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Epitypification of *Ceratocystis fimbriata*

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Abstract: *Ceratocystis* accommodates many important pathogens of agricultural crops and woody plants. *Ceratocystis fimbriata*, the type species of the genus is based on a type that is unsuitable for a precise application and interpretation of the species. This is because no culture or DNA data exist for the type specimen. The aim of this study was to select a reference specimen that can serve to stabilize the name of this important fungus. We selected a strain, CBS 114723, isolated from sweet potato in North Carolina, USA, in 1998 for this purpose. The strain was selected based on the availability of a living culture in a public depository. A draft genome sequence is also available for this strain. Its morphological characteristics were studied and compared with the existing and unsuitable type specimen as well as with the original descriptions of *C. fimbriata*. The selected strain fits the existing concept of the species fully and we have consequently designated it as an epitype to serve as a reference specimen for *C. fimbriata*.

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INTRODUCTION

The genus *Ceratocystis* (*Ceratocystidaceae*, *Microascales*) was established based on *C. fimbriata* (Halsted 1890). The sexual morph is characterized by ascomata with elongated necks and ascospores enveloped in a hat-shaped sheath (Van Wyk & Wingfield 1991, Van Wyk *et al.* 1991). The asexual morph produces three different conidial forms; hyaline cylindrical and barrel-shaped conidia in chains, which are thielaviopsis-like (Paulin-Mahady *et al.* 2002) and dark aleuriospores formed singly or in chains.

In the past two decades, the number of species in *Ceratocystis sensu lato* has increased considerably, driven by the application of DNA-based sequence analyses and broader sampling. Based on phylogenetic inference supported by ecological and morphological differences, *Ceratocystis* spp. were grouped in recognizable lineages including three species complexes; the *C. coerulescens* complex, the *C. fimbriata* complex and the *C. moniliformis* complex (Wingfield *et al.* 1994, 2013, Harrington *et al.* 1996, Johnson *et al.* 2005, Van Wyk *et al.* 2006). These complexes and lineages were subsequently assigned to seven different genera in the *Ceratocystidaceae* based on multi-gene region analyses, and includes *Ceratocystis sensu stricto* (De Beer *et al.* 2014). An additional eight genera have recently been added to the family (De Beer *et al.* 2017, 2018, Mayers *et al.* 2015, 2020, Nel *et al.* 2018). The currently accepted generic concept of *Ceratocystis* is limited to the species that previously resided in the *C. fimbriata* complex and most of these are pathogens of angiosperm plants (Van Wyk *et al.* 2013, Wingfield *et al.* 2013, De Beer *et al.* 2014).

Ceratocystis presently includes 42 species (Marin-Felix *et al.* 2017, Barnes *et al.* 2018, Liu *et al.* 2018, Holland *et al.* 2019, Cho *et al.* 2020). The genus can be distinguished from other closely-related genera by the lack of ornamentations on the ascomatal bases and ellipsoidal ascospores surrounded by a hat-shaped sheath (Van Wyk *et al.* 1991, 1993, De Beer *et al.* 2014).

Ceratocystis fimbriata was introduced to name the causal agent of the important black rot disease of sweet potato in the late 19th century in New Jersey, USA (Halsted 1890). Halsted (1890) provided an extensive description of the fungus without providing measurements of characteristic structures and made no mention of a type specimen being designated. Halsted & Fairchild (1891) subsequently published a more detailed description of the fungus including measurements of the structures. Three herbarium specimens deposited by the original authors are maintained in the US National Fungus Collection (BPI) and the New York Botanical Garden (NY), respectively with the accession numbers BPI 595863, BPI 595869 and NY 01050464. The specimen BPI 595869 was collected by B.D. Halsted on 28 Nov. 1890 without a location being specified. This specimen was examined by Baker-Engelbrecht & Harrington (2005) and annotated as “crumbled powder and useless”. These authors consequently designated the specimen BPI 595863 (Fig. 1A–C) as a neotype with an assumption there was no extant original material for the fungus. This neotype was collected by B.D. Halsted on 12 April 1891 from Swedesboro, New Jersey, USA, and includes dried leaves, stolons and diseased shoots of sweet potato together with the 1890 illustrations of B.D. Halsted. The specimen NY 01050464 collected by B.D. Halsted in September 1890 also from Swedesboro, New Jersey, USA (Fig. 1D, E) was



Fig. 1. Specimen pouches and contents linked to *Ceratocystis fimbriata*. **A–C.** Superseded neotype (BPI 595863). **D, E.** Holotype (NY 01050464). **A.** Envelope containing the specimen designated as neotype. **B.** An original note in the pouch. **C.** Black rot (circles) on stolons of sweet potato. **D.** Envelope containing the holotype specimen. **E.** Sweet potato peels. (Images D, E are from <http://sweetgum.nybg.org/images3/1700/343/01050464.jpg>).

not considered in the study of Baker-Engelbrecht & Harrington (2005). No living cultures have been connected to any of the three fungarium specimens of *C. fimbriata*.

