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Biogeography and ecology of *Cetraria aculeata*, a widely distributed lichen with a bipolar distribution

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Abstract

Ecological and historical biogeography of lichens have rarely been studied in a concerted effort, but both aspects have to be taken into consideration when explaining the distributional patterns of species. This review summarizes, partly preliminary, results from a series of studies on phylogeography, ecophysiology and symbiotic interactions of the lichen *Cetraria aculeata*. This species is not only widespread but also occupies a very wide ecological niche. Evidence suggests that *Cetraria aculeata* has evolved and diversified in the Northern Hemisphere and colonised the Southern Hemisphere during the Pleistocene. Genetic isolation of populations indicates the absence of ongoing long range dispersal and genetic exchange between geographically isolated populations. We observe a hitherto unrecognized genetic diversity that may indicate ecotypic differentiation and speciation processes. Mediterranean and Polar populations differ not only genetically, but also in ecophysiological properties. Ongoing common garden experiments will have to show whether genetically fixed adaptation or acclimation is responsible for these differences. The genetic structure of the photobiont is best explained by climatic differences between localities, but co-dispersal with the mycobiont plays an important role as well. Taken together, these results indicate that a photobiont switch in the past enabled *C. aculeata* to widen its ecological niche, with subsequent genetic isolation of populations. Photobiont switches may play a crucial role in speciation processes of lichens. A combination of ecophysiological and phylogeographic studies with experimental approaches is necessary to better understand the reaction of lichens to changing environmental conditions.

Keywords
Ecological biogeography, phylogeography, population genetics, symbiont interactions, lichens, *Cetraria*

Introduction

Biogeography aims at explaining the distributions of species, which are basically shaped by two factors: their ecological niches and their evolutionary history. Of the two fundamental subdisciplines of biogeography (Cox and Moore 2010), ecological biogeography explains the restricted occurrence of species in terms of environmental conditions (climate, properties of the substratum etc.) and the adaptation or acclimation of the species to these conditions. Historical biogeography, the second important subdiscipline, is focussed on events or processes in the past (plate tectonics, dispersal events etc.) that shaped the extant ranges of species. It is obvious that neither ecological aspects nor history alone are sufficient to understand the geographical patterns that we observe in nature. The close connection between both factors was recognized from the very beginning (e.g. Schimper 1898). Darwin (1872) already invoked glacial climate changes as explanations for the bipolar distribution of taxa. Over the last decades, fossil and genetic data provided compelling evidence for the hypothesis that the distributional ranges of most temperate species shifted widely during the Pleistocene glacial cycles (Hewitt 1999). Their adaptation to certain ecological conditions forced species to migrate or disperse to follow the displacement of their habitats, i.e. ecology influenced the species' population history. On the other hand, adaptation occurs over evolutionary time scales. Ecotypic differentiation (Turesson 1922) in particular, the formation of ecotypes adapted to different ecological conditions within a species' range, may be a direct result of historical population processes, e.g. the fragmentation of habitats or dispersal to areas unconnected with the main distributional range.

Before molecular genetic data became available, historical biogeography relied on fossil evidence or comparisons of distributional ranges of closely related species, often in conjunction with paleoenvironmental reconstructions (e.g. Gray 1859, Firbas 1949). This approach largely excluded taxa for which fossil evidence was sparse. Since the 1990s molecular data have put historical biogeography on an entirely new basis. Phylogenetic methods and model-based approaches using population genetic data allow the reconstruction of past population structure and distributional ranges independent of fossil data, often with high spatial resolution (Ronquist and Sanmartín 2011, see chapter on demography and range shifts below for details). In the field of ecology, ecological niche modelling in combination with spatially explicit reconstructions of paleoclimates has increasingly been used to reconstruct the past distributional ranges of species, e.g. during the last glacial maximum. Comparisons of the results of both methods have shown that phylogeographic methods and approaches using ecological niche modelling often come to similar conclusions regarding the location of ancestral ranges (Waltari et al. 2007; Cordellier and Pfenninger 2009). This is of importance because it suggests that the ecological niches of the studied species have

not changed dramatically over the investigated time scales, a behaviour known as niche conservatism (Peterson et al. 1999; Wiens 2004).

Interpreting the distributional ranges of lichens has always been a challenging endeavour. Due to the sparse fossil record of lichens, historical biogeography was largely speculative before the advent of molecular markers (see e.g. the papers by Lynge 1941 or Poelt 1963). Their large distributional ranges were often attributed to the small size of their propagules that facilitated dispersal (e.g. Galloway and Aptroot 1995). For this reason, Fægri (1950) considered lichens unsuitable objects for historical biogeographical study, perhaps reflecting the microbiological dogma that „everything is everywhere, but the environment selects" (Baas-Becking 1934). Many lichens do not only display large geographical ranges, they also seem to have exceptionally wide ecological niches. The ranges of many predominantly polar species extend into the temperate zone and vice versa (Printzen 2008). Especially for crustose lichens, the true distributional ranges or even species circumscriptions are often not nearly clear, which makes biogeographic inferences difficult if not impossible.

To complicate matters further, the symbiotic nature of lichens impedes the interpretation of results. Distributional ranges may be limited not by ecological restrictions of the lichen or because of dispersal limitations of the mycobiont, but because suitable photobiont(s) are absent. Adaptations to local climatic conditions or acclimation may occur in only one or both of the symbionts. In symbiotic systems, adaptation may not even be the result of mutation and natural selection. A symbiotic host may "outsource" (Gilbert et al. 2010) parts of its stress response to symbiotic partners and respond to changing environmental conditions by habitat-adapted symbiont association (Rodriguez et al. 2008). Such a mechanism has, for example, led to the coral probiotic hypothesis (Reshef et al. 2006) and the hologenome concept (Rosenberg and Zilber-Rosenberg 2011). Many bacterial symbiotic communities are known to vary even in response to short-term environmental changes, e.g. heat stress in corals or the nutritional diet of insects (Littman et al. 2010; Feldhaar 2011). In the case of lichens, it has been demonstrated that species associate with different photobionts in different habitats (Blaha et al. 2006; Yahr et al. 2006) although nothing is known about the time scales over which mycobionts can switch their photobiont partners.

To sum up, the large ecological niches observed in some lichens may be the result of (1) the ability of one or both symbionts to acclimate to widely different ecological conditions, (2) ecotypic differentiation of populations in different parts of a species range, (3) „habitat-adapted symbiosis" by selective association of mycobionts with different photobionts or microbial symbionts, and (4) an overly broad species concept and the presence of unrecognized, possibly cryptic, species. The results summarized in this short review are based on ongoing studies on the phylogeography, population genetics, symbiont interactions and ecophysiology of the widespread bipolar lichen *Cetraria aculeata* (Schreb.) Fr. (Fig. 1). It will become evident that even after years of research focussing on a single species there are more questions open than answered. „Inferring the past to predict the future" (Cordellier and Pfenninger 2009) has been the motivation of many phylogeographic reconstructions. To our mind, it is becom-

Figure 1. *Cetraria aculeata*, habit.

ing increasingly evident that a combination of ecophysiological and phylogeographic approaches and more experimental studies are necessary to understand the reaction of symbiotic systems such as lichens to changing environmental conditions.

Delimiting species within the *Cetraria aculeata* group

Uncertain species delimitations can undermine population-level studies of lichens. If several unrecognized species are included in a study, the assumptions of null-models (e.g. panmixia or certain modes of range expansion) may be violated. Recognizing the presence of different species and restricting the dataset to one of them may lead to a different problem: erosion of the dataset, to an extent that may prevent meaningful statistical inferences. For example, Spribille (2011) found that *Mycoblastus sanguinarius* (L.) Norman, assumed to be a common, easily identified circumboreal species, consists of several genetically, morphologically and chemically distinct lineages. Further, Wirtz et al. (2008) showed that Western North American and Antarctic populations thought to represent the bipolar species *Usnea sphacelata* R. Br. belonged to a different species, *U. lambii* (Imshaug) Wirtz & Lumbsch.

