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NEW FUNGI FROM DASYLIRION (AGAVACEAE)

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ABSTRACT

Dead leaves of Dasyllirion were collected in Texas in Guadalupe Mountains National Park and Big Bend National Park, and in the Valley of Fires, New Mexico. The fungi present were identified and grown in culture to identify teleomorph-anamorph connections. Four new ascomycete species are described—Graphyllium dasylirionis, Splanchnonema dasylirionis, Chaetoplea dasylirionis, and Chaetoplea solifiorum. In addition, a new genus, Parahendersonia, is described to accommodate the coelomycete anamorph of Chaetoplea dasylirionis.

Key words: Agavaceae, ascomycete, Chaetoplea, coelomycete, Dasyllirion, Graphyllium, Parahendersonia, Splanchnonema, systematics.

INTRODUCTION

Spectacular flowering specimens of Yucca L. or Agave L. are almost symbolic of the vegetation of arid regions of the southwest. Nevertheless, in recent years little work has been done on the saprobic foliar fungi of the Agavaceae, the family to which Yucca, Agave, and their lesser-known relatives, Nolina Michx. and Dasyllirion Zucc. belong. Within the boundaries of the United States, approximately 80 fungal species have been described from Yucca, which is widespread. Many fewer species have been described from Agave, which is widely cultivated but more limited than Yucca in natural distribution. Only two species of ascomycetes and three species of coelomycetes have been described from Dasyllirion (Farr et al. 1989). New genera as well as new species of existing genera have been found on some members of the Agavaceae in a continuing study of foliar fungi of that flowering plant family. One new genus and four new species of existing genera, all from Dasyllirion, are described below.

MATERIALS AND METHODS

Measurements are from living materials. Cultures from anamorphs were made by suspending conidia in a drop of sterile tap water on the surface of commercial potato dextrose agar (PDA) in a petri dish and spreading the drop with a glass rod. After 24 hr, single-germinated conidia were placed on PDA. Cultures from teleomorphs were prepared by washing ascii three times in sterile tap water and transferring individual ascii to PDA in a petri dish. Cultures were maintained at room temperature on a table top.

RESULTS AND DISCUSSION

Graphyllium dasylirionis A. W. Ramaley, sp. nov.

Ascomata immersa, subepidermalia, globosa, turbinate, vel oblonga, transectiones mediae circulares vel turbinateae, usque ad ca. 570 μm longa, 440 μm lata et 480 μm alta papilla inclusa; papilla brevis (ca. 60 μm), lata (ca. 90 μm diam), dilute colorata, epidermis tecta, cylindracea, ut circulis, centrale, hyphis repletum, peridion molle et tenue, ex 3–5 stratis cellularum parvarum atipicellaturn, 8–16 μm crassum, et internus 1 vel 2 stratis cellularum dehiscentiae, compressarum constans; hyphae ex peridio in substraturn. Clypeus latus, tenuis, ex cellulis parvis atipicellatis constans, laticolus non superans, ostiolum circulare, centrale, hyphis repletum, peridion tumefaciatum, ex 3–5 stratis cellularum parvarum atipicellaturn, 10–15 μm crassum, ostiolum includens; papilla brevis, tenuis, ex cellulis parvis atipicellatis constans, laticolos non superantibus, ostiolum circulare, centrale, hyphis repletum, peridion tumefaciatum, ex 3–5 stratis cellularum parvarum atipicellaturn, 10–15 μm crassum, ostiolum includens.

