



EFFECTS OF PRIMING AND SALT STRESS ON SEED GERMINATION AND EMERGENCE CHARACTERISTICS OF ASAFETIDA (*FERULA ASSA FOETIDA*); A LABORATORY AND GLASSHOUSE TRIAL

Somayeh Elyasirad^{1,2}, Sied Gholamreza Mousavi² and Gholamreza Sanjari³

¹Agriculture and Natural Resources Research and Training Center, Birjand, Iran

²Islamic Azad University, Faculty of Agronomy and Plant Breeding, Birjand, Iran

³The Research Institute of Forest and Range, Tehran, Iran

E-Mail: g.sanjari@gmail.com

ABSTRACT

The gum of asafoetida (*Ferula assa-foetida*) has a very high demand in market while the supply is limited and fully dependent on natural habitats. The objectives of this research were to evaluate the effects of hydropriming and osmopriming on germination and emergence characteristics of asafetida under salt stress condition at laboratory and glasshouse levels. The results showed that at laboratory experiment, the germination traits (germination percentage, germination rate, vigor index, seedling dry weight, root and shoot lengths) increased to higher significant levels in primed seeds with more successful outcomes by hydroprime and osmopriming (CaCl_2 at -2 MPa) compared with un-primed in salinity condition. In glasshouse experiment and under salinity condition, hydroprime and osmopriming treatments achieved a much higher results than control in almost all the emergence attributes (emergence percentage, emergence rate, vigor index, seedling dry weight, root and shoot lengths, root: shoot ratio, proline content and relative water content). Based on the results of this research we conclude that priming techniques have capacity to significantly improve germination and emergence characteristics of asafoetida and to reduce or in some cases fully overcome the negative effects of the salt stresses.

Keywords: asafetida, emergence, germination, hydroprime, osmopriming, salinity.

INTRODUCTION

Asafetida (*Ferula assa-foetida*) is a perennial herbaceous plant belonging to the family of Apiaceae. The species is native to the steppes in Iran and parts of Afghanistan. Propagation by seed is the only way of reproduction of the plant in natural environment. The gum of asafetida is extracted by cutting the apex producing a valuable income for local farmers in some parts of Iran. Salinity is considered as a major abiotic stress in crop production especially in early stages of growth. It reduces seed germination and seedling growth either by creating an osmotic pressure that prevents water uptake by roots or by ionic toxicity of Na^+ and Cl^- (Almansouri *et al.*, 2001; Munns *et al.*, 1988). Pre-sowing treatments such as priming techniques have shown to assist seed germination and seedling establishment processes (Cantliffe, 2003).

Seed priming refers to the process of water absorption to initiate and progress germination just up to the emergence of the root followed by subsequent seed drying (Farooq *et al.*, 2006). In other words, in primed seeds, germination process advances to some extent and halt at the last stage providing the seeds with ability to start germination shortly after sowing, resulting in more rapid emergence of seedlings (Rashid *et al.*, 2006).

Due to ecological adaptation to environmental conditions, medicinal plants including asafetida commonly have inconsistent germination. Various treatments have been proposed to address the issue, one of which is seed priming (Zangoie *et al.*, 2012). Evidence shows that seed improvement techniques can contribute to rapid germination, uniform emergence and robust plant establishment. Strong seedlings establish better

competition with weeds and enjoy a wider range of tolerance to environmental stresses (Ghiyasi *et al.*, 2008). Akramian *et al.* (2007) found that the treatment of fennel seeds with NaCl, KCl and PEG increased their germination rate, root and shoot lengths, and seedling fresh weight as compared to unprimed seeds. Neamatollahi *et al.* (2009) also reported that priming with NaCl and Na_2SO_4 improved seed germination and seedling establishment of caraway.

At present, the gum of asafetida has a very high demand in market while the supply is limited and fully dependent on natural habitats. Such a pressure has decreased the biological capacity of natural ecosystems leaving the species in danger of complete extinction. Therefore cultivation of asafetida in arable lands is of crucial importance. The objective of this study is to determine the effect of different priming treatments on germination and emergence characteristics of asafetida under different levels of salt stress at laboratory and glasshouse phases.

MATERIALS AND METHODS

The present study was carried out in Mohammadiyah's agriculture research station Southern Khorasan, Iran in 2014. The seeds of asafetida were collected from Mohammadabad natural habitation in Kashmar (Lat. $35^{\circ}57'18''$ N., Long. $58^{\circ}22'18''$ E., Alt. 1900 m). Since asafetida seeds have morpho-physiological dormancy, they were first pre-treated with moist coldness at 5°C for a month (Otroshy *et al.*, 2009). The experiment was carried out in two stages; laboratory and glasshouse trials.



In the lab experiment, a factorial design with two factors including priming at 6 levels (hydropriming with distilled water; osmopriming with CaCl_2 at osmotic potentials of -1 and -2 MPa; osmopriming with KH_2PO_4 at -1 and -2 MPa and unprimed treatment as the control) and salt stress at 4 levels (0, 4, 8 and 12 mmho/cm) was set up based on a completely randomized design (CRD) in four replications. The glasshouse experiment was carried out after the completion of the lab trial. Based on the results of the laboratory work, two superior priming treatments were selected to be used in the glasshouse experiment. The design in glasshouse was the same as the lab experiment but with only three priming treatments including hydropriming with distilled water and osmopriming with CaCl_2 at -2 MPa (selected from the first experiment) plus unprimed treatment as the control).

