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Plectania seaveri (*Ascomycota*, *Pezizales*), a new discomycete from Bermuda

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Abstract— *Plectania seaveri* sp. nov. is described after revision of Bermudian material identified by Seaver under the name *Bulgaria melastoma.* The only known host for this new species is the endemic Bermudian tree *Juniperus bermudiana* (Bermuda cedar), a species whose numbers have greatly reduced over the past 70 years due to the introduction of invasive scale insects. As this new fungus has not been collected since 1940, it may now be extinct. Color pictures of fresh and dried specimens, microscopic morphology, and a black and white drawing of hymenial elements are included. Related, excluded, and doubtful species of *Plectania* are also discussed.

Key words— ascomycetes, *Donadinia*, taxonomy

Introduction

When the senior authors (MC, CA) began their revision of the genus *Plectania* Fuckel, they realized that the Bermudian collections reported by Seaver (1928, 1942) and Seaver & Waterston (1946) under the name *Bulgaria melastoma* [i.e., *Plectania melastoma* (Sowerby) Fuckel] required further study. After publication of Carbone et al. (2012) and Agnello & Carbone (2012), the need to re-examine these collections was even clearer. These early studies strongly indicated that Seaver's Bermudian collections might represent a new taxon, and so, joined by the third author (SLG), we examined all Bermudian material of this taxon in the Plant Pathology Herbarium of Cornell University (CUP).

Our revision rapidly confirmed that Bermudian material of *Bulgaria melastoma* in CUP does not belong to *B. melastoma* but in fact represents a strikingly different, undescribed species. We here name this species in honour

Plate 1. F.J. Seaver in his office at NY Botanical Garden (photographed 1930–35). Photo by H.M. Fitzpatrick, from the Cornell University Plant Pathology Herbarium photograph collection.

of the original researcher for most of this material, the American mycologist Fred Jay Seaver (Plate 1).

Materials & methods

Microscopic characters are based on dry specimens. Two optical microscopes were used: Olympus CX41 trinocular and Optika B353 trinocular with plan-achromatic objectives 4×, 10×, 40×, 60×, and 100× in oil immersion. Microscopic pictures were made using a Nikon 4500 camera and a Nikon Coolpix. Primary mounting media were Melzer's reagent, cotton blue, and Congo red. Water mounts were used for all measurements and observations of pigments. At least 30 spores were measured from each examined apothecium. SEM observations were made using a Hitachi S-4000 scanning electron microscope (SEM) at the University of Florida Core EM facility (Gainesville, Florida, USA). A piece of the hymenium from the holotype was mounted on aluminum stubs with adhesive graphite tabs and sputter coated with a gold-palladium alloy for 60 s.

Taxonomy

Plectania seaveri M. Carbone, Agnello & LaGreca, **sp. nov.** Pl. 2–6

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"*Bulgaria melastoma*" sensu Seaver, N. Amer. Cup-fung., Operc.: 198, 1928, Bermudian records.

Differs from *Plectania melastoma* in the absence of macroscopic orange-red warts, more stipitate apothecia, and ellipsoid and differently ornamented spores; from *P. nannfeldtii* and *P. helvelloides* in a very different spore ornamentation; and from *P. lusitanica* in larger spores and more pronounced ornamentation.

Type: Bermuda, Walsingham, ca. 150 fruitbodies on bark of *Juniperus bermudiana*, 20 January 1922, leg. H.H. Whetzel, Bermuda Fungi no. 188 [**Holotype**, CUP-Whetz. B.F.–0188, as *Bulgaria melastoma*].

Etymology: In honor of the American mycologist, Fred Jay Seaver.

Macrocharacters (based on dried specimens) — apothecia gregarious to subcespitose, mostly long stipitate, nearly globose and closed at first, expanding as they mature; base attached to substrate with very numerous dark black, threadlike filaments (subiculum); cup ≤18 mm across and ≤6 mm high; margin mostly entire: hymenium blackish to black, apparently smooth; external surface blackish to black, with a dark brownish to purplish-amaranth shade, rough, gently wrinkled; stipe ≤16 mm high and ≤2 mm thick, solid, black; the length varying depending on the point of growth from the substrate; flesh leathery, darker near external surfaces.

