

Contents available at ISC and SID

Journal homepage: [www.rangeland.ir](http://www.rangeland.ir)



### Research and Full Length Article:

## Effect of Seed Priming on Enhancement of Seed Germination and Seedling Growth of Annual Sainfoin (*Onobrychis crista-galli* (L.) Lam.) in Medium and Long-term Collections of Gene Bank

Azadeh Kavandi<sup>A</sup>, Ali Ashraf Jafari<sup>B\*</sup> and Mojtaba Jafarzadeh<sup>C</sup>

<sup>A</sup> M.Sc. Graduated in Agronomy, Islamic Azad University, Saveh Branch, Iran

<sup>B</sup> Professor, Research Institute of Forests and Rangelands, Agricultural Research Education and Extension Organization (AREEO), Tehran, Iran, \*(Corresponding author), Email: [aliashrafj@gmail.com](mailto:aliashrafj@gmail.com)

<sup>C</sup> Assistant Professor, Islamic Azad University, Boroujerd Branch, Boroujerd, Iran

Received on: 13/02/2017

Accepted on: 28/06/2017

**Abstract.** Annual Sainfoin (*Onobrychis crista-galli* (L.) Lam.) is widely adapted to moderate and cold regions of Iran and naturally grows in pasture and rangelands used for forage in these areas. In order to study the effects of priming on seed germination and seedling growth in *O. crista-galli*, two factorial experiments were conducted based on a randomized complete design with three replications under laboratory and greenhouse conditions in Research Institute of Forests and Rangelands, Tehran, Iran in 2014-2015. Experimental factors were (A) five conservation methods including medium-term storage (active cold room 4°C for 15 years), long-term storage (basic cold room -18°C for 15 years), regenerated seeds (control) and deteriorated seeds using accelerated ageing techniques (40°C, 98% of RH for 48 and 72h). Levels of factor B were four priming treatments including Control (without priming), two osmopriming (PEG -0.4Mpa and -0.8Mpa), and hydropriming (imbibition with distilled water). Data were collected for germination percent, rate of germination, root length, shoot length, seedling length, vigor index, seedling weight and root/shoot length ratio (R/S). Result of laboratory showed higher mean values of most traits except root length in base store (-18°C). In both experiments, the highest root length was obtained in aged seeds. In greenhouse, higher values of many traits were obtained in osmopriming (PEG -0.4Mpa). In both experiment, higher mean values of many traits were obtained using hydropriming in seeds conserved in both base and active store. In the latter stores, highest root length, seedling length, R/S ratios were obtained by osmopriming (PEG -0.4Mpa and -0.8Mpa). To accelerate aging test, higher mean values of all the traits were obtained by osmopriming (PEG -0.4Mpa). It was concluded that osmopriming could be used as an effective method for the recovery of natural and artificial deteriorated seeds.

**Key words:** Seed deterioration, Seed priming, Seed storage, Annual sainfoin

## Introduction

Sainfoin (*Onobrychis* spp.) is a cross-pollinated legumes used for hay and forage production in Iran. So far, 162 species have been described around the world in the genus *Onobrychis*. This genus extends from the Mediterranean region to Caucasia, the Zagros Mountains of Iran and Asia. The genus is concentrated in Iran (60 species) (Rechinger, 1984) and Turkey (52 species), (Emre *et al.*, 2007 and Çelik *et al.*, 2011). Iran and Turkey appear to be the main centers of genetic diversity. This genus is often growing in conjunction with forage grasses to reduce bloat hazard as well as to improve soil fertility due to its nitrogen fixing ability (Lu *et al.*, 2000). It contains the condensed tannins which reduce its potential to produce bloat and improve protein digestion by grazing animals (Rumball and Claydon, 2005). These plants are widely adapted, especially in temperate and cold regions in Iran. They are resistance to drought and adapted to the conditions of low rainfall (Majidi and Arzani, 2004).

*Onobrychis crista-galli* with common name Cock's comb, Cock's head, and Medick vetch is an annual species belonging to section *Lophobrychis* and is distributed in Aegaea, Rhodes, Cyprus, Syria, Lebanon, Palestine, Jordan, Lower Egypt, Turkey, Iraq, Iran and North Africa (Ghanavati, 2012). It has had 16 or 32 chromosomes that are diploid or tetraploid, respectively. Diploid populations had 3-seeded pods with a spinulose first row of spines while tetraploid populations had 2-seeded pods with the first row of spines simple (Ghanavati, 2012).

Low germination due to hard seed coat in wild *Onobrychis* taxa is the major obstacle to cultivation. On the other hand, the unsynchronized and poor germination permits the survival of wild species under natural conditions. There are very few studies on germination and breaking seed

dormancy in this genus. Majidi and Barati (2011) determined seed dormancy in *Onobrychis viciifolia* Scop., *Onobrychis sintenisii* Bornm and *Onobrychis melanotricha* Boiss. and noted the beneficial effects of acid scarification to overcome the dormancy. Ramezani *et al.* (2013) in *Onobrychis viciifolia* obtained higher mean values of vigor, root length and germination rate using PEG6000 10% for 12 hours.

Many crop species and their wild relatives are preserved in gene banks around the world. The seeds are stored according to the gene bank standards (FAO, 2013); there are no specific standards for the conservation of wild plant species that grow in natural habitats. There are few reports for determining the best times of regeneration of wild species in seeds bank. FAO (2013) recommended monitoring the seeds viability every 5 or 10 years for seeds in medium- or long-term storage, respectively. However, in Iranian natural resource gene bank (Research institute forest and rangeland), there are 45000 accessions that many of them are wild species as range, forage and medicinal plant species. There are many wild accessions that were not regenerated yet preserved in Iranian natural resource gene bank over 30 years. One of the major problems in wild species germplasm is lack of knowledge of how to break dormancy and germinate the deteriorated seeds. In some cases, the deteriorated accessions failed to germinate using the same treatments and/or conditions that were found to be optimum at the start of storage (Probert, 2000). One of the methods that are often used as an invigoration treatment to ensure the seed germination is seed priming. This method is useful particularly if the seeds have already aged during storage (Butler *et al.*, 2009). It is well accepted fact that priming improves germination, reduces seedling

emergence time and improves stand (Nawaz *et al.*, 2013).

