



Original article

The role of nurse functional types in seedling recruitment dynamics of alternative states in rangelands

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ABSTRACT

In arid ecosystems, recruitment dynamics are limited by harsh environmental conditions and greatly depend on the net outcome of the balance between facilitation and competition. This outcome can change as a consequence of degradation caused by livestock overgrazing. Also, distinct plant species may show a differential response to a common neighbour under the same environmental conditions. Therefore, ecosystem degradation could affect the net balance of plant-plant interactions, which can also depend on the functional traits of potential nurse species. The aim of this study is to assess the influence of alternative degradation states on (i) the density of seedlings of perennial species emerging in four microsite types, and on (ii) the relative interaction intensity (RII) between seedlings and potential nurses belonging to three functional types (deep- and shallow-rooted shrubs, and tussock grasses). During three years, we recorded seedling density of perennial species in four alternative degradation states in grass-shrubby steppes from northwestern Patagonia. The density of emerged seedlings of perennial species decreased sharply as degradation increased, showing non-linear responses in most microsites. Seedling density underneath deep-rooted shrubs was higher than underneath shallow-rooted shrubs and tussock grasses. Also, deep-rooted shrubs were the only functional type that recorded seedling emergence in highly degraded states. Deep-rooted shrubs had facilitative effects on the seedlings emerging and surviving underneath them, independently of ecosystem degradation. In contrast, RII between shallow-rooted shrubs and recently emerged seedlings, switched from positive effects in the less degraded states, to negative effects in the most degraded state. Tussock grasses recorded the weakest intensity of facilitative interactions with recently emerged seedlings, switching to competitive interactions as degradation increased. Our results suggest that species with key functional traits should be considered in management and restoration plans for rangelands with different degradation levels, since they have a strong influence in the net outcome of plant-plant interactions and in the recruitment dynamics of arid ecosystems.

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1. Introduction

In arid and semiarid ecosystems, plant recruitment is limited by harsh environmental conditions and greatly depends on plant-plant interactions, as facilitation and competition (Whitford,

2002; Padilla and Pugnaire, 2006). Facilitation occurs when an adult plant (i.e., nurse) increases the emergence, survival, growth and/or fitness of spatially associated seedlings (i.e., protégées) (Callaway, 2008). Nurse plants benefit the individuals growing underneath them by: (i) ameliorating extreme microclimatic conditions (i.e., reducing radiation and temperature at ground level, thereby diminishing evapo-transpiration during the day, and decreasing freezing risk during the night); and/or (ii) protecting against herbivores (i.e., many nurse species have spines or other features that deter herbivores, a mechanism known as 'associational resistance') (Callaway, 2008). Conversely, competition occurs when a plant growing close to another decreases the survival,

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growth and/or fitness of its neighbour as a consequence of sharing limited resources (e.g. water, nutrients, light) (Padilla and Pugnaire, 2006). Positive and negative effects occur simultaneously in arid and semiarid ecosystems (hereafter 'arid ecosystems'), and thus recruitment dynamics depend on the net outcome of the balance between facilitation and competition (Padilla and Pugnaire, 2006).

The magnitude and even the direction (either positive or negative) of net plant-plant interactions can change as a consequence of ecosystem degradation caused by human activities (e.g. livestock grazing). Overgrazing can, directly and/or indirectly, affect seedling recruitment in arid ecosystems (López et al., 2013). As a consequence of livestock overgrazing, the trampling, consumption of flowers and browsing produced by herbivores directly reduce seedling recruitment, and can affect plant-plant interactions since herbivore pressure will be higher for seedlings growing close to palatable species (Graff et al., 2007). Also, the reduction in vegetation cover, and the increase in soil erosion produced by historical overgrazing can decrease soil water availability, by increasing evaporation and decreasing its storage capacity (Paruelo et al., 1993; Reynolds and Stafford Smith, 2002). Thus, ecosystem degradation can indirectly affect seedling recruitment and plant-plant interactions, because as soil water availability decreases, seedling recruitment decreases as a consequence of increased competition for this limiting resource between neighbouring individuals. It has been widely documented that plant-plant interactions can change from negative to positive, or viceversa, in response to increasing abiotic stress or consumer pressure (Bertness and Callaway, 1994; Verwijmeren et al., 2013 and references therein). However, the studies that separately test the effect of abiotic stress and consumer pressure on plant-plant interactions are unrealistic, since abiotic constraints and consumer pressure often co-occur in arid ecosystems (Brooker et al., 2008). In a global context in which a 40% of arid ecosystems are severely degraded (Reynolds and Stafford Smith, 2002; Adeel et al., 2005), mainly as a consequence of livestock overgrazing (Adeel et al., 2005), it would be essential to know the influence of degradation in the recruitment dynamics and in the net balance of plant-plant interactions in rangelands.

Distinct plant species may show a differential response to a common neighbour under the same environmental conditions (Maestre et al., 2009a). This could be because the species that co-occur in a given community have different above- and below-ground traits which make them functionally different. Distinct functional traits could be beneficial, or even detrimental, for the same neighbour. For example, the different degree of palatability of nurse species, determined by the presence or absence of mechanical or chemical defenses, will differentially affect the emergence and/or survival of protégées. Thus, non-palatable nurse species can benefit protégées growing beneath them by deterring herbivores or lowering consumer pressure (Graff et al., 2007). Also, depending on the depth of the radicular system, water competition between neighbouring individuals can increase, or even, decrease. Competition for water will be higher for individuals whose roots are overlapped, than for those whose roots explore different soil layers. Although, the net balance of plant-plant interactions could depend on above- and below-ground traits of plant species, we do not know studies focusing on the functional types of nurse species (*sensu* Sala et al., 1997) as a determinant of the net outcome of plant-plant interactions in arid ecosystems.

While there is broad consensus in the scientific community that facilitation strongly modulates recruitment dynamics in arid ecosystems, facilitative interactions have not yet been explicitly considered in the conceptual framework of degradation (Verwijmeren et al., 2013). In this sense, the aim of this study is to assess the influence of alternative degradation states in seedling

recruitment dynamics considering different functional types of potential nurse plants. To do this, we used grass-shrubby steppes of northwestern Patagonia as a model-system, and during three years we evaluated the recruitment dynamics of perennial species in four alternative degradation states. Specifically, we assessed the influence of alternative degradation states on (i) the interannual density of seedlings emerging in different microsite types and on (ii) the relative interaction intensity between seedlings and potential nurses belonging to three functional types.

2. Materials and methods

2.1. Study area

The study was carried out in Pilcaniyeu, at a 7800 ha field station belonging to Estación Experimental Agropecuaria Bariloche from the Instituto Nacional de Tecnología Agropecuaria (INTA), located in Río Negro, Argentina (41° 01' 42" S, 70° 35' 21" W). Climate is characterized by cold wet winters (coldest month mean temperature is 2.1 °C) with temperate dry summers (hottest month mean temperature is 15 °C). Mean annual precipitation is 265 mm ± 82.5, and more than 70% of the precipitation falls during autumn and winter (López, 2011; Appendix A, Fig. A.1). As in other arid and semi-arid regions, annual precipitations are highly variable (Noy-Meir, 1973; Whitford, 2002), alternating years with precipitations near the historical average, and years with precipitations below- and above-average (Bustos, 2006; López, 2011). We studied the grass-shrubby steppe of *Poa ligularis* and *Mulinum spinosum*, a community of high forage value within the Western Patagonian District of the Patagonian Steppe. Soils are shallow, with a sandy surface layer and a clay-rocky sub-superficial layer (Lores et al., 1983). The main species are tussock-grasses (*P. ligularis* and *Pappostipa speciosa* var. *speciosa*), and deep- and shallow-rooted shrubs (*M. spinosum* and *Senecio filaginoides*, respectively), forming grass and/or shrub patches immersed in a bare soil matrix (Aguar and Sala, 1999; López et al., 2013). Patch dimensions vary from 0.15 to 1.5 m in width and from 0.1 to 1.2 m in height.

