Fungicolous pyrenomycetes 2. Ascocodinaea, gen nov., and reconsideration of Litschaueria

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Abstract: The name Litschaueria corticiorum was found to have been misapplied to a common pyrenomycete that occurs on old basidiomata of Stereum species. The new genus and species Ascocodinaea stereicola is proposed for it; its proven anamorph is a Codinaea species. A second new species, A. polyporicola, which occurs on old basidiomata of Polyporus species, is also proposed; its presumed anamorph is also a Codinaea species.

Key Words: Ascomycetes, *Codinaea*, fungicolous fungi, systematics

INTRODUCTION

While studying pyrenomycetes that occur on basidiomata of the Aphyllophorales, we commonly encountered a pyrenomycete on *Stereum* spp. that, using current literature, could be identified as *Litschaueria corticiorum* (Höhnel) Petrak (Barr, 1976). This fungus was grown in pure culture from ascospores. Further study revealed that the name *L. corticiorum* has been misapplied and that this common pyrenomycete cannot be accommodated in any described genus or species. A second, congeneric, undescribed species on polypores was found. These fungi are discussed and described.

MATERIALS AND METHODS

Single ascospores were isolated with the aid of a micromanipulator on cornmeal agar (Difco) with 2% w/v dextrose added (CMD). Colony characters were taken from CMD cultures grown at 20–22 C under 12 h darkness alternating with 12 h cool white fluorescent light.

Dry specimens were rehydrated briefly in 3% (aq.) KOH and the mounts were then flooded with water. Photographs were taken of ascospores and asci mounted in water. Photographs were taken of perithecial sections mounted in 100% lactic acid.

Five types of microscopy were used in this study. These are indicated in the legends to the illustrations as bright field (BF), fluorescence (FL), interference contrast (IC), phase contrast (PC), and scanning electron microscopy (SEM). The optical brightener calcofluor (0.05% w/v in sodium phosphate buffer at pH 8; Sigma Chemical Co.) was used for FL. Material for SEM was prepared following the procedure in Rogerson and Samuels (1993).

Frequent collectors names are abbreviated as F. Candoussau (FC), C. T. Rogerson (CTR), J.-F. Magni (JFM) and G. J. Samuels (GJS).

DESCRIPTIONS

Ascocodinaea Samuels, Candoussau et Magni, gen. nov.

Hymenoascomycete, perithecia nigra, setosa, ostiolata. Paraphyses anguste filiformes, ramosae. Asci unitunicati, ad apicem cum annulo instructi, J-. Ascosporae 3-septatae, versicoloratae. Anamorph *Codinaea*; dematiaceous; conidia curvatae, enteroblasticae, hyalines, mucosae.

Type species. Ascocodinaea stereicola Samuels, Candoussau & Magni

1. Ascocodinaea stereicola Samuels, Candoussau et Magni, sp. nov. FIGS. 1–9, 11

Perithecia ovoidea, nigra, 143–212–(288) µm alta, 99– 170(–250) µm µm lata. Setosa; setae acutae, nigra, (15-)54– 96(–139) µm longa, sterilia. Paraphyses filiformia, 2 µm lata, septata, ramosa. Asci cylindrici, (54-)73–91(–105) × (4.7-)6.4–8.8(–10.8) µm; ad apicem cum annulo instructi. Ascosporae ellipsoideae vel fusiformes, (7.4-)10.4–13.0(– 15.4) × (3.7-)4.7–6.1(–7.6) µm, 3-septatae; cellulae centrales brunnea, cellulae terminales hyalinae. Anamorphosis *Codinaea* sp.

Holotypus in basidiomata *Stereum* sp., Kentucky, U. S. A., G. J. Samuels 95-186 (BPI).

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FIGS. 1–9. Ascocodinaea stereicola. 1–3. Median, longitudinal sections through the same perithecium showing wall anatomy and arrangement of asci. 4–7. Asci and ascospores. Paraphyses visible in FIGS. 4–6 (arrows). FIGS. 8, 9. Codinaea anamorph, conidiophores from nature. FIGS. 1, 2, 5–9: IC; 3: FL; 4: PC. FIGS. 1–3, 7–9 from GJS 90-245; 4–6 from GJS 95-186. Scale bars: 1–4, 8, 9 = 50 μ m; 5 = 25 μ m; 6, 7 = 10 μ m.

Anamorph. Codinaea sp.

