

A new *Lecidea* species from Dismal Island (Antarctic Peninsula, Antarctica) supported by morphological, chemical, and nrITS phylogenetic evidence

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Abstract

Antarctica remains one of the least explored regions of the world in terms of lichen diversity, and many remote localities still harbor undescribed taxa. In this study, a new saxicolous lichen species belonging to the genus *Lecidea* is described from Dismal Island, located in the East Antarctic Peninsula region, based on an integrative taxonomic approach combining morphological, anatomical, chemical, and nrITS phylogenetic evidence. The examined material was collected from ice-free rocky habitats and deposited in the Erciyes University Lichen Herbarium (ERCH). Morphological and anatomical observations were carried out using stereomicroscopy and light microscopy, while chemical characterization was performed using standard spot tests and thin-layer chromatography (TLC). Molecular analyses were based on the internal transcribed spacer (ITS) region of nuclear ribosomal DNA. The newly generated ITS sequence was analyzed together with 92 reference sequences retrieved from GenBank®, and phylogenetic relationships were inferred using Bayesian analysis. The results demonstrated that the studied specimen forms a distinct and well-supported lineage within *Lecidea*, clearly separated from all currently sequenced congeners. Although phylogenetically closest to members of the *Lecidea atrobrunnea* complex and *L. syncarpa*, the new species differs consistently in several diagnostic characters: (1) dark brown, dull, crustose and areolate thallus, (2) small black lecidine apothecia, (3) dark brown epihymenium, an I– medulla and hymenium, an N+ red hymenial reaction, and (4) the absence of both prothallus and pycnidia. Chemically, TLC revealed (5) norstictic acid as the major secondary metabolite. The combination of these stable, non-overlapping characters supports the recognition of the taxon as a distinct species, here named *Lecidea devrimii*. This study highlights the still underestimated lichen diversity of Antarctica and emphasizes the importance of integrative taxonomy in resolving species boundaries within morphologically complex lichen groups.

Key words: Antarctica, *Lecidea devrimii*, molecular analysis, lichen biodiversity, new species; secondary metabolites, the Faure Islands

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Introduction

Antarctica represents one of the most extreme environments on Earth, characterized by low temperature (Turner *et al.* 2009), strong winds (King and Turner 2007), limited water availability (Convey *et al.* 2008), and high levels of ultraviolet radiation during the austral summer season (Cockell and Blaustein 2000). Despite these harsh conditions, lichens constitute one of the dominant life forms in ice-free terrestrial ecosystems, where they play a crucial role as pioneer organisms in primary succession processes (Øvstedal and Lewis-Smith 2001, Colesie *et al.* 2014). Antarctic lichens exhibit remarkable physiological and morphological adaptations that enable them to survive prolonged desiccation, repeated freeze–thaw cycles, and nutrient-poor substrates (Green *et al.* 2011). These adaptations make them particularly suitable model organisms for studying extremophile biology and ecosystem resilience under climate change scenarios.

The Antarctic Peninsula region, Western coast in particular, has received increasing scientific attention due to its relatively milder climate compared to continental Antarctica and its higher biodiversity potential (Sancho *et al.* 2007). However, many microhabitats and remote localities within this region, such as Dismal Island, remain poorly explored in terms of lichen diversity. Previous studies have demonstrated that even well-studied Antarctic regions continue to yield new taxa, indicating that the current knowledge of lichen biodiversity is still far from complete (Øvstedal and Lewis-Smith 2001, Smykla *et al.* 2011). This underexplored diversity highlights the necessity of continued taxonomic and phylogenetic investigations, particularly in isolated and extreme habitats.

Dismal Island is an island, 1.9 kilometres long and 60 metres (200 ft) high, which is mainly ice covered and is the largest of the Faure Islands, lying in

Marguerite Bay off the west coast of Graham Land. The Faure Islands were discovered and first charted in 1909 by the French Antarctic Expedition under Jean-Baptiste Charcot. The group was visited and surveyed in 1949 by the Falkland Islands Dependencies Survey, who so named this island for its appearance of extreme desolation and lifelessness. Where vegetation occurs, it is mostly in the form of lichens and mosses, along with limited occurrences of microbial communities such as cyanobacterial mats and other microflora adapted to extreme conditions. Based on the available literature, no study has been found that directly addresses or examines lichen biodiversity on Dismal Island.

