DOI: 10.37317/ pbss-2019-0009

Parvin Salehi Shanjani<sup>1</sup>\*, Amir Mousavi<sup>2</sup>, Faeze Rasoulzadeh Farsad<sup>3</sup>

<sup>1</sup> Research Institute of Forests and Rangelands, Agricultural Research, Education and Extension Organization, P.O. Box 13185-116, Tehran, Iran; <sup>2</sup> National Institute of Genetic Engineering and Biotechnology (NIGEB), Tehran, Iran; <sup>3</sup> Islamic Azad Universities, Tehran, Iran; Correspondent author e-mail: psalehi1@gmail.com

## SEED GERMINATION PLASTICITY OF TWO ENDANGERED SPECIES OF *FERULA* IN THE CONTEXT OF CLIMATE CHANGE

## ABSTRACT

*Ferula assa-foetida* and *F. gummosa*, Apiaceae, are important endemic and endangered medicinal plants. Survival of the species is threatened by climate change, overexploiting (as source of oleo-gum resin and forage) and lack of organized cultivation. Cultivation of these valuable medicinal plants is restricted by insufficient domestication knowledge. Germination characteristics of different populations of *Ferula* taxa were studied with the aim of describing and comparing their responses to continuous cold stratification condition. Germination cues for the species were complex, with dormancy mechanisms present to restrict germination until cold stratification are fulfilled. Results indicated that a period of 4 weeks of stratification is sufficient for germination of *F. assa-foetida*, but optimal germination of *F. gummosa* require stratification for periods of 8 weeks. Both species were able to germinate at very low temperatures (4°C). Within-taxon differences in dormancy breaking and seedling emergence may interpret as local adaptations. The continued regeneration and propagation of the species in the wild will depend on the temperature and moisture status of the soil during winter and the maintenance of conditions suitable for stratification for an appropriate length of time.

Key words Dormancy, global warming, Iran, highland, local adaptation

## INTRODUCTION

Seed germination is a critical stage in the life cycle of plants, particularly when considering the effects of global warming on high-altitude species. This is due to the dependence of these species on specific temperature regimes to stimulate germination and ensure seedling development coincides with favorable growing conditions (Mondoni *et al.* 2008, 2011; Milbau *et al.* 2009). The germination emergence stage is a high-risk phase of the plant life cycle, and there-

Communicated by Grzegorz Żurek

fore seed-based research can be useful in identifying species at risk of extinction from climate change, i.e. species with a narrow germination niche in terms of temperature range and/or stratification requirement (Cochrane *et al.* 2011; Walck *et al.* 2011). Information of this type provides a link between environmental change and the mechanisms that control population processes (Ooi *et al.* 2009; Cochrane *et al.* 2011; Walck *et al.* 2011; Ooi 2012), and can thus help to improve the accuracy of models predicting plant response to climate change (Ooi 2012).

*Ferula assa-foetida* and *F. gummosa*, Apiaceae, are important endemic and endangered medicinal plants. The taxa are monocarpic, herbaceous and perennials spread at altitudes of 1500–2500 m, with an average annual precipitation of 350–700 mm of Iran (Safaian and Shokri 1993; Mozaffarian 1996; Ivan 2007; Amiri and Joharchi 2016). Recently, survival of the species is threatened by climate change, overexploiting (as source of oleo-gum resin and forage) and lack of organized cultivation. Cultivation of these valuable medicinal plants is restricted by edaphic and climatic factors, low percentage of seed set and seasonal dormancy, and insufficient domestication knowledge (Golmohammadi *et al.* 2016).

According to Baskin and Baskin (2014), linear embryos in the Apiaceae are under-developed, and seeds have morphological dormancy (MD) or morphophysiological dormancy (MPD). Normally, seeds with MD only need suitable temperature, moisture, oxygen, and of course time to germinate (Baskin and Baskin 2014). However, in many cases, the fully differentiated under-developed embryos also have physiological dormancy (PD), which imposes an additional constraint to germination; such embryos do not germinate in less than one month in suitable germination conditions. In this case, the dormancy is not just morphological but morphophysiological (MPD), and the embryos require additional treatment, such as cold, to complete their growth. Previous studies have classified F. gummosa, and F. asafetida as having deep morphophysiological dormancy, since cold stratification had been suggested as the main dormancybreaking treatment (Otroshi et al. 2009; Rouhi et al. 2012). Formation of deep MPD seems to be an adaptation to regions with a very cold winter and a dry, cool summer. In these areas, temporary sporadic favorable temperatures (elevated temperature) in winter or too early in winter are threatening for seedling establishment. Therefore, the dormancy helps seeds to remain ungerminated throughout the winter. Moreover, the low temperatures alleviate dormancy and once dormancy breaks, two possible scenarios might occur: nondormant seeds either wait for a mild and moist spring to germinate, or they germinate at low temperatures in the middle of winter in cold soil, even covered with heavy snow, until late winter; while the shoots grow and emerge above the soil surface with the increase in temperature (Baskin and Baskin 2014).

Different dormancy breaking and germination stimulating treatments have been tried with seeds of many species of Apiaceae (Baskin and Baskin 1991; Baskin *et al.* 1992, 1995, 1999, 2000; Nadjafi *et al.* 2006; Amooaghaie 2009; Nowruzian *et al.*, 2016; Fasih and Tavakkol Afshari 2018). Results of different treatments including various levels of gibberellic acid, HNO3, chilling and soaking with water at different temperatures showed that moist-chilling and gibberellic acid treatments seem the most promising in *Ferula* species. The best treatments for *F. assa-foetida* was moist-chilling for 4 weeks at  $5 \pm 1$  °C or for 2 weeks of moist-chilling (at  $5 \pm 1$  °C) followed by soaking GA3 (10 mgL<sup>-1</sup>) solution for 24 h (Nowruzian *et al.*, 2016). In similar way treatment of moist-chilling for 6 weeks or 4 weeks followed by 500 ppm gibberellic acid is recommended for *F. ovina* (Fasih and Tavakkol Afshari 2018). Washing and chilling (5±1C) for a period of 14 days was most effective in breaking dormancy in *F. gummosa* (Nadjafi *et al.* 2006).

According field observations of authors, cold stratification causes embryos to complete growth and germinate in the middle of winter in cold soil or covered with heavy snow. Shoots grow and emerge above the soil surface following increasing of temperature in early spring. Therefore aims of the present work were stimulating the cold stratification (the treatment that occurs in nature), study of dormancy termination time and seedling growth in *F. assa-foetida*, *F. gummosa*.

Differences of the present work with earlier are in unlimited cold stratification duration for dormancy breaking; and exposing moist chilling condition for seedling growth. Moreover, studied differences and similarities among closely related taxa in order to increase understanding of adaptations and changes in seed dormancy and germination preferences. One difficulty when comparing seed dormancy and germination between taxa is the intra-taxon variation. Variation within a taxon may depend on genetic differences, local weather during growth of mother plants and maturation of seeds, seed position on the mother plant, soil quality, or other naturally occurring factors. To be able to draw conclusions on a general level, for example for modeling or predicting changes in emergence pattern following climate change, knowledge about a taxon, including its variation, is needed. Therefore for investigation of the impact of the habitat variability, germination characteristics among different populations of F. assa-foetida and F. gummosa were studied under continuous moist chilling conditions. Information about germination can also improve the success rate of using seed for rehabilitation, which is critical to restoration of the high altitude rangelands.

