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Filling gaps in seed germination and species selection: work in progress for dryland restoration in Argentina

Llenando vacíos en la germinación de semillas y la selección de especies: trabajo en proceso para la restauración de tierras secas de Argentina

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Abstract

Ecological restoration and rehabilitation (ERR) practitioners lack comprehensive information on the most effective seed dormancy alleviation treatments to enhance germination of native plants from arid and semiarid regions, as well as on survival and growth rates of these species in the field. In this paper we report on the results of a "professional intelligent tinkering" approach to assess seed germination of species from the Monte Austral, an arid region in southern Argentina. We aim to test a small number of seed dormancy alleviation treatments reported to be effective in other arid regions, and to assess germination results for 16 species against existing information on their performance in the field. This approach proved to be highly effective given that, out of the 16 shrub species evaluated, 11 showed germination rates suitable for ERR (i.e., over 50%). Only four species attained both high survival rates and plant cover values in outplantings, while four other species showed high survival rates but their plant cover values were low or not assessed. We argue that this approach, which involves obtaining and comparing data on seed germination rate with performance of the same species in the field, would be strategic for species selection in ERR.

Resumen

Los practicantes de la restauración y rehabilitación ecológica (RRE) carecen de información completa sobre los tratamientos pregerminativos más efectivos para promover la germinación de plantas nativas de regiones áridas y semiáridas, y sobre la supervivencia y crecimiento de estas *especies a campo. Aquí informamos resultados del enfoque "estrategias profesionales inteligentes" para evaluar la germinación de especies del Monte Austral, una región árida del sur de Argentina. Nuestros objetivos fueron probar una pequeña cantidad de tratamientos pregerminativos que son efectivos en otras regiones áridas y evaluar los resultados de germinación de 16 especies en comparación con la información existente sobre su desempeño a campo. Este enfoque demostró ser altamente efectivo dado que, de las 16 especies de arbustos evaluadas, 11 mostraron tasas de germinación adecuadas para RRE (es decir, más del 50%). Solo cuatro especies alcanzaron altas tasas de supervivencia y altos valores de cobertura vegetal en las plantaciones, y otras cuatro mostraron altas tasas de supervivencia, pero la cobertura vegetal fue baja o no se evaluó. Argumentamos que sería estratégico adoptar este modelo, que incluye obtener y comparar información sobre la germinación de semillas y el desempeño de las mismas especies a campo, para la selección de especies en RRE.*

Kewywords: arid and semiarid lands, germination syndromes, dormancy alleviation treatments, large-scale ecological restoration and rehabilitation, DFSA

Palabras clave: *tierras áridas y semiáridas, síndromes de germinación, tratamientos de alivio de la latencia, restauración y rehabilitación ecológica a gran escala, DFSA*

Introduction

Drylands (arid, semiarid, and dry subhumid lands) cover approximately 46% of the Earth's surface and are home to more than 3 billion people (Mirzabaev et al., 2019). They are also home to surprisingly vulnerable ecosystems sensitive to climate chaos, soil and plant cover loss, and other widespread global changes leading to ecosystem degradation and human misery (Huang et al., 2016). Moreover, most drylands worldwide are demonstrably losing biodiversity and topsoil at a catastrophic rate, as a direct result of poor stewardship and anthropogenic climate change (Cherlet et al., 2018). This is resulting in increased poverty, food insecurity, and disease burden in rural communities that depend more or less entirely upon their local ecosystems for sustenance and survival (Mirzabaev et al., 2019). Thus, drylands, and the people who inhabit them, urgently need large-scale ecological restoration and rehabilitation.

Shrubs are often the dominant lifeform in the vegetation communities of many drylands, and, consequently, represent key elements or candidate 'dryland framework species' (DFS) in kickstarting effective, long-lasting ecological restoration or rehabilitation. However, an overwhelming majority of woody shrubs produce seeds possessing some form of dormancy at maturity (Baskin & Baskin, 2014). This represents a significant obstacle to overcome in the restoration of arid and semiarid lands. Although protocols exist to identify seed dormancy syndromes and appropriate methodologies for dormancy alleviation (Kildisheva et al., 2020; Pedrini & Dixon, 2020), in most dryland regions reliable information about seed dormancy and its effective alleviation remains entirely lacking.

In the Monte Austral, of southern Argentina, an arid and highly degraded region where ERR is notably hampered by scarce information regarding seed dormancy syndromes, we suggest that a "professional intelligent tinkering" approach (Murcia & Aronson, 2014) is needed. This approach suggests "shortcuts" may be taken in ERR decision-making where knowledge is incomplete but inferences can be made from preliminary information. For example, the selection of germination treatments inferred to be likely successful from previous studies in climatically-similar regions.

Even where seed dormancy can be reliably and effectively alleviated, a second practical problem that restoration practitioners face is that successful germination procedures alone may be insufficient as a means for selecting the best species for ERR in severely-degraded drylands. For example, species producing seeds that can be reliably germinated in the nursery may possess traits resulting in poor seedling emergence and establishment (Long et al., 2015), or unsatisfactory rates of growth and development when outplanted as seedlings in drylands (Pérez et al., 2019a; Pérez et al., 2020).