As the type genus of the *Ceratocystidaceae* and the type species of *Ceratocystis*, the specimen of *C. fimbriata* chosen to appropriately represent the type is crucially important (Turland et al. 2018). This raises a problem because the existing specimens linked to the name fail to serve this purpose due to the lack of living cultures or DNA sequence data. We consequently propose an epitype for *C. fimbriata* for which the interpretation of this important taxon can precisely be applied.

MATERIALS AND METHODS

A strain of *Ceratocystis fimbriata* (CBS 114723) was obtained from the culture collection (CBS) of the Westerdijk Fungal Biodiversity Institute, Utrecht, the Netherlands. It was isolated from sweet potato (*Ipomoea batatas*) in North Carolina, USA in 1998 by A. McNew and deposited by C. Baker-Engelbrecht. The specimen designated as neotype of *C. fimbriata* (BPI 595863) was borrowed from the US National Fungus Collections (BPI, Department of Agriculture, Beltsville, Maryland, USA) in order to conduct morphological comparisons between the strain (CBS 114723)

and the type. The specimen NY 01050464 (New York Botanical Garden, Bronx, New York, USA) could not be obtained due to the lockdown of the herbarium commencing in March 2020. It was, however, possible to examine the image of the specimen (<http://sweetgum.nybg.org/images3/1700/343/01050464.jpg>) provided by the C.V. Starr virtual herbarium.

In order to observe the infection process and symptom developed associated with the strain CBS 114723 and to compare its morphological characteristics with the neotype specimen, fresh sweet potato tubers were inoculated under laboratory conditions. Ten locally grown sweet potatoes without blemishes were chosen for this purpose. The tubers were surface-disinfested by immersion in 2.5 % hypochlorite for 10 min and air-dried in a laminar flow cabinet. A syringe needle was then used to make 1 mm-deep small punctures on the surface of the tubers. A spore suspension was prepared in 10 mL sterilized water by diluting an ascospore mass taken from the tip of an ascogonium. A drop of this suspension from a 1 mL Eppendorf tip was then applied to the puncture holes on the surface of the inoculated tubers and these were left exposed. The inoculated sweet potatoes were placed in two plastic containers lined with tissue paper that was dampened with sterile distilled water, covered with a lid, and maintained in the dark at 25 °C. The progression of infection was observed daily.

The strain CBS 114723, used for the inoculation, was grown on sweet potato broth agar medium (SPA: 10 g Difco agar, 250 mL sweet potato broth, 750 mL ionized water) which was used by Halsted & Fairchild (1891) for their morphological observation of *C. fimbriata*. The cultures were incubated in near-UV light at 21 °C.

Fungal structures formed in SPA and the sweet potato tubers were removed from the substrate and mounted on glass slides in water which was later replaced with 85 % lactic acid for observation. Structures from the neotype were mounted in 10 % KOH for microscope examination. Zeiss microscopes (Axioskop 2 Plus and Stemi SV6, Germany) were used to study the microscopic features including fruiting structures and spores. Images were captured using an AxioCam ICc 3 camera mounted on the microscopes. Measurements were made using an Axiovision v. 4.5 software. Twenty-five to 50 measurements were made of each structure based on the availability of the structures and written in minimum–maximum (mean±standard deviation). The dimensions of ascospores were measured excluding the sheath.

To demonstrate the phylogenetic relationship between the sweet potato isolate of *C. fimbriata* epitypified in this paper, other sweet potato isolates and *forma specialis* (Valdetaro *et al.* 2019) available in GenBank, and closely related species (Fourie *et al.* 2015, Marin-Felix *et al.* 2017) a phylogenetic tree was constructed based on the ITS gene regions using maximum parsimony analysis as described in Fourie *et al.* (2015).

RESULTS

Holotype

The designation of the neotype (BPI 595863) by Baker-Engelbrecht & Harrington (2005) should be considered superfluous. This is because the NY specimen (NY 01050464) is an appropriate holotype of *C. fimbriata* linked to the name. The specimen contains well-preserved sweet potato peels from Swedesboro, New Jersey, the same location where the original description was made. The collection date is September 1890, which is prior to the publication of the protologue in November 1890 (Halsted 1890). The two BPI specimens were collected (BPI 595869 in November 1890 and BPI 595863 in April 1891) after Halsted's 1890 paper. The NY specimen pouch is labelled as "*Ceratocystis fimbriata* Ellis & Halsted" as is the case for the protologue. That specimen was originally part of the herbarium of J.B. Ellis, which was incorporated to the New York Botanical Garden Steere 'fungus' fungarium (NY). Although the designation of the type was not explicitly "cited" in the protologue, there is sufficient circumstantial evidence that B.D. Halsted and J.B. Ellis studied this specimen. This makes the NY specimen the only existing material linked to the protologue and consequently is the holotype. This complies with the Code, Art. 9. Note 1 (Shenzhen) "...If the author used only one specimen or illustration, either cited or uncited, when preparing the account of the new taxon, it must be accepted as the holotype...".