FIGS. 8, 9, 11

Sterile setae and conidiophores abundant, arising from the host surface among perithecia. Perithecia forming directly on the hymenial surface, gregarious, superficial with base slightly immersed in the substrate, gray to black, translucent brown in 3% KOH, ovoidal, with an acute apex, 143–212(–288) μ m high, 99–170(–250) μ m wide, collapsing deeply by lateral pinching when dry, with stiff, erect, acute, unbranched, septate, black setae arising as modified cells of the surface of the upper half of the perithecium, (15–)54–96(–139) μ m long, (4-)7–11(–15) μ m wide at base, thick-walled, never conidiogenous. Perithecial wall translucent brown by transmitted light, cells at the surface thin-walled *textura epidermoidea*; lateral perithecial wall ca. 10 μ m wide, of a single region of ca. 5 layers of elliptical cells measuring ca. $5 \times 1.5 \,\mu$ m with walls thin (<0.5 μ m) and pigmented brown. Perithecial apex formed of cells enlarged to 7.5 μ m diam and arranged in files. Ostiolar canal periphysate; periphyses continuous with the paraphyses. Paraphyses abundant among, and overreaching, mature asci, infrequently branched, septate, ca. 2 μ m wide but slightly enlarged at the tip. Asci cylindrical, (54-)73–91(-105) × (4.7-)6.4–8.8(-10.8) μ m, 8-spored; apex with a thin ring pierced by a pore;



FIGS. 10, 11. Codinaea anamorphs of Ascocodinaea species. 10. A. polyporicola, from nature. 11. A. stereicola. All from CMD except the conidiophore and seta on the extreme right, which is from nature. FIG. 10 from GJS 90-245; 11 from 25 June 1978, CTR. Scale bars = $10 \mu m$.

ascospores uniseriate with overlapping ends. Ascospores ellipsoidal to fusiform, slightly curved, (7.4-)10.4–13.0(–15.4) × (3.7-)4.7–6.1(–7.6) µm, 3-septate, central cells translucent brown and end cells hyaline, smooth.

Anamorph in nature. Conidiophores macronematous, mononematous, stiff, erect, unbranched black, morphologically indistinguishable from the sterile setae, $(7-)35-95(-165) \mu m$ long, $(2.3-)3.0-6.4(-9.0) \mu m$ wide at base, each bearing a single, terminal, integrated conidiogenous cell. Conidiogenous cells monophialidic, enteroblastic, proliferating percurrently or sympodially; tip not flared, with slight periclinal thickening at the conidiogenous locus. Conidia broadly ellipsoidal, cylindrical, or inequilateral, often slightly curved, 0–1-septate, hyaline, lacking a visible basal abscission scar, smooth, held in a drop of hyaline slime at the tip of each conidiophore.

Characteristics in culture. Colonies grown 3 wk, 3– 5 mm diam, raised, velutinous, aerial hyphae in densely disposed, acute, rope-like strands, light to dark brown and zonate; conidiophores arising throught the colony in the aerial mycelium. Conidiophores and conidiogenous cells as in nature but conidiophores shorter, 42–105 μ m long. Conidiogenous locus proliferating percurrently for a short distance to produce annellations or for a long distance and then forming an entire conidiogenous cell. Conidia ellipsoidal, oblong or tending to be reniform, lacking a visible basal abscission scar, (5.4)6.0–9.1(–10.8) \times 3.6–4.7(–5.6) μ m, unicellular or with a single median septum, hyaline, forming by replacement-wall building, held in slimy heads at the tip of each conidiophore.

Habitat. On old basidiomata of species of Stereum including S. hirsutum (Willdenow: Fr.) S.F. Gray and S. ostrea (Blume & Nees) Fr.

Known distribution. France. and North America.

HOLOTYPE. UNITED STATES. KENTUCKY: Laurel County, Daniel Boon National Forest, Laurel River Lake Recreation Area, Cane Creek Wildlife Refuge, elev. 350 m, on *Stereum* sp., 27 Sept. 1995, *GJS* 95-186 (BPI).

