Lichen photobiont diversity and selectivity at the southern limit of the maritime Antarctic region (Coal Nunatak, Alexander Island)

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Abstract

Antarctic ice-free inland sites provide an unique perspective on the strategies coevolving organisms have developed for survival at the limits of life. Here, we provide the first combined description of the ecological and genetic diversity of lichen photobionts colonising an isolated Antarctic inland site, Coal Nunatak, on south-east Alexander Island (Antarctic Peninsula). Photobionts of 14 lichen species (42 samples) representing the entire lichen community of Coal Nunatak were investigated using the internal transcribed spacer region (ITS) of the nuclear ribosomal DNA. The study attempted to address the hypothesis that mycobiont selectivity for the photobiont partner is lower in more extreme environments. This hypothesis did not appear to hold true for the entire lichen community except one species. Another aspect focuses on the relevance of the reproduction modus concerning the distribution of photobiont haplotypes in the lichen community. Dispersal of generative mycobiont diaspores depends on lichenisation processes while by dispersal of vegetative diaspores both symbiotic bionts get dispersed.

Keywords: inland site, extreme environment, symbiotic association, community, genetic diversity, photobiont haplotypes

Introduction

Antarctica is the windiest, coldest and highest continent on Earth. Less than 0.5% of the Antarctic continent is permanently or seasonally free of ice cover (British Antarctic Survey, 2004). Many lichens are successful colonisers of extreme environments, and can be found worldwide in deserts, high mountain ranges, tropical and polar regions. Lichens colonising Antarctic habitats are exposed to some of the most extreme environmental conditions faced in terrestrial environments on Earth (Peck *et al*., 2006), including high levels of UV radiation, both extremely low and very variable temperatures, lack of liquid water and desiccation stress, and high wind speeds. With two flowering plants and approximately 50 liverworts and 104 bryophytes known, the roughly 427 recorded lichen species form the dominant element in the diversity of the Antarctic flora (Ochyra, 1998; Bednarek-Ochyra *et al.*, 2000; Øvstedal & Smith, 2001; Convey, 2013).

The success of lichens under extreme environmental conditions is based on a remarkable symbiotic relation between, at least, two bionts. Approximately 21% of all fungi are known to form lichens (Hawksworth, 1988). In this obligate symbiosis the fungus (mycobiont) is associated with one or more photosynthesizing symbionts, the photobionts, which either can be eukaryotic green algae or prokaryotic cyanobacteria (Hawksworth, 1988). The green algae most commonly found as photobionts in lichens belong to the genus *Trebouxia* (Peveling, 1988). Although between 14 000 and 20 000 lichen-forming mycobionts, mainly ascomycota, are estimated to exist (Feuerer & Hawksworth, 2007) they are associated with only a few different photobiont species (Tschermak-Woess, 1988).

To date, studies of the diversity of lichen photobionts in Antarctica have concentrated on coastal regions along the Antarctic Peninsula (Romeike *et al*., 2002; Brinkmann, 2002;

Langohr, 2004; Siegesmund, 2005). Inland sites, where environmental conditions are generally more extreme and terrestrial diversity and levels of community development much lower (Convey & Smith, 1997) have received little attention (Neuburg, 2007; Pérez-Ortega *et al.*, 2012). Until recently most studies have been carried out based on traditional taxonomic approaches. However, few studies have addressed the degree of selectivity between bionts and any correlation this may have with ecological factors in more extreme environments as typified by Antarctic inland sites (Pérez-Ortega *et al.*, 2012).

In this study diversity data are reported from Coal Nunatak (south-eastern Alexander Island, 70°03´S 068°31´W), an inland nunatak ecosystem at the extreme southern limit of the maritime Antarctic. A very limited lichen and bryophyte flora is present with only 14 lichen species recorded. These 14 species were collected on Coal Nunatak, in order to investigate the hypothesis that mycobiont selectivity for the photobiont partner will be lower in more extreme environments (Romeike *et al*. 2002). The slow rates of community development at locations such as Coal Nunatak, which is characterized by very harsh environmental conditions, provide an opportunity to study lichen photobiont diversity. A second hypothesis relates to the reproductive tactics of the mycobionts, where it is postulated that the distribution of photobiont haplotypes is dependent on either the asexual or sexual reproduction of the mycobiont.

