A new species of Reichlingia (Arthoniaceae) from the grasslands of central North America

Caleb A. Morse\textsuperscript{1,3} and Douglas Ladd\textsuperscript{2}

\textsuperscript{1} Biodiversity Institute, R. L. McGregor Herbarium, University of Kansas, 2045 Constant Ave., Lawrence, KS 66047, U.S.A.; \textsuperscript{2} Missouri Botanical Garden, 4344 Shaw Blvd., St. Louis, MO 63110, U.S.A.

ABSTRACT. Reichlingia americana, a new saxicolous species, is described from Oklahoma, U.S.A., representing the first records of the genus in North America. The species is distinguished from other members of the genus by a combination of its epilithic, rimose thallus; submuriform ascospores; presence of 2′-O-methylperl catalolic acid; and unique habitat on sheltered sandstone faces. Distinctions from related species are discussed and a key to members of the genus is provided.

KEYWORDS. Arthonia, Arthothelium, biogeography, Crosstimbers, Great Plains, lichen taxonomy, Oklahoma, 2′-O-methylperl catalolic acid, sandstone.

Reichlingia Diederich & Scheid. was introduced to accommodate a single species, R. leopoldii Diederich & Scheid., initially thought by the authors to represent a sporodochial lichenicolous hyphomycete producing brown, verrucose conidia (Diederich & Scheidegger 1996). The authors proposed that the host was an unknown, sterile, sorediate lichen with a trentepohlioid photobiont. Subsequent studies revealed the putative host of R. leopoldii was actually its thallus (Diederich & Coppins 2009), and phylogenetic analyses by Frisch et al. (2014b, 2020) and Ertz et al. (2020) revised and expanded the genus to include additional, fertile species. Members of Reichlingia are united by similar morphology, septate or submuriform ascospores, and the presence of 2′-O-methylperl catalolic or perl catalolic acid; notably, only the type species bears sporodochia. The genus currently comprises six taxa variously distributed in Africa and western Europe (Cannon et al. 2020; Ertz et al. 2020; Frisch et al. 2014b, 2020). Van den Broeck et al. (2018) and Ertz et al. (2020) showed Reichlingia and unassigned members of Arthonia Ach. s.l. to be sister to Synarthonia Müll.Arg, an exclusively corticolous, mostly tropical genus comprising 20 taxa that produce evernic and psoromic acids, parietin, and unidentified xan-
thones. Collectively, Reichlingia and Synarthonia are sister to Coniocarpon DC. This clade, which has been recovered in several recent studies, has been called the Coniocarpon-Reichlingia clade (Ertz et al. 2020; Frisch et al. 2014a; Van den Broeck et al. 2018).

During fieldwork in the Crosstimbers region of the southeast Great Plains, we discovered an unusual member of the Arthoniaceae, whose morphology and chemistry are indicative of a new species of Reichlingia. We describe the species here.

MATERIALS AND METHODS

Specimens were studied dry using dissecting microscopes. Water mounts were hand sectioned with razor blades. Microscopic characters were observed in water, 10% KOH (K), 10% HNO\textsubscript{3}, 10% H\textsubscript{2}SO\textsubscript{4}, Lugol’s iodine (I) or K followed by I (KI) and images were captured and measured to the nearest 0.1 μm. Measurements are presented as a simple range, or, where sufficient material allowed, as the average (\(\bar{x}\)) ± one standard deviation (SD), bounded by the smallest and largest observed values, and followed by the sample size [n], i.e.: (smallest observed–\(\bar{x}\)–SD–\(\bar{x}\)+SD–largest observed) [n]. Specimens were analyzed using standard spot tests (reagents are abbreviated following Brodo et al. 2001) and thin layer chromatography (TLC). TLC

\textsuperscript{3} Corresponding author’s e-mail: cmorse@ku.edu

DOI: 10.1639/0007-2745-124.1.033
was performed at KANU using solvents A, B’ and C following Orange et al. (2001). Associated taxa are provided below without taxonomic authorities, following the concepts and authorities in Esslinger (2019).

TAXONOMY

**Reichlingia americana** C.A.Morse & Ladd, sp. nov. Fig. 1

**MYCOBANK** MB 838391

A saxicolous member of Reichlingia distinguished by its epilithic, rimose thallus; submuriform ascospores (12.6–)13.8–16.1(–17.9) × (5.6–)6.5–8.2 (–8.8) μm; and the presence of 2’-O-methylperlatolic acid.

