The First Finding of the Lichen *Solorina saccata* at an Algific Talus Slope in Korea

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**ABSTRACT**

An algific talus slope is composed of broken rocks with vents connected to an ice cave, releasing cool air in summer and relatively warmer air in winter to maintain a more stable microclimate all year round. Such geological features create a very unusual and delicate ecosystem. Although there are around 25 major algific talus slopes in Korea, lichen ecology of these areas had not been investigated to date. In this study, we report the first exploration of lichen diversity and ecology at an algific talus slope, Jangyeol-ri, in Korea. A total of 37 specimens were collected over 2017–2018. Morphological and sequencing analysis revealed 27 species belonging to 18 genera present in the area. Of particular interest among these species was *Solorina saccata*, as it has previously not been reported in Korea and most members of genus *Solorina* are known to inhabit alpine regions of the Northern Hemisphere. We provide here a taxonomic key for *S. saccata* alongside molecular phylogenetic analyses and prediction of potential habitats in South Korea. Furthermore, regions in South Korea potentially suitable for *Solorina* spp. were predicted based on climatic features of known habitats around the globe. Our results showed that the suitable areas are mostly at high altitudes in mountainous areas where the annual temperature range does not exceed 26.6°C. Further survey of other environmental conditions determining the suitability of *Solorina* spp. should lead to a more precise prediction of suitable habitats and trace the origin of *Solorina* spp. in Korea.

1. Introduction

Algific talus slopes are composed of broken rocks with vents connected to an ice cave. The air flow through these vents create a meteorologically unique area, where cold air blows out of the vents or “wind-holes” in summer and relatively warmer air is released in winter [1].

During the last ice age in the Pleistocene epoch of the Cenozoic Era, vegetation in the Northern Hemisphere spread southwards. It was reported that due to the special microclimate created by the temperature-stabilizing effect of algific talus slopes, they sheltered various migrating animals and vegetation during interglacial periods [1–3]. Plants, mite-like insects, or snails typically inhabiting higher latitudes were found at algific slopes [4,5], suggesting that these geological features provide a distinctive environment from the surrounding area.

Algific talus slopes are located at latitudes of 35°–45° north [6–11]. They are found in the Northern Hemisphere in Europe, East Asia, and the USA. In the USA alone, more than 400 locations have been reported in Minnesota, Iowa, Wisconsin, Illinois, and Pennsylvania. In Europe, several algific talus areas are distributed in mountainous regions around the Alps [12]. In central Honshu, Japan, there are more than 80 algific talus areas. Although a number of comprehensive studies on climate, topography, surface geology, and vegetation of algific talus areas have been carried out [13–16], only a few studies exist on lichen ecology and distribution in these areas.

In a survey of lichen at White Pine Hollow State Park, Iowa, USA, 71 of 117 samples were collected from algific talus areas and 4 out of 13 newly discovered species were found in these areas. Species such as *Peltigera pononensis*, *Peltigera membranacea*, and *Physconia muscigena* were typically found at higher altitude than in Iowa, but were found at low altitude at White Pine Hollow [17]. In another study, a survey of lichens was conducted in the Spruce Creek Ice Caves, Huntingdon County, Pennsylvania, USA. Many of the lichens identified in this survey, including *Arctoparmelia centrifuga*,...
Cladonia coccifera, Cladonia rangiferina, Porpidia tuberculosa, Protothelenalla corosa, Rhizocarpon subgeminatum, Stereocaulon glaucescens, Vulpicida pinastri are typically native in northern alpine habitats [7].

In South Korea, 25 major algific talus slopes are distributed from Jeju Island to Gangwon province (Figure 1) [2]. Among these areas, vegetation of Jangyeol-ri algific talus slope, in Jeongseon, Gangwon province, was dominated by plant species Astilboides tabularis and Pedicularis resupinata, which are known to be highly vulnerable to climate change, suggesting that the biota typical of the northern regions are specifically present in the area [2].

