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Environmental drivers of seed persistence and seedling trait variation in two *Neltuma* species (Fabaceae)

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Abstract

The endemic tree *Neltuma caldenia* Burk. and the shrub *Neltuma flexuosa* var *depressa* F.A. Roig (Fabaceae; subfam: Mimosoideae) are two promising species from the central region of Argentina, with high potential for use in the restoration of disturbed environments, for extensive livestock grazing and apiculture. Both species have seeds with physical dormancy. Ecological study of native species is important from the point of view of rehabilitation of degraded areas by natural regeneration or via seed-based programmes. The objective of this study was to evaluate soil seed bank persistence and seedling traits to understand variation among different populations of each of the study species growing along an ecological gradient and to identify potential components driving this variation. Home environments influenced seed bank persistence, which was higher in populations originating from more arid and unpredictable environments where it could act as a bet-hedging strategy between years and seasons. We also observed differences associated with seedling growth traits between the species and populations. Populations with higher seed persistence were associated with greater seedling growth in *N. caldenia*. The rapid elongation rate of *N. caldenia* seedlings growing in large populations and unstable environments could help their ability to escape drought by accessing deeper soil moisture and would confer a high relative competitive ability. In contrast, *N. f.* var *depressa* displayed higher seed persistence, which was associated with more arid and unstable conditions and correlated with lower seedling growth, possibly due to an inbreeding depression effect, resulting from the presence of mother plants in low-densities or being isolated from other individuals. There was no effect of population size on seed persistence. To better understand seed persistence and associated seedling trait variation, future studies need to consider the genotype, environment and landscape conditions.

Introduction

The central Argentina ecotone, characterized by an arid-to-subhumid temperate transition climate, encompassing more than 10 M hectares, has been seriously degraded by deforestation, overgrazing and some unsustainable agriculture (González and Pérez, 2017; Zeberio and Pérez, 2020). These high levels of disturbance removed vegetation and exposed areas to erosion, severe processes of desertification and loss of agricultural productivity in arid and semiarid regions (Gabella and Strijker, 2018; Winschel and Pezzola, 2018). In semiarid-to-subhumid environments, continuous grazing contributes to an increased abundance of woody species that may interfere with livestock management and reduce the production of desirable grasses (Villalobos and Peláez, 2001).

Plant species are locally adapted to the environmental conditions in which they occur (Lascoux et al., 2016). *Neltuma* (ex *Prosopis*) *caldenia* Burk. and *Neltuma flexuosa* var *depressa* F.A. Roig (Fabaceae; subfam: Mimosoideae) are two species that occur within the central ecotone region. They are valued as a forage source for ruminants and bees and are suitable candidates for the restoration of degraded areas (Alvarez and Villagra, 2009; Villagra et al., 2010; Farinaccio et al., 2021; Hughes et al., 2022). These species could play an important role in restoration and erosion control, as well as in increasing soil fertility due to their capacity to fix atmospheric nitrogen (Cony, 1996). The tree *N. caldenia* is distributed between 31 and 41°S latitude in a temperate semiarid environment and can form forests where they are dominant or form communities with greater diversity, consisting of other species including *N. flexuosa* DC, *Geoffroea decorticans* Gill. ex H. et A. Burkart and *Jodina rhombifolia* (Hook. & Arn.) Reissek. The shrub- (*N. f.* var *depressa*) occurs predominantly between latitudes 30 and 43°S and has been found to show tolerance to drought, cold, salinity and alkalinity in a temperate arid environment (Roig, 1993; Alvarez and Villagra, 2009; Villagra et al., 2010). Both species are predominantly cross-pollinated and are known to have physical dormancy, caused by water-impermeable palisade cells in the seed coat, a trait which could favour its dispersal via

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endozoochory (Peinetti et al., 1993; Villagra et al., 2002; Campos et al., 2011; Aguilar et al., 2019). From an economic point of view, *Neltuma* species represent the most important plant community in Central Argentina. Unlike other woody legumes, the foliage and pods (~18% of protein) are a good source of forage for domestic herbivores (Somlo et al., 1997). Good adaptation to the extreme environmental conditions, and their ecological and economic importance, give them high potential as candidates for rehabilitating degraded areas.

The study of local adaptation mediated by genotype and environment is relevant to understanding responses to climate change, and for informing conservation and restoration programmes. Seed persistence is one of the key stages in the life history of plants and can be associated with the dormancy strategy. Variation in seed persistence may support bet-hedging strategies, facilitating drought avoidance at xeric sites or those with variable precipitation and temperature regimes (Christie et al., 2022). A bet-hedging strategy acts to reduce establishment risk for the seed produced by an individual plant by spreading germination over the growing season or over several years. This reduces short-term reproductive success in favour of longer-term risk reduction. In contrast, short-term seed persistence occurs in species that may depend on other life-history stages for persistence (Saatkamp et al., 2014) and can make a species more vulnerable for *in situ* conservation in a climate change context (Ooi, 2012; Chen et al., 2023). Species with a long plant lifespan, or high seed fecundity and dispersal, rely comparatively less on persistent soil seed banks (Chen et al., 2020). Arid-semiarid environments seem to evolutionarily select for higher seed persistence in the soil seed bank, and high variability in germination, growth and reproduction as a bet-hedging strategy (Rubio de Casas et al., 2017; Klein and Mitchell, 2024). In *Neltuma* species, there are few studies of seed persistence in the soil bank and seedling traits associated with the environment of origin. In general, past studies have been oriented towards the breaking of physical seed dormancy and germination under controlled laboratory conditions (Villagra, 1997; Vilela and Ravetta, 2001; Villalobos and Peláez, 2001; Galindez et al., 2016; Boeri et al., 2019).

The ecological study of native species is crucial for their informed use in the ecological rehabilitation of degraded areas and the management of grasslands to prevent bush encroachment. The transition from seed to seedling is the most critical stage for survival in a *Neltuma* community (Villalobos and Peláez, 2001). Thus, our objectives were to (i) study the intraspecific variations in persistence in the soil seed bank and early seedling traits in different populations in *N. caldenia* and *N. f. var. depressa* collected in the central region of Argentina; and (ii) assess the relationships between the observed variations and environmental conditions and population size of origin as potential drivers of adaptation.

Materials and methods

Seed collections

The study area comprised the central region of Argentina, including the ecoregions of the central Monte Desert, south Espinal and northern Patagonian steppe (Fig. 1). Ripe pods were collected in bulk (>80 pods) from at least 8 specimens from 8 *N. caldenia* and 12 *N. flexuosa* var. *depressa* native populations separated from each other by ~40 m, during 1 month in the summer of 2020 (Supplementary Table S1). Populations were separated by

>30 km, to ensure they represented independent genetic structure (Bessega et al., 2017). At this period of the season, *N. f. var. depressa* and *N. caldenia* individuals have reached fruit maturity and show active vegetative growth (Roig, 1993; Steibel and Troiani, 1999; Hughes et al., 2022).