**Type:** U.S.A. OKLAHOMA: Osage Co., ca. 13.3–13.8 mi N, 0.25–1 mi E of Pawhuska, Osage Wildlife Management Area: Western Wall Unit: NW part, 36.86°–36.87°N, 96.33°–96.32°W, elev. 850–970 ft, Crosstimbers canyon system with boulders and low cliffs of Pennsylvanian sandstone along South Fork Pond Creek and unnamed tributaries, 14 Mar 2018, C.A. Morse 26007 & D. Ladd (KANU 405788!, holotype; HB. LADD!, isotype; other isotypes to be distributed with *Lichenes Exsiccati Magnicamporum*).

**Description.** Thallus epilithic, thin, rimose, creamy white to greyish or greenish grey, epruinose to finely pruinose; areoles contiguous (less commonly dispersed among protruding grains of substrate), irregularly polygonal, 0.2–0.4 mm in
diam., 0.1–0.3 mm thick, ecorticate, compact felty, smooth to distinctly roughened; pruina rectangular, 2.0–12.0 × 1.1–4.4 μm; white fibrous prothallus occasionally present, or thallus sometimes blackening along contact zone with other lichens; medulla I+ blue (or I+ reddish along outer edges of areoles), KI+ blue, of thick-walled, irregularly branching hyphae, (1.1–)1.3–2.0 μm wide (measured in K), densely inspersed with and largely obscured by fine, POL+ granules (these persisting in 10% H2SO4, clearing in K); photobiont trentepohlioid, the cells 10.8–19.7 μm in diameter, irregularly dispersed through medulla. Ascomata initiating marginally, immersed in areoles, at first discrete, oblong, becoming submoniliform or continuous, linear or crescent shaped, 0.1–0.5 × 0.07 mm, eventually expanding to occupy the areole, plane and level with upper surface of adjacent areoles, 0.1–0.4 mm in diam., angular to rounded, dark brown, usually densely white pruinose and contrasting with creamy white color of areoles or rarely epruinose; epihymenium a dense layer of pruina; hypothecium wetting but with relatively high light intensities. The specific epithet refers to the known distribution of the new species.

**Chemistry.** Spot tests of thallus: K–, KC–, C–, Pd–, UV–. TLC: 2′-O-methylperlatolic acid.

**Etymology.** The specific epithet refers to the known distribution of the new species.

**Discussion.** 2′-O-methylperlatolic acid has been documented from several other genera of Arthonio-mycetes, including Cryptothecia Stirt., Glomerulo- phoron Frisch, Ertz & G.Thor, Inoderma (Ach.) Gray, Myriostigma Kremp., and Sporodophoron Frisch, Y.Ohmura, Ertz & G.Thor, as well as several species currently assigned to Arthothelium A.Massal. (Frisch et al. 2014a, 2015). Reichlingia americana differs from members of these genera in its thallus or spore morphology. However, it shares with fertile species of Reichlingia pruinose ascomata with a basally constricted exciple and a thin microtomentum formed by the free tips of the excipular hyphae, a well-developed pale hypothecium, and oblong-ovoid, hyaline, septate ascospores (Frisch et al. 2014b). A microtomentum of free tips of paraphysoids, discussed by Frisch et al. (2014b), was not evident in the new species, but this feature may be obscured by the dense layer of pruina in the epihymenium. While the new species appears to differ from most other members of Reichlingia in its rimose thallus and immersed ascomata, in other respects, particularly in its microtomentose exciple and chemistry, the species is easily accommodated within the genus.

**Distribution and ecology.** Known only from three collections, on sheltered faces of non-calcareous sandstone cliffs, in areas protected from direct wetting but with relatively high light intensities. Associated lichens include Arthonia cf. madreana, Caloplaca yuchiorum, Chrysothrix xanthina, Crespe- nea premnea var. saxicola, Dirina sp., Dirinaria frostii, Fuscia recensa, Lecanora cf. sulphurescens, Lepraria normandinoides, Pertusaria pseudocorallina and Phyllicita petraea.