To explore how to better integrate seed germination characteristics with other important restoration-relevant metrics, such as successful incorporation in outplanting activities, our objectives in this study were to: a) experimentally evaluate seed germination success in a group of native species from Monte Austral we consider DFS candidate using a suite of well-known, simple, pre-germination treatments, and b) assess the suitability of selected species for ERR in the light of results of germination trials together with available information on field performance (survival and plant cover in outplanting or direct seeding) in our study region.

Material and Method

Study area

Geography and land use

The arid Monte biome covers ca. 466,975 km^2 along a 2400 km NW-SE diagonal, extending from 24°S in a nearly subtropical region to 44°S in a relatively cold region (Morello et al., 2012). The study area is located in the Neuquén Province (Argentina) (**Figure 1**), which harbors the southern, extra-tropical subdivision of the Monte known as the Monte Austral. Here, approximately 32% of the territory suffers from serious ecological and economic degradation, mainly caused by extensive, poorly-managed cattle ranching and poorly or unregulated industrial drilling for gas and oil (Mazzoni & Vazquez, 2009; Morello, 2012).

In Neuquén Province, recent, rampant oil and gas sector activity has led to partial removal and degradation of vegetation over thousands of hectares, highly negatively impacting biodiversity and the health of ecosystems and human communities (Mazzoni & Vazquez, 2009).

Climate and vegetation of the Monte Austral

The mean annual temperature in Monte Austral is 15 °C with high seasonal variation (Morello et al., 2012). Data recorded at our seed collection sites over four years have shown a mean temperature during summer months of c. 25 °C, and c. 9 °C in winter. Over the last 20 years, mean annual rainfall in the study area was 152 ± 60.3 mm, ranging from 52 to 250 mm annually (data from "La Higuera" meteorological station, AIC, personal communication July, 2020).

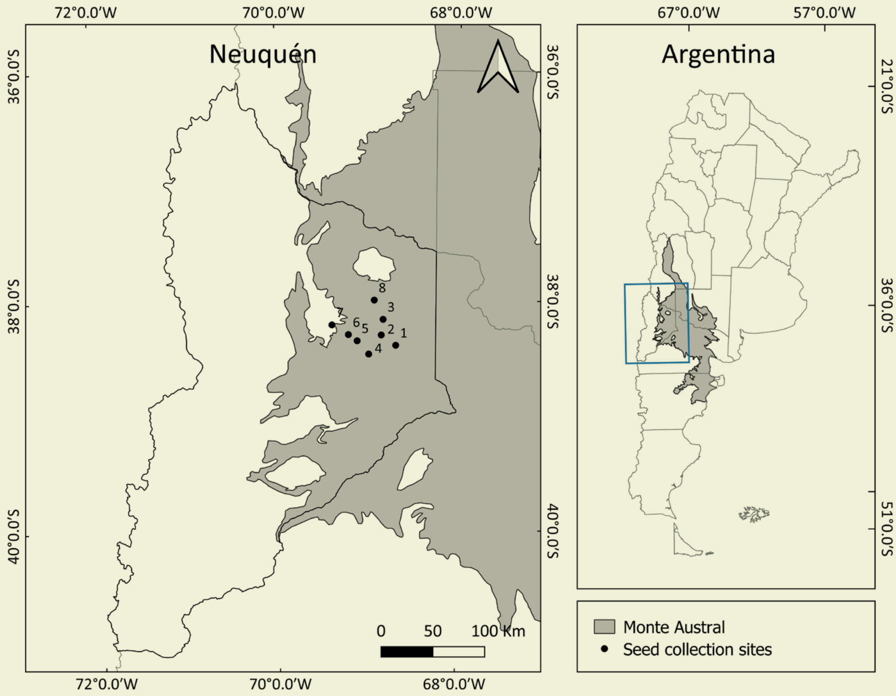

Figure 1. Seed collection sites in Neuquén province, Monte Austral (Oyarzabal et al., 2018). The numbers (1 to 8) refer to the seed collection sites mentioned in **Table 1**. Geographic coordinates system- WGS84

Figura 1. Sitios de recolección de semillas en la provincia de Neuquén, Monte Austral (Oyarzabal et al., 2018). Los números (1 a 8) se refieren a los sitios de recolección de semillas mencionados en la Tabla 1. Sistema de coordenadas geográficas - WGS84

The dominant vegetation type in Monte Austral is steppic, with shrub-dominated patches alternating with patches of very sparse plant cover (Busso & Bonvissuto, 2009). The dominant shrub species include *Larrea divaricata* Cav., *L. cuneifolia* Cav., and *L. nitida* Cav. (Zygophyllaceae), *Monttea aphylla* (Miers) Benth. & Hook. var. *aphylla* (Scrophulariaceae), *Atriplex lampa* (Moq.) D. Dietr. (Chenopodiaceae), *Lycium chilense* Miers ex Bertero (Solanaceae), and *Prosopis flexuosa* DC. var. *depressa* Roig (Fabaceae). Among low-growing perennial life forms, three clump grasses are common, namely *Pappostipa speciosa* var. *speciosa* (Trin. & Rupr.) Romasch, *Panicum urvilleanum* Kunth., and *Poa ligularis* Nees ex Steud, as well as the subshrub *Hyalis argentea* D. Don ex Hook. &Arn. (Asteraceae) (Busso & Bonvissuto, 2009).

Species richness, seed collection and storage

To describe the richness of shrub species, nine 15-meter-long intersecting transects were randomly laid out in seed collection sites (**Figure 1**).

