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Freshwater ascomycetes: new and noteworthy species from aquatic habitats in Florida

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Abstract: As part of a distributional study of freshwater ascomycetes in Florida, a number of new taxa were encountered. The new taxa include six Sordariomycetes, *Aniptodera megaloascocarpa* sp. nov., *Flammispora pulchra* sp. nov., *Hanliniomyces hyaloapicalis* gen. et sp. nov., *Lockerbia striata* sp. nov., *Phomatospora triseptata* sp. nov. and *Physalospora limnetica* sp. nov., and three Dothideomycetes, *Caryospora obclavata* sp. nov., *Lepidopterella tangerina* sp. nov. and *Ophiobolus shoemakeri* sp. nov. These taxa are described and illustrated. Six additional species are reported from Florida for the first time; among them, two species are new reports from freshwater habitats.

Key words: lentic, lotic, saprobic fungi, submerged wood, systematics

INTRODUCTION

As part of a large-scale latitudinal distribution study of freshwater ascomycetes in North and Central America, Raja (2007) investigated the latitudinal, habitat and substrate distribution patterns of freshwater ascomycetes along the Florida Peninsula. Thus far collections from Florida have yielded one new genus (Raja et al 2008) and four new species of freshwater ascomycetes (Raja and Shearer 2006a, b; 2007). In this paper we describe and illustrate additional new taxa from aquatic habitats in Florida.

MATERIALS AND METHODS

Collection sites.—Five sites along the Florida Peninsula were established based on latitude and presence of appropriate aquatic habitats: (i) Blackwater River State Forest (30°N), (ii) Apalachicola National Forest (29–30°N), (iii) Ocala National Forest (29°N), (iv) Big Cypress National Preserve (25–26°N) and (v) Everglades National Park (25°N). These sites comprise southern hardwoods (Blackwater River State Forest and Apalachicola National Forest in the Panhandle), temperate broadleaf evergreens (Ocala National Forest, in the central part of the peninsula) and subtropical forests (Big Cypress National Preserve and Everglades National Park, in the southern tip). In the Big Cypress and Everglades National Parks tropical hardwood hammocks predominate.

Collection methods.—Submerged woody and herbaceous debris was collected randomly from lotic and lentic habitats four times over 2 y at the five collection sites along the Florida Peninsula. Samples were placed in zippered plastic bags containing moist paper towels and transported to the lab in an insulated cooler containing ice to control heat build-up and biological activity. In the lab substrates were gently rinsed with tap water and incubated in plastic storage boxes with moistened paper towels at ambient temperatures (about 24 C) under 12/12 h (light/dark) conditions. Water temperature, pH and latitude and longitude were measured and recorded in the field and are presented in the specimen citations.

Samples were examined with a dissecting microscope within 1 wk of collection and periodically over 6–12 mo. Collectors' names are abbreviated as: CB (Christopher Brown), JLC (J. L. Crane), ANM (Andrew N. Miller), HAR (Huzefa A. Raja), KR (Kevin Robertson), CAS (Carol A. Shearer).

Morphological study.—Ascomata were placed on a large (25 mm) cover slip on a microscope slide and opened in a drop of distilled water with fine dissecting needles and covered with a second smaller (18 mm) cover slip for microscopic examination. Material mounted on slides was preserved with glycerin (100%) or lactic acid (85%) containing azure A using the double cover glass method (Volkmann-Kohlmeyer and Kohlmeyer 1996). Melzer's reagent (MEZ; 0.5 g iodine, 1.5 g IKI, 20 g chloral hydrate, 20 mL distilled water) and aqueous cotton blue was used to determine staining reactions of the ascus apical apparatus. India ink or aqueous nigrosin was added to water mounts to reveal gelatinous sheaths on or around ascospores as well as gelatinous material surrounding the paraphyses or pseudoparaphyses. Measurements were made of material mounted in water, glycerin (100%) or lactic acid containing azure A. When sufficient fruit bodies were available, single ascospore derived cultures were obtained and maintained on cornmeal agar (CMA; Difco, cornmeal agar) or peptone, yeast extract, glucose agar (PYG; 1.25 g Peptone, 1.25 g yeast extract, 3.00 g D-glucose, 18 g agar, 1000 mL distilled water) according to the procedures of Fallah and Shearer (2001) and Shearer et al (2004).

Ascomata were fixed and sectioned with a modification of Huhndorf's technique (Huhndorf 1991, Fallah and Shearer 2001). Holotype slides were deposited in the Herbarium of the University of Illinois at Urbana-Champaign (ILL). Digital micrographs were obtained with an Insight Spot RT digital camera using an Olympus microscope equipped with Nomarski interference and phase optics. Digital images

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were edited with Adobe Photoshop CS2 and assembled with Adobe Indesign CS2.

TAXONOMY

New Taxa

Aniptodera megaloascocarpa Raja et Shearer sp. nov. FIGS. 1–10

Ascomata in ligno 1060–1360 × 430–530 µm, dispersa, immersa, hyalina, membranacea; venter giganteus, globosus, 490–650 × 485–580 µm. Rostrum 540–850 × 70–80 µm longum, cylindricum, periphysatum. Peridium 2–4 stratis, cellulae hyalinae, angulosae. Cellulae e textura epidermoidea composita. Catenophyses hyalinae, septatae, constes ex cellulis elongatis. Asci 100–128 × 20–26 µm, ad apicem applanati, tenuitunicati, clavati, unitunicati, persistes. Porus non-amyloideus. Ascosporae 20–26 × 9–11 µm, ellipsoideae, uniseptatae, hyalinae, sub-brunneae vel fuscae ad maturitates. Tunica gelatinosa, ad apicem.

Ascomata on wood 1060–1360 × 430–530 µm, scattered, immersed, hyaline membranous; venter large, globose, 490–650 × 485–580 µm, with a long, central, hyaline neck protruding through the surface of the wood when venter immersed (FIG. 1). Neck 540–850 × 70–80 µm, cylindrical, periphysate (FIG. 2). Peridium composed of 2–4 layers of hyaline angular cells; cells 10–15 × 2–3 µm, of textura epidermoidea in surface view (FIG. 3). Catenophyses sparse, hyaline, septate, consisting of elongated cells, slightly constricted at the septa, ca. 70–100 × 5–10 µm (FIG. 4). Asci 100–128 × 20–26 µm, (mean = 116 × 24 µm, n = 30), thin-walled, clavate, flattened at apex, tapering to a pointed pedicel, unitunicate, persistent, (FIGS. 5, 6), wall thickened at the apex, ascus apex Melzer's negative (MEZ−), staining blue in aqueous nigrosin, cytoplasm retracted below the ascus apex (FIGS. 5–7); empty asci show an apical pore where the ascospores have been discharged (FIG. 7); containing eight irregularly arranged ascospores. Ascospores 20–26 × 9–11 µm (mean = 23 × 10 µm, n = 60), 1-septate, broadly ellipsoidal, thin-walled, with single large guttule in each cell, surrounded by an irregular gelatinous sheath ca. 2–5 µm wide at the ascospore apex; sheath staining in aqueous nigrosin (FIGS. 8, 9), hyaline when young becoming pale brown to dark brown with age; septum becoming thicker and pigmented brown in older ascospores (FIG. 10).

Etymology. Greek: *megalo* = large; Latin: *ascocarpus* = fruit body of an ascomycete, in reference to the large fruit body compared to those of other described species of *Aniptodera*.

TYPE. USA. FLORIDA: Ocala National Forest, Little Lake Kerr, 29°20′57″N, 81°43′49″W, water 21 C, pH 7, on submerged decorticated woody debris, 2 Feb 2006, *HAR* and *JLC*, *F91-1*. (HOLOTYPE designated here, ILL40109).

Known distribution. USA (FL).

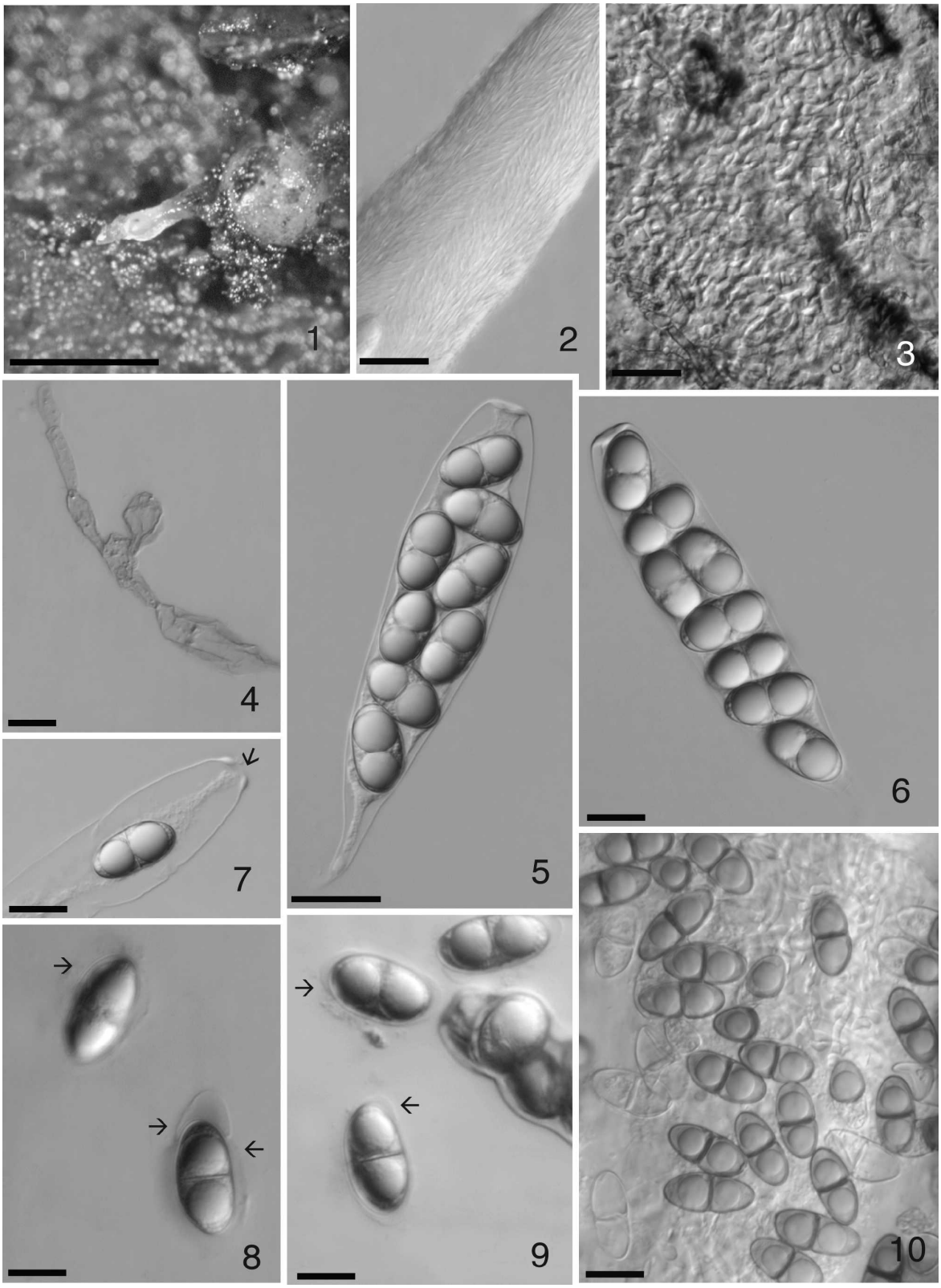
Comments. The genus *Aniptodera* Shearer and Miller is characterized as having membranous, light colored ascomata, presence of catenophyses, apically thickened persistent asci with a distinct pore and subapical retraction of cytoplasm, and hyaline, 1-septate ascospores without appendages (Shearer and Miller 1977). *Aniptodera* species with ascospore appendages were reported later in the literature (Shearer and Crane 1980a, Shearer 1989, Hyde 1992a, Hyde et al 1999, Campbell et al 2003). *Aniptodera megaloascocarpa* fits well within the concept of the genus with respect to ascomal, ascus and ascospore morphology (FIGS. 1, 5, 6, 8). *Aniptodera megaloascocarpa* differs distinctly from other species in the genus because it has the largest ascomata (1060–1360 × 430–530 µm) (FIG. 1). In addition ascospores are surrounded by an irregular mucilaginous sheath (FIGS. 8, 9). The mucilaginous sheath however cannot be seen when the ascospores are fixed in glycerin or lactic acid. Older ascospores of *A. megaloascocarpa* are pale brown and the septum is darkened (FIG. 10); these characters are not observed in other species of *Aniptodera* (Shearer 1989, Volkmann-Kohlmeyer and Kohlmeyer 1994, Hyde et al 1999).

Aniptodera megaloascocarpa is similar to *Phaeonectriella lignicola* R.A. Eaton & E.B.G. Jones (Eaton and Jones 1970), which is characterized by having hyaline to pale brown globose ascomata with a long periphysate neck, clavate asci and hyaline ascospores with germ pores that become gray to brown at maturity. *Aniptodera megaloascocarpa* however differs from *P. lignicola* in having a larger ascoma and in ascospore morphology. Ascospores of *P. lignicola* possess a germ pore at each end, a feature not seen in ascospores of *A. megaloascocarpa*. In addition Hyde et al (1999) reported polar appendages in *P. lignicola* from specimens collected in Mauritius but ascospores of *A. megaloascocarpa* do not have apical appendages.

