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Contributions of ecological facilitation for restoring environments with high conservation value in the Argentine Patagonia

Contribuciones de la facilitación ecológica para la restauración de ambientes de alto valor de conservación en la Patagonia Argentina

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Abstract. The Argentine arid Patagonia, in general, and 'La Payunia' ecosystem in particular, are important areas for conservation due to the high number of endemic species. However, this region presents a high level of degradation as a result of human activities. Thus, ecological rehabilitation strategies need to be implemented to recover certain areas. The main objectives of this study were to (1) evaluate the reintroduction of two selected native species in severely degraded areas, and (2) determine if the survival and growth of an endemic shrub species, *Senna arnottiana*, increase when it is associated with a perennial grass, *Pappostipa speciosa*. These two species were selected based on the results of a reference ecosystem analysis. In addition, two doses of hydrogel (polyacrylamide) were tested to evaluate its moistening effect. After three years, the survival rate of both plant species was high (mean=79.6%), with no significant differences among hydrogel treatments and association between species. However, growth rates of *S. arnottiana* plants nursed by *P. speciosa* were significantly higher ($P<0.05$) than those of isolated plants. Our results suggest that the study, endemic shrub species is able to survive in isolation after its transplanting, and that *P. speciosa* is a suitable species for promoting a higher shrub growth during the first critical years of recovery of the degraded, arid and semiarid environments.

Keywords: Hydrogel; Plant Nursing; Reference ecosystem; Restoration; Survival.

Resumen. La Patagonia Árida Argentina, en general, y el ecosistema 'La Payunia' en particular, son áreas de interés para la conservación debido a su riqueza florística y endemismos. A pesar de ello, la misma está afectada por impactos que generan la necesidad de implementar proyectos de rehabilitación ecológica. Los objetivos principales del trabajo fueron: (1) evaluar especies con potencial para ser introducidas en sitios severamente degradados, y (2) determinar si la supervivencia y crecimiento de una especie arbustiva endémica, *Senna arnottiana*, aumenta cuando se la asocia a un pasto perenne, *Pappostipa speciosa*. Estas dos especies fueron seleccionadas a partir del análisis del ecosistema de referencia. Luego de tres años determinamos si la supervivencia y crecimiento de *S. arnottiana* aumentó cuando estuvo asociada con *P. speciosa* y con agregado hidrogel. Los resultados mostraron que ambas especies tienen alta supervivencia (promedio=79,6%), sin diferencias significativas entre los tratamientos: distintas dosis de hidrogel, y asociación entre las especies. Sin embargo, el crecimiento de *S. arnottiana* fue significativamente mayor ($P<0,05$) cuando estuvo asociada a *P. speciosa* que cuando estuvo aislada. Nuestros resultados mostraron que la especie arbustiva endémica fue capaz de sobrevivir aislada luego de ser trasplantada, y que *P. speciosa* fue una especie capaz de facilitar su crecimiento durante los primeros años de recuperación de ambientes áridos y semiáridos degradados.

Palabras clave: Hidrogel; Plantas nodrizas; Ecosistema de referencia; Restauración; Supervivencia.

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INTRODUCTION

The arid and semiarid Patagonia region in southern Argentina presents high biodiversity levels and significant endemic species richness. This makes this region of great interest and high priority for conservation, and improved planning and management at a landscape scale (Beeskow et al., 2005). Over the past century, this area has been affected by overgrazing, oil prospecting and extraction, continual and excessive firewood extraction, and introduction of non-native species (Morello et al., 2012). As a consequence, several areas of Argentina's Patagonia have reached levels of severe to highly severe desertification (del Valle et al., 1998; Mazzonia & Vazquez, 2009). This situation is aggravated by the extremely slow regeneration of plants (Fiori & Zalba, 2003; Pérez et al., 2010; González et al., 2013). Thus, ecological restoration and/or rehabilitation techniques should be applied to accelerate the recovery of lost ecosystem attributes and the resilience of degraded ecosystems, (Lake, 2013). Facilitation and, particularly, the use of nurse plants in rehabilitation and restoration projects have been implemented in several biomes including tropical rain forests (Kitzberger et al., 2000), seasonally dry forests (Lebrija-Trejos et al., 2010), and arid and semiarid regions (Padilla & Pugnaire, 2006). The importance of a facilitation initiative for plant community restoration theoretically increases as the environmental stress intensifies (Bertness & Callaway, 1994). Thus, in arid and semiarid areas, where environmental conditions are relatively harsh, ecological facilitation is expected to play an important role in supporting and maintaining biodiversity and ecosystem functioning (Pugnaire, 2010).