2.2. Functional types

To study the influence of alternative degradation states on recruitment dynamics of perennial species, we divided the dominant species of the grass-shrubby steppe from the western Patagonian District in functional types based on two criteria, which can influence the net outcome of plant-plant interactions in arid ecosystems: (a) above ground traits mainly associated with the ability to deter herbivores, as the presence of mechanical or chemical defenses; and (b) below ground traits that mainly determine plant-plant water relations which are extremely important in arid ecosystems, because water is the most limiting resource (Sala et al., 1997). Several studies suggest that shrubs can be potential nurses in arid lands (Gómez Aparicio et al., 2004), and it has also been documented facilitation by grasses (Maestre et al., 2001). Therefore, we selected the main shrub (*M. spinosum* and *S. filaginoides*) and tussock-grass (*P. ligularis* and *P. speciosa* var. *speciosa*) species. Shrub species were assigned to different functional types, as they differ in their above- and below-ground functional traits, whereas grass species were grouped together in one functional type due to their functional similarities (Table 1).

2.3. Experimental design

At three sites we identified four alternative degradation states of vegetation (I-IV, *sensu* Bonvissuto et al., 1993), separated by at least one critical threshold (López et al., 2013). Specifically, states I and II

Table 1
Detail of the above- and below-ground traits of the studied functional types.



Functional type	Deep-rooted shrubs	Shallow-rooted shrubs	Tussock grasses	References
Species	<i>Mulinum spinosum</i> (Cav.) Pers.	<i>Senecio filaginoides</i> DC	<i>Poa ligularis</i> Nees ex. Steud <i>Pappostipa speciosa</i> var <i>speciosa</i> Trin. And Rupr.	Godagnone and Bran (2009) Correa (1998)
Family	Apiaceae	Asteraceae	Poaceae	Godagnone and Bran (2009) Correa (1998)
Growth form	Shrub	Shrub	Grass	Godagnone and Bran (2009)
Mechanical defense	Foliar and caulinar spines	None	None	Damascos and Ghermandi (2007) Golluscio et al. (2011)
Chemical defense	Low content of carbon-based secondary metabolites	High content of carbon-based secondary metabolites	None	Golluscio et al. (2011)
Contribution to forage biomass	Low (only buds and flowers are consumed)	Very low (only flowers are consumed)	Very high	Siffredi et al. (2015)
Canopy shape	Hemispherical	Inverted cone	Inverted cone	Aguar and Sala (1998), Damascos and Ghermandi (2007)
Canopy diameter (cm)	50-100	30-50	20-50	López (2011)
Height (cm)	50-70	30-50	20-35	López (2011)
Root depth	The highest proportion of root biomass is found between 40-60 cm of soil depth	The highest proportion of root biomass is found in the upper 30 cm of soil	The highest proportion of root biomass is found in the upper 20 cm of soil	Fernandez and Paruelo (1988), Paruelo and Sala (1995), Aguár and Sala (1998)

(hereafter pre-critical threshold) preserve their ecological integrity and therefore, their resilience under a specific disturbance regime. In contrast, states III and IV (hereafter post-critical threshold) represent situations in which vegetation and soil are severely degraded by overgrazing and thus, the structural and functional integrity of the ecosystem is seriously affected (state III crossed one critical threshold, while state IV crossed two critical thresholds) (López et al., 2013). Pre-critical threshold states had, on average, 48.1% of total plant cover, of which the relative cover of grasses and shrubs is 72.3% and 22.2%, respectively (Table A.1, Appendix A). In these states, the dominant grass species is *P. ligularis*, while *M. spinosum* is the most abundant shrub. In contrast, post-critical threshold states had lower plant cover than pre-critical threshold states (<42%), which is almost co-dominated by grasses and shrubs

(relative cover of 47% and 46%, respectively). In these states, the dominant grass and shrub species are *P. speciosa* and *M. spinosum*, respectively (Table A.1, Appendix A). Specifically, state I has the greatest cover of the main forage species (*P. ligularis* ≈ 35%), whereas state IV has a negligible cover of this species (0.2 ± 0.05). State III has the greatest cover of the secondary forage species (*P. speciosa* var. *speciosa* ≈ 18%), while state IV has the greatest shrub cover (*M. spinosum* and *S. filaginoides*) (Table A.1, Appendix A). Bare soil area is higher in post-than in pre-critical threshold states (>60% and <45%, respectively).

In each of the three sites, we delimited two adjacent sectors: a livestock enclosure over 15 years old (representing state I) and a historically overgrazed area that belongs to an alternative state (II, III or IV, Fig. 1). In order to control the effects of a slight slope (<3%)

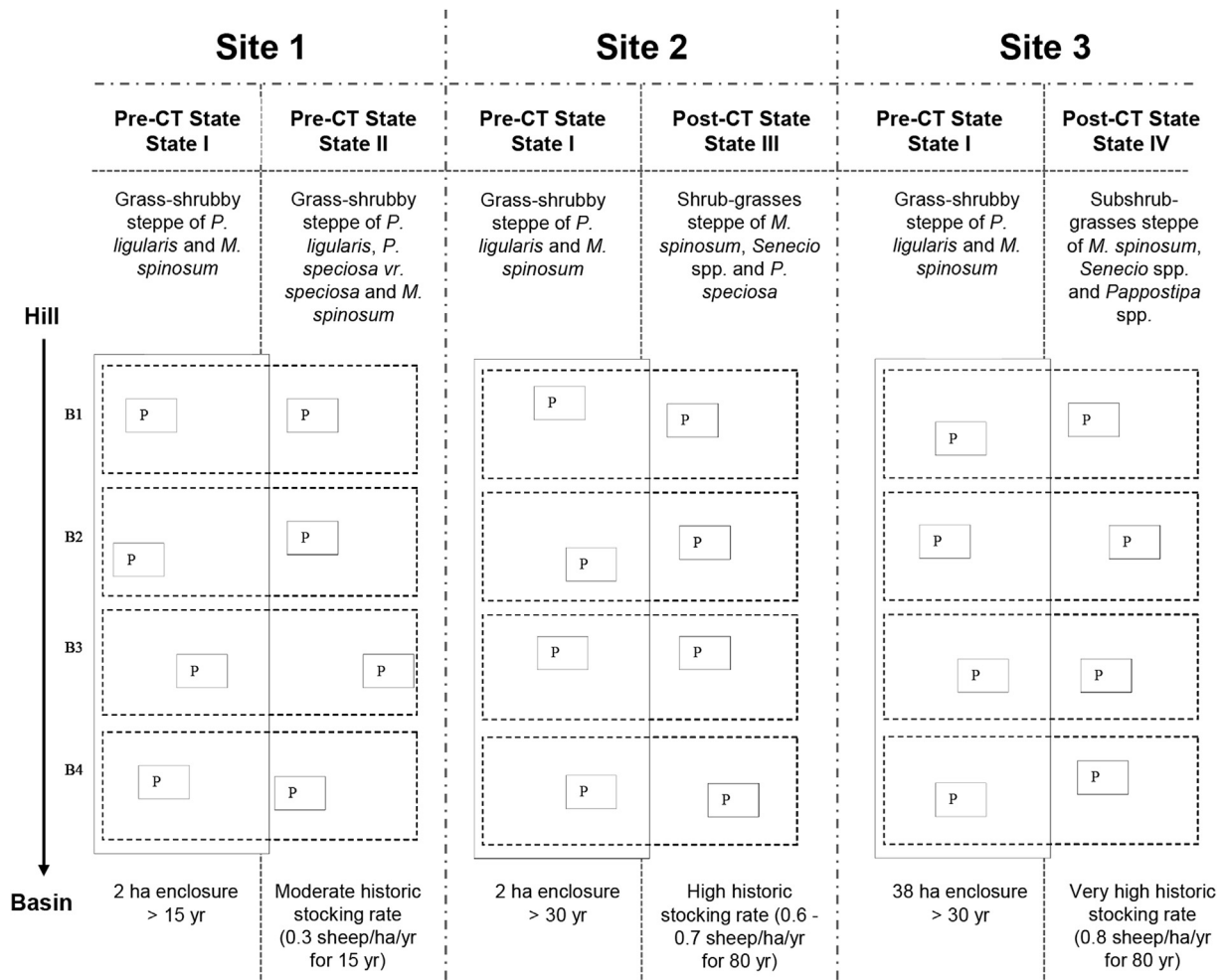


Fig. 1. Scheme of the randomized complete block design at Sites 1, 2 and 3, located in the “Campo Anexo of INTA EEA Bariloche”, Río Negro-Argentina. Each site includes two paired sectors: a livestock enclosure and a historically grazed sector (ranging from 700 to 900 ha). Considering the grazing history (stocking rate and time), sectors were labelled from I to IV based on the state and transition model proposed by Bonvissuto et al. (1993). Site 1: includes states I and II; Site 2: includes states I and III; Site 3: includes states I and IV. CT: critical threshold; B: blocks 1 to 4; P: Plots of 4 m².

a Completely Randomized Block Design (CRBD) was used within each site (Steel and Torrie, 1980). At each site, we placed four blocks including two states (I and II, III or IV) (Fig. 1).