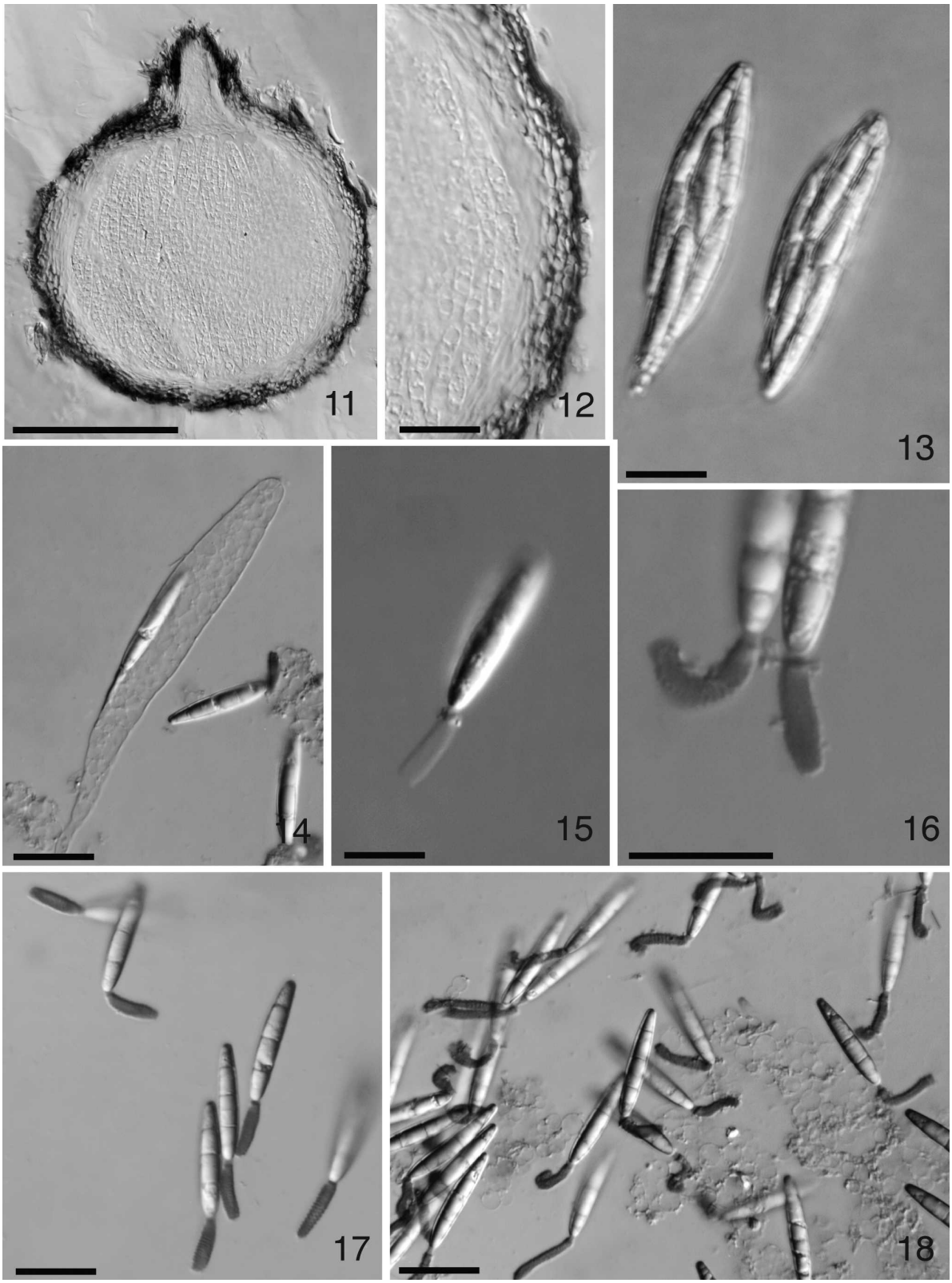
Flammispora pulchra Raja et Shearer sp. nov. FIGS. 11–18

Ascomata in ligno 200–210 × 184–188 µm, solitara, dispersa, immersa, nigra, rostro brevi. Rostrum 50 × 20 µm, conicum, aperiphysatum. Peridium 12–20 µm latum, stratis 4–5 cellularum in sectione longitudinali. Strata interna peridii cellulis hyalinis, elongatis. Strata externa cellulis bruneo, pseudoparenchymato. Physes absentes. Asci 52–107 × 12–18 µm, cylindro-clavati, unitunicati, brevi pedicellati, octospori, deliquescentes. Ascosporae 25–30 × 4–5 µm, tetra-seriatae, angustae, hyalinae, tri-septatae, praeditae appendicibus ad basim.

Ascomata on wood 200–210 × 184–188 µm, solitary, scattered, superficial or partially immersed, black, ostiolate with a small neck (FIG. 11). Neck 50



FIGS. 1–10. *Aniptodera megaloascocarpa* from the holotype. 1. Hyaline, membranous, ascomata on wood, note discharged ascospores around the neck. 2. Periphysate neck. 3. Peridium of *textura epidermoidea* in surface view. 4. Catenophyses. 5. Ascus showing retraction of cytoplasm at apex. 6. Ascus showing thickened wall of the apex. 7. Ascus containing one ascospore, arrow indicates ascus apical pore and apical thickening. 8, 9. Ascospores showing irregular gelatinous sheath. 10. Mature pale brown ascospores with thickened and darkened septum. Bars: 1 = 1 mm, 2–10 = 20 µm.



FIGS. 11–18. *Flammispora pulchra* from the holotype. 11. Longitudinal section through ascoma. 12. Peridium. 13. Asci. 14. Thin-walled nearly empty ascus. 15. Unstained ascospore showing single appendage, note the rod shaped structure at the base of appendage. 16–18. Three-septate ascospores stained in aqueous nigrosin showing single basal appendage, note the remains of deliquenscent asci in 18. Bars: 11 = 50 μm, 12–18 = 20 μm.

× 20 μm, conical, periphyses not seen (FIG. 11). Peridium ca. 12–20 μm wide, in longitudinal section 4–5 cell layers wide; inner layers of hyaline elongated cells, outer layers of pseudoparenchymatic cells with brown walls (FIG. 12). Physes not seen. Asci 52–107 × 12–18 μm, clavate or fusiform, with a short stalk, thin-walled (FIG. 13), separating from the ascogenous hyphae, deliquescent, with eight tetraseriate or irregularly arranged ascospores (FIGS. 13, 14). Ascospores 25–30 × 4–5 μm (mean = 28 × 4 μm, n = 30), narrow, fusiform or cylindrical, hyaline, 3-septate, sometimes 5-septate (FIGS. 15, 16), with a flame shaped appendage protruding through a rod shaped structure 2–3 μm wide at the base of the ascospore (FIGS. 15, 16); appendage 10–15 μm long, ca. 2–3 μm wide, staining in aqueous nigrosin (FIGS. 17, 18).

Etymology. From Latin feminine of *pulcher* = beautiful, referring to the ascospores.

TYPE. USA. FLORIDA: Ocala National Forest, Wildcat Lake, 29°10′14″N, 81°37′40″W, water 35 C, pH 6.1, on submerged decorticated woody debris, 15 Jul 2004, *HAR* and *CB*, *F62-1*. (HOLOTYPE designated here, ILL40119).

Known distribution. USA (FL).

Comments. The genus *Flammispora*, typified by *F. bioteca* U. Pinruan et al, was found originally on submerged leaves of the peat swamp palm, *Licuala longecalycata* Furt. in Thailand (Pinruan et al 2004). This genus is characterized by immersed to semi-immersed, coriaceous ascomata; paraphyses absent; 8-spored, clavate to cylindroclavate, deliquescent asci; and hyaline, 5-septate ascospores with a flame shaped basal gelatinous appendage.

The new species from Florida was found on submerged decorticated woody debris in a lake in Ocala National Forest. The Florida material agrees in all respects with the protolog of *F. bioteca*, the type species of the genus, but differs as follows. The ascomata are immersed to semi-immersed as in the protolog, but medial longitudinal sections through the ascoma revealed a small papilla (FIG. 11), a feature not present in *F. bioteca*. The ascospores of the new species are smaller (25–30 × 4–5 μm) and mostly 3-septate (FIGS. 17, 18), versus the larger (47.5–55 × 5–6.5 μm), 5-septate ascospores of *F. bioteca* (Pinruan et al 2004). *Flammispora pulchra* also differs from *F. bioteca* in substrate (*F. pulchra* was found on woody debris, while *F. bioteca* was found on herbaceous debris) and habitat (*F. pulchra* occurred in a lake, but *F. bioteca* was reported from a peat swamp).

Hanliniomyces Raja et Shearer gen. nov.

Ascomata dispersa, partim immersa vel superficiales, ostiolata, venteribus globosis vel subglobosis, membranacea, atri, collo. Collum cylindricum, atrum ad basem, pallidum versus ad apicem, periphysatum. Peridium e cellulis pseudoparenchymaticis composita, textura angularis. Paraphyses ad basem, angustatae ad apicem septatae, leniter constrictae. Asci anguste fusoidei, stipitati vel sessiles, unitunicati, nonamyloidei ad apices, ascosporis octo uniseriatis vel biseriatis, imbricatis. Apparatibus apicalibus refractivis. Ascosporae ellipsoideae, aseptatae, multiguttulatae ubi juvenes, ad maturitates 3-septatae, brunneae, e vagina irregulari gelatinosa circumcincta.

Etymology. Named in honor of Dr Richard T. Hanlin for his outstanding contributions to ascomycete systematics.

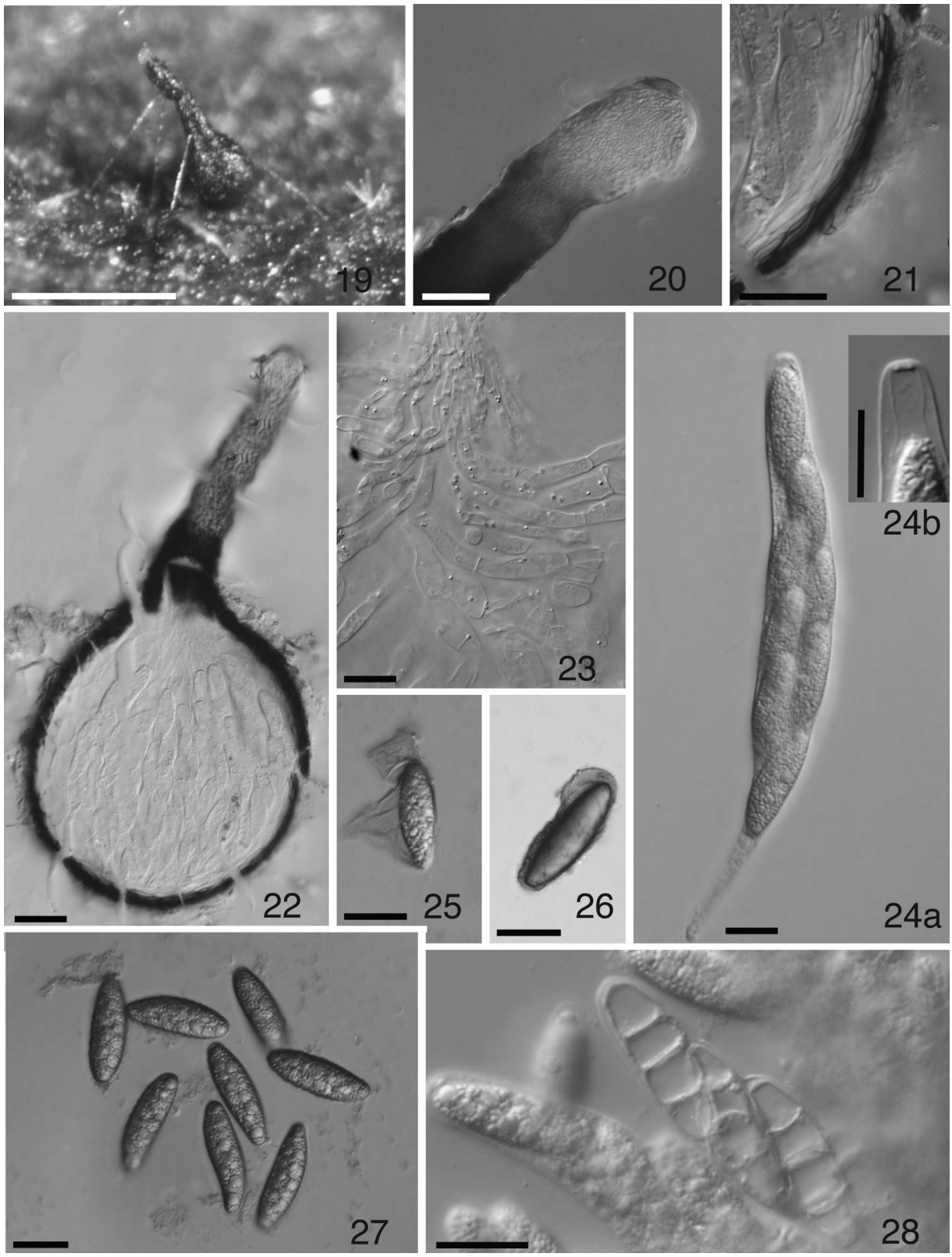
Ascomata scattered, partially immersed to superficial, black, ostiolate, with a cylindrical, periphysate neck; venter globose to subglobose, membranous. Peridium composed of pseudoparenchymatic cells, of textura angularis in surface view. Paraphyses wide at the base, tapering toward the apex, septate, slightly constricted at the septa. Asci unitunicate, narrowly fusoid, with or without a stalk, with a refractive, nonamyloid apical apparatus, containing eight uniseriate to overlapping biseriate ascospores. Ascospores ellipsoidal, aseptate and hyaline when young, becoming 3-septate and brown with age, surrounded by a gelatinous sheath.