In priming, seeds are soaked in different solutions with high osmotic potential. This prevents the seeds from absorbing in enough water for radicle protrusion, thus suspending the seeds in the lag phase (Taylor *et al.*, 1998). Seed priming have an important role in increasing the yield of different crops in relation to enhance 37, 40, 70, 22, 31, 56, 50 and 20% in wheat, barley, upland rice, maize, sorghum, pearl millet, and chick pea, respectively (Harris *et al.*, 2005).

There are some seed priming techniques which are i.e. hydropriming, halopriming, osmopriming and hormonal priming (Nawaz *et al.*, 2013). Osmopriming is a commercially used technique for improving seed germination and vigor. It involves the controlled imbibition of seeds to start the initial events of germination followed by seed drying up to its original weight. Osmopriming has many advantages including rapid and uniform emergence, improved seedling growth and better stand establishment under any environmental and soil conditions (Chiu and Sung, 2002).

One of main problem in maintenance of seed in gene bank is the regeneration of aged seeds that lose their viability over times. For increasing seed germination traits, it is necessary to apply some seed dormancy breaking and seed priming treatments. Therefore, this study was conducted to use the growth regulator substances priming on seeds to increase their germination and seedling growth of wild *Onobrychis crista-galli* seeds naturally preserved in medium (active store) and long-term storage (base store) and accelerate the aged seeds of *O. crista-galli* in laboratory and greenhouse conditions.

## Materials and Methods

### Laboratory experiments

Seeds of three native accessions of *Onobrychis crista-galli* as codes of 3346

(Dehloran), 6595 (Kohdasht), and 11767 (Rodbar) were provided from the natural resource gene bank, Tehran, Iran.

Two separate factorial experiments consisting two factors: A) five conservation methods including medium-term storage (preserved in active cold room 4°C for 15 years), long-term storage (preserved in basic cold room -18°C for 15 years), regenerated seeds in open storage 22°C for 2 years (Control) and deteriorated seeds using the accelerated ageing techniques (40°C, 98% of relative humidity for 48 and 72h) and factor B) four priming treatments including Control (no priming), two osmopriming (PEG -0.4Mpa and -0.8Mpa), and hydropriming (imbibition with distilled water). The treated seeds were sterilized and transferred between sterile moist papers in Petri dishes. The Petri dishes were incubated in a +22°C germinator under light-to-dark cycle of 16 hours light (1000 lux) to 8 hours dark. Next, the germinated seeds were counted and recorded every 3 days (from beginning of germination) until no more seeds germinated.

The germination percent, root length, shoot length, seedling length, Root/Shoot length ratio (RS), and seedling fresh weight were recorded on day 21. The germination rate (Maguire, 1962) and Vigor Index (VI) (Abdul-Baki and Anderson, 1973) were calculated by Equations 1 and 2, respectively.

$$\text{Rate of germination} = \frac{n_1}{d_1} + \frac{n_2}{d_2} + \frac{n_3}{d_3} + \dots$$

(1)

$$VI = (RL + SL) \times GP \quad (2)$$

Where:

n= number of germinated seeds

d= number of days

GP= germination percent

RL= Root length

SL= Shoot length

The factorial experiments were conducted based on a Completely Randomized Design with three

replications. The experimental units were single Petri dishes.

### Greenhouse experiments

Fifteen cm diameter plastic pots were filled with sandy soil. Seeds of various treatments were sown and irrigated with tap water in the greenhouse at  $22\pm 3^{\circ}\text{C}$ . The pots were irrigated and maintained at field capacity. The number of emerged seedlings was recorded and subjected to data analysis. The experimental design was factorial design consisting of two factors as mentioned in laboratory experiment. In the greenhouse, data collection was the same as laboratory.

For a brief presentation of results, the three accessions were considered as replications; therefore, the main and interactions effects of accessions by priming and accessions by conservation methods were not included in the statistical analysis.

Data analysis was carried out using SAS software and the differences between treatment means were tested using Duncan's Multiple Range Test.

## Results and Discussion

### Laboratory experiments

Results of the analysis of variance (ANOVA) in the laboratory experiment showed significant differences among conservation methods and priming treatments for all the traits. There were significant differences between conservation by the priming method for all of traits (Table 1).

The means of traits of five conservation methods under laboratory conditions are presented in Table 2. Results showed that the highest mean values for germination percent, rate of germination, shoot length, seedling length and seedling fresh weight were obtained in the base cold room ( $-18^{\circ}\text{C}$ ). The same trend was observed for accessions with smaller values in active store ( $4^{\circ}\text{C}$ ) and regenerated seeds (control). Higher mean values of base

store ( $-18^{\circ}\text{C}$ ) were compared with active store ( $+4^{\circ}\text{C}$ ) indicating the effect of low temperature in keeping seed viability (Table 2). The seeds preserved in base store with low humidity and temperature had low metabolic activity and led to late deterioration. In contrast, in the active store, there was more traffic of Staff and opening/closing the door and also the repeated power fluctuations and humidity cause the early seed deterioration. Meanwhile, the base temperature of germination in *O. crista-galli* ( $+5^{\circ}\text{C}$ ) (Borreani *et al.*, 2003) is likely to start primary metabolite activities of the seeds in the active store. Similar to our results, Rincker (1983) found that during the 20 years of storing 37 accessions of alfalfa seeds at  $-15^{\circ}\text{C}$  with a relative humidity of 60%, the trend of germination decreases were low from 91 to 81%, whereas in the open storage conditions during 10 years, Priestley (1986) reported that the half of seeds lost their viability.