Ascomata. Immersa, subepidermalia, globosa, turbinate, vel oblonga, transectiones mediae circulares vel turbinateae, usque ad ca. 570 μm longa, 440 μm lata et 480 μm alta papilla inclusa; papilla brevis (ca. 60 μm), lata (ca. 90 μm diam), dilute colorata, epidermis tecta, cylindracea, ut circulis, centrale, hyphis repletum, peridion molle et tenue, ex 3–5 stratis cellularum parvarum atipicellaturn, 8–16 μm crassum, et internus 1 vel 2 stratis cellularum dehiscentiae, compressarum constans; hyphae ex peridio in substraturn. Clypeus latus, tenuis, ex cellulis parvis atipicellatis constans, laticolus non superans, ostiolum circulare, centrale, hyphis repletum, peridion tumefaciatum, ex 3–5 stratis cellularum parvarum atipicellaturn, 10–15 μm crassum, ostiolum includens; papilla brevis, tenuis, ex cellulis parvis atipicellatis constans, laticolos non superantibus, ostiolum circulare, centrale, hyphis repletum, peridion tumefaciatum, ex 3–5 stratis cellularum parvarum atipicellaturn, 10–15 μm crassum, ostiolum includens.
Fig. 1A-D. Graphyllium dasylirionis.—A. Ascus.—B. Ascoma.—C. Ascospores.—D. Conidiogenous cells and portion of wall from conidiomata formed in culture. (Standard line: A = 25 μm; B = 250 μm; C, D = 20 μm.)

except in face view only, the single longitudinal septum in all cells, or the end cells or two end cells may lack a septum.

Cultures with little aerial mycelium, light brown around the inoculum, the rest white to tan, then darkening with age from the inoculum outward, producing small, dark, irregularly shaped multilocular pycnidial conidiomata with several small nonpapillate ostioles, 50–250 μm in diam, in groups up to 500 μm in diam. Locules with a few layers of unevenly brown-pigmented wall cells. Conidiogenous cells phialidic, channel minute, periclinal walls thickened, discrete or integrated on short, 1–2 celled, sometimes branched, conidiophores (Fig. 1D). Conidia mostly 4.8–6.4 × 1.6–2.0 μm, hyaline, smooth, monon- or biguttulate, bacillar, allantoid or sigmoid, released in hyaline to pinkish droplets. Conidia spread on PDA showed no growth, therefore these conidia are likely spermata.

Graphyllium dasylirionis was placed in Graphyllium because it has applanate ascospores with one longitudinal septum in face view, and soft, thin peridia composed of small, dark cells. The generic concept presented by Shoemaker and Babcock (1991) would exclude this species with applanate ascospores from Graphyllium, in the Hysteriaceae, because the peridia do not open by a long slit. Graphyllium dasylirionis cannot be placed in the Pleosporaceae, a second family used to accommodate species with applanate ascospores, because, as delimited by Barr (1987), members of this family have relatively large-celled peridia and the known anamorphs are hyphomycetous. The third family Shoemaker and Babcock use for species with applanate ascospores is the Diademaceae. The opening mechanism characterizing peridia of this family is not found in all species of genera they include in the family, nor are all genera with species opening by a discoid lid included in the Diademaceae. Barr (1993a) therefore rejects using the opening mechanism of ascocoma as the only character on which to base a family or genus. Barr’s revised sense of the Diademaceae includes taxa which may open by a lid, a lysigenous pore, or a slit (1993a). She removes Graphyllium from the Hysteriaceae to the Diademaceae because the thick, three-layered peridium of true members of the Hysteriaceae is quite different from that of species of Graphyllium. Graphyllium dasylirionis can now be properly placed with other species of Graphyllium in the Diademaceae.

Splanchnonema dasylirionis A. W. Ramaley, sp. nov.

