

Two New Species of the Family Acarosporaceae from South Korea

Jung Shin Park , Young-Nam Kwag , Sang-Kuk Han  and Soon-Ok Oh 

Division of Forest Biodiversity, Korea National Arboretum, Pocheon, South Korea

ABSTRACT

Acarosporaceae is a crustose lichen and is known as a species that has more than 50 multi-spores, and has hyaline spores. Those taxa are often found in rock and soil in mountain areas or coastal regions in Korea, and very diverse forms and species are known. However, after an overall genetic phylogenetic analysis of carbonized ascomata in 2015, species consisting only of the morphological base are newly divided, and several species of *Acarosporaceae* in Korea are also being discovered in this situation. As a result of analysis using internal transcribed spacer (ITS) and nuLSU gene analysis, Korean species belonged to *Acarospora* and *Sarcogyne* clade, and *Acarospora* classified as the *Acarospora* clade was mixed with the *Polysporina* group and the *Sarcogyne* clade is mixed with the *Acarospora*. We identified two new species (*Acarospora beangnokdamensis* J. S. Park & S. O. Oh, sp. nov., *Sarcogyne jejuensis* J. S. Park & S. O. Oh, sp. nov.) through morphological, molecular, and secondary metabolite substance and found one new record (*Sarcogyne oceanica* K. Knudsen & Kocourk). We have made a classification key for *Acarospora* and *Sarcogyne* in Korea and reported all information together here.

ARTICLE HISTORY

Received 12 July 2023
Accepted 15 August 2023

KEYWORDS

Lichenized Ascomycota;
carbonized ascomata;
phylogenetic; taxonomy

1. Introduction

Acarosporaceae is one of the taxonomic groups in which movement between genera is becoming active with the recent development of research based on molecular biology. The character of this genera, has an ascus with over 100 spores (polysporous), the hyaline spores of about 3–5 µm in size, the wall of the asci is thick, and the tholus has a clear structure [1]. Currently, seven genera (*Acarospora*, *Caeruleum*, *Myriospora*, *Pleopsidium*, *Sarcogyne*, *Timdalia*, and *Trimmatothelopsis*) are included using the molecular biological classification method along with morphological classification [2]. *Thelocarpella*, which previously belonged to *Acarosporaceae*, was transferred to *Trimmatothelopsis* in 2016 together with some genera of *Acarospora* (*Acarospora rhizobola*, *A. terricola*) and *Melanophloea* (*Melanophloea americana*, *M. coreana*, *M. montana*) in the study of *Trimmatothelopsis* [2]. *Glypholecia* and *Lithoglypha* are also presumed to be included in *Acarosporaceae*, but *Glypholecia* has not been investigated molecularly, but the study confirmed that it is in the *Sarcogyne* clade [3]. *Lithoglypha* also has not been investigated molecularly, but it is thought to belong to *Trimmatothelopsis* because it has the characteristics of long conidia [2]. In addition, although *Acarosporaceae* is classified using molecular biology

study, it is still actively moving from *Acarospora* to other genera only by morphological classification study [4,5]. In Korea, 17 species of *Acarosporaceae*, which are seven species of *Acarospora* (*Acarospora fuscata*, *A. insolata*, *A. hospitans*, *A. nitrophila*, *A. ulleungdoensis*, *A. veronensis*, *A. versicolor*), five species of *Sarcogyne* (*Sarcogyne clavus*, *S. endopetrophila*, *S. privigna*, *S. regularis*, *S. ulleungdoensis*), two species of *Myriospora* (*Myriospora rufescens*, *M. smaragdula*), two species of *Polysporina* (*Polysporina golubkova*, *P. simplex*), and one species of *Trimmatothelopsis* (*Trimmatothelopsis coreana*) have been reported through morphological identification [6–16]. In addition, recently, while arranging *Acarospora* in Korea, a species sample previously thought to be *Acarospora* was identified as a clearly separated genus *Trimmatothelopsis* through molecular biological methods, and two new species were found (on publishing). Recently, many lichens that grow on seashore rocks and on the coast have been collected through investigation of stone cultural heritage and coastal investigation. In particular, the collection of genera belongs to *Acarosporaceae* increased, but overall research was insufficient due to morphological similarity and lack of expert. The genus *Acarospora* is a central genus constituting *Acarosporaceae*, and many genera have been separated from this genus and are moving to

CONTACT Soon-Ok Oh  okkass15@korea.kr

Myriospora, *Pleopsidium*, and *Trimmatothelopsis*. *Sarcogyne* is also a taxon deeply related to *Acarospora*, and research in Korea, which constitutes one axis of East Asia, should be conducted [3,17]. The major difference between two species is the presence or absence of carbonized margin. *Acarospora* is characterized by polyspored asci with simple hyaline ascospores, ascomata that are immersed, pseudolecanorine or lecideine apothecia thallus that are areolate to squamulose growing on rock and soil [18]. *Sarcogyne* is characterized by often immersed and usually inconspicuous thallus, reddish brown to black apothecia with a lecideine exciple, a non-carbonized epihymenium, simple to sparingly branched paraphyses [19]. Some *Sarcogyne* species were segregated into *Polysporina* and divided into the reaction of apothecial wall and the characteristics of the richly branched and anastomosed paraphyses, and the presence of a carbonized epihymenium [20,21]. The two species are worldwide, and *Acarospora* is found xerothermic and arid habitats. *Sarcogyne* is known as a species that occurs in temperate and semi-arid areas, and mainly in the Northern Hemisphere [22,23]. Among the *Acarosporaceae* known in Korea, *Acarospora* and *Sarcogyne*, which are actively moving between genera, will organize Korean *Acarosporaceae* and conduct this study. Our research aims to identify lichens collected from all over the county based on morphological, chemical, and molecular biology, and to report newly discovered species or lichen identified as new species. In addition, based on the taxonomic description and classification keys of past researchers, a Korean classification key was made and reported here.

2. Materials and methods

2.1. Morphological examination

We performed morphological identification on all specimens identified as *Acarospora* and *Sarcogyne* in KH (Korea National Herbarium, Pocheon). Samples of the *Acarospora* and *Sarcogyne* were collected in South Korea from 2009 to 2022. We used air-dried materials overserved under the dissecting microscope (Olympus SZX7; Olympus, Tokyo, Japan) and compound microscope (Olympus CX22LED; Olympus). External structures, such as the thallus and the shape of the apothecia, were confirmed through a stereo microscope. Under the compound microscope, internal structure of the thallus to apothecia was mainly confirmed, and hyphae, algae size, spore morphology, and the structure of the asci were confirmed. Since *Acarospora* is classified according to the presence of gyrophoric acid, the presence of the substance was confirmed through

the Thin-layer chromatography (TLC) method using solvent C (toluene:acetic acid = 85:15).

2.2. DNA extraction and PCR amplification

For molecular analysis, fragments of lichen thallus or apothecia were used as DNA extraction material. DNA extraction was performed on thallus fragments using the DNeasy Plant Mini Kit (Qiagen, Valencia, CA), according to the manufacturer's instructions. PCR amplifications were conducted using the AmpliTaq DNA polymerase (Thermo Fisher Scientific, Waltham, MA). The following primers were used for PCR amplifications: internal transcribed spacer (ITS) and large subunit ribosomal RNA (LSU) for ITS1F, LR5 [24,25]; mtSSU1 and mtSSU3R for mtSSU (mitochondrial small subunit) [26,27]. The following program was used for the amplification of ITS, LSU, and mtSSU: initial denaturation at 95 °C for 5 min, followed by 35 cycles of 95 °C for 20 s, 55 °C for 20 s, and 72 °C for 30 s, and then a final extension step at 72 °C for 3 min. PCR program was performed using PCR pre-mix (AccuPower PCR PreMix, BiONEER, Daejeon, South Korea) according to the manufacturer's recommended protocol. The amplified DNA was concentrated and purified using a Quick-spin PCR Product Purification Kit (INTRON Biotechnology, Inc., Seongnam City, South Korea) for sequencing analysis.

2.3. Sequence alignments and phylogenetic analysis

The sequences were initially aligned using ClustalW ver. 1.83 [28] and edited using the Bioedit program. We drew the phylogenetic tree by adding the sequences obtained from the sequences used in Wedin's study and Knudsen's study [3,19] downloaded from GenBank. All positions containing gaps and missing data were eliminated. The analysis involved 106 nucleotide sequences chosen from the regions of ITS1, 5.8S, ITS2, and nrLSU, which have been deposited in GenBank (listed in Table 1). Seventeen new sequences belonging to *Acarosporaceae* were obtained and added to the constructed phylogenetic tree. There were a total of 1182 positions in the final dataset. Tree inference using Bayesian analysis was performed using MrBayes on XSEDE version 3.2.6. models on the Cipres Web Portal (http://www.phylo.org/sub_sections/portal). Two independent parallel runs of four Metropolis-coupled Monte Carlo Markov chains (MCMC) were run, sampling every 1000 times generation for 10 million total generations. After discarding the first 25% of the sampled trees as burn-in, the remaining trees were determined by calculating a

majority-rule consensus tree with posterior probabilities (PPs). Phylogenetic trees were drawn using Figtree v. 1.3.1 [29].