Type species. *Hanliniomyces hyaloapicalis*

Hanliniomyces hyaloapicalis Raja et Shearer sp. nov. FIGS. 19–28

Ascomata 340–380 × 160–190 μm, dispersa, partim immersa vel superficiales, ostiolata, venteribus globosis vel subglobosis, 115–160 × 100–130 μm, membranacea, atri, collo. Collum 125–160 × 25–30 μm, cylindricum, atrum ad basem, pallidum versus ad apicem, periphysatum. Peridium 8–12 μm latum, e cellulis 4 vel 5 stratis pseudoparenchymaticis composita, textura angularis aspectu externo. Paraphyses 120–150 × 5–10 μm, latae ad basem, angustatae ad 2–3 μm latae; ad apicem septatae, leniter constrictae. Asci 130–160 × 18–20 μm, anguste fusoidei, stipitati vel sessiles, unitunicati, nonamyloidei ad apices, ascosporis octo uniseriatis vel biseriatis, imbricatis. Apparatibus apicalibus refractivis. Ascosporae 28–36 × 9–12 μm, (moda = 32 × 10 μm, n = 40), ellipsoideae, aseptatae, multiguttulatae ubi juvenes, ad maturitates 3-septatae, brunneae, e vagina irregulari gelatinosa circumcincta.

Ascomata 340–380 × 160–190 μm, scattered, partially immersed to superficial, venter globose to subglobose, 115–160 × 100–130 μm, membranous, black, ostiolate, with a neck (FIGS. 19, 22). Neck 125–160 × 25–30 μm, cylindrical, black at the base, lighter toward the apex, periphysate (FIG. 20). Peridium 8–12 μm wide, composed of 4 or 5 layers of pseudoparenchymatic cells, textura angularis in surface view, amorphous, dark material deposited on the outer cell walls, inner layer of hyaline, elongated and compressed cells, ca. 10–15 × 2–4 μm wide (FIG. 21).



FIGS. 19–28. *Hanliniomyces hyaloapicalis* from the holotype. 19. Ascomata on wood. 20. Neck showing hyaline apex. 21. Peridium. 22. Longitudinal section through ascoma. 23. Paraphyses. 24a. Ascus. 24b. Ascus apical apparatus. 25, 26. Ascospores stained in aqueous nigrosin showing gelatinous sheath. 27. Discharged multiguttulate ascospores. 28. Old 3-septate ascospores. Bars: 19 = 500 µm, 20–28 = 20 µm.

Paraphyses 120–150 μm long, 5–10 μm wide at the base, tapering to ca. 2–3 μm wide at the apex, septate, slightly constricted at the septa (FIG. 23). Asci 130–160 × 18–20 μm, unitunicate, narrowly fusoid, with or without a stalk, ascus apical ring MEZ negative, staining blue in aqueous nigrosin, with eight uniseriate to biseriate overlapping ascospores (FIG. 24a, b). Ascospores 28–36 × 9–12 μm, (mean = 32 × 10 μm, n = 40), ellipsoidal, aseptate, multiguttulate and hyaline when young, becoming 3-septate and brown with age, surrounded by an irregular gelatinous sheath that stains in aqueous nigrosin (FIGS. 25–28).

Colonies on PYG agar grown at 25 C, dark gray-black, effuse. Aerial mycelium hyaline to black, septate. Immersed mycelium composed of branched, septate, dark hyphae. No anamorph observed.

Etymology. From Latin *hyalo* = hyaline, and *apicalis* = apical, referring to the hyaline apex of the ascomal neck.

TYPE. USA. FLORIDA: Big Cypress National Preserve, Cypress Swamp Loop Road 5, 25°45′45″N, 80°55′09″W, water 30 C, pH 6, on submerged partially decorticated woody debris, 13 Jul 2006, *HAR* and *JLC*, *F37-3*. (HOLOTYPE designated here, ILL40116).

Additional specimens examined. Blackwater River State Forest, Penny Creek, 30°45′05″N, 86°46′54″W, water 23 C, pH 6.6, on submerged decorticated woody debris, 10 Jul 2004, *HAR* and *CB*, *F37-1*; Blackwater River, north end at Kennedy Bridge, 30°56′01″N, 86°44′07″W, water 11 C, pH 5.5, on submerged decorticated woody debris, 11 Feb 2006, *HAR* and *JLC*, *F37-2*.

Known distribution. USA (FL).

Comments. *Hanliniomyces* is similar to *Lentomitella* Höhnel (Höhnel 1905, Réblová 2006) in the morphology of the ascomata, peridium and paraphyses and in the shape of the ascospores. In addition asci of both *Hanliniomyces* and *Lentomitella* are similar in that they possess a refractive nonamyloid apical ring. However, despite these similarities, *Hanliniomyces* differs from *Lentomitella* in that it has a black ascomal neck with a hyaline apex rather than an entirely black neck and narrowly fusioid rather than cylindrical-clavate asci. The young ascospores of *Hanliniomyces* are smooth-walled, aseptate and surrounded by a gelatinous sheath but become brown and septate at maturity, whereas ascospores of *Lentomitella* are longitudinally striated, aseptate or septate, do not possess a gelatinous sheath and are hyaline throughout their development. In addition the genus *Lentomitella* is associated with a *Phaeoisaria*-like anamorph, but no anamorph was observed in culture for *Hanliniomyces*.

Hanliniomyces also is morphologically closely related to X*ylomelasma* Réblová (Réblová 2006) in having dematiaceous hyphae in culture; dark, nonstromatic perithecial ascomata with a cylindrical neck, a two-layered peridial wall with dark cells on the outside and hyaline cells on the inside; paraphyses that are broad at the base and taper toward the apex; asci that have a refractive nonamyloid apical annulus; and ellipsoidal ascospores that are hyaline and aseptate when young but become brown and septate at maturity. *Hanliniomyces* however differs from *Xylomelasma* in having an ascomal neck with a hyaline apex compared to a dark neck and a peridial wall of textura angularis as opposed to textura prismatica. *Hanliniomyces* has a narrowly fusoid ascus with a long stalk, but *Xylomelasma* has a cylindrical ascus with a short stalk. In addition a gelatinous sheath surrounds the ascospores of *Hanliniomyces* but an ascospore sheath does not occur in *Xylomelasma*.

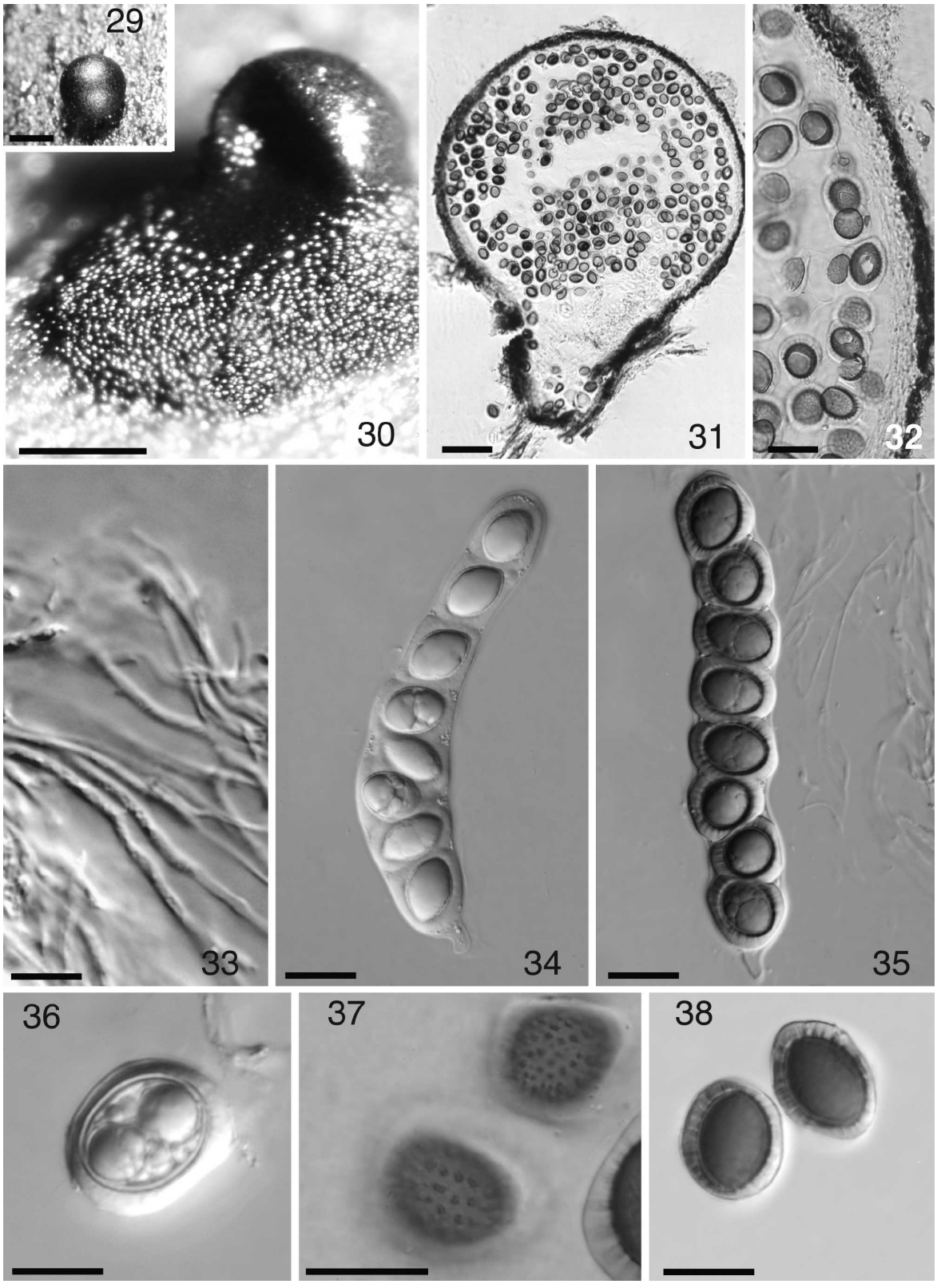
Furthermore *Hanliniomyces* differs from both *Lentomitella* and *Xylomelasma* in its habitat. *Lentomitella* and *Xylomelasma* species have been reported from wood in terrestrial habitats, whereas *Hanliniomyces* was collected on submerged woody debris from lotic and lentic habitats.

Hanliniomyces hyaloapicalis also is similar to *Physalospora citogerminans* Kohlm., Volkm.-Kohlm. & O.E. Erikss. (Kohlmeyer et al 1995) with respect to ascomal, ascus and ascospore morphology. In addition older ascospores of both *H. hyaloapicalis* and *P. citogerminans* become 3-septate. Ascospores of the genus *Physalospora* usually are not 3-septate, and this feature has not been reported in previously described species of *Physalospora* (von Arx and Müller 1954, Barr 1970, Scheuer 1988, Nograsek 1990). *Hanliniomyces hyaloapicalis*, however differs from *P. citogerminans* in having a dark neck with a hyaline apex and ascospores that do not germinate on contact with freshwater. The two species also differ in their ascomal and ascus dimensions, which suggests that they are not conspecific. In addition the two species differ in the type of habitat in which they occur. *Hanliniomyces hyaloapicalis* occurred on submerged woody debris in freshwater habitats in Florida, whereas *P. citogerminans* has been reported from *Juncus roemerianus* Scheele in a salt marsh in North Carolina (Kohlmeyer et al 1995). It is plausible that *P. citogerminans* could be transferred to the genus *Hanliniomyces* in the future; however molecular data would be useful to confirm the relationship of the two taxa.

We establish *Hanliniomyces* as a new genus closely allied to *Lentomitella* and *Xylomelasma*. Using nuclear large ribosomal RNA subunit gene sequence data Réblová (2006) placed *Lentomitella, and Xylomelasma* in the Sordariomycetes incert. sed. We also place *Hanliniomyces* provisionally within the Sordariomycetes incert. sed. pending phylogenetic analyses.

Lockerbia striata Raja et Shearer sp. nov. FIGS. 29–38

Ascomata 500–620 × 400–480 μm, cleistothecialia, glo-



FIGS. 29–38. *Lockerbia striata* from the holotype. 29, 30. Cleistothecial ascoma on wood, note disintegrating peridium in 30. 31. Longitudinal section through ascoma. 32. Peridium. 33. Paraphyses. 34. Young ascus. 35. Mature ascus with a short pedicel. 36. Young, hyaline ascospore in water showing sheath. 37. Ascospores showing warted wall and striated sheath. 38. Mature ascospores showing striated gelatinous sheath. Bars: 29–30 = 500 µm, 31–38 = 20 µm.

bosa vel subglobosa, brunnea, membranacea, pariete ascomatis lavi, superficialia, basi immerso substrato. Peridium 15–20 μm crassum, areolatum aspectu superficiale, bistratum. Paraphyses abundantes, racemosae, septatae, hyalinae. Asci 140–170 × 20–24 μm, unitunicati, cylindrici, brevipedunculis contracto, octo uniseriati ascosporis. Ascosporae 20–24 × 14–18 μm, unicellularis, subglobosae, obovatae vel ovales, hyalinae ubi immaturae, brunneae ad maturitates, sine poro germinali vel fissura, vagina hyalinis, gelatinosis, striatis, circumcinctae.

Ascomata ca. 500–620 × 400–480 μm, scattered, globose to subglobose, tapering to a stalk at the base, superficial with base immersed in the substrate, cleistothecial, dark brown to black, membranous, rupturing and wearing away at maturity (FIGS. 29, 30, 31). Peridium ca. 15–20 μm wide, areolate in surface view, in medial longitudinal section composed of two layers, an outer dark brown amorphous region and an inner hyaline region of short angular cells (FIG. 32). Paraphyses abundant, simple or branched, narrow, ca. 2 μm wide, filiform, septate, hyaline, immersed in gel matrix (FIG. 33). Asci 140–170 × 20–24 μm, unitunicate, cylindrical, with a narrow short pedicel, with eight uniseriate ascospores (FIGS. 34, 35). Ascospores 20–24 × 14–18 μm, 1-celled, subglobose, obovate or oval, hyaline when young (FIG. 36) becoming dark brown with age, without germ pores or slits, ascospore wall thinner at apices, warted (FIG. 37), surrounded by a hyaline, gelatinous sheath ca. 3–5 μm wide; sheath not staining in aqueous nigrosin or India ink striated; striations radiating from ascospore wall (FIGS. 35, 38).

Etymology. From Latin *striatus* meaning marked with striae, referring to the appearance of the ascospore sheath.