The global positioning system (GPS) coordinates were recorded for each population detected (Supplementary Table S1). The descriptive parameters of the populations were (i) latitude and longitude, (ii) altitude, (iii) environmental factors (soil and climate) and (iv) population size, visually by observation of the distribution of the specimens in the field as 'isolated', 'small' or 'large' population size (>~100 specimens per ha) (Leimu and Fischer, 2008). In addition, landscape conditions were assessed through the vegetation cover using satellite image analysis. Forest/shrub cover (0-to-100%) was estimated on cloud-free days, in visually homogeneous zones, with EOS Land Viewer services (<https://eos.com/landviewer>) during the sampling growing period (January-to-February). The images were stored in optical natural colour, and the visual vegetation cover (percent of bare soil) was estimated.

For environmental classification, the WorldClim (<http://worldclim.org>) version 2.0 database was used. Data were extracted by means of DIVA-GIS software from ESRI grids, with a spatial resolution of 30 arc-seconds (~1 km) in the WGS-84 (EPSG: 4326). Nineteen bioclimatic variables were derived from monthly temperature and rainfall values (Fick and Hijmans, 2017). To avoid over-parameterization among the bioclimatic variables, we used Pearson correlation coefficients to measure pairwise correlations between the variables (R, Version 3.5.1), and one of the two paired variables correlated above 0.8 was eliminated. Nine bioclimatic, non-redundant variables used were: annual mean temperature (BIO1), mean diurnal range (mean of monthly (max temp - min temp)) (BIO2), isothermality (BIO3), mean temperature of warmest quarter (BIO10), mean temperature of coldest quarter (BIO11), annual precipitation (BIO12), precipitation seasonality (BIO15), precipitation of warmest quarter (BIO18) and precipitation of coldest quarter (BIO19) (Hijmans et al., 2005). The soil parameters of the populations, such as soil texture, pH and bulk density, were obtained from soil databases (FAO/IIASA/ISSCAS/JRC, 2012) using WGS84 and spatial resolution of 30 arc-seconds. For the details, including basic descriptive statistics of each *Neltuma* population, see Supplementary Table S1.

Seed persistence

After harvest, the pods were incubated under laboratory conditions for 50 days (~22°C) to allow the development of insects throughout the life cycle (from egg to adult) that were potentially infecting the seeds at sampling time (Velez et al., 2018), mainly bruchids (Coleoptera) (Alvarez and Villagra, 2009). Populations were formed from seeds, removed and cleaned by hand from different intact pods. Seed weight was estimated for a sample of 25 seeds ($n = 3$) for each population (Supplementary Table S2).

The soil seed bank persistence for 3 years was evaluated in all the populations. The seeds used were those without any sign of predation (no exit holes, or depressions) and regular in colour and shape (Velez et al., 2018). To determine initial viability, 30 previously scarified seeds per population were incubated in a germination chamber for 7 days (20°C ± 2; 8 h light/16 h dark) and were considered viable with a visible protrusion of the radicle from the seed coat (≥1 mm). Batches of random samples of 25

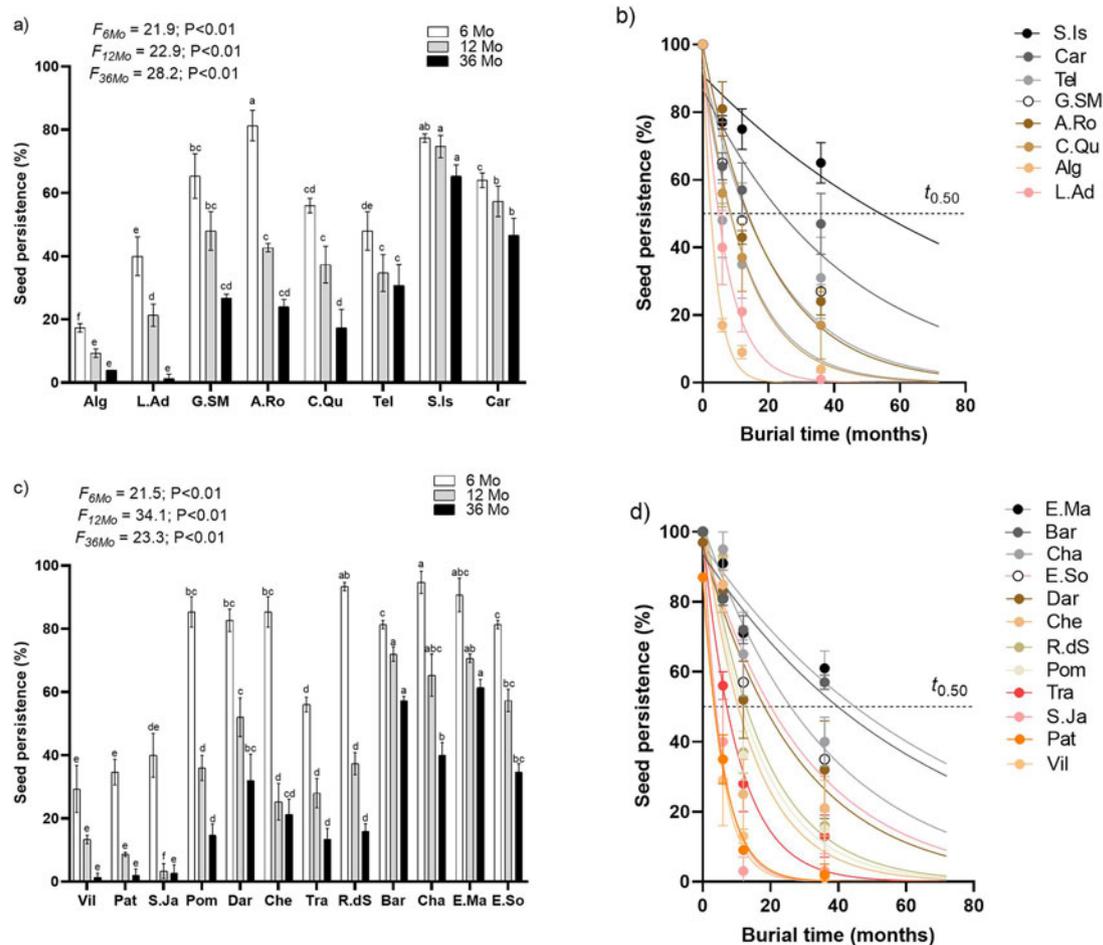


Figure 1. Seed persistence in the soil seed bank during 6, 12 and 36 months (Mo), and asymptotic exponential nonlinear regression in *Neltuma caldenia* Burk. (A, B), and *N. flexuosa* var. *depressa* F.A. Roig populations (D, E). Different letters indicate significant differences between populations for each burial time (Fisher's least significant difference, LSD, $P < 0.05$).

