Perspectives
in Plant Ecology,
Evolution and
Systematics

Volume 30
2018
ISSN 1433-8319

ScienceDirect

Available online at www.sciencedirect.com

Contents

Special issue: Alpine and arctic plant communities: a worldwide perspective
Guest editors: Fabien Anthelme, Montpellier, France Sébastien Lavergne, Grenoble, France

Contents continued on inside back cover

Abstracted/Indexed in
BIOSIS database; Current Contents®/Agriculture, Biology & Environmental Sciences; Ecological Abstracts; Elsevier BIOBASE (Current Awareness in Biological Sciences); GEOBASE; Geo Abstracts; NISC - National Information Services Corporation; Referativnyi Zhurnal; SciExpanded; SciSearch; Science Citation Report; Scopus

Perspectives in Plant Ecology, Evolution and Systematics · Volume **30** · 2018

ELSEVIER

Perspectives in Plant Ecology, Evolution and Systematics

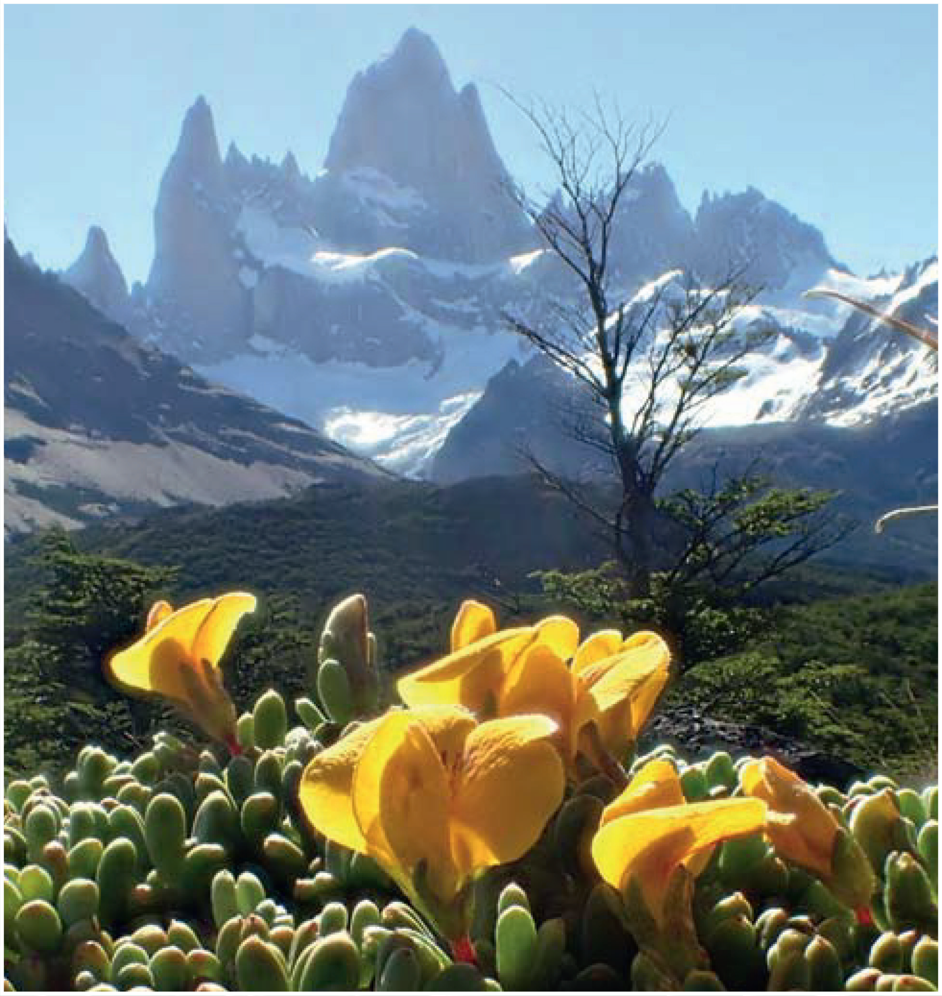
Rübel-Foundation, ***ETH*** Zürich
Supported by the European
Ecological Federation

2018

Special issue:
Alpine and arctic plant communities: a worldwide perspective

Guest editors:
Fabien Anthelme
Montpellier, France

Sébastien Lavergne
Grenoble, France



ISSN 1433-8319
Perspectives in Plant
Ecology, Evolution
and Systematics
30(2018) · pp. 1-132

www.elsevier.com/locate/ppees

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 1–5

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Alpine and arctic plant communities: A worldwide perspective☆

1. Background and context

High-elevation and high-latitude ecosystems experience among the most severe climates on Earth where plant and animal life can thrive (Körner, 2003). Termed arctic and alpine regions, they represent 8% of the global land area and occur on all continents (Chapin and Körner, 1995). Despite their extreme environmental harshness, these ecosystems harbour relatively important biodiversity and endemism, with high diversity turnover along steepest environmental gradients. Some of these ecosystems have long been the focus of ecological studies and captured the attention of some of the most popular naturalists and biologists during the last few centuries (reviewed by Billings and Mooney, 1968; Körner, 2003). The reasons for such a scientific interest are multiple. From a strictly conceptual viewpoint, alpine and arctic ecosystems exhibit relatively simple vegetation patterns, with little vertical stratification, making it easier to characterize stress and disturbance effects on community and ecosystem functioning (Chapin and Körner, 1995). They provide "unsurpassed possibilities for comparative ecological research" because of steep elevation and topographical gradients (Körner, 2003). Their relative isolation allows studying ecological systems at diverse spatial scales, such as local scale communities, along environmental gradients, or entire mountain ranges (e.g. Chalmandrier et al., 2017; Marx et al., 2017). This isolation combined with the common historical setting of most mountain ranges also allows exploring questions related to the origins of biodiversity patterns, local speciation and endemism (Molina-Venegas et al., 2015, 2017). Alpine and arctic ecosystems are key providers and regulators of water resources to human societies (e.g., *water towers*; Körner, 2003), and also serve as biomass reserve for livestock grazing (Jacobsen and Dangles, 2017). Finally, the exceptionally high capacity of some arctic and alpine ecosystems to stock carbon makes them efficient carbon pools (Segnini et al., 2010; Tarnocai et al., 2009).

Although the accumulated knowledge on arctic and alpine environments is now substantial, there remains some unexplored alpine areas, hence current knowledge on alpine systems is geographically unbalanced (Fig. 1). While some important efforts have been devoted to studying arctic and temperate alpine ecosystems, mainly in the northern hemisphere, some important alpine areas such as the Himalayas and the Andes have long remained understudied. Tropical alpine regions are probably the most diverse alpine regions of the world, with unparalleled rates of speciation and resulting high local endemism (Anthelme et al., 2014; Madriñán et al., 2013), but have been particularly overlooked by researchers (Fig. 1). Such a discrepancy would be of minor importance if all arctic and alpine ecosystems were facing similar constraints in comparable climatic and edaphic environments. In fact, despite important common features such as cold temperature, infertile soils and frequent drought (Körner, 2003), a close examination shows strong dissimilarities between alpine regions of the world, mainly in relation to latitudinal position and topography (Nagy and Grabherr, 2009). For instance, prevailing climates in tropical alpine areas are much less seasonal, with daily variations in temperatures exceeding annual variations. As a consequence, winter snow cover, which is considered as a major driver of alpine and arctic vegetation structure (Carlson et al., 2015; Rixen et al., 2014) is almost absent in the tropics, thus affecting freezing and drought frequency. In addition, the inversion of precipitation gradients above 3000–3500 m in most tropical alpine systems generates tropical alpine deserts while annual precipitation increases with elevation in most extra-tropical alpine environments (Anthelme and Dangles 2012). In view of these differences, bridging this latitudinal knowledge gap between alpine regions is an important challenge, for both understanding how climate shape biodiversity patterns in alpine regions and drawing scenarios for the future of alpine and arctic biodiversity.

An important modern challenge for the study of alpine and arctic regions is obviously to understand and predict the consequences of the current high pace and expected high magnitude of global changes on ecosystem properties. A large body of evidence demonstrated that arctic environments are disproportionately impacted by climate warming, a phenomenon known as *arctic amplification* (Pachauri et al., 2014; IPPC report). A less known but now increasingly documented phenomenon is also the expected higher warming at higher elevations (MRI − Mountain Research Initiative EDW Working Group, 2015; Wang et al., 2016). Accordingly, the impact of warming should be exacerbated in tropical alpine systems, which are mostly located above 4000 m a.s.l. These issues are particularly alarming for the conservation of biodiversity and ecosystem functioning. The majority of alpine regions are highly fragmented, isolated and restricted to a very small portion of land surface, thus threatening most species adapted to the narrow environmental conditions of high elevations and high latitudes. Also, climate warming may induce a release of carbon and methane captured in permafrosts, with expected large feedbacks to global climate. This calls for more research about how climatic changes (both past and

---

☆ This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

https://doi.org/10.1016/j.ppees.2017.12.002

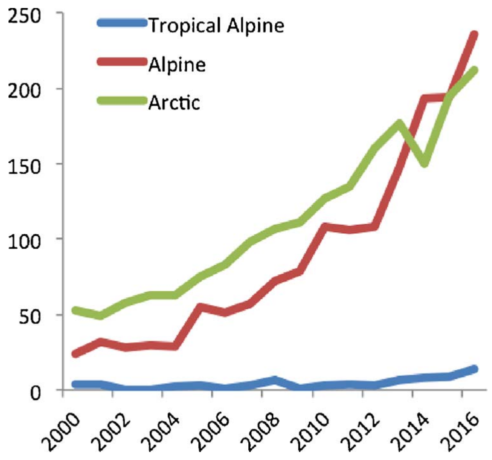


Fig. 1. Number of publications focusing on arctic/alpine ecology between the years 2000 and 2016 (Web of Science). Research strings used for alpine: alpine OR high-elevation OR high-altitude (title) AND ecosystem* (topic). Research strings used for arctic: arctic OR high-latitude OR Antarctic (title) AND ecosystem* NOT marine (topic), excluding field "oceanography. Research strings used for tropical alpine: tropical alpine OR tropical and high elevation OR tropical and high-latitude (title) AND ecosystem* (topic). Results have later been refined manually, excluding papers out of scope. Analysis made on the 29 of June 2017.

contemporary) affect the biodiversity and functioning of alpine and arctic ecosystems. These thus constitute important sentinel ecosystems where much fundamental research is motivated by the need to understand and anticipate the effects of global changes.

Apart from modern societal challenges, alpine and arctic ecosystems raise a number of fundamental questions. The scope of these questions is very large, spanning the entire spectrum of ecological and evolutionary theory, and certainly cannot be tackled within a sole journal special issue, not even a single book. These key fundamental questions concern: (i) the evolutionary origins and historical build-up of species rich biomes and biodiversity hotspots, (ii) the effect of quaternary climatic oscillations on species distributions and biodiversity patterns, (iii) the ecological and micro-evolutionary drivers of species distributions and species range shifts in changing environments, (iv) the mechanisms of community assembly and species coexistence at local spatial scales, and the nature of biotic interactions (both plant–plant and plant-animal interactions). High elevation and latitude ecosystems provide very suitable settings for teasing apart these different mechanisms at varied temporal and spatial scales, and the conceptual contributions of arctic and alpine studies clearly goes beyond the sole study of these biomes.

2. Objectives and themes of this special issue

This special issue presents a selection of papers exploring some of the aforementioned key topics, following a symposium entitled 'High-elevation ecosystems', held at the Ecosummit conference, Montpellier, August 29th – September 1st 2016. The selected papers present cutting-edge research on arctic and alpine plant communities along a double latitudinal and altitudinal gradient, thus reflecting the biogeographic continuum between arctic and alpine ecosystems (see Fig. 2a). These regions include arctic areas in Norway, high alpine areas of Europe, New Zealand, Himalayas, but also high tropical alpine areas. These studies have been selected because they provide significant advances to current ecological theories and improve our understanding of global change effects on plant communities. They also describe a number of original methods, which might be inspiring for future research.

Based on authors' extensive experience in alpine ecology, some papers of the special issue offer conceptual contributions centred on key climatic parameters critical to understanding arctic-alpine ecosystems. By reviewing up to 90 ways to measure temperature, Körner and Hiltbrunner (2018) highlight important pitfalls and propose a narrow selection of indices to depict the effects of temperature on plant distributions. They argue that mean annual temperature may be one of the less pertinent measurements because it does not consider the effects of snow cover and seasonal/diurnal

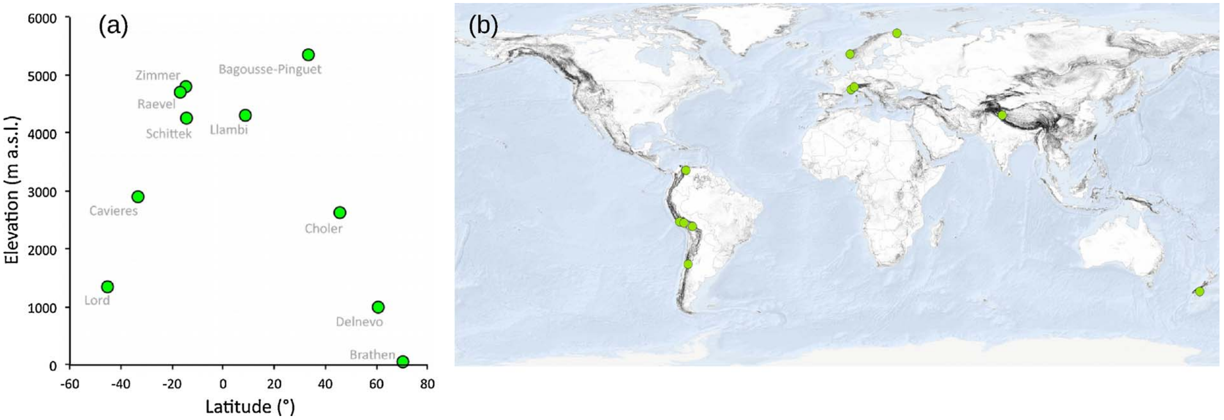


Fig. 2. Biogeographic situation of different empirical contributions to this special issue. (a) Diagram showing the continuous distribution of different study areas along latitude (x-axis) and elevation (y-axis) gradients worldwide. (b) Map depicting different alpine areas of the world according to a ruggedness scale (from light grey to black), following Körner et al. (2011) and the geographic position of all empirical papers included in this special issue. Note that the position of the two conceptual papers of this special issue, namely Körner and Hiltbrunner (2018); Graae et al. (2018), are not depicted in these two figures.

variations in temperature. Given the singularity of each site, the inherent complexity of alpine topography and the frequent occurrence of biotic thermal refuges, the authors recommend researchers to not rely on temperature extracted from extrapolated databases but rather measuring microclimate directly in their own target environments. Along the same line but with a purpose more focused on the structure of alpine vegetation under a warming climate, Choler (2018) demonstrates the need to measure temperature at a finer scale to account for the considerable local-scale environmental heterogeneity of alpine ecosystems. Focusing on the root zone temperature of temperate alpine plants, the author proposes that the combination of two temperature indices, namely growing degree days (GDD) and freezing degree days (FDD), represents a so-far undocumented "leading axis of habitat differentiation in the alpine zone", which allows drawing very realistic scenarios of plant community change in relation to snow cover duration. In a conceptual contribution aiming at characterizing the main survival options for plants in a changing environment ("stay or go"), Graae et al. (2018) combine macro and micro-climatic variables during the growing season and winter. They propose that plant communities in more heterogeneous landscapes are less prone to species range shifts under global warming than communities occurring in homogeneous landscapes. In turn, communities in homogeneous landscapes may face high extinction rates, resulting in unsaturated communities. This is expected by the authors to leave more opportunities for other species to immigrate locally due to lower interspecific competition. Interestingly, these three contributions agree on the fact that local environmental heterogeneity is a major driver of plant distribution and community assembly in a changing world. Because each of the three contributions focuses on alpine and arctic environments periodically covered by snowpack, the authors insist that confronting these models to the reality of tropical environments or dry steppes, where snow cover is very limited in time, is an important challenge for future research in alpine ecology (Choler, 2018).

Along with Choler (2018) and Graae et al. (2018), three other contributions focus on the effects of snow cover dynamics on plant communities. First, Lord et al. (2018) present a 14-year transplant experiment of vegetation turves across a natural snow cover gradient in the mountains of South Island, New Zealand. One interesting result, among others, was that communities were strongly altered by this environmental shift, both by reduced fitness of initial species and by colonization from surrounding vegetation. The authors conclude that biotic interactions and species-specific traits are key drivers of alpine vegetation change following the alteration of snow cover regimes. Second, assuming that changes in temperature and precipitation will impact the timing of snow cover, Delnevo et al. (2018) tested to what extent warmer and wetter conditions (according to IPPC scenarios) impact the phenology and growth of *Ranunculus acris* (Ranunculaceae) through changes in snow cover duration. To do so, they also performed a transplant experiment along environmental gradients in the mountains of southern Norway. Advanced snowmelt resulted in accelerated reproductive phenology, hence a greater exposure of reproductive organs to the risk of frost damage. Interestingly, those phenological changes experienced by the study species were both due to phenotypic plasticity and genetic changes, and were less pronounced at higher elevation (i.e. in the most limiting environment). This second result is echoed by another contribution to this issue by Cavieres and Sierra-Almeida (2018), who study seed germination of fifty Andean alpine plants from Chile. They find that the importance of cold stratification for seed germination was more pronounced for lower elevation species, suggesting that modifications of snow cover duration could affect disproportionately germination phenology of lower alpine species than the ones growing at higher elevations. These latter two studies together suggest that plants facing the harshest conditions at higher elevations may exhibit little phenological responses to global changes. Further research is therefore needed to assess whether this lack of phenological adjustment reflects a greater plasticity or rather an absence of adaptive potential due to strong conservatism in phenological features, and how these potential plastic and genetic responses are potentially mediated by biotic interactions.

Alpine and arctic plants are adapted to high levels of environmental stress with a set of specific traits, well known as stress tolerance syndromes (Cerabolini et al., 2010; Grime, 2006) such as slow growth rate, reduced stature, high individual longevity and varied protective traits. The rapidly changing climate combined with abrupt land use changes may now start favouring different adaptive traits in plants. In this issue, Zimmer et al. (2018) use a 38-year post-glacial chronosequences in the tropical Andes, matching a known period of accelerated climate warming. They observed that most plants having colonized recently unglaciated areas were anemochorous and rapidly growing ruderal species whereas slow-growing, stress-tolerant species were far less abundant. This suggests that the rapidly shrinking glacier cover may favour plants with opportunistic ecological strategies over slow-growing, stress tolerant, plants typical of high alpine environments, likely generating unexpected and likely novel plant communities at such elevations. However, this may not mean that slow growing, stress tolerant plants are immediate candidate to extinction in high alpine and arctic environments. Schittek et al. (2018) examine the longevity of a cushion-forming plant in the dry Andes of Peru. Radiocarbon dating on the peat formed by the cushion identified a chronosequence of 1050 years and permitted reconstructing past plant communities during humid/dry climatic oscillations. The cushion plant was able to cope with extended droughts, demonstrating that stress tolerant species can also support the effects of environmental oscillations in the long term. Nevertheless, the authors indicate that the capacity of the cushion to survive in a changing climate is conditioned by the maintenance of its protective plant surface. It is possible, then, that the intensification of land use will generate higher mortality of this type of plants, resulting in a regression of patrimonial habitats, but also loss of valuable data on past climate and vegetation.

Positive interactions among organisms are a crucial driver of plant community organization in arctic and alpine environments (Cavieres et al., 2014). One major challenge of ecological research is to infer how these interactions will change in response to global changes (Bulleri et al., 2016; HilleRisLambers et al., 2013; Olsen et al., 2016). Several contributions in this special issue have devoted some efforts to answering this question. Lord et al. (2018) demonstrate that plant–plant interactions and plant-invertebrate interactions were very strong drivers of community changes in response to an alteration of snow cover regimes. Zimmer et al. (2018) suggest that the reduced capacity of stress-tolerant nurse plants to colonize recently unglaciated alpine areas might have cascading effects on vegetation structure, by not allowing lower elevation species to colonize these sites, as they would normally do thanks to facilitative effects. Llambí et al. (2018) identify in the Venezuelan Andes a complex network of plant reciprocal interactions where nurses facilitated the presence of a native community and an exotic species. The exotic species had no reciprocal effect on nurse plants but had a negative effect on the rest of community members. By facilitating the presence of an exotic species, the nurse plant hence impact negatively the native community through indirect interactions. In a macro scale study in the Norwegian tundra, Bråthen et al. (2018) characterize an extended network of interactions involving plants and a large herbivore (*Rangifer tarandus*), along a temperature gradient. They show the overwhelming influence of the allelopathic shrub *Empetrum nigrum* on the distribution and abundance of other plant species and demonstrate that this shrub is currently spreading due to a combination of climate warming and lack of herbivory, announcing a scenario of local biodiversity loss in the next decades. Such a scenario would not have been detected by studying only interactions among pairs of plant species only. Focusing on alpine communities dominated by cushion-forming nurse plants, Raevel et al. (2018) examine variations in the functional traits of subordinate plants, which may provide insights into the ecological processes shaping species niches (Albert et al., 2011). They show that graminoids developed a stress avoidance strategy whereas forbs shifted from stress avoidance to stress resistance along an increasing stress gradient. They also demonstrate that cushion plants were able to buffer the effects of biotic and abiotic stresses on subordinate plant traits. Given that stress levels are expected to increase under global changes, these positive effects of nurse plants in the high Andean peatlands may preserve important ecosystem services such as water resource and biomass production.

An outstanding component of alpine biodiversity is constituted by its high rate of endemism. Smyčka et al. (2018) tested two alternative hypotheses to understand the origins of endemic plant species in the European Alps. Through a spatially-explicit bayesian analysis of phylogenetic structure of the high alpine flora, they observe that high endemism occurs in potential glacial refugia, but mainly on calcareous bedrock, and also at high elevations. Plant assemblages in calcareous refugia showed a signature of phylogenetic overdispersion – suggesting non-selective persistence of different lineages during ice ages, whereas those located in high mountain areas showed phylogenetic clustering – indicating repeated recent speciation and environmental filtering. From a conservation viewpoint, the authors conclude that calcareous glacial refugias are more important to conserve, as they host many range restricted species and more diverse evolutionary lineages. It is interesting to note that the primary drivers of mountain endemism cited by these authors, elevation and glacial refugias, are exacerbated in tropic alpine areas (Crawford, 2008; Sklenář and Laegaard, 2003). These regions have been cited to exhibit remarkable rates of endemism in plants and animals, especially in the Andes (Anthelme et al., 2014). A nice research challenge following the approach of Smyčka et al. (2018) would then consist in inferring endemism patterns in the high tropical mountains of the Andes. In another study conducted in the *trans*-Himalayas, Le Bagousse-Pinguet et al. (2018) hypothesized that more constraints at higher elevation should led to higher phylogenetic clustering of local plant communities. They instead observed that plants at higher elevation displayed random phylogenetic patterns, and that no particular mechanism of niche-based species coexistence process could therefore be inferred from the phylogenetic structure of these plant communities. This study echoes a recent one conducted in nival communities of higher summits of western European Alps, where the authors could not reject neutral mechanisms of community assembly by comparing community phylogenetic structure to neutral based simulations of community assembly (Marx et al., 2017).

3. Conclusion and perspectives

The discovery and study of unknown ecosystems and biodiversity have often provided novel knowledge and explicative models in biology (e.g. Rothschild and Mancinelli, 2001). So the position of arctic and alpine ecosystems at the climatic extreme of plant and animal life suggests that their study bears unique findings regarding the ecology and evolution of natural communities. We hope that the paper selection included in this special issue provides a first demonstration of the importance of studying arctic and alpine systems, both for advancing ecological and evolutionary theories, but also for understanding the future of cold-adapted ecosystems under contemporary global changes. A noteworthy progress in artic-alpine studies reflected in this special issue is the increasing integration with research techniques and concepts typically associated with physical sciences (glaciology, paleogeomorphology, cosmogenic nuclid and carbon isotopic dating, micro-climatic monitoring etc…). Also notable is a number of empirical approaches that will need to be undertaken by networked researchers across different mountain ranges of the globe; these are long term monitoring of multi-trophic community changes and whole-community transplant experiments (meadow turves) along key climatic gradients. We thus hope that this special issue will serve inspiring future research in the field of arctic-alpine ecology and beyond.

Acknowledgements

Both guest editors SL and FA thank the journal's editors Silke Dietz and Diethart Matthies for their continuous assistance in putting together this special issue. SL and FA thank Alexia Stokes for welcoming the organization of the symposium entitled “High elevation ecosystems” at the Ecosummit conference in 2016. SL acknowledges funding by the ANR project Origin-Alps (ANR-16-CE93-0004). FA was funded by the Fond Français pour l'Environnement Mondial (FFEM) and the Fondation pour la Recherche sur la Biodiversité (FRB) (BIOTHAW, AAP-SCEN-2011-II). Both guest editors thank all authors for their hard work and their outstanding contributions to this special issue. Julien Renaud helped putting together the map displayed in this paper. Both guest editors contributed equally to this editorial introduction.

References

Albert, C.H., Grassein, F., Schurr, F.M., Vieilledent, G., Violle, C., 2011. When and how should intraspecific variability be considered in trait-based plant ecology? Perspect. Plant Ecol. Evol. Syst. 13, 217–225.

Anthelme, F., Dangles, O., 2012. Plant–plant interactions in tropical alpine environments. Perspect. Plant Ecol. Evol Syst. 14, 363–372.

Anthelme, F., Jacobsen, D., Macek, P., Meneses, R.I., Moret, P., Beck, S., Dangles, O., 2014. Biodiversity patterns and continental insularity in the tropical high Andes. Arctic Antarctic Alp. Res. 46, 811–828.

Billings, W., Mooney, H.A., 1968. The ecology of arctic and alpine plants. Biol. Rev. 43, 481–529.

Bråthen, K.A., Gonzalez, V.T., Yoccoz, N.G., 2018. Gatekeepers to the effects of climate warming? Niche construction restricts plant community changes along a temperature gradient. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.06.005.

Bulleri, F., Bruno, J.F., Silliman, B.R., Stachowicz, J.J., 2016. Facilitation and the niche: implications for coexistence: range shifts and ecosystem functioning. Functional Ecol. 30, 70–78.

Carlson, B.Z., Choler, P., Renaud, J., Dedieu, J.P., Thuiller, W., 2015. Modelling snow cover duration improves predictions of functional and taxonomic diversity for alpine plant communities. Annals Bot. 116, 1023–1034.

Cavieres, L.A., Sierra-Almeida, A., 2018. Assessing the importance of cold-stratification for seed germination in alpine plant species of the high-Andes of central Chile. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.09.005.

Cavieres, L.A., Brooker, R.W., Butterfield, B.J., Cook, B.J., Kikvidze, Z., Lortie, C.J., Xiao, S., Al Hayek, P., Anthelme, F., Cranston, B., Garcia, M.C., le Bagousse-Pinguet, Y., Reid, A.M., le Roux, P., Lingua, E., Nyakatya, M., Touzard, B., Zhao, L., Callaway, R.M., 2014. Facilitative plant interactions and climate simultaneously drive alpine plant diversity. Ecol. Lett. 17, 193–202.

Cerabolini, B.E., Brusa, G., Ceriani, R.M., De Andreis, R., Luzzaro, A., Pierce, S., 2010. Can CSR classification be generally applied outside Britain? Plant Ecol. 210, 253–261.

Chalmandrier, L., Münkemüller, T., Colace, M.-P., Renaud, J., Aubert, S., Carlson, B.Z., Clément, J.-C., Legay, N., Pellet, G., Saillard, A., Lavergne, S., Thuiller, W., 2017. Spatial scale and intraspecific trait variability mediate assembly rules in alpine grasslands. J. Ecol. 105, 277–287.

Chapin, F.S.I., Körner, C., 1995. Arctic and Alpine Biodiversity: Patterns Causes and Ecosystem Consequences, vol. 113 Springer Science.

Choler, P., 2018. Winter soil temperature dependence of alpine plant distribution: implications for anticipating vegetation changes under a warming climate. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.11.002.

Crawford, R.M.M., 2008. Plants at the Margin: Ecological Limits and Climate Change. Cambridge Univ. Press.

Delnevo, N., Petraglia, A., Carbognani, M., Vandvik, V., Halbritter, A.H., 2018. Plastic and genetic responses to shifts in snowmelt time affects the reproductive phenology and growth of *Ranunculus acris*. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.07.005.

Graae, B.J., Vandvik, V., Armbruster, W.S., Eiserhardt, W.L., Svenning, J.C., Hylander, K., Ehrlén, J., Speed, J.D.M., Klanderud, K., Bråthen, K.A., Milbau, A., Opedal, Ø.H., Alsos, I.G., Ejrnaes, R., Bruun, H.H., Birks, H.J.B., Westergaard, K.B., Birks, H.H., Lenoir, J., 2018. Stay or go? How topographic complexity influences alpine plant population and community responses to climate change. Perspect. Plant Ecol. Evol Syst. http://dx.doi.org/10.1016/j.ppees.2017.09.008.

Grime, J.P., 2006. Plant Strategies, Vegetation Processes, and Ecosystem Properties. John Wiley & Sons.

HilleRisLambers, J., Harsch, M.A., Ettinger, A.K., Ford, K.R., Theobald, E.J., 2013. How will biotic interactions influence climate change–induced range shifts? Ann. New York Acad. Sci. 1297, 112–125.

Jacobsen, D., Dangles, O., 2017. Ecology of high altitude waters. Oxford University Press.

Körner, C., Hiltbrunner, E., 2018. The 90 ways to describe plant temperature. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.04.004.

Körner, C., Paulsen, J., Spehn, E.M., 2011. A definition of mountains and their bioclimatic belts for global comparisons of biodiversity data. Alp. Bot. 121, 73–78.

Körner, C., 2003. Alpine Plant Life: Functional Plant Ecology of High Mountain Ecosystems. Springer Science.

Le Bagousse-Pinguet, Y., Liancourt, P., Götzenberger, L., de Bello, F., Altman, J., Brozova, V., Rehakova, K., 2018. A multi-scale approach reveals random phylogenetic patterns at the edge of vascular plant life. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.10.002.

Llambí, L.D., Hupp, N., Saez, A., Callaway, R., 2018. Reciprocal interactions between a facilitator, natives, and exotics in tropical alpine plant communities. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.05.002.

Lord, J.M., Mark, A.F., Humar-Maegli, T., Halloy, S.R., Bannister, P., Knight, A., Dickinson, K.J., 2018. Slow community responses but rapid species responses 14 years after alpine turf transplantation among snow cover zones, south–central New Zealand. Perspect. Plant Ecol. Evol Syst. http://dx.doi.org/10.1016/j.ppees.2017.07.004.

MRI − Mountain Research Initiative EDW Working Group, 2015. Elevation-dependent warming in mountain regions of the world. Nat. Clim. Change 5, 424–430.

Madriñán, S., Cortés, A.J., Richardson, J.E., 2013. Páramo is the world's fastest evolving and coolest biodiversity hotspot. Frontiers Genet. 4, 192.

Marx, H.E., Dentant, C., Renaud, J., Delunel, R., Tank, D.C., Lavergne, S., 2017. Riders in the sky (islands): using a mega-phylogenetic approach to understand plant species distribution and coexistence at the altitudinal limits of angiosperm plant life. J. Biogeogr. 44, 2618–2630.

Molina-Venegas, R., Aparicio, A., Slingsby, J.A., Lavergne, S., Arroyo, J., 2015. Investigating the evolutionary assembly of a Mediterranean biodiversity hotspot: deep phylogenetic signal in the distribution of eudicots across elevational belts. J. Biogeogr. 42, 507–518.

Molina-Venegas, R., Aparicio, A., Lavergne, S., Arroyo, J., 2017. Climatic and topographical correlates of plant palaeo- and neoendemism in a Mediterranean biodiversity hotspot. Annals Bot. 119, 229–238.

Nagy, L., Grabherr, G., 2009. The Biology of Alpine Habitats. Oxford University Press.

Olsen, S.L., Töpper, J.P., Skarpaas, O., Vandvik, V., Klanderud, K., 2016. From facilitation to competition: temperature-driven shift in dominant plant interactions affects population dynamics in seminatural grasslands. Global Change Biol. 22, 1915–1926.

Pachauri, R.K., Allen, M.R., Barros, V.R., Broome, J., Cramer, W., Christ, R., et al., 2014. Climate Change 2014: Synthesis Report. Contribution of Working Groups I II and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. IPCC.

Raevel, V., Anthelme, F., Meneses, R.I., Munoz, F., 2018. Cushion-plant protection determines guild-dependent plant strategies in high-elevation peatlands of the Cordillera Real, Bolivian Andes. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.09.006.

Rixen, C., Wipf, S., Frei, E., Stöckli, V., 2014. Faster, higher, more? Past, present and future dynamics of alpine and arctic flora under climate change. Alp. Bot. 124, 77–79.

Rothschild, L.J., Mancinelli, R.L., 2001. Life in extreme environments. Nature 409, 1092–1101.

Schittek, K., Forbriger, M., Berg, D., Hense, J., Schäbitz, F., Eitel, B., 2018. Last millennial environmental dynamics in the western Peruvian Andes inferred from the development of a cushion-plant peat hillock. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.09.002.

Segnini, A., Posadas, A., Quiroz, R., Milori, D.M.B.P., Saab, S.C., Neto, L.M., Vaz, C.M.P., 2010. Spectroscopic assessment of soil organic matter in wetlands from the high Andes. Soil Sci. Soc. Am. J. 74, 2246–2253.

Sklenář, P., Laegaard, S., 2003. Rain-shadow in the high Andes of Ecuador evidenced by paramo vegetation. Arctic Antarctic Alp. Res. 35, 8–17.

Smyčka, J., Roquet, C., Renaud, J., Thuiller, W., Zimmermann, N.E., Lavergne, S., 2018. Disentangling drivers of plant endemism and diversification in the European Alps–A phylogenetic and spatially explicit approach. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.08.003.

Tarnocai, C., Canadell, J.G., Schuur, E.A.G., Kuhry, P., Mazhitova, G., Zimov, S., 2009. Soil organic carbon pools in the northern circumpolar permafrost region. Global Biogeochem. Cycles 23, GB2023.

Wang, Q., Fan, X., Wang, M., 2016. Evidence of high-elevation amplification versus Arctic amplification. Sci. Reports 6. http://dx.doi.org/10.1038/srep19219.

Zimmer, A., Meneses, R.I., Rabatel, A., Soruco, A., Dangles, O., Anthelme, F., 2018. Time lag between glacial retreat and upward migration alters tropical alpine communities. Perspect. Plant Ecol. Evol. Syst. http://dx.doi.org/10.1016/j.ppees.2017.05.003.

Fabien Anthelme[*]
AMAP, IRD, CNRS, INRA, Université de Montpellier, Montpellier, France
E-mail address: fabien.anthelme@ird.fr

Sébastien Lavergne[**]
Laboratoire d'Écologie Alpine (LECA), CNRS - Univ. Grenoble Alpes - Univ. Savoie Mont Blanc, 38000 Grenoble, France
E-mail address: sebastien.lavergne@univ-grenoble-alpes.fr

---

[*] Corresponding author.

[**] Corresponding author.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 6–15

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Research article

Winter soil temperature dependence of alpine plant distribution: Implications for anticipating vegetation changes under a warming climate[☆]

P. Choler[a,b,*]

[a] Univ. Grenoble Alpes, CNRS, LECA, SAJF, F-38000 Grenoble, France
[b] Univ. Grenoble Alpes, CNRS, LTSER Zone Atelier Alpes, F-38000 Grenoble, France

ARTICLE INFO

Keywords:
Alpine
Root zone temperature
Freezing degree days
Snow cover
Thermal niche
Vegetation dynamics

ABSTRACT

The topographical heterogeneity of mountain landscapes and the associated species turnover over short distances should prompt us to examine the relationships between climate and mountain plant distribution at a much finer scale than is commonly done. Here, I focused on the root zone temperature experienced by low-stature perennial-dominated plant communities of temperate mountains, which are seasonally covered by snow. Based on the analysis of multi-annual recordings of ground temperatures across a broad spectrum of plant communities, I propose a habitat template using Growing Degree Days (GDD) and Freezing Degree Days (FDD). These two indices summarize soil thermal conditions experienced during the favorable and the unfavorable period for growth. This heuristic framework allows refining our working hypotheses on the range shifts of mountain plants in response to recent and future climate change. Regional trends in climate variables controlling GDD and FDD indicate that the combination of earlier snow melt-out and higher summer temperatures have led to an overall increase in GDD over the last decades. However the persistence of cold episodes in spring and in fall along with the shorter snow coverage suggest that the positive effect of an extended growing season might be counteracted by the detrimental effects of increasing FDD. I thus hypothesize (i) a local-scale, downward shift of plant species along mesotopographical gradients, with marked species infilling in sparsely vegetated, long-lasting snow patches that contain vacant niches and (ii) a watershed-scale upward shift of subalpine species inhabiting south-exposed grasslands and able to cope with moderate FDD. This perspective challenges the simplistic view of an overall range shift of mountain plants along elevational gradients and calls for the improvement of models of snow cover dynamics and root zone temperature to draw up realistic scenarios of mountain vegetation changes under a warmer climate.

1. Introduction

In arctic and temperate mountain ecosystems, snow cover dynamics not only determine the length of the favorable period for growth but also tightly control the duration and intensity of freezing temperatures that belowground, perennial tissues have to cope with during the unfavorable period. Shallow or inconsistent winter snowpack causes soil frosts that can persist over weeks or months in cold contexts and this has a long-term ecological significance for the distribution of overwintering organisms (Callaghan et al., 2011; Sutinen et al., 1999). A seasonal reduction of snow cover duration may expose tissues to damaging frosts in spring or in fall and several studies emphasized the impact of this climatic hazard on the performance of organisms (Abeli et al., 2012; Inouye, 2000; Kreyling, 2010; Pauli et al., 2013; Wipf et al., 2009). The legacy of wintertime temperature regime on nutrient availability for plant growth during the following summer is also a key aspect of the functioning and the vegetation dynamics of snow-covered arctic and alpine ecosystems (Kreyling, 2010; Sturm et al., 2005). For example, increased microbial activity and higher N retention are observed in soils that benefit from the insulating effect of the snow (Baptist et al., 2010; Edwards et al., 2007) whereas freezing temperatures may favor the physical degradation of soil organic matter and the amount of inorganic nitrogen (Freppaz et al., 2008).

In mountain landscapes, thermal differentiations among nearby sites are related to the high topographical heterogeneity (Billings, 1973; Scherrer and Korner, 2010; Walker et al., 1993). During winter, these differences might be exacerbated by wind snow redistribution between sheltered sites where snow accumulates and exposed, wind-blown sites

---

[☆] This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author at: Univ. Grenoble Alpes, CNRS, LECA, SAJF, F-38000 Grenoble, France.
E-mail address: philippe.choler@univ-grenoble-alpes.fr.

https://doi.org/10.1016/j.ppees.2017.11.002
Received 31 January 2017; Received in revised form 25 October 2017; Accepted 8 November 2017

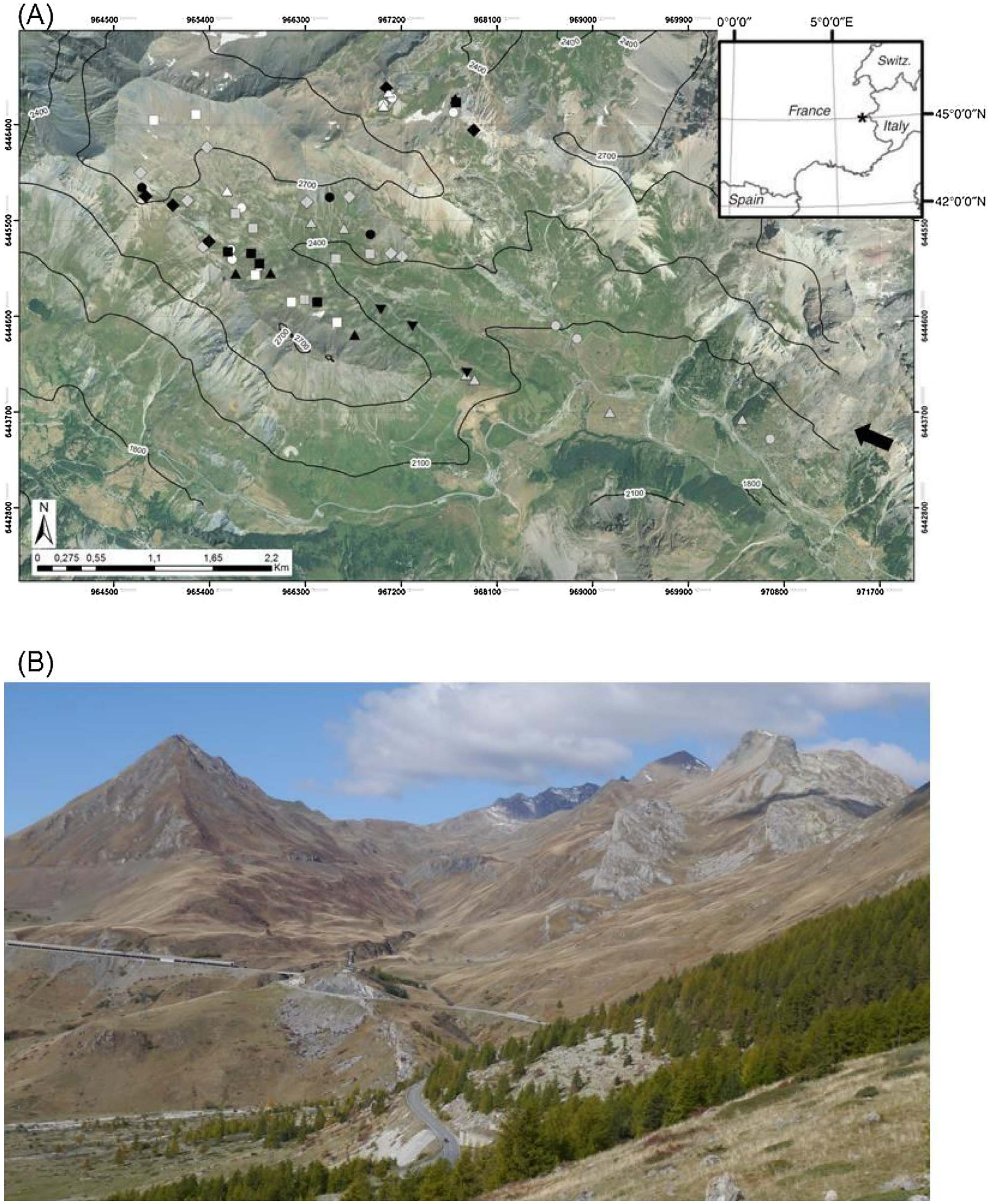


Fig. 1. Location of plots in the study area (A) and overview of the Roche Noire watershed (B). The arrow on the map indicates the position of the photographic shooting. The inset map shows the location of the study area in the French Alps. Symbols for plant communities follow Table 1.

where it is eroded (Essery and Pomeroy, 2004). As a result, highly contrasting soil temperature regimes are reported over mesotopographical gradients, i.e. ridge-to-snow bed gradients which typically represent a distance of a few meters (Baptist and Choler, 2008; Wundram et al., 2010). Consistent patterns of functional and taxonomic beta diversity and for different trophic levels have been documented along these gradients (Heegaard, 2002; Kudo et al., 1999; Zinger et al., 2009). Consequently, the response of alpine plants to ongoing climatic change cannot be properly assessed if this patchiness of the thermal landscape is not properly understood (Graae et al., 2012). In line with this reasoning, a recent study pointed out that the magnitude of predicted change in temperature is relatively weak compared to what plants would experience given a very short-distance range shift (Scherrer and Korner, 2011).

Current models that aim to project the distribution of high elevation species under a warmer climate suffer from the insufficient understanding and representation of the thermal envelopes of alpine plants. Fields of forcing variables are unavailable at the spatial scale that really matters for alpine plant life and the common practice of downscaling global climate gridded data is questionable because the local temperature experienced by low-stature plants is decoupled from atmospheric variables (Graae et al., 2012; Körner, 1999). If the local scale variation of key drivers of plant distribution is overlooked, projected distribution maps cannot be relevant. For example, Graae, (in press) emphasized the pivotal role of topographical complexity to understand how mountain plants will respond to climate change. Another weakness of current models pertains to the insufficient consideration of wintertime climate conditions because snow cover dynamics is poorly represented in these models (Carlson et al., 2013; Pradervand et al., 2014). Studies accounting for snow cover change on ecological processes, mainly focus on the impact of the first snow free day and the growing season length but winter soil temperature remains a missing dimension (Choler, 2015; Ernakovich et al., 2014; Heegaard, 2002). This is at odds with the recognition that winter conditions, and particularly winter soil temperature, are pivotal to capture biodiversity patterns and ecosystem functioning in snow-covered ecosystems (Pauli et al., 2013; Petty et al., 2015).

There is an increasing recognition that snow amount and duration have significantly declined in the Alps since the eighties in relation with a warmer climate (Beniston, 1997; Hantel et al., 2000; Hantel et al., 2012). This trend is clearly observed at intermediate elevations − typically below 1800 m (Hantel and Hirtl-Wielke, 2007) because of the high sensitivity of the snowpack to air temperature in this elevational range. Higher up, there remain large uncertainties essentially because inter-annual and regional variations in winter precipitation are poorly known (Beniston et al., 2003). The consequences of these snowline shifts on mountain vegetation have been mainly discussed from the viewpoint of its positive effect of the growing season length and its likely effect on the upward shift of plants (Carlson et al., 2015; Gottfried et al., 2011; Keller et al., 2005). However, a comprehensive assessment of the effects of snow cover changes on plant performance and distribution should also account for winter conditions and the interplay between snow cover, air temperature, and soil temperatures (Wheeler et al., 2014).

Miniature and standalone sensors enable a much more accurate picture of the thermal conditions really experienced by alpine plants compared to air temperature data provided by weather stations (Graae et al., 2012). Multi-annual soil temperature monitoring has been used to revisit some of the long-standing issues in the biogeography of cold ecosystems, such as the thermal features of the treeline (Körner and Paulsen, 2004; Körner et al., 2003). However studies examining multi-annual variations of summertime and wintertime thermal conditions and across a large range of mountain habitats are lacking. In this manuscript, I report analysis of a previously unpublished database of soil temperature regimes collected in a broad spectrum of above-forest habitats in the French Alps. I build upon this analysis to propose a thermal-based template for alpine plant distribution. More specifically, I show that (i) the duration and intensity of ground freezing temperature is complementary to the Growing Degree Days index to define the thermal niche of dominant alpine plants, (ii) the observed trends in regional climate and snow cover have significantly shifted these thermal domains over the past decades, and (iii) paying further attention to local soil temperature regimes, and especially during wintertime, is pivotal to move beyond the simplistic view of an overall upward shift of mountain plants under a warmer climate.

2. Materials and methods

2.1. Study area

Most of the data were collected in two sites locally known as *Roche Noire* and *Aravo* and located in the Grand Galibier Massif of the South-Western French Alps (Fig. 1). The Roche Noire site is a high elevation watershed with a predominant northwest to southeast orientation (Fig. 1). The main bedrock is represented by flysch sediments, called the ‘Flysch des Aiguilles d'Arves’ (Deharveng et al., 1987). The Aravo site consists of an inclined depression located between 2600 and 2650 m on calcareous shales (Fig. S1, Choler, 2005). A few additional plots were installed in the immediate vicinity of the Galibier pass (Fig. 1). In RON and ARA sites, soils are slightly acidic (pH between 5 and 6.5). Details on soil structure and chemistry are provided in table S1 of Zinger et al. (2011). The treeless vegetation is dominated by a mosaic of heathlands and productive subalpine grasslands in the lower part and by short stature alpine meadows, screes and rocks in the higher part. Based on previous vegetation surveys (Choler and Michalet, 2002) it is hypothesized that the subalpine to alpine ecotone is located between 2300 m and 2500 m depending on slope exposure. Sites are grazed extensively by cattle or sheep from June to September.

2.1.1. Field data

The study area lies between the Lautaret and Galibier passes and is well-known for its rich variety of bedrocks and landforms. Plots were only established on non-acidic sedimentary rocks with base-rich soils to ensure that the targeted plant communities are sharing a common species pool. Based on a regional field survey of vegetation (Choler and Michalet, 2002), I selected 56 plots to cover the most common vegetation types of the area. The sampling design was based on the physiognomy (vegetation cover, grassland *vs*. shrubland) and the presence/absence of dominant species. The resulting ten groups (Table 1) align well with the standard plant classification schemes of the Alps (Theurillat et al., 1994) and are representative of the subalpine, alpine and nival belts of this part of the French Alps.

Sites were equipped with miniaturized and standalone temperature data loggers (Hobo pendant UA, Onset Computer Corporation, Bourne, MA). Loggers were buried at 5 cm below ground level to monitor the near surface ground temperatures on an hourly basis. Most of the time series on which this analysis is based begin in 2007 and end in 2015. Vicissitudes of the monitoring program including battery issues and loss of sensors explain the numerous gaps in time series. Details on data availability per plot are shown in supplementary Fig. S1. Soil temperature monitoring and vegetation surveys are ongoing as part of a long-term program.

Near surface soil thermal conditions are representative of those experienced by the root system of perennial plants, which have a high density of roots in the topsoil layer. Root zone temperatures are highly responsive to snow cover dynamics (Harris et al., 2009; Schmid et al., 2012). Daily mean soil temperatures around 0 °C and showing no circadian thermal amplitude are indicative of the presence of a deep winter snowpack with efficient insulated effect. By contrast, negative daily mean soil temperatures generally occur in snow free conditions or when the snowpack is too shallow to buffer negative air temperatures. Negative soil temperature may eventually occur below a deep snowpack in the case of delayed snowfalls overtopping frozen soils.

2.1.2. Climate data

Time series of temperature, precipitation and snow height were provided by the SAFRAN-SURFEX/Crocus-MEPRA model chain (S2 M), hereafter SAFRAN−Crocus, developed by Météo France for the French Alps (Durand et al., 2009a,b; Vionnet et al., 2012). The model combines observed data from a network of weather stations and estimates from

Table 1
Main characteristics of the twelve surveyed plant communities. *N* is the number of plots per plant community. Mean (+/− 1 standard deviation) are given for elevation and bioclimatic variables (see Material and Methods for details).

Symbol	N	Dominant vascular plant	Elevation (m a.s.l)	Plant cover range (%)	First Snow Free Day (Julian Day)	Growing Degree Days (°C)	Freezing Degree Days (°C)
◍	3	Bromus erectus, Festuca laevigata	2062 (89)	70–90	80 (3)	1688 (117)	−108 (79)
◭	6	Patzkea paniculata	2201 (234)	90–100	116 (20)	1317 (181)	−4 (6)
●	3	Helictotrichon sedenense	2634 (136)	30–50	119 (16)	1082 (230)	−196 (235)
■	5	Carex myosuroides, Dryas octopetala	2538 (30)	50–70	132 (11)	1082 (143)	−536 (105)
▨	5	Carex sempervirens, Trifolium alpinum	2499 (98)	70–90	147 (4)	985 (23)	−42 (58)
◆	5	Carex myosuroides, Sesleria coerulea	2670 (54)	70–90	141 (20)	1029 (168)	−322 (210)
▼	3	Vaccinium myrtillus, Vaccinium uliginosum	2292 (58)	80–100	145 (8)	867 (96)	−178 (166)
▲	5	Salix retusa, Salix reticulata	2562 (51)	40–60	148 (11)	802 (121)	−343 (211)
◈	9	Festuca violacea, Trifolium pratense	2615 (133)	30–60	160 (21)	1010 (144)	−39 (91)
△	3	Salix herbacea, Plantago alpina	2629 (2)	60–80	161 (5)	877 (106)	−13 (15)
○	6	Achemilla pentaphyllea, Carex foetida	2581 (37)	90–100	167 (10)	762 (102)	0 (0)
□	5	Ranunculus glacialis, Poa distichophylla	2649 (165)	5–15	199 (15)	447 (143)	0 (0)

numerical weather forecasting models to provide hourly data of atmospheric parameters and snow amount and duration for 23 massifs of the French Alps. The model accounts for topographic features and climate variables are simulated for 300 m-wide elevational bands, aspect (seven orientations, and slope (flat, 20° and 40°). Because of its physical basis and the unavailability of forcing variables at the relevant scale, the model cannot represent mesotopographical variations of snow cover duration and amount. Therefore, model outputs were essentially used to provide a long-term temporal context of the regional climate and not to predict local thermal conditions. Climate re-analyses corresponding to the period 1959–2015 were extracted for different elevations and for the reference flat surface. The studied sites are located at the limit between the two climate massifs Thabor and Oisans. Because there were no significant differences when comparing long-term temperature and snow cover trends between these two massifs, figures given here are for the Oisans massif.

2.1.3. *Data analysis*

First the daily average of hourly values of soil temperature was calculated as follows

$$\mathrm{Tavg} = 0.5\,\mathrm{x}\ (\mathrm{Tmin} + \mathrm{Tmax})$$

where Tmin and Tmax are the minimum and maximum daily temperature, respectively.

Soil thermal conditions during the favorable period for growth were summarized by calculating a Growing Degree Days (GDD) index corresponding to the sum of average daily degrees above a given threshold (Z_{GDD}) following

$$\mathrm{GDD} = \Sigma_i\, \mathrm{Tavg}_i \text{ with } \mathrm{Tavg}_i = 0 \text{ if } \mathrm{Tavg}_i < Z_{GDD}$$

I used a threshold value of 1 °C instead of 0 °C because slightly positive daily averages can be observed even under a deep snowpack due to measurement errors of the sensor. Other Z_{GDD} values ranging from 1 °C to 5 °C were used without bringing any significant change to the comparative analysis of plots. GDD was estimated either by considering the whole set of days during which Tavg was above 1 °C or by setting a temporal limit at the end of August to restrict the analysis to the productive season. Again, this did not impact the main findings of this study. Noticeably, the GDD-based ranking of plots was not sensitive to this choice. This is mainly because the date of the first snow fall in autumn exhibits low variability among plots. On average, I found a GDD decrease of 20% to 25% when the reduced time frame was used. Because this study mainly focused on spring and summer changes in thermal conditions, results are presented for the period ending on August 31st.

Soil thermal conditions during the unfavorable period were summarized using a Freezing Degree Days (FDD) index. FDD was calculated as the sum of average daily degrees below the threshold Z_{FDD} following

$$\mathrm{FDD} = \Sigma_i\, \mathrm{Tavg}_i \text{ with } \mathrm{Tavg}_i = 0 \text{ if } \mathrm{Tavg}_i > Z_{FDD}$$

Z_{FDD} was set to −1 °C for the reasons explained above. The computation of FDD follows the same logic used to estimate GDD, although a different threshold is used. FDD is used in climate science to capture both frost intensity and the length of the frost episodes for a given period (https://nsidc.org/cryosphere/glossary/term/freezing-degree-days). In this study, FDD was estimated on a yearly basis from the beginning of August of year *n*-1 to the end of July of year n to capture winter harshness.

3. Results and discussion

3.1. *A bi-dimensional template for mountain plant thermal niches*

The two year-long time series of root zone temperatures shown in Fig. 2 illustrate three contrasting soil thermal conditions. A comparison with daily minimum and maximum air temperature at 2500 m is also provided (Fig. 2A). A subalpine grassland dominated by the tall fescue *Patzkea paniculata* and located on gentle south-exposed slopes benefits from long and warm growing season (GDD around 2000 °C) and does not experience severe winter ground freezing temperatures (Fig. 2B). The slightly negative FDD in 2011–2012 is due to a very cold episode in spring 2012 – with minimum air temperatures below – 10 °C (Fig. 2A) – at a time where the melting snowpack was shallow. At a higher elevation, a turf alpine meadow located on wind-blown ridges and dominated by *Carex myosuroides* copes with a reduced GDD (around 1200 °C) and most noticeably with intense and long-lasting ground freezing temperatures during winter (Fig. 2C). At the same elevation although in sheltered locations where snow accumulates, a short stature plant communities dominated by *Alchemilla pentaphyllea* is thermally insulated from the atmosphere all the winter long and shows FDD values equal to 0 (Fig. 2D). Though the growing season is much shorter than in the nearby *Carex myosuroides* community, it is significantly warmer (12.7 °C *vs.* 10.9 °C for the July average temperature). As a

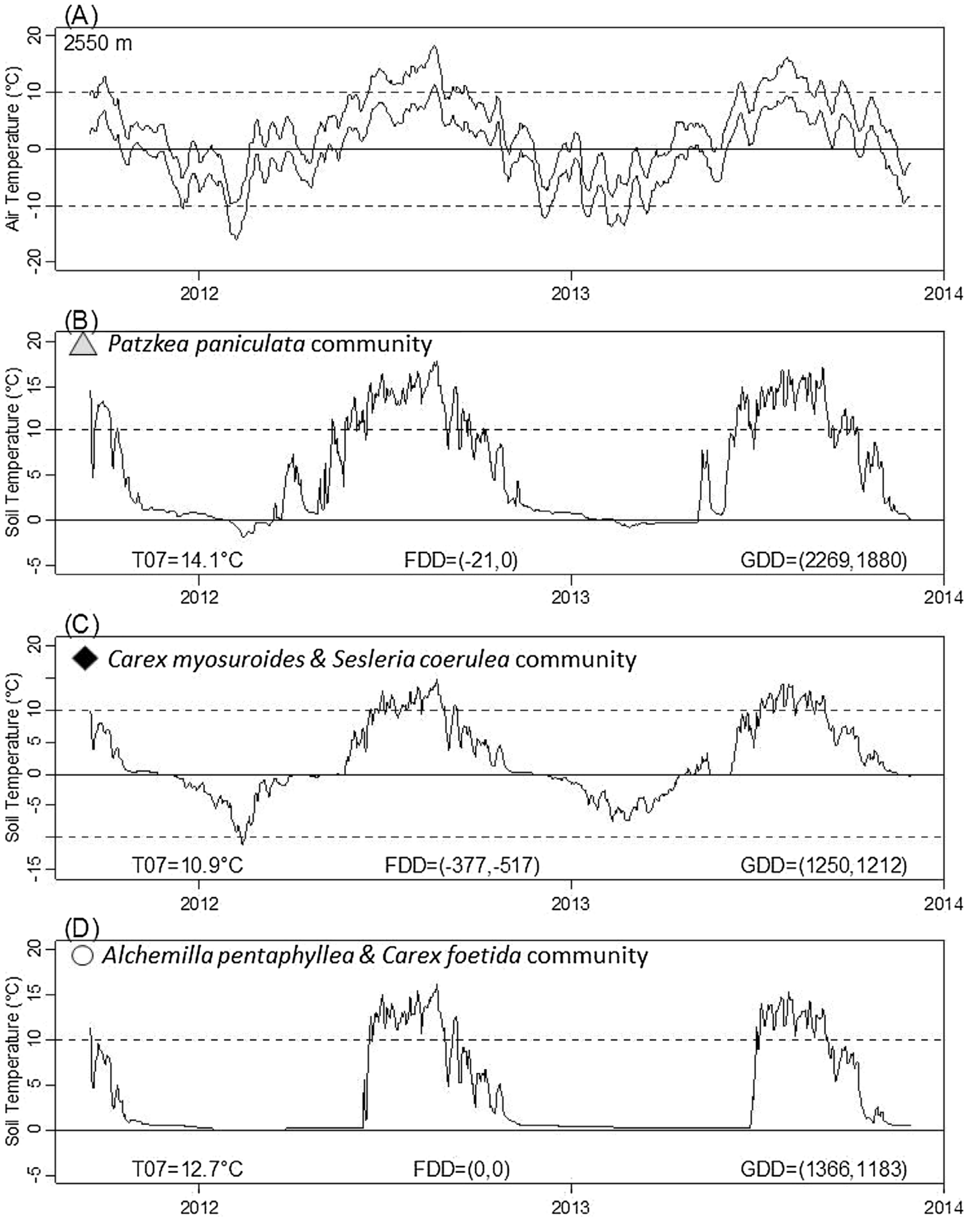


Fig. 2. Annual course of daily minimum and daily maximum air temperature (A) and daily average soil temperature in three contrasting plant communities (B, C, D). Freezing Degree Days (FDD), Growing Degree Days (GDD) and average July temperatures (T07) are given in brackets for each year. Air temperatures are from SAFRAN-SURFEX/Crocus-MEPRA model chain (S2 M) for the Oisans massif.

result, these two alpine sites exhibit similar GDD values. These three examples illustrate how GDD and FDD allow for characterization of thermal habitats of mountain vegetation along elevational and mesotopographical gradients.

Fig. 3 shows the relationships between GDD and FDD for all surveyed plots. Plots belonging to the same community are positioned closed to each other highlighting the ecological relevance of GDD and FDD for defining habitat niches. Plots that do not experience prolonged winter ground freezing, i.e. having FDD values between 0 and − 100 °C, are distributed along a gradient of GDD that ranges from subalpine grasslands to alpine snow beds. In the mid-range of this gradient, i.e. GDD values between 800 °C and 1200 °C, several plant communities experience low FDD values with the most severe conditions occurring in communities dominated by *Carex myosuroides* and *Dryas octopetala*. Overall, the variance within communities is higher for FDD than for GDD (Table 1 and Fig. 3B) and some community types can hardly be distinguished from one another on the sole basis of their thermal profiles. For example, there is a strong overlap in GDD and FDD between the sparsely vegetated communities dominated by *Helictotrichon sedenense* and the turf meadow dominated by *C. myosuroides* and *Sesleria coerulea*. Other ecological factors, such as the substrate stability for example, are to consider for further discriminating the habitat suitability of these dominant species. Further studies are also needed to examine the relevance of this thermal-based habitat template for species that do not account for a high proportion of the aboveground biomass, i.e. subordinates and transients *sensu* Grime (1998). Our previous studies have underlined the importance of biotic interactions to capture the distribution range of these species along elevational and mesotopographical gradients (Choler et al., 2001; Kikvidze et al., 2005). Anthelme et al. (2014) also emphasized the role of positive plant–plant interactions in the upward and downward range shift of alpine plants under a changing climate.

Finally, Fig. 3 also shows unrealized thermal conditions corresponding to the combination of low (or high) GDD and very negative FDD values. Late snow melting plots with low GDD values do not experience prolonged ground freezing because the only period during which this may happen is late fall-early winter in the event of delayed snowfalls. On the other side of the GDD gradient, low elevation plots may experience ground freezing just before or after the snowy period as illustrated in Fig. 2B. However this has never led to strongly negative

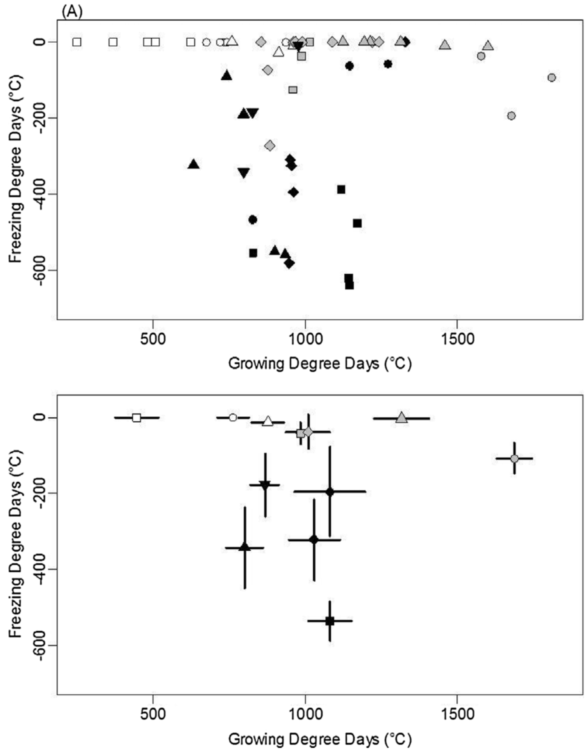


Fig. 3. Location of study plots in a bi-dimensional thermal-based habitat template. Time averages of GDD and FDD are shown for each plot (A) and for each plant community with +/− 1 standard deviation (B). Symbols for plant communities follow Table 1.

FDD values likely because these plots experience rapid warming in the spring.

Overall, these results show that FDD and GDD are two key variables for a first-order habitat filtering of dominant mountain species. The relationship between these two variables delineates a thermal domain occupied by alpine plant communities and provides a heuristic framework to address the effect of climate change on plant community distribution as detailed below.

3.2. Regional climate change

Regional-scale trends of key climate variables controlling GDD and FDD are shown from 1959 onwards and for a reference elevation of 2400 m (Fig. 4). Yearly-based time series were derived from SAFRAN-Crocus re-analyses of air temperature and snow cover duration. The three last decades show a significant positive trend towards warmer summers (Fig. 4A). This is illustrated by examining the sum of average daily degrees above 0 °C for the period July-August. A breakpoint analysis identifies a shifting period in the late seventies – early eighties (Fig. 4A). Over the last thirty years this sum was 100 °C higher that it was during the period 1959–1978. Trends in the First Snow Free Day (FSFD) show similar patterns (Fig. 4B). From 1995 onwards, the average FSFD was the 150th Julian Day (30th of May) whereas it was two weeks later on (14th of June) in the preceding period. The snowy winters of the late seventies – early eighties (1978, 1979 and 1983) have never been observed since then, even if a high year-to-year variability has been noticed recently – see for example the contrast between year 2011 and year 2013. The shift towards low snow winters in the late eighties and the lack of significant trend since then have also been reported for the Swiss Alps (Marty, 2008). Finally, the sum of average daily degrees below −5 °C from April to June was used as a proxy for the duration and intensity of cold episodes in the spring. There have been several years with both cold springs and delayed FSFD in the eighties but overall this sum did not show any significant trend over the period 1959–2016, despite the rise in temperatures (Fig. 4C).

To sum-up, the regional climate trend is marked by warmer and longer snowfree periods and by the persistence of cold events in spring. This means an increasing likelihood of exposure to frost events for early snow melting sites that do no benefit from the insulating effect of the snowpack. Using 37 years-long time series of climate data for the Swiss Alps, Wheeler et al. (2014) reached similar conclusions and pointed out this counterintuitive effect of global warming in mountainous environments. When considering species range shift under climate change, particular attention should thus be dedicated to the balance between the positive effect of increasing GDD (because of the longer and warmer snow free period) and the detrimental effect of a decreasing FDD (because of the increasing risk of ground freezing for soils insufficiently protected by snow). Accounting for these two dimensions of the thermal niche remains a challenge for Species Distribution Models. Predictions of alpine species distribution under climate change scenarios have commonly relied on spatially downscaled air temperatures (Engler et al., 2009). This approach is misleading as the actual temperature experienced by small stature plants strikingly deviates from the air temperature of weather station (Körner and Hiltbrunner, in this issue). In particular, the tight relationship between snow cover dynamics, soil temperature profiles and the distribution of dominant mountain plants has to be accounted for to draw up more realistic scenarios of vegetation changes under a warming climate. In the following section I propose a framework for making progress in this direction.

3.3. Implications for mountain plant distribution under a warming climate

First, I examined the likely effect of regional climate change on the FDD-GDD relationship. To be consistent with observed recent trends over the last three decades, I built up on a scenario with a 15 days shift in the snow melt-out date and a 100 °C increase in the sum of daily average degrees in July-August (Fig. 3). The earlier snow melt-out translates into additional degree days that were estimated using SAFRAN–Crocus times series of air temperature. For each half-month period, I calculated the sum of average daily degrees above 0 °C (hereafter positive degree days) and below −2 °C (hereafter negative degree days) (Fig. 5A). For example, a high-elevation snow bed located between 2400 m and 2700 m, and experiencing a snow melt-out the first of July instead of the 15th of July, would gain 131 positive degree days and no negative degree days. If the whole growing season is considered, this amount of extra degree days due to an advanced snow melting should be added to the 100 °C increase due to the warmer July and August. For a plot located at the same elevation but experiencing a snow melt-out the first of April instead of the 15th of April, the gain of positive degree days would only amount 10 °C and the negative degree days would amount −55 °C (Fig. 5A). As expected, the gain in positive degree days is higher in early summer than in the spring and it decreases with elevation due to the adiabatic lapse rate (Fig. 5A). The impact of an earlier snow melt-out on negative degree days is only noticeable early in the season and vanishes after mid-May (Fig. 5A).

These estimates of degree days based on air temperature need to be converted into GDD and FDD to be ecologically meaningful. As in previous studies (Zheng et al., 1993), I found a significant relationship between the daily average air temperature given by SAFRAN-Crocus and the measured daily average soil temperatures during the snow free period (supplementary Fig. 2A). Based on the estimated slope of 0.65, I hypothesized that an increase of 100 °C in positive degree days will contribute to a 65 °C degree increase in GDD. The effect of negative degree days on FDD is harder to quantify because it is highly dependent on the absence or presence of snow as well as the thermal resistance of the snowpack (Harris et al., 2009; Saccone et al., 2013). Empirical data shown in supplementary Fig. 2 B indicate that daily minimum air temperature in the range −5 °C to −10 °C are generally associated with daily minimum soil temperatures below −2 °C. Although these data can

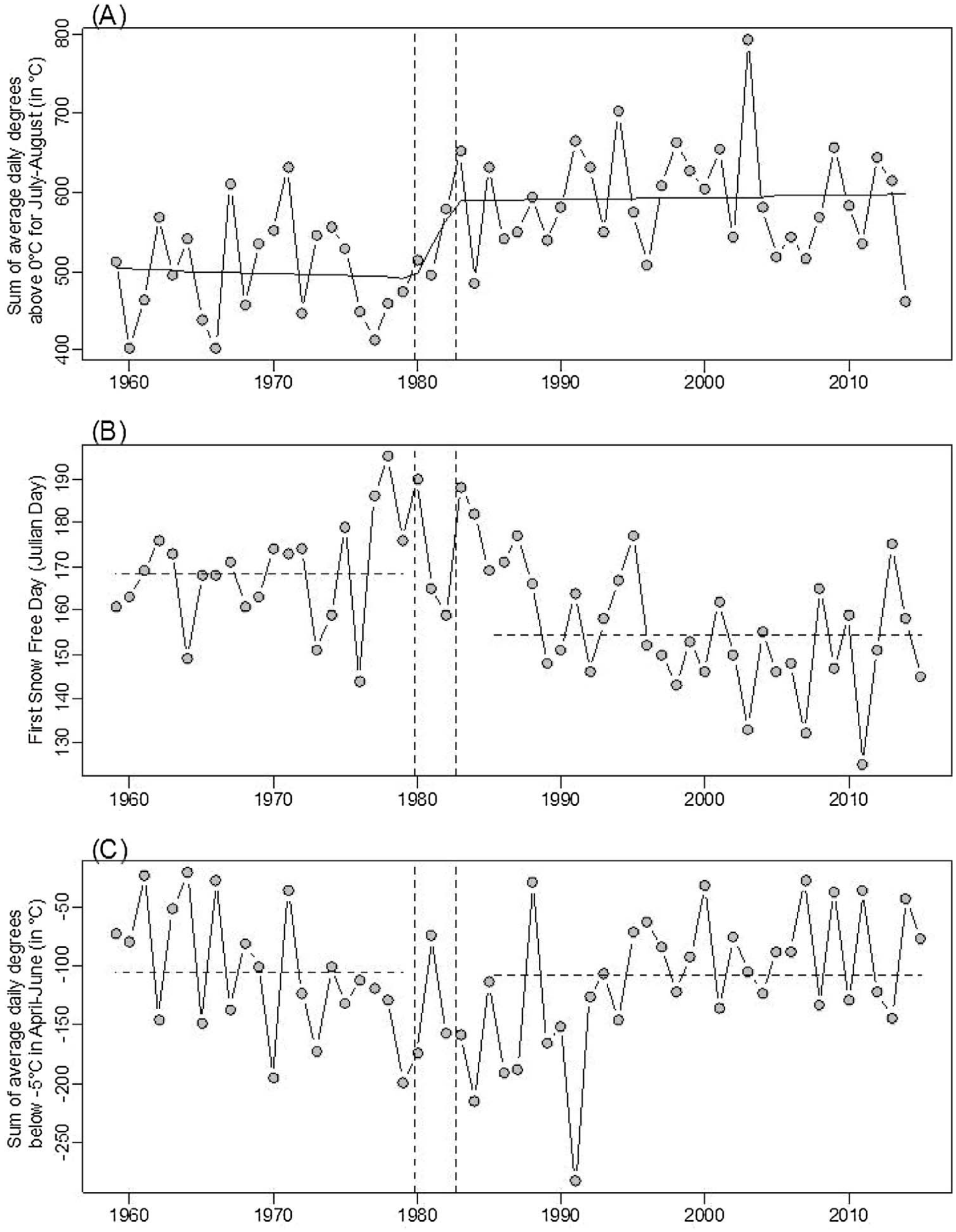


Fig. 4. Long-term trends of the sum of average daily degrees above 0 °C for the period July-August (A), the First Snow Free Day (FSFD in Julian Day) (B), and the sum of average daily degrees below −5 °C for the period April-June (C). Data are derived from the SAFRAN-SURFEX/Crocus-MEPRA model chain (S2 M) for the Oisans massif in the 2400 m–2700 m elevational band and for a flat surface. Regression lines in (A) result from a breakpoint analysis which identifies a period of regime shift from 1979 to 1985. Dotted lines in (B) and (C) represent the long-term averages before and after the regime shift.

be used to provisionally assess the cascading effects of cold air episodes on FDD, biophysical models of soil temperature accounting for snow cover, soil water content and vegetation cover are needed to improve our understanding of this relationship (Wang et al., 2008).

I built on this ensemble of observations and calculations to assess the likely shifts of thermal conditions experienced by the root zone of mountain ecosystems in response to ongoing climate changes (Fig. 5B). At first glance, it appears that calculated changes in GDD and FDD are of ecological significance. A GDD increase of 100 °C, which is a conservative estimate, represents 10% or even more of the average GDD value characterizing most of the plant communities (Table 1, Fig. 5B). It is also the average difference between *Carex sempervirens* communities and *Salix herbacea* communities (Table 1). For late snow melting plots, a GDD increase of more than 150 °C is plausible (Fig. 5) and means a spectacular change in thermal profiles. Taken together, these results suggest a significant amelioration of the thermal conditions experienced by mountain plant communities during the growing season. This might have triggered either a densification of plant coverage or species range shifts along elevational gradient and mesotopographical gradient over the last decades. Unfortunately, there is no available long-term botanical survey to ascertain these changes in the studied area. However, a recent analysis of high-resolution Landsat images in the nearby Ecrins National Park points out to an overall greening of high elevation landscapes (Carlson et al., 2017). This result aligns with other reports on cold ecosystems showing an increasing vascular plant cover at the expense of bare ground over the last decades (Cannone and Pignatti, 2014; Elmendorf et al., 2012; Kullman, 2010; Rundqvist et al., 2011). At high elevation I hypothesize that the positive effect of increasing energy availability might be particularly pronounced in late snow melting habitats offering vacant niches. Aerial photographs dating back to the eighties show that nowadays sparsely vegetated patches in concave microsites were covered by snow even at the end of summer (supplementary Fig. 3). It is likely that the vascular plant cover has significantly increased in these habitats over the last decades with newly established plants coming from a local pool of pioneered species able to cope with low nutrient availability and substrate instability.

The positive effect of a longer growing season can be offset by the detrimental effect of cold-weather episodes if soils are insufficiently

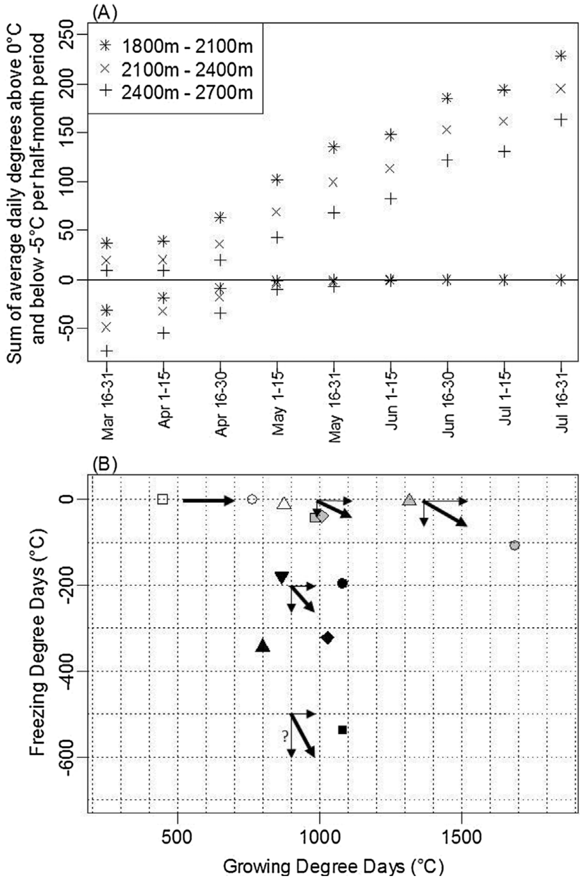


Fig. 5. (A) Sum of average daily degrees above 0 °C and below − 5 °C for each half-month period between March and July. Values are averages over the past 30 years and are calculated using SAFRAN–Crocus air temperatures; (B) Scenario of GDD and FDD shifts in the bi-dimensional habitat template. The magnitude and direction of change indicated by the arrows are consistent with the observed trends in regional climate over the three last decades. Symbols for plant communities follow Table 1. See text for details.

protected by snow. Wipf & al. (2009) showed that this negative effect is species-specific with dominant species of early snow melting sites being the least sensitive to experimental reduction of snow cover. Here, I found that between-plot variation in FDD was high for all plant communities experiencing regular ground freezing temperatures during winter (Fig. 3B). This holds particularly true for communities dominated by *Carex myosuroides*, which are found in the most extreme thermal conditions. For these communities, a FDD decrease of 50 °C – a likely estimate given the scenario above – would be in the same order of magnitude as between-plot variation. By contrast, the same change in FDD may have a more significant impact for low elevation, south-exposed grasslands such as the *Patkea paniculata* communities (Fig. 5B). For these communities I hypothesize that the main beneficiaries of an increasing GDD will be those species able to cope with the associated decrease in FDD. By contrast, species unable to cope with this increased exposure to spring frosts may experience downslope range shift as pointed out by Lenoir et al., (2010). Species-specific long-term surveys or experimental manipulation of the snow cover duration should be conducted to test these hypotheses.

4. Conclusion & perspectives

Here I showed that the duration and intensity of ground freezing, which are summarized by the Freezing Degree Days index, exhibit marked variations along complex gradients of elevation and mesotopography and that this index, along with the Growing Degree Days index, represents a leading axis of habitat differentiation in the alpine zone. Comparative studies are required to examine the relevance of this GDD-FDD habitat template for other alpine areas. In particular, these findings raise questions for alpine ecosystems that do no experience seasonal snow coverage such as the tropical alpine environments (Anthelme and Dangles, 2012; Rundel et al., 1994) or highly continental climates that exhibit reduced snow depth (Brun et al., 2013). The role of soil temperature as a driving factor of beta diversity patterns and ecosystem functioning along complex topographical gradients remains to be elucidated in these bioclimatic contexts.

An in-depth understanding of local bioclimatic change is needed to refine our current models of species distribution and anticipate species range shifts in response to global change (Lenoir and Svenning, 2015). In seasonally snow-covered alpine ecosystems, I point out that a better understanding of snow cover dynamics and root-zone temperature throughout the year is needed to draw up realistic scenarios of vegetation changes. There is a need to better quantifying the relationship between snow cover dynamics and root-zone temperatures. Predicting snowpack characteristics at an ecologically-relevant scale in complex terrain remains a challenge for physical-based models (Brun et al., 2013; Liston and Elder, 2006; Rankinen et al., 2004). Remote sensing approaches using high temporal and spatial resolution are valuable for quantifying snow cover duration at a regional scale (Dedieu et al., 2016; Kivinen et al., 2012). Complementary developments in laser altimetry and infra-red thermometry are also promising to mapping snow depth and surface temperature in mountain catchments (Deems et al., 2013; Revuelto et al., 2014; Scherrer and Korner, 2010). Using these data to calibrate high-resolution models of soil temperatures will provide new opportunities to quantify the thermal niche of alpine plants.

Ackowledgments

Thanks are due to Brad Carlson for helpful discussions and editing of the manuscript. I am indebted to Vincent Vionnet (CNRM – Météo France) who provided me with the latest SAFRAN-Crocus climate reanalyses. Julien Renaud helped me to prepare Fig. 1. I thank my son Romain Choler and all the staff of the Station Alpine J. Fourier for their invaluable assistance in the field. I am particularly grateful to Prof. Serge Aubert (1966–2015) who had always supported my research on alpine plant communities. This research was conducted at the Long-Term Socio-Ecological Research (LTSER) Zone Atelier Alpes, a member of the eLTER-Europe network and at the Station Alpine J. Fourier, a member of the AnaEE-France. It received funding from the program Investissements d'avenir (ANR-11-INBS-0001AnaEE-Services) and from the LTSER Zone Atelier Alpes. LECA is part of Labex OSUG@2020 (ANR10 LABX56).

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.ppees.2017.11.002.

References

Abeli, T., Rossi, G., Gentili, R., Mondoni, A., Cristofanelli, P., 2012. Response of alpine plant flower production to temperature and snow cover fluctuation at the species range boundary. Plant Ecol. 213, 1–13.

Anthelme, F., Dangles, O., 2012. Plant-plant interactions in tropical alpine environments. Perspect. Plant Ecol. Evol. Syst. 14, 363–372.

Anthelme, F., Cavieres, L.A., Dangles, O., 2014. Facilitation among plants in alpine environments in the face of climate change. Front. Plant Sci. 5.

Baptist, F., Choler, P., 2008. A simulation of the importance of length of growing season and canopy functional properties on the seasonal gross primary production of temperate alpine meadows. Ann. Bot. 101, 549–559.

Baptist, F., Yoccoz, N.G., Choler, P., 2010. Direct and indirect control by snow cover over decomposition in alpine tundra along a snowmelt gradient. Plant Soil 328, 397–410.

Beniston, M., Keller, F., Goyette, S., 2003. Snow pack in the Swiss Alps under changing climatic conditions: an empirical approach for climate impacts studies. Theor. Appl. Climatol. 74, 19–31.

Beniston, M., 1997. Variations of snow depth and duration in the Swiss Alps over the last

50 years: links to changes in large-scale climatic forcings. Clim. Change 36, 281–300.

Billings, W.D., 1973. Arctic and alpine vegetations: similarities, differences, and susceptibility to disturbance. Bioscience 23, 697–704.

Brun, E., Vionnet, V., Boone, A., Decharme, B., Peings, Y., Valette, R., Karbou, F., Morin, S., 2013. Simulation of northern eurasian local snow depth, mass, and density using a detailed snowpack model and meteorological reanalyses. J. Hydrometeorol. 14, 203–219.

Callaghan, T.V., Johansson, M., Brown, R.D., Groisman, P.Y., Labba, N., Radionov, V., Bradley, R.S., Blangy, S., Bulygina, O.N., Christensen, T.R., Colman, J.E., Essery, R.L.H., Forbes, B.C., Forchhammer, M.C., Golubev, V.N., Honrath, R.E., Juday, G.P., Meshcherskaya, A.V., Phoenix, G.K., Pomeroy, J., Rautio, A., Robinson, D.A., Schmidt, N.M., Serreze, M.C., Shevchenko, V.P., Shiklomanov, A.I., Shmakin, A.B., Skold, P., Sturm, M., Woo M.-k. Wood, E.F., 2011. Multiple effects of changes in arctic snow cover. Ambio 40, 32–45.

Cannone, N., Pignatti, S., 2014. Ecological responses of plant species and communities to climate warming: upward shift or range filling processes? Clim. Change 123, 201–214.

Carlson, B.Z., Randin, C.F., Boulangeat, I., Lavergne, S., Thuiller, W., Choler, P., 2013. Working toward integrated models of alpine plant distribution. Alp. Bot. 123, 41–53.

Carlson, B.Z., Choler, P., Renaud, J., Dedieu, J.-P., Thuiller, W., 2015. Modelling snow cover duration improves predictions of functional and taxonomic diversity for alpine plant communities. Ann. Bot. 116, 1023–1034.

Carlson, B.Z., Corona, M.C., Dentant, C., Bonet, R., Thuiller, W., Choler, P., 2017. Observed long-term greening of alpine vegetation-a case study in the French Alps. Environ. Res. Lett. 12, 114006. http://dx.doi.org/10.1088/1748-9326/aa84bd.

Choler, P., Michalet, R., 2002. Niche differentiation and distribution of Carex curvula along a bioclimatic gradient in the southwestern Alps. J. Veg. Sci. 13, 851–858.

Choler, P., Michalet, R., Callaway, R.M., 2001. Facilitation and competition on gradients in alpine plant communities. Ecology 82, 3295–3308.

Choler, P., 2005. Consistent shifts in Alpine plant traits along a mesotopographical gradient. Arctic Antarctic Alp. Res. 37, 444–453.

Choler, P., 2015. Growth response of temperate mountain grasslands to inter-annual variations in snow cover duration. Biogeosciences 12, 3885–3897.

Dedieu, J.-P., Carlson, B.Z., Bigot, S., Sirguey, P., Vionnet, V., Choler, P., 2016. On the importance of high-resolution time series of optical imagery for quantifying the effects of snow cover duration on alpine plant habitat. Remote Sens. 8, 481.

Deems, J.S., Painter, T.H., Finnegan, D.C., 2013. Lidar measurement of snow depth: a review. J. Glaciol. 59, 467–479.

Deharveng, L., Perriaux, J., Ravenne, C., 1987. Sédimentologie du flysch des Aiguilles d'Arves (Alpes françaises). Géol. Alp. – Mémoire hors-série 13, 329–341.

Durand, Y., Laternser, M., Giraud, G., Etchevers, P., Lesaffre, B., Merindol, L., 2009a. Reanalysis of 44 yr of climate in the french alps (1958–2002): methodology, model validation, climatology, and trends for air temperature and precipitation. J. Appl. Meteorol. Climatol. 48, 429–449.

Durand, Y., Laternser, M., Giraud, G., Etchevers, P., Lesaffre, B., Mérindol, L., 2009b. Reanalysis of 44 yr of climate in the french alps (1958–2002): methodology, model validation, climatology, and trends for air temperature and precipitation. J. Appl. Meteorol. Climatol. 48, 429–449.

Edwards, A.C., Scalenghe, R., Freppaz, M., 2007. Changes in the seasonal snow cover of alpine regions and its effect on soil processes: a review. Quat. Int. 162, 172–181.

Elmendorf, S.C., Henry, G.H.R., Hollister, R.D., Bjork, R.G., Boulanger-Lapointe, N., Cooper, E.J., Cornelissen, J.H.C., Day, T.A., Dorrepaal, E., Elumeeva, T.G., Gill, M., Gould, W.A., Harte, J., Hik, D.S., Hofgaard, A., Johnson, D.R., Johnstone, J.F., Jonsdottir, I.S., Jorgenson, J.C., Klanderud, K., Klein, J.A., Koh, S., Kudo, G., Lara, M., Levesque, E., Magnusson, B., May, J.L., Mercado-Diaz, J.A., Michelsen, A., Molau, U., Myers-Smith, I.H., Oberbauer, S.F., Onipchenko, V.G., Rixen, C., Schmidt, N.M., Shaver, G.R., Spasojevic, M.J., Porhallsdottir, P.E., Tolvanen, A., Troxler, T., Tweedie, C.E., Villareal, S., Wahren, C.-H., Walker, X., Webber, P.J., Welker, J.M., Wipf, S., 2012. Plot-scale evidence of tundra vegetation change and links to recent summer warming. Nat. Clim. Change 2, 453–457.

Engler, R., Randin, C.F., Vittoz, P., Czaka, T., Beniston, M., Zimmermann, N.E., Guisan, A., 2009. Predicting future distributions of mountain plants under climate change: does dispersal capacity matter? Ecography 32, 34–45.

Ernakovich, J.G., Hopping, K.A., Berdanier, A.B., Simpson, R.T., Kachergis, E.J., Steltzer, H., Wallenstein, M.D., 2014. Predicted responses of arctic and alpine ecosystems to altered seasonality under climate change. Global Change Biol. 20, 3256–3269.

Essery, R., Pomeroy, J., 2004. Vegetation and topographic control of wind-blown snow distributions in distributed and aggregated simulations for an Arctic tundra basin. J. Hydrometeorol. 5, 735–744.

Freppaz, M., Celi, L., Marchelli, M., Zanini, E., 2008. Snow removal and its influence on temperature and N dynamics in alpine soils (Vallee d'Aoste, northwest Italy). J. Plant Nutr. Soil Sci. 171, 672–680.

Gottfried, M., Hantel, M., Maurer, C., Toechterle, R., Pauli, H., Grabherr, G., 2011. Coincidence of the alpine-nival ecotone with the summer snowline. Environ. Res. Lett. 6.

Graae, B.J., De Frenne, P., Kolb, A., Brunet, J., Chabrerie, O., Verheyen, K., Pepin, N., Heinken, T., Zobel, M., Shevtsova, A., Nijs, I., Milbau, A., 2012. On the use of weather data in ecological studies along altitudinal and latitudinal gradients. Oikos 121, 3–19.

Graae, B.J., 2017. Stay or go – how topographic complexity influences alpine plant population and community responses to climate change. Perspect. Plant Ecol. Evol. Syst (in press).

Grime, J.P., 1998. Benefits of plant diversity to ecosystems: immediate, filter and founder effects. J. Ecol. 86, 902–910.

Hantel, M., Hirtl-Wielke, L.M., 2007. Sensitivity of Alpine snow cover to European temperature. Int. J. Climatol. 27, 1265–1275.

Hantel, M., Ehrendorfer, M., Haslinger, A., 2000. Climate sensitivity of snow cover duration in Austria. Int. J. Climatol. 20, 615–640.

Hantel, M., Maurer, C., Mayer, D., 2012. The snowline climate of the Alps 1961–2010. Theor. Appl. Climatol. 110, 517–537.

Harris, C., Arenson, L.U., Christiansen, H.H., Etzemuller, B., Frauenfelder, R., Gruber, S., Haeberli, W., Hauck, C., Holzle, M., Humlum, O., Isaksen, K., Kaab, A., Kern-Lutschg, M.A., Lehning, M., Matsuoka, N., Murton, J.B., Nozli, J., Phillips, M., Ross, N., Seppala, M., Springman, S.M., Muhll, D.V., 2009. Permafrost and climate in Europe Monitoring and modelling thermal, geomorphological and geotechnical responses. Earth-Sci. Rev. 92, 117–171.

Heegaard, E., 2002. A model of alpine species distribution in relation to snowmelt time and altitude. J. Veg. Sci. 13, 493–504.

Inouye, D.W., 2000. The ecological and evolutionary significance of frost in the context of climate change. Ecol. Lett. 3, 457–463.

Körner, C., Hiltbrunner, E., 2017. The 144 ways to describe plant temperature. Perspect. Plant Ecol. Evol. Syst.

Körner, C., Paulsen, J., 2004. A world-wide study of high altitude treeline temperatures. J. Biogeogr. 31, 713–732.

Körner, C., Paulsen, J., Pelaez-Riedl, S., 2003. A bioclimatic characterisation of Europe's alpine areas. Alpine Biodiversity in Europe. Springer, pp. 13–28.

Körner, C., 1999. Alpine Plant Life. Springer Verlag, Berlin.

Keller, F., Goyette, S., Beniston, M., 2005. Sensitivity analysis of snow cover to climate change scenarios and their impact on plant habitats in alpine terrain. Clim. Change 72, 299–319.

Kikvidze, Z., Pugnaire, F.I., Brooker, R.W., Choler, P., Lortie, C.J., Michalet, R., Callaway, R.M., 2005. Linking patterns and processes in alpine plant communities: a global study. Ecology 86, 1395–1400.

Kivinen, S., Kaarlejarvi, E., Jylha, K., Raisanen, J., 2012. Spatiotemporal distribution of threatened high-latitude snowbed and snow patch habitats in warming climate. Environ. Res. Lett. 7.

Kreyling, J., 2010. Winter climate change: a critical factor for temperate vegetation performance. Ecology 91, 1939–1948.

Kudo, G., Nordenhall, U., Molau, U., 1999. Effects of snowmelt timing on leaf traits, leaf production, and shoot growth of alpine plants: comparisons along a snowmelt gradient in northern Sweden. Ecoscience 6, 439–450.

Kullman, L., Richer, A., 2010. Greener and smaller alpine world: review and projection of warming-Induced plant cover change in the swedish scandes. Ambio 39, 159–169.

Lenoir, J., Svenning, J.C., 2015. Climate-related range shifts – a global multidimensional synthesis and new research directions. Ecography 38, 15–28.

Lenoir, J., Gegout, J.-C., Guisan, A., Vittoz, P., Wohlgemuth, T., Zimmermann, N.E., Dullinger, S., Pauli, H., Willner, W., Svenning, J.-C., 2010. Going against the flow: potential mechanisms for unexpected downslope range shifts in a warming climate. Ecography 33, 295–303.

Liston, G.E., Elder, K., 2006. A distributed snow-evolution modeling system (SnowModel). J. Hydrometeorol. 7, 1259–1276.

Marty, C., 2008. Regime shift of snow days in Switzerland. Geophys. Res. Lett. 35.

Pauli, J.N., Zuckerberg, B., Whiteman, J.P., Porter, W., 2013. The subnivium: a deteriorating seasonal refugium. Front. Ecol. Environ. 11, 260–267.

Petty, S.K., Zuckerberg, B., Pauli, J.N., 2015. Winter conditions and land cover structure the subnivium, a seasonal refuge beneath the snow. PLoS One 10.

Pradervand, J.-N., Dubuis, A., Pellissier, L., Guisan, A., Randin, C., 2014. Very high resolution environmental predictors in species distribution models: moving beyond topography? Prog. Phys. Geogr. 38, 79–96.

Rankinen, K., Karvonen, T., Butterfield, D., 2004. A simple model for predicting soil temperature in snow-covered and seasonally frozen soil: model description and testing. Hydrol. Earth Syst. Sci. 8, 706–716.

Revuelto, J., Lopez-Moreno, J.I., Azorin-Molina, C., Vicente-Serrano, S.M., 2014. Topographic control of snowpack distribution in a small catchment in the central Spanish Pyrenees: intra- and inter-annual persistence. Cryosphere 8, 1989–2006.

Rundel, P.W., Smith, A.P., Meinzer, F.C., 1994. Tropical Alpine Environments: Plant Form and Function. Cambridge University Press, Cambridge.

Rundqvist, S., Hedenas, H., Sandstrom, A., Emanuelsson, U., Eriksson, H., Jonasson, C., Callaghan, T.V., 2011. Tree and shrub expansion over the past 34 years at the tree-line near abisko. Sweden Ambio 40, 683–692.

Saccone, P., Morin, S., Baptist, F., Bonneville, J.-M., Colace, M.-P., Domine, F., Faure, M., Geremia, R., Lochet, J., Poly, F., Lavorel, S., Clement, J.-C., 2013. The effects of snowpack properties and plant strategies on litter decomposition during winter in subalpine meadows. Plant Soil 363, 215–229.

Scherrer, D., Korner, C., 2010. Infra-red thermometry of alpine landscapes challenges climatic warming projections. Global Change Biol. 16, 2602–2613.

Scherrer, D., Korner, C., 2011. Topographically controlled thermal-habitat differentiation buffers alpine plant diversity against climate warming. J. Biogeogr. 38, 406–416.

Schmid, M.O., Gubler, S., Fiddes, J., Gruber, S., 2012. Inferring snowpack ripening and melt-out from distributed measurements of near-surface ground temperatures. Cryosphere 6, 1127–1139.

Sturm, M., Schimel, J., Michaelson, G., Welker, J.M., Oberbauer, S.F., Liston, G.E., Fahnestock, J., Romanovsky, V.E., 2005. Winter biological processes could help convert Arctic Tundra to Shrubland. Bioscience 55, 17–26.

Sutinen, M.L., Holappa, T., Kujala, K., 1999. Seasonal changes in soil temperature and in the frost hardiness of Scots pine roots under subarctic conditions: comparison with soil temperature and snow-cover under different simulated winter conditions. Phyton-Annal. Rei Botan. 39, 213–218.

Theurillat, J.-P., Aeschimann, D., Küpfer, P., Spichiger, R., 1994. The higher vegetation units of the alps. Colloq. Phytosociol. 23, 189–239.

Vionnet, V., Brun, E., Morin, S., Boone, A., Faroux, S., Le Moigne, P., Martin, E., Willemet, J.M., 2012. The detailed snowpack scheme Crocus and its implementation in SURFEX v7.2. Geosci. Model Dev. 5, 773–791.

Walker, D.A., Halfpenny, J.C., Walker, M.D., Wessman, C.A., 1993. Long-term studies of snow-vegetation interactions. Bioscience 43, 471–480.

Wang, G., Li, Y., Hu, H., Wang, Y., 2008. Synergistic effect of vegetation and air temperature changes on soil water content in alpine frost meadow soil in the permafrost region of Qinghai-Tibet. Hydrol. Process. 22, 3310–3320.

Wheeler, J.A., Hoch, G., Cortes, A.J., Sedlacek, J., Wipf, S., Rixen, C., 2014. Increased spring freezing vulnerability for alpine shrubs under early snowmelt. Oecologia 175, 219–229.

Wipf, S., Stoeckli, V., Bebi, P., 2009. Winter climate change in alpine tundra: plant responses to changes in snow depth and snowmelt timing. Clim. Change 94, 105–121.

Wundram, D., Pape, R., Loffler, J., 2010. Alpine soil temperature variability at multiple scales. Arct. Antarct. Alp. Res. 42, 117–128.

Zheng, D., Hunt Jr., E.R., Running, S.W., 1993. A daily soil temperature model based on air temperature and precipitation for continental applications. Clim. Res. 2, 183–191.

Zinger, L., Shahnavaz, B., Baptist, F., Geremia, R.A., Choler, P., 2009. Microbial diversity in alpine tundra soils correlates with snow cover dynamics. ISME J. 3, 850–859.

Zinger, L., Lejon, D.P.H., Baptist, F., Bouasria, A., Aubert, S., Geremia, R.A., Choler, P., 2011. Contrasting diversity patterns of crenarchaeal, bacterial and fungal soil communities in an alpine landscape. PLoS One 6, 7.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 16–21

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Review

The 90 ways to describe plant temperature[☆]

Christian Körner[*], Erika Hiltbrunner

Institute of Botany, University of Basel, Schönbeinstrasse 6, 4056 Basel, Switzerland

ARTICLE INFO

Keywords:
Alpine
Climatology
Elevation
Energy balance
Fundamental niche
Plant canopy
Phenology

ABSTRACT

What seems like a trivial task is one of the most difficult ones in functional plant ecology and biogeography: selecting the appropriate measures of temperature for an ecologically meaningful description of habitat conditions and for a mechanistic understanding of responses of plants. The difficulty becomes even more severe at elevations above the climatic tree limit, where plant stature, topography and seasonal snow cover interact in producing temperature conditions that largely deviate from weather station records. Temporal resolution and the distinction between extremes and means for biogeographic applications are emphasized. We summarize the key issues in handling temperature as a driver of plant life in general and in high elevation ecosystems in particular. Future directions in plant-temperature research at high elevation need to resolve the thermal species range limit issues (identify the fundamental temperature niche) and the complex controls of plant development (phenology) in a topography context.

1. Introduction

Together with water and nutrient availability, temperature is a most prominent determinant of the fundamental niche of organisms across all types of terrestrial ecosystems. Temperature is possibly the most widely acknowledged abiotic environmental driver of plant life in high elevation ecosystems. While moisture and nutrients may vary from place to place, with no systematic latitudinal or elevational pattern, air temperature is commonly assumed to follow globally consistent patterns with altitude, associated with the decline in atmospheric pressure (Körner, 2003). For biologists it is common practice to rely on weather station data to describe plant habitat conditions or to define or model a plant species' niche as defined by abiotic drivers such as temperature (e.g., Pellissier et al., 2013; Box, 2016). In this essay, we explore the various ways by which temperature acts upon plants and the various ways the temperature regime can be documented in less or more meaningful ways.

2. Which temperature?

The most basic difference in the action of temperature on plant life is that between its gradual (warmer or colder) and its threshold (extremes) role. Thus, explaining temperature effects on plants best starts with hypotheses related to that dichotomy of action. Different metrics of temperature are required to match these two categories of action. Whatever the focus, the temperature that matters, is that actually imposed on the processes/organs of interest and at a temporal resolution that matches with the biological reactions. For instance, extremes can impact plants within minutes (freezing damage), while gradual influences of temperature (metabolism) may materialize over days or months. Hence, both the time scale and the location of the action of temperature are crucial. When only three major points of action (air-plant-soil) and various measures of temperature are combined, one arrives at 90 ways to describe the temperature regime of a plant (Fig. 1). Some of these are key to test various biological hypotheses, others may be rather meaningless, although widely used. Had we included plant organ type (leaf, bud, flower, stem, shallow versus deep roots), the number of temperature measures would boost to one thousand. Including various statistical calculations such as frequency distribution or recurrence rates, the number would rise further.

Quite obviously, the most inappropriate of all measures of temperatures are those neither related to the critical location of action, nor accounting for the critical time scale of action, best exemplified by mean annual air temperature (MAT) obtained from a weather station or a data base, and neglecting the central role of seasonality of life and phenology. Why could MAT become the most widely used measure of temperature, when life conditions of plants in the field are described, for instance in the material and methods section of papers? We assume this is related to data availability and convenience. Here, we suggest to ban the use of MAT outside the tropics from the scientific literature on

---

[☆] This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author.

E-mail address: ch.koerner@unibas.ch (C. Körner).

http://dx.doi.org/10.1016/j.ppees.2017.04.004
Received 30 December 2016; Received in revised form 27 March 2017; Accepted 9 April 2017

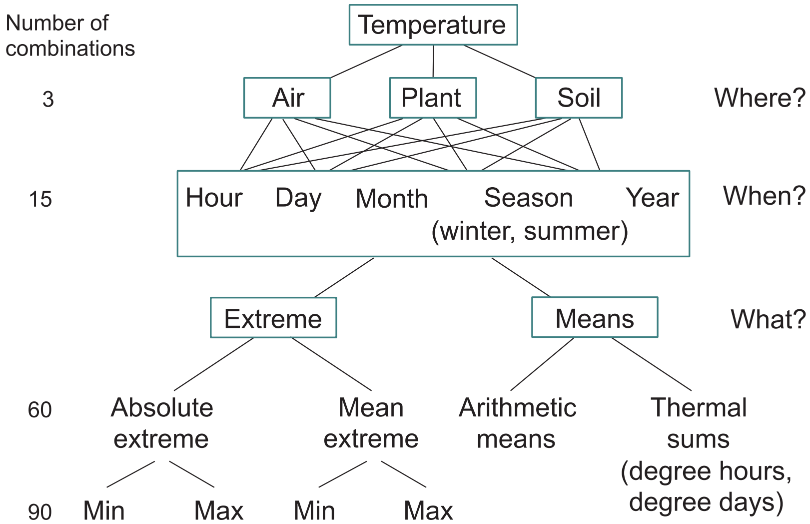


Fig. 1. The combination of temporal resolution and position (where temperature is obtained from) and various measures of temperature leads to 90 ways to describe plant temperature. The catalogue can be further expanded by accounting for temperatures experienced by different plant organs.

plant and animal life, and be it only for the sake of acknowledging the fundamental role of seasonality that led to the so-called climate diagrams that were introduced to comparative ecology ca. 60 years ago (Walter, 1955; Walter and Lieth, 1960–1973).

To illustrate the pitfalls of applying MAT in ecology, we selected data from three weather stations along a W-E gradient from Cork in Ireland (9.4 °C), to Basel in Switzerland (9.9 °C) and Almaty (former Alma Ata) in Kazakhstan (10.0 °C), with almost the same MAT (Fig. 2). While one can grow subtropical palm species and tree *Rhododendrons* in Cork, only few tree taxa survive −38 °C in Almaty, which at the same time have to cope with 43 °C in summer, while the 30-year-hourly record reading for Cork is 29 °C. The main problem with such annual means is that they attribute (arithmetically) similar ecological meaning to the dormant season as to the growing season, with the first, perhaps an issue of freezing tolerance, and the second an issue of metabolism. Such 'average' annual life conditions are disregarding the actual seasonal life cycle (phenology).

Using a more appropriate temporal resolution, ranging from at least seasonal or monthly resolution to temperature extremes, the way temperatures are handled often remains doubtful. For instance, the expression 'minimum temperature' is often applied without specifying whether this refers to the 'absolute minimum' of temperature within a defined period, that is, the lowest ever measured temperature at hourly or shorter intervals, or whether this stands for a 'mean minimum'. The absolute minimum of temperature could be decisive for existence/survival, even if occurring only once in a century. In contrast, the mean minimum temperature that is the lowest daily or monthly mean in a year has a very different, if any, ecological meaning. When a reference is made to absolute minima, this is obviously with survival issues in mind, rendering mean minima irrelevant. The most apt way to refer to minima is the absolute minimum temperature a specific organ or tissue is experiencing, and because such minima are not occurring every year, such extremes can only be obtained from long data series that permit calculating their recurrence rate (Kollas et al., 2014a). Such analysis would, for instance, reveal that a critical temperature of −18 °C would occur, statistically, every 20 years. Not a survival issue for annual or perennial plants that persist in the seed bank, but a fatal event for a long-lived plant with no lasting seed bank and whose meristems (buds, cambium) do not tolerate such a temperature even in a fully hardened state. It also matters in which month of the year (phenological state of the plant) such a temperature extreme does recur (see phenology below).

3. The influence of plant stature

A most critical issue is the difference between the actual climate experienced by plants (often termed microclimate) and the climate reported from weather stations, that is, air temperature in a shelter. In tall stature, narrow leaved plants such as in many tree taxa the difference between air and canopy foliage temperature may be small (commonly < 2.5 K), but with noteworthy exceptions in trees with broad leaves and dense canopies (4–5 K; Leuzinger and Körner, 2007). That means for many tree species, canopy temperature may be readily predicted from weather station data, except for calm and sunny conditions (Kollas et al., 2014b; Paulsen and Körner, 2014). When smaller stature plants are considered (e. g., grasses, herbs, dwarf shrubs), aerodynamic decoupling from free atmospheric circulation creates vast deviations of life conditions from that in ambient air 2 m above the ground (Geiger, 1965; Körner, 2003; Larcher, 2012). This even holds for the interior of flowers, where ovules may be far warmer than what

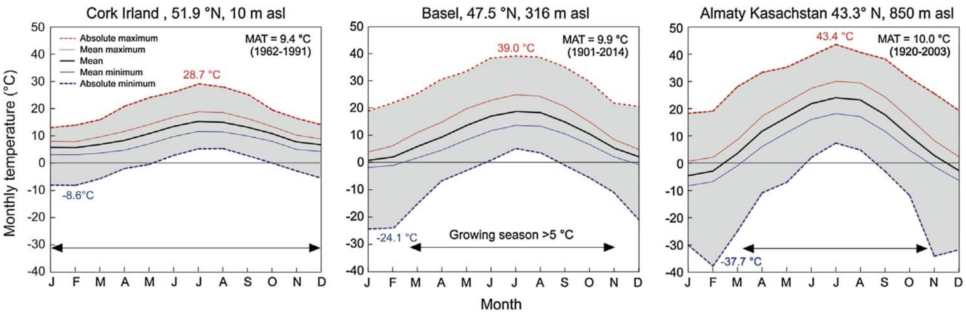


Fig. 2. Climate diagrams for three locations with different life conditions at otherwise similar mean annual temperature (MAT).

would be expected from surrounding air temperature (Dietrich and Körner, 2014). In alpine ecosystems, the growing season means of meristematic (apical) temperatures at or slightly below ground surface, that is, at the location where new tissues are produced, can deviate from the concurrent seasonal mean of air temperature recorded by a weather station at the same elevation by 4–8 K (up to 12 K), depending on topography and plant height (Scherrer and Körner, 2009). Thus, the actual life conditions at 2500 m elevation can be similar to those at 1000 m elevation. This fundamental difference in actual temperature at plant level between small stature (< 20 cm) and tall (> 2 m) plants explains the existence of the high elevation treeline globally. Trees simply cannot decouple their canopy from free atmosphere conditions, whereas small plants effectively do, especially, when solar radiation is heating the plant canopy during daytime (Körner, 2012; Paulsen and Körner, 2014). Seedlings and small stature saplings profit from the warmth near the ground, the reason, why these early life stages of tree taxa often occur high above the tree limit, but do not tell us whether trees could grow at locations where seedlings can.