Seed collection was carried out from November 2010 to January 2013 following standard protocols for ecological restoration work (i.e., harvesting seeds from at least 30 to 50 plants in a population without surpassing 20% of the available seeds per plant; Pedrini & Dixon 2020). Following collection, seeds were air dried at room temperature in a ventilated space, manually cleaned to remove impurities, and then stored at -18°C until germination tests were conducted (between May and August 2013).

Seed dormancy alleviation treatments and experimental design

Prior to carrying out germination tests, seed fill was determined manually by squeezing seeds between fingers or fingernails. All unfilled seeds were discarded. Rates of germination were then evaluated quantitatively using a completely randomized design with four pre-germination treatments and a control group (C), each with three replicates of 30 randomly-selected seeds.

The selected treatments were chosen on the basis of the hypothesis of predominance of physiological (PD) and physical dormancy (PY) in cold- or transitional, warm-cold-deserts such as the Monte Austral. Seed were exposed to either a cold-wet treatment for 7 or 30 days (CW7 and CW30), or chemical scarification for 5 or 45 minutes (CS5 and CS45); the treatment times considered effective for most species of the region tested to date (Paredes et al., 2018).

For CW7 and CW30 treatments, seeds were distributed in a single layer on a polystyrene tray lined with thin paper and a layer of paper napkins moistened with water. These were covered with a second layer of paper, wet cotton, and another tray as a lid. The trays were refrigerated at 4°C for 7 or 30 days. For chemical scarification, the seeds were immersed in sulfuric acid (H_2SO_4; 95-98% purity, Cicarelli laboratory) for periods of 5 or 45 minutes.

After the treatments, seeds were placed in Petri dishes lined with a moistened filter paper and placed in a germination chamber (Mechatronics Services brand) under controlled conditions. We used alternating temperatures between 10 ± 1°C for 12 h in darkness and 20 ± 1°C for 12 h, corresponding to the light period. This temperature and light regime emulates climatic conditions in the study region during fall, when seed germination likely occurs naturally (Páez et al., 2005). The criterion for germination was emergence of the radicle. Germination was monitored every two days for 42 days, until no further germination was recorded.

Statistical analysis

To identify groups of species with a similar germination response to dormancy alleviation treatments, we performed a correspondence analysis on species with over 50% germination rates using InfoStat software (Di Rienzo et al., 2014). A binary logistic regression (SPSS Statistics 25, IBM) was used to assess the main interaction effects of CW and CS treatments on the successful outcome of seed germination. When no significant differences in germination between treatments were found, a mean germination time (MGT) was used as supplementary information to identify the average number of days it took for a single seed to germinate according to the following formula:

$$MGT = \frac{\sum_{i=1}^{n} fi.xi}{\sum_{i=1}^{n} xi}$$

Where *fi* is the number of days elapsed since the start of the germination test and *xi* is the number of seeds that germinated within consecutive time intervals.

MGTs were analyzed using one-way ANOVA with Tukey post-hoc tests using the InfoStat software, with a uniform significance level set at 0.05.

Results

A total of 27 shrub species, belonging to 9 botanical families and 23 genera, were recorded in the study area. However, necessary amounts of seeds for laboratory testing could be collected for 16 of these species (**Table 1**). We also considered results for two native species - one small tree, *Parkinsonia praecox* and one shrub, *Senna aphylla* - collected in the same area, for which germination success have been previously reported using the same experimental procedure and design as we describe here (see Paredes et al., 2018).

Germination response to seed dormancy alleviation treatments

A first response group (G0) was identified for the five species that showed almost no germination even after 42 days. Of these, *Larrea nitida* and *Monttea aphylla* did not germinate at all, while *L. cuneifolia, L. divaricata*, and *Neosparton aphyllum* all had germination probability <0.1 (**Table 2**).

Correspondence analysis made it possible to divide the remaining 11 species into three additional subgroups, named G1, G2, and G3, all showing germination success over 50% with at least one experimental treatment (**Figure 2**). The different probabilities of germination success for each species according to treatment in G0, G1, G2 and G3 are shown in **Figure 3**.

Subgroup G1

The four species in this subgroup showed uniformly medium and high germination rates, and showed no significant differences between treatments. In the case of *B. spinosa*, the lowest germination probability (GP) was observed for treatment CS45 (0.19, 0.11-0.27, Exp(B) = 2.471 [1.357, 4.500], Wald = 0, P < 0.005). All other treatments yielded high GP and did not differ statistically significantly from the control (**Table 2**). Most seeds of G1 species germinated within the 30-day stratification period in CW30, and thus CW30 was not included in MGT calculation. Among other treatments, lowest MGT was reported for all G1 species in CW7 (**Figure 4**).

