



New record and new species of lichenized fungal genus *Candelariella* Müll. Arg. in Antarctica

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Abstract: Previously, only three *Candelariella* species were known from Antarctica: *C. aurella*, *C. flava* and *C. vitellina*. After morphologically and phylogenetically examining our collections on soil from James Ross Island, located in the north-east Antarctic Peninsula region, and Horseshoe Island, a small rocky island in Bourgeois Fjord, Marguerite Bay in the south-west Antarctic Peninsula, we describe the lichen species *Candelariella ruzgarii* as new to science. Sequences of the nrITS, mtSSU and RPB1 gene regions of the new species were amplified and revealed that the phylogenetic position of the new species is in the *C. aurella* group, which is characterised by 8-spored asci and $\pm$ granular thalli. *Candelariella ruzgarii* is phylogenetically most closely related to *C. aurella* s. lat. but differs mainly in ecology as the new species grows on soil or on terricolous lichens, whereas the latter species grows on calcareous rocks, rarely on wood. Morphologically, *C. ruzgarii* is very similar to *C. aggregata*, a Northern Hemisphere species that grows on mosses and plant debris. Apart from the different phylogenetical position, *C. ruzgarii* has a thicker and sometimes slightly crenulated thalline margin and somewhat shorter ascospores than *C. aggregata*. We also report *C. plumbea* for the first time from Antarctica, a species with a thick and grey thallus that was previously known from Europe and Asia.

Keywords: Antarctic, Antarctic Peninsula, biodiversity, lichenized fungi, *Candelariales*, *mtSSU*, *nrITS*, *RPB1*.



Introduction

Our lichen biodiversity studies started in 2016 on James Ross Island, which belongs to the North-East Antarctic Peninsula region and has a special location in the transition zone between maritime and continental climate types (Bednarek-Ochyra *et al.* 2000), and Horseshoe Island located in the south-west Antarctic Peninsula, clearly show that Antarctic lichen biodiversity is far from being fully known. Since 2016, our project team has described using molecular techniques four new lichenized fungal species from James Ross Island; namely *Toniniopsis bartakii* (Halıcı *et al.* 2021a), *Leptogium pirireisii* (Halıcı *et al.* 2022a) *Rhizocarpon ozsoyae* (Halıcı *et al.* 2022b), *Shackletonia backorii* (Halıcı *et al.* 2022c), and reported from this island 12 lichen species as new to Antarctica (Halıcı *et al.*, 2017, 2018, 2020, 2021b, 2022d; Halıcı and Barták 2019; Kahraman and Halıcı 2021).

The genus *Candelariella* is characterized by 8- or polyspored asci with hyaline and aseptate ascospores, biatorine or lecanorine apothecia, crustose or squamulose thalli and a secondary chemistry with pulvinic acid and derivatives, which give the thalli and apothecia its typical yellow to orange-yellow colour. The genus belongs to the family *Candelariaceae* and the order *Candelariales*, which comprises *c.* 50 species world-wide (Westberg 2005). To date, three species of this genus are known from Antarctica: *C. aurella* (Hoffm.) Zahlbr., *C. flava* (C.W. Dodge *et* G.E. Baker) Castello *et* Nimis and *C. vitellina* (Hoffm.) Müll. Arg. (Castello and Nimis 1994; Øvstedal *abd* Lewis Smith 2001). In this paper we describe *C. ruzgarii* as new to science and report *C. plumbea* Poelt *et* Vězda from Antarctica for the first time, increasing the number of *Candelariella* species to five from this continent.

Material and Methods

Morphology. — Lichen samples were collected from James Ross Island and Horseshoe Island in Antarctica. The type specimen and the other specimens detailed below are all deposited in Erciyes University Herbarium Kayseri, Turkey (ERCH). The lichen samples collected from James Ross Island and stored in ERCH have herbarium numbers starting with "JR". Morphological observations were made using an Olympus SZX7 stereomicroscope with macroscopic photographs taken with an Olympus SC30 digital camera. Anatomical observations were made using a Leica DM2500 light microscope, with the microscopic photographs taken with a Flexacam C1 digital camera. Measurements of anatomical structures always refer to water mounts, with at least 20 measurements made for all anatomical structures described in this study. Measurements are followed by the standard deviation (SD), N=the total number

of measurements for all samples of that species and n =the number of samples from which measurements were made.