Among small stature, non-tree taxa, the actual height of buds, leaves or inflorescences above ground and their spatial arrangement exert a strong influence on these microclimatic effects (Figs. 3 and 4). While alpine cushion plants may experience humid tropical conditions on bright summer days, nearby long grass leaves may be coupled to air temperature just like trees are, but their meristems and the expanding leaf bases profit from ground surface heating under solar radiation. A classical example is the influence of plant height among alpine dwarf shrubs (Fig. 3; Cernusca, 1976). While dense 40–60 cm tall *Rhododendron* may heat up by 5–10 K above air temperature (measured at 2 m height), the 3 cm thick carpets of *Loiseleuria* may exceed air temperature by 10–15 K, with intermediate stature *Vaccinium* holding an intermediate position (not shown). Fig. 3 also illustrates the role of snow cover. When plants are covered by snow, weather station signals have no meaning. Hence, snow cover duration becomes crucial. *Loiseleuria* grows at locations that often lack snow cover in winter with air temperature of −10 °C during clear nights, rapidly followed by noon temperatures of up to +30 °C in late winter (March), exerting high demands on stringent endodormancy (see phenology below). Soils rarely freeze under snow in temperate mountains, causing microbial processes to continue throughout winter.

Plant stature also matters in cold nights under clear sky during the growing season (Fig. 4). Under such conditions, sky exposed surfaces may cool substantially below ambient air temperature through heat losses by longwave outgoing radiation (Jones, 2014). The magnitude of such cooling depends on concurrent heat convection. The faster the loss of thermal energy is replaced by convective heat transfer from the surrounding air, the smaller this radiation cooling will be. This explains, why such night time cooling is less pronounced in tall structures compared to prostrate structures in which convective heat transfer is reduced by a large boundary layer resistance. Hence, canopies of small stature plants not only heat up substantially during daytime due to a high boundary layer resistance to heat transfer, but the same aerodynamic boundary layer effects also cause a more negative heat balance during clear nights. The aerodynamic resistance to heat exchange is not only tied to stature (height) but also to leaf shape and the arrangement of foliage (leaf area density), and so is the plant canopy temperature.

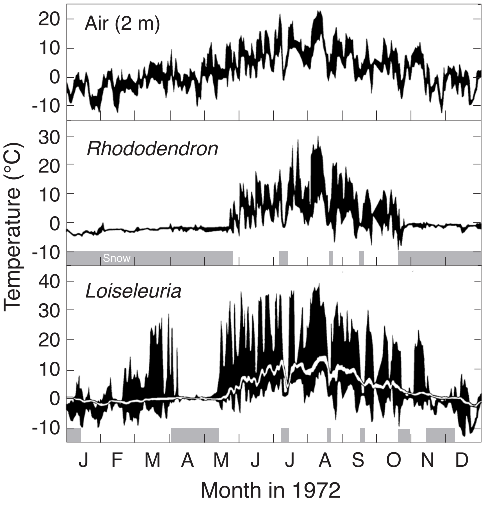


Fig. 3. The influence of plant height and snow cover on life conditions in alpine dwarf shrubs The black area shows the amplitude between daily minimum and maximum temperature, the white strip (lowest diagram) the amplitude in −10 cm soil depth. Snow cover duration is indicated by grey bars (redrawn from Cernusca, 1976).

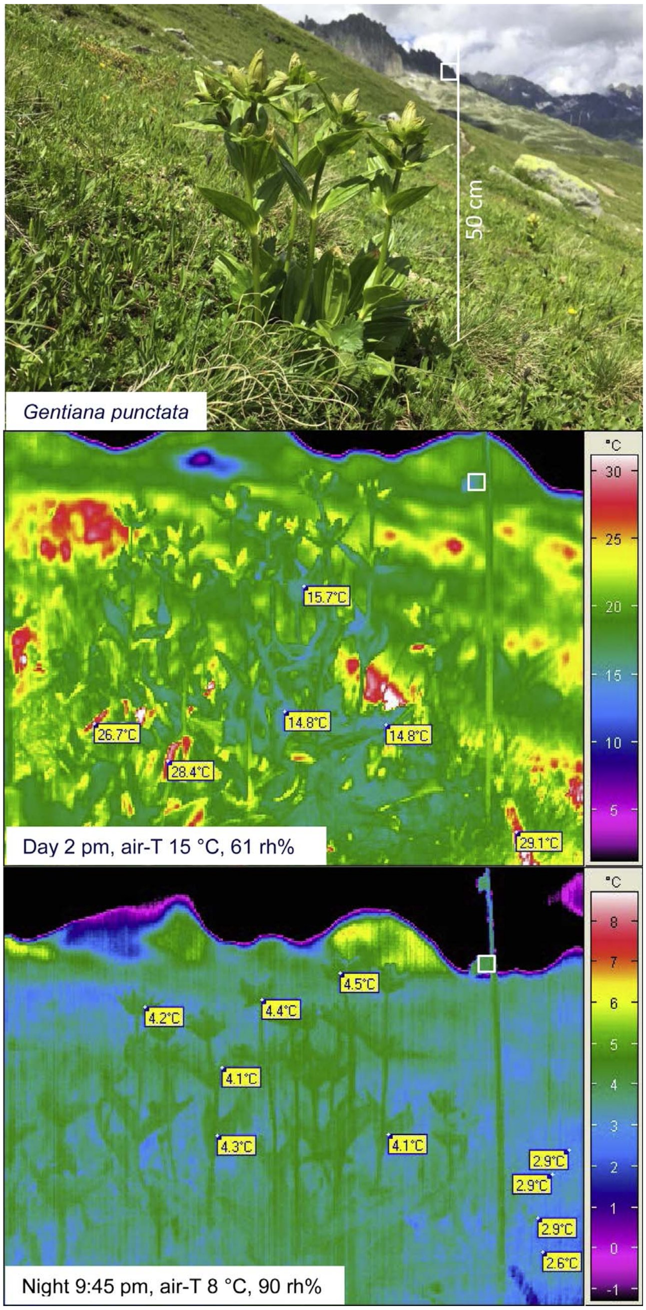


Fig. 4. The effect of plant height on temperature in herbaceous plants. Thermal images of alpine grassland at 2440 m elevation in the Swiss central Alps with clusters of tall *Gentiana punctata* (50 cm plant height). Note the difference between air temperature and plant temperature.

It has long been known that freezing can be more severe near the ground ('ground frost') than in upright structures (Squeo et al., 1991).

Ice formation such as hoar frost may 'interfer' with the heat balance by releasing energy through ice formation (crystallisation energy, see Monteith and Unsworth, 2013). For decades, farmers have employed sprinklers to protect orchard trees from freezing damage during clear, calm nights in early spring when flowers or newly developing fruits are at risk of low-temperature injuries by even small minus temperatures. The heat release during freezing maintains the foliage temperature at around freezing point, which is often sufficient to avoid injury (e.g, Businger, 1965). Similar processes come into play during clear nights in alpine settings, when atmospheric humidity reaches the dew point on cold canopy surfaces. Such surface phenomena are critical for high elevation plant reproductive success, and are not captured by data from weather stations.

4. The role of phenology

From the above, it is obvious that any measure of thermal life conditions needs to account for plant phenology, i.e., the developmental stage and metabolic activity associated with it. Temperatures during the dormant season (endodormancy), during periods of snow cover, during the stage of ecodormancy (ready to go but thermal inhibition of development), the flushing and flowering stage, the stage of leaf maturation and seed set, and the transition to dormancy in autumn have very different meaning and impact. Thus, temperature not only influences these developmental processes, its influence on plant processes also varies with phenology throughout the year. When plants are endodormant, they are little or not responsive to temperature. When plants entered ecodormancy (e.g., via a photoperiod signal), they become receptive to temperature (Keller and Körner, 2003; Li et al., 2016a,b). However, as the photoperiod signal is identical in spring and autumn, chilling requirements during the dormant phase are key for the subsequent development. Winter chill phenomena are well known in fruit trees with effects on yield and fruit quality (Atkinson et al., 2004). For alpine plants, moist-chilling requirements have been reported for seedling emergence in combination with light demands (Shimono and Kudo, 2005). Hardly anything is known in alpine plants about carry-over-effects of chilling experience on later developmental stages. Because phenology differs among co-occurring taxa, they utilize and are adapted to different thermal 'niches' of the season for key developmental steps (Fig. 5). This means, any attempt at modelling a species range limit using meteorological information, needs to account for the species specific phenology. The same low temperature that may kill freshly unfolded foliage or flowers may be harmless for mature organs and tissues a few weeks later. Fig. 5 illustrates the broad spectrum of plant phenologies across an alpine summer, obviously exposing the critical pheno-phases to different temperature regimes in different species (e.g., early *versus* late flowering species).

Thus, phenology is under an internal (genetic) and an external (forcing) control. In alpine plant species that are at risk to experience a periodic lack of snow cover (convex topography), the genetic, internal control must be stronger than in plants with secure snow cover, such as snow bed species. When plants confined to locations with secure winter snow cover are artificially exposed to the direct influence of winter weather conditions, they commonly die (Larcher and Siegwolf, 1985). In addition, the late season transition to winter hardiness before the demanding low temperatures have arrived, requires temperature independent controls of development, in essence using the secure day length signal (photoperiod; Keller and Körner, 2003). Phenology clearly does not show a uniform sensitivity to temperature across the year (e.g., temperature is exerting no advance in phenology when plants are endodormant, which is crucial when snow is lacking in winter). Another central role of phenology is to secure gene-flow among individuals by synchronous flowering. Synchrony of flowering across different snow melt regimes requires a photoperiodic control with temperature having a modulative influence only (e.g., Bienau et al., 2015).

This rises the interesting question of which subpopulations of metapopulations are participating in sexual reproduction across varied alpine topography, with snow release differing by up to two months across habitats of the same species. Can snow melt regimes segregate reproductive guilds among the same taxon? This seems to be the case in the dominant alpine sedge *Carex curvula*, a wind pollinated species that flowers a few days after snow melt irrespective of the date of snow melt within a growing season, in contrast to *Nardus stricta* that leans towards a common flowering date, no matter when snow melted, very similar to what had been observed in insect pollinated *Empetrum hermaphroditum* in northern Sweden (Bienau et al., 2015). *C. curvula* and *N. stricta* are important grassland species in the alpine belt of the European Alps. Most of the mid to late flowing species in Fig. 5 belong to the snow-melt-time independent flowering type (presumably photoperiod controlled). In insect pollinated species, such differences in phenology may

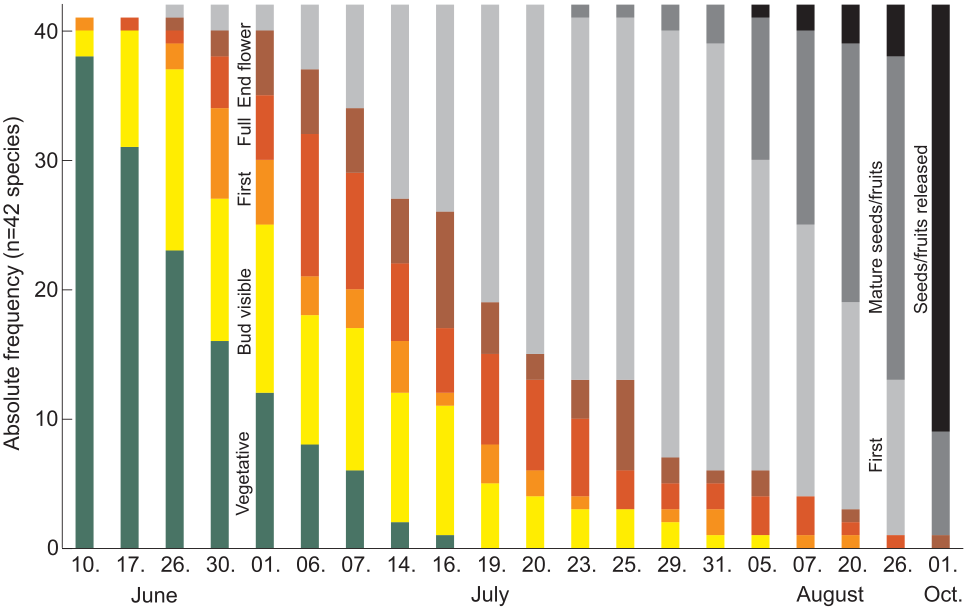


Fig. 5. The seasonal course of flowering and fruiting phenology in an alpine grassland at 2430 m elevation at Furka Pass in the Swiss central Alps (unpublished data). The diagram shows the fraction of species (42 species in total) at different phenological stages between June 6 (5 days after snow melt) and October 1, 2015 (last seed release, complete senescence). To improve readability bars were regularly spaced.

also reflect temperature effects on pollinators, although in high elevation ecosystems, pollinator-insect interactions are commonly less tight (flies becoming increasingly important with increasing elevation).

The net outcome of such different actions of temperature on phenology and the different sensitivity to temperature at different developmental states will be a differential response of species to any climate warming and to snow melt scenarios (Inouye, 2008). So the action of temperature and thus, the measures of temperature used to explain such phenomena will differ for different species, calling for a rather careful selection of temperature measures and their application in any predictive attempt (modelling). A most critical issue is freezing resistance, which undergoes a very pronounced seasonal course and is very species and organ specific (Larcher et al., 2010). When the distribution of species is considered, means of temperature are commonly inept or only indirectly related, with the length of favourable conditions for growth and the extremes during critical periods far more relevant (Zimmermann et al., 2009). For such inferences, detailed ground truth data are needed for growth constraints such as those by Nagelmüller et al. (2017) or phenology-weighted freezing resistance (Körner et al., 2016).

5. Better data handling

In many cases, the actually needed data will not be available, although the collection of such data was never as simple and cheap as today. One way out of applying weather station data to situations where plants definitively experience a climate very different from atmospheric conditions (as is the case above treeline), is the application of empirically obtained probability functions. Scherrer and Körner (2009) provided variance terms for typical alpine settings, that worked similarly well in temperate alpine, arctic and high arctic environments. Once the spectrum of mean temperature deviations across alpine or arctic topography is known (the data produced by Scherrer and Körner, 2009; offer some guidance) a more realistic spectrum of temperature regimes for a given landscape can be applied. While it can reasonably be expected that the entire pattern of thermal mosaics will move upslope under climatic warming, such probability functions will account for topography driven variance, and thus, will help avoiding unfounded predictions of habitat-, and species-losses. Using such a topography driven temperature variation, it could be shown that most alpine thermal habitat types will be retained in a 2 K warming scenario (Scherrer and Körner, 2011), while weather station and lapse rate based estimates would predict great losses (Loarie et al., 2009). This insight also underpins, why spatially fine resolution species distribution models (SDMs) yield far more realistic scenarios for alpine environments than coarse resolution approaches (Randin et al., 2009), although downscaling climatic layers to actual habitat conditions remains to be resolved. Further, there are relatively straight forward routines to model mean snow cover on even terrain (see Paulsen and Körner, 2014), which is the most decisive factor that decouples plants from atmospheric conditions in alpine environments. A more challenging task is to account for snow allocation (e.g. snow bed conditions).

It is highly recommended that researchers employ own networks of temperature loggers for ground-truth information instead of relying on down-scaling weather station data (e.g., Körner et al., 2003; data for European mountains). Yet, some basic meteorological skills are required to obtain unbiased data. For high elevation situations, one advantage is that most species have their vital perennial structures, apical meristems in particular, below ground. This permits collecting most relevant information at ca. 3–5 cm below ground without any visible installations and without running into shelter problems. The problematic part is that at such shallow soil depth, the actual ground cover will exert a very strong influence on the temperatures recorded, particularly during sunshine hours. So, loggers buried at the same site under a tussock or under an open dark soil surface may produce data as different as air temperatures from sites separated by 1000 m in elevation. This calls for very careful and standardised placement and appropriate replication. Since such data will require an on-site air temperature reference to link up with weather stations (see e. g., Kollas et al., 2014b), biologists are advised at employing professional screens, rather than building their own. Self-made shading cups, particularly when placed too close to the ground, can be worse than no shading at all, with its classical radiation bias. A single-walled cup produces re-radiation to the sensor underneath and traps both heat convection and heat radiation from the ground. For assessing actual canopy temperatures, high resolution infra-red imaging at defined weather conditions and view angles are very helpful (e.g., Scherrer and Körner, 2009). Such devices also provide insight into radiation driven surface cooling in clear nights (Fig. 4).

5.1. Future directions of research

Many people think nothing is more central to plant ecology than understanding the potential range limits of taxa, thus, defining their fundamental niche. Knowing the conditions that define species' distribution boundaries is the foundation of a mechanism based projection of potential species distribution, including that under novel environmental conditions. In the context of climatic warming, knowing thermal range limits is absolutely crucial (Walther et al., 2005). The popular space-for-time modelling of plant distribution in a warmer climate presents a lot of power and realism based on plausibility, but it lacks an account of ground truth conditions as well as accounting for the decisive facets of the temperature regime. The onset and duration of the growing season, the actual temperatures in growing tissues, the magnitude of extremes and their timing, and how all these are associated with certain means of air temperature and interact with photoperiod, topography, water and nutrient availability make it very difficult to predict if, and to which new range edge, plants may spread.

For instance, the freezing resistance during flowering (of ovaries in particular) is largely unknown for alpine and arctic plants. As a starting point, we urgently need more temperature records for edge populations for a broad spectrum of alpine species (for a conceptual framework for such surveys see Körner et al., 2016). In trees, including those at treeline, such works profit from the close relationship between air and crown temperatures (Kollas et al., 2014b; Paulsen and Körner, 2014). In alpine plants, air temperature is a poor proxy (as discussed above), but sharp small-scale climatic gradients such as along snow melt gradients offer experimental fields to explore thermal range limits (e.g., Scherrer and Körner, 2011; Sedlacek et al., 2015) and to delineate their causes.

Once such thermal range limits have been identified, the next question is how do plants adjust and control their seasonal activity to meet their specific thermal requirements. Phenology, that is the visual part of development, is the gateway toward escaping unfavourable, and select for favourable life conditions. Some species may be flexible, others follow a more conservative (genetic) program. The most promising avenue toward arriving at a mechanistic understanding of phenology is the combination of (a) collecting high resolution phenological data associated with appropriate temperature records over steep local thermal gradients, (b) transplant experiments, including common gardens (e.g., Prock and Körner, 1996; Scheepens and Stocklin, 2013; Alexander et al., 2015; Sedlacek et al., 2015), and (c) controlled environment studies along the lines of Keller and Körner (2003). Such data would best be associated with the seasonal dynamics of freezing tolerance for vital organs.

More traditional aspects of plant temperature responses such as photosynthetic or respiratory responses reveal very little, if any, explanatory power for plant performance in cold environments (see the discussion in Körner, 2003). There is really no evidence that plants in cold climates are carbon limited (for alpine plants see Inauen et al., 2012), a field, best explored for low temperature treelines (Hoch and Körner, 2012; Fajardo et al., 2012). Due to their stature and thus, aerodynamic coupling, trees arrive at thermal range limits at lower 'isolines' of elevation than small stature plants (Körner, 2007). Whenever studied, processes associated with tissue formation were found far more sensitive to temperature than is known for photosynthesis (for references see Nagelmüller et al., 2017). Plants from cold climates exhibit slightly lower temperature thresholds for growth, not permitting a strong differentiation among taxa (absolute growth limits between 0 and 2 °C, bulk tissue production > 5 °C; see Li et al., 2016a,b; Nagelmüller et al., 2017). Finally, life strategy may exert strong limitations to a rapid range shift in response to temperature. Many alpine taxa show some form of clonal growth, permitting them to inhabit certain locations for thousands of years, irrespective of centennial temperature variations (De Witte and Stöcklin, 2011; De Witte et al., 2012).

6. Conclusions

Temperature is often considered the simplest to obtain environmental information, which clearly is not the case, particularly, in high elevation ecosystems. High elevation vegetation above the tree limit is of low stature and thus creates its own thermal environment in strong interaction with topography. In addition, snow cover decouples plants entirely from atmospheric conditions. This makes annual means or records that do not account for snow cover biologically meaningless. Appropriate judgement of the action of temperature on plants requires information on topographic habitat preferences of the taxa and their phenology. Phenology is the key to understand the action of temperature in all ecosystems, but alpine ones in particular. Of the 90 possibilities of measures of temperature illustrated in Fig. 1, a few really matter, with temporal resolution and the distinction of extremes from means as well as air from organ temperature being the most vital issues. It is recommended that plant ecologists collect their own ground truth data rather than to rely on inferences from data bases and weather stations, which otherwise may provide reliable information for forest trees, including those at high elevation, with their established tight coupling to atmospheric conditions. The most pending questions regarding the action of temperature on plants in cold climates relate to explaining and defining species range limits and the controls of plant phenology. Plant phenology commonly sets the time-frames for plant activity so that temperatures hardly affect metabolism, given the air conditioning effect of low stature vegetation (Körner, 2003). Exceptions are plants that inhabit the most extreme locations, outposts without shelter (e.g., Körner, 2011). At such locations it is possible to explore the ultimate, most basic mechanisms by which plants cope with low temperature.

References

Alexander, J.M., Diez, J.M., Levine, J.M., 2015. Novel competitors shape species' responses to climate change. Nature 525, 515–518.

Atkinson, C.J., Sunley, R.J., Jones, H.G., Brennan, R., Darby, P., 2004. Winter Chill in Fruit. UK Department for Food, Environment and Rural Affairs, London Report No. CTC0206.

Bienau, M.J., Kröncke, M., Eiserhardt, W.L., Otte, A., Graae, B.J., Hagen, D., Milbau, A., Durka, W., Eckstein, R.L., 2015. Synchronous flowering despite differences in snowmelt timing among habitats of Empetrum hermaphroditum. Acta Oecol. 69, 129–136.

Box, E.O., 2016. World bioclimatic zonation. Vegetation Structure and Function at Multiple Spatial, Temporal and Conceptual Scales. Geobotany Studies, Springerpp. 3–52.

Businger, J.A., 1965. Frost protection with irrigation. Agric. Meteorol. 6, 74–80.

Cernusca, A., 1976. Bestandesstruktur, Bioklima und Energiehaushalt von alpinen Zwergstrauchbeständen. Oecol. Plant 11, 71–102.

De Witte, L.C., Stöcklin, J., 2011. Horizontal growth in arctic-alpine clonal plants is not affected by climatic variability among regions. Plant Ecol. Divers. 4, 329–340.

De Witte, L.C., Armbruster, G.F.J., Gielly, L., Taberlet, P., Stöcklin, J., 2012. AFLP markers reveal high clonal diversity and extreme longevity in four key arctic-alpine species. Mol. Ecol. 21, 1081–1097.

Dietrich, L., Körner, C., 2014. Thermal imaging reveals massive heat accumulation in flowers across a broad spectrum of alpine taxa. Alp. Bot. 124, 27–35.

Fajardo, A., Piper, F.I., Pfund, L., Körner, C., Hoch, G., 2012. Variation of mobile carbon reserves in trees at the alpine treeline ecotone is under environmental control. New Phytol. 195, 794–802.

Geiger, R., 1965. The Climate near the Ground. Harvard University Press, Cambridge, MA.

Hoch, G., Körner, C., 2012. Global patterns of mobile carbon stores in trees at the high-elevation tree line. Global Ecol. Biogeogr. 21, 861–871.

Inauen, N., Körner, C., Hiltbrunner, E., 2012. No growth stimulation by CO_2 enrichment in alpine glacier forefield plants. Global Change Biol. 18, 985–999.

Inouye, D.W., 2008. Effects of climate change on phenology, frost damage, and floral abundance of montane wildflowers. J. Ecol. 89, 353–362.

Jones, H.G., 2014. Plants and Microclimate: A Quantitative Approach to Environmental Plant Physiology, 2nd ed. Cambridge University Press, Cambridge.

Körner, C., Paulsen, J., Pelaez-Riedl, S., 2003. A bioclimatic characterisation of Europe's alpine areas. In: Nagy, L., Grabherr, G., Körner, C., Thompson, D.B.A. (Eds.), Alpine Biodiversity in Europe. Springer, Berlin, pp. 13–28 Ecol. Studies 167.

Körner, C., Basler, D., Hoch, G., Kollas, C., Lenz, A., Randin, C.F., Vitasse, Y., Zimmermann, N.E., 2016. Where, why and how? Explaining the low-temperature range limits of temperate tree species. J. Ecol. 104, 1076–1088.

Körner, C., 2003. Alpine Plant Life, 2nd ed. Springer, Berlin.

Körner, C., 2007. Climatic treelines: conventions, global patterns, causes. Erdkunde 61, 315–324.

Körner, C., 2011. Coldest places on earth with angiosperm plant life. Alp. Bot. 121, 11–22.

Körner, C., 2012. Treelines will be understood once the functional difference between a tree and a shrub is. Ambio 41, 197–206.

Keller, F., Körner, C., 2003. The role of photoperiodism in alpine plant development. Arct. Antarct. Alp. Res. 35, 361–368.

Kollas, C., Körner, C., Randin, C.F., 2014a. Spring frost and growing season length co-control the cold range limits of broad- leaved trees. J. Biogeogr. 41, 773–783.

Kollas, C., Randin, C.F., Vitasse, Y., Körner, C., 2014b. How accurately can minimum temperatures at the cold limits of tree species be extrapolated from weather station data? Agr. For. Meteorol. 184, 257–266.

Larcher, W., Siegwolf, R., 1985. Development of acute frost drought in Rhododendron ferrugineum at the alpine timberline. Oecologia 67, 298–300.

Larcher, W., Kainmüller, C., Wagner, J., 2010. Survival types of high mountain plants under extreme temperatures. Flora 205, 3–18.

Larcher, W., 2012. Bioclimatic temperatures in the high Alps. In: Lütz, C. (Ed.), Plants in Alpine Regions. Springer, Wien, pp. 21–27.

Leuzinger, S., Körner, C., 2007. Tree species diversity affects canopy leaf temperatures in a mature temperate forest. Agric. For. Meteorol. 146, 29–37.

Li, L., Li, Z., Cadotte, M.W., Jia, P., Chen, G., Jin, L.S., Du, G., 2016a. Phylogenetic conservatism and climate factors shape flowering phenology in alpine meadows. Oecologia 182, 419–428.

Li, X., Rossi, S., Liang, E., Camarero, J.J., 2016b. Temperature thresholds for the onset of xylogenesis in alpine shrubs on the Tibetan Plateau. Trees 30, 2091–2099.

Loarie, S.R., Duffy, P.B., Hamilton, H., Asner, G.P., Field, C.B., Ackerly, D.D., 2009. The velocity of climate change. Nature 462, 1052–1055.

Monteith, J.L., Unsworth, M.H., 2013. Principles of Environmental Physics, 4th ed. Academic Press, Elsevier.

Nagelmüller, S., Hiltbrunner, E., Körner, C., 2017. Low Temperature Limits for Root Growth in Alpine Species are Set by Cell Differentiation. AoB PLANTS.

Paulsen, J., Körner, C., 2014. A climate-based model to predict potential treeline position around the globe. Alp. Bot. 124, 1–12.

Pellissier, L., Vittoz, P.A., Dubuis, A., Meier, A.S., Zimmermann, N.E., Randin, C.F., Thuiller, W., Garraud, L., et al., 2013. Thermal niches are more conserved at cold than warm limits in arctic-alpine plant species. Global Ecol. Biogeogr. 22, 933–941.

Prock, S., Körner, C., 1996. A cross-continental comparison of phenology, leaf dynamics and dry matter allocation in arctic and temperate zone herbaceous plants from contrasting altitudes. Ecol. Bull. 45, 93–103.

Randin, C.F., Engler, R., Normand, S., Zappa, M., Zimmermann, N.E., Pearman, P.B., Vittoz, P., Thuiller, W., Guisan, A., 2009. Climate change and plant distribution: local models predict high-elevation persistence. Global Change Biol. 15, 1557–1569.

Scheepens, J.F., Stocklin, J., 2013. Flowering phenology and reproductive fitness along a mountain slope: maladaptive responses to transplantation to a warmer climate in Campanula thyrsoides. Oecologia 171, 679–691.

Scherrer, D., Körner, C., 2009. Infra-red thermometry of alpine landscapes challenges climatic warming projections. Global Change Biol. 16, 2602–2613.

Scherrer, D., Körner, C., 2011. Topographically controlled thermal-habitat differentiation buffers alpine plant diversity against climate warming. J. Biogeogr. 38, 406–416.

Sedlacek, J., Wheeler, J.A., Cortes, A.J., Bossdorf, O., Hoch, G., Lexer, C., Wipf, S., Karrenberg, S., van Kleunen, M., Rixen, C., 2015. The response of the Alpine dwarf shrub Salix herbacea to altered snowmelt timing: lessons from a multi-site transplant experiment. PLoS One 10.

Shimono, Y., Kudo, G., 2005. Comparisons of germination traits of alpine plants between fellfield and snowbed habitats. Ecol. Res. 20, 189–197.

Squeo, A., Rada, F., Azocar, A., Goldstein, G., 1991. Freezing tolerance and avoidance in high tropical Andean plants: is it equally represented in species with different plant height? Oecologia 86, 378–382.

Walter, H., Lieth, H., 1960–1973. Klimadiagramm-Weltatlas. In drei Lieferungen mit etwa 8000 Klimastationen (etwa 9000 Diagramme). Gustav Fischer, Jena.

Walter, H., 1955. Die Klimagramme als Mittel zur Beurteilung der Klimaverhältnisse für ökologische, vegetationskundliche und landwirtschaftliche Zwecke. Ber. Dt. Bot. Ges. 68, 331–344.

Walther, G.-R., Berger, S., Sykes, M.T., 2005. An ecological footprint of climate change. Proc. R. Soc. B 272, 1427–1432.

Zimmermann, N.E., Yoccoz, N.G., Edwards, T.C., Meier, E.S., Thuiller, W., Guisan, A., Schmatz, D.R., Pearman, P.B., 2009. Climatic extremes improve predictions of spatial patterns of tree species. Proc. Natl. Acad. Sci. U. S. A. 106, 19723–19728.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 22–30

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Original article

A multi-scale approach reveals random phylogenetic patterns at the edge of vascular plant life[☆]

Yoann Le Bagousse-Pinguet[a,b,*,1], Pierre Liancourt[c,g,*,1], Lars Götzenberger[c], Francesco de Bello[a,c], Jan Altman[c], Viktorie Brozova[a], Zuzana Chlumska[a], Miroslav Dvorsky[c], Katerina Capkova[c], Martin Kopecky[d,e], Klara Rehakova[c], Pavel Riha[c], Jan Leps[a,f], Jiri Dolezal[a,c]

[a] *Department of Botany, University of South Bohemia, Na Zlate stoce 1, 370 05 Ceske Budejovice, Czech Republic*
[b] *Departamento de Biología y Geología, Física y Química Inorgánica, Universidad Rey Juan Carlos, C/Tulipán s/n, 28933 Móstoles, Spain*
[c] *Institute of Botany, Czech Academy of Sciences, Dukelska 135, 379 82 Trebon, Czech Republic*
[d] *Institute of Botany, The Czech Academy of Sciences, Zámek 1, CZ-252 43, Průhonice, Czech Republic*
[e] *Faculty of Forestry and Wood Sciences, Czech University of Life Sciences Prague, Kamýcká 129, CZ-165 00 Prague 6–Suchdol, Czech Republic*
[f] *Institute of Entomology, Biology Centre CAS, 370 05 Ceske Budejovice, Czech Republic*
[g] *Plant Ecology Group, University of Tübingen, Auf der morgenstelle 3, 71076 Tübingen, Germany*

ARTICLE INFO

Keywords:
Alpine communities
Elevation gradient
Extreme environments
Phylogenetic scale
Phylogenetic diversity
Scaling approach

ABSTRACT

Mountain plant diversity results from a myriad of factors, including evolutionary history, species pools, abiotic and biotic constraints. For instance, increasing stress (e.g., elevation) often selects communities with species originating from fewer, and more closely-related clades. We assessed phylogenetic diversity and turnover of plant communities by considering multiple drivers simultaneously: 1) the species pools; 2) the regional context, i.e., two regions of the arid Trans-Himalaya (the Karakoram Range and Little Tibet) with distinct history, climate and species richness (regional scale); 3) the abiotic constraint with communities spread out along an elevation gradient in each region (community scale); 4) the biotic constraint, i.e., species co-existing inside a competitive dominant species (cushion plant) vs. its adjacent open area in each community (neighbourhood scale); and 5) two phylogenetic scales (overall vs. recent events in the assembled phylogeny). We found random phylogenetic patterns for all spatial and phylogenetic scales, irrespective of the regional context, and the abiotic and biotic conditions under consideration. Contrary to expectations, we observed a tendency for phylogenetic evenness in Little Tibet and in the Karakoram Range with increasing elevation. The observed phylogenetic patterns were mostly explained by region, elevation and interactions among the multiple drivers under considerations. Phylogenetic scale, species pools and cushion plants explained patterns to a lower but significant extent. The studied phylogenetic patterns emerge from the complex interplay between multiple drivers, and challenge the common view that increasing spatial and phylogenetic scales, as well as increasing biotic and abiotic constraints, select communities with species originating from fewer, and more closely-related clades.

1. Introduction

Understanding how evolutionary and ecological processes interplay to generate patterns of species diversity and distribution remains a key challenge in ecology (Lavergne et al., 2010; Ricklefs, 1987). Phylogenetic information can shed light on the evolutionary history of a region, past species niche evolution and mechanisms of contemporary species coexistence (Cavender-Bares et al., 2009; Webb et al., 2002). However, inferring the multiple drivers of species coexistence from phylogenetic patterns requires cautious interpretation (Gerhold et al., 2015; Losos, 2008; Mayfield and Levine, 2010; Pausas and Verdu, 2010; Soliveres et al., 2012).

Phylogenetic diversity varies over a wide range of spatial scales (e.g., Cavender-Bares et al., 2009; Graham and Fine, 2008; Münkemüller et al., 2014). Past species diversification, migration events and past and current landscape dynamics primarily shape phylogenetic diversity at biogeographical scales (see Graham and Fine, 2008 for review). Abiotic factors and biotic interactions are more likely

---

[☆] This article is part of a special issue entitled Alpine and arctic plant communities : a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author.

E-mail addresses: y.b-pinguet@orange.fr (Y. Le Bagousse-Pinguet), pierre.liancourt@gmail.com (P. Liancourt).

[1] These authors contributed equally.

https://doi.org/10.1016/j.ppees.2017.10.002
Received 11 November 2016; Received in revised form 2 October 2017; Accepted 14 October 2017

to shape diversity at regional and community scales (Webb et al., 2002). The interplay of these processes can lead to patterns shifting from low (phylogenetic evenness) to high relatedness among co-occurring species (phylogenetic clustering) with increasing spatial scale (e.g., Cavender-Bares et al., 2006; Swenson et al., 2007).

The observed patterns of phylogenetic diversity also vary with the depth of the phylogenetic tree under consideration (phylogenetic scale hereafter, sensu Graham et al., 2016). These variations in patterns of phylogenetic diversity may emerge from different underlying mechanisms operating at different depths (Benton and Emerson, 2007). For instance, deep phylogenetic analyses have suggested that environmental filtering shape communities, while analyses focusing on more recent events often highlight additional mechanisms involved (e.g. biotic interactions). As a result, patterns of phylogenetic diversity are often observed to shift from phylogenetic evenness to clustering with increasing phylogenetic scale (Cavender-Bares et al., 2006; Swenson et al., 2006, 2007).

Multi-scale approaches (spatial and phylogenetic) allow to better understand the interplay between multiple drivers shaping phylogenetic diversity (Emerson and Gillespie, 2008; Graham and Fine 2008; Graham et al., 2016). These approaches are highly needed to develop conservation efforts prioritizing targeted species with respect to their evolutionary uniqueness or distinctiveness (Mouquet et al., 2012). Multi-scale approaches have been successfully used in tropical (e.g., Kembel and Hubbell, 2006) and temperate systems (e.g., Cavender-Bares et al., 2006), but received far less investigations in alpine environments (but see Münkemüller et al., 2014).

Mountain systems exhibit strong environmental changes at various scales, and host many endemic species (Graves, 1985; Körner, 2000). Many recent studies have focused on elevation gradients as a primary driver of phylogenetic diversity. However, elevation gradients encompass a range of underlying factors, such as evolutionary history, climate, or local environmental heterogeneity (see Graham et al., 2014 for a review). Increasing elevation has been associated with phylogenetic clustering for microbes (Bryant et al., 2008), insects (Machac et al., 2011; Smith et al., 2014), birds (Graham et al., 2009), and plants (Jin et al., 2015; but see Bryant et al., 2008). Phylogenetic clustering is often viewed as a signature of habitat filtering, where species from only few clades can withstand environmental conditions of higher elevations. In particular, this should be the case if fitness related traits under selection at high elevations are more similar for closely related species than for species from different clades (Webb et al., 2002).

Mountain phylogenetic diversity has been poorly investigated at neighbourhood scale, i.e., within microhabitats (e.g., Butterfield et al., 2013; Piston et al., 2016). Mountain communities are shaped by local environmental heterogeneity and by foundation species such as cushion plants (Al Hayek et al., 2014; Cavieres et al., 2014; Choler et al., 2001). Cushion plants can profoundly modify the nature and intensity of local abiotic filters, ultimately determining community assembly (Cavieres and Badano, 2009; Reid et al., 2010). Cushion plants can increase plant phylogenetic diversity (e.g., Piston et al., 2016; Soliveres et al., 2012) by creating favourable microhabitats under stressful conditions, in which less tolerant lineages are facilitated and diverge from those of microhabitats without cushion (Butterfield et al., 2013). Cushion plants with compact forms can also have strong competitive effects (Al Hayek et al., 2014), and act as a biotic filter influencing phylogenetic diversity (Piston et al., 2016). Whether and how competitive plant cushions modulate the effect of elevation on phylogenetic diversity remains poorly understood.

In our study, plant diversity was assessed in the arid Trans-Himalaya, one of the highest elevation limits for vascular plants worldwide (Dvorsky et al., 2015). We investigated phylogenetic diversity, and used a complementary approach that explicitly quantifies changes in phylogenetic patterns (phylogenetic turnover hereafter: Graham and Fine, 2008) across multiple spatial scales ranging from the neighbourhood (~1 m^2) up to supra-regional levels (Fig. 1). Our study focused on two geographically distinct regions, i.e., the Karakoram Range and Little Tibet, which differ in terms of history, climate and species richness (Dvorsky et al., 2015). We investigated phylogenetic diversity for each region, as well as between-region and between-site phylogenetic turnovers along elevation gradients within each study region. Observed patterns were all compared with the supra-regional and their respective regional reference pools (Karakoram Range and Little Tibet), to shed light on coarse-scale processes such as biogeographical history (Swenson et al., 2006). In each site, we used the dominant species *Thylacospermum caespitosum* (Caryophyllaceae) — a strong competitive cushion plant that excludes subsets of the local species richness (de Bello et al., 2011; Dvorsky et al., 2013) — and adjacent open areas (i.e., microsites hereafter) to investigate phylogenetic diversity within each microsite, as well as between-microsite turnovers. Phylogenetic patterns were compared to the local reference pool (i.e., the local species pool of a study site) to shed light on processes such as species interactions and fine scale environmental filtering (Swenson et al., 2006). For all spatial scales considered, phylogenetic patterns were finally assessed at two phylogenetic scales (Graham et al., 2016), by comparing phylogenetic diversity observed for the most recent speciation events (tip level) to phylogenetic diversity across the whole tree (e.g., Jin et al., 2015).

We aimed at considering a large panel of potential drivers of phylogenetic patterns and their interactions to test the common assumption that increasing spatial and phylogenetic scales, or increasing abiotic and biotic constraints select communities with species originating from fewer, and more closely-related clades (so called phylogenetic clustering). Specifically, we hypothesized an overall increase in phylogenetic clustering i) from neighbourhood to supra-regional scales, ii) at higher elevation within each region (e.g., Jin et al., 2015; Machac et al., 2011); and iii) at micro-scale due to the competitive effect of the cushion plant considered (Butterfield et al., 2013).

2. Materials and methods

2.1. Study regions

The study supra-region is located in Ladakh, in the Jammu & Kashmir State, NW India. The area is part of the Trans-Himalaya region, delimited by the Eastern Karakoram Range in the north and by the Great Himalaya Range in the south. Ladakh is an arid mountain region that receives little precipitation due to its position in the rain-shadow of the Himalaya Range (Dvorsky et al., 2013; Wang, 1988). Our study focused on two regions of Ladakh that uplifted around 55 Ma ago, i.e., the Karakoram Range and Little Tibet. Their recent history differs substantially since the Karakoram Range was deglaciated more recently than Little Tibet. Climates of the two regions also differ, with Little Tibet experiencing harsher environmental conditions (Dvorsky et al., 2015). Finally, species diversity is higher in the Karakoram Range, while Little Tibet exhibits a high rate of plant endemism (Dvorsky et al., 2015).

2.2. Elevation gradients

Plant phylogenetic diversity and between-site turnovers were assessed using eight sites, i.e., four sites in the Karakoram Range (34°45′N, 77°35′E) and four sites in Little Tibet (32°59′N, 78°24′E). The eight sites were distributed along two elevation gradients ranging from 4850 m to 5250 and from 5350 to 5850 m a.s.l. in the Karakoram Range and in Little Tibet, respectively. In each region, the four sites covered the entire elevation range occupied by *T. caespitosum*. It is also important to note that the two elevation gradients were ecologically comparable. Both gradients covered the complete sequence of mountain vegetation types: steppes at the lowest site of each gradient, alpine communities at the two intermediate sites and subnival communities at the highest site of each gradient (Dvorsky et al., 2013).

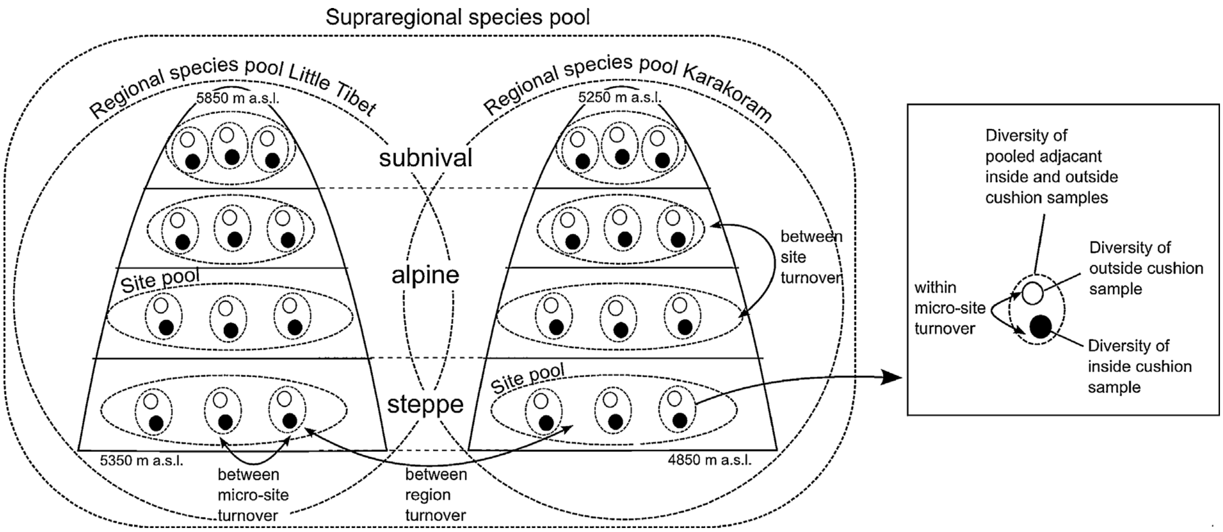


Fig. 1. Graphical representation of the analytical framework used in this study. We represent the different spatial scales, as well as the reference pools used. Note that we applied this framework across the whole-tree and for most recent speciation events (tip level).

2.3. *Impact of the cushion plant*

The phylogenetic diversity within microsites and between-microsite turnovers were assessed using the presence of the cushion plant *T. caespitosum* and the adjacent open areas. *T. caespitosum* is one of the most dominant high-alpine cushion plants in the Himalayas, and one of the few dominant species in the studied area (Klimešová et al., 2011). It is a perennial plant with a woody taproot, forming very dense and solid cushions (Klimešová et al., 2011). The largest cushions can be more than 150 cm in diameter (Dvorsky et al., 2013), suggesting that they may live for decades or even centuries (Le Roux and McGeoch, 2004). *T. caespitosum* occurs along an elevation range of 4600–5900 m, although this range can vary with the geographical locations (Klimešová et al., 2011). *T. caespitosum* is a strong competitor that influences the structure and diversity of plant communities by locally excluding a subset of the site pools (de Bello et al., 2011; Dvorsky et al., 2013).

2.4. *Vegetation survey*

The vegetation survey was carried out during the peak of the growing season (August) in 2011. The eight sites were selected at 4850 m, 5000 m, 5100 m and 5250 m a.s.l. in the Karakoram Range, and at 5350 m, 5600 m, 5750 m and 5850 m a.s.l. in Little Tibet. In each site, cushions were systematically surveyed in an area of ca. 1 ha (n = 66, 61, 69, 77, 70, 70, 70, 73 cushions in each site). The studied cushions represented all size classes with a diameter up to 132 cm. The most common size class was 40–60 cm in the Karakoram Range (n = 79) and 20–40 cm in Little Tibet (n = 102). Two microsites were considered for each cushion: a first microsite covering the cushion itself; a second microsite with the same size and shape randomly placed in the open adjacent area at a distance equal to the cushion diameter. In total, 1112 microsites were surveyed (556 pairs). All vascular plant species rooting in the microsites were recorded and their percentage cover was estimated visually. Because phylogenetic diversity can only be calculated for communities with more than one species, all microsites with fewer than two species were excluded from the data before further analyses, leaving 420 microsites (210 pairs).

2.5. *Species phylogeny*

All sequences used for building the tree were extracted from genBank (www.ncbi.nlm.nih.gov/nuccore/). A combined multigene attitude was applied. Maximum data density was achieved with four loci: internal transcribed spacer (ITS), trnT-trnL intergenic spacer, matK + trnK region, and the gene for rubisco large subunit (rbcL). Missing sequences were filled with Ns (unknown data) or patched by corresponding nucleotides from closely related taxa (*Artemisia minor* was fixed with *A. glacialis*, *Desideria pumila* with *D. linearis*, *Elymus schrenkianus* with *E. dahuricus*, *Potentilla saundersiana* with *P. nivea* and *Sibbaldia tetrandra* with *S. cuneata*).

The L-INS-i algorithm implemented in the online version of MAFFT 6 (http://mafft.cbrc.jp/alignment/server, Katoh and Toh, 2008) was employed to align the sequence datasets. Partial alignments were concatenated, manually adjusted in BioEdit (Hall, 1999) and cleaned with the *automated1* algorithm in trimAll software (Capella-Gutierrez et al., 2009) to exclude highly divergent and gap-rich regions. Prior to the phylogenetic analysis, the best-fit model was selected by Kakusan4 (Tanabe, 2011). Baseml software (Adachi and Hasegawa, 1996) served as the computational core. Both non-partitioned and partitioned models were evaluated. Based on the Bayesian information criterion (Schwarz, 1978), we decided to use the GTR model with rate variation across locations simulated by discrete gamma distribution ($\Gamma 8$), autocorrelated by the AdGamma rates prior and unlinked for particular gene partitions. To reflect the increased probability of transitions over transversions in non-coding loci, the substitution rates prior (revMatPr) was set for the ITS, trnT-L and petB-D partitions to the Dirichlet function with values of 1 and 3.

The phylogenetic analysis was represented by the Bayesian inference (BI), conducted in MrBayes 3.1.2 (Ronquist and Huelsenbeck, 2003). The phylogenetic analysis was provided using two independent runs with four Metropolis-coupled MCMC chains of 1×10^7 generations sampled after every 1000th generation. In each run, one Markov chain was cold and three were incrementally heated by a parameter of 0.3. The first 25% of entries were discarded as burn-in and the rest was used to compute the majority-rule consensus to eliminate trees sampled before reaching apparent stationarity. The resulting tree was edited to be compatible in future ecological studies, since it contained more terminals (unknown multispecies genera were substituted with five terminals). The internal phylogeny of these genera was collapsed to polytomy, branch lengths were averaged and excessive branches were cut out. The nomenclature was unified with the Ladakh plant list (Klimes and Dickore, 2006).

2.6. *Phylogenetic diversity and turnover*

Phylogenetic patterns were investigated by the Mean Pairwise

Distance (MPD) and the Mean Nearest Taxon Distance (MNTD) indices. These two indices allowed assessing the whole-tree species relatedness in each community, and tip-level species relatedness between closely-related species in each community, respectively (Fine and Kembel, 2011; Jin et al., 2015). Comparing MPD and MNTD can therefore inform at which phylogenetic scale patterns of phylogenetic diversity are most evident (Jin et al., 2015). The Mean Nearest Taxon Distance index indicates patterns occurring for the most recent speciation events. The Mean Pairwise Distance indicates patterns occurring across the entire phylogeny (Cadotte and Davies, 2016). While MPD and MNTD allow to investigate the diversity in the studied samples, phylogenetic turnovers assess how much species in a sample are related to all other species in a second sample, either on average, or only for the nearest taxa. Hence, whereas diversities are calculated per plot, phylogenetic turnovers are calculated on a pairwise plot basis.

We investigated phylogenetic diversity for both MPD and MNTD using the functions *mpd* and *mntd* in the *picante* package (Kembel et al., 2010). We computed the phylogenetic turnovers with the functions *comdist* and *comdistnt* that are the analogues of *mpd* and *mntd*. Abundance data were log-transformed after adding 1 to avoid negative values. We used standardized effect sizes (SES) of MPD and MNTD indices. Standardized effect sizes were calculated as $SES_{MPD} = (MPD_{obs} - mean(MPD_{rand}))/sd(MPD_{rand})$, where MPD_{obs} is the observed phylogenetic diversity index, $mean(MPD_{rand})$ is the mean MPD of random communities generated from a null model, and $sd(MPD_{rand})$ is the standard deviation of MPD of those random communities.

For phylogenetic diversity, positive values of SES_{PD} indicate greater distances among species than expected by chance (phylogenetic evenness). Negative values of SES_{PD} indicate lower phylogenetic distances among species than expected by chance (phylogenetic clustering). SES_{PD} values ranging between −1.96 and 1.96 indicate a pattern that is not significantly different from random, under the assumption that indices are normally distributed. For phylogenetic turnover, negative values of SES_{PD} indicate that the species in two plots show less phylogenetic turnover (i.e., are more related to each other) than expected by chance, while positive values indicate higher showing phylogenetic turnover (i.e. species less related to each other) than expected by chance.

To compute SES scores for each community plot, random communities were generated by randomly drawing species from a species pool with the same species richness as in the observed plots, meaning that abundances where shuffled within plots across species, as executed by the “richness” null model of the *randomizeMatrix* function in the *picante* package (Kembel et al., 2010). The used species pools differed in size according to each tested hypothesis (see Fig. 1 and next paragraph): (1) all species sampled in both regions (supra-regional reference pool), (2) all species occurring in a single region (regional reference pool), (3) all species present in a given study site (local reference pool).

The combinations of different diversity indices and reference pools helps testing the strength of different species filters at different scales and according to ecological factors (elevation, cushion plants). SES of alpha diversities and spatial turnovers were achieved by using the supra-regional and regional species pools to test for increased filtering effects with spatial scale, stronger filtering in Little Tibet, and increased filtering with elevation (hypotheses i − ii). Comparing microsite alpha diversities and between-microsites turnovers against randomisations from the local species pool allowed testing the occurrence of biotic filters due to the presence of cushion plants (hypothesis iii).

2.7. Statistical analyses

First, we analysed the effects of region, elevation, the presence of cushions, and their interactions on diversities and turnovers with mixed effect ANOVA models. All predictors were categorical. Pairs of sampled microsites (cushion and adjacent open area) were used as a random factor, and nested within site. We also performed a linear regression model with the between-site turnovers within single region used as the response variable, and with the elevation difference between sites, the region, and their interaction as predictors. We used post-hoc Tukey tests to evaluate if between-site turnover differed between the two regions.

Second, we used a mixed effect ANOVA model to test whether alpha phylogenetic diversity of different sites differed between regions, species pools (supra-regional vs. regional reference pools), elevation, phylogenetic scale (MPD vs. MNTD) and their interactions (Appendix S1 in the Supplementary material). We repeated this analysis at the microsite scale with region, elevation, cushions, phylogenetic scale (MPD vs. MNTD) and their interactions as predictors. At each scale (microsite and site), we evaluated the relative effect of the predictors on phylogenetic diversity by calculating their proportional contribution to the overall explained variance of the model from their sums of square. The obtained relative effects of predictors are grouped into the following identifiable variance fractions: i) region, ii) elevation, iii) phylogenetic scale and iv) interactions among drivers. Note that the following variance fractions v) pool and vi) cushion were also considered at the site and the microsite scales, respectively.

All statistical analyses were performed using R 2.15.1 (R Core Team, 2012).

3. Results

The observed *SES* values of phylogenetic diversity mostly ranged between −1.96 and 1.96, a signature of random patterns (Figs. 2 and 4). Yet, our subsequent analyses revealed significant effects of our drivers and their interactions on phylogenetic diversity. Region and elevation were the most important drivers explaining 45% of the variation in phylogenetic diversity at the site scale and 63% at the microsite scale (Appendix S1 in the Supplementary material). Phylogenetic scale and species pool had lower, but significant effects at the site level (7% and 6%, respectively). Cushion explained 3% of variation at the microsite scale. Finally, interactions among drivers explained 28% of variation in phylogenetic patterns at the site scale and 11% at the microsite scale.

Our results were consistent when comparing the observed phylogenetic patterns to both supra-regional (Fig. 2: upper panels) and regional reference species pools (lower panels). Nonetheless, SES_{MPD} values were generally higher in Little Tibet when considering the regional reference pool rather than the supra-regional pool (Fig. 2a and c).

We found significantly higher − and more often positive − *SES* values in Little Tibet than in the Karakoram Range (Table 1) for both whole-tree (SES_{MPD}: Fig. 2c) and tip level diversity metrics (SES_{MNTD}: Fig. 2b and d). SES values computed at the tip level also increased with elevation in the Karakoram Range (Table 1) (SES_{MNTD}: Fig. 2b and d). These results suggest a tendency for phylogenetic evenness in Little Tibet, and in the Karakoram Range at high elevation.

We also found negative values of $\mathrm{SES_{MPD}}$ for between-region and between-site turnovers, although they all ranged between −1.96 and 1.96 (Fig. 3a). Our results highlight a tendency of phylogenetic similarity between regions for whole-tree diversity indices (SES_{MPD}), i.e., a lower phylogenetic turnover across sites than expected from the supra-regional reference pool. $\mathrm{SES_{MPD}}$ were also significantly lower within regions than between regions, indicating an even lower phylogenetic turnover across sites within regions than between regions (Fig. 3a). Positive SES_{MNTD} were observed between regions, although they were lower than 1.96 (Fig. 3b). This indicated a tendency for phylogenetic turnover at the tip level between regions.

Elevation tended to increase the degree of phylogenetic evenness when considering either the site (Fig. 2, Table 1) or the microsite scales (Fig. 4a and b, Table 2). We also observed an effect of the elevation distance on the between-site turnover along the elevation gradients for both whole-tree (SES_{MPD}) and tip level analyses (SES_{MNTD}, Appendix S2 in the Supplementary material). Closer sites exhibited a phylogenetic turnover of closely related clades (negative *SES* values) while no trend

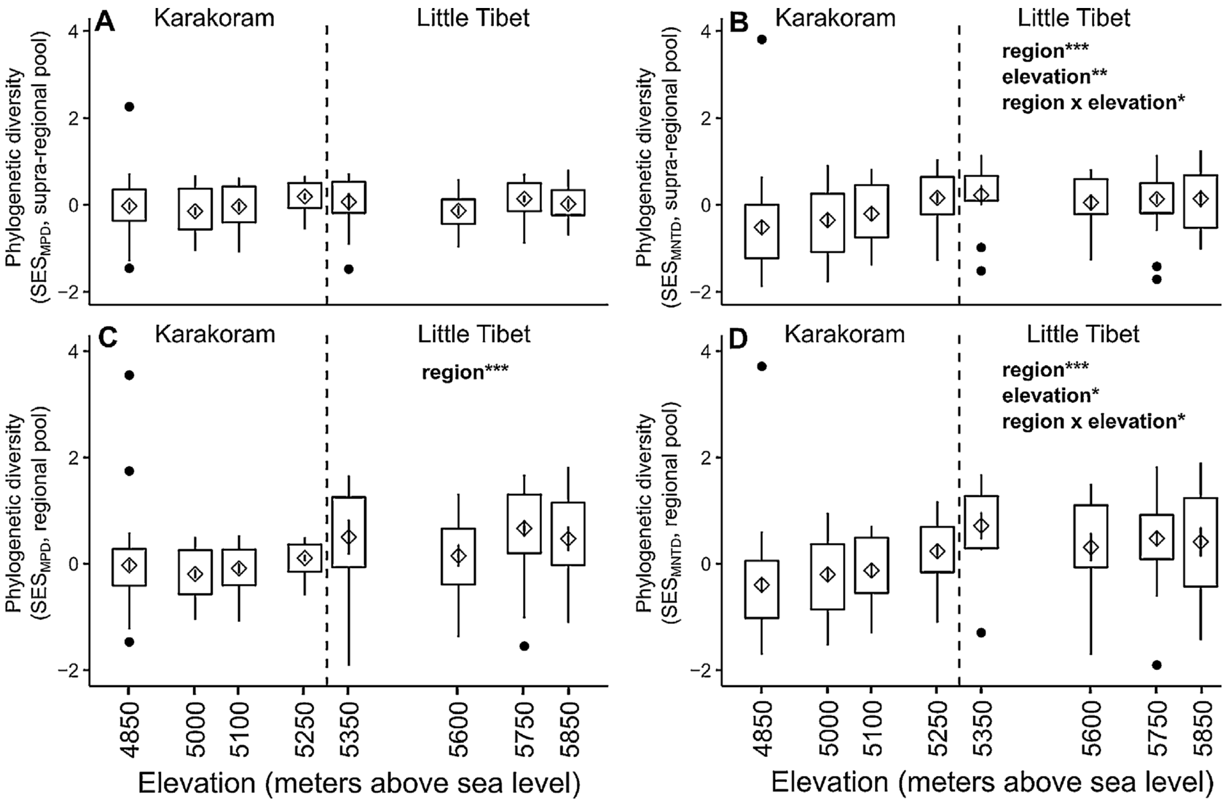


Fig. 2. Boxplot of standardized effect sizes (SES), their mean (diamond) and error around the mean (bar through the diamond) of phylogenetic diversity at the community level (pairs of microsites inside and outside cushions) along the elevation gradient. A and B show SES of mean pairwise phylogenetic distance (MPD) and mean phylogenetic distance to the nearest neighbor (MNTD), respectively, compared to communities randomly drawn from the supra-regional reference pool with the same species richness. C and D show SES of mean pairwise phylogenetic distance (MPD) and mean phylogenetic distance to the nearest neighbor (MNTD), respectively, compared to communities randomly drawn from the regional reference pools with the same species richness. Bold terms describe the significant terms in a linear model with region, elevation, and their interaction as explanatory variables (see Table 1). NS: not significant; * significant with P < 0.05; ** significant with P < 0.01; *** significant with P < 0.001.

Table 1
Results of the mixed effect ANOVA models with phylogenetic diversity expressed as standardized effect size (SES) of MPD and MNTD as response variables. As reference pool for randomizations served either the supra-regional or the regional species pool. Region, elevation, and their interaction were used as fixed factors. Pairs of microhabitats (cushion and adjacent open area) were used as random factor and nested within communities. Significant factors are in bold.

	SES_{MPD}			SES_{MNTD}		
Factor	df	F value	P	Df	F value	P
	Supra-regional reference pool					
Region	1	0.11	0.73	1	13.93	**<0.001**
Elevation	1	0.70	0.40	1	8.89	**<0.01**
Region x Elevation	1	0.21	0.65	1	5.86	**<0.05**
	Regional reference pool					
Region	1	30.71	**<0.001**	1	21.72	**<0.001**
Elevation	1	1.03	0.31	1	6.22	**<0.05**
Region x Elevation	1	0.01	0.94	1	4.42	**<0.05**

occurred for spatially distant communities.

Finally, the presence of the cushion plant slightly increased the degree of phylogenetic evenness (Table 2) for whole-tree diversity metrics (Fig. 4a) and at low elevation for tip level diversity metrics (marginally significant elevation x cushion in Table 2, Fig. 4b, significant elevation x cushion in Appendix S1 in the Supplementary material). However, cushion plant did not seem to drive a significant turnover of plant phylogenetic diversity between sampled microsites (Fig. 4c and d).

4. Discussion

Our multi-scale approach revealed random phylogenetic patterns, irrespectively of the species pool, the regional context (history, climate and species richness), the abiotic and the biotic conditions. Detecting consistent random patterns does not support the common view that phylogenetic patterns depend on spatial scale (e.g., Münkemüller et al., 2014; Swenson et al., 2006), shifting from phylogenetic evenness (Swenson et al., 2007) to clustering with increasing spatial grain and extent (Cavender-Bares et al., 2006; Swenson et al., 2006, 2007). A consistent pattern across multiple spatial scales has also been observed in bee communities (i.e., a consistent phylogenetic clustering in Harmon-Threatt and Ackerly, 2013), and therefore spotlights a possible idiosyncrasy in scaling phylogenetic patterns (Cadotte and Davies, 2016). This discrepancy between our results and other similar studies does not seem to arise from the geographic scale under consideration. Indeed, the range of spatial scales considered in our study (from $\sim 1\ m^2$ to $\sim 20000\ km^2$) fully encompassed the scales of previous studies (e.g., Kembel and Hubbell, 2006 and Swenson et al., 2007: from $< 100 m^2$ to 1 ha).

The observed random patterns of plant community phylogenetic diversity indicate that particular sets of morphological/anatomical adaptations allowing species to cross the environmental filters and to persist into extreme environments can be found within and across different lineages (Butterfield et al., 2013; Dolezal et al., 2016; Webb et al., 2002). For instance, species growing at the highest elevation of Little Tibet recruit mostly from Brassicaceae, Asteraceae, Caryophyllaceae and Poaceae (Appendix S3 in the Supplementary material). Despite their general high phylogenetic dispersion, these species are generally small, exhibit a similar growth form (cushion) with a

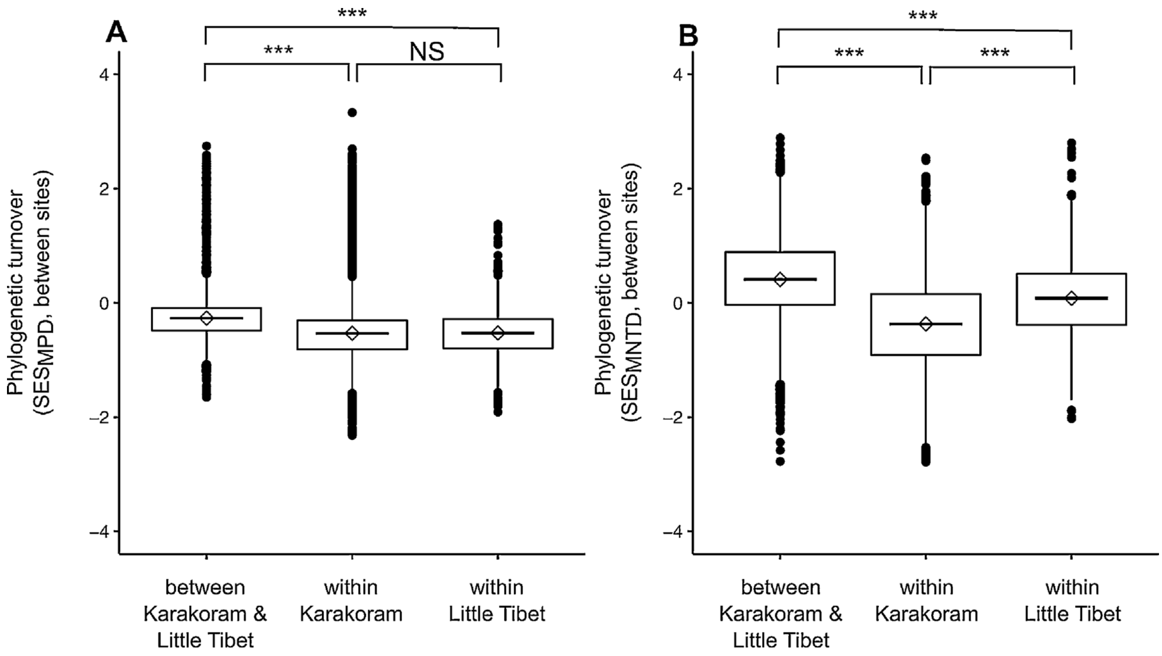


Fig. 3. Phylogenetic turnover based on MPD (A) and MNTD (B), respectively, for all pairwise comparisons between communities (i.e., between pairs of microsites across communities) from the two different regions, and between communities within each of the regions. Boxplots depict the distribution of SES values; the diamond and bar their mean and error around the mean, respectively. Significance above the lines are results of a post-hoc Tukey HSD test to compare the means of the three groups. NS: not significant; * significant with $P < 0.05$; ** significant with $P < 0.01$; *** significant with $P < 0.001$.

relatively high water content and water-use efficiency, and contain more nutrients and soluble carbohydrates than species from lower elevation (Dvorsky et al., 2015, 2016). Further studies simultaneously combining phylogenetic and functional (multi-trait) approaches would certainly allow better understanding the processes governing community assembly in these extreme environments (Swenson, 2013), and addressing the issue of plant adaptation in relation to evolutionary history (Schweingruber et al., 2014).

Elevation was the major driver of community assembly at microsite-scale, explaining 63% of the variation in phylogenetic diversity. The tendency for phylogenetic evenness with increasing elevation in the Karakoram Range is opposite to previous findings generally showing an increase in phylogenetic clustering (Bryant et al., 2008; Graham et al., 2009; Jin et al., 2015; Machac et al., 2011; Smith et al., 2014). Our study system illustrates that communities from extreme environments are not necessarily composed of species originating from fewer, and more closely-related clades (see also Le Bagousse-Pinguet et al., 2017 for similar findings on functional diversity in global drylands).

The regional context was the main driver of our studied communities at the site scale, explaining 45% of the variation in phylogenetic patterns. This suggests that floristic regional context is a major drivers of phylogenetic patterns of plant communities. The difference between the two study regions may stem from the difference in their deglaciation history (Dvorsky et al., 2015). The tendency for phylogenetic evenness in Little Tibet may arise from a longer time for distantly related species to converge in their habitat-use along the elevation gradient. In contrast, the shorter time from deglaciation in the Karakoram Range may have led the species to converge in their habitat-use at the highest elevation only, and more time might be required for this pattern to emerge at low elevation.

Several other underlying processes may explain the observed phylogenetic evenness at high elevation in the Karakoram Range. Elevation gradients are often complex, comprising multiple underlying gradients and notably acting on dispersal limitation (Körner, 2000; Michalet et al., 2014). Phylogenetic turnover of more closely related species between communities of closer elevation (Appendix S2 in the Supplementary material), associated with the absence of phylogenetic clustering at any scale, supports the idea that dispersal limitation could be one of the underlying drivers shaping phylogenetic patterns along the studied elevation gradients (Dvorsky et al., 2016).

The local biotic context, represented by the presence or absence of cushion plants at the neighbourhood scale was also a significant driver of phylogenetic patterns in the studied sites, although it was weaker than the other factors considered (3% of the variations in phylogenetic diversity). The presence of the cushion tended to be associated with phylogenetic evenness when considering the whole tree, although no plant phylogenetic turnover was observed between microsites. These patterns indicate that subsets of species from open areas (similar to the site reference pool) were filtered by the cushion plants, with distantly-related species converging in their use of micro-habitats (Webb et al., 2002). Interestingly, we also found that the net effect of the cushions on phylogenetic diversity changed along the elevation gradient. The competitive effect of the cushion *sensu stricto* remains similar along the entire gradient (Dvorsky et al., 2013). The more pronounced difference between cushions and adjacent open areas observed at low elevation may therefore result from the higher occurrence of competitive species exhibiting the set of morphological/anatomical adaptations to invade the cushions (see also Liancourt et al., 2017 for the role of the species pool on the outcome of biotic interactions). Altogether, our results therefore support the view that considering neighbourhood scale processes is relevant when assessing phylogenetic patterns of plant communities (Valiente-Banuet and Verdu, 2007; Verdu et al., 2009), and that investigating the role of plant–plant interactions may bring interesting insights into the mechanisms driving vegetation structure in extreme habitats (Butterfield et al., 2013; Piston et al., 2016; Soliveres et al., 2012).

The phylogenetic scale explained 7% of the variations in phylogenetic patterns, demonstrating the importance of accounting for different phylogenetic scales to better understand the evolutionary history of regional floras (Fine and Kembel, 2011; Graham et al., 2016; Swenson et al., 2007). The observed changes in patterns were clearer when considering recent speciation events rather than the whole tree, reinforcing the view that the phylogenetic scale under consideration can largely influence our ability to detect non-random patterns in plant

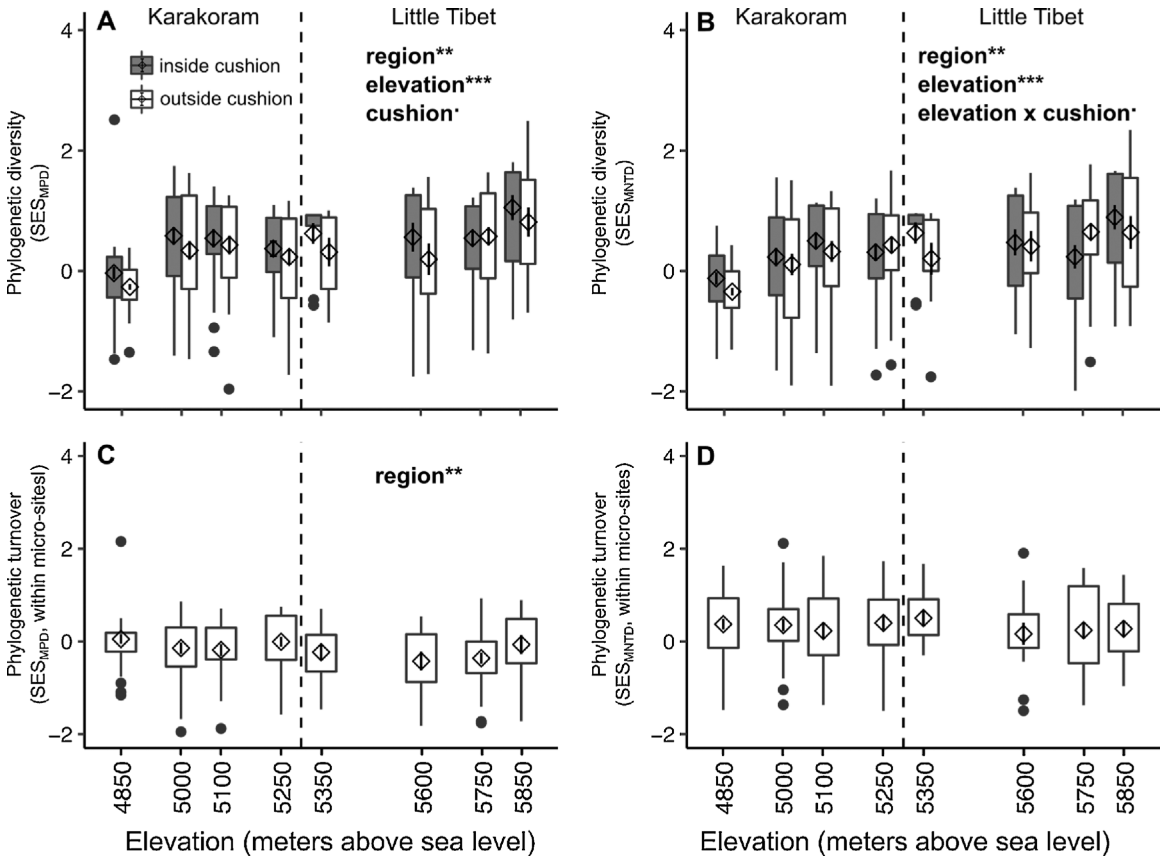


Fig. 4. Phylogenetic diversity and turnover with the local reference pool for randomization (i.e., all species found in the investigated site at a given elevation range). Boxplots depict the distribution of SES values; the diamond and bar their mean and error around the mean, respectively. A and B show the diversity at the community level separately for microsites inside and outside of the cushion plant *Thylacospermum caespitosum* across the elevation gradient as mean pairwise phylogenetic distance (MPD, subfigure A) or mean phylogenetic distance to the nearest neighbor (MNTD, subfigure B), compared to communities randomly drawn from the local reference pool with the same species richness. Bold terms describe the significant terms in a linear mixed effect model with region, elevation, position inside or outside the cushion, and all possible interactions as fixed factor, and pairs of microsites (cushion and adjacent open area) nested within a site as random factor. C and D show phylogenetic turnovers based on MPD and MNTD, respectively, for pairs of adjacent inside and outside cushion microsites along the elevation gradient, compared to communities randomized by shuffling species abundances within a site (see methods for details). Bold terms describe the significant terms in linear models with region, elevation, position outside or inside the cushion and their interactions (for phylogenetic diversity), and with region, elevation, and their interaction (for turnover), as explanatory variables (see Table 2). NS: not significant; * significant with $P < 0.05$; ** significant with $P < 0.01$; *** significant with $P < 0.001$.

communities (e.g., Jin et al., 2015; Swenson et al., 2007). Phylogenetic evenness observed at the tip levels may result from the effect of local abiotic factors and biotic interactions (Cavender-Bares et al., 2006), or alternatively from allopatric speciation (Pigot and Etienne, 2015). This pattern is also likely to become more common when communities are more finely defined taxonomically (Cavender-Bares et al., 2006; Swenson et al., 2006, 2007). We therefore acknowledge that the stronger signals of phylogenetic evenness observed at the tip levels may simply result from a decrease in phylogenetic signal with decreasing the phylogenetic scale of the analysis.

Patterns of whole-tree phylogenetic turnover were low between and within regions, and relatively high at the tip level between regions (Fig. 3). Therefore, our results indicate that patterns of diversity are phylogenetically representative of the supra-regional reference pool and similar between the Karakoram Range and Little Tibet. In both regions, most of the families occurring in the supra-regional reference pool are represented, e.g., Poaceae, Cyperaceae, Asteraceae, Fabaceae, Brassicaceae and Rosaceae. Rather, phylogenetic turnover mostly occurs at the tip level, indicating a recent diversification within families and genera, such as greater diversifications in *Oxytropis, Potentilla* or *Carex* in the Karakorum Range or Caryophyllaceae in Little Tibet, and therefore reflecting the unique phytogeographical history of each region.

Our multi-scale approach, from neighbourhood to supra-regional scales and across phylogenetic scales, revealed random phylogenetic patterns at the edge of vascular plant life. This finding challenges the common view that increasing spatial and phylogenetic scales, or increasing abiotic and biotic constraints select communities with species originating from fewer, and more closely-related clades. Using multiple drivers simultaneously, we also quantified and ranked the importance of commonly investigated drivers of phylogenetic patterns. We showed that phylogenetic patterns could clearly emerge from the complex effects of pools, regional context (history), phylogenetic scale, abiotic and biotic constraints and their interactions (reaching up to 28% of the explained variance). On one hand, our approach illustrates the difficulty to directly infer ecological processes underlying species coexistence and community assembly from phylogenetic patterns (Gerhold et al., 2015; Losos, 2008; Mayfield and Levine, 2010; Pausas and Verdú, 2010; Soliveres et al., 2012). On the other hand, our study clearly illustrates that phylogenetic patterns contain a great deal of information and therefore supports the recent claim that these patterns *"are not only proxies of community assembly processes, they are far better"* (Gerhold et al., 2015).

Table 2

Results of the mixed effect ANOVA models with phylogenetic diversity and turnover expressed as standardized effect size (SES) of MPD and MNTD as response variables. For diversity, region, elevation, location outside or inside the cushion, and interactions among these factors were used as fixed factors. Pairs of microsites (cushion and adjacent open area) were used as random factor and nested within site. For phylogenetic turnover between inside and outside cushions, region, elevation and their interaction were used as fixed factors. Significant factors are in bold.

	SES_{MPD}			SES_{MNTD}		
Factor	df	F value	P	df	F value	P
	Phylogenetic diversity inside and outside cushions, site as the reference pool					
Region	1	9.45	**<0.01**	1	10.20	**<0.01**
Elevation	1	26.32	**<0.001**	1	28.08	**<0.001**
Cushion	1	3.43	0.06	1	0.20	0.65
Region x Elevation	1	0.20	0.65	1	0.56	0.45
Region x Cushion	1	0.11	0.74	1	0.97	0.33
Cushion x Elevation	1	2.37	0.12	1	3.54	0.06
Region x Cushion x Elevation	1	1.19	0.27	1	0.53	0.46
	Phylogenetic turnover between inside and outside cushions, site as the reference pool					
Region	1	9.43	**<0.01**	1	2.30	0.13
Elevation	1	0.001	0.97	1	0.03	0.85
Region x Elevation	1	1.00	0.32	1	0.17	0.68

Acknowledgements

We thank the guest editors for the invitation to participate in this special issue. Y.L.B.P is supported by the project Postdoc USB (reg.no. CZ.1.07/2.3.00/30.0006) realized through EU Education for Competitiveness Operational Programme. Project is funded by European Social Fund and Czech State Budget. Y.L.B.P is also supported by a Marie Sklodowska-Curie Actions Individual Fellowship (MSCA-IF) within the European Program Horizon 2020 (DRYFUN Project 656035). P.L. and L.G. received support from the European Union's Seventh Framework Programme for research, technological development and demonstration under grant agreement no GA-2010-267243–PLANT FELLOWS. J.L. was supported by PLADIAS centre (GACR 14-36079G). The fieldwork was supported by Czech Science Foundation (GACR 13-13368S and 17-19376S). This study was also part of long-term research development project No. RVO 67985939. Finally, we are grateful to Prof. A. Prinzing for fruitful discussions and comments on earlier versions.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.ppees.2017.10.002.

References

Adachi, J., Hasegawa, M., 1996. MOLPHY Version 2.3: Programs for molecular phylogenetics based on maximum likelihood. Comput. Sci. Monogr. 28, 1–150.

Al Hayek, P., Touzard, B., Le Bagousse-Pinguet, Y., Michalet, R., 2014. Phenotypic differentiation within a foundation grass species correlates with species richness in a subalpine community. Oecologia 176, 533–544.

Benton, M.J., Emerson, B.C., 2007. How did life become so diverse?: The dynamics of diversification according to the fossil record and molecular phylogenetics. Palaeontology 50, 23–40.

Bryant, J.A., Lamanna, C., Morlon, H., Kerkhoff, A.J., Enquist, B.J., Green, J.L., 2008. Microbes on mountain sides: contrasting elevational patterns of bacterial and plant diversity. Proc. Natl. Acad. Sci. U. S. A. 105, 11505–11511.

Butterfield, B.J., Cavieres, L.A., Callaway, R.M., Cook, B.J., Kikvidze, Z., Lortie, C.J., et al., 2013. Alpine cushion plants inhibit the loss of phylogenetic diversity in severe environments. Ecol. Lett. 16, 478–486.

Cadotte, M.W., Davies, J., 2016. Phylogeny in Ecology: a Guide to Concepts and Methods. Princeton University Press.

Capella-Gutierrez, S., Silla-Martinez, J.M., Gabaldon, T., 2009. trimAl: a tool for automated alignment trimming in large-scale phylogenetic analyses. Bioinformatics 25, 1972–1973.

Cavender-Bares, J., Keen, A., Miles, B., 2006. Phylogenetic structure of Floridian plant communities depends on taxonomic and spatial scale. Ecology 87, 109–122.

Cavender-Bares, J., Kozak, K.H., Fine, P.V.A., Kembel, S.W., 2009. The merging of community ecology and phylogenetic biology. Ecol. Lett. 12, 693–715.

Cavieres, L.A., Badano, E.I., 2009. Do facilitative interactions increase species richness at the entire community level? J. Ecol. 97, 1181–1191.

Cavieres, L.A., Brooker, R.W., Butterfield, B.J., 2014. Facilitative plant interactions and climate simultaneously drive alpine plant communities. Ecol. Lett. 17, 193–202.

Choler, P., Michalet, R., Callaway, R.M., 2001. Facilitation and competition on gradients in alpine plant communities. Ecology 82, 3295–3308.

de Bello, F., Dolezal, J., Dvorsky, M., Chlumska, Z., Rehakova, K., Klimesova, J., 2011. Cushions of *Thylacospermum caespitosum* (Caryophylaceae) do not facilitate other plants under extreme altitude and dry conditions in the north-west Himalayas. An. Bot. 108, 567–573.

Dolezal, J., Dvorsky, M., Kopecky, M., Liancourt, P., Hiiesalu, I., Macek, M., et al., 2016. Vegetation dynamics at the upper elevational limit of vascular plants in Himalaya. Sci. Rep. 6, 24881.

Dvorsky, M., Dolezal, J., Kopecky, M., Chlumska, Z., Janatkova, K., Altman, J., et al., 2013. Testing the stress-Gradient hypothesis at the roof of the world: effects of the cushion plant *Thylacospermum caespitosum* on species assemblages. PLoS One 8, e53514.

Dvorsky, M., Altman, J., Kopecky, M., Chlumska, Z., Rehakova, K., Janatkova, K., et al., 2015. Vascular plants at extreme elevations of E Ladakh, NW Himalayas. Plant Ecol. Div. 8, 571–584.

Dvorsky, M., Chlumská, Z., Altman, J., Capková, K., Řeháková, K., Macek, M., et al., 2016. Gardening in the zone of death: an experimental assessment of the absolute elevation limit of vascular plants. Sci. Rep. 6, 24440.

Emerson, B.C., Gillespie, R.G., 2008. Phylogenetic analysis of community assembly and structure over space and time. Trends Ecol. Evol. 23, 619–630.

Fine, P.V.A., Kembel, S.W., 2011. Phylogenetic community structure and phylogenetic turnover across space and edaphic gradients in western Amazonian tree communities. Ecography 34, 552–565.

Gerhold, P., Cahill, J.F., Winter, M., Bartish, I.V., Prinzing, A., 2015. Phylogenetic patterns are not proxies of community assembly mechanisms (they are far better). Funct. Ecol. 29, 600–614.

Graham, C.H., Fine, P.V.A., 2008. Phylogenetic beta diversity: linking ecological and evolutionary processes across space in time. Ecol. Lett. 11, 1265–1277.

Graham, C.H., Parra, J.L., Rahbek, C., McGuire, J.A., 2009. Phylogenetic structure in tropical hummingbird communities. Proc. Natl. Acad. Sci. U. S. A. 106, 19673–19678.

Graham, C.H., Carnaval, A.C., Cadena, C.D., Zamudio, K.R., Roberts, T.E., Parra, J.L., et al., 2014. The origin and maintenance of montane diversity: integrating evolutionary and ecological processes. Ecography 37, 711–719.

Graham, C.H., Machac, A., Storch, D., 2016. Phylogenetic scale in ecology and evolution. bioRvix. http://dx.doi.org/10.1101/063560. 063560.

Graves, G.R., 1985. Elevational correlates of speciation and intraspecific geographic variation in plumage in Andean forest birds. Auk 102, 556–579.

Hall, T., 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symp. Ser. 41, 95–98.

Harmon-Threatt, A.N., Ackerly, D.D., 2013. Filtering across spatial scales: phylogeny, biogeography and community structure in bumble bees. PLoS One 8, e60446.

Jin, L.S., Cadotte, M.W., Fortin, M.-J., 2015. Phylogenetic turnover patterns consistent with niche conservatism in montane plant species. J. Ecol. 103, 742–749.

Körner, C., 2000. Why are there global gradients in species richness?: Mountain might hold the answer. Trends Ecol. Evol. 15, 513–514.

Katoh, K., Toh, H., 2008. Recent developments in the MAFFT multiple sequence alignment program. Briefings Bioinf. 9, 286–298.

Kembel, S.W., Hubbell, S.P., 2006. The phylogenetic structure of a neotropical forest tree community. Ecology 2006, 86–99.

Kembel, S.W., Cowan, P.D., Helmus, M.R., Cornwell, W.K., Morlon, H., Ackerly, D.D., et al., 2010. Picante: R tools for integrating phylogenies and ecology. Bioinf. 26, 1463–1464.

Klimešová, J., Doleal, J., Dvorsky, M., de Bello, F., Klimes, L., 2011. Clonal growth forms in eastern Ladakh, Western Himalayas: classification and habitat preferences. Folia Geobot. 46, 191–217.

Klimes, L., Dickore, W.B., 2006. Flora of Ladakh (Jammu & Kashmir, India). A preliminary checklist published at. http://www.butbn.cas.cz/klimes.

Lavergne, S., Mouquet, N., Thuiler, W., Ronce, O., 2010. Biodiversity and climate change: integrating evolutionary and ecological responses of species and communities. Ann. Rev. Ecol. Evol. Syst. 41, 321–350.

Le Bagousse-Pinguet, Y., Gross, N., Maestre, F.T., Maire, V., de Bello, F., Fonseca, et al., 2017. Testing the environmental filtering concept in global drylands. J. Ecol. 105, 1058–1069.

Le Roux, P.C., McGeoch, M.A., 2004. The use of size as an estimator of age in the subantarctic cushion plant, *Azorella selago* (Apiaceae). Arct. Antarct. Alpine Res. 36, 509–517.

Liancourt, P., Le Bagousse-Pinguet, Y., Rixen, C., Doležal, J., 2017. SGH: Stress or Strain Gradient Hypothesis? Insights from an elevation gradient on the roof of the world. Ann. Bot. 120, 29–38.

Losos, J.B., 2008. Phylogenetic niche conservatism, phylogenetic signal and the relationship between phylogenetic relatedness and ecological similarity among species. Ecol. Lett. 11, 995–1003.

Münkemüller, T., Gallien, L., Lavergne, S., Renaud, J., Roquet, C., Abdulhak, S., et al., 2014. Scale decisions can reverse conclusions on community assembly processes. Global Ecol. Biogeogr. 23, 620–632.

Machac, A., Janda, R., Dunn, R.R., Sanders, N.J., 2011. Elevational gradients in phylogenetic structure of ant communities reveal the interplay of biotic and abiotic constraints on diversity. Ecography 34, 364–371.

Mayfield, M.M., Levine, J.M., 2010. Opposing effects of competitive exclusion on the phylogenetic structure of communities. Ecol. Lett. 13, 1085–1093.

Michalet, R., Le Bagousse-Pinguet, Y., Maalouf, J.-P., Lortie, C.J., 2014. Two alternatives to the stress gradient hypothesis at the edge of life: the collapse of facilitation and the switch from facilitation to competition. J. Veg. Sci. 2, 609–613.

Mouquet, N., Devictor, V., Meynard, C.N., Munoz, F., Bersier, L.-F., Chave, J., et al., 2012. Ecophylogenetics: advances and perspectives. Biol. Rev. 87, 769–785.

Pausas, J.G., Verdu, M., 2010. The jungle of methods for evaluating phenotypic ad phylogenetic structure of communities. Bioscience 60, 614–625.

Pigot, A.L., Etienne, R.S., 2015. A new dynamic null model for phylogenetic community structure. Ecol. Lett. 18, 153–163.

Piston, N., Schöb, C., Armas, C., Prieto, I., Pugnaire, F.I., 2016. Contribution of co-occurring shrub species to community richness and phylogenetic diversity along an environmental gradient. Perspect. Plant Ecol. 19, 30–39.

Reid, A.M., Lamarque, L., Lortie, C.J., 2010. A systematic review of the recent ecological literature on cushion plants: champions of plant facilitation. Web Ecol. 10, 44–49.

Ricklefs, R.E., 1987. Community diversity: relative roles of local and regional processes. Science 235, 167–171.

Ronquist, F., Huelsenbeck, J.P., 2003. MRBAYES 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19, 1572–1574.

Schwarz, G., 1978. Estimating the dimension of a model. Ann. Stat. 6, 461–464.

Schweingruber, F.H., Riha, P., Dolezal, J., 2014. Variation in stem anatomical characteristics of Campanuloideae species in relation to evolutionary history and ecological preferences. PLoS One 9, e88199.

Smith, M.A., Hallwachs, W., Janzen, D.H., 2014. Diversity and phylogenetic community structure of ants along a Costa Rican elevational gradient. Ecography 37, 720–731.

Soliveres, S., Torices, R., Maestre, F.T., 2012. Environmental conditions and biotic interactions acting together promote phylogenetic randomness in semi-arid plant communities: new methods help to avoid misleading conclusions. J. Veg. Sci. 23, 822–836.

Swenson, N.G., Enquist, B.J., Pither, J., Thompson, J., Zimmerman, J.K., 2006. The problem and promise of scale dependency in community phylogenetics. Ecology 87, 2418–2424.

Swenson, N.G., Enquist, B.J., Thompson, J., Zimmerman, J.K., 2007. The influence of spatial and size scale on phylogenetic relatedness in tropical forest communities. Ecology 88, 1770–1780.

Swenson, N.G., 2013. The assembly of tropical tree communities – the advances and shortcomings of phylogenetic and functional trait analyses. Ecography 36, 264–276.

Tanabe, A., 2011. Kakusan4 and Aminosan: two programs for comparing non- partitioned, proportional and separate models for combined molecular phylogenetic analyses of multilocus sequence data. Mol. Ecol. Resources 11, 914–921.

Valiente-Banuet, A., Verdu, M., 2007. Facilitation can increase the phylogenetic diversity of plant communities. Ecol. Lett. 10, 1029–1036.

Verdu, M., Rey, P.J., Alcantara, J.M., Siles, G., Valiente-Banuet, A., 2009. Phylogenetic signatures of facilitation and competition in successional communities. J. Ecol. 97, 1171–1180.

Wang, J., 1988. The steppes and deserts of xizang plateau (Tibet). Vegetatio 75, 135–142.

Webb, C.O., Ackerly, D.D., McPeek, M.A., Donoghue, M.J., 2002. Phylogenies and community ecology. Ann. Rev. Ecol. Evol. Syst. 33, 475–505.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 31–40

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Research article

Reprint of: Disentangling drivers of plant endemism and diversification in the European Alps - a phylogenetic and spatially explicit approach☆

Jan Smyčka[a,*], Cristina Roquet[a], Julien Renaud[a], Wilfried Thuiller[a], Niklaus E. Zimmermann[b], Sébastien Lavergne[a]

[a] *Université Grenoble Alpes, CNRS, Laboratoire d'Ecologie Alpine (LECA), 2233 rue de la Piscine, FR-38000 Grenoble, France*
[b] *Swiss Federal Research Institute WSL, Zürcherstrasse 111, CH-8903 Birmensdorf, Switzerland*

ARTICLE INFO

Keywords:
Glacial refugia
High elevation
Mountains
Speciation
Dispersal
Phylogenetic uncertainty

ABSTRACT

Plant endemism in the European Alps is clustered into particular geographic areas. Two contrasted and non exclusive hypotheses have been suggested to explain these hotspots of endemism: (i) those areas were glacial refugia, where endemism reflects survival-recolonisation dynamics since the onset of Pleistocene glaciations, (ii) those are high elevation mountain areas, where endemism was fostered by local speciation events due to geographic isolation and harsh environmental niches, or by low dispersal ability of inhabiting species.

Here, we quantitatively compared these two hypotheses using data of species distribution in the European Alps (IntraBioDiv database), species phylogenetic relationships, and species ecological and biological characteristics. We developed a spatially and phylogenetically explicit modeling framework to analyze spatial patterns of endemism and the phylogenetic structure of species assemblages. Moreover, we analyzed interrelations between species trait syndromes and endemism.

We found that high endemism occurrs in potential glacial refugia, but only those on calcareous bedrock, and also in areas with high elevation. Plant assemblages in calcareous refugia showed phylogenetic overdispersion − a signature of non-selective conservation forces, whereas those located in high mountain areas showed phylogenetic clustering − a signature of recent diversification and environmental filtering. Endemic species were either stress-tolerant, poorly dispersing species, or high elevation specialists with a wide distribution within the European Alps.

While both calcareous refugia and high-elevation hotspots harbour a large portion of plant endemism in the European Alps, the species they host have substantially different characteristics. Our results suggest that hotspots of endemism in calcareous refugia are more important for nature conservation planning, as they host many range restricted endemic species and rather isolated evolutionary lineages.

1. Introduction

Mountain ranges across the world are considered typical examples of endemic-rich regions (Hughes and Atchinson, 2015), but the evolutionary mechanisms and historical factors generating this high endemism are not fully understood. It was early recognized that mountains have a much richer endemic flora than the surrounding lowlands, and also that within mountain systems, there exist specific areas with exceptionally high endemism (de Candolle, 1875; Pawłowski, 1970). Such hotspots of endemism in alpine regions were observed mostly in putative glacial refugia (Pawlowski, 1970; Tribsch and Schönswetter, 2003; Feng et al., 2016) or in high-elevation areas (Aeschimann et al., 2011; Nagy and Grabherr 2009; Tribsch and Schönswetter, 2003). This suggests that their occurrence is coupled with specific evolutionary dynamics: such regions may exhibit lower extinction rates due to climatic stability and reduced glacial extent, or higher speciation rates and poorer dispersal ability of high-elevation species.

Hotspots of plant endemism in the European Alps (Alps hereafter) have traditionally been explained by the presence of refugia on the periphery of glacial cover during the ice age periods. These refugia are assumed to have promoted long term population persistence of many species during glacial periods, which left imprints in the population structure of survivor species (Alvarez et al., 2009; Schönswetter et al., 2005; Stehlik, 2003). The distribution of glacial refugia has also shaped

---

☆ This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

* Corresponding author.
E-mail address: smyckaj@gmail.com (J. Smyčka).

http://dx.doi.org/10.1016/j.ppees.2017.08.003
Received 31 January 2017; Received in revised form 26 May 2017; Accepted 16 June 2017

contemporary species distributions and endemism patterns, since some survivors could not recolonize all adjacent regions after the retreat of glaciers (Dullinger et al., 2012). It is often assumed that refugia with different bedrocks hosted different pool of species, as the majority of plant endemics of the Alps show either a clear affinity or a strong intolerance to calcareous bedrock, with endemic flora of calcareous bedrock being generally richer (Schönswetter et al., 2005; Tribsch and Schönswetter, 2003). It is likely that species survival during glacial periods has generated particular patterns of endemism, and also left a particular phylogenetic signature in local floras. We can thus hypothesize that species surviving glacial cycles in refugia were drawn from a pool of “pre-glacial” species, whereas species occurring outside refugia were filtered for ability of fast recolonisation, resulting in relative phylogenetic overdispersion of species assemblages in glacial refugia. Here, we aim at quantitatively testing across the whole Alps whether the areas predicted as inhabitable during glacial periods *sensu* Schönswetter et al. (2005) match the above described patterns of endemism and phylogenetic structure, and whether patterns of endemism in these potential refugia are influenced by other factors, as is refugium bedrock, topography, or geographic region.

In addition to the influence of Pleistocene historical processes, patterns of plant diversity and endemism in the Alps may also be linked to the characteristics of high-elevation ecosystems, such as geographic insularity and availability of free but hostile niches. Indeed, it was observed that endemic diversity in the Alps grows with elevation (Aeschimann et al., 2011; Tribsch and Schönswetter, 2003). This pattern could be explained by two processes that are not mutually exclusive. First, increased speciation rate in high elevation ecosystems (documented on a global scale and reviewed in Hughes and Atchison, 2015) could induce higher endemism in certain plant clades. This may be due to heterogeneity or niches in high mountain environments and specific life histories of mountain species favouring sympatric speciation (Dixon et al., 2009; Roquet et al., 2013a, 2013b), or due to topographic obstacles in high mountain environments stimulating allopatric speciation (Boucher et al., 2016; Comes and Kadereit, 2003). Repeated speciation events in high-mountain floras could then induce a phylogenetic signature of radiating lineages, producing a phylogenetic clustering in local species assemblages. Second, increased plant endemism in high elevations may have resulted from a reduced dispersal potential itself. Adaptation to high-alpine environments may imply stress tolerance, long lifespan, and preference for vegetative spread (Körner, 2003) to the detriment of dispersal capabilities (i.e. insularity syndrome). High endemism in mountains resulting from increased speciation rates or decreased dispersal capacities of high-elevation species can thus be expected to result in specific signatures of phylogenetic clustering in local species assemblages or in the presence of particular trait syndromes that have improved survival in high mountain environments at the expense of dispersal potential. This has never been tested to date.

We thus argue that patterns of endemism in the Alps could be explained both by local survival-recolonization dynamics following glaciations, and by dynamics of speciation and dispersal in high-elevation ecosystems. In this paper: (i) We quantitatively compare the relative importance of potential glacial refugia on different bedrocks on one side, and elevation on the other side, for patterns of plant endemism in the Alps. To do this, we use two measures of endemism, namely the proportion of endemic species and range size of endemic species, using a grid-based species occurrence data in the Alps (IntraBioDiv; Gugerli et al., 2008). (ii) We test whether the phylogenetic structure (richness-standardized phylogenetic diversity; Faith, 1992) of species assemblages differs between those different types of hotspots, according to hypothesized evolutionary processes. Importantly, we developed here a novel method based on Bayesian imperfect detection framework in order to overcome difficulties when calculating community phylogenetic indices from non-completely resolved phylogenies (Molina-Venegas and Roquet, 2014; Rangel et al., 2015). (iii) To shed more light on processes forming endemism in high elevations, we explore how endemism and range size of endemics are related to species elevational optimum (Landolt et al., 2010), ecological and functional characteristics related to high elevation adaptations and evolutionary distinctiveness of species (Isaac et al., 2007).

2. Methods

2.1. Study region

We focus on the European Alps, which corresponds to the great mountain range system stretching from south-eastern France to Slovenia. The tree line in the European Alps lies at cca. 2000 m a. s. l and upper limits of vascular plant occurrence lie in 3500–4500 m a.s.l., differing by region (Ozenda and Borel, 2003; Tribsch and Schönswetter, 2003). The European Alps belong to one of the coldest biomes on the planet at its highest elevations (Körner, 2011); nevertheless, this region appears to be relatively species rich, with a fairly high rate of plant endemism (about 13%; Aeschimann et al., 2004; Pawlowski, 1970). The Alps thus constitute a well known hotspot of biodiversity in Europe (Väre et al., 2003).

2.2. Species distribution and environmental data

Species distribution data originate from the mapping of the flora across the Alps produced by the IntraBioDiv consortium (IBD; Gugerli et al., 2008). This dataset contains census and expert based presences and absences of all plant species occurring above the tree line on a regular grid with cells of 20′ longitude and 12′ latitude (ca. 25 × 22 km). This grid was used for all subsequent spatial analyses. The restriction of the species pool to species occurring clearly above the treeline may be considered problematic for example for interpreting species richness across the dataset (but see Taberlet et al., 2012). Nevertheless, while investigating evolutionary processes, such restriction removes potential noise generated by lowland species that likely have reduced evolutionary histories related to mountains. We also excluded gymnosperms and ferns, because some of our working hypotheses may not be extended to them, trait definition for angiosperms are not easily applicable to those groups and sampling efforts for ferns was low compared to angiosperms.

To quantify which grid cells might serve as glacial refugia during glacial cycles, we overlaid the IBD grid with distribution of potential siliceous and calcareous refugia based on combination paleoclimatic model with geological data (adapted from Schönswetter et al., 2005) and estimated whether each grid cell contained calcareous, siliceous, none, or both types of potential refugia. The potential refugia, adapted from Schönswetter et al. (2005), are estimated for maximum of the last glacial period (110,000–12,000 years before present), but they can also be considered a proxy information for distribution of refugia in previous glacial periods. These potential refugia are mostly peripheral (along southwestern, southern, eastern and northern borders of the Alps), likely favouring isolation of plant populations for thousands of years (Schönswetter et al., 2005). The calcareous refugia are the ones lying on limestone or dolomitic bedrock, whereas siliceous refugia lie on variety of acidic bedrocks like granite or gneiss. Larger coherent areas of bedrock types not falling into these two categories are relatively rare within the Alps (Schonswetter et al., 2005; Tribsch and Schönswetter 2003). To separate the effect of refugia from the effect of bedrock itself, we also estimated whether each grid cell contained calcareous (limestone or dolomite) or siliceous bedrock (granite, diorite or gneiss) based on dominant parent material map (PARMADO) from European Soil database (resolution 1 × 1 km). To quantify the topography of grid cells, we calculated their mean elevation and their difference between highest and lowest elevation (elevation range, hereafter) based on Global digital elevation model by US Geological Survey (resolution 30″ × 30″, cca 1 × 1 km). All calculations were performed using the statistical environment R (R Core Team, 2016) and the R libraries raster (Hijmans, 2016), rgdal (Bivand et al., 2016) and spatialEco (Evans, 2016). All data used for our analyses are accessible in Dryad repository under accession number (to be completed after attribution of doi).

2.3. Phylogenetic data

A genus-level phylogeny was built for the Alpine flora using the workflow proposed by Roquet et al. (2013a, 2013b). We downloaded from Genbank three conserved chloroplastic regions (rbcL, matK and ndhF) plus eight regions for a subset of families or orders (atpB, ITS, psbA-trnH, rpl16, rps4, rps4-trnS, rps16, trnL-F), which were aligned separately by taxonomic clustering. All sequence clusters were aligned with three programs (MUSCLE, Edgar, 2004; MAFFT, Katoh et al., 2005; Kalign, Lassmann and Sonnhammer, 2005a), then the best alignment for each region was selected using MUMSA (Lassmann and Sonnhammer, 2005b) and depurated with TrimAl (Capella-Gutiérrez et al., 2009) after visual checks. DNA matrices were concatenated to obtain a supermatrix. Maximum-likelihood phylogenetic inference analyses were conducted with RAxML (Stamatakis et al., 2008) applying a supertree constraint at the family-level based on Davies et al. (2004) and Moore et al. (2010). 100 independent tree searches were performed. The 100 ML trees obtained were dated by penalized-likelihood using r8 s (Sanderson, 2003) and 25 fossils for calibration extracted from Smith and Beaulieu (2010) and Bell et al. (2010).

To deal with unknown within-genera structures, we simulated 10 scenarios of within-genera random branchings for each of 100 genera-level trees using a Yule process as implemented in the R library apTreeshape (Bortolussi et al., 2012). This resulted in 1000 trees that represent a distribution of possible hypotheses about evolutionary relations in our dataset *sensu* Rangel et al. (2015).

2.4. Species ecological and biological features

For each species, we extracted from Flora Indicativa (Landolt et al., 2010) the following ecological indicator values and biological traits:

inverted values of Landolt's T (expert based ordinal scale classification of species elevation, ranging from 1 for lowland to 5 for alpine species) as species level information about its elevational optimum domain,

- CSR strategy *sensu* Grime (1977) depicting stress tolerance (S), ruderal strategy (R) and competitive capacity (C) of each species (S and R were coded as independent ordinal variables with values between 0 a 3 referring to amount of “S” and “R” in three letter characteristic of a species, C is a linear combination of S and R and thus was not used separately; e.g. for CSS species, stress tolerance = 2 and ruderal strategy = 0),
- species dispersal capacity (a value of 1 was attributed to anemo- or zoochoric species, and 0 to those with a different dispersal strategy),
- vegetative reproduction (a value of 1 was attributed to species with the ability of any vegetative reproduction, and 0 to species with completely non-vegetative reproduction),
- cushion life form *sensu* Aubert et al., 2014; a value 1 was attributed to species with vegetative reproduction forming tussocks or cushions, 0 for all other species)
- sexuality of the species (a value of 1 was given to species only capable of sexual reproduction, and 0 for species with facultative or obligate asexual seed generation mechanism, as is apomixis or cleistogamy)
- Raunkiaer plant life-forms (a set of binary variables coding for species being a therophyte, geophyte, hemicryptophyte, chamaephyte or phanerophyte)

Species evolutionary distinctiveness (Isaac et al., 2007) was estimated as a so-called fair proportion measurement (as implemented in R package picante; Kembel et al., 2010) averaged across all 1000 phylogenetic trees. The evolutionary distinctiveness describes whether species are positioned in strongly or weakly branching parts of a phylogenetic tree (sometimes referred as “bushy” or ”stemmy” subtrees), and the inverse value of evolutionary distinctiveness may be considered a species-level measure of diversification rate (Jetz et al., 2012).

Endemism distribution and its relationships with potential glacial refugia and topography

Species were classified as endemic or non-endemic to the Alps based on the Flora Alpina (variable referred to as endemic status hereafter; Aeschimann et al., 2011). To explore the relationship between spatial patterns of endemism, potential glacial refugia, bedrock and topography, we modeled the ratio of the number of endemics to the total number of plant species within each grid (proportion of endemics, hereafter) by a binomial process with the following predictors: mean elevation, elevation range, the presence of calcareous or siliceous bedrock, and presence of potential calcareous or siliceous refugia in grid cell. In order to locate region-specific patterns, spatial smoothing was added as an additional term to the model. To control for overdispersion, a random effect from a Gaussian distribution was added. This was done by means of a generalized additive model (GAM), fitted with Bayesian inference. The MCMC sampling was performed using a JAGS sampler (Plummer, 2003; Plummer, 2016), using modified BUGS code generated by the jagam procedure in the R library mgcv (Wood, 2011). The model code is provided in Appendix D in Supplementary material. We ran the model on 5 chains for 70000 iterations, with a 20000 generation burn-in period and we thinned the resulting chains by 50. The convergence of the model was checked visually and by means of the Gelman-Rubin statistic (Gelman and Rubin, 1992).

The endemic rarity was defined as log(1/species number of occurrences in IBD data) for all species endemic to the Alps. For each grid cell, we calculated the mean rarities of all occurring endemic species (variable futher referred to as mean endemic rarity). To investigate links between mean endemic rarity and the presence of potential refugia and topography, we fitted a GAM similar to the one for proportion of endemics. To deal with the fact that mean endemic rarity is poorly estimated in grid cells with fewer endemics, we added a layer of hierarchy and modeled mean endemic rarity of each grid cell as a latent variable representing the mean of a Gaussian distribution from which rarities of different species in grid cell are drawn. The distribution of mean endemic rarity was modeled as a function of mean elevation, elevation range, presence of calcareous and siliceous bedrock, potential calcareous and siliceous refugia and spatial spline as predictors, using the Gaussian error structure. The model code is provided in Appendix D in Supplementary material. The JAGS sampler setup and convergence checking procedure was identical to the one used for modeling species endemism.

The GAM framework is useful for discovering region-specific patterns, does not rely on any pre-defined relationships, and provides easily interpretable visualizations. However, this framework may not necessarily be the most suitable way to control for spatial autocorrelation (Dormann et al., 2007). Because of that, we also tested other methods to account for spatial autocorrelation, namely spatial generalized least square models and conditional autoregressive models. These alternative methods provided qualitatively similar results to those obtained with the spatial GAM (see Appendix A in Supplementary material), and thus only GAM results are presented and discussed in the main text.