In recent decades, integrative taxonomy combining morphological, chemical, and molecular data has become essential for resolving species boundaries in lichenized fungi, especially in morphologically conserved genera such as *Lecidea* (Lumbsch and Leavitt 2011, Schmult *et al.* 2011). Traditional morphological characters alone are often insufficient due to phenotypic plasticity and convergent evolution, whereas molecular markers, particularly the nrITS region, provide robust tools for distinguishing closely related taxa and uncovering cryptic diversity (White *et al.* 1990, Leavitt *et al.* 2013). Consequently, the integration of multiple lines of evidence has significantly improved the understanding of species delimitation within complex lichen groups.

The genus *Lecidea* Ach. (Lecideaceae) that was originally described by Acharius (1803), underwent many systematic changes and its traditional wide circumscription (Zahlbruckner 1926) dramatically changed over the last decades (Hafellner 1984, Hertel 1967, 1977, 2006). Of the 427 species included in *Lecidea* *sensu lato* (Kirk *et al.* 2008), only about 100 belong to *Lecidea* *sensu stricto* based on, amongst other characters, the presence of a *Lecidea*-

type ascus (Hertel 2006). The taxonomy of *Lecidea* species is complex because of the morphological variation within and amongst species. The species circumscription in *Lecidea* needs revision and molecular data are helpful in interpreting subtle morphological differences that have

been considered as intraspecific variability (Schmull et al. 2011).

Morphological, anatomical, and phylogenetic analyses of new species collected from Dismal Island showed that they belong to an undescribed species of *Lecidea* and we describe it here as *L. devrimii*.

Materials and methods

Specimen collection and morphological examination

The new species was collected from Dismal Island (Antarctic Peninsula) by the first author. The collected voucher specimen is deposited in the Erciyes University Lichen Herbarium (ERCH). A stereomicroscope (Olympus S2X7) was used to examine the lichen morphology, and a com-

pound microscope (Leica DM2500) was used to inspect structure inside the apothecia. In order to identify the internal substances of lichen, 5% potassium hydroxide (K) and an aqueous solution of calcium hypochlorite solutions were used to examine the color reaction (chemical color test).

Molecular methods

DNA isolation, PCR, and sequencing

DNA extraction was performed from apothecia fragments using the DNeasy Plant Mini Kit (Qiagen), according to the manufacturer's instructions. PCR amplifications were conducted using 2×EasyTaq® PCR SuperMix. The following primers were used for PCR amplifications: internal transcribed spacer (ITS) for ITS1 and ITS4. The following program was used for the amplification of ITS: initial denaturation at 95°C for 5 min, followed by 35 cy-

cles of 95°C for 30 s, 55°C for 30 s, and 72°C for 90 s, and then a final extension step at 72°C for 8 min. The PCR reactions were visualized on 1.2% agarose gel. Sequence analysis of the lichen sample obtained from the PCR products were performed by the Epigen Biotechnology Company (Ankara, Türkiye). The nrITS sequence generated in this study for *Lecidea devrimii* was deposited in GenBank under the accession number PZ314520.

Sequence alignment and phylogenetic analysis

The obtained sequences were aligned with ClustalW and edited using the Bioedit program. All ambiguous characters were removed and only unambiguous characters were analyzed. Geneious 2023.2.1 program, MrBayes Algorithm, and 1000 boot-

strap branches were used for phylogenetic tree construction. ITS datasets were examined for the phylogenetic positions of new *Lecidea* species and one sequence (*Bellemeria cinereorufescens*) was included as an outgroup.

Phylogenetic tree for the genus *Lecidea* was produced from 92 sequences from GenBank and one new sequence (sequence for ITS) from the new species. Phylogenetic tree was constructed using MrBayes algorithm. All positions with less than 95% site coverage were eliminated. That is, fewer than 5% alignment gaps, missing data, and ambiguous bases were allowed at any position. There were a total of 448 positions in the final dataset. Within the genus *Lecidea*, the new species clearly branched out from other species. The phylogenetic analyses did not designate any species identical to the new species in the genus *Lecidea*. *Bellemeria cinereorufescens* (Ach.) Clauzade & Roux was used as the outgroup (see Fig. 1).

As *Lecidea devrimii* showed the closest phylogenetic affinity to *Lecidea syncarpa* in the ITS-based phylogenetic tree, the two taxa were compared in terms of nucleotide differences at variable positions. This comparison was carried out to reveal the molecular divergence between the new species and its closest relative (Fig. 2).