TYPE. USA. FLORIDA: Blackwater River State Forest, Horns Creek Swamp, 30°46′31″N, 86°54′43″W, water 30 C, pH 6, on submerged partially decorticated woody debris, 10 Jul 2004, *HAR* and *CB*, *F24-1* (HOLOTYPE designated here, ILL40118).

Additional specimens examined. USA. FLORIDA: Apalachicola National Forest, Andrew Lake near Silver Lake Recreation Area, 30°24′09″N, 84°24′27″W, water 33 C, pH 6.5, on submerged herbaceous debris, 13 Jul 2004, *HAR* and *CB*, *F24-2*. MISSISSIPPI: Big Black River at Mississippi 19, east of West, Holmes County, 33°11′38″N, 89°46′14″W, water 7 C, pH 5.5, on submerged decorticated woody debris, 27 Dec 1993, *JLC* and *CAS*, *A247-1*; Edge of Wolf River at I-10 in Harrison County, 30°21′25″N, 89°17′12″W, water 24 C, pH 6.5, on submerged decorticated woody debris, 16 Jun 1997, *KR*, *A247-2*.

Known distribution. USA (FL, MS).

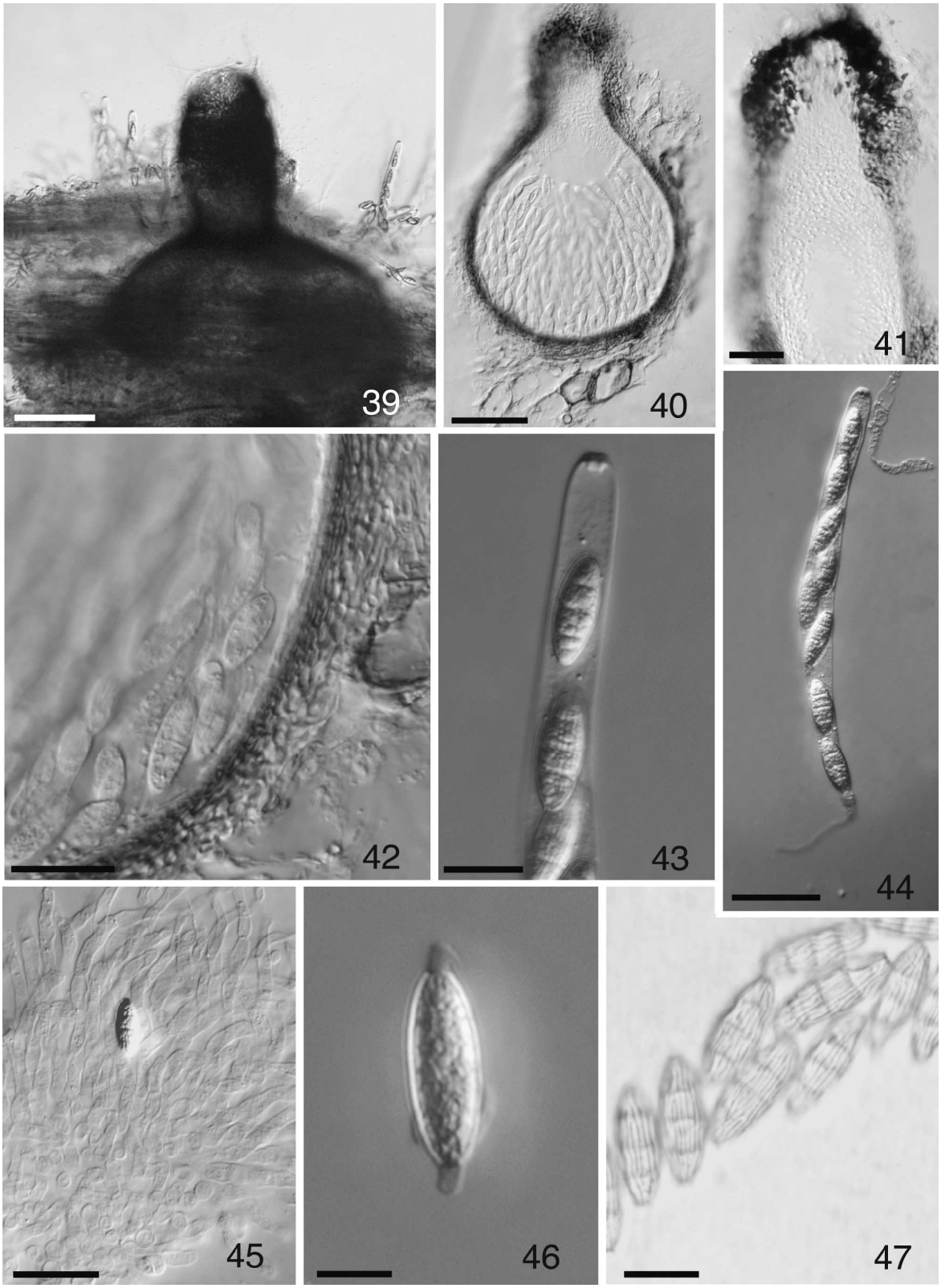
Comments. The genus *Lockerbia* K.D. Hyde, typified by *L. palmicola* K.D. Hyde, originally was found on a terrestrial palm rachis in a rainforest in Queensland, Australia (Hyde 1993). *Lockerbia* is characterized by superficial, cleistothecial, membranous, thin-walled ascomata; 8-spored, apically rounded, cylindrical, unitunicate asci; and brown, limoniform or oval, thick-walled, minutely pitted ascospores with a hyaline sheath.

We collected *L. striata* from submerged decorticated woody debris in a creek as well as on an unidentified piece of herbaceous debris in a lake. During a geographically broader survey of freshwater ascomycetes, we also found *L. striata* on submerged decorticated wood in two lotic habitats in Mississippi. *Lockerbia striata* agrees in most respects with the protolog of *Lockerbia* but differs from the type species, *L. palmicola*, in these ways: the ascomata are large and cleistothecial (FIGS. 29, 30), as in the original protolog, but the medial longitudinal section through the ascomata of *L. striata* reveals a stalk-like structure (FIG. 31) that anchors the base of the cleistothecium to its substrate much like a puffball. This structure was not reported for *L. palmicola*. The ascospores of *L. striata* are warted (FIGS. 32, 37), whereas those of in *L. palmicola* are minutely pitted. In addition the ascospores of *L. striata* are surrounded by a hyaline, striated, gelatinous sheath ca. 3–5 μm wide and having a definite form (FIGS. 35, 37, 38), while those of *L. palmicola* are surrounded by a hyaline mucilaginous sheath (see FIGS. 9–11 in Hyde 1993) lacking a definite form and striations.

Phomatospora triseptata Raja et Shearer sp. nov.
FIGS. 39–47

Ascomata in ligno 290–400 × 190–340 μm, subglobosa, dispersa vel gregaria, immersa in substrato, membranacea, brunnea, osteolata, collo. Collum 180–230 × 60–70 μm, centrale, cylindricum, periphysibus, hyalinum, filiforme, materio atro amorpho circum sectione superno. Peridium 15–20 μm latum, stratis aliquot cellulae hyalinae vel brunneae, pseudoparenchymaticae; cellulae atratiores versus internis, pellucidiores versus externis. Paraphyses numerosae, filamentosae, septatae, ramosae, ca. 3–4 μm latae. Asci 134–164 × 10–12 μm, unitunicati, cylindrici, pedicellati, tenuitunicati, annulo parvo, refracto, apicali, gossypini-caerulei ope tincti. Ascus elongatus aqua, ascosporis octo, imbricatis, uniseriatis. Ascosporae 18–20 × 7–8 μm (modus = 19 × 7 μm, n = 30), ellipsoidea, apicibus applanatis, 3-septatae, striata longitudinales, galeris bipolaribus gelatinus ca. 3–7 μm longis. Medius et basidimidius ascosporae circumcinctus vagina gelatinosa.

Ascomata on wood 290–400 × 190–340 μm, subglobose, scattered to gregarious, immersed in the substrate, membranous, brown, ostiolate, with a neck (FIGS. 39, 40). Neck 180–230 × 60–70 μm, central, cylindrical, covered with black, amorphous material around the upper region (FIG. 41), periphysate; periphyses hyaline, short, filiform. Peridium 15–20 μm wide, composed of 5–10 layers of hyaline to brown pseudoparenchymatic cells; cells darker toward



FIGS. 39–47. *Phomatospora triseptata* from the holotype. 39. Ascomata immersed in wood. 40. Medial longitudinal section through ascoma. 41. Periphysate neck, note darkened area composed of amorphous material around the apex of the neck. 42. Peridium. 43. Ascus with apical ring, in water. 44. Ascus with overlapping uniseriate ascospores. 45. Paraphyses. 46. Ascospore with filamentous apical appendages, and narrow gelatinous sheath, in water. 47. Ascospores fixed in glycerin showing longitudinal striations. Bars: 39 = 50 µm, 40 = 100 µm, 41–47 = 20 µm.

the inside and lighter toward the outside (FIG. 42). Paraphyses numerous, filamentous, septate, broad at the base and tapering toward the apex, branched, ca. 3–4 μm wide (FIG. 45). Asci 134–164 × 10–12 μm, unitunicate, cylindrical, pedicellate, thin-walled, with a small refractive apical ring; apical ring staining blue in aqueous cotton blue, MEZ negative, elongating in water, with eight overlapping uniseriate ascospores (FIGS. 43, 44). Ascospores 18–20 × 7–8 μm (mean = 19 × 7 μm, n = 30), ellipsoidal with slightly flattened apices, 3-septate, with bipolar gelatinous caps ca. 3–7 μm long; appendages staining in aqueous nigrosin; the middle and lower half of the ascospore surrounded by a narrow gelatinous sheath (FIG. 46); wall longitudinally striate, appendages and sheath not visible in glycerin or lactic acid (FIG. 47).

Etymology. From Latin *triseptatus* = three septate, referring to the three septate ascospores.

TYPE. USA. FLORIDA: Big Cypress National Preserve, Cypress Swamp Loop Road 2, 25°45′36″N, 81°02′07″W, water 25 C, pH 8, on submerged soft, decorticated woody debris, 22 Mar 2005, *HAR*, *ANM* and *JLC*, *F67-1*. (HOLOTYPE designated here, ILL40114).

Known distribution. USA (FL).

Comments. Fallah and Shearer (1998) examined the type specimen of the genus *Phomatospora* Sacc., *P. berkeleyi* Sacc, and described it as having immersed, light brown, membranous ascomata, ascoma with a short periphysate beak, which is thick and dark around the ostiole; cylindrical asci with a small chitinoid refractive apical ring, which stains blue in cotton blue; and single-celled, hyaline, longitudinally striate ascospores with bipolar gelatinous caps. The specimen collected from Florida agrees in most respects with the type of *Phomatospora*. *Phomatospora triseptata* differs from the type and 20 other species of *Phomatospora* (Kirk et al 2001) in having an ascoma with a long, cylindrical neck (FIG. 39) and consistently 3-septate ascospores, which are slightly flattened at the apices and equipped with both bipolar gelatinous appendages as well as a gelatinous sheath (FIG. 46). *Phomatospora bellaminuta* Kohlm., Volkm.-Kohl,. & O.E. Erikss. (Kohlmeyer et al 1995) also has a long neck and bipolar appendages but differs from *P. triseptata* in having smaller, one-celled ascospores and no gelatinous sheath. In addition *P. bellaminuta* was isolated from a marine habitat on *Juncus roemarianus* Scheele, whereas *P. triseptata* was found on a piece of submerged wood in freshwater. Multiseptate ascospores occur in *P. radegundensis* Scheuer (Scheuer 1988) and *P. admontensis* Nograsek (Nograsek and Matzer 1991) but *P. triseptata* differs from these two species in ascospore morphology and ecological habitat.

Most species of *Phomatospora* have been reported as saprobic on herbaceous substrates, as well as on old ascomycete stromata (Barr 1994, Yuan and Mohammed 1997). Other than *P. triseptata*, only one other species, *P. aquatica* Minoura & T. Muroi, has been reported from wood. *Phomatospora aquatica* was described from balsa wood submerged in a pond in Japan (Minoura and Muroi 1978).