Fig. 2A–D

Ascomata immersa, subepidermalia, globosa, turbinata, vel oblonga, 225–350 μm lata, 275–450 μm longa, 225–350 μm alta; apex lector depressus, rotundatus, vel brevipapillatus, papilla ex cellulis dilute coloratis composita, ca. 80–100 μm lata et 60–80 μm alta, canalis ostioli hyphis repletae; peridium attribrunneum, molle, ex ca.
Ascomata immersed, subepidermal, globose, turbinate, or oblong, 225–350 μm wide, 275–450 μm long, 225–350 μm high; apex somewhat flattened, rounded, or short papillate, the papilla of light-colored cells, ca. 80–106 μm broad and 60–80 μm high, ostiolar channel stuffed with hyphae; peridium dark brown, soft, composed of about 5 layers of small, dark-walled, slightly compressed, pseudoparenchymatous cells, ca. 20 μm thick at the bottom and sides, somewhat thicker toward the top, brown hyphae from the peridium into the substrate. Asci (Fig. 2C) 125–160 × 20–28 μm, bitunicate, basal and lateral, clavate, 4- or 8-spored. Pseudoparaphyses narrowly cellular, filling the ostiolar channel. Ascospores (Fig. 2A) 28–45.6 × 12.4–17.6 μm, reddish brown, obovoid, ends obtuse, the apex narrower than the base; 5–7-septate, one or two longitudinal septa through most cells but the end cells often aseptate, the first-formed, usually submedian eu-septum constricted, the other septa distoseptate; wall thick, finely punctate to nearly smooth, surrounded by a broad hyaline sheath in water, overlapping biseriate in the ascus.

In culture, mycelium white, becoming light orange centripetally, darkening at the agar surface in older parts of the colony, producing dark, thin-walled, ostiolate, nonpapillate conidiomata singly, or a few grouped together at the agar surface, 200–500 μm in diam. The wall composed of 1–4 layers of brown-walled cells and a variable number of layers of hyaline cells internally. Conidiophores lining the entire conidiomatal cavity, multicellular, branched at the base and above. Conidiogenous cells (Fig. 2D) determinate, phialidic, channel minute, periclinal walls thickened, discrete or integrated, hyaline, smooth, used up in conidium formation, conidiogenesis centrifugal. Conidia released in white to pink droplets, bacillar, hyaline, aseptate, smooth, bigutullate, ca. 4–8 × 1.6 μm. Development of the bacillar Splanchnonema conidia resembles microconidium/spermatium formation in some species of Asteromella, the spermatial synanamorph for many species of Mycosphaerella (Higgins 1929; Ramaley 1991). When spread on PDA, a high percentage of these conidia increased in size and formed ovate or ellipsoid cells, germinated (Fig. 2B), and developed into normal-appearing colonies. Thus, although their size, shape, and formation suggests that their role is as spermatia they may also serve as asexual reproductive structures.

Ascospore dimensions of Splanchnonema dasylirionis are similar to those of three other species with clavate asci and biseriate ascospores, S. carpini (Fuckel) E. M. Barr, S. holochistum (Berk. & Broome) M. E. Barr, and S. vaccini (Barr 1993c). Reddish brown, transversely 5–7-septate ascospores distinguish S. dasylirionis from S. vaccini which has bright yellow brown, 7–9-septate ascospores (Barr 1993b).

Ascomata of *S. holochistum* are much larger than those of *S. dasylirionis*, 500–900 vs. 225–450 μm. In addition, the peridium of *S. dasylirionis* is uniformly narrow at the bottom and sides and slightly thickened at the top whereas the peridium of *S. holochistum* is wide at the sides and narrowed toward both base and apex (Barr 1990). In contrast to the narrow peridium of *S. dasylirionis* ascomata, ca. 20 μm, the peridium of *S. carpini* ascomata is 50–70 μm wide and up to 100 μm wide at the base (Barr 1982). *Splanchnonema carpini* also has brown ascospores which are regularly transversely 5–9-septate (Barr 1982) while the reddish brown ascospores of *S. dasylirionis* are transversely 5–7-septate.

**Chaetoplea sotolifoliorum** A. W. Ramaley, sp. nov.


**Ascomata** (Fig. 3A) immersed, subepidermal, oblong, 250–350 μm long, 225–275 μm wide, and 200–275 μm tall, a small papilla penetrating the host epidermis. Ostiole with periphyses. Peridium 12–28 μm thick, composed of 4–7 layers of small, brown cells, abundant brown hyphae from the peridium into the substrate. Ascii (Fig. 3C) 112–160 × 10.4–14.4 μm, bitunicate, cylindric, 8-spored. Pseudoparaphyses cellular, ca. 3 μm wide. Ascospores (Fig. 3D) 16–21.6 × 7.2–10.4 μm, overlapping uniseriate in the ascus, yellow-brown, symmetric or the upper hemispore slightly broader than the lower hemispore, equilateral, both ends bluntly rounded, thick walled, coarsely punctate to nearly smooth, 3–5-septate, the A1 septum constricted, the other septa less constricted, usually only the middle cells longitudinally 1-septate but sometimes the end cells also longitudinally 1-septate.

