





http://dx.doi.org/10.11646/phytotaxa.176.1.14

# Englerulaceae (Dothideomycetes)

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#### Abstract

The family *Englerulaceae* presently includes seven genera that produce brown to dark-brown colonies on living leaves. Ascomata are superficial and scattered on the colonies and lack ostioles. Asci are 2–8-spored, bitunicate, ovate to globose and ascospores are multi-seriate, oblong to ellipsoid, brown, and smooth-walled with a single septum. This paper brings all genera of the family together in one place with descriptions and illustrations and discusses their current taxonomic placement. The type species of *Englerula*, *Parenglerula*, *Rhizotexis*, *Rhytidenglerula*, *Schiffnerula* and a new species of *Thrauste* are redescribed and illustrated with light photomicrographs, in this paper. It is suggested that *Butleria* should be accommodated in *Myriangiaceae*. *Rhizotexis* is excluded from the family and moved to Dothideomycetes genera *incertae sedis*, as morphological characters are not similar to type genus and other genera in the family. The type specimen of *Goosia* is missing and is redrawn from the protologue. By illustrating and redescribing the genera we expect renewed interest for recollection and molecular study so that these dothideomycetous genera can be placed in a natural taxonomic framework.

Key words: Dothideomycetes, Pleosporales, taxonomy

# Introduction

We are in the process of studying genera of Dothideomycetes in order to provide a natural classification of this large class (Boonmee *et al.* 2011, Liu *et al.* 2011, 2012, Wu *et al.* 2011a,b, Zhang *et al.* 2008, 2009a,b, 2011a,b, 2012, Hyde *et al.* 2013). These studies involved examination of type specimens of the genera, provision of modern descriptions and illustrations or photomicrographs and, wherever possible, linking to their asexual states. In this study, we re-examined the type species of genera placed in *Englerulaceae*.

The family *Englerulaceae* was introduced by Hennings (1904) to accommodate *Englerula* and presently includes *Englerula*, *Goosia*, *Parenglerula*, *Rhizotexis*, *Rhytidenglerula*, *Schiffnerula* and *Thrauste* (Lumbsch & Huhndorf 2010). There is, however, no molecular data available for any of these genera. We redescribe the type species, provide descriptions and illustrations and discussed their possible taxonomic placements in this paper. It is hoped that with redescriptions of all genera in the family, fresh collections will be made so that molecular data can be used to resolve systematic relationships of the genera.

#### Materials and methods

#### Examination of specimens

Type specimens or slides were obtained from B, BPI, FH, NY and S (abbreviations according to Index Herbariorum 2013). The study methods followed are those reported in Chomnunti *et al.* (2011) and Liu *et al.* (2012). Fruiting bodies were rehydrated in water and/or 5% KOH prior to examination and sectioning. Hand sections of the ascomata were mounted in water for microscopic studies and photomicrography. The materials were examined by a Nikon ECLIPSE 80i compound microscope and photographed by Canon 450D digital camera fitted to the microscope. Measurements were made with the Tarosoft (R) Image Frame Work program and images used for figures were processed with Adobe Photoshop CS3, Extended version 10.0 software (Adobe Systems, USA). Hand drawings were made using drawing pens on parchment paper for spores of *Schiffnerula* and illustrations of *Goosia*. Genus and species descriptions are given for all accepted genera; however, if the genus is monotypic then only a species description is provided.

# Results

There is no sequence data for any members of this family in GenBank and all data given below are based on morphology.

# Taxonomy

*Englerulaceae* Henn., Hedwigia Beibl. 43: 353 (1904) MycoBank: MB 80736 Synonym: *Schiffnerulaceae* Hosag., Pl. Pathol. Quarant. 1(2): 132 (2011)

Saprobic or epiphytic on living leaves, primarily tropical. Colonies superficial, thin to dense, brown to dark brown, confluent, velvety, with unicellular appressoria. Hyphae straight to flexuous, septate, irregularly to reticulately branched, brown. Hyphopodia globose, brown, thick-walled, irregular or absent. Sexual state: Ascomata, superficial on hyphae, scattered, ellipsoid to globose, cupulate, brown to black, membranous, non-ostiolate, containing a few asci. Hamathecium of brown, septate, pseudoparaphyses or pseudoparaphyses, sometimes lacking. Asci 8-spored or rarely 2-spored, bitunicate, thick-walled, ovate, globose to subglobose, pedicellate, with or without a well-developed ocular chamber. Ascospores uniseriate to multiseriate, oblong to ellipsoid, or fusiform, hyaline to dark brown, 1-septate, smooth-walled, constricted at the septum. Asexual state: Conidiomata pycnidial, similar to ascomata in shape, superficial, globose, thin-walled. Conidiogenous cells holoblastic, phialidic, integrated or discrete, hyaline, smooth, 1-celled, nearly ampulliform, alternate or absent. Conidia 1-celled, oblong to ovate, globose or nearly globose, hyaline to brown.

**Notes:**—The family *Englerulaceae* was introduced by Hennings (1904) for taxa with brown to dark-brown colonies on host leaves, with superficial, scattered, ascomata lacking ostioles. Lumbsch & Huhndorf (2010) included seven genera in the family, while Hyde *et al.* (2013) considered *Schiffnerulaceae* (Hosagoudar 2011) to be a synonym of *Englerulaceae*. *Schiffnerula*, introduced by Höhnel (1909), was placed in the family *Englerulaceae* because of its globose ascomata and globose to ovate asci. Hosagoudar (2011) introduced *Schiffnerulaceae*, which is typified by *Schiffnerula* and characterized by thyriothecioid ascomata developing below the mycelium, and asci exposed in the central portion of the thyriothecium. Hosagoudar (2011) included four asexual genera in the family with more than one hundred species. *Schiffnerula* has dark colonies on the leaf surface, with brown, superficial, septate mycelium, 8-spored, bitunicate, cylindrical to globose asci, and brown, 1-septate, ascospores which are constricted at the septa. With such characters *Schiffnerula* can be accommodated in *Englerulaceae*.