Coal Nunatak is located on south-eastern Alexander Island off the west coast of the Antarctic Peninsula. It is protected from the direct influence of the open sea, over 200 km due west in summer, by the high landmass of Alexander Island and the ice shelves that fringe its west coast, and by the permanent ice shelf that occupies George VI Sound to the east (6 km from the study site) and south (20 km from the study site). Located at the extreme southern limit of the maritime Antarctic, this region’s climate is considered to be intermediate between that of the more moist maritime region and the colder and drier continental zone (Smith, 1988; Convey & Smith, 1997). Coal Nunatak is snow-free during the Antarctic summer for approximately three months. The ecosystem at this site is characterised by its low developmental level (Brinkmann *et al.*, 2007; Engelen *et al.*, 2008). Small and often barely visible populations of different lichen species, occasionally associated with the few recorded bryophyte species, can be found in microniches restricted to rock surfaces and crevices and to the margins of soil polygons.

Reproductive tactics:

Lichens disperse over long distances by utilising two fundamentally different mechanisms. There may be joint asexual dispersal of both symbionts in specific structures, either by means of a vegetative thallus fragment or by specialized dispersal organs as soredia, which are small (100 - 150 µm in diameter) dispersal units that are produced in specialized cup-like structures called soralia composed of both myco- and photobiont cells. These diaspores can easily be distributed over long distances by wind at high altidtudes. The sexual mechanism of lichen dispersal involves the independent dispersal of the mycobiont (as ascospores) and the photobiont (as vegetative cells). Both can grow individually in a new habitat, before coming into contact through a recognition process and forming a new lichen thallus at that location *de novo* by lichenisation (Ott, 1987). Dispersal of the bionts separately clearly requires the process of relichenisation. Environmental conditions and physiological factors influence the success of the recognition process (Meeßen & Ott, 2013; Meeßen *et al.*, 2013).

Selectivity and specificity:

The species diversity of lichen-forming fungi is much greater than that of the photobionts, especially if only green-algal partners, that constitute the photobionts in the majority of lichens, are considered. Algal lineages are widely shared among taxonomic mycobiont groups.

Previous studies have demonstrated that mycobionts and photobionts cannot simply be combined randomly (Ahmadjian & Jacobs, 1981, 1982, 1983), indicating a degree of selectivity between the two bionts. Successful and complete lichenisation can only take place when both symbionts possess the appropriate adaptations (Schaper & Ott, 2003). The degree of specificity and selectivity of the mycobiont partner for particular photobionts varies between species. Rambold *et al.* (1998) defined specificity as the taxonomic range of photobionts associated with a mycobiont and selectivity as the exclusiveness with which specific photobionts are selected as partners. Galun & Bubrick (1984) defined 'selectivity' as the preferred interaction between two bionts, and 'specificity' as the exclusive interaction between photo- and mycobiont. Some fungi are only able to lichenise if a specific algal species is available (Galun, 1988) while, in contrast, other species of fungi are able to form a lichen thallus with several members of the same genus of photobiont, and sometimes with partners related at an even higher systematic level (Piercey-Normore & DePriest, 2001; Helms *et al*., 2001; Beck *et al.*, 2002; Brinkmann, 2002; Romeike *et al*., 2002). Symbiont selectivity and specificity are not only species-specific, but also can vary during the life-cycle of the partners and due to partner availability and environmental conditions. Graduated selectivity is expressed in the form of symbiotic contact achieved between myco- and photobiont. All stages, ranging from the intimate mutualistic contact of both symbionts in a well-developed lichen thallus to a loose-fitting parasitic contact, where the fungus penetrates the algal cells using haustoria and subsequently even kills the algae, are possible (Schaper & Ott, 2003).

High levels of selectivity shown by a mycobiont are linked with a low diversity of suitable photobionts being present in a lichen genus as, for example, found in the family Cladoniaceae (Piercey-Normore & DePriest, 2001), the genus *Physcia* (Helms *et al*., 2001) and the genus

Letharia (Kroken & Taylor, 2000). In contrast, a lower level of selectivity for photobionts has been reported in the Antarctic species *Umbilicaria antarctica* (Romeike *et al.*, 2002). The lower selectivity was interpreted as a form of flexibility and/or plasticity that acts as an adaptation to extreme environmental conditions. Yahr *et al.* (2004, 2006) concluded that, in particular *Cladonia* species, interactions were highly specific and that locally realized associations were probably influenced by environmental conditions. The more extreme lichen-dominated habitats become, the more it can be expected that environmental factors and community structure and composition will influence individual symbiotic interactions.