In laboratory, The highest values of vigor index, root length and R/S ratio with average values of 4.52, 3.48 cm and 3.84, respectively were obtained in the accelerated aged seeds ( $40^{\circ}\text{C}$ , 98% RH for 72h) (Table 2 and Fig.1). Accelerated aging test is used to evaluate the seed physiological potential of various species (Tekrony, 1995). The principle of this method is based on artificial accelerated aging seeds by placing seeds at high temperature and high relative humidity as environmental factors concerning the intensity and speed of aging (MacDonald, 1999). In this case, low-quality seeds will deteriorate faster than healthy seeds with higher vigor (Marshall and Lewis, 2004). The most important changes that happen in the deteriorated seeds are oxidation reactions such as the production of free radicals, dehydrogenation of enzymes and proteins, reduction of membrane permeability and increased electrolyte leakage under the influence of free radicals, changing molecular structure of

nucleic acids and reduced enzymes activities (Janmohammadi *et al.*, 2008).

In comparison of accelerated aging treatments in the laboratory, the results showed that ageing test had negative and significant effects on seed germination and seedling growth. However, in deteriorated seeds, the root length was increased (Table 3 and Fig.1). Similar to our results, Hampton *et al.* (2004) found more deterioration by increasing time of accelerated aging test in pea and Simic *et al.* (2004) found that increasing temperature of accelerated aging test in corn led to reduce both seed vigor and germination percent. Soltani *et al.* (1996) reported that seed deterioration among populations of wheat was different and each samples had particular seed storability. Reduce seedling growth as a

consequence of seed deterioration is also happened in many studies (Ellis *et al.*, 1988; Basra *et al.*, 2002).

In comparisons between priming treatments in laboratory conditions, the results showed that higher mean values for all of traits except R/S ratio were obtained in regenerated seeds. Results of priming by conservation interaction effects for all the traits under laboratory conditions are presented in Fig. 1. In all of conservation methods, higher rates of germinations were obtained using hydropriming whereas higher values of root length, seedling length, and R/S ratio were obtained in active room (4°C), and basic cold room (-18°C), and accelerated aged seeds using osmopriming treatments (PEG -0.4Mpa and -0.8Mpa) (Fig. 1).

**Table 1.** Analysis of variance and MS of germination percent, rate of germination, root length, shoot length, seedling length, vigor index, seedling weight and root/shoot length ratio in *Onobrychis crista-galli* under laboratory conditions

Source of variation	DF	Germination (%)	Germination rate	Vigor index	Root length (cm)	Shoot Length (cm)	Seedling length (cm)	R/S ratio	Fresh weight (g)
Conservation(C)	4	9896.3**	1802.5**	19.21**	13.30**	0.99**	53.92**	12.66**	1701.08**
Priming (P)	3	8637.2**	1540.7**	18.40**	29.49**	1.66**	16.47**	27.20**	1981.43**
C × P	12	562.5**	120.8**	6.65**	8.33**	0.56**	8.14**	8.93**	668.05**
Error	100	75.63	11.03	0.73	0.57	0.06	0.70	0.49	182.36
CV%		14.17	16.18	23.13	24.64	21.14	27.11	23.68	13.87

ns, \*, \*\*= non-significant and significant at P= 0.05 and 0.01 levels, respectively

**Table 2.** Means comparison of germination percent, rate of germination, root length, shoot length, seedling length, vigor index, seedling weight and root/shoot length ratio in *Onobrychis crista-galli* seeds at five conservation methods under greenhouse conditions

Conservation	Germination (%)	Germination rate	Vigor index	Root length (cm)	Shoot length (cm)	Seedling length (cm)	R/S ratio	Fresh Weight (g)
Control	70.5 b	25.10 b	3.17 b	2.28 c	1.01 b	2.20 b	2.62 c	101.05 b
Aging 48 h	57.8 d	16.50 d	4.18 a	3.14 b	0.95 b	2.33 b	3.20 b	86.76 c
Aging 72 h	24.7 e	5.72 e	4.52 a	3.58 a	0.93 b	1.24 c	3.84 a	98.31 b
Active store	65.6 c	23.24 c	3.24 b	3.19 b	1.35 a	4.48 a	2.70 c	90.27 c
Base store	79.3 a	27.81 a	3.59 b	3.20 b	1.33 a	4.49 a	2.72 c	108.47a

Means with the same letter are not significantly different (P=0.01)

**Table 3.** Means comparison of germination percent, rate of germination, root length, shoot length, seedling length, vigor index, seedling weight and root/shoot length ratio in *Onobrychis crista-galli* at four priming treatments under laboratory conditions

Priming treatments	Germination (%)	Germination rate	Vigor index	Root length (cm)	Shoot length (cm)	Seedling length (cm)	R/S ratio	Fresh weight (g)
Control	86.1 a	28.39 a	4.80 a	4.21 a	1.28 b	5.34 a	3.51 a	104.88 a
Hydropriming	74.4 b	28.19 a	2.83 c	1.67 d	1.47 a	2.44 c	1.51 c	102.85 ab
Osmo -0.4MP	55.2 c	17.86 b	3.54 b	2.94 c	0.94 c	2.59 bc	3.09 b	88.30 c
Osmo -0.8MP	42.8 d	12.47 c	3.86 b	3.65 b	0.98 c	2.84 b	3.71 a	97.72 b

Means with the same letter are not significantly different (P=0.01)