Species delimitation within *Cetraria* s. str. is not unproblematic. *Cetraria aculeata* and *C. muricata* (Ach.) Eckfeldt can be extremely difficult to distinguish in the field. A molecular study by Thell et al. (2000) based on ITS and nucSSU group I intron sequences showed *C. muricata* embedded within a paraphyletic *C. aculeata*. This does

not mean that both belong to the same phylogenetic species (see Wirtz et al. 2012 for a brief outline of species delimitation problems). Several interpretations are possible, including the presence of more than two distinct lineages within a broadly circumscribed *C. aculeata*. Indeed, in a study using ITS and b-tubulin sequences (Thell et al. 2002), two individuals of *C. muricata* appeared as sister to clades of *C. aculeata* and *C. odontella* (Ach.) Ach. In the most recent molecular studies of cetrarioid lichens (Thell et al. 2009; Nelsen et al. 2011) based on more markers, *C. aculeata* and *C. muricata* were always treated as separate species. At least two additional close relatives of *C. aculeata* have so far been described, the corticolous *C. crespoae* (Barreno & Vázquez) Kärnefelt from western Spain and *C. steppae* (Savicz) Kärnefelt from Ukraine containing norstictic acid (Kärnefelt et al. 1993). The latter was later also reported from Spain (e.g. Maestre et al. 2011). The status of these four species has not yet been tested with molecular data. More problems with the circumscription of *C. aculeata* have been reported from Western North America (Goward 1999) and became obvious during our own field work in the region. Most ITS sequences generated in an unpublished population genetic study on *C. aculeata* in Alaska and the Yukon Territory proved to belong to a lineage more or less unrelated to *C. aculeata* (Seifried 2009).

Spanish authors have used the name *C. steppae* for morphologically distinct, vagrant forms of a *Cetraria* with thickened and sometimes almost foliose, unbranched thalli. Apparently, norstictic acid has not been reported from this Spanish material. In a detailed anatomical, physiological and population genetic study, Pérez-Ortega et al. (2012) described anatomical and physiological differences between these forms and „typical" *C. aculeata* from the same habitats. While vagrant morphs differed in several anatomical details (thicker cortex with multilayered cell walls and fibrous material, denser algal layer with smaller algal cells), photosynthetic response curves and dark respiration of both forms were almost identical. Vagrant morphs, however, had an increased water-holding capacity. The genetic comparison revealed that mycobionts and photobionts of both forms shared multilocus haplotypes, but tests of differentiation showed that vagrant haplotypes were not a random subsample of the investigated populations. At present it seems as if the distinct morphology of vagrant thalli is an adaptation to arid conditions, but that both morphs are not differentiated enough to warrant taxonomic delimitation from the typical morphs of *C. aculeata* found in the same locations. In order to further investigate the degree of genetic isolation between these morphs, markers with a higher resolution such as microsatellites will be necessary. Likewise, the degree of isolaton between *C. aculeata* and typical *C. steppae* or the sympatric *C. crespoae* has to be investigated using population genetic approaches. A different case is that of *C. aculeata* in Western North America. Figure 2 shows that the samples from Alaska and the Yukon Territory closely resembling *C. aculeata* that were studied by Seifried (2009) mostly belonged to a group of haplotypes that is well separated from two haplotype groups from Iceland and Svalbard. Only three out of 66 individuals from Beringia belonged to these two groups. In this case, genetic isolation seems to be almost complete.

So, how many species can we distinguish within the *C. aculeata* group and how can they be delimited? The species tree in Fig. 3 is based on a dataset comprising ITS,

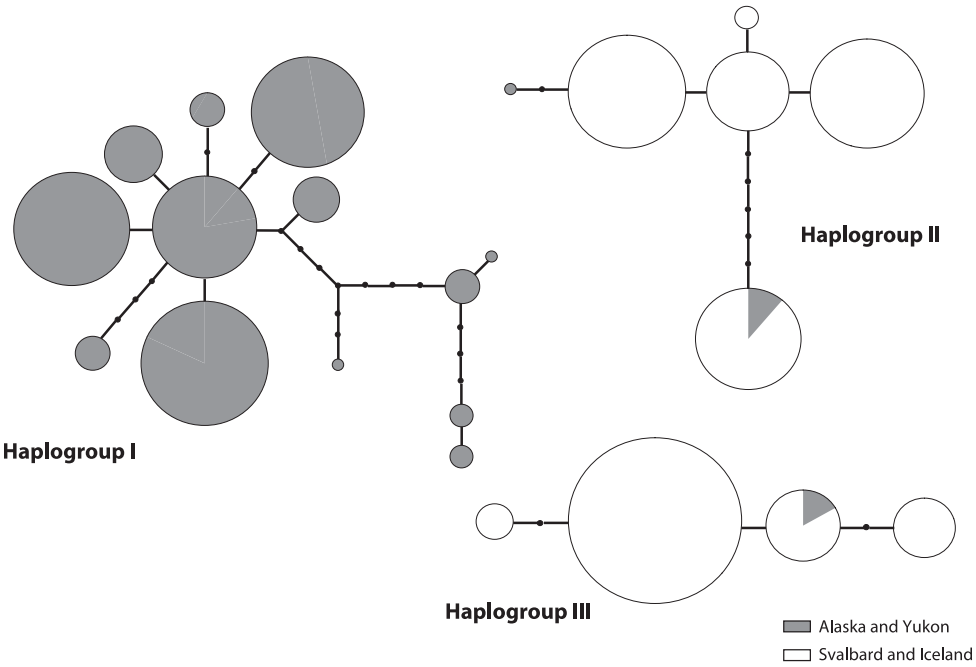

Figure 2. 95 % parsimony probability haplotype network based on ITS sequences of the mycobiont displaying genetic differences between Beringian samples of the *C. aculeata* group (grey) and individuals from two haplotype groups present on Iceland and Svalbard (white). Large circles represent haplotypes, each line a mutational step and black dots missing haplotypes. The size of the circles is proportional to the number of individuals sharing that haplotype. The presence of three unconnected haplogroups indicates that the genetic differences between them are too large to find a connection with a 95 % probability of being the most parsimonious one.

mtLSU and glyceraldehyde-3-phosphate dehydrogenase (GPD) DNA sequences and contains individuals of all currently accepted species of the *C. aculeata* group. Preliminary studies (Fernández-Mendoza et al. 2011; Domaschke et al. 2012) had shown considerable genetic diversity within the group with several relatively well supported clades. When sequences of *C. steppae* and *C. crespoae* were added to these datasets, they usually ended up among the different clades of *C. aculeata* s. lat. We therefore decided to treat each separate clade as a separate OTU (operational taxonomic unit) in a Bayesian species tree approach using *BEAST (for details on methods see below) instead of using a phylogenetic approach to infer the evolutionary relationships among single sequences. In addition to *C. australiensis* Kärnefelt, *C. crespoae, C. muricata, C. odontella* and *C. steppae* we distinguished seven different OTUs within what was potentially *C. aculeata*: (1) material from Western North America and South America, here provisionally called "*C. panamericana*", vagrant morphs from central Spain, Mediterranean and Arctic material from two genetically divergent ITS clades (haplogroups II and III in Figs. 2 and 3), and material from the southern hemisphere that in previous analyses (Fernández-Mendoza et al. 2011; Domaschke et al. 2012) had proved to form a