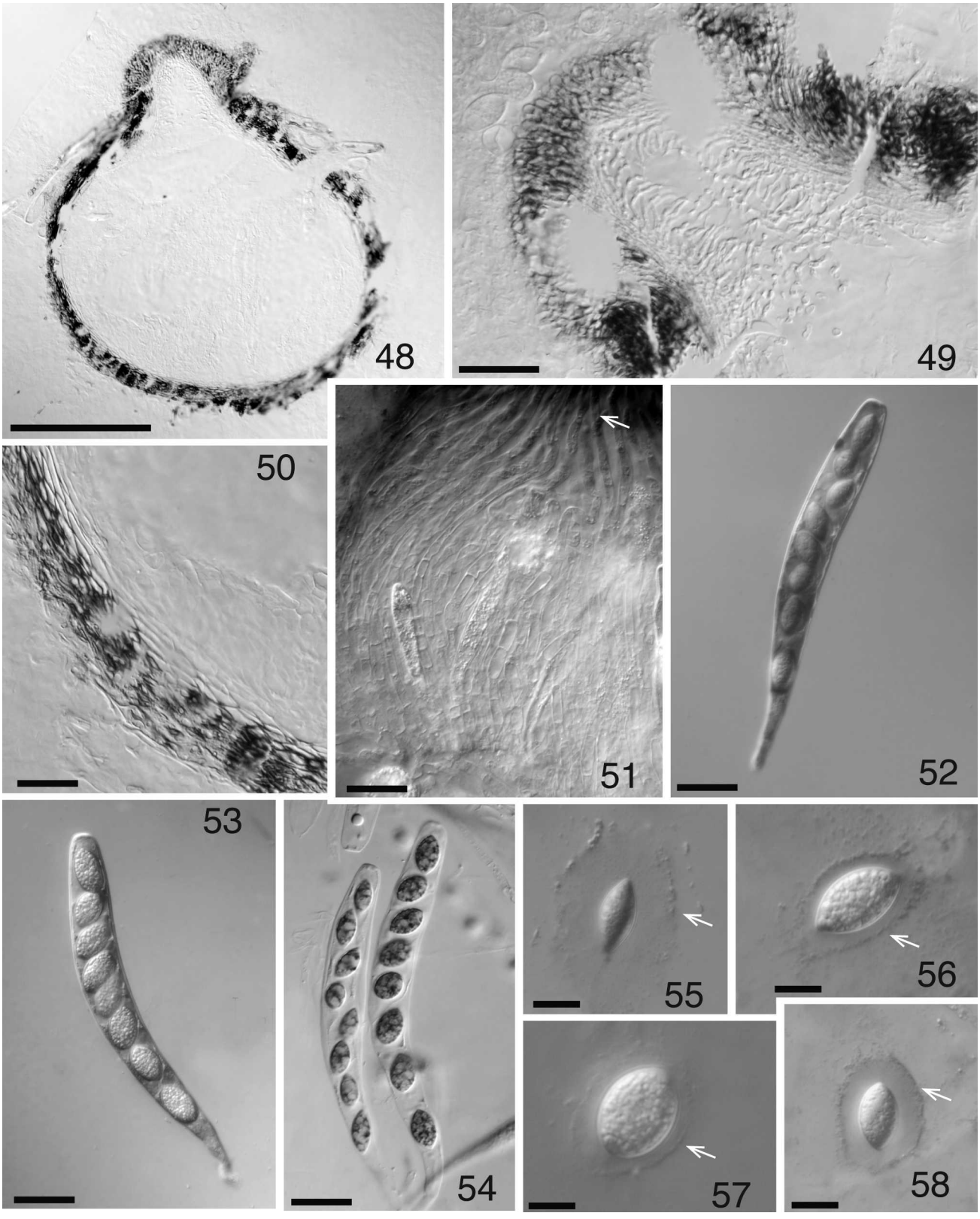
Phomatospora triseptata is the fifth species to be reported from a freshwater habitat. Four other species, *P. aquatica*, *P. berkeleyi*, *P. muskellungensis* Fallah and Shearer and *P. striatigera* Scheuer have been reported previously from freshwater (http://fungi.life.uiuc.edu/). *Phomatospora triseptata* was collected only once, and we were unable to isolate this species in axenic culture. Whether an anamorph is associated with this fungus remains unknown.

Physalospora limnetica Raja et Shearer sp. nov.

FIGS. 48–58

Ascomata 470–490 × 400–450 μm, dispersa, immersa vel partim immersa, vel erumpentes, ostiolata, globosa vel subglobosa, membranacea, fusca vel atra, papillata. Papillae 80–100 × 100–130 μm, conicae, periphysatae. Paries papillae e cellulis atris, extrinsecus divergentibus compositum. Periphyses tenuitunicatae, hyalinae, septatae. Peridium 20–30 μm latum, e 5-vel 6-stratis cellulis elongatis, pseudoparenchymatibus compositum. Strata papillarum fuscior versus exterior, diluta vel hyalina versus interior. Paraphyses simplices, septatae, latae ad basem, angustatae ad apicem, e matrice gelatinosa immersa. Asci 145–190 × 16–20 μm, cylindro-clavati, unitunicati, annulo bipartito, nonamyloideo, apicali. Ascus complanatus ad apicem, contractus in brevistipite ad basem, ascosporis octo, imbricatis. Ascosporae 18–22 × 9–14 μm, aseptatae, rhomboideae ubi juvenes ad maturitates subglobosae vel ovales, hyalinas, multiguttulatae, e vagina gelatinosa ca. 3–4 μm lata circumcincta. Vagina in aqua expansa ca. 10–15 μm.

Ascomata 470–490 × 400–450 μm, scattered, immersed to partially immersed, becoming erumpent, ostiolate, globose to subglobose, membranous, dark brown to black, papillate (FIG. 48). Papillae 80–100 × 100–130 μm, conical, wall of papillae composed of dark brown outwardly diverging hyphae, periphysate; periphyses thin-walled, hyaline, septate (FIG. 49). Peridium 20–30 μm wide, composed of five to six layers of elongated pseudoparenchymatic cells; dark toward the outside, pale brown to hyaline toward the inside; cells ca. 10–15 × 2–4 μm (FIG. 50). Paraphyses abundant, simple, septate, immersed in a gel matrix, broad at the base and narrowing toward the apex, extending above the asci, tips of the paraphyses staining blue in aqueous nigrosin (FIG. 51). Asci 145–190 × 16–20 μm, cylindro-clavate, unitunicate, with a small bipartite, nonamyloid apical ring that stains blue in aqueous nigrosin; ascus flattened at the apex,



FIGS. 48–58. *Physalospora limnetica* from the holotype. 48. Longitudinal section through ascoma. 49. Neck showing periphyses. 50. Peridium. 51. Paraphyses, broad at the base and tapering toward apex; note arrow showing tips of paraphyses stained with aqueous nigrosin. 52. Young ascus in water. 53. Mature ascus in water. 54. Asci fixed in glycerin. 55–58. Ascospores in various stages of development stained in aqueous nigrosin, note arrows showing gelatinous sheath that expands in water. Bars: 48 = 200 µm, 49–58 = 20 µm.

tapering to a short stalk at the base, with eight overlapping uniseriate ascospores (FIGS. 52, 53, 54). Ascospores 18–22 × 9–14 μm, aseptate, rhomboid when young, becoming subglobose to oval when mature, hyaline, multiguttulate, surrounded by a gelatinous sheath ca. 3–4 μm wide, staining blue in aqueous nigrosin, enlarging in water to ca. 10–15 μm wide (FIGS. 55–58).

Colonies on CMA hyaline, producing a pink pigment that diffuses into the surrounding agar. Aerial mycelium white, floccose, immersed hyphae creamy yellow; composed of branched, septate, hyaline hyphae.

Etymology. From Greek *limn* meaning standing water, referring to the habitat of the fungus.

TYPE. USA. FLORIDA: Apalachicola National Forest, Camel Pond, 30°16′36″N, 84°59′20″W, water 33 C, pH 5.5, on submerged herbaceous debris, 10 Jul 2006, *HAR* and *JLC, F108-1.* (HOLOTYPE designated here, ILL40115).

Known distribution. USA (FL).

Comments. *Physalospora* Niessl was established in 1876 and contains 30 species (Kirk et al 2001) that are found on living and dead leaves or sometimes on twigs of angiosperms (Hanlin 1990). The genus, based on *P. alpestris* Niessl, is characterized as having immersed ascomata with an erumpent, periphysate neck; unitunicate, cylindrical to clavate asci, which are broad in the middle; and unicellular, hyaline or pale, obovoid to ellipsoidal ascospores surrounded by a gelatinous sheath (von Arx and Müller 1954, Hanlin 1990, Fröhlich and Hyde 2000).

Physalospora limnetica from Florida fits well within the generic concept of *Physalospora*, subgenus *Physalospora* based on *P. alpestris* (*sensu* Barr 1970). *Physalospora limnetica* is distinguished from all other known species of *Physalospora* (von Arx and Müller 1954, Barr 1970, Scheuer 1988, Hsieh et al 1998, Taylor et al 2001, Sivanesan and Shivas 2002) by its larger ascomata (470–490 × 400–450 μm) and ascospore sheath that enlarges in water to about 10–15 μm wide. In addition *P. limnetica* was found on unidentified herbaceous debris in freshwater, whereas most other *Physalospora* species are reported from terrestrial habitats.

Physalospora limnetica differs from other freshwater species of *Physalospora*, such as *P. aquatica* Ingold (Ingold 1955, Magnes and Hafellner 1991), in having larger ascomata, asci and ascospores. *Physalospora limnetica* also differs from *Physalospora vaccinii* (Shear) von Arx & Müller included in *Physalospora* subgenus *Acanthorhynchus* (*sensu* Barr 1970) in ascomatal, ascus and ascospore morphology. This species originally was thought to be a terrestrial species, but it was reported and redescribed from a freshwater lake in Wisconsin (Fallah and Shearer 2001).

Caryospora obclavata Raja et Shearer sp. nov. (FIGS. 59–70)

Ascomata in ligno 290–320 × 290–340 μm, nigra, carbonacea, dispersa, immersa vel erumpentia, conica, ostiolata, papillata. Papillae 50 × 80 μm, brevis, late-cylindricae sine periphysibus. Peridium 40–45 μm latum, e textura angularis compositum, bistratum. Trabeculae ad apicem anastomosantes. Asci 115–160 × 32–50 μm, fissitunicati, obclavati, pedunculati, octospori. Ascosporae 30–40 × 11–15 μm, late, ellipsoideae vel biconicae, crystallinae, uniseptatae, constrictae sed perraro 1-septo prope apicibus, brunnae, circumcinctae tunica tenui, gelatinosa.

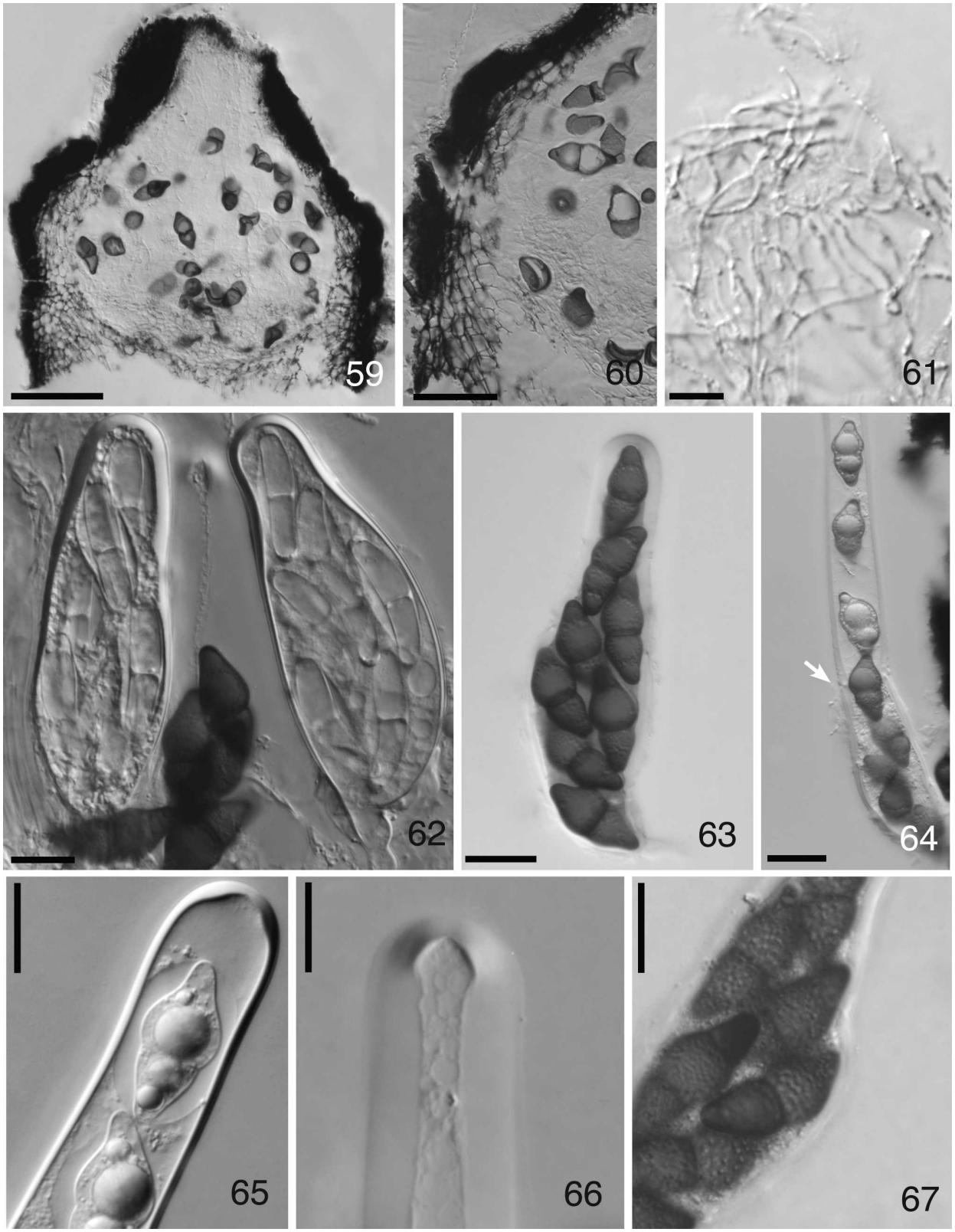
Ascomata on wood 290–320 × 290–340 μm, black, carbonaceous, scattered, partially immersed, erupting from the substrate, broadly conical, ostiolate, papillate (FIG. 59). Papilla 50 × 80 μm, short, broadly cylindrical; periphyses not seen. Peridium 40–45 μm thick; textura angularis in surface view; in longitudinal section composed of two layers; inner layer of pseudoparenchymatic cells 5–10 × 3–5 μm, outer layer of black carbonaceous material (FIG. 60). Pseudoparaphyses trabeculate, branched and anastomosing (FIG. 61). Asci 115–160 × 32–50 μm (mean = 140 × 39 μm, n = 25), basal, bitunicate, fissitunicate, obclavate and broadly rounded at the apex (FIGS. 62, 63), pedicellate, ectoascus splitting at the ascus apex, endoascus thick-walled (FIG. 64), with a broad ocular chamber 13 × 12 μm (FIGS. 65, 66), with eight ascospores, uniseriate at the apex and base and irregularly arranged in the middle (FIGS. 62, 63). Ascospores 30–40 × 11–15 μm (mean = 34.5 × 13 μm, n = 30), broadly ellipsoidal to biconic (FIGS. 68, 69), hyaline to pale green, becoming dark reddish brown at maturity, forming acicular crystals in glycerin or lactic acid, deeply constricted at the midseptum, slightly constricted below the apices; upper cell wider than basal cell, cells with one large guttule and several small ones; surface roughened (FIG. 67); surrounded by a thin gelatinous sheath (FIG. 69); sheath invisible in glycerin or lactic acid, 1-septate, but occasionally additional septa also seen near the tips of the ascospores (FIG. 70).