The lichen genus Solorina is known to be distributed in bipolar [18], boreal, and arctic-alpine environments [19]. The morphological characteristics of the genus Solorina Ach. are said to include terri- culous lichens with a foliose thallus and ascoma impressed in the upper surface [20]. The genus Solorina belongs to the family Peltigeraceae, as does the genus Peltigera, and is also closely related to the genus Nephroma. The relationship between the two genera is supported by similar ascomatal ontogeny and ascus morphology [21]. Unlike members of the genus Peltigera, members of the genus Solorina have laminal apothecia, pigmented and verrucose spores [21].

In a molecular phylogenetic analysis, the genus Solorina was used as the outgroup to assess the specificity of lichen-forming fungi and genus Nostoc in the genus Peltigera section Polydactylon [22]. The genera of Peltigeraceae including Solorina spp. have been used to identify the phylogenetic location of lichen symbiotic members and to improve the classification and phylogeny of Lecanoromycetes [23,24]. In addition, genus Solorina has been classified as a member of family Solorinaceae, and the relationship between species is paraphyletic. Moreover, the phylogenetic relationship between genus Solorina and Peltigeraceae/Pertigerales was identified as monophyletic [25]. Another study on Peltigeraceae claimed that genus Solorina belongs to the monophyletic family Peltigeraceae, and closely related to the sister genus Peltigera [21].

Unfortunately, genus Solorina has been largely disregarded in molecular phylogenetic studies on Peltigeraceae over the last decade and has only been referred to as a sister genus to Peltigera. Genus Solorina consists of about 10 species distributed throughout the world, with 4 species in Japan and 5 species in China [26,27], but had so far not been found in Korea.

The objectives of this study were (i) to survey lichen species in Jangyeol-ri algific talus slope, (ii) to identify the collected lichen species, and (iii) to identify lichen genus Solorina at morphological and molecular levels. In this study, we catalog the variety of lichen species present in Jangyeol-ri algific talus slope and also report the first finding of Solorina saccata, a northern lichen species, in Korea.

2. Materials and methods

2.1. Morphological examination

All lichen specimens were collected from Jangyeol-ri algific talus slope, which is not a protected area and no permit is required, and were deposited at the Korean National Arboretum (KNA). Air-dried samples were observed using a stereomicroscope (Olympus SZX7; Olympus, Tokyo, Japan) and a compound microscope (Olympus CX22LED; Olympus). Water mounts were hand sectioned with a razor blade and microscopic features (ascomatal structure) were observed in water. Color reactions were conducted as described [28].

2.2. DNA extraction and PCR amplifications

Four representative S. saccata specimens were selected and used for further molecular analyses. Lichen thalli with apothecial discs were mainly used for DNA extraction. Samples were ground and extracted with DNeasy plant mini kit (Qiagen,
Valencia, CA, USA) according to the manufacturer's instructions. PCR amplifications were conducted using AmpliTaq DNA polymerase (ThermoFisher, Watham, MA, USA). The following primers were used for PCR amplifications: mtSSU1 and mtSSU3R for mtSSU [29]; NS17UCB, NS20UCB for nuSSU [30]; LIC24R [31] and LR7 [32] for nuLSU; fRPB2-7cf and fRPB2-11aR for RPB2 [33]. PCR conditions for nuSSU are as described in a previous study [34]. The following program was used for amplification of nuLSU: initial denaturation for 4 min at 94°C, followed by 30 cycles of 94°C for 40 s, 52°C for 40 s, 72°C for 50 s, and then a final extension step at 72°C for 8 min. Amplified DNA was concentrated and purified using a PCR quick-spin PCR Product Purification Kit (INTRON Biotechnology, Inc. Sungnam City, Korea) for sequencing analysis.

2.3. Sequence alignments and phylogenetic analysis

Obtained sequences were aligned with Clustal W ver. 1.83 [35] and edited using the Bioedit program. Based on the sequences, we selected commonly present sequence regions of homology, and excluded uncommonly detected sequences. We utilized Gblocks 0.91 b server [36] for deleting ambiguously aligned regions in the concatenated alignment of nuSSU, nuLSU, and RPB1. We prepared a three-locus concatenated dataset containing nuSSU, mtSSU + nuLSU, and nuLSU + RPB1.