Symptom development and morphology

The sweet potato tubers inoculated with the strain CBS 114723 developed black-rot symptoms and fungal structures within 2

wk, on tubers as well as on the newly developing bud sprouts (Fig. 2). Dark sunken lesions developed from the inoculation points and spread to young shoots as the tubers began to sprout.

The strain CBS 114723 produced both sexual and asexual structures readily on both sweet potato and SPA. The morphological characteristics of these structures on both substrates were almost identical, but on SPA the strain produced longer ascomatal necks (Table 1).

The material designated as neotype (BPI 595863) was in poor condition and only a small number of damaged ascomata and asexual structures could be retrieved for observation (Fig. 1C). However, where comparisons could be made, the morphological characteristics of CBS 114723 and BPI 595863 were very similar (Figs 2, 3, Table 1). The holotype specimen (NY 01050464) was viewed in the image and dark sunken lesions were noted (Fig. 1E)

The descriptions and figures prepared by Halsted (1890) and Halsted & Fairchild (1891) were used for comparison (Figs 2, 3, Table 1). Morphological characteristics of fruiting structures did not show notable variation between the "neotype", the strain CBS 114723 on sweet potato and on SPA, and the descriptions by Halsted (1890) and Halsted & Fairchild (1891). This was with the exception of the ascomatal necks, which tend to become longer with prolonged incubation time and the dimensions of ascospores. The ascospores of the "neotype" strain (5–7.5 × 3.5–5.5 μm), the strain CBS 114723 on sweet potato (5.5–7 × 4–5 μm) and on SPA (5–6.5 × 4–5 μm) were slightly smaller than those presented in the description (5–9 × 5–9 μm) by Halsted & Fairchild (1891).

Nomenclature

Ceratocystis fimbriata Ellis & Halst. *Bull. New Jersey Agric. Exp. Sta.* **76**: 14. 1890. Figs 2–4.

Synonyms: Sphaeronaema fimbriatum (Ellis & Halst.) Sacc., *Syll. Fung.* **10**: 215. 1892.

Ceratostomella fimbriata (Ellis & Halst.) Elliott, *Phytopathology* **13**: 56. 1923.

Ophiostoma fimbriatum (Ellis & Halst.) Nannf., *Svenska Skogsvårdsfören. Tidskr.* **32**: 408. 1934.

C. fimbriata f. sp. *ipomoea* Valdetaro *et al.*, *Fungal Biol.* **123**: 181. 2019.

?= *Rostrella coffeae* Zimm., *Meded. Lands Plantentuin, Batavia* **37**: 32. 1900.

Ophiostoma coffeae (Zimm.) Arx, *Antonie van Leeuwenhoek* **18**: 210. 1952.

Ceratocystis moniliformis f. *coffeae* (Zimm.) C. Moreau, *Bull. Sci. Minist. France Outre-Mer* **5**: 424. 1954.

Epitypification

The existing **holotype** NY 01050464 and the superseded **neotype** BPI 595863 of *C. fimbriata* fail to serve the needs of researchers that must consider various questions relating to its taxonomy, biology and population biology using DNA-based methods. An appropriate epitype is consequently provided for *C. fimbriata*. The strain chosen for this purpose originated (broadly) from the area where the species was first described in the USA and from the original host plant, *Ipomoea batatas*. This particular strain sporulates profusely and its genome has been sequenced (Wilken *et al.* 2013).

Epitypus of *Ceratocystis fimbriata* (designated here): **USA**, North Carolina, isolated from sweet potato (*Ipomoea batatas*), 1998, A.

