

***Physconia labrata*, a new species from western North America and Asia**

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**ABSTRACT.** A new species belonging to the lichen genus *Physconia* is described from Alaska and the Canadian and American Rocky Mountains and adjacent forested regions. It is also reported from China, Nepal, India and Siberia. The new species, *Physconia labrata*, is superficially similar to *P. perisidiosa*, but can be distinguished by having a blackened, corticate lower surface and a paraplectenchymatous upper cortex.

**KEYWORDS.** Physciaceae, cortex anatomy, phylogeny, phytogeography, Alberta, Alaska, New Mexico.

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The lichen genus *Physconia* was segregated from *Physcia* by Josef Poelt in 1965 (Poelt 1965), and the first North American checklist in which the genus appeared was the last Hale & Culberson version (1970), where only six species were listed. Two of these were Eurasian species which were later determined to not occur in North America (Esslinger 1994). In the most recent online version of the checklist (Esslinger 2016), the number of *Physconia* species listed has grown to thirteen. Most of the increase is due to the discovery of new or previously described but misunderstood species, some of which are apparently endemic to North America (Esslinger 1994, 2000, 2002).

Over many years, during on-going work on the lichen genus *Physconia* by the first author, occasional specimens of supposed *P. perisidiosa* (Erichsen) Moberg from North America and Asia were found to have an unusual lower surface, which was corticate to the lobe ends and largely blackened, whereas the lower surface of *P. perisidiosa* lacks a well-formed lower cortex, especially at the periphery, which is typically pale and fibrous, often with blackened striations. Because of its distinctive reflexed and labriform soralia, which it shares with *P. perisidiosa*, this new taxon was given the working or herbarium name of *P. labrata*. Over the past several years, during work on lichens in Alaska and Alberta involving the second and third authors, respectively, more specimens of this distinctive taxon were found and communicated to the first author. Most specimens from Alberta were collected during systematic lichen surveys conducted by a province-wide biodiversity monitoring initiative, the Alberta Biodiversity Monitoring Institute, ([www.abmi.ca](http://www.abmi.ca)).

## **METHODS**

***Thin-layer chromatography.*** Attempts to extract and identify secondary compounds were made using the standardized TLC methods of Culberson & Kristinsson (1970) and Culberson

(1972).

**DNA extraction and PCR amplification.** We chose to analyze nuclear internal transcribed spacer (ITS) sequences in keeping with the selection of this locus as the barcoding region for fungi, and its common use by others working on species-level problems in *Physconia* (Cubero et al. 2004; Divakar et al. 2007; Lohtander et al. 2007, 2008). We attempted to obtain DNA sequences for all six recently collected specimens of *P. labrata* that were available to us and of adequate size for sampling. Of these, we had success with only two, although we had higher rates of success with other *Physconia* specimens sequenced for this study (**Table 1**). Although a sampling of two specimens of a new species is fewer than we prefer, the species was sufficiently well defined morphologically to proceed with its description. Sampling outside the new species focused on geographic and phylogenetic neighbors (**Table 1**).

Extraction and amplification protocols are in McCune & Curtis (2012). Primers for amplifying nuclear ITS were ITS1F (Gardes & Bruns 1993) and ITS4 (White et al. 1990). In addition to the amplification primers, the following primers were used for sequencing: ITS2 and ITS3. Staden 1-6-0 (SourceForge.net) was used to quality check the raw sequence, align the two sequencing runs per PCR product and generate a consensus sequence for phylogenetic analyses.

**Phylogenetic analyses.** We inferred phylogenetic relationships from sequences of rDNA from two specimens of the putative new species, along with seven additional *Physconia* specimens (**Table 1**), and 50 *Physconia* specimens already represented in GenBank. We also included *Anaptychia elbursiana* (Szatala) Poelt as an outgroup. ITS sequences selected from GenBank represented species across *Physconia*, with emphasis on taxa with similar morphology, especially *P. grumosa* and *P. perisidiosa*. Sequences were aligned with Geneious alignment using default settings (cost matrix: 65%, gap open penalty 12, gap extend penalty 3), then adjusted manually.

