Symbiotic associations in lecideoid lichens along the Transantarctic Mountains, Antarctica

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# How do symbiotic associations in lecideoid lichens respond to different environmental conditions along the Transantarctic Mountains, Ross Sea region, Antarctica?

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# Abstract

Lecideoid lichens as dominant vegetation-forming organisms in the climatically harsh areas of the southern part of continental Antarctica show clear preferences in relation to environmental conditions (i.e. macroclimate). 306 lichen samples were included in the study, collected along the Ross Sea coast (78°S–85.5°S) at six climatically different sites. The species compositions as well as the associations of their two dominant symbiotic partners (myco- and photobiont) were set in context with environmental conditions along the latitudinal gradient. Diversity values were nonlinear with respect to latitude, with the highest alpha diversity in the milder areas of the McMurdo Dry Valleys (78°S) and the most southern areas (Durham Point, 85.5°S; Garden Spur, 84.5°S), and lowest in the especially arid and cold Darwin Area (~79.8°S). Furthermore, the specificity of mycobiont species towards their photobionts decreased under more severe climate conditions. The generalist lichen species *Lecanora fuscobrunnea* and *Lecidea cancriformis* were present in almost all habitats, but were dominant in climatically extreme areas. *Carbonea vorticosa, Lecidella greenii* and *Rhizoplaca macleanii* were confined to milder areas.

In summary, the macroclimate is considered to be the main driver of species distribution, making certain species useful as bioindicators of climate conditions and, consequently, for detecting climate change.

# **Keywords**

Southernmost areas in Antarctica, latitudinal gradient, polar desert, macroclimate, microhabitat, myco-/photobiont associations

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# Introduction

Polar deserts of the southernmost areas in continental Antarctica are characterized by exceptionally hostile climatic conditions, such as particularly low temperatures and high aridity (Adams et al. 2006; Cary et al. 2010; Magalhaes et al. 2012). Terrestrial life is restricted to ice-free areas, which, apart from a few nunataks, are mainly located along the Transantarctic Mountains forming the west coast of the Ross Sea and Ross Ice Shelf (Monaghan et al. 2005). Because of these special conditions, terrestrial life is rare and can only be found in small areas protected from extreme environmental influences, such as abrasion from windblown particles or high solar radiation, the so-called microhabitats (Hertel 1998; Ruprecht et al. 2012b). They are characterized by sheltered areas in rock crevices or small cavities shielded from the wind and sun that allow life on a small scale in an otherwise hostile environment. The rock surface is often highly weathered which results in a higher water retention capacity, providing the most needed life source for the organisms to survive (Colesie et al. 2014; Green 2009). The only moisture available to rock-dwelling organisms is provided by clouds, fog, dew, sparse precipitation and melting snow (Head and Marchant 2014; Wagner et al. 2020). Additionally, the aspect of the slopes, ridges and depressions as well as the wind regime has an important impact by creating different surface temperatures in small areas (McKendry and Lewthwaite 1990; Yung et al. 2014). However, microhabitats are influenced by both macroclimate and geography, and their life-supporting properties therefore vary along environmental gradients reflected in changing diversity levels and biogeography of Antarctic terrestrial biota (Baird et al. 2019; Lagostina et al. 2021; Magalhaes et al. 2012; Peat et al. 2007; Fig.1; Ruprecht et al. 2012a; Fig.1).

The terrestrial vegetation along the Ross Sea coast (extending from 72°S, Cape Hallett, to 85.5°S, Queen Maud Mountains) is entirely composed of cryptogrammic organisms and dominated by lichens and mosses (Colesie et al. 2014; Ochyra et al. 2008; Peat et al. 2007). Remarkably, the biodiversity of these organisms does not decrease evenly along the latitudinal gradient as one might expect. In fact, the lowest species diversity was recorded at about 79°S at Diamond Hill (Darwin Area), which has by far the harshest climate conditions (lowest humidity; Colesie et al. 2014). However, nonlinear climatic conditions along gradients caused by additional factors, e.g. special wind systems, can be detected effectively with biological systems that act as bioindicators (Dal Grande et al. 2017; Sancho et al. 2019; Singh et al. 2017; Wagner et al. 2020). Additionally, they not only enable the survey of the current state but can also reliably indicate changes in environmental conditions. Due to the structure and diversity of communities, the abundance and distribution of species as well as processes varying along environmental gradients are therefore powerful long-term and large-scale study systems to estimate the consequences of climate change on ecosystems (Sundqvist et al. 2013).

The most abundant vegetation-forming organisms in these areas are lichens, in most of the cases with a crustose thallus fused to the rocky surface or deeply embedded in crevices (Colesie et al. 2014; De los Rios et al. 2004; Hertel 2007; Kappen and Valladares 2007; Ruprecht et al. 2012b). The poikilohydric lifestyle of lichens enables them to survive the harsh climate conditions and the long periods without water and/or light in a dormant state (Schroeter et al. 2011). The symbiotic lifeform of lichens consists of two dominant symbiotic partners: the mycobiont (fungus) and the photosynthetic partner (green algae and/or cyanobacteria: photobiont) and additional associated fungal, algal and bacterial communities forming the holobiome lichen thallus (Aschenbrenner et al. 2016; Grube et al. 2015; Lawrey and Diederich 2003; Ruprecht et al. 2014; Spribille et al. 2016). Therefore, lichens constitute an excellent model for analyzing multi-species associations in one unit to reveal phylogenetic and ecological responses for symbiotic associations.

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Many analyses focused on myco-/photobiont associations have demonstrated that they react sensitively to even small environmental gradients (Dal Grande et al. 2018; Wagner et al. 2020). These results allow the conclusion that mycobionts which are less specialized to specific locations and are able to use a broader range of photobionts, such as the widespread species *Lecidea cancriformis* in continental Antarctica (Ruprecht et al. 2012a; Wagner et al. 2020), are less vulnerable to climate changes. Low photobiont specificity may improve the performance of the lichen symbiosis, e.g. by increasing the adaptive potential to new

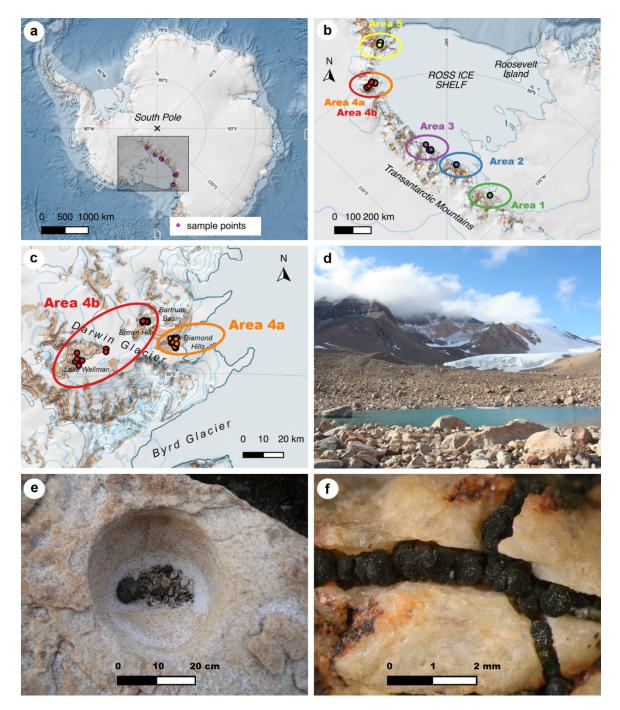


Figure 1. Location of the sample points and lichen habitats. (a) Antarctic continent, investigated area marked with rectangle, (b) location of the six different areas defined in the study, (c) differentiation of area 4 in subareas 4a and 4b, (d) Batrum Basin, (e) microhabitat with crustose lichens at Lake Wellman, (f) chasmolithic growth of *Lecidea cancriformis*. Maps of (a), (b) and (c) are based on the dataset Quantarctica (Matsuoka et al. 2018).

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environmental conditions, and widening the geographical range via ecological niche shifts (Leavitt et al. 2015; Rolshausen et al. 2018; Vančurová et al. 2018). On the other hand, high levels of photobiont specificity are expected under conditions where ecological factors, especially (macro-) climate and/or substrate (e.g. calcareous or siliceous rock), exert a strong selective influence on lichen performance (Peksa and Skaloud 2011; Vančurová et al. 2018; Werth and Sork 2010). Additionally, genetic identity can play a significant role in shaping myco-/photobiont associations along gradients (Dal Grande et al. 2017) or may also lead to turnover zones, suggesting that photobionts are replaced by others as environmental conditions change (Rolshausen et al. 2020). An influence on the selection of *Trebouxia* species due to temperature combined with water availability was suggested in several studies as a key factor of photobiont selection of lichens in Antarctica (Green et al. 2011a; Wagner et al. 2020). Due to the sensitive response of lichen communities to climatic change with modified species compositions and reduced diversity (Ellis 2019; Mayer et al. 2013; Sancho et al. 2017). Consequently, lichens represent excellent bioindicators because of their sensitive responses to environmental changes (Alatalo et al. 2015; Allen and Lendemer 2016; Bassler et al. 2016; Sancho et al. 2019), and especially abundant and cosmopolitan species serve as a valuable model system to record diversity and composition along climatic gradients worldwide.

The current study focuses on the association patterns of the two main symbionts (myco- and photobiont) of the lecideoid lichen group (Ruprecht et al. 2020) that is dominant along the investigated part of the latitudinal gradient (78–85°S) at the Ross Sea coast. The following objectives were addressed: (1) to assess the biodiversity and genetic identity of the symbiotic partners of lecideoid lichens using phylogenic methods; (2) to investigate how the variability of myco-/photobiont associations is related to environmental variables (elevation, temperature, precipitation) using diversity and specificity indices as well as network statistics, and (3) to identify certain myco-/photobiont associations that are representative for climatic conditions and therefore may qualify as bioindicators.

## **Materials & Methods**

#### Study area and investigated lichen specimens

The sample sites were divided in five different main areas (Fig. 1b). Area 4 (Darwin Area) was then subdivided in subareas 4a and 4b, considering the wide range of climate conditions within this region (Figure 1c). Site descriptions of the six regions are given in Table 1, geographical descriptions can be found at the Supplementary Table S1.

Altogether 306 lecideoid lichen specimens were collected on siliceous substrate along a latitudinal gradient (78–85.5°S) of the southwest Ross Sea coast (Antarctica, Fig.1a-c). 147 samples of the genera *Carbonea, Lecanora, Lecidella* and *Lecidea* were collected at 70 different localities from the following sampling areas: Area 1, Scott Glacier/Durham Point; Area 2, Massam Glacier/Garden Spur; Area 3, Mt. Kyffin, Mt. Harcourt, The Gateway; Area 4a, Darwin Area (Diamond Hills, Brown Hills); Area 4b, Darwin Area (Bartrum Basin, Smith Valley and Lake Wellman) (Fig. 1a-c, Supplementary Table S1). To get a better coverage of the latitudinal gradient (Fig. 1a), additionally, 159 lichen samples (collected at 102 different localities) from the Area 5, McMurdo Dry Valleys (MDV), were obtained from the studies of Wagner et al. (2020) and Perez-Ortega et al. (2012), including the solely lecideoid lichen species of the genera *Carbonea, Lecanora, Lecidella, Lecidea* and *Rhizoplaca*. The entire lists of samples can be found at the Supplementary Tables S2-4.

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145.04

	Sampling area	Range of coordinates of sampling sites	Number of sampling sites	Elevation mean (m. a. s. l.)	BIO10: mean temperature, warmest quarter (°C)	BIO12: precipitation, annual mean (mm)
Area 1	Scott Glacier/ Durham Point	S 85.54° W 151.15°	1	370.00	-7.30	190.00
Area 2	Massam Glacier/ Garden Spur	S 84.54°–84.56° W 174.91°–175.01°	2	182.92	-6.96	113.00
Area 3	Mt. Kyffin, The Gateway, Mt. Harcourt	S 83.49°–83.83° E 170.79°–172.76°	6	774.72	-8.20	104.37
Area 4a	Darwin Area: Diamond Hills, Brown Hills	S 79.84°–79.88° E 159.22°–159.39°	30	484.19	-8.23	91.79
Area 4b	Darwin Area: Bartrum Basin, Smith Valley, Lake Wellman	S 79.75°–79.95° E 156.70°–158.67°	31	726.39	-10.18	69.13
Area 5	McMurdo Dry	S 78.02°–78.17°	102	589 /2	-6.24	145.04

**Table 1.** Site descriptions of the six regions defined in the present study, including range of the coordinates of the sampling sites and areas, the number of sampling sites and the BIOCLIM variables BIO10 (mean temp. of the warmest quarter) and BIO12 (annual precipitation) per area.

All voucher specimens are stored in the herbarium of the University of Salzburg (SZU) except for samples collected by Leopoldo G. Sancho which are deposited in the MAF herbarium of the Botany Unit, Fac. Farmacia, in Madrid.

102

E 163.62°-164.10°

589.42

-6.24

#### DNA-amplification, primer-design and sequencing

Valleys

Area 5

Total DNA was extracted from individual thalli by using the DNeasy Plant Mini Kit (Qiagen) following the manufacturer's instructions. For all samples, the internal transcribed spacer (ITS) region of the mycobionts' and photobionts' nuclear ribosomal DNA (nrITS) were sequenced and amplified. Also, additional markers were amplified: for the mycobionts the mitochondrial small subunit (mtSSU) and the low-copy protein coding marker *RPB1*; for the photobionts, the chloroplast-encoded intergenic spacer (psbJ-L) and part of the cytochrome oxidase subunit 2 gene (COX2). This was done using specific primers and PCR-protocols in our project framework (Ruprecht et al. 2020).

The nrITS of the mycobiont was amplified using the primers ITS1F (Gardes and Bruns 1993), ITS4(White et al. 1990), ITS1L (Ruprecht et al. 2020) and ITS4L (Ruprecht et al. 2020). The nrITS of the photobiont was amplified using the primers 18S-ITS unifor (Ruprecht et al. 2012a), ITS4T (Kroken and Taylor 2000), ITS1T (Kroken and Taylor 2000) and ITS4bT\_mod (5'-CCAAAAGGCGTCCTGCA-3'; modified, based on Ruprecht et al. (2014)). For the marker mtSSU, the primer mtSSU rev2 (Ruprecht et al. 2010) and the newly designed primers mtSSU for2 mod1 (5'-AACGGCTGAACCAGCAACTTG-3') and mtSSU rev1 (5'-AGGYCATGATGAACTGTCTT-3') were used. For *RPB1*, gRPB1-A for (Matheny et al. 2002), fRPB1-C rev (Matheny et al. 2002) and RPB1\_for\_Lec (Ruprecht et al. 2020) were chosen. For the marker COX2, COXIIf2 and COXIIr (Lindgren et al. 2014) and COXII\_sense (Ruprecht et al. 2020) were used, for psbJ-L, newly designed psbL\_for1 (5'-GTTGAATTAAATCGTACTAGT-3') psbL-sense and psbJ-antisense (Ruprecht et al. 2014) were chosen.

Unpurified PCR-products were sent to Eurofins Genomics/Germany for sequencing.

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#### Phylogenetic analysis

For both symbionts, the sequences were assembled and edited using Geneious version 8.0.5 (https://www.geneious.com) and aligned with MAFFT v7.017 (Katoh et al. 2002).