seeds ($n = 3$ for each population and exhumation time) were prepared inside permeable nylon mesh bags (10×10 cm), in order to simulate natural soil conditions of air and water diffusion and micro-organism activity. The batches were buried in mid-autumn (23 April 2020) at a depth of 5 cm in sandy loam soil (pH = 7.5, soil organic matter = 1.2%) without any vegetation or leaf litter. The burial site was in the experimental field of INTA EEA Ascasubi (Buenos Aires, Argentina; $39^{\circ}22'S$, $62^{\circ}39'W$). Weather data were recorded at the nearby meteorological station (less than 200 m away) (Supplementary Fig. S1). Seeds were exhumed after 6, 12 and 36 months of burial.

The proportion of PY seeds was assessed. PY seeds (i.e. 'hard' or impermeable) were determined by an imbibition test performed at $20 \pm 2^{\circ}C$ for 2 days (Baskin and Baskin, 2014), after which swollen and germinating seeds were recorded and removed. After the exhumation, seed viability was assessed for the remaining non-germinated seeds by scarifying the opposite end of the embryo and placing them in germination chambers for 4 days ($20^{\circ}C \pm 2$; 8 h' light/16 h' dark). The total number of viable seeds consisted of germinated seeds.

Seedling traits evaluation

Seedling elongation tests in the laboratory were conducted in Petri dishes with two layers of Whatman No. 1 filter paper moistened

with 10 ml of water. Seeds were previously scarified to avoid a lack of germination imposed by physical dormancy. The seed populations were incubated in a germination chamber ($20^{\circ}C \pm 2$; 8 h light/16 h dark). To obtain the necessary seedlings, 30 seeds per dish were incubated. Once seedlings with a radicle length of 1 mm were obtained, six representative seedlings were selected per dish and transferred to new Petri dishes ($n = 6$). The length of seedlings (root + shoot) was measured using a digital calliper following a non-destructive sampling method. The length of each seedling was measured every 24 h for 7 days. The mean seedling elongation rate ($mm\ day^{-1}$) was estimated.

Seedling phenotypic traits were evaluated in each population collected ($n = 20$). The randomly chosen seeds were individually sown in plastic pots (6.5×8.0 cm) filled with 35 g of a sterilized substrate mix (Growmix Multipro, Terrafertil). All seedlings were watered three to four times per week until the time of measurement. Forty days after planting, the leaf number per plant, seedling height (cm), aerial (shoot), subterranean (root) and total dry biomass of each seedling were evaluated ($n = 10$). Then a dry-down experiment to assess seedling survival was conducted in the rest of the replicas ($n = 10$), imposing a severe drought by completely halting daily watering. No additional watering was added during the survival experiment. Seedlings were monitored on a daily basis. Individuals were scored according to the criteria of Engelbrecht and Kursar (2003) and considered dead when all

above-ground parts lost colour and showed no elasticity. To verify seedling death, we subsequently watered each pot to saturation and monitored it for revival (Larson et al., 2020). Thus, seedling survival was measured daily as a function of days without irrigation. The experiments were conducted in a greenhouse, under a completely randomized design. The temperature was controlled at 20/25°C, photoperiod was a 14-h light:10-h dark cycle.

Statistical analysis

For each population, the percentages of PY (i.e. PY seeds = 100 – (germinated (%) + imbibed seeds (%)) at 6, 12, and 36 months were analysed by Generalized Linear Model (GLM) performed with R version 3.6.1 in Infostat. Population means were compared by Fisher's least significant difference test for seed persistence (Di Rienzo et al., 2013). For analyses, the PY data were arcsine -square root transformed, and the untransformed data are presented in the Figures for clarity.

Seed persistence in soil, taking the percentage of initial viability as a starting point, was plotted as a function of burial time (months) with nonlinear models, relating PY seeds remaining over time to an exponential model. The decay function was expressed as:

$$Y = Y_0 * \exp^{(k*x)}$$

where Y = persistent seeds (%), Y_0 = Y value when X (time) is zero, k = rate constant, and X = burial time in months. We estimated the half-lives of seed persistence in the soil seedbank ($t_{0.5}$) for each population. The exponential growth equation was calculated using GraphPad Prism Software version 8.0 (GraphPad).

Correlations between seed persistence at 6, 12, 36 months, and half-lives for seed persistence, and seedling traits and environment conditions of origin were calculated using Pearson's correlation coefficient, with $P < 0.05$ set as the significance level. In addition, a principal component analysis (PCA) was performed with seedling traits and selected bioclimatic variables (BIO1, 2, 3, 10, 11, 12, 15, 18 and 19) in relation to seed persistence using InfoStat software.

Linear regression analysis between vegetation cover and seed persistence ($t_{0.5}$) was performed by species using GraphPad Prism Software version 8.0 (GraphPad).

Results

Seed persistence

Initial seed viability was over 87% in all cases (Supplementary Table S2). The seed persistence in the soil ranged between 0-to-72% in *N. caldenia* and 0-to-64% in *N. f. var depressa* at the third year of burial, with differences found between populations in both species (Fig. 1). The persistence in the soil seedbank obtained at 6 and 12 months correlated with those of the third year in both species ($r > 0.71^{**}$), and with the seed half-life in the soil ($t_{0.5}$) ($r > 0.91^{**}$) (Supplementary Tables S3 and S4).

In both species, we observed a geographical gradient in seed persistence associated with latitude, longitude, and altitude (Fig. 2). Seed persistence increased in the east-to-west (latitude; $r = 0.75^*$, and $r = 0.79^{**}$) and south-to-north (longitude; $r = 0.83^{**}$ and $r = 0.79^{**}$) directions, correlated with higher altitude ($r = 0.72^*$ and $r = 0.80^{**}$) within the range of 11 to 1512 m above sea level.

Seed persistence and seedling traits

In *N. caldenia*, seed persistence was positively associated with seedling elongation rate ($r = 0.73^*$). Higher initial seedling growth was correlated with seedling height ($r = 0.77^*$). Conversely, in *N. f. var. depressa*, we observed a negative association between seed persistence and seedling height ($r = -0.64^*$). Seedling heights were directly associated with shoot, root, and total seedling weight in both species. We did not find a relationship between seed weight and seedling elongation rate for either species ($P > 0.05$) (Fig. 3).