Molecular methods: DNA isolation, PCR and sequencing. — Genomic DNA extraction was performed directly using 5–10 fresh apothecia. Genomic DNAs were extracted from newly collected specimens using the DNeasy Plant Mini Kit from Qiagen (Cat. No./ID: 69204) according to the manufacturer's instructions. Next, they were confirmed by gel electrophoresis of 1.2% agarose (containing 0.05 μ l/ml EtBr). Attempts were made to generate molecular data from the nuclear ribosomal ITS+5.8S region (nrITS), mitochondrial small subunit (mtSSU) and RNA polymerase II subunit (RPB1) to place the new species described below in a phylogenetic context. The nuclear rDNA ITS gene region was amplified by using the fungi-specific primer ITS1-F (5'-CTTGGTCATTTAGAGGAAGTAA-3') and the universal primer ITS4 (5'-TCCTCCGCTTATTGATATGC-3') (Gardes and Bruns 1993; White *et al.* 1990). The mtSSU gene region was amplified by using the primers mtSSU1F (GATGATGGCTCTGATTGAAC) (Shiguo and Stanosz 2001) and mtSSU3R (ATGTGGCACGTCTATAGCCC) (Zoller *et al.* 1999). The *RPB1* gene region was amplified using the primers RPB1-5F pelt 5'-TTCAACAARCTBACVAAR-GATGT-3' (Denton *et al.* 1998) and fRPB1-11aR 5'-GCRTGGATCTTRTCRTC-SACC-3' (Liu *et al.* 1999). PCR amplification for these primers were performed with total 50 μ l standard reaction volume for each sample. Optimum PCR amplification conditions were obtained with 25 μ l 2 $\times$ Taq PCR MasterMix in each tube with 15 μ l of distilled water, 50 ng/ μ l of extracted DNA and 10 μ M of the primers. The thermal cycling conditions included an initial denaturation step of 94°C for 5 min, followed by 35 cycles of 94°C for 60 s (denaturation), 55°C or 56°C and 54°C (nrITS, mtSSU and RPB1, respectively) for 60 s (annealing), and 72°C for 75s (extension) followed by a final extension period of 72°C for 10 min. The amplification results were visualized on 1.5% agarose gel and then sequenced bidirectionally. Sequence analyses of the lichen samples obtained from the PCR products were performed by the Epigen Biotechnology laboratory (Ankara, Türkiye).

Molecular methods: Sequence alignment and phylogenetic analysis. — All the ITS, mtSSU and RPB1 bidirectional sequences were edited in BioEdit V7.2.6.1 (Hall 1999). The final analyses included the newly generated sequences and the most similar *Candelariella* sequences (identity > 90%) according to BLASTN search (Altschul *et al.* 1990) in the GenBank database. All ITS, mtSSU and RPB1 sequences (Table 1) were aligned and edited manually using ClustalW in Bioedit V7.2.6.1. All missing and ambiguously aligned data and parsimony-uninformative positions were removed and only parsimony-informative regions were finally analysed in MEGA XI (Tamura *et al.* 2021). The final ITS alignment contained 550 positions and 66 sequences; the conserved sites contained 285 base pairs, the variable sites 246 base pairs, and the parsimony-informative sites 187 base pairs. The mtSSU alignment had 918 positions and 14 sequences; the

conserved sites contained 334 base pairs, variable sites 403 base pairs, and parsimony-informative sites 253 base pairs. The *RPB1* alignment had 1206 positions and 6 sequences and the conserved sites contained 684 base pairs, variable sites 458 base pairs, and parsimony-informative sites 151 base pairs. Phylogenetic trees with bootstrap values were obtained using the maximum likelihood method with a rapid bootstrap with 1000 bootstrap replications. *Pycnora sorophora* (Vain.) Hafellner was used as the outgroup since *Pycnora* has been recognized as a possible sister clade to *Candelariaceae* (Bendiksby and Timdal 2013).

Results

Sequence alignment and phylogenetic analysis. — Three independent phylogenetic trees for the genus *Candelariella* were produced from 73 sequences (60 for ITS, 9 for mtSSU and 4 for RPB1) from GenBank and 14 new sequences (6 for ITS, 5 for mtSSU and 2 for RPB1) from *C. plumbea* and *C. ruzgarii* (Table 1). All species names are followed by the GenBank accession numbers or voucher information (Table 1). The new species (*C. ruzgarii*) and the new record for Antarctica (*C. plumbea*) are strongly supported as a distinct clade in the ITS, mtSSU and RPB1 phylogeny (Figs. 1–3). The ITS tree shows very clearly that

Table 1.

GenBank accession numbers of specimens used in this study.

New sequences are indicated in bold.