Anamorph: Coelomycetous. Conidia from cultures (Fig. 3B) light brown, smooth or longitudinally striate, 1–3-septate, mostly 8–12 × 3.2–4.8 μm, formed within pycnidial conidiomata. Cultures with a bright red reverse near the center, darkening toward the edges, mycelium dark green near the agar surface and below, white to lighter green above, agar colorless to light green in old cultures.

Five of the eight *Chaetoplea* species with 3–5-septate ascospores (Barr 1990; Ramaley and Barr 1995) are found on hosts not closely related to *Dasylium*. *Chaetoplea nubilosa* (Ellis & Everh.) M. E. Barr and *C. aspera* (Ellis & Everh.) M. E. Barr are superficial on their substrates. The immersed ascomata of *C. pusilla* (Karsten & Malbranche) M. E. Barr are minutely papillate and have a dark, expanded, compact clypeus in close association with the substrate. *Chaetoplea sotolifoliorum* has immersed ascomata associated with the overlying substrate by loosely woven hyphae from the upper wall to the host epidermis. These characters set it apart from *C. nubilosa, C. aspera,* and *C. pusilla*. *Chaetoplea carpincola* (Ellis & Everh.) M. E. Barr, immersed in woody substrates, has cylindric ascii with reddish brown, smooth, uniseriate ascospores that are 15–18 × 7–7.5 μm. *Chaetoplea sotolifoliorum* also has cylindric ascii, but the uniseriate ascospores, measuring 16–21.6 × 7.2–10.4 μm, are
broader than those of C. carpincola, yellowish brown, and often coarsely punctate. Chaetoplea calvescens (Fr. ex Desm.) Clements, immersed or erumpent in herbaceous stems, has yellowish brown to pale brown, smooth ascospores that are (13-)15-23 × (5-)6-8(-9) μm. The asci from American collections are clavate and the spores usually biseriate (Barr 1990), whereas those in European collections (Webster and Lucas 1959) are usually cylindric and the ascospores generally uniseriate. Chaetoplea calvescens and C. sotolifoliorum are similar, but longitudinal septa appear tardily or not at all in the ascospores of C. calvescens, which are smooth and more tapered toward the apex and base than ascospores of C. sotolifoliorum.

The size, shape, and septation of conidia from C. sotolifoliorum cultures are much like those from C. calvescens cultures and on the host (Webster and Lucas 1959). However, mature conidia from C. sotolifoliorum cultures are light brown and most often longitudinally striate, whereas conidia from the anamorph of C. calvescens, Microdiplodia hemingstii Starzic = Chaetodiplodia caudina Karst. (Sutton 1980), are pale yellow and wall sculpturing is not mentioned (Webster and Lucas 1959).

Chaetoplea dasylirionis A. W. Ramaley, sp. nov.

Fig. 4A–C


Ascomata (Fig. 4A) immersa, subependimalis, sphaeroidea, oblonga, vel 300–500 μm longa, 150–350 μm lata, et 175–325 μm alta, apex contra epiderimidem hospitis depressus et ad epiderimidem adhaerens, pars adhaerens nigra et orbiculata vel, saepius, lenticularis a vertice visa, axis longior parallelus ad longitudinem folii, marginem apicis adhaerentia saepe extensus, papilla parva epidermidem stuffed with periphyses and pseudoparaphyses. Peridium 20–40 μm crassum, latera et basis ex 4–6 stratis cellulararum parvarum atiparietarum compositum, hyphae brunneae ex peridio in substratum. Asci 96–136 × 14.4–17.6(-21.6) μm, bitunicati, numerosi, clavati vel subcylindracei, 8-spori. Pseudoparaphyses cellularae, ca. 3 μm latae. Ascosporeae 19.2–28 × 8.8–12 μm, aureae aureobrunnescentes, typice asymmetricae, hemispora supera levior latior quam hemispora infera, equilaterales, externa obtusa roundata vel apex apiculatus, partes crassus, subtiliter punctata, A1 septum subconstrictum, septa alia minus constricta, 3(-5)-septatae, pleuroque mode cellularae mediae longitudinaliter 1-septaetiae sed interdum etiam cellularae terminales longitudinaliter septatae, in aqua tonica lata circumsertae.