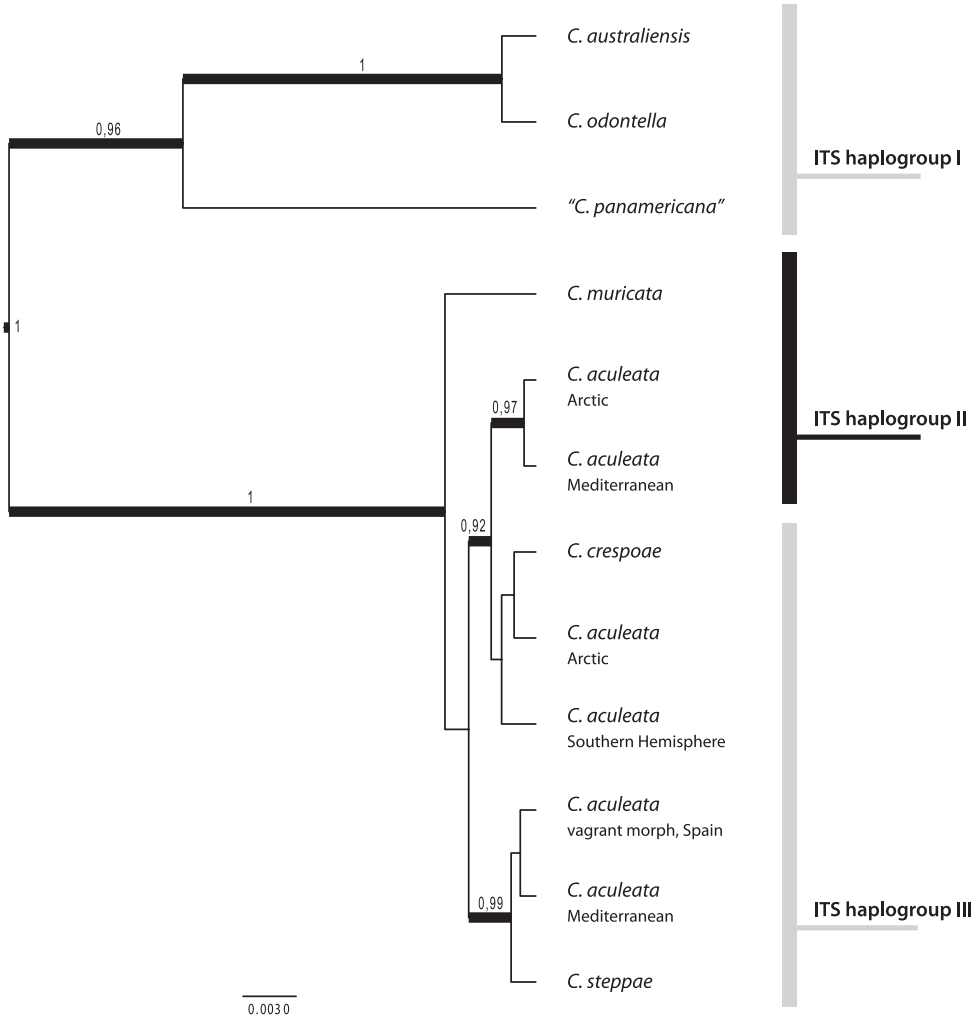

Figure 3. Species tree of the *C. aculeata* group inferred from ITS, GPD and mtLSU DNA sequences. For details of the analysis see under Material and methods.

monophyletic group. A species tree approach has the advantage that it does not require species to form monophyletic groups on gene trees and can account for the discordance between gene trees and species phylogeny that results from incomplete lineage sorting among closely related species.

With a high posterior probability, the Beringian and South American samples ("*C. panamericana*") are closely related to *C. odontella* and *C. australiensis* (Fig. 3). These two taxa are usually easily distinguished from other members of the *C. aculeata* group on account of their flattened thallus lobes. However, Kärnefelt (1986) already pointed out that some specimens from Alaska, Peru and Taiwan were lacking distinctly flattened lobes. We have so far not studied these specimens but assume that they belong to this group which is morphologically characterized by terete densely branched lobes

that are usually more blunt than in *C. aculeata* and have smaller pseudocyphellae. According to our field experience, typical *C. aculeata* is rare in Western North America. Six specimens identified by us as *C. muricata* were included in the analysis and formed an outgroup to the remaining OTUs. Two further clades within *C. aculeata* – *C. crespoae* – *C. steppae* receive high statistical support: (1) a group comprising *C. steppae*, the vagrant morphs and the Mediterranean samples from ITS haplogroup III, and (2) a group consisting of Arctic and Mediterranean individuals from ITS haplogroup II. *Cetraria crespoae*, Arctic samples from ITS haplogroup III and the southern hemispheric group form a third, poorly supported group that is nested between these two clades. The most likely candidate for a separate species within *C. aculeata* s. lat. appears to be a *C. steppae* that also comprises specimens without norstictic acid and the vagrant morphs from Spain. More data are necessary to decide the status of *C. crespoae* and the remaining groups within *C. aculeata*. Because the genetic distances between all of these groups are minimal, we treat them as "*C. aculeata* s. lat." for the time being.

Historical demography and range shifts

Questions about species delimitations and possible cryptic species put aside, the wide distribution of many lichens raises questions about how they managed to colonise their often enormous ranges. *Cetraria aculeata* is widespread in the Arctic and temperate regions of the northern hemisphere and common in Patagonia, Tierra del Fuego, some subantarctic islands and the maritime Antarctic. Furthermore, it has been reported from southeast Australia (Kantvilas 1994) and New Zealand (Galloway 2007) and also occurs at low latitudes in various tropical mountain ranges, including the central Andes (Kärnefelt 1986) and the East African Mountains (Swinscow and Krog 1988). Where did *C. aculeata* first evolve and how did it manage to move between the hemispheres? In the absence of a fossil record, population genetic data offers the most useful source of information about historical range shifts. The basic assumptions of the simplest approaches are that genetic diversity accumulates over time in stable populations and that population size bottlenecks reduce genetic diversity by removing alleles or haplotypes from a population (Nei et al. 1975). Bottlenecks can be caused by different historical events, for example migration or long range colonisation of areas by just a few individuals of a species, population size reductions by habitat fragmentation or climatic changes etc. Many studies have shown that large, refugial areas with a long historical continuity usually harbour more genetic diversity than newly colonised regions (Hewitt 1999). Comparisons of genetic diversity or allelic richness thus help to distinguish between old "source areas" and newly colonised regions within a species' range. The spatial distribution of certain haplotypes might then offer additional information that often helps in identifying dispersal routes.

The comparison of genetic variabilities of the mycobiont of *C. aculeata* (Domaschke et al. 2012) over much of its range showed highest diversity in the Arctic and a marked decline towards the Antarctic (Fig. 4) indicating that *C. aculeata* s. lat. evolved in the

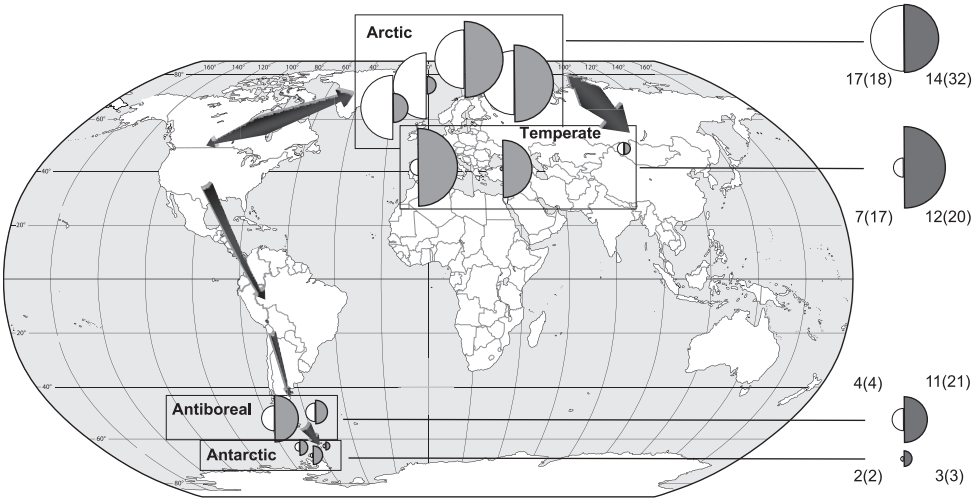

Figure 4. Nucleotide diversity (π) of different populations of photobionts (grey) and mycobionts (white) based on ITS sequences from 222 thalli of *C. aculeata* (data from Domaschke et al. 2012). Size of the semi-circles is proportional to the diversity level. Circles on the right summarize diversity levels within regions. Numbers of observed haplotypes and estimated absolute numbers following Chao 1 (in brackets) are added. Arrows indicate inferred historical migration events.

north and colonised the southern hemisphere from there. Antarctic populations are genetically almost uniform, which suggests that this continent was perhaps colonised in a single event and that genetic exchange with the genetically more diverse southern tip of South America is absent or very rare. The pattern for the photobiont is similar, although the Mediterranean part of the range is considerably more diverse than in the mycobiont. Mediterranean populations of both symbionts are genetically well separated from the rest of the populations (completely for the photobiont). The species tree in Fig. 3 with an entirely Mediterranean group at the base of the *C. aculeata* clade and the Mediterranean *C. crespoae* and ITS haplogroup II spread over the remainder of the clades suggest that *C. aculeata* s. lat. first evolved in the Mediterranean and spread to the Arctic from there.

