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## Patterns of seed germination in Mediterranean mountains: study on 37 endemic or rare species from Sierra Nevada, SE Spain

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### Abstract

LORITE, J., M. RUIZ-GIRELA & J. CASTRO (2007). Patterns of seed germination in Mediterranean mountains: study on 37 endemic or rare species from Sierra Nevada, SE Spain. *Candollea* 62: 5-16. In English, English and French abstracts.

The study of seed germination of endangered flora species is vital for conservation strategies. In this context, performed germination experiments were performed with a set of 37 endemic, rare or often endangered, taxa of the Sierra Nevada high mountain (SE Spain), an area that constitutes a hotspot of diversity in the Mediterranean region. Seed germination varied depending on the species, from 0% to ca. 95%. A group of 10 species belonging to 9 families showed a low or null germination percentage. Dormancy in these species is likely to be complex and to involve some kind of physiological ground. A second group of species showed a germination percentage between 5 and 20%, most of these belonging to families producing hard-coated seeds (e.g. Papillionaceae, Geraniaceae). Treatments to promote their germination were thus easy to apply. A large fraction of taxa had a germination percentage above 20%, often above 50%. The germination percentage did not correlate positively with the speed of germination (estimated as the T<sub>50</sub>) or with altitude, but correlated negatively

### Résumé

LORITE, J., M. RUIZ-GIRELA & J. CASTRO (2007). Modèles de germination de graines dans les montagnes méditerranéennes: étude de 37 espèces endémiques ou rares de la Sierra Nevada, SE Espagne. *Candollea* 62: 5-16. En anglais, résumés anglais et français.

L'étude de la germination des graines d'espèces en danger est fondamentale pour mettre en place des stratégies de conservation. Dans ce contexte, des expériences de germination ont été menées sur 37 taxons endémiques, rares ou souvent en danger, en provenance des hautes montagnes de la Sierra Nevada (SE de l'Espagne), une aire géographique qui constitute un «hotspot» de diversité dans la région méditerranéenne. La germination des graines a varié selon l'espèce de 0% à 95%. Un groupe de 10 espèces appartenant à 9 familles ont montré un pourcentage de germination bas ou nul. La dormance de ces espèces est complexe et dépend de mécanismes physiologiques. Un second groupe d'espèces a montré un pourcentage de germination entre 5 et 20%, la plupart de ces espèces appartenant à des familles produisant des graines à côtes rugueuses (ex. Papillionaceae, Geraniaceae). Les traitements pour promouvoir leur germination ont été faciles à appliquer. Une grande partie de ces taxons ont présenté un pourcentage de germination au dessus de 20%, souvent au dessus de 50%. La corrélation du

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(although weakly) with diaspore mass. Families such as *Compositae*, *Cruciferae*, *Geraniaceae*, *Labiatae* and *Papillionaceae* are identified as families where seed germination may not imply major problems. *Aquilegia vulgaris* subsp. *nevadensis*, *Arenaria nevadensis*, *Carex camposii*, *Gentiana lutea*, *Linaria glacialis*, *Narcissus nevadensis*, *Nardus stricta*, *Odontites granatensis*, *Papaver lapeyrousianum* and *Sorbus hybrida* need studies of dormancy breakage to establish guidelines to optimize management of the scant seed material. As a whole, the results contrast with the generalized idea of a high level of dormancy in alpine species.

### **Key-words**

Sierra Nevada National Park – Seed germination – Endemic species – Rare species – Threatened flora – Plant conservation – Mediterranean high mountain

pourcentage de germination n'a pas été trouvé positive avec la vitesse de germination (estimée comme le T<sub>50</sub>) ou avec l'altitude. Elle a été trouvé négative (faiblement) avec la masse de la diaspore. Les familles comme les *Compositae*, *Cruciferae*, *Geraniaceae*, *Labiatae* et *Papillionaceae* ont été identifiées comme les familles où les germinations de graine ne présentent pas de problèmes majeurs. Des espèces comme *Aquilegia vulgaris* subsp. *nevadensis*, *Arenaria nevadensis*, *Carex camposii*, *Gentiana lutea*, *Linaria glacialis*, *Narcissus nevadensis*, *Nardus stricta*, *Odontites granatensis*, *Papaver lapeyrousianum* et *Sorbus hybrida* ont besoin d'études sur la rupture de dormance pour permettre la mise en place de directives visant à optimiser la gestion de stock peu abondant de graines. Ces résultats contrastent avec l'idée que les espèces alpines ont une dormance élevée.

### Introduction

The knowledge of the germination capacity and germination requirements of any plant species is of interest for its management, but, in particular, the study of the germination of rare and endemic species is of paramount importance for the conservation and management of these species for several reasons. First, seed germination is the only way to preserve the genetic variability of the populations (FENNER & THOMPSON, 2005). Second, these species are in many cases under risk of extinction, so that an accurate understanding of their germination ability is critical to optimize conservation and restoration efforts (Gaston, 1994; Blanca & al., 1998; Moreno-Sáiz & al., 2003). However, the germination of endemic and rare species, in particular from high mountain and alpine habitats, is often scarcely known, as these species have a restricted distribution, are difficult to sample, and are, in many cases, under threat of extinction due to low population levels, which restrict the use of plant material and even make it undesirable in years of low seed production (ESCUDERO & IRIONDO, 2003). In addition, the germination of these species is in many cases subjected to complex physiological controls, given that they inhabit a type of habitat (e.g. high mountain, specific soil types) that has shaped a set of traits related to seed dormancy (KÖRNER, 1999).

In this study, we analyse the germination capacity of 37 rare (i.e. having low abundance and/or small distribution range; see Greuter, 1991; Gaston, 1994) or endemic taxa from Sierra Nevada mountain, SE Spain. Sierra Nevada, together with the Baetic range as a whole, constitutes an important reservoir of diversity and speciation (Heywood, 1995; Domínguez & al., 1996; Médail & Quézel, 1999; Thompson, 2005). It also constitutes a hotspot of endemicity, with 80 exclusive taxa and 125 threatened taxa (Blanca, 2002). There is, however, very little information available on the germination capacity of the species that inhabit Sierra Nevada high mountain, which is a preliminary and necessary step to carry out recovery plans.