Phylogenetic diversity and its relation to potential glacial refugia and topography

We quantified the phylogenetic structure in each grid cell by calculating phylogenetic diversity (Faith, 1992) standardized for species richness effects (ses.PD), as implemented in R package PhyloMeasures (Tsirogiannis and Sandel, 2017). This standardization was used to remove implicit interdependence between species richness and phylogenetic diversity, and resulting values are further interpreted in this way.

The link between ses.PD and the presence of potential refugia and topography was explored with a bayesian GAM similar to the one for proportion of endemism or mean endemic rarity. In order to control for phylogenetic uncertainty, we performed ses.PD calculations for each of 1000 phylogenetic trees within each grid cell. The "real" value of ses.PD was modeled as a latent variable representing mean parameter of Gaussian distribution from which ses.PDs of different trees are drawn. The distribution of the "real" ses.PD per grid cell was modeled as a function of mean elevation, elevation range, presence of calcareous or siliceous bedrock, potential calcareous or siliceous refugia and a spatial spline as predictors, using a Gaussian error structure. For the model code, see Appendix D in Supplementary material. The sampler setup and convergence checking was identical as for the previous Bayesian models.

Endemism-elevation relationships and its link to species traits and ecological properties

In order to explore relationships between species endemism and their elevational niche, we modeled species-level endemic status (endemism to the Alps coded as a binary variable) with a non-spatial generalized linear model (GLM) with a binomial error term, and used each species' elevational optimum (Landolt's T) as a predictor. To evaluate how the other species characteristics affected this endemism-elevation relationship, we fitted the same model with additional predictors: evolutionary distinctiveness, ecological strategies and species traits (see above "Species ecological and biological features"). All predictors were standardized prior to analyses. The important predictors were selected using forward-backward model selection based on Akaike's information criterion (AIC), as implemented in stepAIC procedure in R, starting from a full model containing all variables. In order to tease apart the relative effects of different species features, partial correlations for non-parametric Kendall's correlation coefficient were computed using the package ppcor in R (Kim, 2015). To explore relationships among endemics rarity, elevational optimum and other species characteristics, we fitted models similar to the ones used for endemic status. In this case we used Gaussian error structure in the GLM and parametric Pearson correlation coefficients for estimating partial correlations.

To ascertain that analyses were not biased by a phylogenetic structure in the dataset, we re-ran the final models obtained above by the stepAIC procedure within a phylogenetic regression framework, as implemented in the package phylolm (Ho and Ane, 2014). We obtained an Ornstein-Uhlenbeck alpha = 0.193 in the binomial model of endemic status and Pagel's lambda = 0.037 in the Gaussian model of endemics rarity. These values showed that the phylogenetic signal was weak in both cases, making non-phylogenetic analyses appropriate with our data set (for results of phylogenetic comparative models see Appendix C in Supplementary material).

3. Results

Endemism distribution and its relationships with potential glacial refugia and topography

The proportion of endemics tended to be significantly (in the sense that Bayesian 95% credibility interval did not overlap 0) larger in grid cells that contained calcareous bedrock (effect size = 0.245; (see Table 1 for 95% credibility intervals, and Appendix A in Supplementary material for the results of alternative spatial models). This effect was further augmented by the presence of potential calcareous refugia (ES = 0.195). The proportion of endemics was not affected by the presence of siliceous bedrock and was even significantly lower in cells containing potential siliceous refugia (ES = −0.070). The proportion of endemics increased significantly with mean elevation (ES = 0.256) and elevation range (ES = 0.161) in grid cells; for instance, an increment of 1000 m in mean elevation has an effect comparable to presence of potential calcareous refugia (Fig. 1A). The spatial component of the model showed that the proportion of endemics declined towards the north-western edge of the Alps, and to a lower extent toward the eastern edge of the Alps (Fig. 1 B).

The mean endemic rarity was significantly positively associated with the presence calcareous bedrock (ES = 0.107) and of potential calcareous refugia (ES = 0.068; see Table 1 for 95% credibility intervals).The effect of siliceous bedrock was negative (ES = −0.077) and the effect of potential siliceous refugia was not distinguishable from 0. Mean endemic rarity was influenced neither by mean elevation nor by the elevation range (fig. 1C). Similar as for the proportion of endemics, we found a decline of mean endemic rarity towards the north-western corner of the Alps (Fig. 1D).

Phylogenetic diversity and its relation to potential glacial refugia and topography

Grid cells containing calcareous bedrock presented systematically larger ses.PD (ES = 0.487, see Table 1 for 95% credibility intervals) and this effect was further augmented in potential calcareous refugia (ES = 0.575). Effect of siliceous bedrock on ses.PD is not distinguishable from 0, but potential siliceous refugia exhibited marginally significant increase of ses.PD (ES = 0.255). Large ses.PD in calcareous bedrock, calcareous refugia and possibly siliceous refugia indicates that these areas host species assemblages separated by longer evolutionary branches than the rest of dataset. Sites with a higher mean elevation hosted species assemblages with a relatively lower ses.PD (ES = −0.335); a decrease in 500 m of mean elevation is comparable to the presence of potential calcareous refugia (Fig. 2a). This indicates that areas of higher mean elevation in the Alps host species assemblages that are more closely related than the rest of the dataset. Visualization of the spatial component of the model displayed particular areas with high (Savoy Prealps, Julian Alps) or low ses.PD (Ötztal and Rhaetian Alps). These spatial effects were, however, weak in comparison to the effects of the linear terms of the model (see Fig. 2b).

Endemism-elevation relationship and its link to species traits and ecological properties

The elevational optimum of species (summarized with Landolt's T) was a significant positive predictor of species endemic status (tau = 0.086; Fig. 3A, see Table 2 for *p*-values). When mixed with other species trait predictors, the relationship with elevation became weaker

Table 1
Mean effect size (ES), lower and upper bounds of the 95% credibility intervals of the effects sizes of spatial models in which the proportion of endemics and the mean endemics rarity, as well as species-richness standardized phylogenetic diversity (ses.PD) are explained. Model estimates with credibility intervals not overlapping with 0 are given in bold.

	proportion of endemics			mean endemic rarity			ses.PD		
	lower	ES	upper	lower	ES	upper	lower	ES	upper
intercept	**−3.102**	**−2.962**	**−2.822**	**−4.483**	**−4.368**	**−4.253**	−0.171	0.162	0.501
calc. bedrock	**0.173**	**0.245**	**0.318**	**0.051**	**0.107**	**0.164**	**0.247**	**0.487**	**0.727**
calc. refugia	**0.135**	**0.195**	**0.255**	**0.020**	**0.068**	**0.116**	**0.379**	**0.575**	**0.770**
silic. bedrock	−0.057	0.025	0.108	**−0.142**	**−0.077**	**−0.012**	−0.471	−0.225	0.021
silic. refugia	**−0.140**	**−0.070**	**−0.002**	−0.064	−0.008	0.048	**0.000**	**0.255**	**0.511**
mean elevation	**0.185**	**0.256**	**0.327**	−0.008	0.048	0.103	**−0.596**	**−0.335**	**−0.077**
elevation range	**0.096**	**0.161**	**0.226**	−0.067	−0.015	0.036	−0.046	0.201	0.447

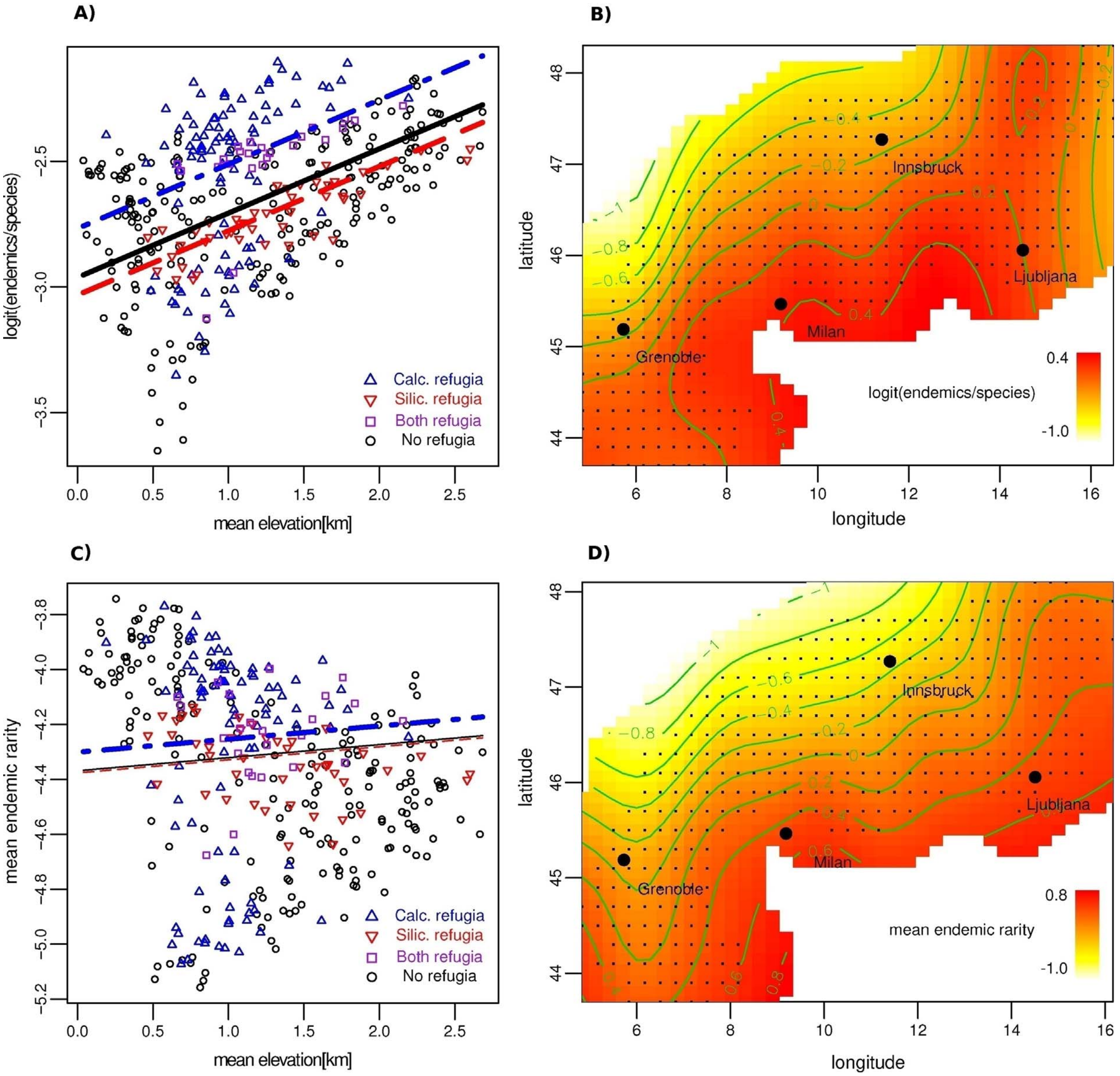


Fig. 1. Spatial models depicting the geographic patterns of proportion of endemics (A, B) and the geographic patterns of mean rarity of endemics (C, D). Panels (A) and (C) show relationship of these two indices with mean elevation for grid cells without refugia (black, solid line, plotted in bold if effect of mean elevation is significant), with calcareous refugia (blue, double dashed line, plotted in bold if effect of calcareous refugia is significant) or with siliceous refugia (red, single dashed line, plotted in bold if effect of siliceous refugia is significant). Note that y-values of points are adjusted to account for the effect of not-displayed model variables and for the mean effect of smooth model component per group. Panels (B) and (D) represent the smooth component of each model, showing geographic areas with overall higher or lower proportion of endemics or mean endemic rarity when simultaneously accounting for parametric model components. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

but remained significant (tau = 0.032). Concerning other species-level predictors, the endemic status is significantly linked to poor dispersal capacity (tau = −0.137), sexuality (tau = 0.080), and stress tolerance (tau = 0.100; Fig. 3A, Table 2). Evolutionary distinctiveness was negatively related to endemic status (tau = −0.054), and its inclusion improved model AIC, but this effect did not reach statistical significance (p = 0.134). The cushion life form was positively related to endemic status (tau = 0.064); the inclusion of this trait also improved the model AIC, but it did not reach statistical significance (p = 0.110) either.

Endemics rarity was significantly negatively linked with elevational optimum (R = −0.110) and the strength of this relationship remained similar in a model that included species traits and ecological predictors (R = −0.102; Table 2, Fig. 3B). Endemics rarity was further associated with sexuality (R = 0.068) and geophyte life form (R = 0.144). Dispersal capacity (R = −0.084) had a weak but significant negative effect on rarity (p = 0.029), but this would turn margnially non-significant if accounting for phylogenetic correction (p = 0.097; see Appendix C in Supplementary material). Stress tolerance (R = 0.046) and hemicryptohyte life form (R = 0.084) were positively related to endemics rarity and improved model fit, yet these effects did not reach significance (p = 0.088, p = 0.104; Fig. 3B, Table 2).

4. Discussion

In this study, we quantitatively compared the importance of endemism hotpots in potential glacial refugia and in areas with high elevation. We showed that the phylogenetic structure of plant assemblages occurring in these two types of hotspots is substantially different and reflects their contrasting evolutionary histories. Moreover, we explored so far undocumented interrelations between plant endemism, elevation, species ecological strategies, biological traits and evolutionary distinctiveness.

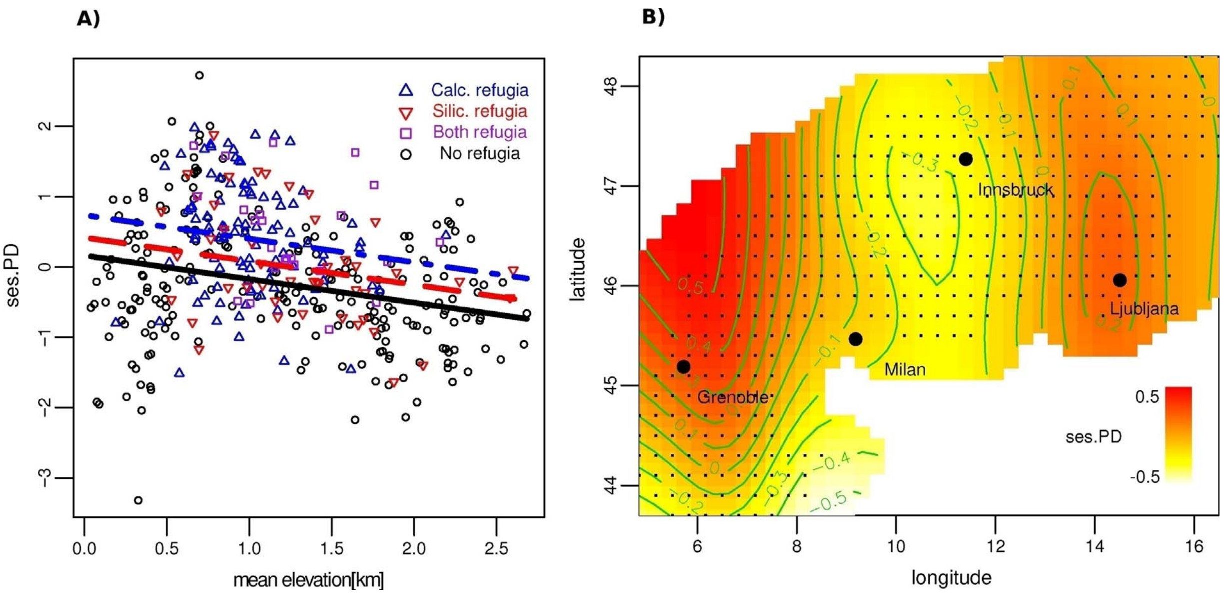


Fig. 2. Spatial model depicting geographic patterns of species-richness standardized phylogenetic diversity (ses.PD). Panel (A) shows the relationship of ses.PD with mean elevation (black, solid line, plotted in bold if effect of mean elevation is significant) and the occurrence of calcareous (blue, double dashed line, plotted in bold if effect of calcareous refugia is significant), siliceous refugia (red, single dashed line, plotted in bold if effect of siliceous refugia is significant). Note that y-values of points are adjusted to account for the effect of not-displayed model variables and for the mean effect of smooth model component per group. Panel (B) represents the smooth component of the model, showing geographic areas with overall higher or lower ses.PD when simultaneously accounting for parametric model components. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

4.1. Hotspots of endemism in calcareous refugia and areas with high elevation

Areas of the Alps comprising potential calcareous glacial refugia hold a substantially high proportion of alpine endemics, and these endemics are typically narrowly distributed (with high rarity of endemics) within the Alps. This is partly caused by generally higher endemism in calcareous areas, but potential calcareous refugia host even larger proportion and rarer endemics that calcareous areas in general in our data. It suggests that potential calcareous refugia indeed host species that did not manage to recolonize larger areas after the retreat of glaciers (Dullinger et al., 2012). Increasing endemism with elevation has an effect per 1000 elevation meters comparable to the presence of calcareous refugia. However, high-elevation endemics are typically widespread within the Alps (with low rarity), in contrast to narrowly distributed endemics of potential calcareous refugia. While high endemism in glacial refugia and high elevations was previously reported from the Alps (Aeschimann et al., 2011; Tribsch and Schönswetter, 2003) and other mountain systems across the world (Feng et al., 2016; Mráz et al., 2016; Nagy and Grabherr, 2009), we quantitatively measured and tested those effects within a spatially explicit modeling framework. We show that endemism in potential calcareous glacial refugia and in high elevations are of comparable importance in the Alps, suggesting that similar patterns might be found in other mountain ranges affected by Pleistonece glaciation dynamics. Our quantitative approach does not only compare importance of refugia and elevation gradient for formation of endemic hotspots in the Alps, but also allows us to shed light on exceptions from this general trend.

Interestingly, we found that the overall patterns of high proportion of endemics and the high rarity of endemics in potential calcareous refugia are not paralleled in potential siliceous refugia. The mean rarity of endemic species occurring in potential siliceous refugia is comparable to the siliceous areas outside the refugia and the proportion of alpine endemics in potential siliceous refugia is even significantly lower than outside refugia. This suggests that high endemism previously reported in some potential siliceous refugia (Tribsch and Schönswetter, 2003) could at least in some cases rather be attributed to the other predictors used in our model than to favourable conditions during glacial periods, i.e. those areas might be endemic-rich due to topographic roughness or because they lie in region that is overally richer in endemics (which is modeled by spatial autocorrelation in our model). A possible reason why potential siliceous refugia are not richer in endemics than non-refugial siliceous areas is that siliceous refugia are typically interconnected with non-refugial siliceous areas in the central Alps, in contrast to calcareous refugial and non-refugial areas, which are scattered at the margins of the Alps. Hence, most siliciceous specialists species that surived glacial periods may have been able to recolonize broader areas after glacier retreat due to lower spatial isolation of habitats on siliceous bedrock (Alvarez et al., 2009; Dullinger et al., 2012). Following this hypothesis, postglacial migration might have erased patterns of endemism, even though the presence of siliceous refugia may be still visible in the spatial genetic structures of some particular species (Schönswetter et al., 2005; Stehlik, 2003).

Spatial models of proportion of endemics and mean endemic rarity suggest that the region of the north-western Alps hosts a lower proportion of endemics and also fewer rare endemics. A possible explanation is that the impact of glaciers was relatively stronger in this area due to combination of lower elevation and higher glacial extent towards the north-western part of the Alps (see maximum extent of glaciers in Schonswetter et al., 2005), making potential refugia in this area uninhabitable. This explanation is in agreement with studies tracking glacial refugia using intraspecific genetic patterns (Schönswetter et al., 2005), counts of endemic species (Tribsch and Schönswetter, 2003), or species rarity (Taberlet et al., 2012);all of these studies found weak or no support for the existence of glacial refugia in the north-western Alps.

4.2. Calcareous refugia as species museums and high elevations as cradles

Areas with potential calcareous refugia contain systematically higher richness-standardized phylogenetic diversity (ses.PD), which suggests that they host a higher proportion of species with long and isolated branches than other sites in the Alps. The ses.PD in potential calcareous refugia is higher also in comparison with calcareous areas outside refugia. Such a phylogenetic signature may result from a random selection of refugial survivors from the pre-glacial species pool

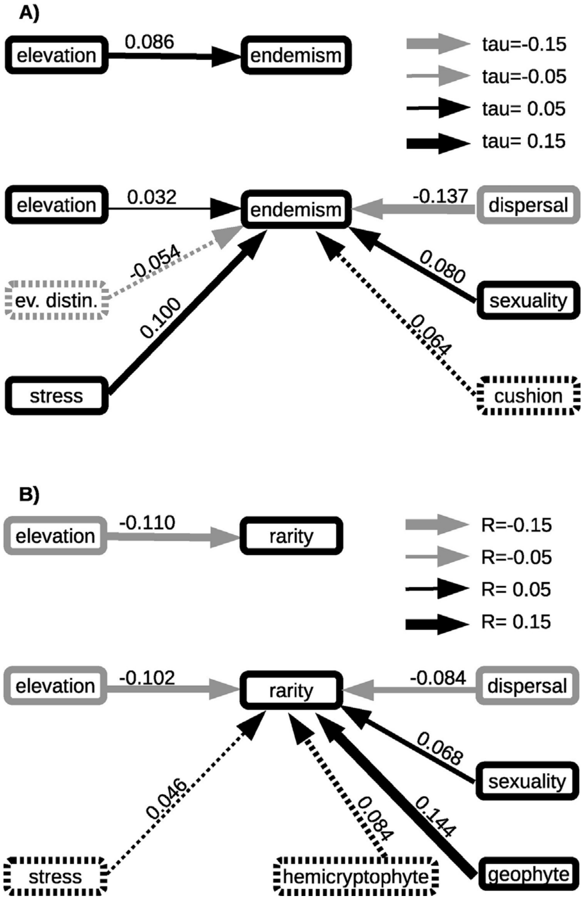


Fig. 3. Relationship of elevational optimum (Landolt's T) and species endemic status (A) or the rarity of endemic species (B) in elevation only models and in models with elevation and the other species level predictors included. Tau is the partial Kendall correlation in binomial model of endemic status (A), R is the partial Pearson correlation in Gaussian model (B) of endemics rarity. All listed predictors contribute to model fit as measured by AIC, predictors significant at $p < 0.05$ appear with solid arrows, non-sigificant with dashed.

combined with selective re-colonizations of previously glaciated sites by well-dispersing species, resulting in assemblages at sites outside refugia being more clustered than in refugia. In addition, resource competition in refugia during glacial periods could have also contributed to the competitive exclusion of related species *sensu* Webb et al. (2002). The ses.PD in glacial refugia might have also been augmented by allopatric speciation among isolated glacial refugia, which would result in sister species seldom sharing one refugial site (Pigot and Etienne, 2015; Warren et al., 2014). With the current resolution of phylogenetic data and without evidence for past migration from and to the Alps it is not possible to estimate the relative importance of those three processes. Still, the observed evolutionary overdispersion in potential calcareous refugia strongly supports the idea that those areas constitute museums of phylogenetic diversity of the Alpine flora *sensu* Stenseth (1984), and served as islands of suitable condition during glacial periods. The long-term conservation of this local flora since glacial periods onwards has thus made them important zones for the protection of plant phylogenetic diversity in the Alps. For potential siliceous refugia, we found a pattern of phylogenetic overdispersion similar to potential calcareous refugia, but only marginally significant. It supports the interpretation that potential siliceous refugia also hosted species during glacial periods. Although the patterns of endemism were erased by postglacial migrations, the imprint of glacial survival may still be detectable in the phylogenetic structure of plant assemblages.

Table 2
Effect size (ES), p-values and correlation coefficients (tau and R) of models explaining endemic status and endemics rarity species level characteristics. Model results are shown with species altitudinalelevational optima (Landolt's T) as a single predictor, and in combination with all other species characteristics. N stands for predictors that did not pass the stepwise AIC optimization procedure and thus did not contribute to the model. Bold are significant model terms ($p < 0.05$).

	endemic status			endemics rarity		
	tau	ES	p	R	ES	p
elevational optimum alone	**0.086**	**0.297**	**< 0.001**	**− 0.11**	**− 0.194**	**0.012**
elevational optimum	**0.032**	**0.179**	**0.043**	**− 0.102**	**− 0.206**	**0.009**
evolutionary distinctiveness	− 0.054	− 0.132	0.134	N	N	N
ruderal strategy	N	N	N	N	N	N
stress strategy	**0.1**	**0.231**	**0.009**	0.046	0.144	0.088
dispersal	**− 0.137**	**− 0.297**	**< 0.001**	**− 0.084**	**− − 0.183**	**0.029**
sexuality	**0.08**	**0.258**	**0.023**	**0.068**	**0.193**	**0.01**
vegetative reproduction	N	N	N	N	N	N
cushions	0.064	0.122	0.11		N	N
chamaephyte	N	N	N	N	N	N
geophyte	N	N	N	**0.144**	**0.245**	**0.003**
hemicryptophyte	N	N	N	0.084	0.136	0.104
phanerophyte	N	N	N	N	N	N
therophyte	N	N	N	N	N	N

Areas in the Alps with high mean elevation exhibit significantly lower ses.PD than other sites. This means that with increasing elevation, species are more likely co-occur with their close relatives, and that this pattern is consistent throughout the Alps. Two classes of processes may explain this pattern. First, the high alpine flora could be strongly shaped by environmental filtering *sensu* Webb et al. (2002) by which related species tend to share adaptations to similar climatic environments and are, thus, jointly sorted across climatic gradients. Second, this first process may have almost certainly been amplified by recent and repeated speciations in clades that have largely diversified in alpine environments (Boucher et al., 2016; Mansion et al., 2012; Molina-Venegas et al., 2015; Roquet et al., 2013a, 2013b). Clearly, environmental filtering alone cannot explain the phylogenetic clustering of high alpine floras, since we found additional evidence that species richness increases with mean elevation (see Appendix B in Supplementary material) and increased filtering should decrease species richness. Our results are thus in line with the extensive evidence that plant clades have rapidly diversified into high-mountain environments in general (reviewed in Hughes and Atchison, 2015). Given increasing evidence that speciation in alpine plants widely occur by allopatric speciation with little niche shift (Boucher et al., 2016; Comes and Kadereit, 2003), the observed signature of phylogenetic clustering suggests that migration during post-glacial recolonization caused closely related species to co-occur in similar mountain environments, a process known as secondary sympatry (Pigot and Tobias, 2013).

Our analyses thus show that high elevation and refugial hotspots of endemism in the Alps are of similar importance, but were formed by different evolutionary processes. It suggests that in the Alps, but possibly also in other temperate mountains affected by Pleistocene glaciation dynamics, endemism should not be considered an indicator of one particular evolutionary process as is glacial survival, altered dispersal dynamics or faster speciation.

4.3. High elevation endemism is shaped by dispersal

Above we discussed that areas with high elevation and calcareous refugia are important hotspots of endemism and here we focus more closely on processes forming endemism in high elevations. Accordingly with spatial models, our species-level analysis provides evidence that species endemic status is positively related to elevational optimum and that endemic species with high elevational optimum have larger distribution ranges (lower rarity) than endemic species from lower elevations. However, once other predictors are included, the effect of species elevational optima on endemism becomes much weaker. The major pattern arising from this analysis is that poorly dispersing and/or stress tolerant endemics tend to be over-represented with increasing elevation. In addition, endemics with restricted distribution ranges (high rarity) seem to be rather poor-dispersing and stress tolerant species, although this last effect is weaker and marginally significant. The relationship between endemism, elevation optimum, dispersal and stress tolerance is in line with previous findings (Aeschimann et al., 2012; Hobohm, 2008; Mráz et al., 2016), and suggests that some of the high-elevation species are endemic, because of their poor colonizing capabilities that are linked to their ecological and trait characteristics. These species are specialists of stressful habitats, such as of rocky outcrops or high-elevation habitats, possibly within glacial refugia, e.g. in Dolomites, Julian Alps or south-western Alps. Typical examples of narrowly distributed high-elevation endemics that are stress tolerant and poor dispersers are: *Rhizobotrya alpina*, *Braya alpina*, *Draba ladina*, *Sempervivum dolomiticum, Saxifraga florulenta, Myosotis gallica*, *Cardaminopsis pedemontana*, *Linaria tonzigii* or *Moehringia concarenae*.

Species life-forms and reproduction strategies also partly explained species endemic status and the rarity of endemic species. Cushion plants are more likely alpine endemics (e.g. *Androsace alpina*), and geophytes are more likely rare endemics (e.g. A*llium kermesinum*) than expected by chance. Our results also indicate that sexually reproducing species are more likely to be endemic and rare than asexual ones. This may appear surprising given that asexual lineages tend to create small but distinct populations, typically considered a species (e.g. *Alchemilla or Sorbus*). This result is thus likely an artefact derived from the fact that, in large compilations such as IntraBioDiv (Gugerli et al., 2008) or Flora Alpina (Aeschimann et al., 2004), asexual and taxonomically problematic species are typically merged to relatively broadly aggregated species definitions. For instance, the genus *Alchemilla* has only 5 species distinguished in the IntraBioDiv data. To shed more light on endemism-sexuality relations, it would be necessary to explicitly control for the species definition, e.g. by including data about species population structures, and perhaps by tackling the issue of species delimitation with modern sequencing techniques. However, given the large number of species of the Alps, this is likely a very demanding, if currently not impossible, task.

In contrast to endemics that are stress-adapted, poorly dispersing or having endemism-related life form, there is another large portion of species whose endemic status is best explained by elevational optimum itself. Such endemics are typically widespread within the Alps (have low rarity), and are likely well adapted to environments such as the subnival zone, that are relatively common within the Alps, but rare or non-existing in other European mountain systems. These species are thus expected to have a dispersal capacity sufficient to spread across the Alps. Yet, their dispersal capacity is not sufficient for colonizing and maintaining viable populations in isolated patches of suitable habitats outside the Alps. Typical examples of well-dispersing, widespread high-elevation endemics are for example: *Festuca interdecens*, *Cerastium pedunculatum*, *Crepis terglouensis*, *Adenostyles leucophylla*, *Erigeron neglectus* or *Artemisia genepi*.

Surprisingly, the evolutionary distinctiveness of species is only weakly linked to endemic status; one would have expected that endemism should be common in species with little evolutionary distinctiveness resulting from repeated recent speciation events. Our analysis on the relationship between endemic status and evolutionary distinctiveness may have been partly weakened by the use of genus-level phylogenies, as recent speciation events would be most apparent at terminal branches, where we are missing accurate information. But if speciation events in general were a main driving force of high mountain endemism patterns, it is unlikely that missing information at the species level would completely remove the signal. The weak relationship between endemic status and evolutionary distinciveness is interesting, because according to other results from our study, the flora of high elevation areas (including non-endemic species) shows a pattern of phylogenetic clustering − a possible indication of faster and recent diversification. Combining our findings, we hypothesize that the high-elevation flora may indeed have faster speciation rate, as was reported in studies from mountains all around the world (reviewed in Hughes and Atchison, 2015). However, even the youngest speciation events in our phylogeny are older than the last glaciation period (all terminal branch lenght estimates are longer than 110,000 years) and most of them is even older than the onset of glaciation-deglaciation cycles in the Quaternary (77% of terminal branch length estimates are longer than 2.58 million years). This suggests that although species may speciate faster in high elevations, the relationship between speciation and endemism is further erased by massive glacial-interglacial migrations and extinctions (see Kadereit et al., 2004). The temporal dynamics of the evolution of new species and drivers of endemism need to be further studied with a well-dated species-level phylogeny of the Alps and possibly beyond such as including the Carpathians and the Pyrenees or the Balkanic mountain systems.

5. Conclusions

Our analyses demonstrate that important hotspots of endemism in the European Alps are situated in potential calcareous glacial refugia, rather than siliceous ones, but also in areas with high elevation: an elevational change of 1000 m contributes to endemism similarly as the presence of potential calcareous refugium. Potential calcareous refugia harbour a large portions of endemics that are rare within the Alps, and plant assemblages with higher phylogenetic diversity than random expectations. It suggests that these areas indeed served as save harbours for distinct evolutionary linages during the periods of glaciation. The assemblages of high-elevation hotspots carry a signature of phylogenetic clustering, which indicates a combination of strong environmental filtering and faster speciation rate. More detailed analyses of high elevation endemism reveal that a large portion of endemics in the Alps are range restricted species with poor dispersal ability, and another large portion of endemics are high elevation specialists that are widespread within the Alps. Our results show that endemic hotspots both in calcareous refugia and high elevations are of high importance, but they are formed by contrasting evolutionary processes. In line with this, future studies of endemism and diversification in the Alps or other mountain ranges affected by past glaciation should consider that endemism in such areas is formed by interplay of migration and different diversification processes acting on different timescales, rather than one dominant force. From a point of view of nature conservation,calcareous refugia in the Alps deserve more attention than high elevation hotspots, because they host primarily range-restricted endemics and phylogenetically distinct species retaining a substantial evolutionary history overall.

Acknowledgements

We would like to acknowledge Tomáš Herben, Zdeněk Janovský, Tamara Münkemüller, Matthew Talluto and Martin Weiser for helpful comments at different stages of preparation of the manuscript, and David Aeschimann for providing digitalized Flora Alpina and useful advices concerning this data source. The research was funded by the ANR project Origin-Alps (ANR-16-CE93-0004), the European Research Council FP7/2007–2013 Grant Agreement no. 281422 (TEEMBIOby a grant from Labex OSUG@2020 (Investissements d'avenir – ANR10LABX56) and a grant from the Pôle départemental de recherches sur la Biodiversité (Conseil Général de l'Isère). Jan Smyčka was supported by doctoral funding of French Ministry of Higher Education and Research.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.ppees.2017.08.003.

References

Aeschimann, D., Lauber, K., Moser, D.M., Theurillat, J.P., 2004. Flora Alpina. Haupt Verlag, Bern, Stuttgart, Wien.

Aeschimann, D., Rasolofo, N., Theurillat, J.P., 2011. Analyse de la flore des Alpes. 2 Diversité et chorologie. Candollea 66, 193–219.

Aeschimann, D., Rasolofo, N., Theurillat, J.P., 2012. Analyse de la flore des alpes. 4: ecologie. Candollea 67, 193–219.

Alvarez, N., Thiel-Egenter, C., Tribsch, A., Holderegger, R., Manel, S., Schönswetter, P., Taberlet, P., Brodbeck, S., Gaudeul, M., Gielly, L., Küpfer, P., Mansion, G., Negrini, R., Paun, O., Pellecchia, M., Rioux, D., Schüpfer, F., Van Loo, M., Winkler, M., Gugerli, F., 2009. History or ecology? Substrate type as a major driver of patial genetic structure in Alpine plants. Ecol. Lett. 12, 632–640. http://dx.doi.org/10.1111/j.1461-0248.2009.01312.x.

Aubert, S., Boucher, F., Lavergne, S., Renaud, J., Choler, P., 2014. 1914–2014: A revised worldwide catalogue of cushion plants 100 years after Hauri and Schröter. Alp. Bot. 124, 59. http://dx.doi.org/10.1007/s00035-014-0127-x.

Bell, C.D., Soltis, D.E., Soltis, P.S., 2010. The age and diversification of the angiosperms re-revisited. Am. J. Bot. 97, 1296–1303.

Bivand, R., Keitt, T., Rowlingson, B., 2016. Rgdal: Bindings for the Geospatial Data Abstraction Library. R Package Version 1.2-5.

Bortolussi, N., Durand, E., Blum, M., Francois, O., 2012. ApTreeshape: Analyses of Phylogenetic Treeshape. R Package Version 1.4-5.

Boucher, F.C., Zimmermann, N.E., Conti, E., 2016. Allopatric speciation with little niche divergence is common among alpine Primulaceae. J. Biogeogr. 43, 591–602. http://dx.doi.org/10.1111/jbi.12652.

Capella-Gutiérrez, S., Silla-Martínez, J.M., Gabaldón, T., 2009. trimAl: a tool for automated alignment trimming in large-scale phylogenetic analyses. Bioinformatics 25, 1972–1973.

Comes, H.P., Kadereit, J.W., 2003. Spatial and temporal patterns in the evolution of the flora of the european alpine systems. Taxon 52, 451–562.

Davies, T.J., Barraclough, T.G., Chase, M.W., Soltis, P.S., Soltis, D.E., Savolainen, V., 2004. Darwin's abominable mystery: insights from a supertree of the angiosperms. Proc. Natl. Acad. Sci. U.S.A. 101, 1904–1909.

Dixon, C.J., Schönswetter, P., Suda, J., Wiedermann, M.M., Schneeweiss, G.M., 2009. Reciprocal Pleistocene origin and postglacial range formation of an allopolyploid and its sympatric ancestors (Androsace adfinis group, *Primulaceae*). Mol. Phylogenet. Evol. 50, 74–83. http://dx.doi.org/10.1016/j.ympev.2008.10.009.

Dormann, C.F., McPherson, J.M., Araújo, M.B., Bivand, R., Bolliger, J., Carl, G., Davies, R.G., Hirzel, A., Jetz, W., Kissling, D.W., Kühn, I., Ohlemüller, R., Peres-Neto, P.R., Reineking, B., Schröder, B., Schurr, F.M., Wilson, R., 2007. Methods to account for spatial autocorrelation in the analysis of species distributional data: a review. Ecography 30, 609–628. http://dx.doi.org/10.1111/j.2007.0906-7590.05171.x.

Dullinger, S., Willner, W., Plutzar, C., Englisch, T., Schratt-Ehrendorfer, L., Moser, D., Ertl, S., Essl, F., Niklfeld, H., 2012. Post-glacial migration lag restricts range filling of plants in the European Alps. Glob. Ecol. Biogeogr. 21, 829–840. http://dx.doi.org/10.1111/j.1466-8238.2011.00732.x.

Edgar, R.C., 2004. MUSCLE: multiple sequence alignment with high accuracy and high throughput. Nucleic Acids Res. 32, 1792–1797.

Evans, J.S., 2016. SpatialEco. R Package Version 0.0. pp. 1–4.

Faith, D.P., 1992. Conservation evaluation and phylogenetic diversity. Biol. Conserv. 61, 1–10.

Feng, G., Mao, L., Sandel, B., Swenson, N.G., Svenning, J.-C., 2016. High plant endemism in China is partially linked to reduced glacial-interglacial climate change. J. Biogeogr. 43, 145–154. http://dx.doi.org/10.1111/jbi.12613.

Gelman, A., Rubin, D.B., 1992. Inference from iterative simulation using multiple sequences. Stat. Sci. 7, 457–511.

Grime, J.P., 1977. Evidence for the existence of three primary strategies in plants and its relevance for ecological and evolutionary theory. Am. Nat. 111, 1169–1194.

Gugerli, F., Englisch, T., Niklfeld, H., Tribsch, A., Mirek, Z., Ronikier, M., Zimmermann, N.E., Holderegger, R., Taberlet, P., 2008. Relationships among levels of biodiversity and the relevance of intraspecific diversity in conservation − a project synopsis. Perspect. Plant Ecol. Evol. Syst. 10, 259–281. http://dx.doi.org/10.1016/j.ppees.2008.07.001.

Hijmans, R.J., 2016. Raster: Geographic Data Analysis and Modeling. R Package Version 2. pp. 5–8.

Ho, L.S.T., Ane, C., 2014. A linear-time algorithm for Gaussian and non-Gaussian trait evolution models. Syst. Biol. 63, 397–408.

Hobohm, C., 2008. Ökologie und verbreitung endemischer gefäßpflanzen in europa. Tuexenia 28, 10–22.

Hughes, C.E., Atchinson, G.W., 2015. The ubiquity of alpine plant radiations: from the Andes to the Hengduan Mountains. New Phytol. 207, 275–282. http://dx.doi.org/10.1111/nph.13201.

Isaac, N.J.B., Turvey, S.T., Collen, B., Waterman, C., Baillie, J.E.M., 2007. Mammals on the EDGE: Conservation priorities based on threat and phylogeny. PLoS One. http://dx.doi.org/10.1371/journal.pone.0000296.

Jetz, W., Thomas, G.H., Joy, J.B., Hartmann, K., Mooers, A.O., 2012. The global diversity of birds in space and time. Nature 491, 444–448. http://dx.doi.org/10.1038/nature11631.

Körner, C., 2003. Alpine Plant Life − Functional Plant Ecology of High Mountain Ecosystems, 2nd ed. Springer, Berlin, pp. 274–290.

Körner, C., 2011. Coldest places on earth with angiosperm plant life. Alp. Bot. 121, 11–22.

Kadereit, J.W., Griebeler, E.M., Comes, H.P., 2004. Quaternary diversification in European alpine plants: pattern and process. Phil. Trans. R. Soc. Lond. B. 359, 265–274.

Katoh, K., Kuma, K., Toh, H., Miyata, T., 2005. MAFFT version 5: improvement in accuracy of multiple sequence alignment. Nucleic Acids Res. 33, 511–518.

Kembel, S.W., Cowan, P.D., Helmus, M.R., Cornwell, W.K., Morlon, H., Ackerly, D.D., Blomberg, S.P., Webb, C.O., 2010. Picante: r tools for integrating phylogenies and ecology. Bioinformatics 26, 1463–1464.

Kim, S., 2015. Ppcor: Partial and Semi-Partial (Part) Correlation. R Package Version 1.1.

Landolt, F., Bäumler, B., Erhardt, A., Hegg, O., Klötzli, F., Lämmler, W., Nobis, M., Rudmann-Maurer, K., Schweingruber, F.H., Theurillat, J.-P., Urmi, E., Vust, M., Wolgemuth, T., 2010. Flora Indicativa − Ökologische Zeigerwerte Und Biologische Kennzeichen Zur Flora Der Schweiz Und Der Alpen. Haupt Verlag, Bern, Stuttgart, Wien.

Lassmann, T., Sonnhammer, E.L., 2005a. Kalign − an accurate and fast multiple sequence alignment algorithm. BMC Bioinf. 6, 289–298.

Lassmann, T., Sonnhammer, E.L., 2005b. Automatic assessment of alignment quality. Nucleic Acids Res. 33, 7120–7128.

Mansion, G., Parolly, G., Crowl, A., Mavrodiev, E., Cellinese, N., Oganesian, M., Fraunhofer, K., Kamari, G., Phitos, D., Haberle, R., Akaydin, G., Ikinci, N., Raus, T., Borsch, T., 2012. How to handle speciose clades? mass taxon-Sampling as a strategy towards illuminating the natural history of campanula (Campanuloideae). PLoS One 7, e50076.

Molina-Venegas, R., Roquet, C., 2014. Directional biases in phylogenetic structure quantification: a Mediterranean case study. Ecography 37, 572–580. http://dx.doi.org/10.2478/s11535-008-0035-4.

Molina-Venegas, R., Aparicio, A., Slingsby, J.A., Lavergne, S., Arroyo, J., 2015. Investigating the evolutionary assembly of a Mediterranean biodiversity hotspot: deep phylogenetic signal in the distribution of eudicots across elevational belts. J. Biogeogr. 42, 507–518. http://dx.doi.org/10.1111/jbi.12398.

Moore, M.J., Soltis, P.S., Bell, C.D., Burleigh, J.G., Soltis, D.E., 2010. Phylogenetic analysis of 83 plastid genes further resolves the early diversification of eudicots. Proc. Natl. Acad. Sci. U.S.A. 107, 4623–4628.

Mráz, P., Barabas, D., Lengyelová, L., Turis, P., Schmotzer, A., Janišová, M., Ronikier, M., 2016. Vascular plant endemism in the Western Carpathians: spatial patterns, environmental correlates and taxon traits. Biol. J. Linn. Soc. 119, 1095–8312.

Nagy, L., Grabherr, G., 2009. The Biology of Alpine Habitats. Oxford University Press, Oxford, pp. 37.

Ozenda, P. Masson Paris, Borel, J.-L., 2003. The alpine vegetation of the Alps. In: Nagy, L., Grabherr, G., Körner, C., Thompson, D.B.A. (Eds.), The Biology of Alpine Habitats. Oxford University Press, Oxford, pp. 53–63.

Pawlowski, B., 1970. Remarques sur l'endémisme dans la flore des Alpes et des Carpates. Vegetatio 21, 181–243.

Pigot, A.L., Etienne, R.S., 2015. A new dynamic null model for phylogenetic community structure. Eco. Lett. 18 1461–0248.

Pigot, A.L., Tobias, J.A., 2013. Species interactions constrain geographic range expansion over evolutionary time. Eco. Lett. 16, 330–338.

Plummer, M., 2003. JAGS: a program for analysis of bayesian graphical models using gibbs sampling. Proceedings of the 3rd International Workshop on Distributed Statistical Computing.

Plummer, M., 2016. Rjags: Bayesian Graphical Models Using MCMC R Package Version. pp. 4–6.

Core Team, R., 2016. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.

Rangel, T.F., Colwell, R.K., Graves, G.R., Fučíková, K., Rahbek, C., Diniz-Filho, J.A.F., 2015. Phylogenetic uncertainty revisited: implications for ecological analyses. Evolution 69, 1301–1312. http://dx.doi.org/10.1111/evo.12644.

Roquet, C., Boucher, F.C., Thuiller, W., Lavergne, S., 2013a. Replicated radiations of the alpine genus Androsace (Primulaceae) driven by range expansion and convergent key innovations. J. Biogeogr. 40, 1874–1886. http://dx.doi.org/10.1111/jbi.12135.

Roquet, C., Thuiller, W., Lavergne, S., 2013b. Building megaphylogenies for macroecology: taking up the Challenge. Ecography 36, 013–026.

Sanderson, M.J., 2003. r8s: inferring absolute rates of molecular evolution and divergence times in the absence of a molecular clock. Bioinformatics 19, 301–302.

Schönswetter, P., Stehlik, I., Holderegger, R., Tribsch, A., 2005. Molecular evidence for glacial refugia of mountain plants in the European Alps. Mol. Ecol. 14, 3547–3555. http://dx.doi.org/10.1111/j.1365-294X.2005.02683. x.

Smith, S.A., Beaulieu, J.M., 2010. An uncorrelated relaxed-clock anaysis suggests an earlier origin for flowering plants. Proc. Natl. Acad. Sci. U.S.A. 107, 5897–5902.

Stamatakis, A., Hoover, P., Rougemont, J., 2008. A rapid bootstrap algorithm for the RAxML web-Servers. Syst. Biol. 75, 758–771.

Stehlik, I., 2003. Resistance or emigration? Response of alpine plants to the ice ages. Taxon 52, 499–510. http://dx.doi.org/10.2307/3647448.

Stenseth, N.C., 1984. The tropics: cradle or museum? Oikos 43, 417.

Taberlet, P., Zimmermann, N.E., Englisch, T., Tribsch, A., Holderegger, R., Alvarez, N., Niklfeld, H., Coldea, G., Mirek, Z., Moilanen, A., Ahlmer, W., Marsan, P.A., Bona, E., Bovio, M., Choler, P., Cieslak, E., Colli, L., Cristea, V., Dalmas, J.P., Frajman, B., Garraud, L., Gaudeul, M., Gielly, L., Gutermann, W., Jogan, N., Kagalo, A.A., Korbecka, G., Küpfer, P., Lequette, B., Letz, D.R., Manel, S., Mansion, G., Marhold, K., Martini, F., Negrini, R., Nino, F., Paun, O., Pellecchia, M., Perico, G., Piekos-Mirkowa, H., Prosser, F., Puscas, M., Ronikier, M., Scheuerer, M., Schneeweiss, G.M., Schönswetter, P., Schratt-Ehrendorfer, L., Schüpfer, F., Selvaggi, A., Steinmann, K., Thiel-Egenter, C., van Loo, M., Winkler, M., Wohlgemuth, T., Wraber, T., Gugerli, F.,

2012. Genetic diversity in widespread species is not congruent with species richness in alpine plant communities. Ecol. Lett. 15, 1439–1448. http://dx.doi.org/10.1111/ele.12004.

Tribsch, A., Schönswetter, P., 2003. Refugia for mountain plants: patterns of endemism and comparative phylogeography confirm palaeo-environmental evidence in the Eastern European Alps. Taxon 52, 477–497. http://dx.doi.org/10.2307/3647447.

Tsirogiannis, K., Sandel, B., 2017. PhyloMeasures: Fast and Exact Algorithms for Computing Phylogenetic Biodiversity Measures. R Package Version 2.1.

Väre, H., Lampinen, R., Humphries, C., Williams, P., 2003. Taxonomic diversity of vascular plants in the European alpine areas. In: Nagy, L., Grabherr, G., Körner, C., Thompson, D.B.A. (Eds.), The Biology of Alpine Habitats. Oxford University Press, Oxford, pp. 133–147.

Warren, D.L., Cardillo, M., Rosauer, D.F., Bolnick, D.I., 2014. Mistaking geography for biology: inferring processes from species distributions. Trends Ecol. Evol. 29, 572–580. http://dx.doi.org/10.1016/j.tree.2014.08.003.

Webb, C.O., Ackerly, D.D., McPeek, M. a., Donoghue, M.J., 2002. Phylogenies and community ecology. Annu. Rev. Ecol. Syst. 33, 475–505. http://dx.doi.org/10.1146/annurev.ecolsys.33.010802.150448.

Wood, S.N., 2011. Fast stable restricted maximum likelihood and marginal likelihood estimation of semiparametric generalized linear models. J. R. Stat. Soc. Ser. B Stat. Methodol. 73, 3–36. http://dx.doi.org/10.1111/j.1467-9868.2010.00749.x.

de Candolle, A.P., 1875. Sur les causes de l'inégale distribution des plantes rares dans la chaîne des Alpes. Atti Del Congresso Internazionale Botanico Tenuto in Firenze 1875, 92–104.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 41–50

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Forum

Stay or go – how topographic complexity influences alpine plant population and community responses to climate change

Bente J. Graae[a,*,1], Vigdis Vandvik[b,1], W. Scott Armbruster[c], Wolf L. Eiserhardt[d,e], Jens-Christian Svenning[e,q], Kristoffer Hylander[f], Johan Ehrlén[f], James D.M Speed[g], Kari Klanderud[h], Kari Anne Bråthen[i], Ann Milbau[j], Øystein H. Opedal[k], Inger G. Alsos[l], Rasmus Ejrnæs[m], Hans Henrik Bruun[n], H. John B. Birks[b], Kristine B. Westergaard[o], Hilary H. Birks[b], Jonathan Lenoir[p]

[a] *Department of Biology, Norwegian University of Science and Technology, Norway*
[b] *Department of Biology, University of Bergen, Norway*
[c] *School of Biological Sciences, University of Portsmouth, United Kingdom, Institute of Arctic Biology, University of Alaska, USA*
[d] *Comparative Plant and Fungal Biology Department, Royal Botanic Gardens, Kew, United Kingdom*
[e] *Section for Ecoinformatics and Biodiversity, Department of Bioscience, Aarhus University, Ny Munkegade 114, DK–8000 Aarhus C, Denmark*
[f] *Dept. of Ecology, Environment and Plant Sciences, Stockholm University, Sweden*
[g] *NTNU University Museum, Norwegian University of Science and Technology, Norway*
[h] *Department of Ecology and Natural Resource Management, Norwegian University of Life Sciences, Norway*
[i] *Department of Arctic and Marine Biology, UiT – The Arctic University of Norway, Norway*
[j] *Research Institute for Nature and Forest INBO, Department of Biodiversity and Natural Environment, Belgium*
[k] *Centre for Biodiversity Dynamics, Department of Biology, Norwegian University of Science and Technology, Norway*
[l] *Tromsø Museum, University of Tromsø – The Arctic University of Norway, Norway*
[m] *Section for Biodiversity and Conservation, Department of Bioscience, Aarhus University, Denmark*
[n] *Department of Biology, University of Copenhagen, Denmark*
[o] *Norwegian Institute for Nature Research – NINA, Norway*
[p] *UR 'Ecologie et Dynamique des Systèmes Anthropisés' (EDYSAN, FRE 3498 CNRS-UPJV), Jules Verne University of Picardy, France*
[q] *Center for Biodiversity Dynamics in a Changing World (BIOCHANGE), Aarhus University, Ny Munkegade 114, DK–8000 Aarhus C, Denmark*

ARTICLE INFO

Keywords:
Adaptation
Dispersal
Microclimate
Niche
Refugia
Resilience

ABSTRACT

In the face of climate change, populations have two survival options − they can remain *in situ* and tolerate the new climatic conditions ("stay"), or they can move to track their climatic niches ("go"). For sessile and small-stature organisms like alpine plants, staying requires broad climatic tolerances, realized niche shifts due to changing biotic interactions, acclimation through plasticity, or rapid genetic adaptation. Going, in contrast, requires good dispersal and colonization capacities. Neither the magnitude of climate change experienced locally nor the capacities required for staying/going in response to climate change are constant across landscapes, and both aspects may be strongly affected by local microclimatic variation associated with topographic complexity. We combine ideas from population and community ecology to discuss the effects of topographic complexity in the landscape on the immediate "stay" or "go" opportunities of local populations and communities, and on the selective pressures that may have shaped the stay or go capacities of the species occupying contrasting landscapes. We demonstrate, using example landscapes of different topographical complexity, how species' thermal niches could be distributed across these landscapes, and how these, in turn, may affect many population and community ecological processes that are related to adaptation or dispersal. Focusing on treeless alpine or Arctic landscapes, where temperature is expected to be a strong determinant, our theorethical framework leads to the hypothesis that populations and communities of topographically complex (rough and patchy) landscapes should

---

[☆] This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author at: Department of Biology, Høgskoleringen 5, NO-7491 Trondheim, Norway.

[1] Graae and Vandvik contributed equally to the writing of the manuscript.

E-mail addresses: bente.j.graae@ntnu.no (B.J. Graae), Vigdis.Vandvik@uib.no (V. Vandvik), wsarmbruster@alaska.edu (W.S. Armbruster), W.Eiserhardt@kew.org (W.L. Eiserhardt), svenning@bios.au.dk (J.-C. Svenning), kristoffer.hylander@su.se (K. Hylander), Johan.Ehrlen@su.se (J. Ehrlén), James.Speed@ntnu.no (J.D.M. Speed), kari.klanderud@nmbu.no (K. Klanderud), kari.brathen@uit.no (K.A. Bråthen), ann.milbau@inbo.be (A. Milbau), oystein.h.opedal@ntnu.no (Ø.H. Opedal), inger.g.alsos@uit.no (I.G. Alsos), rasmus@bios.au.dk (R. Ejrnæs), hhbruun@bio.ku.dk (H.H. Bruun), John.Birks@uib.no (H.J.B. Birks), Kristine.Westergaard@nina.no (K.B. Westergaard), Hilary.Birks@uib.no (H.H. Birks), jonathan.lenoir@u-picardie.fr (J. Lenoir).

https://doi.org/10.1016/j.ppees.2017.09.008
Received 31 December 2016; Received in revised form 9 August 2017; Accepted 28 September 2017

be both more resistant and more resilient to climate change than those of topographically simple (flat and homogeneous) landscapes. Our theorethical framework further points to how meta-community dynamics such as mass effects in topographically complex landscapes and extinction lags in simple landscapes, may mask and delay the long-term outcomes of these landscape differences under rapidly changing climates.

1. Introduction

Our understanding of the magnitude and ecological implications of climatic variation in space and time has greatly developed over the past decades. Studies focusing on the capacity for species to track their climatic niches over large spatial extents, including palaeoecological reconstructions (reviewed in Feurdean et al., 2013) and climate envelope models (Pearson and Dawson, 2003; Sykes et al., 1996), typically suggest high migration rates in response to rapid redistribution of climates at the global scale (Clark et al., 1998; Loarie et al., 2009). For plants, these findings have been challenged by more recent molecular (Westergaard et al., 2010, 2011), palaeoecological (Birks and Willis, 2008; Cheddadi et al., 2014) and meso- to micro-scale climate envelope modelling (Franklin et al., 2013; Lenoir et al., 2017; Randin et al., 2009; Trivedi et al., 2008), all suggesting occasional species persistance within refugia or through short-distance escapes (Hampe and Jump 2011). Here, we discuss how the propensity for species to “stay”, through adaptation processes, or “go”, through dispersal processes, so as to survive in the face of climate change, depends not only on the magnitude of climate-change exposure and the climate-change sensitivity of the constituent species and communities (Dickinson et al., 2014; Bertrand et al., 2016), but also on the spatial structure of the landscapes in which the species occur (Körner, 2004; Slavich et al., 2014).

Topography is a key determinant of climatic variation across spatial scales ranging from regions, covering hundreds of square kilometres, to microsites of less than a square metre, especially in treeless areas like the high Arctic and alpine regions (see Box 1). Across these spatial scales, we can find regions, landscapes, patches, and microsites that are relatively topographically uniform, as well as others that are topographically complex, with associated differences in climatic heterogeneity. For example, there are clear differences in topographic complexity between mountainous *vs.* flat landscapes in high-latitude regions (Lenoir et al., 2013), flat areas *vs.* ridge-snowbed gradients in alpine landscapes (Graae et al., 2011; Körner, 2003), and flat *vs.* microtopographically complex patches within grassland and tundra vegetation (Armbruster et al., 2007; Moeslund et al., 2013; Opedal et al., 2015). The topographic complexity at scales of a few tens of metres can give rise to microclimatic variation in e.g., mean temperatures that often matches what is expected under future climate change scenarios (2–6 °C; Armbruster et al., 2007; Dobrowski et al., 2013; Graae et al., 2012; Lenoir et al., 2013; Opedal et al., 2015; Scherrer and Körner, 2010; Scherrer and Körner, 2011).

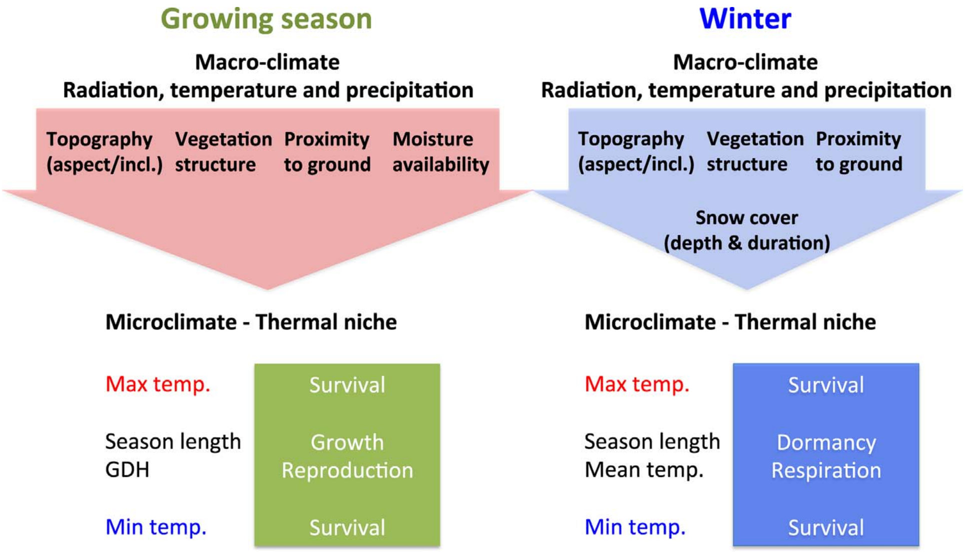
It is important to focus on high-latitude and high-elevation landscapes beyond treeline, not only because the complex topography there provides more spatial heterogeneity in temperature, but especially because temperature itself is expected to be the main determinant of plant distribution in these landscapes (Körner, 2003; Raunkiaer 1934). Indeed, temperature has both direct effects on alpine plant life through setting limits to species' fundamental niches, as well as indirect effects through determining, for instance, decomposition and nutrient cycling, access to water, as well as the abundance of herbivores, pathogens, pollinators, and seed dispersers. Some of these variables are also influenced by other factors – for instance anthropogenic disturbances and herbivore density that are often also regulated by humans. As Box 1 illustrates, we need to incorporate all these various components of temperature into the thermal niche concept of alpine plants. Hence, the thermal niche of an alpine plant species becomes a somewhat theoretical object for which one has to make the often unrealistic assumption of *ceteris paribus* (“other things being equal”). Though the realized niche for a species is difficult to describe because of the complexity of interacting limiting factors, there are good evidence for the existence of microclimatic niches (Lenoir et al., 2013; Scherrer and Körner 2011). In this paper we will make use of this theoretical niche concept, arguing that realised microclimatic niches are important for plants and improving our understanding of the distribution of these thermal niches across the landscape is important for predicting species' capacities to adapt or disperse in response to changing climate.

Here, we synthesise theories relevant for how the topographic complexity of landscapes at high elevations or latitudes influences the

Box 1
The thermal niche of alpine plants.

The thermal niche of plants is often described in a highly simplified manner with a strong focus on synoptic or ambient air temperature characterizing macroclimate. For small-stature and slow-growing alpine and Arctic plants there is a major difference and decoupling between the temperature that the plants experience near the ground and the temperature conditions obtained from weather stations measuring synoptic temperature at 2 m height (Graae et al., 2012; Lenoir et al., 2013; Körner, 2003; Scherrer and Körner, 2010, 2011). During summer, the difference and decoupling between temperature conditions near the ground and synoptic temperature is to a high degree controlled by topography, vegetation structure, proximity to ground and, in the soil, also the moisture level. During winter, difference and decoupling is also caused by topography, vegetation structure and proximity to ground, but this is mostly due to its effect on snow cover and depth that determines the microclimate (temperature and moisture) and light conditions to the plants. Körner (2003) as well as Wipf and Rixen (2010) describes in detail how snow cover and duration matters for alpine and Arctic vegetation.

In addition to these scale effects, it is well established that the multifaceted nature of temperature (maximum, minimum, mean, growing season length, etc.) affects different life cycle and phenological stages to various extent. For instance, extreme temperatures are mostly associated with mortality events and the timing of these extreme events is crucial, whereas mean temperatures are chiefly associated with growth processes. Körner et al. (2016) describe how the many different components of climate affect tree distribution, and this complexity of niche limiting factors and interactions is expected to be even greater for small-stature plants occurring near the ground. Understanding the ecophysiological and ecological mechanisms underlying plant species distribution needs to take such microclimatic considerations into account. Accounting for all these limiting factors to model alpine plant species distribution is rarely done in the scientific literature. The more simplified concept of thermal niche has, however, shown useful because plant species distribution, especially trees for which most studies are done, correlate well with macroclimatic variables such as mean summer and winter temperatures. However, for mechanistic understanding of what is driving these correlations we need to go beyond mean temperatures (Körner et al., 2016) and assess the importance of this topographically-driven heterogeneity in temperature conditions near the ground and its consequences for alpine plant distribution and redistribution under climate change.



Legend: Diagram showing the many factors shaping the microclimatic niche in alpine plant communities. The growing season macroclimate is filtered into microclimate by factors listed in the red arrow and winter macroclimate is filtered into microclimate by factors listed in the blue arrow. The resulting summer microclimatic niche in green for survival, growth and reproduction of plants are determined by temperature extremes (max. and min. temperatures mostly for survival), season length and growing degree hours (GDH) that gives the accumulated temperature for growth and reproduction. Also the winter microclimatic niche in blue is determined by the temperature extremes for survival while season length, that also to a high extent is driven by snow cover, determines important winter processes, for instance respiration and dormancy break.

resistance (the lack of sensitivity and response to perturbation or disturbance) and resilience (the capacity to recover after perturbation or disturbance) of alpine plant populations and communities in response to climate change. Specifically, we explore (1) how populations in landscapes of different topographical complexity are affected by microclimatic heterogeneity under the current climate, and (2) how this may affect their responses to climate change. We then turn to communities, and (3) develop a framework for community response to landscape microclimatic heterogeneity, before we (4) ask how this may affect community-level responses to climate change in landscapes of different topographic complexity. Although we focus on small-stature plants in cold ecosystems, many of the processes we describe here would hold for other groups of organisms, with modifications to account for differences in organism mobility and scale (Roth et al., 2014), as well as other important factors of specific relevance. For example, for small-stature plants in the lowlands, one would also have to consider, in addition to topography, the effect that tree or shrub canopy cover exerts on microclimate (De Frenne et al., 2013; Grimmond et al., 2000) and its consequences for forest plant species distribution (e.g. Wesser and Armbruster 1991; Lenoir et al., 2017).

2. The spatial components of microclimatic niche heterogeneity

Microclimatic heterogeneity affects populations and communities in two general ways. First, by increasing the range of climatic conditions, it increases the climatic niche space that is available within a given surface area and creates potential niche space for more species. At the same time, this inevitably comes at the expense of reduced available habitat area (Kerr and Packer, 1997; Scherrer and Körner, 2011) and thus increases habitat fragmentation (Reino et al., 2013) for species with specialised thermal niches. In alpine and Arctic ecosystems, topography is the main physiographic feature that can enhance microclimatic heterogeneity in space. As a general and simplified example, consider seven hypothetical landscapes of equal size (e.g., 1 km^2), sharing a regional plant species pool (Zobel, 1997), but varying in topographic complexity (Fig. 1). Microclimatic heterogeneity due to topographic complexity has two dimensions: the range of climatic conditions available (increasing from left to right in Fig. 1), and the climatic patchiness or fragmentation (increasing from top to bottom in Fig. 1). The species in the regional pool will be distributed differently among and within the landscapes, depending on niche availability and landscape heterogeneity (in Fig. 1, species are represented by rings and curves of different colours). In addition to experiencing long-term changes in climatic conditions, our hypothetical landscapes can experience different levels of disturbance and seasonal fluctuation, which will naturally influence the population and community dynamics of the plants inhabiting these landscapes. Additionally, alpine plant species have different life histories, sizes and dispersal capacities, involving different spatial scales (cf. the spatial extent and resolution of our hypothetical landscapes). Here, we only focus on the spatial arrangement of microclimatic conditions across a 1-km^2 landscape and the impact of climate change on the “stay” or “go” processes.

Our first example landscape, L0, is climatically homogeneous, with a narrow range of climatic conditions (or niche space), such as can be found for temperature across a flat and smooth landscape. For species whose realised climatic niche requirements are fulfilled in this landscape (species represented by the brown and green curves and rings in Fig. 1), a large and continuous habitat area is available, and the probability of temperature-related local extinction under stable conditions is hence low (Hanski, 1998; Lande, 1993; MacArthur and Wilson, 1963). L1 and L2 encompass increasingly wider ranges of thermal conditions that are distributed in a non-patchy way (positive spatial autocorrelation) such as on a gentle (L1) or a steep (L2) hillside. The available niche space, and hence the potential number of species in the landscape increases from L0 via L1 to L2 (Fig. 1). Depending on the species’ niche width and the climatic niche availability, species may occur in the whole or in parts of the landscape, and there is, for most species, less habitable area available in L2 than in L1 and L0.

L3 has the same available thermal niche space for any given species as L1, but suitable areas for each species are more patchily distributed in space, and the populations will therefore tend to be more spatially scattered, as in a hilly landscape with fine-scale topographic complexity. Landscapes L2 and L4 have the same difference in patchiness as between L1 and L3 but with a larger climatic range. Thus, L3 and L4 have, on

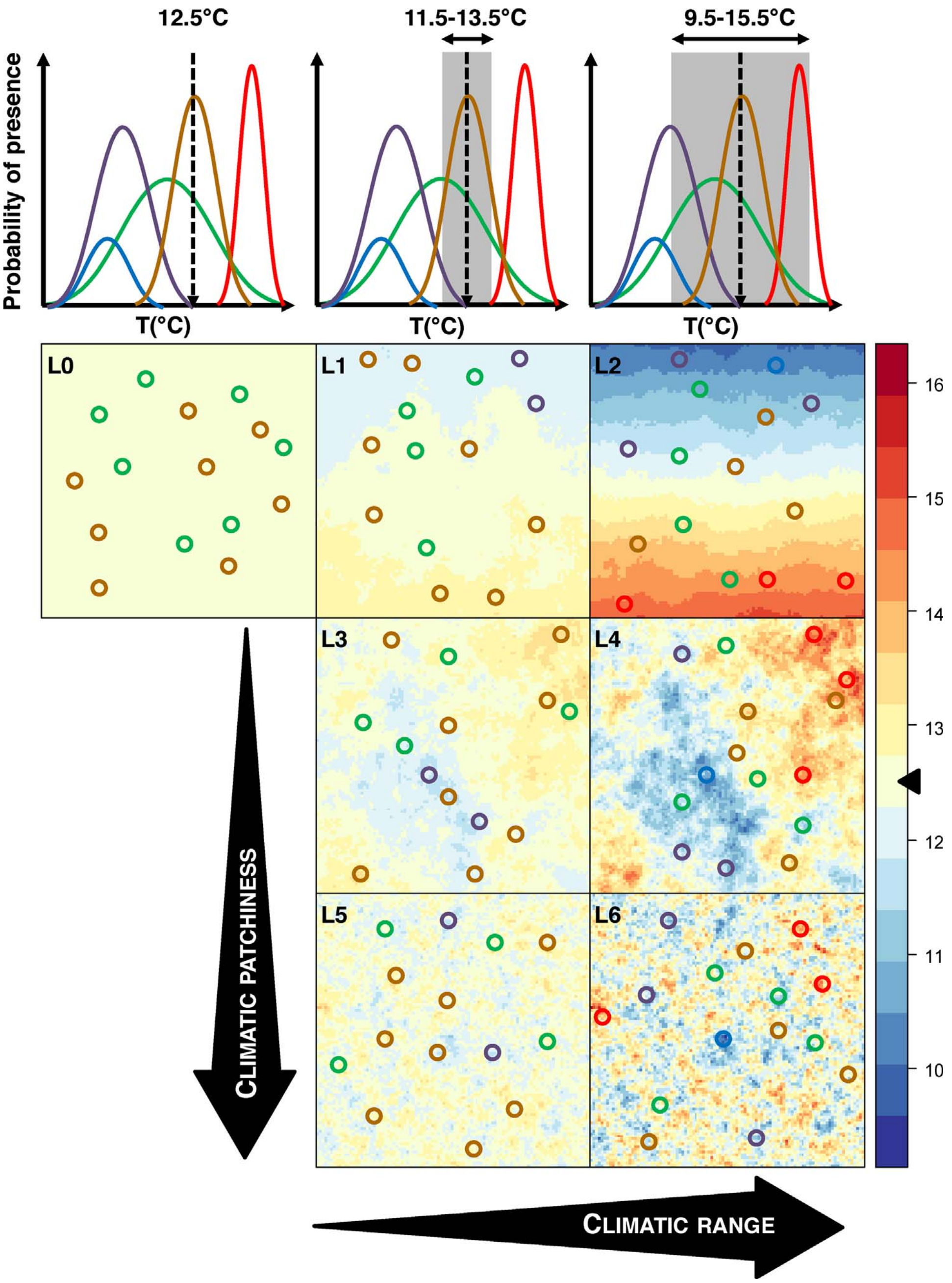


Fig. 1. Species response curves along a temperature gradient (upper panel) and the species' spatial distributions (lower panels) across seven landscapes differing in two important determinants of climatic heterogeneity; the range of climatic conditions available (cf. increasing climatic range from left to right) and the degree of fragmentation in climatic conditions (cf. increasing climatic patchiness from top to bottom). The available climate within the landscapes in each coloumn is represented by a vertical dotted line [mean] and a grey box [range] in the upper panel. For illustrative purpose, a theoretical species pool is provided, containing five different species (represented by colours) with different climatic niches (upper panel). Each of the seven (L0-L6) landscape panels gives exemplified spatial distribution of the climatic niche space (colour scale from cold to warm) and of local populations of the species in the species pool (coloured rings). Note that the mean temperature is similar across all seven landscapes – illustrated by the black triangle on the key to the right.

average, smaller patches, but also shorter distances between patches of suitable habitat, compared to landscapes L1 and L2, respectively. Landscapes L5 and L6 are even more fragmented, to the extent that they may appear quasi-homogeneous.

Below, we evaluate how the increasing landscape heterogeneity affects the populations and communities inhabiting these different landscapes. We outline the consequences of this landscape heterogeneity for the selective pressures within the different landscapes, and for how the populations and communities are equipped to respond to climate change.

3. Populations in landscapes of varying climatic heterogeneity

For species with narrow thermal niches and/or only occurring in part of the climatic range of the landscape (i.e. purple species in L1, L3 and L5 and blue species in L2, L4 and L6), populations will be smaller in size and/or more fragmented in space going from L0 towards L6. This may lead to higher local extinction rates due to stochastic processes in the smaller populations of fragmented landscapes (Fig. 1). However, when moving from L3 to L5 or from L4 to L6 the existence of many small patches will reduce the average distance between patches of suitable habitat in these landscapes, potentially improving connectivity between the fragmented populations and reducing extinction risks via rescue effects (Brown and Kodric-Brown, 1977; Hanski, 1998). Note that this potential increase in connectivity can only happen if the average dispersal distance of the focal plant species within the landscape exceeds the average distance between patches of suitable habitat (i.e., the patches are part of a population or metapopulation, *sensu* Hanski, 1998). In our example with a fixed sized landscape window, the balance between extinctions, caused by reduced patch sizes, and colonisations, caused by reduced distances between the patches and by the area-related colonization capacity, will depend on the organisms' life history. Small sized and well-dispersed plant species will most likely be less affected by decreasing habitat sizes and increasing isolation than plant species with high area requirement or more limited colonisation capacities.

Interestingly, the population processes in climatically variable and patchy landscapes, like L6, may converge towards the situation in homogeneous landscapes such as L0 if distances are so small that individuals can easily move between patches so that populations are no longer fragmented. However, in contrast to L0, highly heterogeneous landscapes as in L6 may allow populations with different niche requirements to coexist, as long as the patch area across the landscape is still large enough for populations to survive locally. Therefore, the constraint due to dispersal limitation towards a climatically suitable location may become less important towards both L6 (i.e. similar microclimates can be very close) and L0 (i.e. homogeneous microclimatic conditions), and may be most important under intermediate microclimatic heterogeneity (relative to the organism under study).

The microclimatic heterogeneity in the landscape will also alter the selective forces acting on populations in the different landscape types. Populations inhabiting climatically heterogeneous landscapes may be under selection for broader niches in order to maintain sustainable population sizes in a heterogeneous environment. When microclimatic heterogeneity increases, either moving from left to right or top to bottom in Fig. 1, species with broad climatic niches, represented by the green curve, will have an advantage compared to the species with narrower niches. Selection for broader niches may result in greater phenotypic plasticity within local populations, with important consequences for the capacity of these populations to respond to environmental changes (Chevin et al., 2010; Jump and Peñuelas, 2005; Nicotra et al., 2010). Whether selection in response to environmental heterogeneity favours phenotypic plasticity, genetic differentiation, or a combination of the two, depends on several factors, including the temporal and spatial scale of climatic variation (Alpert and Simms, 2002; Botero et al., 2015). More effective dispersal in space or time may also counteract the negative effects of fragmentation. Populations in fragmented landscapes may therefore also be under selection towards better dispersal abilities, or they may be under selection towards better survival in dormant or other long-lived stages, thus contributing to extinction time-lags, until opportunities for continued growth and reproduction (re)appear locally (dispersal in time or remnant population strategy *sensu* Eriksson, 1996).

4. Consequences of microclimatic heterogeneity for populations under changing climate

Populations in different landscape types, such as topographically simple *vs.* complex terrains, may be very differently positioned, and also equipped, to meet ecological challenges of climate change. In a climatically homogeneous landscape, like L0, populations can remain within the landscape if they tolerate the new climatic conditions, either through intrinsic ability of individuals to tolerate changing climatic conditions (Bertrand et al., 2016), or through intraspecific variation in the position of the climatic niche optima (Valladares et al., 2014). Alpine plant species within the homogeneous landscapes are expected to have rather narrow niches, but those with the widest climatic niches, represented by the green curve in Fig. 1, will have the highest chance of surviving in this type of landscape and adapting to the new climatic conditions through realised niche shifts (Wasof et al., 2013, 2015). Indeed, in L0, distances to new suitable habitats might be relatively large (i.e., somewhere outside the landscape), thus favouring adaptation ("stay") processes over dispersal and colonisation ("go") processes. In addition, low immigration rates into patches in these landscapes (i.e., long distance to source populations of species with different climatic optima) means that the resident species will have a relatively low risk of being exposed to competition from immigrant species better adapted to the new climate (Ackerly, 2003; Bertrand et al., 2011). This may allow persistence under a new suboptimal climate and hence a longer time during which adaptation to the new climate can occur (Ackerly, 2003; Svenning and Sandel, 2013). Species with high persistence capacity, for instance with very long-lived individuals or dormant stages, may remain for extended periods in this type of landscape (L0) compared to what would be predicted from their climatic niche (Eriksson, 1996, 2000; May et al., 2009; Migliore et al., 2013), contributing to the extinction debt (Tilman, 1994). Related to this, "staying" may also be possible through expansion of the realised niche to encompass the new climate, for example due to changes in biotic interactions (e.g. competitive release (Lenoir et al., 2010)).

In contrast, populations experiencing changing climate in more heterogeneous landscapes (to the right or down in Fig. 1) are more likely to

Box 2
The meta-community paradigm.

The meta-community paradigm defines a meta-community as a set of local communities, linked by dispersal, and describes how the dynamics of the meta-community at large is driven by the interactive effects of local niche processes operating within each patch, and by dispersal between patches (Leibold et al., 2004). **Four general and non-mutually exclusive perspectives** on meta-community dynamics are typically recognized:

- **Neutral processes** assume that all species within a trophic level and all patches are functionally equivalent and coexistence is permitted by stochastic processes and slow competitive exclusion relative to immigration and evolutionary rates (Hubbell, 2001).
- **Patch dynamics** models describe a system where coexistence is permitted by a trade-off between dispersal and competitive ability, so that the most successful colonizers of available patches are relatively poor competitors, and vice versa (Levins and Culver, 1971; Tilman, 1994).
- **Species sorting** models assume an environmentally heterogeneous environment and consider how species' niche requirements 'sort' them into local communities (Chase and Leibold, 2003; Whittaker, 1962).
- **Mass effects** models build on species sorting, but with the added feature that dispersal between communities may allow maintenance of local 'sink' populations also in sites where the niche requirements of that species are not met (Holt, 1993; Mouquet and Loreau, 2003).

In the past decade, the meta-community paradigm has been highly influential in setting the research agenda in community ecology, and it has inspired a substantial literature on the interplay between dispersal and niche processes, covering a wide range of spatial and temporal scales, biomes, and organism groups, and giving rise to both theoretical, observational and experimental advances (Leibold et al., 2010; Murphy and Foster, 2014; Myers and Harms, 2009; Pillar and Duarte, 2010; Telford et al., 2006; Tscharntke et al., 2012; Urban et al., 2008). However, questions of the relative importance of the different meta-community processes in determining the patterns in community composition we observe in nature, and indeed how and if the relative importance of these processes can even be quantitatively assessed, have been highly debated and are far from being resolved (e.g., Logue et al., 2011).

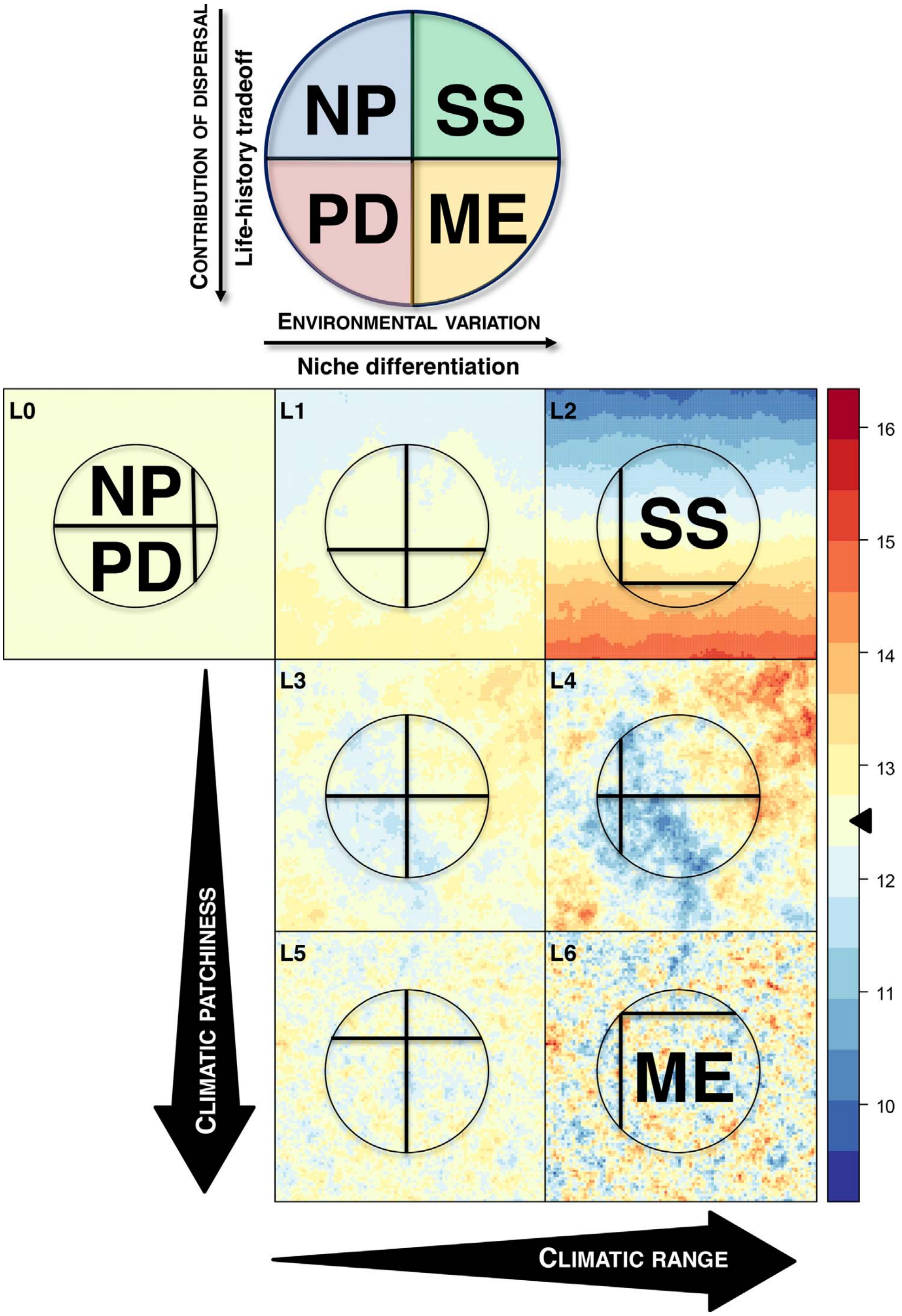


Fig. 2. Prediction of the relative importance of different meta-community dynamics (Neutral processes NP, Patch dynamics PD, Species sorting SS and Mass effects ME (see Box 2 for explanation)) (upper panel) in response to the climatic heterogeneity in the landscape (lower panels). The seven landscapes are the same as in Fig. 1.

have a suitable microclimate patch nearby. At the same time, these populations are likely to have been under selection for better dispersal capacity and wider niches because they have been exposed to such. The populations remaining in these landscapes should thus be better equipped to stay within the landscape. In L1-L6, in contrast to L0, for which species have to migrate outside the landscape if they cannot adapt locally, species can move across the landscape to track the climatic change. Species may go extinct within the landscape if (i) dispersal distances to track the species' niche exceed the species' dispersal capacity, (ii) the available habitat area within the landscape becomes too small to support a viable (meta-) population or (iii) the species' climatic niche is no longer available within the landscape (e.g., very cold-adapted species represented by the blue curve). In L1 the risk of colonisation time-lags and extinctions is expected to be higher than in L2 but this will depend heavily on species climatic tolerance, dispersal capacity and life-history traits (Alsos et al., 2012, 2015; Bertrand et al., 2011; Lenoir et al., 2008).

The average dispersal distance required to track a given climate change within the landscape window decreases from L1 via L3 to L5, requiring successively smaller dispersal capacity for survival. L4 and L6 will offer even better opportunities to disperse between patches under dramatic climate changes, even for dispersal-limited species. There is a high probability of encountering a patch nearby with suitable microclimate unless the microclimatic niche has vanished for that species (i.e. the species represented by blue and purple curves in Fig. 1 may loose their niches after warming), resulting in low dispersal limitation-related extinction rates and short time-lags. New neighbours will colonise at a faster rate. We expect that such rapid changes will pose challenges for species with slow life histories (cf. long-lived species with limited colonisation capacity) (Lenoir and Svenning, 2013; Tscharntke et al., 2012; Vranckx et al., 2012), and they will rely more on their ability to tolerate climate changes (De Witte and Stöcklin, 2010). The more fragmented landscapes, however, will also encompass smaller and more fragmented populations that may be more vulnerable to climatic fluctuations.

Our example landscapes illustrate how the adaptations resulting from the selective pressures that have been shaping the populations inhabiting homogeneous versus heterogeneous landscapes may be the opposite of the adaptations populations will need to survive in those landscapes under a rapid climate change. Populations in homogeneous landscapes have been under selection for traits allowing them to persist under rather homogeneous conditions, but may, in the face of climate change, be required to migrate over large distances (outside the landscape) if they cannot tolerate or adapt to the new conditions. In contrast, populations inhabiting heterogeneous landscapes have better opportunities to “stay” within their landscape throughout short-distance displacements and yet are also better adapted to disperse over longer distances and establish in a wider range of conditions due to historical selection pressures towards better dispersal and wider niches.

5. Communities in landscapes of varying climatic heterogeneity

Landscape structure and the associated differences in climatic range and patchiness will have consequences for community-level processes in the landscape (Tscharntke et al., 2012). The meta-community paradigm (Box 2), as described by Chesson (2000) and Leibold et al. (2004), is a useful starting point for exploring these implications. Here we assume that meta-community dynamics are driven to various degrees by neutral processes, patch dynamics, species sorting, and mass effects (Leibold et al., 2004). Climatically homogeneous landscapes, as exemplified by L0, are not likely to support communities in which climate niche-based processes, such as species sorting or mass effects, play important roles in maintaining species diversity (Fig. 2). There is no climate-driven habitat variation, and the populations that inhabit these landscapes share the same climate niche (see above). The total suitable habitat area is large and climatically homogeneous, which will increase the probability of community assembly based on either neutral processes, where the co-existence results from the very slow stochastic extinction rates of demographically equivalent species within a relatively large population area, or patch dynamics, with species co-existence permitted by a trade-off between dispersal and competitive abilities (Fig. 2).

In contrast, landscapes encompassing a wider range of climatic conditions (L1-L6) have more climate niche space available and there is scope for coexistence based on climate niche partitioning and hence for species sorting and/or mass effects to operate (Fig. 2). The climatic range is equal for all landscapes at the same position along the climate range gradient (for L1, L3, and L5 or for L2, L4, and L6), and the total area of suitable microclimate for any particular species is therefore also equal for the landscapes within each of these columns. It follows that climatic niche-partitioning processes (i.e., species sorting and/or mass effects) is likely to be intermediately important across L1, L3, and L5, and of overriding importance across L2, L4 and L6. As we move from L0 via L1 to L2, the average habitat area available for each species decreases, but for each species the available area is not fragmented (high auto-correlation), leading to an overall decrease in the relative contribution of dispersal to community dynamics.

Towards the lower parts of Fig. 2, both the average patch size of suitable habitats and the dispersal distance between patches decreases, leading to increased probabilities of both local extinction and re-colonisation of locally-extinct populations. The climate gradient length is equal within each column (e.g., L2, L4 and L6) and the importance of climatic niche-partitioning processes (the combined effect of species sorting and mass effects) is hence constant. However, with increased fragmentation, the probability that a dispersed propagule ends up in a ’sink’ population increases, and the relative importance of mass effects is therefore expected to increase at the cost of efficient species sorting (Fig. 2). In L6, however, the decrease in dispersal distances between patches might be so important that, for some species, the landscape is perceived as more homogeneous than L3 and L4. Hence, neutral dynamics could be expected to operate, but within several “parallel communities” each consisting of few species with very specific climatic tolerances. Mass effects are then occurring between these parallel communities, causing all the species to seemingly coexist in the same landscape.

6. Consequences of microclimatic heterogeneity for communities under climate change

In large homogeneous landscapes where diversity is maintained by neutral and patch-dynamics processes, such as L0, there is little climate niche variation among species. Under climate change, persistence due to shifts in species’ realised niche is possible as long as the climate change is within the fundamental niche limits of the species (Lenoir and Svenning, 2015). As the current climate no longer overlaps with the fundamental climatic niches of many of the species in the community, extinction rates are likely to increase sharply, and the ensuing gaps will mostly receive non-suitable recruits. This will result in unsaturated communities, probably with decreased levels of interspecific competitive interactions, which could lead to shifts or expansion of realised niches (cf. Lenoir et al., 2010) and increased probability of persistence for the remaining species. Long-term maintenance of biodiversity and ecological functions in such landscapes will require local extinctions and immigration, and hence remnant population dynamics (Eriksson, 1996, 2000), storage effects (Chesson and Warner, 1981) and dispersal limitation on long-distance dispersal from outside the landscape will result in severe time-lags. Such communities may exhibit considerable unpaid extinction debts (Jackson and Sax, 2010; Kuussaari et al., 2009), as species sorting processes will be inefficient in increasingly unsaturated communities consisting of species poorly adapted to the new climatic conditions. On the other hand, when individuals dispersing in from outside the landscape do eventually arrive, these unsaturated communities are likely to be readily invasible (colonisable) and new species with good dispersal and establishment capacities are likely to be favoured. We therefore expect communities in homogeneous landscapes to experience relatively slow species loss, and low levels of landscape-scale reshuffling over time (cf. time lag and climatic debt, *sensu* Bertrand et al., 2016). In the long term and with dramatic climate change exceeding the tipping point, we expect greater proportional species loss (climatic debt being paid off) here than in heterogeneous landscapes.

In heterogeneous landscapes (L1-L6), climate change is likely to result in species displacement along the climatic gradient, with direction and rate of the realized community change shaped by the interplay between local dispersal and species-sorting processes operating within the landscape (i.e., paralleling the processes operating in the landscape under a stable climate; Fig. 2). In general terms, landscapes with broader climatic ranges will have smaller available habitat area for any given climatic regime and hence higher extinction probability under climate change compared to more homogeneous landscapes. However, the finer-grained spatial heterogeneity of patchy landscapes implies, on average, that a broader range of climatic conditions are available within a given distance from any particular point in the landscape, and hence an influx to patches of species with a broad range of climatic-niche requirements. As discussed above, a species pool adapted to survival in a fragmented landscape may also be better equipped for dispersal within the landscape. The net effect is less dispersal limitation, shorter establishment time-lags, and faster equilibration of the communities to new climatic conditions in patchy compared to homogeneous landscapes. Only the warmest patches may experience problems getting new species from within the landscape. However, at the same time, good dispersal abilities coupled with greater proximity between different habitat types will also result in greater impacts of mass effects on communities within these heterogeneous landscapes (Fig. 2). These mass effects will tend to delay the realized community change in response to climate change in patchy landscapes. Indeed, as long as a sufficient number of source populations are still available within the landscape, communities may appear resistant to climate change (Fig. 2). These contrasting effects of niche availability and patchiness on metacommunity processes within the landscapes thus predicts better climatic-niche tracking across intermediate landscapes (L1 to L5), with shorter time-lags here than in less (L0) or more (L6) fragmented landscapes where greater tolerances to climate change and greater mass effects, respectively, delay community turnover in species composition.

The shift in relative importance of underlying meta-community processes (from neutral processes and patch dynamics via species sorting to mass effects; Fig. 2) as well as the differences in selective pressures (increasing dispersal ability, Fig. 1) may be instrumental in driving differences in community-level response along the gradient from homogeneous to heterogeneous landscapes. At the same time, these same processes (notably, the mass effects) will tend to delay the change in underlying community dynamics in heterogeneous landscapes, resulting in an apparent resistance to climate change.

7. The impact of temporal variation, and non-climatic confounding factors

In addition to the general framework discussed above, other aspects of scale, temporal climatic variation, other niche requirements, biotic interactions, and disturbance will affect populations and communities under climate change. First, climatic heterogeneity varies in time as well as in space, and this also shapes the characteristics of populations and communities, and we may, for example, expect populations and communities with a history of exposure to strong temporal climatic variation due to seasonality or recurring extreme events to cope better with climate changes compared to landscapes in regions with less variable weather and climate. Second, biotic interactions can modify both microclimate and the ability of species to track their climate (Leathwick and Austin, 2001; Wisz et al., 2013). For instance, species colonisation rates may be enhanced by facilitation (Anthelme et al., 2014) or by zoochory (Cunze et al., 2013), and they may be delayed by interference (Pellissier et al., 2010). The strength of biotic interactions are however themselves often dependent on climate (Pellissier et al., 2013; Anthelme et al., 2014; Alexander et al., 2015; Olsen et al., 2016), and may therefore also enforce processes determined by landscape heterogeneity.

The rate and magnitude of climate change will partly determine the need for adaptation or required dispersal capacity for climate tracking (Sandel et al., 2011), and the disturbance frequency in a landscape, whether topographically homogeneous or heterogeneous, also imposes selective pressures on the species. Disturbance creates additional temporal and spatial heterogeneity in plant populations and communities, imposes distinct selective pressures (Tscharntke et al., 2012), and interacts with community dynamics (Levins and Culver, 1971; Tilman, 1994). Communities dominated by disturbance-adapted species will hence change faster than communities dominated by more stress-tolerant or competitive species (*sensu* Grime, 2001). This is not only because the species in the landscape are adapted to rapid changes, but also because the landscape itself will likely be subjected to disturbance in the future providing gaps in the vegetation for new colonisations (Vandvik and Goldberg, 2005, 2006). Many areas with high disturbance are associated with intense use by human or other animals and are often found in flat areas. Therefore, disturbance may cause topographically homogeneous landscapes to change faster than expected from the microclimatic variation patterns outlined above.

8. Conclusion

A growing number of studies points to the importance of landscape topography in modifying the rate of change in populations and communities (Tscharntke et al., 2012). For instance, findings from Bertrand et al. (2011) suggest that the extinction debt in forest plant communities is much more important in the lowlands than in the highlands in France. Spasojevic et al. (2013) and De Frenne et al. (2013) however, showed that alpine and forest plant communities are dynamic through time when studied at a fine spatial scale, even for the species distributions that on a broad scale seem to show extinction debts (Bertrand et al., 2011). We argue that landscapes with high microclimatic heterogeneity will contain populations and communities that have better opportunities for coping with climate change than those of climatically more homogeneous landscapes. However, at the same time, the characteristics of populations in heterogeneous landscapes may also compromise the monitoring of species-environment relationships, due to mass effects. In contrast, populations and communities of climatically more homogeneous landscapes may be relatively more vulnerable to climate change as they can only persist in the long run if they adapt to the new environment, if their realised niches are relaxed, or if they persist through extreme longevity and remnant populations. Nevertheless, lower immigration rates and less-saturated communities may provide opportunities for niche expansion and rapid evolution in homogeneous landscapes under a changing climate. Species and communities in homogeneous landscapes may therefore be more resistant to climate change than predicted solely from the current realised niches of the species and the current community dynamics.

To improve our understanding of population and community responses to climatic change, future studies need to consider the microclimatic heterogeneity of the landscapes in which the species are found and the selective pressures that may have shaped the populations and communities in these landscapes. We here introduced a very simplified theorethical framework to illustrate how the spatial patterns in microclimatic range and patchiness, closely associated to the various effects of topography and variables outlined in Box 1, may affect alpine community dynamics in response to climate change. Synthesis and tests of the importance of temporal climatic variation for the capacity for persistence or migration of populations and communities are also needed. Considering the microclimatic heterogeneity driven by topographic complexity in Arctic and alpine ecosystems may help us better understand the resistance and resilience of populations and communities to changing climate.

Acknowledgments

This paper is the outcome of discussions during network meetings of the Stay or Go network funded by Nordforsk from 2011-2013 (Project number 29662 to BJG). JCS was supported by the European Research Council (ERC-2012-StG-310886-HISTFUNC) and also considers this work a contribution to his VILLUM Investigator project (VILLUM FONDEN, grant 16549).

References

Ackerly, D.D., 2003. Community assembly, niche conservatism, and adaptive evolution in changing environments. Int. J. Plant Sci. 164, S165–S184.

Alexander, J.M., Diez, J.M., Levine, J.M., 2015. Novel competitors shape species' responses to climate change. Nature 525, 515–518.

Alpert, P., Simms, E.L., 2002. The relative advantages of plasticity and fixity in different environments: when is it good for a plant to adjust? Evol. Ecol. 16, 285–297.

Alsos, I.G., Ehrich, D., Thuiller, W., Eidesen, P.B., Tribsch, A., Schonswetter, P., Lagaye, C., Taberlet, P., Brochmann, C., 2012. Genetic consequences of climate change for northern plants. Proc. Royal Soc. B: Biol. Sci. 279, 2042–2051.

Alsos, I.G., Ehrich, D., Eidesen, P.B., Solstad, H., Westergaard, K.B., Schonswetter, P., Tribsch, A., Birkeland, S., Elven, R., Brochmann, C., 2015. Long-distance plant dispersal to North Atlantic islands: colonization routes and founder effect. AoB Plants 7.

Anthelme, F., Cavieres, L.A., Dangles, O., 2014. Facilitation among plants in alpine environments in the face of climate change. Front. Plant Sci. 5. http://dx.doi.org/10.3389/fpls.2014.00387.

Armbruster, W.S., Rae, D., Edwards, M.E., 2007. Topographic complexity and biotic response to high-latitude climate change: variance is as important as the mean. In: Ørbæk, J.B., Kallenborn, R., Tombre, I., Hegseth, E.N., Falk-Petersen, S., Hoel, A.H. (Eds.), Arctic-Alpine Ecosystems and People in a Changing Environment. Springer Verlag, pp. 105–122.

Bertrand, R., Lenoir, J., Piedallu, C., Riofrio-Dillon, G., de Ruffray, P., Vidal, C., Pierrat, J.C., Gegout, J.C., 2011. Changes in plant community composition lag behind climate warming in lowland forests. Nature 479, 517–520.

Bertrand, R., Riofrío-Dillon, G., Lenoir, J., Drapier, J., de Ruffray, P., Gégout, J.C., Loreau, M., 2016. Ecological constraints increase the climatic debt in forests. Nat. Comm. http://dx.doi.org/10.1038/ncomms12643.

Birks, H.J.B., Willis, K.J., 2008. Alpines, trees, and refugia in Europe. Plant Ecol. Divers. 1, 147–160.

Botero, C.A., Weissing, F.J., Wright, J., Rubenstein, D.R., 2015. Evolutionary tipping points in the capacity to adapt to environmental change. Proc. Natl. Acad. Sci. U. S. A. 112, 184–189.

Brown, J.H., Kodric-Brown, A., 1977. Turnover rates in insular biogeography – effect of immigration on extinction. Ecology 58, 445–449.

Chase, J.M., Leibold, M.A., 2003. Ecological Niches. University of Chicago Press, Chicago, IL.

Cheddadi, R., Birks, H.J.B., Tarroso, P., Liepelt, S., Gomory, D., Dullinger, S., Meier, E.S., Hulber, K., Maiorano, L., Laborde, H., 2014. Revisiting tree-migration rates: *Abies alba* (Mill.) a case study. Veg. Hist. Archaeobot. 23, 113–122.

Chesson, P.L., Warner, R.R., 1981. Environmental variability promotes coexistence in lottery competitive-systems. Am. Nat. 117, 923–943.

Chesson, P., 2000. Mechanisms of maintenance of species diversity. Annu. Rev. Ecol. Syst. 31, 343–366.

Chevin, L.M., Lande, R., Mace, G.M., 2010. Adaptation, plasticity, and extinction in a changing environment: towards a predictive theory. PLoS Biol. 8.

Clark, J.S., Fastie, C., Hurtt, G., Jackson, S.T., Johnson, C., King, G.A., Lewis, M., Lynch, J., Pacala, S., Prentice, C., Schupp, E.W., Webb, T., Wyckoff, P., 1998. Reid's paradox of rapid plant migration – dispersal theory and interpretation of paleoecological records. Bioscience 48, 13–24.

Cunze, A., Heidel, F., Tackenberg, O., 2013. Are plant species able to keep pace with the rapidly changing climate? PLoS One 8. http://dx.doi.org/10.1371/journal.pone.0067909.

De Frenne, P., Rodriguez-Sanchez, F., Coomes, D.A., Baeten, L., Verstraeten, G., Vellend, M., Bernhardt-Romermann, M., Brown, C.D., Brunet, J., Cornelis, J., Decocq, G.M., Dierschke, H., Eriksson, O., Gilliam, F.S., Hedl, R., Heinken, T., Hermy, M., Hommel, P., Jenkins, M.A., Kelly, D.L., Kirby, K.J., Mitchell, F.J.G., Naaf, T., Newman, M., Peterken, G., Petrik, P., Schultz, J., Sonnier, G., Van Calster, H., Waller, D.M., Walther, G.R., White, P.S., Woods, K.D., Wulf, M., Graae, B.J., Verheyen, K., 2013. Microclimate moderates plant responses to macroclimate warming. Proc. Natl. Acad. Sci. U. S. A. 110, 18561–18565.

De Witte, L.C., Stöcklin, J., 2010. Longevity of clonal plants: why it matters and how to measure it. Ann. Bot. Lond. 106, 859–870.

Dickinson, M.G., Orme, C.D.L., Suttle, K.B., Mace, G.M., 2014. Separating sensitivity from exposure in assessing extinction risk from climate change. Sci. Rep. 4.

Dobrowski, S.Z., Abatzoglou, J., Swanson, A.K., Greenberg, J.A., Mynsberge, A.R., Holden, Z.A., Schwartz, M.K., 2013. The climate velocity of the contiguous United States during the 20th century. Glob. Change Biol. 19, 241–251.

Eriksson, O., 1996. Regional dynamics of plants: a review of evidence for remnant, source-sink and metapopulations. Oikos 77, 248–258.

Eriksson, O., 2000. Functional roles of remnant plant populations in communities and ecosystems. Glob. Ecol. Biogeogr. 9, 443–449.

Feurdean, A., Bhagwat, S.A., Willis, K.J., Birks, H.J.B., Lischke, H., Hickler, T., 2013. Tree migration-rates: narrowing the gap between inferred post-glacial rates and projected rates. PLoS One 8.

Franklin, J., Davis, F.W., Ikegami, M., Syphard, A.D., Flint, L.E., Flint, A.L., Hannah, L., 2013. Modeling plant species distributions under future climates: how fine scale do climate projections need to be? Glob. Change Biol. 19, 473–483.

Graae, B.J., Ejrnaes, R., Lang, S.I., Meineri, E., Ibarra, P.T., Bruun, H.H., 2011. Strong microsite control of seedling recruitment in tundra. Oecologia 166, 565–576.

Graae, B.J., De Frenne, P., Kolb, A., Brunet, J., Chabrerie, O., Verheyen, K., Pepin, N., Heinken, T., Zobel, M., Shevtsova, A., Nijs, I., Milbau, A., 2012. On the use of weather data in ecological studies along altitudinal and latitudinal gradients. Oikos 121, 3–19.

Grime, J.P., 2001. Plant Strategies, Vegetation Processes, and Ecosystem Properties. John Wiley & Sons, Chichester.

Grimmond, C.S.B., Robeson, S.M., Schoof, J.T., 2000. Spatial variability of micro-climatic conditions within a mid-latitude deciduous forest. Clim. Res. 15, 137–149.

Hampe, A., Jump, A.S., 2011. Climate relicts: past, present, future. Annu. Rev. Ecol. Evol. Syst. 42, 313–333.

Hanski, I., 1998. Metapopulation dynamics. Nature 396, 41–49.

Holt, R.D., 1993. Ecology at the mesoscale: the influence of regional processes on local communities. In: Ricklefs, R.E., Schluter, D. (Eds.), Species Diversity Ecological Communities. University of Chicago Press, Chicago, Illinois, pp. 77–88.

Hubbell, S.P., 2001. The Unified Neutral Theory of Biodiversity and Biogeography. Princeton University Press, Princeton.

Jackson, S.T., Sax, D.F., 2010. Balancing biodiversity in a changing environment: extinction debt, immigration credit and species turnover. Trends Ecol. Evol. 25, 153–160.

Jump, A.S., Peñuelas, J., 2005. Running to stand still: adaptation and the response of plants to rapid climate change. Ecol. Lett. 8, 1010–1020.

Körner, C., Basler, D., Hoch, G., Kollas, C., Lenz, A., Randin, C.F., Vitasse, Y., Zimmermann, N.E., 2016. Where, why and how? Explaining the low-temperature range limits of temperate tree species. J. Ecol. 104, 1076–1088.

Körner, C., 2003. Alpine Plant Life Functional Plant Ecology of High Mountain Ecosystems. Springer-Verlag, Heidelberg.

Körner, C., 2004. Mountain biodiversity, its causes and function. Ambio Spec. Rep. 13, 11–17.

Kerr, J.T., Packer, L., 1997. Habitat heterogeneity as a determinant of mammal species richness in high-energy regions. Nature 385, 252–254.

Kuussaari, M., Bommarco, R., Heikkinen, R.K., Helm, A., Krauss, J., Lindborg, R., Öckinger, E., Pärtel, M., Pino, J., Rodà, F., Stefanescu, C., Teder, T., Zobel, M., Steffan-Dewenter, I., 2009. Extinction debt: a challenge for biodiversity conservation. Trends Ecol. Evol. 24, 564–571.

Lande, R., 1993. Risks of population extinction from demographic and environmental stochasticity and random catastrophes. Am. Nat. 142, 911–927.

Leathwick, J.R., Austin, M.P., 2001. Competitive interactions between tree species in New Zealand's old-growth indigenous forests. Ecology 82, 2560–2573.

Leibold, M.A., Holyoak, M., Mouquet, N., Amarasekare, P., Chase, J.M., Hoopes, M.F., Holt, R.D., Shurin, J.B., Law, R., Tilman, D., Loreau, M., Gonzalez, A., 2004. The metacommunity concept: a framework for multi-scale community ecology. Ecol. Lett. 7, 601–613.

Leibold, M.A., Economo, E.P., Peres-Neto, P., 2010. Metacommunity phylogenetics: separating the roles of environmental filters and historical biogeography. Ecol. Lett. 13, 1290–1299.

Lenoir, J., Svenning, J.C., 2013. Latitudinal and elevational range shifts under contemporary climate change. In: Levin, S.A. (Ed.), Encyclopedia of Biodiversity. Academic Press, 599-611.

Lenoir, J., Svenning, J.C., 2015. Climate-related range shifts – a global multidimensional synthesis and new research directions. Ecography 38, 15–28.

Lenoir, J., Gégout, J.C., Marquet, P.A., De Ruffray, P., Brisse, H., 2008. A significant upward shift in plant species optimum elevation during the 20th century. Science 320, 1768–1771.

Lenoir, J., Gégout, J.C., Guisan, A., Vittoz, P., Wohlgemuth, T., Zimmermann, N.E., Dullinger, S., Pauli, H., Willner, W., Svenning, J.C., 2010. Going against the flow: potential mechanisms for unexpected downslope range shifts in a warming climate. Ecography 33, 295–303.

Lenoir, J., Graae, B.J., Aarrestad, P.A., Alsos, I.G., Armbruster, W.S., Austrheim, G., Bergendorff, C., Birks, H.J.B., Brathen, K.A., Brunet, J., Bruun, H.H., Dahlberg, C.J., Decocq, G., Diekmann, M., Dynesius, M., Ejrnaes, R., Grytnes, J.A., Hylander, K., Klanderud, K., Luoto, M., Milbau, A., Moora, M., Nygaard, B., Odland, A., Ravolainen, V.T., Reinhardt, S., Sandvik, S.M., Schei, F.H., Speed, J.D.M., Tveraabak, L.U., Vandvik, V., Velle, L.G., Virtanen, R., Zobel, M., Svenning, J.C., 2013. Local temperatures inferred from plant communities suggest strong spatial buffering of climate warming across Northern Europe. Glob. Change Biol. 19, 1470–1481.