#### MATERIALS AND METHODS

Seed material of 23 accessions or populations of the two *Ferula* taxa from all over Iran were obtained from Natural Resources Gene Bank, Iran (Table 1).

For each accession 150 seeds were sterilized with 70% ethyl alcohol for five minutes, and then washed with distilled water. Three replicates (50 seeds per replicate) of sterilized seed were placed in Petri dishes on double Whatman papers (TP). For protection against moulds, the water used to moisten the seed samples and substrata contained 0.002% Binomial fungicide. The samples were immediately transferred into a germinator at  $4\pm1$ °C and 12/12 h light (400 lux)/ dark for 60 days. Once the seeds started to germination, the number of germinated seeds were recorded every two days until the end of the experiment (two months). The length of roots and shoots of 10 randomly-selected seedlings

from each replicate were measured in 30 days seedlings. After measuring shoot and root lengths, the caryopses were cut from the seedlings and fresh seedling weight of each replicate was recorded. The seedlings were then placed in an oven at 80°C for 24 hours, after which the dry weight of each replicate was recorded as a percentage of the fresh weight. The vigor index measures seedling performance, relating together the germination percentage and growth of seedlings produced after a given time (Abdul-Baki and Anderson 1973).

e 1

Province	Population	Code	Latitude (decimal)	Longitude (decimals)	Altitude (m above sea level)	Mean annual precipitation (mm)
			F. assa-foetida	a		
Horm	Bandar Abbas	FaBandarA1	28.17	56.83	2200	178
Horm	Bandar Abbas	FaBandarA2	27.88	50.22	1845	179
Khor	Boshroye	FaBoshroye	33.96	57.17	893	94
Horm	Haji Abad	FaHajiAbad	28.94	56.46	1900	179
Esfah	Kashan	FaKashan	33.75	51.48	1800	137
Kerm	Kerman	FaKerman	30.09	57.76	2300	133
Fars	Lar	FaLar	27.46	54.39	2000	200
Yazd	Mehriz	FaMehriz	33.36	57.34	1565	84
Yazd	Tabas	FaTabas1	33.39	57.26	1536	56
Yazd	Tabas	FaTabas2	31.52	54.32	2090	56
Yazd	Taft	FaTaft	31.66	54.18	2122	60
Kerm	Zarand	FaZarand	30.88	56.88	2300	47
			F. gummosa			
K&B	Dena	FgDena	30.50	51.72	2560	760
Elam	Elam1	FgEelam	33.63	46.41	1000	575
Horm	Haji Abad	FgHajiAbad	28.12	56.84	2200	178
Ch B	Lordegan	FgLordegan	31.42	51.26	2683	555
Semn	Shahrod	FgShahrod	35.87	56.65	950	139
Yazd	Tabas	FgTabas	33.36	57.34	1565	84
Yazd	Taft	FgTaft	31.56	54.16	2439	60
K&B	Yasuj	FgYasujl	30.48	51.79	2300	855
K&B	Yasuj	FgYasuj2	31.94	51.44	1950	855
K&B	Yasuj	FgYasuj3	30.45	51.65	2420	855
Kerm	Zarand	FgZarand	30.88	56.87	2400	47

Some details of the studied wild Ferula populations

Horm - Hormozgan; Khor - Khorasan; Esfah - Esfahan; Kerm - Kerman; K&B - Kohkeluye and Boyerahman; Ch B - Charmahal Bakhtiali; Semn - Semnan

#### Data analysis

Variance analysis (ANOVA) were conducted for seed germination traits including dormancy termination, germination period, germination percentage, germination rate, germination index, seed vigor index, radicle length [mm], shoot length [mm], seedling length [mm], radicle/shoot length ratio, seedling fresh weight [mg], seedling dry weight [mg], seedling dry matter %; and seed morphology traits including seed weight [g], seed length [mm], seed width [mm], and 1000 seeds weight [mg] using the SAS9 software (SAS Institute Inc). To assess the relationships among the 13 different traits Pearson's correlation coefficient was analyzed using statistical analysis system software (SAS version 9.1, SAS Institute, 2001). The standardized morphological data were employed to calculate the Euclidean distances among the 23 *Ferula* populations by NTSYS-pc version 2.1 (Rohlf, 2002). Moreover, unweighted pair group methods of arithmetic mean (UPGMA) algorithm and SAHN clustering were utilized to get the genetic relationships. The Principal component analysis (PCA) of 23 *Ferula* populations was determined by Minitab software (version 15).

# RESULTS

Seeds length, width and weight of *F. assa-foetida* (in length: 8-15 mm; in width: 4-7.7 mm; in weight: 9-23 mg) and *F. gummosa* (in length: 9-15 mm; in width: 6.5-10 mm; in weight: 7.7-32 mg) ranged among populations of each species (Table 2). ANOVA suggested significant differences among wild populations of *Ferula* species for the seed traits. A relatively high CV was obtained for seed weight (Table 2).

Table 2

Mean comparisons of seed morph characteristics of 23 populations of *Ferula assa-foetida* (with prefix Fa) and *F. gummosa* (with prefix Fg) constant cold stratification. Different letters indicate significant differences among different populations for the same species. P <0.05

Population	Seed [1	weight ng]	Seed [n	length nm]	Seed [r	l width nm]
			F. assa-foetida			
FaBandA1	12.23	c-f	9.73	f	5.47	с
FaBandA2	13.57	cd	9.30	f	5.67	с
FaBoshro	13.00	cde	15.17	а	7.65	а
FaHajiAb	8.90	f	7.68	g	4.12	d
FaKashan	16.93	b	11.77	bcd	6.97	b
FaKerman	23.00	а	12.21	bc	6.95	b
FaLar	15.23	bc	12.40	b	5.93	с
FaMehriz	9.23	f	11.23	cd	5.77	с
FaTabas1	10.33	def	10.07	ef	5.93	с
FaTabas2	10.13	ef	10.98	ed	5.87	с
FaTaft	10.33	def	11.82	bcd	6.80	b
FaZarand	17.77	b	12.08	bc	6.72	b
Mean	13.37		11.19		6.15	
Cv	43.86		16.95		16.6	

			Continued			
Population	Seed [1	weight mg]	Seed [n	length nm]	Seed [n	width nm]
			F. gummosa			
FgDena	20.07	bc	12.60	b	7.30	bc
FgEelam	15.00	d	12.53	b	6.40	ef
FgHajiAb	14.70	d	10.72	cd	5.97	f
FgLordeg	32.17	а	14.75	а	7.60	b
FgShahro	22.20	b	12.75	b	10.08	а
FgTabas	7.70	e	9.92	d	6.50	def
FgTaft	6.17	e	8.87	e	5.32	g
FgYasuj1	19.77	bc	12.87	b	7.10	bcd
FgYasuj2	22.10	b	13.33	b	7.18	bc
FgYasuj3	17.00	cd	12.43	b	6.40	ef
FgZarand	15.57	d	11.13	с	6.75	ced
Mean	17.49		11.99		6.96	
Cv	37.51		17.10		16.36	