Phylogenetic analyses were conducted with MEGA 7.0 and MrBayes v. 3.2.6. Bayesian analysis was carried out on the data set using the Metropolis-coupled Markov chain Monte Carlo (MCMCMC) method in MrBayes v. 3.2.6 [37,38]. Best fit substitution models were estimated using the Akaike-information as implemented in jModelTest v 2.15. [39]. The TVM + I model in nuSSU, TIM3 + 1 + G model in mtSSU + nuLSU, and JC + G model in nuLSU + RPB1 were selected. Each MCMCMC run was performed with four chains and 10 million generations. Trees were generated 1000 times and the first 25% was discarded. The remaining trees were determined by calculating a majority-rule consensus tree with posterior probabilities (PP).

In addition, we also performed Maximum likelihood (ML) analysis using MEGA 7.0 with 1000 ML bootstrap values (BP) and GTR + G model was applied. We selected outgroup for phylogenetic analysis according to previous research [21,40,41]. Phylogenetic trees were drawn using Figtree v. 1.4.3 [42] with Treeview X v. 0.5.0 and PP above 0.80 were used near the bold branches.

2.4. Estimation of optimum temperature for Solorina spp.

Although Solorina species are native to polar and alpine regions, our specimens were collected at a relatively low altitude of 400–450 m above sea level. To determine climatic requirements for Solorina spp., we first identified all collection sites of Solorina spp. from previously published literature and relevant reports, and then collected 30 years' climate data on the minimum and maximum air temperature for the identified habitats.

A few criteria were applied to filter out unusable data. As the climate in the Southern Hemisphere has different seasonal variations, data from the Southern Hemisphere were excluded. Sites with uncertain sampling location or unspecific records above district level were also excluded. Multiple sites located within 1 km radius were combined and considered as a single site, as we used a 1 km resolution grid dataset to extract corresponding climate data.

As a result, the number of sites was narrowed down from 73 to 63 sites for subsequent climatological analysis. Coordinates (latitude and longitude) of the chosen sites were first estimated based on available site information and using the Google Maps (https://www.google.com/maps). Then monthly minimum and maximum temperature information at the identified coordinates were extracted from the WorldClim, a monthly climatology dataset provided at 1 km resolution (https://www.worldclim.org/). Average monthly minimum and maximum temperatures were calculated and plotted together with all the temperature ranges of the 63 sites (Figure 2).

Site suitability for Solorina spp. in South Korea was determined based on the temperature data of 63 sites. The temperature range for each month was defined by the minimum and maximum values among the 63 sites. The temperature range values were used to identify suitable areas with similar monthly temperature variations in South Korea. If the local temperature of an area satisfies the above defined temperature range profile, it was deemed “Suitable”. Other areas were considered “Unsuitable”. Local temperature profiles to 1 km resolution of South Korea were obtained from a historical climate dataset produced by the Korean Meteorological Administration (KMA). High resolution KMA temperature datasets were produced from 75 KMA Automated Synoptic Observing System (ASOS) stations and 462 KMA Automated Weather Stations (AWS) using the PRIsm-based Downscaling Estimation (PRIDE) model [43], and included daily weather data from 2001 to 2010.
3. Results and discussion

3.1. Survey of lichen species in Jangyeol-ri algific talus slope

Jangyeol-ri algific talus slope is located at 200–450 m above sea level in Jeongseon, Gangwon province (37°27′06.14″N, 128°41′04.18″E). It is 200 m in breadth and sloped at 40 degrees (Figure 3(A–E)). The geological features were consistent with the Paleozoic Ordovician strata, most of which were limestone and dark red forest soils [2].

A total of 37 lichen specimens were collected from 2017 to 2018. From these samples, 27 species belonging to 18 genera were identified by morphological examination and sequencing analysis: genus Amandinea (A. punctata); Caloplaca (C. flavovirescens); Candelaria (C. concolor); Cladonia (C. furcata subsp. furcata and C. pyxidata); Collema (C. japonicum and C. leptaleum var. biliosum); Everniastrum sp.; Flavoparmelia (F. caperata); Graphis (G. scripta); Heterodermia (H. diademata and H. hypoleuca); Lecanora (L. argentata and L. strobilina); Leptogium (L. cyanescens); Lecanora (L. argentata and L. strobilina); Myelochroa (M. aurulenta); Ochrolechia (O. akagiensis); Peltigera (P. elisabethae, P. horizontalis, and P. rufescens); Phaeophyscia (P. primaria); Protoblastenia (P. rupestris); Ramalina sp.; Solorina (S. saccata) (Table 1).

