

SPECIAL FEATURE

FACILITATION IN PLANT COMMUNITIES

Do facilitative interactions increase species richness at the entire community level?

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Summary

1. Although the consequences of facilitation at individual and population levels are well known, the community-level consequences of these processes have received much less attention. In particular, the importance of facilitation in determining richness at the entire community level has seldom been evaluated.

2. In this study, we sampled 11 alpine plant communities along the southern Andes in South America, spanning from tropical (25°S) to sub-antarctic latitudes (55°S). Plant communities were dominated by cushion plants, a particular growth form that acts as a nurse plant for other plant species. Through rarefaction curves, we assessed the effectiveness of community sampling and estimated the number of species present within and outside cushions. Non-metric Multidimensional Scaling ordinations (NMDS) were used to assess differences between the species assemblages growing within and outside cushions. Finally, samples from cushions and open areas were combined in a single matrix accounting for the difference in cover between both microhabitats, and through rarefaction curves we assessed how many more species are added to the community due to the presence of cushions.

3. Samples taken within cushions always contained more species than equivalent samples from open areas. However, the magnitude of this difference varied among communities. NMDS ordination indicated that cushions generate species assemblages structurally different from those found in open areas. Inclusion of samples from cushion and open areas in synthetic analyses – where differences in cover were accounted for – indicated that the presence of cushions consistently increased species richness at the entire community level. The magnitude of these increases in species richness varied with habitat severity, with lower values at both extremes of the environmental severity gradient.

4. *Synthesis.* Facilitative interactions with cushion nurse plants along the high Andes of southern South America changed plant assemblage structure and increased species richness at the entire community level, indicating that facilitative interactions are pivotal in maintaining the diversity of these harsh environments.

Key-words: alpine habitats, alpine plant communities, Andes, community-level interactions, cushion plants, facilitation, positive interactions, stress gradient hypothesis

Introduction

Positive interactions are defined as non-trophic interspecific interactions that increase the average individual fitness of one species (Callaway 2007). The presence of one plant species enhances the chances that another species co-occurs in the

same place, and hence the positive interactions may influence the regulation of biological diversity. Although early ecological research focused on negative interactions (e.g. competition, predation) as the main factors structuring plant communities and regulating biological diversity (e.g. Connell 1978; Grime 1973; Tilman 1982), more recent studies have pointed out the importance of positive interactions for diversity and community structure (Hacker & Gaines 1997; Bruno, Stachowicz &

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Bertness 2003; Michalet *et al.* 2006). Indeed, recent theoretical work has shown that positive interactions among resource competitors can produce species-rich model communities supported by a single limiting resource (Gross 2008).

Positive interactions can increase species diversity when the presence of one species modifies the environment so as to reduce the frequency of some physical disturbance or stress, allowing less tolerant species to survive (Hacker & Gaines 1997). The best example of this phenomenon is facilitation by nurse species (i.e. nurse-protégé, Flores & Jurado 2003), where one species (the facilitator, benefactor or nurse plant) provides shelter from physical stress or herbivory to other plant species (i.e. the beneficiary species; Callaway 1995).

Harsh environments may restrict a plant's resource acquisition, and amelioration of these conditions can favour growth to the extent that the benefits accrued outweigh the negative, competitive impact of growing in close association. Based on this, it has been suggested that positive interactions may have strong impacts in harsh environments, where the mitigation of extreme conditions by facilitator species can benefit many other species (Bertness & Callaway 1994; Callaway & Walker 1997; Brooker & Callaghan 1998). Some studies suggest that facilitative interactions do not always increase in importance with abiotic stress in arid and semi-arid habitats (Maestre, Valladares & Reynolds 2005), but the evidence available so far, from a range of stressful habitats, overwhelmingly supports the importance of positive interactions (e.g. Callaway *et al.* 2002; Lortie & Callaway 2006; Brooker *et al.* 2008).

However, depending on the particular physical stress-limiting survival, the mechanisms involved in the facilitation by a nurse-plant species can differ among habitats (for a complete review of mechanisms, see Callaway 2007). The shelter provided by facilitator plants has been shown to increase the physiological performance, population density and reproduction of some particular beneficiary species (e.g. Pugnaire *et al.* 1996; Choler, Michalet & Callaway 2001; Maestre, Bautista & Cortina 2003; Armas & Pugnaire 2005). Nevertheless, despite these well-known effects of nurses at individual and population levels, community-level consequences (i.e. considering all the species present in the community) of facilitation have received much less attention.

The majority of the studies addressing some community-level consequences of facilitation have compared the diversity of the plant assemblages growing beneath nurses with those growing outside them and report contrasting results among them. While the majority of the studies report higher species richness beneath the canopy of nurse species compared with open areas (e.g. Raffaele & Veblen 1998; Badano *et al.* 2005; Larrea-Alcázar, López & Barrientos 2005), others report that nurses contain similar or lower species richness than that found outside them (e.g. Gutiérrez *et al.* 1993; De Villiers, Van Rooyen & Theron 2001; Rossi & Villagra 2003; Pugnaire, Armas & Valladares 2004), indicating that the facilitative effects on some species do not necessarily generate higher species richness beneath nurses. Nonetheless, nurses and their alternative habitats (open areas among nurses) are part of the

same community. Thus, even when nurse plants contain fewer species than open areas, if nurses allow the persistence of species that otherwise would be excluded from the community, then nurses will generate a net increase in the species diversity at the entire community level. However, the simple question of how many species are added to a community due to the presence of facilitator species has been seldom addressed (Tewksbury & Lloyd 2001).

Alpine habitats are well known for their severe conditions; plants are confronted there by low temperatures, short growing seasons, excessive radiation and unstable substrates (Körner 2003). Cushion-forming plants are one of the most conspicuous plant types found in exposed alpine habitats, and several studies have reported plant species growing inside of cushion plants and suggest a facilitative role for them (e.g. Nuñez, Aizen & Ezcurra 1999; Arroyo *et al.* 2003; Cavieres *et al.* 2002, 2006; LeRoux & McGeoch 2008). The low stature and compact form of cushion plants attenuate the effect of extreme environmental conditions, allowing the persistence of other species inside the cushions (Cavieres *et al.* 2006, 2007). In addition, there are basically no other facilitation effects in these communities (Fajardo, Quiroz & Cavieres 2008), and hence by assessing the impacts of cushions in these systems, we can basically look at the impact of facilitation on the whole community. Further, there is a very clear delimitation of areas with and without a facilitation effect (i.e. inside and outside cushions). Thus, alpine habitats dominated by cushion plants are ideal systems to assess the community-level effects of facilitative interactions. In this particular study, we assessed (i) whether cushion plants contained more species than open areas between them, (ii) whether cushions change species abundances generating assemblages structurally different from those found outside of them, and (iii) whether the presence of cushion-dependent species, which otherwise would be absent from a community with no nurses, produces a significant increase in species richness at the entire community level. The assessment of these effects on more than a single community is important for understanding the generality of the community-level consequences of facilitation. For this, we analysed the community-level effects of nine cushion species on 11 high-elevation plant communities dominated by cushions along the high Andes of Chile and Argentina.