**Distribution.** Reichlingia americana has been documented from several other genera of Arthonio-mycetes, including Cryptothecia Stirt., Glomerulophoron Frisch, Ertz & G.Thor, Inoderma (Ach.) Gray, Myriostigma Kremp., and Sporodophoron Frisch, Y.Ohmura, Ertz & G.Thor, as well as several species currently assigned to Arthothelium A.Massal. (Frisch et al. 2014a, 2015). Reichlingia americana differs from members of these genera in its thallus or spore morphology. However, it shares with fertile species of Reichlingia pruinose ascomata with a basally constricted exciple and a thin microtomentum formed by the free tips of the excipular hyphae, a well-developed pale hypothecium, and oblong-ovoid, hyaline, septate ascospores (Frisch et al. 2014b). A microtomentum of free tips of paraphysoids, discussed by Frisch et al. (2014b), was not evident in the new species, but this feature may be obscured by the dense layer of pruina in the epihymenium. While the new species appears to differ from most other members of Reichlingia in its rimose thallus and immersed ascomata, in other respects, particularly in its microtomentose exciple and chemistry, the species is easily accommodated within the genus.

**Distribution.** Reichlingia americana has been documented from several other genera of Arthonio-mycetes, including Cryptothecia Stirt., Glomerulophoron Frisch, Ertz & G.Thor, Inoderma (Ach.) Gray, Myriostigma Kremp., and Sporodophoron Frisch, Y.Ohmura, Ertz & G.Thor, as well as several species currently assigned to Arthothelium A.Massal. (Frisch et al. 2014a, 2015). Reichlingia americana differs from members of these genera in its thallus or spore morphology. However, it shares with fertile species of Reichlingia pruinose ascomata with a basally constricted exciple and a thin microtomentum formed by the free tips of the excipular hyphae, a well-developed pale hypothecium, and oblong-ovoid, hyaline, septate ascospores (Frisch et al. 2014b). A microtomentum of free tips of paraphysoids, discussed by Frisch et al. (2014b), was not evident in the new species, but this feature may be obscured by the dense layer of pruina in the epihymenium. While the new species appears to differ from most other members of Reichlingia in its rimose thallus and immersed ascomata, in other respects, particularly in its microtomentose exciple and chemistry, the species is easily accommodated within the genus.

**Distribution.** Reichlingia americana has been documented from several other genera of Arthonio-mycetes, including Cryptothecia Stirt., Glomerulophoron Frisch, Ertz & G.Thor, Inoderma (Ach.) Gray, Myriostigma Kremp., and Sporodophoron Frisch, Y.Ohmura, Ertz & G.Thor, as well as several species currently assigned to Arthothelium A.Massal. (Frisch et al. 2014a, 2015). Reichlingia americana differs from members of these genera in its thallus or spore morphology. However, it shares with fertile species of Reichlingia pruinose ascomata with a basally constricted exciple and a thin microtomentum formed by the free tips of the excipular hyphae, a well-developed pale hypothecium, and oblong-ovoid, hyaline, septate ascospores (Frisch et al. 2014b). A microtomentum of free tips of paraphysoids, discussed by Frisch et al. (2014b), was not evident in the new species, but this feature may be obscured by the dense layer of pruina in the epihymenium. While the new species appears to differ from most other members of Reichlingia in its rimose thallus and immersed ascomata, in other respects, particularly in its microtomentose exciple and chemistry, the species is easily accommodated within the genus.

**Distribution.** Reichlingia americana has been documented from several other genera of Arthonio-mycetes, including Cryptothecia Stirt., Glomerulophoron Frisch, Ertz & G.Thor, Inoderma (Ach.) Gray, Myriostigma Kremp., and Sporodophoron Frisch, Y.Ohmura, Ertz & G.Thor, as well as several species currently assigned to Arthothelium A.Massal. (Frisch et al. 2014a, 2015). Reichlingia americana differs from members of these genera in its thallus or spore morphology. However, it shares with fertile species of Reichlingia pruinose ascomata with a basally constricted exciple and a thin microtomentum formed by the free tips of the excipular hyphae, a well-developed pale hypothecium, and oblong-ovoid, hyaline, septate ascospores (Frisch et al. 2014b). A microtomentum of free tips of paraphysoids, discussed by Frisch et al. (2014b), was not evident in the new species, but this feature may be obscured by the dense layer of pruina in the epihymenium. While the new species appears to differ from most other members of Reichlingia in its rimose thallus and immersed ascomata, in other respects, particularly in its microtomentose exciple and chemistry, the species is easily accommodated within the genus.