Maximum likelihood analyses were calculated with IQ-TREE v1.6.12 (Nguyen et al. 2014), using the model selection algorithm ModelFinder (Kalyaanamoorthy et al. 2017). Branch supports were obtained with the implemented ultrafast bootstrap (UFBoot; Minh et al. 2013). Number of bootstrap alignments: 1000, maximum iteration: 1000, minimum correlation coefficient: 0.99. Additionally, a SH-aLRT branch test (Guindon et al. 2010) was performed. Each branch of the resulting tree was assigned with SH-aLRT as well as UFBoot supports. The branches with SH-aLRT < 80 % and/ or UFboot < 95 % were collapsed by adding the command -minsupnew 80/95 to the script.

In order to be able to use all samples with an incomplete marker set, a multi-marker phylogeny with a reduced number of samples and, in comparison, the complete data set with the marker ITS were calculated and compared, respectively for each symbiont.

For the photobiont, the classification and labeling of the different operational taxonomical units (OTUs) followed the concepts of Muggia et al. (2020) and Ruprecht et al. (2020), using automatic barcode gap discovery (ABGD; Puillandre et al. 2012), based on the marker ITS. The threshold of 97.5 % sequence similarity set by Leavitt et al. (2015) and applied by Ruprecht et al. (2020) was used to ensure clear delimitation of OTUs and sub-OTUs.

#### Analysis of spatial distribution

Unless stated otherwise, analysis was conducted in R (R Core Team 2020; version 3.6.3, https://www.r-project.org) using RStudio (RStudio Team 2016; version 1.1.463, https://rstudio.com); figures where produced using the R package ggplot2 (Wickham 2009) and processed using Adobe Photoshop (version 22.2.0, https://www.adobe.com).

Based on data from CHELSA (*Climatologies at high resolution for the earth's land surface areas*; Karger et al. 2017), the 19 BIOCLIM variables (Nix 1986) were calculated for each sample point using the R functions raster() and extract() of the package raster (Hijmans 2020). These variables are derived variables from the monthly minimum, maximum, mean temperature and mean precipitation values, developed for species distribution modeling and related ecological applications (Karger et al. 2017). For the analyses of this study, BIO10 (mean temperature of the warmest quarter) and BIO12 (annual precipitation) were chosen, as these two variables showed the strongest correlations with the diversity and specificity indices (see below).

For analyzing the spatial distribution of the lichen samples, alpha, beta and gamma diversity values were calculated. The concept was developed in 1960 by Whittaker (Whittaker 1960) who distinguished three aspects or levels of species diversity in natural communities: (1) alpha diversity, the species richness within a particular area, (2) beta diversity, the extent of changes in species diversity between the areas, and (3) gamma diversity, a measure of the overall diversity for the different areas within the whole region. These diversity indices were calculated separately for mycobiont species and photobiont OTUs, using the R functions AlphaDiversity(), BetaDiversity() and GammaDiversity() of the package entropart (Marcon and Hérault 2015), which give reduced-bias diversity values (diversity order: q = 1 (Shannon diversity); weights:  $w_i = n/n$  with  $n_i$ , number of samples in area *i* and *n*, total

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number of samples). Next, alpha diversity was analyzed for correlations with the following variables: elevation, latitude, BIO10 and BIO12.

To determine whether mycobiont species or photobiont OTU community composition are related to environmental variables (elevation, BIO10 and BIO12), constrained analyses of principal coordinates were conducted, using the R function capscale() of the package vegan (Oksanen et al. 2019; distance: Bray Curtis). Prior to analysis, to standardize species composition data (convert species abundances from absolute to relative values), a Hellinger transformation was performed on the community matrix, using the R function decostand() of the package vegan (Oksanen et al. 2019). The variance explained by constrained ordination was tested by a Monte Carlo permutation test, using the R function anova() of the package vegan (Oksanen et al. 2019).

A Mantel test was performed to test whether the differences in mycobiont species and photobiont OTU community composition between samples are related to physical distance, using the R function mantel() of the package vegan (Oksanen et al. 2019).

#### Haplotype analysis

In order to ensure that the entire data set could be processed, all further analyses were carried out using only complete sequences of the marker ITS for all calculations. The number of haplotypes, *h*, of the different mycobiont species and photobiont OTUs was determined using the function haplotype() of the R package pegas (Paradis 2010). Haplotype networks were computed, using the function haploNet() of the R package pegas (Paradis 2010) for mycobiont species and photobiont OTUs with  $h \ge 2$  and at least one haplotype with  $n \ge 3$  (*Carbonea* sp. 2, *Lecanora fuscobrunnea, Lecidea cancriformis, Lecidella greenii, Lecidella siplei, Lecidella* sp. nov2 and *Rhizoplaca macleanii,* as well as  $Tr_A02$ ,  $Tr_I01$  and  $Tr_S02$ ). The frequencies were clustered in 10% ranges, for example the circles of all haplotypes making up between 20-30% have the same size. Additionally, for the most common mycobiont *L. cancriformis* and the photobiont OTU  $Tr_S02$ , haplotype networks based on multimarker data sets were calculated, to show that the distribution of haplotypes remains congruent.

#### Diversity and specificity indices of mycobiont species and photobiont OTUs

The haplotype as well as the nucleotide diversity was calculated for each identified mycobiont and photobiont species with more than one sample, using the functions hap.div() and nuc.div() of the R package pegas (Paradis 2010), respectively. The haplotype diversity, *Hd*, represents the probability that two randomly chosen haplotypes are different (Nei 1987), the nucleotide diversity,  $\pi$ , gives the average number of nucleotide differences per site between two randomly chosen DNA sequences (Nei and Li 1979). Additionally, the ratio of the number of haplotypes *h* divided by the number of samples *N* was calculated.

Furthermore, different metrics for quantifying the phylogenetic species diversity and the specificity of the mycobiont species and photobiont OTUs towards their interaction partners were calculated. Those included the indices *NRI* (Net relatedness index), *PSR* (Phylogenetic species richness) and the Pielou evenness index *J*'. (*Note: to make interpretation similar to the other metrics, for further analyses* 1 – J' *instead of* J' *was used.*) An overview of these diversity metrics is given in Supplementary Table S5.

In order to analyze the correlation of these diversity metrics with environment, for every mycobiont species and photobiont OTU with  $n \ge 10$  (*Carbonea* sp. 2, *C. vorticosa, Lecanora fuscobrunnea, Lecidea cancriformis, L. polypycnidophora, Lecidella greenii,* 

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*L. siplei, Rhizoplaca macleanii* and *Trebouxia* OTUs *Tr\_*A02, *Tr\_*I01, *Tr\_*S02, *Tr\_*S15, *Tr\_*S18) the mean values of the sample locations of the following variables were calculated: elevation, latitude, BIO10 and BIO12.

#### Analysis of mycobiont – photobiont associations

To analyze the associations between mycobiont species and photobiont OTUs, bipartite networks were computed, using the R function plotweb() of the package bipartite (Dormann et al. 2008). This was done for each area separately. Additionally, for each bipartite network, the index  $H_2$ ' was calculated.  $H_2$ ' is derived from Shannon entropy and characterizes the degree of complementary specialization of partitioning among the two parties of the network. It ranges from 0 for the most generalized to 1 for the most specialized case und was computed using the R functions H2fun() of the package bipartite (Dormann et al. 2008).

Usually, in the context of bipartite networks, also the *d'* value (specialization index) is computed. This value was originally defined for pollination networks and calculates how strongly a species deviates from a random sampling of interacting partners available (Dormann 2011). Thus, in the case of lichens, the *d'* value of a mycobiont species is based on the assumption that for every site of a sampling area, the whole set of photobiont OTUs basically is available. As this is not true for this study, this index was not included.

# **Results**

#### Phylogenetic analysis

For both the mycobiont and photobiont molecular phylogenies from multi-locus sequence data (nrITS, mtSSU and *RPB1* for the mycobiont (140 samples) and nrITS, psbJ-L and COX2 for the photobiont (139 samples) were inferred (Supplementary Fig. S1 and S3). Additionally, phylogenies based solely on the marker nrITS were calculated (Supplementary Fig. S2 and S4), to include samples where the additional markers were not available. Both analyses include only accessions from the study sites (Fig. 1, Table 1). The phylogenies based on the multi-locus data were congruent to the clades of the phylogenies based on the marker nrITS. Thus, in the following, the focus will be only on the latter.

**Mycobiont:** The final data matrix for the phylogeny based on the marker nrITS comprised 306 single sequences with a length of 550 bp. It included sequences of the families *Lecanoraceae* and *Lecideaceae*. The phylogenetic tree was midpoint rooted and shows a total of 19 strongly supported clades on species level, assigned to five genera. The backbone is not supported and therefore the topology will not be discussed. All genera are clearly assigned to their family level and are strongly supported. Only *Lecanora physicella* forms an extra clade as sister to the families *Lecideaceae* and *Lecanoraeae*, which is not the case at the multimarker phylogeny. *L. physciella* has still an uncertain status, because of morphological similarities to both sister families (Ruprecht et al. 2012b). The clade of the genus *Lecidea* revealed seven species (*L. andersonii, L. polypycnidophora, L.* UCR1, *L.* sp. 5, *L. lapicida, L. cancriformis* and *L.* sp. 6), *Lecanora* five species (*L. physciella, L. sp. 2, L. fuscobrunnea, L. cf. mons-nivis, L.* sp. 3), *Carbonea* three species (*C.* sp. URm1, *C. vorticosa, C.* sp. 2), and *Lecidella* three species (*L. greenii, L. siplei, L.* sp. nov2). The samples allocated to the genus *Rhizoplaca* were monospecific (*R. macleanii*). The taxonomical assignment of the obtained sequences were based on the studies of Ruprecht et al. (2020) and Wagner et al. (2020).

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**Photobiont:** The final data matrix for the phylogeny based on the marker nrITS comprised 281 single sequences with a length of 584 bp. The phylogenetic tree was midpoint rooted and shows six strongly-supported clades, assigned to seven different OTU levels (Puillandre et al. 2012), using the concept of Muggia et al. (2020) and Ruprecht et al. (2020). The backbone is not supported and therefore the topology will not be discussed. All of the OTUs belong to the genus *Trebouxia* (clades A, I, S), comprising *Tr\_*A02, *Tr\_*A04a, *Tr\_*I01, *Tr\_*I17, *Tr\_*S02, *Tr\_*S15 and *Tr\_*S18. Photobiont sequences taken from Perez-Ortega et al. (2012), which were labelled only with numbers, were renamed to assign them to the appropriate OTUs (Ruprecht et al. 2020).

#### Analysis of spatial distribution

In general, the most common mycobionts were *Lecidea cancriformis* (94 of the 306 samples), *Rhizoplaca macleanii* (51 samples) and *Lecidella greenii* (37 samples), followed by *Carbonea* sp. 2 (13 samples), *C. vorticosa* (11 samples), *Lecidea polypycnidophora* (10 samples) and *Lecidella siplei* (10 samples; see Supplementary Fig. S5). Nine mycobiont species were found exclusively in area 5 (MDV, 78°S): *Carbonea vorticosa, Lecanora* cf. *mons-nivis, L.* sp. 2, *Lecidea lapicida, L. polypycnidophora, L.* sp. 5, *L.* sp. 6, *L.* UCR1 and *Rhizoplaca macleanii*. On the other hand, only the mycobiont species *Lecidea cancriformis* was found in all the six areas; *Lecanora fuscobrunnea* was present in all the areas with the exception of area 2.

The most common photobiont OTUs were  $Tr_A02$  (165 of the 281 samples) and  $Tr_S02$  (59 samples), both of them occurring in all the six different areas, followed by  $Tr_S18$  (32 samples),  $Tr_S15$  (10 samples, confined to area 5) and  $Tr_I01$  (10 samples). However, of the 149 photobiont samples of area 5, 134 (89,93 %) were assigned to  $Tr_A02$ . This percentage is much higher than in the other areas (area 1: 44,44 %, area 2: 69,23 %, area 3: 21,74 %, area 4a: 7,69 %, area 4b: 6,67 %), even if those samples with mycobionts occurring exclusively in area 5 (see above) were excluded (76.56 % of the 64 remaining samples are assigned to  $Tr_A02$ ).

			Mycobior	nt species		Photobiont OTUs				
Area	Number of lichen samples	Number of identified species	Alpha diversity	Beta diversity	Gamma diversity	Number of identified OTUs	Alpha diversity	Beta diversity	Gamma diversity	
1	28	7	5.23			3	2.28			
2	13	5	5.48			3	2.20			
3	27	7	5.99	1.00	0.00	5	3.70	1.64	2.25	
4a	48	6	3.55	1.69	9.92	6	4.06	1.64	3.35	
4b	31	2	1.88			4	2.36			
5	159	16	8.93			4	1.50			

 Table 2. Number of lichen samples, number of identified mycobiont species and photobiont OTUs, as well as alpha, beta and gamma diversity values of mycobiont species/ photobiont OTUs for the different areas.

The alpha, beta and gamma diversity values are given in Table 2. For the mycobionts, the value of alpha diversity (species richness within a particular area) was the highest in area 5 (8.93) and the lowest in area 4b (1.88). In contrast, for the photobionts, the lowest alpha diversity value was found in area 5 (1.50) and the highest in area 4a (4.06). Thus, referring to this, area 5 plays a remarkable role: compared to the other areas, it shows the highest richness of mycobiont species on the one hand and the lowest richness of photobiont OTUs on the other hand.

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The beta diversity values (extent in changes of species diversity between the areas) for mycobiont species and photobiont OTUs are quite similar (1.69 and 1.64, respectively). This is in contrast to gamma diversity values: the overall diversity for the different areas within the whole region is much higher for mycobiont species (9.92) than for photobiont OTUs (3.35).

#### Influence of environmental factors (elevation, precipitation and temperature)

First, the proportion of  $Tr_A02$  samples was significantly correlated to BIO10 means of the areas (R = 0.87, p = 0.022; see Supplementary Fig. S6): the higher the temperature mean values of the warmest quarter of an area, the higher the proportion of samples containing photobionts that are assigned to the OTU  $Tr_A02$ .

The alpha diversity values of mycobiont species significantly positively correlated with BIO10 (R = 0.88, p = 0.021; see Supplementary Fig. S7): the higher the temperature mean values of the warmest quarter, the higher the mycobiont diversity within this particular area.

Furthermore, the differences in mycobiont species community composition were significantly related to BIO10 (constrained principal coordinate analysis: F = 14.7137, p = 0.001, see Supplementary Fig. S8), BIO12 (F = 2.7535, p = 0.012), elevation (F = 2.5108, p = 0.025) and the geographic separation of the samples (Mantel statistic r = 0.1288, p = 0.0002).

The differences in community composition of photobiont OTUs were related significantly to BIO10 (constrained principal coordinate analysis: F = 48.5952, p = 0.001, see Supplementary Fig. S9), BIO12 (F = 4.4848, p = 0.008), elevation (F = 6.8608, p = 0.002), and physical distance (Mantel statistic r = 0.4472, p = 0.0001).