Lenoir, J., Hattab, T., Pierre, G., 2017. Climatic microrefugia under anthropogenic climate change: implications for species redistribution. Ecography 40, 253–266.

Levins, R., Culver, D., 1971. Regional coexistence of species and competition between rare species. Proc. Natl. Acad. Sci. U. S. A. 68, 1246–1248.

Loarie, S.R., Duffy, P.B., Hamilton, H., Asner, G.P., Field, C.B., Ackerly, D.D., 2009. The velocity of climate change. Nature 462, 1052–U1111.

Logue, J.B., Mouquet, N., Peter, H., Hillebrand, H., Metacommunity Working G, 2011. Empirical approaches to metacommunities: a review and comparison with theory. Trends Ecol. Evol. 26, 482–491.

MacArthur, R.H., Wilson, E.O., 1963. An equilibrium theory of insular zoogeography. Evolution 17, 373–387.

May, M.R., Provance, M.C., Sanders, A.C., Ellstrand, N.C., Ross-Ibarra, J., 2009. A pleistocene clone of palmer's oak persisting in southern california. PLoS One 4.

Migliore, J., Baumel, A., Juin, M., Fady, B., Roig, A., Duong, N., Medail, F., 2013. Surviving in mountain climate refugia: new insights from the genetic diversity and structure of the relict shrub myrtus nivellei (Myrtaceae) in the sahara desert. PLoS One 8.

Moeslund, J.E., Arge, L., Bøcher, P.K., Dalgaard, T., Ejrnæs, R., Odgaard, M.V., Svenning, J.C., 2013. Topographically controlled soil moisture drives plant diversity patterns within grasslands. Biodivers. Conserv. 22, 2151–2166.

Mouquet, N., Loreau, M., 2003. Community patterns in source-sink metacommunities. Am. Nat. 162, 544–557.

Murphy, C.A., Foster, B.L., 2014. Soil properties and spatial processes influence bacterial metacommunities within a grassland restoration experiment. Restor. Ecol. 22, 685–691.

Myers, J.A., Harms, K.E., 2009. Seed arrival, ecological filters, and plant species richness: a meta-analysis. Ecol. Lett. 12, 1250–1260.

Nicotra, A.B., Atkin, O.K., Bonser, S.P., Davidson, A.M., Finnegan, E.J., Mathesius, U., Poot, P., Purugganan, M.D., Richards, C.L., Valladares, F., van Kleunen, M., 2010. Plant phenotypic plasticity in a changing climate. Trends Plant Sci. 15, 684–692.

Olsen, S.L., Töpper, J.P., Skarpaas, O., Vandvik, V., Klanderud, K., 2016. From facilitation to competition: temperature-driven shift in dominant plant interactions affects population dynamics in semi-natural grasslands. Glob. Change Biol. 22, 1915–1926.

Opedal, O.H., Armbruster, W.S., Graae, B.J., 2015. Linking small-scale topography with microclimate, plant species diversity and intra-specific trait variation in an alpine landscape. Plant Ecol. Divers. 8, 305–315.

Pearson, R.G., Dawson, T.P., 2003. Predicting the impacts of climate change on the distribution of species: are bioclimate envelope models useful? Glob. Ecol. Biogeogr. 12, 361–371.

Pellissier, L., Bråthen, K.A., Pottier, J., Randin, C.F., Vittoz, P., Dubuis, A., Yoccoz, N.G., Alm, T., Zimmermann, N.E., Guisan, A., 2010. Species distribution models reveal apparent competitive and facilitative effects of a dominant species on the distribution of tundra plants. Ecography 33, 1004–1014.

Pellissier, L., Bråthen, K.A., Vittoz, P., Yoccoz, N.G., Dubuis, A., Meier, E.S., Zimmermann, N.E., Randin, C.F., Thuiller, W., Garraud, L., Van Es, J., Guisan, A., 2013. Thermal niches are more conserved at cold than warm limits in arctic-alpine plant species. Glob. Ecol. Biogeogr. 22, 933–941.

Pillar, V.D., Duarte, L.D.S., 2010. A framework for metacommunity analysis of phylogenetic. Ecol. Lett. 13, 587–596.

Randin, C.F., Engler, R., Normand, S., Zappa, M., Zimmermann, N.E., Pearman, P.B., Vittoz, P., Thuiller, W., Guisan, A., 2009. Climate change and plant distribution: local models predict high-elevation persistence. Glob. Change Biol. 15, 1557–1569.

Raunkiaer, C., 1934. The Life Forms of Plants and Statistical Plant Geography: Being the Collected Papers of C. Raunkiaer. Clarendon Press, Oxford.

Reino, L., Beja, P., Araujo, M.B., Dray, S., Segurado, P., 2013. Does local habitat fragmentation affect large-scale distributions?: The case of a specialist grassland bird. Divers. Distrib. 19, 423–432.

Roth, T., Plattner, M., Amrhein, V., 2014. Plants, birds and butterflies: short-term responses of species communities to climate warming vary by taxon and with altitude. PLoS One 9, e82490.

Sandel, B., Arge, L., Dalsgaard, B., Davies, R.G., Gaston, K.J., Sutherland, W.J., Svenning, J.C., 2011. The influence of late quaternary climate-change velocity on species endemism. Science 334, 660–664.

Scherrer, D., Körner, C., 2010. Infra-red thermometry of alpine landscapes challenges climatic warming projections. Glob. Change Biol. 16, 2602–2613.

Scherrer, D., Körner, C., 2011. Topographically controlled thermal-habitat differentiation buffers alpine plant diversity against climate warming. J. Biogeogr. 38, 406–416.

Slavich, E., Warton, D.I., Ashcroft, M.B., Gollan, J.R., Ramp, D., 2014. Topoclimate versus macroclimate: how does climate mapping methodology affect species distribution models and climate change projections? Divers. Distrib. 20, 952–963.

Spasojevic, M.J., Bowman, W.D., Humphries, H.C., Seastedt, T.R., Suding, K.N., 2013. Changes in alpine vegetation over 21 years: are patterns across a heterogeneous landscape consistent with predictions? Ecosphere 4, 117.

Svenning, J.C., Sandel, B., 2013. Disequilibrium vegetation dynamics under future climate change. Am. J. Bot. 100, 1266–1286.

Sykes, M.T., Prentice, I.C., Cramer, W., 1996. A bioclimatic model for the potential distributions of north European tree species under present and future climates. J. Biogeogr. 23, 203–233.

Telford, R.J., Vandvik, V., Birks, H.J.B., 2006. Dispersal limitations matter for microbial morphospecies. Science 312, 1015.

Tilman, D., 1994. Competition and biodiversity in spatially structured habitats. Ecology 75, 2–16.

Trivedi, M.R., Berry, P.M., Morecroft, M.D., Dawson, T.P., 2008. Spatial scale affects bioclimate model projections of climate change impacts on mountain plants. Glob. Change Biol. 14, 1089–1103.

Tscharntke, T., Tylianakis, J.M., Rand, T.A., Didham, R.K., Fahrig, L., Peter, B., Bengtsson, J., Clough, Y., Crist, T.O., Dormann, C.F., Ewers, R.M., Fruend, J., Holt, R.D., Holzschuh, A., Klein, A.M., Kleijn, D., Kremen, C., Landis, D.A., Laurance, W., Lindenmayer, D., Scherber, C., Sodhi, N., Steffan-Dewenter, I., Thies, C., van der Putten, W.H., Westphal, C., 2012. Landscape moderation of biodiversity patterns and processes – eight hypotheses. Biol. Rev. 87, 661–685.

Urban, M.C., Leibold, M.A., Amarasekare, P., De Meester, L., Gomulkiewicz, R., Hochberg, M.E., Klausmeier, C.A., Loeuille, N., de Mazancourt, C., Norberg, J., Pantel, J.H., Strauss, S.Y., Vellend, M., Wade, M.J., 2008. The evolutionary ecology of metacommunities. Trends Ecol. Evol. 23, 311–317.

Valladares, F., Matesanz, S., Guilhaumon, F., Araújo, M.B., Balaguer, L., Benito-Garzón, M., Cornwell, W., Gianoli, E., van Kleunen, M., Nicotra, A.B., Zavala, M.A., 2014. The effects of phenotypic plasticity and local adaptation on forecasts of species range shifts under climate change. Ecol. Lett. 17, 1351–1364.

Vandvik, V., Goldberg, D.E., 2005. Distinguishing the roles of dispersal in diversity maintenance and in diversity limitation. Folia Geobot. 40, 45–52.

Vandvik, V., Goldberg, D.E., 2006. Sources of diversity in a grassland metacommunity: quantifying the contribution of dispersal to species richness. Am. Nat. 168, 157–167.

Vranckx, G., Jacquemyn, H., Muys, B., Honnay, O., 2012. Meta-analysis of susceptibility of woody plants to loss of genetic diversity through habitat fragmentation. Conserv. Biol. 26, 228–237.

Wasof, S., Lenoir, J., Gallet-Moron, E., Jamoneau, A., Brunet, J., Cousins, S.A.O., De Frenne, P., Diekmann, M., Hermy, M., Kolb, A., Liira, J., Verheyen, K., Wulf, M., Decocq, G., 2013. Ecological niche shifts of understorey plants along a latitudinal gradient of temperate forests in north-western Europe. Glob. Ecol. Biogeogr. 22, 1130–1140.

Wasof, S., Lenoir, J., Aarrestad, P.A., Alsos, I.G., Armbruster, W.S., Austrheim, G., Bakkestuen, V., Birks, H.J.B., Bråthen, K.A., Broennimann, O., Brunet, J., Bruun, H.H., Dahlberg, C.J., Diekmann, M., Dullinger, S., Dynesius, M., Ejrnæs, R., Gégout, J.-C., Graae, B.J., Grytnes, J.-A., Guisan, A., Hylander, K., Jónsdóttir, I.S., Kapfer, J., Klanderud, K., Luoto, M., Milbau, A., Moora, M., Nygaard, B., Odland, A., Pauli, H., Ravolainen, V., Reinhardt, S., Sandvik, S.M., Schei, F.H., Speed, J.D.M., Svenning, J.-C., Thuiller, W., Tveraabak, L.U., Vandvik, V., Velle, L.G., Virtanen, R., Vittoz, P., Willner, W., Wohlgemuth, T., Zimmermann, N.E., Zobel, M., Decocq, G., 2015. Disjunct populations of European vascular plant species keep the same climatic niches. Glob. Ecol. Biogeogr. 24, 1401–1412.

Wesser, S.D., Armbruster, W.S., 1991. Species distribution controls across a forest-steppe transition: a causal model and experimental test. Ecol. Monogr. 61, 323–342.

Westergaard, K.B., Jørgensen, M.H., Gabrielsen, T.M., Alsos, I.G., Brochmann, C., 2010. The extreme Beringian/Atlantic disjunction in *Saxifraga rivularis* (Saxifragaceae) has formed at least twice. J. Biogeogr. 37, 1262–1276.

Westergaard, K.B., Alsos, I.G., Engelskjøn, T., Flatberg, K.I., Brochmann, C., 2011. Trans-Atlantic genetic uniformity in the rare snowbed sedge *Carex rufina*. Conserv. Genet. 12, 1367–1371.

Whittaker, R.H., 1962. Classification of natural communities. Bot. Rev. 28, 1–239.

Wipf, S., Rixen, C., 2010. A review of snow manipulation experiments in Arctic and alpine tundra ecosystems. Polar Res. 29, 95–109.

Wisz, M.S., Pottier, J., Kissling, W.D., Pellissier, L., Lenoir, J., Damgaard, C.F., Dormann, C.F., Forchhammer, M.C., Grytnes, J.-A., Guisan, A., Heikkinen, R.K., Høye, T.T., Kühn, I., Luoto, M., Maiorano, L., Nilsson, M.-C., Normand, S., Öckinger, E., Schmidt, N.M., Termansen, M., Timmermann, A., Wardle, D.A., Aastrup, P., Svenning, J.-C., 2013. The role of biotic interactions in shaping distributions and realised assemblages of species: implications for species distribution modelling. Biol. Rev. 88, 15–30.

Zobel, M., 1997. The relative role of species pools in determining plant species richness. An alternative explanation of species coexistence? Trends Ecol. Evol. 12, 266–269.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 51–61

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Research article

Slow community responses but rapid species responses 14 years after alpine turf transplantation among snow cover zones, south–central New Zealand[☆]

Janice M. Lord[a,*], Alan F. Mark[a], Tanja Humar-Maegli[a,1], Stephan R.P. Halloy[b], Peter Bannister[a,2], Allison Knight[a], Katharine J.M. Dickinson[a]

[a] Department of Botany, University of Otago, Dunedin, New Zealand
[b] Ministry for Primary Industries, Wellington, New Zealand

ARTICLE INFO

Keywords:
Celmisia species
Climate change
Herbivory
Reciprocal transplants
Snowbed
Thamnolia vermicularis

ABSTRACT

Alpine ecosystems are particularly vulnerable to the impact of global climate change. Depth and duration of seasonal snow cover are major drivers of variation in alpine plant community composition, so a reduction in snow cover as a result of climate change would expose plants that are currently protected by snow in winter and spring to greater extremes of temperature and increased risk of frost damage. We reciprocally transplanted 64 intact 60 cm × 60 cm × 10 cm (minimum depth) turves of alpine vegetation among four topographic zones on the Rock and Pillar Range, south-central South Island, New Zealand to investigate how shifts along a snow cover gradient affected plant growth, survival and community composition. The four zones: late-melting snowbed and early-melting snowbed in depressions, moderately exposed leeward upper slopes dominated by herbfield, and extremely exposed summit plateau dominated by cushionfield, differed in winter and spring snow cover. As expected, the highest species losses occurred in turves transplanted to very different zones e.g. late snowbed to summit plateau and vice versa. However many snowbed species still survived on the summit plateau seven years following transplantation. The degree to which turves had been colonised after seven years was significantly related to transplant zone rather than turf origin or original species richness; turves transplanted to the most species-rich zones were affected most by colonisation. Measurements of leaf production over three years in three focal *Celmisia* species (Asteraceae), characteristic of the herbfield on leeward slopes, and early and late snowbeds, showed that the late snowbed specialist suffered significantly reduced growth when transplanted to more exposed sites, but its survival was more affected by invertebrate herbivory rather than the direct effects of exposure. The cosmopolitan focal lichen species *Thamnolia vermicularis*, monitored over 14 years, rapidly colonised turves transplanted to cushionfield on the exposed summit plateau, where this and other lichens are abundant, but equally rapidly declined in turves transplanted to snowbeds. These findings add to a growing body of evidence that biotic interactions and species-specific traits will be critical drivers of alpine vegetation change under future climate scenarios.

1. Introduction

Alpine and polar ecosystems are likely to be particularly vulnerable to the impact of global climate change; global increases in air temperature and reduction in snow, ice and permafrost are strongly supported both by climate models and observation (IPCC 2014). However, our understanding of the impact of climate change on alpine areas around the world is less clear than our understanding of climate change in polar regions, due to the effects of temperature, precipitation, topography and geography on the behavior of snow (Gobiet et al., 2014; Musselman et al., 2017). Levels of snow cover in European and western North American mountain regions are significantly decreasing (Mote et al., 2005; Gobiet et al., 2014), and likewise, both maximum snow depth and total snow fall has declined in the Australian Alps (Fiddes et al., 2015). However, in the South American Andes, while the length of the core snow period has decreased generally, snow cover has significantly decreased only in northern portion of the range and instead increased in the southern Andes (Mernild et al., 2017).

[☆] This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author.

E-mail address: janice.lord@otago.ac.nz (J.M. Lord).

[1] Present address: Wabern, Switzerland.

[2] Deceased.

http://dx.doi.org/10.1016/j.ppees.2017.07.004
Received 23 December 2016; Received in revised form 7 June 2017; Accepted 25 July 2017

Globally, increasing temperatures are likely to reduce the extent of alpine habitats, meaning that alpine species unable to migrate to suitable microclimatic niches, are likely to face competition from invading plant species from lower elevations (Halloy and Mark 2003; Hülber et al., 2011; Pauli et al., 2012; Alexander et al., 2016). Actual and experimental increases in temperature have been shown to correlate with changing plant species abundances and distributions (Walker et al., 2006; Cannone et al., 2007; Sebastià et al., 2008; Elmendorf et al., 2015). However, in topographically diverse alpine landscapes the availability of climatic microrefugia will likely provide a buffer against the impacts of warming temperatures (Scherrer and Körner 2010; Patsiou et al., 2014). In addition, a surprising number of alpine species show high levels of phenotypic plasticity (Frei et al., 2014; Sedlacek et al., 2015) and considerable resilience to environment stressors beyond their realised climatic niche (Bannister et al., 2005; Venn et al., 2013). These studies and others, together strongly connect with an emerging theme in the alpine climate change research field; that climate change impacts in alpine regions cannot be readily predicted from large scale climate models of communities. Rather, region-specific interactions between environmental factors, individual species traits, interactions between species, and indirect effects, are paramount in determining outcomes (Debouk et al., 2015; Farrer et al., 2015; Guittar et al., 2016; Alexander et al., 2016).

Variation in the depth and duration of seasonal snow cover is a major driver of alpine plant community composition, and modifications to that cover can produce changes in plant species distributions even over relatively short spaces of time (Carbognani et al., 2014; Mark et al., 2015). Warming temperatures in alpine regions are likely to alter patterns of seasonal snow cover, amount of precipitation falling as rain instead of snow and spring melt water (Lapp et al., 2005; Hendrikx et al., 2012; Gobiet et al., 2014; Klein et al., 2016; Musselman et al., 2017). Seasonal snow cover insulates and protects the underlying vegetation from sub-zero temperatures and damage from wind and frost (Bannister et al., 2005); long-lying snow cover not only acts as an insulator, but also significantly influences soil moisture levels due to the delayed release of melt-water (Sedlacek et al., 2015; Musselman et al., 2017). Furthermore, the timing of release from snow can also significantly delay phenological events such as leaf expansion and flowering (Björk and Molau 2007; Petraglia et al., 2014; Sedlacek et al., 2015), with consequential effects on the availability of resources for herbivores and pollinators (Björk and Molau 2007; Forrest and Thomson, 2011). These effects of snow cover lead to strong correlations between its presence and duration, and variation among alpine species in life-history, growth rate and functional traits (Callaghan et al., 2004; Jonas et al., 2008; Mark et al., 2016; Wheeler et al., 2016). In addition, persistent snowbeds, which are areas that, due to topography, accumulate deep winter snow, usually support a range of species not present elsewhere (Carbognani et al., 2012; Carbognani et al., 2012; Mark et al., 2016). While topographic variation is still likely to provide a mosaic of microclimates, snowbeds will be particularly threatened by spatial reduction and shifts in microclimate (Björk and Molau 2007; Matteodo et al., 2016).

The aim of this study was to investigate the long term impact of altered snow cover on alpine plant community composition on an isolated mountain range in south-central South Island, New Zealand, using reciprocal transplantation of intact plant communities along a natural gradient of snow cover. Transplantation is a powerful, but still relatively uncommon, method for directly testing the impact of altered environmental conditions and potentially novel biotic interactions on species and communities (Alexander et al., 2016; Nooten and Andrew 2017). Within a natural snow cover gradient we aimed to expose alpine species and communities to conditions representing likely and extreme levels of altered snow cover under future climate scenarios. Natural treeline in southern New Zealand is c.1200 m a.s.l. (above sea level) (Cieraad and McGlone 2014). Climate change models currently predict stable to increased precipitation in central and eastern areas of southern New Zealand, but up to 22% reduction in peak snow accumulation below altitudes of 2000 m a.s.l. due to more precipitation falling as rain rather than snow (Mullan et al., 2008; Hendrikx et al., 2012). The New Zealand alpine flora shows a high level of regional and local endemism (c.93%), with highest species richness in the northern and southern regions of South Island. The alpine lichen flora is also very diverse, containing many bipolar species that are also found at high latitudes and altitudes in the Northern Hemisphere (Galloway and Aptroot 1995; Mark, 2012). Specialist alpine plant and lichen species are particularly associated with exposed habitats such as cushionfields and fellfields, characterised by mat-forming vegetation and rocky ground subject to freeze-thaw cycles respectively, as well as persistent snowbeds (McGlone and Heenan 2013). Our study aimed to quantify the resilience of such species and their constituent communities to altered snow regimes by additionally investigating the growth responses of focal species. We predicted that species that are restricted to particular zones along an exposure gradient would be most affected by changes in snow cover as a consequence of transplantation.

2. Methods

2.1. Study area

This study was carried out in the alpine zone on the Rock and Pillar Range (45° 25′ 32″ S, 170° 04′ 15″ E) South Island, New Zealand. The Rock and Pillar Range is one of a series of block faulted, chlorite schistose ranges in Central Otago, with a steeply sloping, heavily dissected, south-eastern scarp face and a summit plateau reaching 1450 m a.s.l. at its highest point. Subalpine shrub species and tall tussock (bunch) grasses occur in scattered patches up to *c.* 1280 m a.s.l., above which alpine herbfield and cushionfield dominate (Fig. 1A). The climate of the Rock and Pillar Range is somewhat maritime as it is 50 km from the Otago coast, but frosts can occur at any time of the year; Bannister et al. (2005) recorded a mean daily minimum air temperature of −0.33° C, an extreme daily minimum of −10.08° C in the same study area, and 42% of days with subzero air temperatures over a 16-month period (February 2003–June 2004). Annual precipitation exceeds 1000 mm, fog and low cloud are common and soil moisture levels remain close to field capacity throughout the growing season (Bliss and Mark 1974; Holdsworth and Mark 1990). Snow can fall at any time of the year, predominantly during southerly storms, and is extensively redistributed by the prevailing westerly winds, which average 7.9 ms^{-1} at 2 m above ground level, reducing to 2–3.5 ms^{-1} at ground level on the summit plateau, and 1.5–2.5 ms^{-1} at ground level on upper leeward slopes (Bliss and Mark 1974). Snow depth and duration vary consistently with topography; winter snow cover on the summit plateau varies between 0 and 0.5 m in depth due to extensive snow redistribution, whereas leeward upper slopes and snowbeds as little as 40 m below the summit plateau, accumulate snow throughout winter to depths of 0.5–4 m and up to 7 m, respectively (Talbot et al., 1992). Winter snow begins to accumulate from mid-May on leeward slopes and in snowbeds. The date that each zone is released from snow cover can vary by several days between successive years, but varies significantly more among zones, with snow in snowbeds persisting more than 50 days longer than on the summit plateau (Talbot et al., 1992). Snowmelt in all zones is usually completed by early– to mid–December.

Alpine vegetation patterns on the Rock and Pillar Range are strongly influenced by variation in snow depth and duration (Bliss and Mark 1974; Talbot et al., 1992). The exposed summit plateau is dominated by very low cushion-mat vegetation with a high abundance and diversity of lichens. The leeward slopes support a low diversity herbfield featuring large patches of *Celmisia viscosa* (Asteraceae), which has 5–8 cm long leaves coated in an epimanool-rich exudate that may provide protection from herbivores and desiccation (Sansom et al., 2013). In contrast, the vegetation of snowbeds is characterised by comparatively high species richness and diversity (Talbot et al., 1992; Table 1). Two

Fig 1. The study area, Rock and Pillar Range, South Island, New Zealand. (A): Catchment 1 showing the relative location of the four topographic zones; (B): late snowbed zone in catchment 2 with an excavated turf containing *Celmisia haastii* var. *tomentosa* ready for transport to its new location. Inset shows the location of the study area in relation to Dunedin, south–eastern South Island.

further *Celmisia* species are restricted to these snowbeds; the summer-green *C. prorepens* occurs in early-melting snowbeds, growing in loose mats with sticky leaves up to 5 cm long and the Rock and Pillar's endemic variety *tomentosa* of *C. haastii* is restricted to late-melting snowbeds, growing in small to large patches with leaves up to 4 cm long (Fig. 1B; hereafter referred to as *C. haastii*). These three *Celmisia* species served as focal species in this study.

2.2. Design of transplant experiment

In the 2002–03 growing season we selected two adjacent south-easterly facing catchments, i.e. exposed to at least some direct sunlight from sunrise to early afternoon, on the upper slopes of the range (catchment 1: 45° 25′ 02″ S, 170° 05′ 05″E, alt. range 1280–1365 m a.s.l; catchment 2: 45° 25′ 05″S, 170° 04′ 45″E, alt. range 1345 – 1380 m), and transplanted two 60 cm × 60 cm × 10 cm deep (min.) turves from each of four topographic zones, reciprocally into all four zones within each of the two catchments. All transplanted turves were surrounded by undisturbed vegetation and their surface was level with that of the surrounding undisturbed community. The four topographic zones consisted of the summit plateau above each catchment, which is dominated by lichen-rich cushionfield, the leeward upper slopes of each catchment, which is dominated by herbfield, early snowbed in shallow depressions/gullies within each catchment, and late snowbed in deep depressions/gullies within each catchment. Two control turves were also excavated and relocated within each topographic zone of each catchment. Vegetation for excavation from the herbfield on leeward slopes and also from snowbeds was selected to include their characteristic *Celmisia* species. The areas available for excavation were constrained by the need to avoid large rocks and obtain a turf of 10 cm

Table 1
Plant community characteristics of topographic zones differing in snow cover, in two catchments on the Rock and Pillar Range, South Island, New Zealand. Species richness is the mean (std. dev.) number of vascular and nonvascular plant species in 0.25 m^2 turves. Unique species are restricted to one zone within a catchment but may or may not be in the same zone in the other catchment. Snow lie is given in days between first persisting snow fall and time of last snow melt, although in some zones winter snow cover is not continuous. The value in brackets is the percent reduction in snow lie compared to the late snowbed zone within each catchment. NA: not available, as snow lie in 2004 was only measured in catchment 1 and equipment failure resulted in the loss of leeward slope data.

	Species richness	Unique species	2003 winter snow lie (days)	2004 winter snow lie (days)
Catchment 1				
Summit plateau	14.75 (3.632)	10	101 (48.5%)	100 (61.2%)
Leeward slope	5.875 (1.965)	3	144 (26.8%)	NA
Early Snowbed	10.63 (3.967)	6	180 (8.2%)	179 (30.6%)
Late Snowbed	13.00 (3.742)	12	196	258
Catchment 2				
Summit plateau	13.88 (2.204)	16	109 (39.4%)	NA
Leeward slope	8.000 (2.398)	4	163 (9.4%)	NA
Early Snowbed	9.375 (4.151)	5	159 (11.7%)	NA
Late Snowbed	14.63 (2.826)	19	180	NA

minimum depth, but were randomly selected within these constraints and randomly allocated to one of the four topographic zones. Turves were excavated by hand and transported in custom-made trays to minimise disturbance (Fig. 1B). Each turf was watered once, immediately after transplantation. No further watering was undertaken for the remainder of the experiment.

2.3. Climate measurements

To record soil temperature differences among topographic zones, Optic StowAway temperature loggers (Onset Computer Corp., Bourne, MA) were buried horizontally 10 cm below the ground surface adjacent to the uppermost turf in each zone of each catchment after all turves were transplanted, and set to record temperature hourly. Dataloggers were removed and downloaded after 14 months and mean, minimum and maximum monthly temperatures calculated.

2.4. Plant community surveys

The initial abundance of all vascular and non-vascular plant species and lichens was assessed by counting the number of 10 cm x 10 cm grid squares each species occupied in the central 9 squares of a permanently marked, 50 cm x 50 cm area (25 squares) on each turf at the time of transplanting. In January 2010, turves were resurveyed, counting the number of grid squares occupied out of 25 in order to quantify peripheral colonisation of the turf by new species. The impact of transplantation on plant community composition was elucidated using nonmetric multidimensional scaling (NMDS). The positions of turves along two ordination axes, based on species count data, were calculated using the Chi-square distance measure. Data from the original survey in the 2002–03 season were analysed to examine the distinctiveness of plant communities naturally occurring in the four topographic zones and between the two replicate catchment systems. Data from the January 2010 survey, seven years after transplantation, were analysed to investigate shifts in compositional similarity among transplanted and control turves. Data from the intervening years are not presented here. Multivariate analysis of variance was used to test if the resulting axis scores differed significantly with either turf source (original zone) or location (zone the turf was transplanted into). Analysis of covariance was used to test if persistence of original species or number of new species colonising after seven years was affected by source or location of turves, independent of original species richness.

2.5. Leaf production in *Celmisia* species

Responses of the focal *Celmisia* species to altered snow cover were assessed by measuring leaf production in each of the three seasons following transplantation (2003–04, 2004–05 and 2005–06). In each *Celmisia*-containing turf transplanted between or within zones (i.e. only turves sourced from the herbfield on leeward slopes, and from late and early snowbeds: the focal *Celmisia* species did not occur in the summit plateau cushionfield), three to five *Celmisia* shoots were marked with coloured pegs. If shoots died, replacement shoots in the same turf were marked if available. Five non-transplanted control shoots were also marked in each of the three *Celmisia*-containing zones adjacent to the uppermost turf in that zone. The leafy shoots of *Celmisia* species grow determinately, with new leaves continually produced from the stem apex throughout the growing season: over-wintering buds are absent in these and most indigenous species (Dumbleton 1967). Inflorescences arise from the axils of the previous season's leaves. In late spring (November) of each year, new season's leaves were counted, the youngest fully expanded leaf present was marked by removing the apical 5 mm of the leaf tip, and developing leaves above the marked leaf were counted. Plants were revisited approximately monthly throughout each growing season until early autumn (March-April) to count further leaf production and re-mark leaves as necessary. The number of leaves produced per shoot was averaged for each turf in each season, and analysed using a General Linear Model treating catchment as a block effect and season, location and species as fixed effects. All 2-way interactions were included in the final model but the 3-way interaction between season, location and species was omitted as it did not contribute significantly to the model. The effect of transplantation was analysed using a Paired T-test, comparing mean leaves per shoot per season for non-transplanted control shoots with the grand mean of leaves per shoot per season of the two turves transplanted within the same zone.

Many shoots of the late snowbed specialist *Celmisia haastii* were observed to suffer from invertebrate herbivory and died during the course of measurement of leaf production. A Chi-squared test for independence was used to examine whether shoot death occurred nonrandomly in this species in relation to zone. All data were inspected for normality and homoscedasticity prior to analysis. Analyses were performed in SPSS v. 24 (IBM) and Statistix v.9 (Analytical Software).

2.6. Abundance of *Thamnolia vermicularis*

To assess the impact of altered snow cover on non-vascular species, the focal lichen species, *Thamnolia vermicularis* was surveyed in detail. *T. vermicularis* is a cosmopolitan, fruticose lichen, with white tubular thalli occurring either in isolation or as interconnected clumps. *T. vermicularis* occurs in alpine areas worldwide as two different chemical variants which differ in UV fluorescence (UV+, UV−) (Sato 1968; Thomson 1984; Kärnefelt and Thell 1995; Galloway 2007), but which have been shown by DNA analysis to be the same species (Lord et al., 2013). Colonisation can occur via wind-blown thalli (Pérez 1991), however this species also produces asexual fungal spores (conidia) capable of long distance dispersal (Lord et al., 2013). *T. vermicularis* is naturally abundant in the exposed cushionfield on the summit plateau of the Rock and Pillar Range and c. 95% of strands here are the UV-variant. *Thamnolia* is also found in moderate abundance in the leeward upper slopes, but is absent from areas of moderate and deep snow accumulation. Its abundance was assessed by counting the total number of thalli > 1 cm in length on all turves in March 2004, February 2011, and February 2017, roughly one, eight and 14 years after

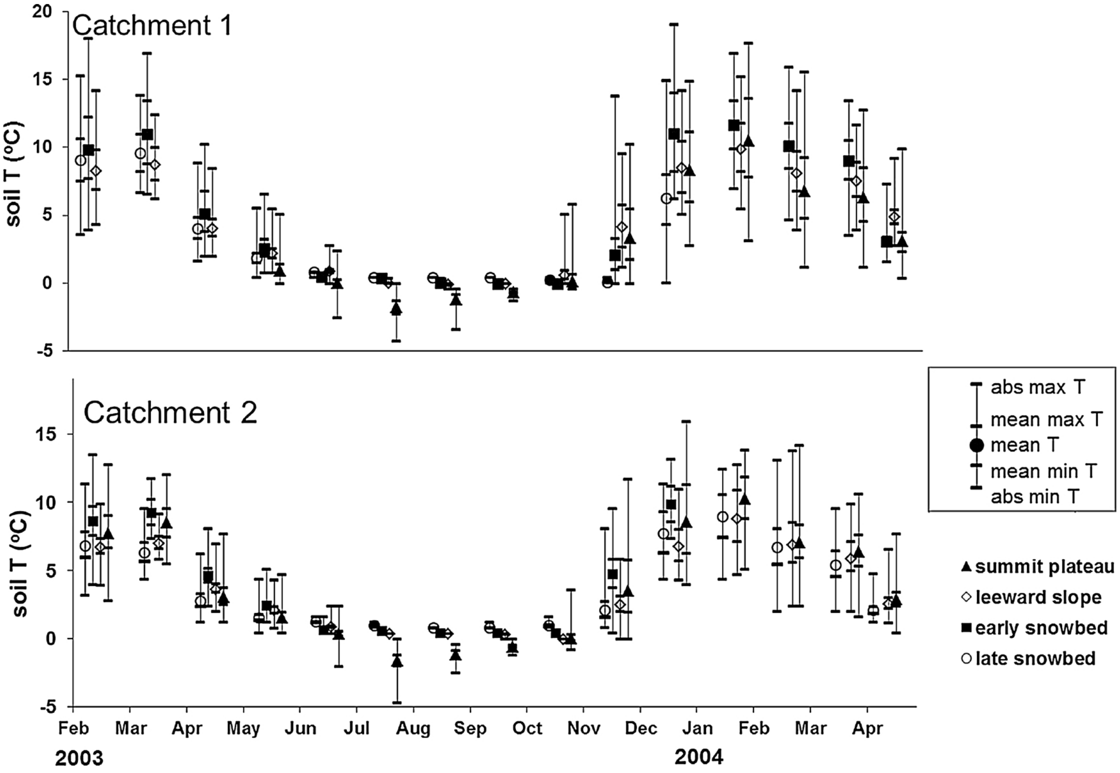


Fig. 2. Monthly soil temperatures in four topographic zones within two catchments on the Rock and Pillar Range, South Island, New Zealand, over the first 14 months of the transplant experiment. Data are missing for Feb–Apr 2003 on the summit plateau and Jan–Apr 2004 in the late snowbed zones of Catchment 1, and Jan-Apr 2004 in the early snowbed zone of Catchment 2 due to equipment failure.

transplantation. No attempt was made to separately count UV+ and UV- strands on turves. Changes in the abundance of *Thamnolia* thalli on turves one, eight and 14 years after transplantation was summed across the two replicate turves within each catchment, sharing the same source and transplant zone, and analysed separately for each catchment by Chi-squared tests of independence. Counts of thalli on turves transplanted into early and late snowbeds were summed to ensure expected values were greater than five.

3. Results

3.1. Climatic variation among topographic zones

Clear differences existed among topographic zones in the consistency of snow cover during winter and spring, and the likelihood of ground frosts. Over the first two winters of the experiment, the duration of snow cover on the summit plateau was reduced by 39–61%, and that of the leeward slopes was reduced by 9–27%, compared with the duration of snow cover in late snowbeds (Table 1). Mean monthly temperatures on the summit plateau were below zero throughout much of winter and early spring in both study catchments (Fig. 2), and the soil was exposed to freezing temperatures on almost 90% of winter days and more than 50% of spring days (Fig. 3). In contrast, soil temperatures on leeward slopes and in snowbeds barely dipped below zero over winter, and late snowbeds in particular were fully protected from ground level frosts throughout winter and spring (Fig. 3). Differences among topographic zones in mean monthly summer temperatures were small compared to the variation between mean minimum and mean maximum monthly temperatures within zones (Fig. 2).

3.2. Plant community responses

Plant community composition prior to transplantation showed very clear differences among topographic zones, with noticeably smaller differences between the two study catchments (Fig. 4A). The most distinctive plant communities were those found on the summit plateau and in late snowbeds; the composition of vegetation in early snowbeds and on the leeward slopes was generally more similar. Seven years after transplantation, turves had become much less distinctive, but species composition was still significantly influenced by the original location of the turf (Wilks Lambda = 0.218, $F_{6,94} = 17.923$, $p < 0.0001$; Fig. 4B). The topographic zone into which the turf had been transplanted had significantly affected species composition after seven years (Wilks Lambda = 0.281, $F_{6,94} = 13.897$, $p < 0.0001$; Fig. 4C), but the effect of zone on species composition was not influenced by the original source of the turf (Wilks Lambda = 0.750, $F_{18,94} = 0.809$, $p > 0.05$).

The number of original species persisting in turves seven years after transplantation was not significantly related to either source or transplant zone (source: $F_{3,47} = 1.69$; transplant: $F_{3,47} = 0.28$; $p > 0.05$ for both; Fig. 5), but was significantly predicted by initial species richness (test for covariate $t = 6.820$, $p < 0.001$), and there was also a significant source × zone interaction ($F_{9,47} = 2.66$, $p < 0.05$, pooled catchments). Likewise the number of extinctions of original species in turves was not significantly related to either source or transplant zone (source: $F_{3,47} = 1.69$; transplant: $F_{3,47} = 0.28$; $p > 0.05$ for both; Fig. 5), but was significantly related to initial species richness (test for covariate $t = 4.55$, $p < 0.001$), and there was a similar source x location interaction ($F_{9,47} = 2.66$, $p < 0.05$, pooled catchments; F values identical as persisting + extinct species = initial species richness, the covariate). Both interactions were driven entirely by significant differences in original species persisting or going extinct on turves from the late snowbed zone transplanted to other zones (LSD pairwise comparisons, $p < 0.05$; Fig. 5). Colonisation showed no effect of source, location or initial species richness when all colonising species were combined (all F values < 2.5, covariate $t = -0.64$, all $p > 0.05$) because vascular and nonvascular species showed very different patterns of colonisation. Colonisation of transplanted turves by vascular plant species was significantly higher in vascular species–rich late

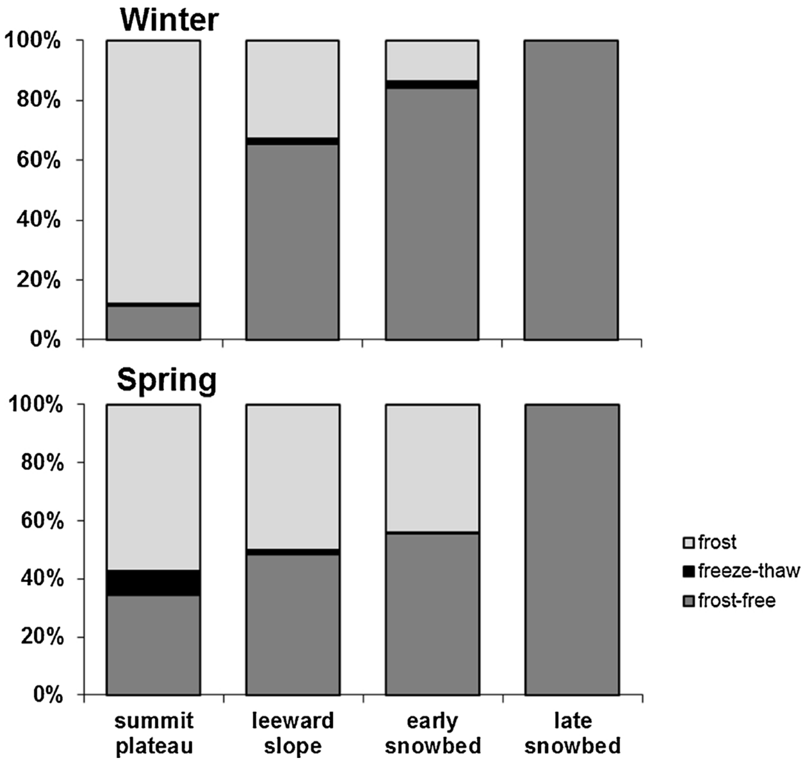


Fig. 3. Frequency of frosts, freeze-thaw days and snow-induced frost-free days in winter and spring, for four topographic zones differing in snow cover, Rock and Pillar Range, South Island, New Zealand. Values are averages for the two adjacent catchments.

snowbed (location $F_{3,47} = 4.03$, $p < 0.05$; Tukey's pairwise tests among locations $p < 0.05$), whereas colonisation by non-vascular species was significantly higher on turves in the lichen–rich summit plateau, following by turves on leeward slopes, and lowest in late snowbeds ($F_{3,47} = 13.19$, $p < 0.001$; Tukey's pairwise tests among locations, $p < 0.05$). Colonisation by either group of species was not affected by initial turf species richness (covariate $t = -0.25$, $p > 0.05$).

3.3. Responses of focal Celmisia species

A total of 303 shoots of the three focal *Celmisia* species were marked and monitored over the course of the three growing seasons; 69 of these survived one season or less, 105 survived two seasons and 129 were followed for the length of the study. Comparison of mean leaf production per shoot per season between transplanted turfs and non-transplanted control shoots indicated that transplantation did not affect leaf production ($T_{17} = 0.10$, $p > 0.05$). However, mean leaf production per shoot differed significantly among seasons, species and topographic zones, in addition to a significant species × zone interaction (Table 2). *A posteriori* tests found no significant difference in leaf production between the herbfield dominant *C. viscosa* and the early snowbed specialist *C. prorepens,* regardless of zone, but both produced significantly more leaves in every zone than *C. haastii*, the late snowbed specialist. Leaf production of the two snowbed species *C. prorepens* and *C. haastii* was, surprisingly, highest on the exposed summit plateau and most reduced in the herbfield on leeward slopes. No species performed best in its original environment.

Plant deaths in each growing season were significantly more frequent in *C. haastii* than in the other two *Celmisia* species for all zones combined (2003–04: $\chi^2 = 10.18$, df = 2, $p < 0.01$; 2004–05: $\chi^2 = 12.59$, df = 2, $p < 0.01$; 2005–06: $\chi^2 = 19.03$, df = 2, $p < 0.001$). In the first growing season after transplantation, the frequency of *C. haastii* deaths did not differ significantly among zones ($\chi^2 = 7.74$, df = 3, $p > 0.05$), however, in the following two growing seasons more *C. haastii* plants in the herbfield on leeward slopes died than expected and fewer died in the original late snowbed zone (2004–2005 season: $\chi^2 = 22.36$, df = 3, $p < 0.001$; 2005–2006 season: $\chi^2 = 36.22$, df = 3, $p < 0.0001$). Many *C. haastii* deaths were associated with invertebrate damage to the leaves, which was identified in a parallel study by Long (2006) as most likely due to feeding by the large weevil *Irenimus posticalis* (Curculionidae), which is endemic to the Rock and Pillar Range and naturally found associated with *C. viscosa* on leeward slopes. Long (2006) conducted *in vitro* experiments on the relative palatability of the three focal *Celmisia* species studied here to *Irenimus posticalis* and found that *C. haastii* was most preferred. When *C. haastii* was transplanted to leeward slopes it was quickly targeted by *Irenimus posticalis* (Long, 2006).

3.4. Response of Thamnolia vermicularis

Colonisation by *Thamnolia vermicularis* of transplanted turves initially lacking this species was surprisingly rapid and significantly affected by location; when transplanted to the summit plateau, turves from other zones were colonised by up to 94 thalli after 14 months and up to 378 thalli after eight years (Fig. 6A,B). Fourteen years after transplantation to the summit plateau, the abundance of *Thamnolia* on snowbed turves was approaching that of control cushionfield turves, transplanted within the summit plateau (Fig. 6C). The abundance of *Thamnolia* also changed rapidly following turf transplantation to late snowbeds (Fig. 6), reducing by 35% in the 11 months from transplantation to early summer, and by a further 15% over the following three months of summer/autumn (mean number of thalli after 14 months = 257.9, std. dev. = 192), compared with control turves (mean = 518.7, std. dev. = 171). After eight years this lichen species had been virtually eliminated from turves in late snowbeds (mean = 1.750, std. dev. 2.872; Fig. 6B). However cushionfield turves from the summit plateau, that were transplanted to leeward slopes and early snowbeds, still supported *Thamnolia* after 14 years (Fig. 6C), although the abundance of thalli in the early snowbeds had reduced to 5.4% of that in control turves. Chi-squared tests of the effect of source and location on the sum of thalli on replicate turves were all highly significant (Catchment 1: 2004, $\chi^2 = 334.2$; 2011, $\chi^2 = 988.6$; 2017, $\chi^2 = 1431$; Catchment 2: 2004, $\chi^2 = 164.2$; 2011, $\chi^2 = 98.80$; 2017,

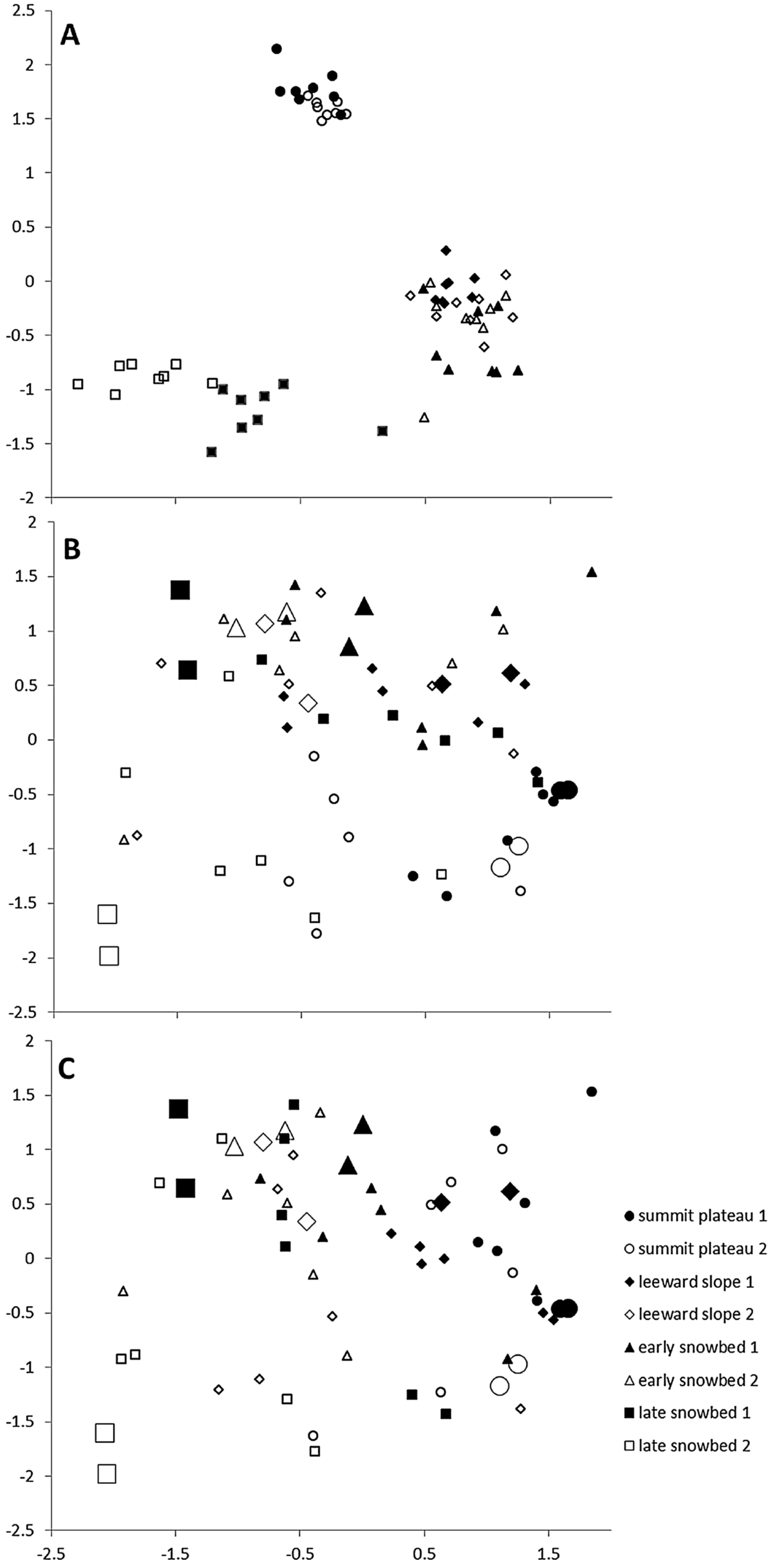


Fig. 4. Non-metric multidimensional scaling plots of variation in plant community composition in 16 turves from each of four topographic zones differing in snow cover, in each of two catchments on the Rock and Pillar Range, South Island, New Zealand. (A): Plant community composition in 2002, at the commencement of the transplant experiment, Stress = 0.2876 after 4 iterations, RSQ for matrix = 0.7977. (B) and (C): seven years after transplantation within and among four zones differing in snow cover. (B): symbols indicate topographic zone from which the turf originated; (C): symbols indicate topographic zone into which turf was transplanted. Larger symbols in B and C denote control turves transplanted within the topographic zone. Stress = 0.33234 after 4 iterations, RSQ for matrix = 0.70522.

$\chi^2 = 438.2$; $p < 0.01$, d.f. = 6 for all analyses).

4. Discussion and conclusions

Arctic and alpine plant communities throughout major parts of the world are undergoing significant transformation consistent with a changing climate (Grabherr et al., 1994; Gottfried et al., 2012; Elmendorf et al., 2015), but we still have a limited understanding of the direct and indirect processes driving these changes (Cooper, 2014; Farrar et al., 2015). Experimental warming, long term observation and space-for-time substitutions consistently highlight the critical role of species' climatic niches in determining responses to actual or inferred

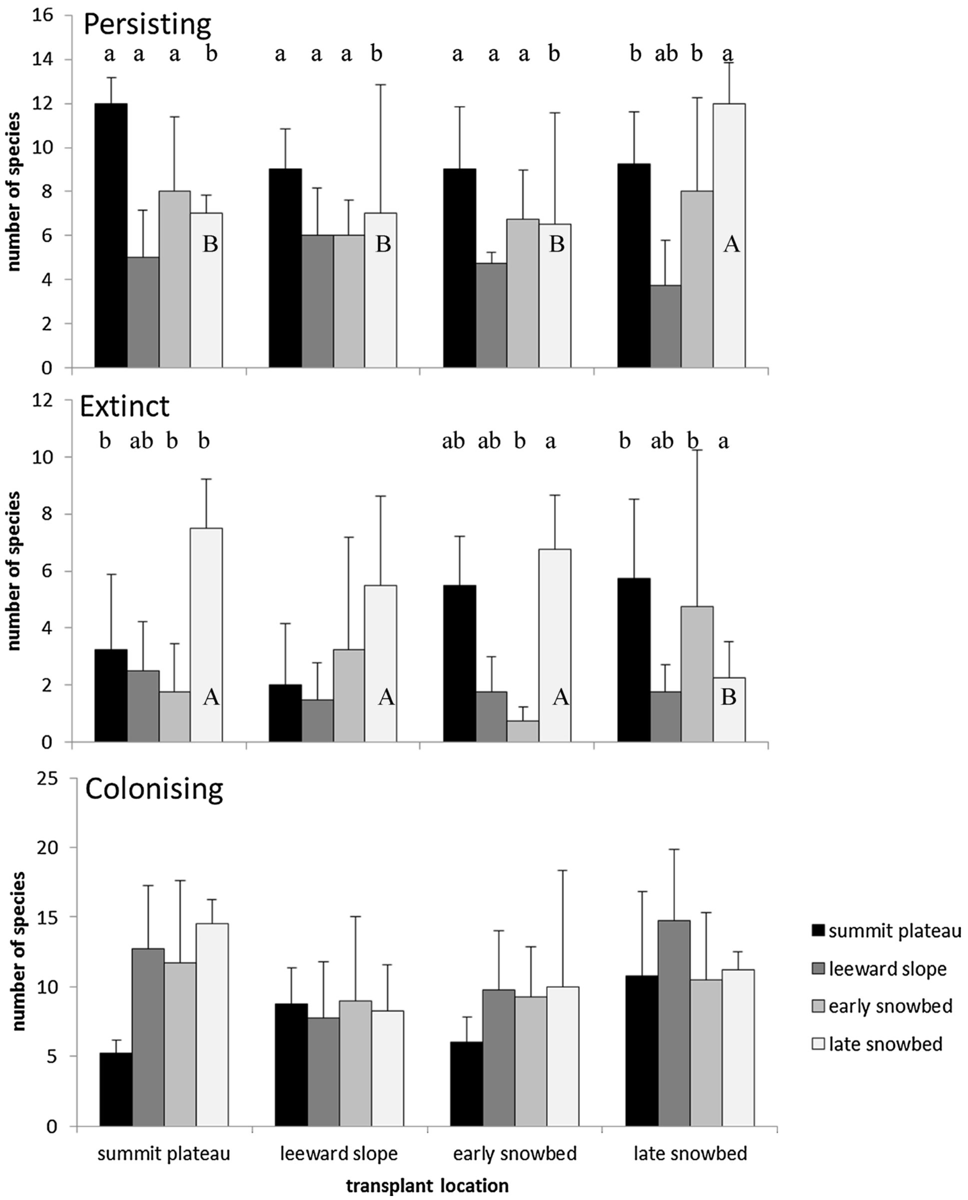


Fig. 5. The origin and fate of species recorded in turves seven years following transplantation among four topographic zones differing in snow cover on the Rock and Pillar Range, South Island, New Zealand. "Persisting" refers to species originally recorded on the turf at the time of transplantation. "Extinct" refers to original species that could no longer be found on the turves. All new species recorded since transplantation are in the "colonising" category. The legend indicates turf origin. Letters indicate significant differences (LSD, $p < 0.05$) following Analysis of Covariance, controlling for original species richness; lower–case indicate differences between turves within a topographic zone, upper–case indicate significant differences between zones for turves derived from the same topographic zone. Analysis of Covariance detected no differences in number of colonising species. Data are averages of four turves per source-location combination from two adjacent catchments. Error bars represent one standard deviation.

Table 2
General Linear Model analysis of mean leaf production per turf over three growing seasons in three focal *Celmisia* species transplanted into four topographic zones differing in snow cover on the Rock and Pillar Range, South Island, New Zealand. The two replicate catchments were treated as a block effect.

Model term	Mean Square	*F, d.f.*	*p*
Intercept	11195.83	$F_{1,119} = 1891.1$	< 0.001
Catchment	47.598	$F_{1,119} = 8.040$	< 0.01
Season	62.211	$F_{2,119} = 5.254$	< 0.01
Zone	185.63	$F_{3,119} = 10.452$	< 0.001
Species	1037.7	$F_{2,119} = 87.639$	< 0.001
Season × zone	56.716	$F_{6,119} = 1.597$	> 0.05
Species × zone	262.29	$F_{6,119} = 7.384$	< 0.001
Season × species	24.223	$F_{4,119} = 1.023$	> 0.05
Error	704.52		

climate change (Elmendorf et al., 2015). However, novel biotic interactions such as competition and herbivory are also emerging as important factors affecting the ability of species to survive *in situ* or migrate with changing climatic patterns (Olofsson et al., 2009; Hülber et al., 2011; Kaarlejärvi and Olofsson 2014; Alexander et al., 2016). Transplant experiments provide a means by which the effects of these interactions can be assessed under natural conditions, yet such experiments are still rare. Our study is one of the few documenting the long–term fates of alpine species and communities following transplantation to different topographic zones along a snow cover gradient, and while we only studied change in two adjacent catchments, our inclusion of four topographic zones in each, in a reciprocal transplant design, allowed for relatively fine scale analyses of species and community responses.

To a certain degree in our study, environment accounted for the observed changes; the snowbed specialist *Celmisia haasti* showed the least growth and snowbed turves demonstrated the greatest number of species extinctions when transplanted to more exposed zones. While the snowbed communities studied here were richer in species and experienced warmer temperatures than in many northern hemisphere snowbeds (e.g. Hülber et al., 2011; Carbognani et al., 2014), our study confirmed that these communities can be strongly affected by reduced snow cover (Kipkeev et al., 2015; Matteodo et al., 2016), making them particularly vulnerable to climate change (Björk and Molau 2006). The difference in snow cover between our most protected late snowbed zone and most exposed summit plateau zone, at the time our experiment was established, was around two times the current predicted reduction in

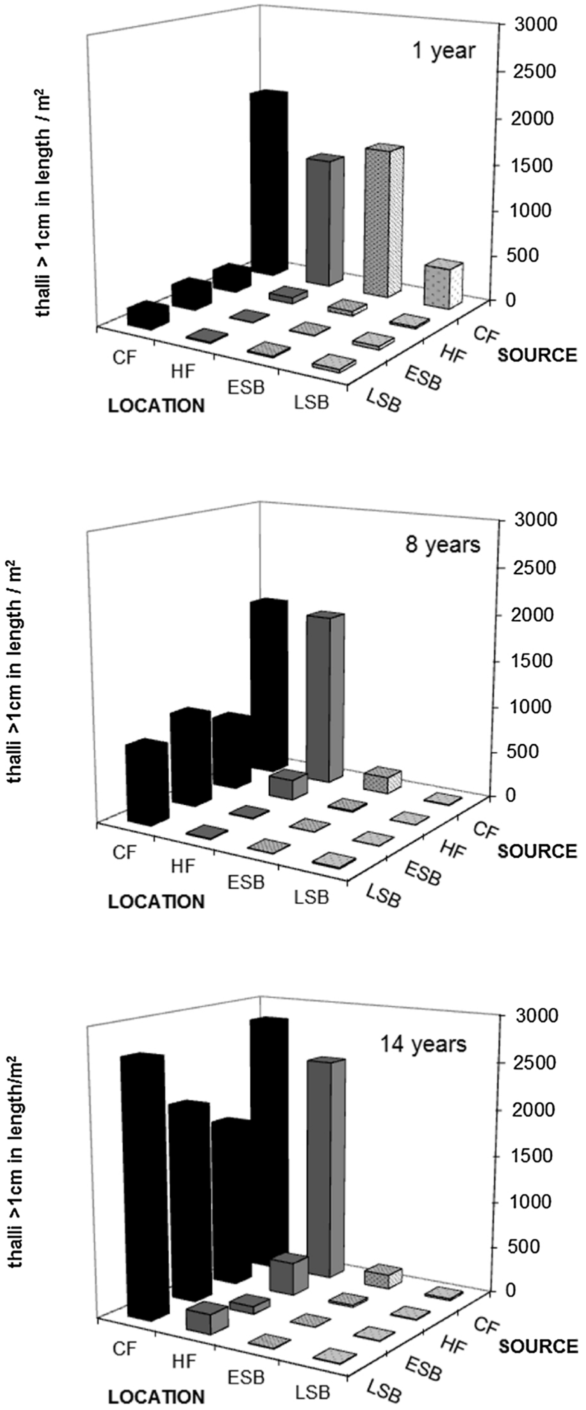


Fig. 6. Changes in the abundance of *Thamnolia vermicularis* thalli > 1 cm in length on turves transplanted within and among topographic zones differing in snow cover on the Rock and Pillar Range, South Island, New Zealand, one, eight, and 14 years after transplantation. Each bar is the sum of four replicate 0.25 m^2 turves. CF: summit plateau cushionfield, HF: leeward slope herbfield, ESB: early snowbed, LSB: late snowbed. Turves with the same source and location are controls that were relocated within the same vegetation type and topographic zone.

snow cover of 22% for alpine southern New Zealand under climate change projections (Mullan et al., 2008; Hendrikx et al., 2012), so represents an extreme scenario. However, differences between the early and late snowbed and leeward slope zones represent realistic levels of future snow cover reduction. Given the current climate change predictions, it is highly unlikely that snowbeds in our study area will completely disappear, but they are likely to contract and fragment, exposing marginal areas. It is reassuring then, that a number of snowbed species were persisting in more exposed zones even after seven years, suggesting that these plant communities may have some inherent resilience to altered snow cover. A growing number of studies, both in New Zealand and elsewhere, have found that alpine and arctic species can demonstrate broad tolerances beyond their apparent microclimatic niche, and respond individualistically to changes in temperature (Bannister et al., 2005; Hülber et al., 2011; Pieper et al., 2011; Venn et al., 2013; Frei et al., 2014; Sedlacek et al., 2015), therefore future plant community distributions are unlikely to be accurately modelled using species' current climate niches. It is highly likely, though, that further changes in species composition will be detected as we continue to follow the fate of our transplanted turves, as it is clear from this and other studies, that altered climatic conditions trigger both rapid responses and long term changes that are species- and process-specific (Pieper et al., 2011; Farrer et al., 2015; Cannone et al., 2007). Even 25 years following transplantation, the composition of alpine plant communities can still be adjusting to a novel environment, although the rate of change may decrease (Kipkeev et al., 2015).

A distinct advantage of transplant experiments, over *in situ* environmental manipulations such as soil warming, is that the transplanted species are exposed to potentially novel species, not just novel climatic factors (Alexander et al., 2016; Nooten and Andrew 2017). In our study system, biotic interactions are likely to be at least as important as abiotic factors in determining the survival of species under altered snow regimes. While increased exposure to freezing temperatures may have affected the survival of some snowbed species transplanted to the exposed summit plateau, Bannister et al. (2005) showed that *C. haastii* at least was capable of surviving even the harshest frosts recorded in that environment. For this plant species, exposure to increased herbivory seemed to be a critical factor in its decline following transplantation. Species-specific herbivory has been shown to counteract the effects of climate-related species range expansions through the preferential browsing of taller or faster growing species (e.g. Olofsson et al., 2009; Kaarlejärvi and Olofsson, 2014). However, in our study herbivory by an endemic weevil appears to have exacerbated the effects of altered environmental conditions. Quite likely this weevil is normally unable to access *C. haastii* due to phenological mismatch driven by differences in snow cover duration; thus a reduction in the extent of snow at critical times of the year could have serious implications for the survival of this, and other palatable species, in snowbed margins. Interactions with insects have been a critical force shaping the plant world (Schoonhoven et al., 2005) and exposure to new insect herbivores is likely to be an important component of plant survival under future climate conditions (Nooten and Andrew 2017). We strongly recommend that experimental controls restricting invertebrate, as well as vertebrate, herbivores are included in future transplant experiments in alpine areas, and also suggest that measures of invertebrate herbivore damage are explicitly included in monitoring methodologies such as GLORIA, which currently only recommends that grazing impacts by mammals be noted (Pauli et al., 2015).

The inclusion in our study of both vascular and non-vascular focal species allowed us to identify opposing effects of altered snow cover on species. *Thamnolia vermicularis* rapidly colonised turves transplanted to the lichen-rich, exposed summit plateau and equally rapidly declined when subject to increased depth and duration of snow cover in the snowbeds, generally matching other studies of lichen responses to increased snow cover (Bidussi et al., 2016). While a scenario of increasing snow cover is unlikely at our study site, it does highlight the potential value of this species as a global indicator of both increasing and decreasing snow cover. Lichens are the dominant photoautotrophs in high alpine and polar regions, often surpassing vascular plants in both species richness and abundance, and contributing key ecosystem services such as soil stabilisation and nitrogen capture (Ahti 1977; Kappen 2000; Cornelissen et al., 2001). Unlike many vascular plants, they are often small, discrete and easily translocatable, so are highly suitable as indicators of climate change both *in situ* (Sancho et al., 2007) and in transplantation experiments (Bidussi et al., 2016). However, the use of cold-climate lichens as indicators of climate change is often hindered by their slow growth rates, for example McCarthy (2003) recorded growth rates of 0.26–0.41 mm/year for the cosmopolitan crustose lichen, *Rhizocarpon geographicum*. Foliose and fruticose lichens generally have higher growth rates than crustose lichens (Sancho et al., 2007); in keeping with this, we have recorded length increases of up to 5 mm in *Thamnolia vermicularis* thallus branches within a single growing season (Knight and Lord unpub. data). As *Thamnolia vermicularis* is globally widespread and distinctive, we strongly encourage alpine ecologists and environmental managers to include it in monitoring programmes, as a rapid and sensitive bioindicator that can be easily compared among alpine and arctic areas worldwide.

Long term transplant studies can provide detailed information not only concerning species' climatic tolerances and the impact of altered environmental conditions on growth and survival, but also provide insights into the types and significance of potential novel interactions that may emerge as species' ranges shift. These types of studies therefore offer ecologists and modelers critical information beyond that which can be provided by *in situ* climate manipulation experiments (e.g. Walker et al., 2006) and more rapidly than can be obtained from long–term monitoring (e.g. Gottfried et al., 2012). The impacts of climate change on alpine, or in fact any, species or communities, cannot readily be predicted from large-scale climate models when effects like novel competitive interactions and local species-specific herbivory are as important to species survival and compositional change as are environmental tolerances. We strongly encourage alpine ecologists to use transplantation, combined with herbivore and competitor exclusion treatments, to test the relative importance of climatic vs biotic interactions to species distributions. We also suggest that the identification of key functional traits and ecological strategies might be the best approach to explain and predict alpine species' responses to the complex web of interacting factors inherent in future climate scenarios (e.g. Mark et al., 2015; Debouk et al., 2015; Guittar et al., 2016), much as plant strategy schemes are now making sense of ecological variation among plant species and communities worldwide (Díaz et al., 2016).

Acknowledgments

This study has benefitted from several University of Otago research grants and postgraduate scholarships, as well as funding from the Miss E.L. Hellaby Indigenous Grasslands Research Trust. Stewart Bell, Justin Blaikie, Terry Chapin, Richard Clayton, Katherine Dixon, Scott Dunavan, Kate Ladley, Adrienne Markey, Norman Mason, Ian Radford, Bernard Smith, Katrina Spencer, and the late Rob Daly assisted with field work. The Department of Conservation gave permission for the experimental work; the McKenzie family permitted access across their property and the Otago Tramping and Mountaineering Club provided use of Leaning Lodge for accommodation during fieldwork.

References

Ahti, T., 1977. Lichens of the boreal coniferous zone. In: Seaward, M.R.D. (Ed.), Lichen Ecology. Academic Press, London, pp. 145–181.

Alexander, J.M., Diez, J.M., Levine, J.M., 2016. Novel competitors shape species' responses to climate change. Nature 525, 515–518.

Bannister, P., Maegli, T., Dickinson, K.J.M., Halloy, S.R.P., Knight, A., Lord, J.M., Mark, A.F., Spencer, K.L., 2005. Will loss of snow cover during climatic warming expose New Zealand alpine plants to increased frost damage? Oecologia 144, 245–256.

Bidussi, M., Solhaug, K.A., Gauslaa, Y., 2016. Increased snow accumulation reduces survival and growth in dominant mat-forming arctic-alpine lichens. Lichenologist 48, 237–247.

Björk, R.G., Molau, U., 2007. Ecology of alpine snowbeds and the impact of global change. Arct. Antarct. Alp. Res. 39, 34–43.

Bliss, L.C., Mark, A.F.M., 1974. High-alpine environments and primary production on the Rock and Pillar Range Central Otago, New Zealand. N. Z. J. Bot. 12, 445–483.

Callaghan, T.V., Bjorn, L.O., Chernov, Y., Chapin, T., Christensen, T.R., Huntley, B., Ims, R.A., Johansson, M., Jolly, D., Jonasson, S., Matveyeva, N., Panikov, N., Oechel, W., Shaver, G., Elster, J., Jonsdottir, I.S., Laine, K., Taulavuori, K., Taulavuori, E., Zockler, C., 2004. Responses to projected changes in climate and UV-B at the species level. Ambio 33, 418–435.

Cannone, N., Sgorbati, S., Guglielmin, M., 2007. Unexpected impacts of climate change on alpine vegetation. Front. Ecol. Environ. 5, 360–364.

Carbognani, M., Petraglia, A., Tomaselli, M., 2012. Influence of snowmelt time on species richness, density and production in a late snowbed community. Acta Oecol. 43, 113–120.

Carbognani, M., Tomaselli, M., Petraglia, A., 2014. Current vegetation changes in an alpine late snowbed community in the south-eastern Alps (N-Italy). Alpine Bot. 124, 105–113.

Cieraad, E., McGlone, M.S., 2014. Thermal environment of New Zealand's gradual and abrupt treeline ecotones. N. Z. J. Ecol. 38, 12–25.

Cooper, E.J., 2014. Warmer shorter winters disrupt Arctic terrestrial ecosystems. Annu. Rev. Ecol. Evol. Syst. 45, 271–295.

Cornelissen, J.H.C., Callaghan, T.V., Alatalo, J.M., 2001. Global change and arctic ecosystems: is lichen decline a function of increases in vascular plant biomass? J. Ecol. 89, 984–994.

Díaz, S., Kattge, J., Cornelissen, J.H.C., Wright, I.J., Lavorel, S., Dray, S., Reu, B., Kleyer, M., Wirth, C., Prentice, I.C., Garnier, E., Bönisch, G., Westoby, M., Poorter, H., Reich, P.B., Moles, A.T., Dickie, J., Gillison, A.N., Zanne, A.E., Chave, J., Wright, S.J., Sheremet'ev, S.N., Jactel, H., Baraloto, C., Cerabolini, B., Pierce, S., Shipley, B., Kirkup, D., Casanoves, F., Joswig, J.S., Günther, A., Falczuk, V., Rüger, N., Mahecha, M.D., Gorné, L.D., 2016. The global spectrum of plant form and function. Nature 529, 167–171.

Debouk, H., de Bello, F., Sebastià, M.-T., 2015. Functional trait changes, productivity shifts and vegetation stability in mountain grasslands during a short-term warming. PLoS One 10, e0141899.

Dumbleton, L.J., 1967. Winter dormancy in New Zealand biota and its paleoclimatic implications. N. Z. J. Bot. 5, 211–222.

Elmendorf, S.C., Henry, G.H.R., Hollister, R.D., Fosaa, A.M., Gould, W.A., Hermanutz, L., Hofgaard, A., Jónsdóttir, I.S., Jorgenson, J.C., Lévesque, E., Magnusson, B., Molau, U., Myers-Smith, I.H., Oberbauer, S.F., Rixen, C., Tweedie, C.E., Walker, M.D., 2015. Experiment, monitoring, and gradient methods used to infer climate change effects on plant communities yield consistent patterns. Proc. Natl Acad. Sci. U. S. A. 112, 448–452.

Farrer, E.C., Ashton, I.W., Spasojevic, M.J., Fu, S., Gonzalez, D.J.X., Suding, K.N., 2015. Indirect effects of global change accumulate to alter plant diversity but not ecosystem function in alpine tundra. J. Ecol. 103, 351–360.

Fiddes, S.L., Pezza, A.B., Barras, V., 2015. A new perspective on Australian snow. Atmos. Sci. Lett. 16, 246–252.

Forrest, J.R.K., Thomson, J.D., 2011. An examination of synchrony between insect emergence and flowering in Rocky Mountains meadows. Ecol. Mon. 81, 469–491.

Frei, E.R., Ghazoul, J., Matter, P., Heggli, M., Pluess, A.R., 2014. Plant population differentiation and climate change: responses of grassland species along an elevational gradient. Global Change Biol. 20, 441–455.

Galloway, D.J., Aptroot, A., 1995. Bipolar lichens: a review. Cryptogam. Bot. 5, 184–191.

Galloway, D.J., 2007. Flora of New Zealand, Lichens, 2nd edition. Government Printer, Wellington.

Gobiet, A., Kotlarski, S., Beniston, M., Heinrich, G., Rajczak, J., Stoffel, M., 2014. 21 st century climate change in the European Alps – a review. Sci. Total Environ. 493, 1138–1151.

Gottfried, M., Pauli, H., Futschik, A., Akhalkatsi, M., Barančok, P., Alonso, J.L.B., Coldea, G., Dick, J., Erschbamer, B., Calzado, M.R.F., Kazakis, G., Krajči, J., Larsson, P., Mallaun, M., Michelsen, O., Moiseev, D., Moiseev, P., Molau, U., Merzouki, A., Nagy, L., Nakhutsrishvili, G., Pedersen, B., Pelino, G., Puscas, M., Rossi, G., Stanisci, A., Theurillat, J.-P., Tomaselli, M., Villar, L., Vittoz, P., Vogiatzakia, I., Grabherr, G., 2012. Continent-wide response of mountain vegetation to climate change. Nat. Clim. Change 2, 111–115.

Grabherr, G., Gottfried, M., Pauli, H., 1994. Climate effects on mountain plants. Nature 369, 448.

Guittar, J.A., Goldberg, D., Klanderud, K., Telford, R.J., Vandvik, V., 2016. Can trait patterns along gradients predict plant community responses to climate change? Ecology 97, 2791–2801.

Hülber, K., Bardy, K., Dullinger, S., 2011. Effects of snowmelt timing and competition on the performance of alpine snowbed plants. Perspect. Plant Ecol. 13, 15–26.

Halloy, S.R.P., Mark, A.F., 2003. Climate-change effects on alpine plant biodiversity: a New Zealand perspective on quantifying threats. Arct. Antarct. Alp. Res. 35, 248–254.

Hendrikx, J., Hreinsson, E.Ö., Clark, M.P., Mullan, A.B., 2012. The potential impact of climate change on seasonal snow in New Zealand: part I – an analysis using 12 GCMs. Theor. Appl. Climatol. 110, 607–618.

Holdsworth, D.K., Mark, A.F., 1990. Water and nutrient input:output budgets – effects of plant cover at seven sites in upland snow tussock grasslands of Eastern and Central Otago, New Zealand. J. R. Soc. N. Z. 20, 1–24.

IPCC, 2014. Climate Change 2014: Synthesis Report. Contribution of Working Groups I, II and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. In: Core Writing Team, Pachauri, R.K., Meyer, L.A. (Eds.), IPCC, Geneva, Switzerland 151 pp.

Jonas, T., Rixen, C., Sturm, M., Stoeckli, V., 2008. How alpine plant growth is linked to snow cover and climate variability. J. Geophys. Res. 113, G03013.

Kärnefelt, E.I., Thell, A., 1995. Genotypical variation and reproduction in natural populations of *Thamnolia*. Bibl. Lichenol. 58, 213–234.

Kaarlejärvi, E., Olofsson, J., 2014.. Concurrent biotic interactions influence plant performance at their altitudinal distribution margins. Oikos 123, 943–952.

Kappen, L., 2000. Some aspects of the great success of lichens in Antarctica. Antarct. Sci.

12, 314–324.

Kipkeev, A.M., Cherednichenko, O.V., Tekeev, D.K., Onipchenko, V.G., 2015. Rate of microsuccessions: structure and floristic richness recovery after sod transplantation in alpine plant communities. Z. Obshch. Biol. 76, 461–474.

Klein, G., Vitasse, Y., Rixen, C., Marty, C., Rebetez, M., 2016. Shorter snow cover duration since 1970 in the Swiss Alps due to earlier snowmelt more than to later snow onset. Clim. Change 139, 637–649.

Lapp, S., Byrne, J., Townshend, I., Kienzle, S., 2005.. Climate warming impacts on snowpack accumulation in an alpine watershed. Int. J. Climatol. 25, 521–536.

Long, T., 2006. The Role of an Insect Herbivore (Curculionidae: *Irenimus posticalis*) in Determining Alpine Plant Distributions. Unpublished Postgraduate Diploma in Science dissertation. University of Otago, Department of Botany.

Lord, J.M., Knight, A., Bannister, J.M., Ludwig, L.R., Malcolm, W.M., Orlovich, D.A., 2013. Rediscovery of pycnidia in *Thamnolia vermicularis*: implications for chemotype occurrence and distribution. Lichenologist 45, 397–411.

Mark, A.F., Korsten, A.C., Guevara, D.U., Dickinson, K.J.M., Humar-Maegli, T., Pascale, M., Halloy, S.R.P., Lord, J.M., Venn, S.E., Morgan, J.W., Whigham, P.A., Nielsen, J.A., 2015. Ecological responses after 52 years to an experimental snow fence in high-alpine cushionfield, Old Man Range, south-central New Zealand. Arct. Antarct. Alp. Res. 47, 751–772.

Mark, A.F., 2012. Above the Treeline. Craig Potton Publishing, Nelson, New Zealand.

Matteodo, M., Ammann, K., Verrecchia, E.P., Vittoz, P., 2016. Snowbeds are more affected than other aubalpine-alpine plant communities by climate change in the Swiss Alps. Ecol. Evol. 6, 6969–6982.

McCarthy, D.P., 2003. Estimating lichenometric ages by direct and indirect measurement of radial growth: a case study of *Rhizocarpon* agg. at the Illecillewaet Glacier, British Columbia. Arct. Antarct. Alp. Res. 35, 203–213.

McGlone, M.S., Heenan, P.B., 2013. Evolution of New Zealand alpine and open-habitat plant species during the late Cenozioc. N. Z. J. Ecol. 37, 105–113.

Mernild, S.H., Liston, G.E., Hiemstra, C.A., Malmros, J.K., Yde, J.C., McPhee, J., 2017. The andes cordillera, Part I: snow distribution, properties, and trends (1979–2014). Int. J. Climatol. 37, 1680–1698.

Mote, P.W., Hamlet, A.F., Clark, M.P., Lettenmaier, D., 2005. Declining mountain snowpack in western North America. Bull. Amer. Meteor. Soc. 86, 39–49.

Mullan, B., Wratt, D., Dean, S., Hollis, M., Allan, S., Williams, T., Kenny, G., 2008. Climate Change Effects and Impacts Assessment: A Guidance Manual for Local Government in New Zealand, 2nd ed. Ministry for the Environment, Wellington, New Zealand.

Musselman, K.N., Clark, M.P., Liu, C., Ikeda, K., Rasmussen, R., 2017. Slower snowmelt in a warmer world. Nat. Clim. Change 7. http://dx.doi.org/10.1038/NCLIMATE3225.

Nooten, S.S., Andrew, N.R., 2017. Transplant experiments –a powerful method to study climate change impacts. In: Johnson, S.N., Jones, T.H. (Eds.), Global Climate Change and Terrestrial Invertebrates. Wiley-Blackwell, Chichester, UK, pp. 46–67.

Olofsson, J., Oksanen, L., Callaghan, T., Hulme, P.E., Oksanen, T., Suominen, O., 2009. Herbivores inhibit climate-driven shrub expansion on the tundra. Global Change Biol. 15, 2681–2693.

Pérez, F.L., 1991. Particle sorting due to off-road vehicle traffic in a high Andean Paramo. Catena 18, 239–254.

Patsiou, T.S., Conti, E., Zimmerman, N.E., Theodoridis, S., Randin, C.F., 2014. Topo-climatic microrefugia explain the persistence of a rare endemic plant in the Alps during the last 21 millennia. Global Change Biol. 20, 2286–2300.

Pauli, H., Gottfried, M., Dullinger, S., Abdaladze, O., Akhalkatsi, M., Alonso, J.L.B., Coldea, G., Dick, J., Erschbamer, B., Calzado, R.F., Ghosn, D., Holten, J.I., Kanka, R., Kazakis, G., Kollár, J., Larsson, P., Moiseev, P., Moiseev, D., Molau, U., Mesa, J.M., Nagy, L., Pelino, G., Puşcaş, M., Rossi, G., Stanisci, A., Syverhuset, A.O., Theurillat, J.-P., Tomaselli, M., Unterluggauer, P., Villar, L., Vittoz, P., Grabherr, G., 2012. Recent plant diversity changes on Europe's mountain summits. Science 336, 353–355.

Pauli, H., Gottfried, M., Lamprecht, A., Niessner, S., Rumpf, S., Winkler, M., Steinbauer, K., Grabherr, G., 2015. The GLORIA Field Manual – Standard Multi-Summit Approach, Supplementary Methods and Extra Approaches, 5th ed. GLORIA-Coordination, Austrian Academy of Sciences & University of Natural Resources and Life Sciences, Vienna.

Petraglia, A., Carbognani, M., Petit Bon, M., Delnevo, N., Chiari, G., Tomaselli, M., 2014. Responses of flowering phenology of snowbed plants to an experimentally imposed extreme advanced snowmelt. Plant Ecol. 7, 759–768.

Pieper, S.J., Loewen, V., Gill, M., Johnstone, J.F., 2011. Plant responses to natural and experimental variations in temperature in alpine tundra Southern Yukon, Canada. Arct. Antarct. Alp. Res. 43, 442–456.

Sancho, L.G., Green, T.G.A., Pintado, A., 2007. Slowest and fastest: extreme range in lichen growth rates supports their use as an indicator of climate change in Antarctic. Flora 202, 667–673.

Sansom, C.E., Larsen, L., Evans, A.C., Perry, N.B., 2013. Epimanool and a new 2,6-dideoxy-hexopyran-3-ulose deriviative from *Celmisia viscosa*. Nat. Prod. Commun. 8, 689–692.

Sato, M.J., 1968. The mixture ratio of the lichen genus *Thamnolia* in Tasmania and New Guinea. J. Jap. Bot. 43, 328–334.

Scherrer, D., Körner, C., 2010. Infra-red thermometry of alpine landscapes challenges climatic warning projections. Global Change Biol. 16, 2602–2613.

Schoonhoven, L.M., van Loon, J.J.A., Dicke, M., 2005. Insect-Plant Biology, 2nd ed. Oxford University Press, Oxford, U.K.

Sebastià, M.-T., Kirwan, L., Connolly, J., 2008. Strong shifts in plant diversity and vegetation composition in grassland shortly after climate change. J. Veg. Sci. 19, 299–306.

Sedlacek, J., Wheeler, J.A., Cortés, A.J., Bossdorf, O., Hoch, G., Lexer, C., Wipf, S., Karrenberg, S., van Kleunen, M., Rixen, C., 2015. The response of the alpine dwarf shrub *Salix herbacea* to altered snowmelt timing: lessons from a multi-site transplant experiment. PLoS One 10.

Talbot, J.M., Mark, A.F., Wilson, J.B., 1992. Vegetation-environment relations in snowbanks on the rock and pillar range central otago, New Zealand. N. Z. J. Bot. 30, 271–301.

Thomson, J.W., 1984. American Arctic Lichens. 1. The Macrolichens. Cambridge University Press, New York.

Venn, S., Morgan, J., Lord, J.M., 2013. Foliar freezing resistance of Australian alpine species over the growing season. Aust. Ecol. 38, 152–161.

Walker, M.D., Wahren, C.H., Hollister, R.D., Henry, G.H.R., Ahlquist, L.E., Alatalog, J.M., Bret-Harteh, M.S., Calef, M.P., Callaghan, T.V., Carrolla, A.B., Epstein, H.E., Jónsdóttirk, I.S., Klein, J.A., Magnússon, B., Molau, U., Oberbauer, S.F., Rewan, S.P., Robinson, C.H., Shaver, G.R., Suding, K.N., Thompson, C.C., Tolvanen, A., Totland, O., Turner, P.L., Tweedie, C.E., Webber, P.J., Wookey, P.A., 2006. Plant community responses to experimental warming across the tundra biome. Proc. Natl. Acad. Sci. 103, 1342–1346.

Wheeler, J.A., Cortés, A.J., Sedlacek, J., Karrenberg, S., van Kleunen, M., Wipf, S., Hoch, G., Bossdorf, O., Rixen, C., 2016. The snow and the willows: earlier spring snowmelt reduces performance in the low-lying alpine shrub *Salix herbacea*. J. Ecol. 104, 1041–1050.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 62–70

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Research article

Plastic and genetic responses to shifts in snowmelt time affects the reproductive phenology and growth of *Ranunculus acris*[☆]

Nicola Delnevo[a,1], Alessandro Petraglia[b], Michele Carbognani[b], Vigdis Vandvik[a], Aud H. Halbritter[a,*]

[a] Department of Biology, University of Bergen, POBox 7803, 5020 Bergen, Norway
[b] Department of Chemistry, Life Sciences and Environmental Sustainability, University of Parma, 11/A Parco Area delle Scienze, 43124 Parma, Italy

ARTICLE INFO

Keywords:
Climate change
Context-dependency
Environmental gradient
Local adaptation
Space-for-time

ABSTRACT

Changes in both temperature and precipitation will affect snowmelt time at high elevation, thereby influencing plant reproduction and growth. Species can respond to changed climate with phenotypic plasticity or genetic adaptation, and these responses might vary at different levels of advanced and delayed snowmelt time. Here we mimicked future climate change projections for western Norway by transplanting individuals of *Ranunculus acris* towards warmer, wetter and warmer & wetter climates. And we replicated the experiment along regional-scale temperature and precipitation gradients. This setup resulted in both advanced (warmer and warmer & wetter transplants) and delayed (wetter transplants) snowmelt in the experimental sites. We recorded phenological development and growth over one growing season.

The reproductive phenology of the transplanted *R. acris* individuals was affected by both phenotypic plasticity and genetic differences between populations of different origins, while growth showed only plastic responses. Plants expressed high plasticity to both advanced and delayed snowmelt time by acceleration of the onset of buds, flowers and fruits. Only the plants from wet and high-elevation sites showed a small response to advanced SMT. The late snowmelt time these populations experience could potentially cause high selection pressure leading to more constrains in plasticity. When grown under common conditions, plants from late snowmelt sites responded with earlier onset of phenological development, suggesting that the timing of snowmelt exerts strong selection on reproduction. To project species fates under future climate we need to consider the interplay between genetic adaptation and plastic responses under different climate contexts, especially towards the species range limits.

1. Introduction

Arctic and alpine ecosystems are characterized by seasonal snow cover, which protects plants against low temperature, desiccation, and ice blast during the cold season, and which also determines the duration of the snow-free period and thus defines the time available for growth and reproduction (Bliss, 1962; Jonas et al., 2008; Körner, 2003; Larcher, 2003). The snowmelt time (hereafter SMT) is regarded one of the most important drivers for growth and reproduction at high latitudes and in the mountains, affecting community composition and distribution (e.g. Braun-Blanquet, 1932; Gjærevoll, 1956; Scott and Billings, 1964). Climate change, such as increased temperature and changes in precipitation patterns, is likely to alter the timing of snowmelt, and hence the time available for plants to grow and reproduce, which can have both immediate and long-term consequences for species composition and distribution (Cooper et al., 2011; Heegaard, 2002).

The combination of temperature and precipitation influence the SMT in arctic and alpine ecosystems in complex ways. Winter accumulation of snow defines the amount of snow that has to melt in spring, while spring temperature affect the rate of snowmelt. Current climate projections predict changes in the mean and variances of both temperature and precipitation (IPCC, 2014), especially for alpine environments at high latitudes (AICA, 2004; Stocker, 2013). Climate projections for the oceanic regions in north-western Europe indicate a

---

[☆] This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author.

E-mail addresses: n.delnevo@ecu.edu.au (N. Delnevo), alessandro.petraglia@unipr.it (A. Petraglia), michele.carbognani@unipr.it (M. Carbognani), vigdis.vandvik@uib.no (V. Vandvik), aud.halbritter@uib.no (A.H. Halbritter).

[1] Present address: School of Science, Edith Cowan University, 270 Joondalup Dr, 6027 Joondalup, WA, Australia.

http://dx.doi.org/10.1016/j.ppees.2017.07.005
Received 31 December 2016; Received in revised form 23 June 2017; Accepted 25 July 2017

warming rate greater than the global average coupled with marked increases in precipitation and in the snow-to-rain ratio (Hanssen-Bauer et al., 2009). These predicted increases in both temperature and snowfall may have opposing effects on SMT, with increased snowfall delaying and higher spring temperature advancing SMT. (Bjorkman et al., 2015). These changes also affect the thermal conditions experienced by plants (Jonas et al., 2008). Indeed, earlier SMT due to warming may expose plants to colder conditions and increase the risk of frost events (e.g. Inouye, 2008), later SMT due to increased snow accumulation in winter provides plants with a shorter, but warmer growing season, as long as the season does not become too short (Carbognani et al., 2016). Consequently, given the complex interplay among the climatic factors, it is challenging to predict how plants will respond to these concurrent climatic changes, and especially their climatic tolerance limits under novel climatic conditions (Bjorkman et al., 2015; Donnelly et al., 2012; Franks et al., 2014).

Plant populations' abilities to respond to fluctuations and change in SMT in space and time is limited by their phenotypic plasticity, genetic adaptations, or a combination of both (Donnelly et al., 2012; Forrest and Miller-Rushing, 2010; Franks et al., 2014). In the short-term, phenotypic plasticity may allow plants to counter the detrimental effects of changing environmental conditions and buffer populations against climate fluctuations (Nicotra et al., 2010). In the long-term population adaptive potential is key for species persistence and community stability (Byars and Hoffmann, 2009). Numerous studies have investigated the plastic response of growth and reproduction to changes in SMT in arctic and alpine habitats (Carbognani et al., 2012; Cooper et al., 2011; e.g. Galen and Stanton, 1995; Heegaard, 2002; Hülber et al., 2010; Petraglia et al., 2014; Totland and Alatalo, 2002). Generally, advanced SMT prolongs the pre-flowering period, while delayed SMT accelerates the phenological development, whereas growth responses are very variable (reviewed in Wipf and Rixen, 2010). However, responses vary among species (Cooper et al., 2011; Totland and Alatalo, 2002; Wipf et al., 2009) and with site-specific factors (Carbognani et al., 2016), for example soil moisture (Gimenez-Benavides et al., 2007; Winkler et al., 2016). Responses to changes in SMT can also be affected by local adaptations exerted from changes in selection pressure along climate gradients. Common garden and reciprocal transplant experiments, the classical approach to disentangle phenotypic plasticity and genetic differentiation (Merilä and Hendry, 2014), provide evidence for adaptation in reproductive phenology to SMT (Haggerty and Galloway, 2011; Hall and Willis, 2006; Kawai and Kudo, 2011; Stinson, 2004). Early-flowering plants might be at a disadvantage in areas where snow melts early, due to risk for late-frost events early in the season (Thomson, 2010). In late snowmelt habitats, on the other hand, flowering early might give a selective advantage, allowing to complete the life cycle during the short growing season (Anderson et al., 2012; Stinson, 2004). However, the evidence of such adaptive responses to SMT are less frequent (Donnelly et al., 2012; Frei et al., 2014; Stanton and Galen, 1997), and typically species-specific (Byars and Hoffmann, 2009).

Although some patterns in species responses to shift in SMT are emerging, the consistently high species- and site-specific variation begs the question of both the generality of the patterns and of the underlying drivers of the variation. One way to address this challenge is to explicitly and systematically address if and how responses vary across species and sites, e.g., by replicating the same experiments along environmental gradients (Dunne et al., 2003), an approach also known as "distributed experiments" (Borer et al., 2014).

This study was conducted in south-western Norway, a fjord landscape characterized by strong regional-scale climate gradients: a precipitation gradient from the continental east to the oceanic west coast, and a temperature gradient from the temperate, boreal sea-level to alpine climates at high elevation. In this region, the combination of the two gradients result in early SMT at drier and low-elevation sites, and late SMT at wet and high-elevation sites (Fig. 1). We replicated our experiments across these two gradients to explore how the climate context, as reflected in variation in local SMT, affects reproductive phenology and growth of an alpine forb. Across these sites, we conducted a transplant experiment to explore how populations originating from different climatic contexts will respond to climate change. Since the climate change projections for our region predict increased temperatures and increased precipitation, we transplanted individual plants of our study species, *Ranunculus acris* towards warmer sites, wetter sites and warmer & wetter sites. We recorded phenological response and growth of the transplanted individuals compared to local populations in the origin site and in the transplanted localities over one growing season. Specifically, we ask: (1) does *R. acris* respond to a change SMT in reproductive phenology and growth? And (2) to what extent are such plastic responses in the reproductive phenology and growth of *R. acris* limited by genetic differences between populations? We expect plants to accelerate phenology (i.e. flower faster after SMT) both when transplanted to sites with advanced SMT due to warming and when transplanted to sites with delayed SMT due to increased snowfall. This is because phenology is affected by temperature, and we expect higher temperatures both at low-elevation sites and later in the season under the same temperature regime. Further, earlier phenology is expected in plants originating from sites with late SMT, as these populations experience strong selection for earlier flowering (Anderson et al., 2012). The predictions for the combined effect of increased temperature and increased precipitation are less clear, because the effects could cancel each other out, or be additive or be multiplicative. Finally, we ask (3) how do these responses in reproductive phenology and growth vary along the climate gradients? We expect plants from harsher conditions, i.e. wet and high-elevation sites, to be more strongly genetically determined in their response to SMT and therefore more constrained in plasticity (Gugger et al., 2015; Vitasse et al., 2013). And we expect the magnitude of response to vary with the magnitude in SMT (Wipf and Rixen, 2010).

2. Material and methods

2.1. Study sites and species

The study was conducted in south-western Norway within semi-natural grassland sites selected to represent a 'climate grid' of sites along broad-scale temperature (three levels, ca. 2 °C apart) and precipitation (four levels ca. 700 mm apart) gradients (Klanderud et al., 2015; Meineri et al., 2014). We selected four sites within the climate grid to represent contrasting climatic contexts (Fig. 1, Table 1); two temperature levels (ca. 7.5 and 9.5 °C mean temperature of the four warmest months), each combined with two precipitation levels (ca. 2000 and 2700 mm annual precipitation). The climate data are interpolated with 100 m resolution from the normal period 1961–1990 (Tveito et al., 2005). All sites are grasslands on calcareous bedrock and were specifically selected to be similar in slope, exposure, grazing pressure, soils, and other ecologically relevant variables, so as to maximize impacts of the regional climate differences (Klanderud et al., 2015). To avoid animal interference with the experimental treatments and the transplanted plants, the experimental areas were fenced during the growing season. At each site, temperature was measured continuously at 30 cm above the ground using UTL-3 version 3.0 temperature loggers (GEOTEST AG, Zollikofen, Switzerland).

SMT advanced along the temperature gradient and was delayed with higher precipitation, with earlier SMT at low-elevation and drier sites (Fig. 1b and c). In 2015, the snow melted in the second half of June in the low-elevation sites and between early July and mid-August at the high-elevation sites (Fig. 1c, Table 1); SMT was 17–50 days earlier at the low-elevation sites and 7–40 days later in wetter sites. Mean daily temperature in the first week after snowmelt was up to 2 °C warmer in the high compared to the low-elevation sites.

Ranunculus acris (Ranunculaceae) is a common perennial herb with a circumpolar distribution; in south-western Norway it grows in a wide range of habitats, but is most abundant on medium dry meadows. *R. acris* flowers are self-incompatible (Totland, 1994). It is an early flowering species and leaves begin to develop precociously after snowmelt

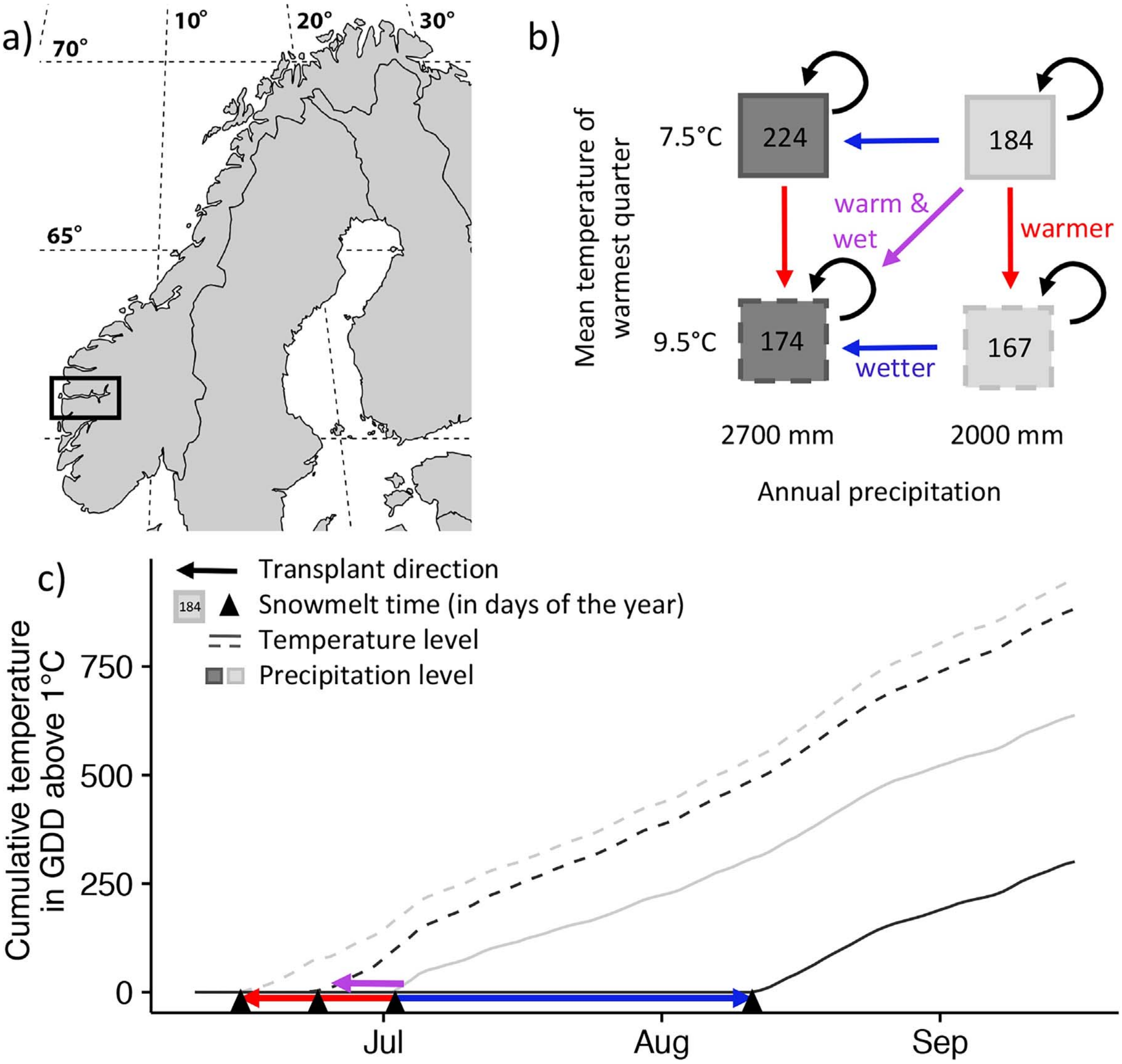


Fig. 1. a) Map of the location of the transplant sites in south-western Norway and b) the transplant design. The four sites shown by boxes, are located in a climate grid representing different temperatures (line type) and precipitation levels (grey shading). The number inside the box indicates the snowmelt date in days of the year. Arrows show the direction of transplant from the origin site (arrow tail) to the destination site (arrow tip) for the warmer (red), wetter (blue) and warmer & wetter transplant treatment (purple) and the local controls (black). Some of the sites thus represent both origin and destination sites. (c) Cumulative temperature (sum of mean daily temperature above 1 °C after snowmelt). The black triangles indicate the timing of snowmelt at each site. The arrows show the direction of transplant from the dry high-elevation site. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 1
Geographic and climatic features of the four transplant sites (according to their temperature and precipitation level); coordinates, elevation, mean annual precipitation, mean temperature of the four warmest months (interpolated with 100 m resolution from the normal period 1961–1990, Tveito et al 2005), snowmelt date in 2015 in day of the year (DOY) and mean daily temperature in the first week after snowmelt are shown.

Temperature level (Elevation) Precipitation level	Cold High dry	Cold High wet	Warm Low dry	Warm Low wet
Latitude (°N)	60.83	60.93	61.09	60.54
Longitude (°E)	7.18	6.41	6.63	6.51
Elevation (m a.s.l.)	1213	1088	769	797
Precipitation (mm)	1925	2725	1848	3029
Temperature (°C)	5.87	6.58	8.77	8.67
Snowmelt date (DOY)	184	224	167	174
Temperature (°C) first week	9.7	9.1	8.5	7.1

and initiate the reproductive development early in the season (Hegland and Totland, 2007). Typically, individuals of high-elevation populations produce a single flower (Totland, 1997). We chose this species because it is one of the most common perennial forbs in the study system, it is dependent on pollinators and well studied (Totland, 1997, 1994, 2001).

2.2. Transplant experiment

In September 2014, at the end of the growing season, individual plants of approximately the same size (6.34 ± 3.07 cm) were collected within populations at the four sites. Each plant was carefully excavated from the ground, surrounded by a soil core of 10 cm in width and 10 cm in depth, to ensure that the roots were not damaged. Plants were randomly allocated to four transplant treatments, representing the expected change in climate: (i) warmer (Fig. 1b, red arrow), (ii) wetter (blue arrow), and (iii) warmer & wetter climate (purple arrow), and a local control (black arrow). Controls were planted back into another location at the original site to control for the transplant effect *per se*. Plants were only transplanted in one direction (not reciprocally), because we were mainly interested in understanding the effects of the projected climate change in the region.

From each site and treatment 30–40 plants were transplanted, adding up to a total of 269 *R. acris* individuals for the entire experiment (see Appendix A for the number of individual plants per transplant site and origin). Fewer plants were transplanted in the wetter transplant treatment at low-elevation sites than originally designed because we were unable to locate enough plants at the dry low-elevation site. Each plant was individually labelled, with labels placed so that they did not shade the plants or interfere with growth.

2.3. Data collection

Phenological observations started in 2015 after SMT and were carried out weekly until mid September, when the plants started to senescence. In these surveys, the phenophases for each target plant was recorded: budding (pedicellate buds emerging from basal rosette on a stalk), flowering (anthesis with completely open flowers) and fruiting (expanding fruits without flowering structures). During the 2015 growing season, 18 plants produced more than one flower, but we considered only the first bud the plant produced. In *R. acris* the different phenophases have limited overlap within individuals and if produced, the second bud appears after the first one is flowering and flowers when the first one is in fruiting (Totland, 1997, 2001). At each site, 6–15 such phenological observations were carried out, depending on the length of the snow-free period. Growth of each individual was assessed by measuring the longest leaf of the rosette at the end of the growing season 2015.

2.4. Data analyses

A generalized linear model was fitted independently for onset of bud, flower and fruits, expressed in days after snowmelt or leaf size as response variables. To account for non-normal distribution of residuals and non-homogeneous variances, we use a Poisson error distributions (appropriate for count data) for the phenological variables and a Gamma error distribution (appropriate for continuous positive data) for leaf size and checked that the assumptions were fulfilled by visual inspection of residual patterns (Zuur et al., 2009). We fitted a separate model for each transplant treatment (warmer, wetter and warmer & wetter), because the transplants are compared to different origin- and destination-control plants, for the different treatments (Fig. 1). Each model contained one transplant treatment and precipitation level (dry vs. wet for warmer treatment) or temperature level (low vs. high elevation for wetter treatment) to define the position in the climate grid and their interaction as predictors. The combined warmer & wetter transplant treatment had no interaction term, because this treatment was replicated only once.

To test for plasticity in the onset of phenology and plant growth, the plants in each transplant treatment were compared to the local control plants from the site of origin (hereafter origin-control). Whereas to test for genetic differentiation between populations in the phenological development and plant growth, the plants in each transplant treatment were compared to the local control plants from the destination site (hereafter destination-control). Only 3 individuals transplanted to wetter sites reached fruiting stage and this phenophase was therefore not analysed for plants transplanted to wetter climate. All the analyses were performed in R version 3.3.2 (R Development Core Team, 2016).

3. Results

3.1. Plastic response in reproductive phenology and growth

Plants in all transplant treatments produced buds, flowers and fruits consistently faster than the corresponding origin-control plants. Budding, flowering and fruiting was advanced on average by 7–11 days when transplanted to warmer climate, compared to the origin-controls (Fig. 2; Appendix B). Plants transplanted to wetter climate advanced the onset of budding and flowering by 9–12 days compared to the corresponding controls. And in the warmer & wetter transplant treatment, plants advanced the onset of each phenophase by 14–21 days relative to the origin-control.

The phenological development of *R. acris* also showed significant differences in relation to SMT. Plants advanced the onset of the phenophases significantly more at drier sites (i.e. where SMT was less advanced; Appendix B; precipitation level term for bud: $P < 0.001$, flower: $P < 0.001$ and fruit: $P < 0.001$) and the onset of flowering at high-elevation sites (i.e. where SMT was more delayed; temperature level term $P = 0.005$). The interactions between the transplant treatments and either precipitation or temperature levels were not significant, indicating consistent plastic responses in the onset of reproduction when transplanted to both warmer and wetter climate irrespective of the change in SMT. The only exception were plants transplanted to wetter climate, where bud swelling was more advanced in high-elevation than low-elevation sites (wetter treatment × temperature level: $P = 0.001$).

Ranunculus acris leaves also responded plastically to the transplant treatments. Plants had twice as large leaves in the warmer treatment and 1.4 times larger in the warmer & wetter treatment, compared to origin-control plants growing at high-elevation sites (Fig. 3 left side). In contrast, leaves of plants transplanted to wetter climate were 0.6-0.8 times smaller compared to the corresponding controls. The negative effect of transplanting to wetter climate on leaf growth was stronger at low compared to high-elevation sites (wetter treatment × temperature level: $P = 0.049$). These changes match patterns in leaf size of *R. acris* in relation to the temperature gradient, as plants at high elevation had significantly smaller leaves than those at low elevation (data not shown).

3.2. Genetic differences in reproductive phenology and growth

Plants transplanted towards warmer climates initiated all phenophases 7–15 days earlier than the local destination-control plants (Fig. 4; Appendix C). In contrast, plants transplanted towards wetter climates initiated budding and fruiting 8–10 days later than the destination-controls. For the warmer & wetter transplant treatment, plants did not differ in the onset of phenology compared to destination-control.

The onset in phenology differed also depending on the delay in SMT. The phenological development was significantly more delayed at low-elevation (i.e. where SMT was less delayed) compared to high-elevation sites (Appendix C; wetter treatment x temperature level for bud: $P = 0.012$; flower: $P = 0.022$). For example, budding was delayed by 15.0 days at low-elevation, but only by 1.1 days at high-elevation sites and flowering was delayed by 17.7 days in the low-, but only by 2.8 days at high-elevation sites.

Leaf size of *R. acris* did not differ from destination-controls under any transplant treatments (Fig. 3 right side, Appendix C). However, low-elevation plants decreased leaf size more compared to corresponding control plants when transplanted to wetter climate than high-elevation plants (wetter treatment × temperature level: $P = 0.015$). Leaf size also depended on the delay in SMT, with longer leaves in low-elevation compared to high-elevation sites (temperature level: $P < 0.001$).

4. Discussion

4.1. Plastic versus genetic responses in ranunculus acris

Ranunculus acris responded plastically in the velocity of the onset of reproductive phenology and growth to both advanced and delayed SMT. Plants transplanted to sites with advanced or delayed snowmelt were exposed to warmer temperature compared to the origin-control plants, which can explain the acceleration in the phenological

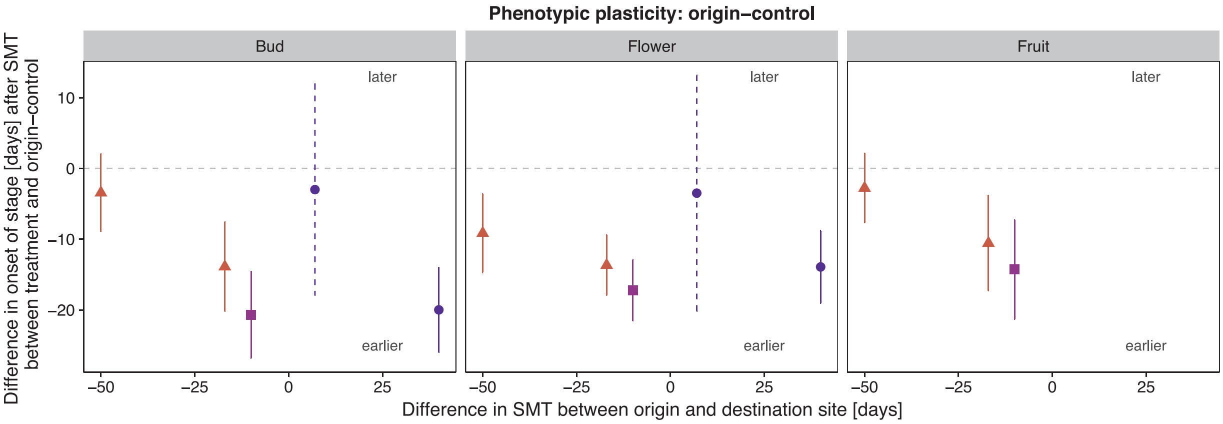


Fig. 2. Plasticity in phenological development as the difference in onset of a phenological stage in days since SMT ± 2SE between treatment and origin-control plants. The x-axis represents the difference in SMT between origin and destination site. The different stages are bud, flower and fruit. The colours and symbols indicate the transplant treatments to: warmer (red triangle), wetter (blue circle) and warmer & wetter (purple square) climate. Points below/above the dashed grey line indicate earlier/later days since SMT for onset in a stage in the transplanted plants compared to the origin-control plants. Closed circles indicate a significant difference between treatment and origin-control plants. Dashed error bars indicate a sample size lower than 6 individuals.

development (Carbognani et al., 2016; Dunne et al., 2003; Petraglia et al., 2014; Wipf and Rixen, 2010). Interestingly, plants in the warmer & wetter transplant treatment responded similarly in the onset of phenology as for the plant in the wetter treatment, but more strongly than the warmer treatment. This suggests that the effect of warmer & wetter conditions may not be fully additive. However, SMT advanced very similarly in both the warm treatment at the dry end of the climate grid and the warmer & wetter treatment and we therefore do not expect a large difference in the onset of phenology between these treatments. Also, leaf size increased in response to advanced SMT, while delayed SMT had a negative effect on growth (Wipf and Rixen, 2010).