morphe, a narrow clypeus often extending beyond the edge of the adherent apex, a small papilla penetrating the epidermis, Ostiole stuffed with periphyses and pseudoparaphyses. Peridium 20–40 μm thick, the sides and bottom composed of 4–6 layers of small, darkwalled cells, brown hyphae from the peridium into the substrate. Asci (Fig. 4C) 96–136 × 14.4–17.6(-21.6) μm, bitunicate, numerous, clavate or subcylindric, 8-spored. Pseudoparaphyses cellularae, ca. 3 μm wide. Ascospores (Fig. 4B) 19.2–28 × 8.8–12 μm, golden becoming golden brown, usually asymmetric, the upper hemispore somewhat broader than the lower hemispore, equilateral, both ends bluntly rounded or the apex with a small apiculus, thick walled, finely punctate, the A1 septum somewhat constricted, the other septa less constricted, 3(-5)-septate, usually only the middle cells longitudinally 1-septate but sometimes the end cells also longiseptate, surrounded by a broad, hyaline sheath in water.
Anamorph: Parahendersonia dasylirionis Ramaley, gen. et sp. nov.

Conidiomata immersa, subepidermalia, sphaeroida, oblonga, vel subgloboa, 250–500 μm longa, 150–350 μm lata, 175–325 μm alta, apex contra epidemidem hospitis depressus et ad epidemidem adhaerens, pars adhaerens nigra et orbiculata vel, saepe, lenticularis a vertice visa, axis longior parallexus ad longitundinem folii, clypeus angustus ultra margo apicis adhaerentis saepere extensus, papilla parva epidemidem penetrans. Pariis 20–40 μm cassus, ex (4)–5–7–9 stratis cellularum bruncearum, et interdum strato cellularum hyalinarum interiorum compositus, hyphae brunceae ex parietis in substratum. Celullae macroconidiogenae ex cellulis interioribus pariis conidiomatis formae, discrete, pterumque determinatae, leves, hyalinae vel paries maculis brunneis, ampulliformes, holoblasticae. Macroconidia 13.6–16.8 × 5.6–8 μm, brunnea, cylindracea, extrema obtusae rotundata, punctata, (1)–3–septata. Ontogenesis conidiis holoblastica constructione ad parietes apicalem; proliferatio cellularum conidiogenarum enteroelasticae; conidia addita ad ca. idem punctum obtuse rotundata, punctata, (1)–3–septata. Conidial ontogeny holoblastic by conidiomatal wall near the apex with a rosy pigment at the tip, and clavate to subcylindric asci containing golden yellow-brown to brown, finely foveolate to smooth ascospores 19.2–30.4 × 8–14.4 μm (Ramaley and Barr 1995). Ascosoma of C. sotoliflorianum lack a rosy pigment at the tip of the papilla, the asci are mostly cylindrical, and the often coarsely punctate ascospores are smaller, 16–21.6 × 7.2–10.4 μm.