Specimen	ITS	Locality	mtSSU	Locality	RPB1	Locality
<i>Candelariella aggregata</i>	MZ243491	USA	-	-	-	-
<i>Candelariella aggregata</i>	MZ243492	USA	-	-	-	-
<i>Candelariella aggregata</i>	MZ243493	USA	-	-	-	-
<i>Candelariella antennaria</i>	MZ922100	USA	-	-	-	-
<i>Candelariella antennaria</i>	MZ922096	USA	-	-	-	-
<i>Candelariella aurella</i>	OK332959	Czech Republic	AY853313	Sweden	DQ915594	Sweden
<i>Candelariella aurella</i>	MZ922102	USA	-	-	-	-
<i>Candelariella aurella</i>	MZ922092	USA	-	-	-	-
<i>Candelariella aurella</i>	MZ243507	USA	-	-	-	-
<i>Candelariella aurella</i>	MZ243506	USA	-	-	-	-
<i>Candelariella aurella</i>	MZ243505	USA	-	-	-	-
<i>Candelariella aurella</i>	MZ243504	USA	-	-	-	-

Specimen	ITS	Locality	mtSSU	Locality	RPB1	Locality
<i>Candelariella aurella</i>	MZ243503	USA	-	-	-	-
<i>Candelariella aurella</i>	MZ243502	USA	-	-	-	-
<i>Candelariella aurella</i>	MK812178	Norway	-	-	-	-
<i>Candelariella aurella</i>	MG271792	Turkey	-	-	-	-
<i>Candelariella aurella</i>	MG271784	Turkey	-	-	-	-
<i>Candelariella aurella</i>	MG271781	Turkey	-	-	-	-
<i>Candelariella aurella</i>	MG271780	Turkey	-	-	-	-
<i>Candelariella aurella</i>	MG271778	Turkey	-	-	-	-
<i>Candelariella aurella</i>	KT695405	Canada	-	-	-	-
<i>Candelariella aurella</i>	EF535163	USA	-	-	-	-
<i>Candelariella aurella</i>	EF535162	USA	-	-	-	-
<i>Candelariella aurella</i>	EF535161	Sweden	-	-	-	-
<i>Candelariella biatorina</i>	EF535164	USA	-	-	-	-
<i>Candelariella biatorina</i>	EF535165	USA	-	-	-	-
<i>Candelariella blastidiata</i>	KX853123	Russia	MH156655	Czech Republic	-	-
<i>Candelariella blastidiata</i>	MN103124	China	-	-	-	-
<i>Candelariella californica</i>	EF535169	USA	-	-	-	-
<i>Candelariella canadensis</i>	MG694271	Canada	-	-	-	-
<i>Candelariella citrina</i>	MZ922104	USA	-	-	-	-
<i>Candelariella citrina</i>	MZ922105	USA	-	-	-	-
<i>Candelariella clarkii</i>	KR052103	USA	-	-	-	-
<i>Candelariella clarkii</i>	KR052104	USA	-	-	-	-
<i>Candelariella complanata</i>	EF535173	Mexico	-	-	-	-
<i>Candelariella complanata</i>	EF535174	Mexico	-	-	-	-
<i>Candelariella coralliza</i>	-	-	AY853314	Sweden	-	-
<i>Candelariella corviniscalensis</i>	GU967377	USA	-	-	-	-
<i>Candelariella deppeanae</i>	EF535179	Mexico	-	-	-	-
<i>Candelariella deppeanae</i>	NR119640	USA	-	-	-	-
<i>Candelariella faginea</i>	MK778596	Russia	MK778524	Russia	-	-
<i>Candelariella flava</i>	MZ919298	Antarctica	-	-	-	-
<i>Candelariella flava</i>	MZ919299	Antarctica	-	-	-	-
<i>Candelariella granuliformis</i>	GU967375	Sweden	-	-	-	-
<i>Candelariella granuliformis</i>	GU967376	Sweden	-	-	-	-
<i>Candelariella kansuensis</i>	EF535181	USA	-	-	-	-

Specimen	ITS	Locality	mtSSU	Locality	RPB1	Locality
<i>Candelariella kansuensis</i>	FJ959349	USA	-	-	-	-
<i>Candelariella placodizans</i>	-	-	KJ766368	-	-	-
<i>Candelariella plumbea</i>	MN103123	China	-	-	-	-
<i>Candelariella plumbea</i> (JR 0.206)	OP003525	James Ross I. Antarctica	-	-	-	-
<i>Candelariella reflexa</i>	EF535189	Norway	DQ912272	-	DQ912354	-
<i>Candelariella reflexa</i>	EF535190	Norway	-	-	-	-
<i>Candelariella rosulans</i>	MZ922106	USA	-	-	-	-
<i>Candelariella rosulans</i>	MZ922108	USA	-	-	-	-
<i>Candelariella ruzgarii</i> (HS 0.200)	OP021662	Horse Shoe I. Antarctica	OP021658	Horse Shoe I. Antarctica	-	-
<i>Candelariella ruzgarii</i> (JR 0.355)	OP021660	James Ross I. Antarctica	OP021661	James Ross I. Antarctica	OP018966	James Ross I. Antarctica
<i>Candelariella ruzgarii</i> (JR 0.370)	OP021666	James Ross I. Antarctica	OP082320	James Ross I., Antarctica	OP018967	James Ross I. Antarctica
<i>Candelariella ruzgarii</i> (JR 0.395)	OP021663	James Ross I., Antarctica	OP021664	James Ross I., Antarctica	-	-
<i>Candelariella ruzgarii</i> (JR 0.396)	OP021665	James Ross I. Antarctica	OP082321	James Ross I. Antarctica	-	-
<i>Candelariella spraguei</i>	EF535194	USA	-	-	-	-
<i>Candelariella spraguei</i>	EF535195	USA	-	-	-	-
<i>Candelariella subdeflexa</i>	MT614341	Sweden	-	-	-	-
<i>Candelariella subdeflexa</i>	MG271794	Turkey	-	-	-	-
<i>Candelariella subsquamulosa</i>	MG694274	Korea	-	-	-	-
<i>Candelariella subsquamulosa</i>	MG694275	Korea	-	-	-	-
<i>Candelariella terrigena</i>	MG271775	Turkey	-	-	DQ986816	-
<i>Candelariella terrigena</i>	HQ650602	USA	-	-	-	-
<i>Candelariella vitellina</i>	-	-	AY853314	Sweden	-	-
<i>Pycnora sorophora</i>	OK333010	Czech Republic	MN508293	Alaska	MH468797	-