An important obstacle for the selection of nurse species in arid and semiarid reference ecosystems is the scarcity of information about both the ecophysiology of native species and the ecological succession (Abella, 2010). In the Patagonian semiarid ecosystems, it has been hypothesized that ecological succession begins with shrubs that colonize bare soils and that, subsequently, generate beneficial conditions that facilitate the establishment of grasses (Soriano et al., 1994; Aguiar & Sala, 1999). This process has also been reported in other deserts of the world (Mauchamp et al., 1993; Schlesinger & Pilmanis, 1998). Thus, several shrub species have been frequently used as nurse plants in restoration projects (Tirado & Pugnaire, 2003; Padilla & Pugnaire, 2009). In other semiarid regions, perennial grasses also showed the ability of acting as facilitators (Gasque & García Fayos, 2004; Soliveres Codina, 2010). Regarding ecophysiological parameters, tolerance to drought varies not only among species but also within a single one (Zavala & Ravetta, 2001), which results in a more complex evaluation of the interactions and the practical application of the facilitation concept in ecological rehabilitation and restoration.

In the semiarid regions of the Argentine Patagonia, ecological rehabilitation with native species has been carried out in sites under high environmental stress using seedlings or transplanting mature plants with moisturizing gel (Pérez et al., 2011). However, further research should elucidate the causes of dissimilar survival values (Altamirano & Pérez, 2010; González, 2010; Becker et al., 2013; Ciano, 2013). The effectiveness and results of hydrogel application are usually variable because the success of seedling survival is generally determined by the occurrence of precipitations higher than the historic values, the growth rate of the root system (Ciano, 2013) or the texture and structure of the disturbed soils (Becker et al., 2013; Beider et al., 2013).

Therefore, the objectives of this study were twofold: 1) evaluate the vegetation of sites with different levels of degradation to select species with the potential to colonize severely degraded areas; and 2) assess the facilitation effect of a perennial grass, *Pappostipa speciosa*, on the survival and growth of reintroduced seedlings of an endemic shrub, *Senna arnottiana*, using different volumes of polyacrylamide (hydrogel).

MATERIALS AND METHODS

Study site. The study site is located in a multiple use protected area of the Auca Mahuida Nature Reserve, Neuquén Province, Argentina. This reserve harbors a variety of taxa such as *Lama guanicoe* (guanaco), a camelid native to South America, several endemic species of lizards (Avila et al., 2013) and a combination of plant communities that represent three phytogeographic provinces, i.e., Monte, Patagonica and Altoandina. In addition, this reserve protects several archaeological sites (Fiori & Zalba 2000, 2003).