In this survey, we could not find any lichen species overlapping with those found in previous studies on lichen inhabiting algific talus slopes in White Pine Hollow, Iowa, USA [17] and Spruce Creek Ice Cave, Pennsylvania, USA [7]. Interestingly, the most predominant lichen species we found in Jangyeol-ri algific talus belonged to genus Solorina (10.8% of all specimens collected), which are mainly observed in circumpolar, arctic alpine and boreal areas including Europe, Asia, and North America [44]. Although there is no overlap between lichen species found in Jangyeol-ri and Spruce Creek Ice Cave, the species collected at these two different algific talus slopes were species common in northern alpine areas, indicating that the vegetation at algific talus slopes are very different from the surrounding areas at an equivalent latitude.

3.2. Phylogenetic analysis of S. saccata using nuSSU, nuLSU, and RPB1

We generated nuSSU, mtSSU, nuLSU, ITS, RPB1, and RPB2 sequences from the four S. saccata

Figure 2. Analysis of minimum and maximum air temperatures at Solorina spp. habitats worldwide. (A) 63 sites where Solorina spp. has been reported, indicated by red dots; Monthly minimum (B); and maximum (C) temperatures of the 63 sites were extracted from the WorldClim dataset (https://www.worldclim.org/). Average (mean) monthly minimum (B) and maximum (C) temperatures are indicated as solid black lines in each graph and temperature range at the 63 sites are indicated in gray.
Additionally, we also generated sequences from three *S. saccata* specimens previously collected in China for comparison. We successfully obtained sequences from all above specimens, except one Korean *S. saccata* specimen, Oh KL17-0241. The five ribosomal genes and ITS region sequences obtained from our specimens were over 99% identical to the *S. saccata* sequences in GenBank.

Figure 3. Photographs of Jangyeol-ri algific talus slope. (A) The collection area is marked by the dashed-line circle and the exposed area of the talus slope is marked by dashed-line rectangle; (B) The collection site of lichens in Jangyeol-ri algific talus slope; (C) Two vents; (D) Lichen species *Solorina saccata* in front of the vent; (E) Exposed area. The smaller insert shows more detail at the ground level.
Table 1. Summary of lichen species in Jangyeol-ri algific talus slope.

| Species                | Family               | No. of specimens |
|------------------------|----------------------|-----------------|
| Amandinea punctata     | Caliciaceae          | 1               |
| Caloplaca flaviscensc  | Teloschistaceae      | 1               |
| Candelaria concolor    | Candelariaceae       | 1               |
| Cladonia furcata       | Cladoniaceae         | 3               |
| Cladonia pyxidata      | Cladoniaceae         | 3               |
| Collema japonicium     | Collemataceae        | 1               |
| Collema leptaleum var. | Collemataceae        | 1               |
| Everniastrum sp.       | Parmeliaceae         | 1               |
| Flavoparmelia caperata | Parmeliaceae         | 1               |
| Graphis scripta        | Graphidiaceae        | 1               |
| Heterodermia diademata | Physciaceae          | 1               |
| Heterodermia hypoleuca | Physciaceae          | 2               |
| Lecanora argentata     | Lecanoraceae         | 1               |
| Lecanora strobilina    | Lecanoraceae         | 3               |
| Leptogium cyanescens   | Collemataceae        | 1               |
| Myelochroa aurulenta   | Parmeliaceae         | 1               |
| Ochrolechia akagiensis | Ochrolechiaceae      | 1               |
| Peltigera elisabethae  | Peltigerae           | 3               |
| Peltigera horizontalis | Peltigerae           | 1               |
| Peltigera rubescens    | Peltigerae           | 2               |
| Phaeophyscia primaria  | Physciaceae          | 1               |
| Protoblastenia rupestris| Pseudepheciae        | 1               |
| Ramalina sp.           | Ramalinae            | 1               |
| Solorina saccata       | Peltigerae           | 4               |
| Total                  |                      | 37              |

3.3. Comparison of average temperatures at Jangyeol-ri algific talus slope and the surrounding area, Jeongseon-gun

To investigate whether Jangyeol-ri algific talus indeed has a distinct temperature profile from the surrounding Jeongseon-gun region, we examined the monthly temperature variation (Figure 7). Because the temperature measurements in Jangyeol-ri algific talus slope were only available from April to November 2017, the data were compared with those of Jeongseon-gun in 2017 (Figure 7).