Different *R. acris* populations grown under common conditions also responded differently in the onset of reproductive phenology, indicating genetic differentiation in the studied populations. However, the warmer and wetter treatment had opposite effects and plants from sites with later SMT had a faster phenological development than plants from early SMT site. This is consistent with other studies (Anderson et al., 2012; Carbognani et al., 2016; Kawai and Kudo, 2011) and might be due to a selective advantage flowering earlier in environments with a short growing season. Consistently, plants transplanted to warmer & wetter climate showed no such responses, suggesting that the opposite effects of temperature and precipitation change cancel each other out. In contrast to reproductive phenology, the leaf size of plants grown at the same site does not vary depending on origin, suggesting less differential selection and local adaptations in growth across sites. Growth has been suggested to be driven by other factors than climate, such as nutrient and water availability (Sedlacek et al., 2015), and especially in alpine plants, there might be trade-offs between growth and reproduction (Kudo, 1992; Molau, 1993; Theurillat and Schlussel, 2000).

Ranunculus acris had the ability to track variation in environmental conditions via phenotypic plasticity and at the same time showed clear indication of genetic differentiation on the phenological development, in accordance with other studies (Forrest and Miller-Rushing, 2010; Franks et al., 2014; Kawai and Kudo, 2011). Overall, the plastic response was stronger for phenology and growth, but not across all transplant treatments. For example, plants exposed to warmer climates reduced the pre-flowering period by approximately 25% compared to the origin-control plants, but also by 30% compared to destination-control plants. The combination of high plasticity and genetic response to warmer climate in high-elevation plants thus resulted in an enormous reduction in the pre-flowering period. This reduction is much larger than the difference in the pre-flowering time between the control

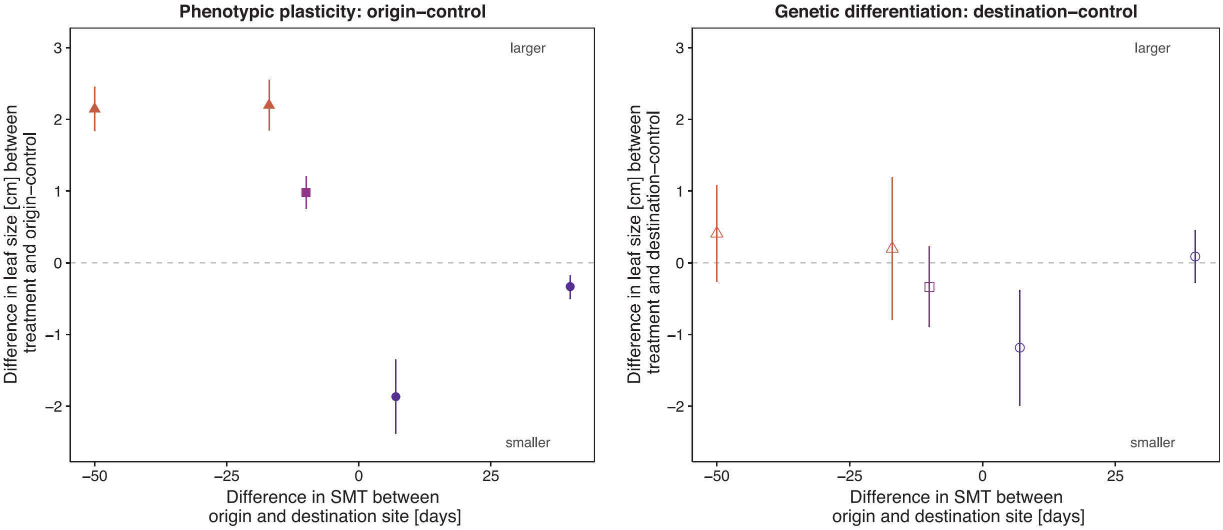


Fig. 3. Plasticity and genetic difference in growth as the difference in leaf size ± 2SE in cm between treatment and origin-control (left) or destination-control (right) plants. The x-axis represents the difference in SMT between origin and destination site. The colours and symbols indicate the transplant treatments to: warmer (red triangle), wetter (blue circle) and warmer & wetter (purple square) climate. Points above/below the dashed grey line indicate larger/smaller leaf size in the transplanted plants compared to the origin-control (left) or destination-control (right) plants. Closed symbols indicate a significant difference between treatment and destination-control plants and open symbols no significant difference.

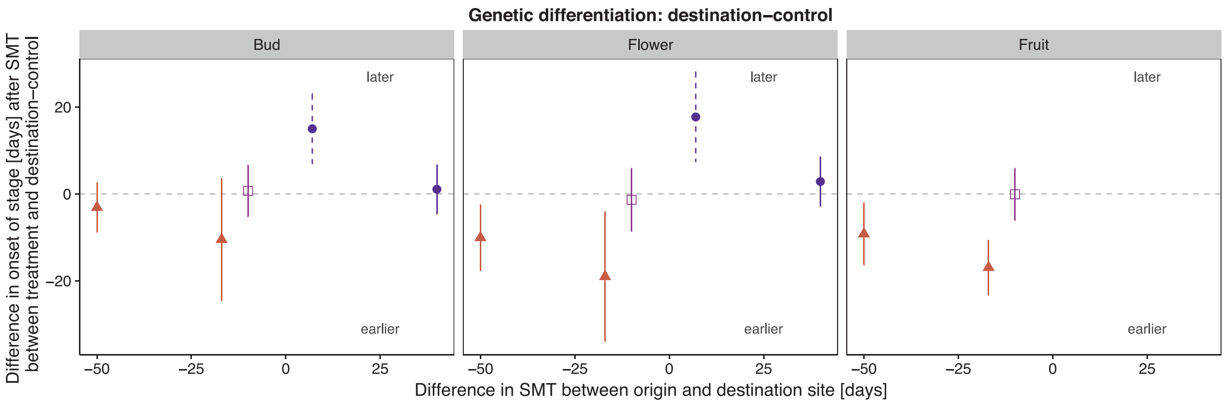


Fig. 4. Genetic difference in phenological development as the difference in onset of a phenological stage in days since SMT ± 2SE between treatment and destination-control site. The x-axis represents the difference in SMT between origin and destination site. The different stages are bud, flower and fruit. The colours and shapes indicate the transplant treatments to: warmer (red triangle), wetter (blue circle) and warmer & wetter (purple square) climate. Points below/above the dashed grey line indicate earlier/later days since SMT for onset in a stage in the transplanted plants compared to the destination-control plants. Closed symbols indicate a significant difference between treatment and destination-control plants and open symbols no significant difference. Dashed error bars indicate a sample size lower than 6 individuals.

plants at high- and low-elevation sites, suggesting that plants from high-elevation sites respond plastically by advancing their phenologies in response to increased temperatures, but also that they are genetically different and progress through the flowering phenology faster than the low-elevation plants. The combined effect is that their phenologies are strongly advanced compared to low-elevation populations in response to warming. However, we did not test the response of the low-elevation plants to warming.

In contrast, plants exposed to wetter climates started the onset of flowering 13% earlier than their origin-control plants, but were still 8% later than destination-control plants. Plants transplanted to warmer & wetter climates reduced the pre-flowering period by 17% compared to origin-control plants and flowered more or less at the same time as destination-control plants. For these two transplant treatments, the plastic response was considerably larger than the genetic response and generally corresponded to the shortened pre-flowering period between the control plants across sites. Overall, *R. acris* responded plastically to the climate treatments in all phenophases, with some exceptions.

4.2. Reproductive phenology and growth across climate contexts

Ranunculus acris showed similar within-population variation in plasticity in the onset of phenology (equally long error bars in Fig. 2) across different SMTs. We expected plants in harsher climate conditions, i.e. wet and high-elevation to be more genetically constrained to the extreme SMT in these sites and therefore exhibit less phenotypic plasticity (Frei et al., 2014; Gugger et al., 2015; Vitasse et al., 2013). Indeed, plants that were exposed to the most severe advance in SMT (i.e. transplanted from wet and high-elevation to wet and low-elevation), shifted the onset in phenology less compared to plants that experienced less severe advance in SMT (Fig. 2). Nine of the plants from wet and high-elevation site transplanted to warmer climate, managed to flower. But four of these did not advance the onset of phenology compared to the origin-control plants (error bar crosses zero line in Fig. 2), suggesting less plasticity in some genotypes of this population. Thus, plants from late SMT sites could be less plastic due to costs to phenotypic plasticity (Ernande and Dieckmann, 2004; Moran, 1992; van Tienderen and van der Toorn, 1991). Also, this population is likely to be close to the range edge of its distribution, which could explain the reduced plasticity. Overall, the largest magnitude in plant response did not correspond to the largest change in SMT the plant experienced, suggesting that plants can be constrained in their responses (see below).

Plants at high- and low-elevation sites also differed in how plastically they responded to delayed SMT. However, we had too few individuals from low-elevation sites to draw any conclusion on the difference in plasticity, which is likely to be due to the low sample size. The strong acceleration in the onset of phenology when SMT was strongly delayed (high-elevation plants) is possibly because later in the season temperatures are higher and speed up plant development (Domènech et al., 2016). Temperature in the first week after snowmelt was 1.6 °C higher in the high than low-elevation site, which could explain this acceleration in development.

Plants exposed to the warmer climate grew twice as tall than the origin-control plants, independent of how much advance in SMT they experienced. The plants exposed to earlier SMT, did not produce larger leaves even though their growing season was 50 days longer, possibly because there is a maximum leaf size that can be reached. Plants transplanted to wetter climate, had 13% shorter leaves at high-elevation and 40% at low-elevation compared to the origin-control plants. Plants at high-elevation generally had smaller leaves, and the negative effect of wetter climate therefore had a smaller effect on leaf size compared to the plants at low-elevation with on average larger leaves.

4.3. Responses to climate change in reproduction phenology

In this study, we were interested in how *R. acris* will respond to future climate, i.e. in a warmer and wetter world. We did not perform a true test of whether or not the species responses to different climates were adaptive, which would require reciprocal transplants experiments (Blanquart et al., 2013; Kawecki, 2008; Merilä and Hendry, 2014). We used individuals grown in their original habitat and can therefore not completely rule out confounding maternal carry-over effect on local adaptation (Kawecki and Ebert, 2004). Local plants did not produce more fruits (data not shown) compared to non-local plants, suggesting no evidence that *R. acris* is locally adapted, however, this interpretation should be taken with caution. *R. acris* plants were taller and produced more fruits when grown at low-elevation, irrespective of origin (controls, warmer and warmer & wetter transplant treatment), suggesting no maladaptation to the predicted future climate in this region. On the contrary, the highly plastic response in the onset of all phenophases, allowed *R. acris* to profit from the longer growing season after advanced SMT. The genetic constraint to start the phenological development earlier did not result in fitness loss, at least not in the year of this study. However, we do not know if early flowering in a less snow-rich year could confer a disadvantage, for example due to frost events (Inouye, 2008; Thomson, 2010). The study sites had a low replication and we can therefore not rule out that some of the effects we saw were due to other abiotic (soil) and biotic factors we did not control for in this experiment. However, the sites were chosen to be as similar as possible to reduce such effects.

All *R. acris* plants drastically reduced the fruit production when grown in the wet high-elevation site in the year of the study, due to late snowmelt. Although the present study was carried out during an extremely snow rich year, further and rapid adaptation might be necessary in wet alpine sites for species to persist (Anderson et al., 2012). Even populations of widespread species may be more vulnerable to extreme climate events (i.e. a snow-rich year) than inferred from their distribution alone. Finally, the interaction of increased precipitation and warmer temperatures are complex and could also result in a delayed SMT, and lead to later flowering in the season and at warmer temperatures. This could accelerate the phenological development and lead to probably much more synchronized flowering across different species within a plant community (Carbognani et al., 2016). Synchrony in flowering among species can attract pollinators and increase reproduction (Moeller and Geber, 2005; Totland, 1993), but may also lead to increased pollen limitation (García-Camacho and Totland, 2009).

Author contribution

ND collected and analysed data and wrote the manuscript, **AP** commented on the manuscript, **MC** analysed data and wrote the manuscript, **VV** designed the experiment and commented on the manuscript and **AHH** designed the experiment, collected data and wrote the manuscript.

Acknowledgements

We thank Jake Alexander and Ørjan Totland for discussions on the experimental design, Deborah Goldberg, Joachim Töpper and three anonymous referees for useful comments on an earlier version of the manuscript and Deborah Davy for checking the language. This work was supported by the Swiss National Science Foundation (SNF) [P2EZP3_155575, 2014] and Olaf Grolle Olsen og Miranda Bødtkers legat [ID 7354, 2015].

Appendix A

Number of individual plants per origin and transplant site.

Transplant site	Origin	Number of plants
High-elevation dry	High-elevation dry	30
Low-elevation dry	High-elevation dry	40
	Low-elevation dry	10
High-elevation wet	High-elevation dry	40
	High-elevation wet	30
Low-elevation wet	High-elevation dry	40
	Low-elevation dry	9
	High-elevation wet	40
	Low-elevation wet	30

Appendix B

Plasticity of reproductive phenology (in days after SMT) and leaf size (in cm) between origin-control plants and transplants to a) warmer, b) wetter and c) warmer & wetter transplant treatment. Analysis of deviance tables on the effects of transplant treatment, precipitation level or temperature level and their interactions on the onset of bud, flower and fruit stage between stages and leaf size of *Ranunculus acris*. Values are χ^2. (Significance codes: *P*-value < 0.001 '***'; < 0.01 '**'; < 0.05 '*'; > 0.05 ' ').

Compare to origin-control	Bud	Flower	Fruit	Leaf size
a) Warmer transplant treatment				
Warmer treatment	45.65 ***	42.36 ***	12.17 ***	83.95 ***
Precipitation level	56.92 ***	55.66 ***	36.22 ***	3.68
Warmer × Precipitation level	1.73	0.002	1.01	1.27
b) Wetter transplant treatment				
Wetter treatment	62.81 ***	19.51 ***	NA	14.73 ***
Temperature level	3.66	7.99 **		36.99 ***
Wetter × Temperature level	10.76 **	2.75		3.86 *
c) Warmer & wetter transplant treatment				
Warmer & wetter treatment	114.73 ***	50.79 ***	24.18 ***	18.38 ***

Appendix C

Genetic response of reproductive phenology (in days after SMT) and leaf size (in cm) between destination-control plants and transplants to a) warmer, b) wetter and c) warmer & wetter transplant treatment. Analysis of deviance tables on the effects of treatment, precipitation level or temperature level and their interactions on the onset of bud, flower and fruit stage between stages and leaf size of *Ranunculus acris*. Values are χ^2. (Significance codes: *P*-value < 0.001 '***'; < 0.01 '**'; < 0.05 '*'; > 0.05 ' ').

Compare to destination-control	Bud	Flower	Fruit	Leaf size
a) Warmer transplant treatment				
Warmer treatment	8.70 **	28.38 ***	16.99 ***	1.23
Precipitation level	49.23 ***	41.93 ***	32.11 ***	3.05
Warmer × Precipitation level	0.73	0.32	0.34	0.19
b) Wetter transplant treatment				
Wetter treatment	8.86 **	7.69 **	NA	2.17
Temperature level	7.58 **	4.68 *		39.37 ***
Wetter × Temperature level	6.33 *	5.22 *		5.87 *
c) Warmer & wetter transplant treatment				
Warmer & wetter treatment	0.12	0.25	< 0.001	1.44

References

AICA, 2004. Impacts of a Warming Arctic, ACIA Overview Report. Cambridge University Press, Cambridge, UK.

Anderson, J.T., Inouye, D.W., McKinney, A.M., Colautti, R.I., Mitchell-Olds, T., 2012. Phenotypic plasticity and adaptive evolution contribute to advancing flowering phenology in response to climate change. Proc. R. Soc. B 279, 3843–3852. http://dx.doi.org/10.1098/rstb.2010.0145.

Bjorkman, A.D., Elmendorf, S.C., Beamish, A.L., Vellend, M., Henry, G.H.R., 2015. Contrasting effects of warming and increased snowfall on Arctic tundra plant phenology over the past two decades. Global Change Biol. 21, 4651–4661. http://dx.doi.org/10.1111/gcb.13051.

Blanquart, F., Kaltz, O., Nuismer, S.L., Gandon, S., 2013. A practical guide to measuring local adaptation. Ecol. Lett. 16, 1195–1205. http://dx.doi.org/10.1111/ele.12150.

Bliss, L.C., 1962. Adaptations of arctic and alpine plants to environmental conditions. Arctic 15, 117–144.

Borer, E.T., Harpole, W.S., Adler, P.B., Lind, E.M., Orrock, J.L., Seabloom, E.W., Smith, M.D., 2014. Finding generality in ecology: a model for globally distributed experiments. Methods Ecol. Evol. 5, 65–73. http://dx.doi.org/10.1111/2041-210X.12125.

Braun-Blanquet, J., 1932. Plant Sociology: The Study of Plant Communities. McGraw-Hill, New York, NY, US.

Byars, S.G., Hoffmann, A.A., 2009. Lack of strong local adaptation in the alpine forb *Craspedia lamicolain* Southeastern Australia. Int. J. Plant Sci. 170, 906–917. http://dx.doi.org/10.1086/599238.

Carbognani, M., Petraglia, A., Tomaselli, M., 2012. Influence of snowmelt time on species richness, density and production in a late snowbed community. Acta Oecol. 43, 113–120. http://dx.doi.org/10.1016/j.actao.2012.06.003.

Carbognani, M., Bernareggi, G., Perucco, F., Tomaselli, M., Petraglia, A., 2016. Micro-climatic controls and warming effects on flowering time in alpine snowbeds. Oecologia 182, 573–585. http://dx.doi.org/10.1007/s00442-016-3669-3.

Cooper, E.J., Dullinger, S., Semenchuk, P., 2011. Late snowmelt delays plant development and results in lower reproductive success in the High Arctic. Plant Sci. 180, 157–167. http://dx.doi.org/10.1016/j.plantsci.2010.09.005.

Domènech, M., Komac, B., Penuelas, J., 2016. Site-specific factors influence the richness and phenology of snowbed plants in the Pyrenees. Plant Biosystems 150, 741–749. http://dx.doi.org/10.1080/11263504.2014.990941.

Donnelly, A., Caffarra, A., Kelleher, C.T., O'Neill, B.F., Diskin, E., Pletsers, A., Proctor, H., Stirnemann, R., O'Halloran, J., Penuelas, J., Hodkinson, T.R., Sparks, T.H., 2012. Surviving in a warmer world: environmental and genetic responses. Clim. Res. 53, 245–262. http://dx.doi.org/10.3354/cr01102.

Dunne, J.A., Harte, J., Taylor, K.J., 2003. Subalpine meadow flowering phenology responses to climate change: integrating experimental and gradient methods. Ecol. Monogr. 73, 69–86. http://dx.doi.org/10.1890/0012-9615(2003)073[0069:SMFPRT]2.0.CO;2.

Ernande, B., Dieckmann, U., 2004. Adaptive changes in harvested populations: plasticity and evolution of age and size at maturation. Proc. R. Soc. Lond. B.

Forrest, J., Miller-Rushing, A.J., 2010. Toward a synthetic understanding of the role of phenology in ecology and evolution. Phil. Trans. R. Soc. B Sci 365, 3101–3112. http://dx.doi.org/10.1111/j.1365-2656.2008.01367.x.

Franks, S.J., Weber, J.J., Aitken, S.N., 2014. Evolutionary and plastic responses to climate change in terrestrial plant populations. Evol. Appl. 7, 123–139. http://dx.doi.org/10.1111/eva.12112.

Frei, E.R., Ghazoul, J., Matter, P., Heggli, M., Pluess, A.R., 2014. Plant population differentiation and climate change: responses of grassland species along an elevational gradient. Global Change Biol. 20, 441–455. http://dx.doi.org/10.1111/gcb.12403.

Galen, C., Stanton, M.L., 1995. Responses of snowbed plant-species to changes in growing-season length. Ecology 76, 1546–1557. http://dx.doi.org/10.2307/1938156.

García-Camacho, R., Totland, Ø., 2009. Pollen limitation in the alpine: a meta-analysis. Arct. Antarct. Alp. Res. 41, 103–111. http://dx.doi.org/10.1657/1523-0430-41.1.103.

Gimenez-Benavides, L., Escudero, A., Iriondo, J.M., 2007. Reproductive limits of a late-flowering high-mountain Mediterranean plant along an elevational climate gradient. New Phytol. 173, 367–382. http://dx.doi.org/10.1111/j.1469-8137.2006.01932.x.

Gjærevoll, O., 1956. The plant communities of the Scandinavian alpine snow beds. Trondheim. The Plant Cover of Sweden.

Gugger, S., Kesselring, H., Stöcklin, J., Hamann, E., 2015. Lower plasticity exhibited by high- versus mid-elevation species in their phenological responses to manipulated temperature and drought. Ann. Bot. 116, mcv155–mcv962. http://dx.doi.org/10.1093/aob/mcv155.

Haggerty, B.P., Galloway, L.F., 2011. Response of individual components of reproductive phenology to growing season length in a monocarpic herb. J. Ecol. 99, 242–253. http://dx.doi.org/10.1111/j.1365-2745.2010.01744.x.

Hall, M.C., Willis, J.H., 2006. Divergent selection on flowering time contributes to local adaptation in *Mimulus guttatus* populations. Evolution 60, 2466–2477.

Hanssen-Bauer, I., Drange, H., Fjørland, E.J., Roald, L.A., Børsheim, K.Y., Hisdal, H., Lawrence, D., Nesje, A., Sandven, S., Sorteberg, A., Sundby, S., Vasskog, K., Aadlandsvik, B., 2009. Klima i Norge 2100. Norsk klimasenter. pp. 1–148.

Heegaard, E., 2002. A model of alpine species distribution in relation to snowmelt time and altitude. J. Veg. Sci. 13, 493–504. http://dx.doi.org/10.1658/1100-9233(2002)013%5B0493:AMOASD%5D2.0.CO;2.

Hegland, S.J., Totland, Ø., 2007. Pollen limitation affects progeny vigour and subsequent recruitment in the insect-pollinated herb *Ranunculus acris*. Oikos 116, 1204–1210. http://dx.doi.org/10.1111/j.2007.0030-1299.15694.x.

Hülber, K., Winkler, M., Grabherr, G., 2010. Intraseasonal climate and habitat-specific variability controls the flowering phenology of high alpine plant species. Funct. Ecol. 24, 245–252. http://dx.doi.org/10.1111/j.1365-2435.2009.01645.x.

IPCC, 2014. Climate Change 2014: Synthesis Report. Contribution of Working Groups I. II and III to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. IPCC, Geneva, Switzerland.

Inouye, D.W., 2008. Effects of climate change on phenology, frost damage, and floral abundance of montane wildflowers. Ecology 89, 353–362. http://dx.doi.org/10.1890/06-2128.1.

Jonas, T., Rixen, C., Sturm, M., Stoeckli, V., 2008. How alpine plant growth is linked to snow cover and climate variability. J. Geophys. Res.: Biogeosci. 113, 377. http://dx.doi.org/10.1029/2007JG000680.

Körner, C., 2003. Alpine Plant Life: Functional Plant Ecology of High Mountain Ecosystems. Springer, Berlin.

Kawai, Y., Kudo, G., 2011. Local differentiation of flowering phenology in an alpine-snowbed herb Gentiana nipponica. Botany 89, 361–367. http://dx.doi.org/10.1139/b11-024.

Kawecki, T.J., 2008. Adaptation to marginal habitats. Annu. Rev. Ecol. Evol. S39, 321–342. http://dx.doi.org/10.1146/annurev.ecolsys.38.091206.095622.

Kawecki, T.J., Ebert, D., 2004. Conceptual issues in local adaptation. Ecol. Lett. 7, 1225–1241. http://dx.doi.org/10.1111/j.1461-0248.2004.00684.x.

Kudo, G., 1992. Performance and phenology of alpine herbs along a snow-melting gradient. Ecol. Res. 7, 297–304. http://dx.doi.org/10.1007/BF02347098.

Larcher, W., 2003. Physiological Plant Ecology: Eco Physiology and Stress Physiology of Functional Groups. Springer Science & Business Media, Berlin.

Merilä, J., Hendry, A.P., 2014. Climate change, adaptation, and phenotypic plasticity: the problem and the evidence. Evol. Appl. 7, 1–14. http://dx.doi.org/10.1111/eva.12137.

Moeller, D.A., Geber, M.A., 2005. Ecological context of the evolution of self-pollination in *Clarkia xantiana*: population size, plant communities, and reproductive assurance. Evolution 59, 786–799. http://dx.doi.org/10.1554/04-656.

Molau, U., 1993. Relationships between flowering phenology and life history strategies in tundra plants. Arct. Alp. Res. 25, 391. http://dx.doi.org/10.2307/1551922.

Moran, N.A., 1992. The evolutionary maintenance of alternative phenotypes. Am. Nat. 139, 971–989. http://dx.doi.org/10.1086/285369.

Nicotra, A.B., Atkin, O.K., Bonser, S.P., Davidson, A.M., Finnegan, E.J., Mathesius, U., Poot, P., Purugganan, M.D., Richards, C.L., Valladares, F., van Kleunen, M., 2010. Plant phenotypic plasticity in a changing climate. Trends Plant Sci. 15, 684–692. http://dx.doi.org/10.1016/j.tplants.2010.09.008.

Petraglia, A., Tomaselli, M., Bon Petit, M., Delnevo, N., Chiari, G., Carbognani, M., 2014. Responses of flowering phenology of snowbed plants to an experimentally imposed extreme advanced snowmelt. Plant Ecol. 215, 759–768. http://dx.doi.org/10.1007/s11258-014-0368-1.

Scott, D., Billings, W.D., 1964. Effects of environmental factors on standing crops + productivity of Alpine tundra. Ecol. Monogr. 34 (243). http://dx.doi.org/10.

2307/1948502.

Sedlacek, J., Wheeler, J.A., Cortés, A.J., Bossdorf, O., Hoch, G., Lexer, C., Wipf, S., Karrenberg, S., van Kleunen, M., Rixen, C., 2015. The response of the alpine dwarf shrub salix herbacea to altered snowmelt timing: lessons from a multi-site transplant experiment. PLoS One 10. http://dx.doi.org/10.1371/journal.pone.0122395.

Stanton, M., Galen, C., 1997. Life on the edge: adaptation versus environmentally mediated gene flow in the snow buttercup, *Ranunculus adoneus*. Am. Nat. 150, 143–178.

Stinson, K.A., 2004. Natural selection favors rapid reproductive phenology in *Potentilla pulcherrima* (Rosaceae) at opposite ends of a subalpine snowmelt gradient. Am. J. Bot. 91, 531–539. http://dx.doi.org/10.3732/ajb.91.4.531.

Stocker, T.F., 2013. Climate Change 2013: The Physical Science Basis. Intergovernmental Panel on Climate Change, Working Group I Contribution to the IPCC Fifth Assessment Report (AR5). Cambridge Univ Press, New York.

R Development Core Team, 2016. R: A Language and Environment for Statistical Computing.

Theurillat, J.P., Schlussel, A., 2000. Phenology and distribution strategy of key plant species within the subalpine-alpine ecocline in the Valaisan Alps (Switzerland). Phytocoenologia 30, 439–456.

Thomson, J.D., 2010. Flowering phenology, fruiting success and progressive deterioration of pollination in an early-flowering geophyte. Phil. Trans. R. Soc. B Sci. 365, 3187–3199. http://dx.doi.org/10.2307/2265776.

Totland, Ø., Alatalo, J.M., 2002. Effects of temperature and date of snowmelt on growth, reproduction, and flowering phenology in the arctic/alpine herb, Ranunculus glacialis. Oecologia 133, 168–175. http://dx.doi.org/10.1007/s00442-002-1028-z.

Totland, O., 1994. Intraseasonal variation in pollination intensity and seed set in an alpine population of *Ranunculus acris* in southwestern Norway. Ecography 17, 159–165.

Totland, O., 1997. Limitations on reproduction in alpine Ranunculus acris. Can. J. Bot. 75, 137–144. http://dx.doi.org/10.1139/b97-016.

Totland, Ø., 2001. Environment-dependent pollen limitation and selection on floral traits in an alpine species. Ecology 82, 2233–2244.

van Tienderen, P.H., van der Toorn, J., 1991. Genetic differentiation between populations of Plantago lanceolata. I. Local adaptation in three contrasting habitats. J. Ecol. 27–42.

Vitasse, Y., Hoch, G., Randin, C.F., Lenz, A., Kollas, C., Scheepens, J.F., Körner, C., 2013. Elevational adaptation and plasticity in seedling phenology of temperate deciduous tree species. Oecologia 171, 663–678. http://dx.doi.org/10.1007/s00442-012-2580-9.

Winkler, D.E., Chapin, K.J., Kueppers, L.M., 2016. Soil moisture mediates alpine life form and community productivity responses to warming. Ecology. http://dx.doi.org/10.1890/15-1197.1.

Wipf, S., Rixen, C., 2010. A review of snow manipulation experiments in Arctic and alpine tundra ecosystems. Polar Res. 29, 95–109. http://dx.doi.org/10.1111/j.1751-8369.2010.00153.x.

Wipf, S., Stoeckli, V., Bebi, P., 2009. Winter climate change in alpine tundra: plant responses to changes in snow depth and snowmelt timing. Clim. Change 94, 105–121. http://dx.doi.org/10.1007/s10584-009-9546-x.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 71–81

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Gatekeepers to the effects of climate warming? Niche construction restricts plant community changes along a temperature gradient☆

Kari Anne Bråthen[a,*], Victoria T. Gonzalez[a,b], Nigel G. Yoccoz[a]

[a] Department of Arctic and Marine Biology, UiT- Arctic University of Norway, N-9037 Tromsø, Norway
[b] NIBIO Svanhovd, 9925 Svanvik, Norway

ARTICLE INFO

Keywords:
Empetrum nigrum
Rangifer tarandus
Herbaceous plants
Structural equation model
Niche construction
Ecosystem engineering

ABSTRACT

Organisms that modify the environment (niche constructors) are likely candidates to mediate the effects of climate warming. Here we assess tundra plant community changes along a temperature gradient and how these are modified in the presence of the common allelopathic dwarf shrub *Empetrum nigrum* and the large herbivore *Rangifer tarandus*.

We developed a structural equation model based on data from a field-based study of 1450 tundra plant communities across Northern Fennoscandia, covering a temperature gradient of 3.5 °C, contrasting *Rangifer* densities, a range of *Empetrum* abundances in addition to gradients in topography and bedrock.

We found temperature to be a significant positive predictor of *Empetrum,* herbaceous and woody plant abundances. However, the effect of temperature as predictor for herbaceous plant abundance was significantly reduced in communities with *Empetrum* present. For woody plant abundance *Empetrum* was a stronger predictor than temperature. In comparison, we found *Rangifer* density to have marginal or no effect on either herbaceous or woody plant abundance. These findings were not modified by either topography or bedrock.

Results from this study indicate that herbaceous plant responses to climate warming are currently reduced in communities where *Empetrum* is present, whereas the abundance of *Empetrum* and other woody plants is promoted. Results also indicate that any future *Empetrum* encroachment is likely to drive tundra communities towards slower process rates and lower biodiversity. As such our results substantiate the importance of understanding the dynamics of niche constructor species and include them in predictive models of climate change.

1. Introduction

The tundra is changing (e.g. Chapin et al., 2005; Elmendorf et al., 2012b; Wookey et al., 2009), with the potential to become more species rich and greener under climate warming (e.g. Aerts et al., 2006; Epstein et al., 2012; Kullman, 2010; Rustad et al., 2001). Environmental modification by organisms, also termed niche construction or ecosystem engineering (Odling-Smee et al., 2013), may, however modify effects of climate warming. Plants can modify the environment in tundra plant communities, for instance by changing nutrient cycling rates (Bråthen and Ravolainen, 2015), subsequently causing plant-based cascade processes that enhance or reduce responses to climate change (Wookey et al., 2009). In addition, herbivores can counteract the greening process (shrub encroachment into circumpolar or alpine areas) (Den Herder et al., 2008; Olofsson et al., 2009; Post, 2013; Ravolainen et al., 2014; Zamin and Grogan, 2013), and hence prevent plant-based cascade processes. The trajectory of change under climate warming may therefore be dependent on the niche constructing abilities of plants and herbivores inhabiting the tundra.

Plant species are tracking their temperature niche as indicated by the upward and northward changes in species distributions in response to a warming climate (Chen et al., 2011; Klanderud and Birks, 2003; Lenoir et al., 2008). A thermophilization of the tundra flora is happening, where warm adapted species replace cold adapted species (Elmendorf et al., 2015; Gottfried et al., 2012). Because warm adapted species are benefited under climate warming and because the species richness is larger among warm adapted species, an increase in species richness is expected (Kullman, 2010). Yet, at the plot scale, no significant increase in species richness has happened in tundra areas in 30 years, whereas there have been changes in the height and abundance of growth forms (Elmendorf et al., 2012b) with increases described for shrubs, graminoids and forbs (Elmendorf et al., 2012a). Shrubs, in particular, are already common in tundra ecosystems (Walker et al., 2005), and are increasing with recent observed climate warming (Elmendorf et al., 2012b; Myers-Smith et al.,

---

☆ This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

* Corresponding author.
E-mail address: kari.brathen@uit.no (K.A. Bråthen).

http://dx.doi.org/10.1016/j.ppees.2017.06.005
Received 31 January 2017; Received in revised form 20 May 2017; Accepted 16 June 2017

2011). However, how this increase in shrub abundance is linked to community species richness and abundance of other growth forms is not evident. Theoretically, shrub encroachment can cause a range of possible outcomes for plant communities depending on the ecological context (Wookey et al., 2009) and empirical evidence of decreasing (Klein et al., 2004; Mod et al., 2016, Post, 2013; Wilson and Nilsson, 2009), stable (Pajunen et al., 2011) and increasing (Bråthen and Lortie, 2016) species richness of vascular plants as a response to shrub encroachment has been found. To advance in the understanding of how ecological contexts influence responses of plant communities to climate warming, large-scale studies on dominant shrub species within a range of habitats and contexts are needed (Christie et al., 2015).

The evergreen dwarf-shrub *Empetrum nigrum* (crowberry) has strong niche constructing capacity (*sensu* Matthews et al., 2014) as indicated by its ability to modify ecosystem properties (Bråthen and Lortie, 2016; Nilsson and Wardle, 2005; Tybirk et al., 2000; Wardle et al., 1998). *Empetrum* is tightly linked to variation in species diversity of tundra plant communities under both high and low intensity *Rangifer tarandus* (reindeer) land use (Ravolainen et al., 2010). The abundance of *Empetrum* has repeatedly been found negatively related to species richness of vascular plants in tundra communities (Bråthen and Lortie, 2016; Bråthen and Ravolainen, 2015; Mod et al., 2016; Wilson and Nilsson, 2009). Accordingly, *Empetrum* is found to be an important predictor along with temperature in species distribution models (Pellissier et al., 2010) and in plant community models (Le Roux et al., 2014). However, although allelopathic to other plant species at their seedling state (González et al., 2015), *Empetrum* does not always have negative effect on coexisting plants. Whereas *Empetrum* is competitively superior to forb and grass species (Mod et al., 2014; Pellissier et al., 2010; Wilson and Nilsson, 2009), it seems to have facilitative effects for species with higher leaf dry matter content (Mod et al., 2014; Pellissier et al., 2010). *Empetrum* may thus modify the environment, thereby driving selection for species with traits causing slow nutrient process rates (cf. Wilson et al., 1999), and potentially reinforcing the filtering effect that *Empetrum* has on the species pool. This is especially pertinent to herbaceous species in tundra plant communities.

Dendroecological studies indicate *Empetrum* is sensitive to climate change (Buntgen et al., 2015; Myers-Smith et al., 2015), and empirical evidence indicates that *Empetrum* is advancing in altitude and abundance in tundra areas in response to recent climate warming (Felde et al., 2012; Klanderud and Birks, 2003; Wilson and Nilsson, 2009). Such observations of a positive response to warming are corroborated by experiments (Buizer et al., 2012; Kaarlejärvi et al., 2012; Wada et al., 2002), and indicate shrub encroachment by *Empetrum* is promoted by climate warming. However, *Empetrum* has been found to be vulnerable to extreme climatic events during winter (Bokhorst et al., 2008) and to attacks by the *Empetrum*-specific parasitic fungus *Arwidssonia empetri* under increased snow cover (Olofsson et al., 2011). Still, *Empetrum* has been found resilient to simulated strong winter damage (Aerts, 2010), has a high tolerance to ice encasement (Preece et al., 2012; Preece and Phoenix, 2014), and it appears that *Empetrum* is relatively resistant to a range of environmental changes, including fertilization, as compared to coexisting species (Tybirk et al., 2000). Hence, although *Empetrum* has been found to be vulnerable to some aspects of climate warming, it seems it mostly thrives.

Empetrum abundance is also affected by herbivores. For instance, close to fences that separate *Rangifer* migration areas, trampling can be intense (Moen and Danell, 2003). As *Empetrum* is vulnerable to trampling (Bell and Tallis, 1973), this can result in reduced abundance (Olofsson et al., 2001; Väisänen et al., 2014). Small rodents have also been shown to control *Empetrum* abundance (Olofsson et al., 2009; Olofsson et al., 2012), but a longer time collapse in small rodent cycles (Ims et al., 2008), may have reduced the capacity of small rodents to do so. Furthermore, while most studies indicate that herbivory are counteracting the encroachment of shrub species, there are empirical indications of *Empetrum* responding differently and that it is rather promoted by *Rangifer* activities (Bråthen et al., 2007b; Francini et al., 2014; Ylänne et al., 2015). Such promotion can potentially happen through apparent competition because *Empetrum* leaves have low palatability (Tybirk et al., 2000) and through endozoochory because *Empetrum* berries are eaten by e.g. *Rangifer* (Bråthen et al., 2007a). Hence, *Empetrum* seems to be both positively and negatively affected by the activities of herbivores.

In sum, an assessment of how *Empetrum* itself is affected by increasing temperatures and *Rangifer* density, and how *Empetrum* affects tundra plant communities in this context, is likely to yield insight into the development of tundra plant communities under climate warming. Therefore, in this study, we ask to what extent *Empetrum* and *Rangifer* are likely candidates for mediating effects of climate warming in tundra plant communities. We use a space for time approach to address climate warming effects (Blois et al., 2013; Elmendorf et al., 2015): the study design covers a temperature gradient of 3.5 °C across a total of 1450 randomly chosen tundra plant communities in the Fennoscandian part of the circumpolar dwarf shrub tundra (*sensu* Walker et al., 2005). The communities are defined as where plant species share a habitat or environment (*sensu* Daubenmire, 1968). *Empetrum* varies in abundance across these communities, from being absent to subordinate and to having supreme dominance. The study design concomitantly covers ten district pairs of high vs. low intensity of *Rangifer* husbandry land use, and has previously been used to assess effects of *Rangifer* density on the ecosystem state of the tundra communities (Bråthen et al., 2007b), focusing on mammalian herbivore abundances (Ims et al., 2007) and plant diversity (Bråthen and Lortie, 2016; Bråthen and Ravolainen, 2015). In this study we hypothesize that (1) *Empetrum* abundance increases along the temperature gradient, and that (2) higher *Rangifer* densities promote *Empetrum* abundance at higher temperatures. We also hypothesize that (3) *Empetrum* reduces the effect that increasing temperatures has in promoting both inflorescences and biomass of growth forms with higher process rates such as forbs and grasses. In addition, since higher *Rangifer* densities have already been shown to reduce biomass of forbs and grasses as well as the abundance of inflorescences (Bråthen et al., 2007b), we hypothesize that (4) *Rangifer* counteract any positive effect of temperature on the abundance of herbaceous plants. Furthermore we hypothesize that (5) *Empetrum* and increasing temperatures facilitate the biomass of other shrub species, but that (6) higher *Rangifer* densities do not modify this relationship because the abundance of these growth forms is mostly independent of *Rangifer* densities (Bråthen et al., 2007b). In order to test these hypotheses in a comprehensive way we developed a structural equation model, and then we tested each hypothesis separately.

2. Materials and methods

2.1. Study area

Field work for this study was conducted in the coastal alpine tundra of northern Fennoscandia between latitudes 69.2–71.2°N and longitudes 20.5–31.3°E (Fig. 1). This is a region characterized primarily as dwarf-shrub tundra (Walker et al., 2005), with *Empetrum nigrum* being both the most common shrub and the most dominant plant (Bråthen et al., 2007b; Ravolainen et al., 2010). The region is typically snow covered until late June and the main vegetation types are heath, meadows and marshes. Besides *Empetrum*, heath is dominated by the dwarf shrubs *Betula nana*, *Salix herbacea*, *Vaccinium myrtillus*, *V. vitis-idaea* and *V. uliginosum*. Forb and grass-rich meadows are often populated with *Bistorta vivipara* and *Viola biflora*, *Avenella flexuosa*, *Deschampsia cespitosa, Anthoxanthum nipponicum*, and sedges of *Carex* spp. Marshes are dominated by *Eriophorum angustifolium* and *E. vaginatum* along with various species of *Carex*. *Empetrum* is often present in both meadows and marshes. Plant names follow the Pan-Arctic Flora (http://nhm2.uio.no/paf/). The main large herbivores in this region are the migratory, semi-domestic *Rangifer tarandus* that have their summer pastures along the coast, which is divided into districts to facilitate management. Other common herbivores include moose (*Alces alces*), domestic sheep (*Ovis aries*), and small rodents (Ims et al., 2007).

The study region is characterized by steep climatic gradients from west to east as well as from coast to inland, caused by the warm NE Atlantic Current that gradually declines from west to east (see temperature map in Fig. 1). The northernmost coast of the study region is classified as Arctic (Walker et al., 2005). Average summer precipitation (June, July and August) is 65, 54 and 48 mm in the west, middle, and east of Finnmark, respectively (calculated from Norwegian Meteorological Institute, http://www.eklima.no). The coastal part of the study region is furthermore characterized by variable bedrock (mainly gabbro in the west, slate and sandstone towards the east [Geological Survey of Norway, http://www.ngu.no/]).

2.2. *Study design*

A total of 1450 plant communities were sampled within the borders of ten pairs of neighbouring *Rangifer* districts (Fig. 2), where each pair represents two decades of high versus low *Rangifer* densities (official numbers provided by the Norwegian Reindeer Husbandry Administration http://www.reindrift.no/) (Bråthen et al., 2007b). Different *Rangifer* densities are to a large extent determined by spatiotemporally heterogeneous management practices in *Rangifer* husbandry (Tveraa et al., 2007). Faeces counts in the field confirmed that each pair of neighbouring *Rangifer* districts represented a contrast in low and high *Rangifer* density (Bråthen et al., 2007b).

Sampling within the *Rangifer* districts was based on *a priori* stratification and plant community selection using GIS as well as on rules set *a priori* to the field sampling (*sensu* Mörsdorf et al., 2015). Each *Rangifer* district was covered with a 2 × 2 km grid (in which each cell is referred to as a “landscape area”). For each district, and in order to avoid landscape areas dominated by glaciers, lakes and boulder fields, satellite image classifications of vegetation types (Johansen et al., 1995) were evaluated to identify landscape areas with more than the district average of mesic and wet vegetation. Among these, landscape areas were retained unless they included > 50% forest, lakes, sea, glaciers or included a fence or a major road (based on http://www.norgeskart.no/). The final landscape areas within each *Rangifer* district were selected randomly. The number of selected landscape areas increased as a function of district area, with the final number of selected landscape areas per district ranging from 6 to 14 units.

The choice of plant communities within the selected landscape areas was random. The landscape areas were subdivided into 100 potential sampling squares of 200 m × 200 m, with a maximum of 21 randomly selected squares (Fig. 1). The centre of each selected square was the starting position for a 50 m long transect whose direction was determined by a random GPS position on a circle with a 50 m radius. If any of the transects had to be discarded because of steep terrain, water (lake, large river or very wet mire), snow cover (more than a 5 m section of the transect running through snow), boulder field (more than half of the transect running over boulders devoid of vegetation), or the transect was below the tree-line, another random direction or start position was identified. If no new acceptable transect placement within the area was available, the entire sampling square was discarded. Each transect was sub-sampled at plots every 5 m along the transect with a triangular sampling frame with sides of 40 cm. Each transect constituted a tundra plant community (Fig. 1). All landscape areas were sampled in July and August 2003. Each landscape area was sampled by two individuals but for a few areas sampled by one person. To avoid differences due to plant phenology, *Rangifer* districts belonging to the same pair were analyzed simultaneously, and sampling was conducted from west to east as the season progressed in order to sample at or just after peak growing season.

2.3. *Environmental variables*

A downscaled WorldClim temperature layer (“Warmest Quarter” Hijmans et al., 2005) of 100 m × 100 m resolution (Pellissier et al., 2013) was applied for average summer temperatures (Fig. 1). We used average summer temperature (across June, July and August) because it is physiologically more meaningful to alpine species than other temperature variables (Körner, 2003).

Bedrock types for each plant community were obtained from maps provided by the Geological Survey of Norway (http://www.ngu.no/). The bedrock type was classified as poor (0), e.g. quartzite, moderate (1) e.g. amphibolite, and rich (2) e.g. mica or limestone, in providing nutrients available to the plant community as based on Tarbuck and Lutgens (1992).

A curvature index, i.e. a geoedaphic factor that serves as a surrogate for the moisture level of vegetation (Moore et al., 1991), was calculated per plant community using GIS and a digital terrain model of 25 m × 25 m pixels (http://www.kartverket.no). Index values from 1 to −1 indicate concave to convex curvature, i.e. moist to dry terrain, respectively. Visual inspection in the field of a few sites verified that the index values correctly depicted concave or convex terrain.

2.4. *Biological variables*

In each plot the point intercept method (Bråthen and Hagberg, 2004) – using three pins attached to the corner of the triangle (Fig. 1) – was used to obtain measures of above ground live vascular plant species biomass. Prior to further analyses, point intercept data per species and plot were converted to biomass estimates in units of grams per m^2 using established calibrations (Ravolainen et al., 2010). To achieve estimates of growth form biomasses and total biomass per plant community, the data were summed across species and averaged across all plots. Point intercept data on inflorescences (including all inflorescence parts and all their phenological stages) were registered separately, and were summed across species and averaged across all plots to achieve estimates of inflorescences abundance per plant community. Estimates of *Empetrum* biomass and *Empetrum* berry abundance were calculated separately.

2.5. *Data analyses*

Structural equation models were developed in the R environment version 3.2.3 (http://www.r-project.org) using in the lavaan package (Rosseel et al., 2013). Data were screened for distributional properties and nonlinear relations which resulted in several variables being log-transformed. The first structural equation model was exploratory, built to test the effect of all predictor variables (temperature, *Rangifer* density, curvature and bedrock) onto the abundance of herbaceous and woody growth forms. The possible mediating effect by the abundance of *Empetrum* was included in the model by having *Empetrum* both as a response variable and as a predictor to the plant growth form abundances. All non-significant predictors were successively removed from the model. The final model was selected based on parsimoniousness and goodness of fit as interpreted from Chi Square tests, RMSEA and Comparative Fit Index provided in the lavaan package (Rosseel et al., 2013). The contribution of each predictor variable in the final model was then assessed in order to possibly simplify the model further. Final models with one predictor variable removed at a time were compared to the final model using anova model comparison. The final model including all predictors had the best AIC score. Lastly, the final model was tested for whether the hierarchical setup of the study design had any consequences to the variance explained by the model using the lavaan survey package and by an assessment of the robust Chi Square test (Oberski, 2014). At this stage, all predictors were retained in the model, including predictors that became non statistically significant but still explained variance above a value of 0.04. Variance explained by the different predictor variables was obtained from model outputs as

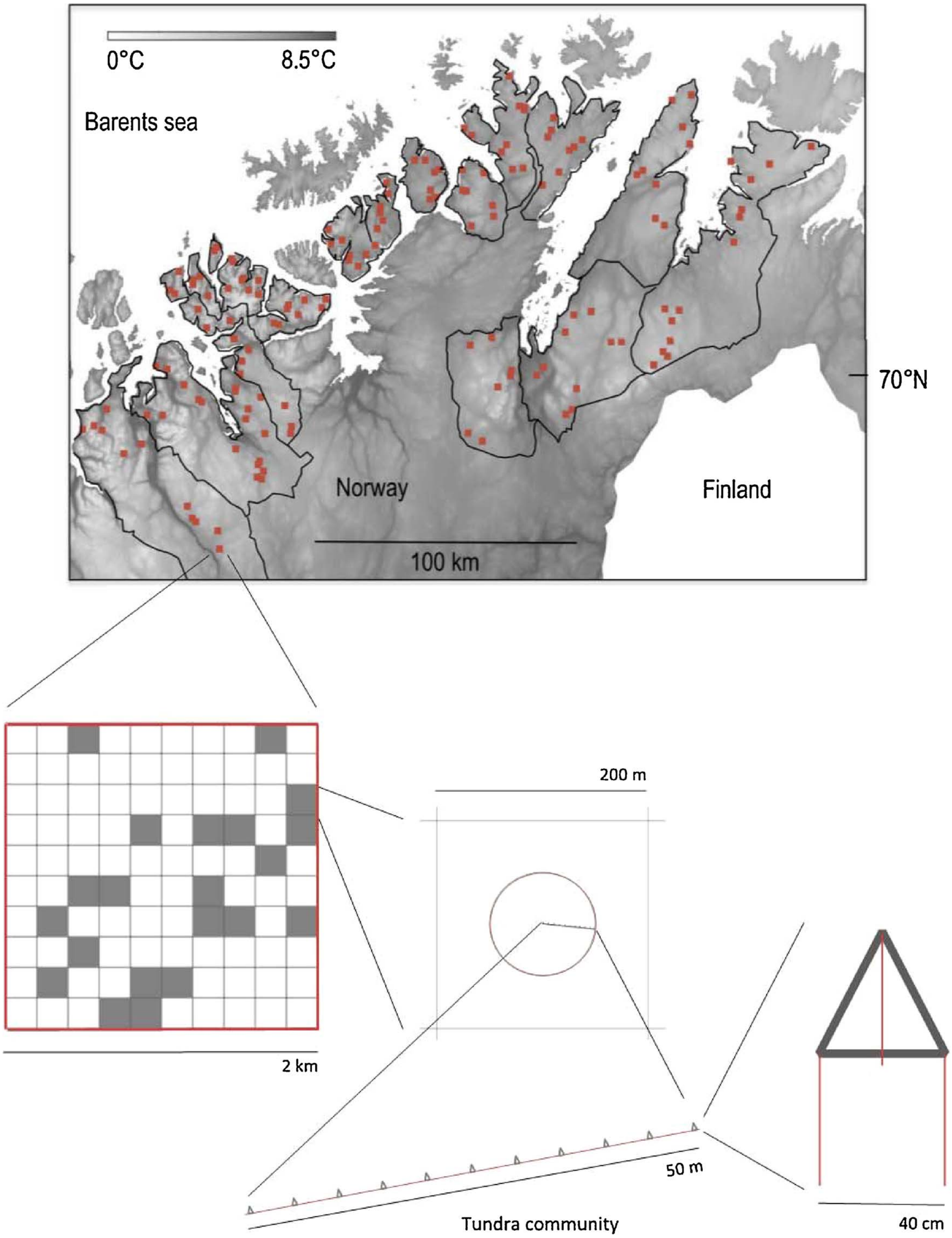
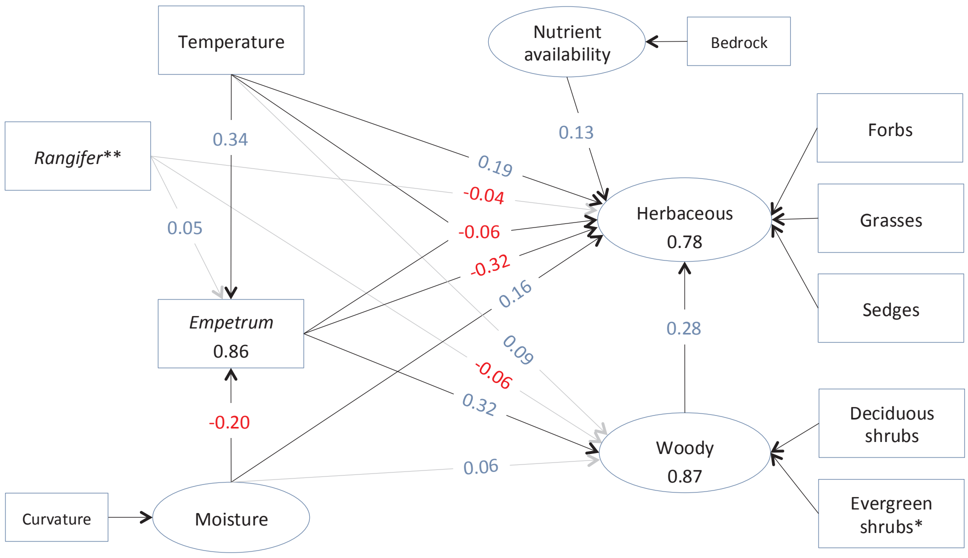


Fig. 1. Summer temperatures across northern Fennoscandia including landscape areas where plant communities were analyzed (upper panel) and an overview of the study design applied in each landscape area (lower panel). In the upper panel the borders represent the 20 *Rangifer* husbandry districts and the red squares represent the landscape areas. In the lower panel the nested design for each landscape area, geographically positioned a priori to the field season, is shown. Within each landscape area a random set of maximum 21 squares of 200 m × 200 m were analyzed by running a transect from the square centre in a random direction. Plots were regularly placed along each transect representing a plant community and each plot was analyzed by the point intercept method using three pins. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

standardized values, and manually entered into a path diagram of the final structural equation model.

In addition we developed linear mixed effects models (Pinheiro and Bates, 2000) using the nlme package in R to test specific hypotheses. In the first set of models fixed factors were *Empetrum* biomass and *Empetrum* berries as response variables and temperature and *Rangifer* density (high and low) as predictor variables. For the model on *Empetrum* berries *Empetrum* biomass was also a predictor. In a second set of models fixed factors were species richness, the abundance of inflorescences, and the biomass of each growth form as response variables along with temperature, *Empetrum* biomass and *Rangifer* density as predictor variables. For all models the random factors were landscape areas nested in pairs of *Rangifer* districts.

To achieve homogeneity of the residual variance of models, all



*excluding Empetrum
**anthropogenically determined

Fig. 2. Structural equation model representing connections between *Empetrum*, temperature, *Rangifer* and herbaceous and woody growth forms supported by the data. Blue text represents positive effects, red text represents negative effects. Black arrows indicate significant effects, grey arrows indicate effects no longer significant under the adjustment of the model when accounting for the hierarchical study design. Only the interaction term significant in both models is presented: the interaction between *Empetrum* and temperature on herbaceous biomass. Model test statistic = 4.825, with 2 model degrees of freedom and p = 0.090 (indicating close model-data fit). The robust test statistic = 1.138 (model also considering the hierarchical design), with 2 model degrees of freedom and p = 0.566 (again indicating close model-data fit). Comparative Fit Index = 0.996 and Root Mean Square Error of Approximation = 0.031, further indicate close model fit. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

abundance variables (i.e. inflorescence abundances and biomasses) were log transformed (x + 1). All predictor variables were standardized (mean = 0 and standard deviation = 1) to facilitate comparison between variables and models (e.g. Iversen et al., 2014).

The standardized coefficients and the confidence intervals of fixed predictors were extracted from model outputs and presented and plotted as predicted values. From the second set of models the predicted values were presented in three different plant community categories; in communities with *Empetrum* biomass constituting 0%, 20–30% or 70–80% out of total community biomass.

All models were extended with nutrient availability (bedrock index) and moisture level (curvature index) as fixed factors to assess any confounding towards main predictors and are presented with their standardized coefficients in supplementary information (Table S1). In addition, standard deviations and confidence intervals of the random factors for each set of models are presented.

3. Results

Empetrum was present in 1230 out of the 1450 randomly chosen tundra plant communities, and composed more than half the biomass across all communities. Among the growth forms, forbs constituted on average the least biomass, followed by sedges, grasses, evergreen shrubs (excluding *Empetrum*) and deciduous shrubs (Table 1). In the communities where *Empetrum* was not present, other shrubs constituted on average 55% of the community biomass.

The structural equation modelling (SEM) showed a significant influence of temperature on the biomass of *Empetrum* and herbaceous plants and a non-significant influence on woody plant biomass (Fig. 2). The influence of *Rangifer* density on either response variable was not significant. The influence by *Empetrum* on herbaceous and woody plant biomass was stronger than that of temperature, but with a negative and a positive relationship respectively. Moreover, there was a significant negative interaction between temperature and *Empetrum* on herbaceous plant biomass, indicating the influence of *Empetrum* was more negative at higher temperatures. Moisture, as presented in the form of the curvature index, had a significant negative influence on *Empetrum* biomass and a significant positive influence on herbaceous plant biomass. Nutrient availability, as presented by the bedrock index, was only influential to herbaceous plant biomass, and in a positive way.

The bivariate model (linear mixed model) result on the influence of temperature on the *Empetrum* biomass was consistent with the SEM results, showing a marked increase in *Empetrum* biomass in response to increasing temperature (Fig. 3, Table 2). However, in contrast to the SEM results, higher *Rangifer* density had significant positive effect on the *Empetrum* biomass in the bivariate model, but with small effect size (Fig. 3, Table 2). The abundance of *Empetrum* berries was only modelled using a bivariate model, with model results showing slightly less berries in communities with high *Rangifer* densities (Fig. 3, Table 2), and a positive relationship with temperature where also the biomass of *Empetrum* was higher (Table 2).

The bivariate model results on the influence of temperature and *Rangifer* on the biomass of the herbaceous and woody growth forms were also mostly consistent with the SEM results; Temperature had a significant positive relationship with both herbaceous and woody plant biomass whereas the effect of *Rangifer* density was insignificant (Fig. 2, Table 2). With increasing biomass of *Empetrum* in the plant communities these positive relationships to increasing temperature were severely reduced for the herbaceous plant biomass, whereas they were enhanced for the woody plant biomass (Fig. 4a, Table 2). This latter interaction effect on woody plant biomass was however not part of the final SEM model (Fig. 2) because the variance explained by the interaction was less than 0.03.

Bivariate models were run to assess responses among the different herbaceous or woody growth forms. The biomass of each of the growth forms were all significantly positively related to the temperature gradient from 4.4 to 7.9 °C (Fig. 4a, Table 2). The effect of *Rangifer* was insignificant for all growth forms except for forbs and grasses where a negative effect of high *Rangifer* densities was similar in effect size and sign to that of *Empetrum* biomass (Table 2).

The bivariate model on the abundance of inflorescences (not including *Empetrum* inflorescences) showed a similar outcome to that of the forbs and grasses, with a positive effect of temperature, a negative effect of *Empetrum* biomass and a negative effect of high *Rangifer* densities (Fig. 4b). The bivariate model on species richness showed a positive effect of temperature and a negative effect of *Empetrum*, but no effect of different *Rangifer* densities (Fig. 4b, Table 2).

The predictor strength and direction of temperature, *Empetrum* biomass and *Rangifer* density was only slightly modified by adding nutrient availability (i.e. bedrock index) and moisture (i.e. curvature

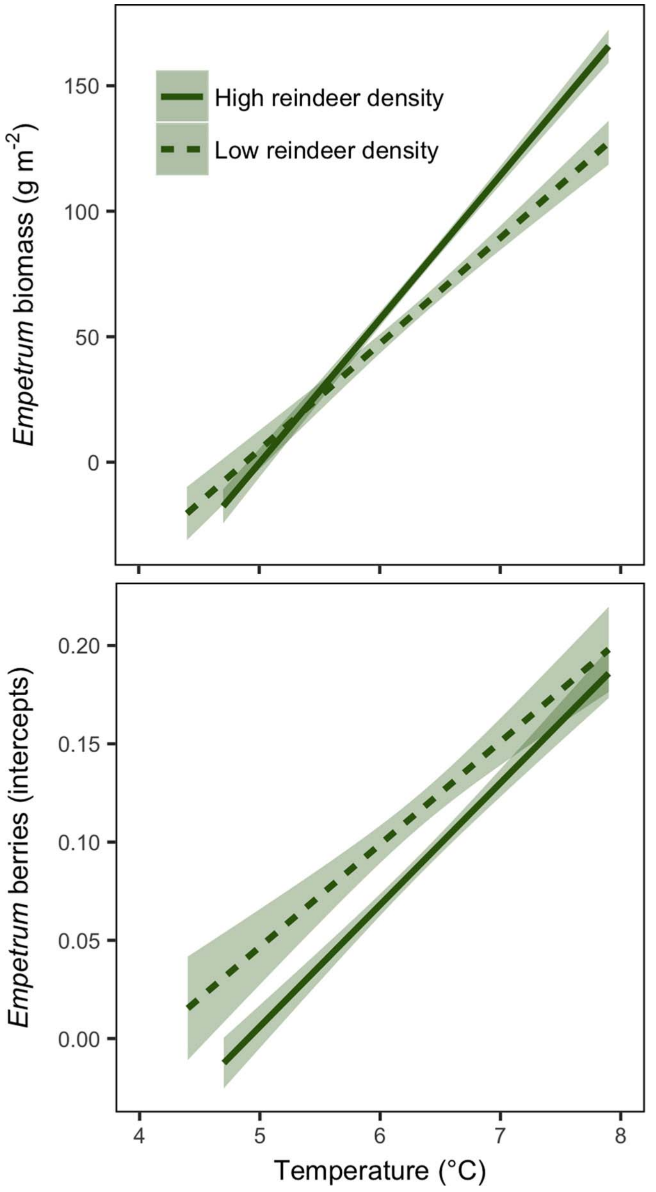


Fig. 3. The effect of increasing temperature (warmest quarter) and *Rangifer tarandus* density (high vs. low) on the predicted values of aboveground biomass (g m^{-2}) and the abundance of berries (point intercept frequency) of *Empetrum nigrum* presented on a log$_e$ (x + 1) scale. The linear regressions are based on predicted values from a lme model output and are presented with ± 95% confidence bands.

index) as predictors of community characteristics (Table S1). This indicates there were no confounding effects between main predictors and the additional environmental predictors, and is consistent with the SEM results that showed temperature and *Empetrum* biomass had a clear influence alongside that of the environmental predictors. Such lack of confounding is also strengthened by the fact that the full range of moisture and nutrient availability regimes were present in all categories of *Empetrum* dominance (Fig. S1 and S2). Nonetheless, the nutrient availability was significantly positively related to species richness, herbaceous and woody shrub biomass. Moisture was significantly negatively related to *Empetrum* biomass (indicating more *Empetrum* in convex and hence drier terrain) and significantly positively related to species richness, inflorescence abundance and herbaceous biomass (indicating higher abundances in more concave and hence moister terrain).

4. Discussion

In this study, we assessed the extent to which *Empetrum nigrum* and *Rangifer tarandus*, two species with niche constructing abilities (*sensu* Matthews et al., 2014; Odling-Smee et al., 2013), mediate the response of tundra plant communities to increasing temperatures. We found *Empetrum* – a common dwarf shrub species in circumboreal-polar areas (http://nhm2.uio.no/paf/) – to have clear mediating effects whereas we found effects of *Rangifer* – the dominant large herbivore in this ecosystem (Huntington, 2013) – to have no or low mediating effects in comparison. Moreover, we found the biomass of *Empetrum* to be positively affected by increasing temperatures and to be slightly positively affected by higher *Rangifer* densities. Because *Empetrum* has allelopathic capacity its proliferation implies a deterioration of habitats to a range of other organisms (Aerts, 2010; Bråthen et al., 2010; Nilsson et al., 1993; Nilsson and Zackrisson, 1992; Wallstedt et al., 2001), with potential wide-reaching consequences to circumpolar areas.

4.1. *Empetrum abundance increases with increasing temperatures*

The increase in abundance of both biomass and berries of *Empetrum* with increasing temperatures is in line with previous studies assessing *Empetrum* biomass (Felde et al., 2012; Kaarlejärvi et al., 2012; Klanderud and Birks, 2003; Wada et al., 2002; Wilson and Nilsson, 2009) and reproduction (Buizer et al., 2012). The ability of *Empetrum* to acquire nitrogen early in the season (Larsen et al., 2012; Wipf, 2010) may give it a competitive advantage over other plant species when responding to increasing temperatures. Also, the increase in berry abundance likely contributes to increased establishment and hence increased abundance of *Empetrum* under warmer temperatures. Increase in *Empetrum* abundance with warming has been found along a temperature gradient ranging from 5 to 15 °C (Buizer et al., 2012) (i.e. at higher temperatures than that provided in the current study), indicating climate warming can promote encroachment by *Empetrum* in ecosystems also beyond circumpolar regions.

Higher intensity land use by *Rangifer* husbandry had a marginally positive effect on the biomass of *Empetrum,* and is consistent with previous findings (cf. Bråthen et al., 2007b). The positive effect of *Rangifer* herbivory might have parallels to *Empetrum rubrum,* a species found in the southern hemisphere and with a high genetic similarity to *Empetrum nigrum* (Popp et al., 2011). Claims are that grazing by sheep over the last 100 years has turned grasslands into *Empetrum rubrum* dominated heathlands (Collantes et al., 1999). In our system, the process by which herbivory causes *Empetrum nigrum* to gain abundance on behalf of other species is most likely apparent competition because it has both low nutrient content (Kaarlejärvi et al., 2012; Sorensen et al., 2008), phenolic rich leaves (Gallet et al., 1999; Väisänen et al., 2013) and low palatability (Tybirk et al., 2000) even during winter when food resources are scarce (Danell et al., 1994). *Empetrum* has also been found to benefit from endozoochory by *Rangifer* (Bråthen et al., 2007a). Hence, the reduced abundance of *Empetrum* berries at higher *Rangifer* densities may ultimately be part of the positive effect of *Rangifer* densities on *Empetrum* abundance: *Rangifer* is likely dispersing *Empetrum* into new habitats. Areas of high *Empetrum* cover are, however, avoided by *Rangifer* (Iversen et al., 2014) and presence of other herbivores is also negatively linked to areas of high *Empetrum* abundance (as shown in a companion study by Ims et al., 2007). There may thus be a threshold in *Empetrum* abundance at which herbivores are no longer influencing its abundance.

4.2. *Climate warming effects on tundra plant communities modified by Empetrum*

Our finding that the abundances of both inflorescences and biomass of herbaceous and woody plants increased with increasing temperatures in the tundra plant communities is in line with expectations of an

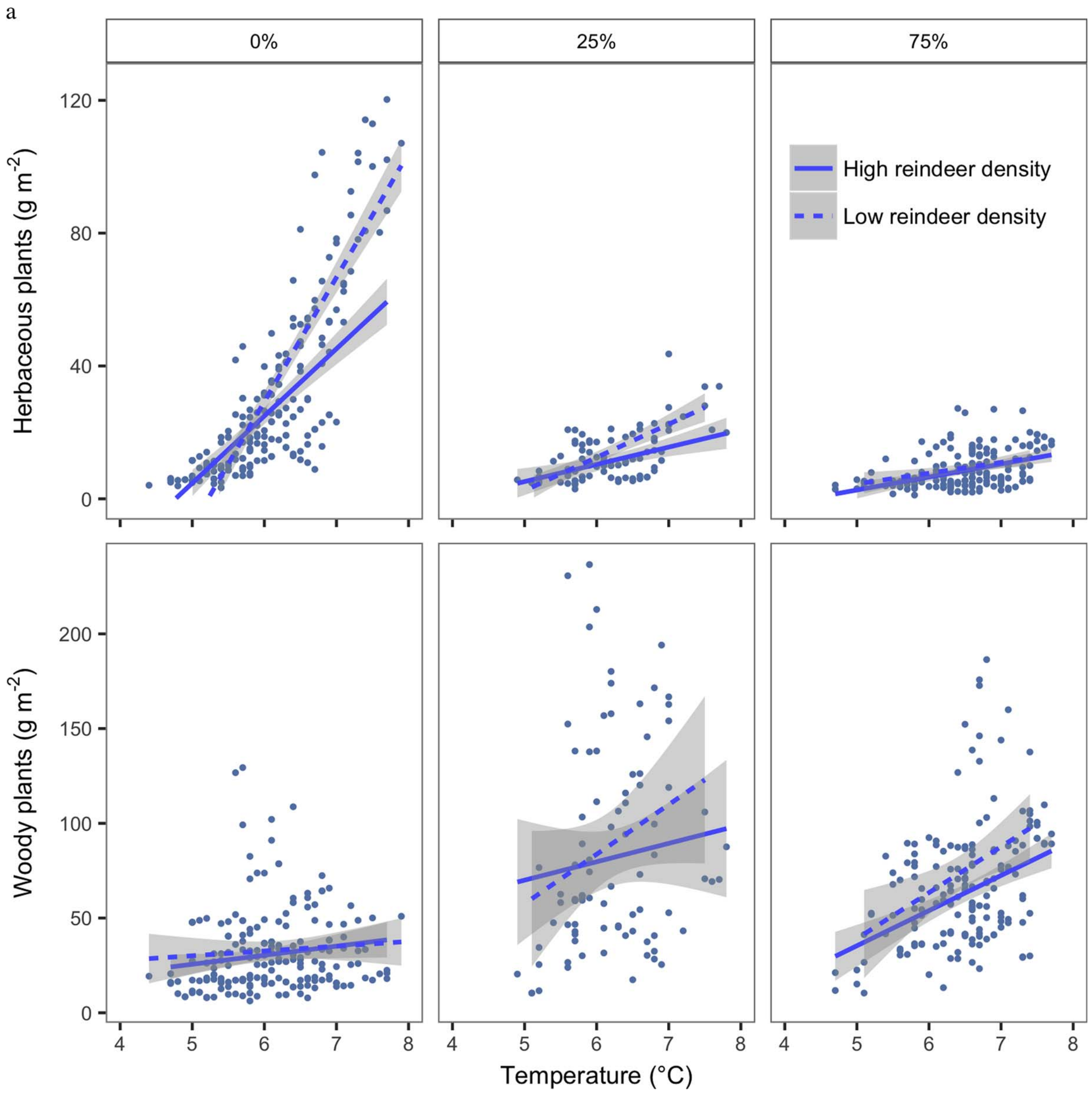


Fig. 4. The effect of increasing temperature (warmest quarter) and *Rangifer tarandus* density (high vs. low) and increasing levels of *Empetrum* biomass on the predicted values of **a)** the aboveground biomass (g m^{-2}) of herbaceous (forbs, grasses and sedges) and woody (deciduous and evergreen shrubs) growth forms and on **b)** the abundance of species richness and inflorescences (point intercept frequency) in the plant communities. The three panels represent communities with 0%, 25% (20–30%) and 75% (70–80%) of community biomass occupied by *Empetrum* biomass. All variables are presented on a log$_e$ (x + 1) scale. The linear regressions are based on predicted values from a lme model output and are presented with ± 95% confidence bands.

increase in ecosystem process rates in response to climate warming (Elmendorf et al., 2012a). Increased biomass in general indicates higher productivity. Also an increase in the abundance of inflorescences will likely improve seed availability and new plant establishment because tundra communities often are seed limited (Graae et al., 2011). The biomass of herbaceous plants, the most productive and palatable growth forms (Chapin et al., 1996; Cornelissen et al., 2004; Cornelissen et al., 2007), were more positively related to the increasing temperatures than that of shrubs. However, in communities with *Empetrum* present the strength of temperature as a predictor of higher herbaceous biomass clearly declined. Although herbaceous growth forms utilize different nutrient pools than *Empetrum* (Michelsen et al., 1996), suggesting low direct competition for nutrients, reduced biomass of herbaceous species in the presence of *Empetrum* is likely. That is, recalcitrant evergreen litter can slow warming-induced increases in soil fertility (De Long et al., 2016; Hobbie, 1992). Additionally, forbs and grasses are especially vulnerable to the allelopathic capacity of *Empetrum* at their germination and seedling stage (Bråthen et al., 2010; González et al., 2015). It is thus likely that niche construction by *Empetrum,* through modifying habitat quality for herbaceous plants, limits the capacity of herbaceous plants in tundra communities to respond positively to climate warming.

In contrast, the biomass of woody plants, i.e. dwarf shrubs, was positively related to *Empetrum* biomass. Increases in the abundance of shrub species alongside increases in *Empetrum* have been shown in previous climate warming studies in dwarf shrub heaths (Buizer et al., 2012; Kaarlejärvi et al., 2012; Wada et al., 2002). Because shrub species in average are long-lived (e.g. Büntgen et al., 2015), they are likely to be promoted by climate warming at an established developmental stage and are hence not as dependent on the seedling stage previously shown to be vulnerable to *Empetrum* interference (González et al., 2015; Nilsson and Zackrisson, 1992). It may rather be that facilitating effects such as sheltering among established shrubs (Wipf et al., 2006) promote shrub species to further encroach upon the tundra in response to

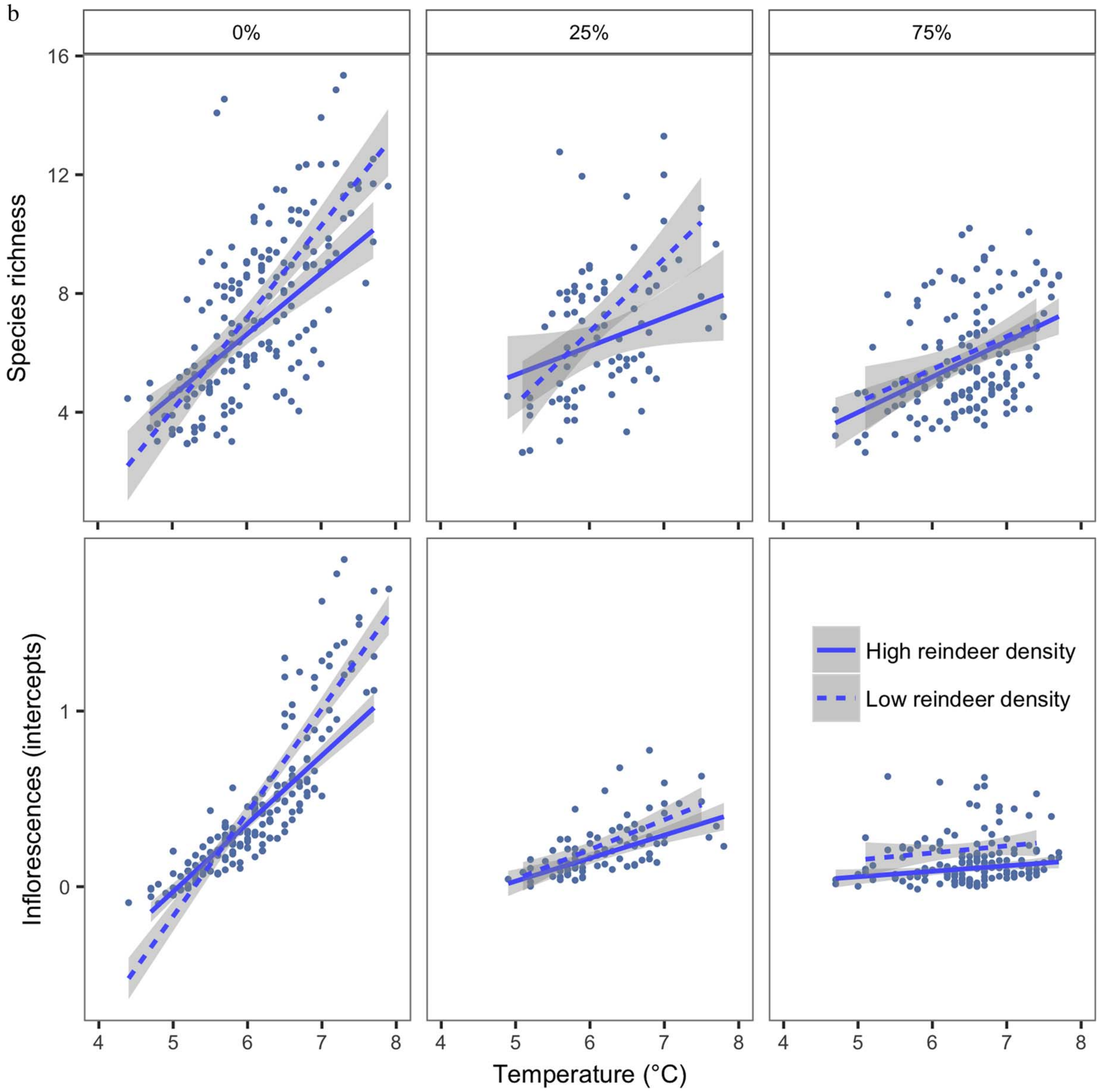


Fig. 4. (*continued*)

climate warming.

4.3. *Climate warming effects on tundra plant communities modified by Rangifer*

The effects of land use by *Rangifer* husbandry on herbaceous growth forms were on average small in comparison to that of *Empetrum,* but when addressing the herbaceous growth forms individually, both forbs and grasses were found to be negatively affected by both higher *Rangifer* densities and *Empetrum* biomass. Hence, our study supports studies addressing *Rangifer* grazing where a general reduction of forbs (Bernes et al., 2015), a general decline in seed production (González et al., 2010) and a decrease in the forb response to climate warming (Kaarlejärvi et al., 2013), have been found.

4.4. *Implications for ecosystem functioning*

Niche construction by *Empetrum* can be so strong that the physical removal of the plant itself has no consequence to aboveground species composition or soil abiotic and biotic properties of its habitat even after ten years (Wardle et al., 2012; Wardle and Jonsson, 2013). Such a legacy effect of *Empetrum* niche construction is plausible due to the bioactivity of leaves accumulated in soil (Bråthen et al., 2010; Wallstedt et al., 2005). *Empetrum* may also have cascading effects on plant-herbivore interactions in tundra ecosystems. For instance, the diet of small rodents consists to a large extent of forbs and grasses (Soininen et al., 2013a; Soininen et al., 2013b), which, as shown in this study, have lower abundance in the presence of *Empetrum*. Also, the reduction of forb and grass biomass at higher *Rangifer* densities (Bråthen et al., 2007b) may come as a consequence of *Empetrum* being a dominant species in most plant communities: The relative grazing pressure on forbs and grasses probably increases when co-occurring with *Empetrum*. Critiques of the ecological sustainability of reindeer husbandry (Moen and Danell, 2003) may thus be valid if they are linked to a trajectory of niche construction by *Empetrum,* promoted by climate warming and more intense *Rangifer* land use, as it may have subsequent cascading effects onto the whole ecosystem.

4.5. *Implications for model predictions*

Shrub expansion following warming in the Arctic is linked to plant longevity and persistence indicating that model assumptions of fast

Table 1
Total and average species pool and aboveground biomass presented per growth form and for *Empetrum nigrum* and in total across the tundra plant communities included in this study.

Growth form	Species pool per growth form	Average species pool (mean ± SD)	Aboveground biomass (mean g m^{-2} ± SD)
Forbs	105	1.18 ± 1.84	5.4 ± 11.6
Grasses	23	1.34 ± 1.47	9.1 ± 17
Sedges	47	1.31 ± 1.37	7.8 + 14.8
Deciduous shrubs	17	2.14 ± 1.14	63.5 ± 58.3
Evergreen shrubs*	9	1.02 ± 0.93	24 ± 32.4
Vascular cryptogams	21	0.16 ± 0.44	0.9 ± 4.0
Empetrum nigrum		*0.85 ± 0.35***	118 ± 106.6
Total per plant community		8.08 ± 4.34	228.7 ± 146.6

* Not including *Empetrum*.
** Average presence of *Empetrum* across all plant communities.

response rates to climate warming are overestimated (Büntgen et al., 2015). The identified linkages between shrub expansion and *Empetrum* and *Rangifer* in this study further indicate that model assumptions of climate responses may be overestimated for areas where species with niche constructing capacity are common. *Empetrum* and to some extent *Rangifer,* as results of this study may indicate, improve niches for long-lived shrubs and deteriorate niches for herbaceous plants. In consequence plant longevity and persistence are favoured over higher process rates and short reaction time to climate warming. Such niche construction may also limit the range expansions of southern species into circumpolar areas (Virtanen et al., 2010) as tundra communities will be less suited as stepping-stones to plants tracking their climatic niche. Accordingly, there is lower variability in warm thermal limits in Fennoscandia as opposed to the Alps, as shown for the climatic niches of species common to the Alps and Fennoscandia (Pellissier et al., 2013). Also, whereas the current assessment of the Arctic acknowledges a controlling impact of herbivores on the encroachment of shrubs under climate warming (Ims et al., 2013), our study shows that the encroachment of a particular shrub, i.e. *Empetrum*, is more likely enforced, although marginally, by herbivory. As such this study substantiates the call for species specific studies on shrub encroachment (Myers-Smith et al., 2011) and further substantiates the conclusion of previous studies to include *Empetrum* (Le Roux et al., 2014; Pellissier et al., 2010) and *Rangifer* (Ims et al., 2013) into predictive models of tundra ecosystem change. In summary, the capacity of *Empetrum* to cause a decoupling with soil processes (Wardle et al., 2012), be unpalatable or avoided by herbivores (Danell et al., 1994; Ims et al., 2007; Iversen et al., 2014) and be competitively superior to herbaceous plants (Mod et al., 2014; Pellissier et al., 2010) suggests that an increase in its abundance is likely to drive a trajectory of tundra communities and ecosystems

Table 2
Standardized coefficients (± 95% C.I.) of fixed effects predictors extracted from mixed effects models on community and *Empetrum* characteristics, along with their random factors (SD ± 95% C.I.). All models include temperature (warmest quarter), *Empetrum* biomass and *Rangifer* density (high vs. low) as predictors along with their significant interactions. The evergreen shrub biomass is not including *Empetrum* biomass, and the inflorescences are not including *Empetrum* inflorescences. Statistically significant predictors have confidence intervals that do not overlap with zero, and non-significant predictors are presented in grey. Growth form names are short for their biomasses, whereas berries and inflorescences are short for their point intercepts.

	Predictors, fixed variables				**Random factors**	
	Temperature	*Empetrum* biomass	*Rangifer* density	Temp x *Empetrum* bm	District pairs	Landscape areas
Response variables						
Empetrum nigrum						
Biomass	0.642 (0.492, 0.792)	-	0.316 (0.005, 0.627)	-	0.308 (0.123, 0.771)	0.792 (0.658, 0.954)
Berries	0.016 (-0.003, 0.140)	0.066 (0.052, 0.081)	-0.032 (-0.064, -0.0001)	0.023 (0.010, 0.036)	0.043 (0.023, 0.081)	0.052 (0.035, 0.077)
Community characteristics						
Species richness	0.204 (0.159, 0.250)	-0.035 (-0.060, -0.010)	-0.057 (-0.150, 0.035)		0.213 (0.126, 0.362)	0.245 (0.208, 0.288)
Inflorescences	0.083 (0.050, 0.117)	-0.079 (-0.101, -0.056)	-0.080 (-0.142, -0.018)	-0.052 (-0.073, -0.030)	0.066 (0.031, 0.139)	0.139 (0,109. 0.175)
Herbaceous plants	0.446 (0.340, 0.553)	-0.343 (-0.407, -0.278)	-0.137 (-0.346, 0.072)	-0.077 (-0.137, -0.017)	0.307 (0.168, 0.562)	0.527 (0.443, 0.628)
Woody plants	0.281 (0.188, 0.375)	0.299 (0.246, 0.352)	-0.038 (-0.226, 0.150)	0.065 (0.016, 0.114)	0.450 (0.265, 0.765)	0.496 (0.422, 0.583)
Growth forms						
Forbs	0.325 (0.221, 0.428)	-0.236 (-0.295, -0.177)	-0.280 (-0.488, -0.072)		0.403 (0.234, 0.693)	0.545 (0.460, 0.646)
Grasses	0.471 (0.363, 0.579)	-0.136 (-0.201, -0.071)	-0.265 (-0.469, -0.062)		0.210 (0.096, 0.462)	0.504 (0.419, 0.607)
Sedges	0.183 (0.082, 0.283)	-0.289 (-0.352, -0.226)	0.165 (-0.032, 0.363)		0.399 (0.233, 0.681)	0.488 (0.406, 0.587)
Deciduous shrubs	0.320 (0.216, 0.425)	0.200 (0.141, 0.259)	-0.025 (-0.233, 0.184)		0.451 (0.260, 0.785)	0.545 (0.464, 0.641)
Evergreen shrubs	0.192 (0.063, 0.322)	0.596 (0.515, 0.676)	0.006 (-0.222, 0.234)		0.617 (0.369, 1.030)	0.527 (0.425, 0.653)

towards slower process rates and lower biodiversity. Such change may be indicative of ecosystem retrogression (Peltzer et al., 2010; Wardle et al., 2004). As such our results substantiate the importance of understanding the dynamics of niche constructor species and include them in predictive models of climate change.

4.6. Conclusion

In this study, tundra plant community changes along a temperature gradient were clearly modified by the presence of the common allelopathic dwarf shrub *Empetrum nigrum* and modified to a marginal degree by the large herbivore *Rangifer tarandus*. More specifically we found the increase in herbaceous plant biomass with increasing temperature to be reduced in the presence of *Empetrum,* whereas the biomass of woody plants was enhanced. Importantly, we also found the biomass of *Empetrum* to increase with increasing temperature. Higher *Rangifer* density negatively affected biomass of forbs and grasses and abundance of inflorescences, and positively, although to a small extent, *Empetrum* biomass. Based on these results we argue for the inclusion of niche constructor species in predictive models of climate change due to their ecosystem-modifying role.

Acknowledgements

This is a study associated to the climate-ecological observatory for Arctic tundra (COAT). We are thankful to Metha Klock and Anna-Katharina Pilsbacher for providing thoughtful comments to a previous version of this study. The data sampling received funding from the Norwegian Research Council through the project Ecosystem Finnmark under leadership by Rolf A. Ims.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.ppees.2017.06.005.

References

Aerts, R., Cornelissen, J.H.C., Dorrepaal, E., 2006. Plant performance in a warmer world: general responses of plants from cold, northern biomes and the importance of winter and spring events. Plant Ecol. 182, 65–77.

Aerts, R., 2010. Nitrogen-dependent recovery of subarctic tundra vegetation after simulation of extreme winter warming damage to Empetrum hermaphroditum. Global Change Biol. 16, 1071–1081.

Büntgen, U., Hellmann, L., Tegel, W., et al., 2015. Temperature-induced recruitment pulses of Arctic dwarf shrub communities. J. Ecol. 103, 489–501.

Bell, J.N.B., Tallis, J.H., 1973. Biological flora of british-isles – empetrum nigrum l. J. Ecol. 61, 289–305.

Bernes, C., Bråthen, K.A., Forbes, B.C., Speed, J.D., Moen, J., 2015. What are the impacts of reindeer/caribou (Rangifer tarandus L.) on arctic and alpine vegetation? A systematic review. Environ. Evid. 4, 1–26.

Blois, J.L., Williams, J.W., Fitzpatrick, M.C., Jackson, S.T., Ferrier, S., 2013. Space can substitute for time in predicting climate-change effects on biodiversity. Proc. Natl. Acad. Sci. 110, 9374–9379.

Bokhorst, S., Bjerke, J.W., Bowles, F.W., Melillo, J., Callaghan, T.V., Phoenix, G.K., 2008. Impacts of extreme winter warming in the sub-Arctic: growing season responses of dwarf shrub heathland. Global Change Biol. 14, 2603–2612.

Bråthen, K.A., Hagberg, O., 2004. More efficient estimation of plant biomass. J. Veg. Sci. 15, 653–660.

Bråthen, K.A., Lortie, C., 2016. A portfolio effect of shrub canopy height on species richness in both stressful and competitive environments. Funct. Ecol. 30, 60–69.

Bråthen, K.A., Ravolainen, V., 2015. Niche construction by growth forms is as strong a predictor of species diversity as environmental gradients. J. Ecol. 103, 701–713.

Bråthen, K.A., González, V.T., Iversen, M., Killengreen, S., Ravolainen, V.T., Ims, R.A., Yoccoz, N.G., 2007a. Endozoochory varies with ecological scale and context. Ecography 30, 308–320.

Bråthen, K.A., Ims, R.A., Yoccoz, N.G., Fauchald, P., Tveraa, T., Hausner, V.H., 2007b. Induced shift in ecosystem productivity? Extensive scale effects of abundant large herbivores. Ecosystems 10, 773–789.

Bråthen, K.A., Fodstad, C.H., Gallet, C., 2010. Ecosystem disturbance reduces the allelopathic effects of Empetrum hermaphroditum humus on tundra plants. J. Veg. Sci. 21, 786–795.

Buizer, B., Weijers, S., Van Bodegom, P.M., et al., 2012. Range shifts and global warming: ecological responses of Empetrum nigrum L. to experimental warming at its northern (high Arctic) and southern (Atlantic) geographical range margin. Environ. Res. Lett. 7, 025501.

Buntgen, U., Hellmann, L., Tegel, W., et al., 2015. Temperature-induced recruitment pulses of Arctic dwarf shrub communities. J. Ecol. 103, 489–501.

Chapin, F.S., Bretharte, M.S., Hobbie, S.E., Zhong, H.L., 1996. Plant functional types as predictors of transient responses of arctic vegetation to global change. J. Veg. Sci. 7, 347–358.

Chapin, F.S., Sturm, M., Serreze, M.C., et al., 2005. Role of land-surface changes in Arctic summer warming. Science 310, 657–660.

Chen, I.-C., Hill, J.K., Ohlemüller, R., Roy, D.B., Thomas, C.D., 2011. Rapid range shifts of species associated with high levels of climate warming. Science 333, 1024–1026.

Christie, K.S., Bryant, J.P., Gough, L., Ravolainen, V.T., Ruess, R.W., Tape, K.D., 2015. The role of vertebrate herbivores in regulating shrub expansion in the arctic: a synthesis. Bioscience 65, 1123–1133.

Collantes, M.B., Anchorena, J., Cingolani, A.M., 1999. The steppes of Tierra del Fuego: floristic and growthform patterns controlled by soil fertility and moisture. Plant Ecol. 140, 61–75.

Cornelissen, J.H.C., Quested, H.M., Gwynn-Jones, D., et al., 2004. Leaf digestibility and litter decomposability are related in a wide range of subarctic plant species and types. Funct. Ecol. 18, 779–786.

Cornelissen, J.H.C., Van Bodegom, P.M., Aerts, R., et al., 2007. Global negative vegetation feedback to climate warming responses of leaf litter decomposition rates in cold biomes. Ecol. Lett. 10, 619–627.

Danell, K., Utsi, P.M., Palo, R.T., Eriksson, O., 1994. Food plant selection by reindeer during winter in relation to plant quality. Ecography 17, 153–158.

Daubenmire, R., 1968. Plant Communities. A Textbook of Plant Synecology. New York, Harper & Row.

De Long, J.R., Dorrepaal, E., Kardol, P., Nilsson, M.-C., Teuber, L.M., Wardle, D.A., 2016. Understory plant functional groups and litter species identity are stronger drivers of litter decomposition than warming along a boreal forest post-fire successional gradient. Soil Biol. Biochem. 98, 159–170.

Den Herder, M., Virtanen, R., Roininen, H., 2008. Reindeer herbivory reduces willow growth and grouse forage in a forest-tundra ecotone. Basic Appl. Ecol. 9, 324–331.

Elmendorf, S.C., Henry, G.H.R., Hollister, R.D., et al., 2012a. Global assessment of experimental climate warming on tundra vegetation: heterogeneity over space and time. Ecol. Lett. 15, 164–175.

Elmendorf, S.C., Henry, G.H.R., Hollister, R.D., et al., 2012b. Plot-scale evidence of tundra vegetation change and links to recent summer warming. Nat. Clim. Change 2, 453–457.

Elmendorf, S.C., Henry, G.H.R., Hollister, R.D., et al., 2015. Experiment, monitoring, and gradient methods used to infer climate change effects on plant communities yield consistent patterns. Proc. Natl. Acad. Sci. 112, 448–452.

Epstein, H.E., Raynolds, M.K., Walker, D.A., Bhatt, U.S., Tucker, C.J., Pinzon, J.E., 2012. Dynamics of aboveground phytomass of the circumpolar Arctic tundra during the past three decades. Environ. Res. Lett. 7, 015506.

Felde, V.A., Kapfer, J., Grytnes, J.-A., 2012. Upward shift in elevational plant species ranges in Sikkilsdalen, central Norway. Ecography 35, 922–932.

Francini, G., Liiri, M., Männistö, M., Stark, S., Kytöviita, M.-M., 2014. Response to reindeer grazing removal depends on soil characteristics in low Arctic meadows. Appl. Soil Ecol. 76, 14–25.

Gallet, C., Nilsson, M.C., Zackrisson, O., 1999. Phenolic metabolites of ecological significance in Empetrum hermaphroditum leaves and associated humus. Plant Soil 210, 1–9.

González, V.T., Bråthen, K.A., Ravolainen, V.T., Iversen, M., Hagen, S.B., 2010. Large-scale grazing history effects on Arctic-alpine germinable seed banks. Plant Ecol. 207, 321–331.

González, V.T., Junttila, O., Lindgård, B., Reiersen, R., Trost, K., Bråthen, K.A., 2015. Batatasin-III and the allelopathic capacity of Empetrum nigrum. Nordic J. Bot. 33, 225–231.

Gottfried, M., Pauli, H., Futschik, A., et al., 2012. Continent-wide response of mountain vegetation to climate change. Nat. Clim. Change 2, 111–115.

Graae, B.J., Ejrnæs, R., Lang, S.I., Meineri, E., Ibarra, P.T., Bruun, H.H., 2011. Strong microsite control of seedling recruitment in tundra. Oecologia 166, 565–576.

Hijmans, R.J., Cameron, S.E., Parra, J.L., Jones, P.G., Jarvis, A., 2005. Very high resolution interpolated climate surfaces for global land areas. Int. J. Climatol. 25, 1965–1978.

Hobbie, S.E., 1992. Effects of plant species on nutrient cycling. Trends Ecol. Evol. 7, 336–339.

Huntington, H., 2013. Provisioning and cultural services. In: Meltofte, H. (Ed.), Arctic Biodiversity Assessement. Status and Trends in Arctic Biodiversity. Conservation of Arctic Flora and Fauna, Akureyri (pp Page).

Ims, R.A., Yoccoz, N.G., Bråthen, K.A., Fauchald, P., Tveraa, T., Hausner, V., 2007. Can reindeer overabundance cause a trophic cascade? Ecosystems 10, 607–622.

Ims, R.A., Henden, J.-A., Killengreen, S.T., 2008. Collapsing population cycles. Trends Ecol. Evol. 23, 79–86.

Ims, R.A., Ehrich, D., Forbes, B.C., et al., 2013. Terrestrial ecosystems. In: Meltofte, H. (Ed.), Arctic Biodiversity Assessment. Status and Trends in Arctic Biodiversity. Conservation of Arctic Flora and Fauna, Akureyri, Iceland (pp Page).

Iversen, M., Fauchald, P., Langeland, K., Ims, R.A., Yoccoz, N.G., Bråthen, K.A., 2014. Phenology and cover of plant growth forms predict herbivore habitat selection in a high latitude ecosystem. PLoS One 9 (6), e100780.

Johansen, B., Tømmervik, H., Karlsen, S.R., 1995. Vegetasjons- og beitekartlegging i Finnmark og Nord-Troms. NORUT Informasjonsteknologi AS, Tromsø, Norway (pp Page).

Körner, C., 2003. Alpine Plant Life: Functional Plant Ecology of High Mountain Ecosystems. Springer, Berlin.

Kaarlejärvi, E., Baxter, R., Hofgaard, A., et al., 2012. Effects of warming on shrub abundance and chemistry drive ecosystem-level changes in a forest?tundra ecotone. Ecosystems 15, 1219–1233.

Kaarlejärvi, E., Eskelinen, A., Olofsson, J., 2013. Herbivory prevents positive responses of lowland plants to warmer and more fertile conditions at high altitudes. Funct. Ecol. 27, 1244–1253.

Klanderud, K., Birks, H.J.B., 2003. Recent increases in species richness and shifts in altitudinal distributions of Norwegian mountain plants. Holocene 13, 1–6.

Klein, J.A., Harte, J., Zhao, X.Q., 2004. Experimental warming causes large and rapid species loss, dampened by simulated grazing, on the Tibetan Plateau. Ecol. Lett. 7, 1170–1179.

Kullman, L., 2010. A richer, greener and smaller alpine world: review and projection of warming-induced plant cover change in the Swedish Scandes. Ambio 39, 159–169.

Larsen, K.S., Michelsen, A., Jonasson, S., Beier, C., Grogan, P., 2012. Nitrogen uptake during fall, winter and spring differs among plant functional groups in a subarctic heath ecosystem. Ecosystems 15, 927–939.

Le Roux, P.C., Pellissier, L., Wisz, M.S., Luoto, M., 2014. Incorporating dominant species as proxies for biotic interactions strengthens plant community models. J. Ecol. 102, 767–775.

Lenoir, J., Gégout, J.C., Marquet, P.A., De Ruffray, P., Brisse, H., 2008. A significant upward shift in plant species optimum elevation during the 20th century. Science 320, 1768–1771.

Mörsdorf, M.A., Ravolainen, V.T., Støvern, L.E., Yoccoz, N.G., Jónsdóttir, I.S., Bråthen, K.A., 2015. Definition of sampling units begets conclusions in ecology: the case of habitats for plant communities. Peer J. 3, e815.

Matthews, B., De Meester, L., Jones, C.G., et al., 2014. Under niche construction: an operational bridge between ecology, evolution, and ecosystem science. Ecol. Monogr. 84, 245–263.

Michelsen, A., Schmidt, I., Jonasson, S., Quarmby, C., Sleep, D., 1996. Leaf 15N abundance of subarctic plants provides field evidence that ericoid, ectomycorrhizal and non-and arbuscular mycorrhizal species access different sources of soil nitrogen. Oecologia 105, 53–63.

Mod, H.K., Le Roux, P.C., Luoto, M., 2014. Outcomes of biotic interactions are dependent on multiple environmental variables. J. Veg. Sci. 25, 1024–1032.

Mod, H.K., Heikkinen, R.K., Le Roux, P.C., Wisz, M.S., Luoto, M., 2016. Impact of biotic interactions on biodiversity varies across a landscape. Journal of Biogeography 43, 2412–2423 n/a-n/a.

Moen, J., Danell, Ö, 2003. Reindeer in the Swedish mountains: an assessment of grazing impacts. Ambio 32, 397–402.

Moore, I.D., Grayson, R.B., Landson, A.R., 1991. Digital terrain modelling: a review of hydrological, geomorphological, and biological applications. Hydrol. Processes 5, 3–30.

Myers-Smith, I.H., Forbes, B.C., Wilmking, M., et al., 2011. Shrub expansion in tundra ecosystems: dynamics, impacts and research priorities. Environ. Res. Lett. 6.

Myers-Smith, I.H., Elmendorf, S.C., Beck, P.S.A., et al., 2015. Climate sensitivity of shrub growth across the tundra biome. Nature Clim. Change 5, 887–891.

Nilsson, M.C., Wardle, D.A., 2005. Understory vegetation as a forest ecosystem driver: evidence from the northern Swedish boreal forest. Front. Ecol. Environ. 3, 421–428.

Nilsson, M.C., Zackrisson, O., 1992. Inhibition of Scots pine seedling establishment by Empetrum hermaphroditum. J. Chem. Ecol. 18, 1857–1870.

Nilsson, M.C., Hogberg, P., Zackrisson, O., Wang, F.Y., 1993. Allelopathic effects by Empetrum hermaphroditum on development and nitrogen uptake by roots and mycorrhizae of Pinus sylvestris. Can. J. Bot. 71, 620–628.

Oberski, D., 2014. lavaan.survey: an R package for complex survey analysis of structural equation models. J. Stat. Softw. 57, 1–27.

Odling-Smee, J., Erwin, D.H., Palkovacs, E.P., Feldman, M.W., Laland, K.N., 2013. Niche construction theory: a practical guide for ecologists. Q. Rev. Biol. 88, 3–28.

Olofsson, J., Kitti, H., Rautiainen, P., Stark, S., Oksanen, L., 2001. Effects of summer grazing by reindeer on composition of vegetation, productivity and nitrogen cycling. Ecography 24, 13–24.

Olofsson, J., Oksanen, L., Callaghan, T., Hulme, P.E., Oksanen, T., Suominen, O., 2009. Herbivores inhibit climate-driven shrub expansion on the tundra. Global Change Biol. 15, 2681–2693.

Olofsson, J., Ericson, L., Torp, M., Stark, S., Baxter, R., 2011. Carbon balance of Arctic tundra under increased snow cover mediated by a plant pathogen. Nat. Clim. Change 1, 220–223.

Olofsson, J., Tømmervik, H., Callaghan, T.V., 2012. Vole and lemming activity observed from space. Nat. Clim. Change 2, 880–883.

Pajunen, A.M., Oksanen, J., Virtanen, R., 2011. Impact of shrub canopies on understorey vegetation in western Eurasian tundra. J. Veg. Sci. 22, 837–846.

Pellissier, L., Bråthen, K.A., Pottier, J., et al., 2010. Species distribution models reveal apparent competitive and facilitative effects of a dominant species on the distribution of tundra plants. Ecography 33, 1004–1014.

Pellissier, L., Bråthen, K.A., Vittoz, P., et al., 2013. Thermal niches are more conserved at cold than warm limits in arctic-alpine plant species. Global Ecol. Biogeogr. 22, 933–941.

Peltzer, D.A., Wardle, D.A., Allison, V.J., et al., 2010. Understanding ecosystem retrogression. Ecol. Monogr. 80, 509–529.

Pinheiro, J.C., Bates, D.M., 2000. Linear Mixed-effects Models: Basic Concepts and Examples. Springer, New York.

Popp, M., Mirré, V., Brochmann, C., 2011. A single Mid-Pleistocene long-distance dispersal by a bird can explain the extreme bipolar disjunction in crowberries (Empetrum). Proc. Natl. Acad. Sci. 108, 6520–6525.

Post, E., 2013. Erosion of community diversity and stability by herbivore removal under warming. Proc. R. Soc. Lond. B: Biol. Sci. 280, 2012–2722.

Preece, C., Phoenix, G.K., 2014. Impact of early and late winter icing events on sub-arctic dwarf shrubs. Plant Biol. 16, 125–132.

Preece, C., Callaghan, T.V., Phoenix, G.K., 2012. Impacts of winter icing events on the growth, phenology and physiology of sub-arctic dwarf shrubs. Physiol. Plant. 146, 460–472.

Ravolainen, V.T., Yoccoz, N.G., Bråthen, K.A., Ims, R.A., Iversen, M., González, V.T., 2010. Additive partitioning of diversity reveals no scale-dependent impacts of large ungulates on the structure of tundra plant communities. Ecosystems 13, 157–170.

Ravolainen, V.T., Bråthen, K.A., Yoccoz, N.G., Nguyen, J.K., Ims, R.A., 2014. Complementary impacts of small rodents and semi-domesticated ungulates limit tall shrub expansion in the tundra. J. Appl. Ecol. 51, 234–241.

Rosseel, Y., Oberski, D., Byrnes, J., Vanbrabant, L., Savalei, V., 2013. Lavaan: Latent Variable Analysis. R Foundation for Statistical Computing (pp Page).

Rustad, L., Campbell, J., Marion, G., et al., 2001. A meta-analysis of the response of soil respiration, net nitrogen mineralization, and aboveground plant growth to experimental ecosystem warming. Oecologia 126, 543–562.

Soininen, E.M., Ravolainen, V.T., Bråthen, K.A., Yoccoz, N.G., Gielly, L., Ims, R.A., 2013a. Arctic small rodents have diverse diets and flexible food selection. PLoS One 8, e68128.

Soininen, E.M., Zinger, L., Gielly, L., et al., 2013b. Shedding new light on the diet of Norwegian lemmings: DNA metabarcoding of stomach content. Polar Biol. 36, 1069–1076.

Sorensen, P.L., Clemmensen, K.E., Michelsen, A., Jonasson, S., Ström, L., 2008. Plant and microbial uptake and allocation of organic and inorganic nitrogen related to plant growth forms and soil conditions at two subarctic tundra sites in Sweden. Arctic Antarctic Alpine Res. 40, 171–180.

Tarbuck, E.J., Lutgens, F.K., 1992. The Earth. An Introduction to Physical Geology. Macmillan Publishing Company, New York.

Tveraa, T., Fauchald, P., Gilles Yoccoz, N., Anker Ims, R., Aanes, R., Arild Høgda, K., 2007. What regulate and limit reindeer populations in Norway? Oikos 116, 706–715.

Tybirk, K., Nilsson, M.C., Michelson, A., et al., 2000. Nordic Empetrum dominated ecosystems: function and susceptibility to environmental changes. Ambio 29, 90–97.

Väisänen, M., Martz, F., Kaarlejärvi, E., Julkunen-Tiitto, R., Stark, S., 2013. Phenolic responses of mountain crowberry (Empetrum nigrum ssp. hermaphroditum) to global climate change are compound specific and depend on grazing by reindeer (Rangifer tarandus). J. Chem. Ecol. 39, 1390–1399.

Väisänen, M., Ylanne, H., Kaarlejärvi, E., Sjögersten, S., Olofsson, J., Crout, N., Stark, S., 2014. Consequences of warming on tundra carbon balance determined by reindeer grazing history. Nat. Clim. Change 4, 384–388.

Virtanen, R., Luoto, M., Rämä, T., Mikkola, K., Hjort, J., Grytnes, J.A., Birks, H.J.B., 2010. Recent vegetation changes at the high-latitude tree line ecotone are controlled by geomorphological disturbance, productivity and diversity. Global Ecol. Biogeogr. 19, 810–821.

Wada, N., Shimono, M., Miyamoto, M., Kojima, S., 2002. Warming effects on shoot developmental growth and biomass production in sympatric evergreen alpine dwarf shrubs Empetrum nigrum and Loiseleuria procumbens. Ecol. Res. 17, 125–132.

Walker, D.A., Raynolds, M.K., Daniels, F.J.A., et al., 2005. The circumpolar arctic vegetation map. J. Veg. Sci. 16, 267–282.

Wallstedt, A., Sommarin, M., Nilsson, M.C., Munson, A.D., Margolis, H.A., 2001. The inhibition of ammonium uptake in excised birch (Betula pendula) roots by batatasin-III. Physiol. Plant. 113, 368–376.

Wallstedt, A., Gallet, C., Nilsson, M.C., 2005. Behaviour and recovery of the secondary metabolite batatasin-III from boreal forest humus: influence of temperature, humus type and microbial community. Biochem. Syst. Ecol. 33, 385–407.