Because only few haplotypes are shared among both Hemispheres (Fig. 3 in Fernández-Mendoza et al. 2011 and Fig. 1 in Domaschke et al. 2012), the disjunction was assumed to result from a single colonisation event. Fernández-Mendoza and Printzen (2013) have recently used coalescent-based migration modelling (Beerli and Palczewski 2010) and a modified version of stochastic character mapping that allows temporally explicit reconstruction of character changes to study the expansion of *C. aculeata* into its current range. They found evidence for a Pleistocene dispersive burst in which a population size expansion led to the acquisition of a South-American range that culminated in the colonization of the Antarctic. Their results suggest that the transition from the Arctic into Patagonia preceded that into the Central Andes. Either Patagonian populations became genetically isolated from an Andean dispersal pathway into South America, while the Central Andes still received immigrants from northern populations. Or the small and scattered patches of suitable habitat in the Central An-

des were extinguished during glacial maxima and later re-colonized from the North, while Patagonia with its widely ice-free areas east of the Andes maintained larger and genetically diverse populations of *C. aculeata*.

Adaptation or acclimation of *C. aculeata* to environmental conditions in temperate and polar environments

In contrast to most other polar organisms, which are highly adapted to the extreme climatic conditions in which they live, most polar lichens are not restricted to cold habitats. Many polar species are widespread at lower latitudes, some are confined to high mountains (Galloway and Aptroot 1995), while others extendend into a wide range of other habitats. *Cetraria aculeata* s. lat. is present not only in polar ecosystems, but also in coastal sand dunes, woodlands and steppes in temperate and semiarid regions around the Mediterranean and in Central Asia. Its occurrence in such widely differing biomes indicates an unusually wide ecological niche which can either result from an extreme physiological plasticity or from niche differentiation between sister taxa. At least to a certain degree, the wide ecological niches of some lichen species could be explained by their poikilohydric life style, avoiding environmental stress by simply deactivating their metabolism (Longton 1988; Pannewitz et al. 2003). However, an ecological niche like that of *C. aculeata* still requires that the species can modulate its physiological performance (Kappen 2000; Pannewitz et al. 2006) and maintain positive net photosynthesis during short phases of biological activity ("resource pulses") under very different climatic regimes (Pannewitz et al. 2003, 2006; Yang et al. 2008). It is tempting to assume that individuals of *C. aculeata* in different parts of its range are genetically adapted to the widely differing environmental conditions. Different temperature optima for photosynthesis have been reported for polar and temperate lichen species (Kappen 1988). Similarly, thalli of the same species sampled in different biomes differed in photosynthetic response and growth rate (Murtagh et al. 2002). Sonesson et al. (1992), for example, concluded that populations of *Nephroma arcticum* (L.) Torss. comprise different ecotypes and Schipperges et al. (1995) suggested high phenotypic plasticity and genetically determined ecophysiological differences between populations of *Flavocetraria nivalis* (L.) Kärnefelt & A. Thell.

So far, the only published ecophysiological study on *C. aculeata* is that by Pérez-Ortega et al. (2012) that compared morphologically divergent thalli within a few localities in Spain (see above). Domaschke and Printzen (2011) studied ecophysiological properties of *C. aculeata* thalli from Antarctica, Svalbard, Spain and Germany and found that Polar and temperate populations differed in maximal net photosynthesis rates (NPR), light compensation points, light saturation points and temperature optima (Fig. 5). NPR was significantly lower in the Polar than in the temperate populations, which could partly be explained by lower chlorophyll concentrations per unit dry mass in the Polar samples. The temperature optima,

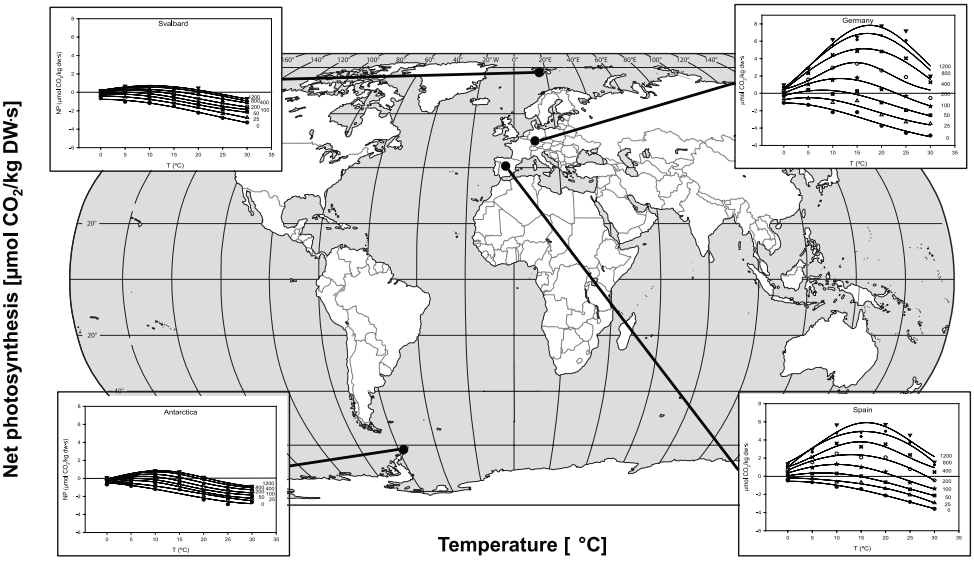

Figure 5. Dependency of net photosynthesis (NP) on temperature at various photon flux densities (0 to 1200 µmol m^{-2} s^{-1}) related to different geographical origins of *C. aculeata*. Figures are based on measurements of 7-8 thalli per location. Further details under Material and methods.

however, are largely unaffected by chlorophyll levels. Because whole lichen thalli were measured, it is not possible to disentangle the contributions of the mycobiont's respiration and the photobiont's photosynthesis and respiration to overall net photosynthesis. Experiments on isolated photobiont cultures are currently under way (Domaschke, in prep.) to assess whether one or both symbionts are responsible for the observed differences. Whether these physiological differences are genetically fixed adaptations or the result of acclimation can only be studied by transplantation experiments which are currently carried out (Domaschke, in prep.). At any rate, the available evidence is so far not in conflict with the hypothesis that genetically different individuals of *C. aculeata* in Polar and temperate biomes are adapted to local environmental conditions.

The symbiotic aspect

Molecular studies have demonstrated that lichen mycobionts can associate with different photobionts (Kroken and Taylor 2000; Opanowicz and Grube 2004; Piercey-Normore 2004; Blaha et al. 2006) and that photobiont switching is common (Piercey-Normore and Depriest 2001; O'Brien et al. 2005; Nelsen and Gargas 2008; Wornik and Grube 2010). It is evident that the association of mycobionts with locally adapted photobionts could contribute greatly to the ability of *C. aculeata* to colonise ecologically different biomes. Evidence for an ecological influence on symbiotic interactions in lichens is indeed accumulating (Blaha et al. 2006; Casano

et al. 2011; Fernández-Mendoza et al. 2011; Peksa and Skaloud 2011). A number of different hypotheses and theories have been developed to explain the dynamics of various symbiotic systems (Reshef et al. 2006; Rosenberg et al. 2007; Rodriguez et al. 2008; Gilbert et al. 2010). They all agree in that the symbiotic lifestyle is likely to increase the adaptive and evolutionary potential of symbiotic holobionts. A symbiotic host may adapt to changing environmental conditions by "outsourcing" (Gilbert et al. 2010) parts of its stress response to the symbiotic partners (habitat adapted symbiosis, Rodriguez et al. 2008). The observed shifting of symbiotic partners in coral-*Symbiodinium* associations has triggered the coral probiotic hypothesis (Reshef et al. 2006) and the hologenome theory of evolution (Rosenberg et al. 2007; Gilbert et al. 2010). At present, there is no evidence for fast and repeated photobiont switches in lichens. But rapid symbiont switches would certainly enable the mycobiont to react much faster to environmental changes than the slow evolutionary processes of mutation and selection.