In *Englerulaceae*, the genus *Englerula* has hyphae without hyphopodia or appressoria, while *Goosia*, *Parenglerula*, *Rhytidenglerula*, *Schiffnerula* and *Thrauste* have hyphopodia (Bin 2003, Castlebury *et al.* 1995, Hosagoudar *et al.* 2011a,b, Höhnel 1909, 1910, 1918, Thesisen 1916). Whether this character is taxonomically important needs to be tested using molecular data, but for now, we accept these genera in the family. In addition, *Parenglerula* has pseudoparaphyses which is different from other genera in the family. *Goosia* has 1-celled, nearly ampulliform and alternate phialides and 2-spored asci, which distinguish it from the other genera (Bin 2003). The

type of *Goosia* is lost, and this genus needs recollecting and neotyping. In *Rhytidenglerula, Schiffnerula* and *Thrauste*, hyphopodia are brown, globose to subglobose and thick-walled. *Thrauste* has large perithecia. *Rhytidenglerula* and *Schiffnerula* have cylindrical and brown ascospores. *Rhytidenglerula* has superficial ascomata with shield-like, radiating hyphal tissue, while *Schiffnerula* differs from other genera by ascomata arising from short lateral branches. In this paper, we suggest to include *Butleria* in *Myriangiaceae*. *Rhizotexis* is excluded from this family and placed in Dothideomycetes genera *incertae sedis*.

Asexual states of this family are coelomyceteous and hyphomyceteous viz. *Capnodiastrum*, *Digitosarcinella*, *Mitteriella*, *Questieriella* and *Sarcinella* (Wijayawardene *et al.* 2012). *Parenglerula* and *Thrauste* lacks known asexual stages, while *Schiffnerula* has four hyphomycetous asexual states, namely *Digitosarcinella*, *Mitteriella*, *Questieriella* and *Sarcinella* (Hosagoudar 2003). *Englerula* and *Rhytidenglerula* have asexual states with pycnidia belonging to *Capnodiastrum* (Wijayawardene *et al.* 2012). Hosagoudar (2003) proposed a new pycnidial genus *Krishnamyces* for an asexual stage of *Rhytidenglerula*. Bin (2003) described *Goosia* with 1-celled, brown conidia. *Butleria* and *Rhizotexis* have no known asexual state (Wijayawardene *et al.* 2012, Saccardo 1913, Theissen & Sydow 1917).

#### Type genus:—Englerula Henn., Bot. Jb. 34: 49 (1904) MycoBank: MB 1820

Saprobic or epiphytic on both sides of living leaves, primarily tropical. *Colonies* on both sides of the leaves, superficial, thin to dense, rounded to irregular, dark brown to black, confluent, velvety. *Hyphae* superficial, straight to curved, septate, irregular to reticulately branched, brown, cells with acute to wide angles. *Hyphopodia* absent. Sexual state: *Ascomata* perithecioid, superficial on hyphae, scattered, globose to subglobose, brown to black, non-ostiolate, containing few asci. *Peridium* thin, membranous. *Pseudoparaphyses* absent. *Asci* 8-spored, bitunicate, thick-walled, obovoid to subclavate, globose to subglobose, with a thick ocular chamber. *Ascospores* multi-seriate, fusiform, broadly ellipsoid to subobovoid, yellowish brown to dark brown, 1-septate, smooth-walled, constricted at the septum, with rounded ends, surrounded by remnants of mucilage. Asexual state: in *Capnodiastrum*. *Conidiomata* pycnidial, perithecioid, superficial, globose, thin-walled. *Conidiogenous cells* holoblastic, discrete, hyaline, smooth-walled. *Conidia* 1-celled, oblong to ovate, brown.

**Notes:**—*Englerula* includes 13 species epithets according to Index Fungorum (2013). This genus is characterized by brown to black colonies with angled hyphae, globose to subglobose ascomata with thin peridium, and obovoid to globose asci (Hosagoudar *et al.* 2011a, von Arx & Müller 1975, Eriksson 1981). The species of this genus need to be recollected.

# Type species:—*Englerula macarangae* Henn., Bot. Jb. 34: 49 (1904) MycoBank: MB 140464 (Fig. 1)

Saprobic or epiphytic on lower surface of living leaves, primarily tropical. *Colonies* superficial, thin to dense, rounded to irregular, dark brown to black, confluent, velvety. *Hyphae* superficial, straight to curved, septate, irregular to reticulately branched, with acute to wide angles, brown. *Hyphopodia* absent. Sexual state: *Ascomata* (49–)59.5–83(–87) µm diam., perithecioid, superficial on hyphae, scattered, globose to subglobose, brown to black, non-ostiolate, containing a few asci. *Peridium* thin, membranous. *Hamathecium* with few asci and lacking pseudoparaphyses. *Asci* (58–)60.5–76 × 40–58(–60) µm ( $\overline{x} = 66 \times 50$  µm, n = 20), 8-spored, bitunicate, fissitunicate, thick-walled, obovoid to subclavate, globose to subglobose, with minute pedicel or apedicellate when mature, with a thick ocular chamber. *Ascospores* (26–)30–35(–37) × 12.5–17(–18) µm ( $\overline{x} = 32 \times 15$  µm, n = 20), 3–4-seriate, fusiform, broadly ellipsoid to subobovoid, yellowish-brown to dark brown, 1-septate, smooth-walled, constricted at the septum, with rounded ends, surrounded by remnants of mucilage. Asexual state: *Conidiomata* pycnidial, superficial, solitary, globose, dark brown. *Conidiophores* absent. *Conidiogenous cells* holoblastic, doliiform, hyaline, smooth. *Conidia* 10–20 × 3–4 µm, fusiform, hyaline, 1–3-septate, smooth-walled.

**Material examined:**—TANZANIA. Tanga: Lushoto, Usambara Mountains, on living leaves of *Macaranga kilimandscharica* Pax., September 1902, *Engler* (Rehm Ascomyceten 1539), (BPI! 566046, syntype).