Materials and methods

STUDY SITES AND TARGET CUSHION SPECIES

We sampled 11 high-elevation plant communities dominated by cushions at six different latitudes along the high Andes of Chile and Argentina (Fig. 1). In total, we analysed the community-level effects of nine different cushion species. From north to south, we sampled plant communities dominated by: *Pycnophyllum bryoides* at 4400 m a.s.l. in northern Chile (23°S); *Azorella madreporica* and *Adesmia subterranea*, both at 4000 m in north-central Chile (30°S); *A. madreporica* at 3600 m and *Laretia acaulis* at 3200 m, both in central Chile (33°S); *Oreopolus glacialis* at 1900 m in south-central Chile (37°S);

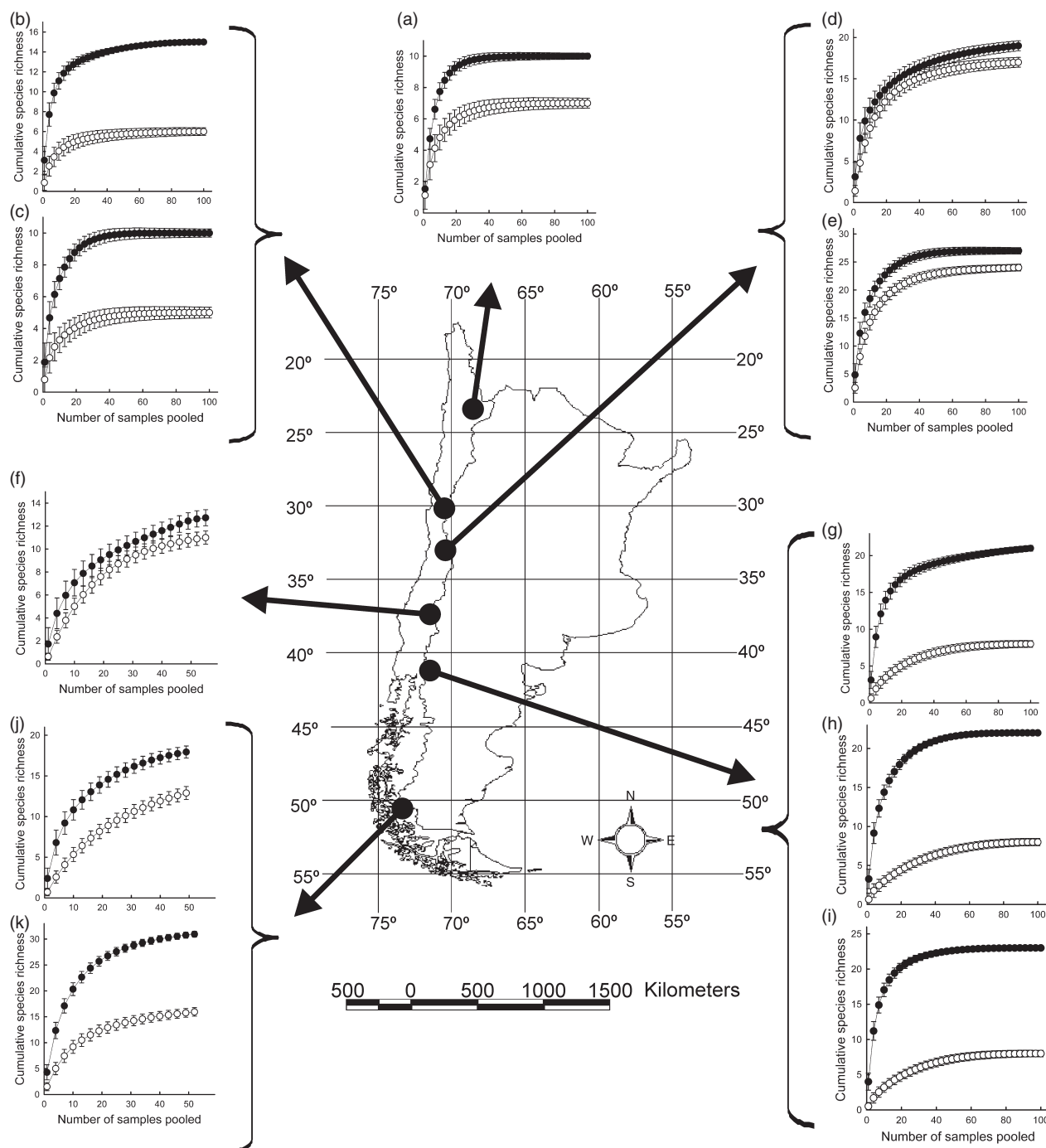


Fig. 1. Map showing the locations of the study sites and the estimated species richness from rarefactions curves at each sampling size ($\pm 95\%$ confidence intervals) for cushions (solid symbols) and open areas (empty symbols) at each study site. (a) *Pycnophyllum bryoides* at 23°S; (b) *Azorella madreporca* at 30°S; (c) *Adesmia subterranea* at 30°S; (d) *Azorella madreporca* at 33°S; (e) *Laretia acaulis* at 33°S; (f) *Oreopolus glacialis* at 37°S; (g) *Mulinum leptacanthum* at 41°S; (h) *Oreopolus glacialis* at 41°S; (i) *Discaria nana* at 41°S; (j) *Bolax gummifera* at 50°S; (k) *Azorella monantha* at 50°S.

Mulinum leptacanthum, *O. glacialis* and *Discaria nana* at 1600 m in southern Argentina (41°S) and *Bolax gummifera* and *Azorella monantha* at 900 m in southern Chile (50°S). All these communities were chosen because of their accessibility, and the sampling sites were located as close as possible to the upper altitudinal limit of cushion distributions. These communities range from sites with low annual precipitation (c. 80 mm) concentrated during the growing season, as found in northern Chile (23°S), to sites with high annual

precipitation (c. 1000 mm) regularly distributed throughout the year, as found in Chilean Patagonia (50°S, see Table 1). Central Chile (30°, 33° and 37°S) is characterized by annual precipitation between 300 and 700 mm from north to south, but most of this is concentrated during the winter months (June–August), generating very dry growing seasons.

In all the sampled communities, cushion plants generate spatially discrete patches isolated from similar units by large open areas. None-

Table 1. Some details of the study sites. The table indicates the cushion species dominating each plant community, the geographical point (latitude and longitude) and elevation of each study site, the total number of samples taken from cushions and open areas, and the estimated percentage cover of each habitat type. The annual precipitation as well as the mean temperature and the precipitation during the growing season are also indicated for each site