The maximum difference in average temperature was observed in August 2017, when Jangyeol-ri was 15.7°C, which was 7.3°C lower than the average temperature of 23.0°C in Jeongseon-gun. In comparison, in November 2017, the average temperature of Jangyeol-ri algific talus slope was 2.93°C, which was 0.87°C lower than the average temperature (3.8°C) in Jeongseon-gun. These results indicate that the greatest differences in temperature between Jangyeol-ri algific talus slope and the rest of Jeongsun-gun is in the summer months. It is presumed that the cooler temperatures in summer may contribute to the continued existence of Solorina spp. in Jangyeol-ri.

3.4. Prediction of suitable areas for Solorina spp. in South Korea

Suitable areas for Solorina spp. in South Korea were predicted using the monthly temperature profiles of 63 sites across the globe where the Solorina spp. were found. Our assumption that monthly temperature variations are a major factor determining habitat suitability was in line with previous findings where bioclimatic or climate-response models developed point to the importance of temperature range and the temperature of the warmest month [45,46]. Our results showed that the highest monthly maximum recorded was 26.6°C in August and the lowest monthly minimum temperature was −43.9°C in January (Figure 2). The minimum temperature of −43.9°C indicates that Solorina spp. may be able to endure the extreme cold temperatures in a state of dormancy like other lichens in polar regions.

On the contrary, warmer temperatures may not be suitable to sustain Solorina spp., as previously reported areas are confined in bipolar, boreal, and arctic-alpine environments [20,27,44–50]. With an assumption that the Solorina spp. in Korea would share the same temperature requirements as those found at other sites around the world, we used the annual temperature ranges of other reported sites to predict potential areas in South Korea that Solorina spp. may exist.
Our result showed that the suitable areas are mostly at high altitudes in mountainous areas, where annual temperature does not exceed 26.6°C. The fact that Solorina spp. in Korea were found in the algific talus area, where the temperature is lower than surrounding areas in the summer months, supports our prediction results. It is highly probable that the Solorina spp. found in the algific talus area may not result from a migration from mountainous areas all the way down to the lower altitudes (Figure 8). Rather, we speculate that Solorina spp. have been inhabiting the algific talus area since thousands of years ago when the algific talus area was at a high altitude before going through diastrophism.

Other algific talus areas in Korea also show similar temperature variations to the predicted areas suitable for Solorina spp. Based on our preliminary analysis using temperature records of those sites over a few years, the highest temperatures recorded are generally 1–2°C less than 26.6°C, which is the highest monthly maximum temperature of the global sites of Solorina spp. Although it is possible that

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**Figure 4.** Phylogenetic relationship within family Peltigerales inferred from Bayesian analysis using nuclear small subunit (nuSSU) sequences. Only posterior probability values higher than 80% are shown. Strongly supported nodes are indicated in bold.
Solorina spp. indeed exist in the mountainous areas as predicted in our study, temperature alone may not be sufficient as an indicator of suitability for the habitat of Solorina spp.; other environmental factors such as relative humidity and rainfall, or substrate preferences may also be necessary determinants [51]. In addition, cross-scale interaction between microhabitat specificity and macroclimatic condition should also play an important role in formulating suitable microclimatic niche for Solorina spp. [52]. However, it is very difficult to find out these complex interactions among multiple environmental factors with limited data available from algific talus areas in Korea. Further survey on other multi-scale environmental factors and their effect on Solorina spp. should result in more reliable prediction of their habitats in Korea and other global areas.

**Figure 5.** Phylogenetic relationship within family Peltigerales inferred from Bayesian analysis of concatenated mitochondrial small subunit (mtSSU) and nuclear large subunit (nuLSU) sequences. Only posterior probability values higher than 80% are shown. Strongly supported nodes are indicated in bold.
Figure 6. Phylogenetic relationship of Peltigerales inferred from Bayesian analysis of concatenated nuLSU and RPB1 sequences. Only posterior probability values higher than 80% are shown. Strongly supported nodes are indicated in bold.