The priming solutions were constructed (Vanthoff, 1887) and the seeds were treated in glassy beakers containing 150 cc of the priming solutions (volumetric ratio of solution to seeds was 5) for 12 hours. They were then washed with distilled water three times and dried at 20°C in an infected-free medium in laboratory for 12 hours.

In the lab experiment, the petri dishes containing two filter papers with 7 ml salt solutions of different concentrations and 15 seeds were used. They were then sealed and kept in a germinator at 14°C with light period of 12 hours for one month. In control treatment, the seeds were placed in petri dishes after chilling without priming. The germination of the seeds was recorded on a daily bases for 30 days and the experiment was continued until germination was fully stopped. The minimum root length of 2 mm was considered as germination (Adam *et al.*, 2007). For the lab experiment, the following traits were recorded: germination percentage (GP), germination rate (GR), seedling dry weight (DW), vigor index (VI), seedling length (SL), root length (RL) and shoot length (Sh.L).

In the glasshouse experiment, the pots with the diameter of 10 cm were filled by fine sands and 15 seeds were placed at the top and were covered with a one centimeter depth of the sand. They were watered with the salt solution treatments from the beginning of the experiment till the end. To make sure that the seedlings have access to nutrients, the pots were watered by hydroponic nutrient solution (Hoagland and Arnon, 1950) once in 3 watering events. In the glasshouse trial, the following traits were recorded: Emergence percentage (EP), emergence rate (ER), seedling dry weight (DW), vigor index (VI), seedling length (SL), root length (RL), shoot length (Sh.L), relative water content (RWC) and proline concentration (Pr).

For germination or emergence percentage Equation 1 (Bajji *et al.*, 2002), for germination or emergence rate Equation (2) (Ellis and Roberts, 1981), for

relative water content Equation (3) (Farooq *et al.*, 2009) and for vigor index equation 4 were used.

$$GP = \left(\frac{N}{N_t}\right) \times 100$$

(1)

where, *GP* is germination (emergence) percentage, *N* and *N_t* are the number of germinated or emerged seeds and total number of seeds, respectively.

$$GR = 1 / \frac{\sum(n \times d)}{N}$$

(2)

where, *GR* is germination (emergence) rate, *n* is the number of seeds germinated (emerged) on the *d*th day and *N* is the total number of germinated (emerged) seeds until the end of the experiment.

$$RWC = \frac{(W_f) - (W_d)}{(W_s) - (W_d)} \times 100$$

(3)

Where, *RWC* (%) is relative water content, *W_f* is fresh weight, *W_s* is saturated weight and *W_d* is dry weight.

$$VI = SL \times GP$$

(4)

Where, *SL* is seedling length, *GP* is germination (emergence) percentage

The proline content was estimated based on Bates method (Bates *et al.* 1973). The lengths of seedling, root and shoot were measured in millimeter with a ruler after the completion of seed germination. Root and shoot were weighed in 3 decimal places accuracy using a digital balance. The seed germination data were transformed, using an arcsine square root transformation for normalization before running statistical analysis tests. However the data presentation in the tables and graphs are all in actual format. The data went through analysis of variance and the means were compared using FLSD test using SAS software package.

RESULTS

Laboratory experiment

The results of the ANOVA illustrate that priming and salt stress treatments had significant effects on all the germination traits of *asafetida* under the laboratory condition.

**Table-1.** Mean comparison of the germination traits for priming and salt stress (laboratory) .

Treatments	GP (%)	GR (days ⁻¹)	DW (gr)	VI ---	S.L (cm)	R.L (cm)	Sh.L (cm)
----- Priming treatments -----							
Control	30.6cd	0.043d	0.0054d	948e	6.92c	3.38b	3.54d
Water	42.9a	0.072ab	0.0080b	2244a	11.19a	5.3a	5.94ab
CaCl ₂ (-1)	30.6cd	0.065bc	0.0092a	1161b	11.17a	5.46a	5.72b
CaCl ₂ (-2)	37.9b	0.079a	0.0086ab	1795b	12.02a	5.48a	6.54a
KH ₂ PO ₄ (-1)	33.7c	0.069bc	0.0067c	1429c	8.3b	3.9b	4.44c
KH ₂ PO ₄ (-2)	31.6c	0.064c	0.0047d	1340c	7.13c	3.3b	3.81cd
----- Salt treatments (mmho/cm) -----							
0	50.2a	0.082a	0.0099a	2624a	14.0a	6.8a	7.2a
4	36.4b	0.067b	0.0081b	1567b	10.4b	4.8b	5.5b
8	28.7c	0.060c	0.0067c	1052c	8.9c	4.1c	4.8c
12	20.5d	0.053d	0.0037d	701d	4.6d	2.1d	2.5d

Germination percentage (GP), Germination rate (GR), Dry weight (DW), Vigorindex (VI), Seedling length (SL), Root length (RL), Shoot Length (Sh.L)

Germination percentage was significantly higher (Table-1) in hydroprime, osmopriming KH₂PO₄ (-1 MPa) and osmopriming CaCl₂ (-2 MPa) than the control treatment. Seed germination in response to salinity showed to have sharp decrease from 50.2% at zero mmho/cm to 20.5% at 12 mmho/cm. The effect of priming at different salt stress levels showed that hydroprime had the highest (60%) effects on germination percentage (Figure-1a) at zero salt that was significantly different with others. At 4 mmho/cm, hydroprime and osmopriming CaCl₂ (-2 MPa) had the higher germinations (48.3% and 43.3% respectively) than the control.