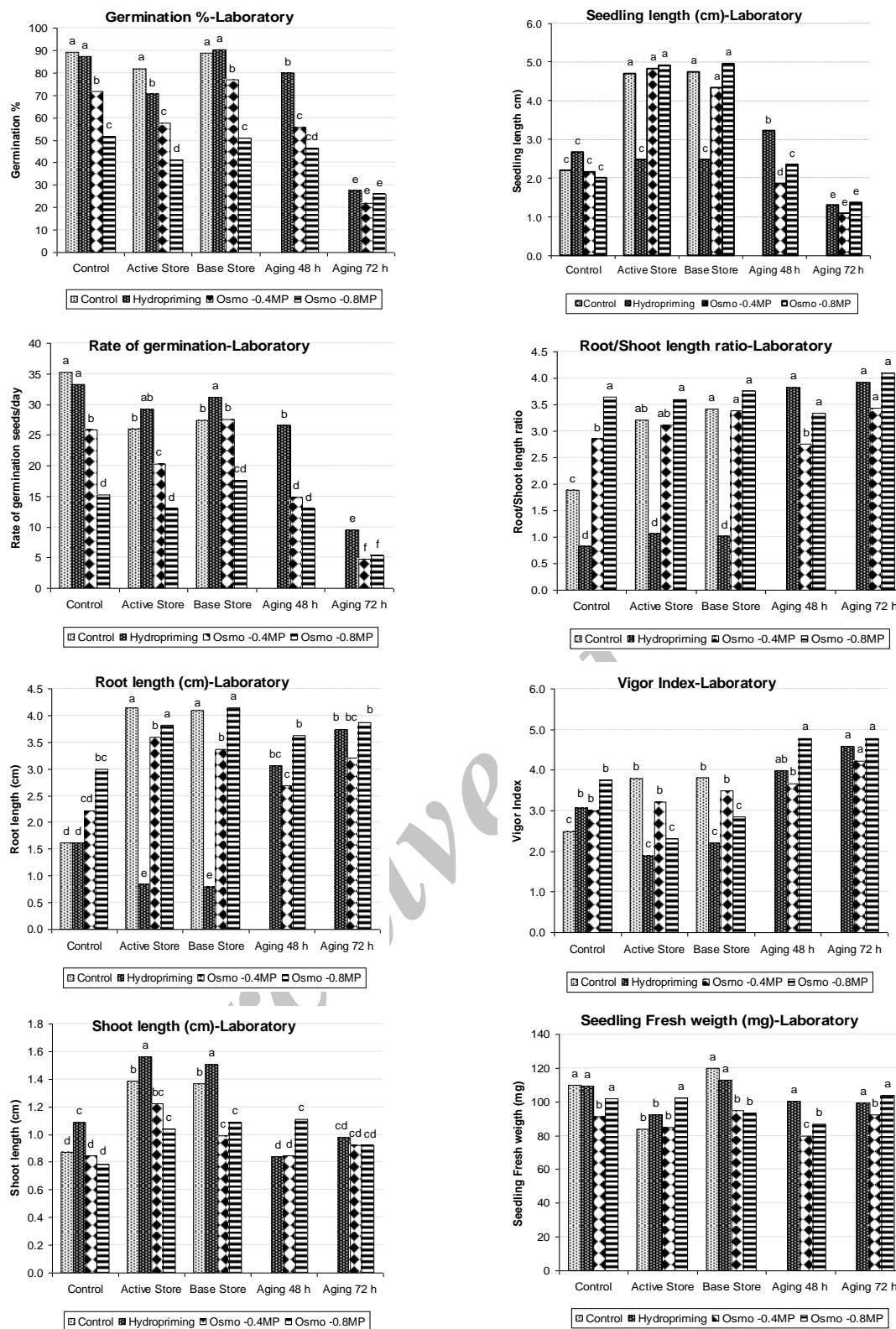


Fig. 1. Priming by conservation interaction effects for seed germination and seedling growth of *Onobrychis crista-galli* under laboratory conditions

## Greenhouse experiments

Results of the analysis of variance (ANOVA) in greenhouse experiments showed significant differences among different levels of conservation methods and priming treatments for all of the traits. There were also significant effects of conservation by priming for all the traits (Table 4).

The means of traits of five conservation methods under greenhouse conditions are presented in Table 5. Results showed that the highest mean values of germination percent, rate of germination and shoot length were obtained in active and basic cold rooms or the regenerated seeds (control). Higher mean values of root, shoot and seedling length were obtained in the accelerated aged seed at 40°C, and 98% RH for 72h (Table 5 and Fig.2). Higher values of regenerated seeds are expected since the regenerated seeds were produced during past two years and they were fresh seeds.

In comparisons between priming treatments for seedling traits in greenhouse, results showed that the hydropriming had significant effects on germination percent, rate of germination and vigor index whereas for other traits, both osmopriming (PEG -0.4Mpa and -0.8Mpa) had increased seedling growth traits (Table 6).

Result of priming by conservation interaction effects (Fig. 2) in both preservation active (4°C), and basic (-18°C) stores showed that hydropriming

had significantly increased the means of germination percent, and rate of germination. In contrast, for seedling growth indices, osmopriming (PEG -0.4Mpa) had a significant impact on the improved vigor index, root length, shoot length, seedling length, and seedling weight as compared to that for control in all of conservation methods (Fig. 2). Similar to our study, Amooaghaie (2011) showed that both osmo and hydro priming improved alfalfa seedling germination and growth as compared to that for control. Farooq *et al.* (2006) studying the effect of seed priming in rice seedling traits found higher effects of priming on root length than shoot length.