3. Results and discussion

3.1. Phylogenetic analysis

We generated a phylogenetic tree containing ITS, nuLSU sequences from the *Acarosporaceae* specimens. For comparison with *Acarosporaceae* in South Korea, existing sequences were used by downloading previously used sequences from GenBank (<https://www.ncbi.nlm.nih.gov/genbank/>) (Table 1). There are 17 newly added sequences in this tree, and the built-up to the new sequences is shown in Figure 1 (*A. beangnokdamensis* (2), *A. fuscata* (4), *A. hospitans* (2), *A. nitrophila* (3), *Sarcogyne endopetrophila* (1), *S. jejuensis* (2), and *S. oceanica* (3)). In this tree, the relationship between each close species was high, but the relationship between non-sister clade was low. Clades were largely divided into *Acarospora* clade and *Sarcogyne* clade, and newly extracted sequences were distributed to each clade according to the genus classification. As in the previous study, the *Sarcogyne* clade contained *A. hospitans*, *A. insolata*, and *A. impressula* classified as genus *Acarospora*, and genus *Polysporina* was conformed to be polyphyletic and spread in the *Acarospora* clade [3]. A new species, *A. beangnokdamensis*, showed a sister clade relationship with *Acarospora wahlenbergii* and showed a high relationship (1). It was confirmed to be in a completely different clade genetically from *A. fuscata*, which has the most similar species and shows a high value (1). *A. fuscata* showed a high relationship with another previous reported *A. fuscata* sequences, and there was no generic difference with *A. umblicata*, which is distinguished by the difference in areoles with fissured unit margins (*A. fuscata*) to areolate-squamulose with lobulate margin (*A. umblicata*) and showed a high similarity (1) [23]. *Acarospora nitrophila* is grouped into one clade, and showed high sequences values with sister clade *Acarospora anomala*, *Acarospora placodiiformis*, and *Acarospora schleicheri* (0.92). Korea *Acarospora hospitans* is a high relationship with *Acarospora impressula*, *A. hospitans* (Norway), and *A. insolata*, which can be compared in previous study (1) [3]. Before classification through DNA, the new species, *Sarcogyne jejuensis*, was thought to have high morphological similarities with *Acarospora badiofusca*. This was because, unlike the genus *Sarcogyne*, which mainly had a black to dark brown apothecia with indistinct thallus, the margin was distinct, and the thallus was present in the brown apothecia. However, as a result of DNA analysis, it belonged to the genus *Sarcogyne*

and was classified as a distinct clade from other species (1). *Sarcogyne oceanica* is a newly reported species in Korea. This species showed a high relationship to *Sarcogyne algoviae*. These two species appear to have generic differences, either margin incised or not, or differences in substrate characteristic (growing on calcareous substrates or non-calcareous substrates such as siliceous rock). This species has morphological similarity with *Sarcogyne hypophaea*, but there is a difference in the color of hypothecium has dark brown to hyaline, and for these reasons, it seems to have been divided into other clades. It can be seen that the two new species and one reported species identified in this study show high support values and are divided from already reported species with high supported value. This study focused on discovering new and unreported species. In the next study, based on reported species, the Korean distribution, morphology, and generic characteristics of *A. hospitans* and *A. insolata*, which are currently thought to have ambiguous clade relationship, will be studied.

4. Taxonomy

4.1. New species

1. *Acarospora beangnokdamensis* J. S. Park & S. O. Oh, sp. nov. (Figure 2).

Mycobank: Similar to *Acarospora fuscata*, but differing in areole contiguousness, angular areole, dark brown upper surface color (vs. light brown), having euamyloid (IKI + blue) reaction, and pycnidia obviously observed.

Type: South Korea, Jeju-do (Prov.), Seogwipo-si, Mt Halla, Baengnokdam Lake, 33°21'35.2"N, 126°32'2.8"E, 1907 m, on rock, July 20 2016, S. O. Oh, C. S. Kim, Y. N. Gwag, J. W. Jo, S. K. Han, KL16-0292 (holotype: KHL0007822).

Etymology: It is named after the type locality, i.e., Baengnokdam lake, Jeju-do, South Korea.

Description: Thallus areolate, indeterminate of areoles often continuous, dispersed toward the margin, flat, areoles 0.5–2 mm wide, 0.25–0.39 mm thick, some areoles round but mostly angular, not rugulose, predominately sterile, replication by division, scattered among other lichen (e.g., *Aspicilia* sp., *Buellia* sp.) or in small patches to less than 1 cm often formed by dividing thalli. Upper surface brown to golden brown, matt, epruinose. Lower surface brown. Epicortex 5–6 µm thick, thin. Cortex 25–30 µm thick, upper layer light brown, lower layer hyaline, cell mostly round, 4–5 µm wide. Algal layer 87.5–125 (–150) µm thick, uniform and even, uninterrupted by hyphal bundles, continuous beneath apothecia, algal cells 4–5 µm in diam. Medulla 100–125 µm thick, medullary hyphae thin-walled, intricate, 2–2.5 µm wide, lower cortex distinct, brown 12.5–

Table 1. A list of GenBank accession number and voucher for sequenced species used in paper.

Name of species with strain number	Origin	Voucher	nITS	mtSSU
<i>Acarospora anomala</i> SAR136	Sweden, Dalarna	Westberg 10-106 (S)	LN810758	.
<i>Acarospora anomala</i> SAR138	Sweden, Dalarna	Westberg 10-108 (S)	LN810759	.
<i>Acarospora atrata</i> SAR69	Norway, Vest-Agder	Westberg 08-125 (S F124797)	LN810761	.
<i>Acarospora atrata</i> WE16	Sweden, Halland	Arup L02737 (LD)	LN810760	.
<i>Acarospora badiofusca</i> WE05	Sweden, Östergötland	Nordin 5552 (UPS L-124833)	LN810762	.
<i>Acarospora badiofusca</i> WE11	Sweden, Jämtland	Nordin & Owe-Larsson 36 (UPS)	LN810763	.
<i>Acarospora beangnokdamensis</i> KL16-0292	South Korea, Seogwipo-si	KHL0007822 (KH)	OQ629804	n/a
<i>Acarospora beangnokdamensis</i> KL22-0363	South Korea, Seogwipo-si	KHL0037531 (KH)	OQ629805	n/a
<i>Acarospora brodoana</i> SAR270	U.S.A., California, San Bernardino Co.	Knudsen 14712 & Kocourková (S F256014)	LN810882	.
<i>Acarospora cervina</i> SAR144	Switzerland, Valais	Westberg 10-172 (S F177758)	LN810764	.
<i>Acarospora cervina</i> SAR200	Sweden, Uppland	Westberg SAR200 (S)	LN810765	.
<i>Acarospora fuscata</i> SAR120	Sweden, Gotland	Westberg SAR120 (LD)	LN810766	.
<i>Acarospora fuscata</i> SAR129	Sweden, Hälsingland	Westberg SAR129 (LD)	LN810767	.
<i>Acarospora fuscata</i> KL16-0146	South Korea, Sinan-gun	KHL0007676 (KH)	OQ629806	OQ641679
<i>Acarospora fuscata</i> KL16-0147	South Korea, Sinan-gun	KHL0007677 (KH)	OQ629807	OQ641680
<i>Acarospora fuscata</i> KL21-0849	South Korea, Yangyang-gun	KHL0036562 (KH)	OQ629808	OQ641681
<i>Acarospora fuscata</i> KL21-1245	South Korea, Yangyang-gun	KHL0036956 (KH)	OQ629809	OQ641682
<i>Acarospora glaucocarpa</i> s. lat. SAR53	Sweden, Härjedalen	Westberg SAR53 (LD)	LN810770	.
<i>Acarospora glaucocarpa</i> s. lat. SAR56	Norway, Østfold	Westberg 08-083 (S)	LN810771	.
<i>Acarospora glaucocarpa</i> s. str. SAR08	Sweden, Gotland	Westberg SAR08 (LD)	LN810768	.
<i>Acarospora glaucocarpa</i> s. str. WE23	Sweden, Öland	Westberg WE23 (LD)	LN810769	.
<i>Acarospora glaucocarpa</i> f. melaniza SAR01	Austria, Steiermark	Hafellner 64533 (GZU 7-2005)	LN810772	.
<i>Acarospora glaucocarpa</i> f. melaniza SAR10	Sweden, Gotland	Westberg SAR10 (LD)	LN810773	.
<i>Acarospora heufferiana</i> SAR166	Switzerland, Valais	Westberg 10-174 (S F177764)	LN810774	.
<i>Acarospora hospitans</i> SAR189	Norway, Oppland	Westberg 08-234 (S)	LN810775	.
<i>Acarospora hospitans</i> K090847	South Korea, Sokcho-si	KHL0002773 (KH)	OQ629810	OQ641683
<i>Acarospora hospitans</i> 152225	South Korea, Namwon-si	KHL0007969 (KH)	OQ629811	OQ641684
<i>Acarospora impressula</i> SAR33	Norway, Oslo	Westberg 08-107 (S F121708)	LN810776	.
<i>Acarospora insolata</i> WE06	Sweden, Bohuslän	Westberg 06-022 (LD)	LN810777	.
<i>Acarospora laqueata</i> SAR143	Switzerland, Vallis	Westberg 10-170 (S F177761)	LN810778	.
<i>Acarospora macrospora</i> SAR156	Norway, Oslo	Westberg 08-109 (S F121710)	LN810779	.
<i>Acarospora macrospora</i> SAR159	Sweden, Gotland	Westberg SAR159 (LD)	LN810780	.
<i>Acarospora moenium</i> SAR223	Sweden, Torne Lappmark	Westberg P116 (S)	LN810782	.
<i>Acarospora moenium</i> SAR93	Sweden, Västmanland	Westberg 09-066 (S F138363)	LN810781	.
<i>Acarospora molybdina</i> SAR121	Sweden, Bohuslän	Westberg & Westberg SAR121 (LD)	LN810783	.
<i>Acarospora murorum</i> ALM10	Spain, Andalusia	Westberg SCIN014 (S)	LN810784	.
<i>Acarospora nevadensis</i> SAR164	U.S.A., California, San Bernardino Co.	Knudsen 9408 (S F223070)	LN810804	.
<i>Acarospora nicolai</i> SAR228	U.S.A., Kansas, Ellsworth Co.	Morse 16136 & Logan (S)	LN810785	.
<i>Acarospora</i> cf. <i>nitrophila</i> SAR113	Sweden, Lule Lappmark	Westberg 3110 (LD)	LN810787	.
<i>Acarospora</i> cf. <i>nitrophila</i> SAR248	Norway, Sør-Trøndelag	Westberg 12-011 (S, under <i>P. subfuscescens</i>)	LN810786	.
<i>Acarospora nitrophila</i> KL21-0677	South Korea, Sokcho-si	KHL0036391 (KH)	OQ629812	OQ641685
<i>Acarospora nitrophila</i> KL21-0930	South Korea, Sokcho-si	KHL0036643 (KH)	OQ629813	OQ641686
<i>Acarospora nitrophila</i> KL21-1310	South Korea, Sokcho-si	KHL0037019 (KH)	OQ629814	OQ641687
<i>Acarospora nodulosa</i> ALM11	Spain, Andalusia	Westberg SCIN032 (S)	LN810788	.
<i>Acarospora nodulosa</i> SAR146	Spain, Madrid	Westberg 10-215 (S F177732)	LN810789	.
<i>Acarospora obpallens</i> SAR163	U.S.A. California, Orange Co.	Knudsen 9325 (S F256015)	LN810790	.
<i>Acarospora oligospora</i> SAR38	Norway, Oslo	Westberg 08-106 (S F121705)	LN810791	.
<i>Acarospora oligospora</i> SAR89	Sweden, Uppland	Westberg 09-659 & Tibell (S)	LN810792	.
<i>Acarospora peliscypha</i> SAR109	Sweden, Uppland	Westberg 09-222 (S F139588)	LN810793	.
<i>Acarospora peliscypha</i> SAR174	Norway, Sogn og Fjordane	Westberg 08-153 (S)	LN810794	.
<i>Acarospora placodiiformis</i> SAR145	Spain, Madrid	Westberg 10-211 (S F177733)	LN810795	.
<i>Acarospora rosulata</i> SAR31	U.S.A., California, Riverside Co.	Knudsen 9509 (S F256011)	LN810796	.
<i>Acarospora rosulata</i> SAR34	Norway, Oppland	Westberg 08-193 (S)	LN810797	.
<i>Acarospora rugulosa</i> SAR172	Norway, Telemark	Westberg 08-119 (S F123671)	LN810798	.
<i>Acarospora rugulosa</i> SAR173	Sweden, Jämtland	Westberg 10-099 (S F177975)	LN810799	.
<i>Acarospora schleicheri</i> SAR222	U.S.A., Arizona	Sweat & Yansky KGS1196 (UPS L-162697)	LN810801	.
<i>Acarospora sinopica</i> A50	Sweden, Bohuslän	Tibell 22676 (UPS L-113079)	DQ374138	.
<i>Acarospora sinopica</i> MW20	Sweden, Härjedalen	Wedin 6617 (UPS)	DQ374148	.
<i>Acarospora socialis</i> SAR165	U.S.A., California, San Bernardino Co.	Knudsen 9392 (S F256016)	LN810802	.
<i>Acarospora strigata</i> SAR169	U.S.A., California, Riverside Co.	Knudsen 9505 (S F256017)	LN810805	.
<i>Acarospora umbilicata</i> CO238	Sweden, Västergötland	Tibell 23532 (UPS L-136981)	LN810808	.
<i>Acarospora wahlenbergii</i> SAR227	Sweden, Torne Lappmark	Westberg P115 (S)	LN810810	.
<i>Acarospora wahlenbergii</i> SAR91	Sweden, Härjedalen	Westberg SAR91 (LD)	LN810809	.
<i>Glypholecia scabra</i> SAR128	Norway, Oppland	Westberg 08-232 (S)	LN810811	.
<i>Myriospora rhagadiza</i> WE04	Sweden, Bohuslän	Westberg 06-040 (LD)	EU870647	.
<i>Myriospora scabrida</i> WE19	Norway, Troms	Westberg 2824 (LD)	EU870643	.
<i>Pleopsidium chlorophanum</i> MW57	Sweden, Torne Lappmark	Nordin 4439 (UPS L-076485)	EU870691	.
<i>Pleopsidium chlorophanum</i> SAR221	Sweden, Jämtland	Nordin 6209 (UPS L-179248)	LN810813	.
<i>Polysporina cyclocarpa</i> SAR246	Sweden, Torne Lappmark	Westberg P117 (S)	LN810815	.
<i>Polysporina cyclocarpa</i> SAR44	Norway, Sør-Trøndelag	Westberg 08-265 (S F123674)	LN810816	.
<i>Polysporina simplex</i> SAR242	Norway, Oppland	Westberg 08-270 (S F122563)	LN810823	.