#### Haplotype analysis

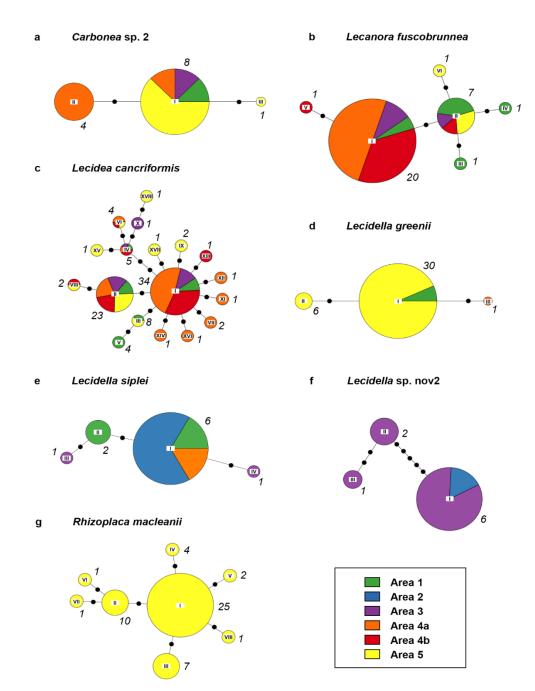
Haplotype networks were computed for the mycobiont species and photobiont OTUs with  $h \ge 2$  and at least one haplotype with  $n \ge 3$  (*Carbonea* sp. 2, *Lecanora fuscobrunnea*, *Lecidea cancriformis*, *Lecidella greenii*, *L. siplei*, *L.* sp. nov2 and *Rhizoplaca macleanii*, as well as  $Tr_A02$ ,  $Tr_101$  and  $Tr_S02$ ), in both cases based on nrITS sequence data (Figs. 2 & 3). The samples of *Carbonea vorticosa* (11) were all assigned to a single haplotype, which was also true for *Lecidea polypycnidophora* (10 samples),  $Tr_S15$  (10 samples) and  $Tr_S18$  (32 samples). Figure 3b and c illustrate the subdivision of  $Tr_101$  (Muggia et al. 2020) into  $Tr_101j$  (Leavitt et al. 2015; Ruprecht et al. 2020) and  $Tr_101k$  (in this study), and the subdivision of  $Tr_S02$  into  $Tr_S02$  (Leavitt et al. 2015), and  $Tr_S02c$  (Ruprecht et al. 2020).

The haplotype networks include pie charts showing the occurrence of the different haplotypes within the different areas. All haplotypes of *Rhizoplaca macleanii* are restricted to area 5, as well as *Lecidella greenii* mainly to area 5 and areas 1 & 4a, and *Lecidella* sp. 2 to areas 2 & 3. However, all other species do not suggest a spatial pattern with different haplotypes being specific for different areas. Moreover, the distribution turned out to be rather unspecific, with a great part of the haplotypes found in multiple areas. For the sake of completeness, additionally, haplotype networks based on multi-locus sequence data were computed for the most abundant mycobiont species and photobiont OTU with multi-locus data available (*Lecidea cancriformis* and *Tr\_S02*). Not

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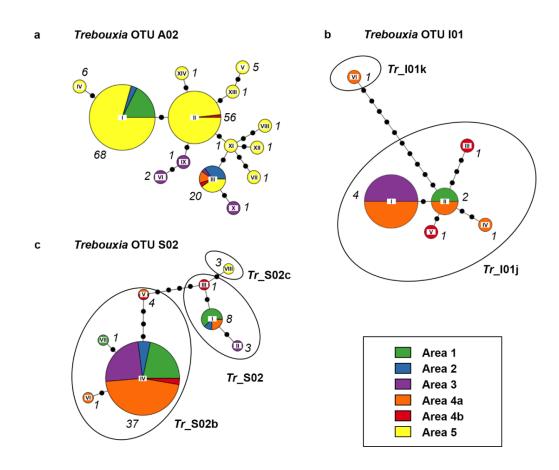
surprisingly, those networks show a greater number of different haplotypes, but they also do not allow conclusions concerning spatial patterns of area specific haplotypes (see Supplementary Fig. S10).



**Figure 2.** Haplotype networks of mycobiont species with  $h \ge 2$  and at least one haplotype with  $n \ge 3$ , showing the spatial distribution within the different areas. Based on nrITS data. (a) *Carbonea* sp. 2, (b) *Lecanora fuscobrunnea*, (c) *Lecidea cancriformis*, (d) *Lecidella greenii*, (e) *Lecidella siplei*, (f) *Lecidella* sp. nov2, (g) *Rhizoplaca macleanii*. Roman numerals at the center of the pie charts refer to the haplotype IDs; the italic numbers next to the pie charts give the total number of samples per haplotype. The circle sizes reflect relative frequency within the species; the frequencies were clustered in ten (e.g. the circles of all haplotypes making up between 20-30 % have the same size). Note: only complete sequences were included.

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**Figure 3.** Haplotype networks of photobiont OTUs with  $h \ge 2$  and at least one haplotype with  $n \ge 3$ , showing the spatial distribution within the different areas. Based on nrITS data. (a)  $Tr_A02$ , (b)  $Tr_101$ , (c)  $Tr_S02$ . Roman numerals at the center of the pie charts refer to the haplotype IDs; the italic numbers next to the pie charts give the total number of samples per haplotype. The circle sizes reflect relative frequency within the species; the frequencies were clustered in ten (e.g. the circles of all haplotypes making up between 20-30 % have the same size). Note: only complete sequences were included.

#### Diversity and specificity indices of mycobiont species and photobiont OTUs

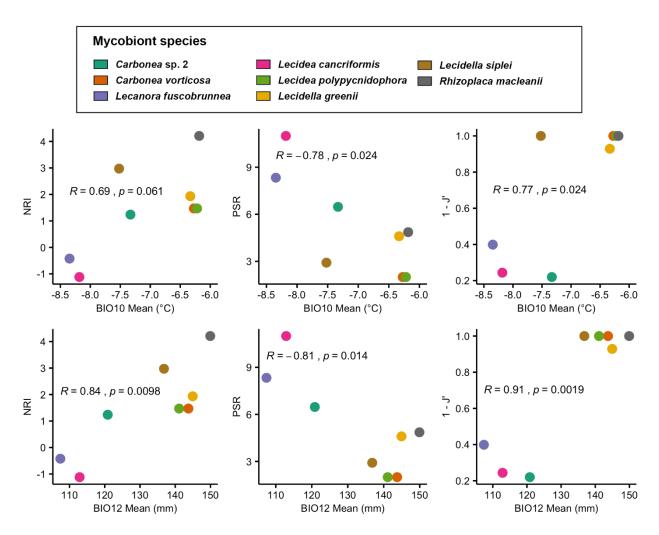
The diversity and specificity indices for the different mycobiont species and photobiont OTUs are given in Supplementary Table S6.

For the sample locations of mycobiont species with  $n \ge 10$ , BIO10 was strongly correlated to the specificity indices *NRI* (net relatedness index) and significantly correlated to *PSR* (phylogenetic species richness) and 1 - J' (Pielou evenness index). BIO12 was significantly correlated to *NRI*, *PSR* and 1 - J'. Figure 4 illustrates these correlations: the higher the BIO10 and BIO12 mean values, the higher the *NRI* (phylogenetic clustering of the photobiont symbiosis partners), the lower the *PSR* (increased phylogenetically relatedness of photobiont symbiosis partners) and the higher 1 - J' (less numerically evenness of the photobiont symbiosis partners). Thus, for the mean values of the sample locations of a mycobiont species, a comparatively high temperature of the warmest quarter and high annual precipitation occurs with associated photobionts that are phylogenetically clustered and closer related to each other. The lowest values of NRI and the highest values of PSR were developed by *Lecidea cancriformis* and

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Lecanora fuscobrunnea, which also showed the lowest BIO10 and BIO12 mean values at their sample sites. On the contrary, the highest values of NRI and PSR were developed by *Rhizoplaca macleanii*, which also had the highest BIO10 and BIO12 means.

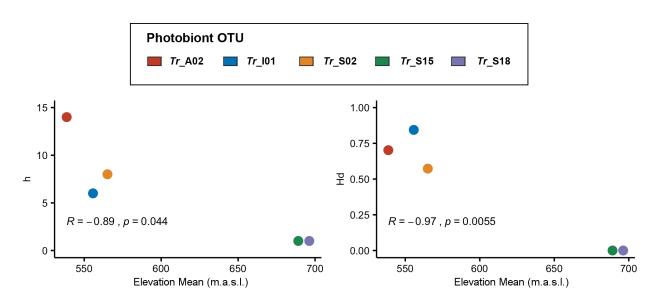


**Figure 4.** Correlation plots. Specificity indices *NRI* (net relatedness index), *PSR* (phylogenetic species richness and 1 - J' (Pielou evenness index) against mean values of BIO10 (mean temperature of warmest quarter) and BIO12 (annual precipitation) for mycobiont species with  $n \ge 10$ .

For the sample locations of photobiont OTUs with  $n \ge 10$ , elevation significantly negatively correlated with h (number of haplotypes) and Hd (haplotype diversity): the higher the mean elevation of sample sites, the lower the number of haplotypes and the lower the probability that two randomly chosen haplotypes are different (Fig. 5). The highest values of h and Hd were for  $Tr_A02$ ,  $Tr_101$  and  $Tr_S02$ , which occurred at sample sites with comparatively low elevations. In contrast,  $Tr_S15$  and  $Tr_S18$  occurred at very high elevations and showed very low values of h and Hd.

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**Figure 5.** Correlation plots. Diversity indices *h* (number of haplotypes) and *Hd* (haplotype diversity) against mean elevation of sample sites for photobiont OTUs with  $n \ge 10$ .

#### Analysis of mycobiont-photobiont associations

Bipartite networks were calculated for all associations between mycobiont species (lower level) and the respective photobiont OTUs (higher level) for all areas (Fig. 6). The  $H_2$ ' value (overall level of complementary specialization of all interacting species) was highest in area 2 (0.921), indicating a network with mostly specialized interactions: within this network, with the exception of *Lecidea andersonii*, the mycobiont species are associated exclusively with one single photobiont OTU. The second highest  $H_2$ ' value was developed by area 4a (0.710); in contrast, area 4a showed the lowest  $H_2$ ' value (0.260), with the most abundant mycobiont species *Lecidea* cancriformis showing associations with five different photobiont OTUs. The  $H_2$ ' values of area 1, area 3 and area 5 indicate medium specification.

In addition, the bipartite networks illustrate the different occurrence of mycobiont species and photobiont OTUs within the different areas: For example, in area 1 (and area 2), five (seven) different mycobiont species are associated with only three different photobiont OTUs. In contrast, in area 4b, only two different mycobiont species are associated with four different photobiont OTUs. In area 5, the number of associated photobiont OTUs is also four, but those four OTUs are associated with 16 different mycobiont species.

The network matrix giving all the associations between the mycobiont species and photobiont OTUs is presented in Supplementary Table S7.

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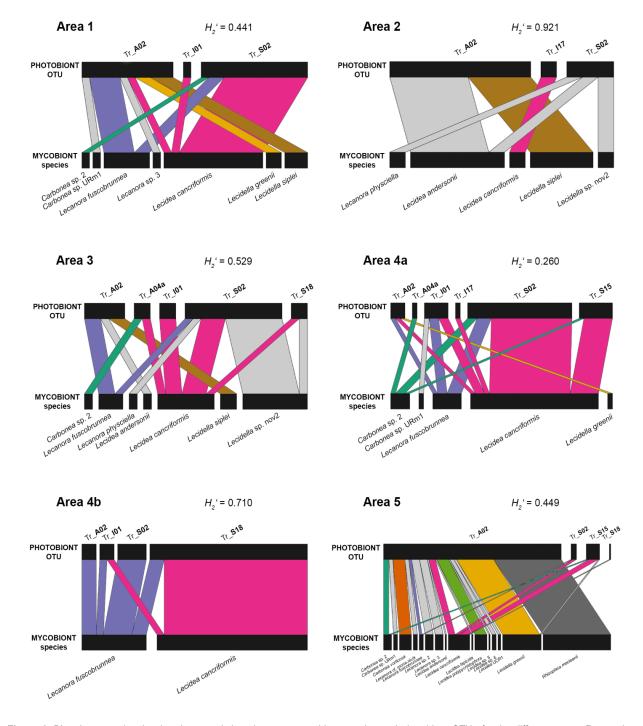


Figure 6. Bipartite networks showing the associations between mycobiont species and photobiont OTUs for the different areas. Rectangles represent species, and the width is proportional to the number of samples. Associated species are linked by lines whose width is proportional to the number of associations.

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# Discussion

The present study investigated the diversity of lecideoid lichens at six different sample areas along a latitudinal gradient (78°S– 85.5°S) along the Transantarctic Mountains, Ross Sea region, at continental Antarctica. The distribution of the different mycobiont species and photobiont OTUs differed considerably between the six sample areas, which is expressed in alpha diversity values (species richness; Supplementary Figure S7). The extreme climate (lowest mean temperatures of the warmest quarter, lowest precipitation) at the areas 4a & b (Darwin Area, ~79.8°S) was reflected in the lowest species richness of mycobionts, and comparatively high species richness of photobiont OTUs. On the other hand, in the climatically mildest area 5 (McMurdo Dry Valleys, 78°S, highest mean temperature, second highest precipitation), the highest species richness of mycobionts and the lowest species richness of photobiont OTUs was found. The number of different photobiont OTUs identified per area is comparatively at a similar level (varying between three and six different groups, Table 2), which is remarkable when considering the great differences in sample sizes (varying between 13 samples (area 2) and 159 samples (area 5), Table 2).

These results are largely consistent with the findings of Colesie et al. (2014), who previously reported that macroclimatic conditions along the latitudinal gradient are not linear. The area at Diamond Hill (79.9°S, part of area 4a, Darwin area, Fig. 1c) addressed in Colesie et al. 2014 showed the lowest species diversity along the latitudinal gradient, which was at least confirmed for area 4b in the current study. Areas 4a and especially 4b are characterized by the harshest climatic conditions such as very low humidity and temperatures, and species numbers in relation to the number of samples are also lowest there; the two generalist species *Lecidea cancriformis* and *Lecanora fuscobrunnea* are dominant. Milder areas, further south (83.5–85.5°S), the McMurdo Dry Valleys (78°S), allow a higher species diversity (Green et al. 2011b; Perez-Ortega et al. 2012). The higher diversity at the southern sites, particularly Mt. Kyffin, appears in part to be due to the occurrence of relic species (Green et al. 2011b). However, Colesie et al. (2014) suggested that physical barriers could be the reason for the low diversity in the Darwin area, but the unspecified haplotype distribution of the widespread species suggest that this is not the case. Since the substrate at all sites is siliceous and there are no other obvious limiting factors, the most likely reason for the limited occurrence of certain species is primarily dependent on abiotic factors, in particular, the environmental conditions caused by geography and macroclimate (Dal Grande et al. 2018).