Germination rate was the lowest (0.043) under unprimed treatment (Table-1) while increased significantly in all the priming treatments, maximized at 0.079 with osmopriming CaCl₂ (-2 MPa). Salt stress treatments had inverse effects on germination rate being highest (0.082) at zero stress and the lowest (0.053) at 12 mmho/cm. Interactions between priming and salt stress showed an

overall significant increase in germination rate for all the priming treatments compared with the control at any single salt stress level (Figure-1b). At zero salinity, CaCl₂ at -2 MPa achieved the highest rate of germination (0.109) that is much higher than the others. The priming treatments also maintained higher germination rates at more salinity stress. For instance at 12 mmho/cm, the rate was at least double in all the priming treatments as compared with their controls.

Seedling dry weight turned out to be the lowest (4.7 mg) in KH₂PO₄ (-2 MPa) and the highest (9.2 mg) in CaCl₂ (-1 MPa). The next high achieving treatments were Hydroprime and osmopriming CaCl₂ (-2 MPa) with 8.6 and 8 mg, respectively. The dry weight of asafetida seedling decreased with the elevated levels of salt stress. The highest effects of priming treatments on dry weight of asafetida (9.9 mg) was obtained under no salt stress treatment which decreased sharply to 3.7 mg when the salt stress increased to 12 mmho/cm.

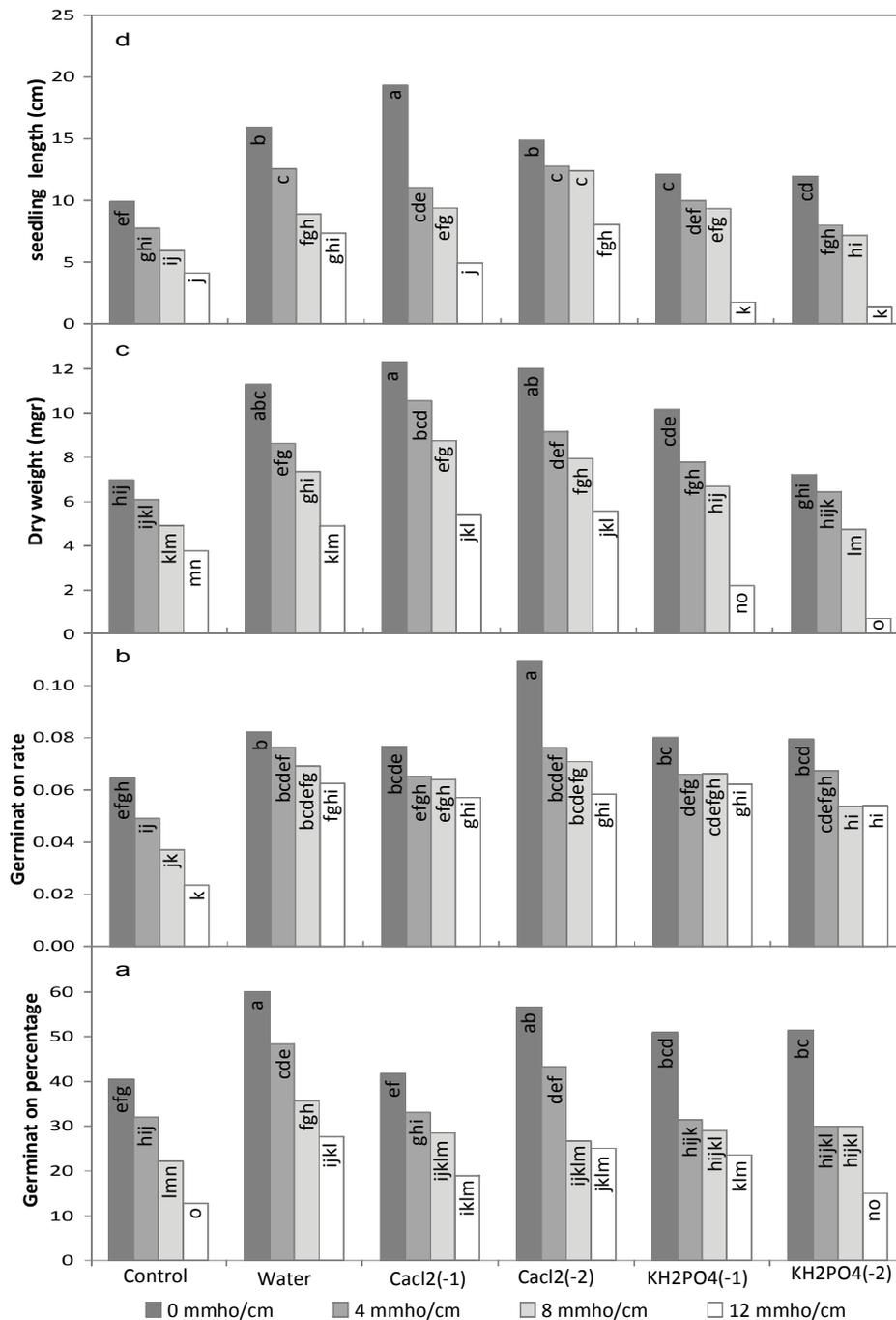


Figure-1. Effect of priming in relation to salt stress on germination traits of asafetida seeds.