**FIGURE 1.** *Englerula macarangae* (syntype). A. Appearance of colonies on lower leaf surface (left), and close up of ascomata on mycelium (right). B. Squash mount of ascoma. C–G. Asci with ascospores. F. Dehisced ascus. H. Immature ascospores. I–J. Brown mature ascospores surrounded by mucilaginous material. Scale bars:  $A = 100 \mu m$ ,  $B-G = 50 \mu m$ ,  $H-J = 20 \mu m$ .

# Other genera included

*Goosia* B. Song, Mycotaxon 87: 413 (2003), MycoBank: MB 28768 **Type species:**—*Goosia melastomatis* B. Song, Mycotaxon 87: 413 (2003) MycoBank: MB 489048 (Fig. 2)

*Parasitic* on both sides of leaves. *Colonies* mostly on lower side, 4 mm diam., superficial, thin, rounded to irregular, black, confluent. *Hyphae* 5–6  $\mu$ m wide, superficial, straight to curved, septate, thick, irregular to reticulately branched, brown. *Hyphopodia* 10–15  $\mu$ m diam., alternate to unilateral, unicellular, globose, dark brown, formed laterally, with thick walls. Sexual state: *Ascomata* solitary to scattered, superficial on the dark hyphae, with immature ascomata 20–35  $\mu$ m long × 12–19  $\mu$ m wide, ellipsoid, dark brown to black, with a short pedicel, mature ascomata not observed in this specimen, containing few asci. *Peridium* dark brown, composed of one layer of large cells of *textura angularis*. *Hamathecium* of asci only. *Asci* 25–29 × 14–16  $\mu$ m, 2-spored, bitunicate, obovoid, ocular chamber not well developed. *Ascospores* 18–24 × 8–11  $\mu$ m, 1-seriate, oblong to ellipsoid, 2-celled, brown, with rounded ends and smooth-walled. Asexual state: *Phialides* sparse, brown, mixed with hyphopodia, alternate, triangular. *Conidia* 7–12 × 6–10  $\mu$ m 1-celled, globose, brown, smooth-walled.



**FIGURE 2.** *Goosia melastomatis.* A. Hyphae with immature ascomata and hyphopodia. B. Phialides and hyphopodia. C. Asci containing ascospores. D. Ascospores with one septum. E. Globose conidia. Scale bars: A,  $B = 30 \mu m$ ,  $C = 15 \mu m$ , D,  $E = 10 \mu m$ . Redrawn from Bin (2003).

**Notes:**—*Goosia,* introduced by Bin (2003) with one species *G melastomatis,* is characterized by dark brown phialides developing directly on the hyphae, and obovoid asci containing two ascospores (Bin 2003). This genus is similar to *Thrauste* in having superficial, ellipsoid ascomata and globose hyphopodia formed on dark brown hyphae. Bin (2003) did not observe mature ascomata and considered that *Goosia* differed from *Thrauste* with the former having phialides and smaller ascospores. No molecular data is available in GenBank for this genus and thus fresh collections are needed in order to confirm if they differ and also show its natural taxonomic relationships.

Material examined:—CHINA. Hainan: Diaolou Shan, on leaves of *Melastoma normale* D. Don (*Melastomataceae*), 28 May 1988, *Y.X. Hu, HMIGD 34601* (holotype lost, redrawn from protologue, Bin 2003).

Parenglerula Höhn., Sber. Akad. Wiss. Wien, Math.-naturw. Kl., Abt. 1 119: 465 [73 repr.] (1910) MycoBank: MB 3727

Synonymy: Linotexis Syd. & P. Syd., Annls mycol. 15(3/4): 197 (1917)

*Epiphytic* on living leaves. *Hyphae* superficial, straight to curved, septate, *Hyphopodia* globose to subglobose, dark brown, with thick walls. Sexual state: *Ascomata* superficial, clustered, gregarious, uniloculate, globose to subglobose, black. *Peridium* thin, composed of dark brown cells of *textura angularis*, brittle. *Hamathecium* of wide, brown to reddish brown, straight or slightly curved, some branched, septate pseudoparaphyses. *Asci* 8-spored, bitunicate, globose to subglobose, obovoid to subclavate, thickened at the apex, short pedicellate, some sessile, ocular chamber not well-developed. *Ascospores* multi-seriate, broadly ellipsoid, subobovoid, some slightly

fusiform, 1-septate, with rounded ends, constricted at the septum, hyaline to yellowish, becoming brown to dark brown at maturity, smooth-walled. Asexual state: Unknown.

**Notes:**—*Parenglerula* was introduced by Höhnel (1910) with seven species and is typified by *Parenglerula macowaniana* ( $\equiv$ *Meliola macowaniana* Thüm.). *Parenglerula* is similar to *Englerula* in having superficial, globose to subglobose, dark ascomata containing obovoid asci and broadly ellipsoid to subobovoid ascospores with one septum, but differs by darker ascomata and ascospores. In addition, this genus has brown, slightly curved pseudoparaphyses which are normally shorter than asci. Such a character is not apparent in other genera of *Englerulaceae* (Bin 2003, Castlebury *et al.* 1995, Hennings 1904, Hosagoudar *et al.* 2011a,b, Höhnel 1909, 1910, 1918, Theisen 1916, 1917). There are nine epithets in Index Fungorum (2013) and the genus has not been recently studied and thus needs recollecting and restudying.