The observation that North and South Polar populations of *C. aculeata* are genetically more similar to each other than to the temperate populations induced Fernández-Mendoza et al. (2011) to study whether the genetic structure of mycobiont and photobiont populations is best explained by geographic distances or by climatic differences between sampling localities. The major problem with such studies is that differences in climate and spatial distance may be correlated over large geographical distances. Variation partitioning (Borcard et al. 1992) showed that climate alone or in combination with geographical distance explained a major part of the genetic variability of the photobiont, but not the mycobiont (Fig. 6). On the other hand, *C. aculeata* is a largely asexual species. Supposedly, it propagates mainly by thallus fragments containing both symbionts. It can therefore be assumed that at least part of the genetic structure of the photobiont is correlated to that of the mycobiont. Indeed co-dispersal with the mycobiont accounts for an almost equally high proportion of the genetic structure of the photobiont (Fig. 6, bottom). This indicates that photobiont switches, which must obviously have occurred because of the partly incongruent genetic patterns of both symbionts, are not happening very often. The fact that the temperate populations of both symbionts are genetically isolated from Polar populations (see above), suggests that a historic photobiont switch enabled *C. aculeata* to colonise its wide ecological niche.

It has recently become clear that the lichen symbiosis also harbours a large diversity and abundance of lichen-associated bacterial communities (reviewed by Grube and Berg 2009). These communities are often dominated by *Alphaproteobacteria* (Cardinale et al. 2008; Bates et al. 2011; Hodkinson et al. 2012) although other bacterial groups may also be fairly abundant. Lichen-associated bacterial communities can be species-specific (Grube et al. 2009; Hodkinson et al. 2012) indicating that they might play an important role in the symbiosis. A preliminary analysis of *Alphaproteobacteria* associated with *C. aculeata* revealed that their community structure is similar to the genetical patterns observed in the photobiont with several OTUs being shared among North and South Polar populations (Printzen et al. 2012). This

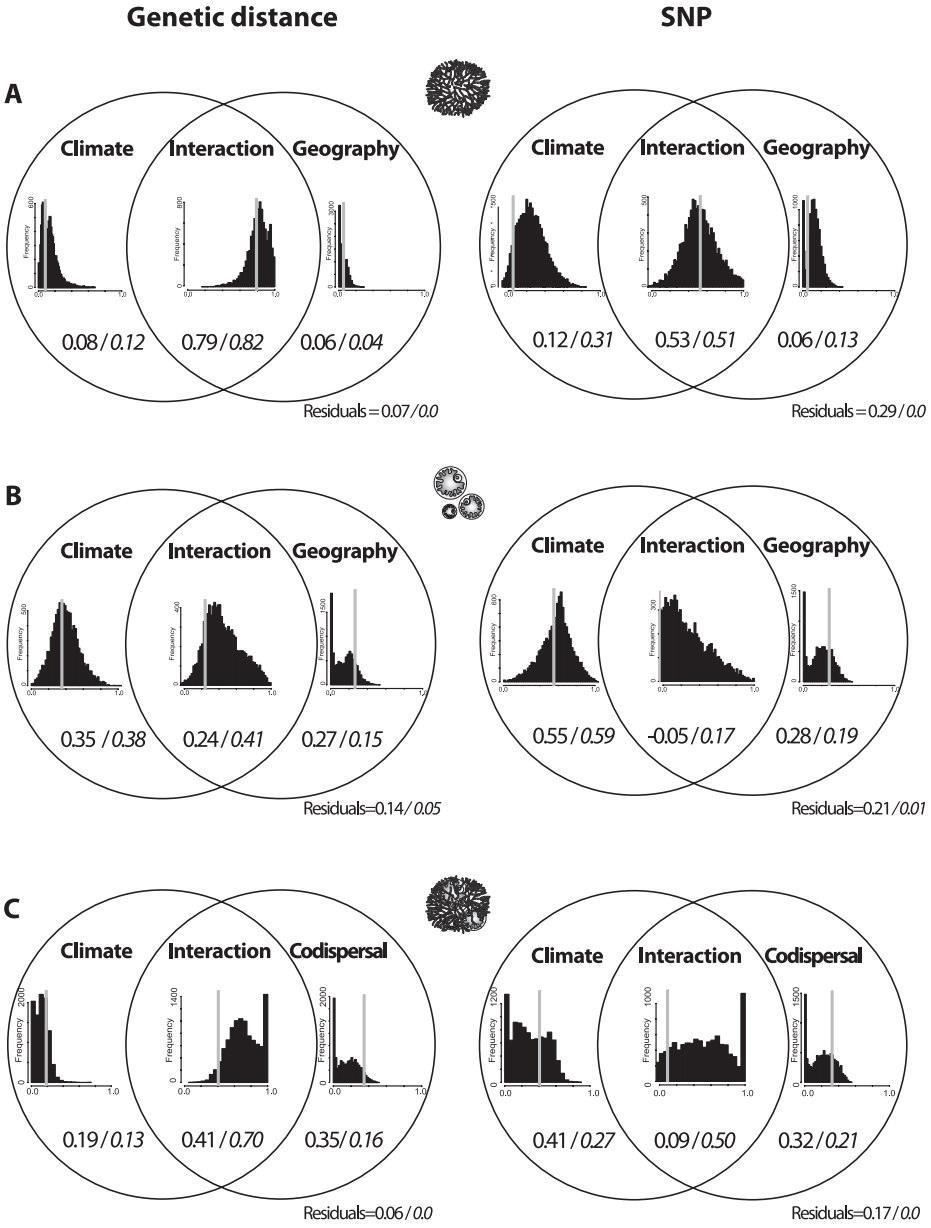

Figure 6. Variation partitioning diagrams using climate, geography and co-dispersal as explanatory components for the genetic structure of mycobiont and photobiont populations of *Cetraria aculeata*. Top, mycobiont; Center, photobiont, explanatory variables climate and spatial distances; Bottom, photobiont, explanatory variables climate and co-dispersal with the mycobiont. Left: genetic structure measured as pair-wise genetic distances between populations. Right: genetic structure measured as SNP composition. Numbers in the Venn diagrams indicate the fraction of the variation that is explained by the respective component or the intersection of components, with medians of the bootstrap analyses in italics. Results of the bootstrap analyses are also displayed in the bar charts with proportions of explained variation on the x-axes. Grey bars indicate results based on the original (non-resampled) data sets. From Fernández-Mendoza et al. (2011).