According to this comparative analysis, 487 nucleotides were determined as conserved sites (C), whereas 17 nucleotides were identified as variable sites (V) within the ITS gene region. The light-coloured regions represent nucleotide differences (Fig. 2). Based on these findings, a sequence divergence of 3.4% was observed between the new species *Lecidea devrimii* and *L. syncarpa*.

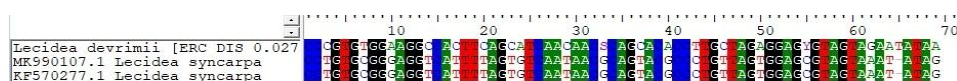


Fig. 2. Shortened ITS alignment, including only variable positions of new species *Lecidea devrimii* and *L. syncarpa*.

Taxonomy

Lecidea devrimii Halıcı, Bişgin & Güllü sp. nov. (see Fig. 3)

MycoBank No: 863510

Type: Antarctica, Antarctic Peninsula, Dismal Island, near Turkish Dismal Global Navigation Satellite System, 68°6' S 68°50' W, alt. 10 m. 12 February 2022, Leg. M. G. Halıcı, ERCH DIS 0.027 (ERCH–holotype).

Diagnosis: *Lecidea devrimii* is closest to *L. syncarpa* and the *L. atrobrunnea* complex but differs by its I– medulla and hymenium (vs. I+ blue in related taxa), dark brown epihymenium (vs. green to olive-green), and the absence of prothallus and pycnidia. Although containing norstictic acid, it shows a K– reaction, unlike *L. syncarpa*. This combination of characters clearly distinguishes the new species.

Etymology: The species is named in memory of the first author's beloved father, who passed away in 2022, in recognition of his lifelong support, encouragement, and valuable contributions to the author's personal and academic development.

Description: Thallus crustose, dark brown, often dull color areoles. Apothecium, black, lecidein, 0.2–0.5 mm. Margin, black or sometimes white, usually persistent. Medulla white, I–, K–. Prothallus absent. Epithecium dark brown, 20–25 µm wide; hymenium colourless to very pale bluish in upper part, 50–60 µm high, I–, N+ red; hypothecium light brown, 15–20 µm wide. Asci 8 spored, thin walled, 40–50 µm. Ascospores simple, hyaline, ellipsoid-oblong, with rounded ends, 7–11.5 × 3–5 µm (n=20). Paraphyses unbranched in upper part, 2–3 µm wide. Photobiont chlorococcoid. Pycnidia are absent.

Chemistry: Medulla, I-,C-, K-, N-, P-. Thallus K- C-. According to TLC results, it contains norstictic acid, a UV screening compound (depsidone), as the major substance.

Ecology: *Lecidea devrimii* inhabits exposed rock surfaces in ice-free terrestrial habitats of Dismal Island, Antarctica. The species occurs in extreme polar environments where vegetation is scarce and largely restricted to lichens and bryophytes. It develops on nutrient-poor mineral substrata subjected to intense abiotic stress, including prolonged cold exposure, repeated freeze-thaw events, desiccation, and high UV radiation. Its crustose, compact, and darkly pigmented thallus likely reflects ecological adaptation to these severe conditions. The species is presumed to form part of the pioneer saxicolous lichen communities that colonize barren Antarctic rock surfaces.