The interaction between priming and salt stress showed that dry weight of asafetida seedlings peaked at zero salt stress for all the priming treatments (Figure-1C) with the maximum values of 11 - 12 mg in hydroprime and osmoprimed CaCl₂ at -1 and -2 MPa. At higher rates of salt stresses, the aforementioned treatments maintained the superiority over the other treatments including control.

Seedling length increased significantly in priming treatments compared with the control (Table-1) which was more prominent in osmoprimed CaCl₂ (-2 MPa),

hydroprime and osmoprimed CaCl₂ (-1 MPa). The seedlings with average length of 14 cm at zero salt stress decreased with salinity getting 4.6 cm at 12 mmho/cm. Taking into account the interaction between priming and salt stress (Figure-1d), seedling length was highest at zero salt stress for all the treatments with higher values recorded under CaCl₂ at -1 MPa (19.5 cm) and -2 MPa (14.9 cm) and hydroprime (16.3 cm). At higher salt stress levels (8 and 12 mmho/cm), CaCl₂ at -2 MPa produced longer seedling length (12.4 and 8 cm) than the others. The minimum



length was recorded at 12 mmho/cm under osmoprime KH_2PO_4 at -2 MPa (1.4 cm) which is much lower than the control's (4.1 cm).

The overall results from the laboratory experiment showed that in comparison to the other priming treatments, hydroprime and osmoprime (CaCl_2 at -2 MPa) have relatively had the highest significant effects on the germination traits at the salt stress levels studied. Therefore the two priming treatments were selected for the second phase of the experiment at glasshouse trial.

Glasshouse experiment

In glasshouse experiment, priming, salt stress and their interactions had significant effects on all emergence

attributes of asafetida. The mean comparisons of the data on priming illustrate that emergence percentage obtained higher significant values in hydroprime (49.2%) and osmoprime (51.2%) than the control (40.8%). In an inverse relation, emergence percentage significantly reduced from 53.1% at zero salt stress to 39.6% at 12 mmho/cm (Table-2). The interaction between priming and salt stress, revealed a higher emergence in priming treatments compared with control in all salt stress treatments (Figure-2a). At 12 mmho/cm, the emergence percentage in osmoprime, hydroprime and the control observed to be significantly varied at 45.1, 40.2 and 33.3 percent, respectively.

Table-2. Mean comparison of the emergence characteristics for priming and salt stress (glasshouse).

Treatments	EP (%)	ER (days ⁻¹)	DW (gr)	VI ---	S.L (cm)	R.L (cm)	Sh.L (cm)	R/Sh ---	Pr mg/gr	RWC %
----- Priming treatments -----										
Control	40.8b	0.139c	0.131b	470b	10.8b	4.96b	5.89b	0.79b	0.4272b	54.32b
Water	49.2a	0.228a	0.181a	839a	16.3a	7.54a	8.68a	0.83b	0.4633a	66.48a
$\text{CaCl}_2(-2)$	51.2a	0.16b	0.181a	863a	16.2a	7.97a	8.26a	0.92a	0.4647a	66.54a
----- Salt treatments (mmho/cm) -----										
0	53.1a	0.236a	0.250a	1272a	23.8a	11.88a	11.9a	1.015a	0.4107d	75.03a
4	49.9b	0.195b	0.193b	988b	19.5b	9.25b	10.3b	0.903b	0.4251c	73.88a
8	45.8c	0.147c	0.127c	393c	8.6c	3.79c	4.8c	0.787c	0.4390b	59.47b
12	39.6d	0.125d	0.091d	243d	5.9d	2.38d	3.5d	0.676d	0.5323a	41.41c

Germination percentage (GP), Germination rate (GR), Dry weight (DW), Vigorindex (VI), Seedling length (SL), Root length (RL), Shoot Length (Sh.L), Root:Shoot ratio (R/Sh), Relative water content (RWC)