In the protected area, the vegetation is abundant and consists of grasses and low-growing plants with an approximate cover of 40% (Fiori & Zalba, 2003). The floristic studies carried out in the Auca Mahuida Reserve report a total of 41 families, 116 genera and 225 species, of which 14 are endemic and distributed in plant communities according to altitude (Long, 2000). Below 1400 m.a.s.l., the plant community is typical of the Monte Province, integrated by species such as *Larrea* spp., *Bougainvillea spinosa, Cercidium praecox, Fabiana peckii, Monttea aphylla* and *Prosopis flexuosa* var. *flexuosa.* At higher altitude, the grass proportion increases, especially with genera such as *Pappostipa* and *Poa*. Above 1500 m.a.s.l., the floristic composition is typical of the Patagonian Province (district Payunia), with species such as *Anarthrophyllum elegans, Mulinum spinosum, Gallardoa fischeri, Ephedra frustillata, Acaena caespitosa, Maihueniopsis darwinii* var. *Hickenii, Maihuenia patagonica, Polygala spinescens, Adesmia boronoides, Patagonian trevoa* and *Schinus roigii*. At the summit of the volcano, species such as *Calceolaria* sp., *Azorella* spp., and *Acaena* spp. belong to the Altoandina Province (Long, 2000).

In relation to the fauna, the reserve is inhabited by several threatened or vulnerable species of birds and mammals. Among the birds are the choique petiso (*Pterocnemia pennata*), condor (*Vultur graphus*) and black-backed eagle (*Garanoaetus* *melanoleucus*). Among mammals are the piche (*Zaedius pichiy*), mara (*Dolichotis patagonum*), puma (*Felis concolor*) and guanaco (*Lama guanicoe*)(Morello et al., 2012).

In the protected area, the average annual rainfall is 140-160 mm (Martínez Carretero, 2004). The hydric deficit of this reserve is the highest of the region (600 mm), and the potential evapotranspiration values range from 700 to 750 mm. The winds are intense and can reach speeds of 80 km/h (Morello et al., 2012). Currently, a large area of the reserve is affected by overgrazing and habitat fragmentation caused by oil prospecting and extraction (Fiori & Zalba, 2000).

Species selection for the facilitation experiment. We evaluated the species composition and cover in habitats with different types of disturbances (low, intermediate and intense) in areas near the sites to be rehabilitated. The sites were as follows: two sites with natural disturbance (e.g., native fauna herbivory, eolian erosion), two sites in a shallow limestone quarry abandoned five years ago, and two road edges. Three 50-m-transects were established on each site using the line intercept method (Mateucci & Colma, 1982). In each transect, all species were identified, and their cover percentage was measured.

Once we obtained these data, we followed several criteria for the selection of species: a) the nurse species could inhabit a wide range of habitats; b) at least one of the species should be endemic to this protected area; c) the selected species should not be too palatable to native herbivores and livestock; d) the nurse plant should grow quickly to provide protection against strong winds; and e) the plants should belong to two functional groups to fulfill different roles in the ecosystem to be rehabilitated (Clewel & Aronson, 2006). Based on these criteria, we selected one shrub and one grass species for the experiments.

Plant facilitation experiment. The site where we performed the experiments is a former one-hectare limestone quarry located at 37° 44' 23" S and 68° 54' 6" W and at 1938 m.a.s.l. It was abandoned 5 years prior to our planting experiment. Previous studies focused on the characteristics of the soil (González & Pérez, 2013) indicated that it presents low organic matter (0.42%), a moderate alkalinity level (pH = 7.72) but no salinity (EC = 1.67 mmho/cm).

We applied four different treatments to the selected plant species: 1) *S. arnottiana* with 1.5 L of hydrogel; 2) *S. arnottiana* nursed by *P. speciosa* with 1.5 L of hydrogel; 3) *S. arnottiana* with 1 L of hydrogel; and 4) *S. arnottiana* nursed by *P. speciosa* with 1 L of hydrogel. For each of the four treatments, we made three replicates separated by 40 meters one from each other. The total number of specimens used was 324 (216 individuals of *S. arnottiana* and 108 of *P. speciosa*). We planted 9-month-old *S. arnottiana* seedlings pruned at 0.1 m height. These seedlings grew in nursery gardens, following several protocols to reach an appropriate genetic diversity for restoration projects (Bischoff et al., 2008; Vander Mijnsbrugge et al., 2010). Likewise, we followed the methods of previous studies (Farinaccio et al., 2013) to obtain *P. speciosa* plants for direct transplants by removing individual plants from well-developed tussocks to minimize detrimental effects on the natural cover.