The ends of the alignments were then trimmed to a nearly equal number of sites for all sequences, including all of ITS1, 5.8S, and ITS2. Alignment gaps were treated as missing data.

Phylogenetic trees were obtained by maximum likelihood analysis of the ITS data, using the GTR (general time-reversible) model and Geneious defaults, except we used the “BEST” (slower) topology search and three substitution rate categories, using the PhyML (Guindon et al. 2010) plug-in to Geneious 5.4.3 (Drummond et al. 2011). Statistical support for branches was evaluated with 1000 bootstrap resamplings. Excluding the outgroup, the alignments had 201 variable sites of 503 in the ITS.

## RESULTS

In addition to the distinctive lower surface color and structure, which drew original attention to this new taxon, subsequent anatomical study of longitudinal lobe sections also revealed that the upper cortex is not scleroplectenchymatous as in *P. perisidiosa*, but rather it is distinctly paraplectenchymatous in mature thallus regions (1.5–2 mm or more from the lobe tips). It appears, therefore, that despite their superficial similarities, this new taxon is not closely related to *P. perisidiosa*. DNA sequencing was successful for two specimens, including the holotype from Alberta and one specimen from Alaska, and this also confirmed the distinct nature of this taxon. In fact, in the phylogenetic tree including these sequences (**Fig. 1**), the new taxon appears closest to *P. grumosa* Kashiw. & Poelt. Interestingly, this closest genetic relative *P. grumosa* does not share the paraplectenchymatous upper cortex of *P. labrata*, but instead has a distinctly scleroplectenchymatous upper cortex. Although all available evidence from previous work has shown that upper cortex type is a good species level character in *Physconia* (e.g., Divakar et al. 2007; Esslinger 1994; Poelt 1966), examination of our tree (**Fig. 1**) or any of those published

previously (Cubero et al. 2004; Divakar et al. 2007; Lohtander et al. 2007, 2008), shows that species with the two types of upper cortex are distributed rather randomly in the tree, and do not clearly identify any large species groupings or possible subgenera.

Based on these morphological and anatomical comparisons as well as phylogenetic analyses of ITS sequences from two specimens, here we describe a new species, *Physconia labrata* Essl., McCune & Haughland.

**Phylogenetic reconstruction.** *Physconia labrata* was readily distinguished from other *Physconia* species by ITS sequences. Phylogenetic reconstruction showed the *P. labrata* specimens to form a supported monophyletic group with 90% bootstrap support (**Fig. 1**). Furthermore, *P. labrata* shows reciprocal monophyly and a supported sister relationship with *P. grumosa*. Both *P. labrata* and *P. grumosa* were placed in a supported clade (91% bootstrap support), which in turn fell in an unsupported clade with *P. kurokawae* Kashiw., *P. leucoleiptes* (Tuck.) Essl., and two clades of *P. muscigena* (Ach.) Poelt.

## TAXONOMY

***Physconia labrata*** Essl., McCune & Haughland, *sp. nov.*

**Figs. 2, 3, 4A & 5A**

MYCOBANK MB 822644

*A sorediate species, bearing soredia on lobe ends and edges, the soralia becoming more or less labriform; differing from the superficially similar species *Physconia perisidiosa* by having a darkened, corticate lower surface and a paraplectenchymatous upper cortex.*

TYPE: CANADA. ALBERTA: ca. 30 km N of Hinton, AB, on Hwy. 40 in William Switzer

Provincial Park, east on Cache Lake & Graveyard Lake gravel access road, at parking lot for wildlife viewing blind, ca. 53.5026°N, 117.82146°W, 1203 m. elev., on trunk of *Picea*

*glauca*, 7 Nov 2015, (holotype: *Diane L. Haughland*, #2015-1a, PMAE; isotypes: #2015-1b, CAN, #2015-1c, herb. Esslinger).