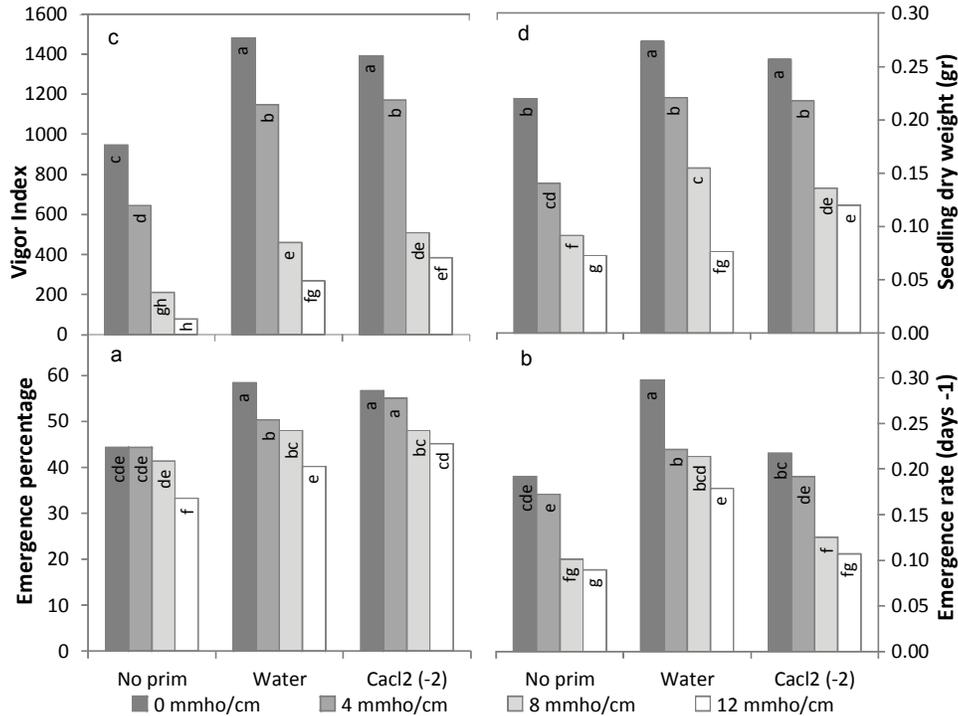


Figure-2. Effects of priming vs. salt stress on some emergence traits of asafetida.

Emergence rates of asafetida's seeds increased significantly in priming treatments especially with hydroprime (0.22) while it remained at 0.16 with the control. Salt stress treatments reduced the emergence rate from 0.236 at zero stress to 0.127 at 12 mmho/cm. Interaction between priming and salt stress was found to have great effects on emergence rate in hydroprime (Figure-2b). Hydroprime treatment not only produced the highest rate of emergence at zero salt stress (0.297) but also presented a much higher rates at 8 and 12 mmho/cm (0.18 - 0.21).

Vigor index under priming treatments was significantly higher than control. Salt stress reduced the index from 1272 at zero stress to 988, 393 and 243 at 4, 8 and 12 mmho/cm, respectively. Hydroprime and osmopriming (CaCl_2 at -2 MPa) increased the vigor index to higher significant levels than control in all the salt stress treatments, which is more prominent at 8 and 12 mmho/cm (Figure-2c).

Seedling dry weight increased to 0.181 gr for the two priming treatments (Table-2) that is significantly different with the control (0.131 gr). Salt stress inversely decreased dry weight from 0.250 gr at zero mmho/cm to the lowest (0.091 gr) at 12 mmho/cm. The interaction between priming and salt treatments showed a higher dry weight for the two priming treatments at 0, 4 and 8 mmho/cm than the control's (Figure-2d). At 12

mmho/cm it was only osmopriming to produce more significant dry weight than control but not hydroprime.

Priming treatments had positive significant effects on root, shoot and seedling lengths and even on root:shoot ratio. Seedling length increased to around the same values of 16.2 to 16.3 cm in the two priming treatments while it remained at 10.8 cm with the control. Root and shoot lengths responded to priming in the same way as the seedling length. But root:shoot ratio was more affected by osmopriming than hydroprime (Table-2). Salt stress negatively affected the lengths of root, shoot and seedling as well as the root:shoot ratio. For instance, the seedling length decreased from 23.8 at zero salt stress to 19.5, 8.6 and 5.9 cm at 4, 8 and 12 mmho/cm, respectively. The root:shoot ratio was highest (1.015) at no salt treatment but decreased significantly with the increase in salt concentration getting the lowest ratio of 0.68 at 12 mmho/cm (Table-2).

The interaction between priming and salt stress on aforementioned attributes showed that priming increased the lengths of the seedling, root and shoot at any single salt stress levels compared with the control (Fig 3a, Fig3c, Fig3d). However in root:shoot ratio, the two priming treatments had only significant effects at zero salt stress with an exception at 4 mmho/cm in osmopriming. In other salt stress levels, priming had no significant effects on root:shoot ratio compared with control (Figure-3b).