Because collection of large number of seeds from these taxa is undesirable in most cases, our aim was to check seed germination ability of a large number of species before treatments to break possible dormancies are used. As far as we know, most of the taxa studied here have never been tested for germination. There are few exceptions, such as *Allium schoenoprasum* L. (SPECHT & KELLER, 1997), *Astragalus granatensis* Steud. (ANGOSTO & MATILLA, 1993), or very closely related taxa such as *Hippocrepis comosa* L. (HENNENBERG & BRUELHEIDE, 2003) or *Primula elatior* subsp. *elatior* (L.) L. (JACQUEMYN & al., 2001). In any case, we include these species in the study because they are threatened in Sierra Nevada and, in addition, the germination capacity may differ greatly among populations (e.g. Albert & al., 2002; Cerabolini & al., 2004; Giménez-Benavides & al., 2005; Fenner & Thompson, 2005).

Three questions were posed here: 1) what is the percentage and speed of germination of untreated seeds of these taxa? 2) is there any relationship between seed germination and traits related either to the seeds (seed mass) or to the habitat (altitude)? 3) which guidelines can be drawn to optimize the use of seeds of these species?

### Material and Methods

Study area

Sierra Nevada is a mountain of 2100 km<sup>2</sup> located in SE Spain (37°N 3°W). It has complex orography, bedrock and soil composition (e.g. areas of dolomitic soils), and reaches a height of 3482 m. Sierra Nevada marks the southernmost limit of the influence of the Quaternary glaciations in Europe, when it was covered with glaciers only in areas above 2500 m (approx.) while large areas of it remained free of permanent ice (GÓMEZ & al., 2001). All this contributed to make of Sierra Nevada a refuge for many plant species during the glacial ages (BLANCA & al., 1998) and, in addition, to harbour isolated populations that have evolved under particular conditions (e.g. soil type or isolated summit areas) which have contributed to speciation and to a high level of endemicity (BLONDEL & ARONSON, 1999; PEÑAS & al., 2005; THOMPSON, 2005). In fact, the area above 2000 m contains about 100 endemic or rare taxa of which 32 are included in this study. Many of these species are threatened by different factors (BLANCA & al., 1998; BAÑARES & al., 2003; see Table 1 for details), with 38 taxa included on the regional protection list (25 taxa with populations above 2000 m; LORITE & al., 2006). Today the distribution areas of the species included in this study lie within National and/or Natural parks. Climatic conditions in the localities of seed collection differ depending on the altitude (localities ranging from 1230 to 3300 m; Table 2). However, most of the species are distributed above ca. 1800 m, which marks the timberline and the start of the orophilous area in Sierra Nevada. Overall, the climate is typically alpine (with mediterranean character), with mean temperatures below 0°C during winter months and snow cover that can persist up to 8 months in the highest localities (occasionally up to 10 months in small, protected areas). Gómez (2002) has reported detailed description of climatic conditions in Sierra Nevada mountain.

### Seed-germination experiments

Ripe fruits of each species studied (see Table 1 for a list of taxa and other relevant characteristics) were collected from natural populations from Sierra Nevada during the summers 2000 to 2003, depending on the species (see Table 2 for

**Table 1.** — Data about life form, distribution and habitat of the 37 taxa from Sierra Nevada high mountain used in this study (Biotype: **Ph** = Phanerophyte, **Ch** = Chamaephyte, **Hc** = Hemicryptophyte, **Ge** = Geophyte, **Th** = Therophyte):

Distribution for endemism (**SN** = Endemic of Sierra Nevada, **BM** = Endemic Baetic Mountains (Sierra Nevada and other nearby mountains such as Sierra de los Filabres, Sierra de Baza, or Sierra de Gádor)).

Threatened categories (according CABEZUDO & al., 2005; following IUCN categories; IUCN, 2001) (**CR** = Critically endangered, **EN** = Endangered, **VU** = Vulnerable, **NT** = Near threatened, — = No threatened. (\*) = less than 2000 reproductive individuals in Sierra Nevada (from Blanca, 2002)).