**Description.** Thallus foliose, irregular to distinctly orbicular, mostly appressed, up to ca. 4 cm in diameter but usually smaller, occasionally composed of numerous small, disconnected lobes, which tend to be ascendant and sorediate. Lobes linear to more often shorter, irregular-rounded or flabellate and partly imbricate, (0.7–) 1–2 mm broad, mostly flat to weakly concave or convex, rarely with a few small, scattered lobules. Upper surface gray to gray-brown or commonly brown to dark reddish brown, with a partial (lobe ends) or sometimes almost complete pruina. Sorediate, the soralia on and under lobe ends or edges, which become ascendant or somewhat reflexed and weakly to more strongly labriform; soredia usually granular, darkening with age, sometimes becoming somewhat isidioid in older parts. Medulla white. Lower surface corticate, entirely black or sometimes almost white to pale tan on lobe ends or immediately behind the soralia, but soon blackening and often becoming rather shiny; rhizines black and squarrosely branched. Upper cortex becoming paraplectenchymatous (lumina up to 5–6  $\mu\text{m}$ ), although somewhat irregular and in scattered places with occasional pairs or trios of interconnected lumina visible in longitudinal lobe sections; lower cortex prosoplectenchymatous. Apothecia rare, up to 2.5 mm in diameter, the margin weakly crenate when young, sometimes becoming lobulate (and lobules sometimes reflexed with terminal soralia); ascospores *Physconia*-type, 25–31  $\times$  15.5–18  $\mu\text{m}$ .

**Chemistry.** Cortex, medulla, and soralia are all K–, C–, KC– and PD–. Secondary products not detected by TLC.

**Substrate and ecology.** Growing on bark and twigs, in North America especially of *Populus* and *Picea*, but also *Pseudotsuga* and *Abies*; occasional on downed logs.

**Discussion.** Because of its distinctive soralia, which occur on and under the reflexed or weakly ascendant ends or edges of lobes and which often become somewhat labriform, this species has most often been identified as *Physconia perisidiosa*, and the two do appear very similar when viewing the upper surface of the thallus. However, the lower surface of *P. labrata* (**Fig. 4A**) has the cortex continuous all the way to the lobe tips (or, on the sorediate lobes, to the edge of the soralia) and is colored black almost throughout, although the lobe ends may be paler, usually pale tan or tawny, occasionally almost white. The same somewhat pale periphery can occur in essentially all the species of *Physconia* that have a blackened, corticate lower surface [e.g., *P. deterosa* (Nyl.) Poelt and *P. grumosa*]. By contrast, the lower surface of the lobes in *P. perisidiosa* (**Fig. 4B–E**) typically lacks a cortex, thus the medulla which is white and obviously fibrous is visible. Usually, in *P. perisidiosa*, more or less distinctive black striations, which consist of darkened individual hyphae, develop at some distance from the lobe end and these become more and more abundant inward until they begin to coalesce into a dark, usually dull pseudocortex, especially in larger, well developed thalli. Another distinction between these two species is the strongly scleroplectenchymatous upper cortex of *P. perisidiosa* (**Fig. 5B**) compared to the paraplectenchymatous cortex of *P. labrata* (**Fig. 5A**). As with other species of *Physconia*, the distinction between these two types of cortex should be made at least 1.5 to 2 mm behind the tip, and preferably even a bit further, because in the young cortex of the first 1–2 mm, the two types look essentially indistinguishable, as very compact tissue without well-defined cells.

Two additional species in North America that usually have similarly reflexed or labriform soralia are *Physconia fallax* Essl. and *P. leucoleiptes* (Tuck.) Essl., both of which also share a similar blackened and corticate lower surface. The labriform soralia of *P. leucoleiptes* can be rather similar to those of *P. labrata*, although they are usually more numerous and can then

sometimes be nearly continuous on the lobe margins. The soralia of *P. fallax* occur in more hooded or “nest-shaped” structures formed by separation of the upper and lower cortex on lobe margins or especially the lobe ends. Both *P. leucoleiptes* and *P. fallax* can also be distinguished from *P. labrata* by their production of secalonic acid A in their soralia, which therefore react K<sup>+</sup> pale to distinctly yellow and KC<sup>+</sup> even stronger yellow to almost yellow orange. These two species also have distributions that differ from the known Alaska-Rocky Mountain distribution of *P. labrata*. *Physconia fallax* is primarily a California species, and *P. leucoleiptes* is most common in central and eastern North America, and in the West it is known by us only from the Southwest, although the online CNALH database includes specimens (questionable, and not seen by the authors) from the Rockies in Montana, Utah, and Colorado.