We planted both species simultaneously during March, 2009. The plantation method had a low impact because we used a mechanic digging machine to make holes at 40 cm depth and 22 cm diameter. On February of 2013, we evaluated the survival and development of *S. arnottiana* plants by measuring their length, from the cotyledon node to the last green leaf. We did not measure the growth and survival of *P. speciosa* because its survival rate was 100%. Moreover, this species does not alter its cover under stress.

Data analysis. A principal component analysis (PCA) was performed to arrange the sites on the axes that explained the greatest variation of the data (PCA; Harris, 2001). This analysis identified the species most associated to the main axis of variation as well as the species with the greatest association among each other. In addition, this analysis determined whether sites with different state of degradation differed along these axes.

We generated contingency tables (chi-square) for the plant survival analyses. As the data did not meet the requirement of homogeneity of variance, we evaluated plant growth with the Mann-Whitney U test (Sokal & Rohlf, 1999) considering the addition of hydrogel and the facilitation with *P. speciosa* as variables. The analyses were performed with SPSS statistics version 20 (IBM Corporation), Info Stat version 2014 (di Renzo et al., 2014) and Statistica 7.0 (StatSoft).

RESULTS

Ecosystem study for species selection. We found 21 species in the reference ecosystem with a cover that ranged between 40 and 50%. In areas subjected to periodic disturbances, we found 10 species with a cover that ranged between 13 and 18%, while in the interior of the quarry, the cover was less than 1% with 4 species: *Astragalus pehuenches*, *Bromus* sp., *Grindelia chiloensis* and *Pappostipa speciosa* (Table 1).

Axis 1 of the PCA explained 43.5% of the total variation and differentiated the sites of the reference ecosystem that had Monte species, which were arranged on the positive side of the axis. The other sites were arranged on the negative side of the axis. The species that contributed the most to the principal component 1 were the following: *A. seriphioiides*, *Boopis* sp., *B. spinosa*, *C. megacarpa*, *E. ochreata*, *Gutierrezia*, *Nassauvia*, *Pappostipa humilis* and *Prosopis denudans*. The species were associated in three groups: species of the Monte (M), species of disturbed environments of the Monte (Md) and a third group of plants associated with the steppe ecosystem. Several species showed no association with a particular environment,

Table 1. Species composition and vegetation cover in each environment: ER1: reference ecosystem in site 1; IC1: quarry interior in site 1; BC1: road edge in site 1; ER2: reference ecosystem in site 2; IC2: quarry interior in site 2; BC2: road edge in site 2.
Tabla 1. Composición de especies y cobertura de vegetación en cada ambiente: ER1: ecosistema de referencia en el sitio 1; IC 1: interior de cantera en sitio 1; BC1: borde de camino del sitio1; ER2: ecosistema de referencia en el sitio 2; IC2: interior de la cantera en el sitio 2; BC2: borde de camino en el sitio 2.

	Environments					
	ER 1	**ER 2**	**I C1**	**IC2**	**BC 1**	**BC 2**
Total cover	44.27%	38.09%	1.23%	0.11%	13.07%	17.89%
Species						
Acantholippia seriphioides	X					
Acaena caespitosa		X				
Astragalus pehuenches	X		X			X
Boopis sp.	X					
Bougainvillea spinosa	X					
Bromus sp.			X			
Condalia megacarpa	X	X				
Ephedra ochreata	X					
Fabiana peckii	X				X	X
Grindelia chiloensis	X	X	X	X	X	X
Gutierrezia sp.	X					
Larrea divaricata						X
Monttea aphylla					X	
Mulinum spinosum		X				X
Nassauvia sp.	X					
Pappostipa humilis	X					
Pappostipa ibarii		X				
Pappostipa speciosa	X	X	X		X	X
Poa dusenii		X				
Poa ligularis	X	X			X	X
Prosopis denudans	X					
Prosopidastrum globosum	X				X	
Schinus johnstonii	X				X	
Senna arnottiana						X
Junellia ligustrina					X	