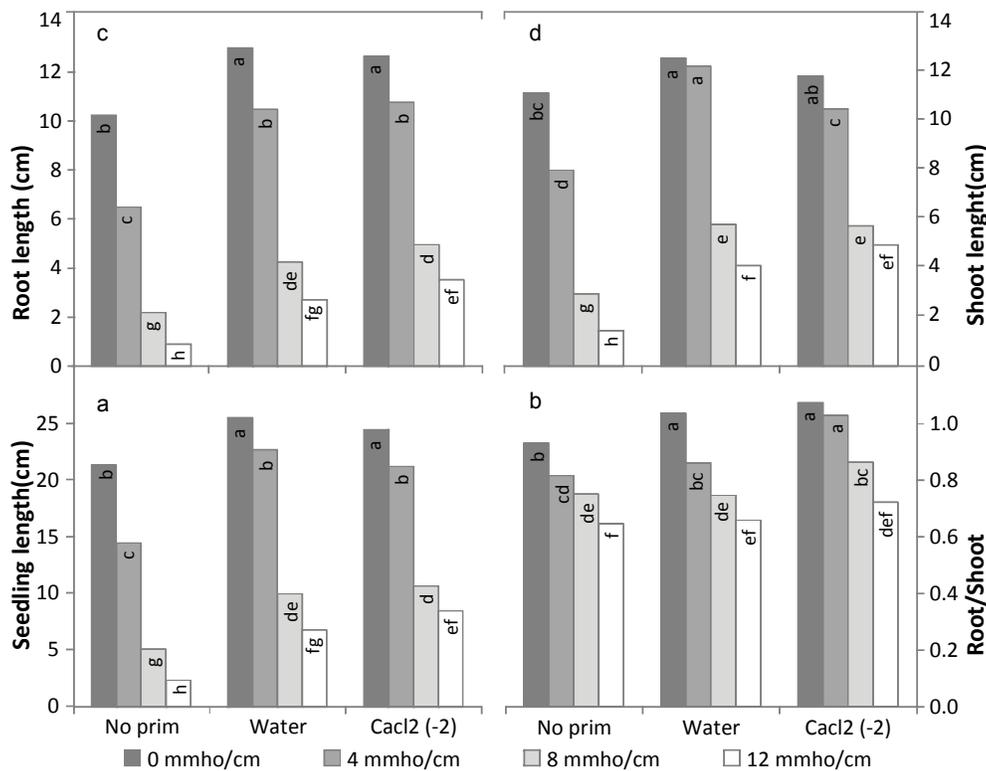


Figure-3. Effects of priming vs. salt stress on seedling, root, shoot and root:shoot ratio of asafetida.

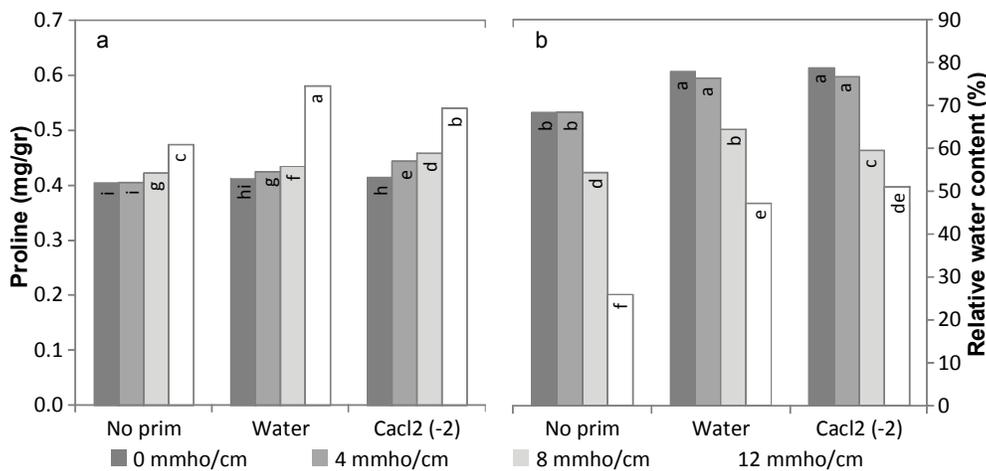


Figure-4. Effects of priming on proline and water content of asafetida in relation to salt stresses.

Seedling proline content increased significantly (Table-2) under the two priming treatments (0.46 mg/gr) compared with the control (0.42 mg/gr). Salt stress also increased proline concentration from 0.41 mg/gr at zero stress to 0.53 mg/gr at 12 mmho/cm). The highest proline concentrations (0.58, 0.54 and 0.47) were obtained at 12 mmho/cm in hydroprime, osmoprime and control, respectively (Figure-4a).

Relative water content (RWC) increased with priming to the significant level of 66.5% compared with 54% in the control but decreased significantly with the increase in salt stress. The RWC declined from 75% at no

salt stress to the minimum amount of 41% at 12 mmho/cm. The interaction (Figure-4b) illustrates significant improvement to RWC in priming treatments at any single salt stress levels especially at 12 mmho/cm as compared with control.

DISCUSSIONS

The results of the laboratory experiment under salt stress condition showed that the priming treatments had positive significant effects on germination attributes of asafetida. Hydropriming and osmopriming with CaCl₂ at both -1 and -2 MPa, were the top three treatments that



have made highest significant improvement to almost all the traits studied. Osmoprime with KH_2PO_4 at -1 and -2 MPa were also observed to have significant effects but not on all the germination traits (Table 1). The positive effects of priming on germination have been reported in literature (Murungu *et al.*, 2003; Demir Kaya *et al.*, 2006; Neamatollahi *et al.*, 2009; Abbasi *et al.*, 2012). Cellular and molecular studies show that priming increases seed capability in improving DNA replication, stimulating RNA activities, enhancing protein synthesis and increasing hormone driven germination like ethylene. Therefore, when the treated seeds are placed in germination conditions, they show significantly higher germination characteristics than control (Azarnivand *et al.*, 2010).