Figure 7. Comparison of average temperatures at the Jangyeol-ri algific talus slope and the surrounding area, Jeongsun, in 2017. X-axis: Temperature/°C, y-axis: Month.
3.5. Key to identify known species of Solorina

1. Photobiont in thallus mainly blue-green (cyanobacterial), apothecia mainly plane, 4-spored in ascus ........................................ S. simensis
2. Lower side of thallus vivid orange ...................................... S. crocea
3. 1-2 spores in each ascus ................................................... 4
3. 4-8 spores in each ascus ................................................... 8
4. 1 spore in each ascus ....................................................... 5
4. 2 spores in each ascus ....................................................... 6
5. Spores 2-septate, spore size 90-128(-161) × 30-44 μm .............................................. S. monospora
5. Spores 2-3 septate, spore length 85-120 μm .............................................. S. embolina
6. Cephalodia immersed in thallus ................................ S. bispora var. subspongiosa
6. Cephalodia not immersed in thallus ................................ 7
7. Spores >90 μm long .................................................. S. bispora var. macrosora
7. Spores <90 μm long ............................................. S. bispora var. bispora
8. 4 spores in each ascus ...................................................... 9
8. 6-8 spores in each ascus ................................................ 10
9. Apothecia urceolate, distinctly sunken in depression in the upper surface ......................... S. saccata
9. Apothecia thinly depressed when immature but becoming level when mature ................ S. platycarpa
10. 6 spores in each ascus, external cephalodia forming a dark spongy mat between and under the apothecia .................................................. S. spongiosa
10. 8 spores in each ascus .................................................. S. octospora

3.6. Characterization for the Korean S. saccata

Solorina saccata (L.) Ach., K. Vetensk Acad. Nya Handl. 29: 228 (1808) (Figure 9).

Thallus well developed, spreading, forming round lobes with waxy margins, smooth, forms rosettes, greenish gray when dry but green when wet, coarse white pruinose at least close to the margin, thin spread; lower surface pale brown, rhizine sparsely present, scattered, 2–2.5 mm long, pale brown, indistinctly veined. Apothecia frequent, reddish brown, to 6.5 mm diam., deeply sunken in depression in the upper surface, with or without margin, margin 0.5 mm thick, concolor with thallus. Asci numerous, 4-spored, epihymenium brown to dark brown, 25–32.5 μm, hymenium hyaline, 155–187.5 μm; ascospores 45–53 μm × 17.5 μm, pale brown to brown,
1-septate, ellipsoid-oblong. No lichen substances detected by TLC.