(continued)

Table 1. Continued.

Name of species with strain number	Origin	Voucher	nITS	mtSSU
<i>Polysporina simplex</i> SAR273	Austria, Salzburg	Westberg SAR273 (S)	LN810826	.
<i>Polysporina simplex</i> SAR71	Norway, Rogaland	Westberg 08-134 (S F123693)	LN810818	.
<i>Polysporina simplex</i> WE29	Sweden, Bohuslän	Westberg 06-020 (LD 1267752)	LN810827	.
<i>Polysporina simplex</i> SAR236	Norway, Aust-Agder	Westberg 08-123 (S F132541)	LN810822	.
<i>Polysporina simplex</i> WE30	Sweden, Bohuslän	Westberg 06-017 (LD)	LN810828	.
<i>Polysporina</i> sp.1 SAR268	U.S.A., Montana, Chouteau Co.	Wheeler 3583 (S)	LN810829	.
<i>Polysporina subfuscescens</i> SAR132	Norway, Rogaland	Westberg 08-136 (S F123694)	LN810833	.
<i>Polysporina subfuscescens</i> SAR161	Norway, Oppland	Westberg 08-240 (S)	LN810836	.
<i>Polysporina subfuscescens</i> SAR185	Sweden, Pite Lappmark	Westberg 09-638 (S)	LN810837	.
<i>Polysporina subfuscescens</i> SAR244	Norway, Sør-Trøndelag	Westberg 08-260 (S F123679)	LN810840	.
<i>Polysporina subfuscescens</i> SAR252	Norway, Sør-Trøndelag	Westberg 12-018 (S)	LN810844	.
<i>Polysporina subfuscescens</i> SAR259	Norway, Vest-Agder	Westberg 08-130 (S F152847)	LN810845	.
<i>Pycnora sorophora</i> CO15	Sweden, Härjedalen	Hermansson 7903a (UPS L-111613)	FJ959357	.
<i>Sarcogyne albothallina</i> SAR268	U.S.A., Montana, Chouteau Co.	Wheeler 3583 (S)	LN810829	.
<i>Sarcogyne algoviae</i> SAR37	Norway, Oppland	Westberg 08-276 (S F122564)	LN810849	.
<i>Sarcogyne algoviae</i> SAR59	Norway, Oppland	Westberg 08-168 (S F122537)	LN810850	.
<i>Sarcogyne arenosa</i> SAR96	U.S.A., California, Los Angeles Co.	Knudsen 11102 & Sagar (S)	LN810851	.
<i>Sarcogyne clavus</i> SAR02	Austria, Steiermark	Obermayer 09129 (GZU 49-2002)	LN810852	.
<i>Sarcogyne clavus</i> SAR220	Sweden, Värmland	Berglund SAR220 (S)	LN810853	.
<i>Sarcogyne distinguenda</i> SAR42	Sweden, Jämtland	Westberg 08-305 (S F120452)	LN810854	.
<i>Sarcogyne distinguenda</i> SAR54	Norway, Hedmark	Haugen H3852 (O L17425)	LN810855	.
<i>Sarcogyne endopetrophila</i> KL21-1274	South Korea, Yangyang-gun	KHL0036984 (KH)	OQ629815	OQ641688
<i>Sarcogyne hypophaea</i> SAR198	Sweden, Uppland	Westberg SAR198 (S)	LN810856	.
<i>Sarcogyne hypophaea</i> SAR209	Finland, Varsinais-Suomi	Pykälä 23561 (H)	LN810857	.
<i>Sarcogyne hypophaeoides</i> SAR36	Sweden, Västmanland	Westberg 08-002 (S F119718)	LN810858	.
<i>Sarcogyne hypophaeoides</i> SAR49	Norway, Rogaland	Westberg 08-139 (S F123697)	LN810859	.
<i>Sarcogyne jejuensis</i> 121402	South Korea, Jeju-si	KHL0007949 (KH)	OQ629816	OQ641689
<i>Sarcogyne jejuensis</i> 140189	South Korea, Jeju-si	KHL0007977 (KH)	OQ629817	OQ641690
<i>Sarcogyne oceanica</i> KL21-0806	South Korea, Yangyang-gun	KHL0036520 (KH)	OQ629818	OQ641691
<i>Sarcogyne oceanica</i> KL21-0835	South Korea, Seoul-si	KHL0036549 (KH)	OQ629819	OQ641692
<i>Sarcogyne oceanica</i> KL21-1002	South Korea, Gangneung-si	KHL0036713 (KH)	OQ629820	OQ641693
<i>Sarcogyne regularis</i> SAR39	Norway, Oslo	Westberg 08-102 (S F121703)	LN810860	.
<i>Timdalia intricata</i> SAR226	Sweden, Torne Lappmark	Westberg P114 (S)	LN810867	.
<i>Timdalia intricata</i> SAR92	Sweden, Härjedalen	Westberg SAR92 (LD)	LN810866	.
<i>Trimmatothelopsis rhizobola</i> WE15	Sweden, Lule Lappmark	Westberg 2994 (LD)	EU870640	.
<i>Trimmatothelopsis terricola</i> SAR94	U.S.A., California, Los Angeles Co.	Knudsen 11216 & Sagar (S F256012)	LN810806	.

Newly produced sequences are given in bold. In most cases, the nLSU (partial 28S rRNA gene) has the same accession number as the nITS. We added mitochondrial information for future research.

25 µm thick. Apothecia occasional to numerous (including specimen), partly gathered such as middle or margin, 1–2 per areoles, rarely 4, immersed, punctiform at first then expanded, varying in round shape, 0.1–0.5 mm wide, rugulose, reddish-brown to black, darker than the thallus, flat, epruinose. Parathecium distinct, 20–25 µm thick, around the apothecial disc. Hymenium (87.5–)100–125 µm high, hyaline, with oil-drops, IKI + blue (euamyloid), epihymenium 10–12.5 µm high, yellowish brown. Paraphyses, 2–2.5 µm wide at mid-level, sparsely branched, apices not expanded. Asci 100–110 × 15–27.5 µm, clavate, over 100 ascospores per asci, ascospore 3–4 × 1.5 µm, ellipsoid. Subhymenium 62.5–75 µm high, hyaline to pale brown. Hypothecium indistinct. Pycnidia punctiform, brown, darker than thallus, immersed with colorless wall, <0.1 mm wide, conidia broadly ellipsoid, ca. 1.5–1.7 × 1 µm. Photobiont chlorococcoid.