Eisvand *et al.* (2011) in carrot (*Daucus carota*) found that hydropriming improved seedling vigor index higher than that for hormonal priming. Similar to our results, El-Araby and Hegazi (2004) stated that osmopriming using PEG was to improve germination traits in tomato. Priming is much effective in dryland farming system in semi-arid regions to improve seed germination and seedling vigor (Finch-Savage *et al.*, 2004). Studies had demonstrated that in primed seeds, the performance and structure of the cell membrane are in a better stability than control seeds. In primed seeds, some biochemical and metabolic reactions improve seed germination (Bittebcourt *et al.*, 2004).

**Table 4.** Analysis of variance and MS of germination percent, rate of germination, root length, shoot length, seedling length, vigor index, seedling weight and root/shoot length ratio in *Onobrychis crista-galli* under greenhouse conditions

Source of variation	DF	Germination (%)	Germination rate	Vigor index	Root length	Shoot Length	Seedling length	R/S ratio	Dry weight
Conservation(C)	4	5514.2**	73.99**	274.93**	138.93**	20.16**	112.02**	2.76**	0.85**
Priming (P)	3	3879.7**	47.30**	119.99**	820.68**	9.06*	726.76**	10.66**	0.41**
C × P	12	2399.0**	19.14**	92.18**	184.65**	10.56**	152.07**	2.74**	0.21**
Error	100	252.51	2.08	21.45	18.55	3.20	23.03	0.17	0.05
CV%		34.60	34.30	35.83	25.10	18.33	17.94	22.53	42.45

ns, \*, \*\* = non-significant and significant at P= 0.05 and 0.01 levels, respectively

**Table 5.** Means comparison of germination percent, rate of germination, root length, shoot length, seedling length, vigor index, seedling weight and root/shoot length ratio *Onobrychis crista-galli* at five conservation methods under greenhouse conditions

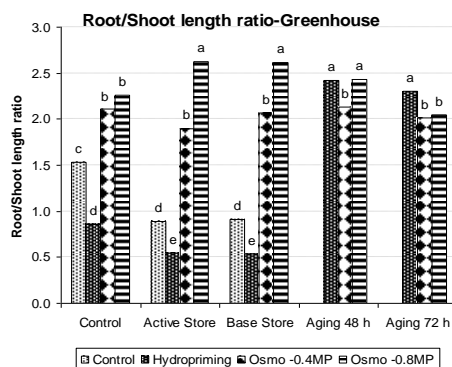
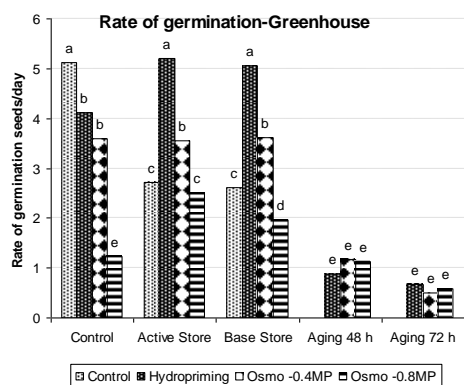
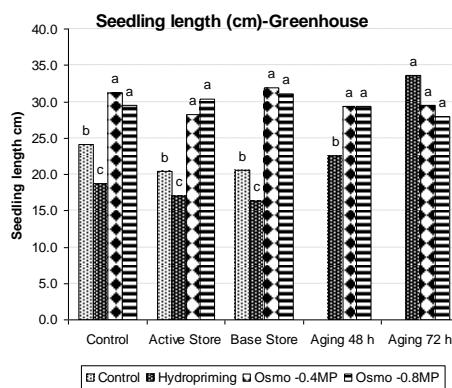
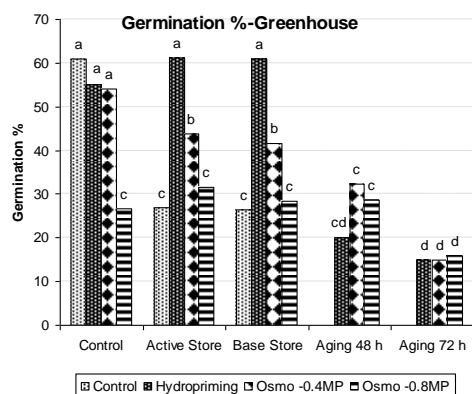
Conservation	Germination (%)	Germination rate	Vigor index	Root length (cm)	Shoot length (cm)	Seedling length (cm)	R/S ratio	Dry Weight (g)
Control	49.67 a	3.30 a	13.20 a	17.16 bc	9.80 a	26.90 b	1.80 c	0.79 a
Aging 48 h	27.83 b	1.08 b	7.69 b	19.34 ab	8.23 b	27.62 b	2.36 a	0.49 bc
Aging 72 h	15.29 c	0.58 b	4.57 c	21.19 a	10.03 a	31.03 a	2.12 b	0.60 b
Active store	43.92 a	3.89 a	10.38 b	15.56 c	9.87 a	25.22 b	1.67 c	0.43 c
Base store	41.51 a	3.58 a	10.14 b	16.80 c	10.11 a	26.66 b	1.75 c	0.48 bc

Means with the same letter are not significantly different (P=0.01)