Cushion species	Geographical point	Elevation (m)	Number of samples	Cushion cover (%)	Open areas cover (%)	Growing season mean temperature (°C)	Annual precipitation (mm)	Summer precipitation (mm)
<i>Pycnophyllum bryoides</i> Rohrb. (Caryophyllaceae)	23°S; 68°W	4300	100	5	95	8.0	42	39
<i>Adesmia subterranea</i> Clos (Fabaceae)	30°S; 70°W	3700	100	11	89	4.3	148	28
<i>Azorella madreporica</i> Clos (Apiaceae)	30°S; 70°W	4000	100	12	88	2.6	164	34
<i>Azorella madreporica</i> Clos (Apiaceae)	33°S; 70°W	3600	140	10	90	3.9	458	46
<i>Laretia acaulis</i> (Cav.) Gill et Hook. (Apiaceae)	33°S; 70°W	3200	140	34	66	5.6	475	42
<i>Oreopolus glacialis</i> (Poepp. & Endl.) Ricardi (Rubiaceae)	37°S; 71°W	1900	55	10	90	10.1	1199	173
<i>Discaria nana</i> (Gay) Weberb. (Rhamnaceae)	41°S; 71°W	1600	100	13	87	8.6	758	154
<i>Mulinum leptacanthum</i> Phil. (Apiaceae)	41°S; 71°W	1600	100	6	94	8.6	758	154
<i>Oreopolus glacialis</i> (Poepp. & Endl.) Ricardi (Rubiaceae)	41°S; 71°W	1600	100	5	95	8.6	758	154
<i>Azorella monantha</i> Clos (Apiaceae)	50°S; 73°W	900	53	30	70	6.5	1117	469
<i>Bolax gummifera</i> (Lam.) Spreng. (Apiaceae)	50°S; 73°W	900	50	10	90	6.5	1117	469

theless, although cushions were the most conspicuous growth form in all communities, small annual and perennial herbs as well as prostrate shrubs were also present (see Arroyo *et al.* 2003; Badano *et al.* 2002; Cavieres *et al.* 2002, 2006; Badano & Cavieres 2006a,b for further details of some communities).

SAMPLING OF SPECIES

At each site, we randomly selected a number of individual cushions (see Table 1 for details of sample size at each site) and all plants growing within these selected cushions were identified to species level and recorded. Since cushions are usually of elliptical form, we also measured the maximum and minimum axes of each cushion to estimate its area. To obtain comparable samples of species richness in surrounding open areas, the shape of each sampled cushion was emulated with wire rings. These rings were randomly placed in open areas away from cushions and all plant individuals within the wire rings were identified to species level and recorded. Further, the percentage cover of cushions and open areas was determined at each site with 50-m long linear transects (see Badano & Cavieres 2006a,b for further details of sampling).

SPECIES RICHNESS COMPARISONS BETWEEN HABITATS

The assessment of differences in species richness between cushions and open areas requires sampling efforts large enough to fully capture the species assemblages associated with each microhabitat (Gotelli &

Colwell 2001). Therefore, before comparing species richness, we constructed a species accumulation curve for cushions and their respective open areas at each study site. In these analyses, the point along the axis of sampling effort where the species accumulation curve reaches an asymptote indicates the number of samples needed to successfully sample the full assemblage richness (Gotelli & Colwell 2001). Since our sampling protocol included samples of different areas, these curves were constructed by using sample-based rarefaction techniques to avoid biases due to the sequence in which samples were added to the curves. For this, we generated a species \times samples matrix for each habitat type, where each cell (i, j) indicated the presence (1) or absence (0) of the i species in the j sample. From these matrices, 500 resamples with replacement were randomly drawn for each sample size (from one sample to the maximum number of samples), then, these 500 values of species richness were averaged. The average values of species richness were then plotted against the respective sample size to construct a rarefaction curve for each habitat. Further, 95% confidence intervals were estimated for each average value of species richness to assess statistical differences between the cushions and open areas. Significant differences were assumed if confidence intervals did not overlap at the asymptote of the rarefaction curves (Gotelli & Colwell 2001). All rarefaction analyses were performed with the software EstimateS v. 8 (Colwell 2000).

To estimate the magnitude of the effects of cushions on species richness, we calculated the Relative Habitat Richness index (RHR) proposed by Badano *et al.* (2006). Relative Habitat Richness is computed as the ratio between the numbers of species found within the nurses and in open areas at the maximum number of samples

included in rarefaction curves. Cushion and open areas will contain similar number of species if $RHR \approx 1$, but cushions will contain more species if $RHR > 1$ and fewer species if $RHR < 1$.

To assess whether the number of species increases with the sample area (i.e. cushion size), and to compare these relationships between cushions and open areas, for each study site we performed a multiple linear regression analysis with categorical variables. In these analyses, the number of species on each sample was the dependent variable, the area of the sample was the continuous predictive variable and the position where the sample was taken (cushions vs. open areas) constituted the categorical variable. Both continuous variables (species richness and sample area) were log-transformed to achieve normality of data. In all the regression models, we also included a multiplicative interaction term between predictive variables to account for differences in the slopes of regression functions obtained for the two levels of the categorical variable (Neter *et al.* 1996).

DIFFERENCES ON SPECIES ASSEMBLAGES

To assess whether composition of species assemblages associated to cushions differed from those in open areas, we performed Non-metric Multidimensional Scaling ordinations (McCune & Mefford 1999) based on species abundance data of each community. Results obtained for the two main ordination axes were used to estimate centroids, with their respective 95% confidence intervals, for cushions and open areas. Statistical differences in species composition between cushions and open areas were assumed, if 95% confidence intervals of centroids did not overlap at least on one ordination axis.

EFFECTS ON SPECIES RICHNESS AT THE COMMUNITY LEVEL

Assessing the effects of facilitation on species richness at the entire community level relies on comparing the number of species a community has due to the presence of nurses with the richness expected in the community without nurses (Badano & Cavieres 2006a; Badano *et al.* 2006). To estimate species richness for the community with cushions, we generated synthetic data sets combining data of cushions and open areas in a single species \times samples matrix for each study site. However, as cushions always represent a smaller fraction of habitat compared with open areas (Table 1), the percentage cover of each habitat type was used to weight these data sets. For instance, if the average cover of cushions in the given community was 10%, and 100 samples were taken in each habitat (e.g. *A. madreporica* at 33°18' S, 70°14' W, see Table 1), the synthetic data set included 10 randomly selected plots from cushions and 90 randomly selected plots from open areas. To avoid biases due to differences in the samples included in the synthetic data set, for each site we constructed 50 randomly derived data sets, and a rarefaction analysis was run for each of these data sets by using the protocol described above. Thus, 50 values of species richness were estimated for each sampling size. These 50 values were averaged and plotted against the respective sampling size, and values at the asymptote of these rarefaction curves were considered maximum-likelihood estimators of species richness (Badano & Cavieres 2006a,b). For each site, the expected species richness of the community without cushions was estimated from the asymptotes of rarefaction curves previously constructed for open areas (Badano *et al.* 2006). The 95% confidence intervals were used to assess whether species richness at the entire community level differed between the samples taken in the open areas and the synthetic data set.

To assess the magnitude of the increase in species richness at the community level due to the presence of nurses, we calculated the

proportion of increase in species richness as: $(S_{+nurses} - S_{-nurses}) / S_{+nurses}$, where $S_{+nurses}$ is the actual number of species in the community, including the species growing both within and outside nurses, while $S_{-nurses}$ is the expected number of species in the absence of nurses. In our case, for each site values of $S_{+nurses}$ and $S_{-nurses}$ were estimated from the asymptotes of rarefaction curves performed with the synthetic data sets (open areas plus cushions) and open areas, respectively.