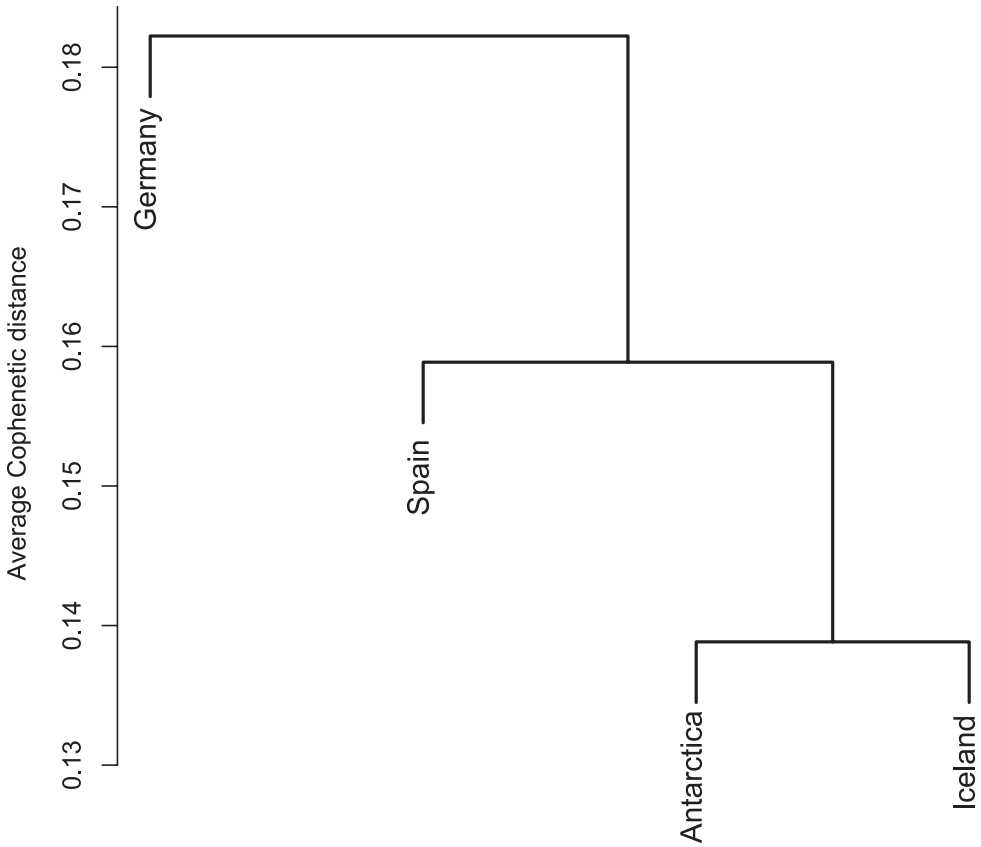

Figure 7. Community classification of four populations of Alphaproteobacteria associated with *C. aculeata*. The bar on the left indicates the value of the cophenetic distances between communities. From Printzen et al. (2012).

was at the same time the first evidence for bipolar distributions of lichen-associated bacteria. A community classification showed that bacterial populations from Iceland and the maritime Antarctic are more similar to each other than to temperate populations from Spain and Germany (Fig. 7).

Hill (2009) recently suggested that "although ... the mycobiont may acquire strains (species, varieties, forms or genotypes) [of photobionts] that are more suitable than others .. this is unlikely [to] have any evolutionary consequence." Our results show that acquiring a new photobiont may have wide-ranging evolutionary consequences for a mycobiont by extending its ecological niche. In the case of *C. aculeata* it has either enabled an ancestral species adapted to arctic conditions to colonise temperate and semi-arid biomes or vice versa. In spite of their relative geographic proximity, extant mycobiont populations of *C. aculeata* from the Mediterranean and the Arctic are genetically almost completely isolated. This suggests that habitat-adapted symbiosis (Rodriguez et al. 2008) might trigger speciation processes in lichens. The role of bacterial symbionts in this process is so far entirely unclear and requires more intensive attention.

Material and methods

Sampling, methods of DNA isolation, sequencing and PCR subcloning of bacterial communities as well as statistical analyses are outlined in Fernández-Mendoza et al. (2011), Domaschke et al. (2012) and Printzen et al. (2012).

Calculation of the species tree in Fig. 2 was based on 83 ITS, 71 mtLSU and 64 GPD sequences from the *C. aculeata* group. Sequences were assigned to eleven species and genetic lineages: i) *Cetraria odontella*, ii) *C. australiensis*, iii) *C. panamericana* ined., iv) *C. muricata*, v) *C. crespoae*, vi) *C. steppae*, and six geographic and genetic groups of *C. aculeata*; vii) Mediterranean haplogroup II, viii) Mediterranean haplogroup III, ix) Southern hemisphere, x) Northern boreoarctic haplogroup II and xi) Northern boreoarctic haplogroup III. We used the species tree ancestral reconstruction method *BEAST (Heled and Drummond 2010) implemented in BEAST v 1.7.1 (Drummond and Rambaut 2007), to reconstruct a mixed species/population tree. Each population/species was modelled under a separate coalescent prior with a constant root and a continuous population size. Optimum substitution models were estimated using jMODELTEST (Guindon and Gascuel 2003; Posada 2008). The analysis of exploratory runs for each locus suggested that using a site heterogeneity model (Gamma) was not adequate which hence was not used in the reconstructions. For all loci, nucleotide frequencies were also estimated in the analysis. Clock like evolutionary models were used without calibration points. The adequacy of imposing a strict molecular clock was assessed for each locus using the ML test implemented in MEGA5 (Tamura et al. 2011). We imposed a strict clock model for mtLSU and GPD, and an uncorrelated exponential relaxed clock for ITS. The analysis was run for 50×10^6 generations; parameters were sampled every 2.5×10^3th generation. Convergence and posterior parameter distributions were examined using Tracer v1.5 (Rambaut and Drummond 2007). The resulting distributions were combined in log-Combiner (Drummond and Rambaut 2007), and after an adequate burn-in fraction was selected, maximum clade credibility trees were calculated for the reconstructed species and gene trees.

CO_2 exchange measurements were performed on lichen thalli that were carefully washed with mineral water and cleaned from fragments of other lichen species. Dry thalli were reactivated for 72 hours in a climate chamber (light intensity 100 µmol m^{-2} s^{-1}, 12 hours light / dark cycle, 15 °C, humidification every 24 h with mineral water). CO_2 measurements were performed with a compact minicuvette system (CMS 400, WALZ, Effeltrich, Germany) under controlled temperature, light and humidity conditions. For each population 7–8 replicates were investigated. Prior to all measurements, samples were soaked in mineral water for 20 minutes to ensure full hydration of thalli. Optimal water contents were determined for 4 individuals per population by studying the change of the photosynthetic performance during the desiccation process of the lichen at a constant temperature of 15 °C and photon flux densities of 0 and 400 µmol m^{-2} s^{-1}. The response of net CO_2 exchange was measured at optimal water content at temperatures between 0 and 30°C and photosynthetic flux densities (PPFD) of 0, 25, 50, 100, 200, 400, 800 and 1200 µmol m^{-2} s^{-1}. PPFD response curves were analysed by statistical fitting of a Smith function (Green et al. 1997) using SigmaPlot 10.0 (Chicago, Illinois, USA).

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References

Baas-Becking GML (1934) Geobiologie of inleiding tot de milieukunde. Van Stockum & Zoon, Den Haag.

Bates ST, Cropsey GWG, Caporaso JG, Knight R, Fierer N (2011) Bacterial communities associated with the lichen symbiosis. Applied and Environmental Microbiology 77: 1309–1314. doi: 10.1128/AEM.02257-10

Beerli P, Palczewski M (2010) Unified framework to evaluate panmixia and migration direction among multiple sampling locations. Genetics 185: 313–326. doi: 10.1534/genetics.109.112532

Blaha J, Baloch E, Grube M (2006) High photobiont diversity associated with the euryoecious lichen-forming ascomycete *Lecanora rupicola* (Lecanoraceae, Ascomycota). Biological Journal of the Linnean Society 88: 283–293. doi: 10.1111/j.1095-8312.2006.00640.x

Borcard D, Legendre P, Drapeau P (1992) Partialling out the spatial component of ecological variation. Ecology 73: 1045– 1055. doi: 10.2307/1940179

Cardinale M, Vieira de Castro J, Müller H, Berg G, Grube M (2008) In situ analysis of the bacterial community associated with the reindeer lichen *Cladonia arbuscula* reveals predominance of *Alphaproteobacteria*. FEMS Microbiology Ecology 66: 63–71. doi: 10.1111/j.1574-6941.2008.00546.x

Casano LM, del Campo EM, García-Breijo FJ, Reig-Armiñana J, Gasulla F, del Hoyo A, Guéra A, Barreno E (2011) Two *Trebouxia* algae with different physiological performances are ever-present in lichen thalli of *Ramalina farinacea*. Coexistence versus competition? Environmental Microbiology 13: 806–818. doi: 10.1111/j.1462-2920.2010.02386.x

Cordellier M, Pfenninger M (2009) Inferring the past to predict the future: climate modelling predictions and phylogeography for the freshwater gastropod *Radix balthica* (Pulmonata, Basommatophora). Molecular Ecology 18: 534–544. doi: 10.1111/j.1365-294X.2008.04042.x

Cox C, Moore P (2010) Biogeography: An ecological and evolutionary approach. 8th ed. John Wiley & Sons, Hoboken.