**Distribution.** Reichlingia americana has been documented from several other genera of Arthonio-mycetes, including Cryptothecia Stirt., Glomerulophoron Frisch, Ertz & G.Thor, Inoderma (Ach.) Gray, Myriostigma Kremp., and Sporodophoron Frisch, Y.Ohmura, Ertz & G.Thor, as well as several species currently assigned to Arthothelium A.Massal. (Frisch et al. 2014a, 2015). Reichlingia americana differs from members of these genera in its thallus or spore morphology. However, it shares with fertile species of Reichlingia pruinose ascomata with a basally constricted exciple and a thin microtomentum formed by the free tips of the excipular hyphae, a well-developed pale hypothecium, and oblong-ovoid, hyaline, septate ascospores (Frisch et al. 2014b). A microtomentum of free tips of paraphysoids, discussed by Frisch et al. (2014b), was not evident in the new species, but this feature may be obscured by the dense layer of pruina in the epihymenium. While the new species appears to differ from most other members of Reichlingia in its rimose thallus and immersed ascomata, in other respects, particularly in its microtomentose exciple and chemistry, the species is easily accommodated within the genus.
exclusively or primarily saxicolous. Of these, four species—*A. evanescens* Övstedal, *A. diffusus* (Nyl.) Imshaug & Fryday, *A. feuereri* Aptroot & Seaward, and *A. halophilum* Follm.—produce larger or more richly muriform ascospores and different secondary metabolites (Follmann 1968; Fryday 2002; Övstedal & Gremmen 2001; Seaward & Aptroot 2004). *Allarthonium elliottii* (Vainio) Zahlbr., known only from the type collection from the Dominican Republic, has larger muriform ascospores and a trebouxiooid photobiont (Vainio 1896). The remaining saxicolous species—*Arthothelium desertorum* Aptroot & Wirth, *A. galapagoense* Huneck & Follman, *A. miesii* Van den Broeck, *A. pacificum* Follmann, *A. saxicolum* Makhija & Patw., and *A. spilomatoides* (Nyl.) Zahlbr.—produce submuriform ascospores of similar size, which differ in having 5 or more transverse septa and in lacking enlarged distal cells; these all also differ from *R. americana* in their chemistry (Aptroot & Wirth 2006; Follmann 1968; Makhija & Patwardhan 1995, 1997; Van den Broeck et al. 2017; F. Bungartz, J. Elix & D. Ertz in litt.).

A larger number of corticolous species of *Arthothelium* produce submuriform, macrocephalic ascospores of similar size to those of *Reichlingia americana*. Most of these are poorly known, bearing scant descriptions and lacking chemistry data, but several species with ascospores bearing fewer than 5 transverse septa warrant comparison. Among them, *A. impolitellum* (Nyl.) Makhija & Patw., known only from the Indian type collection, produces slightly smaller ascospores (12–14 × 4–6 μm) with a thinner hymenium, brown hypothecium, and proximal cells of the ascospores bear a longitudinal division (Makhija & Patwardhan 1995). Secondary metabolites have not been reported for *A. impolitellum*, but two other species—*A. ramosum* Makhijan & Patw. and *A. fuscoroseum* Makhija & Patw.—differ in their chemistry and thalline pigments, respectively (Makhija & Patwardhan 1995). The North American species *Arthothelium leucastrae* Tuck. also produces submuriform, macrocephalic ascospores, which bear 4 transverse septa and are 12–16 × 5–7 μm fide Tuckerman (1872; width 5.5–7.5 μm fide Fink 1935). *Arthothelium leucastrae* has not been the subject of a modern study and was not available for in-person examination by the authors, but reportedly differs in producing a dark brown hypothecium (Tuckerman 1872; Fink 1935). Images of type material (U.S.A. TEXAS: 1848, Wright s.n. [FH, HB TUCK. 3618; MICH 74911]) also indicate that the ascomata of this species differ from those of *R. americana*, although *A. leucastrae* may also belong in *Reichlingia*.

*Reichlingia americana* represents the first member of the genus to be reported from the Americas, although the phylogenetic data in Van den Broeck et al. (2018) indicate that the corticolous species *Arthothelium anglica* Coppins and an undescribed taxon from Florida (*Arthonia* sp. 9090 of Van den Broeck et al. 2018) may also belong to the genus. *Arthothelium anglica* is locally common in more mesic woodlands in regions east of the range of *R. americana*, and differs from other members of *Reichlingia* by its chemistry, producing gyrographic acid. The biogeographic implications of a distribution encompassing western Europe, Africa and central North America are unclear and may be an artifact of low numbers of collections. The lichen biota of the Crosstimbers region includes significant influences from both a better-studied southeast Coastal Plain element, and a comparatively poorly known southwestern element, as well as a smaller, apparently endemic element (e.g., Brodo et al. 2001; Morse & Sheard 2020). Several trentepohlioid saxicolous lichens with similar ecological requirements occurring in western Europe are widely distributed in the southeast Great Plains, southern Ozarks, and Appalachians. Included among these are *Creponea premnea* var. *saxicola* and *Enterographa hutchinsiae*.

