Aptrootia (Dothideomycetes: Trypetheliaceae), a new genus of pyrenocarpous lichens for Thelenella terricola

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Abstract: The new genus *Aptrootia* Lücking & Sipman is described for *Thelenella terricola*, an enigmatic terricolous and muscicolous, pyrenocarpous taxon known from Papua New Guinea and Costa Rica, and the new combination *Aptrootia terricola* (Aptroot) Lücking, Umaña & Chaves comb. nov. is introduced. *Aptrootia* is characterized by completely immersed perithecia with brown-black ostiolar region, surrounded by a white, cartilaginous thallus resembling that of *Gomphillaceae*. The hamathecium is typical of *Trypetheliaceae*, with thin, anastomosing paraphysoids embedded in a gelatinous matrix, but the dark brown ascospores are otherwise unknown within the family. The only known species was tentatively described in *Thelenella*, but hamathecium type and molecular data place *Aptrootia* within *Trypetheliaceae*.

Key words: Costa Rica, loculoascomycetes, mtSSU rDNA, Papua New Guinea, Thelenella, TICOLICHEN

Introduction

Several years ago, Aptroot (1999) described the enigmatic species *Thelenella terricola* from Papua New Guinea. The new taxon was characterized by its growth over bryophytes and debris on the ground, its apparently non-lichenized, cartilaginous thallus, immersed perithecia with carbonized involucrellum, thin anastomosing paraphyses, and large, distoseptate, dark brown ascospores. Aptroot (1999) doubted the position of his new taxon within *Thelenella* and suggested that the species might warrant separation at generic level, but eventually opted for a conservative strategy and described the new species in an existing genus.

During our ongoing TICOLICHEN biodiversity inventory in Costa Rica (Chaves et al. 2004; Lücking et al. 2004, 2006; Nelsen et al. 2006), we collected a strange muscicolous lichen which, at first glance, resembled the thallus and appressed, umbelliform hyphophores of Gyalideopsis lambinonii Vězda. To our surprise, closer examination revealed that the 'hyphophores' were the ostiolar regions of completely immersed perithecia. The trypethelioid hamathecium and large, distoseptate, muriform ascospores suggested placement in the artificial genus Laurera (Harris 1986, 1995), but none of the known species in that genus matched this material. We then sequenced a specimen and included it in a molecular phylogenetic study on pyrenocarpous lichens, which confirmed placement within Trypetheliaceae but outside of any of the others sequenced (Del Prado et al. 2006). We then studied the material from Costa Rica again and discovered that it was conspecific with Thelenella terricola from Papua New Guinea. Our molecular data thus confirmed that the latter is not a genuine

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Thelenella but belongs in Trypetheliaceae, sister to a branch including Bathelium and the Trypethelium floridanum lineage, which includes Campylothelium p.p. and possibly Laurera s. str. We therefore describe the new genus Aptrootia to accommodate Thelenella terricola.

Aptrootia Lücking & Sipman gen. nov.

Genus novum familiae *Trypetheliacearum*. Thallus crustaceus, leviter lichenisatus, cartilagineus, corticatus, albo-cinereus. Alga ad *Trentepohliam* pertinens. Perithecia immersa et strato thallino obtecta, parietibus nigris. Paraphysae ramoso-connexae. Asci bitunicati, unispori. Ascosporae muriformes, distoseptatae, fuscae, juveniliter hyalinae et amyloideae.

Typus: Aptrootia terricola (Aptroot) Lücking, Umaña & Chaves comb. nov. Thelenella terricola Aptroot, Fungal Diversity 2: 45 (1999); type: Papua New Guinea, Simbu Province, Mount Wilhelm, 3600 m, Aug 1992, Aptroot 32649 (CBS—holotype); Bundi Gap on road Keglsugl-Bundi, 2800 m, October 1995, Aptroot 37658 (CBS—paratype).

(Figs 1–3)

Thallus muscicolous or over debris, crustose, lichenized in patches, with thin, cartilaginous, corticiform layer, white-grey, slightly shiny (Fig. 1A–D). *Photobiont Trentepohlia*, with elongate to angular-rounded, green cells in irregular groups and threads (Fig. 1E–H).

Perithecia immersed and covered by a thin thallus layer, pyriform, exposing only the brown-black ostiolar region which appears radiately ridged when hydrated (Figs 1C-D, 2A). Perithecial wall not differentiated into excipulum and involucrellum, inner layer and upper parts carbonized, black, outer layer light brown in thin sections (Fig. 2A). Ostiolar region with short periphysoids (Fig. 2B). Hamathecium of branched and anastomosing paraphysoids, thin and widely spaced, embedded in a gelatinous matrix (Fig. 2C). Asci fissitunicate, clavate, ocular chamber indistinct, when young with apically widening lateral walls (Fig. 2D). Ascospores single, muriform, distoseptate, dark brown when mature within the asci (young ascospores colourless and IKI+ blue), $170-230 \times 40-70 \,\mu\text{m}$ (Fig. 2E–F).