Both species F. assa-foetida and F. gummosa, failed to germinate without prior stratification. However, cold stratification stimulated the germination and growth of seedlings of both species. ANOVA suggested significant differences among wild populations of Ferula species for all the seed germination traits. A relatively high CV was obtained for germination period, germination rate, seed vigor index, seedling fresh weight and seedling dry weight; moderate to low values of CV were obtained for the remaining traits (Table 3). Comparison of means verified that the duration of dormancy termination was significantly longer in F. gummosa (ranged from 31-51 days, with average 42 days) than F. assa-foetida (ranged from 12-28 days, with average 19 days) (Table 3; Fig. 1). Different populations of F. assafoetida species had the significantly higher germination period, germination percentage, germination rate, germination index, seed vigor index and radicle length values (Table 3). In the species F. assa-foetida the highest germination characteristics (germination percentage, rate and index) were obtained in the population Fa-Tabas1 and the highest seedling parameters (radicle and shoot length, and seedling fresh weight) were observed in the populations FaTaft and FaZarand; however, these two populations showed lowest values of seedling dry matter percentage. In the species F. gummosa the highest germination characteristics (germination percentage, rate and index) were obtained in the populations FgLordegan and FgTaft and the highest seedling parameters (radicle and shoot length, and seedling fresh weight and seedling dry matter percent) were obtained in the population FgYasuj2 (Table 3). Populations FgYa-suj1, 2, 3 of the species F. gummosa, with similar habitat and geographical range, have markedly different dormancy and germination characters (Table 3). Variation within a taxon may depend on genetic differences, local weather during growth of mother plants and maturation of seeds, seed position on the mother plant, soil quality, or other naturally occurring factors.

Population	Domanc <sup>i</sup> [d	y terminati [ays]	on Germinat [dɛ	ion period tys]	Germi [%	nation 6]	Germiné	ttion rate	Germinat	tion Index	Seed vig	gor index	Radicl [n	e length nm]
						F. assa	1-foetida							
FaBandA1	14.33	р	10	ab	50.67	cd	8.427	bc	478.2	cd	42.9	abc	28.13	в
FaBandA2	16.33	cd	16.67	ab	70.67	abc	6.300	cde	472.3	cd	32.4	a-d	16.33	abc
FaBoshro	23	ab	10.67	ab	26.67	de	2.797	ed	220.4	de	14.9	dc	18.52	abc
FaHajiAb	13.67	q	9.33	ab	46.67	cde	7.337	bcd	433.6	cd	17.3	bcd	11.17	c
FaKashan	21.67	bc	20.67	а	66.67	abc	7.950	bcd	562.7	bc	46.8	ba	13.20	bc
FaKerman	28.33	а	20.67	а	58.67	abc	4.273	cde	390.6	cd	33.6	a-d	23.43	abc
FaLar	28.33	а	20.67	ŋ	20.00	о	1.343	e	122.9	υ	6.5	q	11.92	c
FaMehriz	26.33	ab	16.00	ab	62.67	abc	5.860	cde	481.8	cd	26.4	a-d	14.20	bc
FaTabas1	13.00	q	10.00	ab	86.67	а	15.190	а	824.5	а	39.8	abc	16.90	abc
FaTabas2	12.33	q	6.67	þ	80.00	ab	14.430	а	771.6	ab	42.9	abc	21.10	abc
FaTaft	12.33	q	8.00	þ	62.67	abc	11.630	ab	607.1	abc	53.4	а	22.40	abc
FaZaran	17.00	cd	13.33	ab	53.33	bcd	7.817	bcd	486.7	cd	46.4	abc	25.73	ab
Mean	18.89		13.56		57.11		7.780		487.7		33.6		18.59	
Cv	16.93		45.19		26.36		36.000		29.1		48.3		37.60	

Parvin	Salehi	Shanjani	et al.
		~	

$\mathbf{c}$
e
[q]
$T_{a}$

ē	
.=	
Ξ	
Ξ	
~	
$\mathbf{U}$	

Population	Dormancy [d	v termination ays]	1 Germinat [dź	tion period ays]	Germi [ <sup>9</sup>	ination %]	Germina	ution rate	Germinat	ion Index	Seed vig	or index	Radicle [m	: length m]
						F. gun	nmosa							
FgDena	45.00	bcd	7.33	cb	20.00	ა	0.763	ы	91.8	p	13.28	ef	22.99	cb
FgEelam	45.00	bcd	8.67	cb	30.67	c	1.133	de	138.5	p	12.98	ef	8.817	р
FgHajiAb	48.33	bc	4.00	c	69.33	ab	2.347	cd	295.8	c	29.25	cd	13.87	cd
FgLordeg	38.33	de	11.33	cb	78.67	53	3.940	ab	439.1	ab	69.25	а	23.13	cb
FgShahro	51.67	ab	4.67	c	29.33	c	0.823	e	106.7	p	13.03	ef	14.20	cd
FgTabas	35.00	e	9.33	cb	65.33	ab	4.110	ab	417.2	abc	33.72	cb	15.80	bcd
FgTaft	32.33	e	14.00	þ	72.00	ab	5.080	в	489.6	53	25.48	cde	11.60	р
FgYasujl	33.00	ы	22.67	а	70.67	ab	2.767	bc	322.4	þc	43.42	þ	25.20	þ
FgYasuj2	56.33	а	2.667	c	16.00	c	0.253	e	36.0	p	16.93	def	42.10	ы
FgYasuj3	43.00	cd	10.00	cb	18.67	c	0.800	e	93.3	p	6.27	f	9.67	p
FgZarand	31.67	Ð	14.67	þ	56.00	q	3.577	bc	359.6	abc	26.12	cde	13.80	cd
Mean	41.79		9.94		47.88		2.330		253.6		26.34		18.29	
Cv	8.72		45.04		24.69		32.93		28.1		28.83		30.16	