Chemistry: Thallus K–, KC–, C+, PD–; gyrophoric found with TLC.

Ecology and distribution: On rocks; from mountain; type species occur in exposed rock, basalt. Newly found only in type locality.

Remarks: *Acarospora beangnokdamensis* species is characterized by brown angular areoles, C positive reaction having gyrophoric acid, round brown to black

apothecia and distinct punctiform pycnidia. The new species is quite similar with *A. fuscata*. It differs in having dark brown areoles and its angular, not subsquamulose, an undulated thallus surface, having euamyloid (IKI + blue) reaction (vs. hemiamyloid (IKI + pale blue turning red or immediately red), apothecia not common, and punctiform pycnidia observed. *A. gallica* is one of the similar species with the new species. The formal one has also having euamyloid hymenium character and having gyrophoric acid on cortex/medulla, but new species differs from rugulose thallus forming contiguous areolate crust never lobulate squamules and occurs in high elevation than lower location (1600–1800 m vs. 100–300 m). *A. beangnokdamensis* is similar to *A. pseudofuscata*, but the latter can be distinguished by epruinose thallus, lower cortex size (50–100 µm vs. 25–30 µm), distinct pycnidia on each areole. Also, ecologically *A. pseudofuscata* only from the Aegean in Greece and collected altitude is also different (50 m vs. 1907 m). *A. beangnokdamensis* is located in altitude of 1600–1800 m or higher place (South Korea's highest altitude is 1950 m), it appears to be concentrated in the region. Several studies have confirmed that the distribution of lichens varies with altitude [30–32]. In particular, through the study of Schroeter et al., it was revealed that not only chlorophyll but also water variation is a major

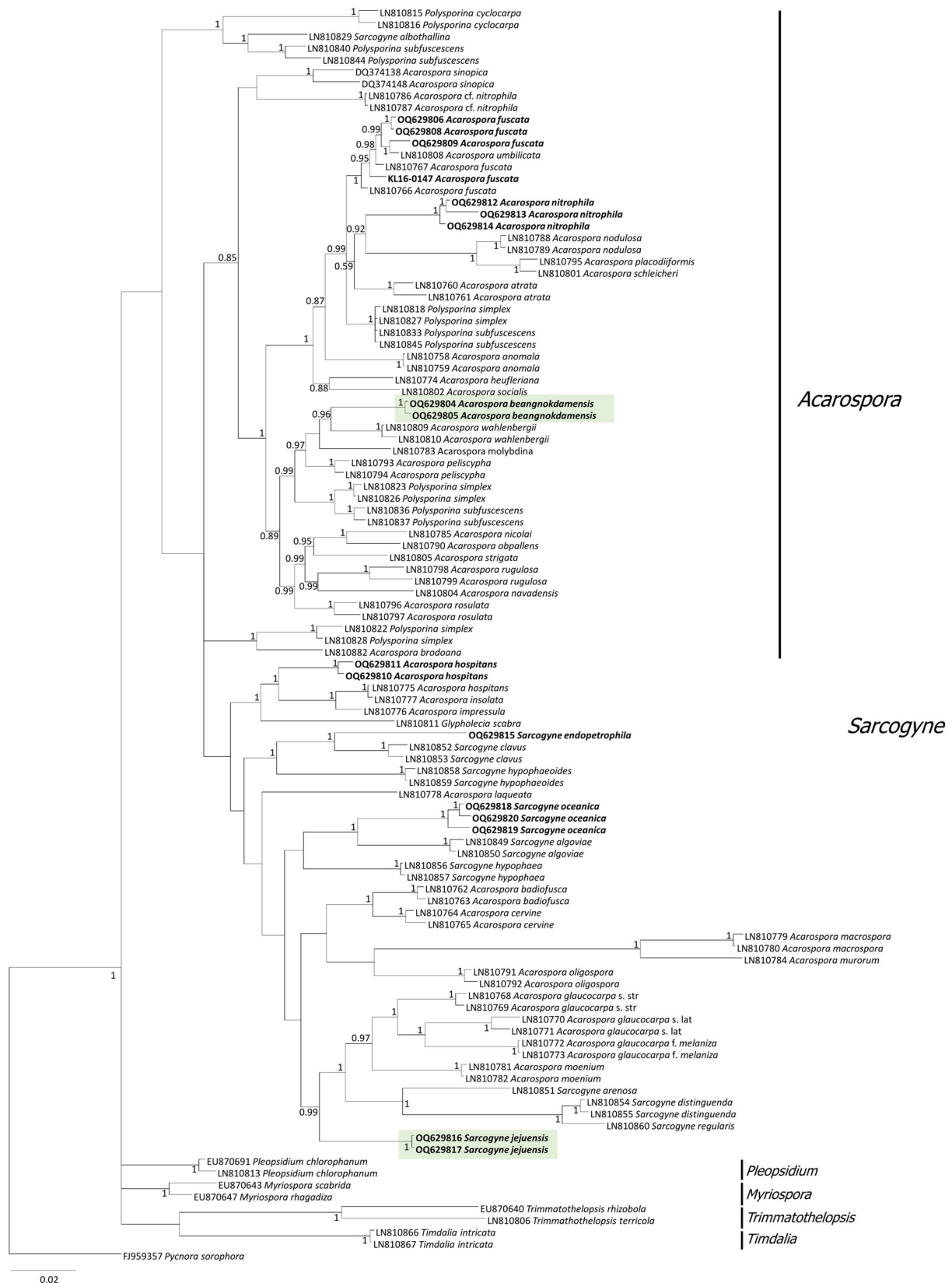


Figure 1. Phylogenetic relationship of *Acarosporaceae*, including *Acarospora* and *Sarcogyne*, inferred from Bayesian analysis of concatenated ITS and nuLSU sequences. Only posterior probability values higher than 80% are shown. The putative new species *Acarospora beangnokdamensis* and *Sarcogyne jejuensis* are highlighted by a green box. Bold letters indicate newly sequenced in this study.

driver in the distribution. Together with these two factors, the substrate specificity of basalt is thought to be the basis for the discovery of new species [31].

Specimens examined: South Korea, Jeju-do, Seogwipo-si, Gwaneumsa Trail, Mt Halla, 33°21'39.7"N, 126°32'8.9"E, 1923 m, on rock, June

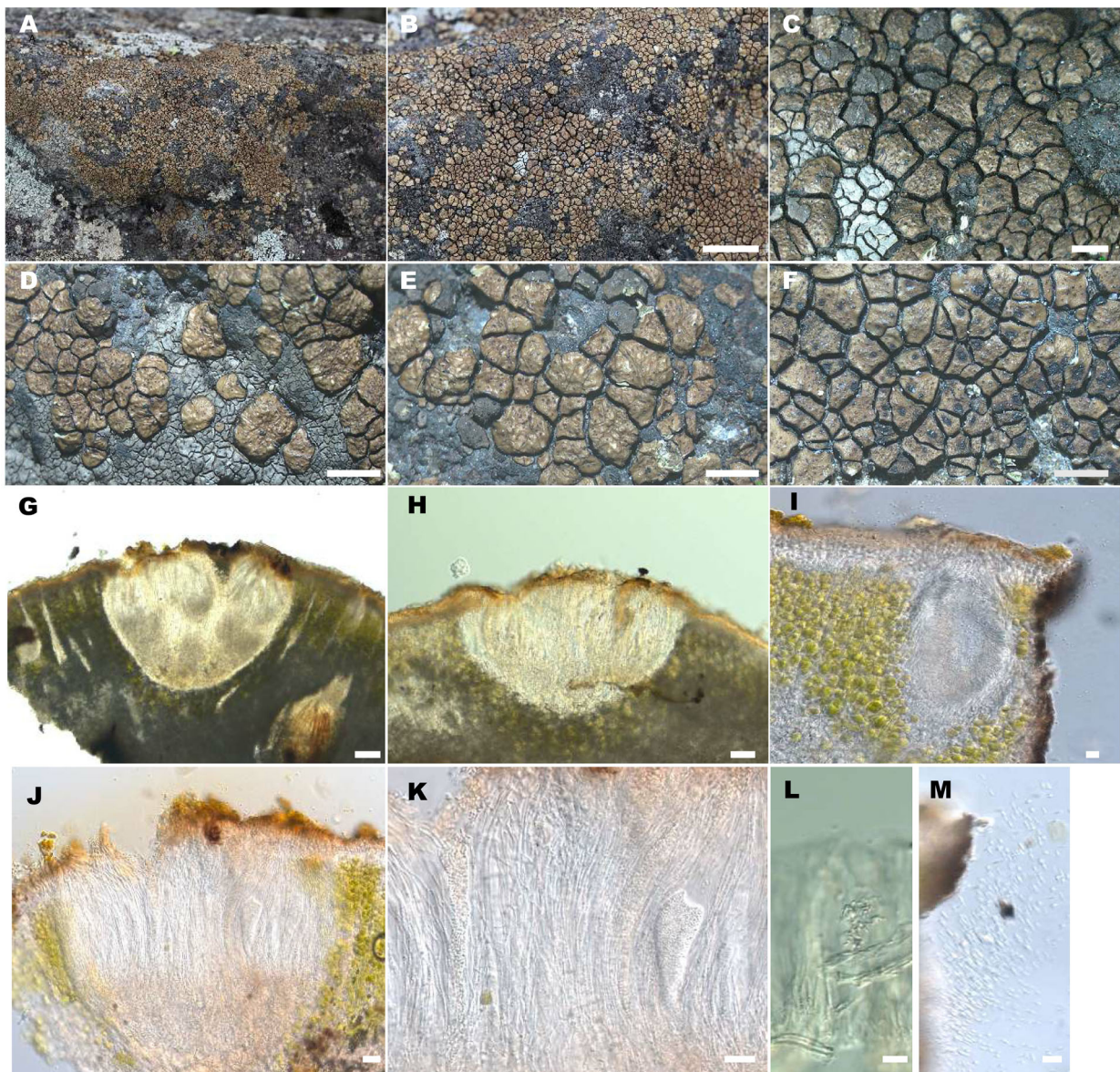


Figure 2. *Acarospora beangnokdamensis* (KL16-0292; KHL0007822); (A) habitat; (B) thallus; (C) enlarged areolate thallus; (D) scattered among other lichen (e.g., *Aspicilia* sp.), lichenicolous; (E) punctiform pycnidia on brown thallus; (F) round black apothecia on thallus, mostly solitary to 1–3 per areoles; (G) section of apothecia, pycnidia coexisting next to apothecia; (H) apothecia, enlarged transverse section of apothecia, no pycnidia around apothecia; (I) transverse section of pycnidia; (J) enlarged transverse section of apothecia; (K) ascus with multispores; (L) ellipsoid to round ascospores; (M) ellipsoid conidia. Scale bars: B = 1 cm; C, D = 2 mm; F = 1 mm; G = 20 μ m; H, I = 10 μ m; J–L = 20 μ m; M = 10 μ m).