**Type species:**—*Parenglerula macowaniana* (Thüm.) Höhn., Sber. Akad. Wiss. Wien, Math.-naturw. Kl., Abt. 1 119: 465 [73 repr.] (1910) MycoBank: MB 239763 (Fig. 3)

Basionym: Meliola macowaniana Thüm., Mycoth. Univ., cent.: no. 568 (1876)

Synonymy: Asterina macowanianum Kalchbr. & Cooke, Grevillea 9(49): 33 (1880)

Dimerosporium macowanianum (Thüm.) Sacc., Syll. fung. (Abellini) 1: 53 (1882)

Englerulaster macowanianus (Thüm.) G. Arnaud, Les Astérinées: 183 (1918)

Meliola macowaniana Thüm., Mycoth. Univ., cent.: no. 568 (1876)

*Epiphytic* on lower surface of living leaves. *Hyphae* superficial, straight to curved, septate, branch, with hyphopodia. Sexual state: *Ascomata* (60–)65–88(–95) µm diam., superficial, clustered, gregarious, uniloculate, globose to subglobose, black. *Peridium* thin, composed of dark brown cells of *textura angularis*, brittle. *Hamathecium* of 32–49(–59) µm long × 5.5–8 µm wide, brown to reddish brown, straight or slightly curved, some branched, septate pseudoparaphyses. *Asci* (49–)55.5–72(–76) × (39–)44–56.5 ( $\overline{x}$  = 61.5 × 50 µm, n = 20), 8-spored, bitunicate, globose to subglobose, obovoid to subclavate, with thickened apex, short pedicellate, some sessile, ocular chamber not well developed. *Ascospores* (29.5–)30–34.5(–37) × 13.5–17(–18.5) µm ( $\overline{x}$  = 31 × 16 µm, n = 20), multi-seriate, broadly ellipsoid, subobovoid, some slightly fusiform, 1-septate, with rounded ends, constricted at the septum, hyaline to yellowish, becoming brown to dark brown at maturity, smooth-walled. Asexual state: Unknown.

**Material examined:**—SOUTH AFRICA. Gauteng: Pretoria, Boschberg Street, Somerset-East, on leaves of *Celastrus buxifolius* L. (synonym of *Gymnosporia buxifolia* (L.) Szyszył.), August 1876, *Mac Owan, No.1252* (BPI! 692058, syntype).

# *Rhytidenglerula* Höhn., Sber. Akad. Wiss. Wien, Math.-naturw. Kl., Abt. 1 127: 386 [58 repr.] (1918) MycoBank: MB 4738

Saprobic on lower surface of living leaves. Colonies superficial, thin, rounded to irregular, brown to dark brown, confluent, velvety, with unicellular hyphopodia. Hyphae long, flexuous reticulately branched, septate, dark brown. Hyphopodia alternate to unilateral, unicellular, ellipsoid, triangular, dark brown, formed laterally, with thick wall. Sexual state: Ascomata orbicular, yellow to brown, with radiating cells on the upper surface, dissolute at the centre at maturity, small, superficial on radiating, a shield–like plate, solitary, globose to subglobose, light yellow to brown, membranous, irregular on the surface, rounded at the top and containing few asci. Peridium thin, composed of a single layer of yellow cells. Hamathecium with few asci, lacking pseudoparaphyses. Asci 8-spored, bitunicate, obovoid, without pedicel, with rounded base, with an ocular chamber at the apex. Ascospores overlapping, oblong, hyaline, 1-septate, with rounded ends, smooth-walled. Asexual state: Conidiomata pycnidial, superficial, solitary, globose, dark brown. Conidiomata with thin wall. Conidiophores absent. Conidiogenous cells holoblastic, determinate, discrete, doliiform, hyaline. Conidia ellipsoid, pale brown, 1-septate, base truncate, apex obtuse to conic, obovoid, upper cell small, pale brown, lower cell much larger.



**FIGURE 3.** *Parenglerula macowaniana* (syntype). A. Black ascomata on host surface. B. Squash of ascoma. C. Brown to light brown pseudoparaphyses. D–G. Asci containing ascospores. H, I. Immature ascospores. J, K. Dark brown mature ascospores. Scale bars: A,  $B = 100 \mu m$ ,  $C-G = 50 \mu m$ ,  $H-K = 20 \mu m$ .

**Notes:**—*Rhytidenglerula* has small yellow ascomata forming on dark hyphae, a character that distinguishes it from other genera with large and dark ascomata in the family. This genus consists of 11 species epithets according to Index Fungorum (2013). *Rhytidenglerula* is similar to *Schiffnerula* in having hyphopodia and globose ascomata, obovoid asci and hyaline ascospores with a single septum. *Rhytidenglerula* is linked to a coelomycetous asexual morph i.e. *Capnodiastrum* Speg. (Kirk *et al.* 2008, Wijayawardene *et al.* 2012) which is characterized by holoblastic, doliiform, conidiogenous cells and ellipsoid, pale brown, 1-celled conidia with a conic apex (Sutton 1980). *Schiffnerula* has been linked to four different hyphomycetous asexual morphs i.e. *Digitosarcinella, Mitteriella, Questieriella* and *Sarcinella* (Hosagoudar 2011, Hosagoudar *et al.* 2011b, Seifert *et al.* 2011). These four genera are characterized by monoblastic to polyblastic conidiogenous cells and sigmoid to ellipsoid, dark brown to reddish brown, or black, 0–4-celled conidia (Hosagoudar 2011, Hosagoudar *et al.* 2011b). Currently no sequence data are available in GenBank and hence natural affinity of these genera is yet to be confirmed.

**Type species:**—*Rhytidenglerula carnea* (Ellis & G. Martin) Höhn., Sber. Akad. Wiss. Wien, Math.-naturw. Kl., Abt. 1 127(4): 386 [58 repr.] (1918) MycoBank: MB 432015 (Fig. 4) Basionym: *Asterina carnea* Ellis & G. Martin, Am. Nat. 17(2): 1285 (1883)



**FIGURE 4.** *Rhytidenglerula carnea* (holotype) A. Colonies on lower surface of leaf. B. Ascomata on mycelium. C. Ascoma containing asci. D. Squash of ascoma. E. Young ascoma on hyphae. F. Hyphopodia and hyphae. G, H. Mature asci with ascospores and showing apical chamber. I, J. Immature asci. K, L. Ascospores with one septum. M. Fractured ascus with ascospores. Scale bars: A = 5 mm,  $B = 100 \text{ \mum}$ .  $C = 50 \text{ \mum}$ ,  $D = 10 \text{ \mum}$ ,  $E-M = 5 \text{ \mum}$ .