**Other specimens examined.** U.S.A. OKLAHOMA: Osage Co., 4.5–5 mi S, 1.5–2 mi E of New Prue, City of Sand Springs Keystone Ancient Forest Preserve: N part, 36.18°N, 96.23°–96.24°W, elev. 850–1050 ft, brushy Crosstimbers forest on steep, rocky, W, E and N-facing slopes, on deeply sheltered, N-facing crevice of W-facing cliff above ravine, 16 Mar 2018, C.A. Morse 26202 & D. Ladd (HB. LADD, KANU); on sheltered face of E-facing cliff, 01 May 2009, C.A. Morse 18874b & D. Ladd (KANU).

**Key to Reichlingia as presently circumscribed**

Note that unidentified substance ‘A’ of Coppins & James (1978), reported for *R. dendritica* (Leight.) Ertz & Sandersson (as *A. altantica* P.James var. *altantica*) and *R. zwackhii* (Sandst.) Frisch & G.Thor (as *Arthonia zwackhii* Sandst.) and ‘anomorphula unknown’ of Coppins (1989) reported for *R. anomorphula* (Coppins & P.James) Frisch (as *Arthonia anomorphula* Coppins & P.James) have
subsequently been determined as 2’-O-methylperlatolic acid (Frisch et al. 2014b, 2020). Recent studies by Ertz et al. (2019) and Thiagaraja et al. (2020) suggest that the lichenicolous taxon Arthothelium arbor vitae Coppins and A. graphidicola Coppins belong to a clade containing Coniocarpon, Reichlingia and Synarthronia, although their position within the clade remains uncertain. Arthothelium arbor vitae inhabits thalli of the sterile sorediate crust Schizocrepis squarosa (Coppins & P James) Ertz, Frisch & Sanderson, while A. graphidicola inhabits the thalli of Graphis Adans. species (Cannon et al. 2020). Both species produce epruinose ascomata and 2–3 transversely septate ascospores.

1a. Thallus bearing dark brown sporochodia-like conidiomata; ascomata unknown.................................................
1b. Thallus lacking sporochodia; ascomata present .......... 2

2a. Ascospores submuriform ................................................. 3
2b. Ascospores transversely septate........................................... 4

3a. Ascospores with 4–5 transverse septa, the terminal cell not enlarged; perlatolic acid plus an unidentified xanthone present; corticolous .................................................................
3b. Ascospores with 3(4) transverse septa, the terminal cell enlarged; 2’-O-methylperlatolic acid present; saxicolous .................................................................

4a. Thallus saxicolous; atranorin, confluentic acid, stictic acid, and unidentified substance ‘B’ present in addition to 2’-O-methylperlatolic acid.................................................................
4b. Thallus corticolous; only 2’-O-methylperlatolic acid present ...... 5

5a. Ascospores 12–14 (16) × 4–5 (6) μm with 1(3) transverse septa....
5b. Ascospores ≥16 μm long, with ≥3 transverse septa .......... 6

6a. Paraphysoids unpigmented; ascospores 19–23 × 7–8 μm with 4–5 transverse septa.....................................................
6b. Paraphysoids brown-pigmented apically; ascospores 16–22 (24) × 5–7 μm with 3–4 transverse septa............................................

Acknowledgments
We thank two anonymous referees for helpful reviews of the manuscript; Garth Holman and Patricia Rogers (Mich) and Michaela Schmull (NH) for preparing images of type material of Arthonia leucostroma; Frank Bungartz (ASU), Jack Elix (CANN) and Damien Ertz (NH) for providing information about the chemistry and morphology of Arthothelium galapagoense; and Grant Gerondale (City of Sand Springs, Oklahoma), Bob Hamilton (The Nature Conservancy) and John Rempe (Oklahoma Department of Wildlife Conservation) for facilitating permissions and site access. Fieldwork was made possible by anonymous contributions to the McGregor Herbarium Endowment Fund and The Nature Conservancy.

Literature Cited


manuscript received September 8, 2020; accepted December 14, 2020.