In this study photobiont diversity was assessed through sequencing of the ITS1, 5.8S and ITS2 region of the ribosomal DNA (Friedl & Rokitta, 1997; Rambold *et al.*, 1998; Beck, 1999; Helms *et al.*, 2001; Kroken & Taylor, 2000; Piercey-Normore & DePriest, 2001; Romeike *et al.*, 2002; Fernandez-Mendoza *et al.*, 2011).

Material and Methods

Study site:

Coal Nunatak is located on south-east Alexander Island off the south-west coast of the Antarctic peninsula (70°03´S 068°31´W) (Fig. 1). The mountain ridge of the nunatak is about 4 km, in a north-east to south-west orientation. Its ice free summit rises 380-424m above sea level. The research site was situated at the north-eastern end of the nunatak, covering approximately 3500 m².

Coal Nunatak belongs to the Le May Group and is composed mainly of greywacke, a coarse grained sedimentary rock type (Burn, 1983). Surface geomorphology is characterised by extensive development of patterned ground and other typical periglacial features (e.g. frostsorted soil polygons, stone stripes) and bare rocks (Brinkmann *et al.*, 2007; Engelen *et al.*, 2008).

Coal Nunatak experiences a continental rather than a maritime climate. From March until mid-December the study site is covered by snow, becoming mostly snow-free during the short summer period from mid-December to early March. Terrestrial ecosystems at this site are at a very low or early stage of development. Much of the ground is barren to the naked eye, with colonisation by macroscopic vegetation restricted to small and generally sheltered micro-niches on rocks, in crevices, and on soil sheltered by rocks or associated with longer-lying snow patches. Investigations of the vegetation on the nunatak revealed a total of 14 lichen species and a small number of mosses are known from the nunatak (Brinkmann *et al.*, 2007; Engelen *et al.*, 2008).

Lichen material:

Most lichen species were collected from the north-eastern part of the study area on the north-east of Coal Nunatak. *Xanthoria elegans* was obtained from the west exposed part of the study area. Three independent samples were obtained for each lichen species, with the quantities sampled being limited by the requirement not to damage the lichen community. After short-term storage at ambient conditions at the field site, lichen samples were transported to the British Antarctic Survey's Rothera Research Station (Adelaide Island). There the samples were stored at -20°C and returned frozen to the laboratory in Düsseldorf. Determination of the lichen species was carried out by taxonomic experts (H. Hertel, Munich; D. Øvstedal, Bergen; N. Wirtz, Frankfurt).

Mycobionts included in the study:

The lichen species examined in this study are listed in Table 1. Seven species of the 14 obtained from Coal Nunatak were epilithic (crustose lichens: *Tephromela disciformis, Tephromela atra, Caloplaca johnstonii, Lecidella pataviana*; macro lichens: *Usnea lambii, Pseudephebe minuscula, Xanthoria elegans*) and seven colonised soil-surface habitats (crustose lichens: *Buellia papillata, Candelariella flava, Caloplaca lewis-smithii, Lepraria cacuminum, Lepraria borealis, Ochrolechia frigida* and *Psoroma cf. tenue*). *Psoroma cf. tenue* was the only lichen colonising soil sites that has a well differentiated thallus.

Laboratory procedures:

Identification of the unicellular green algal photobionts using morphological and anatomical characters is known to be challenging. Therefore, we used a molecular approach to assess photobiont diversity. The nuclear internal transcribed spacer (ITS) region of the rDNA was analysed, including ITS1, ITS2 and the gene coding for the 5.8S ribosomal subunit. The region is located between the genes coding for the 18S and 26S ribosomal units in the ribosomal DNA tandem repeats and has been used routinely in molecular studies of green algal photobionts (Friedl & Rokitta, 1997; Rambold *et al*., 1998; Beck, 1999; Helms *et al.*, 2001; Kroken & Taylor, 2000; Piercey-Normore & DePriest, 2001; Romeike *et al*., 2002; Schaper & Ott, 2003; Yahr *et al*., 2004; Yahr *et al*., 2006).

