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Effect of cold stratification treatments on germination of drought tolerant shrubs seeds

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Abstract: *This study was carried out to determine effects of different durations (20, 40, 60 days and control) of cold stratification treatments on seed germination and to overcome dormancy in seeds of twelve different drought tolerant plants. The species used in this study were Arbutus andrachne L., Cistus creticus L., Colutea armena Bois. Huet., Cotinus coggygria Scop., Cotoneaster numullaria Fisch. and Mey., Elaeagnus angustifolia L., Jasminum fruticans L., Paliurus spina-christii Mill., Punica granatum L., Pyracantha coccinea Roem., Rhus coriaria L., and Ziziphus jujuba Mill. The seeds were sown in polyethylene pots under greenhouse and on seedbeds under open field conditions. Statistical design was a randomized complete block design with three replications. Germinated seeds were counted and observed periodically for 90 days after sowing to determine germination percentages (GP) and germination rates (GR). While the highest GP were determined according to duration of cold stratification under greenhouse conditions for each species were 44.2% for 60 days, 2.5% for 20 days, 85.6% for 60 days, 13.5% for 20 and 60 days, 64.3% for 20 days, 11.2% for 60 days, 8.2% for 20 days and 14.1% for 20 days for Cotinus coggygria, Cotoneaster numullaria, Jasminum fruticans, Paliurus spina-christii, Pyracantha coccinea, Punica granatum, Rhus coriaria and Ziziphus jujuba respectively. The highest GP were 16.5% for Arbutus andrachne and 91.3% for Colutea armena for 20 days cold stratified seeds and sown under open field conditions. Furthermore, the highest GP were obtained from seeds of Cistus creticus (27.5%) and Elaeagnus angustifolia (56.2%) in control sowing*

Key words*: Cold stratification, Drought tolerant, Shrub, Germination percentage*

Introduction

Seeds of many woody plant species can not germinate even if they are sown in proper moisture, oxygen and soil conditions (Urgenc and Cepel, 2001). These seed dormancies are based on different sources. Baskin and Baskin (2004) have classified the types of seed dormancy as physiological, morphological, morpho-physiological, physical and combination dormancies. Some of the biological reasons for dormancy, listed by ISTA (1966, 1993) are hard and impermeable seed coat, immature or dormant embryo, absence of endosperm and fleshy part of fruit. The degree of seed dormancy varies both among and within species. Poulsen (1996) reported by ascribing to Wolf and Kamondo (1993) that dormancy among, and within, seed lots of the same species varies with provenance, crop year and individual trees. There are different methods and techniques to overcome seed dormancy depending on these factors. These methods and techniques carried on different types and periods or if necessary carried out with combinations affiliated with dormancy types and degrees. For example, in general, such pretreatments like floating on hot water, mechanical or chemical scarification and hot aeration are used for seed coat dormancy while the pretreatments of cold and warm stratification are applied to dormancy caused restrictions at the embryo level (Landis *et al*., 1996).

The most important step on taking biopreventive measures for checking soil erosion is the selection of suitable stabilizing plant species taking climatic and slope conditions in consideration. The plant species developing tap root hold excess water and prevent landslides. In order to achieve effective protection in erosion control areas, the problems should be determined correctly and the required plant species should be chosen accordingly (Ucler *et al*., 2002).

Occurring in rocky and steep landscapes *Arbutus andrachne* L., *Cistus creticus* L., *Colutea armena* Bois. Huet., *Cotinus coggygria* Scop., *Cotoneaster numullaria* Fisch.and Mey., *Elaeagnus angustifolia* L., *Jasminum fruticans* L., *Paliurus spina-christii* Mill., *Punica granatum* L., *Pyracantha coccinea* Roem., *Rhus coriaria* L. and *Ziziphus jujuba* Mill. are drought tolerant plants and prevent soil erosion. These species are also effective in increasing the inhabitants' income level using their different parts such as roots, fruits and flowers. According to some researchers, there are germination obstacles in seeds of these plants and thus, there are propagation obstacles in seeds of these species and thus, there are propagation difficulties (Heit, 1967; Urgenc, 1986; Piotto *et al*., 2003). Germination percentage (GP) of the seeds of these species varies approximately between 13% and 100% and cold stratification, soaking in H_2SO_4, GA_3 (gibberellic acid) and KNO_3 (potassium nitrate) are well known methods to increase GP (Belcher and Karrfalt, 1979; Riley, 1981; Kaminski, 1985; Pela *et al*., 2000; Karam and Al Salem, 2001).