Subgroup G2

Treatments CS5 and CS45 yielded significantly greater GP compared with controls for *P. flexuosa* var. *depressa* (hereafter *P. flexuosa*), *P. praecox* and *S. aphylla*. Germination probability for untreated *P. flexuosa* and *P. praecox* seeds was relatively low (0.30, 0.21-0.39, and 0.28, 0.19-0.37, respectively) but was three-fold higher in CS5 and CS45 for *P. flexuosa* (GP = 0.97, 0.93-1.00, Exp(B) = 219.077 [28.976, 1656.38], Wald = 27.265, P < 0.005) and nearly three-fold higher in CS45 for *P. praecox* (0.87, 0.81-0.94, Exp(B) = 22.000 [9.821, 49.281], Wald = 56.430, P < 0.005). GP was very low for untreated *G. chiloensis* seeds (0.03, 0.00-0.07), but improved markedly in CS45 (0.51, 0.41-0.61, Exp(B) =

Table 1. Native species recorded in the study area. Collected and analyzed species are indicated in bold type. For location of collection sites, see **Figure 1**. Site number 8 corresponds to seeds collected and analyzed in a previous study from this same study area (Paredes et al., 2018)
Tabla 1. Especies nativas registradas en el área de estudio. Las especies recolectadas y analizadas se indican en negrita. Para la ubicación de los sitios de recolección, ver la Figura 1. El sitio número 8 corresponde a semillas recolectadas y analizadas en un estudio previo en la misma área de estudio (Paredes et al., 2018)

Species	Family	Collected Species	Collection sites
Baccharis darwinii Hook. &Arn.	Asteraceae	-	-
Chuquiraga erinaceae Don.	Asteraceae	-	-
Gochnatia glutinosa (D. Don) Hook. & Arn.	Asteraceae	-	-
***Grindelia chiloensis* (Cornel.) Cabrera**	Asteraceae	Y	6
Gutierrezia solbrigii Cabrera	Asteraceae	-	-
***Hyalis argentea* D. Don ex Hook. & Arn.**	Asteraceae	Y	5
***Senecio filaginoide*s DC.**	Asteraceae	Y	2
***Senecio subulatus* Don. ex Hook. et Arn.**	Asteraceae	Y	1
***Atriplex lampa* (Moq.) D. Dietr.**	Chenopodiaceae	Y	4
***Atriplex undulata* (Moq.) D. Dietr**.	Chenopodiaceae	Y	3
***Ephedra ochreata* Miers**	Ephedraceae	Y	2
Adesmia guttulifera Sandwith	Fabaceae	-	-
Anarthrophyllum capitatum (Gillies ex Hook.&Arn.)	Fabaceae	-	-
***Parkinsonia praecox* (Ruiz &Pav. ex Hook.) Hawkins**	Fabaceae	Y	5 and 8
***Prosopis flexuosa* var. *depressa* DC**	Fabaceae	Y	5
***Senna aphylla* (Cav.) H.S. Irwin & Barneby**	Fabaceae	Y	5 and 8
***Bougainvillea spinosa* (Cav.) Heimerl**	Nyctaginaceae	Y	2
***Monttea aphylla* (Miers) Grisebach**	Scrophulariaceae	Y	7
Fabiana peckii Niederl.	Solanaceae	-	-
Lycium sp.	Solanaceae	-	-
Mulguraea ligustrina (Lag.) Moldenke	Verbenaceae	-	-
Glandularia crithmifolia (Gillies ex Hook/.) Schnack & Covas	Verbeneceae	-	-
***Neosparton aphyllum* (Gill. et Hook) Kuntze**	Verbeneceae	Y	2
Acantholippia seriphioides(A. Gray) Moldenke	Verbeneceae	-	-
***Larrea cuneifolia* Cav.**	Zygophyllaceae	Y	5
***Larrea divaricata* Cav.**	Zygophyllaceae	Y	5
***Larrea nitida* Cav.**	Zygophyllaceae	Y	4

Table 2. Germination probabilities (confidence interval) for studied species under different dormancy alleviation treatments (C: control, CS5 and CS45: 5 and 45-minute chemical scarification, CW7 and CW30: 7 and 30-day cold-wet treatments). †As in **Table 1**, data from Paredes et al. (2018). * Indicates GP statistically higher than controls (P<0.05)

*Tabla 2. Probabilidades de germinación (intervalo de confianza) para las especies estudiadas bajo diferentes tratamientos pregerminativos (C: control, CS5 y CS45: escarificación química de 5 y 45 minutos, CW7 y CW30: tratamientos frío-húmedo de 7 y 30 días). † Como en la Tabla 1, datos de Paredes et al. (2018). * Indica valores estadísticamente más alto que los controles (P <0.05)*