including *P. speciosa*, *P. ligularis*, *S. arnottiana*, *S. johnstonii* and *G. chiloensis* (Fig 1). Thus, these species met the selection criterion (a). In addition, *S. arnottiana* met the endemism requisite (Martinez Carretero, 2004). *Poa ligularis* and *S. johnstoni* are highly and moderately palatable, respectively (Kröpfl & Villasuso, 2012), while *P. speciosa* and *S. arnottiana* are unpalatable (criterion c; Kröpfl & Villasuso, 2012). *Senna arnottiana* presented low values of palatability due to its xenoxyd content (Arambarri, 2002), while *G. chiloensis* has a moderate palatability (Beider, 2012). *Pappostipa speciosa* has a considerable vegetative growth, it expands laterally and maintains a high percentage of standing dead biomass (Soriano, 1956), which provides protection for other plants against the strong winds and direct solar radiation of the area. Furthermore, this species can perform this protective function (criterion d) because it resists direct transplants from the reference ecosystem (Farinaccio et al., 2013). *Senna arnottiana* is a shrub species that fixes nitrogen (Álvarez et al., 2013), and *P. speciosa* is a grass. Thus, these two species belong to two different functional groups (criterion e). According to the above, we chose *P. speciosa* (Poaceae) as the nurse plant, and *S. arnottiana* (Fabaceae) as the protected plant.

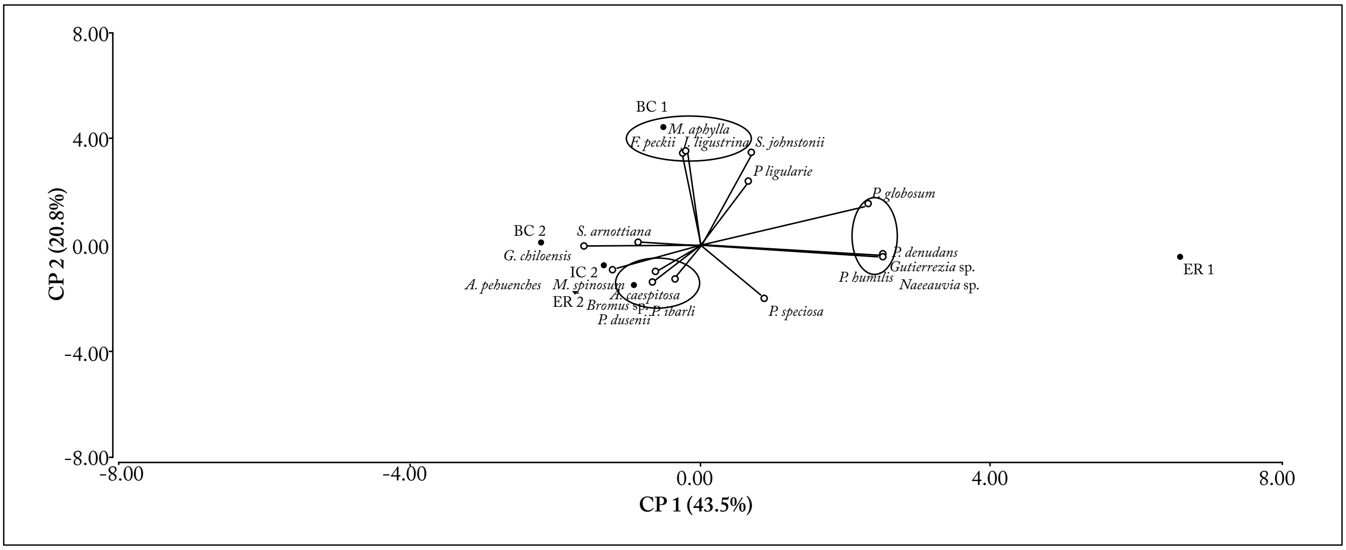

Fig. 1. Biplot graph of the PCA based on species cover in six environments. ER1: reference ecosystem in site 1; IC1: quarry interior in site 1; BC1: road edge in site 1; ER2: reference ecosystem in site 2; IC2: quarry interior in site 2; BC2: road edge in site 2.