In Alberta, *Physconia labrata* is apparently more common than *P. perisidiosa*. *Physconia labrata* is sparsely distributed across moist to mesic ecosites in the Foothills and Boreal natural regions in the northern half of the province (ABMI 2016). The species is most commonly found on the trunks of *Populus* in mature (>100 year old) mixedwood and deciduous forests; it also occurs as an epiphyte in more open sites including pine forests (*Pinus banksiana* and *P. latifolia*), adjacent to forest openings (including anthropogenic disturbances such as roads or cutlines) and in rich fens (ABMI 2016). In Alberta, *P. labrata* is likely decreasing in abundance due to the loss of habitat to forestry and agriculture within its range (ABMI 2016).

Among lichens that exhibit a North America-Asia disjunction, the most frequent and well known pattern is the eastern North America-eastern Asia disjunction, which is also known to occur among species of bryophytes and seed plants (Galloway 2008). The apparent pattern of distribution of this new species as a disjunct between western North America and Asia is much less common. Unfortunately, no recent collections from Asia were available for genetic testing, so the



reality of that distribution pattern remains to be confirmed by genetic studies.

***Additional specimens examined.*** U.S.A. ALASKA: Southeast Alaska, along the Haines highway (Hwy. 7) near Klukwan, 59°24.066'N, 135°53.559'W, *Esslinger 20000* (herb. Esslinger) [small admixture in majority *P. perisidiosa* collection]; Mt. McKinley National Park, north bank of McKinley River, south of Wonder Lake, 63°25'N, 150°55'W, *Weber & Viereck, S7171* (COLO). Lake & Peninsula Co.: Lake Clark National Park, near west end of Portage Lake 60.50782°N, 153.8832°W, *B. McCune 35181* (OSU); Katmai National Park, shore of Naknek Lake, bay just N of Brooks Camp, 58.56264°N, 155.77521°W, *B. McCune 32886* (OSU). Matanuska-Susitna Borough, town of Eklutna, 61.46164°N, 149.3627°W, *H. Root 1763* (OSU). NEW MEXICO: Lincoln Co.: north from the Ski Apache Ski Resort along small stream and trail, 33°24'N, 105°47.4'W, *Esslinger 15435* (herb. Esslinger); Otero Co.: about 13 miles E of Alamogordo, in the Sacramento Mountains, along Hwy 82 near the Cloudcroft Ski Area, 32°38'N, 105°45'W, *Esslinger 12028B* (herb. Esslinger). CANADA. ALBERTA: ca. 9 km SW of Lodgepole, 53.02°N, 115.34°W, *Stairs*, ABMI Site 1208, ABMI Lichen # 248376 (PMAE); ca. 7 km NE of Little Buffalo, 56.49° N, 116.04° *Hogarth*, ABMI Site 589, ABMI Lichen # 26672 (PMAE); ca. 23 km NE of Hinton, 53.56°N, 117.36°W, *Camus*, ABMI Site 1136, ABMI Lichen # 292345 [with *P. perisidiosa* & *P. grumosa* admixture], 300123 & 308210 (PMAE); ca. 17 km NW of Cynthia, 53.37°N, 115.63°W, *Stairs*, ABMI Site 1145, ABMI Lichen # 249705 (PMAE); ca. 16 km SE of Hinton, 53.34°N, 117.36°W, *Bohnen*, ABMI Site 1169, ABMI Lichen # 245211 (PMAE); ca. 30 km N of Hinton, on Hwy. 40 in William Switzer Provincial Park, east on Cache Lake & Graveyard Lake gravel access road, at parking lot for wildlife viewing blind, 53.502693°N, 117.821431°W, *Haughland 2012-133 & 2012-181* (PMAE); ca. 15 km NW of Rocky Mountain House, in Crimson Lake Provincial Park, 52.466984°N, 115.04743°W, *Haughland 2012-350* (PMAE); ca. 18 km SW of