Relationship between seed persistence and environment

Populations of *N. f. var depressa* were located in arid environments (281 ± 77 mm year⁻¹) in contrast to those of *N. caldenia* which were found in semiarid environments (445 ± 98 mm year⁻¹) (Fig. 2). The populations with the highest seed persistence originated in the environment with a highest mean diurnal range ($r = 0.84^{**}$), isothermality ($r = 0.85^{**}$) and mean temperature of the warmest quarter (summer) ($r = 0.70^*$) in *N. caldenia* (Fig. 4A). In *N. f. var depressa*, a higher seed persistence was associated with a higher mean diurnal range ($r = 0.74^{**}$), isothermality ($r = 0.89^{**}$), precipitation seasonality ($r = 0.76^{**}$), precipitation of the coldest quarter ($r = 0.67^*$), lower annual mean temperature ($r = -0.64^*$) and mean temperature of the coldest quarter ($r = -0.71^*$) (Fig. 4B, and Supplementary Table S3).

We observed that the plants of *N. f. var depressa* populations located in the northwest (Bar, Cha, E.Ma, and E.So) showed an isolated distribution, associated with a lower vegetation cover ($\leq 25\%$). Higher seed persistence was associated with a lower vegetation cover (Fig. 5).

Discussion

The climate of the sites varies from subhumid to arid conditions, the latter with only 150 mm of rainfall per year. Seed persistence in both species showed clinal trends increasing in a north-westerly (NW) direction (Fig. 2). This was associated with the natural plant community that also showed an ecological cline between grass steppe with woody species and xerophytic trees (*Larrea* spp. Cav., *Bulnesia* spp. C. Gay, *Neltuma* spp. L., *Cercidium* spp. Tul and *Geoffroea* spp. Jacq.) in the east and arid steppe in the west (Anderson et al., 2011; Torres Robles et al., 2022). The higher seed persistence for north-western populations of *N. caldenia* was found to be associated with strong seasonality with large temperature swings and hot summers, showing a more extreme and less stable environment. In *N. f. var depressa*, we observed that the populations located in the northwest showed greater seed persistence (Fig. 2), associated with cold climates with variable and extreme thermal conditions, and also a marked variability in precipitation, with higher records during the winter. This result is consistent with findings in legume species adapted to aridity, which is likely to be an advantage in unpredictable environments (Villagra et al., 2010; Rubio de Casas et al., 2017; Renzi et al., 2020). We cannot separate the genetic and environmental factors of seed persistence in both species, which involve either local genetic adaptation or phenotypic plasticity associated with maternal effects. However, seed persistence variation underscores the ecological strategies adopted for adapting to diverse environmental constraints. Maternal effects in transgenerational studies in perennial species would probably be smaller in magnitude than in annual species (Yin et al., 2019).

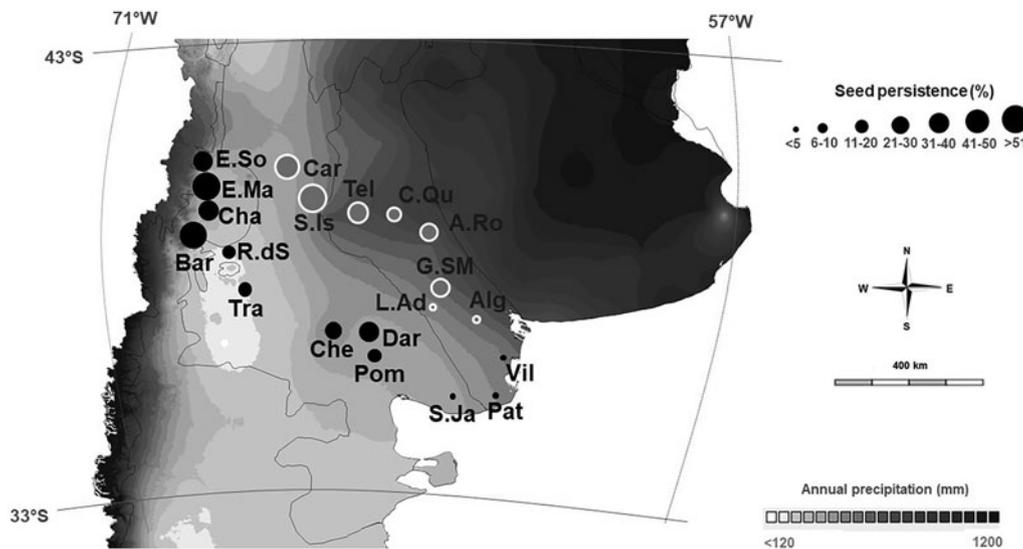


Figure 2. Geographic localization of *Neltuma caldenia* Burk. (grey) and *N. flexuosa* var. *depressa* F.A. Roig populations (black). The size of the circle in each population is related to the seed persistence in the soil bank after 3 years of burial.

The persistent soil seed bank, with more than 30% of seeds intact and viable after 3 years of burial, was observed in the 37% of *N. caldenia* and in 42% of *N. f. var depressa* populations (Fig. 1). Only one *N. caldenia* population and two *N. f. var depressa* populations showed <10% intact seeds after 1 year, forming short-term seed banks (Baskin and Baskin, 2014). Seed bank persistence in *Neltuma* taxa would be species and population-dependent (Lerner and Peinetti, 1996; Villagra et al., 2002; Renzi et al., 2023). Spontaneous regeneration from the soil seed banks will be higher in the northern populations. When the soil seed bank is exhausted, seed-based rehabilitation programmes constitute one of the more promising and economic models for dryland restoration (Golos et al., 2016; Busso and Pérez, 2018; Pérez et al., 2019, 2022). Within the framework of these rehabilitation objectives, candidate taxa should preferably be selected from species of the natural flora of the surrounding non-degraded environment, which would ensure their adaptation to the conditions in which they will be released (Cortina et al., 2004). However, it is important to consider the ecological adaptation of a native species to help define the propagule collection strategy (Woolridge et al., 2022).

We observed a divergent trade-off between soil seed bank persistence and seedling traits in *Neltuma* species, related to the environmental conditions and population size, estimated by vegetation cover. Seed persistence was not associated with seed weight ($P > 0.05$) for the range evaluated in *N. caldenia* (15–39 mg seed⁻¹) and *N. f. var depressa* (18–44 mg seed⁻¹). However, this should be corroborated in future analyses with a larger number of replicates and seeds per replicate in each population and in different years. These results are consistent with the analysis conducted by Chen et al. (2023), who studied 349 species, many of which were legumes (~14%). Although we did not find a relationship between seed weight and persistence or initial seedling growth, it is well established that seed weight plays a significant role in various ecological processes, benefiting early life stage fitness and enhancing seedling survival (Christie et al., 2022). Nonetheless, it is important to note that these relationships are highly context-dependent (Chen et al., 2023).