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The aim of the present study was to overcome seed dormancy, and define the effect of cold stratification treatments with different durations on GP and GR of the seeds of these plants.

Materials and Methods

Arbutus andrachne, *Cistus creticus*, *Colutea armena*, *Cotinus coggygria*, *Cotoneaster numullaria*, *Elaeagnus angustifolia*, *Jasminum fruticans*, *Paliurus spina-christii*, *Punica granatum*, *Pyracantha coccinea*, *Rhus coriaria* and *Ziziphus jujuba* ripe fruits were collected from wild plants growing in Artvin region, located in the north-eastern part of Turkey, between the altitudes of 200 and 1200 m in August, September and October 2003. The seeds were separated from the fruit material, rinsed in tap water, dried in the shade and kept at room temperature in linen sacks. After ratio of filled seed determined, the seeds were stored at ±5°C in plastic bags untill cold stratification.

The seeds were stratified by putting layers of moistened sand and seeds on top of each other. Since there was a risk for some of the seeds to be mixed with the sand because of their small size, linen cloth was placed between the sand and the seeds. The mean temperature of the room where cold stratification was applied on the seeds was 5±1°C. The moisture of the sand and the seeds were checked continuously against drying, heating, and poor aeration and this medium was moistened so that the seeds should not become moldy.

Three different cold stratification durations (20, 40 and 60 days) and control were applied on the seeds and they were sown in polyethylene pots under greenhouse and on seedbed under open field conditions in the spring (March) of 2004. Polyethylene pots were filled with growing medium composed of forest soil, creek sand and manure (1:1:1). The experimental design was a randomized complete block with three replications (30 seeds for each replication) for every treatment.

Number of germinated seeds were counted every day, but recorded for 7, 10, 14 and 21 days and in every week (7 days) after the 21 day counting. Germination percentage (GP) and germination rate (GR) were determined according to duration of cold stratification duration.

Germination rate was calculated as described by (Pieper, 1952) :

$$GR = \frac{(n1 \times t1) + (n2 \times t2) + (n3 \times t3) + ...(ni \times ti)}{T}$$

GR : Germination rate.
n : Number of days for each counting of germinated seeds.
t : Number of germinated seeds in each counting day.
T : Total number of germinated seeds.

The experiment lasted approximately for 90 days when it was observed that the seeds stopped germinating. Filled seed ratios were used to determine GP. Data from the treatments was analyzed by the SPSS version 9.0 and SAS statistical programmes. The ANOVA and Newman Keuls tests were used to compare treatment groups as to whether or not they show any statistically significance differences which was set at p<0.05. Approximate account of Satterthwaite was used to compute the differentials denominator degree of freedom to test greenhouse and open area condition (Satterthwaite, 1946; Milliken and Johnson, 1984). For evaluating data from greenhouse and open field observations and to compare greenhouse and open area, statistical model were used as below :

To analyze greenhouse and open area,

$y_{ijk} = \mu + r_i + \tau_j + rt_{ij} + e_{ijk}$ Models were used.

y_{ijk} = Observed merit of at $k.$ seed of $j.$ pretreatment of $i.$ replication;

μ = General average;

r_i = Random effect of $i.$ replication, $E(r_i) = 0$, $Var(r_i) = s_r^2$;

τ_j = Constant effect of $j.$ pretreatment, $\sum_{J=1}^{n} \tau_j = 0$;

rt_{ij} = Interaction between $i.$ replication and $j.$ treatment, $E(rt_{ij}) = 0$, $Var(rt_{ij}) = s_{rt}^2$;

e_{ijk} = Coincidental error, $E(e_{ijk}) = 0$, $Var(e_{ijk}) = s_e^2$

To compare greenhouse and open field ;

$y_{ijkl} = \mu + \alpha_k + r(a)_{i(k)} + \tau_j + \alpha\tau_{kj} + rt(a)_{ij(k)} + e_{ijkl}$ Models were used.

y_{ijkl} = Observed merit of at $l.$ seed of $j.$ treatment of $i.$ replication of $k.$ area;

α_k = Constant effect of $k.$ area; $E(a_k) = 0$, $Var(a_k) = s_a^2$;