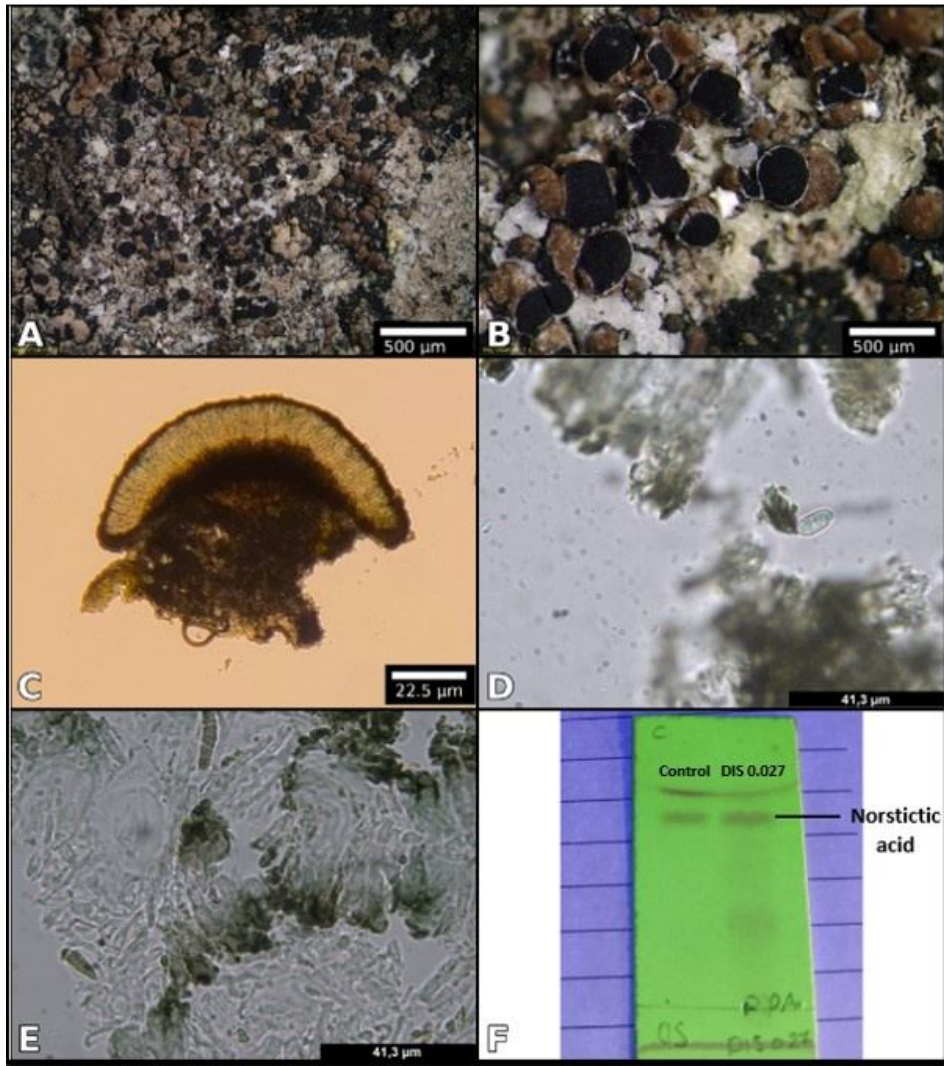


Fig. 3. Morphological, anatomical and chemical characteristics of *Lecidea devrimii*. *Note:* (A) General thallus view. (B) Close-up view of apothecia. (C) Apothecial section. (D) Ascospore. (E) Paraphyses. (F) TLC profile.

Discussion

Our nrITS-based phylogenetic analyses indicate that *Lecidea devrimii* represents a well-delimited lineage that is clearly distinct from currently sampled congeners. In the inferred topology, the new species is placed closest to taxa within and around the *L. atrobrunnea* complex, including *L. atrobrunnea* and its infraspecific taxa, and allied species such as *L. syncarpa*, *L. promiscens*, and *L. confluens*. Despite this proximity in the nrITS tree, *L. devrimii* can be reliably separated from these taxa by a consistent combination of morphology (dark brown, dull areolate crust; small lecideine apothecia), anatomy (hymenium I–, N+ red), and chemistry (norstictic acid as major TLC substance), together with its spot-test profile (medulla I–, K–).

According to the phylogenetic analysis, *Lecidea devrimii* is most closely related to *Lecidea syncarpa* and members of the *Lecidea atrobrunnea* complex; however, it is clearly distinguished from these taxa based on a combination of morphological, anatomical, and chemical characters. The most striking diagnostic feature is the iodine reaction. In *L. devrimii*, both the medulla and hymenium are entirely I– (non-amyloid), whereas in all compared taxa (*L. syncarpa*, *L. atrobrunnea* subsp. *stictica* and *Lecidea atrobrunnea*), the medulla is I+ intensely blue-violet and the hymenium is I+ blue, indicating the presence of amyloid structures (Orange et al. 2001, Hertel and Leuckert 2008). This difference alone provides a strong taxonomic separation. Chemically, *L. devrimii* contains norstictic acid as the major secondary metabolite according to TLC results; however, in contrast to *L. syncarpa*, which shows a characteristic K+ yellow to red reaction, the medulla of *L. devrimii* is K– and I–, an unusual combination that further supports its distinctiveness (Orange et al. 2001, Lumbsch 1998). Moreover, the *L. atrobrunnea* subspecies differ by possessing