Etymology. From Latin *obclavatus* in reference to the shape of the ascus.

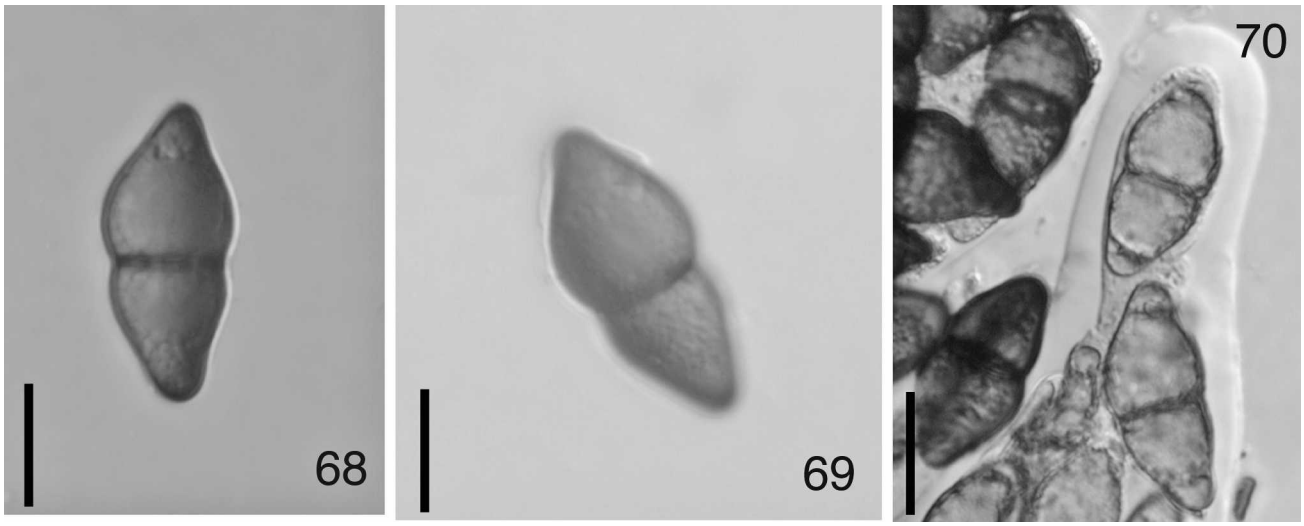
TYPE. USA. FLORIDA: Apalachicola National Forest, Whitehead Lake, 30°09′54″N, 84°40′30″W, water 29 C, pH 7.5, on submerged decorticated woody debris, 13 Jul 2004, *HAR* and *CB, F79-1.* (HOLOTYPE designated here, ILL40110).

Known distribution. USA (FL).

Comments. Following Barr (1979, 1990) our fungus from Florida belongs to the genus *Caryospora* de Not., typified by *Caryospora putaminum* (Schw. Ex Fr.) de Not. (Massariaceae Winter). This genus is characterized as having large, dull black, erumpent ascomata with a papilla; few oblong or inflated, 1-, 2- or 8-spored



FIGS. 59–70. *Caryospora obclavata* from the holotype. 59. Longitudinal section through ascoma. 60. Peridium. 61. Trabeculate pseudoparaphyses. 62. Young asci; note thick-walled apex. 63. Ascus with eight irregularly arranged ascospores. 64. Fissitunicate ascus, arrow indicates area where endoascus extends from ectoascus. 65. Endoascus showing apical chamber. 66. Endoascus with thickened apex showing ocular chamber. 67. Rough-walled ascospores. 68. Mature ascospore. 69. Ascospore showing thin gelatinous sheath. 70. Mature ascospores showing additional septa within endoascus. Bars: 59 = 50 µm, 60–70 = 20 µm.



FIGS. 59–70. Continued.

asci; rich reddish brown, rough walled, ellipsoidal ascospores usually constricted at the midseptum and surrounded by a narrow gelatinous sheath.

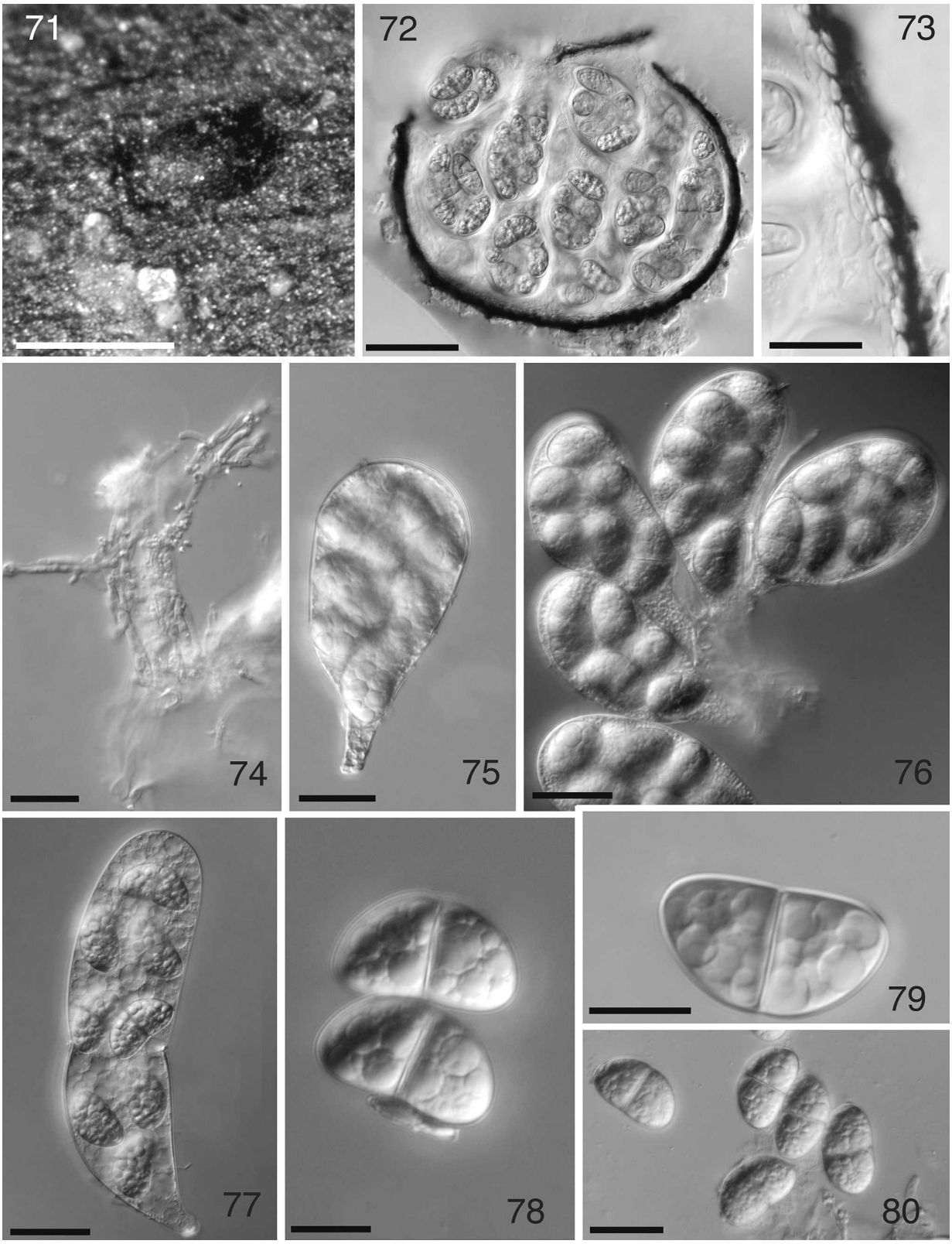
Caryospora obclavata differs from other species in the genus in having smaller ascomata, thick-walled, obclavate asci, as well as broadly ellipsoidal, biconic ascospores. Seven species currently are recognized in the genus *Caryospora* (Barr 1979, 1990; Hawksworth 1982; Abdel-Wahab and Jones 2000). The species from Florida is most similar to *C. putaminum* (Schw. ex. Fr.) de Not (Barr 1979) in that the ascospores of both species have a median septum and thin-walled secondary septa, with a constriction at the midseptum and rough walls. *Caryospora obclavata*, however, differs from *C. putaminum* in the number of ascospores per ascus, ascus shape and size, absence of a wide gelatinous sheath surrounding the ascospores, and the nature of its habitat. The asci of *C. obclavata* are narrow (32–50 μm) versus the wider asci (50–70 μm) of *C. putaminum* (Barr 1979). In addition, *C. obclavata* always has eight ascospores per asci (FIGS. 63, 64), whereas asci in *C. putaminum* usually have 1–4 ascospores per ascus (Jeffers 1940, Barr 1979). The ascospores of *C. obclavata* are surrounded by a narrow gelatinous sheath (FIG. 69), whereas a wide gelatinous sheath is reported for *C. putaminum* (Barr 1979). *Caryospora obclavata* was found on submerged woody debris from a Florida lake, whereas *C. putaminum* originally was collected from endocarps of *Prunus persica* L. (peach) in a terrestrial habitat (Jeffers 1940, Barr 1979).

Most species of *Caryospora* have been reported from a variety of substrates such as wood, stems and fruit endocarps in terrestrial habitats. *Caryospora obclavata* is the third species to be reported from a freshwater habitat. Two other *Caryospora* species, *C. minima* Jeffers and *C. callicarpa* (Currey) Nitschke ex Fuckel, also have been reported from freshwater (Cai et al 2006). These two species however originally were described from terrestrial habitats (Jeffers 1940, Currey 1859). Another species, *C. australiensis* Abdel-Wahab and E.B.G. Jones, was described from marine driftwood (Abdel-Wahab and Jones 2000).

Lepidopterella tangerina Raja et Shearer sp. nov.
(FIGS. 71–80)

Ascomata in ligno 120–190 × 230–300 μm, superficiales vel parti immersi, globosi vel subglobosi. Peridium 8–10 μm crassum, e textura angularis compositum, bistratosum. Pseudoparaphyses septatae, ramosae. Asci 78–130 × 40–58 μm, bitunicati, globosi vel subglobosi, brevi stipitati, octospori. Ascosporae papilioniformes vel reniformae, ad septum 16–19 μm longae, latitudine maxima 30–35 μm, uniseptatae, tangerine.

Ascomata on wood 120–190 × 230–300 μm, cleistothecial, scattered to gregarious, superficial to partially immersed, globose to subglobose, appearing as raised dome-shaped structures on the substrate (FIGS. 71, 72). Peridium 8–10 μm wide, textura angularis in surface view, in median longitudinal section composed of an outer dark brown amorphous layer and an inner hyaline region of 2–3 layers of elongated, thin-walled, angular cells (FIG. 73). Pseudoparaphyses sparse, hyaline, septate (FIG. 74), not covered in gel. Asci 78–130 × 40–58 μm, (mean = 106 × 50 μm, n = 20), produced successively on ascogenous hyphae, then separating from the ascogenous hyphae, globose to subglobose, thick-walled and broadly rounded at the apex, with a short pedicel, containing eight irregularly arranged ascospores (FIGS. 75, 76), fissitunicate, ectoascus rupturing at the apex to release the endoascus; endoascus extend-



FIGS. 71–80. *Lepidopterella tangerina* from the holotype. 71. Cleistothecial ascoma on wood. 72. Longitudinal section through an ascoma. 73. Two layered peridium. 74. Cellular pseudoparaphyses. 75. Ascus with a short pedicel. 76. Asci developing successively from the ascogenous hyphae. 77. Fissitunicate ascus, note granular cytoplasm of endoascus with granules delimiting spheres. 78–80. Mature multiguttulate ascospores. Bars: 71 = 500 µm, 72 = 50 µm, 73–80 = 20 µm.

ing to ca. 90–150 μm long; cytoplasm of discharging endoascus finely granular, with granules delimiting spheres (FIG. 77). Ascospores 16–19 μm long at the septum and 30–35 μm wide at the broadest point (mean = 18 × 32 μm, n = 40), papilionaceous to broadly reniform, 1-septate, bi- to multiguttulate, orange to light orange-brown, smooth-walled (FIGS. 78, 79, 80).

Colonies on PYG agar hyaline to dark greenish brown. Center of the colony composed of aerial, hyaline hyphae and immersed, brown hyphae; mycelium toward the periphery dark green to brown, slightly thick walled, immersed, and effuse; no anamorph observed.

Etymology. From Latin *tangerinus* = orange, referring to the orange ascospores.

TYPE. USA. FLORIDA: Blackwater River State Forest, Blackwater River, north end, at Kennedy Bridge, 30°56′01″N, 86°44′07″W, water 11 C, pH 5, on submerged corticated woody debris, 11 Feb 2006, *HAR* and *JLC, F118-1.* (HOLOTYPE designated here, ILL40112).

Known distribution. USA (FL).

Comments. The characteristics of *L. tangerina* fit within the concept of the genus *Lepidopterella* Shearer & J.L. Crane (Shearer and Crane 1980b). *Lepidopterella tangerina* however differs from the type of the genus, *L. palustris* Shearer & J.L. Crane, in ascospore morphology and size. The ascospores of *L. tangerina* are broadly reniform and shorter at the midseptum (17–19 μm) (FIGS. 78–80) compared to the ascospores of *L. palustris*, which are deeply constricted in proportion to the overall spore morphology (butterfly shaped) and consistently longer at the midseptum (18–25 μm) (Shearer and Crane 1980b). The midseptum of the ascospore is darkened in *L. palustris* but not in *L. tangerina* (FIG. 79). In addition mature ascospores of *L. palustris* are darker than those of *L. tangerina*.