$r(a)_{i(k)}$ = Random effect of $i.$ replication at $k.$ area; $E(r(a)_{i(k)}) = 0$, $Var(r(a)_{i(k)}) = s_{r(a)}^2$;

τ_j = Random effect of $j.$ pretreatment, $\sum_{J=1}^{n} \tau_j = 0$

$\alpha\tau_{kj}$ = Interaction between $k.$ area and $j.$ treatment, $\sum_{k=1}^{2} \sum_{J=1}^{n} \alpha\tau_{kj} = 0$

$rt(a)_{ij(k)}$ = Interaction between $i.$ replication at $k.$ area and $j.$ treatment, $E(rt(a)_{ij(k)}) = 0$, $Var(rt(a)_{ij(k)}) = s_{rt(a)}^2$;

e_{ijkl} = Random error, $E = (e_{ijkl}) = 0$, $Var(e_{ijkl}) = s_e^2$

Table - 1: Germination rates (GR), the days beginning (BG) and ending of germination (EG) of the seeds according to durations of cold stratification

Plant species	Durations of cold stratification (day)	GR, BG and EG in the green house conditions (day)	GR, BG and EG under open field conditions (day)
Arbutus andrachne	Control	42, 34 - 44	45, 39 - 50
	20	31, 30 - 34	39, 34 - 50
	40	23, 16 - 34	43, 34 - 53
	60	42, 39 - 44	39, 39 - 39
Cistus creticus	Control	51, 44 - 68	31, 11 - 41
	20	25, 7 - 68	30, 25 - 37
	40	29, 7 - 51	26, 11 - 41
	60	26, 7 - 61	30, 20 - 44
Colutea armena	Control	37, 14 - 82	42, 14 - 82
	20	28, 7 - 82	39, 14 - 82
	40	28, 7 - 82	32, 14 - 68
	60	30, 7 - 75	46, 14 - 75
Cotinus coggygria	Control	52, 14 - 75	41, 32 - 54
	20	33, 14 - 68	42, 27 - 75
	40	28, 14 - 68	38, 32 - 48
	60	21, 14 - 44	43, 32 - 51
Cotoneaster numullaria	Control	—, — —	25, 25 - 25
	20	24, 14 - 34	30, 30 - 30
	40	24, 14 - 34	25, 25 - 25
	60	30, 30 - 30	—, — —
Elaeagnus angustifolia	Control	35, 21 - 51	42, 24 - 66
	20	30, 16 - 55	42, 28 - 73
	40	26, 14 - 39	36, 24 - 53
	60	19, 14 - 30	36, 24 - 49
Jasminum fruticans	Control	33, 21 - 44	—, — —
	20	24, 16 - 51	41, 32 - 61
	40	21, 14 - 34	38, 32 - 61
	60	19, 14 - 44	36, 32 - 61
Paliurus spina-christii	Control	35, 30 - 55	48, 37 - 56
	20	33, 16 - 51	29, 24 - 37
	40	33, 21 - 58	51, 37 - 66
	60	30, 16 - 61	41, 37 - 49
Punica granatum	Control	55, 34 - 82	—, — —
	20	52, 26 - 89	—, — —
	40	52, 26 - 82	—, — —
	60	38, 21 - 89	—, — —
Pyracantha coccinea	Control	38, 21 - 55	—, — —
	20	32, 16 - 51	—, — —
	40	34, 16 - 55	—, — —
	60	21, 14 - 51	—, — —
Rhus coriaria	Control	82, 82 - 82	27, 8 - 45
	20	43, 16 - 68	45, 45 - 45
	40	42, 16 - 75	73, 73 - 73
	60	—, — —	—, — —
Ziziphus jujuba	Control	39, 26 - 51	46, 37 - 49
	20	23, 14 - 39	48, 37 - 66
	40	22, 14 - 34	37, 37 - 37
	60	24, 16 - 47	43, 37 - 49

Table - 2: Results of statistical analyses showing the relationship of the germination percentage with different cold stratification durations (Means in column with the same letter are not significantly different at p< 0.05)