Group	Species	C	CS5	CS45	CW7	CW30
G0	*L. cuneifolia*	0	0.02 (0.01-0.08)	0.09 (0.05-0.17)	0.03 (0.01-0.10)	0.02 (0.01-0.08)
	L. divaricata	0.01 (0.00-0.07)	0.02 (0.01-0.08)	0.04 (0.02-0.11)	0.01 (0.00-0.07)	0.01 (0.00-0.07)
	L. nitida	0	0	0	0	0
	M. aphylla	0	0	0	0	0
	N. aphyllum	0	0	0.01 (0.00-0.07)	0	0.06 (0.02-0.03)
G1	*A. lampa*	0.39 (0.29-0.49)	0.49 (0.39-0.59)	0.47 (0.37-0.57)	0.43 (0.33-0.53)	0.53 (0.43-0.63)
	B. spinosa	0.98 (0.95-1.00)	0.93 (0.87-0.98)	0.19 (0.11-0.27)	0.98 (0.95-1.00)	0.98 (0.95-1.00)
	E. ochreata	0.93 (0.87-0.98)	0.94 (0.89-0.99)	0.96 (0.92-1.00)	0.97 (0.93-1.00)	0.93 (0.87-0.98)
	H. argentea	0.97 (0.93-1.00)	0.93 (0.87-0.98)	0.94 (0.89-0.99)	0.98 (0.95-1.00)	0.96 (0.92-1.00)
G2	*A. undulata*	0.39 (0.28-0.48)	0.47 (0.37-0.57)	0.60 (0.50-0.70)*	0.40 (0.30-0.50)	0.07 (0.02-0.13)
	G. chiloensis	0.03 (0.00-0.07)	0.23 (0.15-0.32)	0.51 (0.41-0.61)*	0.12 (0.05-0.18)	0.03 (0.00-0.07)
	P. flexuosa	0.30 (0.21-0.39)	0.97 (0.93-1.00)*	0.97 (0.93-1.00)*	0.27 (0.18-0.35)	0.24 (0.16-0.33)
	P. praecox†	0.28 (0.19-0.37)	0.41 (0.32-0.51)*	0.87 (0.81-0.94)*	0.25 (0.17-0.24)	0.25 (0.17-0.24)
	S. aphylla†	0.21 (0.13-0.30)	0.82 (0.74-0.90)*	0.98 (0.95-1.00)*	0.16 (0.09-0.23)	0.16 (0.09-0.23)
G3	*S. filaginoides*	0.06 (0.01-0.11)	0.52 (0.42-0.62)*	0	0.15 (0.08-0.022)	0.11 (0.04-0.17)
	S. subulatus	0.54 (0.044-0.64)	0.77 (0.68-0.85)*	0	0.39 (0.29-0.49)	0.34 (0.24-0.44)

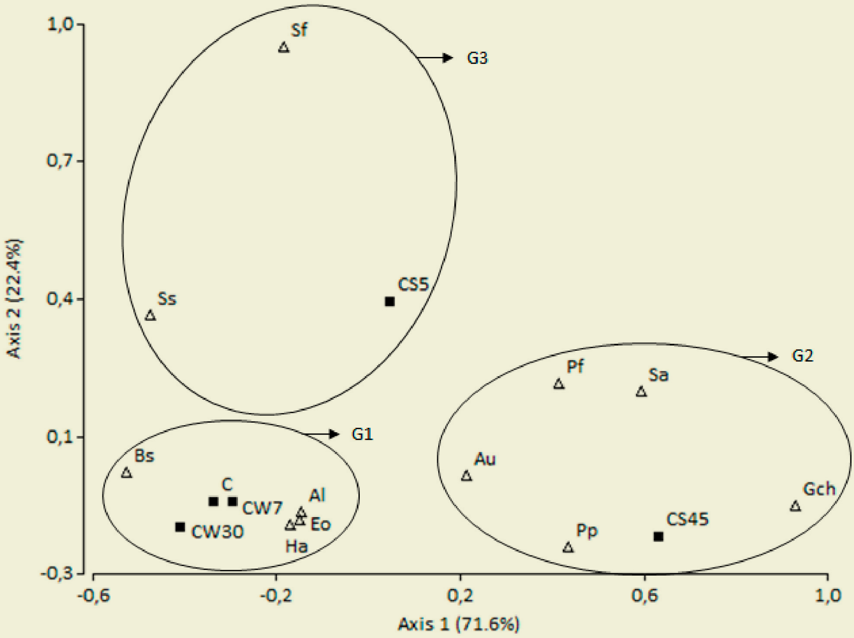

Figure 2. Subgroups of species according to dormancy alleviation treatment (Correspondence analysis). G1: subgroup 1; G2: subgroup 2; G3: Subgroup 3. White triangles represent the species (Al: *Atriplex lampa;* Au: *Atriplex undulata;* Bs: *Bougainvillea spinosa;* Eo: *Ephedra ochreata;* Gch: *Grindelia chiloensis;* Ha: *Hyalis argentea;* Pf: *Prosopis flexuosa;* Pp: *Parkinsonia praecox;* Sf: *Senecio filaginoides;* Sa: *Senna aphylla*); black squares represent treatments (C: control, CS5, CS45, CW7 and CW30). Results for Pp and Sa correspond to data from a previous publication (Paredes et al. 2018)

Figura 2. Subgrupos de especies según respuesta al tratamiento pregerminativo (análisis de correspondencia). G1: subgrupo 1; G2: subgrupo 2; G3: Subgrupo 3. Los triángulos blancos representan la especie (Al: Atriplex lampa*; Au:* Atriplex undulata*; Bs:* Bougainvillea spinosa*; Eo:* Ephedra ochreata*; Gch:* Grindelia chiloensis*; Ha:* Hyalis argentea*; Pf:* Prosopis flexuosa*; Pp:* Parkinsonia praecox*; Sf:* Senecio filaginoides; *Sa:* Senna aphylla*); los cuadrados negros representan tratamientos (C: control, CS5, CS45, CW7 y CW30). Los resultados de Pp y Sa corresponden a datos de una publicación anterior (Paredes et al.2018)*

93.045 [12.420, 697.080], Wald = 19.465, P = 0.002). GP was also higher in CS45 for *A. undulata* (0.60 0.50-0.70, Exp(B) = 2.471 [1.357, 4.500], Wald = 8.743, P = 0.003) than in controls (0.39, 0.28-0.48), but decreased significantly in CW30 (0.07, 0.02-0.13, Exp(B) = 0.097 [0.0036, 0.263], Wald = 21.043, P < 0.005).