Microcharacters — asci 350–400 × 14–15 µm, cylindrical, operculate, with a slightly excentric operculum, inamyloid, eight-spored, with walls ≤1 µm thick and a tapered, flexuous base with a generative hypha that is connected more or less distantly from the basal septum; paraphyses not, or only slightly, exceeding the length of the asci, 1.5–2(–2.5) µm wide, cylindrical, septate, sometimes anastomosing, branched, pale brownish, multiguttulate by very small oil drops; the apex usually more or less diverticulate and lobed; an extracellular, brownish, amorphous pigment present in the upper part (observed in water mounts), uniting them in bundles; setae (hymenial hairs) cylindrical, as long as the paraphyses, (3–)3.5–4.5 µm wide, with a simple to slightly subcapitate apex and single septum at the very base, pale brownish; spores ellipsoid, 22–27 × 10–12 µm, most frequently 23–25 × 10–11 µm, very few spores observed ≤31 µm long, Q = (1.8–)2–2.7(–3), subhyaline to light yellowish, filled with a granular content and sometimes with few slightly bigger oil drops, walls ≤1 µm thick; spore sculpturing of small but well-defined round warts ≤0.8 µm broad and 0.6 µm high, visible also at low magnifications in optical microscopy; confirmed by the SEM, showing round isolated warts; very young spores are round, smooth and thick walled; subhymenium composed of a dense textura intricata of cylindrical, frequently septate, hyphae, with thickened, more or less dark brown walls; at low magnifications, it appears uniformly brownish to brown and not so sharply differentiated from the upper layer of the medullary excipulum; medullary excipulum of textura intricata with cylindrical, septate hyphae, 5–11 µm wide, with walls on average 0.5–0.8 µm thick; two main layers can be recognized: (i) an upper one with more brownish, denser, less gelified hyphae with a parallel orientation to the hymenial surface, (ii) a

Plate 2. *Plectania seaveri*. Upper row: CUP-033234: fresh specimens. Photo by F.J. Seaver. Bottom row: dried specimens of holotype collection. Photos by M. Carbone. Bar = 10 mm valid for bottom row only.

middle-lower one with lighter to yellowish, more loosely interwoven hyphae immersed in a subgelatinous matrix; ectal excipulum of textura subglobulosa-angularis to textura angularis made up of elements ≤20 µm wide and/or high, very dark brown due to the colored walls and the presence of an incrusting

brown pigment. In the very outer part the pigmentation becoming more amber-like and crystalline; EXTERNAL HAIRS mainly of two types, although intermediate forms occurring; (i) cylindrical, septate, hyaline, thin walled, heavily encrusted hyphoid hairs, ≤2.5 µm wide; (ii) cylindrical, infrequently septate, ≤6 µm wide, very long, mainly straight, smooth to very finely encrusted (only in the basal part) by the same kind of crystalline pigment, and brown due to an epimembranaceous pigmentation, with walls thickened ≤0.8 µm; in non-squashed mounts, they are mainly lying on the external surface, and it is not easy to determine their entire length; SUBICULUM made up of cylindrical, sometimes very slightly flexuous and/or notched, 6–9 µm wide, ≤3 mm (or more) long, septate hyphae, brown due to an epimembranaceous pigmentation, with walls thickened ≤0.8 µm, mainly smooth but in places a very light greenish extracellular pigment is present.

ECOLOGY & DISTRIBUTION — gregarious to subcespitose, on bark of Bermuda cedar (*Juniperus bermudiana* L.). January and February. Known only from the Bermuda Archipelago.

ADDITIONAL COLLECTIONS EXAMINED: **Mature collections**: **BERMUDA**, Agricultural Station, eleven fruitbodies on bark of *Juniperus bermudiana* on ground, 4 February 1926, leg. H.H. Whetzel, Ogilvie & F.J. Seaver (CUP-033235); six fruitbodies on *J. bermudiana*, 15 January 1926, leg. H.H. Whetzel, Ogilvie & F.J. Seaver (CUP-034615). Burt's Island, four fruitbodies on fallen tiny twigs of *J. bermudiana*, 9 February 1926, leg. H.H. Whetzel, Ogilvie & F.J. Seaver (CUP-033236).