Darwin C (1872) The origin of species by means of natural selection. 6th ed. John Murray, London.

Domaschke S, Fernández-Mendoza F, García M, Martín M, Printzen C (2012) Low genetic diversity in Antarctic populations of the lichen-forming ascomycete *Cetraria aculeata* and its photobiont. Polar Research 31. doi: 10.3402/polar.v31i0.17353

Domaschke S, Printzen C (2011) Photobiont adaptation to extreme environmental conditions. 4th International Conference on Polar and Alpine Microbiology, Ljubljana, 95.

Drummond AJ, Rambaut A (2007) BEAST: Bayesian evolutionary analysis by sampling trees. BMC Evolutionary Biology 7: 1–214.

Feldhaar H (2011) Bacterial symbionts as mediators of ecologically important traits of insect hosts. Ecological Entomology 36: 533–543.

Fernández-Mendoza F, Printzen C (2013) Pleistocene expansion of the bipolar lichen Cetraria aculeata into the Southern hemisphere. Molecular Ecology 22: 1961–1983. doi: 10.1111/mec.12210

Fernández-Mendoza F, Domaschke S, Garcia MA, Jordan P, Martin MP, Printzen C (2011) Population structure of mycobionts and photobionts of the widespread lichen *Cetraria aculeata*. Molecular Ecology 20: 1208–1232. doi: 10.1111/j.1365-294X.2010.04993.x

Galloway D, Aptroot A (1995) Bipolar lichens: A review. Cryptogamic Botany 5: 184–191.

Galloway DJ (2007) Flora of New Zealand: Lichens, including lichen-forming and lichenicolous fungi. 2nd ed. Manaaki Whenua Press, Landcare Research.

Gilbert SF, McDonald E, Boyle N, Buttino N, Gyi L, Mai M, Prakash N, Robinson J (2010) Symbiosis as a source of selectable epigenetic variation: taking the heat for the big guy. Philosophical Transactions of the Royal Society London, B, Biological Sciences 365: 671–678. doi: 10.1098/rstb.2009.0245

Goward T (1999) The lichens of British Columbia. Illustrated keys. Part 2 - fruticose species. British Columbia, Ministry of Forests.

Gray A (1859) Diagnostic characters of new species of phanogamous plants, collected in Japan by Charles Wright, botanist of the U. S. North Pacific Exploring Expedition. (published by request of Captain John Rodgers, Commander of the Expedition.) with observations upon the relations of the Japanese flora to that of North America, and of other parts of the northern Temperate Zone. Memoirs of the American Academy of Arts, new series 6: 377–452.

Green T, Büdel B, Meyer A, Zellner H, Lange O (1997) Temperate rainforest lichens in New Zealand: light response of photosynthesis. New Zealand Journal of Botany 35: 493–504. doi: 10.1080/0028825X.1987.10410173

Grube M, Berg G (2009) Microbial consortia of bacteria and fungi with focus on the lichen symbiosis. Fungal Biology Reviews 23: 72–85. doi: 10.1016/j.fbr.2009.10.001

Grube M, Cardinale M, de Castro JJ, Mueller H, Berg G (2009) Species-specific structural and functional diversity of bacterial communities in lichen symbioses. Isme Journal 3: 1105–1115. doi: 10.1038/ismej.2009.63

Guindon S, Gascuel O (2003) A simple, fast, and accurate algorithm to estimate large phylogenies by maximum likelihood. Systematic Biology 52: 696–704. doi: 10.1080/10635150390235520

Heled J, Drummond AJ (2010) Bayesian inference of species trees from multilocus data. Molecular Biology and Evolution 27: 570–580. doi: 10.1093/molbev/msp274

Hewitt G (1999) Post-glacial re-colonization of European biota. Biological Journal of the Linnean Society 68: 87–112. doi: 10.1111/j.1095-8312.1999.tb01160.x

Hill DJ (2009) Asymmetric co-evolution in the lichen symbiosis caused by a limited capacity for adaptation in the photobiont. Botanical Review 75: 326–338. doi: 10.1007/s12229-009-9028-x

Hodkinson BP, Gottel NR, Schadt CW, Lutzoni F (2012) Photoautotrophic symbiont and geography are major factors affecting highly structured and diverse bacterial communities in the lichen microbiome. Environmental Microbiology 14: 147–161. doi: 10.1111/j.1462-2920.2011.02560.x

Kantvilas G (1994) *Coelocaulon*. Flora of Australia. Lichens: Lecanorales 2, Parmeliaceae. Vol. 55. CSIRO Publishing, 36–37.

Kappen L (1988) Ecophysiological relationships in different climatic regions. In: Galun M (Ed) Handbook of lichenology. CRC Press, Boca Raton, 37–100.

Kappen L (2000) Some aspects of the great success of lichens in Antarctica. Antarctic Science 12: 314–324. doi: 10.1017/S0954102000000377

Kärnefelt I, Mattsson J, Thell A (1993) The lichen genera *Arctocetraria, Cetraria*, and *Cetrariella* (Parmeliaceae) and their presumed evolutionary affinities. Bryologist 96: 394–404. doi: 10.2307/3243869

Kärnefelt I (1986) The genera *Bryocaulon, Coelocaulon, Cornicularia* and formerly associated taxa. Opera Botanica 86: 1–90.

Kroken S, Taylor J (2000) Phylogenetic species, reproductive mode, and specificity of the green alga *Trebouxia* forming lichens with the fungal genus *Letharia*. Bryologist 103: 645–660. doi: 10.1639/0007-2745(2000)103[0645:PSRMAS]2.0.CO;2

Littman RA, Bourne DG, Willis BL (2010) Responses of coral-associated bacterial communities to heat stress differ with *Symbiodinium* type on the same coral host. Molecular Ecology 19: 1978–1990. doi: 10.1111/j.1365-294X.2010.04620.x

Longton RE (1988) The biology of polar bryophytes and lichens. Cambridge University Press, London and New York. doi: 10.1017/CBO9780511565212

Lynge B (1941) On *Neuropogon sulphureus* (König) Elenk., a bipolar lichen. Skrifter utgitt av det Norske Videnskaps-Akademi i Oslo. I. Matematisk-Naturvidenskapelige Klasse 10: 5–35.

Maestre FT, Bowker MA, Canton Y, Castillo-Monroy AP, Cortina J, Escolar C, Escudero A, Lazaro R, Martinez I (2011) Ecology and functional roles of biological soil crusts in semiarid ecosystems of Spain. Journal of Arid Environments 75: 1282–1291. doi: 10.1016/j.jaridenv.2010.12.008

Murtagh GJ, Dyer PS, Furneaux PA, Crittenden PD (2002) Molecular and physiological diversity in the bipolar lichen-forming fungus *Xanthoria elegans*. Mycological Research 106: 1277–1286. doi: 10.1017/S0953756202006615

Nei M, Maruyama T, Chakraborty R (1975) The bottleneck effect and genetic variability in populations. Evolution 29: 1–10. doi: 10.2307/2407137

Nelsen MP, Chavez N, Sackett-Hermann E, Thell A, Randlane T, Divakar PK, Rico VJ, Lumbsch HT (2011) The cetrarioid core group revisited (Lecanorales: Parmeliaceae). Lichenologist 43: 537–551. doi: 10.1017/S0024282911000508

Nelsen MP, Gargas A (2008) Dissociation and horizontal transmission of codispersing lichen symbionts in the genus *Lepraria* (Lecanorales: Stereocaulaceae). New Phytologist 177: 264–275.