Subgroup G3

GP for untreated *S. filaginoides* seeds was very low (0.06, 0.01–0.11), but increased almost ten-fold in CS5 (0.52, 0.42–0.62, Exp(B) = 23.500 [7.945, 69.511], Wald = 32.554, P < 0.005). Untreated *S. subulatus* seeds germinated relatively well (0.54, 0.044-0.64) but GP was signifi-

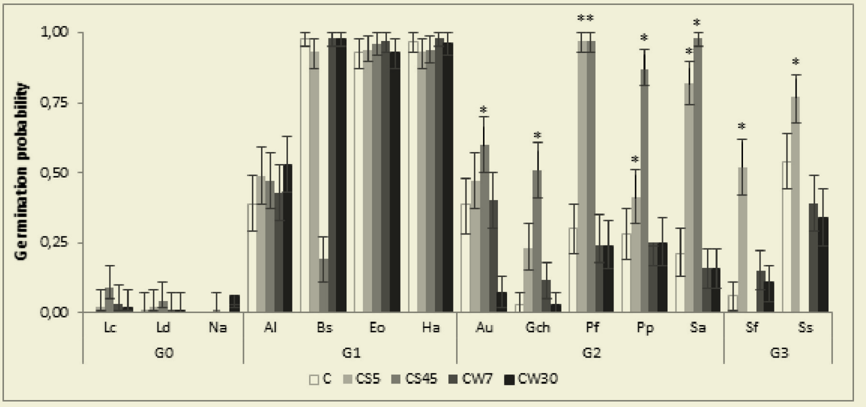

Figure 3. Germination probabilities and confidence intervals according to species and treatments (Species abbreviations Lc: *Larrea cuneifolia*, Ld: *Larrea divaricata*, Na: *Neosparton aphyllum*, Al: *Atriplex lampa*, Bs: *Bougainvillea spinosa*, Eo: *Ephedra ochreata*, Ha: *Hyalis argentea*, Au: *Atriplex undulata*, Gch: *Grindelia chiloensis*, Pf: *Prosopis flexuosa*, Pp: *Parkinsonia praecox*, Sa: *Senna aphylla*, Sf: *Senecio filaginoides*, Ss: *Senecio subulatus*. Treatments abbreviations C: control, CS5 and CS45: chemical scarification during 5 and 45 minutes, CW7 and CW30: cold-wet treatments during 7 and 30 days). Species were separated in groups (G0, G1, G2, G3) according to their response to the treatments. * Indicates GP statistically higher than controls (P<0.05). Data for Pp and Sa are drawn from a previous publication (Paredes et al. 2018)
Figura 3. Probabilidades de germinación e intervalos de confianza según especies y tratamientos (Abreviaturas de especies Lc: Larrea cuneifolia*, Ld:* Larrea divaricata*, Na:* Neosparton aphyllum*, Al:* Atriplex lampa*, Bs:* Bougainvillea spinosa*, Eo:* Ephedra ochreata*, Ha:* Hyalis argentea*, Au:* Atriplex undulata*, Gch:* Grindelia chiloensis*, Pf:* Prosopis flexuosa*, Pp:* Parkinsonia praecox*, Sa:* Senna aphylla*, Sf:* Senecio filaginoides*, Ss:* Senecio subulatus*. Abreviaturas de los tratamientos C: control, CS5 y CS45: escarificación química durante 5 y 45 minutos, CW7 y CW30: tratamientos frío-húmedo durante 7 y 30 días). Las especies se separaron en grupos (G0, G1, G2, G3) según su respuesta a los tratamientos. * Indica GP estadísticamente más alto que los controles (P <0.05). Los datos de Pp y Sa provienen de una publicación anterior (Paredes et al.2018)*

cantly improved in CS5 (0.77, 0.68–0.85, Exp(B) = 2.929 [1.533, 5.595], Wald = 10.584, P = 0.001). GP in *S. subulatus* was markedly decreased in CW7 (0.39, 0.29-0.49, Exp(B) = 0.532 [0.294, 0.963], Wald = 4.338, P = 0.037) and CW30 (0.34 0.24-0.44), Exp(B) = 0.418 [0.229, 0.765], Wald = 8.010, P = 0.005). After 45-minute chemical scarification (CS45) all seeds of all G3 species exhibited embryo damage and, consequently, no germination was recorded (**Table 2**).

Species performances in ERR

Although field survival and growth data were obtained from the literature, most of them were limited to particular soil properties (see Discussion). We found precise information from field studies on survival for 9 of 16 species, all of them with high survival rates (>75%) except for *S. subulatus* whose best performance was associated with non-alkaline soils. As regards to growth or plant cover only