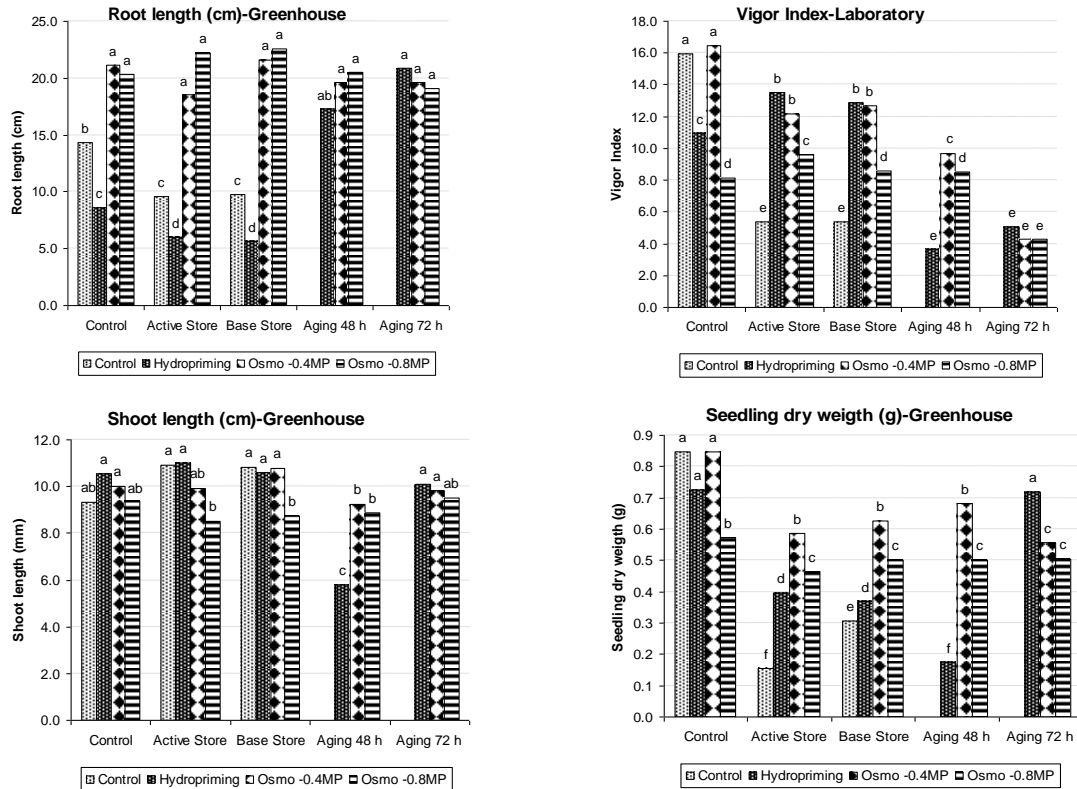
**Table 6.** Means comparison of germination percent, rate of germination, root length, shoot length, seedling length, vigor index, seedling weight and root/shoot length ratio *Onobrychis crista-galli* at four priming treatments under greenhouse conditions

Priming treatments	Germination (%)	Germination rate	Vigor index	Root length (cm)	Shoot length (cm)	Seedling length (cm)	R/S ratio	Dry Weight (g)
Control	36.24 b	3.19 b	8.01 c	10.65 b	10.54 a	21.28 b	1.03 c	0.39 c
Hydropriming	57.50 a	5.02 a	10.35 ab	10.55 b	10.05 a	20.45 b	1.13 c	0.45 bc
Osmo -0.4MP	40.32 b	2.95 b	11.90 a	20.19 a	10.10 a	30.11 a	2.03 b	0.66 a
Osmo -0.8MP	28.20 c	1.80 c	8.47 bc	21.57 a	8.84 b	30.12 a	2.49 a	0.50 b

Means with the same letter are not significantly different (P=0.01)







**Fig. 2.** Priming by conservation interaction effects for seed germination and seedling growth of *Onobrychis crista-galli* under greenhouse conditions

## Conclusion

Our study showed that priming is a useful method to improve quality of deteriorated seeds and an effective way in the recovery of deteriorated seeds. The means of all traits were higher in base cold room ( $-18^{\circ}\text{C}$ ) than active cold room ( $4^{\circ}\text{C}$ ) indicating significant effect of low temperature on seed viability. The root lengths were higher in the accelerated ageing test (48h and 72h). It was due to the improvement of deteriorated seed by priming effect to produce more roots. More seed germination and seedling traits were obtained with regard to the effect of osmopriming (PEG  $-0.4\text{Mpa}$  and  $-0.8\text{Mpa}$ ) followed by hydropriming in both experimental conditions. Regarding our result, it was proved that two priming techniques were effective methods for the improvement of aged seed. To accelerate ageing test, higher mean values of all of traits were obtained by osmopriming (PEG  $-0.4\text{Mpa}$ ). This protocol should be effective for improving the germination

and can be applied by breeders who do not currently have sufficient seed material. The information generated in this research is useful not only for researchers and producers but also for seed companies. In laboratory experiment, the effect of control (no priming) for most of traits was similar and/or higher than priming treatments whereas in the greenhouse experiment, the effects of osmopriming and hydropriming were higher than those for control (no priming) indicating the validity of greenhouse experiment over laboratory.

## References

- Abdual-baki, A. A. and Anderson, J. D., 1973. Relationship between decarboxylation of glutamic acid and vigor in soybean seed. *Crop Science*. 13: 222-226.
- Amooghaie, R., 2011. The effect of hydro and osmopriming on alfalfa seed germination and antioxidant defenses under salt stress. *African Jour. Biotechnology*, 10: 6269-6275.
- Basra, S. M., Ahmad, A. N., Khan, M. M., Iqbal, N. and Cheema, M. A., 2002. Assessment of