					ں ت	ntinued						
opulation	Shoot [m	: length 1m]	Seedlir [n	ıg length 1m]	Radic lengt	le/shoot h ratio	Seedling 1 [r	fresh weight ng]	Seedling [r	dry weight ng]	Seedling [9	dry matter %]
					F. as.	sa-foetida						
3andA1	46.45	abc	74.58	ba	0.613	ab	34.67	ab	2.333	þ	6.72	q
3andA2	29.73	cd	46.07	bcd	0.573	ab	20.73	ab	2.167	þ	5.70	q
3 oshro	37.95	bcd	56.47	a-d	0.503	abc	19.50	ab	2.157	þ	11.84	þ
HajiAb	29.49	cd	40.66	dc	0.397	abc	22.69	ab	7.623	а	5.80	q
Kashan	55.03	ab	68.23	abc	0.253	c	34.00	ab	3.167	þ	9.49	v
Kerman	34.07	bcd	57.50	a-d	0.700	53	34.33	ab	2.833	Ą	8.40	c
Lar	19.36	q	31.28	q	0.627	ab	17.98	þ	2.987	Ą	16.57	5
Mehriz	28.23	cd	42.43	dc	0.510	abc	25.26	ab	1.333	Ą	6.20	q
<b>Fabas1</b>	27.57	cd	44.47	bcd	0.600	ab	13.80	þ	1.233	þ	9.83	c
Fabas2	31.00	cd	52.10	bcd	0.653	ab	12.73	þ	1.000	Ą	8.15	c
Γaft	60.97	в	83.37	в	0.357	bc	29.13	ab	1.833	Ą	6.49	q
Zaran	61.03	а	86.77	а	0.460	abc	40.63	а	2.400	þ	6.07	q
can	38.41		56.99		0.520		25.46		2.590		15.35	
	30.20		28.53		30.360		44.94		60.570		14.36	

$\mathbf{c}$	
O,	
P	
ੁਕ	

P	
e	
n	
n	
÷	
g	
•	
$\Box$	

Population	Shoot [n	t length am]	Seedlin [n	ig length im]	Radicle length	≥/shoot ⊨ratio	Seedling f [n	resh weight ng]	Seedling [r	dry weight ng]	Seedling [9	dry matter 6]
					F. gu	mmosa						
FgDena	42.69	Ą	65.68	q	0.537	ab	36.40	þ	4.067	v	11.53	bcd
FgEelam	36.44	cb	45.26	cq	0.260	þ	17.31	cd	1.577	q	9.33	cde
FgHajiAb	28.40	c	42.27	р	0.540	ab	26.87	cb	1.167	q	4.35	f
FgLordeg	66.33	to	89.47	а	0.347	þ	68.23	ъ	7.567	ъ	11.11	cd
FgShahro	28.59	c	42.78	р	0.483	ab	11.06	q	1.263	q	11.97	cb
FgTabas	36.03	cb	51.83	bcd	0.440	ab	17.90	cd	1.633	q	7.26	Ð
FgTaft	24.13	c	35.73	р	0.483	ab	17.20	cd	1.300	q	7.51	Ð
FgYasujl	36.27	cb	61.47	cb	0.697	а	36.87	þ	3.267	v	8.98	de
FgYasuj2	62.19	53	104.3	а	0.673	а	18.71	cd	5.980	Ą	31.03	а
FgYasuj3	35.81	cb	45.47	cd	0.273	q	21.86	cd	3.167	c	14.20	þ
FgZarand	32.43	cb	46.23	cd	0.457	ab	19.33	cd	1.433	d	7.893	Ð
Mean	39.03		57.32		0.470		26.52		2.950		11.38	
Cv	17.17		16.72		32.750		28.44		27.740		13.34	



Fig. 1. Comparison of germination percentage of different populations of *Ferula assa-foetida* (a; with prefix Fa) and *F. gummosa* (b; with prefix Fg) under constant cold stratification

Using Pearson's correlation, an analysis was done to assess the relationship among the germination and seedling traits. It is useful to determine the relationship among the traits since this information will be useful in the utilization of the germplasm as well in the collection of the germplasm based on the target traits. The correlations among measured traits are shown in Table 4. Dormancy termination, as the most important trait, was positively and significantly correlated with important germination characters including germination percentage, germination rate and seedling vigor index; and negatively correlated with all seed morphological characters. Germination percentage exhibited a positive and significant correlation with germination rate, germination index and seedling vigor index (Table 4; Fig. 2). The correlation analysis indicated that some phenotypic traits had significant correlation ( $p \le 0.05$ ) with climate factors (Table 5). The mean annual precipitation had positive correlation with dormancy termination time, seed weight and length; but showed negative correlation with the germination percentage, germination rate, and germination index. Seedling fresh weight had positive correlation with altitude. There was also positive correlation between seed width and latitude. The above correlations implied that the mean annual precipitation plays an important role in influencing the dormancy and germination traits of the Ferula taxa.

Characters	1.	2.	З.	4	5.	6.	7.	%	9.	10.	11.	12.	13.	14.	15.	16.
1. Dormancy termination	1.000															
2. Germination period	-0.346	1.000														
3. Germination%	-0.497*	0.246	1.000													
4. Germination rate	-0.83**	0.013	$0.678^{**}$	1.000												
5. Germination Index	-0.79**	0.166	0.863**	.944**	1.000											
6. Seed vigor index	-0.45*	0.178	0.778**	0.593**	**607.	1.000										
7. Radicle length	0.066	-0.188	-0.066	0.044	-0.034	0.335	1.000									
8. Shoot length	0.026	-0.172	-0.025	0.064	0.029	0.554**	0.634**	1.000								
9. Seedling length	0.044	-0.195	-0.044	0.063	0.007	0.521*	$0.840^{**}$	0.952**	1.000							
10. Radicle/shoot length ratio	-0.077	0.130	0.100	0.083	0.059	-0.016	0.529**	-0.281	0.012	1.000						
11. Seedling fresh weight	-0.013	0.244	0.224	-0.053	0.067	0.638**	0.340	0.649**	0.589**	-0.149	1.000					
12. Seedling dry weight	0.139	-0.036	-0.216	-0.228	-0.242	0.085	0.327	0.450*	0.445*	-0.128	.537**	1.000				
13. Seedling dry matter%	-0.101	-0.040	-0.194	-0.075	-0.143	-0.274	0.032	-0.092	-0.052	0.052	-0.169	0.533**	1.000			
14. Seed weight	0.506*	0.053	-0.222	-0.464*	-0.430*	0.170	0.397	$0.481^{*}$	0.494*	0.019	0.623**	0.503*	-0.025	1.000		
15. Seed length	0.432*	-0.005	-0.405	-0.446*	-0.478*	-0.005	0.318	0.437*	0.432*	-0.050	0.353	0.173	-0.318	0.700**	1.000	
16. Seed width	$0.511^{*}$	-0.148	-0.293	-0.397	-0410	-010	0 2 7 3	0 7 QR	7070	-0.043	0126	-0.059	208	0.640**	0 731**	1 000

Parvin Salehi Shanjani et al.