Finally, we assessed whether there is a relationship between the magnitude of the increase in species richness due to the presence of cushions and the environmental severity of each particular site. For this, based on the geographical coordinates of each site we obtained their monthly values of precipitation and mean temperature from the data base WorldClim (<http://www.worldclim.com>). WorldClim is a set of global climate layers (climate grids) with a spatial resolution of 1 km² (Hijmans *et al.* 2005). With this information, for each site, we calculated the Effective Precipitation of de Martonne: $EP = 12 * P / (T + 10)$, where P is monthly precipitation and T is monthly mean temperature. EP has been previously used as a surrogate of habitat severity (e.g. Kikvidze *et al.* 2006) because it is highly correlated with habitat productivity (e.g. Stadler 2005), but easy to calculate from simple meteorological data. The Spearman rank correlation was used to test for a relationship between EP and the total number of species and the percentage increase in species richness due to the presence of cushions in each community.

Results

SPECIES RICHNESS BETWEEN HABITATS

Rarefaction curves for the cushion habitat and the open areas reached an asymptote in most of the study sites (Fig. 1), indicating that the sampling effort was large enough to fully capture the composition of species assemblages in both habitat types.

Comparisons of species richness at the maximum number of samples indicated that cushions always contained more species than open areas (Fig. 1). However, the magnitude of these positive effects varied among the study sites. The RHR index showed two peaks at 30° and 41°S, where cushions contained up to 2.5 and 2.9 times more species than open areas, respectively, with decreasing values of RHR towards the north and south extremes (Fig. 2). The lowest effects were detected at middle latitudes (33°–37°S), where cushions contained 1.12–1.18 times more species than the open areas (Fig. 2).

Species richness increased with the area of samples both within cushions and in the surrounding open areas in all study sites (Fig. 3). With the exception of *A. madreporica* at 33°S, intercepts of regression functions for cushions were always significantly higher than those obtained for open areas ($P < 0.01$ in all cases, see Appendix S1 in Supporting Information), indicating that even the smallest cushions support a higher number of species than an equally sized sample in open areas. Slopes of regression functions showed no differences between cushions and open areas for *P. bryoides* at 23°S, *A. madreporica* and *A. subterranea* at 30°S, and *L. acaulis* at 33°S ($P > 0.05$ in all cases, see Appendix S1), indicating that the difference in species richness between cushions and open areas is constant across all the range of sample areas. Most of the sites where the

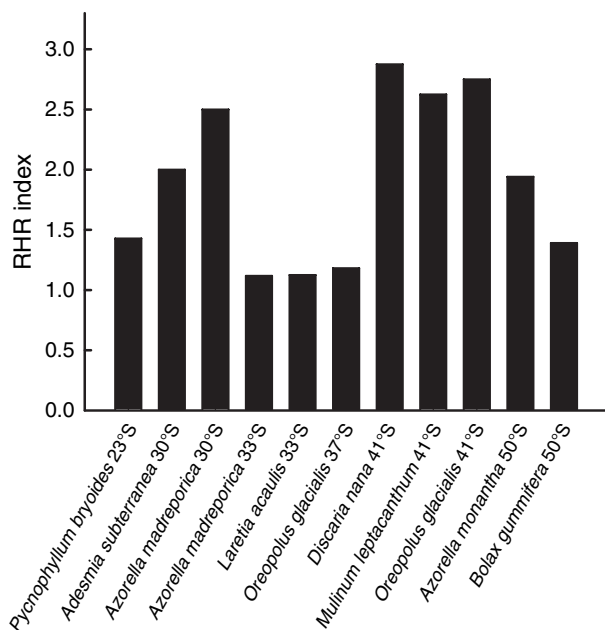


Fig. 2. Values of the Relative Habitat Richness index (RHR) calculated to assess the effects of cushions on species richness at the patch scale for each study site.

slopes of regression functions were significantly higher for cushions than for open areas occurred at latitudes higher than 33°S ($P < 0.01$ in all cases, see Appendix S1), suggesting that at higher latitudes the species accumulation rate of cushions is higher than in open areas.

SPECIES COMPOSITION

In most of the studied communities ordination analyses grouped separately samples from cushion and open areas, where the two main axes of NMS ordinations explained more than 60% of variance between samples (Fig. 4). The exception was the community dominated by *O. glacialis* at 37°27' S. Furthermore, with the exception mentioned above, confidence intervals of centroids for cushions and open areas did not overlap in both axes (Fig. 4), indicating that composition of species assemblages inhabiting cushion patches significantly differed from those inhabiting open areas.

EFFECTS ON SPECIES RICHNESS AT THE ENTIRE COMMUNITY LEVEL

The number of species estimated at the asymptote of rarefaction curves was always significantly higher for the data sets including cushions than for those from open areas only (Fig. 5), indicating that the presence of nurses always increased plant species richness at the entire community level in these high-alpine sites. Nevertheless, strong variation was observed for these entire-community effects of cushions on species richness (Fig. 5). The strongest effect was detected for *M. leptacanthum* at 41°S, where this cushion species appears to support up to 52% of species within the community, and the weakest effect was indicated for *L. acaulis* at 33°S, where only 16% of

the community depends on the presence of this cushion species (Fig. 5).

The total number of species at each site was highly correlated with EP ($R = 0.52$, $P < 0.05$), indicating that higher productivity sites contained higher numbers of species in the entire community (Fig. 6a). However, the percentage increase in species richness due to the presence of cushions did not correlate with EP ($R = 0.1$, $P = 0.7$). Nevertheless, data distribution suggested a nonlinear relationship between both variables (Fig. 6b). A fit to a quadratic model indicated a significant relationship between both variables ($R^2 = 0.3$; $P < 0.05$), indicating that the magnitude of the positive effect of the presence of cushions on species richness at the entire community was lower at both extremes of this climatic gradient (Fig. 6b).

Discussion

Community ecologists recognize that many factors affect the species composition of a given community, with no single factor providing a complete explanation for the observed patterns. Moreover, different factors can interact in a complex hierarchical fashion. The abiotic environmental filter and the regional species pool determine the potential members of a community, dispersal ability decides the identity of those species available to colonize a given community, and interspecific interactions play a fundamental role in the success or failure of species as community members (Lortie *et al.* 2004). Although ecological theory has emphasized the role of negative interactions as the main structuring force of communities at local scales, increasing evidence is accruing for the pivotal role of facilitative interactions in regulating diversity (Michalet *et al.* 2006). In this study, we have shown that the presence of facilitator species (i.e. species that mitigate mortality factors) generates a variety of changes in community structure and species diversity.