Immature collections: **BERMUDA**, Agricultural Station, on bark of *Juniperus bermudiana* on ground, 12 January 1926, leg. H.H. Whetzel, Ogilvie & F.J. Seaver (CUP-033231); on bark of living *J. bermudiana*, 28 January 1926, leg. H.H. Whetzel, Ogilvie & F.J. Seaver (CUP-033234). Harrington house, on fallen bark of *J. bermudiana* on ground, 23 January 1926, leg. H.H. Whetzel, Ogilvie & F.J. Seaver (CUP-033233).

Discussion

PRELIMINARY NOTES — The first hint of the existence of these Bermudian collections can be found in Seaver's note (1928) under *Bulgaria melastoma*: "What appears to be this species is very abundant in Bermuda on the bark of Bermuda cedar, *Juniperus bermudiana*, but never on anything else. Since the host is an endemic species, the fungus may also be found to differ and be itself endemic. The writer has not yet had the opportunity to clear up this point but reference will be made to it in later publications". In Seaver (1942), again under *Bulgaria melastoma*: "Range extended to Bermuda (F.J. Seaver & H.H. Whetzel)". And lastly, in Seaver & Waterston (1946): "However one of us (Seaver 1928: 198, 1942: 320) has shown that the Bermuda species is referable to *Bulgaria melastoma* (Sow.) Seaver. Figure 1 (upper) shows this species on rotten bark of *Juniperus bermudiana* L., from roots of living trees exposed at soil level, Walsingham, Bermuda, Jan. 20, 1922, H.H. Whetzel Bermuda Fungi No. 188. This plant is characterized by apothecia which are frequently stipitate,

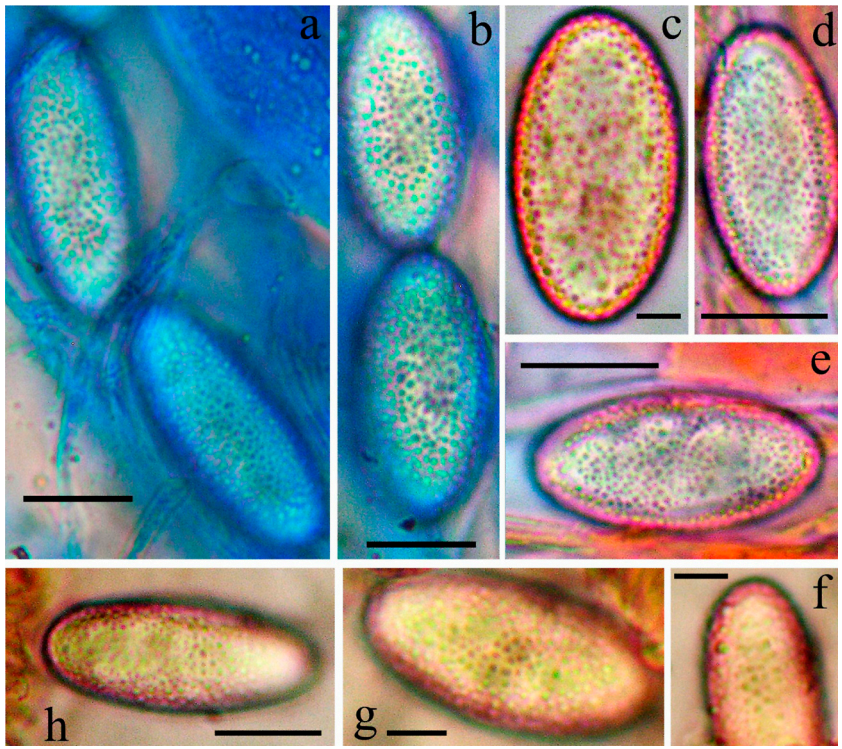

Plate 3. *Plectania seaveri* (holotype). a–b: spores in lactic Cotton blue. c–e: spores in Congo red. f–h: spores in water mount. Bars: a, b, d, e, h = 10 µm; c = 2,5 µm; f, g = 5 µm. Photos by M. Carbone.

with almost smooth, hyaline, ellipsoidal spores, 20–25 × 9–10 µ". In Waterston (1947) we find only a list of the collections studied here, under *Bulgaria melastoma*, and nothing is added from a taxonomic point of view.