Fig. 1. Gráfico Biplot del análisis de componentes principales basado en la cobertura de especies de 6 ambientes: ER1: ecosistema de referencia en el sitio 1; IC 1: interior de cantera en sitio 1; BC1: borde de camino del sitio1; ER2: ecosistema de referencia en el sitio 2; IC2: interior de la cantera en el sitio 2; BC2: borde de camino en el sitio 2.

Survival and growth of *Senna arnottiana*. The average survival of *Senna arnottiana* was 79.6%, with no significant differences among treatments (X^2= 3.42; *df* = 3; P= 0.33; Fig. 2). However, we found significant growth differences between isolated *S. arnottiana* plants and those nursed by *P. speciosa* with 1 L (*U* = 179; P= 0.0035) and 1.5 L (*U* = 174.5; P= 0.0028) of hydrogel. The differences between the two hydrogel treatments were not significant (U = 285; P= 0.17 for isolated plants, and U = 275; P= 0.62 for nursed plants; Fig. 3).

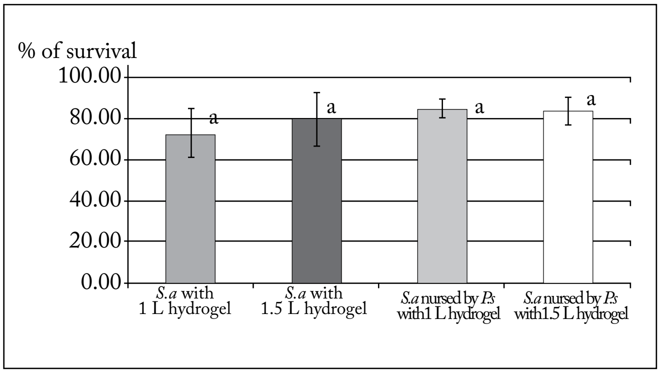

Fig. 2. Percentage of survival of *S. arnottiana* plants under the different treatments: 1) *S. arnottiana* with 1.5 L of hydrogel, 2) *S. arnottiana* nursed by *P. speciosa* with 1.5 L of hydrogel, 3) *S. arnottiana* with 1 L of hydrogel, 4) *S. arnottiana* nursed by *P. speciosa* with 1 L of hydrogel. No significant differences in survival were observed among treatments.

Fig. 2. Porcentaje de supervivencia de las plantas de *S. arnottiana* bajo los distintos tratamientos: 1) *S. arnottiana* con 1,5 L de hidrogel, 2) *S. arnottiana* asociada a *P. speciosa* con 1.5 L de hidrogel, 3) *S. arnottiana* con 1 L de hidrogel, 4) *S. arnottiana* asociada a *P. speciosa* con 1 L de hidrogel. No se observaron diferencias significativas entre tratamientos.

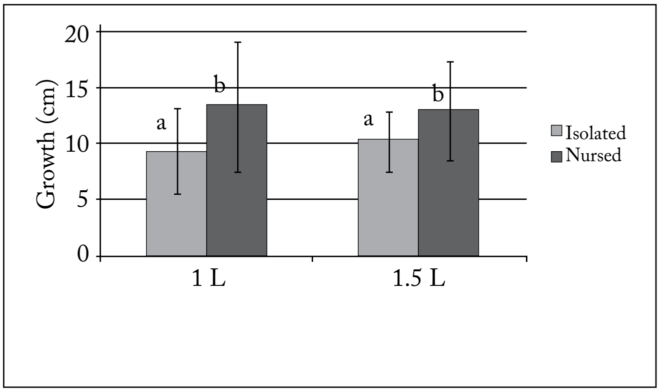

Fig. 3. Average growth and standard deviation of *S. arnottiana* plants isolated or nursed by *P. speciosa* for each hydrogel dose (1 L and 1.5 L). Different letters represent significant differences at the 0.05 level of significance.