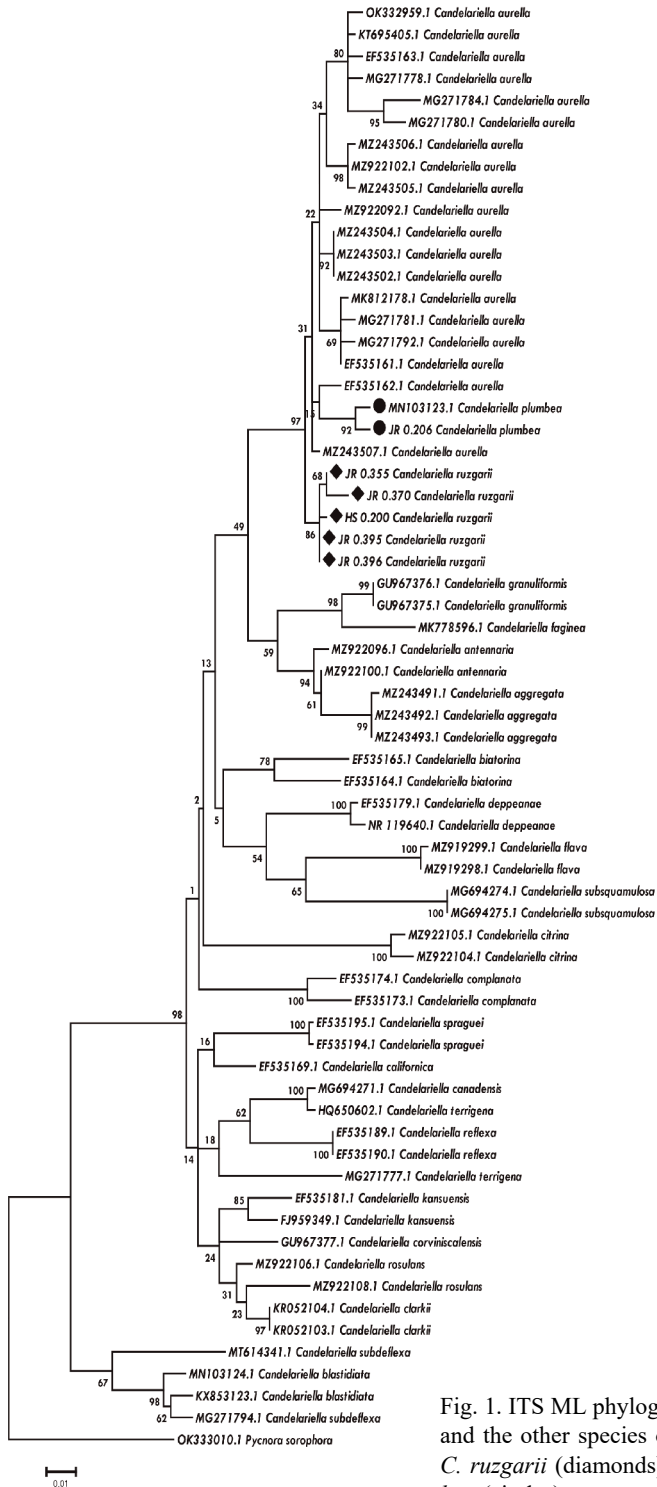


Fig. 1. ITS ML phylogeny of *C. ruzgarii*, *C. plumbea* and the other species of the genus. The new species *C. ruzgarii* (diamonds) and the new record *C. plumbea* (circles) are presented in JR code.