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References

[1] Kong W-S, Lee S, Yoon K, et al. Environmental characteristics of wind-hole and phytogeographical values. J Environ Impact Assess. 2011;20(3):381–395.
[2] Oh SH, Lee Y-M, Kong W-S. Air hole in Korea. Pochon (Korea): Geoobook, KNA; 2013.
[3] Morard S, Delaloye R, Lambiel C. Pluriannual thermal behavior of low elevation cold talus slopes in western Switzerland. Geogr Helv. 2010;65(2):124–134.
[4] Zacharda M, Gude M, Kraus S, et al. The relict Rhagidia gelida (Acari, Rhagidiidae) as a biological cryoindicator of periglacial microclimate in European highland scree. Arct Antarct Alp Res. 2005;37(3):402–408.
[5] Henry C. Refuge for an ice age survivor. Endanger Species Bull. 2003;28:24–26.
[6] Iokawa Y, Ishizawa S. Vascular plants of wind-hole areas in Japan. J Phytogeogr Taxon. 2003;51:13–26.
[7] Lendemer JC, Edenborn HM, Harris RC. Contributions to the lichen flora of Pennsylvania: notes on thelichens of a remarkable talus slope in Huntingdon county. Opusc Philolichenum. 2009;6:125–136.
[8] Nepola JS. Paleorefugia and neorefugia: the influence of colonization history on community pattern and precess. Ecology. 1999;80(8):2459–2473.
[9] Saar R. Eishohlen ein meteorologisch-geophysikalisches Phanomen. Geogr Ann A. 1956;38(1):1–63.
[10] Swarzlow CR. Ice caves in northern California. J Geol. 1935;43:440–442.
[11] Vincent WB. Environmental influence of the glaciers of the Pryor Mountains, Montana. J Caves Karst Stud. 1974;36:13–21.
[12] Kim J-S, Chung J-M, Kim J-H, et al. Floristic study and conservation management strategies of algific talus slopes on the Korean peninsula. Korean J Pl Taxon. 2016;46(2):213–246.
[13] Maki T. Characteristics of topograph, climate and vegetation around Jagaramogara wind cave basin. J Agric Meteorol. 1998;54(3):255–266.
[14] Sasaki H. Air and soil temperature affecting the distribution of plants on a wind-hole site. Ecol Rev. 1986;21:21–27.
[15] Sato K, Kudo G, Uemura S. Cool-spots site vegetation in IZARIIRI-HEIDE, northern Japan. Jpn J Ecol. 1993;43:91–98.
[16] Tanaka HL, Yokoi M, Nohara D. Observation study of summertime ice at the Nakayama wind-hole in Shimogo, Fukushima (Japan): UT; 2000.
[17] Thompson KM. Lichen diversity and conservation of northeast Iowa: White Pine Hollow State preserve and the lichen Lobaria pulmonaria. Ames, Iowa: ISU; 2018.
[18] Smith RI, Øvstedal DO. Solorina spongiosa in Antarctica: an extremely disjunct bipolar lichen. Lichenologist. 1994;26(2):209–213.
[19] Smith CW, Aptroot A, Coppins BJ, et al. The lichens of Great Britain and Ireland. 2nd ed. London (UK): BLS, NHM; 2009. p. 844–846.
[20] Martinez I, Burgaz AR. Revision of the genus Solorina (Lichenes) in Europe based on spore size variation. Ann Bot Fennici. 1998;35:137–142.
[21] Wiklund E, Wedin M. The phylogenetic relationships of the cyanobacterial lichens in the Lecanorales suborder Peltigerineae. Cladistics. 2003;19(5):419–431.
[22] Magain N, Miadlikowska J, Goffinet B, et al. Macroevolution of specificity in cyanolichens of the genus Peltigera section Polydactylon (Lecanoromycetes, Ascomycota). Syst Biol. 2016;66(1):74–99.
[23] Miadlikowska J, Kauff F, Hofstetter V, et al. New insights into classification and evolution of the Lecanoromycetes (Pezizomycotina, Ascomycota) from phylogenetic analyses of three ribosomal RNA and two protein-coding genes. Mycologia. 2006;98(6):1088–1103.
[24] Schmull M, Miadlikowska J, Pelzer M, et al. Phylogenetic affiliations of members of the heterogeneous lichen-forming fungi of the genus Leccidea sensu Zahlbruckner (Lecanoromycetes, Ascomycota). Mycologia. 2011;103(5):1036–1003.
[25] Miadlikowska J, Lutzoni F. Phylogenetic classification of peltigeralean fungi (Peltigerales, Ascomycota) based on ribosomal RNA small and large subunits. Am J Bot. 2004;91(3):449–464.
[26] Ohmura Y, Kashiwadani H. Checklist of lichens of the genus Lobaria. Tokyo (Japan): Hoikusha Publisher; 1974.
[27] Yashimura I. Lichen flora of Japan in color. Osaka (Japan): Hoikusha Publisher; 1974.
[28] Zoller S, Scheidegger C, Sperisen S. PCR primers for the amplification of mitochondrial small subunit ribosomal DNA of lichen-forming ascomycetes. Lichenologist. 2009;31(5):511–516.
[29] Gargas A, Taylor JW. Polymerase chain reaction (PCR) primers for amplifying and sequencing nuclear 18S rDNA from lichenized fungi. Mycologia. 1992;84(4):589–592.
[30] Miadlikowska J, Lutzoni F. Phylogenetic revision of the genus Peltigera (lichen-forming
Ascomycota) based on morphological, chemical, and large subunit nuclear ribosomal DNA data. Int J Plant Sci. 2000;161(6):925–958.