To obtain photobiont DNA, conglomerates of photobiont cells were first carefully removed from the lichen thalli. This avoided the disruption of molecular procedures by secondary lichen metabolites such as phenolic substances. The clusters of photobiont cells were fragmented using liquid nitrogen and quartz sand. For DNA extraction the DNeasy Plant Mini Kit (Qiagen, Hilden, Germany) was used. After extraction the isolated DNA was stored at -20°C.

For a 25 µl PCR reaction, 2.5 µl template, 9 µl sterilized water, 12.5 µl HotStartTaq ™ Master Mix (Qiagen) and 0.5 µl of each primer were used. The green alga specific primer with 5´-3´orientation is Al 1700f (Helms *et al.*, 2001). The primer used with 3´-5´orientation (LR3, http://www.biology.duke.edu/fungi/mycolab/primers.htm) is not specific for green algae (Freidl & Rokitta, 1997). For the amplification of the photobiont ITS-region a thermocycler (Biometra, Goettingen, Germany) was used as follows. The taq-polymerase was activated for one minute 95°C. The DNA was denatured for one minute at 94°C. The annealing temperature of the primers was set to 53°C for one minute. The elongation of the annealed primers by taq-polymerase took place for 1.5 minutes at 72°C. The denaturation, annealing and elongation steps were repeated 35 times, after which the final extension of partially elongated products took 10 minutes at a temperature of 72°C. After final extension the PCR product was cooled at 4°C. The amplified PCR products were purified using the QIAquick PCR Purification Kit (Qiagen, Hilden, Germany).

DNA sequencing was carried out by GATC-Biotech (Konstanz, Germany) using an ABI 3730 XL Sequencer. Non algal specific primers used for sequencing were 1800f (5´-3´orientation) (Friedl, 1996) and ITS4 (3´-5´orientation) (White *et al.*, 1990). The resulting ITS rDNA sequences were edited using the application 'Bioedit for Windows' (http://www.mbio.ncsu.edu/bioedit/bioedit.html). NCBI-BLAST searches of GenBank records were performed to confirm that the amplified and sequenced DNA fragments originated from the photobiont and to identify the taxonomic classification of the closest hit.
The alignment of all sequences was carried out using the online application MAFFT version 7 (http://mafft.cbrc.jp/alignment/server/) based on the HKY substitution model (Hasegawa *et al.*, 1985). The calculation of a phylogenetic maximum likelihood tree using PhyML 3.0 (Guindon *et al.*, 2010) was supported by 1000 bootstrap steps. All sequences obtained from samples of

lichen photobionts from Coal Nunatak have been added to the database of the National Center for Biotechnology Information (acession numbers: FJ426284 - FJ426299).

Results

In 14 lichen species of 11 genera from Coal Nunatak we found seven different haplotypes of the genus *Trebouxia* and one haplotype of the genus *Asterochloris*. Sexual reproduction was noted only in crustose species, with six lichens producing fruiting bodies (Table 1). The reproduction of these lichen species using ascospores in Antarctic habitats has previously been noted by Øvstedal & Smith (2001). In most of the lichens included in this study the mycobiont was associated with green algal photobionts, with the exception of *Psoroma cf. tenue*. This lichen species also forms thallus structures (cephalodia) with cyanobacteria of the genus *Nostoc*. The mycobiont of this lichen is, therefore, associated with both a green algal species and a cyanobacterial species.

In the lichen species examined, haplotype 8 was found as photobiont of three lichen species: *Lepraria borealis*, *Usnea lambii* and *Pseudephebe minuscula. Trebouxia* haplotype 7 was the dominant haplotype in the samples investigated at this locality. Six species (*Lecidella pataviana*, *Lepraria borealis*, *Lepraria cacuminum*, *Tephromela atra*, *Tephromela disciformis* and *Xanthoria elegans*) were associated with this photobiont. Several haplotypes were restricted to a single lichen species, including haplotypes 2 (*Buellia papillata*), 3 (*Candelariella flava*), 4 (*Psoroma cf. tenue*), 5 (*Caloplaca lewis-smithii*) and 6 (*Caloplaca johnstonii*). The algal genus *Asterochloris* (haplotype 1) was found as photobiont in the two lichen species *Lepraria borealis* and *Ochrolechia frigida* (Table 1).