19 2012, S.-O. Oh, U. Jayalal, S. Joshi, J. S. Park, F. T. Tian, J.-S. Hur, 121200 (KHL0007942); Jeju-do, Seogwipo-si, Topyeong-dong, Baengnokdam Lake, Mt Halla, 33°21'34.0"N, 126°37'57.0"E, 1874 m, on rock, October 06 2022, S.-O. Oh, J.-J. Seo, B.-J. Kim, Y.-S. Yang, KL22-0363 (KHL0037531).

2. *Sarcogyne jejuensis* J. S. Park & S.-O. Oh, sp. nov. (Figure 3).

Mycobank: Differs from *Acarospora badiofusca* by its narrow medulla size, not continuous areoles and forming apothecial margin pale brown to same color of areoles.

Etymology: It is named after from type locality Jeju-do, South Korea.

Type: South Korea, Jeju-do, Jeju-si, Hangyeong-myeon, Sinchang-ri, Seashore road, 33°20'31.06"N, 126°10'12.08"E, 82 m, on rock, June 18 2014, Y. Joshi, J. E. So, 140195 (KHL0007955).

Description: Thallus of dispersed areoles, areoles rounded to angular, flat to convex, 0.5–0.8 mm wide, 400–600 μ m thick, broadly attached, replicating by division, not delimited. Upper surface usually brown to dark brown, smooth, epruinose, matt, black edged. Lower surface brown. Upper cortex, 20–30 μ m thick, its upper layer yellowish brown 10–12 μ m thick, lower layer hyaline. Algal layer, uninterrupted by hyphal bundles. 62.5–80 μ m thick, even, algal cells 5–7 μ m in diam. Medulla white, prosoplectenchymatous, 30–50 μ m thick, continuous.

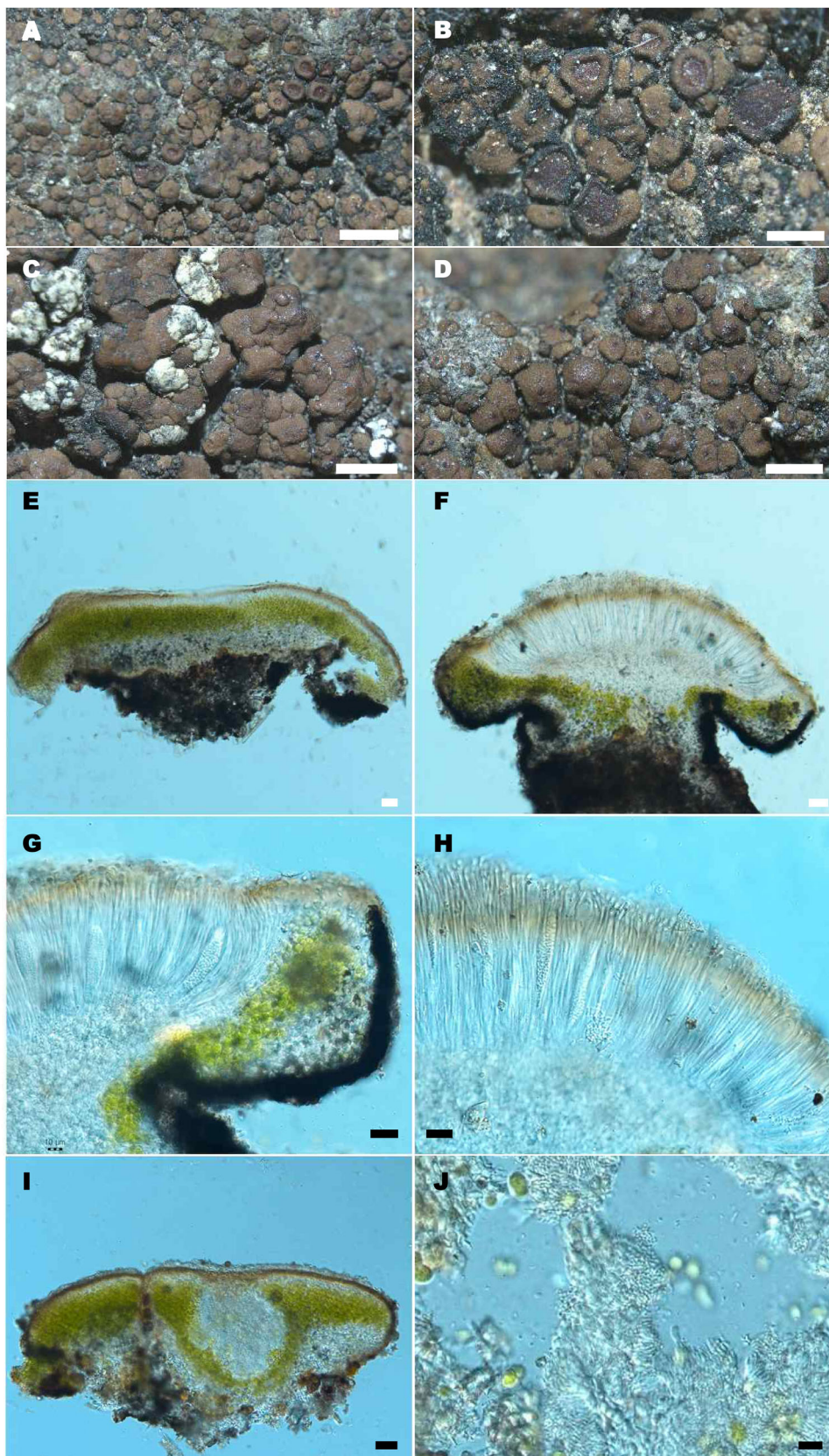


Figure 3. *Sarcogyne jejuensis* (140195; KHL0007955); (A) habitat; (B) enlarged areolate thallus with apothecia; (C) squamulose thallus; (D) punctiform pycnidia; (E) transverse section of thallus, uninterrupted algal layer; (F) transverse section of apothecia; (G) transverse section of apothecia, blackish outer layer; (H) enlarged transverse section of apothecia, ascus with multispores; (I) transverse section of pycnidia; (J) ellipsoid conidia. Scale bars: A, C = 1 mm; B, D = 0.5 mm; E, F = 40 μm; G, H = 20 μm, I = 40 μm, J = 20 μm.

Apothecia usually numerous, scattered, sessile, round to becoming irregular, 0.3–1 mm wide, solitary to one or two apothecia grouped, epruinose.

Disc reddish-brown, smooth, margin formed of the exciple, color brown or same color as disc. Exciple 60–75 μm wide, outer layer 10–12 μm wide, blackish.

Hymenium hyaline, 50–87 μm high, IKI + blue (euamyloid). Epithymenium yellowish brown to reddish brown, 18–20 μm high. Paraphyses 1.8–2 μm wide at mid-level. Asci clavate, 65–80 \times 15–20 μm . Ascospore hyaline, simple, round, over 100–200 ascospores per ascus, round to broadly ellipsoid, 1.5–2 \times 1–2 μm . Pycnidia punctiform, pale brown to brown, flask-shaped, ca 100–112 μm deep by 75–87 μm wide, abundant, conidia, 2 \times 1 μm , globose to slightly ellipsoid.

Chemistry: Thallus K–, KC–, C–, PD–. No acid detected in TLC.

Ecology and distribution: *Sarcogyne jejuensis* was found growing on siliceous rock surface (basalt), together with *Aspicilia*, *Buellia* and *Diploschistes* spp.

Remark: *Sarcogyne jejuensis* is characterized by dispersed areoles with and uninterrupted algal layer and distinct pale to brown apothecial margin. Morphologically, *Acarospora badiofusca* group was similar with new species [33]. *S. jejuensis* differs from *A. badiofusca* in having dispersed areoles, lower size medulla (30–50 μm vs. 100–300 μm), and pale brown apothecial margin. Compared to another *A. badiofusca* group as *A. boulderensis*, *A. irregularis*. Both species separated from *S. jejuensis* by its squamulose thallus (vs. dispersed areoles thallus), interrupted algal layer, and higher hymenium (100–200 μm vs. 50–87 μm). *Sarcogyne saphyniana* is also most similar to new species. Both species has emergent lecideine apothecia with a non-carbonized margin. *S. jejuensis* has a similarly low hymenium but differs its uninterrupted algal layer.

Specimens examined: South Korea, Jeju-do, Jeju-si, Hangeong-myeon, Sinchang-ri, Seashore road, 33°20'31.06"N, 126°10'12.08"E, 82 m, on rock, June 18 2014, Y. Joshi, J. E. So, 140165 (KHL0007975), 140166 (KHL0007970), 140166-1 (KHL0007971), 140189 (KHL0007977); same locality, on rock, July 5 2012, S. Y. Kondratyuk, L. Lökös, 121402 (KHL0007949).