We observed that, in general, cushions contained more species than equivalent open areas, although there were differences in the magnitude of this effect. Evidence from studies performed in other alpine habitats show contrasting results, with some studies reporting either no differences (e.g. Pysek & Liska 1991; Totland, Grytnes & Heegaard 2004) or higher species richness outside nurses (e.g. Cavieres *et al.* 1998). Thus, at a first glance, no clear trends regarding species richness beneath and outside facilitator species emerge from comparisons at this spatial scale. However, these idiosyncratic responses may depend on the intensity of abiotic stress experienced by the plants in the microhabitats away from nurses and the ability of the nurse to mitigate such conditions. For example, studies conducted at two contrasting elevations in the high-alpine zone of the Chilean Patagonian Andes showed that while cushions species and open areas did not differ in species richness at lower elevations, at higher elevations cushions contained three- to fourfold more species than open areas (Cavieres *et al.* 2002; Arroyo *et al.* 2003). Microclimatic amelioration by cushions increased with elevation, which explains the higher positive effect found at higher altitudes (Arroyo

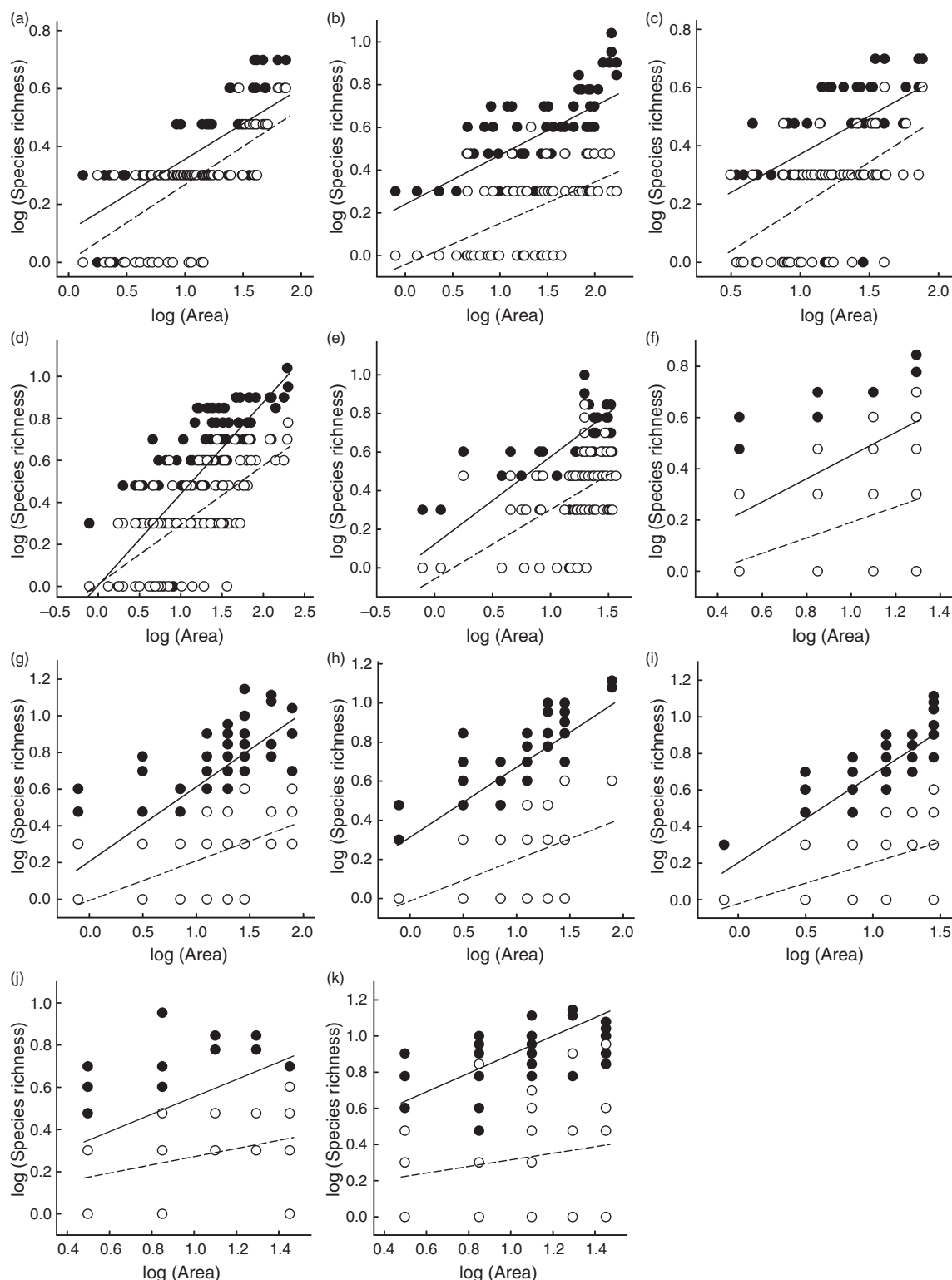


Fig. 3. Species–area relationships for cushions (solid symbols, solid lines) and open areas (empty symbols, dashed lines) on each study site. (a) *Pycnophyllum bryoides* at 23°S; (b) *Azorella madreporica* at 30°S; (c) *Adesmia subterranea* at 30°S; (d) *Azorella madreporica* at 33°S; (e) *Laretia acaulis* at 33°S; (f) *Oreopolus glacialis* at 37°S; (g) *Mulinum leptacanthum* at 41°S; (h) *Oreopolus glacialis* at 41°S; (i) *Discaria nana* at 41°S; (j) *Bolax gummiifera* at 50°S; (k) *Azorella monantha* at 50°S. Statistics for each regression analysis are provided as online Supporting Information.

et al. 2003). Similar effects have been also reported for the Andes of central Chile (Badano & Cavieres 2006b), supporting the idea that the impact of facilitator species on species richness

at a patch scale is higher as the environmental severity increases. More recently, Holzapfel *et al.* (2006) found that while species density (species per area unit) at mesic sites in