Thirteen of the 14 mycobiont species from Coal Nunatak were associated with only a single algal haplotype, the exception being *L. borealis.* This species contained three different photobiont haplotypes 1, 7 and 8 (Table 1). The ITS rDNA sequences of the photobionts detected in *L. borealis* were identical to the photobionts found in lichen species that were colonized by thalli of *L. borealis* (Engelen *et al.*, 2010). Respectively, *L. borealis* when growing in close association with *O. frigida* contained haplotype 1, in association with *T. disciformis* haplotype 7 and in association with *U. lambii* haplotype 8 (Engelen *et al*., 2010).

The phylogenetic tree showed three highly supported basal clades (Fig. 2). The first clade as outgroup only consisted of two sequences of haplotype 1. These sequences closest similarities in a BLAST search were to the genus *Asterochloris*. The sequences of the second clade belonging to haplotype 8 showed highest BLAST hits with taxa identified as *Trebouxia jamesii*. The third clade consisted of two subclades. One included haplotypes 6 and 7, and had highest BLAST similarities with taxa also identified as *T. jamesii*, and the other consisted of haplotypes 2 to 5, which were most similar to *Trebouxia impressa*. Most substitutions were found in the subclade consisting of haplotypes 2 to 5 (*T. impressa)*. The other clades, involving the two different groups of *Trebouxia jamesii* and one group of *Asterochloris* sp., were composed of almost identical sequences within each clade.

With the exception of *Lepraria borealis*, the photobiont clades were correlated to groups of lichens characterised by sharing particular morphological and ecological features. The clade consisting of the photobiont haplotype 8 (*T. jamesii*) was associated with fruticose lichens growing on rocks, whereas photobionts of the clade consisting of haplotypes 6 and 7 (*T. jamesii*) were found in crustose lichens on rocks. Crustose lichens growing on soil and/or mosses were either associated with photobionts of the clade consisting of haplotypes 2-5 (*T.*

impressa) or with the photobiont clade of haplotype 1 (*Asterochloris*). Photobionts of *L. borealis* were present in all clades with the exception of the *T. impressa* group.

Discussion

This study is the first to document lichen photobiont diversity in the southern region of the Antarctic Peninsula. Molecular studies on photobionts have shown that identical haplotypes are widespread and can be found across geographic regions (Kroken & Taylor, 2000; Yahr *et al.*, 2004; Yahr *et al.*, 2006), continents (Piercey-Normore & DePriest, 2001) and even hemispheres. *Trebouxia jamesii* identified here at Coal Nunatak, has been described from a range of other localities in the maritime and continental Antarctic (Romeike *et al.*, 2002; Pérez-Ortega *et al.*, 2012) as well as from Europe (Beck, 1999), supporting effective dispersal across distances up to intercontinental and global scales.

Patterns of haplotype distribution being unique to specific habitats suggest a process of local adaptation or might be pre-adapted to local environmental conditions and communities (Pérez-Ortega *et al.*, 2012). Such an interpretation is in line with the conclusions drawn by Yahr *et al.* (2004) in an extensive community study, who found a homogeneous photobiont pool across geographic distances and proposed that local adaptation of the photobiont was of importance at some sites.

All localities analyzed by Yahr *et al.* (2004) shared the same habitat and vegetation type, reflected by the occurrence of similar *Cladonia sp.* communities. Such a sampling design enhances the detection of environmental selection acting in photobiont lineages that might cause ecological specialization.

The ITS sequence of photobiont haplotype 8 found on Coal Nunatak in this study was also found in lichens from the maritime Antarctic sites Lagoon Island, Rothera Point and Charcot Island by Romeike *et al*. (2002). It is also identical to the sequence of *T. jamesii* cultured from *Lecidea silacea*, collected from siliceous and heavy-metal containing rocks at localities in Austria (Beck, 1999). It seems that haplotype 8 (*T. jamesii)* is a generalist with a bi-hemispherical distribution that occurs ranging from extreme habitats at the limits of vegetation to more moderate maritime and polar habitats to comparably benign temperate localities.