Korf (1957) seems to have been the first mycologist to realize that these collections might represent a new taxon: "...the species of "*Bulgaria*" described by Seaver and Waterston (1946: 182) from Bermuda, which appears to be undescribed..." While revising the cited material we found that Denison (Korf's student) also noted Seaver's misinterpretation, for in collection CUP-033235, there is a handwritten annotation noting, "This is not *Bulgaria* (*Plectania*) *melastoma*. Note, for example, the delicately sculptured spores, the larger apothecia and longer stipe. W C Denison 2 Nov 1961".

Many years before, Boedijn (1932) suggested that Seaver's Bermudian *Bulgaria melastoma* (Seaver 1928) might be conspecific with *Sarcosoma thwaitesii* (Berk. & Broome) Petch [≡ *Galiella thwaitesii* (Berk. & Broome) Nannf.], mainly

because Petch (1910) recorded *S. thwaitesii* from Ceylon, growing on *Juniperus bermudiana*. Although Seaver & Waterston (1946) definitely misinterpreted *Bulgaria melastoma*, they demonstrated as well that Boedijn (1932) was wrong. *Galiella thwaitesii* is undoubtedly a very distinct and different taxon. For further details about *Galiella thwaitesii* see Le Gal (1953).

Species delimitation — *Plectania melastoma*, in its original sense, is definitely a very different species from the material studied here. As demonstrated by Agnello & Carbone (2012) the macroscopic, vivid orange-red, external warts are always present, and still visible to the naked eye even after 150 years; in addition, the apothecia are at most substipitate and never definitely stipitate as in *P. seaveri*. Microscopically, there are more numerous and immediately appreciable differences. For example, in *P. seaveri* the spore sculpturing is delicate but, at the same time, very distinct and pronounced. It is easily visible in water mounts (1000×, oil immersion). In contrast, in *P. melastoma,* the spores are far less sculptured and have very different warts that are barely visible, even in 1000× Cotton blue mounts. The spore shapes are also quite different — mostly subfusoid in *P. melastoma,* ellipsoid in *P. seaveri*. Lastly, *P. melastoma* produces only one kind of external hairs and a differently coloured crystalline pigmentation that is mostly a vivid orange-red-amber, not pale brownish to dark amber as in *P. seaveri*.

Unlike *Plectania melastoma*, *P. seaveri* surely belongs to *Plectania* sect. *Donadinia* (Bellem. & Mel.-Howell) M. Carbone & Agnello. In this section, Carbone et al. (2012) recognize only three stipitate species with more or less defined ornamented ellipsoid spores: *P. helvelloides* (Donadini et al.) Donadini, *P. nannfeldtii* Korf, and *P. lusitanica* (Torrend & Boud.) M. Carbone et al. As pointed out by Carbone et al. (2012), while all these species have extremely similar gross macroscopic morphologies, excipular anatomies, and external hairs, they can clearly be distinguished based on the combination of spore size and spore sculpturing. In fact, if on one hand we observe that spore measurements show a continuum from a minimum of 16 µm (in *Plectania lusitanica*) to a maximum of 38 µm (in *Plectania nannfeldtii*), on the other hand we are presently unaware of any populations that show a simultaneous overlap of spore size range and sculpturing. SEM and optical observations also support this morphological distinction.

The well-known American "snowbank mushroom," *Plectania nannfeldtii,* can be immediately differentiated by its different ecology, geographical distribution, longer spores, and different episporium (e.g. Seaver 1928; Miller 1967; Smith et al. 1981; Arora 1986; Tylutki 1993; Li & Kimbrough 1995). *Plectania lusitanica* surely shares a similar ornamentation but differs in its smaller spores and a much less pronounced spore sculpturing that is barely visible in optical microscopy (data taken from many spore prints). Lastly, *P. helvelloides* spores