The uniformity of photobiont OTUs in area 5 is mainly due to a strong dominance (89.93 %) of the OTU *Trebouxia* A02 which occurred in all the six sampling areas. The proportion of photobiont samples assigned to *Tr\_*A02 was significantly correlated to the mean value of BIO10 (mean temperature of the warmest month) of the areas. Thus, higher temperatures are related to a higher relative abundance of *Tr\_*A02, and colder temperatures to a higher relative abundance of other *Trebouxia* OTUs. This result is in basic agreement with the previous study of Wagner et al. (2020). The community composition of both, mycobiont species as well as photobiont OTUs is significantly related to elevation, BIO10 and BIO12 (annual precipitation). Thus, as sampling sites become more dissimilar in terms of elevation, BIO10 or BIO12, they also become more dissimilar in terms of community composition. These findings are partially supported by Rolshausen et al. (2020) who surveyed *Trebouxia* communities in temperate climates, suggesting that photobionts are replaced by others as environmental conditions change. In addition, significant correlations emerged between the composition of mycobiont and photobiont OTU communities and the geographic separation of the samples: The further the sampling sites are spatially separated, the more dissimilar the corresponding communities become, which is in agreement with Fernandez-Mendoza et al. (2011).

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Furthermore, the specificity of mycobiont species towards their photobiont partners was shown to be related to environmental variables; these findings are partially in agreement with the studies of Peksa and Skaloud (2011), Singh et al. (2017) and Rolshausen et al. (2018), who reported climate as well as substrate a selective pressure in terms of increased specificity of mycobiont-photobiont interactions. However, the current study has shown that a higher value of BIO10 correlated with a higher phylogenetic clustering of the symbiotic partners of a single mycobiont species (higher 1 - J' and NRI values) and a closer phylogenetic relatedness of these photobionts (lower PSR values). Similarly, the specificity of mycobiont species towards their photobiont symbiosis partners also correlated with BIO12 mean values: A higher value of BIO12 is related to higher values of 1 - J'and NRI and to lower values of PSR. Consequently, the mycobiont species with  $n \ge 10$  showing the highest BIO10 and BIO12 mean values at its sample locations (Rhizoplaca macleanii) also had the highest value of NRI and a rather low value of PSR, as it was solely associated with  $Tr_A02$ . On the other hand, the two mycobiont species with  $n \ge 10$  showing the lowest mean values of BIO10 and BIO12 (Lecanora fuscobrunnea and Lecidea cancriformis) exhibited the lowest values of NRI and the highest values of PSR, as they had associations with the phylogenetically distinct Trebouxia OTUs A02, 101, S02 and S18 (Lecanora fuscobrunnea) or all seven Trebouxia OTUs of this study (Lecidea cancriformis), respectively. Additionally, Lecanora fuscobrunnea and Lecidea cancriformis were the two most widespread species that occurred in five of the six (L. fuscobrunnea) or all the six different areas (L. cancriformis). This result is in agreement with former studies that had shown that L. cancriformis is able to associate with all known photobiont species, and is one of the most widespread lichens in continental Antarctica (Castello 2003; Ruprecht et al. 2012a; Ruprecht et al. 2010; Wagner et al. 2020). Previous studies suggested that a higher photobiont diversity within a single lichen species is indicative of a lower selectivity by the mycobiont, and that this condition is related to enhanced colonization ability (Blaha et al. 2006; Guzow-Krzeminska 2006; Wirtz et al. 2003). According to a model developed by Yahr et al. (2006), selectivity may vary between habitats and may enable lichens to select a photobiont that is well adapted to conditions of the local environmental. These photobiont switches were suggested to increase the geographical range and ecological niche of lichen mycobionts, but may also lead to genetic isolation between mycobiont populations and thus drive their evolution (Fernandez-Mendoza et al. 2011). More generally, flexibility concerning the partner choice has been considered to be an adaptive strategy to survive harsher environmental conditions (Engelen et al. 2016; Leavitt et al. 2013; Singh et al. 2017; Werth and Sork 2010).

Dal Grande et al. (2018) reported elevational preferences for some *Trebouxia* taxa at the OTU level at a mountain range in central Spain, covering an elevational gradient of 1400 m. Additionally, in the present study, the mean elevation of photobiont OTUs were negatively correlated to differences in diversity indices: the dominant photobiont OTU  $Tr_A02$ , occurring in all the six different areas, exhibited the lowest mean value of elevation of sample sites and had the highest number of haplotypes and the highest value of haplotype diversity. On the other hand, the OTUs  $Tr_S15$  and  $Tr_S18$  had the highest mean elevations and the lowest values of *h* and *Hd*. Thus, higher mean elevation of photobiont OTUs were significantly related to a lower number of haplotypes (*h*) and a lower haplotype diversity (*Hd*).

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# Conclusions

Lichens and their myco-/photobiont associations clearly show environmental preferences and therefore are useful as bioindicators. The *Trebouxia* OTU A02 occurred in all the six different areas and was dominant in milder areas, whereas in colder areas, a higher relative abundance of other *Trebouxia* OTUs was found. Accordingly, mycobiont species occurring in milder areas (like *Carbonea vorticosa, Lecidella greenii* and *Rhizoplaca macleanii*) are almost exclusively associated with *Tr\_A02*, while the generalist mycobiont species *Lecidea cancriformis* und *Lecanora fuscobrunnea*, occurring in a broad range of climatically different environments, show associations with phylogenetically distinct photobiont OTUs. However, if they are the only lecideoid lichen species present in certain areas, then they are also meaningful bioindicators of extreme climatic conditions.

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# **Additional information**

**Competing interests:** The authors declare no competing interests.

Supplementary information: Supplementary Material 1: Tables, Supplementary Material 2: Figures

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# Supplementary Material 1: Tables

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Supplementary Table S1. Geographical description of sampling sites.

	Sampling area	Range of coordinates of sampling sites	Collected by	Geographical description
Area	Scott Glacier/	S 85.54°	Leo Sancho (2011)	Durham Point emerges as a big cliff closed to the confluence of Scott Glacier with the Ross Ice Shelf and it is surrounded by frozen
1	Durham Point	W 151.15°		lakes. The substrate is predominantly made up of crystalline plutonic (granite) or metamorphic rocks.
Area	Massam Glacier/	S 84.54°–84.56°		Garden Spur is a narrow rocky ridge at the lowest end of Shackleton Glacier. The substrate is predominantly made up of crystalline
2	Garden Spur	W 174.91°-175.01°	Leo Sancho (2011)	plutonic (granite) or metamorphic rocks.
Area	Mt. Kyffin, The	S 83.49°–83.83°	Leo Sancho (2011)	The investigated area of Mt. Kyffin, Gateway, Mt. Harcourt and surroundings is located at the southern edge of Beardsmore Glacier.
3	Gateway, Mt. Harcourt	E 170.79°–172.76°	Roman Türk (2003)	The mountains are formed by Goldie Formation greywacke (Gunn and Walcott 1962) and schist as well as crystalline plutonic (granite) or metamorphic rocks.
Area 4a	Darwin Area: Diamond Hills,	S 79.84°–79.88° E 159.22°–159.39°	RomanTürk (2004, 2009)	<b>Diamond Hill</b> is located at the eastern edge of the Transantarctic Mountains, close to the Ross Ice Shelf and north from the Darwin Glacier. Climate conditions are characterized by higher air humidity and precipitation that support a higher diversity and abundance of lichens.
40	Brown Hills	L 109.22 -109.09	2003)	The <b>Brown Hills</b> are located in the north of Darwin Glacier. The Carlyon Granodiorite makes up most of the Brown Hills and includes a variably foliated, biotite-hornblende granodiorite and granite (Simpson and Cooper 2002). This site appears to be a particularly dry part of the continental Transantarctic Range.
Area	Darwin Area: Bartrum Basin,	S 79.75°–79.95°	RomanTürk (2004,	Bartrum Basin is a very dry area, located in the north-west of the Brown Hills very dry area. The dominant rock types are dolerite and granite.
4b	Smith Valley, Lake Wellman	E 156.70°–158.67°	2007, 2009)	The surroundings of the <b>Smith Valley</b> and <b>Lake Wellman</b> are characterized by a very dry climate, caused by a high evaporation rate due to low average air humidity and/or continuous winds originating from the cold glacier regions. The bedrock surrounding this area is sandstone from the Beacon Group and dolerite from the Ferrar dolerite sills.
Aroc	MoMurdo Dr.	S 78.02°–78.17°	Roman Türk (2010)	The landscape of the <b>McMurdo Dry Valleys</b> is a mosaic of glacially formed valleys with intervening high ground, ice-covered lackes, ephemeral streams, arid rocky soils, ice-cemented soils, and surrounding glaciers along the steep scree and boulder slopes (Doran et
Area 5	McMurdo Dry Valleys	E 163.62°–164.10°	Ulrike Ruprecht (2009, 2011)	al. 2002; Stichbury et al. 2011; Yung et al. 2014). There are four main valleys (Miers Valley, Garwood Valley, Hidden Valley, and Marshall Valley) and some other extensive ice-free areas (Shangri-La). The valleys have the typical glaciated form with a U-cross-section with steep sides, often with scree slopes, and the valley floors are covered with glacial drift.

Supplementary Table S2. Samples used in this study, with information on collecting localities and Genbank accession numbers of different markers.

				Mycobiont					Associated green micro algae (Trebouxia)				
					A	ccession numb	oers		Ac	Accession numbers			
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	OTU ID	nrITS2	psbJ-L	COX2		
MAF_DP1_01	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208709	MK205016	MK226962	Tr_S02	MK226844	MK226760	MK227045		
MAF_DP1_02	1	-85.539	-151.150	Lecanora fuscobrunnea Dodge & Baker	MK208710	MK205017	MK226963	Tr_S02	MK226845	MK226761	MK227046		
MAF_DP1_04	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208711	MK205018	MK226964	Tr_S02	MK226846	MK226800	MK227047		
MAF_DP1_06	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208712	MK205019	MK226965	Tr_S02	-	MK226812	MK227048		
MAF_DP1_07	1	-85.539	-151.150	Lecanora sp. 3	MK208713	-	-	Tr_A02	MK226847	MK226736	MK227049		
MAF_DP1_08	1	-85.539	-151.150	Lecanora fuscobrunnea Dodge & Baker	MK208714	MK205021	MK226966	Tr_A02	MK226848	MK226737	MK227050		
MAF_DP1_09	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208715	MK205022	-	Tr_S02	MK226849	MK226801	MK227051		
MAF_DP1_10	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208716	MK205023	MK226967	Tr_S02	MK226850	MK226802	MK227052		
MAF_DP1_11	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208717	MK205024	MK226968	Tr_S02	MK226851	MK226762	MK227053		
MAF_DP1_15	1	-85.539	-151.150	Lecidella greenii Ruprecht & Türk	MK208718	-	MK226969	Tr_A02	MK226852	MK226738	MK227054		
MAF_DP1_19	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208719	MK205025	MK226970	-	-	-	-		
MAF_DP1_20	1	-85.539	-151.150	Lecanora fuscobrunnea Dodge & Baker	MK208720	MK205026	MK226971	Tr_A02	MK226853	MK226739	MK227055		
MAF_DP1_22	1	-85.539	-151.150	Lecidella siplei Dodge & Baker	MK208721	-	-	Tr_A02	MK226854	MK226740	MK227056		
MAF_DP1_24	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208722	MK205027	MK226972	<i>Tr</i> _I01	MK226855	-	-		
MAF_DP1_25	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208723	MK205028	MK226973	Tr_A02	MK226856	MK226741	MK227057		
MAF_DP1_28	1	-85.539	-151.150	Lecanora fuscobrunnea Dodge & Baker	MK208724	MK205029	MK226974	Tr_S02	MK226857	MK226771	MK227058		
MAF_DP1_30	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208725	MK205030	-	Tr_S02	MK226858	MK226764	MK227059		
MAF_DP1_32	1	-85.539	-151.150	Lecanora fuscobrunnea Dodge & Baker	MK208726	MK205031	MK226975	Tr_A02	MK226859	MK226742	MK227060		
MAF_DP1_33	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208727	MK205032	MK226976	Tr_S02	MK226860	MK226765	MK227061		
MAF_DP1_34	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208728	MK205033	MK226977	Tr_S02	MK226861	MK226813	MK227062		
MAF_DP1_35	1	-85.539	-151.150	Lecidella siplei Dodge & Baker	MK208729	-	-	Tr_A02	MK226862	MK226743	MK227063		
MAF_DP1_36	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208730	MK205034	MK226978	Tr_S02	MK226863	MK226815	MK227064		
MAF_DP1_39	1	-85.539	-151.150	Lecidea cancriformis Dodge & Baker	MK208731	MK205035	MK226979	Tr_S02	-	MK226816	MK227065		
MAF_DP1_50	1	-85.539	-151.150	Lecanora fuscobrunnea Dodge & Baker	MK208732	MK205036	MK226981	Tr_A02	-	MK226745	MK227067		
MAF_DP1_51	1	-85.539	-151.150	Lecidella greenii Ruprecht & Türk	MK208733	MK205037	MK226982	Tr_A02	MK226865	MK226746	MK227068		
MAF_DP1_52	1	-85.539	-151.150	Carbonea sp. URm1	MK208734	MK205038	-	Tr_A02	MK226866	MK226747	MK227069		
MAF_DP1_54	1	-85.539	-151.150	Carbonea sp. 2	MK208735	-	MK226984	Tr_S02	MK226868	MK226772	MK227071		
MAF_DP1_57	1	-85.539	-151.150	Lecidella siplei Dodge & Baker	MK208736	-	-	Tr_A02	MK226869	-	MK227072		
MAF_GR1_29	3	-83.487	170.790	Lecidea cancriformis Dodge & Baker	MK208737	MK205039	MK226985	-	-	-	-		
MAF_GS1_12	2	-84.535	-174.954	Lecidea andersonii Filson	MK208738	MK205040	-	Tr_S02	MK226871	MK226763	MK227075		
MAF_GS1_13	2	-84.535	-174.954	Lecidella siplei Dodge & Baker	MK208739	-	-	Tr_A02	MK226872	-	MK227076		