Romeike *et al*. (2002) noted that this haplotype has only been described to date from iron-rich sites (Beck, 1999). However, the study on Coal Nunatak does not have atypically high iron concentration. Haplotype 8 was the second most abundant photobiont at Coal Nunatak.

At Coal Nunatak photobionts are associated with a relatively low number of mycobionts. Haplotype 7 was the most abundant photobiont amongst 14 lichen species recorded on Coal Nunatak, being present in 6 different species while haplotypes 1-6 and 8 were distributed amongst 9 lichen species (Table 1). This is suggestive of selectivity and specificity of the symbionts of the respective lichen species. Eight different photobiont haplotypes were found in the 14 lichen species (Table 1). With one exception *Lepraria borealis* (Engelen *et al.*, 2010) all mycobionts were associated with a single photobiont haplotype.
This might be due to the harsh environmental conditions and the overall short growing season at the inland site that limits photosynthetic activity and thus photobiont productivity. In such a life-averse habitat which effects a limited primary production (Sadowsky & Ott, 2012) symbiont interactions can be expected to be fine-tuned for the holobiont to survive and to successfully colonise and keep the habitat.

To form an own thallus *L. borealis* takes over the photobiont haplotypes 1, 7 and 8. Our data suggest that the species may be able to obtain these photobionts from physically adjacent thalli of other lichen species, such as *Ochrolechia frigida* (Pertusiales), *Tephromela disciformis* and *Usnea lambii* (Lecanorales s. str.) (Table 1). When growing close to *T. disciformis* or *U. lambii*, *L. borealis* incorporated the identical *Trebouxia jamesii* haplotypes 7 and 8 as the photobionts of these immediately adjacent lichens. Similarly, when growing in association with *Ochrolechia frigida* the same *Asterochloris* haplotype 1 was present in both lichens (Engelen *et al.*, 2010). Thus, only the *L. borealis* mycobiont shows low selectivity towards potential photobionts consistent with the prediction of Romeike *et al*. (2002).

The diversity of the photobiont haplotypes within the lichen community may be influenced by the mechanism of reproduction. Sexual reproduction requires a lichenisation process during which the mycobiont must encounter a suitable algal partner amongst those available in its immediate vicinity. The data obtained in the current study give no indication of any difference in diversity of photobiont haplotypes between the mycobionts reproducing asexually or sexually (Table. 1).

For the degree of selectivity of the mycobiont to the photobiont partner characteristic features of the respective lichen symbiosis may be primarily responsible at extreme habitats. Environmental conditions might also effect the degree of selectivity (Romeike *et al.* 2002). However, based on the results presented the potential of the symbiotic state of lichens seems to be the more relevant factor considering the success of colonisation processes particularly at extreme environments.

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Tab. 1: Reproductive mode and distribution of photobiont haplotypes in lichen species found on Coal Nunatak.

* haplotype 1: genus *Asterochloris*; haplotypes 2-8: genus *Trebouxia*

lichen species	**reproductive mode**		**haplotypes***							
	sexual	asexual	1	2	3	4	5	6	7	8
Buellia papillata	**X**			**X**						
Caloplaca johnstonii	**X**							**X**		
Caloplaca lewis-smithii	**X**						**X**			
Candelariella flava		**X**			**X**					
Lecidella pataviana	**X**								**X**	
Lepraria borealis		**X**	**X**						**X**	**X**
Lepraria cacuminum		**X**							**X**	
Ochrolechia frigida		**X**	**X**							
Pseudephebe minuscula		**X**								**X**
Psoroma cf. tenue	**X**					**X**				
Tephromela atra	**X**								**X**	
Tephromela disciformis		**X**							**X**	
Usnea lambii		**X**								**X**
Xanthoria elegans		**X**							**X**	