Conidiomata not observed.

Chemistry. No substances detected by TLC.

Etymology. Named after our colleague and collaborator in the TICOLICHEN project, the Dutch lichenologist and mycologist André Aptroot, in recognition of his numerous contributions to tropical lichenology.

Ecology and distribution. The taxon grows muscicolous and over debris in wet tropical montane regions in (semi-)exposed situations, with (micro-)habitat preferences very similar to those of *Gomphillus ophiosporus* and certain species of *Gyalideopsis*. Thus far, it is known from Papua New Guinea and Costa Rica, but certainly overlooked and probably pantropical.

Remarks. Thelenella terricola was already recognized by Aptroot (1999) as an anomalous element within Thelenella. It agrees with the latter (Mayrhofer 1987) in the perithecioid ascomata with periphysoids and anastomosing paraphysoids, the fissitunicate, non-amyloid asci producing muriform ascospores, the corticiform layer, and the absence of secondary substances. However, our study of the Costa Rican material confirmed that the species differs from Thelenella in significant details: the trentepohlioid photobiont, the carbonized perithecial wall, the structure of the paraphysoids, and the single-spored asci with apically widening walls, producing very large, distoseptate, dark brown ascospores.

Because of the trypethelioid hamathecium and large, distoseptate ascospores, we had tentatively identified the Costa Rican material as a *Laurera* sp., and as such it was included in a molecular phylogenetic analysis, which strongly supported placement of the taxon within *Trypetheliaceae* (Del Prado *et al.* 2006). *Laurera* itself is an artificial assemblage of taxa defined by having solitary ascomata with apical ostiole producing muriform ascospores (Harris 1986, 1995) and contains many species more closely related to taxa within other genera (*Astrothelium, Bathelium, Campylothelium, Cryptothelium, Trypethelium*) than within *Laurera*. If



FIG. 1. Aptrootia terricola (Costa Rica, Lücking 17211). A–D, thallus with perithecia, note the light greenish, lichenized patches and the erumpent, brown-black ostiolar areas (arrows); E–H, trentepohlioid photobiont, note the close association with the fungal hyphae and the partially filamentous arrangement of the photobiont cells (arrows). Scales A–D=1 mm; E–H=10 µm.



FIG. 2. *Aptrootia terricola* (Costa Rica, *Lücking* 17211). A, section through perithecium showing perithecial wall structure, hamathecium, and asci with ascospores; B, periphysoids; C, anastomosing paraphysoids; D, young ascus, showing apically widening lateral walls and basal stipe; E, immature, still hyaline ascospore; F. mature, dark brown ascospore. Scales A=200 μm; B–F=50 μm.

applying more natural criteria for the delimitation of genera, as has been done in Pyrenula (Harris 1989) or more recently in Graphidaceae and Thelotremataceae (Staiger 2002; Frisch et al. 2006), Laurera s. str., which as well as the type L. varia (Fée) Zahlbr. includes the common and widespread L. megasperma (Mont.) Riddle, is probably characterized by solitary to rarely aggregate perithecia covered by thick thallus tissue and with an apical to lateral ostiole, producing both transversely septate and muriform ascospores. As such, Laurera s. str. would include many species presently included in Campylothelium and Cryptothelium (Harris 1995), but possibly also Trypethelium (T. floridanum group) and even Astrothelium. Although Laurera varia and L. megasperma have not yet been sequenced, the terminal position of taxa with Laurera morphology, viz. Trypethelium floridanum and Campylothelium cf. superbum, distant to Aptrootia terricola (Fig. 3), suggests that the latter is not closely related to Laurera s. str. This is supported by the dark brown ascospores and the very different ecology and thallus and perithecial morphology.

Aptrootia also differs from the other named genera in its peculiar ecology and the dark brown ascospores, the latter being a unique feature within the family. Except for Architrypethelium, which has very large 3-septate ascospores with longitudinal folds and a very different thallus and perithecial morphology compared to Aptrootia, members of Trypetheliaceae develop brown ascospores only exceptionally after ejection through the ostiole, when they are deposited on the surrounding thallus (Harris 1986). In Aptrootia, the dark pigmentation also develops late, but dark brown ascospores can be readily observed within the asci. The young amyloid ascospores are also a rare feature within the family, otherwise known only from Polymeridium proponens (Nyl.) R. C. Harris. The position of the Costa Rican material on its own branch within the Trypetheliaceae (Fig. 3) confirms that it deals with a separate lineage, which justifies establishing a new genus for this taxon. In a parallel phylogenetic study (Schmitt et al. 2005), *Thelenella* itself was shown to be part of the Ostropomycetidae within the Lecanoromycetes, thus unrelated to both the *Pyrenulales* (Eurotiomycetes) and the *Trypetheliaceae* (Dothideomycetes).