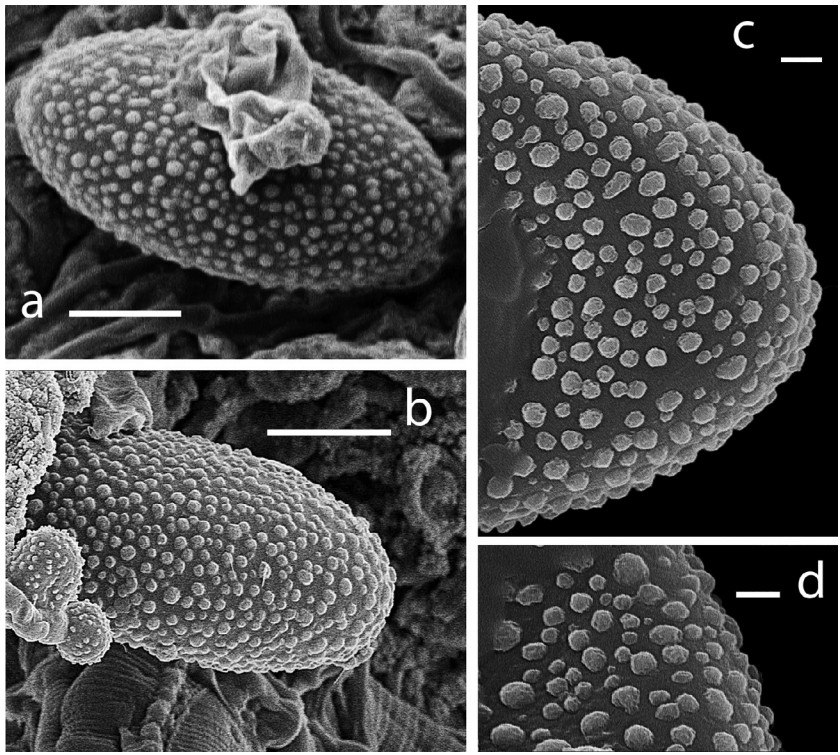

Plate 4. *Plectania seaveri* (holotype). a–b: spores. c: pole of the spore. d: spore sculpturing. Bars a–b = 5 µm; c–d = 1 µm. SEM courtesy of Mark Whitten and the Electron Microscopy Core Facility at the University of Florida (Gainesville, USA).

(22–28 × 9–13 µm; Donadini et al. 1973, Carbone et al. 2012), which might be regarded as the closest to *P. seaveri* in size, have a different ornamentation type — relatively easy to see in cotton blue (1000×), but smaller, differently shaped, and not easily detectable in outline.

The spore sculpturing seen in *Plectania seaveri* is extremely similar to that found in *Galiella* Nannf. & Korf, which, however, produces very different fleshy large gelatinous apothecia; for a quick survey, see Boedijn (1932, as *Sarcosoma*), Le Gal (1953, as *Sarcosoma*), Korf (1957), Cao et al. (1992), and Zhuang & Wang (1998). The same can be said for the genus *Neournula* Paden & Tylutki, previously placed in *Sarcosomataceae* Kobayasi but now transferred into a different family, *Chorioactidaceae* Pfister. *Neournula* does indeed possess similarly warted spores but is distinct from *Plectania* in many other features (see Pfister et al. 2008).