- cotton seed deterioration during accelerated ageing. *Seed Science and Technology*, 31: 531-540.
- Bittebcourt, M.L.C., Dais, D.C.F.S., Dias, L.A.S. and Araujo, E.F., 2004. Effect of priming on asparagus seed germination and vigor under water and temperature stress. *Seed Science and Technology*, 32: 607-616.
- Borreani, G., Peiretti, P.G. and Tabacco, E., 2003. Evolution of yield and quality of Sainfoin (*Onobrychis viciifolia* Scop.) in the spring growth cycle *Agronomie*, 23:193-201.
- Butler, L.H., Hay, F.R., Ellis, R.H., Smith, R.D., Murray, T.B., 2009. Priming and re-drying improve the survival of mature seeds of *Digitalis purpurea* during storage. *Ann Bot*, 103: 1261-1270.
- Çelik, A. Karakaya, A., Avci, S., Sancak, C. and Özcan, S., 2011. Powdery mildews observed on *Onobrychis spp.* in Turkey. *Australasian Plant Disease Notes*, 6(1): 49-53.
- Chiu, K.Y. and Sung, J.M., 2002. Effect of priming temperature on storability of primed sh-2sweet corn seed. *Crop Sci.* 42: 1996-2003.
- Eisvand, H.R., Shahrosvand, S, Zahedi, B., Heidari, S. and Afrougheh, S., 2011. Effects of hydro-priming and hormonal priming by gibberellins and salicylic acid on seed and seedling quality of carrot (*Daucus carota var. sativus*) *Iranian Jour. Plant Physiology*, 1(4): 233-239. (In Persian).
- El-Araby, M. and Hegazi, A.Z., 2004. Response of tomato seed to hydro and osmopriming: and possible relation of some antioxidant enzymes and endogenous polyamine fractions. *Egyptian Jour. Biology*, 6: 81 -93.
- Ellis, R. H., Agrawal, P. K. and Roose, E. E., 1988. Harvesting and storage factors that affect seed quality in pea, lentil, Faba bean and chickpea. R.J. Summer field. (ed.) *World crops: Cool Season Food Legumes*.
- Emre, I., Turgut-Balk, D., Sahin, A. and Kursat, M., 2007. Total electrophoretic band patterns of some *Onobrychis* species growing in Turkey. *American-Eurasian Jour. Agric. Environ. Sci.*, 2: 123-126.
- FAO, 2013. Draft gene bank standards for plant genetic resources for food and agriculture. <http://www.fao.org/agriculture/crops/core-themes/theme/seeds-pgr/conservation/gbs/en/> (accessed April 2013).
- Farooq, M., Basra, S.M.A., Tabassum, R. and Afzal, I., 2006. Enhancing the performance of direct seeded fine rice by seed priming. *Plant Prod. Sci.* 9: 446-456.
- Finch-Savage, W.E., Dent, K.C. and Clark, L.J., 2004. Soak conditions and temperature following sowing influence the response of maize (*Zea mays* L.) seeds to on-farm priming (Pre-Sowing Seed Soak). *Field Crops Research*. 90: 361- 374.
- Ghanavati, F., 2012. Notes on the *Onobrychis crista-galli* (L.) Lam. (Fabaceae) in Iran. *Iran. Jour. Bot.* 18 (1): 104-107.
- Hampton, J. G., Brunton, B. J., Pemberton, G. M. and Powarth, J. S., 2004. Temperature and time variables for accelerated again Vigor testing of Pea seed. *Seed science and Technology*.32: 261-264.
- Harris, D., Rashid, A., Arif, M., Yunas, M., 2005. Alleviating micronutrient deficiencies in alkaline soils of the North-West Frontier Province of Pakistan: on-farm seed priming with zinc in wheat and chickpea. In: Andersen, Tuladhar P, Karki JK, Maskey KB. S.L. (Eds) *Micronutrients in South and South East Asia*, pp 143-151. Kathmandu: ICIMOD.
- Janmohammadi, M., Fallahnezhad, Y., Golshan, M. and Mohammadi, H., 2008. Controlled ageing for storability assessment and predicting seedling early growth of canola cultivars (*Brassica napus* L.). *Jour. Agricultural and Biological Science*, 3: 22-26. (In Persian).
- Lu, Y., Sun, Y., Foo, Y., McNabb, W.C., Molan, A.I., 2000. Phenolic glycosides of forage legume *Onobrychis viciifolia*, *Phytochemistry*, 55: 67-75.
- MacDonald, M. B.. 1999. Seed deterioration: physiology, repair and assessment. *Seed Sci. Technol.* 27:177-237.
- Maguire, J. D.. 1962. Speed of germination-aid in selection and evaluation for seedling emergence and vigor. *Crop Science*. 2: 176-177.
- Majidi. M.M. and Barati, M., 2011. Methods for breaking seed dormancy in one cultivated and two wild *Onobrychis* species. *Seed Sci. Tech.* 39: 44-53. (In Persian).
- Majidi, M. M., and Arzani, A. 2004. Study of induced mutation via Ethyl- Methan Sulphonat (EMS) in Sainfoin (*Onobrychis viciifolia* Scop.). *Jour. Agricultural Science and Technology*, 18: 167-180. (In Persian).
- Marshall, A.H. and Lewis, D.N., 2004. Influence of seed storage conditions on seedling emergence, seedling growth and dry matter production of temperate forage grasses. *Seed Science and Technology*, 32: 493-501.
- Nawaz, J., Hussain, M., Jabbar, A., Nadeem, G.A., Sajid, M., Subtain, M., Shabbir, I., 2013. Seed Priming A Technique. *International Jour. Agriculture and Crop Sciences*. 6 (20): 1373-1381.