Saprobic on the lower surface of living leaves. Colonies 2–6 mm wide  $\times$  3–12 mm long, superficial, thin, round to irregular, brown to dark brown, confluent, velvety, with unicellular hyphopodia. Hyphae long,  $(2.5-)3-5(-7) \mu m$ wide, flexuous reticulately branched, septate, dark brown. *Hyphopodia*  $5-7 \times 6-7.5 \mu m$  alternate to unilateral, unicellular, elliptical, triangular, dark brown, formed laterally, with thick wall. Sexual state: Immature ascomata 15–25.7 µm diam., orbicular, yellow, with radiating cells on the upper surface, dissolute at the centre at maturity. *Mature ascomata* (45–)56.5–95(–105) µm diam., small, superficial on hyphae, solitary, globose to subglobose, light yellow to brown, membranous, irregular on the surface, rounded containing few asci. Peridium thin, composed of one layer of yellow tissue. *Hamathecium* with few asci, lacking pseudoparaphyses. Asci 30–38(-40.5)  $\times$  (20–)25–33.5(–35.5) µm ( $\overline{x}$  = 34.5  $\times$  28.9 µm, n = 10), 8-spored, bitunicate, obvoid, without pedicel, with rounded base, with a thick ocular chamber. Ascospores (17–)18.5–21(–22) × (5.5–)7–9.5(–10)  $\mu$ m ( $\overline{x}$ = 19.8 × 8.3  $\mu$ m, n = 10), multi-seriate, oblong, hyaline, 1-septate, with rounded ends, wall smooth. Asexual state: *Conidiomata* pycnidial, superficial, solitary, globose, dark brown, unilocular, without ostiole. Conidiomata wall thin composed of one layer of cells textura angularis. Conidiophores absent. Conidiogenous cells holoblastic, determinate, discrete, doliform, hyaline, smooth. Conidia pale brown, 1-septate, base truncate, slightly protuberant, apex obtuse to conic, obovoid, thick-walled, with a central guttule and a median transverse hyaline band; upper cell small, pale brown, lower cell much larger.

**Material examined:**—USA. New Florida, on leaves of *Persea palustris*, *B. Ellis & G. Martin* (NY! 01089642, holotype).

Schiffnerula Höhn., Sber. Akad. Wiss. Wien, Math.-naturw. Kl., Abt. 1 118: 867 [55 repr.] (1909) MycoBank: MB 4886

Synonymy: Clypeolella Höhn., Sber. Akad. Wiss. Wien, Math.-naturw. Kl., Abt. 1 119: 403 [11 repr.] (1910)

Coniosporiella Bat., in Batista, Maia & Bezerra, Atas Inst. Micol. Univ. Pernambuco 3: 113 (1966)

*Diathrypton* Syd., Philipp. J. Sci., C, Bot. 21(2): 137 (1922)

Mitteriella Syd., in Sydow & Mitter, Annls mycol. 31(1/2): 95 (1933)

Phaeoschiffnerula Theiss., Brotéria, sér. bot. 12: 21 (1914)

Questieria G. Arnaud, Thèses Fac. Sci. Paris, Sér. A 805: 186 (1918)

Questieriella G. Arnaud ex S. Hughes, Can. J. Bot. 61(6): 1729 (1983)

Sarcinella Sacc., Michelia 2(6): 31 (1880)

*Colonies* formed on surface of living leaves, thin to dense, circular to irregular, black, confluent. *Hyphae* superficial, straight to curved, septate, irregular to reticulately branched, brown, appressoriate, with smooth outer surface, tuberculate to crenulate. *Hyphopodia* alternate to unilateral, unicellular, elliptical, globose to subglobose, dark brown, formed laterally, wall thick. Sexual state: *Immature ascomata* orbicular, pale to dark brown, with radiating cells on the upper surface, dissolute at the centre at maturity. *Mature ascomata* superficial on hyphae, scattered, globose, dark brown, membranous, with a few exposed asci. *Peridium* thin. *Hamathecium* with few asci, lacking *pseudoparaphyses*. *Asci* 8-spored, bitunicate, broadly clavate, globose to ovate, exposed. *Ascospores* multiseriate, broadly fusiform, ellipsoid to oblong, hyaline, 1-septate, smooth-walled, constricted at the septum, ends rounded to point. Asexual state: *Conidiophores* macronematous to micronematous, mononematous, lateral, 0–2-septate. *Conidiogenous cells* monoblastic to polyblastic, lateral or integrated, terminal, lateral, intercalary, determinate. *Conidia* solitary, acrogenous or acropleurogenous, 4-celled, narrowly ellipsoidal to obovoid, subglobose, sarciniform, dark brown to reddish brown, curved, falcate, sigmoid, truncate at the base, constricted at the septum (Hosagoudar 2011, Hosagoudar *et al.* 2011b ).

**Notes:**—*Schiffnerula* includes more than 100 species epithets (Index Fungorum 2013). Asexual states of this genus are reported in *Mitteriella*, *Questieriella* and *Sarcinella* (Hyde *et al.* 2013, Hosagoudar 2011, Seifert *et al.* 2011). Hughes (1983) mentioned that practically all *Sarcinella* species have a *Questieriella* synanamorph, and the sexual morphs of species with synanamorphic combination are in *Schiffnerula*. Hughes (1984) introduced a new asexual genus *Digitosarcinella* (with single species *D. caseariae*) synanamorphic to *Questieriella*. Some species of *Schiffnerula* produced *Questieriella* or *Mitteriella* alone, or no asexual morphs (Hughes 1983). Hosagoudar (2011) considered *Schiffnerula* as a new family, because *Schiffnerula* has young ascomata which look like a shield-plate on hyphae. According to Höhnel (1909, 1910), both *Rhytidenglerula* and *Schiffnerula* have thin, shield-plate-like immature ascomata and should belong to the family *Englerulaceae*. In addition, *Schiffnerula* has globose, ascomata

when matured. The ascomata develop on brown hyphae and contain a few globose to ovate asci with multi-seriate, ellipsoid, 1-septate, ascospores. Therefore, at present we retain *Schiffnerula* in *Englerulaceae*.

