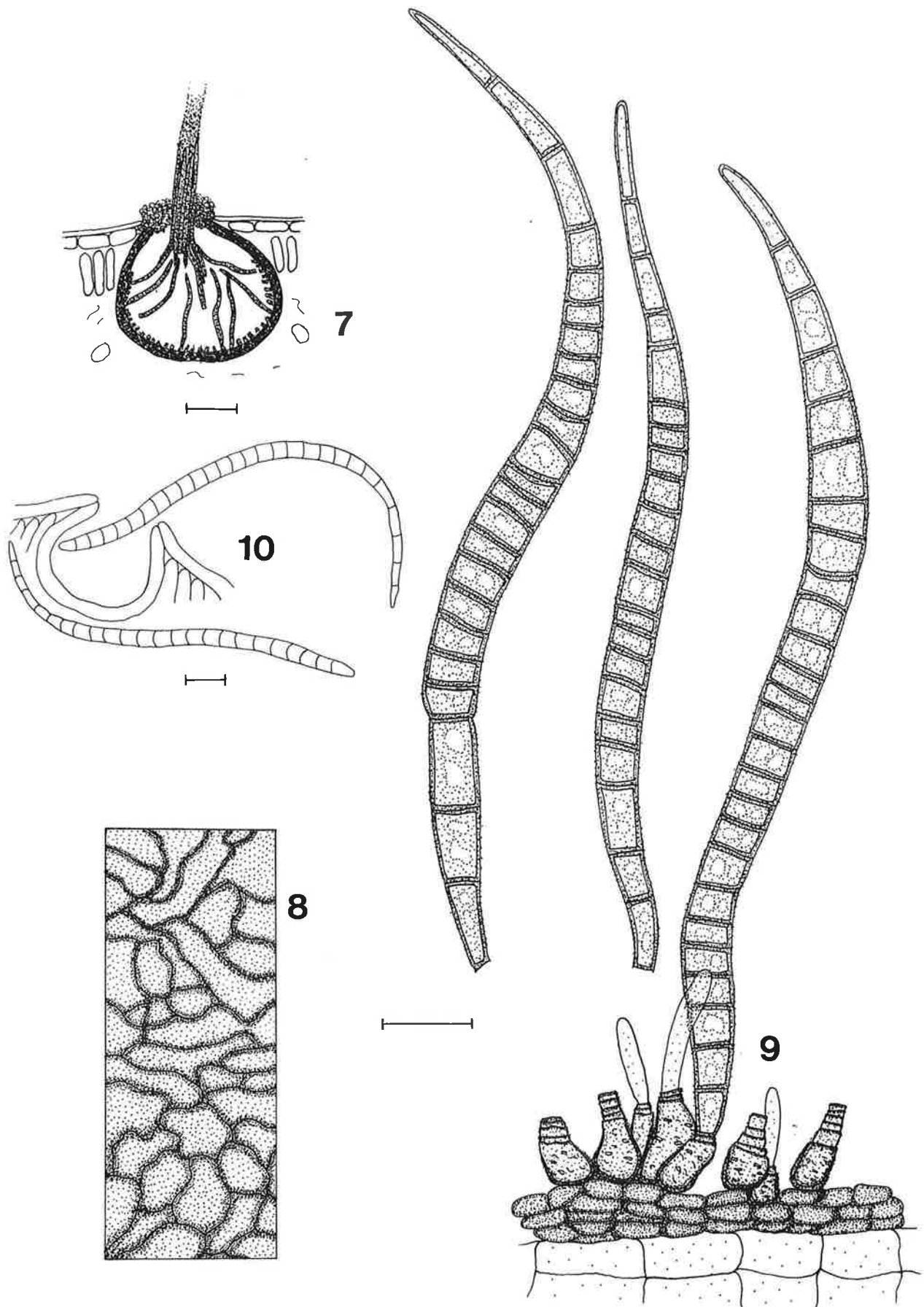
Phaeophleospora eucalypti (Cooke & Masee) Crous, F.A. Ferreira & B. Sutton, comb. nov.

Basionym: *Cercospora eucalypti* Cooke & Masee apud Cooke, Grevillea 18: 7 (1889).

Additional synonyms listed in Walker *et al.* (1992).

Phaeophleospora hebes (W. Wu, B. Sutton & A.C. Gange) Crous, F.A. Ferreira & B. Sutton, comb. nov.

Basionym: *Kirramyces hebes* W. Wu, B. Sutton & A.C. Gange, Mycol. Res. 100: 1208 (1996).



Figures 7–10 *Phaeophleospora eugeniae*. 7. Vertical section through a pycnidium. 8. Texture of outer pycnidial wall (surface view). 9. Conidia and conidiogenous cells. 10. Original illustration provided by Rangel (1916). (Scale bars = 10 μ m, except Figure 10 = 25 μ m).

Phaeophleospora lilianiae (J. Walker, B. Sutton & I. Pascoe) Crous, F.A. Ferreira & B. Sutton, comb. nov.

Basionym: *Kirramyces lilianiae* J. Walker, B. Sutton & I. Pascoe, *Mycol. Res.* 96: 919 (1992).

Phaeophleospora proteae (B. Sutton) Crous, F.A. Ferreira & B. Sutton, comb. nov.

Basionym: *Kirramyces proteae* B. Sutton, *Mycol. Pap.* 167: 35 (1993).

Phaeophleospora phormii (Naito) Crous, F.A. Ferreira & B. Sutton, comb. nov.

Basionym: *Hendersonia phormii* Naito, *Science Reports of the Kagoshima Univ.* 1: 77 (1952).

Kirramyces phormii (Naito) M.E. Palm, *Mycol. Res.* 100: 374 (1996).

Of the genera remaining to be re-appraised based on future collections, it is unlikely that *Scoleciasis* with its rather thin conidogenous cells ($8\text{--}24 \times 2 \mu\text{m}$) would represent an earlier name for these taxa. Although only eight species are presently known to occur in *Phaeophleospora*, several additional specimens with similar characteristics are presently being studied, and it appears that especially in the Myrtaceae, *Phaeophleospora* spp. will be of significant interest to pathologists in future.

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