Rocky Mountain House, in/near Mitchell Lake Provincial Recreation Area, 52.22°N, 114.98°W, *Nobert*, ABMI Site 1326, Lichen # 653038 & 653422 (PMAE); ca. 15 km NE of MacKay, in/near Highway Natural Area, 53.76°N, 115.47° W, *Lewis*, ABMI Site 1077, Lichen # 26954 (PMAE); ca. 61 km NE of Fort MacKay, 57.73°N, 111.55°W, *Harding*, ABMI Site 328, Lichen # 588621, 588626 & 588633 (PMAE); ca. 12 km NW of Sexsmith, 55.44°N, 118.89°W, *De Carvalho*, ABMI Site 802, Lichen # 39172 (PMAE); ca. 22 km NW of Calling Lake, in/near Otter-Orloff Lakes Wildland Provincial Park, 55.35°N, 113.45°W, *O'Sullivan*, ABMI Site 755, Lichen # 21461 & 21502 (PMAE); ca. 45 km NW of Notikewin, 57.36°N, 117.82°W, *Hillman*, ABMI Site 459, Lichen # 69728 & 121862 (PMAE); within 6 km of Busby, 53.96°N, 113.97°W, *O'Sullivan*, ABMI Site OG-ABMI-1015-1, Lichen # 652862 (PMAE); ca. 66 km SW of Fort McMurray, in/near Grand Rapids Wildland Provincial Park, 56.55°N, 112.45°W, *Birkigt*, ABMI Site 538, Lichen # 157973 (PMAE); within 6 km of Leslieville, 52.392°N, 114.64°W, *Nobert*, ABMI Site 1298, Lichen # 463588 & 463618 (PMAE); Harlech Recreation Area, 15 km E of Nordegg, 52°31'N, 115°58'W, *Marsh*, B 86.141.116 (PMAE);. BRITISH COLUMBIA: ca. milepost 223 along the Alaska Highway, near Bougie Creek, 58°01.779'N, 122°43.365'W, *Esslinger 19160* (herb. Esslinger). YUKON TERRITORY: banks of the LaBiche River, 4 km N of Kootaneelee Natural Gas Drilling Camp, 60°09.223'N, 124°04.468'W, *Spribille 28508* (GZU); along the Alaska Highway (Hwy. 1) ca. 1 mile W of Koidern, 61°59.103'N, 140°30.724'W., *Esslinger 19916* (herb. Esslinger). CHINA. YUNNAN: Zhongdian County. 27.48°N, 99.42°E, *Wang Xian-ye et al.* 4335 (herb. Esslinger, HMAS-L); XIZANG: southeastern Tibet, lower Tsangpo Valley, ca. 10 km SW of Tzela Dzong (Gyamda/Nijang-Tsangpo confluence), 29°24'N, 94°25'E, *Dickoré L-10* (GZU). NEPAL. Langtang Area, near Kyangjin, *Poelt N86-L241* (GZU); Marsyandi-Tal, nahe Pisang Low, *Kirschbaum*, 30 Sep 1979 (GZU); Kali Gandaki-Gebiet, Tukuhe, *Kirschbaum*, 28 Mar 1979,

(GZU). INDIA. NW Himalaya, Pangi, *Stoliczka 442* (H). RUSSIA. SAKHA REPUBLIC (Yakutia): near mouth of Aldan River on lower course of Lena River, *Cajander*, 17 Jul 1901 (H).