				Mycobiont					ociated green mi	cro algae ( <i>Treb</i>	ouxia)
					A	ccession numb	oers		Ad	ccession numbe	ers
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	OTU ID	nrITS2	psbJ-L	COX2
MAF_GS1_44	2	-84.535	-174,954	Lecidea andersonii Filson	MK208740	MK205041	-	Tr_A02	MK226873	-	MK227077
MAF_GS1_45	2	-84.535	-174.954	Lecanora physciella (Darb.) Hertel	MK208741	-	MK226986	Tr_S02	MK226874	MK226766	MK227078
MAF_GS1_58	2	-84.535	-174.954	Lecidella siplei Dodge & Baker	MK208742	-	-	Tr_A02	MK226875	-	MK227079
MAF_GS1_60	2	-84.535	-174.954	Lecidella siplei Dodge & Baker	MK208743	-	-	Tr_A02	MK226876	-	-
MAF_GS1_61	2	-84.535	-174.954	Lecidea andersonii Filson	MK208744	MK205042	MK226987	Tr_A02	MK226877	MK226749	MK227080
MAF_GS1_62	2	-84.535	-174.954	Lecidella siplei Dodge & Baker	MK208745	-	-	Tr_A02	MK226878	MK226750	MK227081
MAF_GS1_64	2	-84.535	-174.954	Lecidella sp. nov2	MK208746	MK205043	MK226988	Tr_S02	MK226879	MK226775	MK227082
MAF_HS7_59	3	-83.806	172.262	Lecidella sp. nov2	MK208747	MK205044	MK226989	Tr_S02	MK226880	MK226810	MK227083
MAF_MG1_16	2	-84.559	-175.009	Lecidea andersonii Filson	MK208748	MK205045	MK226990	Tr_A02	MK226881	MK226751	MK227084
MAF_MG1_17	2	-84.559	-175.009	Lecidea andersonii Filson	MK208749	MK205046	-	Tr_A02	MK226882	MK226752	MK227085
MAF_MG1_23	2	-84.559	-175.009	Lecidea andersonii Filson	MK208750	MK205047	MK226991	Tr_A02	-	-	MK227086
MAF_MG1_47	2	-84.559	-175.009	Lecidea cancriformis Dodge & Baker	MK208751	-	-	Tr_l17	MK226883	-	-
MAF_MK1_03	3	-83.775	171.828	Lecanora fuscobrunnea Dodge & Baker	MK208752	MK205048	MK226992	Tr_A02	MK226884	MK226753	MK227087
MAF_MK1_18	3	-83.775	171.828	Lecidella sp. nov2	MK208753	-	-	Tr_S02	MK226886	MK226767	MK227089
MAF_MK1_21	3	-83.775	171.828	Lecidella sp. nov2	MK208754	MK205049	-	Tr_S02	MK226887	MK226788	MK227090
MAF_MK1_26	3	-83.775	171.828	Lecidea cancriformis Dodge & Baker	MK208755	MK205050	MK226993	Tr_S02	MK226888	MK226776	MK227091
MAF_MK1_27	3	-83.775	171.828	Lecidella sp. nov2	MK208756	MK205051	MK226994	Tr_S02	MK226889	MK226777	MK227092
MAF_MK1_31	3	-83.775	171.828	Lecidea cancriformis Dodge & Baker	MK208757	MK205052	MK226995	<i>Tr_</i> I01	MK226890	-	-
MAF_MK1_37	3	-83.775	171.828	Lecidea cancriformis Dodge & Baker	MK208758	MK205053	MK226996	<i>Tr_</i> I01	MK226891	-	-
MAF_MK1_38	3	-83.775	171.828	Lecidea cancriformis Dodge & Baker	MK208759	MK205054	-	Tr_S02	MK226892	MK226778	MK227093
MAF_MK1_40	3	-83.775	171.828	Lecanora fuscobrunnea Dodge & Baker	MK208760	MK205055	MK226997	Tr_A02	MK226893	MK226754	MK227094
MAF_MK1_43	3	-83.775	171.828	Lecidella sp. nov2	MK208761	MK205056	-	Tr_S02	MK226894	MK226779	MK227095
MAF_MK1_48	3	-83.775	171.828	Lecidea cancriformis Dodge & Baker	MK208762	MK205057	MK226998	Tr_S02	MK226895	MK226780	MK227096
MAF_MK1_49	3	-83.775	171.828	Lecidella sp. nov2	MK208763	MK205058	MK226999	Tr_S02	MK226896	MK226799	MK227097
MAF_MK1_55	3	-83.775	171.828	Lecidea cancriformis Dodge & Baker	MK208764	MK205059	MK227000	-	-	-	-
MAF_MK1_56	3	-83.775	171.828	Lecanora fuscobrunnea Dodge & Baker	MK208765	MK205060	MK227001	Tr_S02	MK226897	MK226781	MK227098
MAF_MK1_63	3	-83.775	171.828	Lecanora physciella (Darb.) Hertel	MK208766	MK205061	MK227002	Tr_S02	MK226898	MK226789	MK227099
MAF_Sancho	3	-83.761	172.755	Lecidea cancriformis Dodge & Baker	GU074439	GU074489	MK227003	Tr_A04a	JN204838	-	-
MAF_Sancho	3	-83.761	172.755	Carbonea sp. 2	MK208767	-	-	Tr_A04a	JN204839	-	-
T33335	3	-83.803	172.207	Lecidella sp. nov2	MK208768	MK205062	-	<i>Tr</i> _S18	MK226899	-	-
T33338	3	-83.803	172.207	Lecidella sp. nov2	MK208769	MK205063	-	Tr_S02	MK226900	-	-
T33346	3	-83.803	172.207	Lecanora physciella (Darb.) Hertel	JN873878	-	-	-	-	-	-
T33348	3	-83.803	172.207	Lecidea cancriformis Dodge & Baker	-	MK205064	-	-	-	-	-

				Mycobiont				Ass	ociated green mi	icro algae ( <i>Treb</i>	ouxia)
					A	ccession numb	bers		Ad	ccession numbe	ers
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	OTU ID	nrITS2	psbJ-L	COX2
T33446	3	-83.828	172.749	Lecidea cancriformis Dodge & Baker	-	MK205065	-	<i>Tr_</i> S18	MK226901	-	-
T33449	3	-83.761	172.755	Lecidella siplei Dodge & Baker	JN873897	-	-	Tr_A02	JN204729	-	-
T33456	3	-83.761	172.755	Lecidea andersonii Filson	-	MK205066	MK227004	Tr_A02	-	-	MK227100
T33457	3	-83.761	172.755	Lecidella siplei Dodge & Baker	JN873898	-	-	<i>Tr_</i> A02	JN204731	-	-
T35540	4a	-79.842	159.363	Lecidella siplei Dodge & Baker	MK208770	-	-	-	-	-	-
T35544	4a	-79.838	159.341	Lecanora fuscobrunnea Dodge & Baker	JN873873	-	-	-	-	-	-
T35559	4a	-79.838	159.221	Lecidea cancriformis Dodge & Baker	MK208771	MK205067	MK227005	<i>Tr</i> _S18	-	MK285375	-
T35604	4a	-79.836	159.317	Lecidea cancriformis Dodge & Baker	EU257671	GU074480	MK227006	<i>Tr</i> _S18	JN204749	-	-
T35620	4a	-79.835	159.385	Lecidea cancriformis Dodge & Baker	EU257672	-	-	<i>Tr</i> _I01	JN204750	-	-
T35622	4a	-79.835	159.392	Lecidea cancriformis Dodge & Baker	MK208772	MK205068	MK227007	-	-	-	-
T35647	4a	-79.851	159.341	Carbonea sp. 2	JN873866	-	-	Tr_S02	JN204751	-	-
T35650	4a	-79.851	159.341	Carbonea sp. 2	JN873867	-	MK227008		JN204752	-	-
T35662	4a	-79.840	159.332	Lecidea cancriformis Dodge & Baker	EU257673	-	-	Tr 101	JN204753	-	-
T35664	4a	-79.851	159.341	Lecanora fuscobrunnea Dodge & Baker	JN873874	-	-	-	-	-	-
T35686	4a	-79.842	159.363	Carbonea sp. 2	JN873868	-	-	-	-	-	-
T42988	4b	-79.889	156.764	Lecidea cancriformis Dodge & Baker	GU074435	GU074481	MK227009	Tr S18	MK226902	MK226818	MK227101
T42990	4b	-79.926	156.890	Lecidea cancriformis Dodge & Baker	GU170841	-	MK227010		JN204770	MK226819	MK227102
T42991	4b	-79.917	156.759	Lecidea cancriformis Dodge & Baker	GU170842	MK205069	MK227011	<i>Tr</i> S18	MK226903	_	-
T42992	4b	-79.917	156.751	Lecidea cancriformis Dodge & Baker	GU074436	GU074482	MK227012	<i>Tr</i> _S18	JN204771	MK226820	MK227103
T42994	4b	-79.879	157.539	Lecanora fuscobrunnea Dodge & Baker	GU170839	MK205070	MK227013		MK226904	-	-
T44625	4a	-79.868	159.360	Lecanora fuscobrunnea Dodge & Baker	MK208773	MK205071	MK227014	 TrA02	MK226905	MK226755	MK227104
T44626	4a	-79.882	159.361	Carbonea sp. URm1	JN873865	-	MK227015	 Tr101	JN204797	-	-
T44628	4a	-79.865	159.352	Lecanora fuscobrunnea Dodge & Baker	JN873875	MK205072	MK227016	Tr_S02	-	MK226804	MK227105
T44632	4a	-79.868	159.352	Lecanora fuscobrunnea Dodge & Baker	MK208774	MK205073	MK227017	Tr_101	JN204800	-	-
T44633	4a	-79.868	159.358	Lecidea cancriformis Dodge & Baker	MK208775	MK205074	-		-	MK226817	MK227106
T44634	4a	-79.869	159.341	Lecidea cancriformis Dodge & Baker	GU074434	GU074486	MK227018		JN204801	KF907601	-
T44636	4a	-79.869	159.358	Lecidella greenii Ruprecht & Türk	MK208776	-	-	Tr_A02	MK226906	_	-
T44638	4a	-79.869	159.358	Lecidea cancriformis Dodge & Baker	-	MK205075	-	<i>Tr_</i> S18	MK226907	MK226827	MK227107
T44640	4a	-79.869	159.358	Lecidea cancriformis Dodge & Baker	MK208777	MK205076	MK227019	Tr S02	MK226908	MK226809	MK227108
T44641	4a	-79.866	159.365	Carbonea sp. 2	JN873871	-	MK227020	Tr A04a	JN204803	KF907602	-
T44643	4a	-79.859	159.238	Lecidea cancriformis Dodge & Baker	MK208778	MK205077	-	<i>Tr_</i> S18	-	MK226831	MK227109
T44645	4a	-79.857	159.283	Lecidea cancriformis Dodge & Baker	MK208779	MK205078	MK227021	Tr_S02	MK226909	MK226790	MK227110
T44646	4a	-79.857	159.283	Lecidea cancriformis Dodge & Baker	MK208780	MK205079	-	Tr_S02	MK226910	MK226793	MK227111

				Mycobiont					ociated green mi	cro algae ( <i>Treb</i>	ouxia)
					A	ccession numb	bers		Ac	ccession numbe	ers
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	OTU ID	nrITS2	psbJ-L	COX2
T44647	4a	-79.858	159.288	Lecidea cancriformis Dodge & Baker	MK208781	MK205080	-	Tr_S02	MK226911	MK226794	MK227112
T44648	4a	-79.858	159.288	Lecidea cancriformis Dodge & Baker	MK208782	MK205081	-	-	-	-	-
T44649	4a	-79.858	159.288	Lecidea cancriformis Dodge & Baker	MK208783	MK205082	-	Tr_S02	MK226912	MK226791	MK227113
T44650	4a	-79.858	159.288	Lecanora fuscobrunnea Dodge & Baker	MK208784	MK205083	MK227022	<i>Tr_</i> I01	MK285376	-	-
T44651	4a	-79.858	159.288	Lecidea cancriformis Dodge & Baker	MK208785	MK205084	-	Tr_S02	MK226913	MK226795	MK227114
T44652	4a	-79.858	159.288	Lecidea cancriformis Dodge & Baker	MK208786	MK205085	-	Tr_S02	MK226914	MK226796	MK227115
T44655	4a	-79.874	159.339	Carbonea sp. 2	MK208787	-	MK227023	Tr_S02	MK226915	MK226805	MK227116
T44656	4a	-79.863	159.373	Lecidea cancriformis Dodge & Baker	MK208788	MK205086	-	Tr_S02	MK226916	MK226797	MK227117
T44657	4a	-79.863	159.373	Lecanora fuscobrunnea Dodge & Baker	JN873876	MK205087	MK227024	Tr_S02	JN204804	MK226806	MK227118
T44659	4a	-79.864	159.368	Lecidea cancriformis Dodge & Baker	MK208789	MK205088	-	Tr_S02	MK226917	MK226798	MK227119
T44665	4a	-79.869	159.345	Lecidea cancriformis Dodge & Baker	MK208790	MK205089	-	Tr_S02	MK226918	MK226814	MK227120
T44666	4a	-79.869	159.345	Lecidea cancriformis Dodge & Baker	MK208791	MK205090	-	-	-	-	-
T44667	4a	-79.869	159.345	Lecidea cancriformis Dodge & Baker	MK208792	MK205091	MK227025	Tr_S02	MK226919	MK226782	MK227121
T44669	4a	-79.863	159.378	Lecidea cancriformis Dodge & Baker	MK208793	-	-		MK226920	MK226768	MK227122
T44670	4a	-79.863	159.378	Lecidea cancriformis Dodge & Baker	MK208794	MK205092	-		MK226921	-	-
T44674	4a	-79.877	159.326	Lecidea cancriformis Dodge & Baker	MK208795	-	-	Tr_S02	MK226923	MK226783	MK227124
T44675	4a	-79.877	159.326	Lecanora fuscobrunnea Dodge & Baker	-	MK205093	-	-	JN204806	-	-
T44676	4a	-79.883	159.348	Lecidea cancriformis Dodge & Baker	MK208796	MK205094	MK227026	Tr_S18	MK226924	MK226832	MK227125
T44677	4a	-79.883	159.348	Lecidea cancriformis Dodge & Baker	MK208797	MK205095	MK227027	Tr_A02	MK226925	MK226756	MK227126
T44679	4a	-79.877	159.331	Lecidea cancriformis Dodge & Baker	MK208798	MK205096	-	Tr_S02	MK226927	MK226769	MK227128
T44687	4a	-79.863	159.379	Lecanora fuscobrunnea Dodge & Baker	MK208799	-	-	-	-	-	MK227130
T44688	4a	-79.863	159.382	Lecanora fuscobrunnea Dodge & Baker	JN873877	MK205097	MK227028	Tr_S02	JN204807	MK226807	MK227131
T44690	4a	-79.863	159.382	Lecidea cancriformis Dodge & Baker	MK208800	MK205098	-	Tr_S02	MK226930	MK226786	MK227133
T44692	4a	-79.866	159.366	Lecidea <i>cancriformis</i> Dodge & Baker	MK208801	MK205099	MK227029		JN204809	KF907603	_
T44694	4b	-79.755	158.503	Lecidea cancriformis Dodge & Baker	MK208802	MK205100	-	 Tr101	MK226931	-	_
T44695	4b	-79.761	158.503	Lecanora fuscobrunnea Dodge & Baker	MK208803	MK205101	MK227030	-	-	-	_
T44697	4b	-79.758	158.598	Lecanora fuscobrunnea Dodge & Baker	MK208804	MK205102	-	Tr_S02	MK226932	MK226770	MK227134
T44698	4b	-79.755	158.627	Lecidea cancriformis Dodge & Baker	MK208805	MK205103	MK227031	Tr_S18	MK226933	MK226825	MK227135
T44699	4b	-79.762	158.636	Lecanora fuscobrunnea Dodge & Baker	MK208806	MK205104	MK227032	Tr_A02	MK226934	MK226758	MK227136
T44700	4b	-79.762	158.637	Lecanora fuscobrunnea Dodge & Baker	MK208807	MK205105	MK227033	Tr_101	MK226935	_	-
T44701	4b	-79.757	158.608	Lecidea cancriformis Dodge & Baker	MK208808	MK205106	-	Tr_S18	MK226936	MK226833	MK227137
T44702	4b	-79.756	158.610	Lecanora fuscobrunnea Dodge & Baker	MK208809	MK205107	MK227034	Tr A02	MK226937	MK226759	MK227138
T44703	4b	-79.756	158.614	Lecidea cancriformis Dodge & Baker	-	MK205108	MK227035	<i>Tr_</i> S18	MK226938	MK226828	MK227139