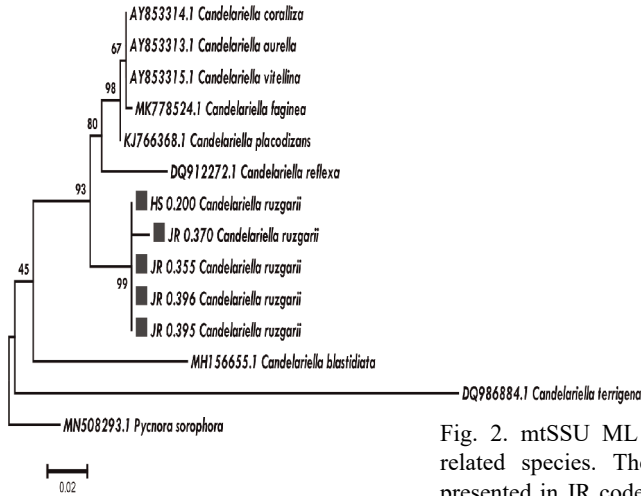


Fig. 2. mtSSU ML phylogeny of *C. ruzgarii* and related species. The new species *C. ruzgarii* is presented in JR code.

the new species is located in a separate clade represented by a bootstrap value of 86 (Fig. 1). According to the *ITS* data, the new species is closely related to *C. aurella* (Hoffm.) Zahlbr. and *C. plumbea* Poelt et Vězda. As seen in the *ITS* phylogenetic tree, the new record *C. plumbea* matched with the GenBank number MN103123, *C. plumbea* (Fig. 1). The mtSSU and RPB1 tree shows clearly that the new species is located in a separate clade represented by a bootstrap value of 100 (Figs. 2 and 3). The phylogenetic analyses did not designate any species identical to the new species in the genus *Candelariella*.

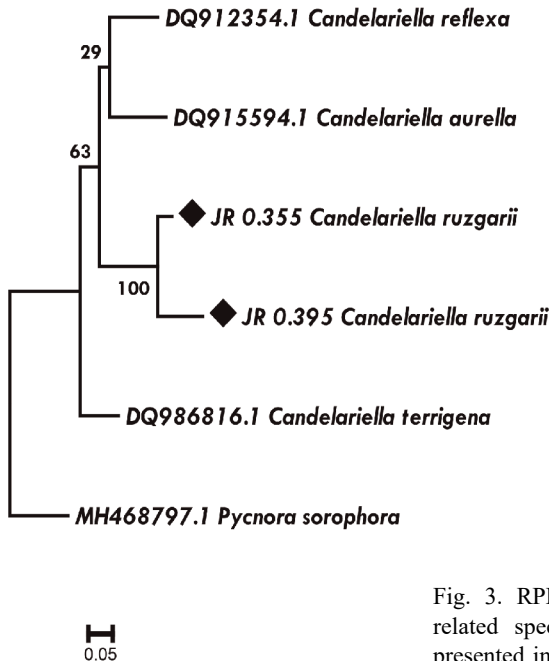


Fig. 3. RPB1 ML phylogeny of *C. ruzgarii* and related species. The new species *C. ruzgarii* is presented in JR code.