All the *N. caldenia* surveyed were found at sites with a large population size and high vegetation cover (Fig. 4). Higher persistence in an unpredictable environment observed in *N. caldenia* may require greater vigour at emergence to ensure reproductive success. It is during seedling establishment that most mortality occurs, and this could be an adaptive mechanism (Villalobos and Peláez, 2001; Lewandrowski et al., 2021). Rapid seedling growth may help escape drought by enabling a greater depth of soil moisture to be reached and would confer a high relative competitive ability, especially if thermal conditions for initial growth are limited. In arid environments with *Plantago patagonica*, it was also observed that higher seedling height was a significant predictor of survival under early-season drought (Christie et al., 2022). Studies under natural conditions should be performed on *Neltuma* species in the future.

The northwest populations of *N. f. var depressa*, situated at the ecological niche boundary (Renzi et al., 2023), exhibited higher seed persistence associated with lower seedling height (–18.5%; Fig. 3B). In sites with low vegetation cover (isolated habitat), seedlings are likely to face relatively little competition during establishment and will therefore require less height in the early stages of life (Christie et al., 2022). However, a more plausible explanation could be that animal-pollinated trees or shrubs in small populations or isolated conditions tend to decrease outcrossing rates, thereby increasing the inbreeding coefficients of the generated offspring (Angeloni et al., 2011). This did not occur in our study of *N. caldenia* because the populations collected were in a continuous forest in the middle of its ecological niche distribution of species (Dussart et al., 2015). Consistent with our results, Aguilar et al. (2019) observed that the progeny of fragmented and isolated populations of *N. caldenia* showed lower seedling growth compared to continuous forest areas due to inbreeding depression. The extent of progeny vigour decrease may depend on landscape fragmentation, mating system, presence of potential pollinators in outcrossing species (obligate > facultative), and the number of generations post-fragmentation (Rocha and Aguilar, 2001; Broadhurst et al., 2008; Aguilar et al., 2019; Doyle et al., 2023). The effect on population size would not be evident in seed

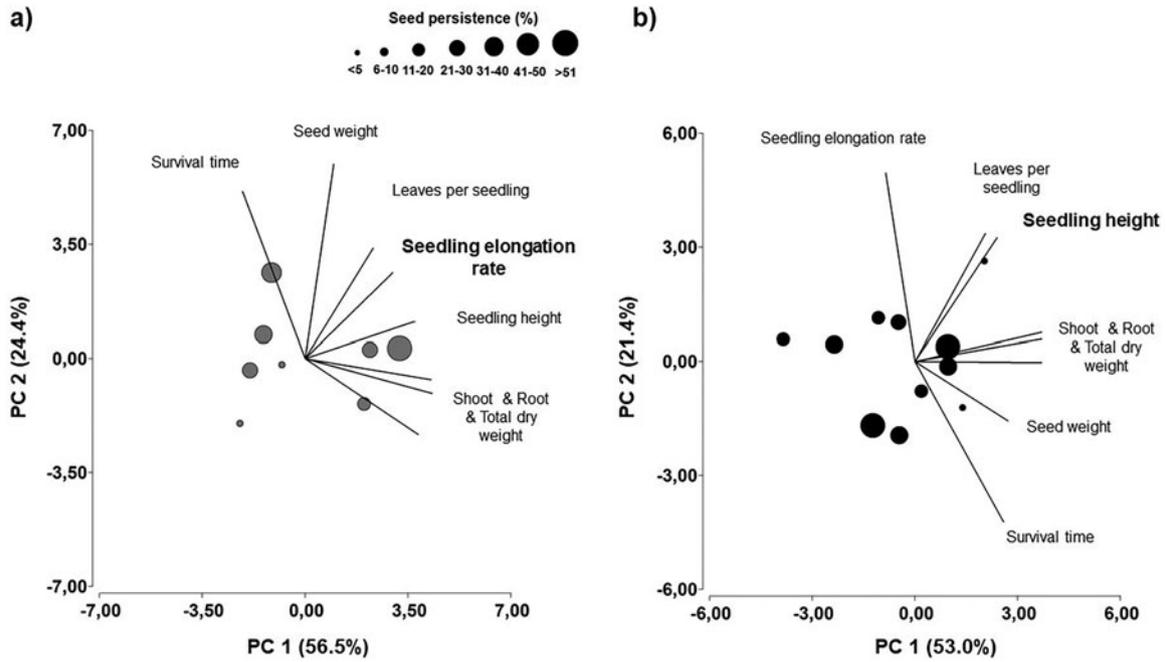


Figure 3. Biplot of *Neltuma caldenia* Burk. (A), and *N. flexuosa* var. *depressa* F.A. Roig (B) seed persistence ranges on the first two principal components (80.9 and 74.4%) based on seedling traits in early growth stage. In bold letters the trait that shows a significant correlation with seed persistence ($P < 0.05$).

germination (Baskin and Baskin, 2024) and seed dormancy, which could be attributed to maternal inheritance and environmental control (Bewley and Black, 1994; Iwasaki et al., 2022).

The rehabilitation of degraded or small populations of *Neltuma* specimens will usually require the collection of seeds from similar environmental settings (Cortina et al., 2004). The seed-collection environment has strong effects on seedling performance (Klein and Mitchell, 2024). More arid and unstable

environments will have seeds with greater persistence in the soil seed bank and with the suitable seedling traits for establishment in the face of environmental variability. In addition to the environment, population size must be considered during collection, ensuring it corresponds to a large population to avoid the effects of inbreeding depression in the progeny fitness and the possibility of maladaptation effects (Leimu and Fischer, 2008; Alves et al., 2018).