Aptroot (1999) describes 'Thelenella' *terricola* as principally non-lichenized and growing over miscellaneous bryophyte material and debris including various algae. However, careful examination of the Costa Rican material showed the thallus to have a patchily light greenish colour when fresh or hydrated (Fig. 1A-B). These patches uniformly contain the same alga, which belongs to the genus Trentepohlia (Fig. 1E-H). In the material from Papua New Guinea, the lichenized patches are less obvious but also present. We therefore conclude that Aptrootia terricola is lichenized, although the algal cells may be difficult to observe, and the taxonomic position of the photobiont is consistent with the placement of the new genus within the Trypetheliaceae, whose members are lichenized with Trentepohlia (Harris 1986, 1995).

Most pyrenocarpous, lichenized fungi are corticolous, foliicolous, or saxicolous, and only few species are found growing over bryophytes and debris, a substratum more commonly inhabited by discocarpous Lecanoromycetes. Muscicolous species can be found in the genus Chromatochlamys, which is related to *Thelenella* and differs from Aptrootia by its chlorococcoid photobiont, non-carbonized perithecial wall, and usually 8-spored asci producing hyaline, thin-walled ascospores, among other details. Species of Porina might also be found overgrowing bryophytes, and while these have a trentepohlioid photobiont and sometimes a carbonized perithecial wall, their paraphyses are unbranched, their asci entirely thin-walled and always 8-spored, and their ascospores hyaline and thin-walled.

As already mentioned, at first glance the thallus of *Aptrootia terricola*, with its immersed perithecia displaying only the brown-black ostiolar area, closely resembles the thallus of *Gomphillaceae*, where several species in the genera *Gomphillus* and *Gyalideopsis* frequently overgrow bryophytes.



FIG. 3. Phylogenetic position of Aptrootia (Costa Rica, Lücking 17211) within the Trypetheliaceae, based on mtSSU rDNA data (adapted from Del Prado et al. 2006). Only the Dothideomycetes branch is shown; for rest of the tree, as well as details on phylogenetic analysis, see Del Prado et al. (2006). Thick branches indicate 75% or higher bootstrap support and 95% or higher Bayesian posterior probabilities.

When hydrated, the ostiolar area of the perithecia of *Aptrootia terricola* displayes radiating ridges and hence almost 'mimics' perfectly the adnate, umbelliform hyphophores of *Gyalideopsis lambinonii* Vězda. Microscopic examination is thus necessary to reveal the true nature of this enigmatic lichen.

Additional specimens examined. Costa Rica: Guanacaste: Volcán Tenorio National Park, Pilón Biological Station (Arenal-Tempisque Conservation Area), Tilarán Ridge, 140 km NW of San José, 25 km NNW of Tilarán, near Bijagua, access road to station and river, 84°59'W, 10°43'N, 700 m, lower montane cloud forest zone, exposed trees and fence posts along pasture, over bryophytes on bark (lower trunk), exposed, 2004, Lücking 17211 (F, INB-4004850), Sipman 51849 (B, INB); Alto Masis Section (Arenal-Tilarán Conservation Area), Tilarán Ridge, 125 km NW of San José, 15 km NNW of Tilarán, near Tierras Morenas, access road to station, 85°00'W, 10°36'N, 850-900 m, lower montane moist forest zone, exposed trees and fence posts and forest regrowth along savanna-like pasture, over bryophytes on bark (lower trunk), semi-exposed, 2004, Lücking 17304d (F); shaded, Mar 2004, Lücking 17308a (ABL, F, INB-4008173).—**Papua New Guinea:** Simbu Province: Mount Wilhelm area, Bundi Gap, on road Keglsugl-Bundi, 2800 m, 1992, Sipman 35503 (B). Northern Province: Owen Stanley Range, Myola, 3 km NE of guesthouse, 2700 m, 1995, Sipman 38333 (B).

The Ticolichen biodiversity inventory in Costa Rica was supported by a grant from the National Science Foundation (DEB 0206125 to The Field Museum; PI Robert Lücking) and by funds from the World Bank and the Dutch Government to the Instituto Nacional de Biodiversidad (INBio). We appreciate the support of the Sistema Nacional de Áreas de Conservación (SINAC) and the Ministerio de Ambiente y Energía (MINAE) in receiving the necessary collection permits. Enia Navarro, Eida Fletes, Ronald Gómez, and Eduardo Alvarado (INBio), Susan Will-Wolf, Marie Trest, and Matthew Nelsen (University of Wisconsin-Madison), as well as André Aptroot (ABL Herbarium), Martin Grube (University of Graz, Austria) and William Buck (New York Botanical Garden), participated in the field work, and their support and company are warmly acknowledged.

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Accepted for publication 7 October 2006