2.4. Recruitment patterns in alternative states

We sampled seedling density of perennial species (shrubs and grasses) in 2 × 2 m plots randomly distributed in each state within each block. In each plot, we counted all seedlings younger than one year, and registered the microsite in which they emerged and/or survived (deep-rooted shrubs, shallow-rooted shrubs, tussock grasses, and open, which includes litter and bare soil). We estimated the cover of each microsite in each plot by using four 1 × 1 m quadrants divided into 100 grid cells of 0.1 × 0.1 m. Seedling counts were done in April because the emergence pulse of most species occurs in early autumn (López, 2011). Seedling density was sampled during three years, which recorded mean precipitations far below and near the historical average. While in the study region the mean precipitation during the emergence period (i.e., december–april) is 60.9 ± 13.5 mm (Bustos, 2006), in 2007–2008 precipitation was 4.35 times lower than the historical average (i.e., 14 mm), whereas in 2006–2007 and 2008–2009 precipitation was near the historical average (i.e., 55.1 mm and 51.1 mm,

respectively). Finally, in April 2009, we evaluated the survival percentage of each cohort.

2.5. Micro-environmental quality of microsites in alternative states

To assess if the changes in recruitment patterns in response to degradation can be associated to changes in the quality of recruitment microsites, as additional information we recorded soil temperature and moisture in the most contrasting microsites (deep-rooted shrubs and bare soil) in two alternative states (state I and III from Site 2). Microclimatic variables were recorded in two microsites on two alternative states due to logistical limitations. Between December 2007 and 2009 four data-loggers (each one with two temperature sensors: TC1047A Microchip Inc.) and two moisture sensors: ECH₂O) were installed in each state (4 replicates by state). In 2 × 2 m plots, temperature at 3 cm below the soil and moisture in the first 10 cm of soil was recorded every hour in two microsites: deep-rooted shrub and bare soil. We selected this microsites because they recorded the highest and lowest seedling densities, respectively. Also, bare soil was the most abundant microsite in degraded states. The response variables assessed were: mean, standard deviation, minimum and maximum monthly temperature, number of hours with temperatures below 0 °C and soil

moisture percentage. We used the standard deviation of mean monthly temperature to estimate thermal amplitude and variability. Soil moisture (measured with ECH₂O sensors) was corrected based on field data. In October 2009, three, six, ten, and 20 days after a rain event, we extracted soil samples of 10 × 10 cm in the microsites where the sensors were installed (n = 40, 10 samples in bare soil and 10 in plant patches, in both S2-I and S2-III), and recorded moisture percentage by gravimetry. We fitted an exponential regression with the data from the ECH₂O sensors and those gravimetrically obtained (adjusted R² = 0.79; y = 0.5723·e^{0.0059x}).

2.6. Data analysis

To estimate the degradation level of each alternative state we calculated a Structural Degradation Index (SDI, López et al., 2011; 2013). To calculate SDI we constructed a Mahalanobis Distance (MD) matrix (Legendre and Legendre, 1998) between the alternative states (four blocks per state, n = 24) based on structural variables (bare soil area, plant, grass and shrub cover; patch basal length, width, height, density, and symmetry; inter patch length; and soil depth; see Appendix A for sampling procedure). The Structural Degradation Index was calculated as follows: $SDI_i = [(MD_i \times 100) \times (MD_{max})^{-1}]$, where MD_i is the MD between the ith sampling point and the sampling point that had the greatest total vegetation cover. The MD_{max} corresponds to the maximum value of the registered MD. Based on MD_{max}, all the MD values were standardized, determining a variation range from 0 to 100%.

To assess the influence of degradation on the recruitment dynamics in the dominant recruitment microsites (deep-rooted shrubs, shallow-rooted shrubs, tussock grasses and open), we divided the number of seedlings recorded in a given microsite by the area of the microsite, to obtain seedling density (m² of microsite)⁻¹. To assess the influence of degradation on the net outcome of plant-plant interactions we calculated the Relative Interaction Index (RII; adapted from Armas et al., 2004). We calculated RII as: $RII = (P_{nurse} - P_{open}) \cdot (P_{nurse} + P_{open})^{-1}$, where P_{nurse} is the mean seedling density under the canopy of a nurse functional type (NFT) divided by the cover of the NFT (e.g. seedling density under tussock grasses · (tussock grasses cover)⁻¹) and P_{open} is the mean seedling density in bare soil and/or litter divided by bare soil and/or litter cover. The RII values range from -1 to +1, are symmetrical around zero, are negative for competitive species interactions and positive for facilitative species interactions (Armas et al., 2004). To assess the influence of degradation (i.e., SDI) on (i) seedling density of perennial species emerging underneath potential nurses belonging to different functional types, and on (ii) the relative interaction intensity (RII) of seedlings of perennial species emerging and surviving underneath potential nurses belonging to different functional types, we fitted simple, quadratic and non-linear regressions. Quadratic and non-linear regressions were judged to better fit the data when significant differences were detected between models or when the Akaike Information Criterion (AIC) was at least 2 units lower than the AIC of simple regressions (Crawley, 2007). Regressions were performed with R software (lm package; R Development Core Team, 2006). Response variables were transformed with ln(y + 1) when did not met normality, heteroscedasticity, and residuals independence assumptions.

To evaluate the effect of the degradation state and microsite type on micro-climatic variables we performed Linear Mixed Models (LMM). Each factor had two levels: state I (pre-critical threshold; hereafter 'pre-CT') vs. state III (post-critical threshold; hereafter 'post-CT') for State, and deep-rooted shrubs vs. bare soil for Microsite. We used a split plot design with the main plot in blocks, and assigned the State factor to the main plot and the Microsite factor to subplots, with a crossed structure between

them. To calculate the mean monthly temperature and its standard deviation we used the daily average from each temperature sensor, whereas to calculate mean monthly minimum and maximum temperatures we used the minimum and maximum daily values. We used the daily average from soil moisture sensors to calculate the mean monthly soil moisture percentage.

3. Results

3.1. Recruitment patterns in alternative states

During the three years of the study, the density of emerged seedlings of perennial species decreased sharply as degradation increased, showing non-linear responses in most cases (Fig. 2, Appendix C). However, the influence of degradation on the density of emerged seedlings depended on the functional type of potential nurses. In 2007, in the less degraded states (I and II), seedling density underneath deep-rooted shrubs was, on average, 1.4 and 4 times higher than underneath shallow-rooted shrubs and tussock grasses, respectively (Fig. 2a–c). Also, deep-rooted shrubs were the only functional type that recorded seedling emergence in highly degraded states (III and IV; Fig. 2a). In 2008, we could not perform statistical analyses because seedling densities were very low in all states. Nevertheless, the scarce seedlings that emerged in this year, were located underneath deep-rooted shrubs. Finally, the density of emerged seedlings was lower in 2009 than in 2007, independently of microsite type. Thus, for the cohort 2009, we could not find significant associations between the density of seedlings emerging underneath shallow-rooted shrubs, tussock grasses or in open microsites; and ecosystem degradation. Conversely, the density of seedlings emerging underneath deep-rooted shrubs showed a non-linear response to ecosystem degradation. Specifically, seedlings emerged only in un-degraded states (i.e., pre-critical threshold: states I and II) (Fig. 2a).