4.2. New record

1. *Sarcogyne oceanica* K. Knudsen & Kocourk., in Knudsen, Kocourková, Cannon, Coppins, Fletcher &

Simkin, Revisions of British and Irish Lichens 12: 23 (2021). (Figure 4).

Description: Thallus endolithic. Apothecia lecideine, occurring on substrate, dispersed, emergent, 1–2 mm in diam., rarely replicating by division, broadly attached with thin algal layer beneath the hypothecium. Disc smooth, plane, blackish red to brownish red, epruinose. Margin black, slightly elevated above the disc, not becoming convex, thin to thick, prominent, not incised, mostly smooth to rugulose. Exciple 70–100 μm thick laterally, outer layer blackish, carbonized 10–12.5 μm wide, internally delimited, pale brown to reddish brown. Epithymenium yellowish-brown, not carbonaceous, 10–13 μm high. Hymenium hyaline, 50–75 μm thick, IKI + blue (euamyloid); paraphyses in yellowish gelatinous gel above, 2–2.5 μm wide at mid-level, apices not expanded. Asci 62.5–67.5 \times 12.5–17.5 μm , ascospores ellipsoid, 4–4.5 \times 1–1.5 μm , mostly 100 per ascus. Subhymenium pale brown, 25–38 μm thick. Hypothecium hyaline, 25–30 μm thick. Pycnidia not seen.

Chemistry: Thallus K–, KC–, C–, PD–. No acid detected in TLC.

Ecology and distribution: On sunny exposed siliceous rock, species mostly occurring on north side of South Korea.

Remark: *Sarcogyne oceanica* has apothecia with smooth to rugulose apothecial margin and hyaline hypothecium. The species could be mistaken for *S. hypophaea* when apothecial margin is not incised and segmented. *Sarcogyne lapponica* looks similar to the *S. oceanica*, but latter one has 1–2 mm apothecia (vs. 0.3–0.5 mm diam.), smooth reddish brown to brown disc (vs. often umbonate). *S. oceanica* can be confused with its calcareous substrates, margin incised of *Sarcogyne algoviae*. The species of *Sarcogyne clavus* is one of the species with very high morphological similarities to the *S. oceanica* due to the similarity of the thalline margin of the apothecia. The two species both have red to black disc with epruinose, but *S. oceanica* features smooth to rugulose margin. Comparatively, *S. clavus* has verrucose and crenulate margins, so the differences



Figure 4. *Sarcogyne oceanica* K. Knudsen & Kocourk. (KL21-1002; KHL0007955); (A) habitat; (B) transverse section of apothecia. Scale bars: B = 50 μm .

between them can be compared. And the color of the hypothecium is different, but the *S. oceanica* is hyaline and *S. clavus* is different in that it has a dark brown hypothecium. *Sarcogyne praetermissa* is one of the similar to *S. oceanica*. Both species have a hymenium with paraphyses 2–3 µm wide at mid-level and red disc with smooth margin, but the formal one occurs in mainly in calcareous substrates.

Specimens examined: South Korea, Gangwon-do, Yangyang-gun, Seo-myeon, Pagoda in Seorimsaji Temple site, 37°59'17.30"N, 128°31'48.43"E, 152 m, on rock, August 11 2021, S.-O. Oh, J. S. Park, J.-J. Seo, S.-G. Son, KL21-0806 (KHL0036520); Seoul Metropolitan Government, Dobong-gu, Tomb of King Yeonsangun, 37°39'41.58"N, 127°1'21.35"E, 53 m, on rock, September 13 2021, S.-O. Oh, J. S. Park, J.-J. Seo, S.-G. Son, KL21-0835 (KHL0036549); Gangwon-do, Gangneung-si, Sacheon-myeon, Pagoda in the Yongyeonsa temple, 37°47'23.63"N, 128°46'49.64"E, 270 m, on rock, August 12 2021, S.-O. Oh, J. S. Park, J.-J. Seo, S.-G. Son, KL21-1002 (KHL0036713); Gangwon-do, Inje-gun, Inje-eup, The Standing stone Buddha statue in Sangdongri, 37°36'5.89"N, 128°0'4.78"E, 194 m, on rock, August 18 2021, S.-O. Oh, J. S. Park, J.-J. Seo, S.-G. Son, KL21-1068 (KHL0036779).

4.3. Reported species

1. *Acarospora fuscata* (Ach.) Arnold.

Description: Thallus areolate, forming either a continuous crust or dispersed, indeterminate of areoles often continuous, occasionally imbricate. Areoles 1–1.5 mm wide, 0.3–0.4 mm thick, sometimes lobulate with edges free, irregular, replicating by division, margin irregular to round. Upper surface light brown to brown, epruinose, matte, smooth. Lower surface light brown. Epicortex 5 µm thick, thin. Cortex 25–37.5 µm thick, upper layer yellow brown, paraplectenchymatous, lower layer hyaline. Algal layer 65–87.5 µm thick, uniform and even and uninterrupted by hyphal bundles, algal cells 7.5–8 µm diam. Medulla white, 62.5–100 µm thick, hyphae thin-walled, septate, 3–4 µm wide. Apothecia solitary to up to 3 per areole, immersed, punctiform, 0.1–0.5 mm wide, dark brown to reddish-brown, disc round to irregular, epruinose. Parathecium expanding up to 25 µm wide, hyaline. Hymenium (65–)70–120 µm high, Epihymenium 12.5–25 µm high, reddish-brown. Paraphyses 1.5–2 µm wide, septate, not branching, apices rarely expanded or capitate. Asci 50–55 × 10–15 µm, clavate, over 100 ascospores per ascus, ascospore 4–4.5 × 1.5–2 µm, ellipsoid. Subhymenium 40–50 µm high, hyaline to pale brown. Hypothecium 15–20 µm high, indistinct.

Chemistry: Thallus K–, KC–, C+, PD–. Gyrophoric acid detected in TLC.

Ecology and distribution: On rocks. According to Knudsen's paper (2022), the world distribution of *A. fuscata* has been reported worldwide such as Europe, Asia, North to North America, Africa, Australia. Korea *A. fuscata* was distributed in sea-shore to mountain and altitude also variable (0–1677 m).

Remark: *Acarospora fuscata* is common species of Europe and it has large variation world wide [34]. According to Knudsen et al., it is shade or full sun that causes the polymorphism of species [2]. The characteristic of *A. fuscata* has pale brown color irregular areoles, immersed punctiform apothecia (0.1–0.3 mm wide) containing gyrophoric acid. According to specimens given to taxonomist, hymenium description was variable. The type collection ((80–)100–120(–140) µm) to Magnusson's description (70–)85–100 µm, a range of 30 µm and depending on local conditions, the British flora cites (70–)80–120 µm, a range of 50 µm [34,35].

Specimens examined: South Korea, Gangwon-do, Yangyang-gun, Hyeonbuk-myeon, 37°56'56.76"N, 128°40'35.12"E, 210 m, on rock, September 2 2021, S.-O. Oh, J. S. Park, J.-J. Seo, S.-G. Son, KL21-1245 (KHL0036956); same locality, on rock, S.-O. Oh, J. S. Park, J.-J. Seo, S.-G. Son, KL21-0849 (KHL0036562); Jeollanam-do, Sinan-gun, Amtaemyeon, 34°50'02.12"N, 126°04'34.71"E, 156 m, on rock, May 12 2016, S.-O. Oh, KL16-0146 (KHL0007676); same locality, on rock, May 12 2016, S.-O. Oh, KL16-0147 (KHL0007677); Jeju-do, Jeju-si, Odeung-dong, Mt Halla, 33°22'12.05"N, 126°31'58.05"E, 1677 m, on rock, June 19 2012, S.-O. Oh, 121121 (KHL0007941).

2. *Acarospora hospitans* H. Magn.

Description: Thallus areolate to subsquamulose, dispersed to continuous, forming small patches, slightly shining. Areoles 0.5–1 mm wide, 0.3–0.4 mm thick, angular to rounded, slightly convex, Upper surface reddish-brown, epruinose. Lower surface dark brown to black. Epicortex 10–12.5 µm thick, distinct. Cortex 25–50 µm high, yellowish-brown to reddish-brown, cortical cells mostly round, 4–5 µm diam., lower layer hyaline, Algal layer 100–125 µm thick, uninterrupted by hyphal bundles, continue under apothecia, algal cells 3–5 µm diam. Medulla 75–90 µm thick, white. Hyphae thin-walled, 1.5–2 µm diam. Apothecia 1–2 per areole, sometimes 3 per areole, immersed the surrounding disc area is slightly raised, same color as thallus, brown, disc to 0.2–0.3 mm wide, mostly round, epruinose. Parathecium hyaline, 10–20 µm diam., poorly developed. Hymenium 75–87.5 µm high, epihymenium

15–25 µm thick, reddish-yellow. Paraphyses 1.5–2 mm wide at midlevel, apices expanded 2.5–3 mm wide, slightly branched. Asci Subhymenium pale brown to hyaline, 30–37.5 µm high. Asci 62.5–87.5 µm high, ascospore 4–5 × 2–2.5 µm, ellipsoid. Hypothecium not distinct, 10–15 µm thick, hyaline. Pycnidia conspicuous or not, punctiform, rising above the thallus like pimple, darker than thallus, conidia hyaline, 0.4–0.5 × 0.8–1 µm.

Chemistry: Thallus K–, KC–, C–, PD–. No acid detected in TLC.

Ecology and distribution: It grows on rock surface and crustose lichen *Pertusaria* sp., *Ropalospora* sp., etc.

Remark: *Acarospora hospitans* is characterized by the presence of brown, disperses to continuous areolate thallus with slightly shining. This species is similar to *A. insolata* in appearance; however, the latter one has thick areoles (0.3–2.4 µm vs. 0.3–0.4 µm) and having numerous apothecia.