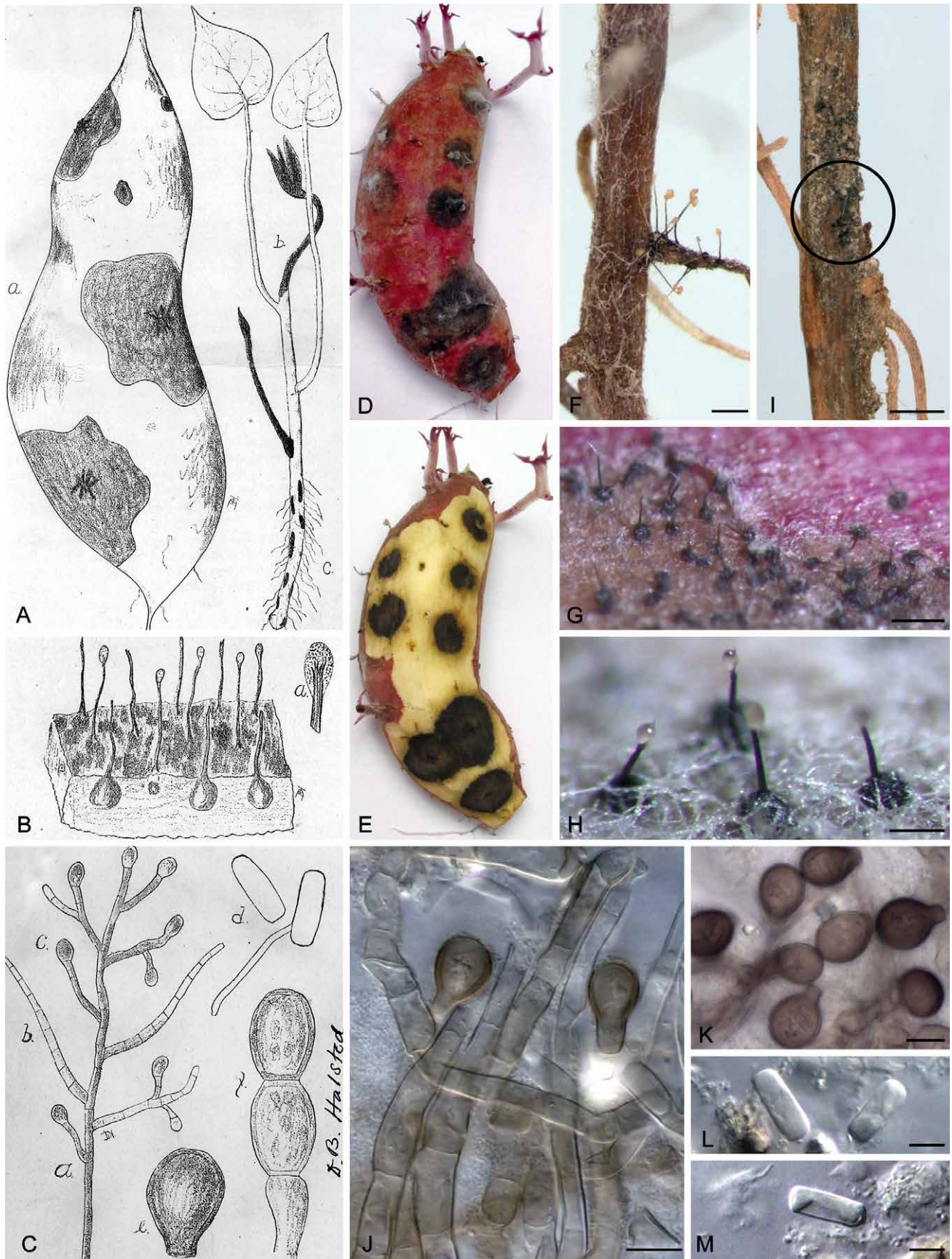


Fig. 2. *Ceratocystis fimbriata* infecting a sweet potato. **A–C.** Drawings from Halsted (1890). **D–H.** Sweet potato tuber inoculated with *C. fimbriata* (Ex-epitype, CBS 114723). **I–M.** Superseded neotype (BPI 595863) of *C. fimbriata*. **A.** Sweet potato and young shoot showing the black rot symptom. **B.** Ascomata with a long neck exuding a slimy droplet of spores at its tip, and the ascomatal base embedded in the substrate. **C.** Aleuriospores and their conidiophores, and thielaviopsis-like conidia (marked as 'd'). **D, E.** Sweet potato showing fully developed black-rot lesions 2 wk after inoculation. **F.** Ascomata on young shoot. **G.** Ascomata produced in the lesion on the surface of the tuber. **H.** Close-up of ascomata with a slimy droplet of ascospores. **I.** Ascomata (circle) with broken necks in dried shoot. **J.** Pigmented thielaviopsis-like conidiophores. **K.** Aleuriospores. **L, M.** Thielaviopsis-like conidia. Scale bars: F = 250 μ m; I = 1 mm; G, H = 200 μ m; J, K = 10 μ m; L, M = 5 μ m.