Fig. 2. Comparison of dormancy termination with germination rate (a) and with 1000 seeds weight (b) of different populations of *Ferula assa-foetida* (with prefix Fa) and *F. gummosa* (with prefix Fg)

char	acteristics	within differ	ent populat	ions of Feru	<i>la</i> taxa and	some ecolog	ical parame	ters
Parameters	Dormancy termination	Germination period	Germination	Germination rate	Germination Index	Seed vigor index	Radicle length	Shoot length
Latitude	0.200	-0.218	-0.017	0.032	0.037	-0.002	-0.084	0.140
Longitude	-0.321	-0.028	0.203	0.286	0.307	0.052	-0.004	-0.265
Altitude	-0.028	0.195	0.224	0.037	0.116	0.361	0.275	0.264
Mean annual precipitation	0.61**	-0.112	-0.45*	-0.6**	-0.6**	-0.224	0.319	0.274
	Seedling length	Radicle/ shoot length ratio	Seedling fresh weight	Seedling dry weight	Seedling dry matter [[%	Seed weight	Seed length	Seed width
Latitude	0.065	-0.328	-0.218	-0.238	-0.345	0.016	0.343	0.574**
Longitude	-0.187	0.331	-0.157	-0.271	-0.247	-0.315	-0.263	-0.057
Altitude	0.294	0.131	0.56**	0.330	-0.079	0.262	-0.106	-0.263

Pearson correlation analyses for the relationship between seed (germination and morph) characteristics within different populations of *Ferula* taxa and some ecological parameters

\*: significant at 0.05 level; \*\*: significant at 0.01 level

The Euclidean distances matrix was subjected to agglomerative hierarchical clustering utilizing UPGMA method to construct a dendrogram (Fig. 3). 23 populations of the *Ferula* taxa were classified into two main groups. Cluster I consisted of 10 populations of *F. gummosa* and 4 populations of *F. assa-foetida*; cluster II included eight populations of *F. assa-foetida* and only one population of *F. gummosa* (Fig. 3). Comparison of means of two clusters indicated that populations in cluster I have significantly higher dormancy termination time, germination period and seed weight, however populations cluster II showed higher germination percentage, germination rate, germination index, seed vigor index and seedling length (Table 6). UPGMA trees of seed germination andmorphological characters partially separated the two species, a behavior also supported by PCA plot (Fig. 4). However, almost within each species cluster, the populations differed somewhat from each other and were joined together with different distances.

Therefore, there was no obvious relationship between phenotypic traits and the origin of these *Ferula* populations. PCA analysis of seed germination and morphological data revealed that the first 4 components comprise about 77% of total variance (Table 7). The first component accounted for 34.4% of the total variation in the data set while the second and third principal components contributed 21.2% and 14.4%, respectively. Together, these three components could explain 68% of the total variation in the characterized the *Ferula* populations. Analysis of the factor loadings of the characters in the retained PCs indicated that any of seed germination and morphological traits showed positive loadings in PC 1-3 (Table 7).



Fig. 3. Dendrograms of the 23 populations of *Ferula assa-foetida* (with prefix Fa) and *F. gummosa* (with prefix Fg) based on studied traits



Fig. 4. Scatter diagram of the 23 populations of *Ferula assa-foetida* (with prefix Fa) and *F. gummosa* (with prefix Fg) based on studied traits

## Mean comparisons of seed (germination and morph) characteristics of populations that separated in two clusters of Fig. 4. Different letters indicate significant differences among different populations for the same species (P ≤0.05)

Group	Dormancy termination [days]	Germination period [days]	Germination [%]	Germination rate	Germination Index	Seed vigor index	Radicle length [mm]	Shoot length [mm]
Ι	36.85a	12.40a	46.53b	2.78b	274.18b	22.80b	17.55a	34.14b
II	17.28b	11.67a	66.70a	9.38a	570.14a	42.43a	19.45a	43.71a
	Seedling	Radicle/	Seedling fresh weight	Seedling dry weigh	Seedling dry matter	Seed weight	Seed length	Seed width
	[mm]	ratio	[mg]	[mg]	[%]	[mg]	[mm]	[mm]
I	[mm] 51.69b	0.52a	[mg] 23.18b	[mg] 2.38a	[%] 10.78b	[mg] 18.26a	[mm] 12.17a	[mm] 6.87a

Factor loadings (eigenvectors) for the different seed characteristics of the <i>Ferula</i> populations for the principal components retained										
Variable	PC1	PC2	PC3	PC4						
Dormancy termination	-0.295	-0.267	0.048	0.174	-					
Germination period	0.068	0.105	-0.028	0.204						
Germination [[%	0.236	0.24	0.271	0.177						
Germination rate	0.308	0.268	0.101	-0.133						
Germination Index	0.31	0.285	0.154	0.01						
Seed vigor index	0.323	-0.03	0.312	0.197						
Radicle length	0.105	-0.3	0.363	-0.345						
Shoot length	0.214	-0.392	0.1	0.075						
Seedling length	0.192	-0.393	0.214	-0.084						
Radicle/shoot length ratio	-0.056	0.089	0.403	-0.438						
Seedling fresh weight	0.172	-0.293	0.189	0.265						
Seedling dry weight	0.057	-0.343	-0.1	-0.233						
Seedling dry matter [%]	-0.005	-0.057	-0.296	-0.537						
Seed weight	-0.322	0.067	0.274	-0.015						
Seed length	-0.267	0.144	0.261	-0.131						
Seed width	-0.263	0.069	0.315	-0.053						
Eigenvalue	6.1832	3.8224	2.2375	1.6753						
Proportion	0.344	0.212	0.124	0.093						
Cumulative	0.344	0.556	0.68	0.773						

# DISCUSSION

Germination cues for *F. assa-foetida* and *F. gummosa* were complex, with dormancy mechanisms present to restrict germination until cold stratification or other requirements are fulfilled (Nadjafi *et al.* 2006; Amooaghaie, 2009; Nowruzian *et al.* 2016; Fasih and Tavakkol Afshari, 2018). The existence of morphophysiological dormancy (MPD) is very frequent in the Apiaceae (Baskin *et al.* 1992, 1995, 2000; Phartyal *et al.* 2009; Vandelook *et al.* 2008, 2009; Scholten *et al.* 2009; Yaqoob and Nawchoo 2015; Fasih and Tavakkol Afshari 2018). Cold stratification temperature used in this experiment (4°C) provides an adequate moist chilling treatment. The temperature is also within the range of

soil temperatures likely to be encountered in the field in high altitude Iran (Tabari and Talaee 2011; Ghasemi, 2015; Aghajanlou and Ghorbani 2016; Shirvani *et al.* 2018). This cold stratification temperature has been reported as successful in breaking dormancy in studies of alpine and high altitude species (Baskin and Baskin 2014).

Results indicated that the duration of dormancy termination was significantly longer in F. gummosa than F. assa-foetida. A period of 4 weeks of stratification is sufficient for germination of F. assa-foetida, but F. gummosa require cold stratification for periods of 8 weeks for optimal germination. The final germination percentage of Ferula taxa at present study was higher than the previous experiences (Nadjafi et al. 2006; Amooaghaie 2009, Nowruzian et al. 2016; Fasih and Tavakkol Afshari 2018), in which Ferula seeds transferred to standard germination condition following limited cold stratification treatment. Sommerville et al. (2013) by studding of several species of Australian Alps suggested species requiring stratification for periods of 8 weeks or more for optimal germination may be particularly sensitive to climate change. High altitude ecosystems are considered to be among the most sensitive to climate changes (Hughes 2003; Laurance et al. 2011), and recent declines in average snow depth have been observed in alpine and high altitude areas in both the Northern and Southern Hemispheres (Hughes 2003; Nicholls 2005; Hennessy et al. 2007; Rosenzweig et al. 2007; Amiri and Eslamian 2010). For species in Apiaceae depend on cold moist conditions (wet stratification) to break dormancy; reduced snow cover during winter may threaten the survival of these species, even if subsequent temperatures are suitable for germination (Liu et al. 2011). Although the seed of some species may be able to tolerate winter temperatures in the absence of snow, a reduction in snow cover may also mean a reduction in the amount of available water (in total precipitation in winter and spring). As the level of seed hydration plays a role in breaking seed dormancy (Hoyle et al. 2008; Walck et al. 2011; Baskin and Baskin 2014), relative drought during winter and spring may prove to be more important in limiting the germination of these species than the lack of snow cover per se (Liu et al. 2011). Results of this research also indicated significant correlation between precipitation and germination traits.