Taxon	Biotype	Distribution	tribution Habitat			
Allium schoenoprasum L.	Ge	Holartic	Clearings of cushions scrubs	1800-2300	VU	
Anthyllis tejedensis Boiss.	Ch	Baetic mountains and North Africa				
		(Morocco and Algeria)	Dolomitic sandy areas	1000-2000	_	
Aquilegia vulgaris subsp. nevadensis						
(Boiss. & Reut.) T. E. Díaz	Нс	Endemic SN	Siliceous wet meadows	1200-2500	VU	
Arabis margaritae Talavera	Нс	Endemic SN	Hygrophilous high-mountain pasturelands	2000-2500	CR	
Arenaria nevadensis Boiss. & Reut.	Th	Endemic SN	Siliceous screes	2950-3200	CR*	
Armeria filicaulis subsp. trevenqueana						
Nieto Feliner	Нс	Endemic SN	Dolomitic sandy areas	1700-2000	EN	
Artemisia alba subsp. nevadensis (Willk.)						
Blanca & Morales Torres	Ch	Endemic BM	Limestone montane cushion scrubs	1800-2200	EN*	
Artemisia umbelliformis Lam.	Ch	Alps, Apennines and Sierra Nevada	Rocky fissures and siliceous screes	2800-3000	EN*	
Astragalus granatensis Lam.	Ch	southern half of Spain and North Africa				
		(Atlas)	Montane cushion scrubs	1800-2700	_	
Carex camposii Boiss. & Reut.	Нс	Endemic BM	Hygrophilous meadows on siliceous substrates	1300-3000	NT	
Centaurea gadorensis Blanca	Ch	Endemic BM	Scrublands on calcareous substrates	1300-2000	VU	
Chaenorrhinum glareosum (Boiss.) Willk.	Нс	Endemic SN	Rocky fissures and siliceous screes	2400-3300	_	
Chamaespartium undulatum (Ern)						
Talavera & L. Sáez	Ch	Endemic SN	Dolomitic sandy areas	1300-1700	VU	
Delphinium nevadense Kunze	Нс	Endemic BM	Mesophilous woodlands and thorny Shrublands	1100-1600	VU	
Draba hispanica subsp. laderoi						
Rivas Martínez, M. E. García & Penas	Ch	Endemic SN	Rocky fissures	2400-3200	_	
Echinospartum boissieri (Spach) Rothm.	Ch	Endemic BM	Scrublands on calcareous substrates	1000-1900	_	
Erigeron frigidus Boiss.	Нс	Endemic SN	Psychroxerophilous pasturelands	2900-3400	VU	
Erodium boissieri Coss.	Ch	Endemic SN	Dolomitic sandy areas	1300-2000	VU	
Erodium rupicola Boiss.	Нс	Endemic BM	Rocky fissures	1500-1900	VU	
Gentiana lutea L.	Нс	Mountains of C-S Europe	Wet meadows	1900-2300	CR*	
Hippocrepis prostrata Boiss.	Ch	Endemic SN	Montane cushion scrubs	1800-2300	CR*	
Hormathophylla purpurea (Lag. & Rodr.)						
P. Küpfer	Нс	Endemic SN	Psychroxerophilous pasturelands	2500-3400	VU	
Hormathophylla spinosa (L.) P. Küpfer	Ch	Iberian Peninsula and N Africa	Montane cushion scrubs	1700-3300	_	
Lavandula lanata Boiss.	Ch	Endemic BM	Scrublands on calcareous substrates	1200-1900	_	
Linaria glacialis Boiss.	Нс	Endemic SN	Siliceous screes	2400-3400	VU	
Moehringia fontqueri Pau	Нс	Endemic SN	Rocky fissures	1800-2400	EN	
Narcissus nevadensis Pugsley	Ge	Endemic SN	Hygrophilous meadows	1400-2000	EN	
Nardus stricta L.	Нс	Holartic	Hygrophilous high-mountain pasturelands	2000-3200		

Taxon	on Biotype Distribution		Habitat	Altitudinal range [m]	Threatened
Odontites granatensis Boiss.	Th	Endemic SN	Juniper-genista patches over calcareous substrate	1900-2200	CR
Papaver lapeyrousianum Guterm.	Нс	Sierra Nevada and Pyrenees	Psychroxerophilous pasturelands	3200-3450	EN
Plantago nivalis Boiss.	Нс	Endemic SN	Hygrophilous high-mountain pasturelands	2400-3100	_
Primula elatior subsp. lofthousei					
(HeslHarr. f.) W. W. Sm. & Fletcher	Нс	Endemic BM	Hygrophilous meadows	1500-2900	VU
Senecio elodes Boiss.	Нс	Endemic SN	Hygrophilous meadows	1800-2500	EN*
Senecio nevadensis Boiss.	Ch	Endemic SN	Siliceous screes	2600-3300	VU
Sorbus hybrida L.	Ph	Mountains of Europe	Mixed deciduous forests	1800-2000	CR*
Teucrium oxylepis Font Quer & Pau					
subsp. <i>oxylepis</i>	Ch	Endemic BM	Scrublands	1700-1850	CR*
Thymus serpylloides Bory					
subsp. serpylloides	Ch	Endemic BM	Montane cushion scrubs	1900-2900	_
Vella spinosa Boiss.	Ch	southern Iberian Peninsula	Montane cushion scrubs	1800-2200	_

collection dates). Fruits were collected from at least 50 individuals per species whenever possible, except for Sorbus hybrida, as population size was below 50 individuals. Seeds were manually cleaned and separated from the fruit coats, eliminating any visually malformed seeds. Three subsamples of 100 seeds were weighed to determine mean propagule mass (except in two species; Table 2). Seeds were thereafter stored in darkness in paper bags under ambient conditions (ca. 20°C and ca. 30% relative humidity) for ca 5-7 months until the start of germination tests (10-11 months in Narcissus nevadensis and Aquilegia vulgaris, see Table 2). Seeds were the target of this study in all cases except in Compositae (where achenes were used) or *Plumbaginaceae* (seeds with calyx); these do not differ functionally from seeds from the standpoint of managing these species, and thus hereafter we refer to all propagules types as seeds.

Seeds were germinated in an incubator chamber (ASL,  $\pm 0.1^{\circ}$  C) with a photoperiod of 16 h and alternating temperatures of 15°C during dark periods and 20°C during light periods. Alternating day-night temperatures between 10 and 20°C have been reported as promoter of highest germination rates in alpine species (e.g. BLISS, 1971; KÖRNER, 1999). Prior to germination experiments, seeds were disinfected by immersion in a 1% sodium hypochlorite solution for 10 min, followed by thorough rinsing with sterile distilled water. Seeds were thereafter placed in plastic Petri dishes of 9 cm diameter containing a disk of filter paper resting on a layer of vermiculite in order to maintain the moisture level, all material being previously sterilised. Each Petri dish contained from 25 to 100 seeds, with 4 (occasionally 3 or 5) replicates (see Table 2 for details). Dishes were randomly repositioned within the chamber every 5 days.

Germination, identified as visible radical protrusion, was recorded at 2-3 days interval during a 25-85 days period (Table 2). The germination rate was estimated as  $T_{50}$ , i.e. the time needed for manifestation of half of the final germination level.

### Data analysis

The relationship between 1) germination percentage and the speed of germination  $(T_{50})$ , 2) germination percentage and altitude, 3) T<sub>50</sub> and altitude, and 4) germination percentage and diaspore mass, were analysed using the Pearson's productmoment correlation. For this, all the species with germination values lower than 5% were eliminated from the analysis, assuming that those values are indicative of deep dormancy and could bias the correlations. In all these analyses, the values of germination or T<sub>50</sub> were the mean values of the four replicates per species (Table 2). The T<sub>50</sub> values were not calculated in case lower than 5% germination. Seeds that were attacked by fungi were removed from the plates and eliminated from analysis; in those cases the germination percentage was calculated considering the number of non-contaminated seeds. Analyses were performed with JMP 5.0 software (SAS Institute). Throughout the paper, means are followed by  $\pm$  SE.