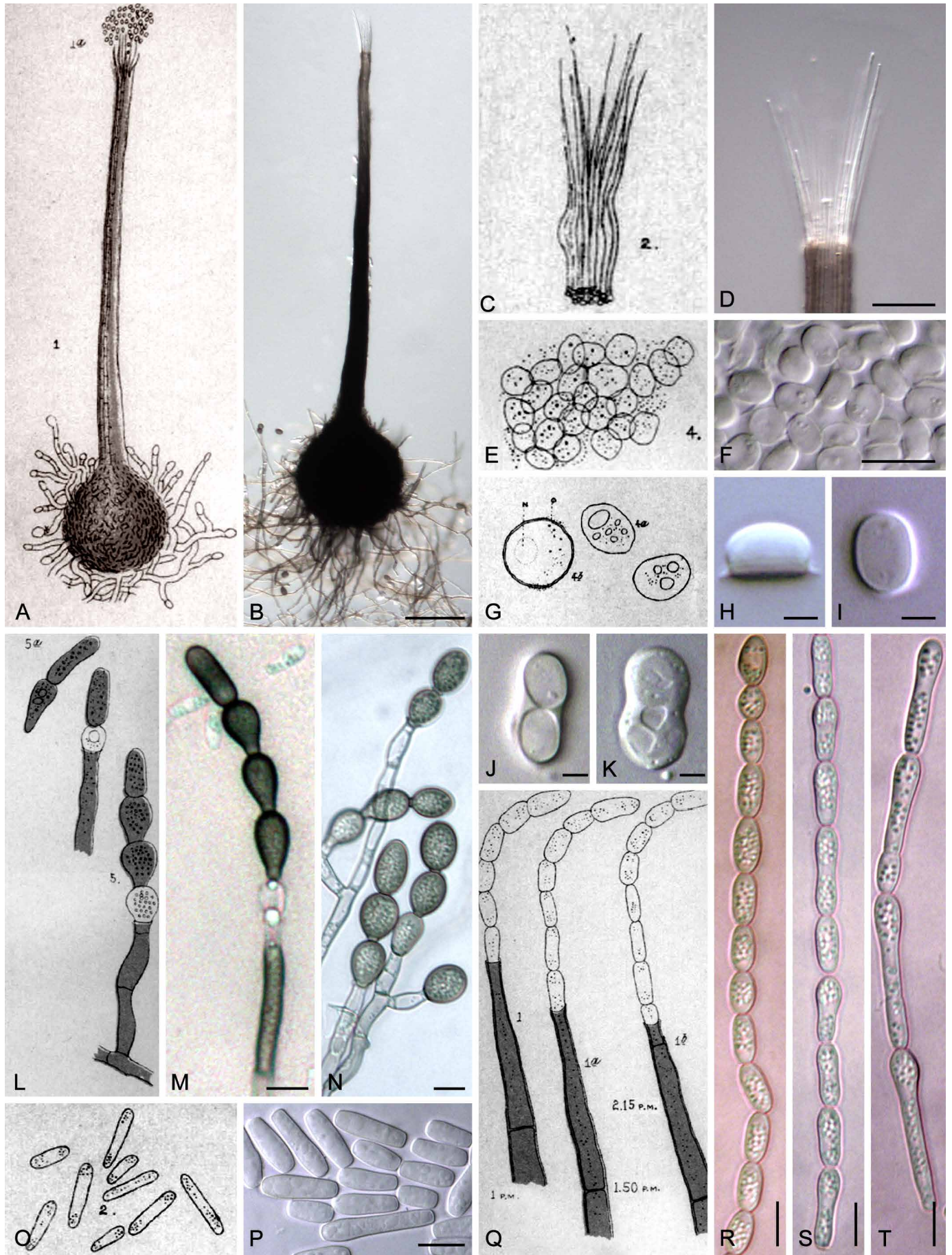


Fig. 3. Microscopic features of *Ceratocystis fimbriata* growing on sweet potato broth agar medium (SPA). A, C, E, G, L, O, Q: Drawings of Halsted & Fairchild (1891). B, D, F, H–K, M, N, P, R–T: Images of the ex-epitype (CBS 114723). A, B. Ascusa. C, D. Divergent ostiolar hyphae. E, F. Clustered ascospores. G–K. Ascospores (K focused on a sheath of J). L–N. Aleuriospores in chains. O, P. Thielaviopsis-like conidia. Q–T. Thielaviopsis-like conidia in chains. Scale bars: B = 100 μm; D = 25 μm; F, M, N, P, R–T = 10 μm; H–K = 2.5 μm.

Table 1. Comparison of morphological characteristics of the type specimens and ex-epitype strains of *Ceratocystis fimbriata*.