Wardle, D.A., Jonsson, M., 2013. Long-term resilience of above- and belowground ecosystem components among contrasting ecosystems. Ecology 95, 1836–1849.

Wardle, D.A., Nilsson, M.C., Gallet, C., Zackrisson, O., 1998. An ecosystem-level perspective of allelopathy. Biol. Rev. Camb. Philos. Soc. 73, 305–319.

Wardle, D.A., Walker, L.R., Bardgett, R.D., 2004. Ecosystem properties and forest decline in contrasting long-term chronosequences. Science 305, 509–513.

Wardle, D.A., Gundale, M.J., Jäderlund, A., Nilsson, M.-C., 2012. Decoupled long-term effects of nutrient enrichment on aboveground and belowground properties in subalpine tundra. Ecology 94, 904–919.

Wilson, S.D., Nilsson, C., 2009. Arctic alpine vegetation change over 20 years. Global Change Biol. 15, 1676–1684.

Wilson, P.J., Thompson, K., Hodgson, J.G., 1999. Specific leaf area and leaf dry matter content as alternative predictors of plant strategies. New Phytol. 143, 155–162.

Wipf, S., Rixen, C., Mulder, C.P.H., 2006. Advanced snowmelt causes shift towards positive neighbour interactions in a subarctic tundra community. Global Change Biol. 12, 1496–1506.

Wipf, S., 2010. Phenology, growth, and fecundity of eight subarctic tundra species in response to snowmelt manipulations. Plant Ecol. 207, 53–66.

Wookey, P.A., Aerts, R., Bardgett, R.D., et al., 2009. Ecosystem feedbacks and cascade processes: understanding their role in the responses of Arctic and alpine ecosystems to environmental change. Global Change Biol. 15, 1153–1172.

Ylänne, H., Stark, S., Tolvanen, A., 2015. Vegetation shift from deciduous to evergreen dwarf shrubs in response to selective herbivory offsets carbon losses: evidence from 19 years of warming and simulated herbivory in the subarctic tundra. Global Change Biol. 21, 3696–3711.

Zamin, T.J., Grogan, P., 2013. Caribou exclusion during a population low increases deciduous and evergreen shrub species biomass and nitrogen pools in low Arctic tundra. J. Ecol. 101, 671–683.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 82–88

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Reciprocal interactions between a facilitator, natives, and exotics in tropical alpine plant communities[☆]

Luis D. Llambí[a,*], Nicole Hupp[b], Agustin Saez[c], Ragan Callaway[d]

[a] Instituto de Ciencias Ambientales y Ecológicas, Universidad de Los Andes, Mérida 5101, Venezuela
[b] Division of Biological Sciences and Institute on Ecosystems, University of Montana, Missoula, MT, 59812, USA
[c] Universidad Nacional del Comahue and INIBIOMA-CONICET, 8400 San Carlos de Bariloche, Río Negro, Argentina
[d] Division of Biological Sciences and Institute on Ecosystems, University of Montana, Missoula, MT, 59812, USA

ARTICLE INFO

Keywords:
Competition
Facilitation
Feedback effects
Invasive species
Indirect interactions
Nurse plants

ABSTRACT

Facilitation by nurse plants has received considerable attention, but the feedback effects of beneficiaries on the benefactor fitness remain comparatively unexplored. In particular, to our knowledge there have been no studies of feedback effects of exotic species on nurse plants. Furthermore, there have been few studies of how exotic plants interact with native beneficiaries, potentially generating indirect effects on the nurse. Here, through spatial association analyses, we estimate the reciprocal interactions between the cushion nurse-plant *Arenaria musciformis,* native beneficiaries, and the dominant exotic *Rumex acetosella* (a fast growing heliophile species) in a tropical alpine ecosystem of the Venezuelan Andes, locally known as *páramos*. We recorded the density of natives and the exotic within *Arenaria* cushions and in paired open areas at three sites and calculated Relative Interaction Indices (RII) to explore the effects of the cushion. Then, we analyzed the feedback effects of *Rumex* and the natives on the density of flowers produced by *Arenaria*. Finally, we analyzed how *Rumex* abundance is related to the abundance of native species inside and outside cushions. RIIs indicated a facilitative effect of cushions on the abundance of native plants, but no effect on *Rumex*. We found a negative relationship between the density of natives and cushion flower density, but no such relationship for *Rumex*. However, at high densities, *Rumex* was negatively correlated with the abundance of the natives both inside and outside of cushions. Our results suggest the possibility of complex reciprocal interactions between nurses, natives and exotics, and that native and exotic plants can differ in their feedback effects on nurse cushions. Native plants appeared to be facilitated by cushions and then develop a parasitic relationship with their nurses. In contrast, the exotic *Rumex* had a neutral interaction with the nurse, but competitive effects on the native beneficiary community, which could indirectly modulate the feedback effects of natives on the nurse. These results highlight the importance of analyzing direct and indirect reciprocal interactions and the impacts of exotic invaders for understanding alpine community assembly.

1. Introduction

Facilitative interactions are important processes in plant community assembly (Lortie et al., 2004; Brooker et al., 2008). In particular, there is a large body of evidence for positive interactions driving increases in species abundance and diversity in low-productivity environments (Callaway 2007; Cavieres et al., 2014). We know much about the effects of facilitators on other species, but we know little about the reciprocal feedback effects (RFE) of the beneficiaries on the fitness or performance of the facilitator, or “nurse” species (Schöb et al., 2014a, 2014b; García et al., 2016). Understanding the nature of the bi-directional interaction between nurses and beneficiaries (i.e. parasitism, commensalism or mutualism) has implications for the evolution of these interactions (Bronstein 2009; Brooker and Callaway 2009). For example, if beneficiaries have negative feedback effects on the fitness of benefactors, this could select for mechanisms employed by the nurse plant to reduce the abundance of beneficiaries or to tolerate their negative effects (Schöb et al., 2014b).

A few studies in semi-arid ecosystems have shown that the RFEs of beneficiaries on benefactors can be positive (e.g. Pugnaire et al., 1996; Holzapfel and Mahall 1999). However, most research in desert and alpine communities has reported negative effects of beneficiaries on the

---

[☆] This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author.

E-mail addresses: ldllambi@gmail.com (L.D. Llambí), nicole.hupp@umontana.edu (N. Hupp), agustinsaezmail@gmail.com (A. Saez), ray.callaway@mso.umt.edu (R. Callaway).

http://dx.doi.org/10.1016/j.ppees.2017.05.002
Received 7 December 2016; Received in revised form 26 April 2017; Accepted 24 May 2017
Available online 30 May 2017
1433-8319/

growth or reproduction of the nurse (Valiente-Banuet et al., 1991; Flores-Martínez et al., 1994; Michalet et al., 2011; Cranston et al., 2012; Schöb et al., 2014a). In alpine ecosystems, where cushion plants commonly facilitate other species (e.g. Arroyo et al., 2003; Cavieres and Badano 2009; Yang et al., 2010; Cavieres et al., 2014), research at a global scale showed that beneficiary species predominantly have negative effects on the flower production and seed set of the cushions that host them (Schöb et al., 2014b). These RFEs were variable in their directionality and intensity depending on the environmental context, with high-productivity sites showing weaker negative effects of beneficiaries on the benefactor (Schöb et al., 2014c).

However, to our knowledge, there have been no studies of the RFEs of exotic species on nurse plants. These interactions may be particularly important in ecosystems where exotic species have become invasive. Exotic species are generally uncommon in alpine ecosystems and global research indicates that their richness decreases with altitude in mountains regions (Pauchard et al., 2009; Alexander et al., 2011; Seipel et al., 2012). This may be due to more frequent introductions at low elevations, strong environmental filtering, and low disturbance at higher elevations. However, close to two hundred exotic plant species have been reported in alpine areas worldwide and their impacts on native alpine plant communities remain largely unexplored (Alexander et al., 2016). Although many of these species are not considered invasive, this may change with increasing effects of climate and land use change in alpine areas, which are expected to promote invasion (Theurillat and Guisan, 2001; Alexander et al., 2016).

Natives and exotics could differ in their direct impact on nurse species due to differences in resource use strategies, and thus the intensity of their competitive effects on the benefactor (Callaway, 2007). For example, if exotics grow faster and have higher resource demand than natives in general, they could have a stronger negative feedback effect on the benefactor. This is reasonable to expect for introduced species with ruderal strategies in unproductive environments where stress tolerant strategies are likely to be favored in the natives (Tilman 1982). However, having evolved in environments with different selective pressures, there could be lower temporal or spatial niche overlap in resource use between exotic species and the nurse, which would reduce their impact on the benefactor fitness relative to natives. Differences in the effects of natives and exotics on the reproductive output of the nurse could also occur if they have different interactions with pollinators. This would depend on the degree to which natives and exotics share the same pollinators with the nurse, and whether exotics, natives and the nurse compete for pollinators (e.g. Muñoz and Cavieres, 2008Muñoz and Cavieres 2008; Morales and Traveset 2009) or alternatively, there are net facilitative effects through mutual attraction of pollinators (e.g. Moragues and Traveset, 2005).

Exotic species could also have important RFEs on nurse plants indirectly through their interactions with the native beneficiary community. In many invaded systems exotic plants suppress native species (Vilà et al., 2011; Besaw et al., 2011; Shah et al., 2014), and thus create scenarios in which indirect effects might be strong. In the temperate Chilean Andes, cushion nurses positively affect the abundance and performance of abundant invaders (Cavieres et al., 2005; Badano et al., 2007; Cavieres et al., 2008). In turn, this facilitation of invaders could ultimately suppress native species within cushion plants, and indirectly dampen the negative feedback effects of natives on the nurse (i.e. "a competitoŕs enemy is a friend", Levine, 1999; Callaway and Pennings, 2000). These types of indirect facilitation effects between nurses, natives and exotics have not been explored, especially in the context of studies on RFEs. More generally, indirect facilitation and non-transitive plant-plant interactions have received limited empirical attention, although they can be an important mechanisms for promoting species coexistence (particularly in species rich communities), and considerably expand our understanding of community assembly rules and species interdependence (Callaway 2007; Brooker et al., 2008; Filazzola and Lortie 2014; Aschehoug et al., 2016).

Here, we analyze the RFEs of native species and a dominant exotic (*Rumex acetosella* L.) on the flowering of a cushion nurse plant, *Arenaria musciformis* Triana & Planch, in a tropical alpine ecosystem in the high Andes of Venezuela. There has been little previous research on the role of facilitation and nurse plants on community structure in the tropical alpine of the Andes (see review by Anthelme and Dangles 2012). This represents an important knowledge gap since these systems, known as *páramos* in the Andes, comprise more than 90% of the global tropical alpine biome (Jacobsen, 2008). They also host a very diverse flora with outstanding levels of endemism (Simpson and Toddzia, 1990). In the páramos of Ecuador and Venezuela, cushion plants can increase local species richness and facilitate other species in the community, with facilitation increasing in importance at higher elevations (Sklenář, 2009; Anthelme et al., 2011; Hupp et al., 2017). In the high Venezuelan páramos, nurse shrubs also increase local species diversity, modify community structure and improve the performance of other associated species (Cáceres et al., 2015; Ramírez et al., 2015).

To our knowledge, RFEs of beneficiaries on nurses have only been studied at two sites in the páramo, in Ecuador and Venezuela, in the broader context of a global study on RFEs (Schöb et al., 2014c). In these tropical alpine sites, no feedback effect of beneficiaries on the flower production of cushions was found. However, this study did not distinguish between the effects of natives vs. exotic beneficiaries. In Venezuela, the exotic forb *Rumex acetosella* (called *Rumex* hereafter) has become a dominant herbaceous species both in cushions and in open spaces. *Rumex* is also one of the most widespread exotic species in the high tropical Andes. Consequently, this is an ideal study system to explore potential direct and indirect interactions among nurse cushions, native beneficiaries, and exotic beneficiaries. Here we investigate how native and exotic beneficiaries may differ in their feedback effect on nurses. We asked the following questions: 1) Does the cushion species *A. musciformis* facilitate the abundance of other native plants and the exotic *Rumex*? 2) Do the feedback effects of native beneficiaries on flower production of *A. musciformis* differ than those of *Rumex*? 3) What is the relationship between *Rumex* abundance and the density of native plants? And 4) Do these effects differ inside vs. outside of the cushions? We investigated these questions by comparing the abundance of native species and *Rumex* inside and outside of cushions. Then we reanalyzed data from Venezuela included in Schöb et al. (2014c) on the relationship between the number of flowers produced by *Arenaria* cushions and the density of beneficiaries inside cushions, but separating the effects of *Rumex* and the natives. Finally, we analyzed the relationship between the abundance of *Rumex* and the abundance of the natives inside and outside the nurse, to evaluate the possibility of indirect effects of the exotic on the nurse.

2. Methods

2.1. Study site

The study was conducted in the northern tropical Andes, in the Piedras Blancas páramo, Sierra de La Culata National Park, Venezuela, during January 2014, the dry season. Sites were located between 4250–4350 m, on northeast facing slopes with inclinations ranging between 15 and 25°. We sampled at three similar sites within a 5 km^2 area: Rio Azul (Lat 8.8866,Long −70.8685), Avenida (8.8847, −70.8666), and Gloria (8.8928, −70.8714). Mean annual temperature is 3 °C ± 2.7 and relatively constant throughout the year, but daily topsoil temperature can range from 40 °C during the day to less than −5 °C at night (Ramirez et al., 2015). This region has an average annual precipitation of 860 mm with a dry season between December and March (*Pico El Águila* weather station, 4118 m). The soils at the study sites are entisols, shallow, with a coarse sandy texture, acid and with low soil organic matter levels, and subject to constant disturbance by needle-ice formation due to daily freeze-thaw cycles (Pérez, 1987).

The plant community in these high páramo areas generally attains less than 40% cover and the vegetation is highly clustered. Cover is partitioned to some degree into two strata, one dominated by giant rosettes and shrubs, and the other consisting of cushions, grasses, forbs, and acaulescent rosettes (Cáceres et al., 2015). Our focal nurse plant, *Arenaria musciformis* (Caryophyllaceae, called *Arenaria* hereafter), is one of the most abundant cushion species in the region. It is endemic to the northern páramos of Colombia and Venezuela (3500 and 4500 m, Briceño and Morillo, 2002). The exotic invader forb *Rumex acetosella* has become the dominant species in terms of cover and density within the lower vegetation stratum in our study region, and it is the only exotic species present at our study sites (Cáceres et al., 2015). *Rumex* was introduced into the Venezuelan páramos from Europe with wheat cultivation during the 1800's (Salgado-Labouriau and Schubert, 1977). Sarmiento et al. (2003) and Llambí et al. (2003) described *Rumex* as a ruderal, fast growing heliophile species in páramos at lower elevations (between 3300 and 3800 m), which strongly dominates early stages of secondary succession in agricultural old-fields, being almost completely displaced by the natives after about 10 years. However, *Rumex* has expanded its distribution from the agricultural belt into high sub-nival environments, probably taking advantage of the increase in bare soil cover with elevation and reduced competition with tall native plants at high altitudes.

2.2. Sampling design

At each of the three study sites we haphazardly selected 35 individual *Arenaria* cushions within a 200 × 200 m area. We only sampled individuals with their maximum length measuring at least 20 cm. We placed a wire ring 20 cm in diameter on each cushion plant and recorded the identity and abundance (number of individuals) of all vascular plant species within. We defined individuals as ramets, as some species in the páramo, including the exotic *Rumex*, are interconnected underground. Then, we randomly sampled open substrate 1 m from each cushion (in 35 paired samples at each site) and measured the abundance of all plant species within the rings in the same way we did inside the cushion. Hence, a total of 105 samples were collected inside and 105 outside of cushions across the three sites. The average maximum length and width of the 105 *Arenaria* individuals included in the sample across the three sites was 51.6 × 29.3 cm. At each of the three sites we also quantified the cover of our focal cushion species using the line-intercept method (Greig-Smith, 1983).

To estimate the density of flowers within *Arenaria* cushions, we haphazardly placed five 10 × 2.5 cm micro-quadrats within each of the 105 cushion plants sampled, and counted all the flowers present within each micro-quadrat. The majority of the cushions in the research sites were in flower during our study.

2.3. Data analysis

We first calculated the total number of individuals (density) of all native vascular plant species found inside and outside of *Arenaria* cushions. We also calculated the density of *Rumex*. Then, to analyze the relationship of the cushion with the abundance of natives and *Rumex*, we calculated the Relative Interaction Index (*RII*, Armas et al., 2004) for each replicate pair using the formula $RII = N_{cushion} - N_{open}/N_{cushion} + N_{open}$, where N represents the density of the natives or invader species. We then averaged these values and calculated the 95% confidence interval across all replicate pairs combining the three sites and for each site separately. RII values range from 1 to −1, where positive values suggest a net facilitative effect (with the 95% confidence interval not including zero) and negative values indicate a net antagonistic effect of cushions on native/exotic densities. The average density of both natives and *Rumex* inside vs. outside of the cushions was also compared using a two-way permutational analysis of variance (PERMANOVA) using the Permanova + for Primer 6.0 software (Anderson et al., 2008). Here, we defined the local sampling situation as a fixed factor with two levels (inside vs. outside *Arenaria*) and the sampling site as a random factor with three levels (the three study sites).

To evaluate the feedback effect of both the density of natives and invaders on the number of flowers produced by *Arenaria,* we used a generalized linear mixed-effect model. Data were analyzed with the *glmmadmb* function from the *glmmADMB* package (Bolker et al., 2012) of the R software (version 2.15.1). Given that the response variable (i.e. number of flowers) corresponds to counts, we assumed a negative binomial error distribution with a log-link function. Density of native plants and the invader *Rumex* were included as additive fixed effects, and the sampling site as a random effect, allowing the intercept to vary among sites. Since we had five replicate flower density measurements *per* cushion plant, we defined the individual cushion identity as a random factor nested within the sites.

Finally, to analyze the effect of the density of the exotic *Rumex* on the total density of native plants, both inside and outside the cushions, we used linear mixed-effects models. Data analysis was carried out using the *lme* function from the *nlme* package (Pinheiro et al., 2015) of the R software (version 2.15.1), assuming a Gaussian error distribution, and a quadratic relationship between predicted and response variables. The density of *Rumex* and type of habitat (i.e. inside *vs* outside cushions) were included as interactive fixed effects, to evaluate if the quadratic relation between *Rumex* and Native densities depends on whether the plants are inside or outside cushions; the sampling site was included as a random effect. We log transformed the response variable (i.e. density of native pants) as *ln + 1* to fulfil the normality assumption (Shapiro-Wilk test).

3. Results

At the *Rio Azul* and *Avenida* sites, *Arenaria musciformis* comprised 2% of the total landscape cover, while at the *Gloria* site the cover of the cushion was 5%. The average size of each of the 105 cushions of *Arenaria* included in our sample was 0.49 ± 0.04 m^2 (with no differences between the three replicate sites).

3.1. Effect of cushions on native and invader densities

The Relative Interaction Index (RII) indicated a positive effect of *Arenaria* cushions on the density of native plants, but no effect on *Rumex* (Fig. 1). The average (± SE) density of native plants inside *Arenaria* was 75.8 ± 9.1 individuals m^{-2} *versus* 37.3 ± 6.2 per m^{-2} in open areas around cushions (*pseudoF* = 25.32; *p* = 0.0384). There

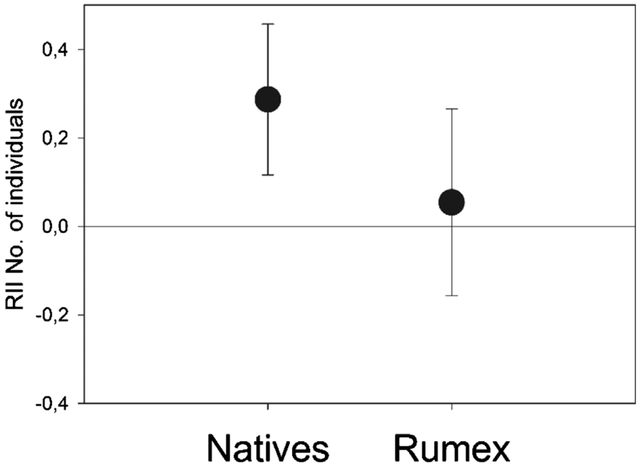


Fig. 1. Mean relative interaction index (RII) and ± 95% confidence levels for local total density of natives plants and the exotic invader *Rumex acetosella* sampled in rings placed inside and outside *Arenaria musciformis* cushion plants at three sites in the Venezuelan high páramo.

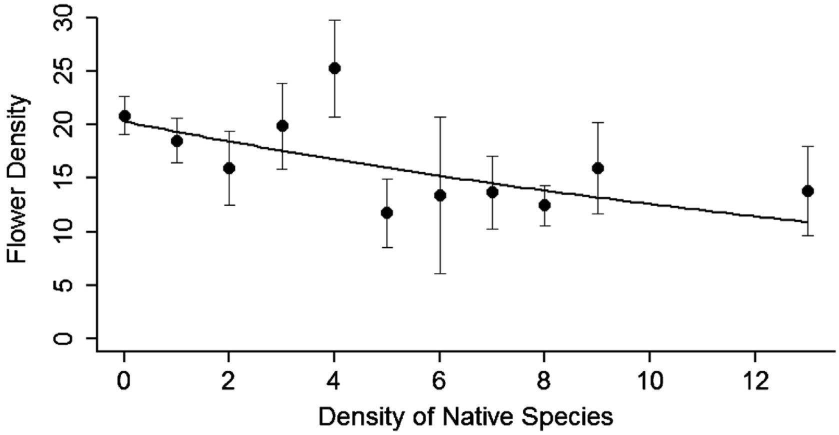


Fig. 2. Relationship between *Arenaria musciformis* flower density (flowers per micro-quadrat) as a function of the total density of native plants within the sampling rings. Symbols represent mean ± SE, while the solid curve the regression equation estimated from the negative binomial model.

was no significant site effect ($p = 0.3251$) and no interaction between site and microhabitat ($p = 0.6244$; i.e. the positive effect of cushions on native plants was consistent across the three sites). For *Rumex*, the average density inside the cushions was 27.6 ± 4.4 per m^2 versus 30.9 ± 5.1 per m^2 in the open areas, and this difference was not statistically significant (*pseudoF* = 1.33; $p = 0.3608$). There was a significant difference in the exotiće's abundance between the study sites (*pseudoF* = 15.90; $p = 0.001$), and no interaction between site and microhabitat (i.e., *Rumex* densities did not differ inside vs. outside of *Arenaria* in any of the three sites).

Both inside and outside of the cushions, species accumulation curves approached asymptotes indicating that our sampling represented most of the local species pool (see Hupp et al., 2017). Across the three sampling sites, more vascular plant species accumulated inside *Arenaria* cushions (30 spp.) than in the associated open samples (23 spp.).

3.2. Effects of native and invader densities on cushion flower density

The mixed-effects model for flower production by *Arenaria* as a function of the density of natives and invaders, showed a significant negative relationship between the density of natives and flower produced by the cushion (Fig. 2, $\beta_1 = -0.047$, $SE = 0.023$, $p = 0.038$), but not for the density of *Rumex* ($\beta_2 = 0.075$, $SE = 0.055$, $p = 0.137$). The estimated mean number of flowers produced by the cushions decreased 53%, from 20.3 flowers per micro-quadrat^{-1} when native plants were absent to 10.9 flowers per micro-quadrat^{-1} at the maximum native plant density observed within the cushions.

3.3. Effect of invader densities on native densities

Across our three study sites, *Rumex* represented in average 26.7% of the total plant density within *Arenaria* cushions and 45.3% of the total density in open areas. This made *Rumex* the dominant species in open areas, and co-dominant with the grass *Agrostis breviculmis* within cushions. There was a complex density-dependent relationship between the exotic *Rumex* and the density of native plants, both inside and outside the cushions. In both cases, the abundance of native species showed a hump-shaped relationship with the abundance of *Rumex* (Fig. 3). This was explained by a quadratic function, with parameters $\beta_1 = 0.676$, $SE = 0.084$, $p < 0.001$, $\beta_2 = 0.650$, $SE = 0.154$, $p < 0.001$, $\beta_3 = -0.142$, $SE = 0.037$, $p < 0.001$, inside the cushions, and $\beta_1 = 0.297$, $SE = 0.084$, $p < 0.001$, $\beta_2 = 0.599$, $SE = 0.154$, $p < 0.001$, $\beta_3 = -0.099$, $SE = 0.037$, $p = 0.001$, outside the cushions (Fig. 3). The only parameter of the quadratic function that was different between the two fitted curves was the intercept (i.e. β_1) (estimated ± SE = −0.37 ± 0.11, $p = 0.001$), which was higher inside the cushions. This indicates that when *Rumex* was not present, the density of native plants is higher inside cushions than outside. Aside from this, the linear (i.e. β_2) and quadratic (i.e. β_3) parameters of the polynomial function were not statistically different inside *vs* outside the cushions (estimated ± SE = −0.51 ± 0.203, $p = 0.802$, and 0.043 ± 0.045, $p = 0.345$, respectively).

4. Discussion

The general aim of this study was to evaluate if the feedback effects on the reproductive output of nurse cushions differed between native plants and a common exotic invader (*Rumex acetosella*) in tropical alpine communities. We found that native beneficiaries had a direct negative effect on the cushion flower production, but the exotic did not. However, because at high densities the exotics show a negative effect on the abundance of natives within cushions, they could indirectly modulate the native's effect on the cushion fitness. Hence, our results highlight the need to consider both direct and indirect interactions for interpreting the complex reciprocal relations that can develop between nurses and beneficiaries of contrasting adaptive strategies and/or origins (i.e. natives *vs* exotics). Moreover, it offers an original way to explore indirect non-trophic interactions between plants, and their importance for community organization.

4.1. Cushion effects on the abundance of natives and exotics

To evaluate the RFEs of natives and exotics on the nurse, we first needed to analyze their relationship with *Arenaria*. Our results indicated that *Arenaria* cushions had a positive effect on the density of native plant species, but no effect on the exotic *Rumex*. At these same study sites Hupp et al., 2017 found a positive effect of *Arenaria* cushions on local plant species richness, a significant increase in soil organic matter, and a decrease in temperature amplitudes within these cushions. Interestingly, the other abundant cushion species at these sites, *Azorella julianii*, had a positive association with *Rumex*, and a positive effect on soil water, while *Arenaria* did not (Hupp et al., 2017). *Rumex* has a lower water use efficiency than many native páramo species (Llambí et al., 2003), and this may explain its positive association with *Azorella* but not with *Arenaria*. More generally, these results suggest that interactions between the exotic and cushions can be species-specific, which in turn might drive different kinds of reciprocal interactions than those we report here. Other studies in the high páramos of Ecuador reported positive effects of *Arenaria* and *Azorella* cushions on plant abundance and local richness, but only at sites above 4600 m (Sklenár, 2009; Anthelme et al., 2011).

4.2. Feedback effects of natives and the exotic Rumex on cushion reproduction

Previous results from the high tropical Andes of Ecuador and Venezuela indicated a neutral effect of benefactor densities within cushions on the nurse reproductive output (Schöb et al., 2014c).

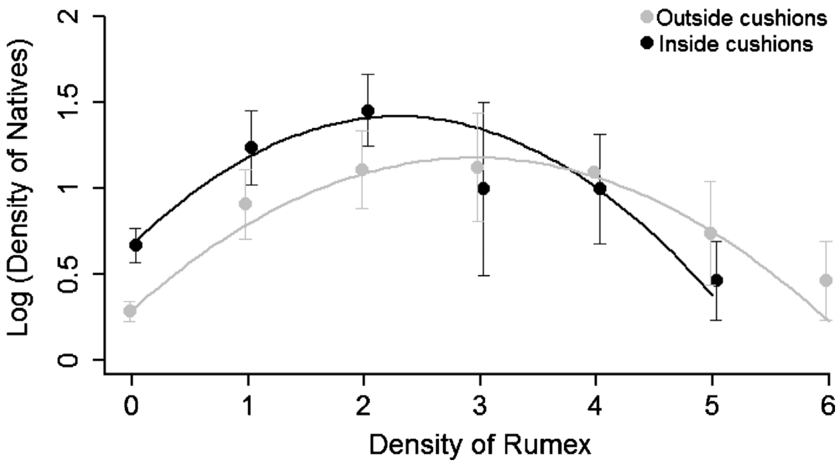


Fig. 3. Relationship between the density of native plants as a function of the density of the invader *Rumex acetosella,* both inside and outside the cushions. Symbols represent mean ± SE, while the solid curves the quadratic equations estimated from the mixed effect model for inside (gray) and outside (black) cushions.

However, when we separated the RFE of natives and exotics, our results indicated a significant negative effect of native abundance on the flower density of *Arenaria*. We observed 53% fewer flowers in cushions where the maximum native density was observed, compared to cushions with no natives. In contrast, *Rumex* density had no significant correlation with flower density. This suggests that the native community has a 'parasitić relationship with nurse cushions in our systems, as has been recently reported across many temperate alpine sites (Schöb et al., 2014b, 2014c). However, we evaluated RFE using flower density, and further research is needed to explore how this relates to fruit and seed set and seed quality. Even so, previous research on feedback effects of beneficiaries on cushion reproduction indicates that a reduction in flower densities is consistently associated with a reduction in seed production (but not seed quality or fruit set), in both experimental and observational contexts (Schöb et al., 2014b; but see García et al., 2016). This would be compatible with the reproductive feedback effects being induced by resource competition between the natives and the cushion (Schöb et al., 2014a). Negative feedback effects mediated by pollinator competition seem less likely, as many of the dominant species in the native community growing within the nurse in our system are wind-pollinated grasses (Hupp et al., 2017), whereas *Arenaria* cushions are pollinated by insects (Wyat, 1986).

The lack of a significant effect of *Rumex* on *Arenaria* flower densities may have been due to an indirect positive effect derived from the substantial decrease in abundance of natives at high densities of *Rumex* (see below). This may also explain why Schöb et al. (2014c) did not find significant feedback effects from beneficiaries on the flower density of other cushion species. To explore this would require experimental manipulation of the densities of natives and exotics. Interestingly, interactions with native species mediated by pollinators are also unlikely in the case of *Rumex*, as it is wind pollinated (Friedman and Barret 2009). This contrasts with the case of the alien *T. officinalis* in Chile, which competes with native species for insect pollinators (Muñoz and Cavieres 2008).

4.3. Effects of Rumex on native plant densities

Rumex was the dominant species outside cushions, and co-dominant with the grass *Agrostis breviculmis* inside cushions. Moreover, *Rumex* seemed to have a density-dependent effect on the abundance of natives. We found a quadratic relationship between the density of *Rumex* and native plants species as a group both inside and outside the cushions. Both natives and *Rumex* increased in abundance at low densities, but at high densities of the exotic, the natives showed a marked decrease in abundance, indicating a competitive effect of *Rumex* (and the possibility of an indirect facilitation effect on the nurse). Interestingly, the same quadratic relationship was found between *Rumex* density and native species richness (results not shown). We did not find a significant reciprocal effect of native abundance on the density of *Rumex*. We suspect that at low densities of *Rumex*, both native species and the exotic increase in abundance in response to microhabitat quality, inside and outside of cushions. Inside cushions, this could be due to observed differences in the cover of dead-tissue patches within different *Arenaria* individuals, which could have influenced the quality of cushions as facilitators, modifying how other plant species occupy them (e.g. Anthelme et al., 2017).

The contrasting behavior of *Rumex* and the natives could be linked both with its exotic character (offering an advantage through mechanisms such as the scape from natural enemies) and several adaptive traits which could make it a strong competitor in high tropical alpine ecosystems. *Rumex* has been found to behave as a ruderal in páramo old-fields at lower elevations and to have a higher SLA, higher leaf N, CO_2 assimilation and growth rates than other native páramo species (Llambí et al., 2003). It is also very effective in expanding both clonally (thorough rhizomes and an extensive root system) and via seeds (Escarre and Houssard, 1989). This could explain why *Rumex* is able to displace the natives when it can attain high densities in favorable microsites, such as in open subnival environments dominated by low-stature, slow growing, stress-tolerant plants, and where soil disturbance by solifluction is common (i.e. outside cushions, where it is the dominant species).

Though our research provides evidence that high exotic densities can have a negative effect on native species, this does not always seem to be the case. For example, in the Australian Alps *Rumex* has been common in the alpine zone for at least 60 years, but there is no evidence of negative effects on native species richness (Alexander et al., 2016). Furthermore, in a Mediterranean ecosystem in the central Andes of Chile, Cavieres et al. (2005) found a positive correlation between the exotic *Taraxacum officinale* and the density and species richness of natives within cushions. In open areas, there were no differences in overall native density or richness with or without the exotic, but several species showed negative associations with *T. officinale*.

5. Conclusions

Our results indicate the potential existence of complex reciprocal effects between nurses, natives and exotic plants in the high tropical Andes, and that natives and exotics differ in their feedback effects on the nurses (Fig. 4). Native plant species were facilitated by *Arenaria* cushions, and the native community growing within *Arenaria* had a negative feedback effect on this nurse planťs flower production, suggesting a parasitic relationship. In turn, we found no significant relationships between the exotic *Rumex* and *Arenaria*. But at high densities, the invader was associated with much lower densities of native species both inside and outside cushions. This may have an indirect positive effect on the cushiońs reproduction through the reduction of

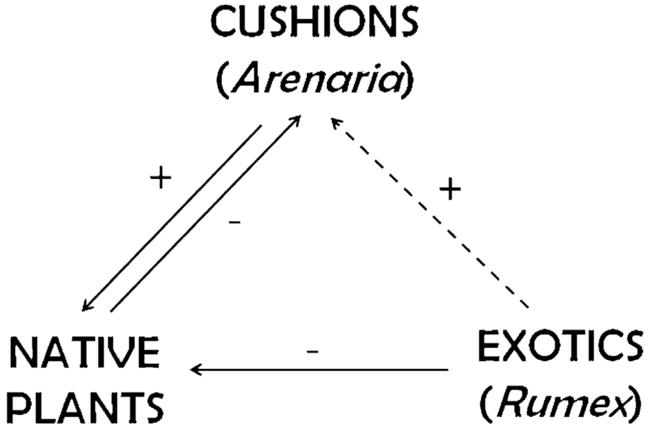


Fig. 4. Schematic diagram representing the potential nature of the interactions between our focal cushion plant (*Arenaria musciformis*), the other native species in the community and the exotic *Rumex acetosella*. Solid arrows indicate direct positive (+) or negative effects (−), while the dotted arrow indicates the possibility of an indirect positive effect of *Rumex* on the cushiońs reproduction mediated by the exotićs negative effect on native beneficiaries.

the density of natives within them. These conclusions are based on spatial associations and correlative patterns, and need to be further explored through manipulative experiments. Even so, our results suggest the possibility of complex direct and indirect reciprocal interactions between facilitators and beneficiaries and emphasize the importance of documenting the impacts of exotic invaders on alpine plant communities.

Acknowledgments

We thank the Fulbright Scholars Program (grant No. 68150764) for support for LDLL, NSF EPSCoR Track-1 EPS-1101342 (INSTEP 3) for support for NH, and Fulbright-BEC.AR Program for support for AS. Special thanks to F. Anthelme and S. Lavergne and for their invitation to participate in this special issue on High Elevation Ecosystems and to the editor and two anonymous referees for their excellent comments and suggestions which greatly improved the manuscript. We also are grateful for the assistance with field data collection of N.J. Márquez and Y. Cáceres, insights on data analysis provided by C. Schöb and I. Suárez and discussions on the pollination strategies of páramo plants with R. Pelayo.

References

Alexander, J.M., Kueffer, C., Daehler, C.C., Edwards, P.J., Pauchard, A., Seipel, T., MIREN Consortium, 2011. Assembly of nonnative floras along elevational gradients explained by directional ecological filtering. Proc. Natl. Acad. Sci. U. S. A. 108, 656–661.

Alexander, J.M., Lembretchts, J.J., Cavieres, L.A., Daehler, C., Haider, S., Kueffer, C., Liu, G., McDougall, K., Milbau, A., Pauchard, A., Rew, L.J., Seipel, T., 2016. Plant invasions into mountains and alpine ecosystems: current status and future challenges. Alp. Bot. 126, 89–103. http://dx.doi.org/10.1007/s00035-016-0172-8.

Anderson, M., Gorley, R., Clarke, K., 2008. PERMANOVA+ for PRIMER: Guide to Software and Statistical Methods. PRIMER-E, Plymouth.

Anthelme, F., Dangles, O., 2012. Plant–plant interactions in tropical alpine environments. Persp. Plant Ecol. Evol. Syst. 14, 363–372.

Anthelme, F., Buendina, B., Mazoyer, C., Dangles, O., 2011. Unexpected mechanisms sustain the stress gradient hypothesis in a tropical alpine environment. J. Veg. Sci. 23, 62–72.

Anthelme, F., Meneses, R.I., Valero, N.H., Pozo, P., Dangles, O., 2017. Fine nurse variations explain discrepancies in the stress-interaction relationship in alpine regions. Oikos. http://dx.doi.org/10.1111/oik.04248.

Armas, C., Ordiales, R., Pugnaire, F.I., 2004. Measuring plant interactions: a new comparative index. Ecology 85, 2682–2686.

Arroyo, M.T.K., Cavieres, L.A., Peñaloza, A., Arroyo-Kalin, M.A., 2003. Positive associations between the cushion plant *Azorella monantha* (Apiaceae) and alpine plant species in the Chilean Patagonian Andes. Plant Ecol. 169, 121–129.

Aschehoug, E.T., Brooker, R., Atwater, D.Z., Maron, J.L., Callaway, R.M., 2016. The mechanisms and ecological consequences of interspecific competition among plants. Annu. Rev. Ecol. Evol. Syst. 47, 263–281.

Badano, E.I., Villarroel, E., Bustamante, R.O., Marquet, P.A., Cavieres, L.A., 2007. Ecosystem engineering facilitates invasions by exotic plants in high-Andean ecosystems. J. Ecol. 95, 682–688.

Besaw, L.M., Thelen, G.C., Sutherland, S., Metlen, K., Callaway, R.M., 2011. Disturbance, resource pulses and invasion: short-term shifts in competitive effects, not growth responses, favor exotic annuals. J. Appl. Ecol. 48, 998–1006.

Bolker, B.M. et al., 2012. Generalized linear mixed models using ad model builder. R package version 0.7.7.

Briceño, B., Morillo, G., 2002. Catálogo abreviado de las plantas con flores de los páramos de Venezuela: Parte I. Dicotiledóneas (Magnoliopsida). Acta Botánica Venezuélica 25, 1–46.

Bronstein, J.L., 2009. The evolution of facilitation and mutualism. J. Ecol. 97, 1160–1170.

Brooker, R.W., Callaway, R.M., 2009. Facilitation in the conceptual melting pot. J. Ecol. 97, 1117–1120.

Brooker, R.W., Maestre, F.T., Callaway, R.M., Lortie, C.L., Cavieres, L., Kunstler, G., Liancourt, P., Tielbörger, K., Travis, J.M.J., Anthelme, F., Armas, C., Coll, L., Corcket, E., Delzon, S., Forey, E., Kikvidze, Z., Olofsson, J., Pugnaire, F., Quiroz C. l. Saccone, P., Schiffers, K., Seifan, M., Touzard, B., Michalet, R., 2008. Facilitation in plant communities: the past, the present, and the future. J. Ecol. 96, 18–34.

Cáceres, Y., Llambí, L.D., Rada, F., 2015. Shrubs as foundation species in a high tropical alpine ecosystem: a multi-scale analysis of plant spatial interactions. Plant Ecol. Divers. 8, 147–161.

Callaway, R.M., Pennings, S.C., 2000. Facilitation may buffer competitive effects: indirect and diffuse interactions among salt marsh plants. Am. Nat. 156, 416–424.

Callaway, R.M., 2007. Positive Interactions and Interdependence in Plant Communities. Springer, Berlin.

Cavieres, L.A., Badano, E.I., 2009. Do facilitative interactions increase species richness at the entire community level? J. Ecol. 97, 1181–1191.

Cavieres, L.A., Quiroz, C., Molina-Montenegro, M., Pauchard, A., 2005. Nurse effect of the native cushion plant *Azorella monantha* on the invasive non-native *Taraxacum officinale* in the high-Andes of central Chile. Persp. Plant Ecol. Evol. Syst. 7, 217–226.

Cavieres, L.A., Quiroz, C.L., Molina-Montenegro, M.A., 2008. Facilitation of the non-native *Taraxacum officinale* by native nurse cushion species in the high Andes of central Chile: are there differences between nurses? Funct. Ecol. 22, 148–156.

Cavieres, L.A., Brooker, R.B., Butterfield, B.J., et al., 2014. Facilitative plant interactions and climate simultaneously drive alpine plant diversity. Ecol. Lett. 17, 193–202.

Cranston, B.H., Callaway, R.M., Monks, A., Dickinson, K., 2012. Gender and abiotic stress affect community-scale intensity of facilitation and its costs. J. Ecol. 100, 915–922.

Escarre, J., Houssard, C., 1989. Variations de populations de *Rumex acetosella*. L. le long d'une succession secondaire: I. Allocation de biomasse. Acta Oecol. 10, 3–19.

Filazzola, A., Lortie, C.J., 2014. A systematic review and conceptual framework for the mechanistic pathways of nurse plants. Global Ecol. Biogeogr. 23, 1335–1345.

Flores-Martínez, A., Ezcurra, E., Sánchez-Colón, S., 1994. Effect of *Neobuxbaumia tetetzo* on growth and fecundity of its nurse plant *Mimosa luisana*. J. Ecol. 82, 325–330.

Friedman, J., Barret, S.H.C., 2009. Wind of change: new insights into the ecology and evolution of pollination and mating in wind-pollinated plants. Ann. Bot. 103, 1515–1527.

García, M.C., Bader, M.Y., Cavieres, L., 2016. Facilitation consequences for reproduction of the benefactor cushion plant *Lauretia acaulis* along an elevation gradient: costs or benefits? Oikos 125, 434–442.

Greig-Smith, P., 1983. Quantitative Plant Ecology. University of California Press, Berkeley.

Holzapfel, C., Mahall, B.E., 1999. Bidirectional facilitation and interference between shrubs and annuals in the Mojave dessert. Ecology 80, 1747–1761.

Hupp, N., Llambí, L.D., Ramírez, L., Callaway, R.M., 2017. Alpine cushion plants have species-specific effects on microhabitat and community structure in the tropical Andes. J. Veg. Sci. http://dx.doi.org/10.1111/jvs.12553.

Jacobsen, D., 2008. Tropical high-altitude streams. In: Dudgeon, D. (Ed.), Tropical Stream Ecology. Elsevier Science, pp. 219–256.

Levine, J.M., 1999. Indirect facilitation: evidence and predictions from a riparian community. Ecology 80, 1762–1769.

Llambí, L.D., Fontaine, M., Rada, F., Saugier, B., Sarmiento, L., 2003. Ecophysiology of dominant plant species during old-field succession in a high tropical Andean ecosystem. Arct. Antarct. Alp. Res. 35, 447–453.

Lortie, C.J., Brooker, R.W., Choler, P., Kikvidze, Z., Michalet, R., Pugnaire, F.I., Callaway, R.M., 2004. Rethinking plant community theory. Oikos 107, 433–438.

Michalet, R., Xiao, S., Touzard, B., Smith, D.S., Cavieres, L.A., Callaway, R.M., Whitham, T.G., 2011. Phenotypic variation in nurse traits and community feedbacks define an alpine community. Ecol. Lett. 14, 433–443.

Moragues, E., Traveset, A., 2005. Effect of *Carpobrotus* spp. on the pollination success of native plant species of the Balearic Islands. Biol. Conserv. 122, 611–619.

Morales, C.L., Traveset, A., 2009. A meta-analysis of impacts of alien vs. native plants on pollinator visitation and reproductive success of co-flowering native plants. Ecol. Lett. 12, 716–728.

Muñoz, A.A., Cavieres, L.A., 2008. The presence of a showy invasive plant disrupts pollinator service and reproductive output in native alpine species only at high densities. J. Ecol. 96, 459–467.

Pérez, F.L., 1987. Needle-ice activity and the distribution of stem-rosette species in a Venezuelan páramo. Arct. Antarct. Alp. Res. 19, 135–153.

Pauchard, A., Kueffer, C., Dietz, H., Daehler, C.C., Alexander, J., Edwards, P.J., Arévalo, J.R., Cavieres, L.A., Guisan, A., Haider, S., Jakobs, G., McDougall, K., Millar, C.I., Naylor, B.J., Parks, C.G., Rew, L., Seipel, T., 2009. Aińt no mountain high enough: plant invasions reaching new elevations. Front. Ecol. Environ. 7, 479–486.

Pinheiro, J., Bates, D., Deb Roy, S., Sarkar, D., R Core Team, 2015. nlme: Linear and Nonlinear Mixed Effects Models. R package version 3. 1–121, http://CRAN.Rproject.

org/package=nlme.

Pugnaire, F.I., Haase, P., Puigdefabregas, J., 1996. Facilitation between higher plant species in a semiarid environment. Ecology 77, 1420–1426.

Ramírez, L., Rada, F., Llambí, L.D., 2015. Linking patterns and processes through ecosystem engineering: effects of shrubs on microhabitat and water status of associated plants in the high tropical Andes. Plant Ecol. 216, 213–225.

Salgado-Labouriau, M.L., Schubert, C., 1977. Pollen analysis of a peat bog from Laguna Victoria (Venezuelan Andes). Acta Científica Venezolana 28, 328–332.

Sarmiento, L., Llambí, L.D., Escalona, A., Márquez, N., 2003. Vegetation patterns, regeneration rates and divergence in an old-field succession of the high tropical Andes. Plant Ecol. 166, 63–74.

Schöb, C., Prieto, I., Armas, C., Pugnaire, F.I., 2014a. The consequences of facilitation: one planís benefit is another planís cost. Funct. Ecol. 28, 500–508.

Schöb, C., Michalet, R., Cavieres, L.A., Pugnaire, F.I., Brooker, R.W., Butterfield, B.J., Cook, B.J., Kikvidze, Z., Lortie, C.J., Xiao, S., Al Hayek, P., Anthelme, F., Cranston, B.H., García, M.A., Le Bagousse-Pinguet, Y., Reid, A.M., le Roux, P.C., Lingua, E., Nyakatya, M., Touzard, B., Zhao, L., Callaway, R.M., 2014b. A global analysis of bidirectional interactions in alpine plant communities shows facilitators experiencing strong reciprocal fitness costs. New Phytol. 202, 95–105.

Schöb, C., Callaway, R.M., Anthelme, F., Brooker, R.W., Cavieres, L.A., Kikvidze, Z., Lortie, C.J., Michalet, R., Pugnaire, F.I., Xiao, S., Cranston, B.H., García, M.C., Hupp, N., Llambí, L.D., Lingua, E., Reid, A.M., Zhao, L., Butterfield, B.J., 2014c. The context dependence of beneficiary feedback effects on benefactors in plant facilitation. New Phytol. 204, 386–396.

Seipel, T., et al., 2012. Processes at multiple scales affect richness and similarity of non-native plant species in mountains around the world. Global Ecol. Biogeogr. 21, 236–246.

Shah, M.A., Callaway, R.M., Shah, T., Houseman, G.R., Pal, R.W., Xiao, S., Luo, W., Rosche, C., Reshi, Z.A., Khasa, D.P., Chen, S., 2014. *Conyza canadensis* suppresses plant diversity in its nonnative ranges but not at home: a transcontinental comparison. New Phytol. 202, 1286–1296.

Simpson, B.B., Toddzia, C.A., 1990. Patterns and processes in the development of the high Andean flora. Am. J. Bot. 77, 1419–1432.

Sklenář, P., 2009. Presence of cushion plants increases community diversity in the high equatorial Andes. Flora 204, 270–277.

Theurillat, J.P., Guisan, A., 2001. Potential impact of climate change on vegetation in the European Alps: a review. Clim. Change 50, 77–109.

Tilman, D., 1982. Resource Competition and Community Structure. Princeton University Press, Princeton.

Valiente-Banuet, A., Vite, F., Zavala-Hurtado, J.A., 1991. Interaction between the cactus Neobuxbaumia tetetzo and the nurse shrub Mimosa luisana. J. Veg. Sci. 2, 11–14.

Vilà, M., Espinar, J.L., Hejda, M., Hulme, P.E., Jarošík, V., Maron, J.L., Pergl, J., Schaffner, U., Sun, Y., Pyšek, P., 2011. Ecological impacts of invasive alien plants: a meta-analysis of their effects on species, communities and ecosystems. Ecol. Lett. 14, 702–708.

Wyat, R., 1986. Ecology and evolution of self-pollination in *Arenaria uniflora* (Caryophyllaceae). J. Ecol. 74, 403–418.

Yang, Y., Niu, Y., Cavieres, L.A., Sun, H., 2010. Positive associations between the cushion plant *Arenaria polytrichoides* (Caryophyllaceae) and other alpine plant species increase with altitude in the Sino-Himalayas. J. Veg. Sci. 21, 1048–1057.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 89–102

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Research article

Time lag between glacial retreat and upward migration alters tropical alpine communities☆

Anaïs Zimmer[a,b,c], Rosa I. Meneses[b], Antoine Rabatel[d], Alvaro Soruco[e], Olivier Dangles[f,g,h], Fabien Anthelme[a,b,c,*]

[a] AMAP, IRD, CNRS, INRA, Université de Montpellier, Montpellier, France
[b] Museo Nacional de Historia Natural, Herbario Nacional de Bolivia (LPB), Cota Cota, Casilla 10077 Correo Central, La Paz, Bolivia
[c] Instituto de Ecología, Universidad Mayor San Andrés, Calle 27, Cota Cota, Campus Universitario, La Paz, Bolivia
[d] Université Grenoble Alpes, CNRS, IRD, Institut des Géosciences de l'Environnement (IGE, UMR 5001), F-38000 Grenoble, France
[e] Instituto de Geología y del Medio Ambiente, Universidad Mayor San Andrés, Calle 27, Cota Cota, Campus Universitario, La Paz, Bolivia
[f] Institut de Recherche pour le Développement (IRD), EGCE, 91198 Gif-sur-Yvette Cedex, France
[g] Université Paris-Sud 11, 91405 Orsay Cedex, France
[h] Pontificia Universidad Católica del Ecuador, Facultad de Ciencias Exactas y Naturales, Quito, Ecuador

ARTICLE INFO

Keywords:
Biological soil crust
Climatic debt
Chronosequence
Nurse plant
Species range shifts
Tropical andes

ABSTRACT

Species range shifts and possible species extinctions in alpine regions are hypothesized being influenced by the increasing time lag between the velocity of global warming and the slowness of primary succession. We tested this hypothesis in tropical alpine environments above 4700 m a.s.l. (Central Andes) and we explored the underlying mechanisms at work by using four sites gradually deglaciated since the acceleration of warming in the late 1970's. These post-glacial chronosequences, made available by a multidisciplinary approach combining glaciology and ecology, are extremely rare and provide a pertinent space-for-time substitution for the study of climate change effects. We found consistent patterns in plant succession (abundance, species richness and functional strategies) along the four chronosequences. Dispersal limitation was a prominent constraint for succession, even at the end of the chronosequences, leading to an overrepresentation of anemochorous species in comparison with adjacent ecosystems. Nurse plants were infrequent and their low maturity seemed to make them poorly efficient as facilitators, contrarily to the expectations made by the stress-gradient hypothesis in alpine regions. This suggests that, despite the accelerating rate of warming, the dynamics of primary succession remains slow, generating a climatic debt and hampering the adaptation to climate change in alpine plant communities.

1. Introduction

Under the pervasive effects of climate warming on ecosystems, characterizing the biodiversity-climate change relationship has become a major scientific challenge (Lavergne et al., 2010; Chen et al., 2011; Urban, 2015). Especially, a detailed description of patterns and mechanisms of vegetation succession is required to identify the ecological processes that will shape future biodiversity (Walker and Wardle, 2014). In mountain systems, despite the local availability of biotic and abiotic refuges in heterogeneous areas (Scherrer and Körner, 2011; Anthelme et al., 2014a), the majority of organisms need to perform upward range shifts to adapt to the direct and indirect effects of warming (Gottfried et al., 2012; Lenoir and Svenning, 2015; Harsch and HilleRisLambers, 2016). Located at the upper limit of life, alpine species are particularly sensitive to these range shifts because new, upward habitats are often devoid of life and soil, or they are simply absent. The migration lag experienced by alpine plants, *i.e.* the time spent between a climatic fluctuation and the moment when plants effectively reach the new site, has been shown to cause a significant reduction in their spatial distribution in a study centred on the Holocene scale (Dullinger et al., 2012). Given the high magnitude and velocity of current and predicted climate change, it is expected that this migration lag might affect negatively the altitudinal distribution of alpine species (Svenning and Sandel, 2013). A key issue is to know whether plant migration lag can

---

Abbreviations: CSR, competitor, stress-tolerant, ruderal; BSC, biological soil crust; LIA, little ice age

☆ This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

* Corresponding author at: UMR AMAP/DIADE, Boulevard de la Lironde, TA A-51/PS2, F-34398 Montpellier Cedex 5, France.
E-mail addresses: zimmer.anais@gmail.com (A. Zimmer), rosaiselameneses11@gmail.com (R.I. Meneses), antoine.rabatel@univ-grenoble-alpes.fr (A. Rabatel), alvaro.soruco@gmail.com (A. Soruco), oliver.dangles@ird.fr (O. Dangles), fabien.anthelme@ird.fr (F. Anthelme).

http://dx.doi.org/10.1016/j.ppees.2017.05.003
Received 11 October 2016; Received in revised form 24 May 2017; Accepted 30 May 2017

result in a climatic debt for natural communities that would be no longer in equilibrium with climate (Devictor et al., 2012; Svenning and Sandel, 2013).

Elevation has been shown positively correlated with the intensity of warming in mountainous areas, with tropical alpine ecosystems being among the most heavily affected by global warming because of their high elevation, generally above 4000 m a.s.l. (Bradley et al., 2006; MRI, 2015). This is observable with the tropical Andean glaciers, which have experienced a more pronounced shrinking than other glaciers worldwide since the late 1970's (Rabatel et al., 2013). The impacts of these changes on biodiversity seem to be rapid and severe, as demonstrated with aquatic and terrestrial invertebrates (Jacobsen et al., 2012; Cauvy-Fraunié et al., 2016; Moret et al., 2016). They are in line with the recent evidence that the biodiversity of South America −the continent that shelters more than 90% of tropical alpine regions worldwide- is the most affected by changing climate with 23% of its species at risk *vs.* 7.9% worldwide, under an optimistic warming scenario (Urban, 2015). Tropical alpine environments are also impacted by climate warming at ecosystem level, through glacier shrinking, which has been shown to modify water input and increase ecosystem fragmentation (Dangles et al., 2017) and by increasing plant growth and organic carbon production (Cooper et al., 2015). All in all, this makes tropical Andean ecosystems flagship descriptors for the characterization of the biodiversity-climate change interactions in a changing world. However, although secondary succession in the alpine tropics has received considerable attention (e.g. Sarmiento et al., 2003; Bueno and Llambí, 2015), primary succession has been overlooked, so far, especially its characterization in a changing world (but see Suárez et al., 2015).

Our first specific hypothesis is that the unrivalled velocity of warming in tropical alpine regions may exacerbate the dispersal filter with new species assemblages being even more dominated by anemochorous species than what has been observed along longer post-glacial chronosequences, so far (Stöcklin and Bäumler, 1996; Erschbamer and Caccianiga, 2016; Marta et al., 2016). Indeed, wind is expected to be the most efficient dispersal vector for seeds to reach rapidly upwards alpine sites, whereas water streams would preferentially disperse seeds downwards. Additionally, we expect that recently deglaciated sites will not be sufficiently visited by dispersers such as mammals, birds and insects to allow animal-dispersed plants to be correctly represented.

Our second specific hypothesis is that the direction and intensity of plant–plant interactions, a crucial mechanism of community organization in alpine environments, will be influenced by rapid warming, thus having a significant impact on the future alpine plant communities. Indeed, once seeds have reached a new site, a suitable environment is required to proceed with establishment. At this stage, the presence of refuges for plants, either biotic or abiotic, may be critical (Klanderud and Totland, 2007; Anthelme et al., 2014a) especially because soil is generally absent, or at best composed of mineral ashes and rests of bacterial activity in scattered microsites/safe sites (Caccianiga et al., 2006; Sattin et al., 2009). Environment amelioration by nurse plants has been evidenced in a large panel of stressful environments (Filazzola and Lortie, 2014), including tropical alpine environments (Sklenář, 2009; Anthelme et al., 2012; Cáceres et al., 2015), and has been included into the conceptual framework on species interactions (Callaway et al., 2002; Maestre et al., 2009; *niche concept*: Bulleri et al., 2016). However, there are no available studies questioning a possible limitation in nurse effects induced by a lack of time consecutive to accelerated warming (HilleRisLambers et al., 2013; Anthelme et al., 2014a). We address two specific questions related to plant–plant interactions in rapidly changing environments: (1) can the altitudinal migration lag mentioned above reduce the presence/abundance of nurse plants, thus indirectly reducing plant diversity through cascade effects (*sensu* Malatesta et al., 2016)? And (2) would the short time available for the development of alpine nurse plants in recently deglaciated sites impact negatively plant communities because of limited ontogenic variations between the nurse and the beneficiary (Anthelme and Dangles, 2012)?

More generally, how the various pioneer organisms interact early after glacial retreat is poorly known and requires further investigation (Matthews and Vater, 2015; Erschbamer and Caccianiga, 2016). Among these organisms, biological soil crust (BSC), an association of bacteria, lichens, algae, mosses and fungi, has been proposed to be another driver of primary succession after glacial retreat, through facilitative interactions with other plants (Türk and Gärtner, 2001; Breen and Lévesque, 2008; Matthews and Vater, 2015). Can they ensure successful primary succession under a rapidly changing climate irrespective of nurse plant effects?

By characterizing the abiotic environment, the species diversity, plant–plant and plant-rock spatial associations along primary succession in four sites in the tropical Andes gradually deglaciated since the late 1970's (post-glacial chronosequences) we aimed at testing these specific hypotheses and their related research questions. Our results were discussed into the recent conceptual framework on species range shifts consecutive to global changes (Lenoir and Svenning, 2015), exploring future scenarios for the tropical alpine biodiversity.

2. Material and methods

2.1. Study area and study sites

At elevations above 500 m a.s.l., the tropical Andes extend over 1.5 millions km^2. They are one of the most important biodiversity hotspots worldwide; by, with a high proportion of endemic plants (Anthelme et al., 2014b). Tropical alpine regions in the Andes range between 3200 m a.s.l. and more than 5000 m a.s.l., even if tropical alpine vegetation *stricto sensu*, i.e. that develops above the natural treeline, is primarily observed above 4200 m a.s.l. (Smith, 1994; Meneses et al., 2015). The Andes gather 99% of the tropical glaciers worldwide, 71% of which in Peru and 21% in Bolivia (Rabatel et al., 2013).

To test our hypothesis under different environmental conditions, we studied four sites distributed in two countries. In Bolivia, three sites were located in three different valleys of the Cordillera Real (Fig. 1). This mountain range includes 11% of tropical glaciers and 55% of Bolivian glaciers (Soruco et al., 2009). 338 vascular plants have been described above 4200 m a.s.l. (Meneses et al., 2015) and glaciers lost 48% of their surface area between 1975 and 2006 (Soruco et al., 2009). In Peru, we selected one site in the Cordillera Blanca, Ancash region, which comprises more than 755 glaciers covering approximately 527 km^2 in 2003. Glaciers in Peru showed a reduction of 43% their surface over the past 40 years (UGRH, 2014).

The southernmost Bolivian site was located on the foreland of glacier Zongo (4900 m a.s.l.; 16°16'28.57"S; 68°8'45.50"W). The second site was located in the foreland of glacier "10" (4812 m a.s.l.; 16°7'36.13"S; 68°15'4.42"W; see labelling inSoruco et al., 2009). The third Bolivian site was located on the foreland of glacier Tarija (4690 m a.s.l.; 16°10'42.78"S; 68°13'18.67"W). Annual precipitations reach between 600 mm/yr in sites Tarija and 10 and 860 mm/yr in site Zongo. Precipitations are essentially distributed between November and March (Rabatel et al., 2012). The Peruvian site was located on the foreland of glacier Yanamarey (4720 m a.s.l.; 9°39'9.45"S; 77°16'16.30"W) where precipitations reach 1000 mm/yr. These precipitations are mainly distributed between October and April, denoting a longer rainy season than in the Bolivian sites (Tarazona Santos, 2005; Rabatel et al., 2012).

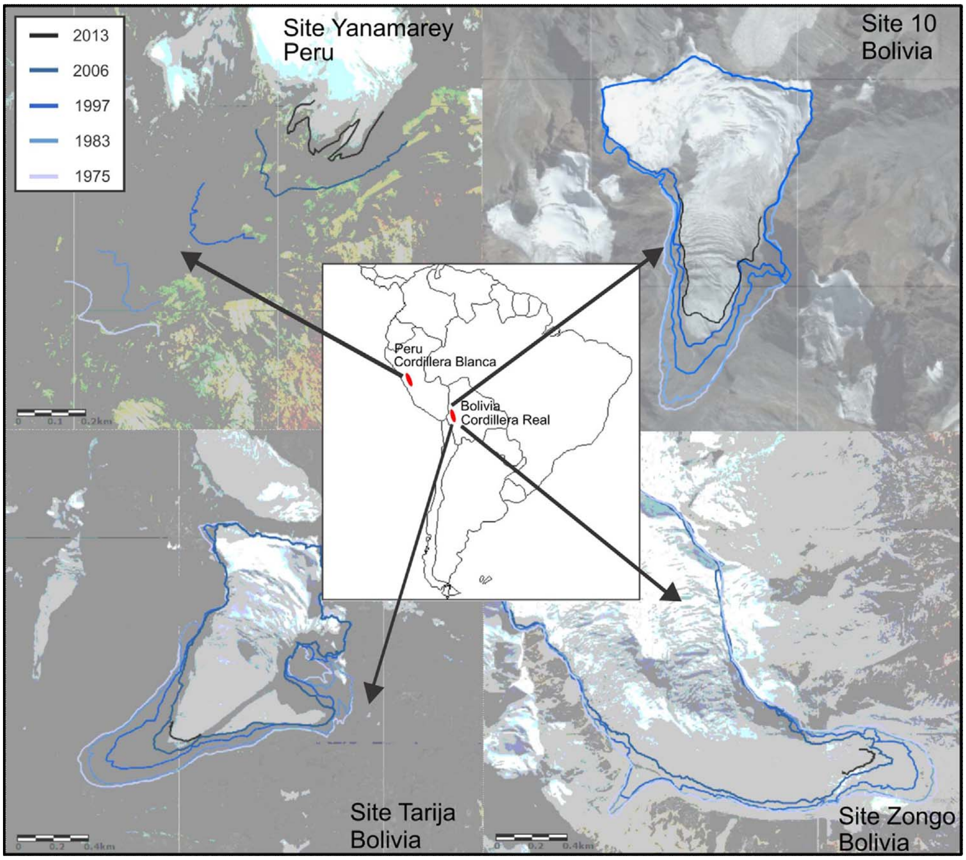


Fig. 1. Four sites located in tshe central Andes (three in the Cordillera Real in Bolivia and one in the Cordillera Blanca in Peru), each of which providing a 38-years post-glacial chronosequence. Lines represent the date of glacial retreat at five periods: 2013, 2006, 1997, 1983 and 1975.

Each of the four sites comprises a glacier in the upper part of the catchment. These glaciers are of valley-glacier type within the altitudinal range 4800–6000 m a.s.l., and a surface area between 0.6 and 1.9 km^2 in 2006. For the tropical Andes, these are middle-size glaciers and their forelands show a sufficiently wide deglaciated area to make repetitions of vegetation plots while being more rapidly responding to climate change than larger glaciers (Rabatel et al., 2013). Glaciers were also selected upon the following criteria: good accessibility, available data on glacial retreat, and absence of large stone slabs and steep slopes in the deglaciated areas that are expected to influence colonization processes.

2.2. Chronosequences under rapid warming

Chronosequences provide reliable space-for-time substitution designs for predicting climate change effects on biodiversity (Blois et al., 2013; Walker and Wardle, 2014), especially post-glacial chronosequences in alpine-arctic environments (Matthews, 1992; Cannone et al., 2008). Indeed, glacial retreat is recognized as a powerful and integrative indicator of recent climate change (IPCC, 2013), especially within the Tropics where ablation occurs all year round resulting in a short-time response of glacier extension and volume to changing climate (Rabatel et al., 2013). By releasing newly colonisable areas free of soil and plant propagules, glacial retreat provides consistent sites for studying primary succession (Walker and Del Moral, 2003; Matthews and Vater, 2015).

The high Andes experienced an average warming of 0.04 °C per decade since the Little Ice Age in Bolivia (LIA; 1.2 °C between years 1675 and 2000). This value increased to 0.1 °C per decade since the 1970’s (Rabatel et al., 2013). Accordingly, considering the warming since LIA maximum as a control, the rate of warming has been multiplied by 2.5 during the last 38 years. It means that temperature changes during the 38 years of our post-glacial chronosequences would have taken (38 × 2.5) 95 years before the 1970’s and the acceleration of human-induced CO2 emissions, even without considering the probable higher warming at high elevation (MRI, 2015). Meanwhile, scenarios of changes in precipitations are much less developed and do not permit to define structured trends during the future decades in the Andes (IPCC, 2013). For these reasons, they were not considered into our framework. However, it has to be kept in mind that the combination of warming and precipitation variations, in dry mountainous environments, has been shown recently to have unexpected impacts on plants (Harsch and HilleRisLambers, 2016; Grossiord et al., 2017) and some studies in the tropical Andes propose structured scenarios of future precipitation changes at regional or local scale (e.g. Tovar et al., 2013).

For each of the four selected sites we considered four zones defined by the time since deglaciation: 0–7 years, 7–16 years, 16–30 years and 30–38 years. At the time of the plant observation (2013), this matched chronosequences starting in 1975, *i.e.* the period when atmospheric CO_2 started to increase rapidly worldwide (IPCC, 2013) and when the acceleration of glacial retreat started in the tropical Andes (Rabatel et al., 2013). Each line delimiting zones was extracted from existing data in 2013, 2006, 1997, 1983 and 1975 (1976 in glacier Yanamarey) through satellite imagery, aerial photographs and direct *in-situ* observations (Rabatel et al., 2008; Soruco et al., 2009; L. Dávila, unpublished data; Soruco and Rabatel unpublished data).

2.3. Sampling, data collection and data analysis

At each site, between 15 and 38 square plots of 1 m^2 were selected randomly in each deglaciated zone (total: four sites, 325 plots) once the following two key environmental conditions were met: 1) absence of dominant rock or slab stone within –or adjacent to- each plot; and 2) absence of water-dominated patch (e.g. lake) in the vicinity of plots (< 5 m). Each plot was divided into 100 subplots of 0.1 m^2 each in order to yield a more precise data collection.

Within each plot, biotic and abiotic data were collected to identify succession patterns and mechanisms. We listed all the vascular species observed in our sampling (Table A.1) and each individual of each vascular species was assigned to one subplot. We estimated visually the relative cover of each vascular plant and the relative cover of the biological soil crust (BSC). Each individual of each vascular species was assigned to one subplot. We calculated the area of each individual at soil surface by multiplying its maximum length by the perpendicular width. Then we calculated the volume of each individual (1) by multiplying its area with its vegetative height and (2) by multiplying this product with the visually-estimated vegetation cover of each individual at soil surface (%). This individual-based approach permitted to yield data at individual level, species level, and at community level. Other abiotic data measured in each plot were: slope gradient (clinometer Suunto Tandem, Vantaa, Finland), granulometry (making difference between sand, gravel (< 2 cm), rock (between 2 and 20 cm) and block (> 20 cm), and including gravel, rock and block as coarse elements), soil temperature in a subsample of 10 plots at each zone of site Zongo (using 40 Tidbit V2 Temp data loggers each 30 min during 27 days), and soil moisture, extracted from the difference between moist and dry samples of soil (%, 500 g of soil, 11–13 samples at each zone). Methods are detailed in Zimmer et al. (2014). Wind speed was not measured. From our experience it was higher in site Zongo, located at the level of a pass between the Bolivian Altiplano and Amazonia.

To evaluate and compare the role played by biotic refuges (plants, biological soil crust) and abiotic refuges (rocks) on the presence of other vascular plants we reported each spatial association (1) between vascular plants (as soon as the aerial parts of two individuals touched/overlapped), (2) between plants and BSC (as soon as an individual was located on BSC), and (3) between plants and rocks (as soon as rock height being superior to plant height and the distance between plant and rock not exceeding plant height). We assumed that root interactions did not exceed aerial interactions among plants spatially because of the short time available for plants to grow, although plants are expected to produce extended belowground organs in cold environments (Körner, 2003). Spatial associations do not necessarily reflect positive interactions. For example, two competing species can be spatially associated because competing for the same local resource. In our sample, we increased the probability that associations represent positive interactions (1) by selecting smooth areas where resources are expected to be equally distributed, and (2) because positive interactions involving a nurse and a beneficiary were identified as soon as one of the two interacting individuals had a volume at least two times larger than the other individual, which reflects complementarity in resource acquisition and/or ontogenic shifts (Anthelme and Dangles, 2012). These specific spatial associations were extracted as a subsample. The fact that they represent positive interactions was highly probable but not certain.

We estimated the constraint played by dispersal limitation by assigning a dispersal mode to each species observed in our samples. Taxonomic identifications and dispersal strategies were both determined using Meneses et al. (2015) and Kolff and Kolff (2005).

Given the lack of general patterns on plant functional traits of alpine plants in relation with climate change, the C (competitor) S (stress-tolerant) R (ruderal) strategies by Grime (1977, 2002) have been proposed as a relevant way to infer the functional response of plants to climate change in alpine zones (Grytnes et al., 2014; Parmesan and Hanley, 2015). Using criteria proposed by Grime (2002, taking into account arctic and alpine regions in this edition), Hodgson et al. (1999) and Caccianiga et al. (2006 in an alpine region), we built a local CSR classification in our sites based on the assignment of one of the following strategies for each species: C (value: 1), S (1), R (1), SR (0.5/0.5), CS (0.5/0.5), CR (0.5/0.5) and CSR (0.33/0.33/0.33). Criteria for the determination of strategies were extracted from field observations, bibliography (Meneses et al., 2015) and specimen in the Herbarium LPB (Table A.2). Accordingly, they related mostly to morphological and life history traits and their relative weight in the determination of strategies was function of their deviation from the mean value observed in the community. For example, erect shrubs were predominantly C-strategists because of their dominant structure, even if producing micro-leaves, whereas grasses were predominantly R-strategists because of their fast growth rate and despite their high longevity during establishment phase (a trait shared by the overwhelming majority of species, locally). At each zone of each site, we calculated C, S and R community weighted means, *i.e.* plot-level strategy values weighted by species abundance (see Violle et al., 2007; Sonnier et al., 2010 for calculation details). Note that this classification is still exploratory with the objective to serve as a solid base for future CSR classifications in tropical alpine regions.

To test statistically the variations of biotic and abiotic variables observed along with sites and chronosequences, we used mixed effect models. For this purpose we used a generalized linear model with the variable "chronosequence" (fixed, four zones) being nested within the variable "site" (random, four sites, see Fig. 1). The quantitative variables being explained by this model were species richness, plant cover, number of individuals (abundance) and dispersal modes. We computed linear regressions to quantify the effects of the chronosequences on the average CSR strategies (%) by calculating an average time since glacial retreat (semi-quantitative data) at each zone, resulting in the following quantitative values: 3.5; 11.5; 23 and 34 years. Significant differences between pairs of zones in the chronosequence were identified with post-hoc Tukey tests. Similarities among plant communities between

Table 1

The abiotic environment along the chronosequences. Site effect and chronosequence effect nested within site were calculated with mixed effect models. ***: $P < 0.001$; *: $P < 0.05$; (a) data available for site Zongo only.

	0–7 yrs	7–16 yrs	16–30 yrs	30–38 yrs	Site Effect	Chronosequence Effect
Soil moisture (%)	4.97 ± 0.37	4.52 ± 0.38	4.18 ± 0.31	4.42 ± 0.46	*	–
Coarse elements (%)	75.82 ± 2.47	73.20 ± 2.64	61.07 ± 3.02	54.61 ± 3.08	***	***
Slope coefficient (°)	7.89 ± 1.03	12.79 ± 1.87	8.74 ± 1.28	14.11 ± 1.88	***	***
Temperature min. (C°)	−1.19 ± 0.09	−1.73 ± 0.11	−2.25 ± 0.13	−0.68 ± 0.11	(a)	***
Temperature max. (C°)	8.69 ± 0.59	12.17 ± 0.55	11.64 ± 0.50	16.26 ± 0.62	(a)	***

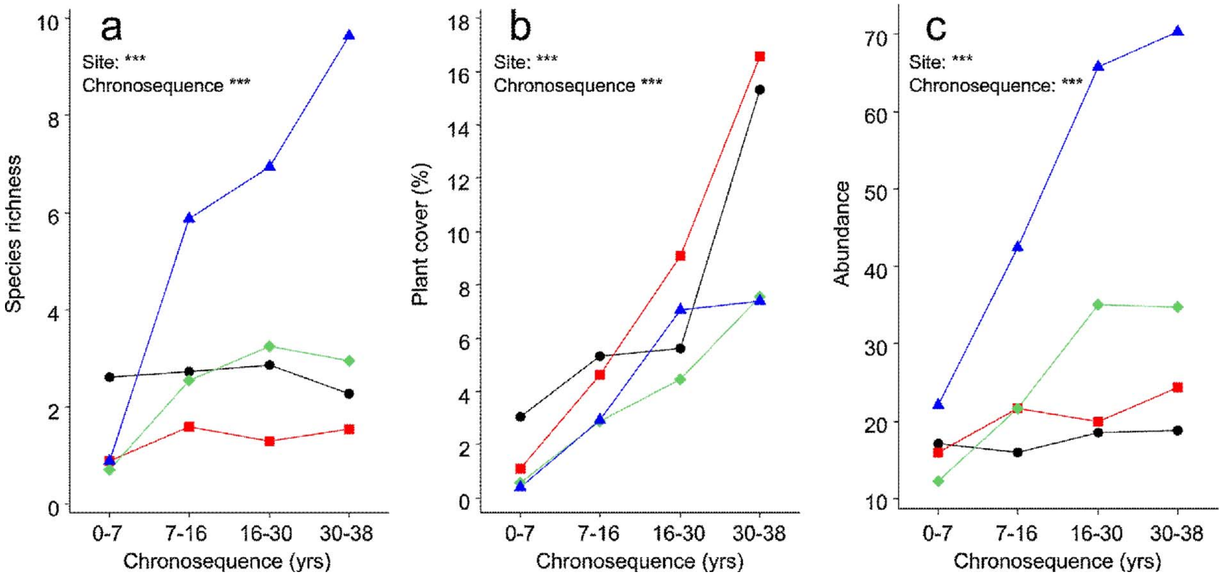


Fig. 2. Succession patterns along the chronosequences, at each site. Each symbol represents the mean value at one site and one period of a chronosequence. (a) Species richness; (b) plant cover at soil surface and (c) abundance (number of individuals at community level). Green losanges: site Yanamarey; black circles: site 10; red squares: site Tarija; blue triangles: site Zongo. Significance of site and chronosequence (nested within site) effects: ***: $P < 0.001$. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

deglaciated zones were tested with the Sørensen index of similarity. Statistics were run with Minitab.

3. Results

3.1. Abiotic environment

The overall granulometry at soil surface changed along the chronosequences, with coarse elements (> 2 mm) reducing significantly in abundance ($P < 0.001$; Table 1). This reduction was negatively correlated with the increasing cover of BSC ($R^2 = 0.29$, $P < 0.001$). Minimum and maximum belowground temperatures significantly increased along the chronosequence (mixed effects model: $P < 0.001$). Maximum temperatures were strongly impacted by the chronosequence, increasing from 8.69 ± 0.59 °C after 0–7 years of glacial retreat to 16.26 ± 0.62 °C after 30–38 years. In contrast we did not observe significant variations in slope along the chronosequences at the four study sites ($P > 0.05$). Soil moisture was also relatively constant and low, between 4 and 5% ($P > 0.05$).

3.2. Species diversity

On a total of 4743 individuals, 64 species were observed along the four chronosequences, 34 in site Zongo, 9 in site Tarija, 10 in site 10 and 29 in site Yanamarey (Table A.1). Four groups of species were distinguished, by order of first appearance: those first established between 0 and 7 years (13 species), those first established between 7 and 16 years (16), those first established between 16 and 30 years (22) and those first established between 30 and 38 years (13). The overall species richness, relative cover and abundance of plants increased at plot scale along the chronosequences, despite significant site effects (mixed effect model: $P < 0.001$; Fig. 2). Site Zongo displayed both the highest species richness and the highest abundance of individuals. Maximum variation in species composition was observed between zone 0–7 and zone 30–38, as revealed by a minimum Sørensen index of 0.18 in site Yanamarey (Table A.3).

3.3. Dispersal

The overwhelming majority of observed species were dispersed by wind although their number decreased significantly from 99% to 92% from 0 to 7 to 30–38 years ($P < 0.001$; Fig. 3a). However, these results differed among sites ($P < 0.05$) with sites 10 and Tarija displaying almost exclusively species dispersed by wind at each zone along the chronosequence (Fig. 3d). Conversely, species dispersed by animals increased significantly and monotonously along the chronosequences, from 0% to 5% (Fig. 3b). Species dispersed by water averaged 2% and remained constant along the chronosequences (Fig. 3c).

3.4. CSR strategies

We observed an overall shift from R-strategists (ruderals) to S-strategists (stress-tolerants) and, to a lesser extent, C-strategists (competitors) along the chronosequences: at community level R-strategy decreased significantly from 80% to 37% ($R^2 = 0.46$; $P < 0.001$) whereas S-strategy increased from 1% to 32% ($R^2 = 0.21$; $P < 0.001$; Fig. 4). Meanwhile, C-strategists shifted from 19% to 35% ($R^2 = 0.05$; $P < 0.001$). These significant differences concerning C, S and R strategies were confirmed at each site separately, with two exceptions: S-strategists did not increase in site Tarija whereas C-strategists did not increased significantly in sites Yanamarey and Zongo.

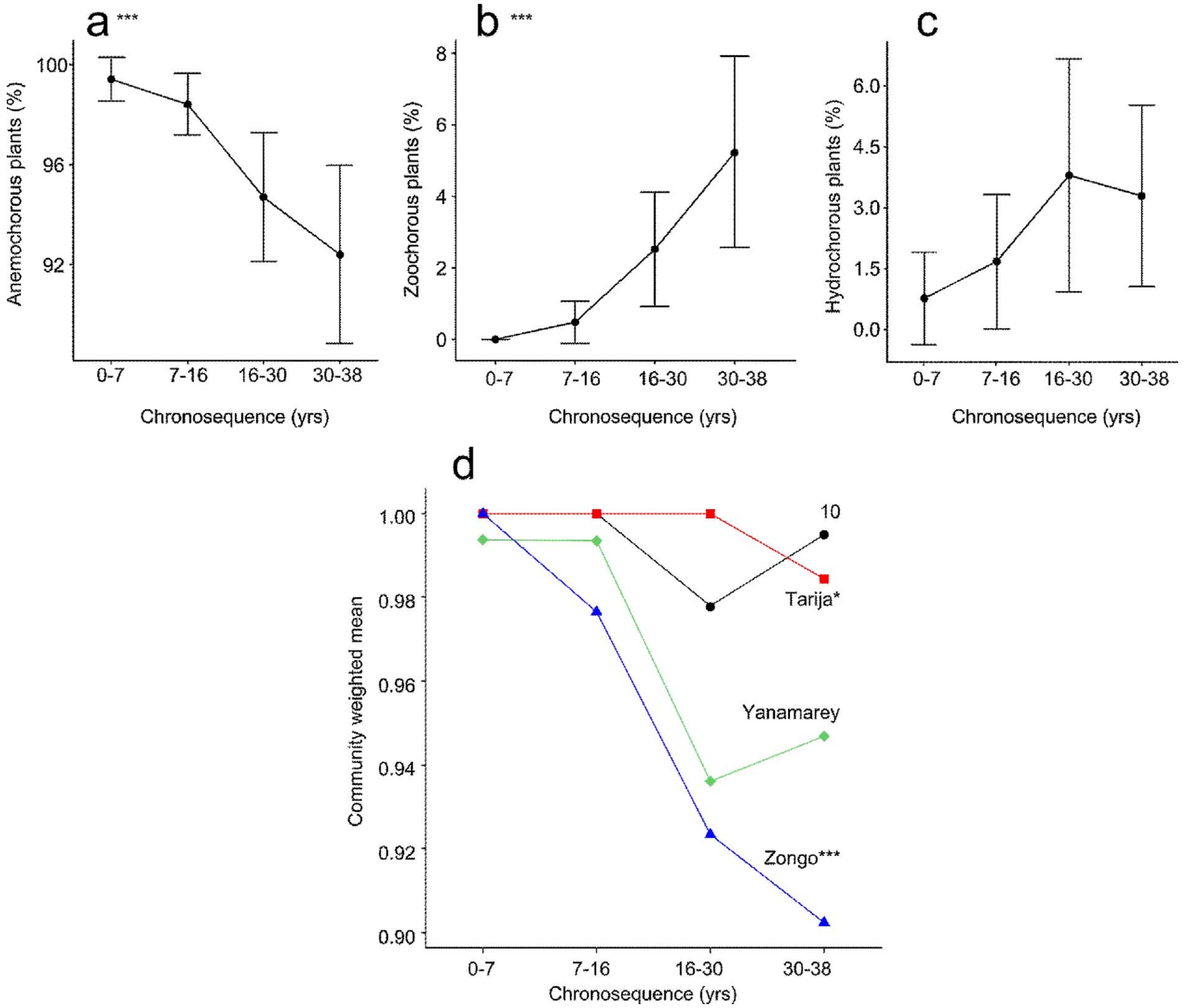


Fig. 3. Plant dispersal strategies along early succession. Each symbol represents the mean value at one site and one period of a chronosequence. Relative proportions of dispersal syndromes: (a) anemochory, (b) zoochory, (c) hydrochory. (d) Community proportion of anemochorous species at each of the four sites. ***: $P < 0.001$; *: $P < 0.05$.

3.5. Spatial associations

At the beginning of the chronosequences (0–7 years), 11 ± 2% of plants were associated with other plants and 86 ± 2% were associated with rocks (Fig. 5a). Plant-plant associations increased monotonously up to 24 ± 1% after 30–38 years whereas associations with rocks decreased significantly while remaining more frequent than plant–plant associations at the end of the chronosequence (49 ± 1%). 205 spatial associations between pairs of plants were clearly sustained by nurse/beneficiary interactions. Within this subsample, the efficiency of the three most frequent nurses, i.e. the average number of beneficiary individuals observed in one nurse individual, reached 0.43 ± 0.24 with *Senecio rufescens*, 0.23 ± 0.08 with *Xenophyllum dactylophyllum* and 0.17 ± 0.05 with *Deyeuxia nitidula*, which was the most frequent nurse in the sampling (Fig. 5b). The three species were infrequent at the beginning of the chronosequence (0–7 years: 60 on a total of 891 individuals).

Fig. 5a shows that plants were poorly associated with BSC 0–7 years after glacial retreat (1.9 ± 1.4%) but this association reached a peak after 16–30 years (39 ± 4%). They remained more frequent than plant–plant associations at the end of the chronosequences. The relative cover occupied by BSC was positively correlated with the frequency of associations between BSC and vascular plants ($R^2 = 0.18$; $P < 0.001$).

4. Discussion

4.1. Common succession patterns at regional scale

Patterns of primary succession are strongly influenced by stochastic factors (e.g., Del Moral et al., 2010), hiding partly assembly rules (Walker and Del Moral, 2003). Reducing their importance to quantify environmental effects on plants is a challenging task during early succession (Marteinsdóttir et al., 2013; Mori et al., 2013). By combining four Andean sites at regional scale, we observed an overall increase in

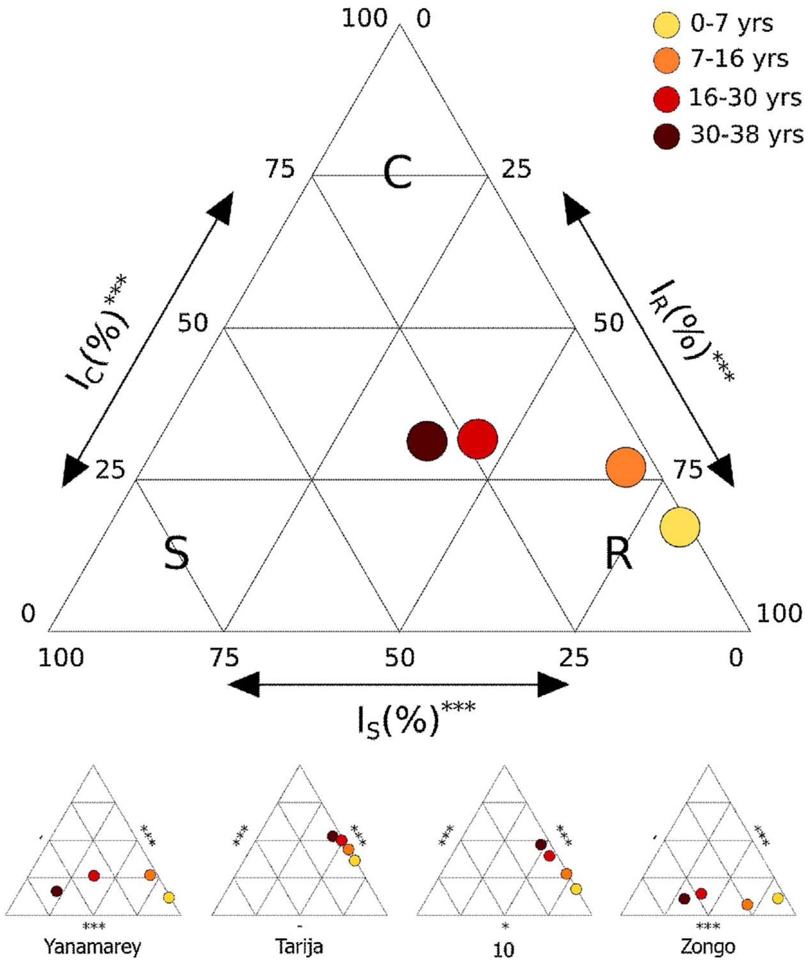


Fig. 4. Functional CSR strategies of plants along the chronosequences. At each deglaciated zone, a bullet represents the combination of the three strategies at community level. Statistical significance extracted from Mixed effect model (site & chronosequence nested within site); ***: $P < 0.001$; **: $P < 0.01$. For each strategy, different letters indicate significant differences (Tukey post-hoc tests). I_c: intensity of competition; I_S: intensity of stress; I_R: intensity of perturbation.

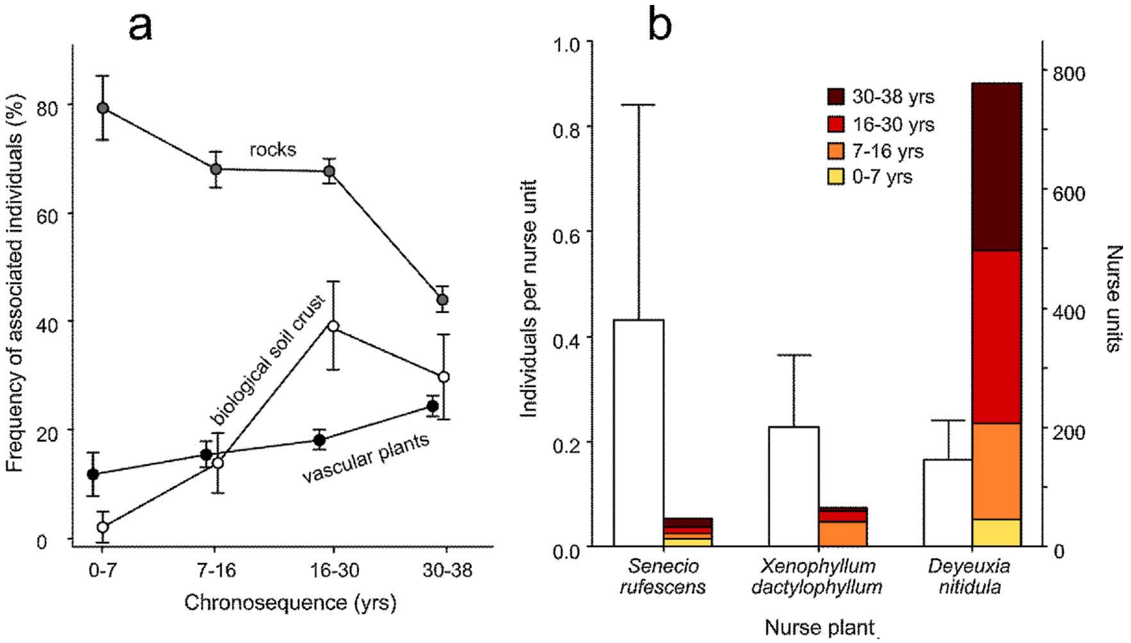


Fig. 5. Spatial associations between plants and refuges along the chronosequence of site Zongo (Bolivia). (a) Associations with biotic and abiotic refuges; (b) ranking of the three principal nurse plants by efficiency and by number of individuals sheltered. Error bars represent 95% confidence intervals.

species richness, abundance, and plant cover along chronosequences. Also, plant communities separated by a larger time frame displayed low similarities. All these results are in line with general patterns reported in primary successions after glacial retreat (Matthews, 1992; Walker and Del Moral, 2003). Another common pattern among sites, although less documented, is the significant shift from R-strategists to S-strategists during succession. As observed and interpreted in the Alps, this shift may indicate a first colonization step by R-strategists taking advantage of glacial till deposit thanks to their rapid phenological development (Caccianiga et al., 2006; Erschbamer and Caccianiga, 2016). Later, stress-tolerant species may dominate the community by maximizing nutrient use before soil development. The slight, but significant, increase in the abundance of C-strategists is in line with later successional stages described during primary succession (e.g. Chapin et al., 1994). These common trends among sites reveal a structured succession pattern in our data, which is not hidden by stochastic factors. This justifies *a posteriori* our choice to build a regional sampling, adding relevance to the discussion developed below.

4.2. Dispersal filter during upward migration

Congruently with our hypotheses, our data revealed several discrepancies with the general knowledge on primary succession. The dominance of anemochorous species early after glacial retreat is a classical feature observed along post-glacial chronosequences (Stöcklin and Bäumler, 1996; Erschbamer and Caccianiga, 2016). However, during plant primary succession centred on 50–350 years (little ice age scale), a decrease in anemochorous species and/or an increasing frequency of species not dispersed by wind is generally observed (e.g. Matthews, 1992; Erschbamer et al., 2008). Under a temporal frame equivalent in terms of temperature warming (our chronosequences since the late 1970s correspond to a warming of approximately 0.4 °C; *i.e.* what would have taken 95 years before the acceleration of warming; see details in the section “Chronosequences under rapid warming” in the methods), the overwhelming majority of species observed at the end of the chronosequences remained anemochorous (92%). Interestingly, a publication on the tropical flora of the Cordillera Real (Bolivia) above 4200 m a.s.l. and concerning 338 species pointed out that 33% were anemochorous (Meneses et al., 2015). This proportion might increase up to 55–60% given that the majority of species with multiple strategies (32%) and with no dispersal data (12%) should be anemochorous. This is roughly two thirds of what we observed at the end of our chronosequences. At the same time the proportion of zoochorous species was slightly higher (6.5% instead of 5%) and the proportion of hydrochorous species reached 7% instead of 2% at the end of the chronosequence. Accordingly, from a strictly dispersal “viewpoint”, tropical alpine plants seem to be confronted to a climatic debt, as shown for animals under the effects of warming (Devictor et al., 2012). This debt mainly concerns tropical alpine plants that are dispersed by water, which are generally observed in aquatic and semi-aquatic tropical alpine ecosystems (Meneses et al., 2015). These species, and to a lesser extent a number of zoochorous species, are not able to match the new rate of warming and are outcompete by wind-dispersers during upward migration.

The most abundant sites in terms of plant individuals (Zongo and, to a lesser extent, Yanamarey) displayed more pronounced successional trends along the chronosequences than sites Tarija and 10, either in terms of dispersal modes, but also in terms of plant species richness or CSR strategies. Interpreting these differences would be speculative at this stage given the limited number of sites and our observational approach. Seed trap experiments may be a relevant method in future studies to further assess the role played by local and regional environmental drivers on the dispersal of alpine plants along primary succession under warming.

4.3. Establishment constrained by low nurse effects

A second discrepancy in our data when compared with general patterns of primary succession concerns plant–plant interactions. After reaching sites, plants require habitat amelioration to achieve a successful establishment phase and compensate the near absence of soils (Erschbamer et al., 2008). In alpine environments, abiotic heterogeneities such as rocks and small crevices can facilitate plant recruitment by acting as seed traps, but also by generating local gradients in soil moisture and soil nutrients (Mori et al., 2013). However, by having biological nurse effects, plants are expected to be better nurses than rocks along primary succession (Haussmann et al., 2010) and they have been shown more associated with plants than abiotic areas early after glacial retreat (Erschbamer et al., 2008). Nurse plants mitigate extreme temperatures, increase soil moisture, soil nutrients and they can decrease the negative effects of wind (Callaway, 2007; Filazzola and Lortie, 2014), a feature that has also been reported in tropical alpine environments (Sklenář, 2009; Anthelme et al., 2012; Ramírez et al., 2015). For these reasons, alpine habitats have been recognized as one of the environments where plant–plant facilitation takes greater importance in assembly rules and increases species diversity (Cavieres et al., 2016).

However, in our sampling, plant-rock associations were more frequent than plant–plant associations. As soon as we consider that the majority of these associations reflects habitat amelioration (see methods), this indicated that established plants do require habitat amelioration (frequent nurse rocks), in line with the stress-gradient hypothesis in alpine regions (Callaway et al., 2002). But, at the same time, the infrequent plant–plant associations were not able to fully provide this habitat amelioration. This low frequency results from the lack of time for plants to colonize the recently deglaciated terrains, as shown with a low plant cover at soil surface.

Importantly, the three most efficient nurse species of the sampling (*Senecio rufescens*, *Xenophyllum dactylophyllum*, *Deyeuxia nitidula*) did not facilitate more than 0.43 individuals per nurse unit. This is extremely low in comparison with other tropical alpine nurses which averaged, e.g., 3.06 individuals per unit with the compact cushion species *Azorella compacta* (N = 60), and 1.60 individuals with the loose cushion species *Pycnophyllum spathulatum* (N = 45) in another alpine site in Bolivia not constrained by recent glacial cover (Sajama National Park, 4900 m a.s.l.; Anthelme et al., 2017). We explain this poor facilitative efficiency –not related with dispersal filter- by the low maturity of nurses as increased ontogenetic variations between nurses and beneficiaries are expected to increase facilitation intensity (Armas et al., 2013). The short time frame offered by accelerated glacial retreat for plant colonization is probably not sufficient for expressing their full nurse potential. This would be especially true for cushion-forming plants like *Xenophyllum dactylophyllum*, which are stress-tolerant species that display slow growth rates (Caccianiga et al., 2006). Consequently, the climatic debt experienced by alpine plants may express not only through dispersal filter, but also during establishment phase through the time lag indebt to the development of a biotic substrate provided by nurse plants. It has been recently suggested that deficient facilitation among plants could be responsible for lagged species range shifts under rapid warming but without field example (HilleRisLambers et al., 2013). Our study evidences it for the first time in the field, adding shape to the conceptual framework on plant–plant interactions under the effects of climate change.

Interestingly, by rapidly developing a fairly high relative cover at the soil surface (indirectly diminishing the proportion of coarse elements) BSC promoted plant diversity and biomass shortly after glacial retreat. The decrease of the BSC nurse effects on plants coincided with the increasing positive effects by vascular plants and temperature increase. As shown by Breen and Levesque (2006), the decreasing positive associations between BSC and plant at the end of the chronosequences may represent a shift from net facilitation to net competition between the two groups. This agrees with observations of BSC developing rapidly after glacial retreat, especially in windswept places and periglacial areas (Türk and Gärtner, 2001; Matthews and Vater, 2015). Accordingly, rapid terrain availability consecutive to warming may generate a transient deficit in vascular nurse plants. In this context, BSC might take greater importance in the success of primary succession by facilitating the establishment phase of the plants that achieved successful dispersal.

4.4. Altered species assemblages in the future

Geographical shortfalls have been identified when building scenarios of climate-related species range shifts and tropical regions like the Andes are among these research gaps (Lenoir and Svenning, 2015). Based on the comprehensive conceptual framework by these authors, among the 64 species observed in our sampling the overwhelming majority were capable of performing upward migration. Accordingly, they belong either to type “march” (upward colonization combined with downward retraction) or “expand” (upward colonization). A small portion of these species may actually not migrate upward and rather move from moraines adjacent to our study sites (“lean” species; see Suárez et al., 2015). This was not distinguishable with our design.

In contrast with these “march” and “expand” species, which found a way to adapt the effects of warming, the species absent from our sampling were not capable of migrating upward rapidly. For example, cushion-forming plants of high Andean wetlands (*Distichia muscoides*, *D. filamentosa*, *O. andina*, *Phylloscirpus deserticola*, *Plantago tubulosa*; Loza Herrera et al., 2015) were absent in our plots (see Table A.1) despite the presence of wetlands at short distance below each site (Meneses et al., 2015; pers. obs.). In parallel, growing evidence shows that the trailing edge distribution of these species is negatively impacted by the combined effects of thermophilization and the intensification of human activities (Squeo et al., 2006; Meneses et al., 2015; Dangles et al., 2017). As such they may be categorized within the “retract” type of species range shift, a type of distribution with no upward migration and a retraction of the trailing edge distribution that makes species highly vulnerable to extinction (Lenoir and Svenning, 2015).

5. Conclusions

At a time when data from long-term experiments are needed to determine the effects of global warming on plant succession (Walker and Wardle, 2014) post-glacial chronosequences constitute a promising alternative approach as soon as they are sufficiently replicated to provide structured patterns at regional scale. We provided an exploratory insight on its ability to determine to what extent communities can turn unbalanced under accelerated warming and, to a certain extent, why. The time lag generated between increased temperature and constant dispersal constraints combined with constant time for nurse plant establishment creates altered plant communities, as suggested in other alpine studies (Dullinger et al., 2012; Svenning and Sandel, 2013). The consecutive deficit in positive interactions among plants is not congruent with current literature, which predicts facilitation interactions to be a crucial driver of plant community organization in alpine systems, even under warming (Callaway, 2007; Cavieres and Sierra-Almeida, 2012; Metz and Tielbörger, 2016). It deserves being taken into account when generalizing the stress-gradient hypothesis. Taking into account the last IPCC scenarios, global temperature by 2055 will rise between 0.2 °C (RCP2.6) and 0.4 °C (RCP8.5) per decade (IPCC, 2013) thus extending the climatic debt born from the time lag between climate warming and ecological processes. This study is mostly exploratory although it provides precise data on a regional scale. Much is to be done and a next topical challenge will be quantifying the respective influences of dispersal filter and nurse limitation on the climatic debt that constrains plant distribution.

Acknowledgments

We thank Ariel Lliully, Arely Palabral, Edwin J. Palomino, Luzmila Dávila Roller, Marlene Rosario Guerrero, Selwyn Valverde, Martin Salvador Gianny Rodriguez y Noriza Garcia Romero for outstanding help in the field, plant determination and data on glacial retreat. We also thank the Editor and two anonymous reviewers for their valuable comments. This work was funded by the *Fond Français pour l’Environnement Mondial* (FFEM) and the *Fondation pour la Recherche sur la Biodiversité* (FRB) (Modeling BIOdiversity and land use interactions under changing glacial water availability in Tropical High Andean Wetlands” – BIOTHAW, AAP-SCEN-2011-II). Antoine Rabatel acknowledges the support of LabEx OSUG@2020 (*Investissements d’avenir* – ANR10LABX56), GLACIOCLIM and LMI GREAT-ICE funded by the French *Institut de Recherche pour le Développement* (IRD). The Pléiades satellite images from 2013 were acquired through the CNES program ISIS_Pléaides-FC18473.

Appendix A

Table A.1
List of the species observed in our sampling, with their zone of first establishment along the chronosequence and their dispersal mode(s).

Species code	Genus	Species	Family	Zone of first establishment	Dispersal
Afol	*Agrostis*	*foliata*	Poaceae	7–16 yrs	A
Alep	*Anthochloa*	*lepidula*	Poaceae	0–7 yrs	A
Are1	*Arenaria*	*sp1*	Caryophyllaceae	0–7 yrs	Z
Are2	*Arenaria*	*sp2*	Caryophyllaceae	30–38 yrs	Z
Aspl	*Asplenium*	sp.	Aspleniaceae	30–38 yrs	A
Atol	*Agrostis*	*tolucensis*	Poaceae	7–16 yrs	A
Bcre	*Bartsia*	*crenoloba*	Orobanchaceae	7–16 yrs	Z
Bdif	*Bartsia*	*diffusa*	Orobanchaceae	16–30 yrs	AZ
Bmon	*Brayopsis*	*monimocalyx*	Brassicaceae	30–38 yrs	Z
Bpic	*Belloa*	*pickeringii*	Asteraceae	7–16 yrs	A
Cala	*Calamagrostis*	sp.	Poaceae	16–30 yrs	A
Camp	*Calamogrostis*	*ampliflora*	Poaceae	7–16 yrs	A
Ccra	*Cerastium*	crassipes	Caryophyllaceae	30–38 yrs	Z
Cfib	*Calamagrostis*	*fibrovaginata*	Poaceae	30–38 yrs	A
Clag	*Calamagrostis*	*lagurus*	Poaceae	16–30 yrs	A
Cnit	*Calamagrostis*	*nitidula*	Poaceae	0v7 yrs	A
Cort	*Cortaderia*	sp.	Poaceae	16–30 yrs	A
Cova	*Calamagrostis*	*ovata*	Poaceae	0–7 yrs	A
Covo	*Chersodoma*	*ovopedata*	Asteraceae	16–30 yrs	A
Cscl	*Calamagrostis*	*sclerantha*	Poaceae	30–38 yrs	A
Cspi	*Calamagrostis*	*spicigera*	Poaceae	16–30 yrs	A
Dcal	*Dissanthelium*	*calycinum*	Poaceae	16–30 yrs	A
Ddis	*Draba*	*discoidea*	Brassicaceae	16–30 yrs	A
Dflo	*Dielsiochloa*	*floribunda*	Poaceae	7–16 yrs	A
Drab	*Draba*	sp.	Brassicaceae	30–38 yrs	AZ
Edent	*Epilobium*	*denticulatum*		7–16 yrs	A
Elap	*Elaphoglossum*	sp.	Dryopteridaceae	16–30 yrs	A
Erup	*Ephedra*	*rupestris*	Ephedraceae	7–16 yrs	Z
Gcor	*Galium*	*corymbosum*	Rubiaceae	30–38 yrs	Z
Gent	*Gentianella*	sp.	Gentianaceae	16–30 yrs	A
Gery	*Gamochaeta*	*erythractis*	Asteraceae	16–30 yrs	A
Glac	*Gnaphalium*	*lacteum*	Asteraceae	7–16 yrs	A
Glul	*Gamochaeta*	*lulioana*	Asteraceae	30–38 yrs	A
Gpun	*Gynoxys*	sp.	Asteraceae	16–30 yrs	A
Hobt	*Hypolepsis*	*obtusata*	Dennstaedtiaceae	0–7 yrs	AH
Kkur	*Koeleria*	*kurtzii*	Poaceae	7–16 yrs	A
Lfer	*Loricaria*	*ferruginea*	Asteraceae	30–38 yrs	A
Lrac	*Luzula*	*racemosa*	Juncaceae	16–30 yrs	A
Lvul	*Luzula*	*vulcanica*	Juncaceae	16–30 yrs	A
Mvol	*Muehlenbeckia*	*volcanica*	Polygonaceae	16–30 yrs	Z
Orth	*Orthrosanthus*	sp.	Iridaceae	16–30 yrs	AZ
Pkur	*Poa*	*kurtzii*	Poaceae	16–30 yrs	A
Poa	*Poa*	sp.	Poaceae	7–16 yrs	A
Pper	*Poa*	*perligulata*	Poaceae	0–7 yrs	A
Ppro	*Pernettya*	*prostrata*	Ericaceae	16–30 yrs	Z
Pspi	*Poa*	*spicigera*	Poaceae	16–30 yrs	A
Sade	*Senecio*	*adenophyllus*	Asteraceae	7–16 yrs	A
Saur	*Senecio*	sp1.	Asteraceae	0–7 yrs	A
Sene	*Senecio*	sp2.	Asteraceae	30–38 yrs	A
Seva	*Senecio*	*evacoides*	Asteraceae	16–30 yrs	A
Shoh	*Senecio*	*hohenackeri aff.*	Asteraceae	0–7 yrs	A
Smag	*Saxifraga*	*magellanica*	Saxifragaceae	0–7 yrs	AH
Sman	*Silene*	*mandonii*	Caryophyllaceae	16–30 yrs	ZH
Smod	*Senecio*	*modestus*	Asteraceae	7–16 yrs	A
Sp	Undetermined			16–30 yrs	A
Srhi	*Senecio*	*rhizomatus*	Asteraceae	7–16 yrs	A
Sruf	*Senecio*	*rufescens*	Asteraceae	0–7 yrs	A
Sser	*Senecio*	*serratifolius*	Asteraceae	0–7 yrs	A
Vniv	*Valeriana*	*nivalis*	Caprifoliaceae	30–38 yrs	A
Vpet	*Valeriana*	*petersenii*	Caprifoliaceae	7–16 yrs	A
Wnub	*Werneria*	*nubigena*	Asteraceae	30–38 yrs	A
Xdac	*Xenophyllum*	*dactylophyllum*	Asteraceae	0–7 yrs	A

A: anemochory; Z: zoochory; H: hydrochory (Meneses et al., 2015).

Table A.2
CSR strategy for each species in our sampling, with CSR coefficients (see Methods for calculation) and CSR attributes. Full name of species available in Table A.1.