Parahendersonia dasylirionis was thought to be the anamorph of Chaetoplea dasylirionis because ascomata and conidiomata were found separately or intermixed on the host and were so similar in appearance that the contents could be ascertained only by opening them. Colonies in cultures from conidia and asci had mixed on the host and were so similar in appearance that only of these genera (Sutton 1980; Swart and Walker 1988; Sutton and Dyko 1989; Walker et al. 1992). As presently delimited, none of these genera comfortably accommodates P. dasylirionis. Members of nine of these genera have hyaline conidia or are acervular. Seven more genera regularly have percurrent proliferation when more than one conidium is formed on a single conidium. Ascosoma appearing at all similar to ascomata of C. dasylirionis. However, C. dasylirionis asci are much larger than those of C. pusilla, 96–136 × 14.4–17.6(–21.6) μm vs. 60–90 × 10–15 μm. In addition, the ascospores of C. dasylirionis are 19.2–28 × 8.8–12 μm, golden, becoming golden brown, and finely punctate, whereas ascospores of C. pusilla are 14–18(–20) × 6.5–8(–9) μm, and pale brown with smooth walls. The three Chaetoplea species from hosts in the Agavaceae are quite easily distinguished. The tiny papilla and expanded upper wall or clypeus flattened against the host epidermis distinguishes ascomata of C. dasylirionis from those of C. roshipapilla A. W. Ramaley & M. E. Barr and C. sotoliflorianum. Ascosoma of C. roshipapilla from Yucca, have a broadly papillate apex with a rosy pigment at the tip, and clavate to subcylindric asci containing golden yellow-brown to brown, finely foveolate to smooth ascospores 19.2–30.4 × 8–14.4 μm (Ramaley and Barr 1995). Ascomata of C. sotoliflorianum lack a rosy pigment at the tip of the papilla, the asci are mostly cylindrical, and the often coarsely punctate ascospores are smaller, 16–21.6 × 7.2–10.4 μm.

Conidiomata immersed, subepidermal, sphaeroidal, oblong, or nearly globose, 250–500 μm long, 150–350 μm wide, 175–325 μm tall, the top flattened against, and adhering to, the host epidermis, the adherent portion black and circular or, more often, lenticular from above, long axis parallel to the length of the leaf, a narrow clypeus often extending beyond the edge of the adherent apex, a small papilla penetrating the epidermis. Wall 20–40 μm thick, composed of (4)–5–7–9 layers of brown cells, sometimes with a layer of hyaline cells at the interior, brown hyphae from the wall into the substrate. Macroconidiogenous cells (Fig. 3D) formed from the interior of the conidiomatal wall, discrete, mostly determinate, smooth, hyaline or with brown spots on the walls, ampulliform, holoblastic. Macroconidia (Fig. 3E) 13.6–16.8 × 5.6–8 μm, brown, cylindric, both ends obtusely rounded, punctate, (1)–3–septate. Conidial ontogeny holoblastic by apical wall building, conidiogenous cell proliferation enteroblastic, additional conidia produced at approximately the same level, rarely additional conidia produced at successively higher levels. Microconidia ca. 5 μm in diam, approximately globose, smooth, hyaline, formed on the upper conidiomatal wall near the ostiole.

Among the Chaetoplea species with (3–5)–septate ascospores (Barr 1990), only C. pusilla has immersed, clypeate ascomata appearing at all similar to ascomata of C. dasylirionis.
conidiogenous cell. Like Parahendersonia, the nine remaining genera have pycnidal conidiomata and conidial secession is at approximately the same level on the conidiogenous cells. These genera may be separated from Parahendersonia by a variety of characters. Massariotheca Syd., and Camarographium Bub. conidia are smooth and disoseptate. Conidiomata of Wojnowicia Sacc. are setose and bear smooth, pale brown, several-septate conidia. Neottiospora Desm. conidia are hyaline to olivaceous, aseptate, and fusiform, with a gelatinous sheath everted toward the base. Sphaerellopsis Cooke conidia are 0–1-septate and the apex has a gelatinous cap. Sienocarpella Syd. conidia are smooth, pale brown, fusiform, and (0–)1–3-septate. Hendersoniopsis Höhn. conidiomata are multilocular and eusporic. Macroconidia are pale brown, smooth, and fusiform, and macroconidia are falcate. Hendersonula Speg. has been recently revised (Sutton and Dyko 1980; Ramaley and Barr 1995). Although some conidia of Macrospora, and similar fungi on Agavaceae. Can. J. Bot. 70: 1617–1658.


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