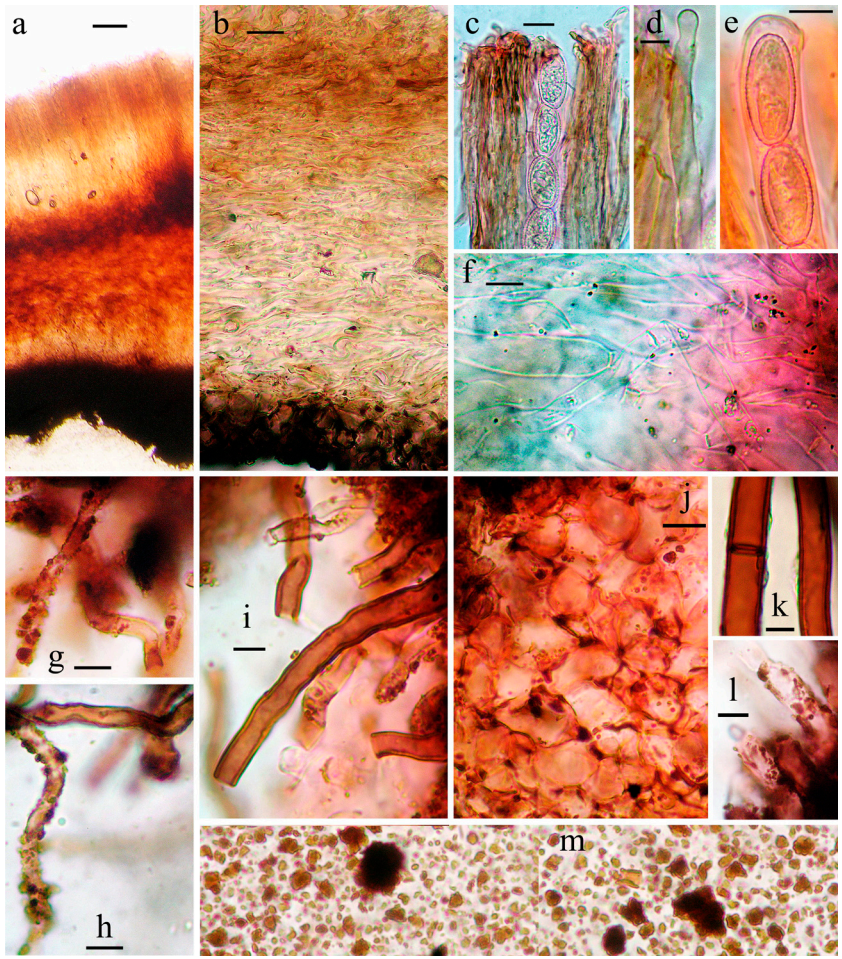

Plate 5. *Plectania seaveri* (holotype). a: section of the apothecium; b: medullary excipulum in water mount; c: bundles of paraphyses; d: seta (hymenial hair); e: ascus tip; f: medullary excipulum in Congo red (magnified); g–h: hyphoid excipular hairs; i: excipular hairs; j: ectal excipulum; k: subiculum; l: encrusted cells of the margin; m: crystals of excipular pigment. Bars: a = 100 µm; b = 20 µm; c, e, f, j = 10 µm; d, g, h, i, k, l = 5 µm. Photos by M. Carbone.

Extralimital and doubtful species — Mention must be made here of two other *Plectania* species described from the Americas. *Plectania coelopus* (Mont.) Sacc. was described from Chile and, according to the iconotype and original description, it surely belongs to sect. *Donadinia*. We agree with Le Gal (1958) and Cabello (1988) that *P. coelopus* must be regarded as a doubtful species until