Species code	CSR strategy	C coefficient	S coefficient	R coefficient	C attibutes	S attributes	R attributes
Afol	CR	0.5	0	0.5	High stature, robust leaves, lateral spread.		Large production of seeds, fast growth rate
Alep	R	0	0	1			Fast growth rate, high production of seeds
Are1	S	0	1	0		Low stature, small leaves	
Are2	S	0	1	0		Low stature, small leaves	
Aspl	CSR	0.33	0.33	0.33	Large leaves	High tolerance to dessication	High production of spores, leaves thin
Atol	CSR	0.33	0.33	0.33	Lateral spread	Compact foliage (protection against stress)	Fast gtowth rate, probable high SLA
Bcre	SR	0	0.5	0.5		Small, thick and hairy leaves	Limited lateral spread, relatively fast growth rate
Bdif	SR	0	0.5	0.5		Small, thick and hairy leaves	Limited lateral spread, relatively fast growth rate
Bmon	S	0	1	0		Dense rosette structure, low stature, small, thick leaves	
Bpic	S	0	1	0		Small, cushion-forming species	
Cala	S	1	1	1		Low, compact stature	Fast growth rate
Camp	CR	0.5	0	0.5	High, extensive lateral spread		Large production of seeds, fast growth rate
Ccra	S	0	1	0		Small, thick leaves, cushion-forming species	
Cfib	CSR	0.33	0.33	0.33	Relatively high stature, lateral spread	Thick leaves (probably low SLA)	Fast growth rate
Clag	S	0	1	0		Low stature, small leaves, high longevity of leaves	
Cnit	CR	0.5	0	0.5	High stature, lateral spread, robust leaves		Fast growth rate, high production of seeds
Cort	CR	0.5	0	0.5	Tussock, high stature, robust leaves		Fast growth rate, present in areas with human intervention
Cova	R	0	0	1			Fast growth rate, high producton of seeds
Covo	CS	0.5	0.5	0	High stature, extensive lateral spread	Moderate/slow growth rate	
Cscl	CR	0.5	0	0.5	Tussock, high stature, robust leaves		Fast growth rate
Cspi	S	0	1	0		Low stature	
Dcal	SR	0	0.5	0.5		Low stature, small leaves	Present in area with human intervention
Ddis	S	0	1	0		Very low stature, small and sturdy leaves, basal leaves rounded, raceme compact	
Dflo	SR	0	0.5	0.5		Low, compact stature	Fast growth rate, relatively large and thin leaves
Drab	S	0	1	0		Low stature, small, thick leaves, low growth rate	
Edent	R	0	0	1			Large, thin leaves, fast growth rate
Elap	CS	0.5	0.5	0	Large leaves	Leathery leaves, resistance to dessication	
Erup	CS	0.5	0.5	0	Shrub with vertical and lateral spread, robust leaves	Prostrate shrub, needle-like leaves, low growth rate	
Gcor	S	0	1	0		Small, thick, needle-like leaves	
Gent	SR	0	0.5	0.5		Small stature, small thick leaves	Fast growth rate
Gery	S	0	1	0		Small cushion-forming species, small leaves, low growth rate	
Glac	S	0	1	0		Low, compact stature, thick leaves	
Glul	S	0	1	0		Thick, hairy leaves, low stature, cushion-like shape	
Gpun	C	1	0	0	Large shrub (dominant growth form)		
Hobt	SR	0	0.5	0.5		Resistant to dessication	Large and thin leaves
Kkur	CS	0.5	0.5	0	Lateral spread	Compact shape, leaves with long life span	
Lfer	C	1	0	0	Erect shrub		
Lrac	S	0	1	0		Thick, needle-like leaves, low growth rate	
Lvul	CS	0	0.5	0	Lateral spread (stoloniferous species)	Thick, needle-like leaves, low growth rate	
Mvol	S	0	1	0		Small plant, low stature, leathery leaves	
Orth	S	0	1	0		Needle-like leaves	
Pkur	CS	0.5	0.5	0	large, wide and robust leaves	Cushion-like growth form	
Poa	CSR	0.33	0.33	0.33	Dense shoots, relatively high stature	Low stature	fast growth rate
Pper	SR	0	0.5	0.5		Small plant, cushion-like stature	Seeds all year long
Ppro	CS	0.5	0.5	0	Lateral spread	Prostrate shrub, small linear leaves, low growth rate	
Pspi	SR	0	0.5	0.5		Small leaves, low stature	Seeds all year long
Sade	CS	0.5	0.5	0	Erect shrub	Leathery leaves	
Saur	CR	0.5	0	0.5	Large, erect shrub		Fast growth rate
Sene	S	0	1	0		Dense, leathery leaves, cushion-forming species, low stature	
Seva	S	0	1	0		Cushion-forming species, leaves small and thick, cushions	
Shoh	CR	0.5	0	0.5	Erect, tall herb		Fast growth rate

(continued on next page)

Table A.2 *(continued)*

Species code	CSR strategy	C coefficient	S coefficient	R coefficient	C attibutes	S attributes	R attributes
Smag	S	0	1	0		Dense, leathery leaves, low growth rate, cushion-forming species	
Sman	R	0	0	1			leaves large and thin, fast growth rate (probable high SLA)
Smod	CS	0.5	0.5	0	Erect shrub	Low stature, dense foliage	
Sp	S	1	1	1		Dense, leathery leaves, cushion-forming species, low stature	
Srhi	SR	0	0.5	0.5		Rhizomatous species	Fast growth rate, large leaves
Sruf	CR	0.5	0	0.5	Erect shrub		Fast growth rate,
Sser	CR	0.5	0	0.5	Lateral spread		Rosette,extended seed availability, relatively fast growth rate
Vniv	CR	0.5	0	0.5	Lateral spread		Large, thin leaves, plant protected by rocks, fast growth rate
Vpet	R	0	0	1			Large, thin leaves, plant protected by rocks, fast growth rate
Wnub	CSR	0.33	0.33	0.33	Robust plant with large leaves, invasive behaviour	Leaves leathery or thick	Abundant in disturbed areas (tolerant to grazing)
Xdac	SR	0	0.5	0.5		Dense, leathery leaves, cushion-forming species	Large production of seeds, relatively fast growth

Table A.3
Sørensen index of similarity among deglaciated zones at each of the four chronosequences.

Pairs of zones	Zongo	Tarija	10	Yanamarey
0–7 yrs *vs.* 7–16 yrs	0.53	0.86	0.83	0.73
0–7 yrs *vs.* 16–30 yrs	0.39	0.86	0.77	0.17
0–7 yrs *vs.* 30–38 yrs	0.29	0.36	0.50	0.18
7–16 yrs *vs.* 16–30 yrs	0.71	1.00	0.80	0.30
7–16 yrs *vs.* 30–38 yrs	0.73	0.50	0.57	0.24
16–30 yrs *vs.* 30–38 yrs	0.76	0.50	0.80	0.53

References

Anthelme, F., Dangles, O., 2012. Plant–plant interactions in tropical alpine environments. Perspect. Plant Ecol. Evol. Syst. 14, 363–372.

Anthelme, F., Buendia, B., Mazoyer, C., Dangles, O., 2012. Unexpected mechanisms sustain the stress gradient hypothesis in a tropical alpine environment. J. Veg. Sci. 23, 62–72.

Anthelme, F., Cavieres, L.A., Dangles, O., 2014a. Facilitation among plants in alpine environments in the face of climate change. Front. Plant Sci. 5. http://dx.doi.org/10.3389/fpls.2014.00387.

Anthelme, F., Jacobsen, D., Macek, P., Meneses, R.I., Moret, P., Beck, S., Dangles, O., 2014b. Biodiversity patterns and continental insularity in the high tropical Andes. Arct. Antarct. Alp. Res. 46, 811–828.

Anthelme, F., Meneses, R.I., Huaman Valero, N.N., Pozo, P., Dangles, O., 2017. Fine nurse variations explain discrepancies in the stress-interaction relationship in alpine regions. Oikos. http://dx.doi.org/10.1111/oik.04248.

Armas, C., Schöb, C., Gutiérrez, J.R., 2013. Modulating effects of ontogeny on the outcome of plant–plant interactions along stress gradients. New Phytol. 200, 7–9.

Blois, J.L., Williams, J.W., Fitzpatrick, M.C., Jackson, S.T., Ferrier, S., 2013. Space can substitute for time in predicting climate-change effects on biodiversity. Proc. Natl. Acad. Sci. 110, 9374–9379.

Bradley, R.S., Vuille, M., Diaz, H.F., Vergara, W., 2006. Threats to water supplies in the tropical Andes. Science 312, 1755–1756.

Breen, K., Lévesque, E., 2008. The influence of biological soil crusts on soil characteristics along a High Arctic glacier foreland Nunavut, Canada. Antarct. Arct. Alp. Res. 40, 287–297.

Breen, K., Levesque, E., 2006. Proglacial succession of biological soil crusts and vascular plants: biotic interactions in the High Arctic. Botany 84, 1714–1731.

Bueno, A., Llambí, L.D., 2015. Facilitation and edge effects influence vegetation regeneration in old-fields at the tropical Andean forest line. Appl. Veg. Sci. 18, 613–623.

Bulleri, F., Bruno, J.F., Silliman, B.R., Stachowicz, J.J., 2016. Facilitation and the niche: implications for coexistence: range shifts and ecosystem functioning. Funct. Ecol. 30, 70–78.

Cáceres, Y., Llambí, L.D., Rada, F., 2015. Shrubs as foundation species in a high tropical alpine ecosystem: a multi-scale analysis of plant spatial interactions. Plant Ecol. Divers. 8, 147–161.

Caccianiga, M., Luzzaro, A., Pierce, S., Ceriani, R.M., Cerabolini, B., 2006. The functional basis of a primary succession resolved by CSR classification. Oikos 112, 10–20.

Callaway, R.M., Brooker, R.W., Choler, P., Kikvidze, Z., Lortie, C.J., Michalet, R., Paolini, L., Pugnaire, F.I., Newingham, B., Aschehoug, E.T., Armas, C., Kikodze, D., Cook, B.J., 2002. Positive interactions among alpine plants increase with stress. Nature 417, 844–848.

Callaway, R.M., 2007. Positive Interactions and Interdependence in Plant Communities. Springer, Dordrecht, Netherlands.

Cannone, N., Diolaiuti, G., Guglielmin, M., Smiraglia, C., 2008. Accelerating climate change impacts on alpine glacier forefield ecosystems in the European Alps. Ecol. Appl. 18, 637–648.

Cauvy-Fraunié, S., Andino, P., Espinosa, R., Calvez, R., Jacobsen, D., Dangles, O., 2016. Ecological responses to experimental glacier-runoff reduction in alpine rivers. Nat. Commun. 7. http://dx.doi.org/10.1038/ncomms12025.

Cavieres, L.A., Sierra-Almeida, A., 2012. Facilitative interactions do not wane with warming at high elevations in the Andes. Oecologia 170, 575–584.

Cavieres, L.A., Hernández-Fuentes, C., Sierra-Almeida, A., Kikvidze, Z., 2016. Facilitation among plants as an insurance policy for diversity in alpine communities. Funct. Ecol. 30, 52–59.

Chapin, F.S., Walker, L.R., Fastie, C.L., Sharman, L.C., 1994. Mechanisms of primary succession following deglaciation at Glacier Bay, Alaska. Ecol. Monogr. 64, 149–175.

Chen, I.C., Hill, J.K., Ohlemüller, R., Roy, D.B., Thomas, C.D., 2011. Rapid range shifts of species associated with high levels of climate warming. Science 333, 1024–1026.

Cooper, D.J., Kaczynski, K., Slayback, D., Yager, K., 2015. Growth and organic carbon production in peatlands dominated by *Distichia muscoides*, Bolivia, South America. Arct. Antarct. Alp. Res. 47, 505–510.

Dangles, O., Rabatel, A., Kraemer, M., Zeballos, G., Soruco, A., Jacobsen, D., Anthelme, F., 2017. Ecosystem sentinels for climate change? Evidence of wetland cover changes over the last 30 years in the tropical Andes. PLoS One 12, e0175814. http://dx.doi.org/10.1371/journal.pone.0175814.

Del Moral, R., Saura, J.M., Emenegger, J.N., 2010. Primary succession trajectories on a barren plain, Mount St. Helens, Washington. J. Veg. Sci. 21, 857–867.

Devictor, V., Van Swaay, C., Brereton, T., Chamberlain, D., Heliölä, J., Herrando, S., Julliard, R., Kuussaari, M., Lindström, A., Reif, J., Roy, D.B., Schweiger, O., Settele, J., Stefanescu, C., Van Strien, A., Van Turnhout, C., Vermouzek, Z., WallisDeVries, M., Wynhoff, I., Jiguet, F., 2012. Differences in the climatic debts of birds and butterflies at a continental scale. Nat. Clim. Change 2, 121–124.

Dullinger, S., Willner, W., Plutzar, C., Englisch, T., Schratt-Ehrendorfer, L., Moser, D., Ertl, S., Essl, F., Niklfeld, H., 2012. Post-glacial migration lag restricts range filling of plants in the European Alps. Glob. Ecol. Biogeogr. 21, 829–840.

Erschbamer, B., Caccianiga, M.S., 2016. Glacier Forelands: Lessons of Plant Population and Community Development. Springer, Berlin Heidelberg, pp. 1–26.

Erschbamer, B., Niederfriniger Schlag, R., Winkler, E., 2008. Colonization processes on a central Alpine glacier foreland. J. Veg. Sci. 19, 855–862.

Filazzola, A., Lortie, C.J., 2014. A systematic review and conceptual framework for the mechanistic pathways of nurse plants. Glob. Ecol. Biogeogr. 23, 1335–1345.

Gottfried, M., Pauli, H., Futschik, A., Akhalkatsi, M., Barancok, P., Benito Alonso, J.L., Coldea, G., Dick, J., Erschbamer, B., Fernandez Calzado, M.R., Kazakis, G., Krajci, J., Larsson, P., Mallaun, M., Michelsen, O., Moiseev, D., Moiseev, P., Molau, U., Merzouki, A., Nagy, L., Nakhutsrishvili, G., Pedersen, B., Pelino, G., Puscas, M., Rossi, G., Stanisci, A., Theurillat, J.-P., Tomaselli, M., Villar, L., Vittoz, P., Vogiatzakis, I., Grabherr, G., 2012. Continent-wide response of mountain vegetation to climate change. Nat. Clim. Change 2, 111–115.

Grime, J.P., 1977. Evidence for the existence of three primary strategies in plants and its relevance to ecological and evolutionary theory. Am. Nat. 111, 1169–1194.

Grime, J.P., 2002. Plant Strategies, Vegetation Processes, and Ecosystem Properties, second edition. John Wiley & Sons, Chichester.

Grossiord, C., Sevanto, S., Adamas, H.D., Collins, A.D., Dickman, L.T., McBranch, N., Michaletz, S.T., Stockton, E.A., Vigil, M., McDowell, N.G., 2017. Precipitation, not air temperature, drives functional responses of trees in semi-arid ecosystems. J. Ecol. 105, 163–175.

Grytnes, J.A., Kapfer, J., Jurasinski, G., Birks, H.H., Henriksen, H., Klanderud, K., Odland, A., Ohlson, M., Wipf, S., Birks, H.J.B., 2014. Identifying the driving factors behind observed elevational range shifts on European mountains. Glob. Ecol. Biogeogr. 23, 876–884.

Harsch, M.A., HilleRisLambers, J., 2016. Climate warming and seasonal precipitation change interact to limit species distribution shifts across western North America. PLoS One 11, e0159184.

Haussmann, N.S., McGeoch, M.A., Boelhouwers, J.C., 2010. Contrasting nurse plants and nurse rocks: the spatial distribution of seedlings of two sub-Antarctic species. Acta Oecol. 36, 299–305.

HilleRisLambers, J., Harsc, M.A., Ettinger, A.K., Ford, K.R., Theobald, E.J., 2013. How will biotic interactions influence climate change–induced range shifts? Ann. N. Y. Acad. Sci. 1297, 112–125.

Hodgson, J.G., Wilson, P.J., Hunt, R., Grime, J.P., Thompson, K., 1999. Allocating CSR plant functional types: a soft approach to a hard problem. Oikos 85, 282–294.

IPCC, 2013. Climate Change 2013: the Physical Science Basis. Contribution of Working Group I to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge and New York.

Jacobsen, D., Milner, A.M., Brown, L.E., Dangles, O., 2012. Biodiversity under threat in glacier-fed river systems. Nat. Clim. Change 2, 361–364.

Körner, C., 2003. Alpine Plant Life. Springer.

Klanderud, K., Totland, Ø., 2007. The relative role of dispersal and local interactions for alpine plant community diversity under simulated climate warming. Oikos 116, 1279–1288.

Kolff, H., Kolff, K., 2005. Wildflowers of the Cordillera Blanca. Instituto de Montaña, Huaraz, Peru.

Lavergne, S., Mouquet, N., Thuiller, W., Ronce, O., 2010. Biodiversity and climate change: integrating evolutionary and ecological responses of species and communities. Ann. Rev. Ecol. Evol. Syst. 41, 321–350.

Lenoir, J., Svenning, J.C., 2015. Climate-related range shifts–a global multidimensional synthesis and new research directions. Ecography 38, 15–28.

Loza Herrera, S., Meneses, R.I., Anthelme, F., 2015. Comunidades vegetales de los bofedales de la Cordillera Real Bolivia bajo el calentamiento global. Ecol. Bolivia 50, 39–56.

MRI–Mountain Research Initiative EDW Working Group, 2015. Elevation-dependent warming in mountain regions of the world. Nat. Clim. Change 5, 424–430.

Maestre, F.T., Callaway, R.M., Valladares, F., Lortie, C.J., 2009. Refining the stress-gradient hypothesis for competition and facilitation in plant communities. J. Ecol. 97, 199–205.

Malatesta, L., Tardella, F.M., Piermarteri, K., Catorci, A., 2016. Evidence of facilitation cascade processes as drivers of successional patterns of ecosystem engineers at the upper altitudinal limit of the dry puna. PLoS One 11, e0167265.

Marta, S., Lacasella, F., Gratton, P., Cesaroni, D., Sbordoni, V., 2016. Deciphering range dynamics: effects of niche stability areas and post-glacial colonization on alpine species distribution. J. Biogeogr. 43, 2186–2198.

Marteinsdóttir, B., Thórhallsdóttir, T.E., Svavarsdóttir, K., 2013. An experimental test of the relationship between small scale topography and seedling establishment in primary succession. Plant Ecol. 214, 1007–1015.

Matthews, J.A., Vater, A.E., 2015. Pioneer zone geo-ecological change: observations from a chronosequence on the Storbreen glacier foreland, Jotunheimen, southern Norway. Catena 135, 219–230.

Matthews, J.A., 1992. The Ecology of Recently Deglaciated Terrain. A Geological Approach to Glacier Forelands and Primary Succession. Cambridge University Press, Cambridge, UK.

Meneses, R.I., Beck, S., Anthelme, F., 2015. La Cordillera Real y sus Plantas. IRD & Herbario Nacional de Bolivia, La Paz, Bolivia.

Metz, J., Tielbörger, K., 2016. Spatial and temporal aridity gradients provide poor proxies for plant–plant interactions under climate change: a large-scale experiment. Funct. Ecol. 30, 20–29.

Moret, P., Aráuz, M.D.L.Á., Gobbi, M., Barragán, Á., 2016. Climate warming effects in the tropical Andes: first evidence for upslope shifts of Carabidae (Coleoptera) in Ecuador. Insect Conserv. Divers. 9, 342–350.

Mori, A.S., Uchida, M., Kanda, H., 2013. Non-stochastic colonization by pioneer plants after deglaciation in a polar oasis of the Canadian High Arctic. Polar Sci. 7, 278–287.

Parmesan, C., Hanley, M.E., 2015. Plants and climate change: complexities and surprises. Ann. Bot. 116, 849–864.

Rabatel, A., Jomelli, V., Francou, B., Naveau, P., Grancher, D., 2008. The Little Ice Age in the tropical Andes of Bolivia (16°S) and its implication for a climate reconstruction. Quat. Res. 70, 198–212. http://dx.doi.org/10.1016/j.yqres.2008.02.012.

Rabatel, A., Bermejo, A., Loarte, E., Soruco, A., Gomez, J., Leonardini, G., Vincent, C., Sicart, J.E., 2012. Can the snowline be used as an indicator of the equilibrium line and mass balance for glaciers in the outer tropics? J. Glaciol. 58, 1027–1036. http://dx.doi.org/10.3189/2012JoG12J027.

Rabatel, A., Francou, B., Soruco, A., Gomez, J., Caceres, B., Ceballos, J.L., Basantes, R., Vuille, M., Sicart, J.-E., Huggel, C., Scheel, M., Lejeune, Y., Arnaud, Y., Collet, M., Condom, T., Consoli, G., Favier, V., Jomelli, V., Galarraga, R., Ginot, P., Maisincho, L., Mendoza, J., Ménégoz, M., Ramirez, E., Ribstein, P., Suarez, W., Villacis, M., Wagnon, P., 2013. Current state of glaciers in the tropical Andes: a multi-century perspective on glacier evolution and climate change. Cryosphere 7, 81–102. http://dx.doi.org/10.5194/tc-7-81-2013.

Ramírez, L.A., Rada, F., Llambí, L.D., 2015. Linking patterns and processes through ecosystem engineering: effects of shrubs on microhabitat and water status of associated plants in the high tropical Andes. Plant Ecol. 216, 213–225.

Sarmiento, L., Llambí, L.D., Escalona, A., Marquez, N., 2003. Vegetation patterns, regeneration rates and divergence in an old-field succession of the high tropical Andes. Plant Ecol. 166, 145–156.

Sattin, S.R., Cleveland, C., Hood, E., Reed, S.C., King, A.J., Schmidt, S.K., Robeson, M.S., Ascarrunz, N., Nemergut, D.R., 2009. Functional shifts in unvegetated, perhumid, recently-deglaciated soil do not correlate with shifts in soil bacterial community composition. J. Microbiol. 47, 673–681.

Scherrer, D., Körner, C., 2011. Topographically controlled thermal-habitat differentiation buffers alpine plant diversity against climate warming. J. Biogeogr. 38, 406–416.

Sklenář, P., 2009. Presence of cushion plants increases community diversity in the high equatorial Andes. Flora 204, 270–277.

Smith, A.P., 1994. Introduction to Tropical Alpine Vegetation. In: Rundel, P.W., Smith, A., Meinzer, F.C. (Eds.), Cambridge University Press, Cambridge, pp. 1–19.

Sonnier, G., Shipley, B., Navas, M.-L., 2010. Quantifying relationships between traits and explicitly measured gradients of stress and disturbance in early successional plant communities: trait-environment linkages in early successional communities. J. Veg. Sci. 21, 1014–1024.

Soruco, A., Vincent, C., Francou, B., Gonzalez, J.F., 2009. Glacier decline between 1963 and 2006 in the Cordillera Real, Bolivia. Geophys. Res. Lett. 36, L03502. http://dx.doi.org/10.1029/2008GL036238.

Squeo, F.A., Warner, B., Aravena, R., Espinoza, D., 2006. Bofedales: high altitude peatlands of the central Andes. Revista Chilena de Historia Natural 79, 245–255.

Stöcklin, J., Bäumler, E., 1996. Seed rain, seedling establishment and clonal growth strategies on a glacier foreland. J. Veg. Sci. 7, 45–56.

Suárez, E., Orndahl, K., Goodwin, K., 2015. Lava flows and moraines as corridors for early plant colonization of glacier forefronts on tropical volcanoes. Biotropica 47, 645–649.

Svenning, J.C., Sandel, B., 2013. Disequilibrium vegetation dynamics under future climate change. Am. J. Bot. 100, 1266–1286.

Türk, R., Gärtner, G., 2001. Biological soil crusts of the subalpine, alpine, and nival areas in the Alps. In: Belnap, J., Lange, O.L. (Eds.), Biological Soil Crusts: Structure, Function, and Management. Springer, Berlin, Heidelberg, New York, pp. 67–73.

Tarazona Santos, N.E., 2005. Generación de descargas mensuales en subcuencas de la cuenca del río Santa utilizando el método de Lutz Scholz (No. P10 T37-T). Universidad Nacional Agraria La Molina, Lima (Peru) Master.

Tovar, C., Arnillas, C.A., Cuesta, F., Buytaert, W., 2013. Diverging responses of tropical Andean biomes under future climate conditions. PLoS One 8, e63634.

UGRH, 2014. Inventario Nacional de Glaciares y Lagunas. Unidad de Glaciología y Recursos Hídricos, Autoridad Nacional del Agua, Huaraz, Peru.

Urban, M.C., 2015. Accelerating extinction risk from climate change. Science 348, 571–573.

Violle, C., Navas, M.L., Vile, D., Kazakou, E., Fortunel, C., Hummel, I., Garnier, E., 2007. Let the concept of trait be functional!. Oikos 116, 882–892.

Walker, L.R., Del Moral, R., 2003. Primary Succession and Ecosystem Rehabilitation. Cambridge University Press, Cambridge, UK.

Walker, L.R., Wardle, D.A., 2014. Plant succession as an integrator of contrasting ecological time scales. Trends Ecol. Evol. 29, 504–510.

Zimmer, A., Meneses, R.I., Rabatel, A., Soruco, A., Anthelme, F., 2014. Caracterizar la migración altitudinal de las comunidades vegetales altoandinas frente al calentamiento global mediante cronosecuencias post-glaciales recientes. Ecol. Bolivia 49, 27–41.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 103–114

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Research article

Cushion-plant protection determines guild-dependent plant strategies in high-elevation peatlands of the Cordillera Real, Bolivian Andes[☆]

Valérie Raevel[a,b,*], Fabien Anthelme[a,c,d], Rosa Isela Meneses[c,d], François Munoz[b,e]

[a] Institut de Recherche pour le Développement (IRD), UMR-AMAP, TA A-51/PS2, 34398 Montpellier Cedex 05, France
[b] French Institute of Pondicherry (IFP), 11 Saint Louis Street, 605001 Pondicherry, India
[c] Instituto de Ecología, Universidad Mayor San Andrés, Casilla 10077, Correo Central, La Paz, Bolivia
[d] Museo Nacional de Historia Natural, Herbario Nacional de Bolivia (LPB), Cota Cota, Casilla 10077 Correo Central, La Paz, Bolivia
[e] Université Grenoble Alpes, LECA, Laboratoire d'Écologie Alpine, CS 40700, 38058 Grenoble Cedex 9, France

ARTICLE INFO

Keywords:
Tropical Andean peatland
Giant cushion-plants
Graminoid and forb guilds
Emergent and understory plants
Between-species and intraspecific functional trait variation
Herbivore pressure

ABSTRACT

The functional trait composition of plant communities reflects the influence of ecological constraints on their assembly. In high-elevation peatlands of the Andes, multiple abiotic and biotic stresses are exerted on herbaceous plant communities. We hypothesized that dominant cushion-plant species found in these ecosystems strongly influence the dynamics of their subordinate plants by providing protection against external stress. To test the hypothesis, we addressed the variation of functional traits reflecting how subordinate plants invest in durable tissues and develop below and above the canopy of two cushion-plant species dominating the ecosystem, with varying intensity of herbivore pressure. We expected different functional composition of plants living below (understory stratum) and above (emergent stratum) the canopy, related to cushion-plant protection. In addition, we considered the functional composition of basic morphological guilds, forbs and graminoids, as they invest differently in plant development and respond differently to herbivore pressure. We found contrasted functional compositions among morphological guilds and according to the position relatively to cushion-plant canopy. These variations support a protective influence of cushion-plants on subordinate plants for both strata, with investment into less costly leaves and a larger stature when protection is greater, depending on cushion-plant species and cushion-plant height. External stress mainly affected emergent plants and led to different responses according to the guild. Graminoids showed avoidant strategy, while forbs shifted from avoidant to tolerant strategy with increasing herbivore pressure. Both between-species and intraspecific trait variations contributed to explain functional changes of guilds, but the relative contributions depended on morphological guild and stratum. A basic trade-off in physiological and morphological variability determined different relative contribution in forbs and graminoids. While functional ecology classically focuses on the composition of spatially delimited communities, we here underline the need to address separately the functional composition and assembly dynamics of distinct guilds making a community. In addition, our survey is the first to investigate the functional ecology of high-elevation tropical peatlands and provides novel insights into vegetation ecology in this stressful ecosystem.

1. Introduction

Understanding the role of biotic and abiotic constraints on species survival and coexistence is a central issue of community and functional ecology (Ackerly and Cornwell, 2007; McGill et al., 2006; Westoby and Wright, 2006). Alpine ecosystems are especially relevant to broach the issue, as they are subject to multiple and intense stresses like high solar radiation, cold, water limitation and herbivore pressure (Anthelme and Dangles, 2012; Körner, 2003). Interactions among plants also play a crucial role in the dynamics of these ecosystems (Callaway et al., 2002). Specifically, cushion-plants are common in cold and arid environments (Aubert et al., 2014) and can have protective influence on the

---

Abbreviations: H, Height; LDMC, Leaf Dry Matter Content; LW, leaf Length/leaf Width; DM, *Distichia muscoides*; OA, *Oxychloe andina*; HP, Herbivore Pressure; BTV and ITV, Between-species and Intraspecific Trait Variation; GWM, Group Weighted Mean; UG and EG, Understory and Emergent Graminoids; UF and EF, Understory and Emergent Forbs

[☆] This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author.

E-mail address: vraevel@gmail.com (V. Raevel).

https://doi.org/10.1016/j.ppees.2017.09.006
Received 21 December 2016; Received in revised form 15 August 2017; Accepted 28 September 2017

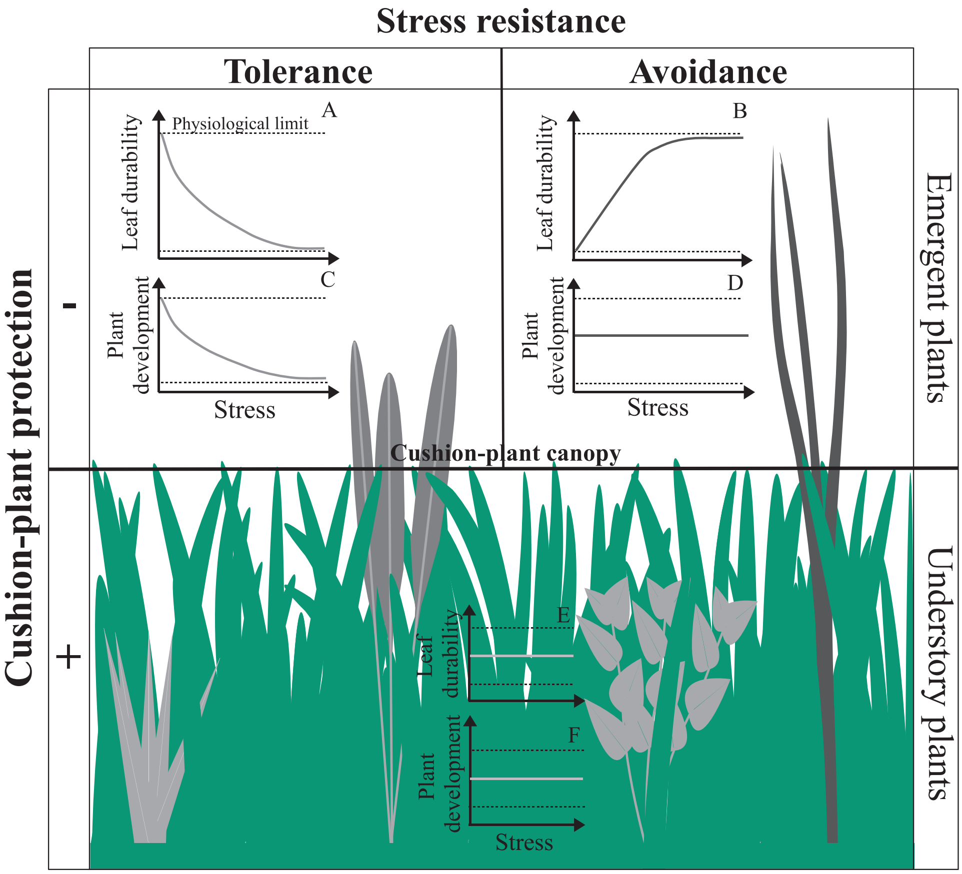


Fig. 1. Influence of environmental stresses on leaf durability (A & B for emergent and E for understory plants) and plant development (C & D for emergent and F for understory plants) in subordinate plant communities, depending on plant position relative to the cushion-plant canopy. The cushion-plant is represented in green, while the subordinate plants are in grey. The scale of grey represents a gradient in leaf durability investment. Changing abiotic or biotic conditions are expected to influence differently leaf functional strategies and plant development below and above the cushion-plant canopy. The stress gradient represented on abscissa can reflect the influence of abiotic (e.g., hydric and temperature stress) or biotic (e.g., herbivore pressure) factors. For understory plants (bottom part), protection by the cushion-plant should limit the influence of abiotic and biotic stresses, so that we expect less investment into leaf durability (lighter grey) than emergent plants, and that leaf durability (E) and plant development (F) should be less if not influenced by stress variation. Conversely, emergent plants benefit less from cushion-plant protection, and two alternative ways to survive under increasing stress are expected: stress avoidance (B & D) means increased investment into leaf structure and durability (more conservative strategy, darker grey), which limits the damages due to stress, while stress tolerance (A & C) means producing less durable leaves (lighter grey) that are more damaged but are less expensive to renew. In A, increasing stress entails less leaf durability, while in B increasing stress entails greater leaf durability, within physiological limits. In D, limited damages due to stress entail less influence of stress intensity on plant development. Conversely, plants in C invest more into tissue renewal, which means that plant development should decrease with stress, due to more frequent and more extensive damages. (Any colour code mentioned in the legend refers to the figure layout in the web version of this article.)

subordinate plants establishing and living within (Cavieres et al., 2014; Reid et al., 2010). They accumulate litter, increase water and nutrient availability (Anthelme and Dangles, 2012; Cavieres et al., 2007; Körner, 2003) and their protection can limit impact of grazing and trampling by herbivores (Acuña-Rodríguez et al., 2006). Previous surveys basically investigated how cushion-plants influence the assembly of subordinate plants in terms of taxonomic (Arroyo et al., 2003; Badano and Marquet, 2009; Cavieres et al., 2002; Sklenář, 2009) and phylogenetic composition (Butterfield et al., 2013), and determine long-term stability and diversity in high-elevation alpine ecosystems (Badano and Cavieres, 2006). However, the influence of cushion-plant on the functional composition of subordinate plants is little studied (Körner, 2003; Schöb et al., 2012). The diversity of functional traits among coexisting organisms provides insights into the ecological processes driving community dynamics (Albert et al., 2011; McGill et al., 2006). We then expect that the functional diversity of subordinate plants inhabiting cushion-plants reflect the combined influence of environmental stress and micro-environmental conditions (Violle and Jiang, 2009) generated by the cushion-plants. We thus investigated the functional diversity of basic physiological and morphological traits representing strategies of resource acquisition and spatial development (Westoby, 1998) of subordinate plants living in giant cushion-plants of high-Andean peatlands (bofedales, Squeo et al., 2006).

Figs. 1 and 2 summarize our hypotheses on how environmental constraints and cushion-plant protection interplay and determine the functional composition of subordinate plants. Of primary importance is the spatial organization of subordinate plants occupying different microhabitats within cushion-plants (Schöb et al., 2012). Cushion-plants in high-elevation ecosystems display compact architecture and limited vertical development, with clear canopy boundary, so that the environmental context differs below and above the canopy. Plants located below the cushion-plant canopy (i.e., understory plants) are expected to be more protected against external stress (Acuña-Rodríguez et al., 2006). Conversely, the plants located above the canopy (i.e., emergent plants) benefit less from this protection, as only the basis of the plant can be protected (Fig. 1, top). We then expect different functional trait values of subordinate plants according to their position below or above cushion-plant canopy (Fig. 1). Plants above the canopy subject to greater stress are expected to invest into more conservative and more durable leaves (Garnier et al., 2016; Grime, 2002). In addition, owing to the protection, the functional traits of plants below the canopy are expected to be less influenced by the variation of external stress.

For emergent plants, abiotic stress and herbivore pressure entail damages to stems and leaves, loss of invested resources, and reduced

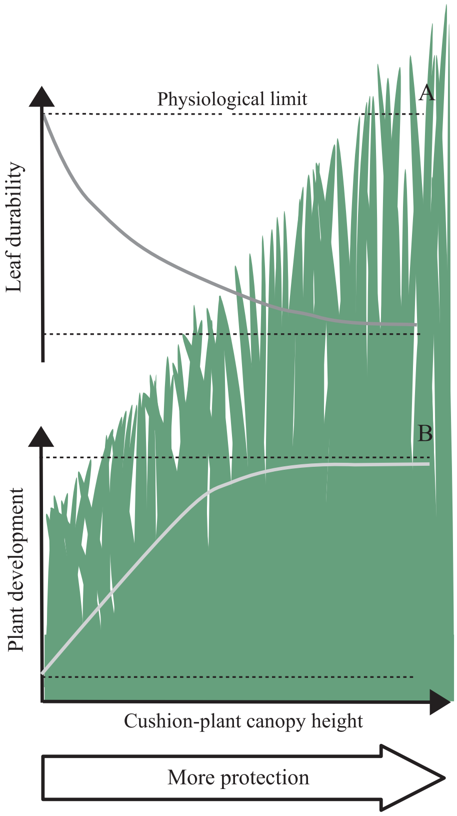


Fig. 2. Influence of cushion-plant canopy height on the leaf durability (A) and vegetative plant development (B) of subordinate plant species. For both understory and emergent plants, a taller cushion-plant is expected to protect more, so that investment in durable leaves is expected to decrease (A) while development of subordinate plants (B) is expected to increase. The cushion-plant is represented in green.

primary production (Grime, 1974). Two alternative strategies of resistance to such damages are possible. Avoidant species invest more into defenses through physiological and morphological adaptations, which allow greater leaf longevity, while tolerant species build cheaper and less durable tissues but need to renew them more often due to greater loss (Fig. 1, Briske, 1996; Del-Val and Crawley, 2005). We expect that avoidant emergent plants invest into more durable structure and conservative strategy when exposed to greater stress (Fig. 1A), while tolerant species cannot develop as much due to greater loss of tissues when stress increases (Figs. 1B and D). The protective influence of the cushion-plant, highly dependent on its morphology, is expected to increase with canopy height (Schöb et al., 2013), allowing less investment in durable leaves (Fig. 2A) and greater development of subordinate plants (Fig. 2B).

From this premise, we expected (i) more conservative strategies in emergent than in understory plants if the protective influence of cushion-plant predominates, (ii) more conservative strategies in stress-avoidant emergent plants vs. more acquisitive strategies in stress-tolerant emergent plants (Fig. 1), (iii) taller subordinate plants and less conservative strategies in taller cushion-plants for both emergent and understory plants (Fig. 2). We questioned whether and how the influence of cushion-plant protection could differ between two cushion-plant species dominating the high-elevation peatlands of the Bolivian Andes, *Distichia muscoides* Phil. and *Oxychloe andina* Nees & Meyen (Meneses et al., 2015; Munoz et al., 2014), which present different canopy height and substrate properties (Loza Herrera et al., 2015).

The influence of ecological processes on the functional composition of communities is classically tackled by analyzing how the community average trait values change with environmental conditions (Garnier et al., 2004). The variation can be due to trait variation between species (BTV) or within species (i.e., intraspecific, ITV). Greater trait variability across populations of a species along environmental gradients is expected to inflate the contribution of ITV, while the sorting of species with contrasted strategies is expected to increase the contribution of BTV. The relative contribution of ITV and BTV thus represents the extent to which environmental constraints filter species with different adaptations or entail intraspecific phenotypic variation across communities (Albert et al., 2011). In addition, guilds are groups of species displaying distinctive ecological strategies, and the assembly of guilds can differ depending on their specific attributes. For instance, forbs or graminoids are two basic morphological guilds of herbaceous plants with contrasting ways to explore space and to respond to stress (McIntyre et al., 1999). Graminoids have the capacity to elongate their leaves (Wilson and Roxburgh, 1994), which contributes to their spatial development and allows persistence and continuing development of leaves under grazing (Coughenour, 1985). We expect that different investment into leaf tissue and plant development of forbs and graminoids should influence their response to environmental stress and to cushion-plant protection. We thus analyzed separately the functional structure of the two guilds, and determined the contribution of BTV and ITV to the changing functional composition of guilds among subordinate communities. We tested our hypotheses of Figs. 1 and 2 by analyzing the variation of leaf durability and plant development for each morphological guild, depending on cushion-plant species (*Distichia muscoides* or *Oxychloe andina*), their canopy height, herbivore pressure and on the position of subordinate plants from cushion-plant canopy, i.e., understory or emergent.

2. Material and methods

2.1. Study area

High-elevation cushion-plant peatlands are found in the central Andes of Peru, Bolivia, Chile and Argentina. They are semi-aquatic ecosystems surrounded by a matrix of dry ecosystems (Ostria, 1987). They undergo intense abiotic stresses, i.e., low temperature with broad daily variations, daily frost or snow, seasonally water-saturated soils and intense solar radiation (Ruthsatz, 2012; Squeo et al., 2006).

The study area is the Huayna Potosi valley located in the Cordillera Real mountain range close to La Paz, Bolivia. Rainfall is concentrated from December to March (wet season): 410.4 mm with mean temperature of the period around 6.4 °C. Rainfall during the dry season from April to November reaches 184 mm, and the mean temperature of the period is around 4.5 °C (Loza Herrera et al., 2015). The Huayna Potosi valley is surrounded by glaciers, and includes numerous peatland ecosystems. These ecosystems are a key resource for domestic herbivores and are thus subject to intense herbivore pressure (Buttolph and Coppock, 2004; Dangles et al., 2014; Muñoz et al., 2015).

We selected four spatially isolated peatland sites above 4700 m of altitude, each of which was formed by a mosaic of cushion-plants dominated by *Distichia muscoides* Nees & Meyen (abbreviated as DM; Fig. 3A) and *Oxychloe andina* Phil. (abbreviated as OA; Fig. 3E). These cushion-plant species display distinctive substrate compaction, spatial development and ecophysiological properties, e.g., leaf nutrient contents (Loza Herrera et al., 2015), which represent distinct abiotic and biotic conditions for the subordinate plant communities (Fig. 3).

OA has longer spiny leaves and forms taller and less compact cushion-plants than DM (Loza Herrera et al., 2015). In this regard, we expected that OA cushion-plants could provide greater protection against herbivore pressure and/or cold stress (Ruthsatz, 2012).

2.2. Sampling of subordinate communities

We sampled in April 2015 at least twelve cushion-plants per peatland (54 in total), with equal proportion of OA and DM species. We selected cushion-plants occurring in comparable hydric conditions and

Fig. 3. *Distichia muscoides* Nees & Meyen (DM; A, B, C & D) and *Oxychloe andina* Phil. (OA; E, F, G & H), two Juncaceae forming dense cushion-plants in bofedal ecosystems, and some of their subordinate species. A: a DM cushion-plant; B: DM ramets; C: *Lachemilla diplophylla* (Diels) Rothm., an understory forb in DM cushion-plant; D: *Carex maritima* Gunnerus, an emergent graminoids in DM cushion-plant; E: a OA cushion-plant; F: OA leaves; G: *Carex maritima* Gunnerus, an emergent graminoids in OA cushion-plant; H: *Oritrophium limnophilum* (Sch.Bip.) Cuatrec., an understory forb in OA cushion-plant.

located far from peatland border. The sampled cushion-plants were large enough to allow laying a 1 m^2 quadrat at their center, without influence of cushion edge. The 1 m^2 quadrats were subdivided in sub-quadrats of 10 × 10 cm. In each quadrat, we visually estimated the total cover of all species making the subordinate plant community (Fig. 4). The species were identified according to Meneses et al. (2015) and belonged to two morphological guilds, i.e., graminoids and forbs (Wilson and Roxburgh, 1994).

2.3. Trait sampling and measurement

To test our hypotheses on subordinate plant community assembly (Figs. 1 and 2), we considered two sets of functional traits related to (i) resource acquisition strategies, and (ii) spatial development. First, the dry mass of a leaf divided by its water-saturated fresh mass (expressed in $mg.g^{-1}$, Pérez-Harguindeguy et al., 2013), LDMC, is a physiological trait related to photosynthesis activity, water budget, growth rate and durability (Weiher et al., 1999). Second, vegetative height (H in cm), is a morphological trait related to plant competitive ability for light interception (Westoby, 1998). We also considered the ratio of the leaf major (length) to minor axis (width), called LW (leaf length/leaf width), another morphological trait, to characterize leaf elongation (Niinemets et al., 2007). Because the stems of subordinate plants were very short, LW was expected to be an important index of the ability of

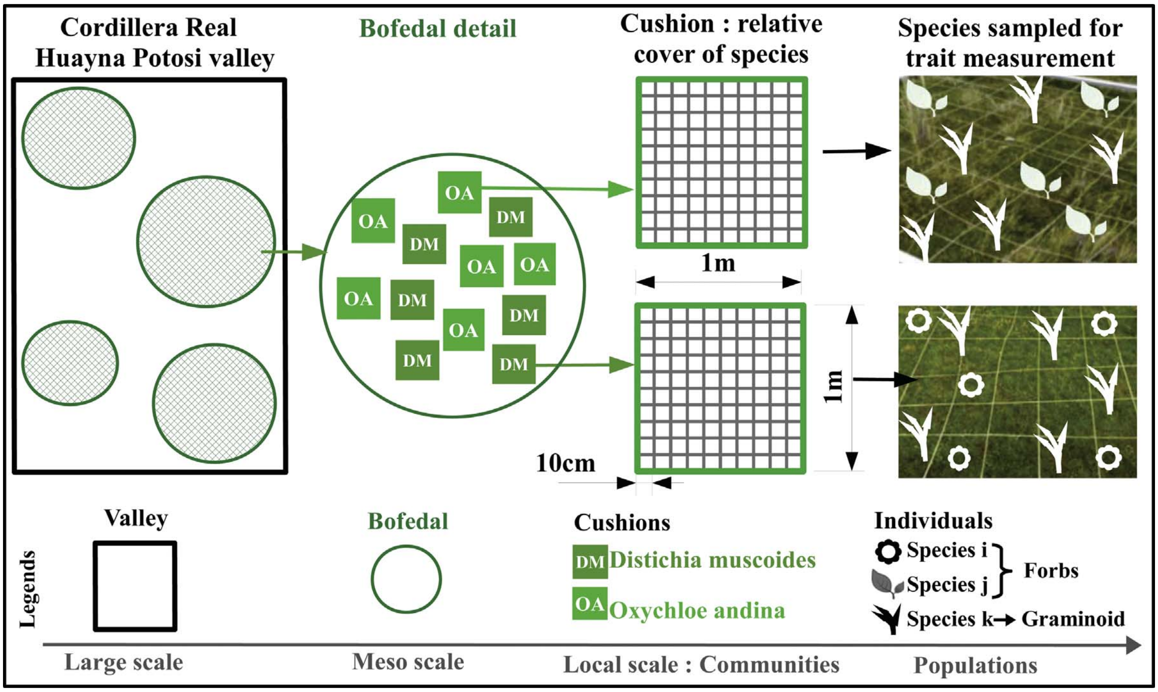


Fig. 4. Sampling scheme of subordinate plant communities. In each bofedal, we selected a balanced number of *Oxychloe andina* (OA) and *Distichia muscoides* (DM) cushion-plants, and measured the relative cover of subordinate plant species in 1m x 1m quadrats. For each of the fifteen species selected for trait measurement, and in each quadrat where it occurred, three entire individuals were collected for leaf trait analysis and the height of five individuals was measured.

plants to explore space and intercept light. Morphological traits like H and LW have often been related to response to disturbance including grazing (Cingolani et al., 2005; McIntyre et al., 1999). We performed trait sampling and measurement following the standardized protocols of Pérez-Harguindeguy et al. (2013). In each selected cushion-plant, functional traits were measured for both subordinate and cushion-plant species. Furthermore, we measured and averaged the cushion-plant canopy height at five random points in each quadrat, and found that canopy height was significantly lower for DM (1.5 cm ± 0.12) than for OA cushion-plants (3.8 cm ± 0.8, one-tailed Wilcoxon test, $P < 0.001$).

Based on preliminary inventories, we selected fifteen subordinate species for trait measurement (see Table A1), which were (i) present in both OA and DM cushion-plant species, (ii) belonged to the two basic guilds (graminoid and forb) and (iii) represented on average more than 85% cover of subordinate plant communities (Pakeman and Quested, 2007).

We measured the trait values of selected species in each quadrat where they occurred. We selected five and three individuals of each species per quadrat for height and leaf trait measurement, respectively. Sampling was performed after the wet season, during April 2015. Cooper et al. (2015) evidenced biomass production of *Distichia muscoides* all year round in Bolivia, with a peak during the extended austral summer between October and April. We selected only reproductively mature individuals at similar vegetative stage, which were not damaged by freezing or by herbivores. Only well-developed and non-senescent leaves were collected for leaf trait measurement (Pérez-Harguindeguy et al., 2013). In addition, we sampled as many leaves per individual (and at least 3 leaves) needed to get at least 0.02 g fresh material. Leaf elongation (LW) was measured on the fresh leaves sampled for LDMC measurement. For subsequent analyses, we averaged the trait values of each species in each sampled cushion-plant.

We subdivided the subordinate community in each cushion-plant into 4 ecological groups representing the combination of two basic morphological guilds (forbs and graminoids), and two vertical strata, representing the position of individuals from cushion-plant canopy (understory and emergent plants). A species in a given cushion-plant belonged to only one stratum, emergent or understory, but the species stratum could change among cushion-plants. For each group in each cushion-plant, we calculated the average of trait values of subordinate species weighted by their relative cover. We coined this average value Group Weighted Mean (GWM), by analogy with the Community Weighted Mean value calculated at community level (Garnier et al., 2004):

$$GWM_t(j) = \sum_{i=1}^{S} p_{ij}\, t_{ij} \tag{1}$$

where p_{ij} is the relative cover and t_{ij} is the value of trait t for species i in the group j, and S is the number of species of the group.

We thus calculated four GWM values per cushion-plant, namely, for emergent graminoids (EG), understory graminoids (UG), emergent forbs (EF), and understory forbs (UF). We then modeled the variation of GWM according to the biotic and abiotic conditions expected to drive the dynamics of ecological groups (Figs. 1 and 2).

2.4. Herbivore pressure

Intense herbivore pressure has been referred as one on the main threats in bofedal ecosystems (Salvador et al., 2014). During the dry season, the majority of herbivores rest and feed in the peatlands yielding biomass all year round (Buttolph and Coppock, 2004). We estimated dung density in each bofedal as a proxy of the residence time and feeding activity of herbivores (Howison et al., 2015; Whitworth-Hulse et al., 2016). Dung density has proved well related to herbivore presence (Von Müller et al., 2012), and shepherds did not collect dungs for fuel in our area (Meneses pers. com.). In each bofedal, we counted the dung clusters of cows, lamas, sheeps and alpacas in a transect 2 m wide and 100 m long. We defined three categories of dung clusters: small, medium and large. On average, large clusters were twenty times and medium clusters ten times larger than small clusters. The herbivore pressure (HP) in each bofedal was then calculated as the sum of clusters sampled along the transect, weighted by the relative size of clusters, i.e., weighted by 1, 10 and 20 for small, medium and large clusters, respectively. We thus obtained a quantitative index of herbivore pressure in each bofedal.

2.5. Analysis of GWM variation

First, we tested the variation in GWM values by performing ANOVA analysis and a post-hoc Tukey's HSD test. We expected that the two guilds had different GWM values because of distinctive morphological characteristics, and that cushion-plant protection from abiotic and biotic stresses led to lower LDMC of understory plants. Second, for each of the four groups taken separately, we built three linear models of the variation of GWM values across cushion-plants according to (i) the intensity of herbivore pressure (HP), (ii) cushion-plant species (OA or DM) and (iii) cushion-plant canopy height:

(a) For plant height, $GWM_H \sim (HP + HP^2)$ x Cushion-plant species + Cushion-plant canopy height
(b) For plant LDMC, $GWM_{LDMC} \sim (HP + HP^2)$ x Cushion-plant species + Cushion-plant canopy height + GWM_H
(c) For plant LW, $GWM_{LW} \sim (HP + HP^2)$ x Cushion-plant species + Cushion-plant canopy height + GWM_H

We analyzed the variation of GWM_H across cushion-plants (model a) depending on herbivore pressure (HP), cushion-plant species and cushion-plant canopy height, under the hypotheses that the height of plants varied or not with herbivore pressure depending on tolerance or avoidance strategy (Fig. 1), that the development and height depended on cushion-plant species, and that taller cushion-plants allowed greater development by providing more protection against external stress for both emergent and understory plants (Fig. 2). For GWM_{LDMC} (model b), we further included GWM_H as a predictor, under the hypothesis that taller emergent plants were subject to more abiotic stress and/or herbivore pressure, which both should entail larger LDMC. In model (c), GWM_{LW} (leaf elongation), a component of plant spatial development, was expected to vary in the same way as plant height, depending on herbivore pressure, cushion-plant species and canopy height. GWM_H was also a predictor in model (c), in order to assess whether taller plants could elongate more their leaves.

We expected different directions of trait variation with increasing stress for stress-avoidant and stress-tolerant strategies (Fig. 1). However, at group level, the relative proportion of individuals expressing one or the other strategy could vary along the gradient (Laliberté et al., 2012). Specifically, more importance of one strategy at an extreme of the gradient and of the other at the other extreme should yield a hump-shaped curve. In order to acknowledge this combined influence, we included both a linear and a quadratic term of herbivore pressure in each model (Albert et al., 2010b). We also included in the model an interaction term of herbivore pressure and cushion-plant species, to examine whether there was different influence of herbivore pressure between the two cushion-plant species.

We checked the validity of the models by analyzing the distribution of their residuals. For each model, we checked the normality (Shapiro-Wilk Normality test, $P > 0.05$) and the homogeneity (visually) of the residuals with the R package gvlma (Peña and Slate, 2006). In addition, non-independence of the residuals could arise when, for instance, unmeasured environmental variation among bofedales influenced trait variations and thus the variation of residuals in our models (Zuur et al., 2010). As suggested in Zuur et al. (2009), we checked the absence of such dependency in the residuals by analyzing the variation among bofedales of their average value, with ANOVA tests, and of their variance, with Levene's tests (function leveneTests of package heplots, Fox et al., 2009). All the P-values of these tests were higher than 0.05, indicating homogeneity of the variance and of the average residual values among bofedales. We could thus perform ANOVA tests (type 1) of the variance explained by the predictors.

2.6. Influence of intraspecific trait variation (ITV) in assembly of ecological groups

Changes in GWM values among cushion-plants could be related to (i) intraspecific trait variation of species in common (ITV), and/or (ii) between-species trait variation due to the presence of different species (BTV, between-species). For each guild in each stratum (i.e., each ecological group), we decomposed the influence of each predictor on the variation of GWM values in models (a), (b) and (c) into ITV and BTV contributions (Lepš et al., 2011; function traitflex.anova of package cati, Taudiere and Violle, 2016).

This method compared GWM values derived from the trait values of species measured in each cushion-plant (t_{ij} in Eq. (1)), and the GWM_BTV value derived from the average trait values of species across cushion-plants for a considered group (t_i replacing t_{ij} in Eq. (1)). For a species present in both understory and emergent strata, two t_i values were calculated, one for emergent and another for understory individuals. Significant variation of GWM_BTV across cushion-plants depending on environmental predictors indicated that different species with different average trait values were found in distinct environmental contexts, while significant variation of GWM could further encompass the contribution of intraspecific trait variation (ITV). Analyses of GWM_BTV, GWM and their difference thus allowed characterizing and testing the contributions of BTV and ITV to changing functional composition (Lepš et al., 2011).

All statistical analyses were performed with R 3.3.0 (Team, 2016).

3. Results

We found 29 subordinate plant species (12 graminoids and 17 forbs), distributed in 13 families and 24 genera (see Table A1). The most abundant families were Poaceae and Asteraceae. Quadrat richness ranged from 7 to 18 species. The total cover of subordinate species ranged from 4.5 to 46.5% of the quadrat and was greater in DM cushion-plants (Wilcoxon test, $P \leq 0.001$). The relative cover of graminoids represented from 4.2 to 97.5% (mean = 54.3%) of the total cover of subordinate species. Among the fifteen species selected for trait analyses, nine were forbs and six were graminoids. The selected forb and graminoid species represented on average 81.9% and 87.2% of the cover of forbs and graminoids per quadrat, respectively. We found the same relative cover per quadrat of the two guilds when considering only selected species or all the species occurring there (both Wilcoxon test, $P > 0.05$). The selected species were thus representative of the relative guild importance in subordinate plant communities.

3.1. Variation of subordinate plant traits at guild-level

We found broad variation of trait values between and within subordinate plant species, from 113.9 to 437.7 mg.g^{-1} for LDMC, 1.3–60.8 for LW, and 0.38–10.3 cm for height (Fig. A1). Both the between and within species variation contributed to the changing functional composition of subordinate plants among cushion-plants, namely, 73% and 21% for LDMC, 20% and 46% for height and 45% and 38% for LW, respectively (function decompCTRE in cati package, Taudiere and Violle, 2016). Graminoid species displayed higher LDMC and LW than forbs (Figs. 5A and B). Emergent graminoids displayed higher LDMC and larger LW than understory graminoids, while understory forbs did not differ from emergent forbs for these traits (Figs. 5A and B). Height of understory graminoids and forbs did not differ, while emergent graminoids were taller than emergent forbs (Fig. 5C).

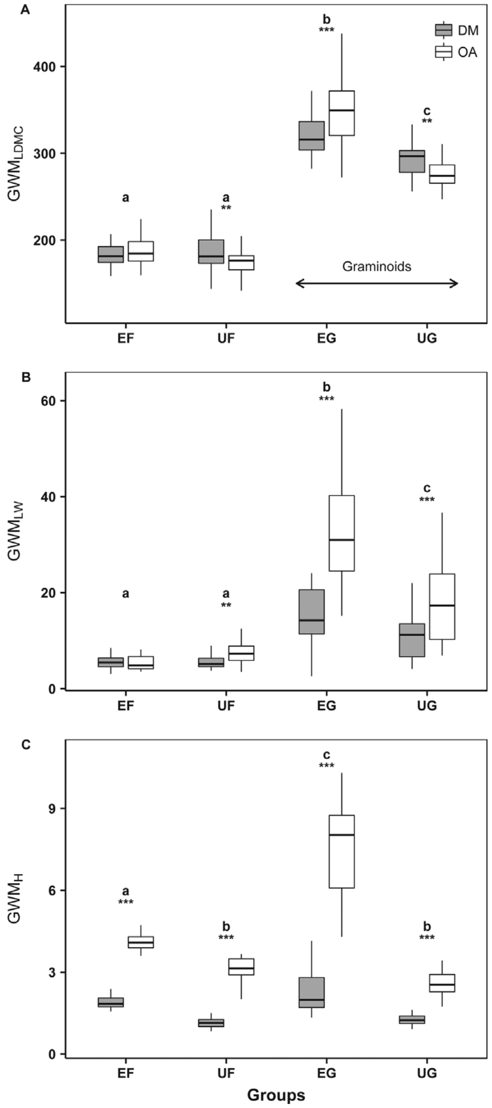


Fig. 5. Group Weighted Mean (GWM) of (A) leaf dry matter content (LDMC), (B) leaf elongation (LW) and (C) height in OA and DM cushion-plants. GWM trait values in DM and OA cushion-plants are presented in grey and white boxplots, respectively. Boxplots include the median value, the inter-quartile range (horizontal edges of the box) and an estimated 95% confidence interval (vertical lines). Different lower case letters indicate significant differences ($P < 0.01$) between groups based on post hoc Tukey's HSD test. Furthermore, the asterisks represent significant differences between OA and DM for a given group, with * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$ (results from Tables 1 A, B and A2).

For each of the emergent and understory strata, we found significant variation in functional properties of the two guilds across cushion-plants depending on herbivore pressure, subordinate plant height, cushion-plant species, and cushion-plant canopy height (Tables 1 A, B and A2). The selected predictors explained a large part of the total variation of subordinate plant traits: more than 83.5% for plant height (Table 1A), and from 30% to 76.5% for LDMC (Table 1B). For leaf elongation (LW), we found only significant changes for graminoids and the predictors explained more than 58.3% of the total variation for this guild (Table A2).

3.2. Influence of cushion-plant species and canopy height

When significant, the influence of cushion-plant species explained a

Table 1
Variation of Group Weighted Mean (GWM) values of plant height (A) and LDMC (B) depending on herbivore pressure (HP), cushion-plant species, GWM of subordinate plant height (GWM$_H$) for LDMC, and cushion canopy height. GWM values are calculated for each of the four groups, emergent forbs, emergent graminoids, understory forbs and understory graminoids. The analysis decomposes the contribution of between-species, intraspecific variation and their covariance. Each contribution is expressed in percentage of total trait variation; hence a total of 1 appears at bottom right of each table. The residuals quantify the part of variation not explained by the explanatory variables. "^{2}" represents a quadratic effect of herbivore pressure. The asterisks represent significant contributions in type 1 ANOVA, with the thresholds: * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$. Whenever there is significant effect of predictor on GWM total, the significant contributions to this effect are in bold. The sign of the coefficient of the significant predictors is provided within brackets for between-species, intraspecific and total variation, while the cushion-plant species with higher value is indicated when significant (OA: *Oxychloe andina*, DM:*Distichia muscoides*).

A.		FORBS				GRAMINOIDS			
		GWM of Height				GWM of Height			
	Explanatory variables	Between-species	Intraspecific	Covariation	Total	Between-species	Intraspecific	Covariation	Total
EMERGENT	HP	0.001	0.001	−0.002	0.000	0.015	0.001	0.006	**0.021*(+)**
	HP^2	**0.005*(−)**	0.001	0.005	**0.011***(−)**	**0.029*(−)**	0.002	−0.016	**0.015*(−)**
	Cushion-plant species	**0.075***(OA)**	**0.481***(OA)**	0.381	**0.937***(OA)**	**0.334***(OA)**	**0.088***(OA)**	0.343	**0.765***(OA)**
	Cushion canopy height	0.001	**0.008**(+)**	0.006	**0.015***(+)**	0.017	0.001	−0.009	0.009
	HP : Cushion	0.001	0.002	−0.002	0.000	**0.036*(OA)**	0.002	−0.016	**0.022**(OA)**
	HP^2 : Cushion	0.000	0.003	−0.002	0.001	0.015	0.003	−0.014	0.004
	Residuals	0.042	0.031	−0.038	0.035	0.279	0.038	−0.152	0.165
	Total	0.125	0.527	0.348	1	0.724	0.135	0.141	1
UNDERSTORY	HP	0.001	0.003	0.004	0.008	**0.014***(+)**	0.000	0.003	**0.018*(+)**
	HP^2	0.001	0.000	0.000	0.001	0.000	**0.053***(−)**	0.005	**0.058***(−)**
	Cushion-plant species	**0.212***(OA)**	**0.208***(OA)**	0.419	**0.839***(OA)**	0.000	**0.763***(OA)**	−0.014	**0.749***(OA)**
	Cushion canopy height	**0.017*(+)**	**0.004*(+)**	0.018	**0.039***(+)**	0.001	**0.066***(+)**	0.013	**0.079***(+)**
	HP : Cushion	0.005	0.000	0.002	0.007	0.002	0.005	0.005	**0.012*(OA)**
	HP^2 : Cushion	0.005	0.002	−0.007	0.001	0.000	0.002	−0.001	0.001
	Residuals	0.106	0.037	−0.038	0.105	0.025	0.072	−0.014	0.083
	Total	0.348	0.254	0.398	1	0.041	0.961	−0.002	1

B.		FORBS				GRAMINOIDS			
		GWM of LDMC				GWM of LDMC			
	Explanatory variables	Between-species	Intraspecific	Covariation	Total	Between-species	Intraspecific	Covariation	Total
EMERGENT	HP	0.014	**0.044*(+)**	0.050	**0.108**(+)**	**0.044**(+)**	**0.043**(+)**	0.087	**0.174***(+)**
	HP^2	0.017	**0.059**(−)**	0.064	**0.140**(−)**	**0.065***(−)**	0.007	−0.042	**0.029*(−)**
	Cushion-plant species	0.166***(OA)	0.092**(DM)	−0.247	0.011	**0.156***(OA)**	0.002	−0.035	**0.123***(OA)**
	GWM$_H$	0.029	0.001	0.010	0.040	**0.337***(+)**	0.001	0.034	**0.372***(+)**
	Cushion canopy height	**0.063*(+)**	0.012	−0.055	0.020	**0.024**(−)**	0.008	0.027	**0.060**(−)**
	HP : Cushion	0.020	**0.047*(OA)**	0.061	**0.127**(OA)**	0.000	0.006	−0.003	0.003
	HP^2 : Cushion	0.034	0.001	−0.012	0.023	0.006	0.018	−0.021	0.003
	Residuals	0.430	0.315	−0.213	0.531	0.149	0.170	−0.084	0.235
	Total	0.773	0.570	−0.343	1	0.783	0.254	−0.037	1
UNDERSTORY	HP	0.037*(+)	0.005	−0.027	0.015	0.036***(−)	0.097*(+)	−0.118	0.015
	HP ^{2}	0.031*(−)	0.001	−0.012	0.021	0.000	0.018	−0.005	0.013
	Cushion-plant species	**0.025*(DM)**	**0.034*(DM)**	0.058	**0.116**(DM)**	0.000	**0.171**(DM)**	0.011	**0.182**(DM)**
	GWM$_H$	0.004	0.007	0.011	0.022	0.012*(+)	0.004	−0.014	0.002
	Cushion canopy height	**0.093***(−)**	0.010	0.062	**0.165**(−)**	0.002	0.012	−0.010	0.004
	HP: Cushion	0.002	**0.060*(OA)**	0.020	**0.081*(OA)**	0.001	0.007	0.004	0.012
	HP^2: Cushion	0.008	0.001	0.004	0.013	0.001	0.055	0.016	0.072
	Residuals	0.223	0.436	−0.093	0.567	0.055	0.613	0.034	0.701
	Total	0.423	0.554	0.023	1	0.108	0.975	−0.083	1

large fraction of the total variation of GWM values, and was stronger for plant height and LW (Table 1A and A2) than for LDMC (< 20%, Table 1B). GWM$_H$ was higher in OA than in DM cushion-plants for the four ecological groups (Fig. 5C). Leaf elongation (LW) was also greater in OA cushion-plants, except for emergent forbs (Fig. 5B). LDMC was higher in DM than in OA for understory species and higher in OA than DM for emergent graminoids (Fig. 5A).

We also found influence of cushion-plant canopy height on subordinate plant traits, apart from the influence of cushion-plant species (type 1 ANOVA). Taller cushion-plants sheltered taller subordinate plants. However the height of cushion-plant canopy did not influence the GWM$_H$ of emergent graminoids (Table 1A). Lower GWM$_{LDMC}$ was observed in taller cushion-plants for emergent graminoids and understory forbs (Table 1B). Cushion-plant canopy height only influenced GWM$_{LW}$ of understory graminoids.

3.3. *Influence of herbivore pressure*

Herbivore pressure affected trait variation of the two guilds but mainly the emergent stratum. GWM$_H$ of understory graminoids and emergent forbs decreased slowly with herbivore pressure. GWM$_{LDMC}$ of emergent graminoids basically increased with herbivore pressure (predominant linear effect, Table 1B and Fig. 6B). GWM$_H$ of emergent graminoids and GWM$_{LDMC}$ of emergent forbs first increased and then decreased neatly at high herbivore pressure (negative quadratic effect, Fig. 6). Herbivore pressure affected more strongly the variation of GWM$_{LDMC}$ (between 20.3–46.9%; Table 1B) than the variation of plant height (less than 10%; Table 1A). Herbivore pressure did not affect the

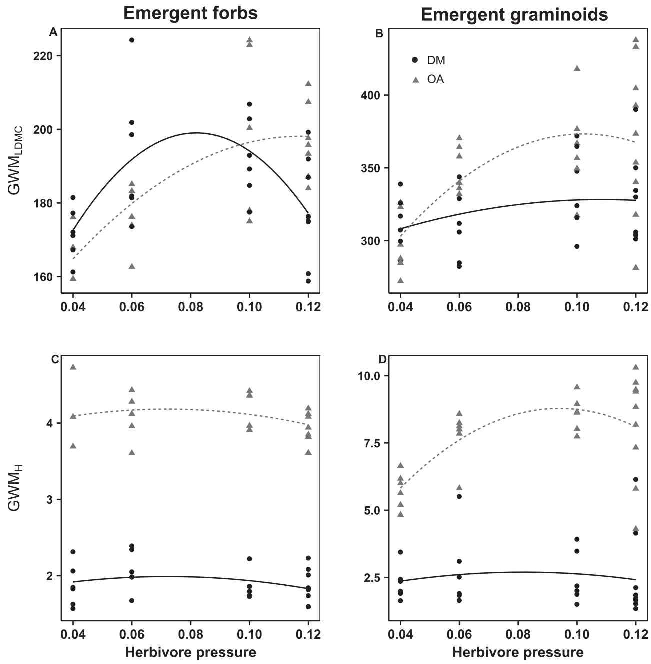


Fig. 6. Influence of herbivore pressure on Group Weighted Mean (GWM) values of emergent forbs (A & C) and graminoids (B & D), for leaf dry matter content (LDMC, top) and height (bottom). The figures represent the cases where the quadratic term of herbivore pressure has a significant effect on GWM variation, for LDMC (A, B) and height (C, D). Grey triangles and black dots represent GWM values in DM and OA, respectively. The fitted quadratic relationship of group-level trait with herbivore pressure is represented by two lines, plain for DM and dotted for OA (Table 1).

total variation of GWM$_{LDMC}$ of understory species. GWM$_{LW}$ of graminoids decreased linearly, while GWM$_{LW}$ of forbs did not vary with increasing herbivore pressure (Table A2). We found significant interaction of cushion-plant and herbivore pressure only for GWM$_{LDMC}$ of forbs and GWM$_H$ of graminoids (Table 1B).

3.4. *Influence of subordinate plant height on LDMC and LW*

GWM$_H$ was also used as a predictor of GWM$_{LDMC}$ and GWM$_{LW}$ models, as we expected that taller emergent plants should be more exposed to abiotic stress and expected that taller plants would elongate more their leaves. GWM$_H$ affected strongly and positively GWM$_{LDMC}$ (37.2%) and GWM$_{LW}$ (28.4%) of emergent graminoids.

3.5. *Contribution of between species (BTV) and intraspecific (ITV) trait variation*

Both between-species (BTV) and intraspecific (ITV) trait variation contributed to GWM variation (Tables 1 and A2). Variation of GWM ofgraminoids was more often due to BTV for emergent plants and to ITV for understory plants. For emergent graminoids, ITV also contributed to variation of GWM between cushion-plant species for H and LW, and to the response to herbivore pressure for LDMC and LW. For understory graminoids, BTV contributed to GWM variation with herbivore pressure, especially for H and LW (Tables 1A and A2). Contribution of BTV and ITV to the variation of GWM$_{LDMC}$ with herbivore pressure cancelled each other out and led to no variation of GWM of understory graminoids (Table 1B).

For both emergent and understory forbs, significant effects of predictors on GWM variation were more often due to ITV. However, for understory forbs, BTV contributed to the variation of GWM$_{LDMC}$ (Table 1B) and GWM$_H$ (Table 1A) between cushion-plant species and with cushion-plants canopy height. For emergent forbs, BTV contributed to GWM$_H$ between cushion-plant species and to the quadratic effect of herbivore pressure. As for understory graminoids, opposite contribution of BTV and ITV to the variation of GWM$_{LDMC}$ of emergent forbs between cushion-plant species cancelled each other out (Table 1B).

4. Discussion

We determined whether and how two dominant cushion-plant species of the bofedal ecosystem, *Distichia muscoides* (DM) and *Oxychloe andina* (OA), protect and influence the assembly of their subordinate plants. First, we found distinctive functional attributes of subordinate plants according to their position below or above the cushion-plant canopy, supporting a protective influence of cushion-plants against external stress (Tables 1 and A2). Understory plants invested into less dense and less conservative leaf tissues than emergent plants (Fig. 5). Second, two basic morphological guilds with distinct developmental capacities, forbs and graminoids, showed dissimilar functional responses to cushion-plant species and herbivore pressure, supporting the role of developmental strategies in stress resistance (Table 1, Hodgson et al., 1999; McIntyre et al., 1999). Varying influence of cushion-plant protection according to morphological guild and vertical stratum calls for investigating the functional composition of ecologically coherent groups within communities. Third, we found contributions of both between- and within-species functional trait variations to the changing composition of these groups between cushion-plant species and with herbivore pressure, although their relative influence depended on morphological guild and functional trait. An overall trade-off of physiological and morphological variability was found in response in the functional responses of guilds, namely, forbs had greater physiological and lower morphological variability, while graminoids had lower physiological but greater morphological variability. Our results underline a hierarchy in the response of subordinate plant communities to stress and cushion-plant protection, with significant variation of functional attributes among ecological groups, among species within ecological groups, and with phenotypic variability within species.

4.1. *Broad variation of functional traits related to environmental drivers*

We considered key functional traits related to resource acquisition and conservation on one hand (physiological LDMC), and plant development and spatial exploration on the other hand (morphological height and LW; Westoby, 1998). In general, higher LDMC is expected to reflect investment into denser and more durable leaf tissues. Conversely, less conservative strategies and faster growth rate are associated to lower LDMC (Garnier et al., 2016). We found remarkably broad variation of LDMC values among subordinate species, covering more than 50% of the worldwide range of this trait in global databases (Pérez-Harguindeguy et al., 2013), and exceeding the range of almost all existing studies at a local scale (Jung et al., 2010; Sides et al., 2014). This large range could indicate contrasted environmental filters in bofedal ecosystem (Schöb et al., 2012). However, the physiological LDMC trait was sensibly less variable within species (21%) than the morphological height and LW traits (46 and 38%; Albert et al., 2010b).

Cushion-plants characteristics, namely here species and height, and herbivore pressure explained more than 83.5% of the variation of subordinate plant height for each ecological group (Table 1A), while they explained with subordinate plant height on average 52.8% of the variation of LDMC (Table 1B) and LW (Table A2). These high percentages underline strong filtering of subordinate plants under the influence of key biotic and abiotic environmental factors and over a broad range of plant functional trait values.

4.2. *Cushion-plant protection below and above the canopy*

We hypothesized that cushion-plant protection entailed different functional composition of emergent and understory plants, as these plants undergo distinct abiotic and biotic conditions (Fig. 1). We indeed evidenced contrasted functional composition of subordinate plants depending on their relative position to cushion-plant canopy (Fig. 5 and Table 1). Lower LDMC in understory than in emergent plants implied less investment into durable and resistant leaf structure (Cornelissen et al., 2003) in this compartment due to reduced exposure to external stress. Furthermore, GWM_{LDMC} of understory plants was globally not affected by variations of external stress like herbivore pressure, while herbivore pressure influenced GWM_{LDMC} of emergent plants (Table 1B). These results support an overall protective influence of cushion-plants on understory plants. LDMC of understory plants was higher in DM than in OA, indicating that DM could offer less protection against external stress than OA cushion-plants. OA cushion-plants sheltered taller plants with more elongate and less durable leaves (lower LDMC) than DM (Tables 1 and A2). Because of its spiny leaves and taller canopy, OA could better protect against environmental stress and especially herbivore pressure while allowing plant development under the canopy. DM was clearly smaller than OA, grew basically more laterally than in height and had dense and intricate ramets, which could entail less protection to external stress and also limit subordinate plant development under canopy.

Apart from the influence of cushion-plant species, a taller cushion-plant could enhance protection, allowing plants to grow taller and to have less durable leaves for both strata (effect of cushion-plant canopy height in addition to cushion-plant species, Tables 1 and A2). The protective influence of cushion-plants did not only concern plants living under the canopy, as cushion-plant species could also influence the traits of emergent plants. Part of emergent plants was located below the canopy and thus the protective influence of cushion-plant could improve establishment and survival of individuals until adult stage (Cavieres et al., 2007; Körner, 2003). We thus evidenced a protective influence of cushion-plants depending on their characteristics (Schöb et al., 2013) for both strata. The contrasted patterns between the two strata, as demonstrated by the influence of herbivore pressure on emergent plants, reflected different exposure to external stress.

4.3. Guild-dependent responses to herbivore pressure

We also hypothesized that the two morphological guilds of perennial graminoids and forbs responded differently to stress and cushion-plant protection due to their distinct developmental capacities (Wilson and Roxburgh, 1994). Previous works showed different responses of the guilds in terms of relative abundance (McIntyre and Lavorel, 2001) and functional traits (McIntyre et al., 1999) along grazing gradients. Graminoids display long leaves with parallel ribs, which can more easily elongate and contribute to plant morphological variability. Graminoids are adapted to grazed ecosystems (Coughenour, 1985), and Adler et al. (2004) showed that graminoids of arid ecosystems in South America display avoidant strategies against herbivore pressure, with fibrous, less palatable and less digestible tissues. Consistent with this hypothesis, graminoids in bofedal ecosystem had higher LW and LDMC than forbs (Fig. 5). The main driver of the Group Weighted Mean variation of LDMC (GWM_{LDMC}) in emergent graminoids was their height (Table 1B). Taller emergent graminoids thus displayed higher LDMC, supporting stress avoidant strategy (Fig. 5). The taller was the species the stronger was the stress it could stand, and more avoidant species could grow taller. In addition, increasing GWM_{LDMC} of emerging graminoids with greater herbivore pressure was in line with the hypothesis of increasing avoidance (Figs. 1 and 6 B). Tallest *Festuca* graminoids in bofedal ecosystems also occur in surrounding arid and overgrazed environments and are typical stress avoiders with low palatability and high tissue density and durability (Muñoz et al., 2015). Conversely, forbs invested less in durable tissues (lower GWM_{LDMC} values) and showed less morphological variability through leaf elongation than graminoids (Fig. 5). Forbs emerged little from the canopy, and GWM_{LDMC} of emergent forbs first increased with herbivore pressure, as for emergent graminoids, but ultimately decreased at highest herbivore pressure, resulting in a hump-shaped curve (negative quadratic effect, Table 1B and Fig. 6A). This tendency could represent a shift from stress-avoidant to stress-tolerant strategy in forbs at highest herbivore pressure. Previous studies reported shifts from slower-growing, less palatable plants (grazing avoidant) in case of extensive and selective grazing, to faster-growing, more palatable plants (grazing tolerance) in case of intense and non-selective grazing (Laliberté et al., 2012; Westoby et al., 1999).

The Group Weighted Mean of height (GWM_H) also varied with herbivore pressure. Decreasing plant height under more intense grazing was reported at species and functional group levels in sub-humid mountain ecosystems (McIntyre and Lavorel, 2001; Whitworth-Hulse et al., 2016), and in other environmental contexts (Golodets et al., 2009; Gross et al., 2007). At group level, emergent and understory graminoids, and emergent forbs were smaller under higher herbivore pressure. However the height of emergent forbs globally decreased, while the height of emergent and understory graminoids first increased and then decreased with herbivore pressure, indicating again a guild-dependent response. Hump-shaped curve of some traits (e.g., LDMC, height; Fig. 6) along environmental gradients has already been reported in literature (Albert et al., 2010b; Garnier et al., 2016). For emergent graminoids, increasing GWM_{LDMC} with herbivore pressure suggested greater stress avoidance, but morphological traits, height (negative quadratic effect, Table 1A & Fig. 6) and LW (negative effect, Table A2) decreased at high pressure and indicated lesser development.

We also found interaction of herbivore pressure and cushion-plant species. For graminoids, plant height increased more in OA than in DM when herbivore pressure increased (Fig. 6D). Likewise, GWM_{LDMC} of forbs increased slightly more in OA then in DM with herbivore pressure (Table 1B). Subordinate plants thus tended to become more stress avoidant in OA than in DM cushion-plants when the herbivore pressure increased. Although the most active herbivores, camelids, can graze both cushion-plant species, the guilds responded differently to herbivore pressure depending on cushion-plant species (Fig. 6). Therefore, different cushion-plant characteristics can modulate the influence of herbivore pressure on subordinate plants.

4.4. Trade-off in leaf physiological and morphological variability

The variation of functional composition at group level could be due to changing trait values within species (ITV) or to changing trait values between species (BTV) (Kichenin et al., 2013; Lepš et al., 2011). While much emphasis was initially put on the role of BTV in functional ecology (Garnier et al., 2004), recent studies have underlined the importance of ITV to explain community assembly and species coexistence (Albert et al., 2011; Lepš et al., 2011; Violle et al., 2012). We found contrasted contributions of BTV and/or ITV depending on the functional trait, the position to the canopy, and the morphological guild. Our results underline a basic trade-off of leaf physiological and morphological variability among guilds (McIntyre et al., 1999) in emergent plants. Emergent forb species could adjust their leaf density in response to herbivore pressure and cushion-plant characteristics (greater ITV contribution), while the response of emergent graminoids was primarily related to between-species variations (greater BTV contribution; Table 1B). Conversely, graminoid species could adjust their leaf morphology through the LW ratio in response to external stress (greater ITV contribution), while forb species could less (Table A2). Morphological variability in graminoids species has been shown to represent an important avoidance strategy against grazing (Adler et al., 2004).

For emergent graminoids and for all traits, there was a turnover of species with different values, between cushion-plant species and with varying herbivore pressure. For understory graminoids, there was greater contribution of ITV to the influence of cushion-plant species and herbivore pressure, but it could basically reflect the variability of two widespread species, *Zameioscirpus muticus* Dhooge & Goetgh. and

Phylloscirpus deserticola (Phil.) Dhooge & Goetgh. These species could stay present in different environmental conditions but accommodate the varying constraints through phenotypic variability. Stronger environmental filtering for graminoids emerging from the cushion-plants could generate greater species turnover and BTV contribution, while protection within the cushion-plants could mitigate environmental heterogeneity and allow species persistence in different contexts with greater contribution of ITV (Albert et al., 2011). Despite a greater contribution of ITV, changes in species composition also contributed to the functional changes of understory graminoids, meaning that species filtering occurred even under the canopy. We also found noticeable opposite contributions of ITV and BTV to the variation of GWM_{LDMC} of understory graminoids with herbivore pressure, leading to unchanged functional composition of this group (Table 1B). Evidence for opposing responses of between-species and intraspecific trait variation in plant communities along environmental gradient have been also reported in previous works (Kichenin et al., 2013; Lepš et al., 2011). It underlines that between-species and intraspecific variation, as well as possible (negative or positive) covariation, can simultaneously control community-average trait values (Violle et al., 2012).

By contrast to emergent graminoids, ITV mostly contributed to the variation of GWM_{LDMC} of emergent forbs with herbivore pressure alone and in interaction with the cushion-plant species. Significant contribution of ITV to functional trait response under grazing was also reported by Howison et al. (2015). BTV still contributed to the variation of height of emergent forbs, indicating varying species composition of forbs between the two cushion-plant species and with herbivore pressure. We again found opposite contributions of ITV and BTV to the effect of cushion-plant species on the variation of LDMC for emergent forbs, leading to unchanged functional composition of this group (Table 1B). For understory forbs, both ITV and BTV could contribute to the influence of cushion-plant species depending on the trait, supporting both turnover of species and phenotypic variability of some species between cushion-plant species.

4.5. Limits and perspectives

We found non-monotonic variations of LDMC and height for some groups with herbivore pressure, but this trend would require further investigation, as there were only four distinct values of herbivore pressure, one per bofedal site. The response of groups to finer variation of herbivore pressure should thus be investigated to confirm the conclusions. We also lacked replication of bofedales for a given level of herbivore pressure, and the effect of herbivore pressure could then be confounded with other environmental variation among bofedales. However, we did not find a variation of the structure of the residuals depending on bofedales, so that there was no hidden influence of environmental variation across bofedales in our models of GWM variations (Zuur et al., 2010). The variation of selected functional traits across bofedales could thus be primarily related to herbivore pressure. Another limitation concerns the reliability of using dung cluster density as an index of herbivore pressure in cushion-plants. Even though previous studies showed that it could be related to actual herbivore activity (Von Müller et al., 2012), dung density can still remain inaccurate, so that alternative and direct measurements of herbivore activity could be considered to address the influence of herbivore pressure in greater details. In addition, an ongoing exclosure experiment should complement our study by providing a case of subordinate community assembly without herbivore pressure. Greater replication and additional measurements of varying environmental conditions across bofedales, such as soil characteristics or diurnal amplitude of soil temperature, should also be conducted in the future to address in further details the influence of abiotic vs. biotic conditions on subordinate plants assembly.

Although the selected traits showed rich and contrasted patterns of variation among groups and cushion-plants, they could not represent all the components of subordinate plant ecology. Reproduction is also likely to be greatly constrained by little pollinator availability and harsh conditions (García et al., 2016), so that mating systems, flowering and fruiting phenology of cushion and subordinate plants would deserve a specific survey. It is noteworthy that graminoid plants (including cushion-plants) were all wind-pollinated (Cyperaceae, Juncaceae and Poaceae), while forbs were more often entomophilous. Other functional traits like specific leaf area (SLA), seed mass, and leaf chemical traits (carbon, nitrogen and phosphorus) are known to respond to herbivore pressure (Golodets et al., 2009; McIntyre and Lavorel, 2001) and should also be analyzed in the bofedal ecosystem. In addition, bofedal ecosystems are highly fragmented and isolated in a matrix of hostile arid environment. In this context, dispersal and establishment success is likely to be greatly constrained, and analyzing dispersal strategies and gene flow would be needed to characterize possible metacommunity dynamics in bofedal ecosystems (see Bertin et al., 2015 in comparable ecosystems in Chile).

5. Conclusions

Our study provides original insights into the functional ecology of one of the most stressful and sensitive peatland ecosystems of the world (Salvador et al., 2014; Schittek, 2014) and underlines the key role of giant cushion-plant species dominating these ecosystems. It should allow better understanding the functioning of this ecosystem, crucial for water regulation and cattle resources in the area. Considerable functional trait variation of subordinate plants was found at both inter- and intraspecific level, reflecting the combined influence of environmental stress, cushion-plant protection, and guild-specific dynamics. Strong ecological segregation of guilds underlines distinctive responses to micro-environmental conditions. In this regard, the outcome of cushion-plant protection, intrinsically related to cushion-plant characteristics (Schöb et al., 2013), appears to be highly guild-dependent and can be altered in a context of highly grazed ecosystems (Smit et al., 2009).

While functional ecology classically investigates the composition of complete communities in delimited spatial units (McGill et al., 2006), our study shows that ecological groups sharing specific biological attributes and/or occurring in different micro-environmental conditions can have distinct assembly dynamics and functional responses within and among communities (Lavorel et al., 1997). Pooling these groups together in communities can blur the signature of their distinct functional responses (Albert et al., 2010a). Our results thus call for a decomposition of assembly dynamics within communities, by analyzing group-level statistics such as the GWM metric proposed here. Apart from the context of guild-dependent dynamics in plant communities of high-elevation peatlands, our approach can be valuable for addressing plant community ecology in a broad range of ecosystems.

Acknowledgements

This work was funded by the Fond Français pour l'Environnement Mondial (FFEM) and the Fondation pour la Recherche sur la Biodiversité (FRB) (Modeling BIOdiversity and land use interactions under changing glacial water availability in Tropical High Andean Wetlands – BIOTHAW, AAP-SCEN-2011-II). We thank the National Herbarium of Bolivia (LPB) and the Institute of Ecology of the university Mayor San Andres (UMSA) in Bolivia for their logistic support and Ariel Lliully and Susi Loza Herrera for their assistance during field work. We finally thank the editor and the anonymous reviewers for their helpful comments and recommendations.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.ppees.2017.09.006.

References

Ackerly, D.D., Cornwell, W.K., 2007. A trait-based approach to community assembly: partitioning of species trait values into within- and among-community components. Ecol. Lett. 10, 135–145.

Acuña-Rodríguez, I.S., Cavieres, L.A., Gianoli, E., 2006. Nurse effect in seedling establishment: facilitation and tolerance to damage in the Andes of central Chile. Rev. Chil. Hist. Nat. 79, 329–336.

Adler, P.B., Milchunas, D.G., Lauenroth, W.K., Sala, O.E., Burke, I.C., 2004. Functional traits of graminoids in semi-arid steppes: a test of grazing histories. J. Appl. Ecol. 41, 653–663.

Albert, C.H., Thuiller, W., Yoccoz, N.G., Douzet, R., Aubert, S., Lavorel, S., 2010a. A multi-trait approach reveals the structure and the relative importance of intra- vs. interspecific variability in plant traits. Funct. Ecol. 24, 1192–1201.

Albert, C.H., Thuiller, W., Yoccoz, N.G., Soudant, A., Boucher, F., Saccone, P., Lavorel, S., 2010b. Intraspecific functional variability: extent, structure and sources of variation. J. Ecol. 98, 604–613.

Albert, C.H., Grassein, F., Schurr, F.M., Vieilledent, G., Violle, C., 2011. When and how should intraspecific variability be considered in trait-based plant ecology? Persp. Plant Ecol. Evol. Syst. 13, 217–225.

Anthelme, F., Dangles, O., 2012. Plant-plant interactions in tropical alpine environments. Perspectives in Plant Ecology. Evol. Syst. 14, 363–372.

Arroyo, M.T.K., Cavieres, L.A., Peñaloza, A., Arroyo-Kalin, M.A., 2003. Positive associations between the cushion plant Azorella monantha (Apiaceae) and alpine plant species in the Chilean Patagonian Andes. Plant Ecol. 169, 121–129.

Aubert, S., Boucher, F., Lavergne, S., Renaud, J., Choler, P., 2014. 1914-2014: A revised worldwide catalogue of cushion plants 100 years after Hauri and Schröter. Alp. Bot. 124, 59–70.

Badano, E.I., Cavieres, L.A., 2006. Impacts of ecosystem engineers on community attributes: effects of cushion plants at different elevations of the Chilean Andes. Divers. Distrib. 12, 388–396.

Badano, E.I., Marquet, P.A., 2009. Biogenic habitat creation affects biomass?diversity relationships in plant communities. Perspectives in Plant Ecology. Evol. Syst. 11, 191–201.

Bertin, A., Alvarez, E., Gouin, N., Gianoli, E., Montecinos, S., Lek, S., Gascoin, S., Lhermitte, S., 2015. Effects of wind-driven spatial structure and environmental heterogeneity on high-altitude wetland macroinvertebrate assemblages with contrasting dispersal modes. Freshwater Biol. 60, 297–310.

Briske, D., 1996. Strategies of Plant Survival in Grazed Systems: a Functional Interpretation. The Ecology and Management of Grazing Systems. CAB International, Wallingford, pp. 37–67.

Butterfield, B.J., Cavieres, L.A., Callaway, R.M., Cook, B.J., Kikvidze, Z., Lortie, C.J., Michalet, R., Pugnaire, F.I., Schöb, C., Xiao, S., Zaitchek, B., Anthelme, F., Björk, R.G., Dickinson, K., Gavilán, R., Kanka, R., Maalouf, J.P., Noroozi, J., Parajuli, R., Phoenix, G.K., Reid, A., Ridenour, W., Rixen, C., Wipf, S., Zhao, L., Brooker, R.W., 2013. Alpine cushion plants inhibit the loss of phylogenetic diversity in severe environments. Ecol. Lett. 16, 478–486.

Buttolph, L.P., Coppock, D.L., 2004. Influence of deferred grazing on vegetation dynamics and livestock productivity in an Andean pastoral system. J. Appl. Ecol. 41, 664–674.

Callaway, R.M., Brooker, R., Choler, P., Kikvidze, Z., Lortie, C.J., Michalet, R., Paolini, L., Pugnaire, F.I., Newingham, B., Aschehoug, E.T., 2002. Positive interactions among alpine plants increase with stress. Nature 417, 844–848.

Cavieres, L., Arroyo, M.T.K., Peñaloza, A., Molina-Montenegro, M., Torres, C., 2002. Nurse effect of Bolax gummifera cushion plants in the alpine vegetation of the Chilean Patagonian Andes. J. Veg. Sci. 13, 547–554.

Cavieres, L.A., Badano, E.I., Sierra-Almeida, A., Molina-Montenegro, M.A., 2007. Microclimatic modifications of cushion plants and their consequences for seedling survival of native and non-native herbaceous species in the high andes of central Chile. Arctic, Antarctic Alp. Res. 39, 229–236.

Cavieres, L.A., Brooker, R.W., Butterfield, B.J., Cook, B.J., Kikvidze, Z., Lortie, C.J., Michalet, R., Pugnaire, F.I., Schöb, C., Xiao, S., Anthelme, F., Björk, R.G., Dickinson, K.J.M., Cranston, B.H., Gavilán, R., Gutiérrez-Girón, A., Kanka, R., Maalouf, J.-P., Mark, A.F., Noroozi, J., Parajuli, R., Phoenix, G.K., Reid, A.M., Ridenour, W.M., Rixen, C., Wipf, S., Zhao, L., Escudero, A., Zaitchik, B.F., Lingua, E., Aschehoug, E.T., Callaway, R.M., 2014. Facilitative plant interactions and climate simultaneously drive alpine plant diversity. Ecol. Lett. 17, 193–202.

Cingolani, A.M., Posse, G., Collantes, M.B., 2005. Plant functional traits, herbivore selectivity and response to sheep grazing in Patagonian steppe grasslands. J. Appl. Ecol. 42, 50–59.

Cooper, D.J., Kaczynski, K., Slayback, D., Yager, K., 2015. Growth and organic carbon production in peatlands dominated by distichia muscoides, Bolivia, S outh america. Arctic, Antarctic Alp. Res. 47, 505–510.

Cornelissen, J.H.C., Lavorel, S., Garnier, E., Diaz, S., Buchmann, N., Gurvich, D.E., Reich, P.B., ter Steege, H., Morgan, H.D., van der Heijden, M.G.A., Pausas, J.G., Poorter, H., 2003. A handbook of protocols for standardised and easy measurement of plant functional traits worldwide. Aust. J. Bot. 51, 335–380.

Coughenour, M.B., 1985. Graminoid responses to grazing by large herbivores: adaptations, exaptations, and interacting processes. Ann. Mo. Bot. Gard 852–863.

Dangles, O., Meneses, R.I., Anthelme, F., 2014. BIOTHAW: Un proyecto multidisciplinario que propone un marco metodologico para el estudio de los bofedales altoandinos en un contexto de cambio climatico. Ecol. Bolivia 49, 6–13.

Del-Val, E., Crawley, M.J., 2005. Are grazing increaser species better tolerators than decreasers? An experimental assessment of defoliation tolerance in eight British grassland species. J. Ecol. 93, 1005–1016.

Fox, J., Friendly, M., Monette, G., 2009. Visualizing hypothesis tests in multivariate linear models: the heplots package for R. Comput. Stat. 24, 233–246.

García, M.C., Bader, M.Y., Cavieres, L.A., 2016. Facilitation consequences for reproduction of the benefactor cushion plant Laretia acaulis along an elevational gradient: costs or benefits? Oikos 125, 434–442.

Garnier, E., Cortez, J., Billes, G., Navas, M.L., Roumet, C., Debussche, M., Laurent, G., Blanchard, A., Aubry, D., Bellmann, A., Neill, C., Toussaint, J.P., 2004. Plant functional markers capture ecosystem properties during secondary succession. Ecology 85, 2630–2637.

Garnier, E., Navas, M.-L., Grigulis, K., 2016. Plant Functional Diversity: Organism Traits, Community Structure, and Ecosystem Properties. Oxford University Press.

Golodets, C., Sternberg, M., Kigel, J., 2009. A community-level test of the leaf-height-seed ecology strategy scheme in relation to grazing conditions. J. Veg. Sci. 20, 392–402.

Grime, J.P., 1974. Vegetation classification by reference to strategies. Nature 250, 26–31.

Grime, J.P., 2002. Plant Strategies, Vegetation Processes, and Ecosystem Properties. John Willey & Sons LTD, New York, USA.

Gross, N., Suding, K.N., Lavorel, S., 2007. Leaf dry matter content and lateral spread predict response to land use change for six subalpine grassland species. J. Veg. Sci. 18, 289–300.

Hodgson, J.G., Wilson, P.J., Hunt, R., Grime, J.P., Thompson, K., 1999. Allocating C-S-R plant functional types: a soft approach to a hard problem. Oikos 85, 282–294.

Howison, R.A., Olff, H., Steever, R., Smit, C., 2015. Large herbivores change the direction of interactions within plant communities along a salt marsh stress gradient. J. Veg. Sci. 26, 1159–1170.

Jung, V., Violle, C., Mondy, C., Hoffmann, L., Muller, S., 2010. Intraspecific variability and trait-based community assembly. J. Ecol. 98, 1134–1140.

Körner, C., 2003. Alpine Plant Life: Functional Plant Ecology of High Mountain Ecosystems, 2nd ed. Springer Science & Business Media.

Kichenin, E., Wardle, D.A., Peltzer, D.A., Morse, C.W., Freschet, G.T., 2013. Contrasting effects of plant inter- and intraspecific variation on community-level trait measures along an environmental gradient. Funct. Ecol. 27, 1254–1261.

Laliberté, E., Shipley, B., Norton, D.A., Scott, D., 2012. Which plant traits determine abundance under long-term shifts in soil resource availability and grazing intensity? J. Ecol. 100, 662–677.

Lavorel, S., McIntyre, S., Landsberg, J., Forbes, T.D.A., 1997. Plant functional classifications: from general groups to specific groups based on response to disturbance. Trends Ecol. Evol. 12, 474–478.

Lepš, J., de Bello, F., Šmilauer, P., Doležal, J., 2011. Community trait response to environment: disentangling species turnover vs intraspecific trait variability effects. Ecography 34, 856–863.

Loza Herrera, S., Meneses, R., Anthelme, F., 2015. Comunidades vegetales de los bofedales de la Cordillera Real (Bolivia) bajo el calentamiento global. Ecol. Bolivia. 50, 39–56.

McGill, B.J., Enquist, B.J., Weiher, E., Westoby, M., 2006. Rebuilding community ecology from functional traits. Trends Ecol. Evol. 21, 178–185.

McIntyre, S., Lavorel, S., 2001. Livestock grazing in subtropical pastures: steps in the analysis of attribute response and plant functional types. J. Ecol. 89, 209–226.

McIntyre, S., Lavorel, S., Landsberg, J., Forbes, T.D.A., 1999. Disturbance response in vegetation −towards a global perspective on functional traits. J. Veg. Sci. 10, 621–630.

Meneses, R., Beck, S., Anthelme, F., 2015. La Cordillera Real Y Sus Plantas.

Muñoz, M.A., Faz, A., Acosta, J.A., Martínez-Martínez, S., Zornoza, R., 2015. Effect of South American grazing camelids on soil fertility and vegetation at the Bolivian Andean grasslands. Agriculture. Agric. Ecosyst. Environ. 207, 203–210.

Munoz, F., Anthelme, F., Raevel, V., 2014. Procesos ecológicos a múltiples escalas que afectan a las dinámicas de comunidades de plantas en los humedales altoandinos de Bolivia – Multi-scale ecological processes driving plant community dynamics in high-elevation peatlands of Bolivia. Ecol. Bolivia 49, 104–119.

Niinemets, Ü., Portsmuth, A., Tobias, M., 2007. Leaf shape and venation pattern alter the support investments within leaf lamina in temperate species: a neglected source of leaf physiological differentiation? Funct. Ecol. 21, 28–40.

Ostria, C., 1987. La végétation d'une vallée alto-andine (Hichu kkota, cordillère orientale, bolivie). I- données générales. bulletin de la société botanique de France. Lett. Bot. 134, 385–397.

Pérez-Harguindeguy, N., Diaz, S., Garnier, E., Lavorel, S., Poorter, H., Jaureguiberry, P., Bret-Harte, M.S., Cornwell, W.K., Craine, J.M., Gurvich, D.E., 2013. New handbook for standardised measurement of plant functional traits worldwide. Aust. J. Bot. 61, 167–234.

Pakeman, R.J., Quested, H.M., 2007. Sampling plant functional traits: what proportion of the species need to be measured? Appl. Veg. Sci. 10, 91–96.

Peña, E.A., Slate, E.H., 2006. Global validation of linear model assumptions. J. Am. Stat. Assoc. 101, 341–354.

Reid, A.M., Lamarque, L.J., Lortie, C.J., 2010. A systematic review of the recent ecological literature on cushion plants: champions of plant facilitation. Web Ecol. 10, 44–49.

Ruthsatz, B., 2012. Vegetacion y ecologia de los bofedales altoandinos de Bolivia. Phytocoenologia 42, 133–179.

Salvador, F., Monerris, J., Rochefort, L., 2014. Peatlands of the Peruvian Puna ecoregion: types, characteristics and disturbance. Mires Peat 15, 1–17.

Schöb, C., Butterfield, B.J., Pugnaire, F.I., 2012. Foundation species influence trait-based community assembly. New Phytol. 196, 824–834.

Schöb, C., Armas, C., Guler, M., Prieto, I., Pugnaire, F.I., 2013. Variability in functional traits mediates plant interactions along stress gradients. J. Ecol. 101, 753–762.

Schittek, K., 2014. Cushion peatlands in the high Andes of northwestern Argentina as archives for palaeoenvironmental research. Dissertationes Botanicae. Schweizerbart Science Publishers, pp. 176.

Sides, C.B., Enquist, B.J., Ebersole, J.J., Smith, M.N., Henderson, A.N., Sloat, L.L., 2014. Revisiting Darwin's hypothesis: does greater intraspecific variability increase species'

ecological breadth? Am. J. Bot. 101, 56–62.
Sklenář, P., 2009. Presence of cushion plants increases community diversity in the high equatorial Andes. Flora − Morphology Distribution. Funct. Ecol. Plants 204, 270–277.
Smit, C., Rietkerk, M., Wassen, M.J., 2009. Inclusion of biotic stress (consumer pressure) alters predictions from the stress gradient hypothesis. J. Ecol. 97, 1215–1219.
Squeo, F.A., Warner, B.G., Aravena, R., Espinoza, D., 2006. Bofedales: high altitude peatlands of the central Andes. Rev. Chil. Hist. Nat. 79, 245–255.
Taudiere, A., Violle, C., 2016. cati: an R package using functional traits to detect and quantify multi-level community assembly processes. Ecography 39, 699–708.
R Core Team, 2016. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.
Violle, C., Jiang, L., 2009. Towards a trait-based quantification of species niche. J. Plant Ecol.-Uk 2, 87–93.
Violle, C., Enquist, B.J., McGill, B.J., Jiang, L., Albert, C.H., Hulshof, C., Jung, V., Messier, J., 2012. The return of the variance: intraspecific variability in community ecology. Trends Ecol. Evol. 27, 244–252.
Von Müller, A.R., Cingolani, A.M., Vaieretti, M.V., Renison, D., 2012. Estimación de carga bovina localizada a partir de frecuencia de deposiciones en un pastizal de montaña. (Estimation of localized cattle stocking rate from dung frequency in a mountain grassland.). Ecol. Aust. 22, 178–187.
Weiher, E., van der Werf, A., Thompson, K., Roderick, M., Garnier, E., Eriksson, O., 1999. Challenging Theophrastus: a common core list of plant traits for functional ecology. J. Veg. Sci. 10, 609–620.
Westoby, M., Wright, I.J., 2006. Land-plant ecology on the basis of functional traits. Trends Ecol. Evol. 21, 261–268.
Westoby, M., Eldridge, D., Freudenberger, D., 1999. The LHS strategy scheme in relation to grazing and fire. In: Proceedings of the VIth International Rangeland Congress. International Rangeland Congress Townsville, AU. pp. 893–896.
Westoby, M., 1998. A leaf-height-seed (LHS) plant ecology strategy scheme. Plant Soil 199, 213–227.
Whitworth-Hulse, J.I., Cingolani, A.M., Zeballos, S.R., Poca, M., Gurvich, D.E., 2016. Does grazing induce intraspecific trait variation in plants from a sub-humid mountain ecosystem? Aust. Ecol. 41, 745–755.
Wilson, J.B., Roxburgh, S.H., 1994. A demonstration of guild-based assembly rules for a plant community, and determination of intrinsic guilds. Oikos 69, 267–276.
Zuur, A.F., Ieno, E.N., Walker, N.J., Saveliev, A.A., Smith, G.M., 2009. Mixed Effects Models and Extensions in Ecology with R. Springer.
Zuur, A.F., Ieno, E.N., Elphick, C.S., 2010. A protocol for data exploration to avoid common statistical problems. Methods Ecol. Evol. 1, 3–14.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 115–124

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Last millennial environmental dynamics in the western Peruvian Andes inferred from the development of a cushion-plant peat hillock☆

Karsten Schittek[a,*], Markus Forbriger[a], Dominik Berg[b], Jonathan Hense[c], Frank Schäbitz[b], Bernhard Eitel[a]

[a] *Institute of Geography, Universität Heidelberg, Im Neuenheimer Feld 348, 69120 Heidelberg, Germany*
[b] *Institute for Geography Education, Universität zu Köln, Gronewaldstr. 2, 50931 Köln, Germany*
[c] *Nees Institute, Department of Biology, Universität Bonn, Meckenheimer Allee 170, 53115 Bonn, Germany*

ARTICLE INFO

Keywords:
Andean peatlands
Cushion plants
Palaeoecology
Late Intermediate Period
Little Ice Age
Palynology

ABSTRACT

For the first time, an isolated cushion-plant peat hillock was investigated, a so far neglected feature of high-Andean spring ecosystems. These small hillocks typically cluster around springs within the upper catchment areas of larger cushion-plant peatlands at altitudes ranging from 4000 m to 5000 m a.s.l.

The size of the investigated peat hillock, located within the Río Viscas catchment area (Lucanas province, District of Ayacucho) at 4250 m a.s.l., is relatively small (about 10 m in diameter). Due to its dome-shaped and densely green habitus, it overlooks the surrounding vegetation by about 1–2 m and stands out by its color. Terrestrial laser scanning (TLS) techniques are used in order to provide insights into the spatial extension of the *Distichia muscoides*-dominated vegetation stand.

For the reconstruction of environmental dynamics during the past millennium, plant micro-/macrofossil analyses and total carbon/total nitrogen measurements were applied. Based on radiocarbon dating, the peat archive provides a chronology for the past 1050 years. We interpret phases of relatively high abundances of Poaceae pollen in our record as an expansion of Andean grasslands during humid phases. Drier conditions are indicated by a decrease of Poaceae pollen and higher abundances of Asteraceae pollen. The results reflect significant climate oscillations and provide evidence for a sustained dry phase between AD 900 and AD 1100. A more humid and cooler phase prevailed from around AD 1300 to AD 1825, during the Little Ice Age.

Our data provide evidence that such a spatially defined peat-accumulating ecosystem, as represented by the studied peat hillock, is capable to survive pronounced climatic oscillations as long as it does not lose its protective cushion-plant surface. As peat hillocks are heavily affected by grazing, multitemporal studies should be carried out to document changes and to provide new insights into adaption strategies of vegetation to changing environmental conditions.

1. Introduction

The central Andes are characterized by a great variety of landscapes, which is expressed in a diverse flora and fauna. The high-mountain landscape between the Eastern and Western Cordilleras, which commonly is referred to as the “Altiplano” or “Puna”, includes broad continuous, 3500–4100 m a.s.l. high plains. In these high-mountainous areas, the Andean flora is exposed to exceptionally harsh environmental conditions, including high solar radiation, pronounced diurnal temperature variation with frequent frosts, a prolonged dry season and low oxygen concentration (Schittek et al., 2012). Some plants show specific morphological adaptions, which facilitate the survival under these conditions. Impressive representatives of the typical, highly adapted plant growth-forms in high-altitude ecosystems of South America are the cushion plants. Their closely packed shoots with very short internodes grow so densely that they can form extensive, stable mats, ranging in shape from almost flat to hemispherical. The compact architecture protects them from strong winds and reduces the risk of water shortage (Sklenář, 2009).

Nevertheless, the occurrence of cushion plants is not necessarily

---

Abbreviations: TLS, terrestrial laser scanning; SASM, South American Summer Monsoon; TC, total carbon; TN, total nitrogen; MCA, medieval climate anomaly; LIA, little ice age; LIP, late intermediate period

☆ This article is part of a special issue entitled Alpine and arctic plant communities: a worldwide perspective published at the journal Perspectives in Plant Ecology, Evolution and Systematics 30C.

[*] Corresponding author.