Both species were able to germinate at very low temperatures (4°C). The ability to germinate at very low temperatures has been observed in several high altitude species (Wardlaw *et al.* 1989; Sommerville *et al.* 2013). The capacity to germinate at low temperatures may provide an advantage during a short growing season by allowing germination to begin under snow banks (Meyer *et al.* 1995; Forbis and Diggle 2001; Walck and Hidayati 2004). *Aciphylla glacialis* (Apiaceae) germinated optimally at low temperatures, similar to the Asian and North American *Osmorhiza* species (Walck *et al.* 2002; Baskin *et al.* 1995; Walck and Hidayati 2004) in the same family (Apiaceae). Cold stratification response having similar effects to high altitude and alpine species: improving final germination, widening the range of temperatures for germination, decreasing germination time, and synchronizing germination by reducing variability in time to germination (Shimono and Kudo 2005).

The study species were highly variable in their dormancy and germination response to the moist chilling treatment. Variation of the dormancy termination duration parameter was significant among different populations of each species; ranging from 31 to 51 days in the *F. gummosa*, and from 12 to 28 days in the *F. assa-foetida*. Dormancy is a genetic seed characteristic, but it strongly interacts with environmental factors. Dormancy intensity depends on age, nutritive conditions and water supply of the plant, as well as the weather conditions during seed ripening (Andersson and Milberg 1998). Ecological factors, such as temperature, humidity, oxygen and light, greatly influence the seed's dormancy discontinuance among species (Podrug *et al.* 2014; Mahmoudi *et al.* 2015; Mazangi *et al.* 2016; Mirzaei Mossivand *et al.* 2018; Aghajanlou *et al.* 2018). In concordance with the researches significant correlation were found between germination characteristics (including dormancy termination) and precipitation.

The germination responses of F. assa-foetida, F. gummosa seeds was significantly affected by seed origin. Several studies have been published of attempts to interrelate the germination responses of populations of a particular species collected in different parts of its range. Haasis and Thrupp (1931) and Skordilis and Thanos (1995) working with coniferous species, and McNaughton (1966) with Typha species all reported variations in germination of different ecotypes. Lauer (1953), on the other hand, failed to distinguish notable differences between populations of Agrostemma githago and Datura stramoniam collected in various locations in Europe. The variety of observed responses to germination is expected, as high altitude environments exhibit significant spatiotemporal variability (Kaye 1997; Shimono and Kudo 2005; Noroozi et al. 2013, 2015). Even within a particular habitat, germination responses are unlikely to be consistent. For each species, germination is likely to vary between altitudes and populations. Variability in germination is an important strategy to ensure species survival in unpredictable environments, reducing the risk of exposing the entire seedling cohort to poor growing conditions (Giménez-Benavides et al. 2005; Venn, 2007; Mondoni et al. 2008). For example, in the genus Penstemon, Meyer (1995) suggests that germination of most species combines predictive mechanisms (e.g. fulfillment of cold stratification requirements) with the potential for development of a persistent seed bank.

### CONCLUSIONS

Cold stratification is the main prerequisite for breaking deep complex dormancy in *F. assa-foetida* and *F. gummosa*. A period of 4 weeks of stratification is sufficient for germination of *F. assa-foetida*, but *F. gummosa* require stratification for periods of 8 weeks for optimal germination. Both species were able to germinate at very low temperatures (4°C). The characteristics of deep MPD in the taxa are part of the plant's adaptation to its environment. Highly significant intraspecific population differences in the germination parameters of the taxa might reflect local adaptation to a particular environment. Pronounced differences occurred within both *F. assa-foetida* and *F. gummosa*, even though the some studied sites in each taxon were adjacent sites. Variation within a taxon may depend on genetic differences, local weather during growth of mother plants and maturation of seeds, seed position on the mother plant, soil quality, or other naturally occurring factors. To be able to draw conclusions on a general level, for example for modelling or predicting changes in emergence pattern following climate change, knowledge about a taxon, including its variation, is needed. Therefore, studies of germination behavior should include several populations from the same species.

The continued regeneration of the species in the wild will depend on the temperature and moisture status of the soil during winter and the maintenance of conditions suitable for stratification for an appropriate length of time. In this context temperature is a critical driver of plant regeneration, directly influencing seed dormancy, germination and vegetative reproduction. Therefore changing climate not only affect the dormancy and germination traits, but also is likely to impact on the germination response of these species through maternal effects on the developing seed. These species could be targeted for conservation in *ex situ* collections, whilst monitoring their response in the field.

### ACKNOWLEDGEMENTS

This work was supported by the Agricultural Research, Education and Extension Organization, and Research Institute of Forests and Rangelands (RIFR), Iran; Project no. 14-09-09-9354-93198.

#### REFERENCES

- Abdul Baki A. A., Anderson J. D. 1973. Vigor determinations in soybean seed multiple criteria. Crop Sci. 13: 630-633.
- Aghajanlou F., Ghorbani A. 2016. A study on the effects of some environmental factors on the distribution of *Ferula gummosa* and *Ferula ovina* in Shilander mountainous rangelands of Zanjan. *Iran J. Rangeland.* 9: 407-419.
- Aghajanlou F., Ghorbani A., Zare Chahoki M. A., Hashemi Majd K., Mostafazadeh R. 2018. The impact of environmental factors on distribution of *Ferula ovina* (Boiss.) Boiss. in northwest Iran. *Appl. Ecol. Envi*ron. Res. 16: 977-992.
- Amiri M. S., Joharchi M. R. 2016. Ethnobotanical knowledge of Apiaceae family in Iran: A review. Avicenna J. Phytomed. 6: 621-635.
- Amiri M. J., Eslamian S. 2010. Investigation of Climate Change in Iran. J. Environ. Sci. Technol. 3: 208-216. doi:10.3923/jest.2010.208.216
- Amooaghaie R. 2009. The Effect Mechanism of Moist-Chilling and GA on Seed Germination and Subsequent Seedling Growth of *Ferula ovina* Boiss. Open Plant Sci. J. 3: 22-28.
- Andersson L., Milberg P. 1998. Variation in seed dormancy among mother plants, populations and years of seed collection. Seed Sci. Res. 8: 29–38.
- Baskin C. C., Baskin J. M. 1991. Non-deep complex morphophysiological dormancy in seeds of Osmorhiza claytonii (Apiaceae). Amer. J. Bot. 78: 588-593.