Plant species, durations of cold stratification (C*) (day) and sowing conditions (G or O**)	Number in sample (N)	Germination percentage (%)	ANOVA (F ratio)	Newman Keuls test results
C. numullaria (C, G)	90	0.00		a
C. numullaria (40, G)	90	0.00		a
C. numullaria (60, O)	90	0.00		a
J. fruticans (C, O)	90	0.00		a
R. coriaria (60, O)	90	0.00	9.83***	a
R. coriaria (60, G)	90	0.00		a
R. coriaria (40,O)	90	1.03		ab
R. coriaria (20, O)	90	1.03		ab
A. andrachne (C, G)	90	1.14		ab
C. numullaria (60, G)	90	1.25		ab
C. numullaria (40, O)	90	1.25		ab
C. numullaria (20, O)	90	1.25		ab
C. numullaria (C, O)	90	1.25		ab
A. andrachne (60, O)	90	1.37		ab
R. coriaria (C, G)	90	1.37		ab
Z. jujuba (40, O)	90	1.41		ab
R. coriaria (C, O)	90	2.06		ab
A. andrachne (60, G)	90	2.29		ab
C. numullaria (20, G)	90	2.50		ab
Z. jujuba (60, O)	90	2.81		ab
P. spina-christii (60, O)	90	3.37		abc
C. creticus (40, O)	90	4.42		abcd
J. fruticans (C, G)	90	4.44		abcd
A. andrachne (C, O)	90	4.81		abcde
C. creticus (60, O)	90	5.44		abcde
R. coriaria (40, G)	90	5.49		abcde
P. spina-christii (40, O)	90	5.61		abcde
Z. jujuba (C, O)	90	5.63		abcde
C. creticus (20, O)	90	6.12		abcde
A. andrachne (40, O)	90	6.19		abcde
P. granatum (C, G)	90	6.73		abcdef
A. andrachne (20, G)	90	6.87		abcdef
Z. jujuba (C, G)	90	7.03		abcdef
P. spina-christii (C, O)	90	7.85		abcdef
C. creticus (C, G)	90	7.94		abcdef
R. coriaria (20, G)	90	8.23		abcdef
Z. jujuba (20, O)	90	8.44		abcdef
P. spina-christii (C, G)	90	8.97		abcdef
P. spina-christii (20, O)	90	8.97		abcdef
P. granatum (40, G)	90	8.98		abcdef
P. granatum (20, G)	90	8.98		abcdef
C. coggygria (40, O)	90	9.07		abcdef
P. granatum (60, G)	90	11.22		abcdef
P. spina-christii (40, G)	90	11.23		abcdef
A. andrachne (40, G)	90	11.45		abcdef
Z. jujuba (60, G)	90	12.66		abcdefg
Z. jujuba (40, G)	90	12.66		abcdefg
P. spina-christii (60, G)	90	13.46		abcdefg
P. spina-christii (20, G)	90	13.47		abcdefg
Z. jujuba (20, G)	90	14.07		abcdefgh
C. coggygria (60, O)	90	15.87		abcdefgh
C. coggygria (C, O)	90	15.87		abcdefgh
A. andrachne (20, O)	90	16.49		abcdefghi
C. creticus (40, G)	90	18.14		abcdefghij
C. coggygria (20, O)	90	19.27		abcdefghijk
E. angustifolia (20, O)	90	21.42		abcdefghijk
C. creticus (20, G)	90	23.81		abcdefghijk
C. coggygria (C, G)	90	23.81		abcdefghijk
C. creticus (60, G)	90	24.94		abcdefghijk
E. angustifolia (40, O)	90	25.43		abcdefghijk
J. fruticans (20, G)	90	25.53		abcdefghijk
E. angustifolia (60, O)	90	26.77		abcdefghijk
C. creticus (C, O)	90	27.54		abcdefghijk
C. armena (60, O)	90	27.77		abcdefghijk
E. angustifolia (C, O)	90	28.11		abcdefghijk
P. coccinea (C, G)	90	29.24		abcdefghijkl
C. armena (40, O)	90	29.76		abcdefghijkl
C. coggygria (20, G)	90	30.61		abcdefghijkl
J. fruticans (40, G)	90	38.89		abcdefghijkl
C. coggygria (40, G)	90	39.68		abcdefghijkl
C. armena (C, O)	90	39.68		abcdefghijkl
E. angustifolia (40, G)	90	41.50		bcdefghijkl
C. armena (C, G)	90	41.66		bcdefghijkl
E. angustifolia (20, G)	90	42.84		cdefghijkl
J. fruticans (60, O)	90	43.33		defghijkl
C. armena (60, G)	90	43.65		defghijkl
C. coggygria (60, G)	90	44.21		defghijkl
P. coccinea (40, G)	90	44.45		efghijkl
C. armena (20, G)	90	45.63		fghijkl
E. angustifolia (60, G)	90	49.53		ghijkl
J. fruticans (40, O)	90	51.11		hijkl
J. fruticans (20, O)	90	53.34		ijkl
P. coccinea (60, G)	90	54.97		jkl
C. armena (40, G)	90	55.56		kl
E. angustifolia (C, G)	90	56.22		kl
P. coccinea (20, G)	90	64.32		l
J. fruticans (60, G)	90	85.55		m
C. armena (20, O)	90	91.27		m