E-mail address: schittek@uni-heidelberg.de (K. Schittek).

http://dx.doi.org/10.1016/j.ppees.2017.09.002
Received 8 February 2017; Received in revised form 15 August 2017; Accepted 8 September 2017

restricted to desert-like ecosystems. The high-altitude cushion-plant peatlands of the Central Andes are a typical element of the high-Andean vegetational belt at altitudes of 4000–5000 m a.s.l. The main peat-accumulating species of these soligenous peatlands are the Juncaceae *Distichia muscoides* and *Oxychloe andina* (Ruthsatz, 2012, 2000; Squeo et al., 2006). The shoots of *Distichia*- and *Oxychloe*-cushion plants continue to grow at their tops, but die off from the bottom (Rauh, 1988). On the cushion surface, they form dense mats with a strong apical dominance (Balslev, 1996). The cushions probably started to grow as a single individual producing numerous shoots and rhizomes, which over time transformed into smaller groups as the underground parts died off. The rhizomes continuously produce short aerial branches with small terminal rosettes. Due to anaerobic conditions and cold temperatures, the dead remains do not decay immediately. The older tissue is overgrown and deposited in the form of peat (Schittek, 2014; Benavides et al., 2013). High-Andean cushion peatlands can accumulate several meters of peat sediment (Schittek et al., 2012; Squeo et al., 2006; Earle et al., 2003). The palaeoenvironments, which facilitated peat deposition in the past, can be reconstructed using the peat structure retrieved from the peat column. The few studies available on high-altitude cushion peatlands in the Central Andes have shown that they can indeed be very suitable archives to retrieve palaeoecological and palaeoclimatic time series (Schittek et al., 2016, 2015; Schittek, 2014; Engel et al., 2014; Kuentz et al., 2011; Earle et al., 2003). Up to now, little is known about the establishment and growth habits of the characteristic, cushion-forming and peat-accumulating Juncaceae.

In this study, we investigate a single cushion-plant peat hillock, which is situated on a spring at an altitude of 4250 m a.s.l. So far, cushion-plant peat hillocks have only been reported by Schittek (2014), who observed similar structures in the spring areas within an extensive high-altitude peatland in the Eastern Cordillera of NW Argentina. In contrast, the here-described peat hillock is a markedly isolated feature, being surrounded by a sparse vegetation of low stature woody shrubs and tussock grasses.The approach of this study combines a plant inventory and a terrestrial laser scan of the hillock to investigate the current vegetation. We further aimed to conduct an analysis of palaeobotanical and geochemical patterns of peat sections retrieved from the peat hillock to check if the peat accumulation represents a continuous archive for palaeoenvironmental research. This was of special interest to possibly supplement the current knowledge about climatic and environmental changes in the western Andes of southern Peru, especially concerning the past centuries. A previously investigated peat record from the nearby Cerro Llamoca peatland (Schittek et al., 2015) lacked palaeoecological information about the past 500 years. Therefore, a special objective of our research was to gather more palaeoecological details about those missing centuries.The current state of knowledge on the climate history of the youngest period of the Holocene is still very limited, although indispensable for archaeological research. Especially in the Palpa/Nasca area and the valleys of the Río Grande drainage in southern Peru, further information on past climate variabilities concentrating on the time span from AD 1000-1550 is necessary for a better understanding of what happened during that cultural period (Sossna, 2016; Unkel et al., 2012; Reindel, 2009). Only few information is available about the environmental changes in the central Andes during the Little Ice Age (ca. AD 1500–1880) (Liu et al., 2005).

2. Study area

2.1. Geographical setting

The studied peat hillock is situated in the western cordillera of the Andes of southern Peru (14° 10′ S, 74° 44′ W) at an altitude of 4250 m a.s.l., within the water catchment area of Cerro Llamoca peatland (Schittek et al., 2015; Schittek et al., 2012) (Fig. 1). It is located west of the continental watershed in the Río Viscas drainage. Cerro Llamoca (4450 m a.s.l.), the overlooking and name-giving peak, is the highest point of the whole Río Grande drainage system.

The study site is characterized by a tropical climate (Lauer, 1993), shaped by distinct temperature and precipitation fluctuations on daily and yearly patterns. The ERA-Interim model (ECMWF, 2014) renders a mean annual precipitation of 349 mm per year for the location of Cerro Llamoca peatland, with about 90% of the annual precipitation falling during the austral summer (December to March). Rain tends to cluster in “rainy episodes” lasting about a week, interrupted by dry episodes of the same length (Garreaud, 2000). The duration and intensity of wetter and drier episodes in the summertime is highly sensitive to large-scale circulation anomalies (Garreaud and Aceituno, 2001). This concentration of precipitation during distinct periods of the year could be termed as a “monsoon-like” precipitation pattern (Zhou and Lau, 1998).

In total, 17 vascular plants were identified from the studied peat hillock (Table 1). Because our collections took place within the dry season, it is possible that some herbaceous species were not registered. The floristic composition is dominated by species, which, according to Ruthsatz (2012, 2000), typically occur in the high-altitude cushion-plant peatlands (bofedales) of the more humid, tropical central Andes, within the range of the humid Puna (*sensu* Troll, 1968), like *Distichia muscoides* and *Plantago tubulosa*. In contrast, the occurring *Phylloscirpus deserticola* is a typical representative of the drier western central Andean peatlands (Ruthsatz, 2012). *Distichia muscoides* is the most characteristic cushion-forming and peat-accumulating species of the hillock. Nowadays, superficial drying and the impact by the trampling of the hoofed animals (mainly cows) limit the growth of its cushions significantly. Dense mats of *Phylloscirpus deserticola* dominate degraded sections and drier cushion tops. *Plantago tubulosa* prevails where the surface is characterized by well-saturated conditions, but repeatedly trampled. Sections that are less frequented by the grazing animals are still grown with sporadic tussocks of *Deyeuxia rigida*, bordering well-saturated swards of *Werneria pygmaea*.

Especially the steep sides of the peat hillock originating to the west are highly degraded. This degradation is caused by cattle climbing on top of the hillock in order to reach fresh plants to feed on. Through this severe damage, the cushion-forming vegetation dies off. The loss of the protective vegetation accelerates water loss (Schittek et al., 2012). If the vegetation cover once is lost, the brown peat surface warms and dries quickly during daytime. Once if the peat dries out, the peat substrate is easily eroded by wind.

2.2. Geomorphometric mapping

Terrestrial laser scanning (TLS) techniques were applied for the first time on a single cushion-plant peat hillock in order to provide new insights into its spatial characteristics. Data acquisition was performed by using a time-of-flight scanner (Riegl VZ-400) with online waveform processing technology according to the methods described in Höfle et al. (2013). Four scan positions were necessary for capturing the whole peat hillock.

TLS allows to determine and to quantify exactly the extent of the investigated peat hillock. Fig. 2 shows the captured point cloud of the peat hillock. Different grey scales represent reflectance properties of the peat surface. The black color (i.e. no data values) results from either shading effects or open water areas. Assuming an underlying inclined plane, the total calculated volume of the hillock is $\sim 24.5\,m^3$ (Forbriger et al., 2011).

The use of the laser reflection values was tested for derivation of plant segmentation, but due to distorting effects of partly dried plant material, backscatter values resulting from different species and reflection properties give only sparse information on vegetation units. Water content, with its high reflection variation, overlays the radiometric information of single plants (Fig. 3).

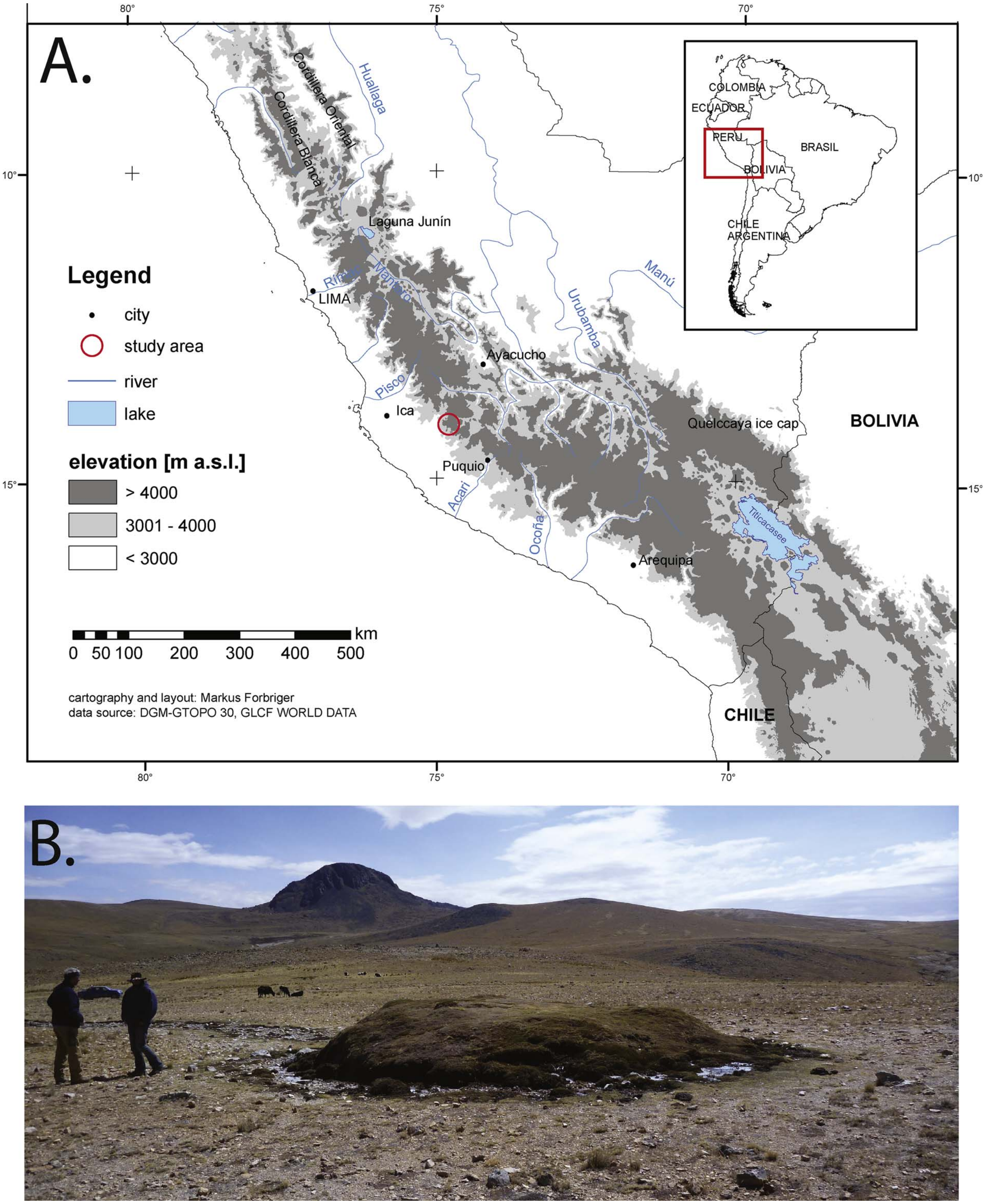


Fig. 1. A. The study area (red circle), located in the western cordillera of the Peruvian Andes. B. Studied peat hillock surrounded by high-Andean steppe and overlooked by Cerro Llamoca mountain (4450 m a.s.l.). Cattle in the background cause heavy degradation of steppe and cushion vegetation. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

3. Material and methods

3.1. Sampling techniques

The field work was carried out in August 2010 and October 2011. During both campaigns, plant species lists of the peat hillock and the surrounding vegetation were created and specimens of characteristic plants were herbarized. The nomenclature of plant species follows Zuloaga et al. (2008).

Two parallel cores of 4 cm diameter were extracted from the top of the peat hillock by using a newly designed sampler suitable for extremely dense and fibrous Andean peat types (Schittek, 2014). In this study, palaeoecological analyses focus on the deepest core (P103B), which reached a depth of 66 cm. In the Palaeoecology Laboratory of the Institute for Geography Education (University of Cologne), the core was split into two core halves, photographed and described

Table 1
List of plant species inhabiting the peat hillock.

Species	Family
Aciachne pulvinata Bentham	Poaceae
Arenaria spec.	Caryophyllaceae
Castilleja pumila (Benth.) Wedd. ex Herrera	Orobanchaceae
Deyeuxia brevifolia J.Presl	Poaceae
Deyeuxia curvula Wedd.	Poaceae
Deyeuxia rigescens (J.Presl) Türpe	Poaceae
Distichia muscoides Nees & Meyen	Juncaceae
Gentiana prostrata Haenke	Gentianaceae
Gentianella spec.	Gentianaceae
Hypochoeris taraxacoides (Walp.) Benth. & Hook.f.	Asteraceae
Lachemilla diplophylla (Diels) Rothm.	Rosaceae
Lachemilla pinnata (Ruiz & Pav.) Rothm.	Rosaceae
Phylloscirpus deserticola (Phil.) Dhooge & Goetgh.	Cyperaceae
Plantago rigida H.B.K.	Plantaginaceae
Plantago tubulosa Decne.	Plantaginaceae
Werneria pygmaea Gillies ex Hook. & Arn.	Asteraceae
Zameioscirpus muticus Dhooge & Goetgh.	Cyperaceae
Total 17	9

sedimentologically. Both core halves were subsampled at 1 cm intervals for micro-/macrofossil analyses and for geochemical analyses.

3.2. Chronology

For age control, seven sediment samples were dated by Bernd Kromer and Susanne Lindauer (Klaus Tschira Centre for Archaeometry, Heidelberg University) (Table 2). All radiocarbon dates were calibrated using CALIB 7.0.4. The IntCal13 data set for Northern Hemisphere calibration (Reimer et al., 2013) was applied due to the south shift of the intertropical convergence zone during the austral spring and summer seasons. This brings atmospheric CO_2 from the Northern Hemisphere to the Andes, which is primarily taken up by the vegetation. The age-depth model (Fig. 4) is based on a Monte Carlo approach to generate confidence intervals that incorporate the probabilistic nature of radiocarbon dates by using the MCAgeDepth software (Higuera et al., 2009). The software generates a cubic smoothing spline through all the dates. A total of 1000 Monte Carlo simulations were used to generate confidence intervals. The final probability model is based on the median of all the simulations.

3.3. Total carbon and total nitrogen content

For the measurement of total carbon (TC) and total nitrogen (TN) contents, samples were dried at 105 °C, milled with a high speed mill grinder and dried again at 105 °C for at least 24 h. The Elementar Vario EL CHNOS elemental analyzer (Elementar Analysensysteme GmbH) at the Institute of Geography (University of Cologne) was used to measure the TC and TN contents according to the amounts of CO_2 and N_2 released after sample combustion (peak integration) and calibrated against elemental standards. As stated in Schittek et al. (2016) and Ruthsatz (1993), the amounts of inorganic carbon and nitrogen in peats from high-Andean peat-accumulating ecosystems usually are very low. Therefore, the TC content is treated as an equivalent of the apparent total organic carbon content and is used for the calculation of the C/N ratio.

3.4. Analysis of micro-/macrofossils and charred particles

For macrofossil and microfossil sample preparation, we applied the extended protocol according to Schittek et al. (2015). Subsamples were taken at 4 cm intervals. After KOH treatment for deflocculation, the samples were sieved in order to separate three size fractions (> 2 mm, 2 mm–250 µm, 250–125 µm) for the study of macrofossils. The further microfossil preparation followed standard techniques described in Faegri et al. (1989). Microfossil samples (< 112 µm) were mounted in glycerine for the counting of pollen and non-pollen palynomorphs under x400 and x1000 magnification. For identification, we used our own reference collection and published atlases (Torres et al., 2012; Kuhry, 1988; Hooghiemstra, 1984; Markgraf and D'Antoni, 1978; Heusser, 1971). Regional pollen types were counted to sums of at least 300 in each sample. For macroscopic analyses, the samples > 125 µm were disaggregated in deionized water. Plant tissues were determined in the 2 mm fraction under a dissecting microscope. Macroscopical charcoal analysis was performed at 1 cm intervals and followed the methods described in Schittek (2014).

4. Results

The investigated cushion-plant peat hillock represents a so-far undescribed spring ecosystem feature of the central Andes. The hillock seems to consist of only a single cushion covering a punctiform spring. However, in fact, the relatively homogenous giant cushion is the product of the accumulation of multiple cushion-forming plants over

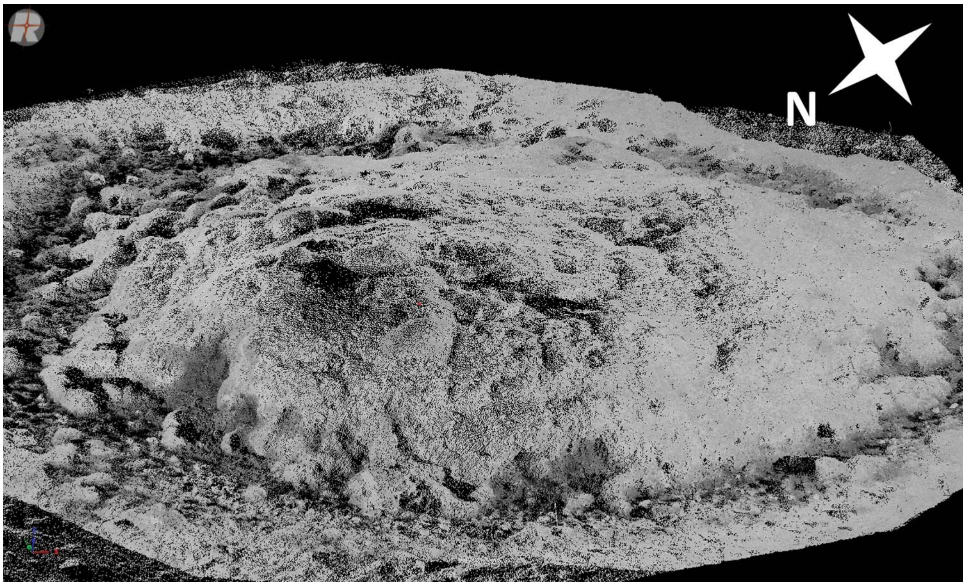


Fig. 2. Point cloud (appr. 3.5*10^6 points) with reflectance values. Holes (black areas) result either from shading effects or from absorption within water areas.

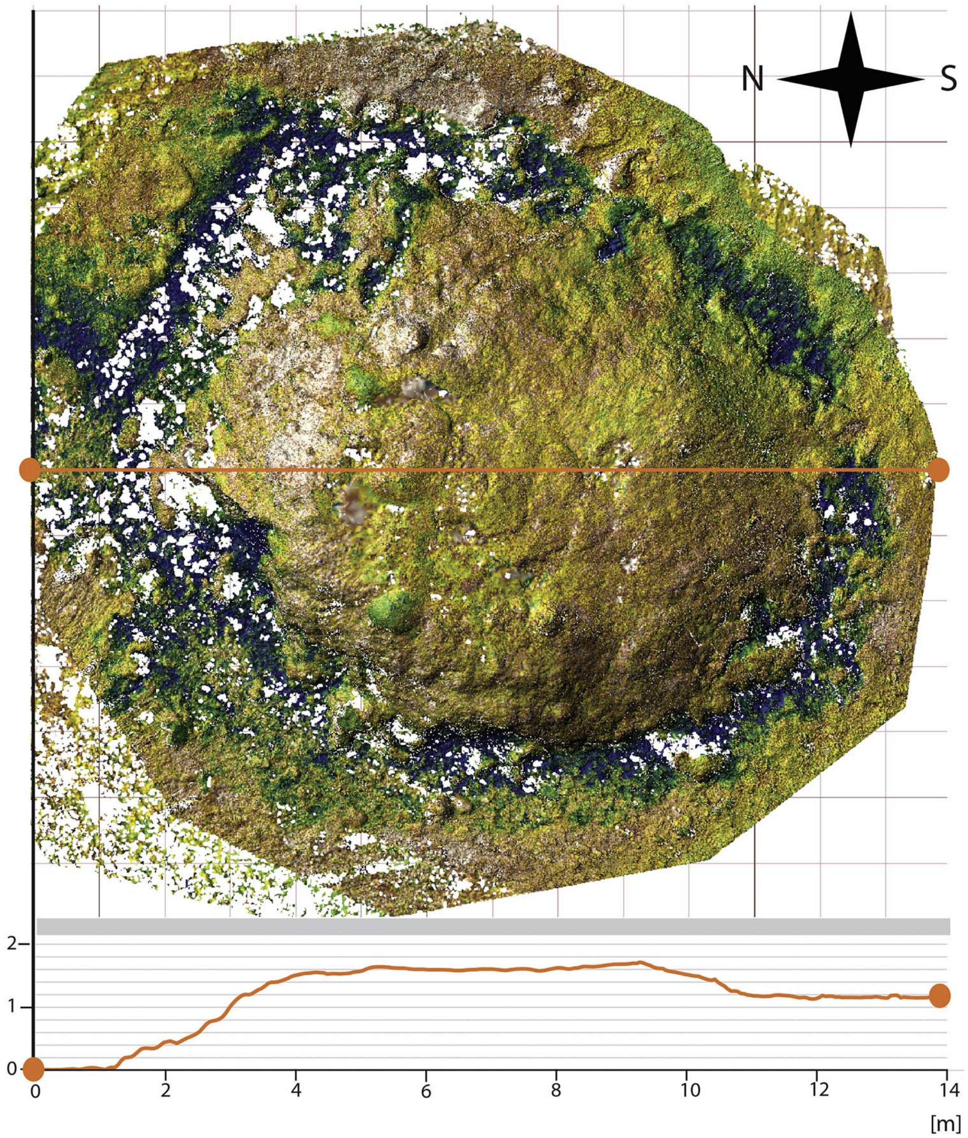


Fig. 3. Derivation of plant segmentation computed with GigaMesh and contour of the peat hillock. We used a hypsometric color ramp ranging from dark blue (minimum) to white (maximum). To maximize the contrast of the visualization, we chose a non-linear mapping of reflectance to color. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

centuries. With an extension of about 10 m in diameter and a height of 1,60 m, the nearly circular peat hillock (Fig. 3) represents a distinctly small type of a soligenous peat-accumulating ecosystem.

4.1. Stratigraphy and chronology

The stratigraphical and palaeoecological approaches derived from the peat cores help to reconstruct the growth history of the peat hillock through time, in dependence on the environmental conditions influenced by larger-scale climatic variabilities. The sedimentary deposit represents a continuous, homogenous and high-resolute peat record. The peat matrix, with variable contents of embedded silt, mainly consists of fibrous *Distichia* rhizome remains.

In contrast to the heavily degraded cushion-plant vegetation of nearby Cerro Llamoca peatland at the valley bottom (Schittek et al., 2015), which lacked palaeoenvironmental information about the past 500 years, the isolated peat hillock appeared markedly less degraded than other *Distichia muscoides*-dominated peat areas within the Cerro Llamoca valley. It therefore represented a unique opportunity to deliver further information on the landscape's environmental history.

Table 2
Radiocarbon ages of core P103B, retrieved from the peat hillock. The calibrated age ranges were calculated using CALIB 7.0.4 and the IntCal13 data set (Reimer et al., 2013). The modelled ages are the result of a probabilistic age-depth model using MCAgeDepth (Higuera et al., 2009). The range represents the 2σ values, and the median ages are in parentheses.

Lab #	core depth (cm)	measured ^{14}C	measured error (±)	MCAgeDepth modelled age (cal yr BP)
MAMS-11625	23,5	90	25	31–(106)–257
MAMS – 11622*	38,5	554	24	527–(554)–632
MAMS-11626	38,5	627	23	557–(597)–657
MAMS-14773	44,5	971	26	801–(860)–932
MAMS – 11623*	57,5	729	25	658–(675)–704
MAMS-11627	61,5	1124	25	970–(1020)–1129
MAMS-14774	65,5	1142	25	979–(1036)–1167

* Ages not used for the age-depth model.

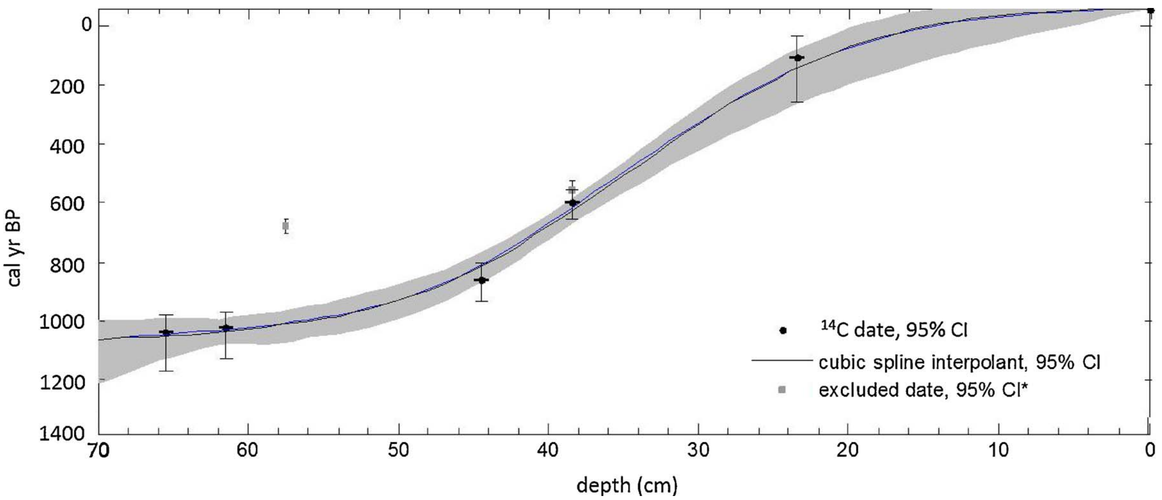


Fig. 4. Age-depth model for core P103 B retrieved from the peat hillock based on seven ^{14}C dates. The grey band represents the modelled range of dates and the black line the 50th percentile of all runs.

The retrieved 66 cm long peat core gives an insight to the past 1050 years of the peat hillock's growth. The age-depth model is based on seven radiocarbon dates. Due to possible contamination and/or redeposition effects from the surrounding steppe, two dates were omitted from the model. The remaining dates allow a high-resolute palaeoenvironmental reconstruction with a sample resolution ranging between 10 and 35 years per cm.

4.2. Geochemical analyses

The core sequence shows variable percentual total carbon (TC) and total nitrogen (TN) contents (Fig. 5). The mean TC percentages range between 40 and 50%, with maximum values of 55% at around AD 1550. Here, TN percentages also peak with the highest measured value of 2.7%. Mean values range between 1.5-2%, but after AD 1850 until the present, they decline to mean values ranging between 1 and 1.5%.

TC/TN ratios constantly decline from values of 30–35 at AD 930–1000 towards values below 25 from AD 1300-1600. At AD 1600–1650, TC/TN ratios rise to values of 27, but constantly retreat to values of around 23 starting from AD 1650. After AD 1830, TC/TN ratios rise and fluctuate between values of 30–40.

4.3. Analyses of microfossils and charred particles

The pollen record (Fig. 6) is mainly dominated by *Senecio*-type Asteraceae (mostly shrubs and herbs) and Poaceae (grasses), which reflects the dominating vegetation surrounding the peat hillock. The remaining pollen flora is composed of typical regional taxa of the Altoandean and Puna altitudinal belts (Reese and Liu, 2005; Kuentz et al., 2007). Cyperaceae (including *Zameioscirpus*) and *Plantago* represent local peatland and semi-aquatic vegetation (Ruthsatz, 2000) and were excluded from the pollen sum. Other pollen types comprise extra-regional tree species (*Alnus, Polylepis*) from the eastern Andean forests below 3500 m a.s.l., which occurred only sporadically in low abundances. In addition to pollen, green algae (*Spirogyra* spec.) and coprophilous fungal spores represent further microfossils other than pollen. Charred particles were counted separately in the 250–125 μm size fraction of the sieved samples.

Senecio-type Asteraceae show highest abundances at AD 950–1150. This period is further characterized by a high abundance of *Plantago* pollen and high amounts of fungal spores. The accumulation rates of charred particles reach the highest values in the whole record and tend to fluctuate significantly during that period. After AD 1150 until about AD 1550, the pollen spectrum is marked by a steady decline in *Senecio*-type Asteraceae and a coinciding rise in Poaceae percentages. Charred particles remain at low levels between AD 1050 and 1300, but are highly abundant afterwards until about AD 1500. The period between AD 1450 and 1550 shows a distinct change, when Poaceae begin to dominate and remain highly abundant until AD 1700. Higher abundances of Cyperaceae after around AD 1600, and very low values of charred particles characterize the younger part of the record. Beginning at around AD 1700, Poaceae decline and *Senecio*-type Asteraceae begin to increase again. The period between AD 1880 and 1980 proved to be palynologically barren. Only in the most recent samples, Chenopodiaceae, Cyperaceae, trees and most fungal spores occurred in higher abundances.

5. Discussion

In the light of the current global warming, we need past analogues and comprehensive documentation of the present situation to be able to interpret observed and modelled future changes. This might open the door to a much more precise understanding of the reaction of sensitive high-mountainous ecosystems towards climatic changes, and make it possible to link observed ecology-based evidence with palaeoecology-based reconstruction.

The investigated cushion-plant peat hillock fulfills all prerequisites to serve as a climate archive. It is characterized by a continuous growth, and a lot of palaeoecologic information can be retrieved from the accumulated peat. It further represents a very sensitive ecosystem, which responds to climate-induced and human-induced changes.

5.1. Clues from the past

Based on the investigation of lake sediments from Laguna Pumacocha in Peru, Bird et al. (2011) provided evidence for drier conditions at AD 900-1100 and linked this event to the Northern Hemisphere Medieval Climate Anomaly (MCA) and a considerable weakening of the South American Summer Monsoon (SASM) at the same time. Chepstow-Lusty et al. (2003) also observed markedly drier conditions for that period for the Cuzco region, as well as ice core data derived from Quelccaya ice cap by Thompson et al. (1986). A fully established dry period at AD 900 is reported by Schittek et al. (2015), based on evidence from Cerro Llamoca peatland, which is situated in close vicinity to the peat hillock investigated in this study. After AD 900, humidity slowly increased. TC/TN ratio values remain high during that period. In Fig. 7, selected proxies of this study are compared with published records from Laguna Pumacocha (Bird et al., 2011) and

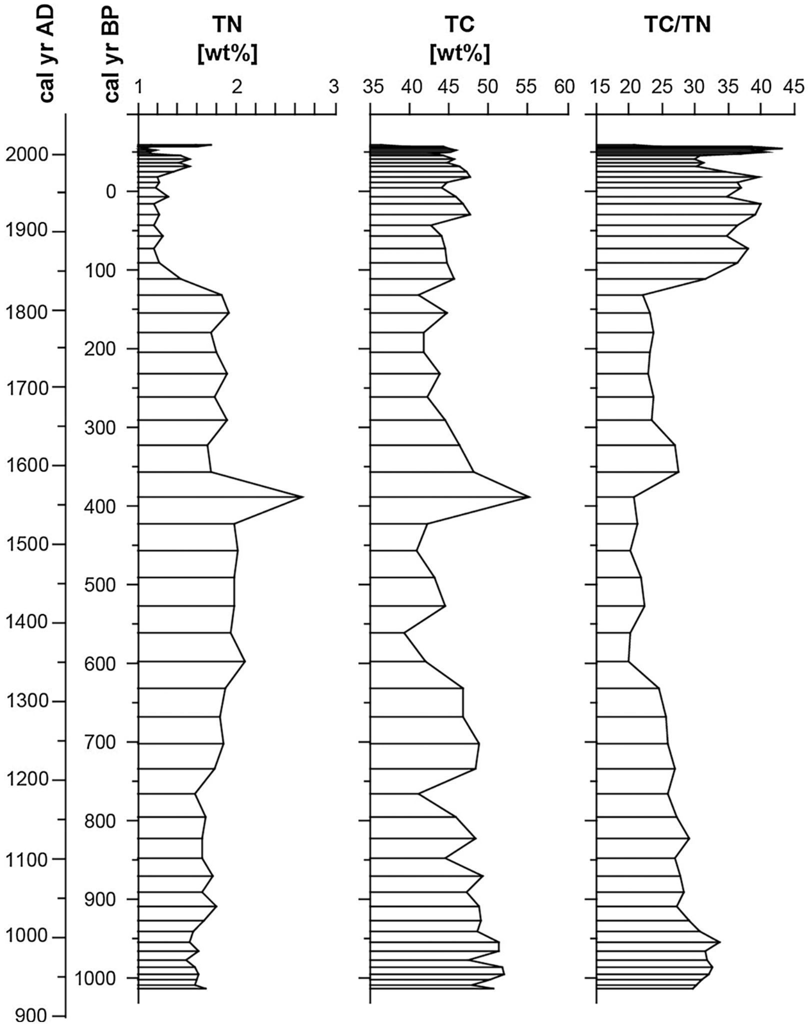


Fig. 5. Total carbon and total nitrogen elemental contents for core P103B.

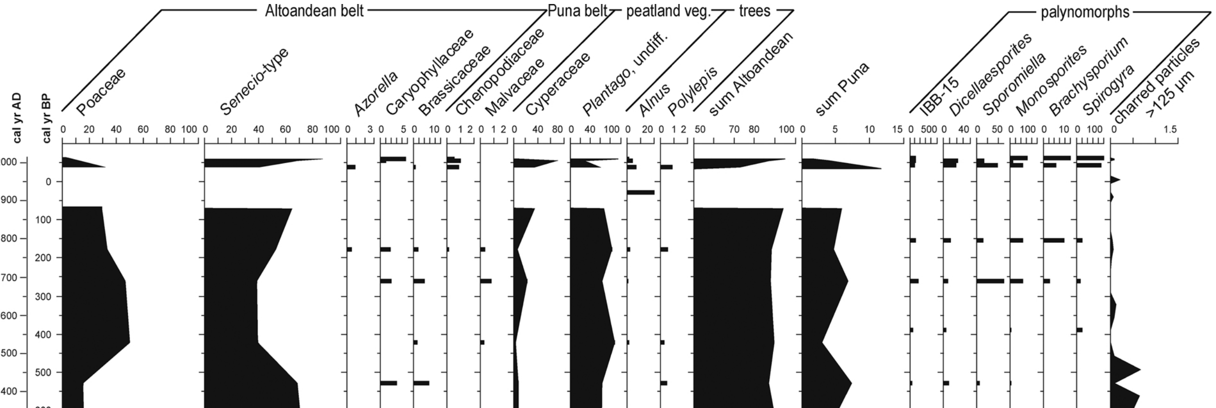


Fig. 6. Pollen, palynomorphs and charred particles diagram for core P103 B retrieved from the peat hillock. Peatland vegetation was excluded from the pollen sum.

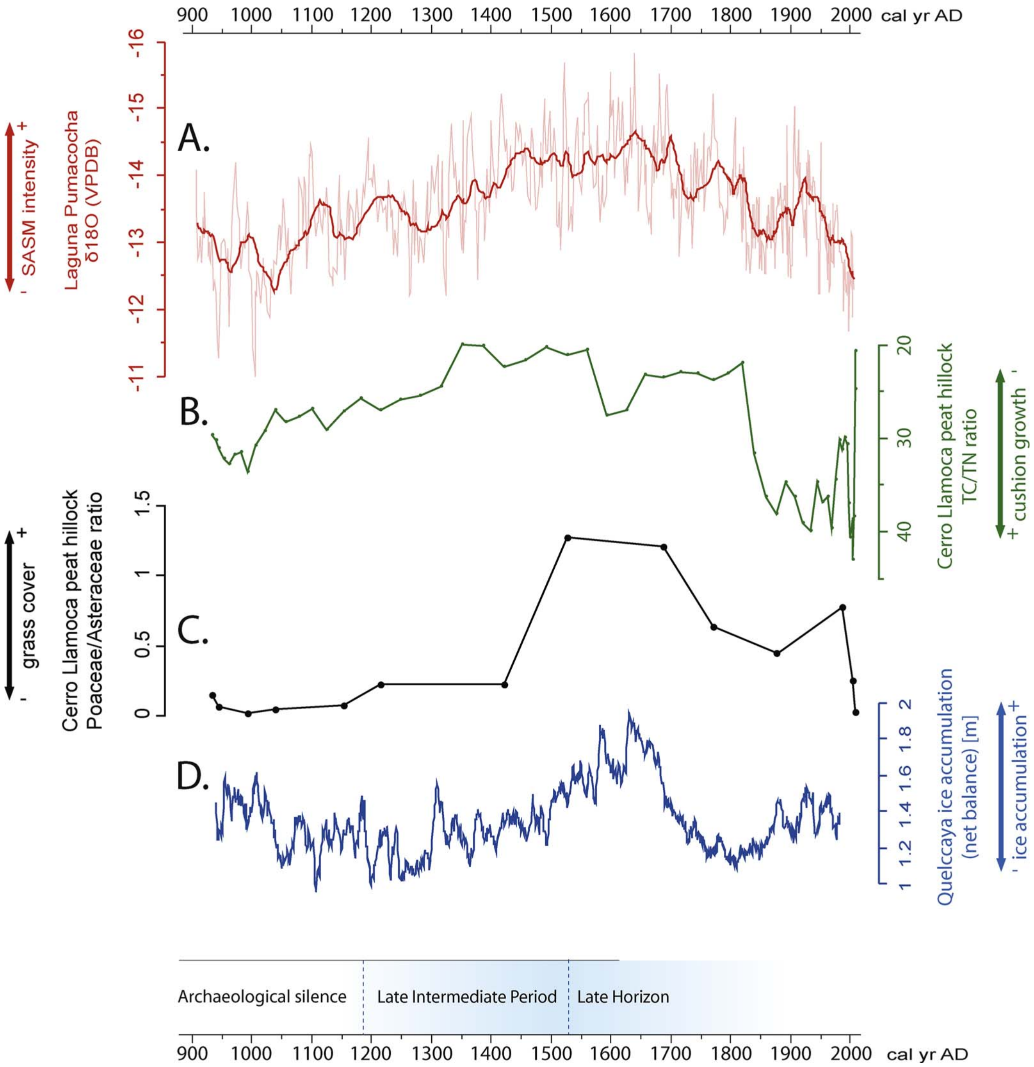


Fig. 7. The last millennial Poaceae/Asteraceae ratio and TC/TN ratio sequences of the peat hillock compared with other palaeoclimate records from the Peruvian Andes and the archaeological chronology by Unkel et al. (2012). A. The Laguna Pumacocha $\delta^{18}O$ record (11° S) of SASM intensity (Bird et al. (2011) with 11-year mean values. B. TC/TN ratios of the peat hillock sequence. C. Poaceae/Asteraceae ratio of the peat hillock sequence. D. Quelccaya ice cap (14° S): ice accumulation (net balance) curve (Thompson et al., 1986) (11-year mean values).

Quelccaya ice core (Thompson et al., 1986). The Poaceae/Asteraceae ratio gives evidence that the abundance of grasses was rather low at that time. During drier periods, the predominance of grasses in the high-Andean vegetation belt diminishes, and better-adapted high-mountainous vegetation components like Asteraceae (mostly shrubs), Brassicaceae, Malvaceae and Apiaceae (*Azorella*) become more evident in the pollen spectrum (Schittek et al., 2015; Kuentz et al., 2011).

The high abundance of charred particles points to repeatedly occurring fire events in the high-Andean steppe, which is promoted by rather unstable conditions with frequent climate oscillations. The proposed occurrence of unstable conditions is additionally supported by strongly fluctuating $\delta^{18}O$ values derived from Laguna Pumacocha (Bird et al., 2011), showing the largest amplitudes of the past 1000 years during that period. This indicates that the overall weaker SASM was frequently interrupted by spells of intensified monsoonal activity.

The period of drought lasts until about AD 1150-1200, when grass pollen become more abundant in the peat. After a relatively stable period, which prevails until about AD 1300, overall more humid, but highly unstable conditions occur. The SASM significantly intensified until about AD 1400 (Bird et al., 2011), and Rodbell et al. (2008) reported heavy clastic influx into Andean lakes around AD 1450. Furthermore, this period is outlined with evidence of glacial advance starting at around AD 1350 (Stansell et al., 2013), which indicates a significant lowering of temperatures. Ice accumulation at Quelccaya ice cap also increased after AD 1350. A corresponding change in the isotopic composition of Laguna Pumacocha record (Bird et al., 2011) suggests the onset of the Little Ice Age (LIA).

This colder period is characterized by a significant dominance of grass pollen in the peat hillock's peat samples. TC/TN ratios lowered, which is due to less favorable conditions for plant growth. SASM activity, according to Bird et al. (2011), was most intense during that period. This heavy monsoonal forcing seems to be linked to the destabilization of slopes within the Cerro Llamoca peatland catchment area as observed by Schittek et al. (2015). Here, more than 3 m of debris were deposited upon the peatland's sediments between AD 1450 and 1700.

The transition towards the current warm period is marked as an abrupt change in the peat hillock record. Around AD 1800/1850, TC/TN ratios increased significantly and grass pollen abundance decreases again. Bird et al. (2011) also mention an abrupt change from the LIA to the prevailing warmer period and suggest a generally weaker SASM as observed during the MCA. Several cold spells occurred during the 19th century, which influenced plant growth, and therefore, TC/TN ratios repeatedly lowered.

The peat hillock's palaeoecological information corresponds well to other palaeoclimate records from the Peruvian Andes. This similarity underlines the suggestion that climatic changes during the past millennium occurred synchronously across the central Andes (Liu et al., 2005). It further supports the assumptions made by Bird et al. (2011), who suggest that an intensification of the SASM, which results in a stronger moisture transport towards the western range of the Andes, is driven by a more southerly position of the Intertropical Convergence

Zone (Haug et al., 2001).

The palaeoenvironmental record further evidences a remarkable coincidence with changes in population density of local people in the highlands and at the foothills of the whole Río Viscas and Río Grande drainage area at the beginning of the Late Intermediate Period (LIP), at around 1180 AD (Unkel et al., 2012). The previous dry period, which was fully developed around 900 AD (Schittek et al., 2015), corresponds to an archaeological silence, when the foothills may have been depopulated almost completely (Reindel, 2009). A rapid shift to more humid conditions at the beginning of the LIP triggered a massive repopulation of the lower valleys and river oases in the Palpa-Nasca area. The extremely high population density in the area during the height of the LIP required an optimum of agricultural production, which in turn suggests abundant and reliable rainfall providing sufficient irrigation water for the highland terraces and for the cultivation of the valley floors (Sossna, 2016). Our findings therefore extend the knowledge about pre-Columbian human-environment interactions in the Palpa-Nasca area and show that an intensification of the SASM had a pivotal influence on cultural development during the LIP.

5.2. The influence of climate changes on peat accumulation

Down-core analyses of carbon and nitrogen contents are a simple proxy of the varying degree of peat decomposition (Chambers et al., 2011). The investigated peat hillock represents a sink of carbon. Layers with high carbon contents indicate periods that promoted a rapid accumulation of organic matter. Since climatic changes have a direct influence on temperature and soil moisture, climate-induced changes are also reflected in the carbon and nitrogen mineralization and can be read off well in the carbon/nitrogen ratio. Temperature and water level directly controls the amount of accumulated carbon. Climatic changes can influence the labile carbon and nitrogen reserves in the peat in such a way, that they trigger structural changes of the associated plant communities and, in relation, changes in decomposition processes (Keller et al., 2004).

Regarding the growth of the peat hillock, cooler temperatures are most probably a limiting factor. Presumably, the peat hillock was always sufficiently water-saturated during the past 1000 years, and thus, dry climate conditions (to a certain degree) would play a minor role as long as the spring delivers enough water. However, cooler temperatures could significantly decelerate the growth of the peat-accumulating vegetation. According to Scherrer and Körner (2011), plants with cushion growth forms are able to modify their surrounding temperatures creating pockets of more favorable microclimatic conditions. This feature suggests a tolerance of the plants to higher temperatures than those that would naturally occur in such a high-elevation habitat (Benavides et al., 2013).

The data presented here highlight that *Distichia*-dominated cushions and their accumulated peat are indicators of the degree of environmental changes that are induced by climate, on one hand. It further provides evidence that such a spatially-defined peat-accumulating ecosystem is capable to survive pronounced climatic oscillations as long as it does not lose its protective cushion-plant surface. Presently, the most conspicuous impact on the peat hillock's structure is overgrazing, and hence, damage by the trampling of hoofed animals. Where *Distichia muscoides* as main cushion-former is once destroyed, the peat rapidly erodes (Schittek et al., 2012).

For the documentation of future changes, the methods of long-term ecological research, mainly by installing permanent vegetation plots, should be applied. As vegetation mapping by TLS proves particularly effective in documenting the current situation of the vegetation stand, it represents an important prerequisite for future studies concerning long-term vegetation dynamics.

6. Conclusions and perspectives

Cushion-plant peat hillocks represent a unique, so-far neglected vegetation component of the Andean highlands. In the western Andes of Peru, these spring ecosystems are dominated by *Distichia muscoides*, a cushion-forming Juncaceae, which is capable to accumulate peat.

The investigated peat hillock, spatially-defined by the spring area, revealed an astonishing age of about 1000 years. During that time, the cushion plants constantly accumulated peat and survived pronounced climatic oscillations. The past environmental changes could be tracked and further analyzed by applying geochemical and palynological methods to the retrieved peat sediments. Our data show that moisture fluctuations can be correlated to shifts in the intensity of the SASM. The cooling of the Little Ice Age at AD 1300–1825 decelerated peat accumulation. A higher abundance of grass pollen provides evidence of vegetational changes in the environment of the peat hillock during that period.

Nowadays, the peat hillock is heavily affected by grazing. To document future changes, permanent vegetation plots should be installed, accompanied by TLS technologies. As the high resolution of the acquired data allows the precise measurement, it becomes possible to detect minor changes over time by repeated measurements. Taken the given dataset as a basis for future investigations, a recapture of the peat hillock is planned for 2020–ten years after the first data acquisition. Further ecological and palaeoecological approaches from peat cores will help to achieve a better understanding of past climatic changes and to decipher the mechanisms of cushion-plant succession.

Acknowledgements

The authors cordially thank Markus Dzakovic for the preparation of the pollen slides and help in the laboratory. Funding by the German Federal Ministry for Education and Research (BMBF) within the project “Andean transect – Climate Sensitivity of pre-Columbian Man-Environment Systems” (BMBF-01UA0804B) is greatly acknowledged.

References

Balslev, H., 1996. Juncaceae. Flora Neotropica. Monograph. pp. 68.

Benavides, J.C., Vitt, D.H., Kelman Wieder, R., 2013. The influence of climate change on recent peat accumulation patterns of *Distichia muscoides* cushion bogs in the high-elevation tropical Andes of Colombia. J. Geophys. Res.: Biogeosci. 118, 1627–1635.

Bird, B.W., Abbott, M.B., Vuille, M., Rodbell, D.T., Stansell, N.D., Rosenmeier, M.F., 2011. A 2,300-year-long annually resolved record of the South American summer monsoon from the Peruvian Andes. P. Natl. Acad. Sci. Usa 21, 8583–8588.

Chambers, F.M., Beilman, D.W., Yu, Z., 2011. Methods for determining peat humification and for quantifying peat bulk density, organic matter and carbon content for palaeostudies of climate and peatland carbon dynamics. Mires Peat 7, 1–10.

Chepstow-Lusty, A., Frogley, M.R., Bauer, B.S., Bush, M., Herrera, A.T., 2003. A late Holocene record of arid events from the Cuzco region, Peru.J. Quaternary Sci. 18 (6), 491–502.

ECMWF, 2014. European Reanalysis Interim. Retrieved from http://www.ecmwf.int/.

Earle, L.R., Warner, B.G., Aravena, R., 2003. Rapid development of an unusual peat-accumulating ecosystem in the Chilean Altiplano. Quaternary Res. 59, 2–11.

Engel, E., Skrzypek, G., Chuman, T., Šefrna, L., Mihaljevič, M., 2014. Climate in the Western Cordillera of the Central Andes over the last 4300 years. Quaternary Sci. Rev. 99, 60–77.

Faegri, K., Kaland, P.E., Krzywinski, K., 1989. Textbook of pollen analysis.

Forbriger, M., Schittek, K., Siart, C., Höfle, B., 2011. New approaches in vegetation mapping – use of terrestrial laser scanning on high Andean cushion peatlands. Geophys. Res. Abstr. 13, 12268.

Garreaud, R.D., Aceituno, P., 2001. Interannual rainfall variability over the South American Altiplano. J. Climate 14, 2779–2789.

Garreaud, R.D., 2000. Intraseasonal variability of moisture and rainfall over the South American Altiplano. Mon Weather Rev. 128, 3337–3346.

Haug, G.H., Hughen, K.A., Sigman, D.M., Peterson, L.C., Röhl, U., 2001. Southward migration of the intertropical convergence zone through the Holocene. Science 293, 1304–1308.

Heusser, C.J., 1971. Pollen and Spores of Chile. (167 p).

Higuera, P.E., Brubaker, L.B., Anderson, P.M., Sheng Hu, F., Brown, T.A., 2009. Vegetation mediated the impacts of postglacial climate change on fire regimes in the south-central Brooks Range. Alaska. Ecol Mono. 79 (2), 201–219.

Höfle, B., Griesbaum, L., Forbriger, M., 2013. GIS-based detection of gullies in terrestrial LiDAR data of the Cerro Llamoca peatland (Peru). Remote Sens. 5, 5851–5870.

Hooghiemstra, H., 1984. Vegetational and climatic history of the high plain of Bogotá, Colombia : a continuous record of the last 3.5 million years. Dissertat. Botanicae 79 138 p.

Keller, J.K., White, J.R., Bridgham, S.D., Pastors, J., 2004. Climate change effects on carbon and nitrogen mineralization in peatlands through changes in soil quality. Global Change Biol. 10, 1053–1064.

Kuentz, A., Galán de Mera, A., Ledru, M., Thouret, J., 2007. Phytogeographical data and modern pollen rain of the puna belt in southern Peru (Nevado Coropuna, Western Cordillera). J. Biogeogr. 34, 1762–1776.

Kuentz, A., Ledru, M.P., Thouret, J.C., 2011. Environmental changes in the highlands of the western Andean Cordillera, southern Peru, during the Holocene. Holocene 22 (11), 1215–1226.

Kuhry, P., 1988. Palaeobotanical-palaeoecological studies of tropical high Andean peatbog sections (Cordillera Oriental, Colombia). Dissertat. Botanicae 116, 241p.

Lauer, W., 1993. Human development and environment in the Andes: a geoecological overview. Moun. Res. Develop. 13, 157–166.

Liu, K.B., Reese, C.A., Thompson, L.G., 2005. Ice-core pollen record of climatic changes in the central Andes during the last 400 yr. Quaternary Res. 64 (2), 272–278.

Markgraf, V., D'Antoni, H.L., 1978. Pollen Flora of Argentina. (208 p).

Rauh, W., 1988. Tropische Hochgebirgspflanzen: Wuchs- Und Lebensformen. (206p).

Reese, C.A., Liu, K., 2005. A modern pollen rain study from the central Andes region of South America. J. Biogeogr. 32, 709–718.

Reimer, P.J., Bard, E., Bayliss, A., Beck, J.W., Blackwell, P.G., Ramsey, C.B., Buck, C.E., Cheng, H., Edwards, R.L., Friedrich, M., Grootes, P.M., Guilderson, T.P., Haflidason, H., Hajdas, I., Hatté, C., Heaton, T.J., Hoffmann, D.L., Hogg, A.G., Hughen, K.A., Kaiser, K.F., Kromer, B., Manning, S.W., Niu, M., Reimer, R.W., Richards, D.A., Scott, E.M., Southon, J.R., Staff, R.A., Turney, C.S.M., van der Plicht, J., 2013. INTCAL13 and MARINE13 radiocarbon age calibration curves 0–50,000 years cal BP. Radiocarbon 55 (4), 1869–1887.

Reindel, M., 2009. Life at the edge of the desert – archaeological reconstruction of the settlement history in the valley of palpa, Peru. In: Reindel, M., Wagner, G.A. (Eds.), New Technologies for Archaeology, Natural Science Archaeology, pp. 439–461.

Rodbell, D., Seltzer, G., Mark, B., Smith, J., Abbott, M., 2008. Clastic sediment flux to tropical Andean lakes: records of glaciation and soil erosion. Quat. Sci. Rev. 27, 1612–1626.

Ruthsatz, B., 1993. Flora and ecological conditions of high Andean peatlands of Chile between 18°00' (Arica) and 40°30' (Osorno) south latitude. Phytocoenologia 25, 185–234.

Ruthsatz, B., 2000. Die Hartpolstermoore der Hochanden und ihre Artenvielfalt. Berichte der Reinhold-Tüxen-Gesellschaft 12, 351–371.

Ruthsatz, B., 2012. Vegetacion y ecologia de los bofedales altoandinos de Bolivia. Phytocoenologia 42, 133–179.

Scherrer, D., Körner, C., 2011. Topographically controlled thermal-habitat differentiation buffers alpine plant diversity against climate warming. J. Biogeogr. 38 (2), 406–416.

Schittek, K., Forbriger, M., Schäbitz, F., Eitel, B., 2012. In: Weingartner, H., Blumenstein, O., Vavelidis, M. (Eds.), Cushion Peatlands – Fragile Water Resources in the High Andes of Southern Peru. In: Water-Contributions to Sustainable Supply and Use, Landscape and Sustainable Development. Workinggroup Landscape and Sustainable Development, Salzburg, Austria, pp. 63–84.

Schittek, K., Forbriger, M., Mächtle, B., Schäbitz, F., Wennrich, V., Reidel, M., Eitel, B., 2015. Holocene environmental changes in the highlands of the southern Peruvian Andes (14° S) and their impact on pre-Columbian cultures. Clim. Past. 11, 27–44.

Schittek, K., Kock, S., Lücke, A., Hense, J., Ohlendorf, C., Kulemeyer, J.J., Lupo, L.C., Schäbitz, F., 2016. A high-altitude peatland record of environmental changes in the NW Argentine Andes (24 °S) over the last 2100 years. Clim. Past 12, 1165–1180.

Schittek, K., 2014. Cushion Peatlands in the High Andes of Northwestern Argentina as Archives for Palaeoenvironmental Research, Dissertat. Botanicae 412 Schweizerbart Science Publishers 176 p.

Sklenář, P., 2009. Presence of cushion plants increases community diversity in the high equatorial Andes. Flora – Morphology Distribution. Fun. Ecol. Plants 204, 270–277.

Sossna, V., 2016. Climate and settlement in southern Peru. the northern Río grande de nasca drainage between 1500 BCE and 1532 CE. Forschungen zur Archäologie Außereuropäischer Kulturen 13 320 p.

Squeo, F.A., Warner, B.G., Aravena, R., Espinoza, D., 2006. Bofedales: high altitude peatlands of the central Andes. Rev. chil. Hist. nat. 79, 245–255.

Stansell, N., Rodbell, D., Abbott, M., Mark, B., 2013. Proglacial lake sediment records of Holocene climate change in the western Cordillera of Peru. Quat. Sci. Rev. 70, 1–34.

Thompson, L.G., Mosley-Thompson, E., Dansgaard, W., Grootes, P.M., 1986. The Little Ice Age as recorded in the stratigraphy of the tropical Quelccaya ice cap. Science 234, 361–364.

Torres, G.R., Lupo, L.C., Sánchez, A.C., Schittek, K., 2012. Aportes a la flora polínica de turberas altoandinas, Provincia de Jujuy, noroeste argentino. Gayana Bot. 69, 30–36.

Troll, C., 1968. In: Troll, C. (Ed.), The Cordilleras of the Tropical Andes. Aspects of Climatic, Phytogeographical and Agrarian Ecology. In: Geo-Ecology of the Mountainous Regions of the Tropical Americas. Colloquium Geographicum, 9, Bonn.

Unkel, I., Reindel, M., Gorbahn, H., Isla Cuadrado, J., Kromer, B., Sossna, V., 2012. A comprehensive numerical chronology for the pre-Columbian cultures of the Palpa valleys south coast of Peru. J. Archaeol. Sci. 39, 2294–2303.

Zhou, J., Lau, K.M., 1998. Does a monsoon climate exist over South America? J. Climate 11, 1020–1040.

Zuloaga, F.O., Morrone, O., Belgrano, M.J., 2008. Catálogo De Las Plantas Vasculares Del Cono Sur (Argentina, Sur De Brasil, Chile, Paraguay Y Uruguay) 107 Monographs in Systematic Botany from the Missouri Botanical Garden 3350 p.

Perspectives in Plant Ecology, Evolution and Systematics 30 (2018) 125–131

Contents lists available at ScienceDirect

Perspectives in Plant Ecology, Evolution and Systematics

journal homepage: www.elsevier.com/locate/ppees

Research article

Assessing the importance of cold-stratification for seed germination in alpine plant species of the High-Andes of central Chile

Lohengrin A. Cavieres[a,b,*], Angela Sierra-Almeida[a,b]

[a] ECOBIOSIS, Departamento de Botánica, Facultad de Ciencias Naturales y Oceanográficas, Universidad de Concepción, Casilla 160-C, Concepción, Chile
[b] Instituto de Ecología y Biodiversidad (IEB), Casilla 653, Santiago, Chile

A R T I C L E I N F O

Keywords:
Seed germination
Cold stratification
Alpine
High-Andes of central Chile

A B S T R A C T

Snow cover duration, which depends on winter precipitation and temperature, is decreasing along several alpine areas around the world as a consequence of the global warming and further increases are expected in the future. Thus, alpine species that requires cold stratification to initiate seed germination may be threatened in the future. Here we report the results of an exploratory experimental survey to assess the importance of cold stratification for the seed germination in alpine plant species from the High-Andes of central Chile. In addition, we assessed how this requirement varies with elevation at the community level. Seeds of a total of fifty plant species were collected along an altitudinal range from 2200 to 3600 m a.s.l. Eighteen species corresponded to the Low-Andean vegetation belt, while thirteen and nineteen species corresponded to the Mid- and High-Andean vegetation belts, respectively. For each species, seeds that experienced cold-stratification at 4 °C for 3 months and control seeds (stored at room temperature and dry conditions for 3 months) were placed on moist paper in 4 Petri dishes containing 50 seeds each, and arranged in a growth chamber set at 20 °C/10 °C and a photoperiod of 14/10 h. While 36% of the studied species were able to germinate without cold-stratification, after this pretreatment 74% of these species germinated, suggesting that in general cold-stratification promotes seed germination of these alpine plant species. This positive effect was particularly evident on the species from the lower elevations. At highest elevation both the number of species that showed seed germination and the percentage and velocity of seed germination were low. Nonetheless, non-germinated seeds remained viable, suggesting a deeper dormancy. Our results suggest that cold stratification could be an important requirement for successful seed germination in species from lower elevations, while species from higher elevations could require other factors than cold-stratification to break seed dormancy. Nonetheless, as we did not use temperatures as those experienced in the field, further studies are needed to gain insights into the importance of the duration of cold stratification and the underlying mechanisms involved in the seed germination in the field of high-Andean plant species.

1. Introduction

The germination of seeds, that is the transition from seed to seedling, is a high-risk period in the life cycle of many plant species (Baskin and Baskin, 2014); hence, the mechanisms regulating the timing of this transition are expected to be under strong selective pressure (Angevine and Chabot, 1979; Willis et al., 2014). If seeds germination timing is an adaptive process, then natural selection should favor seed germination requirements that reduce the probability of facing environmental conditions that are not appropriate for seedlings establishment (Vleeshouwers et al., 1995; Willis et al., 2014; Baskin and Baskin, 2014). Thus, seed germination requirements should vary depending on the environmental risks associated to seedlings mortality (Meyer and Monsen, 1991; Meyer et al., 1989, 1990, 1995).

Alpine environments are characterized by low temperatures, strong winds, unstable substrates, and short growing seasons (Körner, 2003). The short growing season of alpine habitats is a major barrier for plants recruitment as it constrains seedlings growth and the period favorable for their establishment (Chambers et al., 1990; Forbis, 2003) because seedlings have to attain a critical biomass by the end of growing season to withstand the harsh and long-lasting winter conditions (Schütz, 2002).

It has been suggested that seeds from plant populations that normally experience long snow cover periods and adverse winter conditions, as occur in alpine habitats, would require an amount of time experiencing the moderately low and constant temperatures that occur

---

[*] Corresponding author at: ECOBIOSIS, Facultad de Ciencias Naturales y Oceanográficas, Departamento de Botánica, Universidad de Concepción, Casilla 160-C, Concepción, Chile.
E-mail address: lcaviere@udec.cl (L.A. Cavieres).

http://dx.doi.org/10.1016/j.ppees.2017.09.005
Received 29 November 2016; Received in revised form 20 July 2017; Accepted 28 September 2017

under the snow cover to allow or promote their germination (Meyer and Monsen, 1991; Meyer et al., 1989; Meyer et al., 1990; Schütz and Rave, 1999). In the seed germination literature the process of seeds experiencing low and constant temperatures to simulate natural winter conditions as if they were under snow is called cold stratification (Bewley et al., 2013; Baskin and Baskin, 2014). The requirement of a cold stratification period would allow seeds to sense the presence of snow, thereby timing the germination to a period appropriate for seedling survival and establishment. This way, cold stratification requirements would prevent precocious germination under autumn conditions where appropriate soil moisture and temperatures to complete seed germination and seedling establishment are not likely to persist for more than a few days (Meyer and Monsen, 1991). In addition, cold stratification requirements allow germination to occur only in spring after the snow melt, preventing young seedlings from being damaged by freezing temperatures (Billings and Mooney, 1968). Thus, it may be expected that a cold stratification period would be a common requirement for seed germination in alpine plant species. In an early review, Amen (1966) concluded that cold stratification is not a common requirement for the seed germination of several alpine plant species from different mountains in the USA (see also Sayers and Ward, 1966; Marchand and Roach, 1980; Kaye 1997). In contrast, Söyrinki (1938, cited in Körner, 2003) experimenting with 91 alpine species from the Alps, found that storage at winter temperatures (cold stratification) increased seed germination in the great majority of species. More recently, Sommerville et al. (2013) and Hoyle et al. (2015) showed that in 19 and 54 Australian alpine plant species, respectively, a cold stratification period significantly increased seed germination only in half of the species tested. Shimono and Kudo (2005) reported that cold stratification increased the seed germination over a range of temperatures in most of the 27 plant species studied in the alpine zone of Japan. Baskin and Baskin (2014) showed that seeds of most tundra species are dormant, and only those with physiological dormancy require cold stratification to release dormancy. Therefore, how important is cold stratification for seed germination in alpine plant species remains elusive. Further, although studies conducted in species with broad altitudinal distribution (e.g., Cavieres and Arroyo, 2000; García-Fernández et al., 2015) and in species living in habitats with contrasting snow cover duration (Meyer et al., 1989, 1995), have found a positive relationship between the duration of snow cover and duration of cold stratification for maximum seed germination (but see Schütz and Milberg, 1997; Schütz,2002), few studies have analyzed seed germination patterns in high elevation species at the community level, and as far as we are aware, none has evaluated the altitudinal variation of cold stratification requirements for seed germination at this scale.

Snow cover duration, which depends on winter precipitation and temperature, is decreasing on several alpine areas around the world as a consequence of the recent climate change (Beniston, 1997, 2012), and further decreases are expected to occur in the future due to global warming (op. cit.). Thus, alpine species that require to experience a time of low and constant temperatures as occur under snow (a cold stratification) to initiate their seed germination may be threatened in the future, whilst species with no such requirements may be indirectly favored, generating important changes in the composition and dynamics of alpine plant communities (Briceño et al., 2015; Walder and Erschbamer, 2015). In this study we evaluated the importance of a cold stratification period for seed germination in 50 high elevation plant species from the Andes of central Chile, and whether the requirement of cold stratification varies with elevation at the community level. We expected that a cold stratification period is an important factor for seed germination in these species, and that this importance increases with the altitude (i.e., higher duration of snow cover) at which a community develops.

2. Materials and methods

2.1. Seeds sources

Seeds of the studied alpine plant species were collected in the central Chilean Andes, in the surroundings of La Parva (33°21′S, 70°19′W) and Valle Nevado (33°19′S, 70°15′W) Ski resorts, distanced at 50 and 80 km east of Santiago, respectively. This area is characterized by an alpine climate with strong influence of the Mediterranean-type climate that prevails in the lowlands (di Castri and Hajek, 1976). Precipitation mainly occurs as snow during winter, with summer months usually receiving very few or no rain (Santibáñez and Uribe, 1990). Snow cover remains for 3–5 months depending on the altitude and slope aspect (Rozzi et al., 1989).

The alpine vegetation of the central Chile Andes can be found from immediately above the *Kageneckia angustifolia*'s tree line at 2200 m up to 3600 m a.s.l. (Cavieres et al., 2000). Based on the altitudinal distribution of plant life-forms, this vegetation has been divided in three altitudinal vegetation belts (Cavieres et al., 2000). The Low-Andean belt span from 2200 to 2500 m a.s.l., and is dominated by prostrate shrubs species such as *Chuquiraga oppositifolia* D. Don (Asteraceae), and *Anarthrophyllum cumingii* Hook. et Arn. J.F. Phil. (Fabaceae), with several annual species growing in open spaces between shrubs (Cavieres et al., 2000). The Mid-Andean belt extends from 2600 to 3200 m a.s.l., and is characterized by the dominance of cushion plants (e.g., *Laretia acaulis* Gillies & Hook. (Apiaceae), *Anarthrophyllum gayanum* (A. Gray) B.D. Jacks (Fabaceae)), and perennials herbs such as *Phacelia secunda* J.F. Gmel (Boraginaceae), *Nassauvia aculeata* (Less.) Poepp. et Endl. (Asteraceae), and *Melosperma andicola* Benth. (Plantaginaceae) (Cavieres et al., 2000). The High-Andean belt can be found from 3200 to 3600 m elevation, and is characterized by a low plant cover, where the dominant species are small rosette-forming herbs such as *Pozoa coriacea* Lag. (Apiaceae), *Chaetanthera* sp. (Asteraceae) and *Montiopsis potentilloides* (Barneoud) Ford (Montiaceae) (Cavieres et al., 2000).

The studied species were selected on basis of their seeds availability, to represent different growth-forms and families, and for to be important component of the plant communities in the study area (Table 1). Seeds of fifty plant species were collected along an altitudinal range from 2200 to 3600 m a.s.l.; eighteen species corresponded to the Low-Andean belt, while thirteen and nineteen species corresponded to the Mid- and High-Andean belts, respectively.

All seeds were collected during the natural period of seed dispersal (January-March), stored in paper bags, and transported to the laboratory at the University of Concepción. To assure high intra-specific representation for each species seeds were collected from at least 50 different individuals sparsely distributed within the corresponding vegetation belt. Depending on the species, some fruits (e.g., caryopses, achenes) were treated as seeds. In the laboratory, seeds were immediately cleaned, removing non-seed material by hand and blow away. Then visually-healthy seeds were selected, discarding those aborted and/or predated. The selected seeds of each species were allocated to treatments of cold stratification and control as follow.

2.2. Cold stratification

A widely used standard protocol for the stratification of seeds was used (Baskin and Baskin 2014). For each species, 200 seeds were placed in plastic boxes (20 × 15 × 6 cm) between two layers of paper previously wet. For this, we added distilled water until the papers were saturated and the excess of water was drain out from the plastic boxes. The boxes were completely wrapped with aluminum foil to avoid the passage of light, and were arranged into a growth chamber (Pitec, Bioref-19L) at 4 °C in darkness during three months. We monthly checked that papers remained moist during the cold stratification treatment, and we added distilled water when it was necessary. This procedure was done in dark conditions to avoid light effects on seed

Table 1
Effect of cold stratification on the seed germination of plant species from the Andes of central Chile. Germination parameters: **FPSG**, Final percentage of seed germination (%); **CVG**, coefficient of velocity of germination. Seeds of studied species were collected along three altitudinal vegetation belts: Low-Andean (2100–2500 m), Mid-Andean (2600–3200 m) and High-Andean belt (3200–3600 m). Growth forms: **S**, Shrub; **C**, Cushion plant; **PH**, Perennial herb; **A**, Annual. Statistical differences are shown as: (*) $P < 0.05$; (**) $P < 0.01$; (***) $P < 0.001$; (ns) not significant. Plant species nomenclature follows Teillier et al. (2011):.

			FPSG		CVG	
Species	Family	Growth form	Control	Cold-stratified	Control	Cold-stratified
Low-Andean belt						
Acaena alpina	Rosaceae	PH	0	2.2^{ns}	–	2.4
Acaena splendens	Rosaceae	PH	0	70^{***}	–	14.3
Acaena pinnatifida	Rosaceae	PH	0	5.6^{ns}	–	5.5
Adesmia papposa	Fabaceae	S	13.3	100^{***}	4.0	33.3^{*}
Anarthrophyllum cumingii	Fabaceae	S	23.3	0^{***}	5.2	–
Chaetanthera microphylla	Asteraceae	A	100	100^{ns}	33.3	33.3^{ns}
Diplolepis mucronata	Asclepiadaceae	S	0	0^{ns}	–	–
Galium sp.	Rubiaceae	PH	0	100^{***}	–	33.3^{*}
Lupinus microcarpus	Fabaceae	A	16.7	23.3^{ns}	3.7	12.8^{*}
Madia sativa	Asteraceae	A	2	100^{***}	7.0	33.3^{***}
Mutisia sinuata	Asteraceae	S	4	100^{***}	2.6	33.3^{*}
Nicotiana corymbosa	Solanaceae	A	0	20^{*}	–	2.9
Rhodophiala rhodolirion	Amaryllidaceae	PH	1.3	100***	2.2	33.3*
Sanicula graveolens	Apiaceae	PH	0	100***	–	33.3
Schizanthus hookeri	Solanaceae	PH	0	44.4**	–	33.3
Senecio eruciformis	Asteraceae	S	2	85.6***	1.9	15.3*
Sisyrinchium arenarium	Iridaceae	PH	0	0^{ns}	–	–
Stachys albicaulis	Lamiaceae	PH	0	23.3*	–	28.1
Mid-Andean belt						
Anarthrophyllum gayanum	Fabaceae	C	5.2	8.9^{ns}	3.9	8.7^{ns}
Astragalus looseri	Fabaceae	PH	7.7	16.7^{ns}	5.7	6.6^{*}
Calceolaria biflora	Calceolariaceae	PH	0	23.3^{*}	–	2.5
Calceolaria purpurea	Calceolariaceae	PH	0	0^{ns}	–	–
Chaetanthera euphrasioides	Asteraceae	A	0	24.4^{*}	–	33.3
Euphorbia collina	Euphorbiaceae	PH	0	0^{ns}	–	–
Laretia acaulis	Apiaceae	C	0	6.7^{ns}	–	3.6
Latace andina	Amaryllidaceae	PH	0	18.9^{*}	–	33.3
Microsteris gracilis	Polemoniaceae	A	92.7	100^{*}	3.2	33.3^{*}
Schizanthus grahamii	Solanaceae	A	0	0^{ns}	–	–
Taraxacum officinale	Asteraceae	PH	72	49.3^{ns}	4.4	5.3^{*}
Tristagma bivalve	Amaryllidaceae	PH	0	13.3^{*}	–	33.3
Tristagma sp.	Amaryllidaceae	PH	0	5.6^{*}	–	13.9
High-Andean belt						
Azorella madreporica	Apiaceae	C	0	2.2^{ns}	–	2.5
Caiophora coronata	Loasaceae	PH	34.5	54.5^{**}	4.8	5.1^{ns}
Calandrinia caespitosa	Montiacea	PH	0	2.2^{ns}	–	1.6
*Cerastium arvense**	Caryophyllaceae	PH	54.5	56.7^{ns}	6.1	3.0^{*}
Chaetanthera euphrasioides	Asteraceae	A	0	0^{ns}	–	–
Cistanthe sp.	Montiaceae	PH	0	47.8^{**}	–	3.3
Draba gilliesii	Brassicaceae	PH	1.3	13.3^{***}	0.9	5.6^{*}
Erigeron andicola	Asteraceae	PH	36.5	90^{*}	5.8	6.2^{ns}
Gamochaeta sp.	Asteraceae	PH	0	0^{ns}	–	–
Loasa sigmoidea	Loasaceae	A	0	3.3^{ns}	–	2.9
Melosperma andicola	Plantaginaceae	S	0	0^{ns}		
Montiopsis potentilloides	Montiaceae	PH	12.5	40.7^{**}	10.1	10.3^{ns}
Nassauvia pinnigera	Asteraceae	PH	0	16.7^{*}	–	8.8
Nastanthus ventosus	Calyceraceae	PH	0	0^{ns}	–	–
Nicotiana corymbosa	Solanaceae	A	0	0^{ns}	–	–
Noccaea magellanica	Brassicaceae	PH	0	2^{ns}	–	5.6
Phacelia secunda	Boraginaceae	PH	0	0^{ns}	–	–
Pozoa coriacea	Apiaceae	PH	0	0^{ns}	–	–
*Taraxacum officinale**	Asteraceae	PH	37	40.7^{ns}	6.3	5.7^{ns}
* non-native species						

germination responses.

We selected three months for stratification because it is the minimum amount of time that seeds experience snow cover in the field, especially those from the low elevation vegetation belt. In addition, other studies have proved that this period is enough to promote germination in alpine species with physiological dormancy requiring cold stratification to initiate germination (e.g., Schwienbacher et al., 2011; Sommerville et al., 2013; Hoyle et al., 2015; Fernández-Pascual et al., 2017). Although 4 °C is a higher temperature than those recorded below the snow cover (that usually fluctuate around 0 °C, Körner, 2003), we chose this temperatures because it is an efficient and widely used treatment to the break the seed dormancy in many species (Bewley et al., 2013; Baskin and Baskin, 2014). In addition, several studies addressing the role of cold stratification in the seed germination of alpine species have used temperatures between 3 and 5 °C for the cold stratification allowing our results to be comparable with those studies (e.g. Kaye, 1997; Liu et al., 2011; Schwienbacher et al., 2011; Sommerville et al., 2013; García-Fernández et al., 2015; Fernández-Pascual et al., 2017).

For controls, we also followed standard protocols where other group of 200 clean seeds per species was air-dried for 48 h. Then, they were placed into paper bags (without substrate), and stored in plastic boxes (20 × 15 × 6 cm) at room temperature (20 °C) for three months, allowing the maintenance of low moisture content in the seeds as it occurs when they are dispersed from the mother plants (Bewley et al., 2013). We used these control conditions because we wanted to assess the importance of to experience constant low temperature and not to compare germination responses mimicking sites with and without snow cover.

2.3. Germination assays

After three months, cold stratification and control boxes were open to initiate seed germination assays. For each species and experimental condition, 50 seeds were placed in a Petri dish between two layers of wet filter paper Petri dishes (4 replicates per species and condition) were arranged in a growth chamber set with a thermoperiod of 20°/10 °C (day/night) and a photoperiod of 14/10 h (light/dark). We chose these germination conditions because they had produced good results in seed germination experiments with other alpine plant species from many different mountains (Chabot and Billings, 1976; Cavieres and Arroyo, 2000; Liu et al., 2013; Sommerville et al., 2013; Fernández-Pascual et al., 2017). During the germination assay seeds were checked every two days for germination, and germinated seeds were removed from the Petri-dishes to reduce mistakes. Seeds were scored as germinated if the radicle and at least 1 mm of the green epicotyls or cotyledons were visible (Schütz, 2002). Germination assays lasted 45 days.

After the end of the germination assay, non-germinated seeds were placed on moist filter papers at room temperature and sliced along its longitudinal axis with a scalpel. Both seed sections were incubated in a 0.1% aqueous solution of Tetrazolium chloride for 12 h in darkness to assess their viability (Hendry and Grime, 1993). Seeds showing a strong red-stained embryo were considered viable (Appendix A in Supplementary material).

2.4. Seed germination parameters and statistical analyses

For each replicate per species we calculated the final percentage of seed germination (FG) both for control and cold-stratified seeds. The FG was arcsine transformed to improve normality and stabilize variances. Statistical differences for FG between treatments were assessed for each species with one-way analysis of variance (ANOVA).

In addition, for each replicate per species a coefficient of velocity of germination (CVG) was calculated with and without cold stratification as follow:

$$CVG = 100[\Sigma N_i / \Sigma N_i T_i]$$

Where N_i is the number of seeds germinated on day i and T_i is the number of days elapsed from the start of the experiment (Scott et al., 1984). CVG increases as more seeds germinate in a shorter time, indicating how fast germination occurs (Scott et al., 1984). Given that assumptions for parametric tests were not met even after data transformations, significant differences for CVG between treatments were assessed with Mann-Whitney *U* tests.

To assess the altitudinal variation in the importance of cold stratification for germination we conducted a two-way ANOVA using the FG as the response variable, and vegetation belt (Low, Mid and High) and treatment (with and without cold stratification) as fixed factors. In the case of CVG, statistical differences were evaluated with a Kruskal-Wallis ANOVA by ranks. Further, three indicators of germination at the community level were calculated for each vegetation belt: 1) the percentage of species that did not require cold stratification for germination, 2) the percentage of species that germinated after a cold-stratification period, and 3) the percentage of species where seed germination was significantly increased (final percentage or speed of germination) after cold stratification. These three indicators were statistically compared among altitudinal vegetation belts with multiple proportions tests (Zar, 1999).

3. Results

In the Low-Andean belt, eight out of the eighteen tested species showed some germination without cold stratification, with very low FG (< 10%) in four of them (Table 1). After cold stratification however, fifteen species germinated and only two showed FG < 10% (Table 1). Two species (*Diplolepis mucronata* and *Sisyrinchium arenarium*) did not germinate during the evaluated period neither with nor without cold stratification, whereas *Anarthrophyllum cumingii* germinated only without cold stratification. Eleven of the fifteen species that germinated after cold stratification significantly increased their FG compared with no-stratification (Table 1). Among the seven species that germinated both with and without stratification, six significantly increased their CVG after cold stratification (Table 1), indicating that in these species germination was faster in cold-stratified seeds.

In the Mid-Andean belt, only four out of the thirteen studied species showed seed germination without cold stratification, and only two of them with FG > 10% (Table 1). Seeds of ten species germinated after cold stratification, where seven of them showed a FG > 10% (Table 1). Only *Calceolaria purpurea, Schizanthus grahamii* and *Euphorbia collina*, did not germinate in any of the evaluated conditions. Viability tests showed that only the seeds of *E. collina* were non-viable (see Appendix A in Supplementary material). Cold stratification significantly increased the FG of six species (Table 1). Only three out of the four species that germinated both with and without stratification significantly increased their CVG after cold stratification (Table 1).

In the High-Andean belt, only six out of the nineteen species tested showed some seed germination without cold stratification (Table 1). After cold stratification, seeds of twelve species germinated (Table 1), although most of these species showed FG < 60% (Table 1). Seven species did not germinate during the germination assays (Table 1), viability test however revealed that all those seeds that did not germinate were viable (Appendix A in Supplementary material). Cold stratification significantly increased FPSG in seeds of six species, whereas CVG slightly increased in *Draba gilliesii* seeds but it decreased in *Cerastium arvense* seeds (Table 1).

When species were grouped by vegetation belt, there was not effect of vegetation belt on the final seed germination ($F_{2,127} = 2.2$, $P = 0.34$), but cold stratification significantly affected seeds germination ($F_{2,127} = 18.1$, $P < 0.0001$; Fig. 1). Whilst the mean FG of cold-

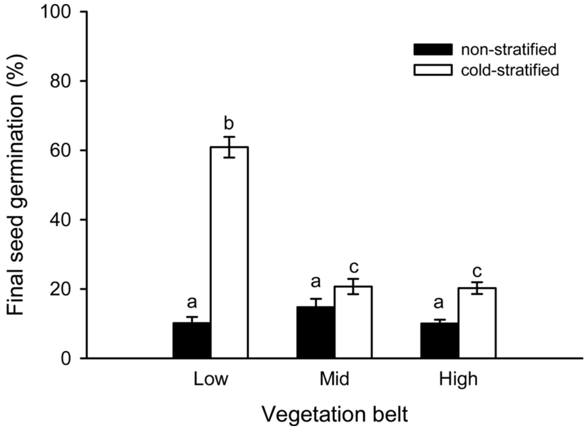


Fig. 1. Final Percentage of seed germination of alpine plant species from the central Chilean Andes. Values are shown as mean ± 2 standard error (n = 13–19 species). Vegetation belts correspond to **Low-Andean**, 2100–2500 m; **Mid-Andean**, 2600–3200 m; and **High-Andean**, 3200–3600 m above sea level. Lowercase letters indicate significant differences ($P < 0.05$).

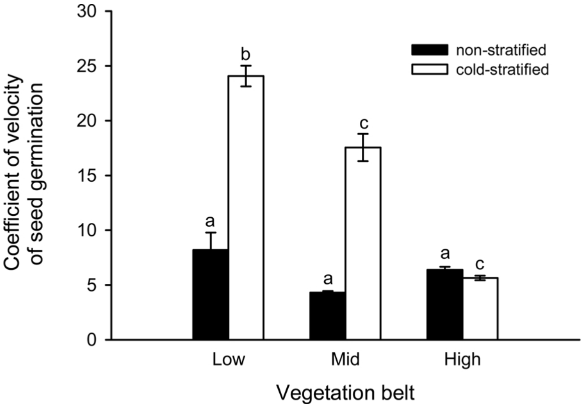


Fig. 2. Coefficient of velocity of seed germination of alpine plant species from the central Chilean Andes. Values are shown as mean ± 2 standard error (n = 13–19 species). Vegetation belts correspond to **Low-Andean**, 2100–2500 m; **Mid-Andean**, 2600–3200 m; and **High-Andean**, 3200–3600 m above sea level. Lowercase letters indicate significant differences ($P < 0.05$).

stratified seeds did not differ between species from the Mid- and High-Andean belts, those from the Low-Andean belt showed a significantly greater FG (*c.* three-fold; Fig. 1). Likewise, while the CVG of non-cold stratified seeds was similar among vegetation belts ($H_{2,56} = 2$, $P = 0.44$), after cold stratification the germination of seeds decreased with elevation ($H_{2,56} = 6.8$, $P < 0.05$; Fig. 2).

The percentage of species that germinated without cold stratification was similar among the three altitudinal vegetation belts (35% on average; Fig. 3a). In contrast, the percentage of species that germinated after a cold-stratification period was 32% greater in the Low- compared to the High-Andean belt, whereas the Mid-Andean belt showed no difference with the two others vegetation belts (Fig. 3b). Likewise, the percentage of species that increased their seeds germination after a cold-stratification period was almost twice in the Low- compared to the High-Andean belt, whilst the Mid-elevation belt showed no differences with the two others (Fig. 3c).

4. Discussion

Although one third of the studied species showed seed germination without cold-stratification, after three months of cold stratification a higher amount of species showed germination, and there were important increases in the final number of seeds germinated. Thus, our results suggest that, in general, a cold and wet period is important for promoting seed germination in alpine plant species from the central Chilean Andes. This is in line with several other studies that have shown that a cold-stratification period is important for seed germination of different alpine plant species (Cavieres and Arroyo, 2000; Schütz, 2002; Shimono and Kudo, 2005; Giménez-Benavides et al., 2005; Sommerville et al., 2013; García-Fernández et al., 2015; Hoyle et al., 2015; Fernández-Pascual et al., 2017). In the field, the cold stratification of the seeds naturally occurs when they are in the soil covered with snow during the winter, experiencing constant temperatures around 0–1 °C. Although we chose a higher temperature for our cold-stratification (4 °C) compared to that the seeds may experience in field, stratification temperatures of 3–5 °C are widely used in seed germination studies because they break the dormancy in many different species (Bewley et al., 2013; Baskin and Baskin, 2014). Although temperatures close to 0 °C would have been more realistic and has been used for cold-stratification of seeds in studies with alpine plant species (Schütz, 2002; Shimono and Kudo 2005; Milbau et al., 2009), the majority of studies addressing the role of cold stratification in the seed germination of alpine species have used temperatures between 3 and 5 °C for the cold stratification, (e.g. Kaye, 1997; Giménez-Benavides et al., 2005; Liu et al., 2011; Schwienbacher et al., 2011; Sommerville et al., 2013; García-Fernández et al., 2015) allowing our results to be comparable with all of them. Further, as the maximum density of pure water is reached at 4 °C, stratification temperatures between 0 and 5 °C may have similar effects on the germination of seeds. Thus, is seems likely that a cold stratification period *sensu lato* is more important to trigger germination than its temperature *per se.* Nevertheless, a note of caution has to be considered as for both stratified and control seeds we did not use temperatures as those that seeds experience in the field. Thus, further studies are needed considering field temperatures from exposed and snow protected areas to gain insights on the importance of different duration of cold stratification and the mechanisms involved.

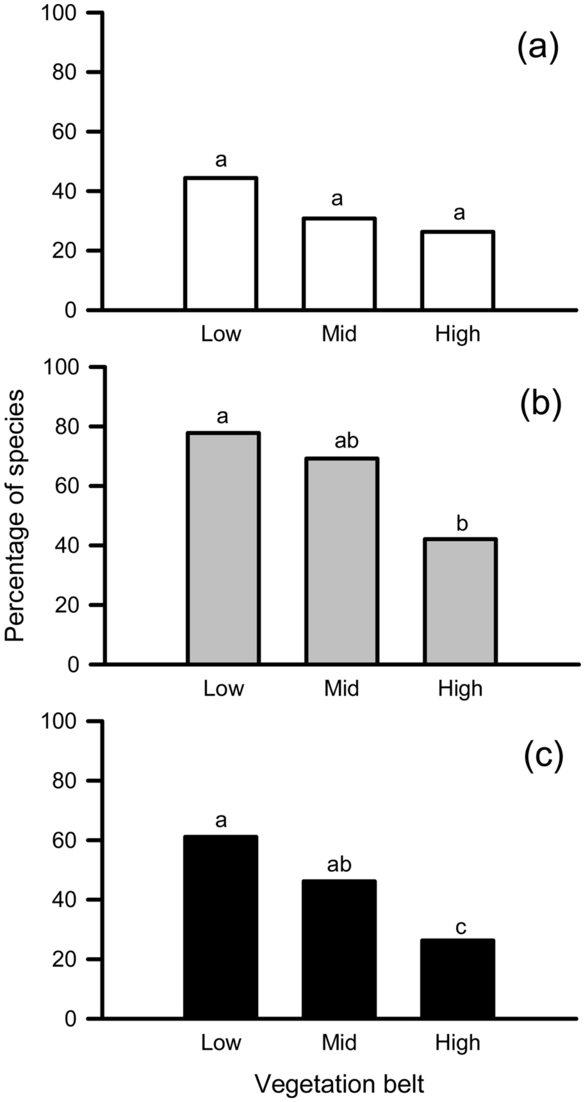


Fig. 3. Altitudinal variation of the importance of cold stratification for seed germination in alpine plant species of the central Chilean Andes. Vegetation belts correspond to **Low-Andean**, 2100–2500 m; **Mid-Andean**, 2600–3200 m; and **High-Andean**, 3200–3600 m above sea level. **a**) Percentage of species whose seeds did not require cold-stratification for germination; **b**) Percentage of species whose seeds germinated after cold-stratification; and **c**) Percentage of species whose seeds significantly increased their germination after cold-stratification. Different lowercase letters indicate significant differences ($P < 0.05$).

The importance of cold stratification for the seed germination of alpine species of the central Chilean Andes varied across the altitudinal vegetation belts, but in contrast to our expectations, the positive effect of cold stratification on seeds germination did not increase with altitude. The percentage of species showing seeds germination after the cold stratification period decreased from 83% at the Low-Andean to 63% at the High-Andean vegetation belt. Moreover, the positive effect of the cold stratification in the final percentage of germination and the coefficient of velocity were greater on seeds of species from the lower Andean vegetation belt. Several not mutually exclusive explanations can be suggested to account for this interesting altitudinal trend.

On one hand, it has been shown that a cold stratification period can synchronize germination (Schütz, 2002; Baskin and Baskin, 2014). One of the main sources of seedlings mortality in alpine habitats is drought (Bell and Bliss, 1980; Stöcklin and Bäumler, 1996; Forbis, 2003; Briceño et al., 2015). The central Chilean Andes are located in a Mediterranean-type climate zone where the lower elevations of the Andes experience the long summer drought that characterizes this climate type zone, and where this drought is an important factor of seedling mortality (Cavieres et al., 2006, 2007). The frequent occurrence of summer storms at high elevations implies that the summer drought is much less severe at higher elevations (op. cit). Hence, faster and more abundant seed germination immediately after snow melts might assure a longer period with soil water availability enabling seedlings to develop deeper roots and to attain a critical biomass to successfully cope with the dry summer conditions (see also Giménez-Benavides et al., 2005). In our study area, seedlings density decreased through the growing season, but only at the lower elevations (see Appendix B in Supplementary material), where this seedlings mortality coincides with a decrease of the soil water potentials below −3 MPa towards the second half of the growing season (Jan–Mar; Sierra-Almeida et al., 2009). Although seedlings emergence in early spring could increase mortality risk due to the freezing temperatures that are frequent in the first part of this period, seedlings of the great majority of these species are able to resist very low temperatures (Sierra-Almeida et al., 2010). Therefore, cold stratification requirements would be crucial for species at low elevations because it allows seed germination to occur immediately after the snow-melts when soil moisture is high, increasing the chances of a successful seedlings establishment despite the occurrence of freezing temperatures.

On the other hand, the low positive effect of cold stratification on seed germination found on species from high-Andean belt could be related with additional requirements for seed germination. Although cold stratification produced seed germination in 63% of the species at the High-Andean belt, the average germination was lower than 20%, and no changes in the coefficient of velocity were observed after cold stratification. Interestingly, the seeds of these species that did not germinate remained viable (Appendix A in Supplementary material), suggesting a deeper dormancy that was not broke after three months of cold stratification. Many of the studies that report increased seed germination of alpine species after cold stratification have used stratification periods of 1–3 months (e.g., Shimono and Kudo, 2005; Giménez-Benavides et al., 2005; Sommerville et al., 2013). Nonetheless, several studies have found that species from sites where the snow cover lasts for long periods need longer cold stratification periods for germination than species from sites with shorter duration of snow cover (Meyer and Monsen, 1991; Meyer et al., 1989, 1990, 1995; Schütz and Rave, 1999; Cavieres and Arroyo, 2000), although other studies have found no relationship (Schütz and Milberg, 1997; Schütz,2002). The three months of cold-stratification used in this study corresponded to the average duration of snow cover in the Low- and Mid-Andean vegetation belt. Snow duration in the High-Andean belt can span from 3 to 5 months, suggesting that longer cold-stratification periods may enhance germination. However, Cavieres and Arroyo (2000) studied the seed germination of *Phacelia secunda* populations at 2900 and 3400 m a.s.l. in the same study area, and found that there were no important differences in the germination after 3 or 5 months of cold stratification in this species. In addition, an assessment with six species from the High-Andean belt that were exposed to three and five months of cold stratification and then exposed to the same germination assay conditions reported here, showed that only in two species a longer stratification period increased germination (see Appendix C in Supplementary material). Hence, it seems unlikely that longer stratification periods could enhance germination on seeds of the species from the high-Andean belt, suggesting that seeds of these species need other factors to overcome dormancy. Among the factors that overcome dormancy, physical scarification has been found to increase germination in some alpine species (Amen, 1966; Baskin and Baskin, 2014). For instance, Bell and Amen (1970) reported the presence of germination inhibitors in the seed testa of *Luzula spicata* and *L. parviflora*, which were liberated after mechanical scarification through repeated freeze/thawing of water in the testa (cryofracturation). Dorne (1981) found that seed dormancy in seeds of the alpine *Chenopodium bonus-henricus* was imposed by phenols present in the testa, which also required a combination of low temperature, humidity and cryoturbation to disable its inhibitory action on germination. Cavieres and Arroyo (2001) also suggested that scarification via cryoturbation could be involved in the dormancy release of *Phacelia secunda* seeds from very high elevations in the Chilean Andes. Further work is needed to unravel the dormancy mechanisms involved in species from the highest elevations of the central Chilean Andes.

As the species of the High Andean belt showed low germination compared to the Low Andean belt but the ungerminated seeds remained viable, our results also suggest that deep seed dormancy becomes more important towards higher elevations. Similar results have been reported by Bu et al. (2007) who studied the seed germination of 570 alpine plant species collected at different elevations in the Tsinghai-Tibet plateau, China, reporting a negative correlation between germination percentage (but still viable seeds) and elevation. It has been proposed that seed dormancy would be advantageous in habitats spatially and temporary unpredictable such as alpine habitats (Cavieres, 1999; Schwienbacher et al., 2011; Jaganathan et al., 2015). In alpine habitats, seed germination depends on the seeds produced in the previous growing seasons (Körner, 2003). Unseasonably cold years and/or short growing seasons can produce strong reductions in seed output, while also decreasing seed germination and seedling establishment (Bliss, 1985; Galen and Stanton, 1999). If a long and favorable growing season was preceded by a very short growing season where species did not produce seeds, only species that remained dormant in the soil will have the possibility to germinate and recruit (Cavieres, 1999; Jaganathan et al., 2015). Thus, the deep dormancy found here in the higher elevations species as well as in other alpine species (e.g., Schwienbacher et al., 2011; Baskin and Baskin, 2014) suggests that this strategy is very important to cope with the interannual fluctuation of the growing season length in high-elevation environments.

In conclusion, our exploratory study suggest that cold stratification is important to release seed dormancy in alpine plant species from the high-Andes of central Chile, and it seems to be particularly important in most of the species from lower elevations, increasing their final seed germination and the velocity of this process. This strategy seems to be related with a synchronization of seed germination with a period when soil moisture is not limiting for seedling growth. Moreover, the lower percentage of species that germinate after cold stratification, the small positive effect of cold stratification on the seed germination, and the high viability of the seeds that no germinated in these species suggest that break dormancy mechanisms other than cold stratification are required. However, further research is needed to unravel the dormancy mechanisms of these high elevation species and their importance in assuring a successful establishment in the high-Andes of central Chile. In addition, germination trials that properly simulate the environmental conditions that seeds experience in the field (e.g. duration of snow cover, temperature to which seeds are exposed post-dispersal, etc.) are needed to increase our understanding of the mechanisms involved in this important process (see Briceño et al., 2015 for further suggestions). Global climate change is causing important changes in the snow cover duration in several alpine areas around the world (Beniston, 1997, 2012; Gobiet et al., 2014), threatening those alpine species that requires cold stratification to initiate their seed germination, and indirectly favoring those with no such requirement (Sommerville et al., 2013). The species inhabiting the lower elevations of alpine areas are those that naturally experience shorter snow cover periods, and those where the major changes in snow cover duration have been observed during the last decades, and also those where the major changes are expected (Beniston, 1997, 2012; Gobiet et al., 2014). Thus, major impacts on the natural regeneration process of the species in the Low-Andean vegetation belt may be expected with the current trends in climate change.

Acknowledgements

We thank Ana María Humaña, Carol Peña, and María José Parra for their help in the experiments and the staffs of La Parva and Valle Nevado Ski Resorts for logistic help and the permission for the use of private roads. This work was funded by FONDECYT Projects1030821, 1060710, 1130592 and F ICM 05-002 and CONICYT PFB 0-23 supporting the Institute of Ecology and Biodiversity.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.ppees.2017.09.005.

References

Amen, R.D., 1966. The extent and role of seed dormancy in alpine plants. Q. Rev. Biol. 41, 271–281.

Angevine, M.W., Chabot, B.F., 1979. Seed germination syndromes in higher plants. In: Solbrig, O.T., Jain, S., Johnson, G.B., Raven, P. (Eds.), Topics in Plant Population Biology. Columbia University Press, New York, pp. 188–206.

Baskin, C.C., Baskin, J.M., 2014. Seeds: Ecology, Biogeography, and Evolution of Dormancy and Germination, second ed. Academic Press, New York.

Bell, K., Amen, R., 1970. Seed dormancy in *Luzula spicata* and *L. parviflora*. Ecology 51, 492–496.

Bell, K.L., Bliss, L.C., 1980. Plant reproduction in high arctic environment. Arctic Alpine Res. 12, 1–10.

Beniston, M., 1997. Variation of snow depth and duration in the Swiss Alps over the last 50 years: links to changes in large-scale climatic forcings. Clim. Change 36, 281–300.

Beniston, M., 2012. Is snow in the Alps receding or disappearing? WIREs Clim. Change 3, 349–358.

Bewley, J.D., Bradford, K.J., Hilhorst, H.W., Nonogaki, H., 2013. Seeds: Physiology of Development, Germination and Dormancy, 3rd ed. Springer, New York.

Billings, W.D., Mooney, H.A., 1968. The ecology of arctic and alpine plants. Biol. Rev. 43, 481–529.

Bliss, L.C., 1985. Alpine. In: Billings, W.D., Mooney, H.A. (Eds.), Physiological Ecology of North American Plant Terrestrial Communities. Chapman & Hall, New York, pp. 41–65.

Briceño, V.F., Hoyle, G.L., Nicotra, A.B., 2015. Seeds at risk: how will a changing alpine climate affect regeneration from seeds in alpine areas? Alpine Bot. 125, 59–68.

Bu, H., Chen, X., Xu, X., Liu, K., Jiu, P., Du, G., 2007. Seed mass and germination in an alpine meadow on the eastern Tsinghai-Tibet plateau. Plant Ecol. 191, 127–149.

Cavieres, L.A., Arroyo, M.T.K., 2000. Seed germination response to cold stratification period and thermal regime in *Phacelia secunda* (Hydrophyllaceae): altitudinal variation in the Mediterranean Andes of central Chile. Plant Ecol. 149, 1–8.

Cavieres, L.A., Arroyo, M.T.K., 2001. Persistent soil seed banks in *Phacelia secunda* (Hydrophyllaceae): experimental detection of variation along an altitudinal gradient in the Andes of central Chile (33°S). J. Ecol. 89, 31–39.

Cavieres, L.A., Peñaloza, A., Arroyo, M.T.K., 2000. Altitudinal vegetation belts in the high Andes of central Chile (33°S). Rev. Chil. Hist. Nat. 73, 331–344.

Cavieres, L.A., Badano, E.I., Sierra-Almeida, A., Gómez-González, S., Molina-Montenegro, M.A., 2006. Positive interactions between alpine plant species and the nurse cushion plant *Laretia acaulis* do not increase with elevation in the Andes of central Chile. New Phytol. 169, 59–69.

Cavieres, L.A., Badano, E.I., Sierra-Almeida, A., Molina-Montenegro, M., 2007. Microclimatic modifications of cushion plants and their consequences for seedling survival of native and non-native plants in the high-Andes of central Chile. Arc. Antarct. Alp. Res. 39, 229–236.

Cavieres, L.A., 1999. Bancos de semillas persistentes: modelos de germinación retardada y su aplicación en ambientes alpinos. Rev. Chil. Hist. Nat 72, 457–466.

Chabot, B., Billings, W., 1976. Origin and ecology of the Sierran alpine vegetation. Ecol. Monogr. 42, 163–199.

Chambers, J.C., MacMahon, J.A., Brown, R.W., 1990. Alpine seedling establishment: the influence of disturbance type. Ecology 71, 1323–1341.

di Castri, F., Hajek, E., 1976. Bioclimatología de Chile. Ediciones de la Pontificia Universidad Católica de Chile, Santiago.

Dorne, A., 1981. Variation in seed germination inhibition of *Chenopodium bonus-henricus* in relation to altitude of plant growth. Can. J. Bot. 59, 1893–1901.

Fernández-Pascual, E., Jiménez-Alfaro, B., Bueno, Á., 2017. Comparative seed germination traits in alpine and subalpine grasslands: higher elevations are associated with warmer germination temperatures. Plant Biol. 19, 32–40.

Forbis, T.A., 2003. Seedling demography in an alpine ecosystem. Am. J. Bot. 90, 1197–1206.

Galen, C., Stanton, M., 1999. Seedling establishment in alpine buttercups under experimental manipulations of growing-season length. Ecology 80, 2033–2044.

García-Fernández, A., Escudero, A., Lara-Romero, C., Iriondo, J.M., 2015. Effects of the duration of cold stratification on early life stages of the Mediterranean alpine plant *Silene ciliata*. Plant Biol. 17, 344–350.

Giménez-Benavides, L., Escudero, A., Pérez-García, F., 2005. Seed germination of high mountain Mediterranean species: altitudinal, interpopulation and interannual variability. Ecol. Res. 20, 433–444.

Gobiet, A., Kotlarski, S., Beniston, M., Heinrich, G., Rajczak, J., Stoffel, M., 2014. 21 st century climate change in the European Alps − a review. Sci. Total Environ. 493, 1138–1151.

Hendry, G.A., Grime, J.P., 1993. Methods in Comparative Plant Ecology: A Laboratory Manual. Chapman and Hall, London.

Hoyle, G.L., Steadman, K.J., Good, R.B., McIntosh, E.J., Galea, L.M.E., Nicotra, A.B., 2015. Seed germination strategies: an evolutionary trajectory independent of vegetative functional traits. Front. Plant Sci. 12. http://dx.doi.org/10.3389/fpls.2015.00731.

Jaganathan, G.K., Dalrymple, S.E., Liu, B., 2015. Towards an understanding of factors controlling seed bank composition and longevity in the alpine environment. Bot. Rev. 81, 70–103.

Körner, C., 2003. Alpine Plant Life, second ed. Springer-Verlag, Berlin.

Kaye, T., 1997. Seed dormancy in high elevation plants: implications for ecology and restoration. In: Kaye, T.N., Liston, A., Love, R.M., Luoma, D.L., Meinke, R.J., Wilson, M.V. (Eds.), Conservation and Management of Native Plants and Fungi. Native Plant Society of Oregon, Corvallis, Oregon, pp. 115–120.

Liu, K., Baskin, J.M., Baskin, C.C., Bu, H., Liu, M., Liu, W., Du, G., 2011. Effect of storage conditions on germination of seeds of 489 species from high elevation grasslands of the eastern Tibet Plateau and some implications for climate change. Am. J. Bot. 98, 12–19.

Liu, K., Baskin, J.M., Baskin, C.C., Bu, H., Du, G., Ma, M., 2013. Effect of diurnal fluctuating versus constant temperatures on germination of 445 species from the Eastern Tibet plateau. PLoS One 8 (7), e69364.

Marchand, P.J., Roach, D.A., 1980. Reproductive strategies of pioneering alpine species: seed production, dispersal and germination. Arctic Alpine Res. 12, 137–146.

Meyer, S.E., Monsen, S.B., 1991. Habitat-correlated variation in mountain big sagebrush (*Artemisia tridentata ssp. vaseyana) seed germination patterns*. Ecology 72, 739–742.

Meyer, S.E., McArthur, E.D., Jorgensen, G.L., 1989. Variation in germination response to temperature in rubber rabbitbrush (*Chrysothamnus nauseosus*: Asteraceae) and its ecological implications. Am. J. Bot. 76, 981–991.

Meyer, S.E., Monsen, S.B., McArthur, E.D., 1990. Germination response of *Artemisia tridentata* (Asteraceae) to light and chill: patterns of between-population variation. Bot. Gaz. 151, 176–183.

Meyer, S.E., Kitchen, S.G., Carlson, S.L., 1995. Seed germination timing patterns in intermountain *Penstemon* (Scrophulariaceae). Am. J. Bot. 82, 377–389.

Milbau, A., Graae, B.J., Shevtsova, A., Nijs, I., 2009. Effects of a warmer climate on seed germination in the subarctic. Ann. Bot. 104, 287–296.

Rozzi, R., Molina, J.D., Miranda, P., 1989. Microclima y períodos de floración en laderas de posición ecuatorial y polar en los Andes de Chile central. Rev. Chil. Hist. Nat. 62, 75–84.

Santibáñez, F., Uribe, M., 1990. Atlas Agroclimático de la V Región y Región Metropolitana Ediciones Facultad de Agronomía. Universidad de Chile, Santiago.

Sayers, R.L., Ward, R.T., 1966. Germination responses in alpine species. Bot. Gaz. 127, 11–16.

Schütz, W., Milberg, P., 1997. Seed dormancy in *Carex canescens*, regional differences and ecological consequences. Oikos 78, 420–428.

Schütz, W., Rave, G., 1999. The effect of cold stratification and light on the seed germination of temperate sedges (*Carex*) from various habitats and implications for regenerative strategies. Plant Ecol. 144, 215–230.

Schütz, W., 2002. Dormancy characteristics and germination timing in to alpine *Carex* species. Basic Appl. Ecol. 3, 125–134.

Schwienbacher, E., Navarro-Cano, J.A., Neuner, G., Erschbamer, B., 2011. Seed dormancy in alpine species. Flora 206, 845–856.

Scott, S.J., Jones, R.A., Williams, W.A., 1984. Review of data analysis methods for seed germination. Crop Sci. 24, 1192–1199.

Shimono, Y., Kudo, G., 2005. Comparisons of germination traits of alpine plants between fellfield and snowbed habitats. Ecol. Res. 20, 189–197.

Sierra-Almeida, A., Cavieres, L.A., Bravo, L.A., 2009. Freezing resistance varies within the growing season and with elevation in high-Andean species of central Chile. New Phytol. 182, 461–469.

Sierra-Almeida, A., Cavieres, L.A., Bravo, L.A., 2010. Freezing resistance of high-elevation plant species is not related to their height or growth-form in the Central Chilean Andes. Environ. Exp. Bot. 69, 273–278.

Sommerville, K.D., Martyn, A.J., Offord, C.A., 2013. Can seed characteristics or species distribution be used to predict the stratification requirements of herbs in the Australian Alps? Bot. J. Linn. Soc. 172, 187–204.

Stöcklin, J., Bäumler, E., 1996. Seed rain, seedling establishment and clonal growth strategies on a glacier foreland. J. Veg. Sci. 7, 45–56.

Vleeshouwers, L.M., Bouwmeester, H.J., Karssen, C.M., 1995. Redefining seed dormancy: an attempt to integrate physiology and ecology. J. Ecol. 83, 1031–1037.

Walder, T., Erschbamer, B., 2015. Temperature and drought drive differences in germination responses between congeneric species along altitudinal gradients. Plant Ecol. 216, 1297–1309.

Willis, C.G., Baskin, C.C., Baskin, J.M., Auld, J.R., Venable, D.L., Cavender-Bares, J., Donohue, K., Rubio de Casas, R., 2014. The evolution of seed dormancy: environmental cues, evolutionary hubs, and diversification of the seed plants. New Phytol. 203, 300–309.

Zar, J., 1999. Biostatistical Analysis, fourth ed. Prentice Hall, New Jersey.

Perspectives in Plant Ecology, Evolution and Systematics

Editors-in-Charge
Peter J. Edwards, Zürich, Switzerland, e-mail: peter.edwards@env.ethz.ch
Claus Holzapfel, Newark, NJ, USA, e-mail: holzapfe@andromeda.rutgers.edu
Elvira Hörandl, Göttingen, Germany, e-mail: ehoeran@uni-goettingen.de
Florian Jeltsch, Potsdam, Germany, e-mail: jeltsch@uni-potsdam.de
Diethart Matthies, Marburg, Germany, e-mail: matthies@staff.uni-marburg.de
Kirk A. Moloney, Ames, IA USA, e-mail: kmoloney@iastate.edu
George Perry, Auckland, New Zealand, e-mail: george.perry@auckland.ac.nz

Managing Editor
Silke Dietz, Eidgenössische Technische Hochschule (ETH) Zürich,
Department of Environmental Systems Science,
Zürich, Switzerland
e-mail: ppeesjournal@gmail.com

Editors
Dana Blumenthal, Fort Collins, CO, USA
Regine Claßen-Bockhoff, Mainz, Germany
Rob Colautti, Queens University, Canada
Birgit Gemeinholzer, University Giessen, Germany
Itamar Giladi, Ben Gurion University of the Negev, Israel
Antoine Guisan, Lausanne, Switzerland
Erik Hamerlynck, Burns, OR, USA
Tomas Herben, Academy of Sciences of the Czech Republic, Pruhonice, Czech Republic
Hans Jacquemyn, Leuven, Belgium
Jasmin Joshi, Potsdam, Germany
Gudrun Kadereit, Mainz, Germany
Marcus Koch, Heidelberg, Germany
Alexander Kocyan, Potsdam, Germany
Robert H. Marrs, Liverpool, UK
Patrick G. Meirmans, Amsterdam, The Netherlands
Christer Nilsson, Umeå, Sweden
Anibal Pauchard, University of Concepcion, Chile
Roberto Salguero-Gomez, Oxford, UK
Peter Schönswetter, Innsbruck, Austria
Alfredo Valido, Spanish National Research Council, Spain
Joseph Veldman, Texas, USA
Joseph W. Veldman, Iowa State University, Ames, Iowa, USA
Mark F. Watson, Edinburgh, UK
Kerstin Wiegand, Göttingen, Germany
Gerhard Zotz, Oldenburg, Germany