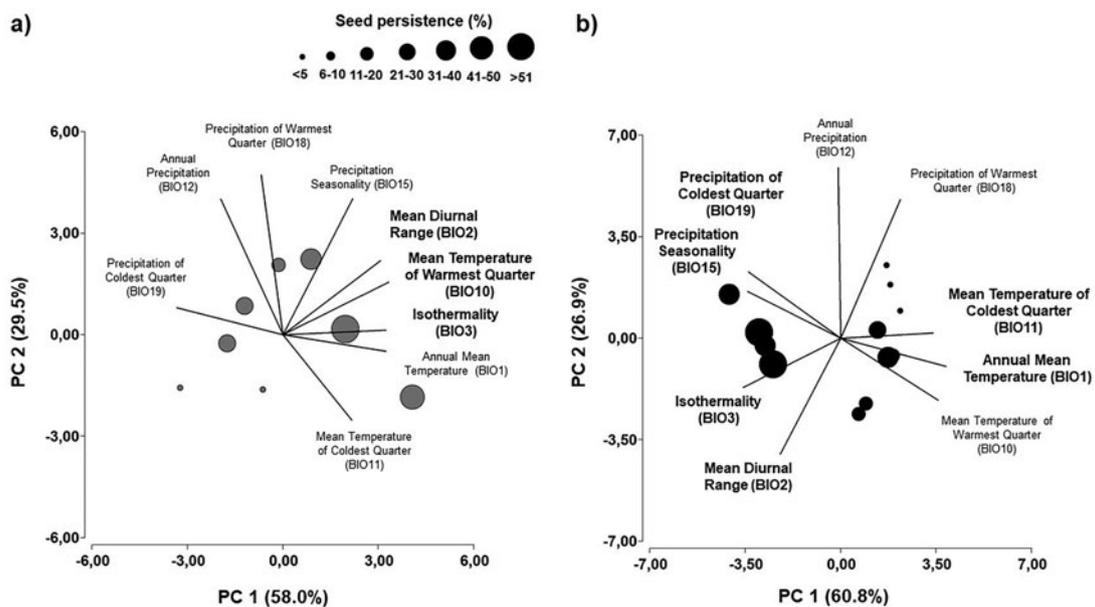


Figure 4. Biplot of *Neltuma caldenia* Burk. (A), and *N. flexuosa* DC. var. *depressa* F.A. Roig (B) seed persistence ranges on the first two principal components (80.9 and 74.4%) based on bioclimatic variables (BIOS 1, 2, 3, 10, 11, 12, 15, 18 and 19). In bold letters the BIO that shows a significant correlation with seed persistence ($P < 0.05$).

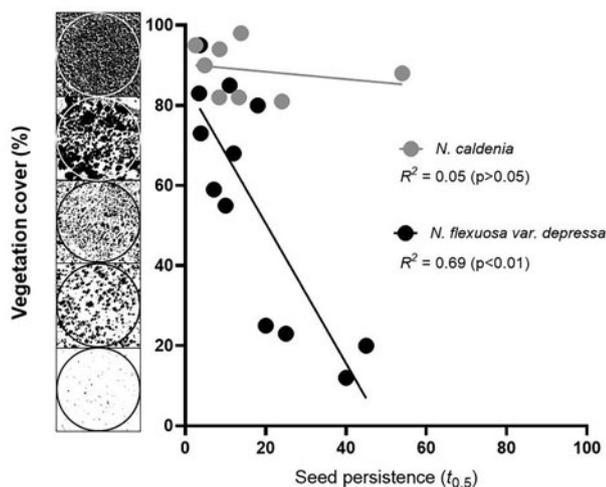


Figure 5. Relationship between half-lives for seed persistence in the soil seedbank ($t_{0.5}$) in *Neltuma caldenia* Burk. (grey), and *N. flexuosa* var. *depressa* F.A. Roig populations (black) and vegetation cover estimated in a hectare area (circle).

Conclusions

Neltuma populations in central Argentina are expected to continue to face severe precipitation and thermal variability associated with global climate change (Soliani et al., 2024). Our results show a different response depending on the species and populations associated with environmental conditions and population size. In arid and unstable environments, it is expected that the seed bank will be more persistent. Natural regeneration could form small plant patches that would improve the microenvironment and facilitate the establishment of a ring of pasture around the trees or shrubs (Busso and Pérez, 2018). However, when complete exhaustion of the soil seed bank occurs, it will require seed-based rehabilitation programmes. In this scenario, seed collection would have to be undertaken in large populations close to the rehabilitation area, to avoid collection of seeds from isolated specimens (Alves et al., 2018). Regeneration mediated by the seed bank could vary depending on the environment and seed persistence. The collection of isolated plants or small populations of *Neltuma* adapted to an arid environment and used as germplasm may have undesirable implications in the progeny performance, due to inbreeding depressions (Bessegga et al., 2017). Further studies on seed persistence and associated plant traits under natural conditions are needed with a holistic approach considering the interactions between genotype, environment, and landscape conditions. A finer description of population size and habitat heterogeneity would be desirable in subsequent studies.

Supplementary material. The supplementary material for this article can be found at <https://doi.org/10.1017/S0960258524000205>.

Data availability statement. The data that support the findings of this study are openly available in the supplementary material.

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Competing interests. None.