Specimens examined: Gangwon-do, Sokcho-si, Mt Sorak, 38°09′58.14″N, 128°27′49.86″E, 673 m, on rock, May 24 2009, Y. Joshi, X. Y. Wang, K090847 (KHL0002773); Jeju-do, Jeju-si, Gwaneumsa Temple trail, Mt Halla, 33°21′39.07″N, 126°32′08.09″E, 1923 m, on rock, June 19 2012, S.-O. Oh, U. Jayalal, S. Joshi, J. S. Park, F. H. Tian and J.-S. Hur, 121201 (KHL0013871).

3. *Acarospora nitrophila* H. Magn.

Description: Thallus areole, areoles forming a continuous crust or dispersed toward the edge, areoles quite variable in size, 0.5–1 mm wide, 0.2–0.25 mm thick, mostly margins rounded and having irregular shape, flat, not convex, large areoles replicating by division. Upper surface pale brown to brown, dull, epruinose, smooth, no black edged. Epicortex indistinct, cortex paraplectenchymatous, 15–20 µm thick, of rounded cells, upper layer yellowish brown, lower layer hyaline with 8–10 µm thick. Algal layer 37–140 µm thick, uninterrupted by hyphal bundle, algal cells 5–7(–10) µm wide. Medulla 62.5–125 µm thick, medullary hyphae intricate, thin-walled, 2–2.5 µm thick, septate. Apothecia usually 1 per areoles, rarely 2, immersed, punctiform, disc rounded, 0.2–0.5 mm wide, brown to reddish brown, without a distinctly visible proper exciple. Parathecium 10–12.5 µm wide, not expanding around the disc. Hymenium hyaline, 100–125 µm high, without oil-drops. Epihymenium yellowish brown, 20–25 µm high, paraphyses 1.5–2 µm wide at mid-level, sparsely branched. Asci clavate, 65–75 × 16.5–17.5 µm, with about 100 ascospores, ascospore hyaline, simple, ellipsoid 3.5–4 × 1.5–2 µm. Subhymenium hyaline, 35–37.5 µm high. Hypothecium 25–50 µm high. Pycnidia not observed. Photobiont chlorococcoid.

Chemistry: Thallus K–, KC–, C–, PD–. Lichen products not detected by TLC.

Ecology and distribution: On rocks; from mountain, species occur in exposed rock, basalt.

Remarks: *Acarospora nitrophila* has been misidentified as several species due to its morphological characteristics [36]. *A. nitrophila* is characterized by the dark brown squamules thallus (0.3–1 mm), in having hemiamyloid hymenium, which has (–90)100–130(–150) µm height. *Acarospora normanii* differs from *A. nitrophila* in having a thallus of areoles, in having an amyloid subhymenium, and hymenium height (80–100 µm vs. (–90)100–130(–150) µm). *Acarospora praeruptorum* and *Acarospora irregularis* were also similar species, both species have large squamulose (1–2 µm vs. 0.5–1 µm). *A. muddii* and *A. inaequalis* have areolae thallus beside squamulose. *A. suzai* is also quite similar to *A. nitrophila*, but differs in its deep blue amyloid hymenium character.

Specimens examined: South Korea, Gyeongsangnam-do, Namhae-gun, seashore, 34°43′30.9″N, 127°53′40.68″E, 16 m, on rock, April 28 2011, X. Y. Wang, 110244 (KHL0013902); Gyeongsangnam-do, Sancheong-gun, Peak Ungseokbong, 35°22′51.15″N, 127°52′32.56″E, 270 m, on rock, June 22 2015, S. Y. Kondratyuk, 150204 (KHL0007940); Gangwon-do, Sokcho-si, Temple Sinheung, 38°10′23.94″N, 128°29′35.45″E, 205 m, on rock, August 10 2021, S.-O. Oh, J. S. Park, J.-J. Seo, S.-G. Son, KL21-0677 (KHL0036391); Gangwon-do, Yangyang-gun, Temple Myeongju, 37°56′56.72″N, 128°40′35.34″E, 221 m, on rock, August 11 2021, S.-O. Oh, J. S. Park, J.-J. Seo S.-G. Son, KL21-0930 (KHL0036643); same locality, on rock, August 11 2021, S.-O. Oh, J. S. Park, J.-J. Seo S.-G. Son, KL21-0950 (KHL0036663); same locality, on rock, August 11 2021, S.-O. Oh, J. S. Park, J.-J. Seo S.-G. Son, KL21-1310 (KHL0037019).

4. *Sarcogyne endopetrophila* Tokizawa & Y. Ohmura.

Description: Thallus crustose, inconspicuous, Apothecia sessile, irregular to round, 0.5–0.9 mm wide. Disc black, epruinose, margin black, mostly prominent, smooth to rugulose, rare. Pycnidia black, sessile, verrucose to globose. 0.5–1 mm wide. Conidia ellipsoid, 2.0–2.5 × 1.0–1.2 µm.

Remark: Almost no apothecia were seen in Korea with *S. endopetrophila*, and most of collected samples were those with only pycnidia. It was recognized as a genus *Sarcogyne* because of morphological appearance (thallus indistinct, lecidine apothecia with black color), but the species could not be identified because the apothecia was not found. However, a type sequence (ITS: NR_154390) was provided and was easy to find. The

species was reported in 2018 and was confirmed to be in Pyeongchang-gun, Gangwon-do. The elevation has been reported to be at 700 m, but our species have been found below 200 m. *S. endopetrophila* has the endolithic thallus, apothecia and pycnidia black as *S. clavus*. However, the size of the pycnidia is large (0.5–1.3 vs. 0.1–0.3) and the color of the hypothecium is pale brown (vs. dark brown). *S. endopetrophila* is similar to *S. hypophaea*, but differs from hymenium is its large size (90–150 µm thick), and larger pycnidia (0.5–1.3 mm in diameter).

Specimens examined: South Korea, Seoul-si, Dobong-gu, Tomb of King Yeonsangun, 37°39'41.0"N, 127°1'21.0"E, 287 m, on rock, September 1 2021, S.-O. Oh & J. S. Park, KL21-0826 (KHL0036540); Gangwon-do, Yangyang-gun, Temple Myeongju, 37°56'56.72"N, 128°40'35.34"E, 221 m, on rock, September 2 2021, S.-O. Oh, J. S. Park, J.-J. Seo S.-G. Son, KL21-1274 (KHL0036984)

4.4. Key to the genera in Acarosporaceae in South Korea

1. Having globose apothecia with a hymenium as high as 250 µm and thin paraphyses usually 1 µm wide *Trimmatothelopsis*
 1. No having globose apothecia with high hymenium (not over 200 µm).....2
 2. Apothecia with a thalline exciple in section or immersed and bordered by thallus, apical dome well develops..... 3
 2. Apothecia lacking of thalline exciple in section, having carbonized outer margin.....5
 3. Blue stain of the tholus in Lugol's iodine after pretreatment in potassium hydroxide (K) (IKI + blue).....*Caeruleum*
 3. No reaction above in tholus (IKI-).....4
 4. Having high hymenium (100–200 µm), distinguished by the chambered medulla with a discontinuous algal layer..... *Myriospora*
 4. Mostly having narrow hymenium (up to 100–110 µm), distinguished by the chambered medulla with a continuous algal layer..... *Acarospora*
 5. Apothecia disc flat, smooth, not warty, paraphyses simple..... *Sarcogyne*
 5. Apothecia disc umbonate, often becoming gyrose; paraphyses richly branched and anastomosing..... *Polysporina*

4.5. Key to *Acarospora* species of South Korea (follow Knudsen 2021)

1. Thallus C + red (gyrophoric acid)..... 2
1. Thallus C-.....3

2. Upper surface dull yellow-brown, flat to slightly convex, pycnidia not obviously observed.....*A. fuscata*
 2. Upper surface dark brown, undulated to angular surface, pycnidia obviously observed..... *A. beangnokdamensis*
 3. Areoles densely or partly pruinose..... *A. versicolor*
 3. Areoles not or sparingly pruinose.....4
 4. Thallus indistinct, if found areoles, areoles 0.2–0.3 (–0.6) mm in diam., apothecia solitary..... *A. ulleungdoensis*
 4. Thallus distinct, areoles over 0.2–0.3 (–0.6) mm in diam., dispersed to continuous, apothecia not solitary..... 5
 5. Thallus areoles thick (0.35–0.9 mm in diam.), apothecia 5–12 per areoles..... *A. insolata*
 5. Thallus areoles thin (0.2–0.4 mm in diam.), Apothecia 1–6 per areoles..... 6
 6. Areole surface uneven-nodular, apothecial disc plane to concave, slightly roughened, variable in shape.....*A. nitrophila*
 6. Areole surface plane; not nodular..... 7
 7. Upper cortex not over 15 µm, thin..... *A. veronensis*
 7. Upper cortex over 15 µm, thick..... *A. hospitans*

4.6. Key to *Sarcogyne* species of South Korea with similar species (refer to the taxonomic key of Acarosporaceae in Revisions of British and Irish Lichens in 2021)

1. On calcareous substrates..... 2
1. On non-calcareous substrates..... 4
2. Margin incised..... *S. algoviae*
 2. Margin not incised..... 3
 3. Hymenium usually (65–)90–100(–120) µm high..... *S. regularis*
 3. Hymenium (50–)70–80(–85) µm high..... *S. praetermissa*
 4. Thallus distinct, dispersed to continuous, apothecia on the thallus..... *S. jejuensis*
 4. Thallus indistinct (endolithic lichen), apothecia on the substrate..... 5
 5. Apothecia 1–3 (–6) mm diam.; true exciple strongly crenulate; maritime..... *S. clavus*
 5. Apothecia 0.3–1 (–1.5) mm diam.; true exciple not crenulate (though sometimes split into segments); maritime or inland.....6
 6. True exciple broad, ca. 300 µm thick, often split into angular segments; hypothecium dark brown..... *S. hypophaea*
 6. True exciple narrow, to 100 µm thick, entire; hypothecium indistinct or distinct. If having distinct hypothecium, pale brown to hyaline..... 7

7. Margin incised.....8
 7. Margin not incised..... 9
 8. Having higher hymenium (100–110 µm) and larger asci (75–80 µm × 16–18 µm)..... *S. ulleungdoensis*
 8. Having narrow hymenium (65–85 µm) and small asci (45–55 µm × 10–12 µm)..... *S. privigna*
 9. Hypothecium broad, 90–150 µm thick.....*S. endopetrophila*
 9. Hypothecium narrow to indistinct, if having hypothecium 20–30 µm thick..... *S. oceanica*

Acknowledgements

We are grateful to Jun-Yeong Kim for assistance with DNA sequencing, Young-Nam Kwag for helpful comments. We also thank to herbarium of National Institute of Biological Resource (NIBR), Korea for loan of specimens.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

This work was supported by “Distribution of Lichen on Street Trees on Selecting Bioindicators of Air Pollution” funded by the Korea National Arboretum (KNA1-1-27, 21-1).