Overall (including all cohorts), the 94% of emerged seedlings were of forage grass species, mainly of *P. speciosa*, *Hordeum comosum* and *Bromus setifolius*. In the less degraded states (I and II), the main forage species (*H. comosum*, *B. setifolius*, *P. ligularis*) accounted for more than 50% of the seedlings, whereas the secondary forage species (*P. speciosa*) accounted for less than 40% of the seedlings. Conversely, in the more degraded states (III and IV), *P. speciosa* was the most abundant species, accounting for 50–70% of the seedlings. Finally, bare soil and litter (i.e., open) were the microsites that recorded the lowest density of emerged seedlings during the three years of the study.

The influence of ecosystem degradation on the Relative Interaction Intensity (RII), between potential nurses and seedlings, was different for the three functional types. In 2007, deep-rooted shrubs had facilitative effects on the seedlings emerging and surviving underneath them, independently of ecosystem degradation (Fig. 3a; Appendix C). In contrast, RII between shallow-rooted shrubs and recently emerged seedlings, switched from positive effects in the less degraded states (I and II), to negative effects in the most degraded state (Fig. 3b; Appendix C). However, only the 0.6% of the seedlings whose emergence had been nursed by shallow rooted shrubs had survived two years later (Fig. 3b), making impossible to fit regressions. Tussock grasses recorded the weakest intensity of facilitative interactions with recently emerged seedlings, switching to competitive interactions as degradation increased (Fig. 3c; Appendix C). Also, two years later, none seedling survived underneath this functional type (Fig. 3c). Finally, in 2009, the response of RII to ecosystem degradation showed the same pattern than in 2007. The deep-rooted shrubs recorded positive effects on emerged seedlings independently of ecosystem degradation, with an average RII value of 0.971 (±0.047). Conversely, the

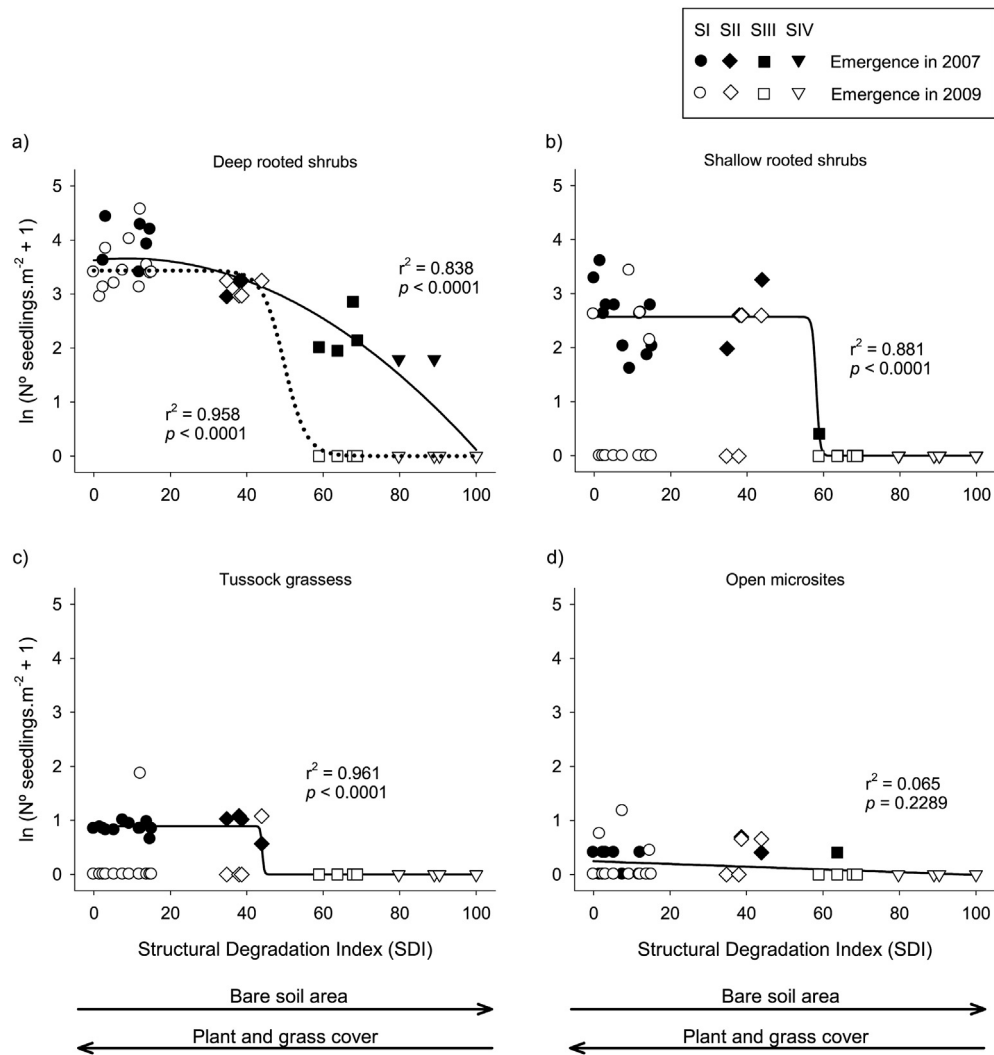


Fig. 2. Density of seedlings that emerged in 2007 and in 2009 underneath deep-rooted shrubs (a), shallow-rooted shrubs (b), tussock grasses (c) and in open microsites (d) in function of ecosystem degradation (SDI). Continuous lines show the regressions fitted for seedling emergence in 2007 and the dotted line shows the regression fitted for seedling emergence in 2009. In (a) regression diagnostics (r^2 and p -value) are shown at top right for seedling emergence in 2007, and at bottom left for seedling emergence in 2009.

effects of shallow-rooted shrubs and tussock grasses on spatially associated seedlings did not show a clear pattern, due to the very low seedling densities that emerged in that year (Fig. 2). Nevertheless, the average RII values were higher for shallow-rooted shrubs than for tussock grasses (0.448 ± 0.838 and -0.44 ± 0.812 , respectively).

3.2. Micro-environmental quality of microsites in alternative states

Mean monthly temperature (mmT) in each state depended on the microsite type for almost every month. Bare soil in both states recorded the highest mmT values, while deep-rooted shrubs of pre-CT state recorded the lowest values (Fig. 4a). Also, deep-rooted shrubs from the pre-CT state were the microsite with the lowest temperature variability (Fig. 4b), reaching the lowest values of mean monthly maximum temperature (<36.3 °C) during the 2008–09 summer season (Appendix B). In contrast, bare soil from post-CT state recorded the highest temperature variability, mainly during the 2008 winter season (i.e. from May to September) and 2009 summer season (i.e., January–February) (Fig. 4b). In this sense, during the 2008 winter season the bare soil from post-CT state registered the lowest values of mean monthly minimum

temperature (reaching -3.6 °C in July 2008), whereas during most of the summer 2009, registered the highest values for mean monthly maximum temperature (Appendix B).

In March and April 2008 the bare soil from the post-CT state had the highest number of hours with temperatures below 0 °C ($p < 0.05$). In March 2008 deep-rooted shrubs from the pre-CT state did not record hours with temperatures below 0 °C, while deep-rooted shrubs from post-CT state showed, on average (\pm SE), 1.3 (± 0.7) hours with temperatures below 0 °C. In contrast, the bare soil from both pre- and post-CT states registered an average of 4.7 (± 0.9) and 6.8 (± 1.2) hours with temperatures below 0 °C, respectively. In April 2008 the average number of hours (\pm SE) with temperatures below 0 °C was 76 (± 17.1) and 95.5 (± 7.5) for the bare soil from pre- and post-CT states, and 44.3 (± 11.0) and 59.2 (± 6.5) for deep-rooted shrubs from pre- and post-CT states, respectively. Finally, soil moisture percentage was similar between states and microsites, reaching on average 15% in the winter season and 2% in the summer season.