### **Results**

Contamination of plates was low (0 to < 5% of seeds contaminated in most cases), with only *Linaria glacialis* and *Gentiana lutea* showing severe fungal attack (11.5  $\pm$  2.0% and 28.0  $\pm$  7.6% of seeds contaminated, respectively). Seed germination varied considerably, from 0% germination to around 95%

**Table 2.** — Seed-germination percentage and some details related to germination experiments for the 37 taxa from Sierra Nevada high mountain used in this study (**Germ.** = Date of the start of the germination test, **N** = Number of replicates used for germination experiments (Petri dishes), **SPD** = Seeds per Petri dish, **DE** = Days of experiment (number of days in which germination was monitored), **Seed mass** = Weight for 100 seeds, **T**<sub>50</sub> = Time (in days) needed for reaching 50% of the final germination percentage, —— = not calculated. \*Seed mass from ANGOSTO & MATILLA (1993)).

	Family	Taxon	Date collection	Altitude [m]	Germ.	N	SPD	DE	Seed mass	Germination [%]	т <sub>50</sub>
Group 1	Amarillydaceae	Narcissus nevadensis	6/2002	2100	5/2003	5	100	55	0.573	0.0±0.0	_
	Caryophyllaceae	Arenaria nevadensis	8/2000	3000	12/2000	5	50	55	0.014	0.0±0.0	_
	Сурегасеае	Carex camposii	7/2002	2350	3/2003	4	100	30	0.121	0.5±0.5	_
	Gentianaceae	Gentiana lutea	8/2002	2250	3/2003	4	100	55	0.010	0.3±0.3	_
	Papaveraceae	Papaver lapeyrousianum	9/2002	3300	4/2003	4	100	35	0.008	2.0±0.6	_
	Poaceae	Nardus stricta	9/2002	2730	4/2003	4	100	55	0,040	4.5±1.0	_
	Ranunculaceae	Aquilegia vulgaris									
		subsp. nevadensis	7/2002	2010	5/2003	4	100	55	0.133	$0.0 \pm 0.0$	_
	Rosaceae	Sorbus hybrida	10/2002	1850	3/2003	5	25	55	1.200	$0.0 \pm 0.0$	_
	Scrophulariaceae	Linaria glacialis	8/2002	3050	4/2003	4	100	40	0.051	1.1±0.4	_
	Scrophulariaceae	Odontites granatensis	8/2000	2160	12/2000	4	50	30	0.054	$0.0 \pm 0.0$	_
Group 2	Geraniaceae	Erodium boissieri	7/2002	1850	3/2003	4	42	60	0.350	13.6±2,7	12.3±1.9
	Labiatae	Thymus serpylloides									
		subsp. serpylloides	8/2002	2360	5/2003	4	100	30	0.008	18.3±4.4	$3.5 \pm 0.5$
	Liliaceae	Allium schoenoprasum	9/2000	2450	12/2000	4	100	45	0.175	13.8±2.0	13.5±1.0
	Papilionaceae	Anthyllis tejedensis	7/2002	1758	3/2003	4	110	55	0.301	20.2±1.4	16.0±0.7
	Papilionaceae	Echinospartum boissieri	7/2002	1680	3/2003	4	100	55	_	5.0±1.0	35.8±3.5
	Papilionaceae	Hippocrepis prostrata	8/2002	1850	3/2003	4	100	55	0.283	8.5±1.0	8.0±1.3
Group 3	Caryophyllaceae	Moehringia fontqueri	8/2002	2169	4/2003	4	100	40	0.008	76.3±0.9	9.0±0.0
	Compositae	Artemisa alba									
		subsp. nevadensis	8/2000	1890	11/2000	4	100	25	0.022	31.2±2.4	9.5±1.4
	Compositae	Artemisia umbelliformis	9/2000	2900	12/2000	3	100	30	0.032	90.7±1.2	12.0±0.0
	Compositae	Centaurea gadorensis	7/2002	1380	3/2003	4	100	85	0.446	54.7±2.8	14.8±2.7
	Compositae	Erigeron frigidus	9/2000	3100	11/2000	4	100	30	0.020	93.0±1.2	4.0±0.4
	Compositae	Senecio elodes	8/2000	2044	11/2000	4	100	30	0.068	32.0±1.7	9.0±0.4
	Compositae	Senecio nevadensis	9/2000	3189	01/2001	4	100	25	0.041	22.8±2.5	11.0±0.4
	Cruciferae	Arabis margaritae	7/2002	2350	5/2003	5	100	35	0.015	88.8±1.9	no data
	Cruciferae	Draba hispanica									
		subsp. <i>laderoi</i>	8/2002	2580	3/2003	4	100	50	0.014	93.8±0.9	12.3±0.3
	Cruciferae	Hormathophylla									
		purpurea	8/2002	2980	3/2003	5	110	40	0.124	43.6±2.3	5.6±0.2
	Cruciferae	Hormathophylla									
		spinosa	8/2002	2050	3/2003	4	100	30	0.070	94.8±1.7	3.0±0.0
	Cruciferae	Vella spinosa	8/2002	2100	4/2003	4	100	35	_	35.8±2.6	7.3±0.2
	Geraniaceae	Erodium rupicola	6/2002	2000	3/2003	3	75	60	0.245	54.7±4.0	14.0±1.1
	Labiatae	Lavandula lanata	9/2002	1600	4/2003	4	100	35	0.143	31.1±3.8	15.0±0.4
	Labiatae	Teucrium oxylepis									
		subsp. <i>oxylepis</i>	11/2002	1750	5/2003	4	100	65	0.092	49.5±9.6	7.8±0.3