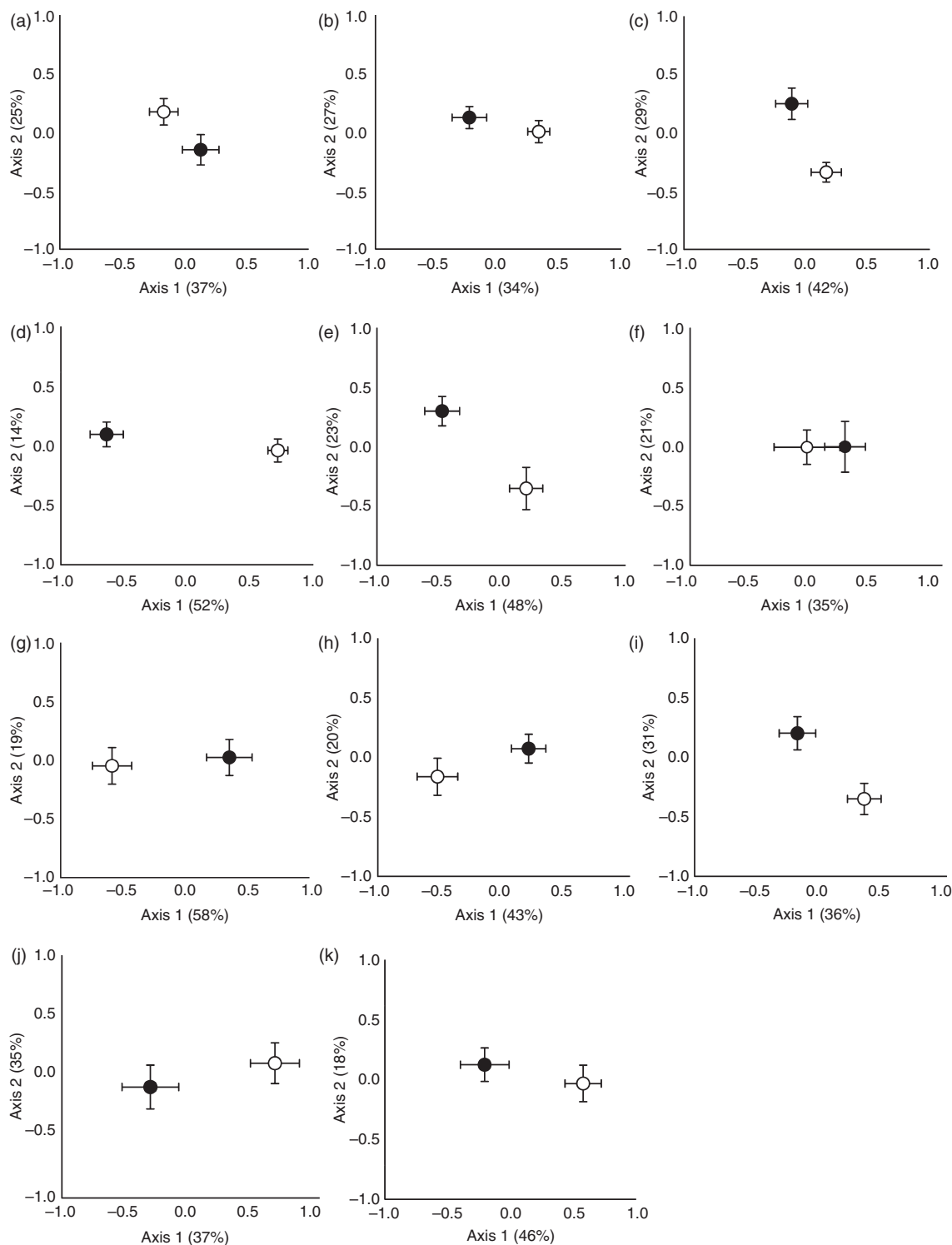


Fig. 4. Non-metric Multidimensional Scaling ordinations (NMDS) analyses for samples taken within cushions (solid circles) and in equivalent open areas (open circles). Error bars are ± 2 SE. Percent variance explained by each axis is indicated on each figure. (a) *Pycnophyllum bryoides* at 23°S; (b) *Azorella madreporica* at 30°S; (c) *Adesmia subterranea* at 30°S; (d) *Azorella madreporica* at 33°S; (e) *Laretia acaulis* at 33°S; (f) *Oreopolus glacialis* at 37°S; (g) *Mulinum leptacanthum* at 41°S; (h) *Oreopolus glacialis* at 41°S; (i) *Discaria nana* at 41°S; (j) *Bolax gummifera* at 50°S; (k) *Azorella monantha* at 50°S.

Israel did not differ between shrub understoreys and open areas, species density beneath shrubs was twice as high as in the open in arid sites. In our study, we sampled communities dominated by cushions at the upper altitudinal limits of cush-

ions distributions, hence it seems likely that in all cases we were working in the most stressful conditions for these plants. From this, it is important to note that the effect of cushions on species richness at this spatial scale was always positive.

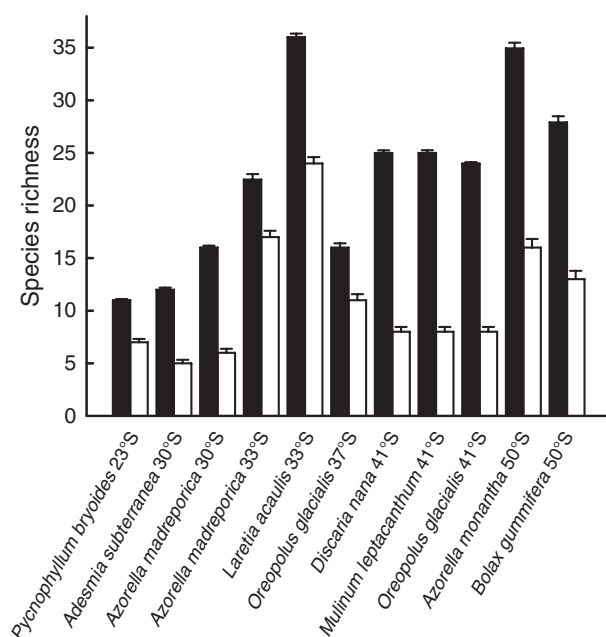


Fig. 5. Estimated values (and 95% confidence intervals) of species richness for entire communities comprised of cushions plus open areas (solid bars) and communities only comprised of open areas (empty bars) at each study site.

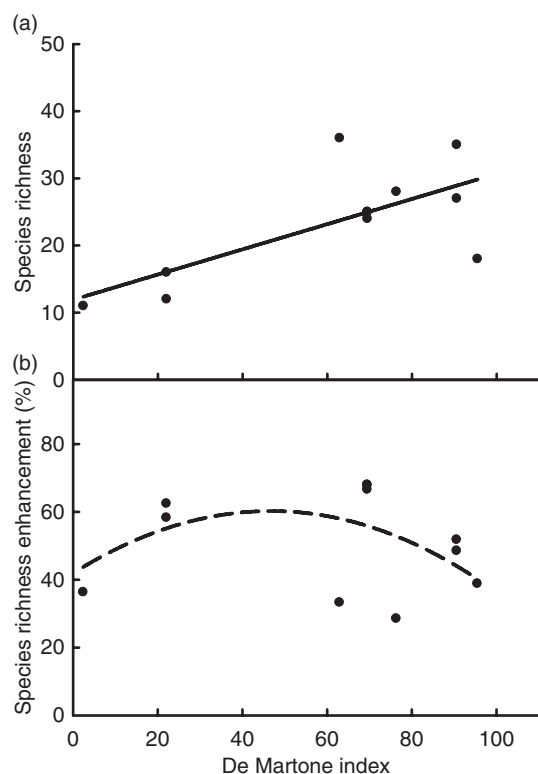


Fig. 6. Relationship between habitat severity assessed by the De Martonne index and species richness at the entire community (a) and percentage increase in species richness at the entire community due to the presence of cushions (b) for 11 sites along the southern Andes of South America.

Besides changes in species density, cushions also produced changes in the abundance of species, generating assemblages structurally different from those found in open areas as shown in the ordination analyses. Interestingly, these changes in the plant assemblages also occurred in those communities where cushions had lower values of the RHR index (e.g. *A. madreporica* and *L. acaulis* in central Chile, 33°S), indicating that in these cases changes in species abundances were more important than changes in species composition. Similar effects of nurse species were reported by Tewksbury & Lloyd (2001), who showed that *Olneya tesota* trees harbour structurally different species assemblages than open areas, and that this effect occurred mostly in xeric rather than mesic sites (see also Suzán, Nabhan & Patten 1996). These results highlight the important impact that positive interactions with nurse species have on the structure of plant communities, particularly in severe habitats (Tirado & Pugnaire 2005).