Figure captions

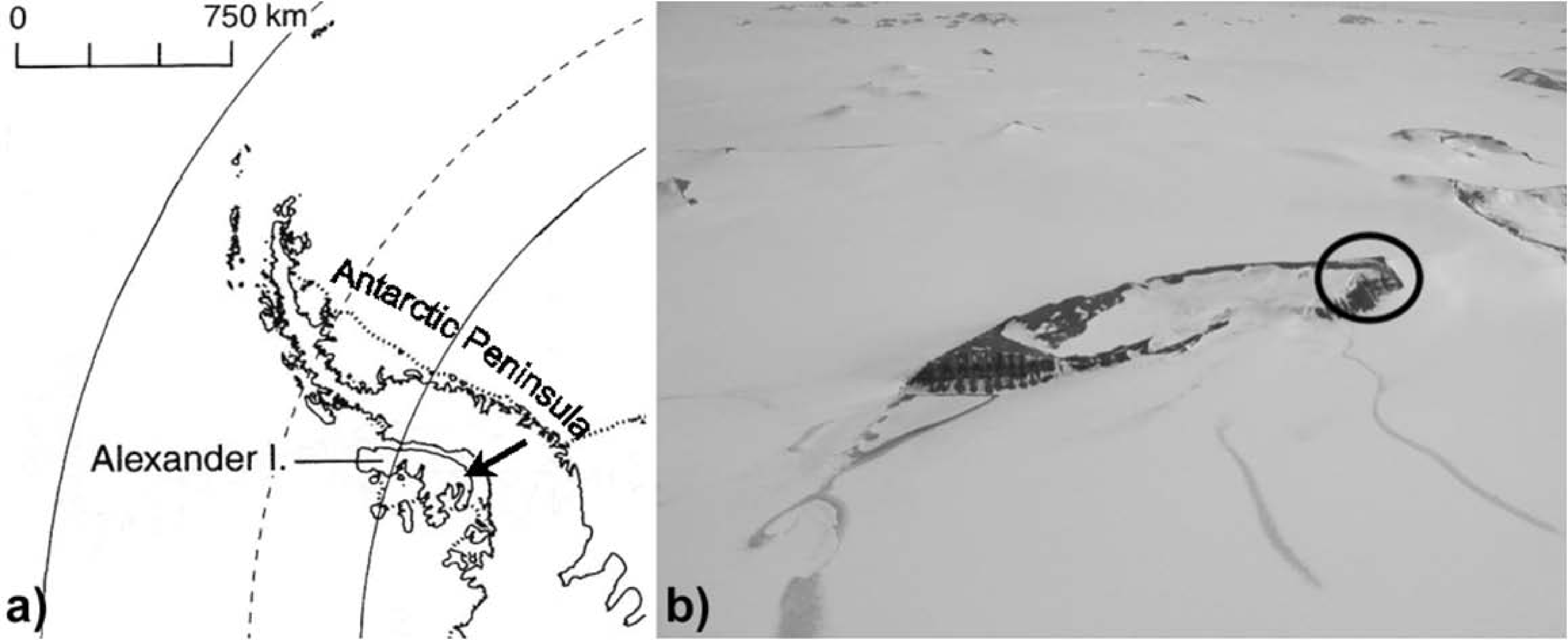
Fig. 1: a: arrow=location of Coal Nunatak on Alexander Island (70°03’S 68°31’W). b: circle=location of the research area on Coal Nunatak.