Baskin C., Baskin J. M. 2014. Seeds: Ecology, Biogeography, and Evolution of Dormancy and Germination. Academic Press, San Diego: Elsevier/Academic Press.

Baskin C. C., Baskin J. M., Chester E. W. 1999. Seed dormancy in the wetland winter annual *Ptilianium nuttalli* (Apiaceae). Wetland. 19: 23-29.

Baskin C. C., Chester E. W., Baskin J. M. 1992. Deep complex morphophysiological dormancy in seeds of *Thaspium pinnatifidum* (Apiaceae). Int. J. Plant Sci. 153: 565–571.

Baskin C. C., Meyer S. E., Baskin J. M. 1995. Two types of morphophysiological dormancy in seeds of two genera (*Osmorhiza* and *Erythronium*) with an arctotertiary distribution pattern. Amer. J. Bot. 82: 293– 298. Baskin C. C., Milberg P., Andersson L., Baskin J. M. 2000. Deep complex morphophysiological dormancy in seeds of *Anthriscus sylvestris* (Apiaceae). *Flora*. 195: 245–251.

Cochrane A., Daws M. I., Hay F. R. 2011. Seed-based approach for identifying flora at risk from climate warming. Austral. Ecol. 36: 923–935.

- Fasih M., Tavakkol Afshari R. 2018. The morphophysiological dormancy of *Ferula ovina* seeds is alleviated by low temperature and hydrogen peroxide. Seed Sci. Res. 28: 52 –62. doi:10.1017/ S0960258517000356.
- Forbis T. A., Diggle P. K. 2001. Subnivean embryo development in the alpine herb Caltha leptosepala (Ranunculaceae). Can. J. Bot. 79: 635–642.
- Ghasemi A. R. 2015. Changes and trends in maximum, minimum and mean temperature series in Iran. Atmos. Sci. Lett. 16: 366–372.

Giménez-Benavides L., Escudero A., Pérez-Garcia F. 2005. Seed germination of high mountain Mediterranean species: altitudinal, interpopulation and interannual variability. Ecol. Res. 20: 433–444.

- Golmohammadi F., Ghoreyshi S. E., Parvaneh H. 2016. Ferula assa-foetida as a main medical plant in east of Iran (harvesting, main characteristics and economical importance). Int. J. Farm. Alli. Sci. 5-6: 453-475.
- Haasis F. W., Thrupp A. C. 1931. Temperature relations of lodgepole pine seed germination. *Ecology*. 12: 728 -744.
- Hennessy K., Fitzharris B., Bates B. C., Harvey N., Howden S. M., Hughes L., Salinger J., Warrick, R. 2007. Australia and New Zealand. In: M. L. Parry, O. F. Canziani, J. P. Palutikof, P. J. van der Linden, C. E. Hanson (Eds.) Climate change: impacts, adaptation and vulnerability. Contribution of working group II to the fourth assessment report of the intergovernmental panel on climate change, (pp. 507–540). Cambridge: Cambridge University Press.
- Hoyle G. L., Daws M. I., Steadman K. J., Adkins S. W. 2008. Mimicking a semi-arid tropical environment achieves dormancy alleviation for seeds of Australian native Goodeniaceae and Asteraceae. Ann. Bot. 101: 701–708.

Hughes L. 2003. Climate change and Australia: trends, projections and impacts. Aust. Ecol. 28: 423-443.

- Ivan A. R. 2007. Ferula assafoetida. Med. Plant World. 3: 223-234.
- Kaye T. N. 1997. Seed dormancy in high elevation plants: implications for ecology and restoration. In: T. N. Kaye, A. Liston, R. M. Love, D. L. Luoma, R. J. Meinke, M. V. W ilson (Eds.) Conservation and management of native plants and fungi, (pp. 115–120). Eugene, OR: Native Plant Society of Oregon.
- Lauer E. 1953. Über die Keimtemperatur von Ackerun-kräutern und deren Einfluß auf die Zusammensetzung von Unkrautgesellschaften. Flora. 140: 551-595.
- Laurance W. F., Dell B., Turton S. M., Lawes M. J., Hutley L. B., McCallum H., Dale P., Bird M., Hardy G., Prideaux G., Gawne B., McMahon C. R., Yu R., Hero J. M., Schwarzkopf L., Krockenberger A., Douglas M., Silvester E., Mahony M., Vella K., Saikia U., Wahren C. H., Xu Z., Smith B., Cocklin C. 2011. The 10 Australian ecosystems most vulnerable to tipping points. Biol. Conserv. 144: 1472–1480.
  Liu K., Baskin J. M., Baskin C. C., Bu H., Liu M., Liu W., Du G. 2011. Effect of storage conditions on germination of seeds of 489 species from high elevation grasslands of the eastern Tibet Plateau and some
- Liu K., Baskin J. M., Baskin C. C., Bu H., Liu M., Liu W., Du G. 2011. Effect of storage conditions on germination of seeds of 489 species from high elevation grasslands of the eastern Tibet Plateau and some implications for climate change. Amer. J. Bot. 98: 12–19.
- Mahmoudi J., Mahdavi S. Kh., Mansouri B. 2015. Examination of effect of topography (elevation and aspect) on distribution of medicinal plant *Ferula gummosa* case study: rangelands of Khombi and Saraii Germeh city in Khorasan Shomali Province. Bull. Environ. Pharmacol. Life Sci. 4: 108-113.
- Mazangi A., Ejtehadi H., Mirshamsi O., Ghassemzadeh F., Hosseinian Yousefkhani S. S. 2016. Effects of climate change on the distribution of endemic *Ferula xylorhachis* Rech.f. (Apiaceae: Scandiceae) in Iran: Predictions from ecological niche models. Russ. J. Ecol. 47: 349-354, doi:10.1134/ s1067413616040123.
- McNaughton S. J. 1966. Ecotype functions in the Typha community-type. Ecol. Monographs. 36: 297–325.
- Meyer S. E., Kitchen S. G., Carlson S. L. 1995. Seed germination timing patterns in intermountain *Penstemon* (Scrophulariaceae). Amer. J. Bot. 82: 377–389.
- Milbau A., Graae B. J., Shevtsova A., Nijs I. 2009. Effects of a warmer climate on seed germination in the subarctic. Ann. Bot. 104: 287–296.
- Mirzaei Mossivand A., Ghorbani A., Zare Chahoki M. A., Keivan Behjou F., Sefidi K. 2018. Compare the environment factors affecting the distribution of species *Prangos ferulacea* and *Prangos pabularia* in rangelands of Ardabil Province. *Iran. J. Range Desert Res.* 25: 200-210.
- Mondoni A., Probert R., Rossi G., Hay F., Bonomi C. 2008. Habitat-correlated seed germination behavior in populations of wood anemone (*Anemone nemerosa* L.) from northern Italy. Seed Sci. Res. 18: 213–222.
- Mondoni A., Probert R. J., Rossi G., Vegini E., Hay F. R. 2011. Seeds of alpine plants are short lived: implications for long-term conservation. Ann. Bot. 107: 171–179.
- Mozaffarian V. 1996. A dictionary of Iranian plant names: Latin, English, Persian: Farhang Mo'aser, 505p.
- Nadjafi F., Bannayan M., Tabrizi L., Rastgoo M. 2006. Seed germination and dormancy breaking techniques for *Ferula gummosa* and *Teucrium polium*. J. Arid Environ. 64: 542–547.