4. Discussion

The seedling recruitment decreased sharply as degradation

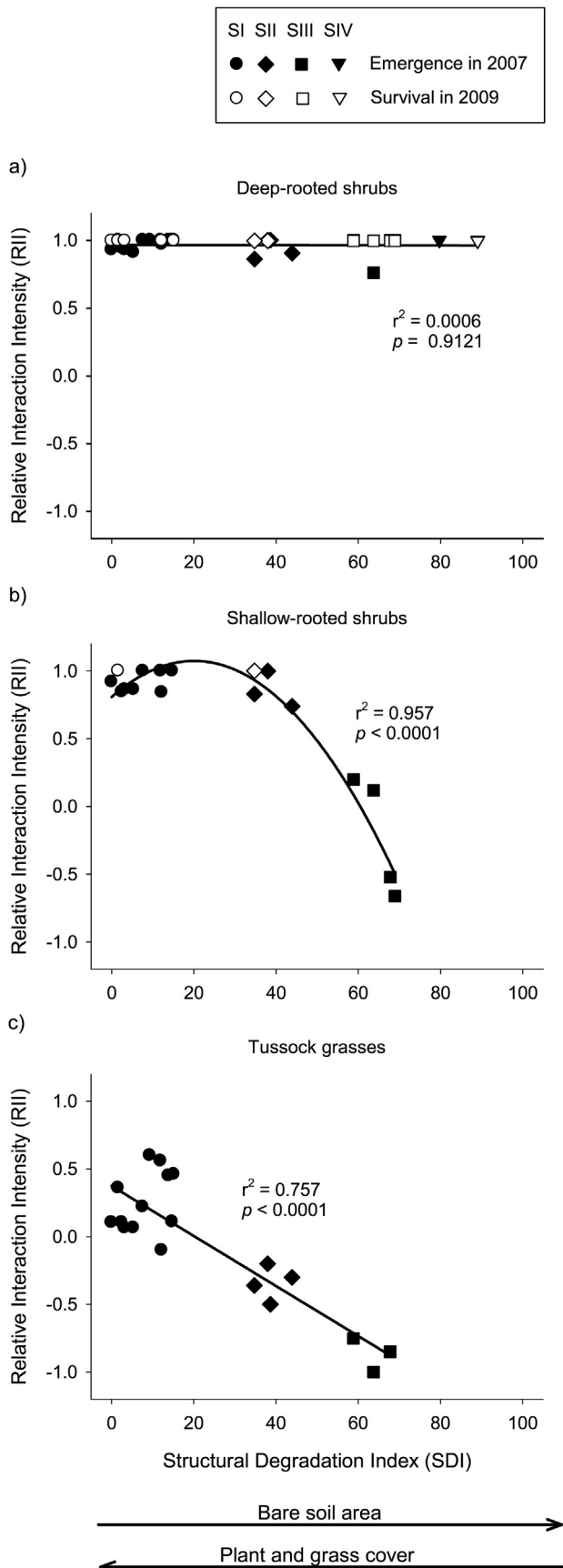


Fig. 3. Relative Interaction Intensity (RII) between nurses, of three functional types, and seedlings along a degradation gradient (SDI). Nurse functional types: Deep rooted

increased, independently of the microsite, being significantly lower in the states that crossed, at least one, critical threshold (Fig. 2). Consequently, degradation would affect ecosystem resilience, limiting the ability of the community to reconstitute its original (or pre-disturbance) composition, structure and patch-configuration, even after livestock removal (Briske et al., 2006; Folke et al., 2004; López et al., 2013). However, recruitment dynamics and the net outcome of plant-plant interactions also depended on the functional type of the potential nurse. Deep-rooted shrubs were the main nurse because facilitated seedling emergence and survival even in the most degraded state (Fig. 3a). In contrast, underneath the other functional types the seedling recruitment was null in post-CT states, and the net balance of plant-plant interactions changed from positive to negative as degradation increased. Therefore, in arid ecosystems, above- and below-ground traits of nurse species have a strong influence in the net outcome of plant-plant interactions.

Degradation caused by historical overgrazing could affect seedling recruitment by changing the quantity and/or the quality of recruitment microsites. We found that seedling density was significantly lower in post- than in pre-critical threshold states (Fig. 2). Our results suggest that the scarce or null recruitment in highly degraded states would be due to a decline in microsite quality rather than quantity, because nurse shrub cover (i.e., deep-rooted shrubs) increased in post-CT states (Appendix A). Also, thermal conditions were different in pre- and post-CT states. The post-CT state recorded more variable and extreme temperatures than the pre-CT state (Fig. 4, Appendix B). This could be because post-CT states had not only lower plant cover and greater bare soil area than pre-CT states (Appendix A), but also lower patch connectivity (López et al., 2013). A lower plant cover and patch connectivity in highly degraded states, increased the edge-effect of bare soil over plant patches, which were more influenced by microclimatic conditions of bare soil. For instance, soil temperature during the growing season was significantly greater in the plant patches of the post-CT state than in those of the pre-CT state (Fig. 4a, Appendix B). This result suggests that a greater bare soil area could decrease the quality of microsites underneath nurse plants in degraded states (Wickens, 1998; Snyman and du Preez, 2005). Therefore, historic overgrazing would generate alternative states with a more dry and unstable microclimate, reducing the quality of microsites available for seedling recruitment.

Plant patches could have greater capacity to retain soil moisture than bare soil due to the greater amount of organic matter and litter in soil and to the buffering effect of vegetation on micro-climatic conditions (Wickens, 1998; Snyman and du Preez, 2005; Gaitán, 2009). However, moisture content was similar between plant patches and bare soil. This could be because plant patches sustain adult and seedling individuals (López et al., 2013; Aguiar and Sala, 1999) which are expected to consume water, thereby increasing water loss by transpiration in plant patches compared to bare soil patches (Whitford, 2002). On the other hand, the soils of the studied ecosystem have a surface sand layer which would interrupt water conduction by capillarity from clay textured deeper layers to the soil surface (Noy-Meir, 1973). For this reason, although bare soil recorded the highest temperatures (Fig. 2), only the first few centimetres of soil (<10 cm) would dry up quickly (D. Lopez, unpublished data) decreasing water evaporation in bare soil. However, the scarce or even null seedling recruitment in open microsites (Fig. 2d) suggest that the desiccation of the surface sand layer would be one

shrubs (a), Shallow rooted shrubs (b) and Tussock grasses (c). RII was calculated for seedling emergence in 2007 and for the seedlings that survived two years later (in 2009).