ORCID

Jung Shin Park  <http://orcid.org/0000-0001-5450-7358>
 Young-Nam Kwag  <http://orcid.org/0000-0001-7333-2303>
 Sang-Kuk Han  <http://orcid.org/0000-0003-4549-8896>
 Soon-Ok Oh  <http://orcid.org/0000-0001-5170-0371>

References

- [1] Crewe AT, Purvis OW, Wedin M. Molecular phylogeny of Acarosporaceae (Ascomycota) with focus on the proposed genus *Polysporinopsis*. *Mycol Res*. 2006;110(5):521–526. doi: 10.1016/j.mycres.2006.01.010.
- [2] Knudsen K, Kocourková J, Cannon P, et al. Acarosporales: Acarosporaceae including the genera *Acarospora*, *Caeruleum*, *Myriospora*, *Pleopsidium*, *Sarcogyne* and *Trimmatothelopsis*. *Revisions Br Irish Lichens*. 2021;12:1–25.
- [3] Knudsen K, Adams J, Kocourková J, et al. The monophyletic *Sarcogyne canadensis-wheeleri* clade, a newly recognized group sister to the European *Acarospora glaucocarpa* group. *Bryologist*. 2020; 123(1):11–30. doi: 10.1639/0007-2745-123.1.011.
- [4] Knudsen K, Hollinger J, Götz A, et al. A new name for a common desert lichen. *Bull Calif Lichen Soc*. 2021;28(1):8–11.
- [5] Knudsen K, Kocourková J, Hodková E. Four species from New Mexico and Europe. *Arch Liche*. 2022;32:1–10.
- [6] Kondratyuk S, Lőkös L, Tschabanenko S, et al. New and noteworthy lichen-forming and lichenicolous fungi. *Acta Bot Hung*. 2013;55(3–4):275–349. doi: 10.1556/ABot.55.2013.3-4.9.
- [7] Zhang L, Wang X, Zhao Z, et al. Lichens newly recorded from the South Korean Coast. *Mycotaxon*. 2013;122(1):421–432. doi: 10.5248/122.421.
- [8] Kondratyuk S, Lőkös L, Halda J, et al. New and noteworthy lichen-forming and lichenicolous fungi 5. *Acta Bot Hung*. 2016;58(3–4):319–396. doi: 10.1556/ABot.58.2016.3-4.7.
- [9] Kondratyuk S, Lőkös L, Halda J, et al. New and noteworthy lichen-forming and lichenicolous fungi 6. *Acta Bot Hung*. 2017;59(1–2):137–260. doi: 10.1556/034.59.2017.1-2.7.
- [10] Kondratyuk S, Lőkös L, Halda J, et al. New and noteworthy lichen-forming and lichenicolous fungi 4. *Acta Bot Hung*. 2016;58(1–2):75–136. doi: 10.1556/034.58.2016.1-2.4.
- [11] Kondratyuk S, Lőkös L, Halda J, et al. New and noteworthy lichen-forming and lichenicolous fungi 7. *Acta Bot Hung*. 2018;60(1–2):115–184. doi: 10.1556/034.60.2018.1-2.8.
- [12] Kondratyuk S, Lőkös L, Farkas E, et al. New and noteworthy lichen-forming and lichenicolous fungi 3. *Acta Bot Hung*. 2015;57(3–4):345–382. doi: 10.1556/034.57.2015.3-4.7.
- [13] Kim S. Floral studies on the lichens in Korea. *Bull Kongju Teach Coll*. 1981;17:279–305.
- [14] Joshi Y, Nguyen T, Wang X, et al. Contribution to the lichen mycota of South Korea. *Mycotaxon*. 2011;116(1):61–74. doi: 10.5248/116.61.
- [15] Aptroot A, Moon K. New lichen records from Korea, with the description of the Lichenicolous *Halecania parasitica*. *Herzogia*. 2015;28(1):193–203. doi: 10.13158/hea.28.1.2015.193.
- [16] Aptroot A, Moon K. 114 new reports of microlichens from Korea, including the description of five new species, show that the microlichen flora is predominantly Eurasian. *Herzogia*. 2014;27(2):347–365. doi: 10.13158/hea.27.2.2014.347.
- [17] Knudsen K, Kocourková J. Lichenological Notes 7: on taxa of *Acarospora* and *Sarcogyne*. *Opusc Philolichenum*. 2020;19:158–162.
- [18] Knudsen K. Lichen flora of the greater Sonoran desert region. Tempe (AZ): Arizona State University Lichen Herbarium; 2007.
- [19] Westberg M, Millanes A, Knudsen K, et al. Phylogeny of the Acarosporaceae (Lecanoromycetes, Ascomycota, Fungi) and the evolution of carbonized ascomata. *Fungal Divers*. 2015;73(1):145–158. doi: 10.1007/s13225-015-0325-x.
- [20] Vězda A. Neue oder wenig bekannte Flechten in der Tschechoslowakei. II. *Folia Geobot Phytotax*. 1978;13(4):397–420. doi: 10.1007/BF02851943.
- [21] Seppelt R, Nimis P, Castello M. The genus *Sarcogyne* (Acarosporaceae) in Antarctica. *Lichenologist*. 1998;30(3):249–258. doi: 10.1006/lich.1998.0135.
- [22] McCarthy PM, Kantvilas G. Two new species of *Sarcogyne* (lichenised Ascomycota:

- Acarosporaceae) from Central and Southern Australia. *J Adelaide Bot Gard.* 2013;26:15–21.
- [23] Knudsen K, Tsurukau A, Hodková E, et al. *Acarospora fuscata* and *A. umblicata* (Acarosporaceae, Ascomycota) in Belarus. *Bot Zh.* 2022;107(1):38–46. doi: [10.31857/S0006813622010057](https://doi.org/10.31857/S0006813622010057).
- [24] Vilgalys R, Hester M. Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. *J Bacteriol.* 1990;172(8):4238–4246. doi: [10.1128/jb.172.8.4238-4246.1990](https://doi.org/10.1128/jb.172.8.4238-4246.1990).
- [25] Gardes M, Bruns T. ITS primers with enhanced specificity for basidiomycetes—application to the identification of mycorrhizae and rusts. *Mol Ecol.* 1993;2(2):113–118. doi: [10.1111/j.1365-294x.1993.tb00005.x](https://doi.org/10.1111/j.1365-294x.1993.tb00005.x).
- [26] White T, Bruns T, Lee S, et al. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: *PCR protocols: a guide to methods and applications*. New York (NY): Academic Press, Inc.; 1990. p. 315–322.
- [27] Zoller S, Scheidegger C, Sperisen C. PCR primers for the amplification of mitochondrial small subunit ribosomal DNA of lichen-forming ascomycetes. *Lichenologist.* 1999;31(5):511–516. doi: [10.1006/lich.1999.0220](https://doi.org/10.1006/lich.1999.0220).
- [28] Thompson J, Higgins D, Gibson T. CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res.* 1994;22(22):4673–4680. doi: [10.1093/nar/22.22.4673](https://doi.org/10.1093/nar/22.22.4673).
- [29] Rambaut A. FigTree: tree figure drawing tool. v.1.3.1. Institute of Evolutionary Biology. Edinburgh: University of Edinburgh; 2009.
- [30] Pinokiyo A, Singh K, Singh J. Diversity and distribution of lichens in relation to altitude within a protected biodiversity hot spot, North-east India. *Lichenologist.* 2008;40(1):47–62. doi: [10.1017/S0024282908007214](https://doi.org/10.1017/S0024282908007214).
- [31] Schroeter B, Green T, Pintado A, et al. Summer activity patterns for a moss and lichen in the Maritime Antarctic with respect to altitude. *Polar Biol.* 2021;44(11):2117–2137. doi: [10.1007/s00300-021-02939-9](https://doi.org/10.1007/s00300-021-02939-9).
- [32] Wang Y, Zheng Y, Wang X, et al. Lichen-associated fungal community in *Hypogymnia hypotrypa* (Parmeliaceae, Ascomycota) affected by geographic distribution and altitude. *Front Microbiol.* 2016;7:1231. doi: [10.3389/fmicb.2016.01231](https://doi.org/10.3389/fmicb.2016.01231).
- [33] Knudsen K, Kocourková J, Nordin A. Conspicuous similarity hides diversity in the *Acarospora badiofusca* group (Acarosporaceae). *Bryologist.* 2014;117(4):319–328. doi: [10.1639/0007-2745-117.4.319](https://doi.org/10.1639/0007-2745-117.4.319).
- [34] Magnusson A. A monograph of the genus *Acarospora*. *Almqvist & Wiksells boktryckeri-a.-b.*; 1929.
- [35] Smith C, Aptroot B, Coppins B, et al. *Lichens of Great Britain and Ireland*. 2nd ed. London: BLS; 2009. p. 125–132.
- [36] Knudsen K, Kocourková J. What is *Acarospora nitrophila* (Acarosporaceae)? *Bryologist.* 2017;120(2):125–129. doi: [10.1639/0007-2745-120.2.125](https://doi.org/10.1639/0007-2745-120.2.125).