Additional specimens examined. FRANCE. AIN: Bois d'Evieu 01, on Stereum sp., 30 Oct. 1994, JFM A94119 (BPI). PYRÉNÉES ATLANTIQUES: Bois Bastard de Pau 64, on Stereum sp., 11 Nov. 1994, FC 312 (BPI). UNITED STATES. DELAWARE. Wilmington, on old Corticium [=Stereum] on an oak log lying on the ground in the woods, 15 Nov. 1894, Commons 2667 (NY). FLORIDA: Alachua County, 6 mi NW of Gainesville, University of Florida Horticultural Farm, on Stereum, 10 Aug. 1985, CTR (NY); 4.5 mi NW of Gainesville, Devil's Millhopper Geological Site, on Stereum ostrea, 26 Aug. 1977, CTR 77-101, 77-103 (NY). Wakulla County, Apalachicola National Forest, W side of Bradwell Bay Wilderness Area, entrance to Florida National Scenic Trail, W side of Forest Sevice Rd. 314, 30°08'N, 84°37'W, on Stereum sp., 1 Jan. 1991, GJS 90-245 & CTR (BPI 1107395). GEORGIA: Walker County, Chattahoochee National Forest, Keown Falls Recreation Area, on Stereum sp., 27 Sept. 1992, CTR & GJS (NY). ILLINOIS: Mclean County, Funk Grove, University of Illinois Timber Woods, on Stereum ostrea, 13 Aug. 1965, CTR (NY). KENTUCKY: Laurel County, Daniel Boon National Forest, Laurel River Lake Recreation Area, Cane Creek Wildlife Refuge, elev. 350 m, on Stereum sp., 27 Sept. 1995, GJS 95-184 (BPI; culture ATCC 200439). LOUISIANA: St. Tammany Parish, Honey Island Swamp, near Pearl River, on Stereum ostrea, 6 June 1976, CTR (NY). NORTH CAROLINA: Jackson County, Nantahala National Forest, woods near Upper Falls of Whitewater River, on Stereum hirsutum, 5 Oct. 1972, CTR & GJS (NY). Macon County, Nantahala National Forest, along Ellicott Rock Trail from Bull Pen Rd along Chattooga River, on Stereum ostrea, 4 Oct. 1972, CTR & GJS (NY); along Scotsman Creek, Branch of Chattooga River, Bull Pen Road, on Stereum sp., 3 Aug.

1961, R.H. Petersen & CTR (NY). Transylvania County, along Corbin Creek, branch of Whitewater River, on Stereum, 9 Aug. 1961, R.H. Petersen & CTR (NY); along Toxaway River near junction with Bear Wallow Creek, on old Stereum on log, 29 Jul. 1961, R.H. Petersen & C. T. Rogerson (NY, BPI 1107396). Wake County, Lake Johnson Park, on Stereum, 12 Nov. 1972, J. Julis, Menge 622 (NY). SOUTH CAROLINA: Oconee County, 3.2 mi S of state line, 4 mi S of Upper Falls of Whitewater River, on Stereum sp., 14 Aug. 1961, R.H. Petersen & CTR (NY).

Notes. We have isolated this species into pure culture from ascospores twice (*GJS 90-245, GJS 95-186*). C. T. Rogerson has also grown the species in pure culture. Because perithecia form in cultures derived from more than one ascospore, but not in monospore cultures, the species is heterothallic.

2. Ascocodinaea polyporicola Samuels, Candoussau et Magni, sp. nov. FIGS. 10, 12–20

Ascocodinaeae stereicolae sed ascosporae $(10.4-)12.7-15.7(-18.0) \times (4.2-)5.0-6.4(-7.3) \ \mu m$ et conidia $(5.2-)9.5-13.3(-17.3) \ \mu m$. Anamorphosis *Codinaea* sp.

Holotypus in basidiomata *Polyporus pargamenus*, U.S.A., Vermont, 30 Aug. 1981, C. T. Rogerson 81-106 (NY).

Anamorph. Codinaea sp. (presumed) FIG. 10

Sterile setae and conidiophores abundant, arising from the host surface among perithecia. Perithecia forming directly on the hymenial surface, gregarious, superficial with base slightly immersed in the substrate, gray to black, translucent brown in 3% KOH, ovoidal, with an acute apex (142-)155–177(–186) μm high, (99-)104-126(-147) µm high, collapsing deeply by lateral pinching when dry, with stiff, erect, acute, unbranched, septate, black setae arising as modified cells of the surface of the upper half of the perithecium, (46-)60-100(-123) µm long, (5-)8-12(-19) µm wide at base, thick-walled never conidiogenous. Perithecial wall translucent brown by transmitted light, cells at the surface thin-walled textura epidermoidea; lateral perithecial wall 25-30 µm wide, of a single region of several layers of nondescript cells or intertwined hyphae, the hyphal aspect predominating toward the perithecial apex, walls of cells toward the exterior pigmented and tending to have thinner walls than cells toward the interior; perithecial apex not anatomically distinct from the wall below. Ostiolar canal periphysate; periphyses continuous with the paraphyses. Paraphyses abundant among, and overreaching, mature asci, frequently branched and forming a reticulum around asci, septate, 1.2-1.7 µm wide, apically free but tips not enlarged. Asci cylin-