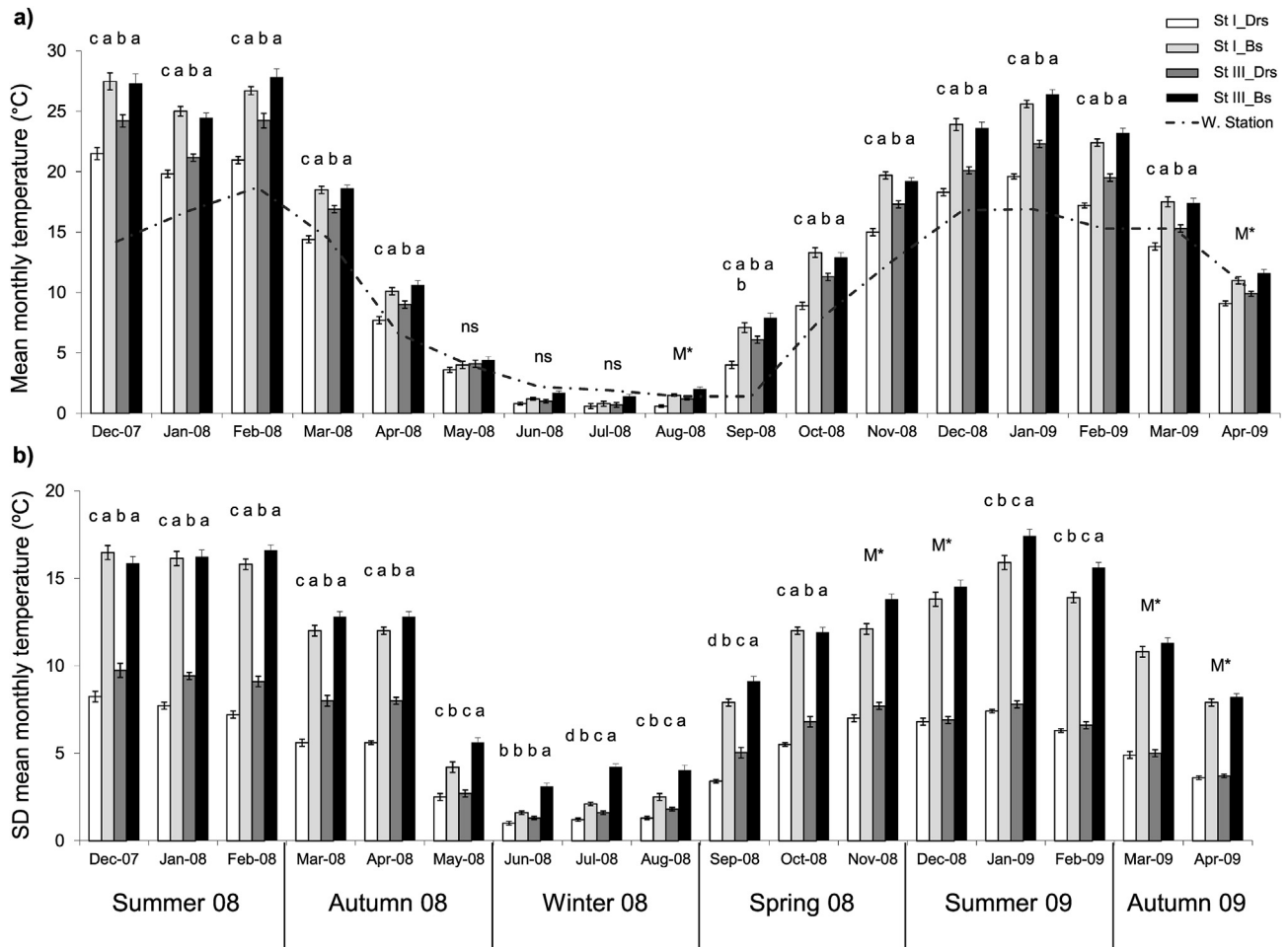


Fig. 4. Mean monthly (\pm SE) sub-superficial soil temperature (a), and mean monthly standard deviation (\pm SE) of sub-superficial soil temperature (b) recorded from December 2007 to April 2009, in two microsites: deep-rooted shrubs (Drs) and bare soil (Bs) in states I and III from Site 2. In (a) W-Station shows the mean monthly air temperature recorded by an automatic weather station (Davis), located in Site 2. Statistical analyses were performed separately for each month. S* indicates differences detected for State factor (State I vs. State III) and M* indicates differences detected for Microsite factor (Drs vs. Bs) ($p < 0.05$). Significant interaction between factors is indicated by different lowercase letters ($p < 0.05$). ns: no significant differences.

of the main constraints to seedling establishment in those microsites. Therefore, any factor that decreases evapotranspiration (e.g. shade of deep rooted shrubs), during the period in which the radicular system of seedlings reach deeper soil layers, will increase the establishment and survival likelihood of these seedlings (Fig. 3a).

The influence of degradation sates on the net outcome of plant-plant interactions was different for the three nurse functional types. Deep-rooted shrubs were the only functional type that recorded positive interactions with seedlings independently of ecosystem degradation (Fig. 3a). This could be due to the following functional traits: (i) hemispherical shape (Damascos et al., 2008), with a high capacity to trap seeds and sediments and to capture and redistribute rainwater (Aguar and Sala, 1999; Whitford, 2002); (ii) spiny triset leaves (Damascos et al., 2008) that protect spatially associated seedlings from herbivore browsing (Aguar and Sala, 1998, 1999; Rebollo et al., 2002); and (iii) deep roots that can absorb water from deep soil layers, reducing competition with seedlings that absorb water from superficial soil layers (Aguar and Sala, 1998; Golluscio et al., 1998). In contrast, the net outcome of plant-plant interactions changed from positive to negative underneath shallow rooted shrubs and tussock grasses as degradation increased (i.e., State II, III and IV) (Fig. 3). This could be because

these species have a superficial radicular system increasing competition for water with spatially associated seedlings (Fig. 3) (Aguar and Sala, 1998, 1999). Besides, both species would not provide an efficient protection against herbivory due to their conical shaped canopy, leaving the seedlings growing under their canopy more accessible to herbivore detection and browsing. Consequently, our results suggest that the hemispherical shape of the canopy, the presence of mechanical defenses and the deep roots were key traits that could explain the persistence of facilitative interactions underneath deep-rooted shrubs, even in highly degraded states.

Recruitment is a dynamic process which shows an interannual variation depending on the occurrence of wet, normal or dry years (Tielbörger and Kadmon, 2000; Armas and Pugnaire, 2005) (Fig. 3). In arid zones, changes in water availability can affect interactions between plants, which can switch from facilitation to competition and viceversa (Holmgren et al., 1997; Tielbörger and Kadmon, 2000). This switching between positive and negative effects may be relevant to the success of the recruitment process, since different results can be obtained in the same microsite but in different years, depending on annual precipitations. In this context, in 2007 rainfall was relatively close to the historical average (López, 2011) and almost all seedlings survived underneath deep-rooted shrubs

(Fig. 3b). In contrast, between October 2007 and April 2008 a severe drought occurred, during which few seedlings emerged, mainly below deep-rooted shrubs, and survival was null. Finally, in 2009 precipitation was around the historical average (López, 2011), and seedlings emerged mostly underneath deep-rooted shrubs (Fig. 3c). In summary, during years with climatic conditions oscillating around the historical average, seedling recruitment would be strongly associated to deep-rooted shrubs, since underneath other functional types competition would prevail. Conversely, in extremely dry years competition for water would be more relevant even underneath deep-rooted shrubs, since water availability becomes so low that positive effects (i.e., reduction of thermal amplitude, soil water evaporation and transpiration caused by nurse shade) cannot outweigh negative effects (i.e., water competition and rainfall interception) that adult individuals cause on the seedlings micro-environment (Holmgren et al., 1997; Maestre et al., 2009a; Holmgren and Scheffer, 2010). Although long term studies are needed to establish if the patterns we found are consistent over time, the variability in annual rainfall (i.e., the occurrence of years with precipitation near to the historical average is not greater than that of dry or wet years; Bustos, 2006, Godagnone and Bran, 2009) is the rule rather than the exception in many arid and semi-arid ecosystems (Noy-Meir, 1973; Whitford, 2002).

4.1. Implications for management and restoration in arid ecosystems

Many rangelands are degraded due to the interaction of overgrazing, fuel wood extraction and recurrent droughts (del Valle et al., 1998; Reynolds and Stafford Smith, 2002; Adeel et al., 2005). This scenario raises the need to implement plans to restore the ecological and productive functionality of degraded ecosystems. Our results suggest that the functional traits of the

dominant species of the plant community could be used to select key nurse species which should be specially considered in management and/or restoration practices in arid ecosystems. Thus, we propose different management or restoration strategies depending on the level of ecosystem degradation, which could be applied in regions where climate regime, soil and/or water availability are marginally suitable for grasslands, and where shrubs are a structural component of the ecosystem (i.e., which can be found in conserved and degraded states as well):

(i) In conserved environments (pre-CT states), hemispheric deep-rooted shrubs should be preserved, as they have a key role on ecosystem structuring and functioning (Whitford, 2002; Aguiar and Sala, 1999). Although some Patagonian-local farmers and field technicians believe that reducing the abundance of shrubs can increase forage grass production; ensuring shrub persistence may allow seedling recruitment of grass species by facilitation.

(ii) In degraded environments (post-CT states), deep-rooted shrubs are essential elements for ecosystem restoration (Gómez-Aparicio et al., 2004; Maestre et al., 2009b; Howard et al., 2012), because they could nurse native forage species. However, restoration practices would depend on the post-CT state in which the ecosystem is found. For example, in degraded states with low soil erosion (e.g. soil loss <5 cm, such as state III) we suggest to plant the foundational species, *P. ligularis* underneath deep-rooted shrubs (restoration pathway between states III and II; Fig. 5). In highly degraded states, with high soil erosion (e.g. 10–15 cm of soil loss, such as state IV), recovery to an original or previous state (i.e., state I or II) would be very unlikely and may take too much time, cost and energy (Bonvissuto et al., 1993; López, 2011). In such states, we suggest to plant native species resistant to water stress conditions (e.g.