				Мус		Associated green micro algae (Trebouxia)					
					A	Accession numbers			Ac	ers	
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	OTU ID	nrITS2	psbJ-L	COX2
T44704	4b	-79.756	158.617	Lecidea cancriformis Dodge & Baker	-	MK205109	-	<i>Tr_</i> S18	MK226939	MK226829	MK227140
T44705	4b	-79.756	158.611	Lecanora fuscobrunnea Dodge & Baker	MK208810	-	-	Tr_S02	MK226940	MK226811	MK227141
T44707	4b	-79.758	158.598	Lecidea cancriformis Dodge & Baker	MK208811	MK205110	MK227036	<i>Tr_</i> S18	MK226941	MK226834	MK227142
T44708	4b	-79.753	158.549	Lecidea cancriformis Dodge & Baker	MK208812	MK205111	-	<i>Tr_</i> S18	-	MK226823	MK227143
T44709	4b	-79.753	158.541	Lecidea cancriformis Dodge & Baker	MK208813	MK205112	MK227037	<i>Tr_</i> S18	MK226942	MK226826	MK227144
T44712	4b	-79.759	158.507	Lecidea cancriformis Dodge & Baker	GU074438	GU074487	MK227038	<i>Tr_</i> S18	JN204811	-	-
T44713	4b	-79.759	158.511	Lecidea cancriformis Dodge & Baker	MK208814	-	-	<i>Tr_</i> S18	MK226945	MK226836	MK227147
T44714	4b	-79.763	158.497	Lecidea cancriformis Dodge & Baker	MK208815	MK205113	-	<i>Tr_</i> S18	MK226946	MK226830	MK227148
T44715	4b	-79.758	158.602	Lecidea cancriformis Dodge & Baker	-	MK205114	MK227039	<i>Tr_</i> S18	MK226947	MK226837	MK227149
T44716	4b	-79.758	158.606	Lecidea cancriformis Dodge & Baker	-	MK205115	MK227040	<i>Tr_</i> S18	MK226948	MK226841	MK227150
T44717	4b	-79.755	158.620	Lecanora fuscobrunnea Dodge & Baker	MK208816	MK205116	MK227041	Tr_S02	MK226949	MK226787	MK227151
T44719	4b	-79.924	156.814	Lecanora fuscobrunnea Dodge & Baker	MK208817	MK205117	MK227042	<i>Tr_</i> S18	MK226950	MK226821	MK227152
T44720	4b	-79.949	156.789	Lecanora fuscobrunnea Dodge & Baker	GU170840	MK205118	MK227043	Tr_S02	MK226951	MK226808	MK227153
T44721	4b	-79.754	158.636	Lecidea cancriformis Dodge & Baker	MK208818	MK205119	MK227044	<i>Tr_</i> S18	MK226952	MK226842	MK227154
T44723	4b	-79.878	157.527	Lecidea cancriformis Dodge & Baker	MK208819	MK205120	-	<i>Tr_</i> S18	MK226953	MK226838	MK227155
T44727	4b	-79.892	157.524	Lecidea cancriformis Dodge & Baker	GU074437	GU074488	-	<i>Tr_</i> S18	JN204812	-	-
T44787	4b	-79.929	156.705	Lecidea cancriformis Dodge & Baker	MK208820	MK205122	-	<i>Tr_</i> S18	MK226955	MK226839	MK227157

Supplementary Table S3. Additional samples taken from Perez-Ortega et al. (2012) (Perez-Ortega et al. 2012) and used in this study, with information on collecting localities and Genbank accession numbers.

				Mycobiont	Associated green micro algae (Trebouxia)		
Laboratory code	Area	Latitude	Longitude	Species name	Accession numbers nrITS	OTU ID	Accession numbers nrITS
s106	5	-78.113	163.782	Lecanora sp. 2	JX036037	Tr_A02	JX036159
s113	5	-78.114	163.854	Lecidea cancriformis Dodge & Baker	JX036044	Tr_S15	JX036166
s114	5	-78.114	163.854	Lecidea cancriformis Dodge & Baker	JX036045	Tr_A02	JX036167
s115	5	-78.066	163.870	Lecidella greenii Ruprecht & Türk	JX036046	Tr_A02	JX036168
s120	5	-78.083	163.768	Lecidea polypycnidophora Ruprecht & Türk	JX036051	Tr_A02	JX036172
s121	5	-78.024	163.900	Carbonea vorticosa (Flörke) Hertel	JX036052	Tr A02	JX036173
s122	5	-78.110	163.787	Carbonea vorticosa (Flörke) Hertel	JX036053	Tr_A02	JX036174
s123	5	-78.110	163.787	Carbonea vorticosa (Flörke) Hertel	JX036054	Tr_A02	JX036175

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				Mycobiont	Associated green micro algae (Trebouxia)		
Laboratory code	Area	Latitude	Longitude	Species name	Accession numbers nrITS	OTU ID	Accession numbers nrITS
s124	5	-78.057	163.844	Rhizoplaca macleanii (Dodge) Castello	JX036055	Tr_A02	JX036176
s125	5	-78.057	163.844	Rhizoplaca macleanii (Dodge) Castello	JX036056	Tr_A02	JX036177
s171	5	-78.036	163.837	Lecanora sp. 2	JX036076	Tr_A02	JX036197
s173	5	-78.114	163.854	Lecanora sp. 2	JX036078	<i>Tr_</i> A02	JX036199
s175	5	-78.033	163.849	Lecanora sp. 2	JX036080	Tr_A02	JX036201
s179	5	-78.063	163.809	Lecanora sp. 2	JX036084	Tr_A02	JX036205
s181	5	-78.111	163.858	Lecidella greenii Ruprecht & Türk	JX036086	Tr_A02	JX036207
s190	5	-78.025	163.899	Lecanora sp. 3	JX036095	Tr_A02	JX036216
s191	5	-78.025	163.900	Lecanora sp. 3	JX036096	Tr_A02	JX036217
s192	5	-78.030	163.834	Lecidella greenii Ruprecht & Türk	JX036097	Tr_A02	JX036218
s197	5	-78.061	163.791	Rhizoplaca macleanii (Dodge) Castello	JX036101	Tr_A02	JX036222
s198	5	-78.061	163.791	Rhizoplaca macleanii (Dodge) Castello	JX036102	Tr_A02	JX036223
s201	5	-78.024	163.900	Rhizoplaca macleanii (Dodge) Castello	JX036105	Tr_A02	JX036226
s202	5	-78.027	163.839	Lecanora cf. mons-nivis Darbishire	JX036106	Tr_A02	JX036227
s203	5	-78.027	163.839	Lecidella greenii Ruprecht & Türk	JX036107	Tr A02	JX036228
s205	5	-78.068	163.861	Rhizoplaca macleanii (Dodge) Castello	JX036108	Tr_A02	JX036229
s206	5	-78.068	163.861	Rhizoplaca macleanii (Dodge) Castello	JX036109	Tr_A02	JX036230
s207	5	-78.068	163.861	Rhizoplaca macleanii (Dodge) Castello	JX036110	Tr_A02	JX036231
s208	5	-78.068	163.861	Rhizoplaca macleanii (Dodge) Castello	JX036111	Tr_A02	JX036232
s209	5	-78.068	163.861	Rhizoplaca macleanii (Dodge) Castello	JX036112	Tr_A02	JX036233
s212	5	-78.034	163.845	Rhizoplaca macleanii (Dodge) Castello	JX036115	Tr_A02	JX036236
s213	5	-78.034	163.845	Rhizoplaca macleanii (Dodge) Castello	JX036116	Tr A02	JX036237
s214	5	-78.034	163.845	Rhizoplaca macleanii (Dodge) Castello	JX036117	Tr_A02	JX036238
s215	5	-78.070	163.711	Lecanora fuscobrunnea Dodge & Baker	JX036118	Tr_A02	JX036239
s230	5	-78.034	163.845	Lecidella greenii Ruprecht & Türk	JX036133	Tr_A02	JX036252
s232	5	-78.034	163.845	Rhizoplaca macleanii (Dodge) Castello	JX036135	Tr_A02	JX036254
s233	5	-78.034	163.845	Rhizoplaca macleanii (Dodge) Castello	JX036136	Tr_A02	JX036255
s235	5	-78.113	163.778	Rhizoplaca macleanii (Dodge) Castello	JX036138	Tr_A02	JX036257
s236	5	-78.113	163.778	Rhizoplaca macleanii (Dodge) Castello	JX036139	Tr A02	JX036258
s237	5	-78.113	163.778	Rhizoplaca macleanii (Dodge) Castello	JX036140	<i>Tr</i> _A02	JX036259
s266	5	-78.075	163.791	Lecidella greenii Ruprecht & Türk	JX036141	<i>Tr</i> _A02	JX036260
s271	5	-78.047	164.104	Rhizoplaca macleanii (Dodge) Castello	JX036145	Tr_A02	JX036264
s272	5	-78.035	163.978	Rhizoplaca macleanii (Dodge) Castello	JX036146		JX036265
s273	5	-78.030	163.949	Rhizoplaca macleanii (Dodge) Castello	JX036147		JX036266
s274	5	-78.036	163.990	Rhizoplaca macleanii (Dodge) Castello	JX036148	Tr_A02	JX036267

				Mycobiont	Associated green micro algae (Trebouxia)		
Laboratory code	Area	Latitude	Longitude	Species name	Accession numbers nrITS	OTU ID	Accession numbers nrITS
s300	5	-78.030	163.834	Lecidella greenii Ruprecht & Türk	JX036150	Tr_A02	JX036269
s301	5	-78.030	163.834	Lecidella greenii Ruprecht & Türk	JX036151	Tr_A02	JX036270
s95	5	-78.024	163.900	Rhizoplaca macleanii (Dodge) Castello	JX036152	Tr_A02	JX036271

Supplementary Table S4. Additional samples taken from Wagner et al. (2020) (Wagner et al. 2020) and used in this study, with information on collecting localities and Genbank accession numbers.

			Mycobiont					Ass	ociated green mic	ro algae ( <i>Treb</i>	ouxia)	
					Ac	Accession numbers			Ac	Accession numbers		
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	OTU ID	nrITS	psbJ-L	COX2	
T46643	5	-78.031	163.865	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970698	-	-	
T46647b	5	-78.033	163.898	Rhizoplaca macleanii (Dodge) Castello	MK970665	MN023039	MN023053	Tr_A02	MK970698	-	-	
T46651	5	-78.028	163.851	Carbonea vorticosa (Flörke) Hertel	MK970656	-	-	-	-	-	-	
T46659	5	-78.023	163.903	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023055	Tr_A02	MK970698	-	-	
T46672	5	-78.024	163.898	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970698	-	-	
T46673	5	-78.027	163.851	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970699	-	-	
T46676	5	-78.028	163.851	Lecidella greenii Ruprecht & Türk	MK970671	-	-	Tr_A02	MK970699	-	-	
T46677	5	-78.027	163.848	Lecidea cancriformis Dodge & Baker	MK970681	-	-	Tr_A02	MK970696	-	-	
T46678	5	-78.028	163.850	Rhizoplaca macleanii (Dodge) Castello	MK970669	-	-	Tr_A02	MK970699	-	-	
T46679	5	-78.036	163.971	Rhizoplaca macleanii (Dodge) Castello	MK970664	MN023039	-	Tr_A02	MK970696	-	-	
T46680	5	-78.036	163.971	Lecidea polypycnidophora Ruprecht & Türk	MK970663	MN023043	MN023053	Tr_A02	MK970699	-	-	
T46681	5	-78.032	163.951	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970698	-	-	
T46684	5	-78.044	163.986	Lecidea cancriformis Dodge & Baker	MK970677	-	-	Tr_S02	MK970693	-	-	
T46685	5	-78.044	163.986	Lecidea cancriformis Dodge & Baker	MK970677	-	MN023056	Tr_S02	MK970693	-	MN023030	
T46701	5	-78.020	163.805	Rhizoplaca macleanii (Dodge) Castello	MK970663	MN023034	-	Tr_A02	MK970698	-	-	
T46706	5	-78.028	163.821	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023054	Tr_A02	MK970698	-	-	
T46710	5	-78.073	163.717	Rhizoplaca macleanii (Dodge) Castello	MK970666	-	-	Tr_A02	MK970698	-	-	
T46713	5	-78.028	163.843	Lecidea polypycnidophora Ruprecht & Türk	MK970663	MN023043	MN023061	Tr_A02	MK970698	-	-	
T46716	5	-78.040	163.802	Lecidea polypycnidophora Ruprecht & Türk	MK970663	MN023043	MN023061	Tr_A02	MK970698	-	-	
T46717	5	-78.040	163.806	Lecidea polypycnidophora Ruprecht & Türk	MK970663	-	-	Tr_A02	MK970698	-	-	
T46718	5	-78.040	163.807	Carbonea vorticosa (Flörke) Hertel	MK970656	-	-	Tr_A02	MK970698	-	-	
T46719	5	-78.038	163.804	Carbonea sp. URm1	MK970657	-	-	Tr_A02	MK970698	-	-	