FIGS. 12–20. Ascocodinaea polyporicola. 12. General morphology of a perithecium. 13. Submedian section of a mature perithecium. 14. Median longitudinal section through a perithecial apex showing periphyses lining the ostiolar canal and continuous with the paraphyses lining the perithecial locule (arrow). 15. Perithecial setae. 16–20. Asci, ascospores and paraphyses (arrow). Note the ascal ring in FIGS. 19, 20. FIGS. 12, 15: BF; 13, 14, 16–19: IC; 14d, 20: PC. FIGS. 12–16 from CTR 81-106; 17 from CTR 76-44. Scale bars: $10 = 200 \mu m$; $13-18 = 50 \mu m$; $19, 20 = 10 \mu m$.

drical, $(103-)111-135(-143) \times (6.5-)7.0-8.5(-9.8)$ µm, 8-spored; apex apex with a thin ring pierced by a pore; ascospores uniseriate with overlapping ends. Ascospores ellipsoidal, $(10.4-)12.7-15.7(-18.0) \times (4.2-)5.0-6.4(-7.3)$ µm, 3-septate, central cells translucent brown and end cells hyaline, smooth.

Anamorph in nature. Conidiophores macronematous, mononematous, stiff, erect, unbranched black, morphologically indistinguishable from the sterile setae, (53-)86–138(–202) μ m long, (5.5-)6.0–9.0(–12.0) μ m wide at base, each bearing a single, terminal,integrated conidiogenous cell. Conidiogenous cells monophialidic, enteroblastic, proliferating percurrently, sympodial proliferation not observed; tip not flared, with slight periclinal thickening at the conidiogenous locus. Conidia allantoid, 0–1-septate, hyaline, lacking a visible basal abscission scar, smooth, held in a drop of hyaline slime at the tip of each conidiophore.

Habitat. On old basidiomata of species of Polyporus, including P. pargamenus and P. versicolor.

Known distribution. France. United States (Maine, New York, Virginia, Vermont).

HOLOTYPE. UNITED STATES. VERMONT: 7 mi SE of Bennington, Burgess Rd., on *Polyporus pargamenus*, 30 Aug. 1981, *CTR 81-106* (NY).

Additional specimens examined. FRANCE. ALLE-GRE (Haute Loire): Forêt de Menteyres, elev. 1200 m, on Trichaptum fuscoviolaceum, 5 Oct. 1996, FC 457 (BPI). UNITED STATES. MAINE: Franklin County, Flagstaff Rd., on Polyporus pargamenus, 7 Sep. 1971, H.E. & M.E. Barr Bigelow MEBB 5903b (NY). NEW YORK: Fulton County, woods NE of Cranberry Creek, W side Sacandaga Reservoir, on tubes of Polyporus pargamenus, 1 Oct 1970, CTR, K. P. Dumont, J. H. Haines & GJS (NY). VIRGINIA: Craig County, Jefferson National Forest, Potts Mountain, on old Polyporus pargamenus, 25 June 1978, CTR (NY).

Notes. We have not had fresh material of *C. polyporicola*, thus the *Codinaea* anamorph is presumed on the basis of juxtaposition and also similarity of the two species of *Ascocodinaea*.

Ascocodinaea polyporicola and C. stereicola are easily distinguished. Ascospores, conidia and conidiophores of C. polyporicola are larger than are those of C. stereicola. Paraphyses of C. polyporicola are finer and much more intricately branched than in C. stereicola. Finally, the presumed anamorph of C. polyporicola is much more densely disposed around the perithecia and conspicuous on the host than is the anamorph of C. stereicola.

DISCUSSION

Petrak (1923) introduced Litschaueria for Helminthosphaeria corticiorum Höhnel. He distinguished the new genus from *Helminthosphaeria* Fuckel, and its type *H. clavariarum* (L.-R. Tulasne) Fuckel, citing differences in host, anamorph, ascospore septation, and perithecial anatomy. Petrak considered the two genera to be closely related. Barr (1976) examined the type specimen of *H. corticiorum* and, interpreting the material as being largely immature, identified a common pyrenomycete that occurs on *Stereum* as *L. corticiorum*. Samuels et al. (1996) concluded that *Helminthosphaeria* is the correct genus for *H. corticiorum*; it is common on basidiomata of corticiaceous basidiomycetes in Europe. They did not accept *Litschaueria* as distinct from *Helminthosphaeria*.

The type collection of *H. corticiorum* has young perithecia and few discharged ascospores. Ascospores in asci are unicellular and are olivaceous or grey-greenish. Discharged ascospores have three septa, a minute pore in each end, and are uniformly pigmented. Conidia of a *Diplococcium* Grove anamorph, a characteristic of *Helminthosphaeria*, are present (Samuels et al., 1996). There is no doubt that this is a species of *Helminthosphaeria*.