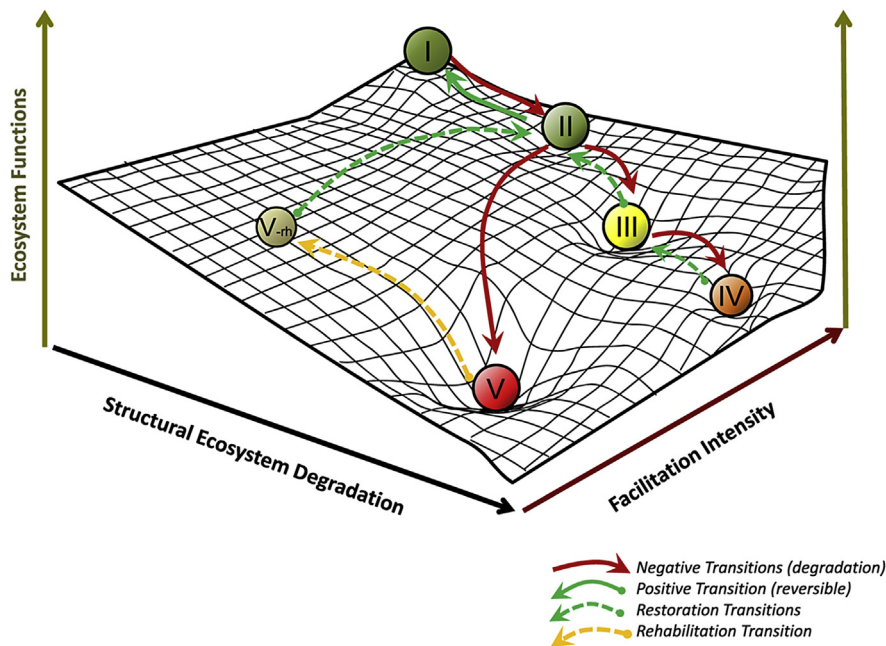


Fig. 5. Schematic representation of the states and transitions for the studied ecosystem based on three axes: Structural Ecosystem Degradation (x axis), Ecosystem Functions (y axis) (see more on López et al., 2013), and Facilitation intensity (z axis). States I to IV are those considered in this study, state V was described by Gaitan (2009) and state Vrh is hypothetical. The transitions between states are represented with different arrows. Negative transitions (continuous red arrows) are associated with degradation caused by overgrazing (stoking rate and time). Positive transition (reversible, continuous green arrow) is based on the elimination of the disturbance factor (grazing exclusion). Restoration transitions (dashed green arrows) rely on re-vegetation of native forage-grasses under deep-rooted shrubs; and the rehabilitation transitions (dashed yellow arrows) should be aimed at increase the cover or abundance of key nurses as deep-rooted shrubs. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

P. speciosa var. *speciosa*, which is the dominant grass in state III; restoration transition between states IV and III, Fig. 5) underneath deep-rooted shrubs. Finally, in extremely degraded ecosystems, that have undergone a very fast degradation process (>20 cm of soil loss), which allowed the survival and expansion of unpalatable sub-shrubs and short-grasses, and caused a reduction in shrub cover (<5%) (described by Gaitán, 2009), restoration practices should be aimed at increase the cover of deep-rooted shrubs (*M. spinosum*). Revegetation practices with this shrub would increase the quantity of microsites favourable for grass recruitment (e.g. as they trap sediments, organic matter and seeds, and provide shade). This rehabilitation practice may increase the structural-functional levels (e.g. greater plant cover and productivity) (rehabilitation transition between states V and V_{rh} , Fig. 5). Finally, when the re-habilitated state (V_{rh} , Fig. 5) has accumulated enough sediments and litter underneath shrubs, it could be restored by sowing the forage grass species that originally dominated that ecosystem (e.g. *P. ligularis*; restoration transition between IV_{rh} and II, Fig. 5).

In the last decades a debate about shrub encroachment (or thickening) into rangelands and grasslands has gained attention because this phenomenon was considered by several authors as a consequence of degradation and, at the same time, a limitation to natural recovery or restoration practices (Maestre et al., 2009b and references therein). However, new evidence suggests that shrubs could be essential components to prevent or even reverse degradation in arid ecosystems (Maestre et al., 2009b; Eldridge et al., 2011). In this sense, our results show that shrubs with key functional traits play an essential role in ecosystem structuring and functioning, facilitating seedling emergence and survival of perennial species (mainly grasses). Thus, shrubs could play a key role not only in the maintenance of ecosystem integrity, but also in the recovery and restoration of some degraded arid and semi-arid ecosystems.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.actao.2017.01.003>.

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APPENDIX A: Complementary information on the physiognomy and composition of States I, II, III and IV of the grass-shrubby steppes of *Poa ligularis* and *Mulinum spinosum* (Western Patagonian District- Argentina).

Sampling procedure to estimate grass and shrub cover and density in each alternative state:

We estimated plant, grass and shrub cover by performing physiognomic-floristic surveys (Daubenmire 1968) in 0.1 m² plots placed at 0.25m intervals along 30 m transects (one in each state of each block). In addition, for each state in each block, we sampled the density of perennial tussock grasses in four 1 x 10 m plots, and shrub density in four 5 x 10 m plots. To evaluate the effect of degradation on vegetation cover variables, we performed linear mixed models (LMM), comparing each enclosure (state I) with its paired historically grazed sector (states II-IV; Fig. 1). In these LMM models, the State factor was analyzed with two levels per site: Site 1= State I and II; Site 2= State I and III; and Site 3= State I and IV. Also, to assess the influence of degradation on physiognomy we performed LMMs comparing the three sites. In these analyses, within each block (B), we considered the difference between the value of each variable from the historically grazed state and the value of its paired enclosure (see Fig. 1). For example, in Site 1, this difference (Δ) variable is calculated as follows: (S1:II_{B1} - I_{B1}), (S1:II_{B2} - I_{B2}), (S1:II_{B3} - I_{B3}), (S1:II_{B4} - I_{B4}). The sub-indexes B1 to B4 represent the block where experimental units were placed, applying the same procedure for difference (Δ) calculation in Sites 2 and 3.

Results:

Post-critical threshold states (hereafter 'post-CT') had lower plant cover than pre-critical threshold states (hereafter 'pre-CT'). Likewise, post-CT states recorded the lowest grass cover, and a very low cover of the main forage species, *P. ligularis* (Table A.1). Whereas, the secondary forage species cover, *P. speciosa* vr. *speciosa*, was highest in state III. Shrub cover was highest in the most degraded state (State IV), associated with high *M. spinosum* and *S. filaginoides* cover (Table A.1).

Post-CT states had higher woody species density (shrubs and sub-shrubs) than pre-CT states. *Senecio filaginodes* recorded higher density in post- than in pre-CT states, whereas *M. spinosum* densities were similar between states. The most degraded state (State IV) had the lowest total grass density. *Poa ligularis* density was lower in post- than in pre-CT states. The highest density of *P. speciosa* vr. *speciosa* was registered in state III, and the lowest in state IV (Table A.1).

Table A.1. Mean values (\pm SE) of vegetation and litter cover and density of individuals in different states (I, II, III and IV) from Site 1 (SI and SII), Site 2 (SI and SIII) and Site 3 (SI and SIV). Significant differences within each site are indicated by *. Significant differences between Sites (Δ variable) are indicated with different lowercase letters ($\alpha = 0.05$). Pre-CT: pre-critical threshold; Post-CT: post-critical threshold.