References

- Aguilar R, Cristóbal-Pérez J, Balvino-Olvera J, Aguilar-Aguilar MJ, Aguirre-Acosta N, Ashworth L, Lobo JA, Martín-Rodríguez S, Fuchs EJ, Sanchez-Montoya G, Bernardello G and Quesada M (2019) Habitat fragmentation reduces plant progeny quality: a global synthesis. *Ecology Letters* **22**, 1163–1173.
- Alvarez JA and Villagra PE (2009) *Prosopis flexuosa* DC. (Fabaceae, Mimosoideae). *Kurtziana* **35**, 49–63.
- Alves FM, Sartori ALB, Zucchi MI, Azevedo-Tozzi AMG, Tambarussi EV and de Souza AP (2018) A high level of outcrossing in the vulnerable species *Prosopis rubriflora* in a Chaco remnant. *Australian Journal of Botany* **66**, 360–368. doi:10.1071/BT17195
- Anderson DL, Bonvissuto GL, Brizuela MA, Chiossone G, Cibils AF, Cid MS, Feldman I, Fernández Grecco RC, Kunst C, Oesterheld M, Oliva GE, Paruelo JM, Peinetti HR and Villagra ES (2011) Perspectives on rangeland management education and research in Argentina. *Rangelands* **33**, 2–12. doi:10.2111/rangelands-d-10-00016.1
- Angeloni F, Ouborg NJ and Leimu R (2011) Meta-analysis on the association of population size and life history with inbreeding depression in plants. *Biological Conservation* **144**, 35–43.
- Baskin CC and Baskin JM (2014) *Seeds: Ecology, Biogeography, and Evolution of Dormancy and Germination*, 2nd Edn. San Diego: Academic Press.
- Baskin JM and Baskin CC (2024) Population size is not a reliable indicator of seed germination. *Seed Science Research* **33**, 213–225. doi:10.1017/S0960258523000247
- Bessegga C, Pometti C, Campos C, Saidman BO and Vilardi JC (2017) Implications of mating system and pollen dispersal indices for management and conservation of the semi-arid species *Prosopis flexuosa* (Leguminosae). *Forest Ecology and Management* **400**, 218–227.
- Bewley JD and Black M (1994) *Seeds: Physiology of Development, Germination and Dormancy*, 3rd Edn. New York: Springer.
- Boeri P, Cedrés Gazo M, Failla M, Barrio DA, Dalzotto DC and Sharry SE (2019) Optimum germinative conditions of a multipurpose shrub from Patagonia: *Prosopis alpataco* (Fabaceae). *Darwiniana* **7**, 199–207.
- Broadhurst LM, Young AG and Forrester R (2008) Genetic and demographic responses of fragmented *Acacia dealbata* (Mimosaceae) populations in south-eastern Australia. *Biological Conservation* **142**, 2843–2856. doi:10.1016/j.biocon.2008.08.021
- Busso CA and Pérez DR (2018) Opportunities, limitations and gaps in the ecological restoration of drylands in Argentina. *Annals of Arid Zone* **57**, 191–200.
- Campos CM, Campos VE, Mongeaud A, Borghi CE, De Los Rios C and Giannoni SM (2011) Relationships between *Prosopis flexuosa* (Fabaceae) and cattle in the Monte desert: seeds, seedlings and saplings on cattle-use site classes. *Revista Chilena de Historia Natural* **84**, 289–299.
- Chen S-H, Poschod P, Antonelli A, Liu U and Dickie JB (2020) Trade-off between seed dispersal in space and time. *Ecology Letters* **23**, 1635–1642.
- Chen S-H, Hu W-H, Baskin CC and Baskin JM (2023) A long-term experiment reveals no trade-off between seed persistence and seedling emergence. *New Phytologist* **241**, 521–522.
- Christie K, Pierson NR, Lowry DB and Holeski LM (2022) Local adaptation of seed and seedling traits along a natural aridity gradient may both predict and constrain adaptive responses to climate change. *American Journal of Botany* **109**, 1–16. doi:10.1002/ajb2.16070
- Cony MA (1996) Genetic variability in *Prosopis flexuosa* D.C., a native tree of the Monte phytogeographic province, Argentina. *Forest Ecology and Management* **87**, 41–49.
- Cortina J, Bellot J, Vilagrosa A, Caturra R, Maestre FT, Rubio E, de Urbina JM O and Bonet A (2004) Restauración en semiárido. In Vallejo VR and Alloza JA (eds), *Avances en el Estudio de la Gestión del Monte Mediterráneo*. Valencia: Fundación CEAM, pp. 345–406.
- Di Rienzo JA, Casanoves F, Balzarini MG, Gonzalez L, Tablada M and Robledo CW (2013) *Infostat Version 2013 grupo Infostat*. FCA. Argentina: Universidad Nacional de Córdoba.
- Doyle CAT, Yap JYS, Bragg J, Rossetto M, Orme A and Ooi MJK (2023) Reproductive characteristics, population genetics, and pairwise kinship inform strategic recovery of a plant species in a fragmented