*C: Control; ** G: Greenhouse, O:Open field, ***Significant at 95% significance level

Results and Discussion

Results showed that the seeds of *Cistus creticus*, *Colutea armena*, *Cotinus coggygria*, *Cotoneaster numullaria*, *Elaeagnus angustifolia*, *Jasminum fruticans*, *Paliurus spina-christii*, *Rhus coriaria, Ziziphus jujuba* and *Arbutus andrachne* were germinated both under greenhouse and open field conditions while *Punica granatum* and *Pyracantha coccinea* seeds were germinated only in the greenhouse conditions.

In our study, it was found that the GP and GR were better under the greenhouse condition than under open field conditions. Germinations resulted earlier in the greenhouse than under the open field. The best GR according to durations of cold stratification and sowing conditions (greenhouse or open field) can be seen in Table 1. The results indicated that the duration of cold stratification was positively effective on germination rate of the seeds (Table 1). All findings and discussions on GP and GR of each species were evaluated and summarized.

While the highest GP were determined according to duration of cold stratification under greenhouse conditions for each species were 44.2% for 60 days, 2.5% for 20 days, 85.6% for 60 days, 13.5% for 20 and 60 days, 64.3% for 20 days, 11.2% for 60 days, 8.2% for 20 days and 14.1% for 20 days for *Cotinus coggyria*, *Cotoneaster numullaria*, *Jasminum fruticans*, *Paliurus spina-christii*, *Pyracantha coccinea*, *Punica granatum*, *Rhus coriaria* and *Ziziphus jujuba* respectively. The highest GP were 16.5% for *Arbutus andrachne* and 91.3% for *Colutea armena* for 20 days cold stratified seeds and sown under open field conditions. Furthermore, the highest GP were obtained from seeds of *Cistus creticus* (27.5%) and *Elaeagnus angustifolia* (56.2%) in control sowing (Table 2).

The highest GP (16.5%) was obtained from *Arbutus andrachne* seeds stratified for 20 days and sown under under open field conditions. This ratio was lower than the results of Gultekin (2004); Karam and Al Salem (2001) and Tilki (2004) for *A. unedo*. Gultekin (2004) found 85% of germination percentage using H_2SO_4 and 86% was found using GA_3 by Karam and Al Salem (2001) for *A. andrachne* seeds. It was observed that *A. andrachne* seeds germinated while they were in stratification medium. So it can be said that the germination of the seeds in stratification medium affected GP negatively.

Cistus creticus seeds also germinated like the seeds of *A. andrachne* while they were in stratification medium by the 14 day. The highest GP (27.5%) was determined from control sowing in these seeds. Pela *et al*. (2000) obtained 96% of GP under laboratory condition from the seeds soaked in hot water (100°C) for 35-60 seconds.

Colutea L. seeds do not germinate readily unless the impermeable seed coat is ruptured by mechanical or chemical scarification. Soaking the seeds in concentrated H_2SO_4 for 30 to 60 minutes, before sowing in nursery beds, results in good germination (Dirr, 1990; Dirr and Heuser, 1987). In this study, we obtained 91.3% of GP in seeds of *C. armena* applied cold stratification for 20 days and sown under open field conditions. Some of *C. armena* seeds also germinated in stratification medium by the 20 day. As Piotto *et al*. (2003) implied, it was estimated that approximately 40% of the seeds were empty because of insect damage. Consequently these seeds were cleaned and disinfected before storage and sowing.

The highest GP (44.2%) was obtained in *Cotinus coggygria* seeds that were cold stratified for 60 days and sown under greenhouse conditions. Piotto *et al*. (2003) noticed that 60 day cold stratification soaking in H_2SO_4 treatments increased GP and decreased GR of *C. coggygria* seeds.