F	Family	Taxon	Date collection	Altitude	Germ.	N	SPD	DE	Seed mass	Germination	T <sub>50</sub>
				[m]					[g]	[%]	
F	Papilionaceae	Astragalus granatensis	10/2002	2180	4/2003	4	100	30	0.520*	41.5±7.2	8.8±0.3
F	Plantaginaceae	Plantago nivalis	8/2002	2857	2/2003	4	100	60	0.121	20.7±1.2	7.3±0.3
F	Plumbaginaceae	Armeria filicaulis									
		subsp. trevenqueana	7/2002	1600	5/2003	5	50	35	0.072	94.8±1.7	t
F	Primulaceae	Primula elatior									
		subsp. lofthousei	8/2002	2073	4/2003	4	100	55	0.084	79.5±2.5	14.7±0.9
K	Ranunculaceae	Delphinium nevadense	8/2002	1230	4/2003	5	50	40	0.138	96.0±1.7	t
9	Scrophulariaceae	Chaenorrhinum									
		glareosum	8/2002	3150	3/2003	4	100	60	0.006	79.3±4.7	12.5±0.7

(Table 2). Similarly,  $T_{50}$  varied considerably across species (Table 2), ranging from 3 (*Hormathophylla spinosa*) to 35.8 (*Echinospartium boissieri*). No correlation was found between  $T_{50}$  and germination percentage (P = 0.19; Fig. 1).

The percentage of germination was not correlated with the altitude where the population grows (P = 0.74), nor was there any correlation between altitude and  $T_{50}$  values (P = 0.17). By contrast, there was a marginally significant, negative correlation between propagule mass and germination percentage ( $r^2 = 0.15$ , P = 0.056; Fig. 2).

### Discussion

Differences in germination percentages across species

The results reveal substantial differences in germination among species, as might be expected due to the heterogeneity of the genera and families studied. However, three main groups of taxa can be distinguished according to germination percentages. First, 10 species had low (below 5%) or null germination (Table 2). Some of these species belong to genera or families with a characteristic physiological dormancy. For instance, Carex species germinates better after cold stratification, and many of them even fail to germinate without chilling (SCHÜTZ & RAVE, 1999). Seeds of Gentiana species also need stratification for germination (i.e. Gentiana kurroo; RAINA & al., 2003; see also BASKIN & BASKIN, 1998, for other species of Gentianaceae). Seeds of most temperate Sorbus species are dormant at maturity and needs moist chilling to enable germination (e.g. Yagihashi & al., 1998; Jensen, 2003; TAKOS & EFTHIMIOU, 2003), and similarly seeds of Aquilegia species also require stratification for germination (e.g. FINNERTY & al., 1992). For species closely related to Narcissus nevadensis (N. longispathus and N. bugei), germination values of around 10% were registered after cold stratification,

and about 50% after one year of sowing in a nursery (BLANCA & al., 1999). This suggests that Carex camposii, Gentiana lutea, Sorbus hybrida, Aquilegia vulgaris subsp. nevadensis and Narcissus nevadensis may require moist chilling to promote germination. There are still other possibilities to explain the low germination percentages in this group of species. For instance, some Linaria species show higher germination or emergence percentages both after chilling or coat scarification (e.g. Saner & al., 1995; Stöcklin & Bäumler, 1996; Schwien-BACHER & ERSCHBAMER, 2002). This suggests that both physical and physiological dormancy may be operating in the dormancy of *Linaria glacialis* seeds here studied. BLISS & GOLD (1999) found a low germination percentage (7%) for seeds of the arctic species Papaver radicatum after 40 days (thus five days more than in our experiment), but germination rose to 23% after 90 days of experiment, pointing out that the germination of species such as P. lapeyrousianum may require longer time. We are not aware of any information for Arenaria nevadensis, Nardus stricta or Odontites granatensis (or closely related species), all of these with low germination percentages in the present study. However, a cold, moist stratification would probably help to encourage their germination, as is common in many alpine and arctic species with dormancy (BASKIN & BASKIN, 1998; KÖRNER, 1999).

A second group of species can be formed with those having a germination percentage between 5 and 20% (Table 2). This is relatively low, but germination in these species is not likely to be a major problem for their management and conservation for several reasons. First, some belong to genera and/or families with a characteristic physical dormancy imposed by hard coats, as is the case of *Papilionaceae* (see BASKIN & BASKIN, 1998; LÓPEZ & al., 1999, for discussion of physical dormancy in this family). It is thus likely that high germination percentages in *Echinospartium boissieri*, *Hippocrepis prostrata*, *Anthyllis tejedensis* and *Astragalus* 

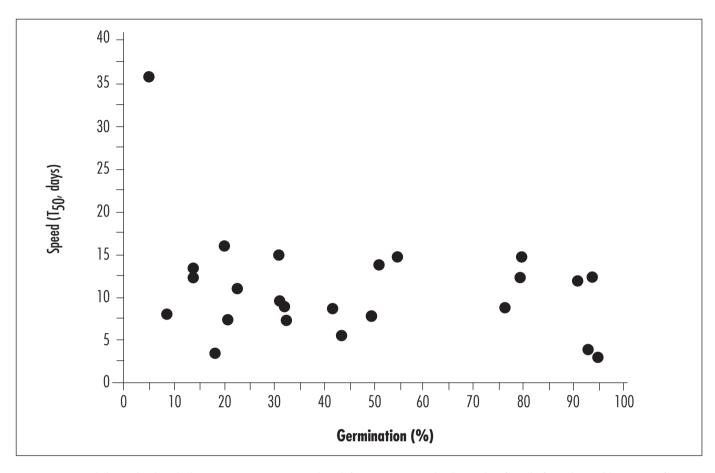


Fig. 1. — Scatter graph showing the relationship between germination percentage and speed of germination (estimated as the T<sub>50</sub>, days) for seeds of 24 endemic and/or rare species from Sierra Nevada high mountain germinated without promoting treatments. For each species, values are the mean of the replicates used in the germination experiments (see Table 2 for details). All the species used had a germination rate of 5%.