other chemical profiles, including stictic acid (subsp. *stictica*) and confluent acid (*atrobrunnea*) (Hertel and Leuckert 2008, Nimis 2026). Morphologically, the epihymenium of *L. devrimii* is dark brown, clearly differing from the characteristic green to olive-green epihymenium observed in the compared taxa (Hertel 1995, Orange et al. 2001). The thallus of the new species is composed of dark brown, dull, and relatively homogeneous areoles, whereas the other lecideoid taxa typically exhibit paler, often shiny, and more variable areoles with distinct margins (Hertel and Schuhwerk 2010). Additionally, *L. devrimii* lacks a prothallus, while a conspicuous dark prothallus is usually present in the other species (Nimis 2026). Reproductive structures also provide distinguishing characters: pycnidia are absent in *L. devrimii*, in contrast to the typically present, immersed, graphidoid pycnidia in the related taxa (Orange et al. 2001). Taken together, these consistent differences in iodine reaction, chemistry, pigmentation, thallus morphology, and reproductive structures clearly support the recognition of *L. devrimii* as a distinct and independent species within the genus.

In summary, while nrITS data place *Lecidea devrimii* closest to the *L. atrobrunnea* lineage and a set of allied species, *L. devrimii* is diagnosable by a stable, non-overlapping character combination: a dark brown, dull areolate crustose thallus; small black lecideine apothecia (0.2–0.5 mm) with a usually persistent margin; a hymenium that is I– and N+ red; a white medulla that is I– and K–; absence of pycnidia; and TLC chemistry dominated by norstictic acid. The table below summarizes the key similarities and distinguishing characters between *L. devrimii* and these related taxa, highlighting the diagnostic features that support its recognition as a distinct species (Table 1).

Compared species	Main similarities to <i>Lecidea devrimii</i>	Main differences from <i>Lecidea devrimii</i>	Diagnostic value for <i>L. devrimii</i>
<i>Lecidea atrobrunnea</i> (including the <i>L. atrobrunnea</i> complex)	Phylogenetically close; both belong to the broader <i>atrobrunnea</i> lineage and may share a crustose thallus with lecideine apothecia.	<i>L. atrobrunnea</i> s. lat. commonly shows a medullary I+ reaction (often violet to blue-violet), whereas <i>L. devrimii</i> has a white medulla that is consistently I– and K–. <i>L. devrimii</i> also has very small apothecia (0.2–0.5 mm) and lacks a prothallus.	The I– medulla, small apothecia, and absence of prothallus separate <i>L. devrimii</i> from taxa in the <i>L. atrobrunnea</i> complex.
<i>Lecidea syncarpa</i>	Phylogenetically close; both may contain norstictic acid, which can make them appear chemically similar.	<i>L. syncarpa</i> is generally characterized by I+ reactions in the medulla and/or hymenium (blue to violet tones), while <i>L. devrimii</i> is I– in both medulla and hymenium and has a hymenial N+ red reaction. In addition, pycnidia may be present in <i>L. syncarpa</i> , but they are absent in <i>L. devrimii</i> .	The combination of I– medulla + I– hymenium + N+ red hymenium + absent pycnidia is a strong diagnostic set for <i>L. devrimii</i> .

Table 1. Comparative morphological, anatomical, chemical, and phylogenetic differences between *Lecidea devrimii* and closely related *Lecidea* species.

Conclusion

In this study, an integrative taxonomic approach combining morphology, anatomy, secondary chemistry, and nrITS-based phylogenetics demonstrated that the material collected from Dismal Island represents a distinct, previously undescribed species of *Lecidea*. This finding is particularly noteworthy because the available background information indicates that no prior work has directly addressed lichen biodiversity on this island, where vegetation is otherwise limited primarily to lichens and mosses. The voucher material has been deposited in the Erciyes University Lichen Herbarium (ERCH), ensuring long-term accessibility for verification and future comparative studies.

The phylogenetic results obtained in this study provided strong molecular evidence for the taxonomic distinctiveness of the studied material. The nrITS dataset, which included 92 reference sequences retrieved from GenBank together with one

newly generated sequence from the Dismal Island specimen, yielded a final alignment of 448 informative positions after the exclusion of ambiguous regions. Bayesian phylogenetic reconstruction recovered the studied specimen as a clearly separated lineage within the genus *Lecidea*, and no currently available sequence was found to be identical or conspecific with the newly generated sequence. This result is particularly important because it indicates that the studied material cannot be accommodated within any currently sequenced or recognized *Lecidea* taxon. Although the inferred topology placed the new species in proximity to taxa related to the *Lecidea atrobrunnea* complex, including *L. syncarpa*, *L. promiscens*, and *L. confluens*, the phylogenetic tree nevertheless supported its status as an independent lineage, rather than a divergent population or local morphotype of an already known species.