Fig. 3. Crecimiento y desvío estándar promedios de *S. arnottiana* plantada aislada o asociada a *P. speciosa* para cada dosis de hidrogel (1 L y 1,5 L). Letras distintas indican diferencias significativas a un nivel de significancia de 0,05.

DISCUSSION

The vegetation analysis showed a high environmental variability in the vegetation cover and species composition of the Auca Mahuida protected area. This highlights the need for studies on reference ecosystems to design an appropriate rehabilitation/restoration project (Tongway et al., 2004; Pérez et al., 2010).

The criteria for selecting species for restoration projects are highly debated. For instance, several authors recommend using species present in the late successional stages, which would enhance the ecosystem resilience (Bonet, 2004). Others authors propose using species present in intermediate successional stages (Miranda et al., 2004). This latter idea is particularly advised for highly degraded areas because these plants are able to establish in harsh environments and boost the plant succession. However, knowledge on the successional stages is limited at our research area. Hence, we established selection criteria based on habitat limitations and ecological roles of the species.

We found a significant survival percentage in the chosen species, and we did not detect differences on the survival of *S. arnottiana* when plants grew in isolation *versus* with *P. speciosa* clumps (Fig. 2). This indicates that *S. arnottiana* could inhabit areas with harsh environmental conditions in isolation. However, *S. arnottiana* plants nursed by *P. speciosa* showed higher growth rates than those grown in isolation (Fig. 3). The increased length of nursed *S. arnottiana* plants constitutes evidence of the positive interaction with *P. speciosa* clumps. Oesterheld & Oyarzábal (2004) demonstrated the facilitating effect of *P. speciosa* on another grass species (*Bromus setifolius*) because it provided protection from grazing, which outweighed the competitive interactions for resources. In our study, the two species involved, *S. arnottiana* (derived from nursery specimens) and *P. speciosa* (derived from transplants), had different life strategies. This niche separation resulted in lower negative interactions (Soriano & Sala, 1984).

Information about the initial establishment and the first years of growth is fundamental in rehabilitation projects that require a large cover to protect the soil. However, it is necessary to conduct long-term follow-up studies to assess whether such differences would result in positive effects on the development of both biomass and reproductive structures as well as on the integration of other native species and soil enhancement in the planting microsites.

In addition, the results of several experiments with hydrogel in soils with different textures and under controlled conditions suggest that higher doses increase seedling survival (Akhter et al., 2004; Al-Humaid & Moftah, 2007). However, we did not find significant differences in seedling survival or growth between the two applied doses. This discrepancy might be due to the fact that, in our study, temperature and humidity conditions were not controlled, and humidity was not a limiting factor during the performance of this experiment. In contrast, when humidity levels in the soil are low, high doses of hydrogel result in a decreased seedling growth because hydrogel might compete for water with the plants (Al-Harbi et al., 1996; Mangold & Sheley, 2007).

CONCLUSIONS

Of all the species described for this area and studied in the surveys, few could meet the basic requirements for their reintroduction in semiarid zones with high environmental variability as the studied area.

The two selected species had high survival values. *Pappostipa speciosa* quickly adapted to the transplants from the reference ecosystem; it was a suitable nurse plant and was able to inhabit at several altitudes a.s.l. The other species, the endemic *Senna arnottiana*, was able to get established in highly degraded sites from nursery gardens both in isolation or with nurse plants.

Although *Senna arnottiana* can survive in degraded sites with no facilitation, its growth was higher when it was associated with nurse plants.

The hydrogel doses used in this study did not show advantages for the survival or growth of *S. arnottiana* specimens.

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