				Mycobiont				Ass	sociated green mic	ro algae ( <i>Treb</i>	ouxia)
					Accession numbers			Ac	cession numb	ers	
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	otu id	nrITS	psbJ-L	COX2
T48769	5	-78.126	163.700	Lecidella greenii Ruprecht & Türk	MK970671	-	-	Tr_A02	MK970699	-	-
T48770	5	-78.127	163.690	Lecanora sp. 3	MK970659	-	-	Tr_A02	MK970701	-	-
T48773	5	-78.127	163.674	Lecidea polypycnidophora Ruprecht & Türk	MK970663	MN023043	-	Tr_A02	MK970699	MN023065	-
T48774	5	-78.135	163.626	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970698	-	-
T48776	5	-78.165	163.753	Lecidea cancriformis Dodge & Baker	MK970679	MN023046	MN023057		MK970702	-	-
T48777	5	-78.166	163.755	Lecidea cancriformis Dodge & Baker	MK970679	MN023046	MN023058		MK970692	-	MN023031
T48778a	5	-78.149	163.769	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-		MK970698	-	-
T48779	5	-78.164	163.755	Rhizoplaca macleanii (Dodge) Castello	MK970664	MN023039	-		MK970696	-	-
T48781	5	-78.128	163.620	Lecidea sp. 6	MK620097	-	-		MK970699	-	-
T48782	5	-78.123	163.642	Lecidea cancriformis Dodge & Baker	MK970679	MN023046	-	Tr_S18	MK970695	MN023070	MN023032
T48784	5	-78.121	163.683	Lecidea polypycnidophora Ruprecht & Türk	MK970663	MN023043	-		MK970699	MN023065	-
T48785	5	-78.133	163.666	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023054		MK970692	-	-
T48787	5	-78.127	163.678	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023054		MK970699	-	-
T48788	5	-78.120	163.684	Carbonea vorticosa (Flörke) Hertel	MK970656	MN023033	MN023050		MK970699	-	-
T48789	5	-78.152	163.739	Carbonea sp. 2	MK970654	-	MN023051	Tr_S15	MK970692	-	MN023031
T48790a	5	-78.120	163.682	Carbonea vorticosa (Flörke) Hertel	MK970656	MN023033	-		MK970699	-	-
T48790b	5	-78.120	163.682	Lecanora sp. 3	MK970659	-	-		MK970703	-	-
T48791a	5	-78.120	163.686	Lecidella greenii Ruprecht & Türk	MK970671	-	-		MK970699	-	-
T48793a	5	-78.153	163.731	Lecidea cancriformis Dodge & Baker	MK970677	-	MN023056		MK970692	-	MN023031
T48793b	5	-78.153	163.731	Carbonea sp. 2	MK970654	-	-		MK970692	-	MN023031
T48794b	5	-78.151	163.735	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970698	-	-
T48795a	5	-78.161	163.714	Rhizoplaca macleanii (Dodge) Castello	MK970668	-	-		MK970698	-	-
T48797	5	-78.156	163.689	Rhizoplaca macleanii (Dodge) Castello	MK970667	-	-	Tr_A02	MK970698	-	-
T48798	5	-78.150	163.736	Rhizoplaca macleanii (Dodge) Castello	MK970667	-	-		MK970698	-	-
T48799b	5	-78.145	163.620	Lecidea cancriformis Dodge & Baker	MK970679	-	-	Tr_A02	MK970698	-	-
T48799c	5	-78.145	163.620	Lecidea cancriformis Dodge & Baker	MK970677	-	-	Tr_S15	MK970692	-	-
T48800	5	-78.146	163.631	Lecanora sp. 2	MK970662	MN023036	MN023052	Tr_A02	MK970698	-	-
T48801a	5	-78.144	163.626	Lecanora sp. 3	MK970659	-	-	Tr_A02	MK970699	-	-
T48801c	5	-78.144	163.626	Lecidella greenii Ruprecht & Türk	MK970671	MN023040	-	Tr_A02	MK970699	-	-
T48803b	5	-78.148	163.630	Carbonea sp. URm1	MK970657	MN023034	-	Tr_A02	MK970699	-	-
T48804	5	-78.148	163.630	Rhizoplaca macleanii (Dodge) Castello	MK970670	-	MN023053	Tr_A02	MK970699	-	-
T48805	5	-78.144	163.626	Lecidella greenii Ruprecht & Türk	MK970671	-	-	Tr_A02	MK970698	-	-
T48806	5	-78.142	163.628	Carbonea sp. URm1	MK970657	MN023034	-	Tr_A02	MK970699	-	-

				Mycobiont				Ass	ociated green mic	ro algae ( <i>Treb</i>	ouxia)
					Ad	Accession numbers			Accession numbers		ers
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	otu id	nrITS	psbJ-L	COX2
T48807	5	-78.141	163.631	Lecidea andersonii Filson	MK970673	MN023042	MN023060	Tr_A02	MK970702	-	-
T48809	5	-78.148	163.657	Lecidella greenii Ruprecht & Türk	MK970671	-	-	<i>Tr_</i> A02	MK970699	-	-
T48811a	5	-78.142	163.657	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023055	Tr_A02	MK970698	-	-
T48812a	5	-78.138	163.619	Lecidella greenii Ruprecht & Türk	MK970671	-	-	Tr_A02	MK970699	-	-
T48812b	5	-78.138	163.619	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023055	Tr_A02	MK970699	-	-
T48813	5	-78.142	163.657	Lecidea UCR1	MK970675	MN023044	-	Tr_A02	MK970699	-	-
T48817a	5	-78.113	163.785	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970698	-	-
T48817b	5	-78.113	163.785	Carbonea sp. 2	MK970655	-	-	Tr_A02	MK970702	-	-
T48820	5	-78.114	163.780	Carbonea sp. 2	MK970654	-	-	Tr_A02	MK970698	-	-
T48821	5	-78.114	163.779	Rhizoplaca macleanii (Dodge) Castello	MK970665	-	-	Tr_A02	MK970698	-	-
T48823a	5	-78.098	163.710	Carbonea vorticosa (Flörke) Hertel	MK970656	-	-	-	-	-	-
T48825	5	-78.097	163.691	Carbonea sp. 2	MK970654	MN023035	-	Tr A02	MK970699	-	-
T48826	5	-78.097	163.717	Lecanora fuscobrunnea Dodge & Baker	MK970661	-	-		MK970694	MN023068	-
T48828a	5	-78.110	163.858	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023054	-	-	-	-
T48828b	5	-78.110	163.858	Lecidella greenii Ruprecht & Türk	MK970671	-	-	Tr_A02	MK970698	-	-
T48829	5	-78.111	163.858	Lecidella greenii Ruprecht & Türk	MK970671	-	-		MK970699	-	-
T48831	5	-78.111	163.858	Carbonea vorticosa (Flörke) Hertel	MK970656	MN023033	-		MK970699	-	-
T48832	5	-78.112	163.824	Carbonea sp. 2	MK970654	-	MN023051		MK970702	-	-
T48836	5	-78.099	163.778	Lecanora cf. mons-nivis Darbishire	MK970658	-	-		MK970699	-	-
T48837	5	-78.098	163.777	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023055		MK970699	-	-
T48839	5	-78.114	163.854	Lecidella greenii Ruprecht & Türk	MK970671	-	-	Tr_A02	MK970699	-	-
T48841a	5	-78.068	163.861	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-		MK970698	-	-
T48841b	5	-78.068	163.861	Carbonea vorticosa (Flörke) Hertel	MK970656	-	MN023050	Tr A02	MK970698	-	-
T48843a	5	-78.066	163.870	Lecidea cancriformis Dodge & Baker	MK970677	-	-		MK970693	-	-
T48843c	5	-78.066	163.870	Lecidea cancriformis Dodge & Baker	MK970678	-	-	-	-	-	-
T48843d	5	-78.066	163.870	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970699	-	-
T48843e	5	-78.066	163.870	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970702	-	-
T48844	5	-78.067	163.863	Lecidella greenii Ruprecht & Türk	MK970671	-	-	Tr_A02	MK970698	-	-
T48851	5	-78.074	163.793	Lecanora fuscobrunnea Dodge & Baker	MK970660	MN023037	-	Tr A02	MK970698	-	-
T48855b	5	-78.036	163.837	Lecidea cancriformis Dodge & Baker	MK970680	MN023046	MN023059	Tr S15	MK970692	-	MN023031
T48857a	5	-78.036	163.827	Lecidea sp. 6	MK970684	MN023045	-	Tr_A02	MK970699	-	-
T48858	5	-78.025	163.975	Lecidella greenii Ruprecht & Türk	MK970671	-	-	-	-	-	-
T48859	5	-78.025	163.986	Lecidea polypycnidophora Ruprecht & Türk	MK970663	-	MN023061	Tr_A02	MK970698	-	-

				Mycobiont				Associated green micro algae (Trebouxia)				
					Ac	Accession numbers			Ac	Accession numbers		
Voucher ID	Area	Latitude	Longitude	Species name	nrITS	mtSSU	RPB1	otu id	nrITS	psbJ-L	COX2	
T48860b	5	-78.036	163.836	Lecidea sp. 6	MK970684	MN023045	MN023064	Tr_A02	MK970699	-	-	
T48861	5	-78.024	163.892	Lecanora sp. 3	MK970659	-	-	Tr_A02	MK970699	-	-	
T48862	5	-78.037	163.978	Lecidea cancriformis Dodge & Baker	MK970682	-	-	-	-	-	-	
T48864	5	-78.039	163.989	Lecidea cancriformis Dodge & Baker	MK970679	-	-	<i>Tr_</i> A02	MK970702	-	-	
T48865a	5	-78.036	163.990	Rhizoplaca macleanii (Dodge) Castello	MK970666	-	-	Tr_A02	MK970698	MN023067	-	
T48867a	5	-78.043	164.104	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	<i>Tr_</i> A02	MK970696	-	-	
T48867c	5	-78.043	164.104	Rhizoplaca macleanii (Dodge) Castello	MK970666	-	-	<i>Tr_</i> A02	MK970702	-	-	
T48869	5	-78.037	163.978	Lecidella greenii Ruprecht & Türk	MK970671	-	-	-	-	-	-	
T48872	5	-78.030	163.951	Lecidella greenii Ruprecht & Türk	MK970671	-	-	-	-	-	-	
T48873	5	-78.024	163.893	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023054	Tr_A02	MK970698	-	-	
T48874	5	-78.024	163.899	Lecidea UCR1	MK970676	MN023044	MN023062	Tr_A02	MK970697	MN023066	-	
T48875	5	-78.024	163.900	Lecidea lapicida (Ach.) Ach. subsp.	MK970683	-	-	Tr_A02	MK970699	-	-	
T48876a	5	-78.024	163.900	Lecidella greenii Ruprecht & Türk	MK970671	MN023040	-	Tr_A02	MK970698	-	-	
T48876b	5	-78.024	163.900	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023055	Tr_A02	MK970699	-	-	
T48877	5	-78.024	163.900	Carbonea vorticosa (Flörke) Hertel	MK970656	-	MN023050	Tr_A02	MK970698	-	-	
T48879	5	-78.057	163.747	Lecidella greenii Ruprecht & Türk	MK970671	-	MN023055	-	-	-	-	
T48880	5	-78.058	163.740	Lecidea polypycnidophora Ruprecht & Türk	MK970674	-	-	-	-	-	-	
T48881b	5	-78.061	163.791	Rhizoplaca macleanii (Dodge) Castello	MK970666	-	-	<i>Tr_</i> A02	MK970699	-	-	
T48882	5	-78.057	163.817	Lecidea polypycnidophora Ruprecht & Türk	MK970663	-	-	<i>Tr_</i> A02	MK970699	MN023065	-	
T48883b	5	-78.058	163.847	Lecidea sp. 5	MK620099	-	MN023063	Tr_A02	MK970700	-	-	
T48885	5	-78.057	163.844	Rhizoplaca macleanii (Dodge) Castello	MK970666	-	-	Tr_A02	MK970698	-	-	
T48887a	5	-78.114	163.854	Lecidea cancriformis Dodge & Baker	MK970677	-	-	Tr_S15	MK970692	-	-	
T48887a	5	-78.070	163.711	Lecidea cancriformis Dodge & Baker	MK970679	MN023046	-	Tr_S15	MK970692	-	-	
T48888b	5	-78.073	163.717	Lecanora cf. mons-nivis Darbishire	MK970658	-	-	Tr_A02	MK970699	-	-	
T48900	5	-78.034	163.845	Rhizoplaca macleanii (Dodge) Castello	MK970663	-	-	Tr_A02	MK970698	-	-	

Supplementary Table S5. Diversity metrics compared in this study, citations, descriptions and interpretation of each, and the used R functions.

Metric	Definition	Citations	Description and interpretation of values	R functions, R package
NRI	Net relatedness index	Webb, 2000; Webb, 2002 (Webb 2000; Webb et al. 2002)	Comparison of phylogenetic distances among all members of a community (pos. values = phylogenetic clustering; neg. values = phylogenetic evenness)	ses.mpd(), picante (Kembel et al. 2010)
PSR	Phylogenetic species richness	Helmus, 2007 (Helmus et al. 2007)	PSV (phylogenetic species variability; degree to which species in a community are phylogenetically related) multiplied by species richness SR (number of species in a sample); SR after discounting species relatedness (values range from 0 = increased relatedness to SR = decreased relatedness)	psd(), picante (Kembel et al. 2010)
J'	Pielou evenness index	Pielou, 1969 (Pielou 1969)	Measure of how evenly distributed abundance is numerically among the species that exist in a community (values range from 0 = no evenness to 1 = complete evenness)	diversity(), vegan (Oksanen et al. 2019)
1 – J'	1 – Pielou evenness index		Values range from 0 = complete evenness to 1 = no evenness	

Supplementary Table S6. Diversity indices (left), specificity indices (middle) and BIO10, BIO12, elevation and latitude means (right) for the different mycobiont species and photobiont OTUs: N, number of sequences; h, number of haplotypes; h/N, ratio of h and N; Hd, haplotype diversity;  $\pi$ , nucleotide diversity; NRI, net relatedness index; PSR, phylogenetic species richness; J', Pielou evenness index. (Note: the specificity indices were calculated for the respective symbiosis partners: 1 - J' on the basis of species/OTUs, NRI and PSR on the basis of haplotypes. As a consequence, only samples where both mycobiont as well as photobiont could be identified are included.)

Mycobiont species	N	h	h/N	Hd	π	NRI	PSR	1 – J'	BIO10 mean	BIO12 mean	Elevation mean (m. a. s. l.)	Latitude mean
Carbonea sp. 2	13	3	0.231	0.564	0.0014	1.238	6.473	0.220	-7.33	120.85	518.83	-79.79
Carbonea sp. URm1	5	2	0.400	0.400	0.0128	-0.297	2.913	0.743	-6.78	138.00	524.40	-79.95
Carbonea vorticosa	11	1	0.091	-	-	1.471	2.000	1.000	-6.27	143.73	436.09	-78.08
Lecanora cf. mons-nivis	3	2	0.667	0.667	0.0000	1.413	2.000	1.000	-6.30	137.33	389.33	-78.07
Lecanora fuscobrunnea	31	6	0.194	0.546	0.0013	-0.423	8.332	0.399	-8.35	107.41	574.81	-81.11
Lecanora physciella	3	2	0.667	0.667	0.0014	1.473	0.011	1.000	-7.70	103.67	610.33	-84.04
Lecanora sp. 2	6	1	0.167	-	-	2.305	3.000	1.000	-6.23	141.00	606.83	-78.08
Lecanora sp. 3	7	1	0.143	-	-	3.002	2.727	1.000	-6.47	153.86	450.43	-79.14
Lecidea andersonii	8	1	0.125	-	-	-0.307	1.764	0.806	-7.19	117.88	282.50	-83.65
Lecidea cancriformis	93	18	0.194	0.797	0.0032	-1.119	10.994	0.244	-8.18	112.87	601.86	-80.82
Lecidea lapicida	1	1	1.000	-	-	-	-	1.000	-5.90	151.00	375.00	-78.02
Lecidea polypycnidophora	10	1	0.100	-	-	1.471	2.000	1.000	-6.22	141.10	411.20	-78.06
Lecidea sp. 5	1	1	1.000	-	-	-	-	1.000	-6.50	172.00	671.00	-78.06
Lecidea sp. 6	3	2	0.667	0.667	0.0106	-	-	1.000	-6.33	148.67	709.00	-78.07
Lecidea UCR1	2	1	0.500	-	-	1.450	2.000	1.000	-6.25	140.00	466.00	-78.08
Lecidella greenii	37	3	0.081	0.324	0.0007	1.933	4.600	0.929	-6.33	144.95	488.57	-78.53
Lecidella siplei	10	4	0.400	0.644	0.0012	2.974	2.916	1.000	-7.52	136.80	322.30	-84.21
Lecidella sp. nov2	9	3	0.333	0.556	0.0073	1.643	0.403	0.821	-7.90	100.33	787.67	-83.87
Rhizoplaca macleanii	51	8	0.157	0.707	0.0019	4.209	4.860	1.000	-6.18	149.92	684.53	-78.07
Photobiont OTU	N	h	h/N	Hd	π	NRI	PSR	1 – J'	BIO10 mean	BIO12 mean	Elevation mean (m. a. s. l.)	Latitude mean
Tr_A02	165	14	0.085	0.702	0.0025	1.739	16.265	0.220	-6.47	144.24	538.61	-79.20
Tr_A04a	3	3	1.000	1.000	0.0023	-0.759	3.000	0.784	-8.70	116.67	491.00	-82.46
Tr_101	10	6	0.600	0.844	0.0084	0.036	1.955	0.695	-8.12	98.60	555.70	-81.19
Tr_I17	2	1	0.500	-	-	-	-	1.000	-7.60	101.50	358.50	-82.21
Tr_S02	58	8	0.138	0.573	0.0049	0.538	7.970	0.580	-7.89	120.25	565.10	-82.12
Tr_S15	10	1	0.100	-	-	0.578	2.596	0.728	-6.16	139.10	689.10	-78.12
<i>Tr</i> _S18	32	1	0.031	-	-	2.529	3.431	0.828	-9.91	77.88	696.25	-80.02

Supplementary Table S7. Network matrix giving the number of associations between the mycobiont species and photobiont OTUs.