Germination attributes showed inverse relations with salt stress being highest at zero mmho/cm and lowest at 12 mmho/cm. Nevertheless priming treatments significantly increased germination traits almost at any single salt stress levels compared with control. Such effect was more pronounced at salt stress 8 and 12 mmho/cm in germination rate in all the priming treatments (Figure-1(b)). Although the decrease in germination characteristics of asafetida under salt stress can be attributed to the decreasing osmosis potential of solution, increasing toxin ions and changing in remobilization balance of seed reservoirs, however priming can reduce or eliminate such effects through the enhancement of plant salt tolerance (Cantliffe, 2003).

At the laboratory stage of the experiment we found that under salt stress condition, hydroprime and osmoprime using CaCl_2 are more effective than osmoprime with KH_2PO_4 on improving germination traits. In our glasshouse trial, hydropriming and osmoprime using CaCl_2 at -2 MPa were also found to have significant effects on wide range of emergence attributes studied under the same salt condition as the laboratory trial. Iqbal *et al.*, (2006) who used CaCl_2 as a priming solution, found that priming has been effective in alleviating the negative impacts of salt stress on wheat through their effects on changing the levels of the plant phytohormones. The success of hydroprime on germination and emergence of asafetida is also in line with the results of Vajanti *et al.*, (2013) who found that hydroprime can fully eliminate the negative effects of salt stress at concentration up to 100 mM and reduce the effects from 100 mM to 200 mM in all attributes of sunflower.

In glasshouse experiment, emergence percentage and emergence rate were both increased with priming as compared with control in all salt stress levels (Figures 2(a), 2(b)). The results are in agreement with Vajanti *et al.* (2013), Tavili *et al.* (2010) and Neamatollahi *et al.* (2009) who observed hydroprime to exhibit significant effects on germination percentage and the rate in sunflower, woolly broom and cumin. It is pointed out that priming improves seed performance by encouraging rapid, uniform, and vigorous germination which helps seedlings to grow in stressed conditions resulted in a relatively higher plant emergence (Carbineau and Come 2006; Ashraf and Kinet, 2005; Cantliffe 2003).

Seedling vigor index was significant in hydroprime and osmoprime (CaCl_2 at -2 MPa) under salt stress condition as compared with control (Table-2 and Figure 2(c)). Such a result was reported by Khan *et al.* (2009) who observed that seeds of hot pepper primed with NaCl improved vigor index and seedling establishment under salinity condition. In our study, the two priming treatments both increased the index in even higher salt concentration over the control which is the same as the results obtained by Mohammadi *et al.* (2008). He reported that hydropriming could significantly increase the vigor index of maize mainly at high osmotic potential. Vigor index is generally affected by germination percentage and seedling length thus it can be said that treatments with relatively high germination percentage and seedling length would have a higher seed vigor.

Seedling dry weight increased with priming treatments in salinity condition as compared with the control, being highest with osmoprime at 12 mmho/cm (Fig 2d). Obtaining higher seedling dry weight under primed treatments seems normal, because the primed seeds usually germinate faster and start expanding their root systems sooner than unprimed seeds giving the chance of having more time for taking up water and nutrient as well as assimilation of organic materials. Moreover the establishments of the primed seedlings are quick and have better chance to compete weeds. Murungu *et al.* (2003) and kaur *et al.* (2005) reported significant increase in dry weight of wheat and pea due to seed priming compared with control.

The lengths of seedling, root, shoot and root:shoot ratio increased with priming under salt stress condition. The priming treatments increased the four attributes to significant levels higher than control at all the salt stress levels. The effects of priming on root:shoot ratio showed that in salt stress levels of 4 and 8 mmho/cm, hydroprime and osmoprime promoted root length more than shoot length. The positive effects of priming on root and shoot lengths have been also reported by Midaoui *et al.* (2003) and Vajanti *et al.* (2013) on sunflower. Increase in root and shoot length is attributed to the increase in germination and emergence rate of primed seeds (Khalil *et al.* 1997). For instance, in a study on wild rye (*Elymus chinensis*), it was found that seeds osmoprime by PEG 30% for 24 hours enhanced the activities of superoxide dismutase and per-oxidase and rapid increase in respiration rate which usually results in a higher seedling length (Jie *et al.*, 2002).

Proline content of the seedlings increased significantly in both hydroprime and osmoprime treatments under salt stress condition. The effects of priming on proline content increased with the increase in salt stress. Plant species synthesize proline to protect themselves against salinity and drought stress. Therefore plants that synthesize more proline are more tolerant to the stresses (Demiral and Turkan 2005). In line with our results, accumulation of proline under stress condition has been documented by Mohammadi and Nassiry (2015), Farahmandfar *et al.* (2013) and Elouaer *et al.* (2014) who worked on basil, fenugreek and safflower respectively.



Proline acts as an osmotic regulator associated with mechanism of salt tolerance under salinity stresses (Yu and Shaozheng 2000) therefore increasing in synthesis of proline is known as common metabolic reaction to plant under stress which enhances plant salt tolerance to salinity stress.