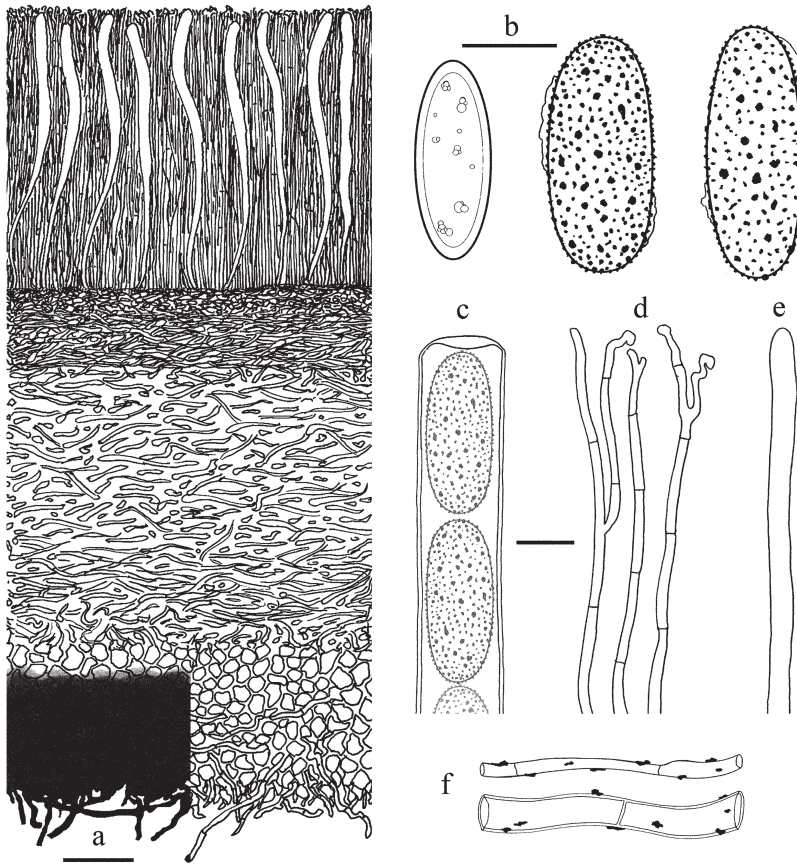

Plate 6. *Plectania seaveri* (holotype). a: section of the cup; b: spores; c: ascus; d: paraphyses; e: seta (hymenial hair); f: excipular hairs. Bars: a = 100 µm; b = 10 µm; c–f = 10 µm. Drawing by C. Agnello.

its real identity is investigated and/or it is typified with topotypical (or other Chilean) material.

Another doubtful American taxon is *Plectania rimosa* Peck, described from California (Peck 1903). Seaver (1942) treated it as a "doubtful species", and due to the (apparent) lack of original material, he stated that this species might represent *Paxina corium* (O. Weberb.) Seaver [≡ *Helvella corium* (O. Weberb.) Massee]. As the protologue does not fit *Helvella corium,* we disagree and regard *Plectania rimosa* as a nomen dubium.

During our literature searches, we have found other unclear species seemingly in need of revision (e.g., *Neournula helvelloides* sensu Zhuang & Wang (1998) and *Plectania nannfeldtii* in the sense of the Chinese authors Teng (1963), Tai (1979), and Bi et al. (1990) and the Japanese author Otani (1973). Future studies will address the misapplications of these names.

Ecology — All known *Plectania seaveri* specimens were collected from rotting bark of *Juniperus bermudiana*, an endemic Bermudian tree that historically dominated the upland forests of the Bermuda Archipelago (Thomas 2004). Most collections of this fungus were made in the 1920s, with the last known specimen collected in 1940 (Waterston, 1947). A search of 125 fungus specimens from the last major Bermudian mycological expedition collected by Richard Korf and colleagues during the week of January 17–23, 1980 (unpublished; specimens deposited at CUP) revealed no additional *P. seaveri* specimens.

Between 1946 and 1953, introduced scale insects decimated the Bermuda cedar, killing an estimated 95% (Phillips 1984), thus deforesting the islands and altering the ecology of many other Bermudian species in major ways (Thomas 2004). With no known *Plectania seaveri* specimens collected since 1940, it seems probable that this fungus is now extinct. While it may still occur on some of the dead, non-decorticated trunks of Bermuda cedar that now dot the Bermudian landscape (and perhaps on the few living Bermuda cedars which still persist), it seems likely that the fate of *P. seaveri* may eventually follow that of other endemic Bermudian organisms dependent on Bermuda cedar, such as the Bermuda cicada (*Tibicen bermudiana*) and the ochre-banded looper moth (*Semiothisa ochrifascia*) (Sterrer 1998).

As far as we know, all species of *Plectania* sect. *Donadinia*, including *P. seaveri*, associate as saprophytes with coniferous trees. Future collections of representatives of this section would help us understand whether this coniferous association is taxonomically relevant.

Final considerations — This study demonstrates that *Bulgaria melastoma* as interpreted by Seaver (1928) needs an in-depth revision. Paden & Tylutki (1969) observed that "in Seaver, *P. milleri* will key out to *P. melastoma* (as *Bulgaria melastoma*)." We can speculate that, at least in the case of *Plectania seaveri*, Seaver might have misinterpreted the encrusting pigment of the external surface, indicating that he was not aware of the true diagnostic characters of *P. melastoma*. In any case, we are convinced that Seaver misapplied the name *P. melastoma* to other specimens, such as one from Puerto Rico (CUP-PR-001281, examined as part of the current study), and probably also the Jamaican collection cited by Seaver & Waterston (1946). The latter specimen, supposedly deposited in NY, could not be located during a recent visit to that herbarium (SLG pers. obs., April 2, 2012). Our future studies will attempt to answer these lingering questions.

Acknowledgments

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