**Type species:**—*Schiffnerula mirabilis* Höhn., Sber. Akad. Wiss. Wien, Math.-naturw. Kl., Abt. 1 118: 868 [56 repr.] (1909) MycoBank: MB 211121 (Fig. 5)



**FIGURE 5.** *Schiffnerula mirabilis* (holotype). A. Superficial ascomata on brown hyphae with brown conidia. B. Globose to ovate asci surrounding by thin peridium. C. Alternate to unilateral, subglobose, dark brown hyphopodia. D, E. Ascospores (Drawn based on slides). Scale bars: A,  $B = 10 \mu m$ ,  $C-E = 5 \mu m$ .

*Colonies* forming superficially on the upper of living leaves, thin to dense, round to irregular, black, confluent. *Hyphae* long, (4.5-)5-6.5(-7.5) µm wide, superficial, straight to curved, septate, irregular to reticulately branched, brown, appressoriate, outer surface smooth, tuberculate to crenulate. *Hyphopodia*  $8.5-12 \times 7-9$  µm, alternate to unilateral, unicellular, elliptical, globose to subglobose, dark brown, formed laterally, with thick wall. Sexual state: *Immature ascomata* 17.5–23.5 µm diam., orbicular, pale, with radiating cells on the upper surface, dissolute at the centre at maturity. *Mature ascomata* (37–)31–50.5(–62) µm diam., superficial on hyphae, scattered, globose, dark

brown, membranous, with few exposed asci. *Peridium* thin, composed of one layer of light brown tissue. *Hamathecium* with few asci, lacking pseudoparaphyses. *Asci* (26.5–)29.5–39.5(–40) × 21–28  $\mu$ m ( $\overline{x}$ = 34 × 24.5  $\mu$ m, n = 10), 8-spored, bitunicate, globose to ovate, exposed, without pedicel, with rounded base, with an ocular chamber. *Ascospores* (9–)14–22.5 × (6–)7.5–11.5  $\mu$ m ( $\overline{x}$ = 18.5 × 9.5  $\mu$ m, n = 10), multi-seriate, broadly fusiform, ellipsoid to oblong, hyaline, 1-eptate, smooth-walled, constricted at the septum, with rounded ends. Asexual state: *Conidia* few, fusiform, curved, 3-septate, brown.

**Material examined:**—INDONESIA. West Java: in the forest of Depok at Bogor, on the leaves of *Passiflora* sp., 1894, *V. Schiffner* (FH! Slides: 3372, 3375, holotype).

#### Thrauste Theiss., Verh. zool.-bot. Ges. Wien 66: 337 (1916) MycoBank: MB 5455

Saprobic on lower surface of living leaves. Colonies black, thin, round to irregular, confluent. Hyphae superficial, straight, septate, brown, branched. Hyphopodia alternate to unilateral, unicellular, elliptical, globose to subglobose, dark brown, formed laterally, wall thick. Sexual state: Ascomata solitary to scattered, superficial on the black hyphae and easily detachable, globose to subglobose, black, membranous, containing few asci. Peridium dark brown, composed of one layer of large cells of textura angularis. Hamathecium with few asci, lacking pseudoparaphyses. Asci 8-spored, bitunicate, globose to subglobose, pyriform to clavate, thick-walled, with an ocular chamber. Ascospores 2–3-seriate to multi-seriate, ellipsoid to slightly fusiform, two-celled, median to supra median septate, hyaline to pale brown. Asexual state: Unknown.

**Notes:**—The genus *Thrauste*, typified by *Thrauste medinillae* ( $\equiv$  *Balladyna medinillae* Racib.), was introduced by Theissen (1916) with two species and the other species being *T. affinis* Sydow (1917). Original description mentions size and shape of only immature ascomata (Saccardo 1913). *Thrauste* is similar to *Goosia* in having dark hyphopodia and ellipsoid immature ascomata growing directly on hyphae (Bin 2003, Theissen 1916). However, *Thrauste* differs by its 8-spored, pyriform to clavate asci with an ocular chamber, while *Goosia* has 2-spored, obovoid asci and the ocular chamber is not well developed.

We examined a collection (No.: F: 111745), labelled *T. medinillae*, obtained from S. There are marked differences in the shape and size of asci and ascospores between the exciccatae we studied and the original description of the species (Saccardo 1913, p. 41) under the genus. The asci in *T. medinillae* and the second species *T. affinis* are globose to subglobose, wide, (one per ascoma in *T. medinillae*) and ascospores median 1-septate,  $25-30 \times 13-15 \mu m$  and  $20-25 \times 10-13 \mu m$ , respectively. In *T. parva* asci are clavate, narrow and ascospores 1 supra median septate,  $10-15 \times$  $3-5 \mu m$  (Table 1). In the light of these differences, we propose a new species for this specimen.

Species	Ascomata	Asci	Ascospores
Thrauste medinillae	Immature ascomata: 40–48 diam., subglobose, with pedicel $24-40 \times 7-9$ $\mu$ m; Mature ascomata: not mentioned	Sometime single in ascomata (no size mentioned)	25–30 × 13–15 $\mu$ m, median septate
T. affinis	Immature ascomata: with pedicel 15–25 × 7–9 μm; Mature ascomata: 30–50 μm diam., subglobose	28–35 diam., globose to subglobose,	$20-25 \times 10-13 \ \mu\text{m}$ , median septate, multi-seriate
T. parvii	Immature ascomata: $33-50 \times 10-16 \mu m$ , ellipsoid, with pedicel $10-24 \times 5-8\mu m$ ; Mature ascomata: $50-80 \mu m$ diam., subglobose	$30-50 \times 9-12 \ \mu\text{m}$ , few in ascomata, clavate	10–15 $\times$ 3–5 $\mu$ m, supra median septate, 2–3-seriate