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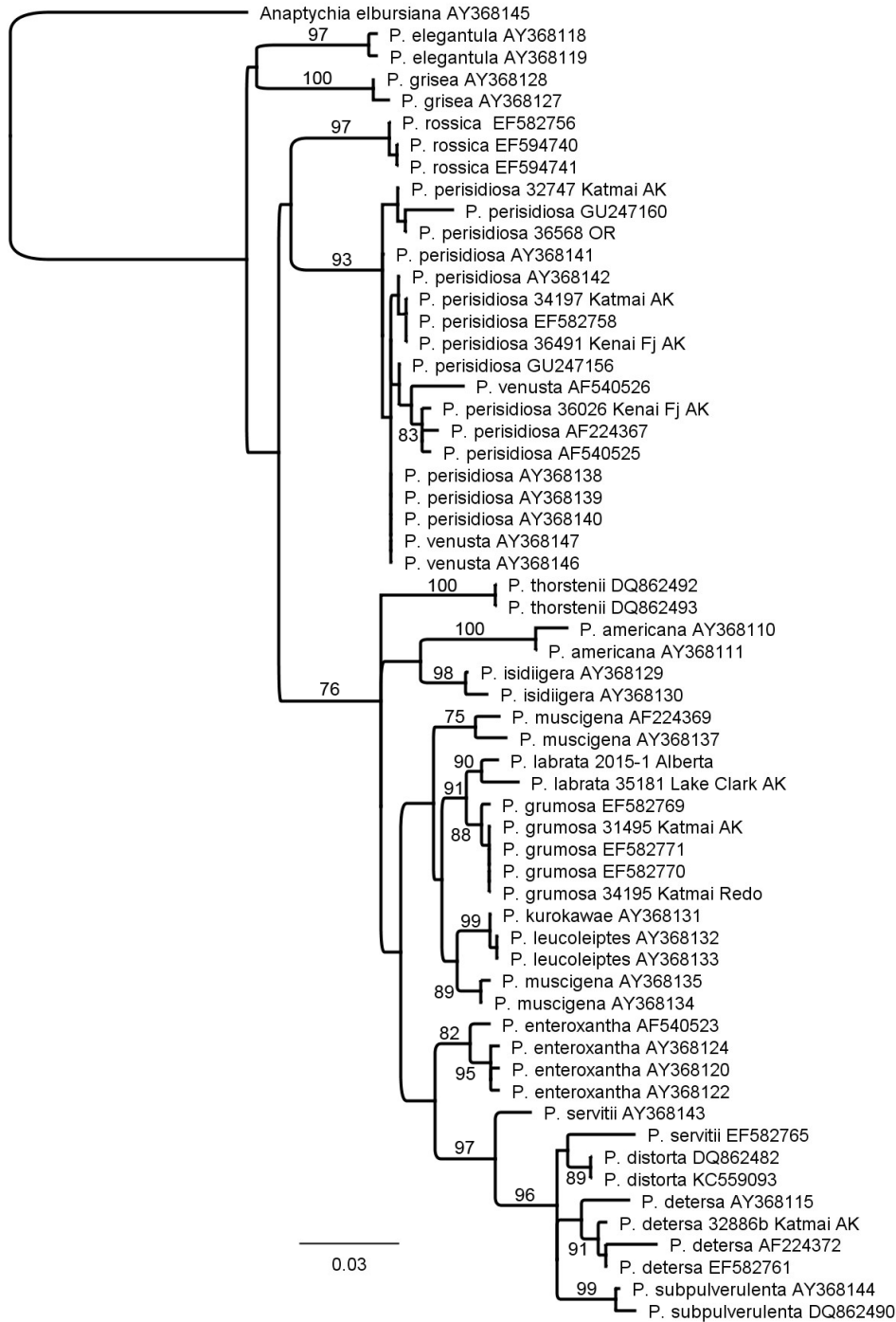
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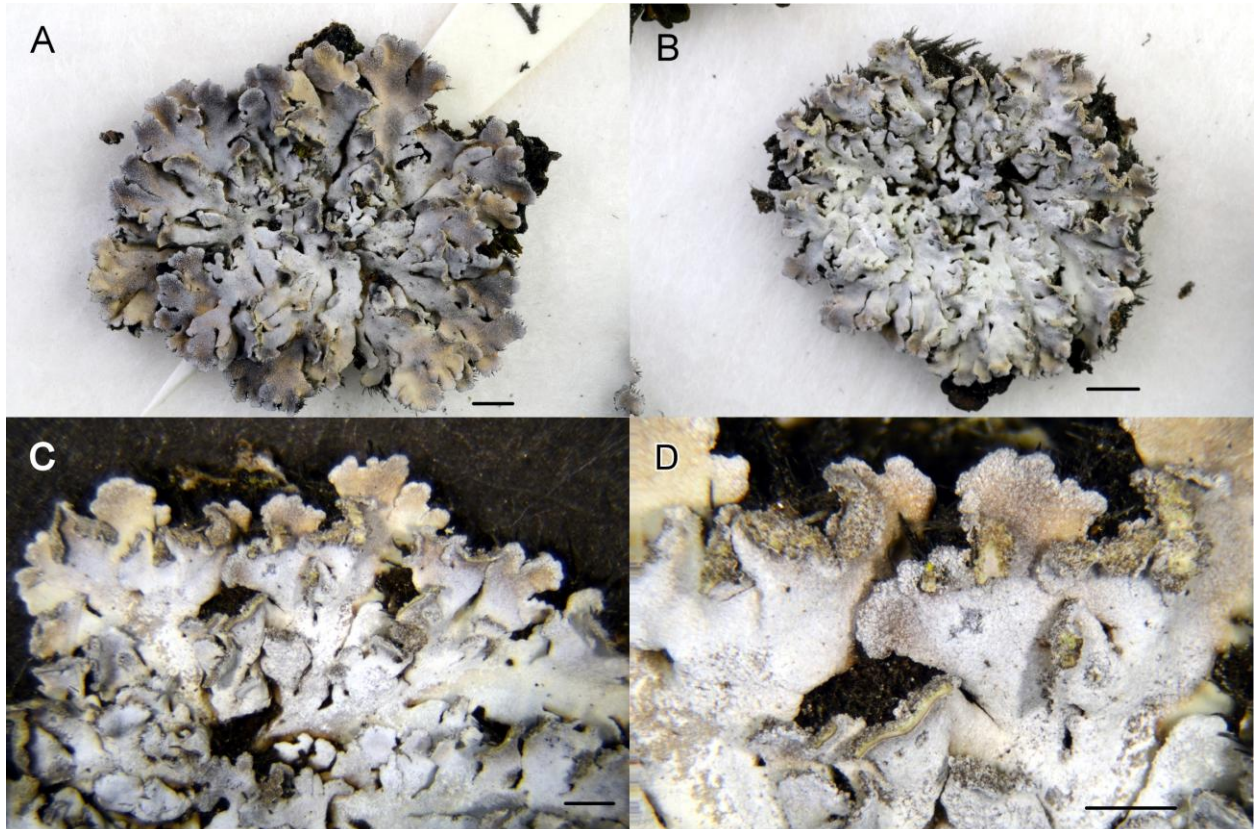
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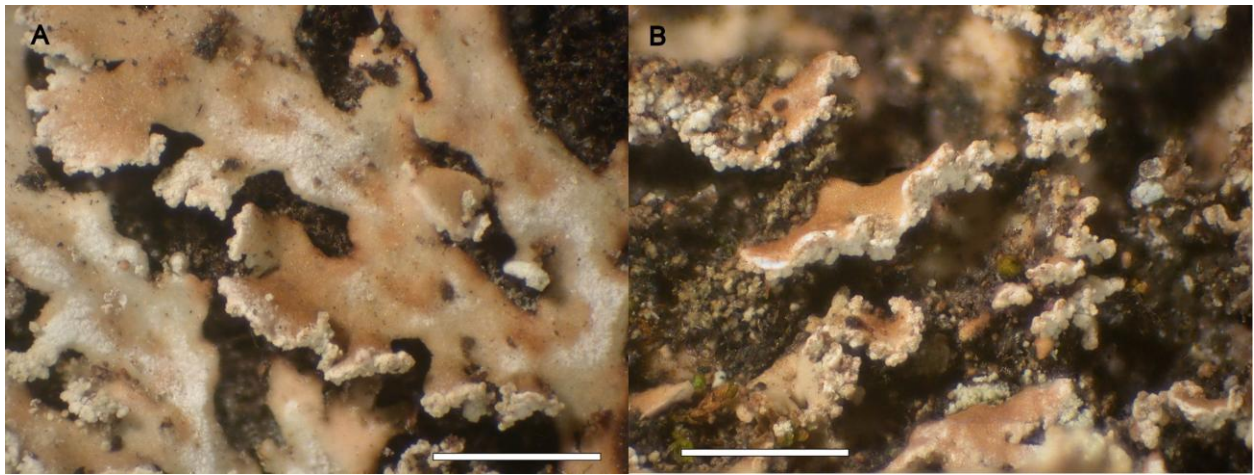
**Figure 1.** Phylogenetic reconstruction. Most likely phylogenetic relationships among sampled *Physconia* species were inferred from ITS sequences and based on rooting with *Anaptychia elbursiana* as outgroup. Bootstrap percentages above 75% are shown above branches; branch lengths are based on estimated number of substitutions per site assuming a GTR model of substitution. GenBank accession numbers are given for previously published sequences. GenBank numbers for new sequences are given in Table 1.