Reference ¹	Superseded neotype	Ex-epitype	Ex-epitype	Holotype (NY 01050464)
Substrate or medium	Young stems of sweet potatoes	Sweet potato tuber and stems (9 d)	Sweet potato broth agar medium (SPA, 30 d)	Sweet potato broth agar medium (SPA)
Origin of the strain	New Jersey, USA	North Carolina, USA	North Carolina, USA	New Jersey, USA
Figures in the photoplates	Figs 1A–C, 2I–M	Fig. 2D–H	Fig. 3B, D, F, H–K, M, N, P, R–T	Figs 1D, E, 2A–C, 3A, C, E, G, L, O, Q
Ascomata (ascomatal base)	Not examined due to a limited number of structures	175.5–266.5 × 174–269.5 µm when pressed	157–230.5 × 141–228.5 µm	96–224 × 96–224 µm, described as “pycnidia”
Necks	Not examined due to a limited number of structures	129–422 µm long (380–850 µm long in 27 d), 17–34 µm wide at base, 15–26 µm wide at apex	367.5–876.5 µm long, 31.5–45 µm wide at base, 17.5–25 µm wide at apex	395–608 µm long, 24–34 µm wide at base, 14–20 µm wide at apex
Ostolar hyphae	Not examined due to a limited number of structures	28–120 µm long, 1.5–4 µm wide at base, 1.5–3 µm wide at apex	46.5–90 µm long, 1.5–3 µm wide at base, 1–1.5 µm wide at apex	30–60 µm long
Asci	Not observed	Not observed	Not observed	Not mentioned
Ascospores ²	5–7.5 × 3.5–5.5 µm, hyaline, subglobose, a mass of ascospores present as a thin film	5.5–7 × 4–5 µm in face view, subglobose, 3.5–4 µm high in side view, hyaline, hat-shaped in sheath	5–6.5 × 4–5 µm in face view, subglobose, 3–4 µm high in side view, hyaline, hat-shaped in sheath	5–9 × 5–9 µm and swell to 12–17 × 9–15 µm in water for several hours (possibly lateral extension of sheath), hyaline, globose or oblong, described as “pyncospores”
Conidiophores	Not observed	Not observed	Terminal or intercalary, sub-hyaline to pale brown, cylindrical, straight or curved, consisting of a few cells	60–160 µm long, 6–7 µm wide at widest, multiseptate, somewhat fusiform, greenish brown with paler colored tips, described as “primary sporophores”
Conidiogenous cells (phialides)	Not observed	47–76 µm long, 4.5–6 µm wide at base	36.5–68 µm long, 4.5–8.5 µm wide at base, 3–5 µm wide at apex	not separately described by authors but description was included in conidiophores
Conidia of thielaviopsis-like asexual morph	8.5–17.5 × 3.5–5.5 µm, hyaline, oblong to cylindrical with truncate ends	8.5–20.5 × 3.5–5.5 µm, hyaline, oblong to cylindrical with truncate or slightly round ends	6–26 × 3–5 µm, hyaline, oblong to cylindrical with truncate or slightly round ends, at times inflated through the whole cell or partly	16–30 × 4–9 µm, hyaline, bacillary, sometimes oblong or clavate, described as “hyaline or microconidia”
Aleuriospores	15–18 × 10.5–13 µm	11.5–19 × 9.5–14.5 µm, pale brown, ellipsoidal to subglobose, base truncated or often elongated in oblong or rectangular shape	9.5–18.5 × 6–11.5 µm, pale brown, oblong or ellipsoidal to subglobose with base truncated or often elongated in oblong or rectangular shape, formed single or in chains	12–19 × 6–13 µm (mostly 10–11 × 12–15 µm), hyaline becoming brown with age, first conidium oblong-ovate, succeeding ones globose or elliptical with a small pedicel or extension at lower extremity, described as as “olive or macroconidia”
Conidiophores of aleuriospores	Not observed	Not observed	Micronematous or semi-macronematous, terminal or intercalary, hyaline to pale brown, oblong to cylindrical, 0–several septated	Simple, septate, branching, frequently indistinguishable from vegetative hyphae or “primary sporophores”

Table 1. (Continued).

Reference ¹	Superseded neotype	Ex-epitype	Ex-epitype	Holotype (NY 01050464)
(BPI 595863)	(CBS 114723 = CMW 14799)	(CBS 114723 = CMW 14799)	(CBS 114723 = CMW 14799)	Halsted (1890), Halsted & Fairchild (1891) (isolate not specified)
Conidiogenous cells of aleuriospores	Not observed	4.5–26 × 4.5–8.5 µm, sub-hyaline to pale brown, doliiform to cylindrical phialides	9–30 × 3.5–6.5 µm	Not mentioned

¹BPI = US National Fungus Collections, Beltsville, USA; CBS = the culture collection of the Westerdijk Fungal Diversity Institute, Utrecht, the Netherlands; CMW = the culture collection of the Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Pretoria, South Africa; NY = New York Botanical Garden, Bronx, New York, USA.

²The measurements represent the ranges without sheath except for the holotype.

McNew (**epitype** CBS H-21516, dried culture of CBS 114723, culture ex-epitype CBS 114723 = C1421 = CMW 14799, Genome Accession No. APWK03000000, MycoBank MBT392678).

Geographical distribution: Australia, China, Haiti, Hawaii, Indonesia, Japan, Republic of Korea, Malaysia, New Zealand, Papua New Guinea, Southern Caribbean (Saint Vincent and Grenadines), USA (Baker 1926, Harter & Weimer 1929, Sy 1956, Baker *et al.* 2003, Steimel *et al.* 2004, Johnson *et al.* 2005, Kajitani & Masuya 2011, Li *et al.* 2016, Paul *et al.* 2018, Cho *et al.* 2020).