Ophiobolus shoemakeri Raja et Shearer sp. nov. (FIGS. 81–88)

Ascomata 540–580 × 295–300 μm, elongata, globosa vel late ellipsoidea, dispersa, superficiales vel demi-immersa, atra, coriacea, ostiolata. Ostiolum ca. 15–20 μm latum, collo. Collum 140–160 × 80–100 μm, cylindraceum, truncatum, paraphysatum. Peridium ca. 40–50 μm latum, glabrum e bistratis compositum; stratum externum cellulis atro-brunneis, interspersis materis amorphis atris; stratum internum 8–10 seriebus cellulis hyalinis, elongatibus, angulari, cellulae 13–18 × 2–3 μm. Pseudoparaphyses ca. 2 μm latae, cellulosae, hyalinae, constrictae ad septis, matrice gelatinosis. Asci 200–300 × 12–20 μm, longi, fissitunicati, cylindrici, apice rotundato, brevi-pedunculo contracto, 8–ascosporis, fasciculo solitaris. Ascosporae 230–270 × 4–6 μm, parallelae, e fasciculo spiratim torta dispositum ad basem asci, hyalinae vel pallidae-luteae, longae-cylindraceae, ad apicem leviter rotundatam, ad basem contracta, 25–30-septatae, circumcinctae vagina tenui gelatinosa. In quoque cellula ascosporae monoguttalata.

Ascomata 540–580 × 295–300 μm, scattered, superficial to partially immersed, black, coriaceous; venter elongated globose to broadly ellipsoidal, ostiolate; ostiole ca. 15–20 μm wide, with a neck (FIG. 81). Neck 140–160 × 80–100 μm, cylindrical, truncate, periphysate (FIG. 82). Peridium ca. 40–50 μm wide, glabrous, composed of two layers; outer layer of dark brown cells interspersed with amorphous black material; inner layer of 8–10 rows of thin-walled, hyaline, elongated, angular cells; cells 13–18 × 2–3 μm (FIG. 83). Pseudoparaphyses ca. 2 μm wide, hyaline, septate, constricted at the septa, in a gel matrix (FIG. 84). Asci 200–300 × 12–20 μm, fissitunicate, long, cylindrical, apex rounded, tapering to a short stalk, with eight ascospores in a single fascicle (FIG. 85). Ascospores 230–270 × 4–6 μm, parallel, arranged in a spirally twisted fascicle at the base of the ascus, hyaline to pale yellow, long-cylindrical, scolecosporous, slightly rounded toward the apex and tapering toward the base, 25–30 septate, multiguttulate at first (FIG. 86), then with a single guttule in each cell, surrounded by a narrow gelatinous sheath, sheath staining in aqueous nigrosin, invisible in glycerin or lactic acid (FIGS. 87, 88).

Colonies on PYG agar gray; reverse side black. Mycelium composed of branched, septate, dark hyphae; no anamorph observed.

Etymology. Named in honor of Dr Robert A. Shoemaker for his early work on the genus *Ophiobolus*, as well as his outstanding contributions to the taxonomy and systematics of the Dothideomycetes.

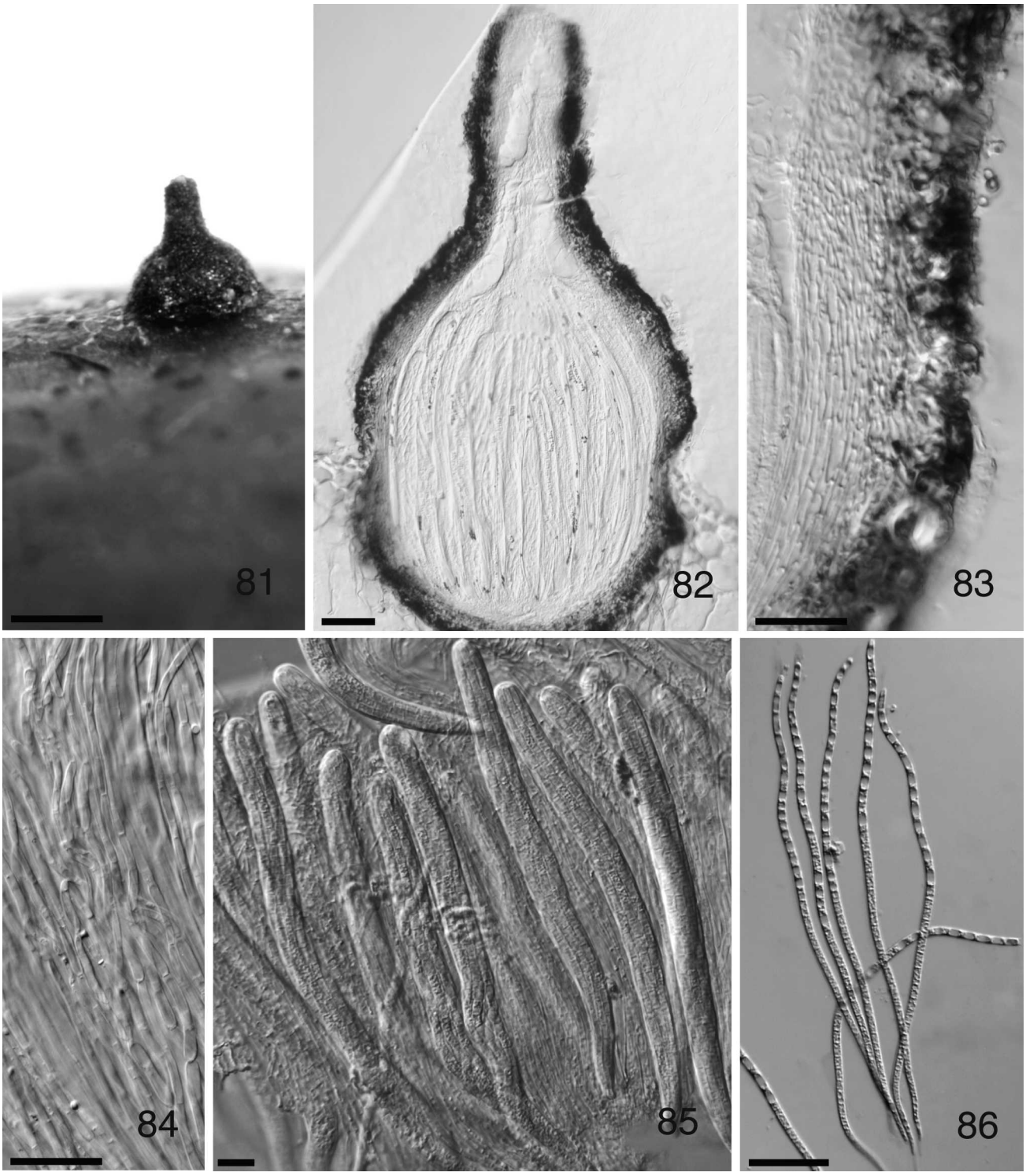
TYPE. USA. FLORIDA: Ocala National Forest, Fore Lake, 29°16′17″N, 81°55′03″W, water 37 C, pH 6.6, on submerged herbaceous debris, 16 Jul 2004, *HAR* and *CB, F22-1.* (HOLOTYPE designated here, ILL40113).

Additional specimens examined. Ocala National Forest, Lake Eaton, 29°15′18″N, 81°51′55″W, water 36 C, pH 7, on submerged herbaceous debris, 16 Jul 2004, *HAR* and *CB, F22-2.*

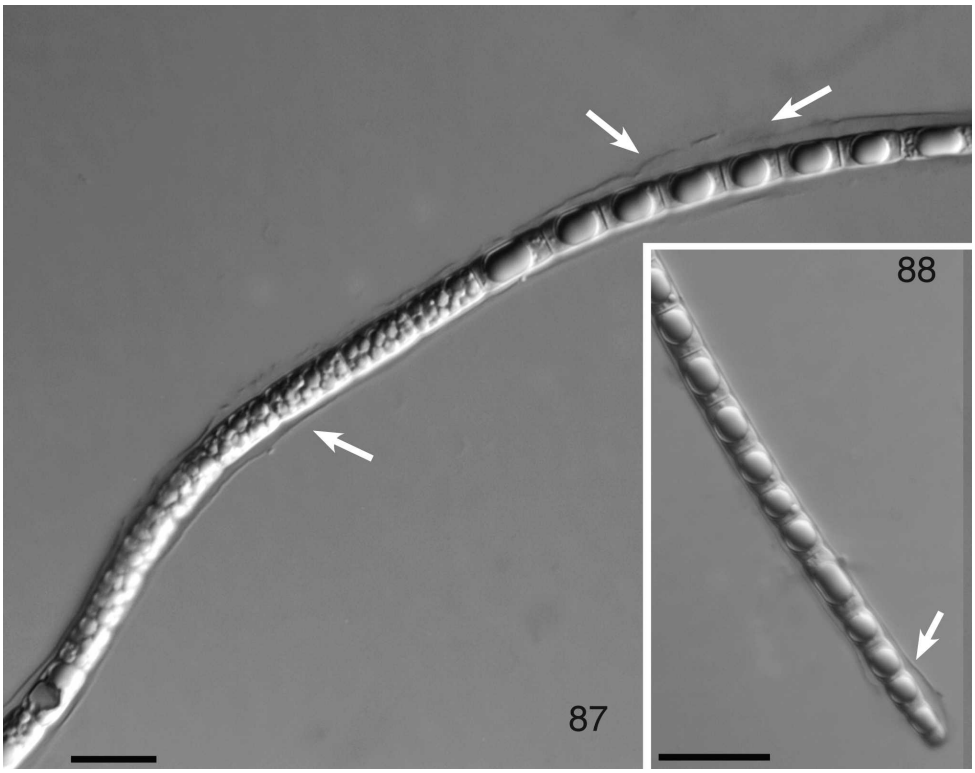
Known distribution. USA (FL).

Comments. *Ophiobolus* Riess, based on *Ophiobolus acuminatus* (Sow. ex Fr.) Duby (Leptosphaeriaceae Barr), is characterized by its solitary to clustered ascomata, immersed to erumpent, spherical to obpyriform, brown to black, with a conical papilla; numerous, long-cylindrical, fissitunicate asci; and multiseptate, yellow to brown scolecosporous ascospores (Shoemaker 1976, Walker 1980). The characteristics of *O. shoemakeri* fit within the concept of the genus *Ophiobolus*.

The ascospores of our specimen were carefully compared with other species in the genus because ascospores seem to provide useful key morphological



FIGS. 81–88. *Ophiobolus shoemakeri* from the holotype. 81. Ascoma on herbaceous substrate. 82. Longitudinal section through ascoma. 83. Peridium. 84. Cellular pseudoparaphyses in gel matrix. 85. Asci. 86. Ascospores fixed in glycerin; note ascospores rounded at the apex and tapering toward the base. 87, 88. Ascospores stained in aqueous nigrosin; note arrows showing narrow gelatinous sheath surrounding entire ascospore. Bars: 81 = 200 μm; 82–85, 87, 88 = 20 μm; 86 = 50 μm.



FIGS. 81–88. Continued.

characters to delineate species within *Ophiobolus* (Shoemaker 1976). Ascospores of the Florida specimen are similar to those of *O. lonicerae* Fabre in that they are >20 septate and are broad at the apex and taper gradually toward the base (Shoemaker 1976, FIG. 30). However *O. shoemakeri* differs from *O. lonicerae* in that the ascospores of *O. shoemakeri* have a narrow gelatinous sheath surrounding the ascospore (FIGS. 87, 88), a character absent in *O. lonicerae*. In addition the two species also differ in their ecological habitat. *Ophiobolus shoemakeri* was collected from submerged unidentified herbaceous debris from lentic habitats in Florida, whereas *O. lonicerae* originally was described from *Lonicera hispidula* (Lindl.) Douglas ex Torr. & A. Gray (Caprifoliaceae) in a terrestrial habitat in California.

In addition to *O. shoemakeri* three other *Ophiobolus* species, *O. gracilis* (Niessl) E. Mueller, *O. herpotrichus* (Fries) Sacc. and *O. typhae* Feltgen, have been reported from freshwater (http://fungi.life.uiuc.edu/). These taxa differ from *O. shoemakeri* in the size and septation of the ascospores, as well as in ascomal morphology. None of the aforementioned species collected from fresh water possess a gelatinous ascospore sheath as in *O. shoemakeri*. Another species, *O. australiensis* Johnson & Sparrow, was reported from an aquatic habitat on dead roots of *Avicennia marina* (Forssk.) Vierh var. *resinifera* (G. Forst.) Bakh. from Queensland, Australia. Kohlmeyer and Kohlmeyer (1979) think *O. australiensis* might not belong in the genus because it lacks septate ascospores and pseudoparaphyses.

Ophiobolus shoemakeri was tested for the production of extracellular enzymes in vitro and was found positive for cellulase, endoglucanase, beta-glucosidase, xylanase, laccase, amylase, pectic lyase, and polygalacturonase (Simonis, Raja, Shearer unpubl), suggesting it has the potential to decay herbaceous substrates in freshwater. Although positive for xylanase *O. shoemakeri* was negative for the production of the lignin modifying enzymes, peroxidase and tyrosinase, and it did not cause soft-rot in balsa wood. The forgoing results are interesting because most of the species of *Ophiobolus* have been isolated from herbaceous substrates. It is plausible that lack of lignin modifying enzymes and ability to form soft-rot cavities might prevent species of *Ophiobolus* from being competitive on woody substrates. More species in the genus should be tested for production of extracellular enzymes to determine whether additional herbaceous species lack lignin-modifying enzymes.

NEW RECORDS

(For descriptions and illustrations see Raja 2007, http://fungi.life.uiuc.edu/)

Aniptodera inflatiascigera K.M. Tsui, K.D. Hyde & I.J. Hodgkiss, Sydowia 49:188. 1997.