granatensis could be achieved after coat scarification with abrasive methods, either sulphuric acid or sandpaper (see Aparicio & Guisande, 1997; Ibáñez & Passera, 1997; López & al., 1999 for increased germination after coat scarification in related species; and ANGOSTO & MATILLA, 1993 for similar results with A. granatensis). Physical dormancy imposed by coats is also characteristic in Geraniaceae (BASKIN & BASKIN, 1998; MEISERT, 2002), suggesting that the germination of Erodium boissieri (14%) could be easily promoted. The germination percentage for Allium schoenoprasum (14%) was similar to that reported by GIMÉNEZ-BENAVIDES & al. (2005) (seeds from central Spain) and close to values around 20% reported by SPECHT & KELLER (1997), supporting the idea that the populations from Sierra Nevada have no particular impediments for germination. In addition, germination of this species was boosted by cold-wet stratification (up to 98%; GIMÉNEZ-

BENAVIDES & al., 2005). *Thymus serpylloides* registered 18% germination, which is below the commonly high values of germination in other *Thymus* species; for instance, *T. pulegioides* ca. 60% (WIERZBICKA & OBIDZINSKA, 1998), *T. loscosii*, *T. mastichina*, *T. vulgaris* and *T. zygis*, above 70% (ALBERT & al., 2002; GONZÁLEZ-BENITO & al., 2004). This endemic species, however, is abundant in Sierra Nevada, and collection of an amount of seeds large enough to supply nursery and conservation demands is not a problem.

Finally, there is a third group of 21 taxa in which the germination percentage was above 20%, and even more, 15 out of the 37 species studied had a germination percentage above 40%, many actually above 75% (Table 2). These species therefore need no particular treatments for their germination, and may be propagated soon after seed collection. This should be taken into account in order to optimise time and efforts in

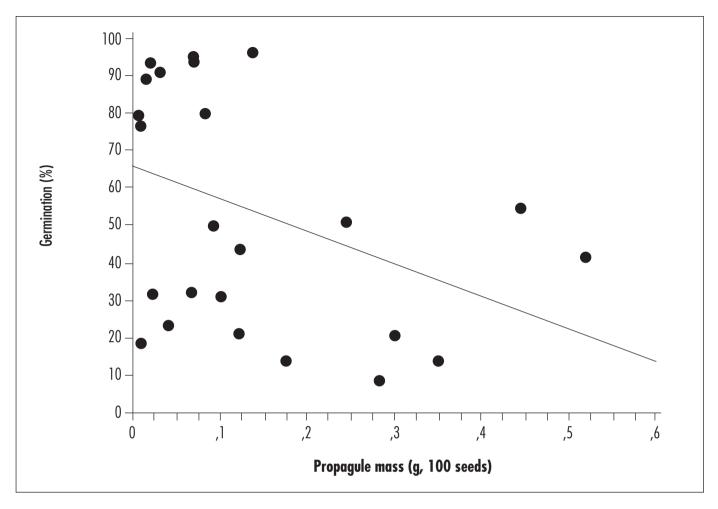


Fig. 2. — Correlation between propagule mass and germination percentage for 25 endemic and/or rare species from Sierra Nevada high mountain germinated without promoting treatments. See Table 2 for species identity and propagule mass. Germination values are the mean of the replicates used in the experiments (see Table 2 for details). All the species used had a germination rate of 5%.

conservation and restoration programs. For instances, *Primula* species are commonly cold stratified before germination (MCKEE & RICHARDS, 1998; WATANABE & al., 2003; CERABOLINI & al., 2004), including the taxa *Primula elatior* (JACQUEMYN & al., 2001), closely related to *P. elatior* subsp. *lofthousei* studied here. However, our results reflect a germination percentage of 79% without chilling, even above the values of around 55% found by JACQUEMYN & al. (2001) after 8 weeks of moist chilling (see also Shimono & Washitani, 2004, for 20-60% germination without chilling in *Primula modesta*). The endemic *Plantago nivalis* showed the lowest germination percentage in this group of species (21%), but its relatively high abundance guarantees seeds for propagation and conservation. Some endangered species (e.g. *Arabis margaritae*, *Armeria fiilicaulis* subsp. *trevenqueana*, *Artemisia* 

umbelliformis or Moehringia fontqueri) have the advantage of a high germination percentage, which may compensate the scant seed material available. Members from the Geraniaceae, Labiatae and Papilionaceae may likely increase their germination percentage after coat stratification (see above), although in any case their germination may be high enough. Overall, this group of taxa with high germination percentage was dominated by members from Compositae and Cruciferae.

### High germination percentage in alpine species

The overall high germination percentage in the species here studied (15 out of the 37 taxa with above 40% germination, see above) is somewhat surprising given the generally assumed requirements of cold stratification for germination of seeds

from alpine habitats (e.g. BASKIN & BASKIN, 1998; KÖRNER, 1999). This may be explained by two non-exclusive possibilities. First, seeds from high mountain species may not require dormancy (or have dormancy requirements lower than populations form lower altitudes) given that germination will be blocked by the low temperatures registered in the site after seed dispersal, which will prevent germination until the next growing season (KÖRNER, 1999; JENSEN, 2003; FENNER & THOMPSON, 2005). Second, there is an overrepresentation of Cruciferae and Compositae species in this group of taxa, all of them with a high germination percentage that might be characteristic of the family, or at least of the genera studied here (see below), creating a possible phylogenetic bias. In any case, high germination percentages where found across families and genera (see Table 2), implying that seeds from the Mediterranean high mountain are in many cases physiologically prepared for germination just or soon after dispersal (see GIMÉNEZ-Benavides & al. (2005) for similar results with orophilous species from central Spain).

### Patterns of germination across species

The analysis of the relationship between germination percentage, speed of germination and altitude indicates some trends of ecological and management interest. First, the lack of a relationship between T<sub>50</sub> and germination percentage (Fig. 1) support the idea that the speed of germination is an intrinsic characteristic for each species. A low germination speed is therefore not indicative of low germination ability, and even seeds with a low initial germination may finally reach high percentages (e.g. BLISS & GOLD, 1999). Second, there was a weak but significant negative correlation between germination percentage and propagule mass, which could be used as a preliminary screening to plan conservation efforts (e.g. preferential selection of species with higher seed mass as those putatively showing more problems for germination). In any case, this trend was too weak to postulate generalizations, and further studies with a wider pool of alpine species are necessary to drawn conclusions (a phylogenetic effect related to the abundance of fast germinating Compositae and Cruciferae might be present; see below).