**TABLE 1.** Morphological features of *Thrauste* spp.

**Type species:**—*Thrauste medinillae* (Racib.) Theiss., Verh. zool.-bot. Ges. Wien 66: 338 (1916) MycoBank: MB 145100

Basionym: Balladyna medinillae Racib., Bull. int. Acad. Sci. Lett. Cracovie, Cl. sci. math. nat. Sér. B, sci. nat. 3: 373 (1909)

Thrauste parvii D.Q. Dai, Q. Tian, D.J. Bhat & K.D. Hyde, sp. nov. MycoBank: MB 805414 (Fig. 6)

**Etymology:**—With reference to the smaller conidia.



**FIGURE 6.** *Thrauste medinillae* (holotype) A, B. Herbarium material. C, D. Ascomata on hyphae which form on the lower surface of leaves (arrow). E. Dark brown hyphae with hyphopodia. F. Squash mount of brown ascoma. G. Squash mount of cells of peridium. H. Immature ascomata and hyphopodia. I–K. Asci containing ascospores. L–O. Hyaline ascospores with single septum. Scale bars:  $C = 1000 \ \mu m$ ,  $D = 100 \ \mu m$ ,  $E = 50 \ \mu m$ ,  $F = 20 \ \mu m$ ,  $G = 10 \ \mu m$ ,  $H = 10 \ \mu m$ ,  $I–O = 5 \ \mu m$ .

Saprobic on lower surface of living leaves. Colonies 3–5 mm diam., superficial, solitary to scattered, black, thin, round to irregular, confluent. Hyphae 6–8 µm wide, thick, superficial, septate, dark brown, branched, straight to curved. Hyphopodia (7–)9.5–12.8(–13) µm diam., alternate to unilateral, unicellular, globose to subglobose, dark brown, formed laterally, thick-walled. Sexual state: Ascomata solitary to scattered, superficial on the black hyphae, immature ascomata (24–)33–50 × (7–)10–16 µm, ellipsoid, dark brown, with a short pedicel 10–24 × 5–8µm, mature ascomata (37–)50–80 µm diam., globose to subglobose, black, membranous, easy to remove, containing few asci. Peridium dark brown, composed of one layer of large cells of textura angularis. Hamathecium with few asci, lacking pseudoparaphyses. Asci (30–)32.5–51.5(–60) × (8.5–)9.0–12.5(–13) µm ( $\bar{x}$ = 42.1 × 10.5 µm, n = 10), 8-spored, bitunicate, pyriform to clavate, thick-walled, with an ocular chamber. Ascospores (10–)11–13(–15) × (3.5–)4–5.5(–6.5) µm ( $\bar{x}$ = 12.1 × 4.8 µm, n = 20), 2–3-seriate, ellipsoid to slightly fusiform, supra median 1-septate, normally unequal, upper cell wider, lower cell narrow, hyaline to pale brown, with oil droplets in each cell.

**Material examined:**—PHILIPPINES. Laguna: Los Banos, Mount Maquiling, on living leaves of *Medinilla myriantha* Merr., 13 December 1913 (S! F: 111745, holotype).

Key to genera of Englerulaceae			
1.	Hyphopodia and appressoria absent	Englerula	
1.	Hyphopodia or appressoria present		
2.	Pseudoparaphyses brown, septate, straight to curved	Parenglerula	
2.	Pseudoparaphyses absent		
3.	Phialides brown, 1-septate, nearly ampulliform, alternate	Goosia	
3.	Phialides absent		
4.	Perithecia, large, greenish brown	Thrauste	
4.	Perithecia, small, yellowish brown		
5.	Ascomata superficial on hyphae or radiating, shield-like plate	Rhytidenglerula	
5.	Ascomata arise from short lateral branches	Schiffnerula	

*Myriangiaceae* Nyl., Mém. Soc. Sci. nat. Cherbourg 2: 9 (1854) MycoBank: MB81866 Synonymy: Phymatosphaeriaceae Speg., Anal. Soc. cient. argent. 26(1): 57 (1888)

**Notes:**—*Myriangiaceae*, was introduced by Nylander (1854) and are saprobes, occasionally causing leaf spots (Hyde *et al.* 2013). This family is characterized by superficial, solitary, aggregated to gregarious ascostromata containing multi-locules, globose to subglobose asci and oblong to fusiform, hyaline ascospores with 4–7 transverse septa or muriform (Eriksson 1981, Hyde *et al.* 2013).

Butleria Sacc., Annls mycol. 12(3): 302 (1914) MycoBank: MB 692

**Notes:**—The genus *Butleria* was established by Saccardo (1914) as a monotypic genus, characterized by small and black ascomata which develop superficially on leaves without any hyphae. Barr (1979), Lumbsch & Huhndorf (2010), Kirk *et al.* (2001, 2008) listed *Butleria* in *Elsinoaceae*. Li *et al.* (2011) redescribed the type species and illustrated the genus *Butleria* in detail based on holotype specimen and suggested *Butleria* could be placed in the family *Elsinoaceae* as it is a parasite on leaves and has single asci locules scattered throughout a somewhat reduced, but pulvinate ascomata. *Butleria* however, has small, globose and black ascomata with a membranous peridium, and subglobose asci in several locules and has 1-septate, oblong to ovoid ascospores (Li *et al.* 2011). Available evidence suggests that *Butleria* is more similar to genera of *Myriangiaceae* and hence we propose to move *Butleria* to this family.

Type species:—Butleria inaghatahani Sacc., Annls mycol. 12(3): 302 (1914) MycoBank: MB 156750

Dothideomycetes genera *incertae sedis* 

There are 184 genera accommodated in Dothideomycetes genera *incertae sedis* (Lumbsch & Huhndorf 2010) because familial placements are unclear. These doubtful genera lack molecular data in GenBank and need phylogenetic analyses to establish which families they belong to. Below we include *Rhizotexis* in this group as it is not apparent where we can place this genus.