Specimens examined. USA. FLORIDA: Ocala National Forest, Bock Lake, 29°05′52″N, 81°39′11″W, water 37 C, pH 6.5, on submerged decorticated woody debris, 15 Jul 2004, *HAR* and *CB*, *F48-1*; Lake Kerr, 29°21′19″N, 81°48′45″W, water 21 C, pH 7, on submerged decorticated woody debris, 3 Mar 2005, *HAR*, *ANM* and *JLC*, *F48-5*; Lake Eaton, 29°15′49″N, 81°52′11″W, water 36 C, pH 7, on submerged herbaceous debris, 15 Jul 2006, *HAR* and *JLC*, *F48-6*; Big Cypress National Preserve, Cypress Swamp Loop Road 2, Monroe County, 25°45′36″N, 81°02′07″W, water 25 C, pH 8, on submerged decorticated woody debris, 22 Mar 2005, *HAR*, *ANM* and *JLC*, *F48-2*.

Known distribution. Brunei, Hong Kong, Philippines, USA (FL).

Comments. The specimens from Florida fit well within the concept of *Aniptodera* and are similar to *A. inflatiascigera* K.M. Tsui, K.D. Hyde & I.J. Hodgkiss (Tsui et al 1997). *Aniptopdera inflatiascigera* can be distinguished from other members of the genus by its characteristic ascus, which inflates in water on release from the ascomata. Apical appendages were not observed in the Florida specimens but were reported for the type specimen (Tsui et al 1997). If the ballooning of the ascus in water is disregarded, the thick-walled ascospores of *A. inflatiascigera* resemble those of *A. limnetica* Shearer (Shearer 1989). *Aniptodera inflatiasciger* however has larger asci and ascospores compared to *A. limnetica*.

Aniptodera inflatiascigera was found from central and south Florida on submerged decorticated woody debris as well as herbaceous debris from lentic habitats. These collections are the first reports from North America. In earlier studies this fungus was reported from submerged decorticated wood in lotic habitats in the paleotropics (Tsui et al 1997, Hyde et al 1999, Tsui et al 2000, 2001).

Annulatascus velatisporus K.D. Hyde, [as '*velatispora*'], Aust. Syst. Bot. 5:118. 1992b.

Specimens examined. USA. FLORIDA: Blackwater River State Forest, Horns Creek Swamp, 30°46′31″N, 86°54′43″W, water 30 C, pH 6, on submerged decorticated woody debris, 10 Jul 2004, *HAR* and *CB*, *F002-1*; Sweetwater Creek, 30°51′21″N, 86°51′03″W, water 26 C, pH 6.8, on submerged decorticated woody debris, 10 Jul 2004, *HAR* and *CB*, *F002-2*; 6 Jul 2006, *HAR* and *JLC*, *F002-13*; Hurricane Lake, 30°56′19″N, 86°45′12″W, water 34 C, pH 8.7, on submerged decorticated woody debris, 11 Jul 2004, *HAR* and *CB*, *F002-4*; Hurricane Creek, 30°56′36″N, 86°44′50″W, water 30 C, pH 8, on submerged decorticated woody debris, 11 Jul 2004, *HAR* and *CB*, *F002-6*; Big Coldwater Creek East Fork, 30°50′47″N, 86°59′02″W, water 25 C, pH 5, on submerged decorticated woody debris, 7 Jul 2006, *HAR* and *JLC*, *F002-15*; Apalachicola National Forest, Leon Sinks Gopher Hole, 30°18′35″N, 84°20′46″W, on submerged decorticated woody debris, 15 Feb 2006, *HAR* and *JLC*, *F002-13*; Ocala National Forest, Bock Lake, 29°05′52″N, 81°39′11″W, water 37 C, pH 6.5, on submerged decorticated woody debris, 15 Jul 2004, *HAR* and *CB*, *F002-3*; Mill Dam Lake, 29°10′42″N, 81°50′03″W, water 37 C, pH 6.7, on submerged decorticated woody debris, 15 Jul 2004, *HAR* and *CB*, *F002-5*; Redwater Lake, 29°11′49″N, 81°53′28″W, water 36 C, pH 6.4, on submerged decorticated woody debris, 14 Jul 2004, *HAR* and *CB*, *F002-7*; Marion County, Quarry Fishing Pond, swampy area, 29°12′45″N, 81°53′45″W, water 21 C, pH 7, on submerged decorticated woody debris, 26 Mar 2005, *HAR*, *ANM* and *JLC*, *F002-8*; Lake County, Alexander Springs, 29°04′51″N, 81°34′02″W, water 18 C, pH 7, on submerged decorticated woody debris, 20 Feb 2006, *HAR* and *JLC*, *F002-11*; Juniper Creek at Juniper Springs, 29°10′59″N, 81°42′46″W, water 22 C, pH 6, on submerged palm rachis, 20 Feb 2006, *HAR* and *JLC*, *F002-12*; Doe Lake, 29°02′24″N, 81°49′15″W, water 30 C, pH 5, on submerged decorticated pine wood, 16 Jul 2006, *HAR* and *JLC*, *F002-16*; Big Cypress National Preserve, Tamiami Canal, swampy area, 25°52′36″N, 81°13′40″W, water 29 C, pH 7, on submerged decorticated woody debris, 24 Mar 2005, *HAR*, *ANM* and *JLC*, *F002-9*; Turner River Canal, 25°54′21″N, 81°15′43″W, water 30 C, pH 6, on submerged decorticated woody debris, 24 Mar 2005, *HAR*, *ANM* and *JLC*, *F002-10*.

Known distribution. Australia, Brunei, Hong Kong, Malaysia, Seychelles, South Africa, USA (FL, IL, NC, NH), Venezuela.

Comments. *Annulatascus velatisporus* K.D. Hyde (Hyde 1992b) is the type species of the genus *Annulatascus* K.D. Hyde, and the family Annulatascaceae (Wong et al 1998). Populations of this species from Florida agree well in all aspects with the protolog of *A. velatisporus* (Hyde 1992b). Hyde (1992b) in the original description of the genus mentioned the ascospore sheath as being thin and irregular. The morphology of the ascospore sheath was redescribed and illustrated with TEM and SEM by Wong et al (1999). The Florida material agrees with the description and illustrations of Wong et al (1999) in that the ascospores are surrounded by a mucilaginous sheath, which is condensed at first but later becomes wide and spreads in water and is sticky.

Annulatascus velatisporus has been reported widely from lotic habitats on submerged decorticated woody debris from temperate and tropical latitudes (http://www.life.uiuc.edu/fungi/). We have collected this species from four of the five collection sites within Florida at water temperatures of 21–37 C and at pH (5–)6–8(–8.7). It also occurs in both lotic and lentic habitats and on submerged woody debris and palm rachis. These reports suggest that *A. velatisporus* is a warm-water generalist species with respect to the substrates and habitats it colonizes.

Ayria appendiculata Fryar & K.D. Hyde, Cryptogamie Mycologie 25:248. 2004.

Specimens examined. USA. FLORIDA: Apalachicola National Forest, Camel Pond, 30°16′36″N, 84°59′20″W, water 33 C, pH 5, on submerged herbaceous debris, 10 Jul 2006, *HAR* and *JLC, F116-1.*

Known distribution. Brunei, USA (FL).

Comments. The fungus collected from Florida is morphologically similar in almost all respects to the protolog of *A. appendiculata* (Fryar and Hyde 2004). The specimen collected from Florida however has a hyaline, periphysate neck, a character not reported for the type species (Fryar and Hyde 2004). The asci of *A. appendiculata* were described as persistent; asci from the Florida material although present at first, start to deliquesce after a few minutes in water. The ascospores of *A. appendiculata* appear to be surrounded by gelatinous material because the ascospores seem to be equally apart from each other; staining with India ink or nigrosin however does not seem to indicate the presence of a gelatinous sheath. The Florida specimen of *A. appendiculata* occurred on a piece of unidentified herbaceous debris in a freshwater lake, whereas the type specimen was reported from submerged wood in brackish water and seawater from Brunei. This species is reported from North America and freshwater for the first time.

Falciformispora lignatilis K.D. Hyde, Mycological Research 96(1):27. 1992c.

Specimens examined. USA. FLORIDA: Everglades National Park, Royal Palm Pond, 25°22′58″N, 80°36′36″W, water 20 C, pH 7, on submerged, decorticated woody debris, 16 Feb 2006, *HAR* and *JLC, F88-1*; Ocala National Forest, Red Water Lake, 29°11′49″N, 81°53′28″W, water 18 C, pH 5, on submerged, decorticated woody debris, 19 Feb 2006, *HAR* and *JLC, F88-2.*

Known distribution. Mexico, USA (FL).

Comments. Hyde (1992c) described and illustrated *F. lignatilis* as a new genus and species from intertidal mangrove wood in Mexico. The specimens collected from central and southern Florida represent the first reports of *F. lignatilis* from a freshwater habitat and from North America. The specimens agree well in size and morphology with the type description of *F. lignatilis* with a few exceptions. The ascomata of *F. lignatilis* were described as ''epapillate'' in the type description (Hyde 1992c), but a wide papilla was observed in the medial longitudinal section of the Florida specimen. In addition the pseudoparaphyses and asci in our collections are surrounded by a gel matrix, a characteristic not reported in the original description.

Hyde (1992c) compared *F. lignatilis* to *Setosphaeria* Leonard & Suggs (Leonard and Suggs 1974), but we believe that *F. lignatilis* is more closely related to the genus *Chaetomastia* (Sacc.) Berlese, which was reinstated by Barr (1989) for fungi having immersed to erumpent ovoid, obpyriform to globose ascomata with a wide papilla; peridium relatively wide, composed of pseudoparenchymatic cells; bitunicate, clavate to cylindrical asci, with pseudoparaphyses embedded in a gel matrix; and multiseptate obovoid, elongate, yellowish brown to dark brown ascospores surrounded by gel coating. *Falciformispora* however differs from *Chaetomastia* in having hyaline ascospores.

Fluviatispora reticulata K.D. Hyde, Mycological Research 98:724. 1994.

Specimens examined. USA. FLORIDA: Blackwater River State Forest, Bone Creek, 30°44′19″N, 86°46′29″W, water 25 C, pH 6.7, on submerged decorticated woody debris, 10 Jul 2004, *HAR* and *CB, F11-5*; Horns Creek swampy area, 30°46′31″N, 86°54′43″W, water 30 C, pH 6, on submerged decorticated woody debris, 10 Jul 2004, *HAR* and *CB, F11-7*; Apalachicola National Forest, unnamed lake east of Lost Lake, 30°21′48″N, 84°22′56″W, water 35 C, pH 7.3, on submerged decorticated woody debris, 13 Jul 2004, *HAR* and *CB, F11-1*; Andrew Lake, 30°24′09″N, 84°24′27″W, water 33 C, pH 6.5, on submerged decorticated woody debris, 13 Jul 2004, *HAR* and *CB, F11-2*; Rowletts Creek, 30°03′32″N, 85°01′11″W, water 35 C, pH 5, on submerged decorticated woody debris, 10 Jul 2006, *HAR* and *JLC, F11-8*; Camel Pond, 30°16′36″N, 84°59′20″W, water 33 C, pH 5, on submerged, decorticated woody debris, 10 Jul 2006, *HAR* and *JLC, F11-9*; Ocala National Forest, Mary Lake, 29°04′23″N, 81°49′57″W, water 33 C, pH 5, on submerged decorticated woody debris, 10 Jul 2006, *HAR* and *CB, F11-3*; 16 Jul 2006, *HAR* and *JLC, F11-10*; Fore Lake, 29°16′17″N, 81°55′03″W, water 37 C, pH 6.6, on submerged decorticated woody debris, 16 Jul 2004, *HAR* and *CB, F11-4.*

Known distribution. Brunei, Papua New Guinea, USA (FL).

Comments. Currently three species are accepted in the genus *Fluviatispora* K.D. Hyde (Hyde 1994, Fryar and Hyde 2004). Following Fryar and Hyde (2004) the species from Florida best fits the description of *F. reticulata* K.D. Hyde (Hyde 1994). The ascospores of the specimen from Florida are slightly longer and narrower (20–25 × 8–12 μm) than those of the type species (17–23 × 10–13 μm) (Hyde 1994). In addition the ascospores of the Florida material are biguttulate, as opposed to a single large eccentric guttule reported in the type species (Hyde 1994).

We have collected *F. reticulata* nine times from three of the five collection sites in northern and central Florida in lotic and lentic habitats. It occurred during the summer in two successive years at water temperatures of 25–37 C and at pH 5–7, only on woody substrates. This species is reported from North America for the first time.

Lepidopterella palustris Shearer & J.L. Crane, Trans-

actions of the British Mycological Society 75(2):194. 1980b.

Specimens examined. USA. FLORIDA: Apalachicola National Forest, Owls Creek Backwater Swamp, 30°03′31″N, 85°01′11″W, water 30 C, pH 7.8, on submerged decorticated woody debris, 12 Jul 2004, *HAR* and *CB*, *F32-1*. (ILL); Fisher Creek, Leon County, 30°18′51″N, 84°23′57″W, water 25 C, pH 5, on submerged corticated woody debris, 9 Jul 2006, *HAR* and *JLC*, *F32-2*.

Known distribution. USA (FL, IL).

Comments. The specimens from Florida fit well within the genus *Lepidopterella* and agree well with the description of *L. palustris*, the type species of the genus (Shearer and Crane 1980b). This genus has butterfly-shape ascospores and globose to subglobose asci, which show fissitunicate dehiscence when left in water for few minutes. Although fissitunicate dehiscence occurs in this fungus, the ascospores are not forcefully discharged from the endoascus, (Shearer and Crane 1980, Raja pers obs) instead the endoascus wall deliquesces to release the ascospores.

Lepidopterella palustris originally was described from a cypress swamp in southern Illinois (Shearer and Crane, 1980b). Thus far this species has not been reported elsewhere from temperate and tropical regions worldwide (Cai et al 2006, http://www.life.uiuc.edu/fungi/). Shearer and Crane (1980b) suggested that this fungus is probably a warm water species. Our collections from Florida add support to their premise because the fungus from Florida was collected only in the summer at water temperatures of 25–30 C.

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