Ascocodinaea stereicola, on Stereum, differs from H. corticiorum in several respects. Most conspicuously, ascospores in asci of A. stereicola are 4-celled, and the middle cells are brown while the end cells are hyaline (FIG. 7). These ascospores do not have pores. Unlike species of Helminthosphaeria, ascospores of which may develop septa late in development or-as in H. corticiorum-only after discharge from asci, ascospores of Ascocodinea develop septa at a very early stage in their development. Perithecia of H. corticiorum are carbonaceouos whereas perithecia of A. stereicola are leathery and easily crushed. This is a reflection of differences in cells of the perithecial wall. Cells of the wall of H. corticiorum, and of Helminthosphaeria in general, are thick-walled, polyhedral and more or less heavily melanized (Samuels et al., 1996) while the cells of the wall of Ascocodinaea species are more hyphal or at least not thick-walled and not melanized (FIGS. 2, 14). The perithecial apex in Helminthosphaeria is formed of conspicuous clavate cells while the perithecial apex in Ascocodinaea is less sharply differentiated from the cells that form the rest of the perithecial wall (FIG. 2). Finally, the anamorph A. stereicola is a species of Codinaea, with phialides and hyaline conidia (FIG. 8, 9, 11). The putatative anamorphs of Helminthosphaeria species are Diplococcium species with pigmented conidia that form tretically (Ellis, 1971). Because of these differences, we see little relationship between Helminthosphaeria and Ascocodinaea.

We were surprised that A. stereicola has not previously been described because it is so common on basidiomata of Stereum. Petrak (1940) synonymized Sphaeria porothelia Berk. & M. A. Curtis sensu Kirschstein (1939, i.e. *Melanostsigme* Kirschst.) and *Litschaueria corticiorum*, but *S. porothelia* is a loculoascomycete, *Capronia porothelia* (Berk. & M. A. Curtis) M. E. Barr (Barr, 1991; Dothideales: Herpotrichiellaceae), and thus is not an earlier name for this fungus. The second species of *Ascocodinaea*, *A. polyporicola*, is known from only four collections on old basidiomata of thin-fleshed *Polyporus* species.

Ascocodinaea is a genus of the Sordariales family Lasiosphaeriaceae (*sensu* Barr, 1990) for the following reasons: perithecia are black, setose, superficial, have a pseudoparenchymatous wall and a hamathecium of apically free paraphyses (FIGS. 4, 5; 16–20); ascospores are septate, nonporose and in part translucent brown (FIGS. 5, 7; 17–20); anamorphs are dematiaceous and phialidic (FIGS. 10, 11).

Two additional genera of the Lasiosphaeriaceae are characterized, in part, by having versicolorous ascospores and setose ascomata. *Ascocodinaea* can be distinguished from them by combined teleomorph, anamorph and substrate characters. The anamorphs of *Chaetosphaerella* Müller & Booth (1972) species are species of *Oedemium* Link, and the perithecial wall is formed of thick-walled, polyhedral cells. *Melanochaeta* Müller et al. (Müller et al., 1969; Müller and Samuels, 1982) species have *Sporoschisma* anamorphs, and the perithecial hairs are capitate. Species of neither of these genera occur on other fungi.

Barr (1976) reported finding a Cylindrotrichum Bonorden anamorph associated with perithecia of A. stereicola (as L. corticiorum). Later, S. J. Hughes studied cultures that had been derived from ascospores of the fungus by C. T. Rogerson and concluded that the anamorph is a species of Codinaea Maire. It was distinguished from Cylindrotrichum by having curved and not straight conidia (in litt. to C. T. Rogerson, 20 Jan. 1977, NY). The only ascomycetes known to be linked to either Codinaea or Cylindrotrichum are species of Chaetosphaeria L-R. & C. Tul. (Hughes and Kendrick, 1968; Gams and Holubová-Jechová, 1976). Ascocodinaea is distinguished from Chaetosphaeria in the Lasiosphaeriaceae on ascospore characters and on the presence or absence of perithecial setae. Ascospores of Chaetosphaeria are bicellular or phragmosporous and hyaline. Any vestiture on perithecial walls in species of Chaetosphaeria is composed of conidiophores. While Chaetosphaeria species may be found on basidiomycetes, the only known species of Ascocodinaea occur only on basidiomycetes. The anamorph of Ascocodinaea, with darkly pigmented, mononematous conidiophores and enteroblastically produced conidia, is entirely consistent with anamorphs of other members of the Lasiosphaeriaceae (e.g. Hughes and Kendrick, 1968; Gams and Holubová-Jechová, 1976; Barr and Crane, 1979; Constantinescu et al., 1995).

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