Description of the epitype

Colonies on SPA at 30 d. Sexual morph present. *Ascomatal bases* partly immersed in medium, subglobose, dark-brown to black, covered with dark hyphae, 157–230.5 (187.44 ± 18.71) µm long, 141–229 (184.73 ± 22.33) µm wide. *Ostiolar necks* cylindrical gradually tapering towards apex, straight or slightly curved, composed of dark hyphae, becoming pale brown towards apex, 367.5–876.5 (659.85 ± 124.51) µm long, 31.5–45 (37.70 ± 3.26) µm wide near base, and tapering to 17.5–25 (20.85 ± 2.38) µm wide just below ostiolar hyphae. *Ostiolar hyphae* divergent, straight with blunt apex, hyaline, 46.5–90 (61.06 ± 9.96) µm long, 1.5–3 (2.12 ± 0.3) µm wide near base and tapering towards apex to 1–1.5 (1.19 ± 0.09) µm wide. *Asci* evanescent. *Ascospores* hyaline, pale yellow in mass, subglobose in face view, 5–6.5 × 4–5 (5.91 ± 0.32 × 4.54 ± 0.23) µm, hat-shaped in side view, 3–4 (3.61 ± 0.32) µm high, surrounded by sheath. Asexual morph present, producing 2 types of conidia: thielaviopsis-like and aleuriospores. Thielaviopsis-like. *Conidiophores* macronematous, terminal or intercalary, sub-hyaline to pale brown, cylindrical, straight or curved, consisting of a few cells. *Conidiogenous cells* endoblastic, mostly occurring single, hyaline to sub-hyaline, collarette indistinct, 36.5–68 (53.82 ± 7.34) µm long, 4.5–8.5 (5.76 ± 0.95) µm wide near base, gradually tapering towards apex to 3–5 (4.04 ± 0.42) µm wide. *Conidia* hyaline, oblong to cylindrical with truncate or slightly round ends, at times inflated throughout the whole cell or partly, 6–26 (13.31 ± 4.46) µm long, 3–5 (4.04 ± 0.51) µm wide, in chains, guttulated. Aleuriospores. *Conidiophores* micronematous or semi-macronematous, terminal or intercalary, hyaline to pale brown, oblong to cylindrical, 0–several-septate. *Conidiogenous cells* sub-hyaline to pale brown, terminal or intercalary, discrete or less often integrated, doliiform to cylindrical, straight or curved, 9–30 (16.97 ± 5.30) µm long, 3.5–6.5 (4.93 ± 0.71) µm wide. *Conidia* pale brown, oblong or ellipsoidal to subglobose with base truncated or often elongated in an oblong or rectangular shape, occurring single or in chains, some immersed in medium, 9.5–18.5 × 6–11.5 (13.99 ± 2.02 × 9.19 ± 1.20) µm.

Notes: The newly selected ex-epitype strain was inoculated on sweet potato tuber and cultured on SPA to provide a basis for comparisons of morphological characteristics with the “neotype” (Baker-Engelbrecht & Harrington 2005) and the original descriptions (Halsted 1890, Halsted & Fairchild 1891). The overall morphological characteristics provided in the original descriptions, the “neotype” and the epitype are almost identical with only minor variation in the dimensions of ascomatal necks and ostiolar hyphae. The ascospore dimensions of the ex-epitype strain were smaller than those in the original description (Halsted & Fairchild 1891), which might be due to the fact that we excluded the ascospore sheath from our measurements.

Phylogenetic analysis

The sweet potato isolates of *C. fimbriata* from various geographical locations grouped in a clade with the ex-epitype strain (CMW 14799, GenBank accession no. KC493160) in a phylogenetic tree based on sequences of the internal transcribed spacer regions (Fig. 4). This provides supporting evidence for their common identity.