GP of 2.5% was determined from *Cotoneaster numullaria* seeds, cold strafied for 20 days and sown under greenhouse conditions. So it can be said that only stratification treatment is not enough to overcome seed dormancy for this species. Kaminski (1985) found 80% of GP for *C. divaricata* seeds soaked in H_2SO_4 for 30-45 minutes with cold stratification for 180 days.

It was observed that *E. angustifolia* seeds were moulded while they were in the stratification medium and the seeds were germinating while the stratification treatment was continued for 50 days. Piotto *et al*. (2003) suggested that peat should be used as a stratification medium because it provides aeration to seeds. The highest GP of 56.2% was achieved from *E. angustifolia* seeds in control sowing.

GR of *E. angustifolia* seeds sown in the greenhouse conditions showed better results (germinated in fewer days) than those sown in the open field conditions. A 60-day cold stratification (CS) pretreatment gave the shortest GR (19 days) in greenhouse conditions (Table 1).

In general, *Jasminum fruticans* is used as an ornament plant in gardens and it is propagated by cuttings (Gungor *et al*., 2002). In this study, it was found that cold stratification treatment was successful in overcoming seed dormancy of *J. fruticans*. Cold stratified seeds for 60 days showed the highest germination percentage (85.6%) in the greenhouse (Table 2). The best GR (19 days) was also obtained from the seeds that were cold stratified for 60 days. When germination rates of *J. fruticans* seeds were considered it could be said that increasing of duration of stratification improved germination rates under both greenhouse conditions and open field conditions.

The highest GP (13.5%) was obtained in *Paliurus spina christii* seeds that were cold stratified for 60 days and sown under greenhouse conditions. Takos *et al*. (2001) reported that increasing the duration of stratification resulted in a significant increase in GP of seeds of *P. spina-christii*. Piotto *et al*. (2003) implied that soaking in H_2SO_4 for 40-120 minutes could increase GP in *P. spina-christii* seeds.

Punica granatum seeds germinated only in the greenhouse. The highest GP was 11.2% in seeds that were cold stratified for 60 days. Germinations in the stratification medium can be a result of low GP for these seeds.

The highest GP was 64.3% in *Pyracantha coccinea* seeds that were cold stratified for 20 days and sown in the greenhouse conditions. Dirr and Heuser (1987) determined that stratification for 90 days treatment lowered the duration of germination and increased GP.

The degree of seedcoat hardness and embryo dormancy varies within and among seedlots for most species (Hartmann *et al*., 1997). This is true for *Rhus* species and consequently treatments to overcome seed dormancy also varies among *Rhus* species (Li *et al*., 1999). In addition, previous studies showed that the *R. coriaria* seeds have double dormancy, hardcoated seed and additional embryonic dormancy (Heit, 1967; Doussi and Thanos, 1994; Takos and Efthimiou, 2002). In this study, the best GP (8.2%) was obtained from *R. coriaria* seeds that were applied for 20 day cold stratification. As a result of our findings it can be said that only stratification method is not enough to achieve a high GP. Farmer *et al*. (1982) reported that without scarification, <5% of seeds of *R. glabra* germinated, but 3 to 4 hr of scarification in concentrate H_2SO_4 promoted an average of 58% germination. Scarification with H_2SO_4 for about 1 hr followed by cold stratification for 1 to 3 months is recommended for seeds of *R. aromatica* (Heit, 1967; Weber *et al*., 1982). Boiling water treatments were reported effective on breaking dormancy to some *Rhus* species (Li *et al*., 1999).

The highest GP was 14.1% for *Ziziphus jujuba* seeds that were 20 days strafied and sown under greenhouse conditions. Some researchers recommended scarification in H_2SO_4 for 120 to 360 minutes followed by stratification at 5°C for 60 to 90 days (Lyrene, 1979).

Overall, it can be said that there is an affirmative effect of cold stratification treatments on GP and GR of the seeds used in this study. The results also showed that the durations of cold stratification are insufficient to overcome seed dormancy of *Arbutus andrachne*, *Cotoneaster numullaria*, *Rhus coriaria* and *Ziziphus jujuba*. For that reason, the durations of cold stratification or some chemical and mechanical scarification pretreatments need to be determined according to characteristics of the species. It can also be concluded that the greenhouse conditions were more effective on GP and GR of all the species used in this study over open field conditions.

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