Nicholls N. 2005. Climate variability, climate change and the Australian snow season. Austral. Meteorol. Magazne. 54: 177–185.

Noroozi J., Akhani H., Breckle S. W. 2013. Biodiversity and phytogeography of the alpine flora of Iran. Biodivers. Conserv. doi:10.1007/s10531-007-9246-7.

- Noroozi J., Dietmar M., Franz E. 2015. Diversity, distribution, ecology and description rates of alpine endemic plant species from Iranian mountains. Alp. Bot. 126. doi:10.1007/s00035-015-0160-4.
- Nowruzian A., Masoumian M., Ebrahimi M., Bakhshi Khaniki G. 2016. Effect of Breaking Dormancy Treatments on Germination of *Ferula assafoetida* L. Iran. J. Seed Res. 3: 155-169, doi:10.29252/yujs.3.2.155.
   Ooi M. K. J., Auld T. D., Denham A. J. 2009. Climate change and bet-hedging: interactions between in-
- creased soil temperatures and seed bank persistence. Glob. Change Biol. 15: 2375–2386. Ooi M. K. J. 2012. Seed bank persistence and climate change. Seed Sci. Res. 22: S53–S60.
- Otroshi N. Zamani A., Khodambashi M., Ebrahimi M., Struik P. C. 2009. Effect of exogenous hormones and chilling on dormancy breaking of seeds of asafoetida (*Ferula assafoetida* L.). J. Seed Sci. 21: 9–15.
- Phartyal S. S., Kondo T., Baskin J. M., Baskin C. C. 2009. Temperature requirements differ for the two stages of seed dormancy break in *Aegopodium podagraria* (Apiaceae), a species with deep complex morphophysiological dormancy. Amer. J. Bot. 96: 1086–1095.
- physiological dormancy. Amer. J. Bot. 96: 1086–1095.
   Podrug A., Gadžo D., Muminović Š., Grahić J., Srebrović E., Đikić M. 2014. Dormancy and germination of johnsongrass seed (Sorghum halepense L.). Herbologia. 14(2):1-10, doi: 10.5644/Herb.14.2.01.
- Rohlf F. 2002. NTSYS-pc: Numerical Taxonomy and Multivariate Analysis System (2.1 Ed.), Department of Ecology and Evolution, State University of NY, Stony Brook.
- Rosenzweig C., Casassa G., Karoly D. J., Imeson A., Liu L., Menzel A., Rawlins S., Root T. L., Seguin B., Tryjanowski P. 2007. Assessment of observed changes and responses in natural and managed systems. In: M. L. Parry, O. F. Canziani, J. P. Palutikof, P. J. van der Linden, C. E. Hanson, (Eds.), Climate change 2007: impacts, adaptation and vulnerability. Contribution of working group II to the fourth assessment report of the intergovernmental panel on climate change. (pp. 79–131). Cambridge: Cambridge University Press.
- Rouhi H. R., Rahmati H., Saman M., Shahbodaghloo A. R., Karimi F. A., Moosavi S. A., Rezaei M. E., Karimi F. 2012. The effects of different treatments on dormancy-breaking of Galbanum seeds (*Ferula gummosa* Boiss). Int. J. Agric. Sci. 27: 598–604.
- Safaian N. and Shokri M. 1993. Botanical and ecological study of species of the genus *Ferula* (Medicinal Plants) in Mazandaran province. Acta Hort. 333: 159-167.
- SAS Institute. 2001. SAS InstituteSAS/Stat user's guide, Version 9.1 SAS Institute, Cary, NC, USA.
- Scholten M., Donahue J., Shaw N. L., Serpe M. D. 2009. Environmental regulation of dormancy loss in seeds of *Lomatium dissectum* (Apiaceae). Ann. Bot. 103: 1091–1101.
- Shimono Y., Kudo G. 2005. Comparisons of germination traits of alpine plants between fellfield and snowbed habitats. Ecol. Res. 20: 189–197.
- Shirvani A., Moradi-Choghamarani F., Zand-Parsa S., Moosavi A. A. 2018. Analysis of long-term trends in air and soil temperature in a semi-arid region in Iran. Environ Earth Sci. 77: 173-177.
- Skordilis A., Thanos C. 1995. Seed stratification and germination strategy in the Mediterranean pines *Pinus brutia* and *P. halepensis*. Seed Sci. Res. 5: 151–160.
- Sommerville K. D., Martyn A. J., Offord C. A. 2013. Can seed characteristics or species distribution be used to predict the stratification requirements of herbs in the Australian Alps? Bot. J. Linn. Soc. 172: 187– 204.
- Tabari H., Talaee P. H. 2011. Analysis of trends in temperature data in arid and semi-arid regions of Iran. Glob. Planet Change. 79: 1–10.
- Vandelook F., Bolle N., Van Assche J. A. 2008. Seasonal dormancy cycles in the biennial *Torilis japonica* (Apiaceae), a species with morphophysiological dormancy. Seed Sci. Res. 18: 161–171.
- Vandelook F., Bolle N., Van Assche J. A. 2009. Morphological and physiological dormancy in seeds of Aegopodium podagraria (Apiaceae) broken successively during cold stratification. Seed Sci. Res. 19: 115– 123.
- Venn S. E. 2007. Plant recruitment across alpine summits in south-eastern Australia. DPhil Thesis, LaTrobe University.
- Walck J. L., Hidayati S. N., Dixon K. W., Thompson K., Poschlod P. 2011. Climate change and plant regeneration from seed. Glob. Change Biol. 17: 2145–2161.
- Walck J. L., Hidayati S. N., Okagami N. 2002. Seed germination ecophysiology of the Asian species Osmorhiza aristata (Apiaceae): comparison with its North American congeners and implications for evolution of types of dormancy. Amer. J. Bot. 89: 829–835.
- Walck J. L., Hidayati S. N. 2004. Germination ecophysiology of the western North American species Osmorhiza depauperata (Apiaceae): implications of preadaptation and phylogenetic niche conservatism in seed dormancy evolution. Seed Sci. Res. 14: 387–394.
- Wardlaw I. F., Moncur M. W., Totterdell C. J. 1989. The growth and development of *Caltha introloba* F. Muell. II. The regulation of germination, growth and photosynthesis by temperature. Austral. J. Bot. 37: 291–303.
- Yaqoob U., Nawchoo I. A. 2015. Conservation and cultivation of *Ferula jaeschkeana* Vatke: a species with deep complex morphophysiological dormancy. Proceedings of the National Academy of Sciences, India Section B: Biological, 1–11.