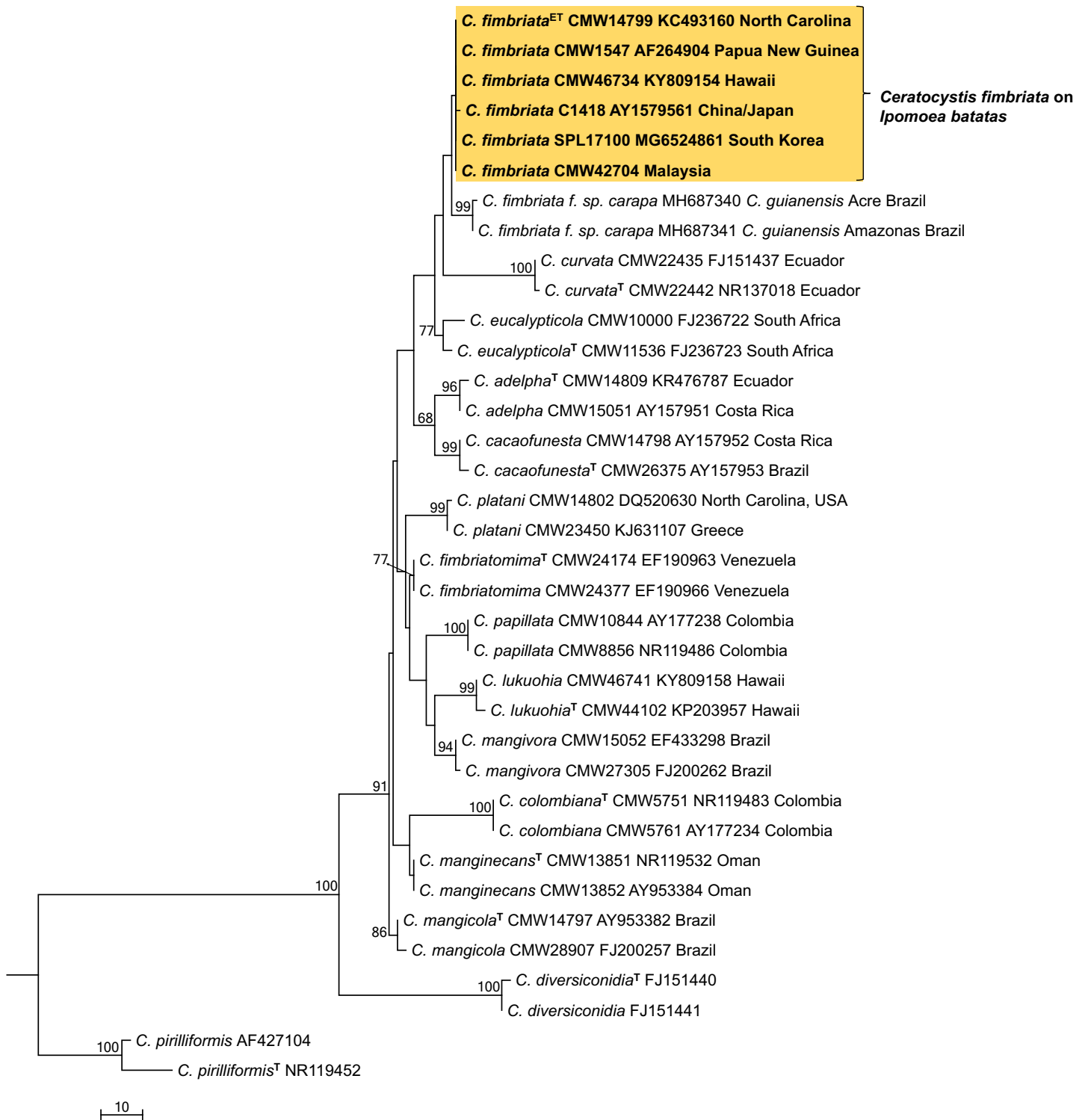


Fig. 4. *Ceratocystis fimbriata* and closely related species in *Ceratocystis*. One of the most parsimonious trees selected from maximum parsimony analysis of ITS sequence data of *Ceratocystis fimbriata* from *Ipomoea batatas*, and closely related species. Bootstrap support values above 60 % are indicated above or below branches. The ex-epitype or ex-type strain is indicated by ET or T, respectively.

DISCUSSION

No ex-type strain exists for *C. fimbriata*. When Baker-Engelbrecht & Harrington (2005) neotypified the fungus, three strains of the fungus from sweet potato were studied. None of these strains were designated as a type strain to be used as an interpretative specimen for the species and none were deposited in a publicly available culture collection. This motivated the present study to select a specific strain to serve

as an epitype for *C. fimbriata*, which, as the type species, must provide a basis for comparison with all other species in the genus. The strain CBS 114723 was selected as the epitype based on the fact that it comes from a similar origin and the same host as the strain used in the original descriptions (Halsted 1890, Halsted & Fairchild 1891). It is also the strain that was selected for full genome sequencing (Wilken *et al.* 2013) and has been featured in 13 publications representing *C. fimbriata* (Google Scholar dated on 1 April 2020).

Inoculation of sweet potato tubers in this study showed that the *C. fimbriata* epitype strain is not only viable but it has retained a high level of pathogenicity to its original host. Inoculated tubers produced both the sexual and asexual states that match the holotype (see Halsted 1890, Halsted & Fairchild 1891 for a full description) and the “neotype” (Baker-Engelbrecht & Harrington 2005).

The strain chosen to establish an epitype for *C. fimbriata* was obtained from North Carolina. While this is on the same continent and from the same host as that representing the original description, it would have been preferable to use material from New Jersey where the species was first collected (Ariyawansa *et al.* 2014). The following explanations justify the selection of the strain CBS 114723 as the epitype. At the time of its first report of black rot of sweet potato caused by *C. fimbriata*, the disease was already widespread and found “... in nearly all portions of the State where sweet potatoes were grown...” (Halsted & Fairchild 1891). Contemporary studies on *C. fimbriata* from sweet potato have shown that all strains from this host show minor genetic variation, possibly representing a single clonal lineage (Li *et al.* 2016). There would consequently be very little difference between them, assuming that they have not hybridized with other species.

The morphological characteristics of the epitype chosen in this study were compared with the material designated as neotype specimen BPI 595863 and the original descriptions provided by Halsted (1890) and Halsted & Fairchild (1891). There was limited access to the holotype NY 01050464, although the virtual image of that specimen provided confidence that it represents the same fungus. Furthermore, the original description in the protologue (Halsted 1890), the measurements of characteristic structures provided in the subsequent study of Halsted & Fairchild (1981) and a careful study of one of the two existing specimens (BPI 595863) linked to the name, *C. fimbriata* provided confidence that all three specimens are of the same fungus. This is also consistent with recent molecular genetic data showing that isolates of *C. fimbriata* from sweet potato represent a clonal lineage (Li *et al.* 2016), which has also been designated (Valdetaro *et al.* 2019) as a host-specific form referred to as *C. fimbriata f. sp. ipomoea*.

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