Although the species richness of nurses can be lower than that of open areas, the important assessment is whether many more species are added to the community due to the presence of the nurse. Previous studies have failed to examine this simple, but important question. For example, De Villiers, Van Rooyen & Theron (2001) studied seedling emergence and survival beneath and between the canopy of five shrub species in the Strandveld Succulent Karoo in South Africa and found that species richness and seedling densities were significantly higher in open areas than underneath shrubs. These authors concluded, therefore, that there was no evidence of facilitation at that site. Nevertheless, they reported 15 species that were only found growing beneath shrubs, indicating that *c.* 23% of the local species richness is due to the presence of shrubs. Interestingly, most of the species that were restricted to shrub understoreys were perennial herbaceous species, whereas species growing in open areas away from shrubs were ephemeral (annual) species. Similar results were reported by Gutiérrez *et al.* (1993) for a semi-desert area in northern Chile, where 19 species were found growing beneath the canopy of the shrub *Porlieria chilensis*, whereas 29 species were reported for open areas between shrubs. Nonetheless, although there were approximately fourfold more plant individuals outside than underneath shrub canopies, there were no differences in total biomass, indicating that those individuals growing underneath shrubs attained a higher biomass than those growing outside. Interestingly, six species (*c.* 17% of the local richness) were found only beneath shrubs. Likewise Pugnaire, Armas & Valladares (2004), despite reporting that shrub patches did not contain more species than patches of similar size in open areas, found 14 species (*c.* 40% of the total species reported for that site) that grew exclusively beneath shrubs. In all the sites of our study, the presence of cushions increases the species richness at the entire community level, with some cases where this increase in species richness accounts for approximately half of the species present in the entire community. Therefore, considering the field evidence reported in this study, and the re-evaluation of published studies, it is clear

that facilitative interactions can increase species richness at the community level.

Hacker & Gaines (1997) suggested that the impact of a facilitator species on the enhancement of species richness will be higher as the environment becomes more stressful (see also Michalet *et al.* 2006). Very few studies have tested this expectation at the entire community level. Tewksbury & Lloyd (2001) visited locations along a gradient in the Sonora desert that differed in water availability and conducted a comprehensive study of the consequences of facilitation by *O. tesota* trees on perennial and ephemeral herbs. They found that on mesic sites, while species richness in perennial plants beneath *O. tesota* did not differ from that found outside the nurse, richness of ephemeral species was higher outside the nurse. In contrast, on xeric sites, while species richness of ephemeral plants did not differ between the nurse understorey and areas outside the understorey, richness of perennial species was higher beneath the nurse. Interestingly, while on the mesic sites only c. 25% of the species grew exclusively in association with the nurse, on xeric sites this figure increased to c. 45%, supporting the proposition that in more severe environmental conditions the effect of nurse species in increasing the species richness of the entire community is higher. However, Tewksbury & Lloyd (2001) only had two categories of severity (i.e. xeric and mesic sites). Our results seem to support a nonlinear relationship between the magnitude of the positive effect on species richness and environmental severity (Fig. 6), where higher impacts are found in the middle of this severity gradient. This result is in line with the model proposed by Michalet *et al.* (2006), which shows that at both extremes of a severity gradient the importance of facilitative interactions in determining species richness decreases. Although our study was focused on severe habitats, our results also suggest that under extremely stressful conditions fewer species are present in the entire community, decreasing the pool of species which can benefit from the environmental mitigation performed by the nurse-cushion plants. However, more samples spanning a higher productivity range are needed to further explore this relationship. Notwithstanding, our analyses clearly show that cushions always increase species richness at the entire community level.

Greater species diversity within communities may enhance several key ecosystem functions (e.g. carbon and nitrogen fixation); hence our study is relevant to understand the impact of facilitation on ecosystem function (see Callaway 2007). The positive effects of facilitation on species richness at the community level emerge more clearly in studies that evaluate several communities along wide geographical gradients (e.g. Holzapfel *et al.* 2006; Tewksbury & Lloyd 2001; this study), where exploration of both environmental conditions and plant community compositional change enable a clearer distinction of the key processes that drive community assembly. Environments in which facilitation is a key process (e.g. alpine and arid systems) are particularly sensitive to major anthropogenic drivers of ecosystem change, including climate change, land use change and invasive non-native species (Brooker 2006). Thus, for proper management and conservation of these environments,

it is vital to understand the importance of facilitation in the regulation of their diversity.