*Rhizotexis* Theiss. & Syd., Annls mycol. 15(1/2): 140 (1917) MycoBank: MB 4715

**Type species:**—*Rhizotexis bauhiniarum* (Henn.) Theiss. & Syd., Annls mycol. 15(1/2): 140 (1917) MycoBank: MB 202256 (Fig. 7) *Basionym: Parodiella bauhiniarum Henn., Hedwigia 43(6): 359 (1904)* 



**FIGURE 7.** *Rhizotexis bauhiniarum* (syntype) A. Herbarium material. B, C. Black ascomata formed on lower side of leaves. D. Section of ascomata. C. Peridium of ascomata. F–I. Asci containing eight ascospores. J–L. Brown ascospores with one septum. Scale bars: A = 10mm, B, C = 500µm, D, E = 50 µm, F–L = 10 µm.

*Epiphytic* on lower surface of living leaves. Sexual state: *Ascostromata* 150–500 µm long, 100–400 µm wide, solitary to gregarious, irregular, black, coriaceous, stromatic. *Ascostromata wall* composed of brown to dark brown cells of *textura angularis*, with side wall 12–38 µm thick, cell 4–9 × 4–6 µm, lower wall 14–37 µm thick, cell 2.5–6.5 × 2.5–7.5 µm. *Locules* (50–)70–170(–180) µm diam., (60–)70–110(–115) µm high, superficial, globose to subglobose to irregular, black, non-ostiolate, containing a few asci. *Hamathecium* with asci and remnants of thinwalled, pseudoparanchymatous cells; pseudoparaphyses not seen. *Asci* (35–)36.5–60(–65) × (14–)15–20(–22) µm ( $\bar{x} = 52.5 \times 17.5 \mu$ m, n = 20), 8-spored, bitunicate, broadly cylindrical, ellipsoidal to clavate, pedicellate, apically rounded and thickened, with an ocular chamber. *Ascospores* (14–)15–20(–21.5) × 5–6 µm ( $\bar{x} = 17.5 \times 5.8 \mu$ m, n =

20), 2–4-seriate, brown to dark brown, oblong,1-septate, septum supra median, upper cell broad and shorter, lower cell longer and oblong, ends rounded to slightly narrow, smooth-walled, constricted at the septum. Asexual state: Unknown.

**Material examined:**—BRAZIL. Amazonas: Jurua, Jurua-Miry, on leaves of *Bauhinia longipetala* (Benth.) Walp. (synonym of *Bauhinia* glabra Jacq.), June 1901, *E. Ule*, No: 2915 (B! 700014085, syntype).

**Notes:**—*Rhizotexis* was introduced by Theissen & Sydow (1917) with a single species, *R. bauhiniarum* ( $\equiv$  *Parodiella bauhiniarum* Henn.). This genus has cylindrical to clavate asci with an ocular chamber and brown to dark brown, oblong ascospores with a single septum. These characters are atypical of those found in other genera of *Englerulaceae* which have globose asci. *Rhizotexis* also differs in its ascostromata with mostly single locules, and lacking superficial hyphae or hyphopodia. Such a combination of characters is atypical of *Englerulaceae*, thus we exclude *Rhizotexis* from this family (Bin 2003, Castlebury *et al.* 1995, Hennings 1904, Hosagoudar *et al.* 2011a,b, Höhnel 1909, 1910, 1918, Thesisen 1916, Theissen & Sydow 1914, 1917).

*Rhizotexis* is similar to other genera in *Parmulariaceae* (i.e. *Parmularia, Parmulariopsis, Rhipidocarpon*) with its locules developing in black ascostromata. *Rhizotexis* is distinct with small, irregular, protuberant ascostromata without ostioles and hamathecium without hyphal filaments; while, other three genera in *Pamulariaceae* have large, globose to subglobose, flattened pseudothecia with long ostioles and hamathecium with hyaline to dark brown filaments (Inácio 2008). *Rhizotexis bauhiniarum* is most similar to *Asterotexis cucurbitacearum* (presently placed in *Asterinaceae*) which has black stromatic ascomata growing gregariously on leaf surfaces (Rehm 1897, Theissen & Sydow 1914) and a similar stromatic wall (von Arx & Müller 1975). *Rhizotexis bauhiniarum* has smaller ascomata (less than 500 µm) and is only known from *Bauhinia longipetala* (Rehm 1897). *Asterotexis cucurbitacearum* has more than 1 mm diam. ascomata and found on *Cucurbita pepo* (gourd) and *Sechium edule* (chayote) (Guerrero 2011). With these morphological characters, we opine that *Rhizotexis* and *Asterotexis* are more closely related to each other. *Rhizotexis* is placed in Dothideomycete genera *incertae sedis* (Lumbsch & Huhndorf 2010) pending fresh collections.

#### Discussion

Small-sized, globose ascomata growing superficially on brown hyphae is characteristic of the genera in *Englerulaceae* (Eriksson 1981, Hyde *et al.* 2013, von Arx & Müller 1975). Species of *Goosia, Parenglerula, Rhytidenglerula, Schiffnerula* and *Thrauste* have dark colonies, developing on living or dead leaves. These taxa have small, ellipsoid to globose ascomata, growing superficially on hyphae, and pyriform, ovate to globose, asci containing 2–8 ascospores and are typical of *Englerulaceae. Rhizotexis* on the other hand has stromatic ascomata which are gregarious on leaf surfaces and lack hyphae and thus this genus is excluded from the family and placed in *Dothideomycetes* genera *incertae sedis*. Molecular data is required to clarify the relationships in *Englerulaceae*. This calls for fresh collections of these fungi from as many and distant localities worldwide.

# Acknowledgments

The Mushroom Research Foundation, Chiang Rai Province, Thailand is thanked for providing a postgraduate scholarship to Dong-Qin Dai, Qin Tian and Saranyaphat Boonmee. Mae Fah Luang University grant for studying Dothideomycetes (no. 56101020032) and international cooperative project of Guizhou province ([2013]7004) to Yong Wang, and the National Natural Science Foundation of China (Project ID: 31000013 and 31360014) to Rui-Lin Zhao are thanked for support. Putarak Chomnunti and Hiran A. Ariyawansa are thanked for assistance in requesting herbarium specimens. The curators of the herbaria B, BPI, FH, NY and S are thanked for providing herbarium material on loan for this study.

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