- landscape. *Conservation Science and Practice* 5, e12910. doi:10.1111/csp2.12910
- Dussart E, Medina A and Bogino S** (2015) Dendroecología en la pampa Argentina: investigaciones actuales, pasadas y futuros desafíos. *Ecosistemas* 24, 51–59. doi:10.7818/ECOS.2015.24-2.08
- Engelbrecht BMJ and Kursar TA** (2003) Comparative drought-resistance of seedlings of 28 species of co-occurring tropical woody plants. *Oecologia* 136, 383–393.
- Farinaccio FM, Ceccon E and Pérez DR** (2021) Starting points for the restoration of desertified drylands: puesteros' cultural values in the use of native flora. *Ecosystems and People* 17, 476–490. doi:10.1080/26395916.2021.1968035
- Fick SE and Hijmans RJ** (2017) Worldclim 2: new 1 km spatial resolution climate surfaces for global land areas. *International Journal of Climatology* 37, 4302–4315. doi:10.1002/joc.5086
- Gabella JI and Strijker D** (2018) Sustainability or resilience? A case study in the semi-arid Pampean region of Argentina. *Resilience*, 7, 1–20. doi:10.1080/21693293.2018.1446298
- Gálvez G, Ceccato D, Malagrina G, Pidal B, Chilo G, Bach H, Fortunato R and Ortega-Baes P** (2016) Physical seed dormancy in native legume species of Argentina. *Boletín de la Sociedad Argentina de Botánica* 51, 73–78.
- Golos PJ, Dixon KW and Erickson TE** (2016) Plant recruitment from the soil seed bank depends on topsoil stockpile age, height, and storage history in an arid environment. *Restoration Ecology* 24, 53–61. doi:10.1111/rec.12389/supinfo
- González FM and Pérez DR** (2017) Contributions of ecological facilitation for restoring environments with high conservation value in the Argentine Patagonia. *Phyton* 86, 332–339.
- Hijmans RJ, Cameron SE, Parra JL, Jones PG and Jarvis A** (2005) Very high resolution interpolated climate surfaces for global land areas. *International Journal of Climatology* 25, 1965–1978. doi:10.1002/joc.1276
- Hughes CE, Ringelberg JJ, Lewis GP and Catalano SA** (2022) Disintegration of the genus *Prosopis* L. (Leguminosae, Caesalpinoideae, mimosoid clade). In Hughes CE, de Queiroz LP and Lewis GP (eds), *Advances in Legume Systematics 14. Classification of Caesalpinoideae Part 1: New generic delimitations. PhytoKeys* 205, 147–189. doi:10.3897/phytokeys.205.75379
- Iwasaki M, Penfield S and Lopez-Molina L** (2022) Parental and environmental control of seed dormancy in *Arabidopsis thaliana*. *Annual Review of Plant Biology* 73, 355–378. doi:10.1146/annurev-arplant-102820-090750
- Klein Z and Mitchell RM** (2024) Seed source environment predicts response to water availability in *Plantago patagonica*. *Restoration Ecology* 32, e14002. doi:10.1111/rec.14002
- Larson JE, Anacker BL, Wanous S and Funk JL** (2020) Ecological strategies begin at germination: traits, plasticity, and survival in the first four days of plant life. *Functional Ecology* 34, 968–979.
- Lascoux M, Glémin S and Savolainen O** (2016) Local adaptation in plants. In: *eLS*. Chichester: John Wiley & Sons, Ltd. online doi:10.1002/9780470015902.a0025270.
- Leimu R and Fischer M** (2008) A meta-analysis of local adaptation in plants. *PLoS ONE* 3, e4010. doi:10.1371/journal.pone.0004010
- Lerner P and Peinetti R** (1996) Importance of predation and germination on losses from the seed bank of calden (*Prosopis caldenia*). *Journal of Range Management* 49, 147–150.
- Lewandrowski W, Stevens JC, Webber BL, Dalziel E, Trudgen MS, Bateman AM and Erickson TE** (2021) Global change impacts on arid zone ecosystems: seedling establishment processes are threatened by temperature and water stress. *Ecology and Evolution* 11, 8071–8084. doi:10.1002/ece3.7638
- Ooi MKJ** (2012) Seed bank persistence and climate change. *Seed Science Research* 22, 53–60. doi:10.1017/S0960258511000407
- Peinetti R, Pereyra M, Kin A and Sosa A** (1993) Effects of cattle ingestion on viability and germination rate of caldén (*Prosopis caldenia*) seeds. *Journal of Range Management* 46, 483–486.
- Pérez DR, Pilustrelli C, Farinaccio FM, Sabino G and Aronson J** (2019) Evaluating success of various restorative interventions through drone- and field-collected data, using six putative framework species in Argentinian Patagonia. *Restoration Ecology* 28, 1–10. doi: 10.1111/rec.13025
- Pérez DR, Ceballos C and Oneto ME** (2022) Outplanting and direct seeding costs of *Prosopis flexuosa* var. *depressa* (Fabaceae) for ecological restoration. *Acta Botánica Mexicana* 129, e1888. doi:10.21829/abm129.2022.1888
- Renzi JP, Brus J, Duchoslav M, Pirintso S, Erdős L and Smykal P** (2020) Release of *Medicago truncatula* and *Pisum sativum* subsp. *elatius* seed dormancy tested in soil conditions. *Agronomy* 10, 1026. doi:10.3390/agronomy10071026
- Renzi JP, Reinoso O, Quintana M and Cantamutto MA** (2023) Natural distribution and phenotypic traits of *Neltuma flexuosa* var. *depressa*, a candidate taxon for the rehabilitation in the central arid region of Argentina. *Journal of Arid Environments* 209, 104889. doi:10.1016/j.jaridenv.2022.104889
- Rocha OJ and Aguilar G** (2001) Reproductive biology of the dry forest tree *Enterolobium cyclocarpum* (Guanacaste) in Costa Rica: a comparison between trees left in pastures and trees in continuous forest. *American Journal of Botany* 88, 1607–1614.
- Roig FA** (1993) *Informe nacional para la selección de germoplasma en especies del género Prosopis*. Mendoza: Contribuciones mendocinas a la V Reunión Regional para América Latina y el Caribe del CIID, pp. 1–36.
- Rubio de Casas R, Willis CG, Pearse WD, Baskin CC, Baskin JM and Cavender-Bares J** (2017) Global biogeography of seed dormancy is determined by seasonality and seed size: a case study in the legumes. *New Phytologist* 214, 1527–1536. doi:10.1111/nph.14498
- Saatkamp A, Poschlod P, Venable DL and Gallagher RS** (2014) The functional role of soil seed banks in natural communities. In Gallagher RS (ed.), *Seeds: The Ecology of Regeneration in Plant Communities*, CABI, pp. 263–295. doi:10.1079/9781780641836.0263.
- Soliani C, Ceccarelli V, Lantschner MV, Thomas E and Marchelli P** (2024) Predicting the distribution of plant species from Southern South America: are the hotspots of genetic diversity threatened by climate change? *Biodiversity and Conservation* 33, 725–757. doi:10.1007/s10531-023-02770-0
- Somlo R, Cohen L, Giorgetti H, Montenegro O and Rodriguez G** (1997) *Tabla de valor nutritive de especies forrajeras, Monte Pampeano*. Comunicación Técnica No6. Buenos Aires. Ed. INTA, p. 13.
- Steibel PE and Troiani HO** (1999) El género *Prosopis* (Leguminosae) en la Provincia de La Pampa (República Argentina). *Revista de la Facultad de Agronomía* 10, 25–48.
- Torres Robles SS, Arturi MF, Contreras C, Peter G and Zebedio JM** (2022) Variaciones geográficas de la estructura y composición de la vegetación leñosa en el límite entre el espinal y el monte en el Noreste de la Patagonia (Argentina). *Boletín de la Sociedad Argentina de Botánica* 50, 209–215.
- Velez S, Chacoff NP and Campos CM** (2018) Pre-dispersal seed loss in two *Prosopis* species (Fabaceae: Mimosoidea) from the Monte Desert. *Argentina. Ecología Austral* 28, 361–373.
- Vilela AE and Ravetta DA** (2001) The effect of seed scarification and soil-media on germination, growth, and survival of seedlings of five species of *Prosopis* L. (Mimosaceae). *Journal of Arid Environments* 48, 171–184.
- Villagra PE** (1997) Germination of *Prosopis argentina* and *P. alpataco* seeds under saline conditions. *Journal of Arid Environments* 37, 261–267.
- Villagra PE, Marone L and Cony MA** (2002) Mechanisms affecting the fate of *P. flexuosa* seeds during secondary dispersion. in the Monte desert. *Austral Ecology* 27, 416–421.
- Villagra PE, Vilela A, Giordano C and Álvarez JA** (2010) Ecophysiology of *Prosopis* species from the arid lands of Argentina: what do we know about adaptation to stressful environments?. In Ramawat KG (ed.), *Desert Plants*. Berlin, Germany: Springer, pp. 321–340. doi:10.1007/978-3-642-02550-1_15.
- Villalobos AE and Peláez DV** (2001) Influences of temperature and water stress on germination and establishment of *Prosopis caldenia* Burk. *Journal of Arid Environments* 49, 321–328.
- Winschel C and Pezzola A** (2018) *Avance de la frontera agrícola sobre el monte nativo en Villarino y Patagones, provincia de Buenos Aires, 1975–2018*. Informe técnico N°60. Buenos Aires. Ed. INTA, p. 34.
- Woolridge CB, Fant JB, Flores AI, Schultz K and Kramer AT** (2022) Variation in overall fitness due to seed source: projections for predictive provenancing. *Restoration Ecology* 31, 1–10. doi: 10.1111/rec.13717
- Yin J, Zhou M, Lin Z, Li QQ and Zhang Y** (2019) Transgenerational effects benefit offspring across diverse environments: a meta-analysis in plants and animals. *Ecology Letters* 22, 1976–1986. doi: 10.1111/ele.13373
- Zeberio JM and Pérez CA** (2020) Rehabilitation of degraded areas in north-eastern Patagonia, Argentina: effects of environmental conditions and plant functional traits on performance of native woody species. *Journal of Arid Land* 12, 653–665. doi:10.1007/s40333-020-0021