Germination patterns across species were presumably determined by phylogenetic relationship among species, at least in some cases. There are clearly some families where germination was consistently high, mostly *Compositae* and *Cruciferae*. Species of *Papilionaceae*, *Labiatae* and *Geraniaceae* showed lower germination percentages, but germination in these species is likely to be promoted with easy and fast treatments (see above). These results are of great interest for conservation plans of the Sierra Nevada, endemic and/or endangered flora: more than 30% of the whole flora and a similar percentage of the threatened flora (40 taxa of the 125 threatened

species, according to Blanca, 2002) belong to these five families. Thus, although the germination behaviour of any species may not be ascertained solely by its phylogeny and may in fact be determined by other traits related to the natural history of the species (e.g. Baskin & Baskin, 2003), the results found here support the contention that, in the alpine flora of Sierra Nevada, conservation efforts related to seed germination may be applied to members of other families if a choice must be made.

### Summary of guidelines proposed

In summary, our results demonstrate that the germination percentages of these species from Sierra Nevada vary widely but may reach high values in a considerable number of species. Taxa from *Compositae*, *Cruciferae*, *Geraniaceae*, *Labiatae* and *Papillionaceae* are prone to easy germination. Major efforts should be addressed to those species which, in addition to low germination percentages, have a critical conservation status (critically endangered, endangered, or vulnerable, according IUCN 2001 categories, see Table 1), which in this set of 37 taxa are mainly *Aquilegia vulgaris* subsp. *nevadensis*, *Arenaria nevadensis*, *Carex camposii*, *Gentiana lutea*, *Linaria glacialis*, *Narcissus nevadensis*, *Odontites granatensis*, *Papaver lapeyrousianum* and *Sorbus hybrida*, most of them endemic of the area.

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### References

ALBERT, M. J., J. M. IRIONDO & F. PÉREZ-GARCÍA (2002). Effects of temperature and pretreatments on seed germination of nine semi-arid species form NE Spain. *Israel J. Pl. Sci.* 50: 103-112.

ANGOSTO, T. & A. J. MATILLA (1993). Germination, seed-coat structure and protein patterns of seeds from Adenocarpus decorticans and Astragalus granatensis growing at different altitudes. *Seed Sci. Technol.* 21: 317-326.

APARICIO, A. & R. GUISANDE (1997). Replenishment of the endangered *Echinospartum algibicum* (Genisteae, Fabaceae) from the soil seed bank. *Biol. Conservation* 81: 267-273.

Bañares, A., G. Blanca, J. Güemes, J. C. Moreno & S. Ortiz (ed.) (2003). *Atlas y libro rojo de la flora vascular amenazada de España*. Ciutat.

BASKIN, C. C. & J. H. BASKIN (1998). Seeds - Ecology, biogeography and evolution of dormancy and germination. Academic Press.

BASKIN, C. C. & J. H. BASKIN (2003). Biogeography, and phylogenetic relationships of seed dormancy. *In:* SMITH, R. D., J. B. DICKIE, S. H. LININGTON, H. W. PRITCHARD & R. J. PROBERT (ed.), *Seed conservation: turning science into practice:* 519-545. The Royal Botanic Gardens, Kew.

BLANCA, G. (ed.) (2002). Flora amenazada y endémica de Sierra Nevada. Consejeria de Medio Ambiente, Junta de Andalucía.

BLANCA, G., B. CABEZUDO, E. HERNÁNDEZ-BERMEJO, C. M. HERRERA, J. MOLERO-MESA, J. MUÑOZ, & B. VALDÉS (1999). Libro Rojo de la Flora Silvestre Amenazada de Andalucía. Tomo I: Especies en Peligro de Extinción. Consejeria de Medio Ambiente, Junta de Andalucía.

BLANCA, G., M. CUETO, M. J. MARTÍNEZ-LIROLA, & J. MOLERO (1998). Threatened vascular flora of Sierra Nevada (Southern Spain). *Biol. Conservation* 85: 269-285.

BLISS, L. C. (1971). Arctic and alpine plant life cycles. *Annual Rev. Ecol. Syst.* 2: 405-438.

BLISS, L. C. & W. G. GOLD (1999). Vascular plant reproduction, establishment, and growth and the effects of cryptogamic crusts within a polar desert ecosystem, Devon Island, N.W.T., Canada. *Canad. J. Bot.* 77: 623-636.

BLONDEL, J. & J. Aronson (1999). *Biology and wildlife of the Mediterranean region*. Oxford university Press.

CABEZUDO, B., S. TALAVERA, G. BLANCA, C. SALAZAR, M. CUETO, B. VALDÉS, E. HERNÁNDEZ-BERMEJO & C. M. HERRERA (2005). *Lista roja de la flora vascular de Andalucía*. Consejeria de Medio Ambiente, Junta de Andalucía.

CERABOLINI, B., R. DE ANDREIS, R. M. CERIANI, S. PIERCE & B. RAIMONDI (2004). Seed germination and conservation of endangered species from the Italian Alps: Physoplexis comosa and Primula glaucescens. *Biol. Conservation* 117: 351-356.

Domínguez, F., D. Galicia, J. L. Moreno-Rivero, J. C. Moreno-Sáiz & H. Saínz-Ollero (1996). Threatened plants in Peninsular and Balearic Spain: a report based on the EU Habitats Directive. *Biol. Conservation* 76: 123-133.

ESCUDERO, A. & J. M. IRIONDO (2003). Restauración de poblaciones de especies amenazadas. *In:* REY, J. M., T. ESPIGARES & J. M. NICOLAU (ed.), *Restauración de ecosistemas mediterráneos:* 113-139. Universidad de Alcalá.

Fenner, M. & K. Thompson (2005). *The ecology of seeds*. Cambridge University Press.

FINNERTY, T. L., J. M. ZAJICEK & M. A. HUSSEY (1992). Use of seed priming to bypass stratification requirements of three Aquilegia species. *HortScience* 27: 310-313.

GASTON, K. J. (1994). Rarity. Chapman & Hall.