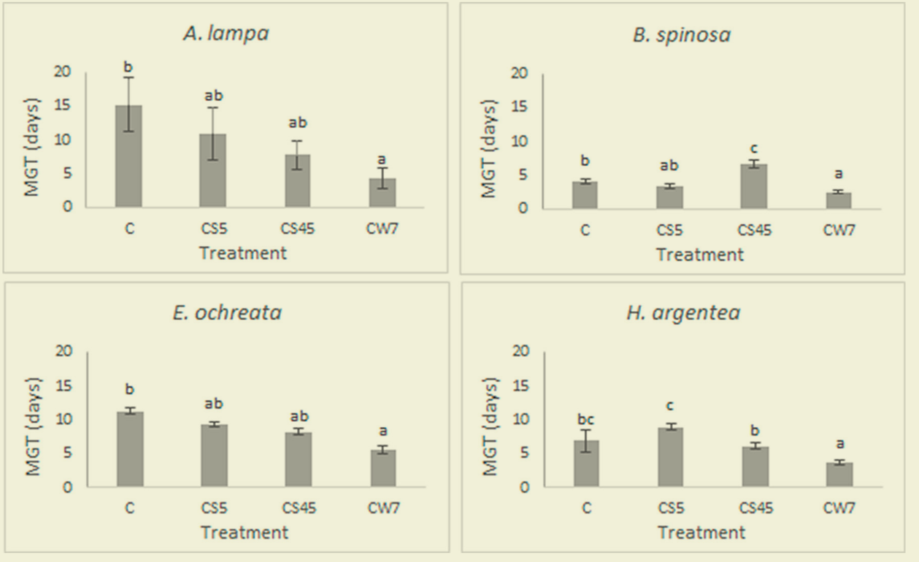

Figure 4. Mean germination time (MGT) expressed in days for subgroup 1G species according to different dormancy alleviation treatments (C: control, CS5 and CS45: chemical scarification during 5 and 45 minutes, CW7 and CW30: cold-wet treatments during 7 and 30 days). Values represent the mean and the standard deviation of 3 replicates per treatment. Means marked with the same letter are not statistically different (p> 0.05)

Figura 4. Tiempo medio de germinación (MGT) expresado en días para las especies del subgrupo G1 según diferentes tratamientos de pregerminativos (C: control, CS5 y CS45: escarificación química durante 5 y 45 minutos, CW7 y CW30: tratamientos frío-húmedo durante 7 y 30 días). Los valores representan la media y el desvío estándar de 3 repeticiones por tratamiento. Las medias marcadas con la misma letra no son estadísticamente diferentes (p> 0.05)

five species have been evaluated, four attained relatively high plant cover compared to other species planted simultaneously (**Table 3**).

Discussion

With respect to seed collection, great variability in fruit production has been observed in tree and shrub species native to arid and semiarid lands. In the Monte region, Dalmasso & Anconetani (1993) reported seed production of *P. flexuosa* varying from 80,000-800,000 seeds·ha^{-1} in different years. Resource limitation, the time of rainfall, frost and wind occurrence, and soil water content have all been suggested as proximate causes of differing seed abortion rates (Villagra, 2000). Possibly, some of these reasons are the causes that determined that we could not gather enough seeds to carry out more detailed experiments for 11 of the 27 candidate species.

The "G0" subgroup

Five out of the 16 species tested showed negligible response to the dormancy alleviation treatments applied. Recent stud-

Table 3. Best dormancy alleviation treatment for studied species and availability (Y: available, -: not available) of field performance information in ecological restoration or rehabilitation. C: control, CS5: 5-minute chemical scarification, CS45: 45-minute chemical scarification, CW7: 7-day cold-wet treatment, CW30: 30-day cold-wet treatments, MS: mechanical scarification. [1] Fernández et al. 2019. [2] Hernández et al. 2020. * Non applicable for large-scale ecological restoration or rehabilitation

*Tabla 3. Mejor tratamiento pregerminativo para las especies estudiadas y disponibilidad (Y: disponible, -: no disponible) de información de desempeño en restauración o rehabilitación ecológica a campo. C: control, CS5: escarificación química durante 5 minutos, CS45: escarificación química durante 45 minutos, CW7: Tratamiento frío-húmedo durante 7 días, CW30: tratamientos frío-húmedo durante 30 días, MS: escarificación mecánica. [1] Fernández et al. 2019. [2] Hernández et al. 2020. * No aplicable para restauración o rehabilitación ecológica a gran escala*

Species	Family	Best dormancy alleviation treatment	Survival studies	Plant cover studies
Grindelia chiloensis (Cornel.) Cabrera	Asteraceae	CS45	Y	Y
Hyalis argentea D. Don ex Hook. & Arn.	Asteraceae	C - CW7	Y	Y
*Senecio filaginoide*s DC.	Asteraceae	CS5	-	-
Senecio subulatus Don. ex Hook. et Arn.	Asteraceae	CS5	Y	-
Atriplex lampa (Moq.) D. Dietr.	Chenopodiaceae	C - CW7	Y	Y
Atriplex undulata (Moq.) D. Dietr.	Chenopodiaceae	CS5	-	-
Ephedra ochreata Miers	Ephedraceae	CW7	-	-
Parkinsonia praecox (Ruiz &Pav. ex Hook.) Hawkins	Fabaceae	CS45	Y	Y
Prosopis flexuosa var. *depressa* DC	Fabaceae	CS5	Y	Y
Senna aphylla (Cav.) H.S. Irwin & Barneby	Fabaceae	CS45	Y	-
Bougainvillea spinosa (Cav.) Heimerl	Nyctaginaceae	C - CW7	Y	-
Monttea aphylla (Miers) Grisebach	Scrophulariacea	-	-	-
*Neosparton aphyllu*m (Gill. et Hook) Kuntze	Verbeneceae	-	-	-
Larrea cuneifolia Cav.	Zygophyllaceae	MS[1*]	-	-
Larrea divaricata Cav.	Zygophyllaceae	MS[2]	Y	-
Larrea nitida Cav.	Zygophyllaceae	MS[1*]	-	-

ies have indicated that for *Larrea* species, mechanical scarification can be effective (Fernandez et al., 2019), and a novel low-cost scarification technique with a drill and sandpaper shows promise for application for large volumes of seed such as will be needed for large-scale ERR (Hernandez et al., 2020). There are no previously reported studies on germination for *N. aphyllum* or *M. aphylla*. In relation to outplanting performance, only *L. divaricata* has been studied, with reports of very strong survival results of ca. 79% (Pérez et al., 2019b). Thus, this is the only species in this group that we can safely called suitable and promising for large-scale ERR restorative practices.