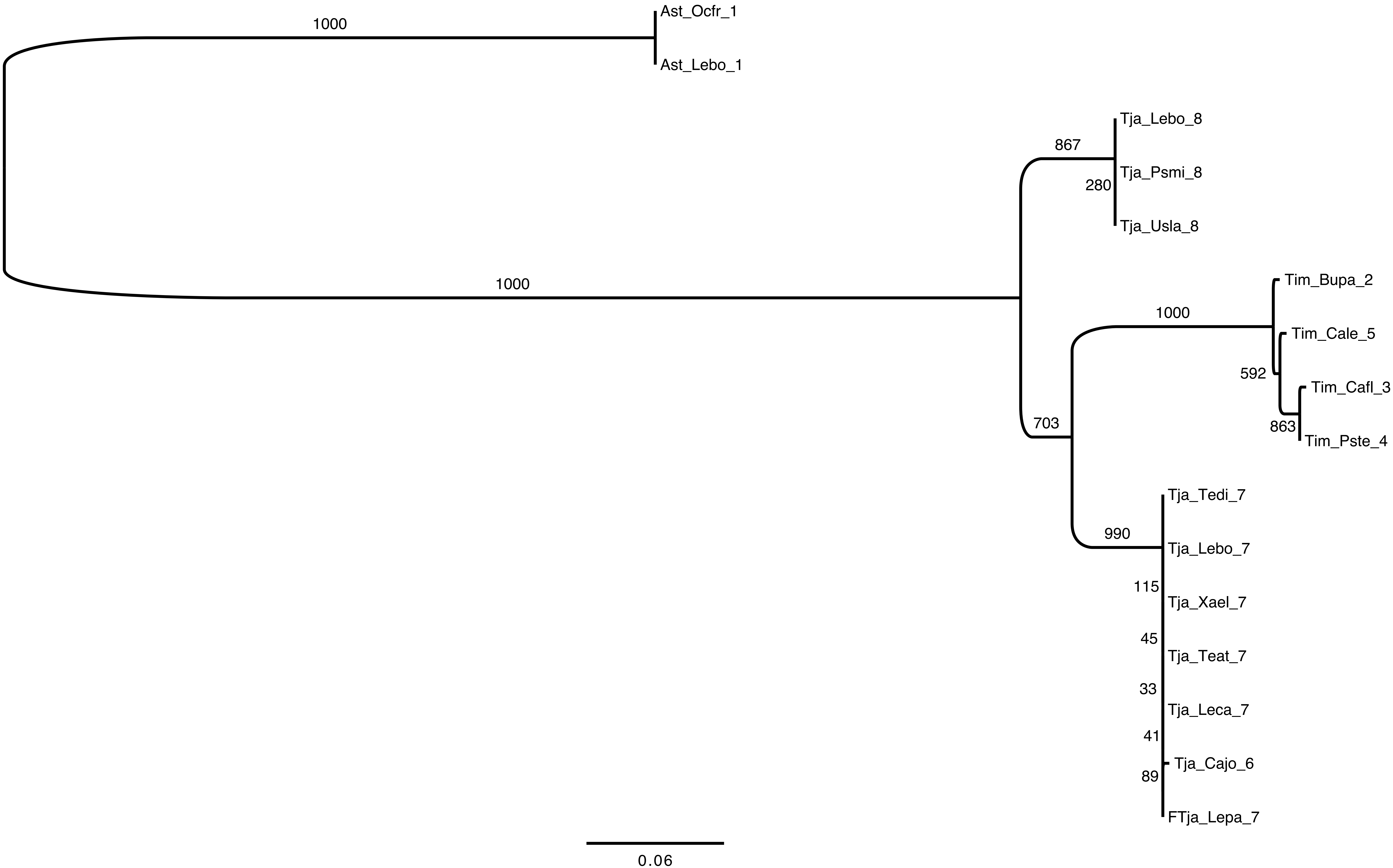
Fig. 2: ML-tree of the *Asterochloris* haplotype and the 7 *Trebouxia* haplotypes as shown in Tab. 1.

The photobiont sequences are named as follows: abbreviation of the photobiont_abbreviation of the mycobiont_number of the haplotype as in Tab.1

Abbreviations of the mycobiont: Bupa: *Buellia papillata*, Cafl: *Candelariella flava*, Cajo: *Caloplaca johnstonii*, Cale: *Caloplaca lewis-smithii*, Lebo: *Lepraria borealis*, Leca: *Lepraria cacuminum*, Lepa: *Lecidella pataviana*, Ocfr: *Ochrolechia frigida*, Psmi: *Pseudephebe minuscula*, Pste: *Psoroma cf. tenue*, Teat: *Tephromela atra*, Tedi: *Tephromela disciformis*, Usla: *Usnea lambii*, Xael: *Xanthoria elegans*

Abbreviations of the photobionts: Ast: *Asterochloris spec.*, Tja: *Trebouxia jamesii*, Tim: *Trebouxia impressa*


0
750 km
Antarctic Peninsula
Alexander I.
a)
b)



Ast_Ocfr_1
Ast_Lebo_1
Tja_Lebo_8
Tja_Psmi_8
Tja_Usla_8
Tim_Bupa_2
Tim_Cale_5
Tim_Cafl_3
Tim_Pste_4
Tja_Tedi_7
Tja_Lebo_7
Tja_Xael_7
Tja_Teat_7
Tja_Leca_7
Tja_Cajo_6
FTja_Lepa_7
1000
1000
867
280
1000
592
863
703
990
115
45
33
41
89
0.06