		Photobiont OTU								
Mycobiont species	Tr_A02	Tr_A04a	<i>Tr_</i> 101	Tr_ 17	Tr_S02	<i>Tr_</i> S15	<i>Tr_</i> S18			
Carbonea sp. 2	4	2	-	-	3	2	1			
Carbonea sp. URm1	4	-	1	-	-	-	-			
Carbonea vorticosa	9	-	-	-	-	-	-			
Lecanora cf. mons-nivis	3	-	-	-	-	-	-			
Lecanora fuscobrunnea	11	-	3	-	11	-	2			
Lecanora physciella	-	-	-	-	2	-	-			
Lecanora sp. 2	6	-	-	-	-	-	-			
Lecanora sp. 3	7	-	-	-	-	-	-			
Lecidea andersonii	7	-	-	-	1	-	-			
Lecidea cancriformis	7	1	6	2	34	7	28			
Lecidea lapicida	1	-	-	-	-	-	-			
Lecidea polypycnidophora	9	-	-	-	-	-	-			
Lecidea sp. 5	1	-	-	-	-	-	-			
Lecidea sp. 6	3	-	-	-	-	-	-			
Lecidea UCR1	2	-	-	-	-	-	-			
Lecidella greenii	31	-	-	-	-	1	-			
Lecidella siplei	9	-	-	-	-	-	-			
Lecidella sp. nov2	-	-	-	-	8	-	1			
Rhizoplaca macleanii	51	-	-	-	-	-	-			

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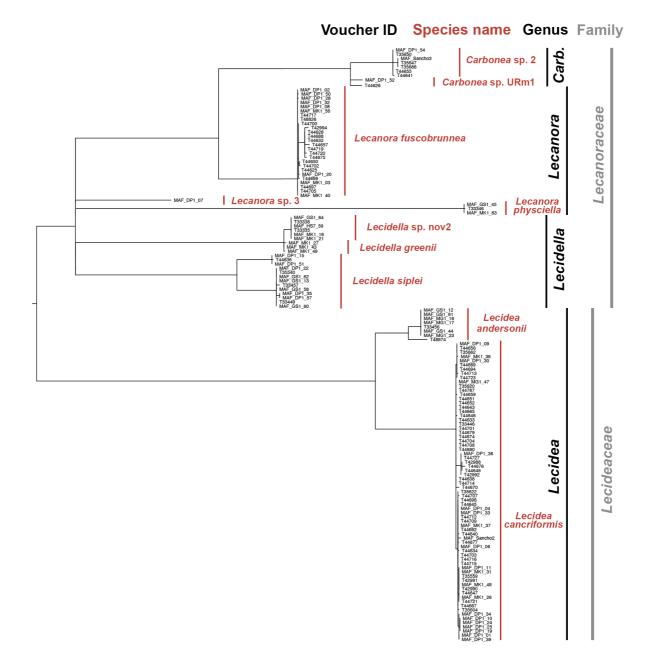
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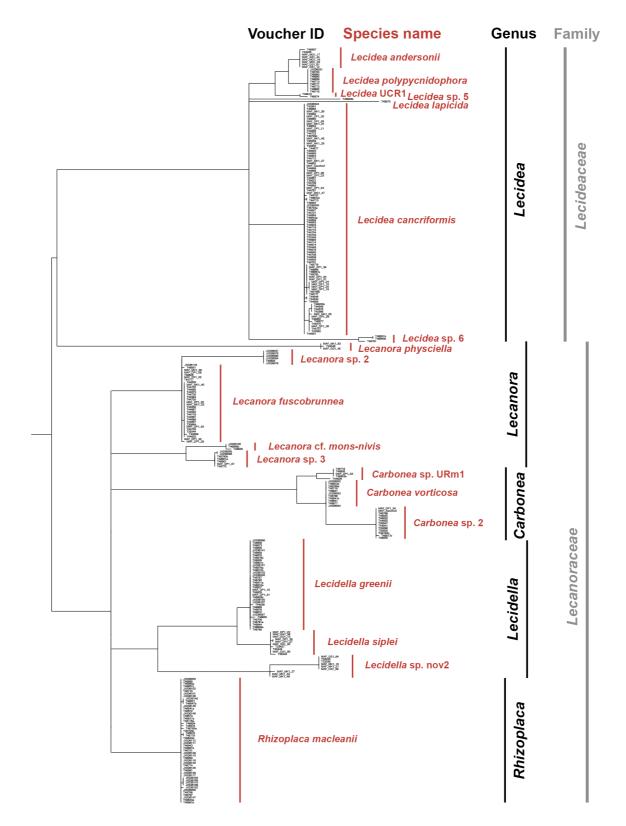
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**Supplementary Figure S1.** Phylogeny of mycobiont specimen based on multi-locus sequence data (nrITS, mtSSU and RPB1; calculated with IQ-TREE (Nguyen et al. 2014); branches with SH-aLRT < 80 % and UFboot < 95 % were collapsed).

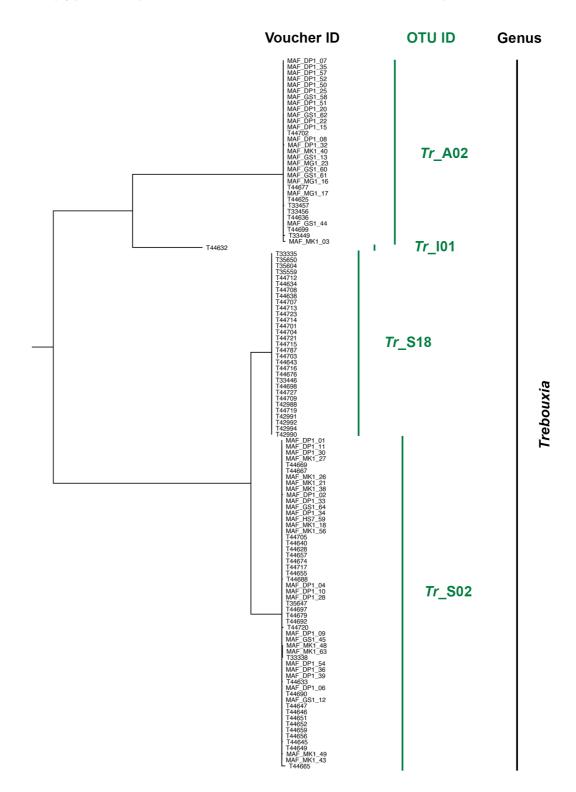


Supplementary Material 2: Figures

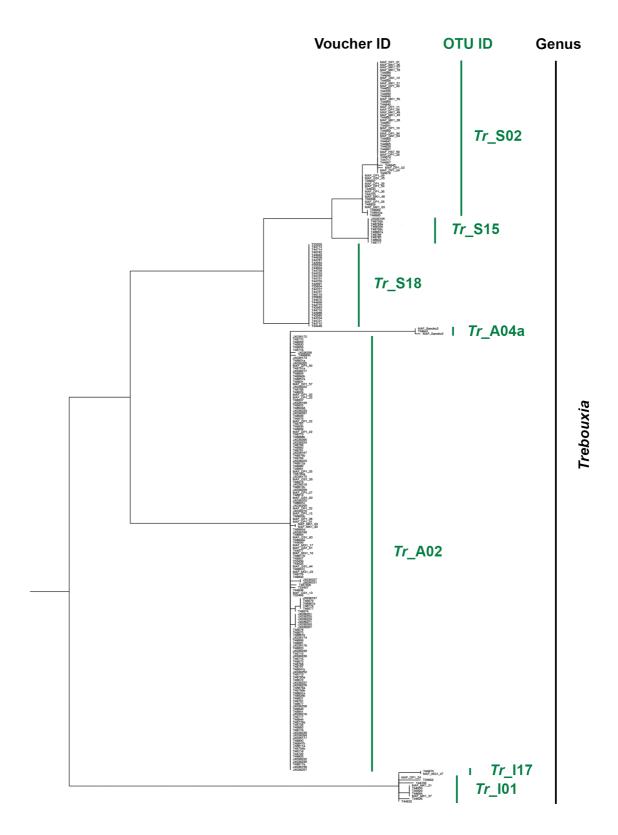
**Supplementary Figure S2.** Phylogeny of all mycobiont specimen based on the marker nrITS (calculated with IQ-TREE (Nguyen et al. 2014); branches with SH-aLRT < 80 % and UFboot < 95 % were collapsed).



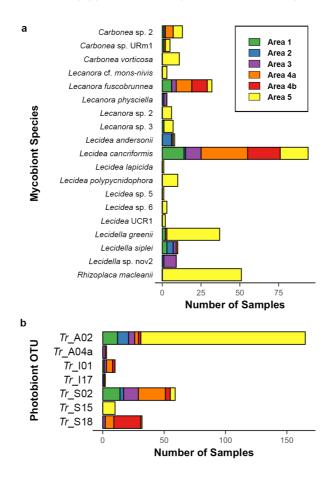
**Supplementary Figure S3.** Phylogeny of photobiont specimen based on multi-locus sequence data (nrITS, psbJ-L and COX2; calculated with IQ-TREE (Nguyen et al. 2014); branches with SH-aLRT < 80 % and UFboot < 95 % were collapsed).



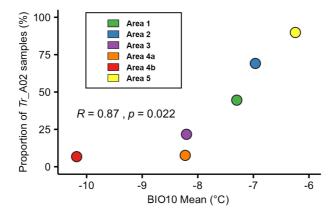
**Supplementary Figure S4.** Phylogeny of all photobiont specimen based on the marker nrITS (calculated with IQ-TREE (Nguyen et al. 2014); branches with SH-aLRT < 80 % and UFboot < 95 % were collapsed).



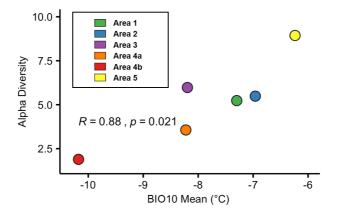
**Supplementary Figure S5.** Barplots giving the number of samples per mycobiont species/ photobiont OTU and area included in this study. (a) Mycobiont species (total sample size: n = 306), (b) photobiont OTUs (total sample size: n = 281).



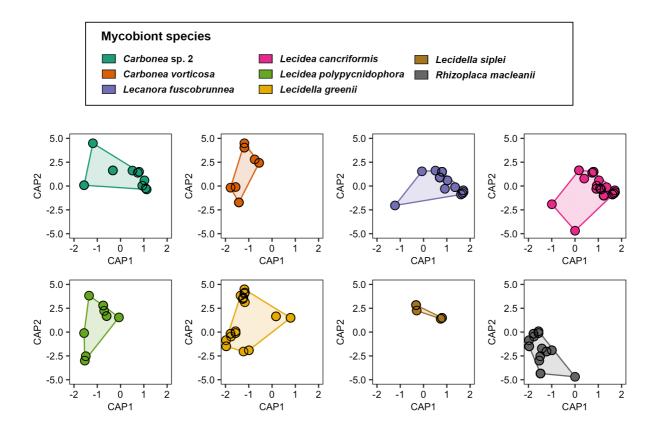
**Supplementary Figure S6.** Correlation plot. Percentage of *Trebouxia* OTU A02 samples against mean values of BIO10 (mean temperature of warmest quarter) for the different areas.



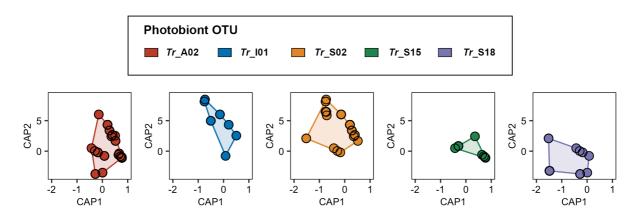
Supplementary Figure S7. Correlation plot. Alpha diversity values of mycobiont species against BIO10 (mean temperature of warmest quarter) mean values of the different areas.



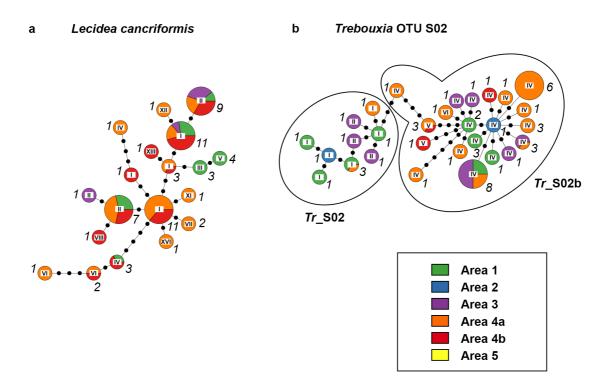
**Supplementary Figure S8.** Ordination plots showing the similarity of mycobiont samples with  $n \ge 10$  after constrained analysis of principal coordinates. Samples located closer to each other are also more similar in terms of the environmental factors elevation, BIO10 and BIO12. The first constrained axis CAP1 explained 13.77 % of the variance, the second constrained axis CAP2 1.76 % of the variance. Only the first axis was significant (F = 17.1640, p = 0.001).



**Supplementary Figure S9.** Ordination plots showing the similarity of photobiont OTUS with  $n \ge 10$  after constrained analysis of principal coordinates. Samples located closer to each other are also more similar in terms of the environmental factors elevation, BIO10 and BIO12. The first constrained axis CAP1 explained 36.87 % of the variance, the second constrained axis CAP2 1.46 % of the variance. Only the first axis was significant (F = 57.0275, p = 0.001).



**Supplementary Figure S10.** Haplotype networks based on multi-locus sequence data, showing the spatial distribution within the different areas. (a) *Lecidea cancriformis*, (b) *Trebouxia* OTU S02. Roman numerals at the center of the pie charts refer to the haplotype IDs based on ITS data (cf., Fig. 2 and Fig. 3 of main text). The italic numbers next to the pie charts give the total number of samples per haplotype. The circle sizes reflect relative frequency within the species; the frequencies were clustered in ten (e.g. the circles of all haplotypes making up between 20-30 % have the same size).



### References

Nguyen L-T, Schmidt HA, von Haeseler A, Minh BQ (2014) IQ-TREE: A Fast and Effective Stochastic Algorithm for Estimating Maximum-Likelihood Phylogenies Molecular Biology and Evolution 32:268-274 doi:10.1093/molbev/msu300