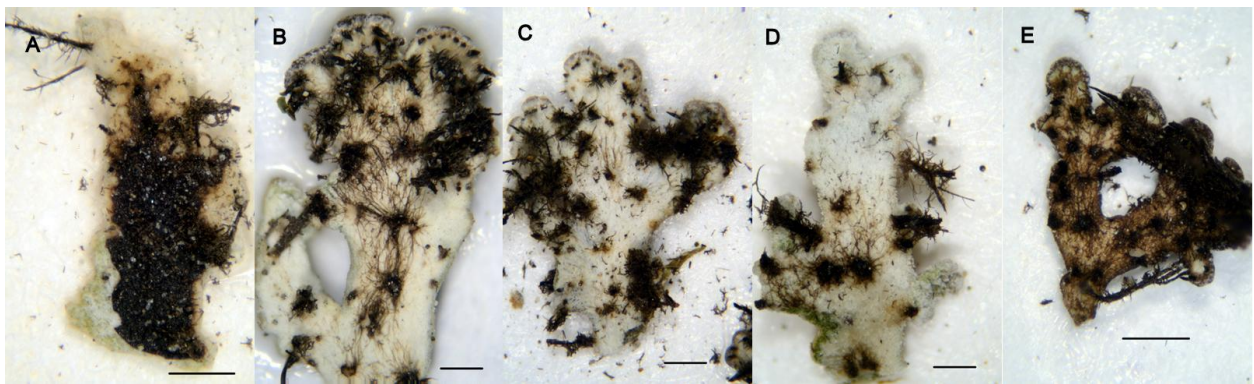
**Figure 2.** *Physconia labrata*, from the holotype specimen [*Haughland*, #2015-1a (PMAE)]. **A & B.** Regular, rosette-forming thalli. **C & D.** Closer view of soralia on 2B. Scale bars: A & B = 2 mm, C & D = 1 mm.



**Figure 3.** *Physconia labrata*, close-up of lobes and soralia on a less rosette-forming specimen, also with sparser pruina than in Fig. 2 [*McCune 32886a* (OSU)]. Scale bar = 1 mm].



**Figure 4.** Lower surface on lobes of *Physconia*. spp. **A.** *P. labrata* [*McCune 32886a* (OSU)]. **B–E.** Various *P. perisidiosa*. **B.** *Esslinger 20384* (herb. Esslinger). **C.** *Esslinger 15485* (herb. Esslinger). **D.** *Esslinger 16467* (herb. Esslinger). **E.** *Esslinger 19214* (herb. Esslinger). Scale bars A–E = 0.5 mm.





**Figure 5.** Upper cortex anatomy of *Physconia* spp. as observed in longitudinal sections,  $\times 1000$ . **A.** *P. labrata*, Haughland 2015-1 (PMAE). **B.** *P. perisidiosa*, Stairs 243925 (PMAE).