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References

- Armas, C. & Pugnaire, F.I. (2005) Plant interactions govern population dynamics in a semi-arid plant community. *Journal of Ecology*, **93**, 978–989.
- 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 Ecology*, **169**, 121–129.
- Badano, E.I. & Cavieres, L.A. (2006a) Ecosystem engineering across ecosystems: do engineer species sharing common features have generalized or idiosyncratic effects on species diversity? *Journal of Biogeography*, **33**, 304–313.
- Badano, E.I. & Cavieres, L.A. (2006b) Impacts of ecosystem engineers on community attributes: effects of cushion plants at different elevations of the Chilean Andes. *Diversity and Distribution*, **12**, 388–396.
- Badano, E.I., Molina-Montenegro, M.A., Quiroz, C. & Cavieres, L.A. (2002) Efectos de la planta en cojín *Oreopolus glacialis* (Rubiaceae) sobre la riqueza y diversidad de especies en una comunidad alto-andina de Chile central. *Revista Chilena de Historia Natural*, **75**, 757–765.
- Badano, E.I., Cavieres, L.A., Molina-Montenegro, M. & Quiroz, C. (2005) Slope aspect influences plant association patterns in the Mediterranean matorral of central Chile. *Journal of Arid Environment*, **62**, 93–108.
- Badano, E.I., Jones, C.G., Cavieres, L.A. & Wright, J.P. (2006) Assessing impacts of ecosystem engineers on community organization: a general approach illustrated by effects of a high-Andean cushion plant. *Oikos*, **115**, 369–385.
- Bertness, M.D. & Callaway, R.M. (1994) Positive interactions in communities. *Trends in Ecology and Evolution*, **9**, 191–193.
- Brooker, R.W. (2006) Plant–plant interactions and environmental change. *New Phytologist*, **171**, 271–289.
- Brooker, R.W. & Callaghan, T.V. (1998) The balance between positive and negative interactions and its relationship to environmental gradient: a model. *Oikos*, **81**, 196–207.
- Brooker, R.W., Maestre, F.T., Callaway, R.M., Lortie, C.J., Cavieres, L.A., Kunstler, G. *et al.* (2008) Facilitation in plant communities: the past, the present and the future. *Journal of Ecology*, **96**, 18–34.
- Bruno, J.F., Stachowicz, J.J. & Bertness, M.D. (2003) Inclusion of facilitation into ecological theory. *Trends in Ecology and Evolution*, **18**, 119–125.
- Callaway, R.M. (1995) Positive interactions among plants. *Botanical Review*, **61**, 306–349.
- Callaway, R.M. (2007) *Positive Interactions and Interdependence in Plant Communities*. Springer, Berlin.
- Callaway, R.M. & Walker, L.R. (1997) Competition and facilitation: a synthetic approach to interactions in plants communities. *Ecology*, **78**, 1958–1965.
- Callaway, R.M., Brooker, R.W., Choler, P., Kikvidze, Z., Lortie, C.J., Michalet, R. *et al.* (2002) Positive interactions among alpine plants increase with stress. *Nature*, **417**, 844–848.
- Cavieres, L.A., Peñaloza, A.P.G., Papic, C. & Tambutti, M. (1998) Efecto nodriza de *Laretia acaulis* en plantas de la zona andina de Chile central. *Revista Chilena de Historia Natural*, **71**, 337–347.
- Cavieres, L.A., 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. *Journal of Vegetation Science*, **13**, 547–554.
- Cavieres, L.A., Badano, E.I., Sierra-Almeida, A., Gómez-González, S. & Molina-Montenegro, M. (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 Phytologist*, **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 seedlings survival of native and non-native plants in the high-Andes of central Chile. *Arctic, Antarctic and Alpine Research*, **39**, 229–236.
- Choler, P., Michalet, R. & Callaway, R.M. (2001) Facilitation and competition on gradients in alpine plant communities. *Ecology*, **82**, 3295–3308.
- Colwell, R.K. (2000) *EstimateS: Statistical Estimation of Species Richness and Shared Species From Samples*. University of Connecticut, Storrs, CT.
- Connell, J.H. (1978) Diversity in tropical rain forests and coral reefs. *Science*, **199**, 1302–1310.
- De Villiers, A.J., Van Rooyen, M.W. & Theron, G.K. (2001) The role of facilitation in seedling recruitment and survival patterns, in the Strandveld Succulent Karoo, South Africa. *Journal of Arid Environments*, **49**, 809–821.
- Fajardo, A., Quiroz, C. & Cavieres, L.A. (2008) Spatial structures in cushion-dominated plant communities of the high-Andes of central Chile: how frequent are positive associations? *Journal of Vegetation Science*, **19**, 87–96.
- Flores, J. & Jurado, E. (2003) Are nurse-protégé interactions more among plants from arid environments? *Journal of Vegetation Science*, **14**, 911–916.
- Gotelli, N.J. & Colwell, R.K. (2001) Quantifying biodiversity: procedures and pitfalls in the measurement and comparison of species richness. *Ecology Letters*, **4**, 379–391.
- Grime, J.P. (1973) Competitive exclusion in herbaceous vegetation. *Nature*, **242**, 344–347.
- Gross, K. (2008) Positive interactions among competitors can produce species-rich communities. *Ecology Letters*, **11**, 929–936.
- Gutiérrez, J.R., Meserve, P.L., Contreras, L.C., Vásquez, H. & Jaksic, F.M. (1993) Spatial distribution of soil nutrients and ephemeral plants underneath and outside the Canopy of *Porlieria chilensis* shrubs (Zygophyllaceae) in arid coastal Chile. *Oecologia*, **95**, 374–382.
- Hacker, S.D. & Gaines, S.D. (1997) Some implications of direct positive interactions for community species diversity. *Ecology*, **78**, 1990–2003.
- 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. *International Journal of Climatology*, **25**, 1965–1978.
- Holzapfel, C., Tielbörger, K., Parag, H.A., Rigel, J. & Sternberg, M. (2006) Annual plant–shrub interactions along an aridity gradient. *Basic and Applied Ecology*, **7**, 268–279.
- Kikvidze, Z., Khetsuriani, L., Kikodze, D. & Callaway, R.M. (2006) Seasonal shifts in competition and facilitation in subalpine plant communities of the central Caucasus. *Journal of Vegetation Science*, **17**, 77–82.
- Körner, C. (2003) *Alpine Plant Life*, 2nd edn. Springer, Berlin.
- Larrea-Alcázar, D.M., López, R.P. & Barrientos, D. (2005) The nurse-plant effect of *Prosopis flexuosa* D.C. in a dry valley of the Bolivian Andes. *Ecotropics*, **16**, 89–95.
- LeRoux, P.C. & McGeoch, M.A. (2008) Spatial variation in plant interactions across a severity gradient in the sub-arctic. *Oecologia*, **155**, 831–844.
- Lortie, C.J. & Callaway, R.M. (2006) Re-analysis of meta-analysis: support for the stress-gradient hypothesis. *Journal of Ecology*, **93**, 7–16.
- 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.
- Maestre, F.T., Bautista, S. & Cortina, J. (2003) Positive, negative and net effects in grass-shrub interactions in Mediterranean semi-arid grasslands. *Ecology*, **84**, 3186–3197.
- Maestre, F.T., Valladares, F. & Reynolds, J.F. (2005) Is the change of plant–plant interactions with abiotic stress predictable? A meta-analysis of field results in arid environments. *Journal of Ecology*, **93**, 748–757.
- McClune, B. & Mefford, M.J. (1999) *Multivariate Analysis of Ecological Data*. MjM Software Design, Gleneden Beach, OR.
- Michalet, R., Brooker, R.W., Cavieres, L.A., Kikvidze, Z., Lortie, C.J., Pugnaire, F.I., Valiente-Banuet, A. & Callaway, R.M. (2006) Do biotic interactions shape both sides of the humped-back model of species richness in plant communities? *Ecology Letters*, **9**, 767–773.
- Neter, J., Kutner, M.H., Nachtsheim, C.J. & Wasserman, W. (1996) *Applied Linear Statistical Models*, 4th edn. WBC/McGraw-Hill, Boston, MA.
- Núñez, C.I., Aizen, M.A. & Ezcurra, C. (1999) Species associations and nurse plant effects in patches of high-Andean vegetation. *Journal of Vegetation Science*, **10**, 357–364.
- Pugnaire, F.I., Armas, C. & Valladares, F. (2004) Soil as a mediator in plant–plant interactions in a semi-arid community. *Journal of Vegetation Science*, **25**, 85–92.
- Pugnaire, F.I., Haase, P., Puigdefábregas, J., Cueto, M., Clark, S.C. & Incoll, D. (1996) Facilitation and succession under canopy of a leguminous shrub, *Retama sphaerocarpa*, in a semi-arid environment in south-east Spain. *Oikos*, **76**, 455–464.
- Pysek, P. & Liska, J. (1991) Colonization of *Sibbaldia tetrandra* cushions on alpine scree in the Pamiro-Alai Mountains, Central Asia. *Arctic and Alpine Research*, **23**, 263–272.
- Raffaele, E. & Veblen, T.T. (1998) Facilitation by nurse shrubs of resprouting behavior in a post-fire shrubland in northern Patagonia, Argentina. *Journal of Vegetation Science*, **9**, 693–698.
- Rossi, B.E. & Villagra, P.E. (2003) Effects of *Prosopis flexuosa* on soil properties and the spatial pattern of understory species in arid Argentina. *Journal of Vegetation Science*, **14**, 543–550.
- Stadler, S. (2005) Aridity Indexes. *Encyclopedia of World Climatology* (ed. J.E. Oliver), pp. 89–94. Springer, Berlin.
- Suzán, H., Nabhan, G.P. & Patten, D.T. (1996) The importance of *Olneya tesota* as a nurse plant in the Sonoran Desert. *Journal of Vegetation Science*, **7**, 635–644.
- Tewksbury, J.J. & Lloyd, J.D. (2001) Positive interactions under nurse-plants: spatial scale, stress gradients and benefactor size. *Oecologia*, **127**, 425–434.
- Tilman, D. (1982) *Resource Competition and Community Structure*. Princeton University Press, Princeton.
- Tirado, R. & Pugnaire, F.I. (2005) Community structure and positive interactions in constraining environments. *Oikos*, **111**, 437–444.
- Totland, O., Grytnes, J.A. & Heegaard, E. (2004) Willow canopies and plant community structure along an alpine environmental gradient. *Arctic, Antarctic, and Alpine Research*, **36**, 428–435.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Appendix S1. Results of linear regression analyses for species richness and the area sampled inside cushions and open areas at different site along the southern Andes of South America.

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