- Priestley, D. A., 1986. Seed aging. Cornell University Press.
- Probert, R.J., 2000. The role of temperature in the regulation of seed dormancy and germination. In Fenner M, editor., ed., Seeds: The Ecology of Regeneration in Plant Communities, Ed 2 CAB International, Wallingford, UK, pp 261-292.
- Ramezani, M., Rezaei, R., Abandani, S., 2013. Effect of Priming and its Duration on Quality of In-pod Seeds Germination and Seedling Vigor of Sainfoin 'Eski' under Laboratory Conditions *Jour. Crop Improvement* 15 (2): 1-15. (In Persian).
- Rechinger, K.H., 1984. *Onobrychis* in Flora Iranica. Akademische Druck and Verlagsanstalt. Graz, Austria, 157: 387-484.
- Rincker, C. M., 1983. Germination of forage crop seeds after 20 years of subfreezing storage. *Crop Science*, 23: 229-231.
- Rumball, W. and Claydon, B., 2005. Germplasm release 'G35' Sainfoin (*Onobrychis viciifolia*). *Jour. Agri. Res.*, 48: 127-128.
- Simic, B., Popovic, S., Tucak, M., 2004. Influence of corn (*Zea mays* L.) inbred line seed processing on their damage. *Plant, Soil and Environment*, 50, 157-161.
- Soltani, A., Kamkar, B., Galeshi, S.A. and Akram Qadiri, S. F., 1996. The effect of seed deterioration on seed reserves depletion and heterotrophic seedling growth of wheat. *Jour. Agricultural Sciences and Natural Resources*. 15, 61-75. (In Persian).
- Taylor, AG., Allen, PS., Bennett, MA., Bradford, KJ., Burrisand, JS., Misra, MK., 1998. Seed enhancements. *Seed Sci. Res.* 8: 245-256.
- Tekrony, D.M., 1995. Accelerated aging. In: Van de venter, H.A. (Ed.) Seed vigor testing seminar. Copenhagen: ISTA. Pp. 53-72.

Archive of SID

## اثر پرایمینگ بذر بر بهبود شاخص‌های جوانه‌زنی بذر و رشد گیاهچه اسپرس یکساله (*Onobrychis crista-galli*) در شرایط نگهداری میان مدت و بلند مدت بانک ژن

آزاده کاوندی<sup>الف</sup>، علی اشرف جعفری<sup>ب</sup>، مجتبی جعفر زاده<sup>ج</sup>

<sup>الف</sup>فارغ التحصیل کارشناسی ارشد در رشته کشاورزی دانشگاه آزاد اسلامی واحد ساوه، ساوه، ایران

<sup>ب</sup>استاد موسسه تحقیقات جنگل‌ها و مراتع کشور، سازمان ترویج، آموزش و تحقیقات کشاورزی، تهران، ایران \* (نگارنده مسئول)، پست الکترونیک:

aliashrafj@gmail.com

<sup>ج</sup>استادیار دانشگاه آزاد اسلامی واحد بروجرد، بروجرد، ایران

تاریخ دریافت: ۱۳۹۵/۱۱/۲۵

تاریخ پذیرش: ۱۳۹۶/۰۴/۰۷

**چکیده.** اسپرس یکساله *Onobrychis crista-galli* L. سازگاری خوبی به مناطق معتدل و سرد ایران دارد و به صورت طبیعی در مراتع رویش دارد و از آن برای تولید علوفه و چرای دام استفاده می‌شود. به منظور بررسی تاثیر پرایمینگ بذر بر بهبود جوانه‌زنی و رشد گیاهچه در *O. crista-galli*، دو آزمایش فاکتوریل جداگانه در قالب طرح کاملا تصادفی با ۳ تکرار در سال ۱۳۹۴ در آزمایشگاه و گلخانه موسسه تحقیقات جنگل‌ها و مراتع، تهران، انجام گرفت. فاکتور A شامل ۵ روش نگهداری بذر ذخیره‌سازی میان مدت (دمای °C ۴ به مدت ۱۵ سال)، طولانی مدت (دمای °C ۱۸- مدت ۱۵ سال)، بذرهای احیاء شده (شاهد) و تیمار پیری زودرس با قرار دادن بذور در دمای °C ۴۱ و رطوبت ۱۰۰٪ در دو بازه زمانی ۴۸ و ۷۲ ساعت بودند. فاکتور B، پرایمینگ بذر در ۴ سطح شامل اسموپرایمینگ با پلی اتیلن گلیکول PEG6000 (۰/۴- و ۰/۸- مگاپاسکال)، هیدروپرایمینگ (خیساندن بذر به مدت ۲۴ ساعت در آب مقطر) و شاهد (بدون پرایم) بودند. بذرهای پرایم شده اسپرس و شاهد در آزمایشگاه و گلخانه کشت شدند و پس از ۲۱ روز رشد در ژرمیناتور و ۴۵ روز رشد در گلخانه صفات درصد جوانه زنی، شاخص بنیه بذر، طول ریشه‌چه، طول ساقچه، طول گیاهچه و وزن تر گیاهچه اندازه‌گیری شد. داده‌ها با استفاده از نرم افزار SAS مورد تجزیه واریانس قرار گرفتند و میانگین اثرات اصلی و اثرات متقابل با روش دانکن مورد مقایسه قرار گرفتند. نتایج نشان داد که در آزمایشگاه، بیشترین میانگین صفات جوانه زنی بجز طول ریشه‌چه در حفاظت طولانی مدت (دمای °C ۱۸-) بدست آمد. در گلخانه بیشترین رشد رویشی گیاهچه با تیمار اسموپرایمینگ (۰/۴- مگاپاسکال) مشاهده شد. در هر دو محیط آزمایشی هیدروپرایمینگ نیز اثر معنی‌داری بر افزایش میانگین صفات جوانه زنی و رشد گیاهچه در هر دو سیستم حفاظت شده میان مدت و طولانی مدت داشت. در هر دو سیستم حفاظت بذر بیشترین طول ریشه‌چه از طریق اعمال اسموپرایمینگ (۰/۴- و ۰/۸- مگاپاسکال) بدست آمد. در تیمارهای پیری زودرس بیشترین میانگین صفات جوانه‌زنی و رشد گیاهچه از طریق اعمال اسموپرایمینگ (۰/۴- مگاپاسکال) بدست آمد. نتیجه‌گیری کلی نشان داد که اسموپرایمینگ روشی کارآمد در بازیافت بذور زوال یافته طبیعی و مصنوعی می‌باشد.

**کلمات کلیدی:** زوال بذر، پرایمینگ بذر، نگهداری بذر، اسپرس یکساله