In addition to the molecular evidence, chemotaxonomic data further support this distinction. The detection of norstictic acid in the studied material is noteworthy, as this secondary metabolite is known to be associated with specific chemotypes within the *Lecidea atrobrunnea* complex, particularly in *Lecidea atrobrunnea* subsp. *saxosa*, where norstictic acid represents a characteristic compound. However, despite this chemical similarity, the phylogenetic placement of the studied specimen does not cluster within the *L. atrobrunnea* clade, indicating that the presence of norstictic acid alone is not sufficient for species delimitation in this group. This highlights the importance of integrating molecular, morphological, and chemical data in resolving species boundaries within *Lecidea*.

Furthermore, the observed pattern can also be interpreted in the context of the “syndrome” concept sensu Anantaprayoon et al. (2023), which emphasizes that certain combinations of morphological and chemical traits may recur across phylogenetically distinct lineages due to convergent adaptation to similar environmental conditions. In this regard, the shared occurrence of norstictic acid and related phenotypic traits within the *L. atrobrunnea* complex and the studied material may reflect ecological convergence rather than close evolutionary relatedness. Therefore, both phylogenetic independence and the lack of complete congruence with known chemotype–lineage associations further support the recognition of the studied specimen as a distinct taxon (Anantaprayoon et al. 2023).

The morphological and anatomical findings further corroborated this molecular separation. The species is characterized by a dark brown, dull, crustose and areolate thallus, together with small black lecidine apothecia measuring 0.2–0.5 mm in diameter, typically with a persistent margin. Anatomically, the species is distinguished by its dark brown epihymenium, colourless to slightly bluish hymenium, light

brown hypothecium, and 8-spored asci bearing simple, hyaline, one-celled, ellipsoid-oblong ascospores. Particularly noteworthy is the reaction pattern of the internal structures: both the medulla and hymenium are consistently I–, while the hymenium shows an N+ red reaction, providing a highly informative set of diagnostic characters. In addition, the absence of pycnidia and the lack of a prothallus represent further stable features that help to distinguish the species from closely related taxa. These characters, taken together, form a coherent and reproducible phenotypic profile that is not attributable to ordinary intraspecific variation.

The chemical evidence also played an important role in supporting the delimitation of the species. Spot test reactions showed that the medulla is I–, C–, K–, N–, P–, and the thallus is K– and C–, while thin-layer chromatography (TLC) revealed norstictic acid as the major secondary metabolite. This chemical profile is taxonomically meaningful because it helps separate the species from morphologically or phylogenetically similar taxa that differ in their secondary metabolite composition and associated reaction patterns. In this respect, the chemistry of *Lecidea devrimii* is not merely supplementary, but forms an integral component of the species concept established in this study.

When all lines of evidence are evaluated together, the present study demonstrates that *Lecidea devrimii* represents a well-circumscribed and independently diagnosable taxon within *Lecidea*. Its recognition is supported not by a single isolated feature, but by a stable, non-overlapping combination of characters, including its phylogenetic distinctness, I– medulla and hymenium, N+ red hymenium, norstictic acid chemistry, dark brown areolate thallus, small lecidine apothecia, and the absence of pycnidia and prothallus. This combination clearly differentiates it from its nearest relatives, particularly *Lecidea syncarpa*, *Lecidea atrobrunnea* (includ-

ing its infraspecific taxa such as subsp. *stictica*), as well as closely related species within the *L. atrobrunnea* complex such as *L. promiscens* and *L. confluens*, and supports its treatment as a separate species rather than as part of an already described species complex

Overall, the discovery of *Lecidea devrimii* underlines the fact that the lichen biota of Antarctica, even within relatively well-known genera such as *Lecidea*, still contains hidden and unresolved taxonomic

diversity. The results of this study reinforce the value of integrative taxonomy for resolving species boundaries in morphologically conservative and chemically variable lichen groups. Future studies incorporating broader Antarctic sampling, additional molecular markers, and expanded comparative material will be important for further clarifying species relationships within the genus and for improving our understanding of the evolutionary and biogeographical diversity of Antarctic lichens.

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