Taxonomy

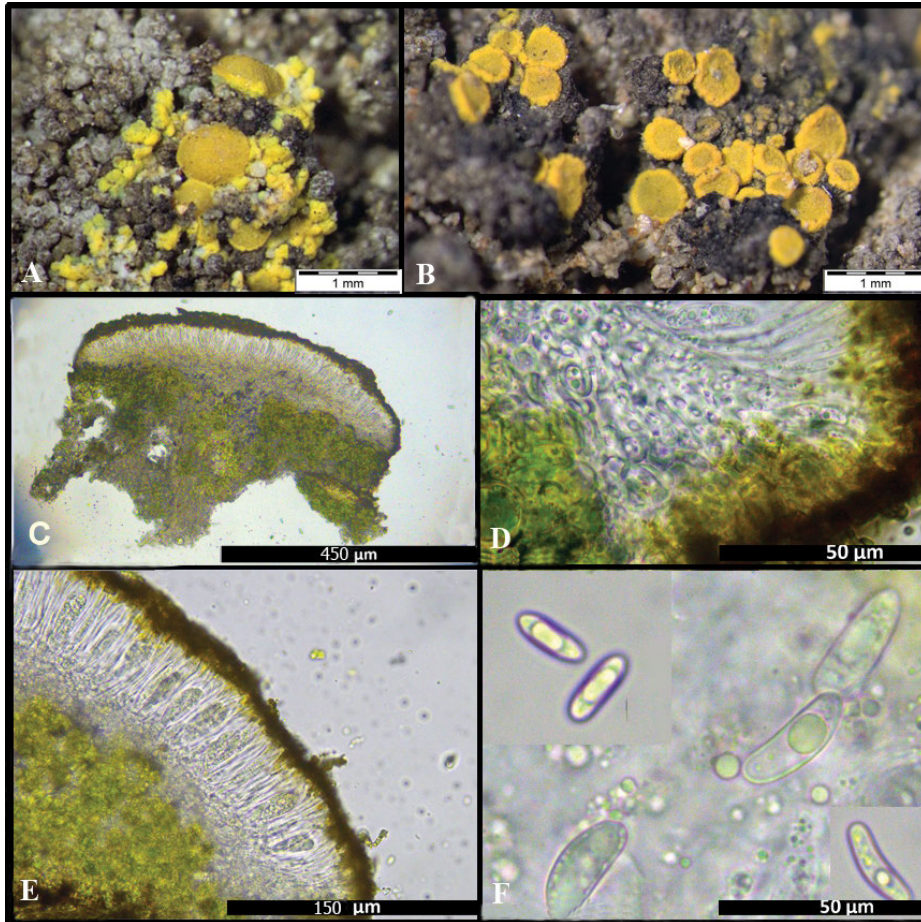


Fig. 4. *Candelariella ruzgarii*. **A.** Specimen growing on soil. **B.** Specimen growing on other terricolous lichens. **C.** Section of the apothecium (taken from the middle of the apothecium), showing the algal layer continuous below the hypothecium. **D.** Close-up showing the isodiametric cells of the proper margin. **E.** Close-up showing that proper margin not forming a distinct stipe. **F.** Ascospores

Mycobank No.: MB 845684

Type: Antarctic Peninsula, James Ross Island: south-east part of Johnson Mesa, 63°49'46.2"S 57°54'21.6"W, alt. 292 m, 21 January 2017, leg. M. G. Halıcı, *ERCH JR 0.395* (ERCH—holotype).

Diagnosis: It is similar to *C. aggregata* M. Westb. in its ecology and in the proper margin not forming a distinct, compact stipe at the base of the apothecia, but it differs in having smaller ascospores. It differs from the phylogenetic sister species *C. aurella* s. lat. since it grows on soil or other terricolous lichens and does not form a distinct, compact stipe at the base of the apothecia.

Etymology: Named in honour of Rüzgar Kağan Halıcı, dear son of the first author, because of the sacrifices he made during his father's Antarctic expeditions.

Description: Thallus crustose, granular or not well developed. Granules, when present, scattered to continuous, the older ones yellow to orange yellow, the younger ones with a greenish tinge, 0.1–0.2 mm wide. Cortex pseudoparenchymatous, up to 15–20 μm thick, consisting of gelatinized hyphae with isodiametric cells, $4\text{--}8 \times 3.5\text{--}6 \mu\text{m}$. Algal layer 60–75 μm thick, algal cells chlorococcoid, $10\text{--}15 \times 11\text{--}12 \mu\text{m}$. Apothecia lecanorine, abundant, scattered to aggregated, sessile, (0.2–)0.25–0.6–0.95(–1.2) mm (n=25). Disc flat or slightly concave and with a greenish tinge when young, dark yellow to orange-brownish yellow and more convex when older, rounded to angular shaped. Thalline margin persistent, up to 0.2 mm thick, sometimes slightly crenulate, with a pseudoparenchymatous cortex. Proper margin indistinct to rarely visible as a greyish rim, not forming a distinct stipe at the base of the apothecia, 50–60 μm thick consisting of isodiametric cells, $3.5\text{--}9 \times 3\text{--}6 \mu\text{m}$. Epihymenium golden brown or yellowish brown, (25–)25.5–40–55.5(–60) μm (n=15). Hymenium hyaline or sometimes with golden brown tinge, (30–)40–60–80(–95) μm (n=15). Paraphyses simple, thin, unbranched, sometimes branched near the tips, septate with oil droplets, tips capitate, 3–7 μm . Hypothecium hyaline, sometimes with golden brown tinge, (25–)30–80–120(–175) μm (n=15). Asci 8-spored, (25–)27–35.5–44(–55) $\times$ (8–)12–15–18(–20) μm (n=15). Ascospores hyaline, aseptate, ellipsoid or oblong, sometimes slightly narrowing at one end, straight or mostly curved, oil droplets present or not, (9–)11–13.5–16(–19) $\times$ (4–)4.5–6–7.5(–9) μm (n=50), length/width ratio (1.43–)1.78–2.24–2.7(–3.8) μm (n=50). Pycnidia not observed. Spot tests all negative.

Specimens studied: Antarctic Peninsula, Sally Cove, Horseshoe Island: Bourgeois Fjord, Marguerite Bay, near Base Y, 67°48'30"S 67°17'39"W, 17 February 2022, leg. M. G. Halıcı, *ERCH JR 0.200* (ERCH); James Ross Island: Berry Hill Mesa, 63°48'42.0", 57°50'5.4" W, alt. 345 m., 23 January 2017, leg. M. G. Halıcı, *ERCH JR 0.355* (ERCH); Puchau, 63°48'24.9" S, 57°50'27.6" W, alt. 142 m., 24 January 2017, leg. M. G. Halıcı, *ERCH JR 0.370* (ERCH); Johnson Mesa, 63°49'46.2" S, 57°54'21.6" W, alt. 292 m., 27 January 2017, leg. M. G. Halıcı, *ERCH JR 0.396* (ERCH).