[32] White TJ, Bruns T, Lee S, et al. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ, editors. PCR protocols: a guide to methods and applications. New York: Academic Press; 1990. p. 315–322.

[33] Liu YJ, Whelen S, Hall BD. Phylogenetic relationships among ascomycetes: evidence from an RNA polymerase II subunit. Mol Biol Evol. 1999;16(12):1799–1808.

[34] Miadlikowska J, Kauff F, Högnabba F, et al. A multigene phylogenetic synthesis for the class Lecanoromycetes (Ascomycota): 1307 fungi representing 1139 infragenetic taxa, 317 genera and 66 families. Mol Phylogenet Evol. 2014;79:132–168.

[35] Thompson JD, Higgins DG, Gibson TJ. 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.

[36] Castresana J. Selection of conserved blocks from multiple alignments for their use in phylogenetic analysis. Mol Biol Evol. 2000;17(4):540–552.

[37] Huelsenbeck JP, Ronquist F. MRBAYES: Bayesian inference of phylogenetic trees. Bioinformatics. 2001;17(8):754–755.

[38] Ronquist F, Huelsenbeck JP. MrBayes 3: Bayesian phylogenetic inference under mixed models. Bioinformatics. 2003;19(12):1572–1574.

[39] Zarrit R, Boumaza MS, Kerrou S, et al. L’Effet du Rapport de Forme sur la Convection Naturelle dans une Cavité Rectangulaire Inclinée Remplie d’Air. The 3rd International Seminar on New and Renewable Energies; Ghardaïa – Algérie; 2014. p. 1–7.

[40] Wedin M, Tehler A, Gargas A. Phylogenetic relationships of Sphaerophoraceae (Ascomycetes) inferred from SSU rDNA sequences. Pl Syst Evol. 1998;209(1-2):75–83.

[41] Wedin M, Wiklund E, Jørgensen PM, et al. Slippery when wet: phylogeny and character evolution in the gelatinous cyanobacterial lichens (Peltigerales, Ascomycetes). Mol Phylogenet Evol. 2009;53(3):862–871.

[42] Rambaut A. FigTree: tree figure drawing tool, v.1.4.0. Institute of Evolutionary Biology, University of Edinburgh; 2012. Available from: http://tree.bio.ed.ac.uk/software/figtree/

[43] Kim MK, Han MS, Jang DH, et al. Production technique of observation grid data of 1km resolution. J Clim Res. 2012;7:55–68.

[44] Sinigla M, Lőkös L, Molnár K, et al. Distribution of the legally protected lichen species Solorina sacca-ta in Hungary. Studia Bot Hung. 2018;49(1):47–70.

[45] Ellis CJ, Coppins BJ, Dawson TP, et al. Response of British lichens to climate change scenarios: trends and uncertainties in the projected impact for contrasting biogeographic groups. Biol Conserv. 2007;140(3-4):217–235.

[46] Ellis CJ, Geddes H, McCheyne N, et al. Lichen epiphyte response to non-analogue monthly climates: a critique of bioclimatic models. Perspect Plant Ecol. 2017;25:45–58.

[47] Thomson NF, Thomson JW. Spore ornamentation in the lichen genus Solorina. Bryologist. 1984;87(2):151–153.

[48] Krog H, Swinscow T. Solorina simensis and S. sacca-ta. Lichenologist. 1986;18(1):57–62.

[49] Jahns HM, Klöckner P, Ott S. Development of thalli and ascocarps in Solorina spongiosa (Sm.) Anzi and Solorina sacca-ta (L.) Ach. Bibl Lichenol. 1995;57:241–251.

[50] Gärtner G, Dablander A, Kofer W. Zur Taxonomie von Solorina bispora NYL. ssp. bispora (Ascolichenes) nach Sporenmerkmalen. Ber Naturwiss-med Ver Innsb. 2011;97:27–33.

[51] Martin L, Randlane T, Martin J. Lichens and their substrate preferences on the Pakri Peninsula (Northwest Estonia). Folia Cryptog Estonica. 2011;48:45–58.

[52] Ellis CJ. A risk-based model of climate change threat: hazard, exposure, and vulnerability in the ecology of lichen epiphytes. Botany. 2013;91(1):1–11.