O'Brien H, Miadlikowska J, Lutzoni F (2005) Assessing host specialization in symbiotic cyanobacteria associated with four closely related species of the lichen fungus *Peltigera*. European Journal of Phycology 40: 363–378. doi: 10.1080/09670260500342647

Opanowicz M, Grube M (2004) Photobiont genetic variation in *Flavocetraria nivalis* from Poland (Parmeliaceae, lichenized Ascomycota). Lichenologist 36: 125–131. doi: 10.1017/S0024282904013763

Pannewitz S, Schlensog M, Green TGA, Sancho LG, Schroeter B (2003) Are lichens active under snow in continental Antarctica? Oecologia 135: 30–38.

Pannewitz S, Green TGA, Schlensog M, Seppelt R, Sancho LG, Schroeter B (2006) Photosynthetic performance of *Xanthoria mawsonii* C. W. Dodge in coastal habitats, Ross Sea region, continental Antarctica. Lichenologist 38: 67–81. doi: 10.1017/S0024282905005384

Peksa O, Skaloud P (2011) Do photobionts influence the ecology of lichens? A case study of environmental preferences in symbiotic green alga *Asterochloris* (Trebouxiophyceae). Molecular Ecology 20: 3936–3948. doi: 10.1111/j.1365-294X.2011.05168.x

Peterson A, Soberon J, Sanchez-Cordero V (1999) Conservatism of ecological niches in evolutionary time. Science 285: 1265–1267. doi: 10.1126/science.285.5431.1265

Pérez-Ortega S, Fernández-Mendoza F, Raggio J, Vivas M, Ascaso C, Sancho L, Printzen C, De Los Ríos A (2012) Extreme phenotypic variation in *Cetraria aculeata* (lichenized Ascomycota): adaptation or incidental modification? Annals of Botany 109: 1133–1148. doi: 10.1093/aob/mcs042

Piercey-Normore MD, Depriest PT (2001) Algal switching among lichen symbioses. American Journal of Botany 88: 1490–1498. doi: 10.2307/3558457

Piercey-Normore MD (2004) Selection of algal genotypes by three species of lichen fungi in the genus *Cladonia*. Canadian Journal of Botany 82: 947–961. doi: 10.1139/b04-084

Poelt J (1963) Flechtenflora und Eiszeit in Europa. Phyton (Horn) 10: 206–215.

Posada D (2008) jModelTest: Phylogenetic model averaging. Molecular Biology and Evolution 25: 1253–1256. doi: 10.1093/molbev/msn083

Printzen C, Fernández-Mendoza F, Muggia L, Berg G, Grube M (2012) Alphaproteobacterial communities in geographically distant populations of the lichen *Cetraria aculeata*. FEMS Microbiology Ecology. 82: 316–325. doi: 10.1111/j.1574-6941.2012.01358.x

Printzen C (2008) Uncharted terrain: the phylogeography of arctic and boreal lichens. Plant Ecology and Diversity 1: 265–271. doi: 50870802328702

Rambaut A, Drummond A (2007) Tracer. http://beast.bio.ed.ac.uk/Tracer

Reshef L, Koren O, Loya Y, Zilber-Rosenberg I, Rosenberg E (2006) The coral probiotic hypothesis. Environmental Microbiology 8: 2068–2073. doi: 10.1111/j.1462-2920.2006.01148.x

Rodriguez R, Henson J, Van Volkenburgh E, Hoy M, Wright L, Beckwith F, Kim Y, Redman R (2008) Stress tolerance in plants via habitat-adapted symbiosis. Isme Journal 2: 404–416. doi: 10.1038/ismej.2007.106

Ronquist F, Sanmartín I (2011) Phylogenetic Methods in Biogeography. Annual Review of Ecology, Evolution and Systematics 42: 441–464. doi: 10.1146/annurev-ecolsys-102209-144710

Rosenberg E, Koren O, Reshef L, Efrony R, Zilber-Rosenberg I (2007) The role of microorganisms in coral health, disease and evolution. Nature Reviews Microbiology 5: 355–362. doi: 10.1038/nrmicro1635

Rosenberg E, Zilber-Rosenberg I (2011) Symbiosis and development: The hologenome concept. Birth Defects Research Part C: Embryo Today: Reviews 93: 56–66. doi: 10.1002/bdrc.20196

Schipperges B, Kappen L, Sonesson M (1995) Intraspecific variations of morphology and physiology of temperate to arctic populations of *Cetraria nivalis*. Lichenologist 27: 517–529.

Seifried J (2009) Genetische Diversität in beringischen und extraberingischen arktischen Populationen der Strauchflechte *Cetraria aculeata*. Diploma thesis, Frankfurt am Main: Goethe-Universität, Frankfurt.

Sonesson M, Schipperges B, Carlsson B (1992) Seasonal patterns of photosynthesis in alpine and subalpine populations of the lichen *Nephroma arcticum*. Oikos 65: 3–12. doi: 10.2307/3544881

Spribille T (2011) Circumboreal Lichen Diversification: Phylogenetic and phylogeographic studies in the genus *Mycoblastus*. Ph.D. thesis, Institute of Plant Sciences, Karl-Franzens-Universität Graz, Graz.

Swinscow T, Krog H (1988) Macrolichens of East Africa. British Museum (Natural History), London.

Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S (2011) MEGA5: Molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Molecular Biology and Evolution 28: 2731–2739. doi: 10.1093/molbev/msr121

Thell A, Högnabba F, Elix JA, Feuerer T, Kärnefelt I, Myllys L, Randlane T, Saag A, Stenroos A, Ahti T, Seaward MRD (2009) Phylogeny of the cetrarioid core (Parmeliaceae) based on five genetic markers. Lichenologist 41: 489–511. doi: 10.1017/S0024282909990090

Thell A, Stenroos S, Feuerer T, Kärnefelt I, Myllys L, Hyvönen J (2002) Phylogeny of cetrarioid lichens (Parmeliacdae) inferred from ITS and b-tubulin sequences, morphology, anatomy and secondary chemistry. Mycological Progress 1: 335–354. doi: 10.1007/s11557-006-0031-x

Thell A, Stenroos S, Myllys L (2000) A DNA study of the *Cetraria aculeata* and *C. islandica* groups. Folia Cryptogamica Estonica 36: 95–106.

Turesson G (1922) The genotypical response of plants to the habitat. Hereditas 3: 211–350. doi: 10.1111/j.1601-5223.1922.tb02734.x

Waltari E, Hijmans R, Peterson A, Nyári Á, Perkins S, Guralnick R (2007) Locating pleistocene refugia: Comparing phylogeographic and ecological niche model predictions. Plos One 2:e563. doi: 10.1371/journal.pone.0000563

Wiens J (2004) Speciation and ecology revisited: Phylogenetic niche conservatism and the origin of species. Evolution 58: 193–197.

Wirtz N, Printzen C, Lumbsch HT (2008) The delimitation of Antarctic and bipolar species of neuropogonoid *Usnea* (Ascomycota, Lecanorales): a cohesion approach of species recognition for the *Usnea perpusilla* complex. Mycological Research 112: 472–484. doi: 10.1016/j.mycres.2007.05.006

Wirtz N, Printzen C, Lumbsch HT (2012) Using haplotype networks, estimation of gene flow and phenotypic characters to understand species delimitation in fungi of a predominantly Antarctic *Usnea* group (Ascomycota, Parmeliaceae). Organisms Diversity and Evolution 12: 17–37. doi: 10.1007/s13127-011-0066-y

Wornik S, Grube M (2010) Joint dispersal does not imply maintenance of partnerships in lichen symbioses. Microbial Ecology 59: 150–157. doi: 10.1007/s00248-009-9584-y

Yahr R, Vilgalys R, Depriest PT (2006) Geographic variation in algal partners of *Cladonia subtenuis* (Cladoniaceae) highlights the dynamic nature of a lichen symbiosis. New Phytologist 171: 847–860. doi: 10.1111/j.1469-8137.2006.01792.x

Yang LH, Bastow JL, Spence KO, Wright AN (2008) What can we learn from resource pulses? Ecology 89: 621–634. doi: 10.1890/07-0175.1