The Antarctic specimen was collected from James Ross Island on basaltic rocks in a locality with a lot of skua nests. It has a thick subsquamulose to squamulose grey thallus. Apothecia lecanorine, abundant, sessile, 0.1–0.2 mm (n=10). Disc flat or slightly concave, dark yellow to orange-brownish yellow, rounded to $\pm$ angular shaped. Proper margin not forming a distinct stipe at the base of the apothecia unlike *C. aurella* s. lat. Asci 8-spored, (25–)27–35.5–44(–55) $\times$ (8–)12–15–18(–20) μm (n=15). Ascospores hyaline, aseptate, ellipsoid or oblong, sometimes slightly narrowing at one end, straight or mostly curved, oil droplets present or not, (9–)11–13.5–16(–19) $\times$ (4–)4.5–6–7.5(–9) μm (n=50),

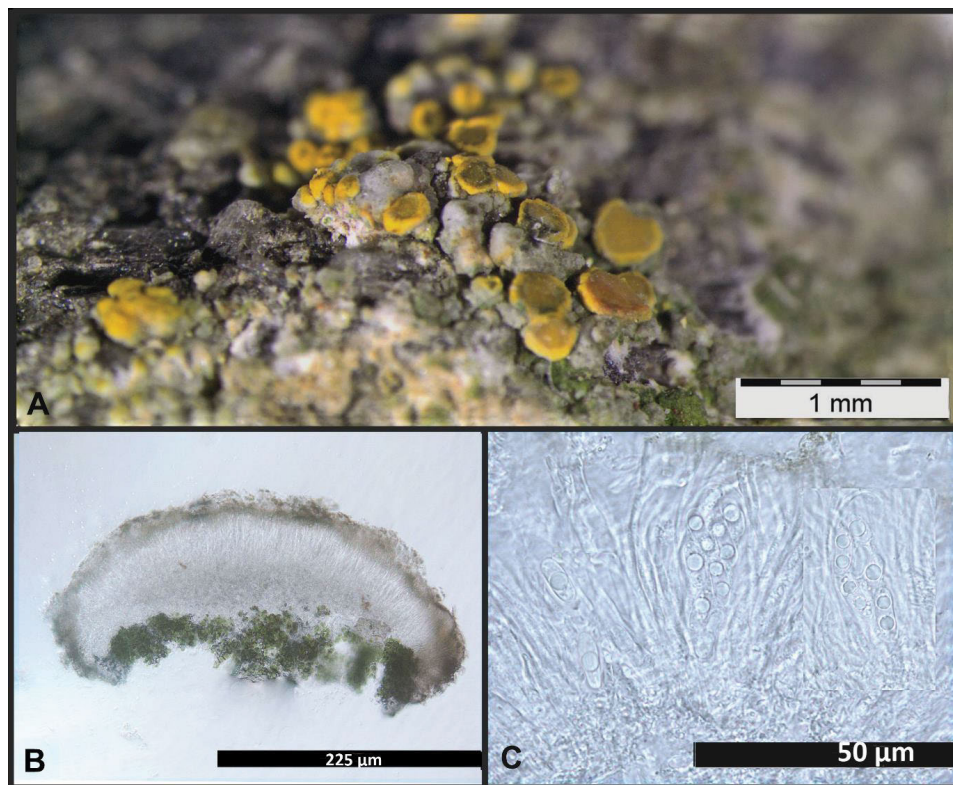


Fig. 5. *Candelariella plumbea*. A. Habitus. B. Section of the apothecium, showing the algal layer continuous below the hypothecium. C. Asci and Ascospores.

length/width ratio (1.43–)1.78–2.24–2.7(–3.8) µm (n=50). Pycnidia not observed. Spot tests are all negative.

Specimen examined: Antarctic Peninsula, James Ross Island: around Lachman Lake, 63°48'36"S, 57°48'29"W, alt. 36 m, 17 January 2017, leg. M. G. Halıcı, *ERCH JR 0.206*.