GIMÉNEZ-BENAVIDES, L., A. ESCUDERO & F. PÉREZ-GARCÍA (2005). Seed germination of high mountain Mediterranean species: altitudinal, interpopulation and interannual variability. *Ecol. Res.* 20: 433-444

GÓMEZ, A., A. PALACIOS, M. RAMOS, L. M. TANARRO, L. SCHULTE & F. SALVADOR (2001). Location of Permafrost in Marginal Regions: Corral del Veleta, Sierra Nevada, Spain. *Permafrost & Periglacial Process* 12: 93-110.

GÓMEZ, A. (2002). Geomorphological map of Sierra Nevada; glacial a periglacial geomorphology. Consejería de Medio Ambiente. Junta de Andalucía.

GONZÁLEZ-BENITO, M. E., M. J. ALBERT, J. M. IRIONDO, F. VARELA & F. PÉREZ-GARCÍA (2004). Seed germination of four thyme species after short-term storage at low temperatures at several moisture contents. *Seed Sci. Technol.* 32: 247-254.

GREUTER, W. (1991). Botanical diversity, endemism, rarity, and extinction in the Mediaterranean area: an analysis based on the published volumes of Med-Checklist. *Bot. Chron.* 10: 63-79.

Hennenberg, K. J. & H. Bruelheide (2003). Ecological investigations on the northern distribution range of Hippocrepis comosa L. in Germany. *Pl. Ecol.* 166: 167-188.

HEYWOOD, V. (1995). The mediterranean flora in the context of world biodiversity. *Ecol. Medit.* 21: 11-18.

IBAÑEZ, A. N. & C. B. PASSERA (1997). Factors affecting the germination of albaida (Anthyllis cytisoides L.), a forage legume of the Mediterranean coast. *J. Arid Environm.* 35: 225-231.

IUCN (2001). *IUCN Red List Categories and Criteria: Version* 3.1. IUCN Council.

JACQUEMYN, H., R. BRYS & M. HERMY (2001). Within and between plant variation in seed number, seed mass and germinability of Primula elatior: effect of population size. *Pl. Biol. (Stuttgart)* 3: 561-568.

JENSEN, M. (2003). Effect of seed maturity and pretreatment on dormancy and germination of Sorbus mougeotii seeds. *Scand. J. Forest Res.* 18: 479-486.

KÖRNER, C. (1999). Alpine plant life. Functional plant ecology of high mountains ecosystems. Springer.

LÓPEZ, J., J. A. DEVESA, T. RUIZ & A. ORTEGA-OLIVENCIA (1999). Seed germination in Genisteae (Fabaceae) from South-West Spain. *Phyton (Horn)* 39: 107-129.

LORITE, J., F. B. NAVARRO & F. VALLE (2006). Estimation of threatened orophytic flora and priority of its conservation in the Baetic range (S. Spain). *Pl. Biosyst.* 141: 1-14.

MCKEE, J. & A. J. RICHARDS (1998). The effect of temperature on reproduction in five Primula species. *Ann. Bot. (Oxford)* 82: 359-374.

MÉDAIL, F. & P. QUÉZEL (1999). Biodiversity hotspots in the Mediterranean Basin: setting global conservation priorities. *Conservation Biol.* 13: 1510-1513.

Meisert, A. (2002). Physical dormancy in Geraniaceae seeds. *Seed Sci. Res.* 12: 121-128.

Moreno-Sáiz, J. C., F. Domínguez & H. Saínz-Ollero (2003). Recent progress in conservation of threatened Spanish vascular flora: a critical review. *Biol. Conservation* 113: 419-431.

Peñas, J., F. Pérez-García & J. F. Mota (2005). Patterns of endemic plants and biogeography of the Baetic high mountains (south Spain). *Acta Bot. Gallica* 152: 247-360.

RAINA, R., M. C. BEHERA, R. CHAND & Y. SHARMA (2003). Reproductive biology of Gentiana kurroo Royle. *Curr. Sci.* 85: 667-670.

SANER, M. A., D. R. CLEMENTS, M. R. HALL, D. J. DOOHAN & C. W. CROMPTON (1995). The biology of Canadian weeds. 105. Linaria vulgaris Mill. *Canadian J. Pl. Sci.* 75: 525-537.

SCHÜTZ, W. & G. RAVE (1999). The effect of cold stratification and light on the seed germination of temperate sedges (Carex) from various habitats and implications for regenerative strategies. *Pl. Ecol.* 144: 215-230.

SCHWIENBACHER, E. & B. ERSCHBAMER (2002). Longevity of seeds in a glacier foreland of the Central Alps – a burial experiment. *Bull. Geobot. Inst. E. T. H.* 68: 63-71.

SHIMONO, A. & I. WASHITANI (2004). Seedling emergence patterns and dormancy/germination physiology of Primula modesta in a subalpine region. *Ecol. Res.* 19: 541-551.

Specht, C. E. & E. R. J. Keller (1997). Temperature requirements for seed germination in species of the genus Allium L. *Genet. Resources Crop Evol.* 44: 509-517.

STÖCKLIN, J. & E. BÄUMLER (1996). Seed rain, seedling establishment and clonal growth strategies on a glacier foreland. *J. Veg. Sci.* 7: 45-56.

TAKOS, I. A. & G. S. EFTHIMIOU (2003). Germination results on dormant seeds of fifteen tree species autumn sown in a northern Greek nursery. *Silvae Genet*. 52: 67-71.

Thompson, J. D. (2005). *Plant evolution in the Mediterranean*. Oxford University Press.

WATANABE, A., K. GOKA, & I.WASHITANI (2003). Effects of population spatial structure on the quantity and quality of seeds set by Primula sieboldii (Primulaceae). *Pl. Spec. Biol.* 18: 107-121.

WIERZBICKA, M. & J. OBIDZINSKA (1998). The effect of lead on seed imbibition and germination in different plant species. *Pl. Sci.* (*Elsevier*) 137: 155-171.

YAGIHASHI, T., M. HAYASHIDA & T. MIYAMOTO (1998). Effects of bird ingestion on seed germination of Sorbus commixta. *Oecologia* 114: 209-212.