The "G1" subgroup

In this subgroup, medium germination rates were obtained (ca. 50%) for *A. lampa* in all treatments applied. Previous researchers have reported higher germination rates for this species (ca. 80%) after manually removing the bracteoles (Bonvissuto & Busso, 2007). However, this procedure would be prohibitively expensive for the large volume of seeds required in large-scale ERR. The other three species in this subgroup (*B. spinosa*, *E. ochreata*, and *H. argentea*) showed very high germination rates (>95%) without treatments of any kind. High rates of germination from seeds in this manner may favor natural regeneration where reproductively-mature individuals of these species could be established to provide a source of seeds in the restoring site.

All the species in this subgroup germinated within 30 days of the cold-wet treatment, with the shortest mean germination time (MGT) in the CW7 treatment (**Figure 4**). This supports a seed recruitment niche in these species matching the thermal and moisture conditions observed during the autumn period where natural seed germination has been observed. With respect to species performance in the field, *A. lampa* and *H. argentea* showed very high survival rates (84–91%), and relatively good plant cover compared to other species planted at the same time (Pérez et al., 2019a; 2020). Outplanting of *B. spinosa* in compacted soils yielded high survival rates (85%; Pérez et al., 2020), although this species generally grows quite vertically with few ramifications and bearing only very small leaves, and thus covers very little ground even when mature (Pérez et al., 2020).

The "G2" subgroup

In this group all the species, even from different families, such as Chenopodiaceae, Fabaceae, and Asteraceae, showed germinations rates over 50% with a single treatment (CS). Previous studies reported higher germination for *A. undulata* following manual removal of the bracteoles (Piovan et al., 2014). However, this labour-intensive approach is likely impractical for large-scale ERR. Acid scarification of seeds in the manner we tested yielded acceptable seed germination results, and partially resolves issue of scale in application to large quantities of seeds.

Regarding species suitability for ERR, *G. chiloensis* and *P. flexuosa* both exhibited high survival rates and relatively high growth rates compared to other species outplanted concurrently (Becker et al., 2013; Pérez et al., 2019b). However, *P. praecox* and *S. aphylla* had notably higher survival rates after being outplanted, although *P. praecox* showed relatively little plant cover in very compacted soils after five years (Pérez et al., 2020). No information could be found on field survival or plant cover for *A. undulata* and *S. aphylla*, and we suggest further study is required for these taxa.

The "G3" subgroup

CS5 yielded the highest germination rate for two congeneric species in this group (50% and 77% for *Senecio filaginoides* and *S. subulatus*, respectively), and this treatment should be preferred over the standard procedure of nicking seeds (i.e. cutting the testa with a scalpel; Kildesheva et al., 2020), which achieves similar results but it is very time-consuming (Masini et al., 2016). *Senecio subulatus* plantations have been recommended for non-alkaline soils (Pérez et al., 2019a) and new studies are required to assess survival and growth attributes for *S. filaginoides* in order to assess their suitability for large-scale plantations and holistic restorative programs.

Final remarks

In many arid and semiarid regions, especially those in developing countries, embarking on the long process of ERR can be extremely complex and costly, especially in the face of extreme and unrelenting desertification processes. Fortunately, more and more easily accessible protocols and recommendations are being developed to address and solve technical and strategic difficulties faced by ERR practitioners. Furthermore, the positive benefit—cost ratios of such investments, and the worldwide trend to step up national commitments to ERR—are changing the picture. But, for now, the problems of getting restoration processes in gear remain daunting. In this case study, we employed a tool that we consider very useful for dryland restoration practitioners, namely *professional intelligent tinkering* (Murcia & Aronson, 2014). Simple trial and error experiments on a small scale are the hallmark of this approach. When good results are obtained, then more detailed scientific experiments can be undertaken as a follow up in view to solid validation.

In this manner ERR activities can progress concurrently with on-site research undertaken to fill knowledge gaps in best practice, and/or with current practices modified and improved through input of knowledge and know-how from external sources. We applied this conceptual framework to evaluate germination, using hypotheses about seed dormancy type and dormancy alleviation methodologies for groups of species rather than empirically evaluating each and every candidate species. This approach allowed us to achieve acceptable germination success (50–100%) in 11 out of 16 studied species using only two simple dormancy alleviation treatments. These results, coupled with field performance data and recently-published information on seed germination for three additional species, allowed us to propose eight of the 16 studied species as DFS candidates for our region based on their seed germination traits, high survival rates when outplanted, and potential to rapidly achieve high rates of plant cover.

In conclusion, our study offers one example of how some typical knowledge gaps constraining ERR can be sidestepped to allow restorative activities to progress even when complete information on best-practice approaches is lacking.

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