Discussion

According to our *ITS* phylogeny, the new species, *C. ruzgarii*, is the phylogenetic sister species to *C. aurella* and *C. plumbea*. Both of these species are epilithic species unlike the terricolous *C. ruzgarii*. *Candelariella aurella* is a cosmopolitan species known from all continents growing on calcareous rocks and rarely on wood (Westberg 2007). This species differs from *C. ruzgarii* in the anatomy of the proper margin. Whereas the proper margin forms a distinct, compact stipe composed of strongly gelatinized, mostly thick-walled hyphae with elongated to rectangular cells at the base of the apothecia in *C. aurella* (illustrated in Westberg 2007 page 110), the stipe is indistinct in *C. ruzgarii* and composed of

thin-walled hyphae with $\pm$ isodiametric cells. In addition, the ascospores appear to be wider in *C. ruzgarii* [(4–)4.5–6–7.5(–9) μm] than in *C. aurella* (4–5(–6) μm ; Westberg 2007). The other phylogenetic sister species, *Candelariella plumbea*, has a thick grey thallus, unlike that of *C. ruzgarii*, which is yellow.

Morphologically and ecologically *C. ruzgarii* is most similar to *C. aggregata*, a species growing on mosses and plant debris in alpine heaths and arid steppe habitats (Westberg and Sohrabi 2012). The latter species was described from North America (Westberg 2007) and is also known from the Murmansk Region in Russia (Urbanavichus *et al.* 2008), Europe (Westberg and Clerc 2012), Iran (Westberg and Sohrabi 2012) and Turkey (Halıcı *et al.* 2012). These two species and *C. antennaria* Räsänen, a species growing on bark with a grey thallus, share a similar proper margin anatomy as the proper margin does not form a distinct, compact stipe. Apart from the different phylogenetical position, *C. ruzgarii* also has a thicker and sometimes slightly crenulated thalline margin and somewhat shorter ascospores [(9–)11–13.5–16(–19) μm] than *C. aggregata* [(10.5–)13–19(–24) μm ; Westberg 2007].

The type specimen of *C. cerinella* var. *unilocularis*, which is known to grow on rocks and salted soil in southern Russia (Elenkin 1907), was considered to be synonym of *C. aurella* by Khodosovtsev (2005) and Westberg and Sohrabi (2012). Westberg and Sohrabi (2012) indicated that the specimens reported as “var. *unilocularis*” may also belong to an undescribed species and/or to *C. aggregata*. Later, Otte *et al.* (2013) described *C. commutata* Otte *et al.* M. Westb., a species growing on soil and plant debris on calcareous ground from the alpine belt of the Caucasus and the Alps, and also from Scandinavia. They suggested that some of the specimens previously reported as *C. aurella* var. *unilocularis* actually belonged to *C. commutata*, species with much larger apothecia (up to 2 mm in diam.) and much longer ascospores (20–28(–32) μm) than *C. ruzgarii*.

Another terricolous species, *C. canadensis* H. Magn., known from Arctic North America, Greenland and the Himalayas differs from *C. ruzgarii* by having a much better developed, lobate thallus (Westberg 2010). Westberg (2010) considered *C. nepalensis* and *C. hudsonica* as synonyms of *C. canadensis*. *Candelariella citrina* de Lesd. is a common terricolous lichen in south-west North America and differs from *C. ruzgarii* by having greenish squamules that are pruinose (Westberg 2007). *C. terrigena* Räsänen was considered as a synonym of *C. citrina* by Westberg (2007).

Candelariella placodizans (Nyl.) H. Magn., an Arctic-Alpine terricolous species known from the Himalayas in Asia and the Rocky Mountains in North America (Westberg 2007, 2010) differs from *C. ruzgarii* by having polyspored asci and a greenish yellow, subsquamulose thallus with a “pruinose” surface (Westberg and Sohrabi 2012). *Candelariella kuusamoensis* Räsänen and *C. vitellina* can sometimes grow on soil but unlike *C. ruzgarii* has polyspored asci (Räsänen 1939).

Candelariella flava, a species known from Antarctica and Falkland Islands also grows on mosses and on other lichens but it has a well-developed thallus and

unlike *C. ruzgarii* apothecia are rarely seen in this species, (Castello and Nimis 1994; Øvstedal and Lewis Smith 2001; Fryday *et al.* 2019).

Candelariella spraguei (Tuck.) Zahlbr., a species known from North America and Russia sometimes grows on soil or mosses in rock crevices, but differs from *C. ruzgarii* by having acicular and much longer ascospores [(30.5–)34.5–48(–57) μm vs. (9–)11–13.5–16(–19) μm] (Westberg 2007).

Candelariella plumbea was described from Romania by Poelt *et Vězda* (1976) from lowlands, but according to Nimis (2003), this species is an Arctic-Alpine lichen. This is supported by Halıcı *et al.* (2012) as this species was reported from 2300 m altitude in Turkey. *C. plumbea* was previously reported from Europe, Asia and North America (Weber 1990). The first author's collection from James Ross Island has a thick subsquamulose to squamulose grey thallus and 8-spored asci and we did not hesitate to include it under *Candelariella plumbea*, which is here reported as new to the Southern Hemisphere and Antarctica.

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