Variables	SITE 1		SITE 2		SITE 3	
	State I (Pre-CT)	State II (Pre-CT)	State I (Pre-CT)	State III (Post-CT)	State I (Pre-CT)	State IV (Post-CT)
Total plant cover (%)	60.0 (2.9)	50.5 (3.9) a	58.3 (2.3)	42.0 (2.2)* b	62.0 (3.0)	40.0 (1.5)* c
Shrub cover (%)	13.3 (0.5)	14.5 (1.0)	12.9 (0.5)	16.7 (0.7)	10.5 (0.7)	21.1 (0.5)*
^a <i>Mulinum spinosum</i>	7.9 (0.3)	8.1 (0.4) b	7.0 (0.3)	8.2 (0.3) b	6.4 (0.3)	10.0 (0.2)* a
^a <i>Senecio filaginoides</i>	3.9 (0.3)	4.0 (0.3) b	3.9 (0.3)	6.9 (0.3) b	3.3 (0.3)	11.0 (0.4)* a

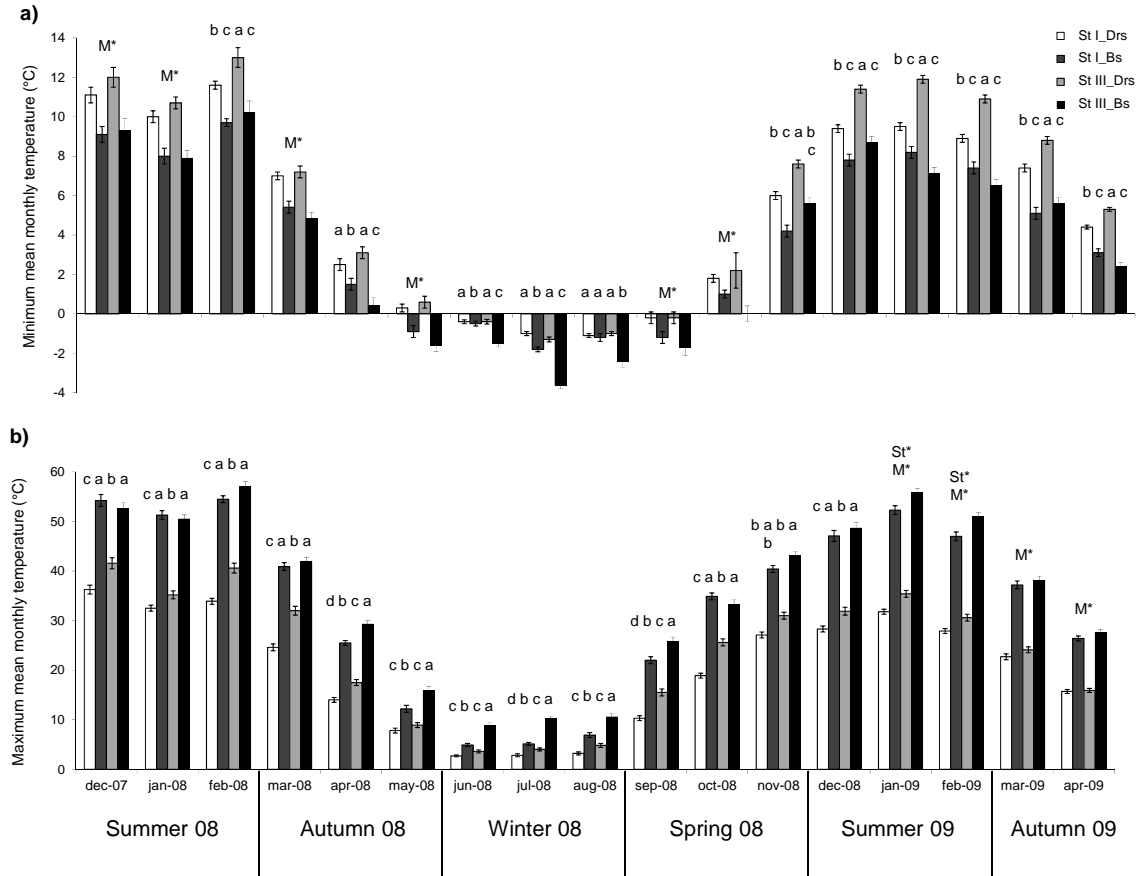
Grass cover (%)	45.0 (2.5)	30.6 (2.0)* a	48.3 (2.3)	23.3 (1.2)* ab	43.0 (2.3)	15.2 (0.9)* b
^b <i>Poa ligularis</i>	39.9 (1.5)	10.1 (1.8)* a	30.5 (1.4)	1.7 (0.1)* b	29.5 (1.3)	0.2 (0.05)* c
^c <i>Pappostipa speciosa</i> var. <i>speciosa</i>	9.0 (0.3)	14.1 (0.9)* b	5.8 (0.5)	17.9 (1.0)* a	9.9 (0.4)	5.6 (0.4)* c
Litter cover (%)	5.0 (2.9)	4.5 (0.9) a	4.6 (1.0)	2.2 (0.4) a	7.5 (0.5)	2 (0.3)* b
Shrub density (n°.m ⁻²)	0.7 (0.02)	0.6 (0.07) b	0.6 (0.02)	1.2 (0.1)* a	0.6 (0.07)	1.1 (0.09)* a
^a <i>M. spinosum</i>	0.2 (0.02)	0.2 (0.01)	0.3 (0.03)	0.4 (0.02)	0.3 (0.06)	0.2 (0.03)
^a <i>S. filaginoides</i>	0.3 (0.02)	0.2 (0.02) b	0.1 (0.01)	0.4 (0.07)* a	0.1 (0.05)	0.4 (0.04)* a
Grass density (n°.m ⁻²)	5.0 (0.4)	6.1 (0.5) a	8.3 (0.5)	9.4 (0.9) a	7.8 (0.2)	3.2 (0.4)* b
^b <i>P. ligularis</i>	2.3 (0.1)	1.7 (0.3) a	4.0 (0.4)	0.4 (0.03)* b	3.4 (0.2)	0.1 (0.04)* b
^c <i>P. speciosa</i> var. <i>speciosa</i>	1.9 (0.2)	3.1 (0.5)* b	2.7 (0.3)	8.3 (0.4)* a	2.6 (0.7)	1.2 (0.1)* c

^aMain shrub species (*M. spinosum*: deep-rooted shrub; *S. filaginoides*: shallow-rooted shrub); ^bmain forage species; ^csecondary forage species.

Reference:

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APPENDIX B. Minimum (a) and maximum (b) mean monthly temperature of sub-superficial soil temperature (\pm SE) from December 2007 to April 2009, recorded underneath deep-rooted shrubs (Drs) and in Bare soil (Bs) in State I and III from Site 2. Statistical analyses were performed for each month separately. St* indicates significant differences for State factor (State I vs. State III) and M* significant differences for Microsite factor (Drs vs. Bs) ($p < 0.05$). Significant interactions between factors are indicated by lowercase letters ($p < 0.05$).



Appendix C. Adjusted determination coefficients and Akaike Information Criterion (AIC) of the simple and quadratic or sigmoid regressions of response variables in function of the Structural Degradation Index (SDI). P-value for comparison between models (simple vs. quadratic or sigmoid regression). Difference between Akaike Information Criterion for both models (Δ AIC). Quadratic or sigmoid regressions were performed when significant differences between models were detected and when Δ AIC between models was greater than 2.

Response variable	Trans-formation	Adjusted r^2 simple regression	AIC simple regression	Adjusted r^2 best fit model	AIC best fit model	Fitted curve	Model comparison (p)	Δ AIC
Seedling density underneath deep-rooted shrubs (2007)	ln ($y + 1$)	0.744	47.06	0.823	39.04	Quadratic	0.0034	8.02
Seedling density underneath deep-rooted shrubs (2009)	ln ($y + 1$)	0.806	57.38	0.937	33.55	Sigmoid	<0.0001	23.83
Seedling density underneath shallow-rooted shrubs	ln ($y + 1$)	0.730	52.92	0.881	42.13	Sigmoid	0.0051	10.79
Seedling density underneath tussock grasses	ln ($y + 1$)	0.731	0.47	0.961	-47.91	Sigmoid	<0.0001	48.38
RII between deep-rooted shrubs and associated seedlings	none	-0.049	-55.18	-0.076	-53.75	Linear	0.4913	1.43
RII between shallow-rooted shrubs and associated seedlings	none	0.686	11.30	0.935	-13.21	Quadratic	<0.0001	24.51
RII between tussock grasses and associated seedlings	none	0.747	3.37	0.767	2.68	Linear	0.1385	0.69