Relative water content (RWC) was significantly decreased in asafetida seedling in response to increasing salinity stress. Meanwhile RWC was higher in plants derived from seeds primed with the treatments than plants derived from no primed seeds. Priming treatments at any single salt stress level showed to have higher significant RWC than control especially at 12 mmho/cm. Drought and salt stress reduce RWC but plants tolerant to the stresses have abilities to store more water in their bodies to compensate the water that is lost through transpiration. Mattioni *et al.* (1997) pointed out that under salt stress condition when proline is accumulated, RWC acts the same. The positive effects of priming on RWC in our experiment is also in line with reports of Mehmet and Kaydan (2008) and Vajanti *et al.* (2013) who found significant effects of hydroprime on RWC in triticale and sunflower.

CONCLUSIONS

Asafetida could be recognized as a sensitive plant to salinity at germination and early growth stages. Seed priming in asafetida has capacity to improve germination and emergence attributes and to reduce or overcome salinity at concentrations up to 12 mmho/cm. Based on the results obtained in this research we introduce hydroprime and osmopriming (CaCl₂ at -2 MPa) as the most successful treatments in seed priming for the plant although more trials and repetition are needed to confirm such statement. It is advised to conduct similar experiments using a range of different ecotypes of asafetida at a wider range of salinity as well as priming solutions.

ACKNOWLEDGEMENTS

The authors thank Mr Mohsen Zangoie for his comments and acknowledge the research and training center of South Khorasan for agriculture and natural resources for the supports during laboratory and glasshouse experiments.

REFERENCES

- Abbasi M., Pouzesh. H., Enayati A., Hedayati A. 2012. Investigation the effect of hydropriming and osmopriming treatments on seeds germination of Tall Wheatgrass (*Agropyronelongatum*) under drought stress, *Annals of Biological Research*. 3(10): 4874-4879.
- Adam N., R. Dierig D., A. Coffelt T., A. and Wintermeyer M., J. 2007. Cardinal temperatures for germination and early growth of two *Lesquerella* species. *Industrial Crops and Products*. 25: 24-33.
- Akramian M., Hosseini H., Kazerooni Monfared A., Rezvani Moghaddam P. 2007. Effect of seed osmopriming on germination and seedling development of fennel (*Foeniculum vulgare Mill.*). *Iranian Journal of Agricultural Researches*. 5(1): 37-46.
- Almansouri M., J. M. Kinet and S. Lutts. 2001. Effect of salt and osmotic stress on germination in durum wheat (*Triticum durum Desf.*). *Plant and Soil*. 231: 243- 254.
- Ashraf M., Foolad M. 2005. Pre-sowing seed treatment-A shotgun approach to improve germination, plant growth, and crop yield under saline and non-saline conditions. *Advances in Agronomy*. 88:223-271.
- Azarnivand H., Abasi M. & Enayati A. 2010. Evaluation and determination of the best hydro and osmopriming treatments for germination properties of tall wheatgrass (*Agropyron elongatum*). *Iranian Journal of Natural Resources*. 62(4):431-444.
- Bajji M., J. Kinet M. and Lutts S. 2002. Osmotic and ionic effects of NaCl on germination, early seedling growth, and ion content of *Atriplexhalimus*. *Canadian Journal of Botany*. 80: 297-304.
- Bates I. S., R. P. Waldern & I. D. Teare 1973. Rapid determination of free proline for water stress studies. *Plant and Soil*. 39:205-207.
- Cantliffe D. 2003. Seed enhancements, *Acta Horticulturae* 607:53-62.
- Carbineau F., Come D. 2006. Priming: a technique for improving seed quality, *Seed Testing International*. (132).
- Demir Kaya M., Okcu G., Atak M., Cikili Y. And Kolsarici O. 2006. Seed treatments to overcome salt and drought stress during germination in sunflower (*Helianthus annuus L.*). *European Journal of Agronomy*. 24:291-295.
- Demiral T. and İ. Türkan. 2005. Comparative lipid peroxidation, antioxidant defense systems and proline content in roots of two rice cultivars differing in salt tolerance. *Environmental and Experimental Botany*. 53:247-257.
- Ellis R., H. and Roberts E., H. 1981. The quantification of aging and survival in orthodox seeds. *Seed Science & Technology*. 9:373-409.
- Elouaer M.A, Ben F.M, Zhani K, Hannachi C. 2014. Influence of NaCl Seed Priming on Growth and Some Biochemical Attributes of Safflower Grown under Saline Conditions, *Research on Crop Ecophysiology*. 9(1):13-20.
- Farahmandfar E, Bagheri M, Azimi S, Hoseinzadeh D. 2013. Effect of seed priming on morphological and physiological parameters of fenugreeek seedlings under salt stress, *International Journal of Agriculture and Crop Sciences*. 5(8):811-815.



- Farooq M., Basra S.M.A, Wahid A. 2006. Priming of field-sown rice seed enhances germination, seedling establishment, allometry and yield. *Plant Growth Regulation*. 49:285-294.
- Farooq M., Wahid A and D.J. Lee. 2009. Exogenously applied polyamines increase drought tolerance of rice by improving leaf water status, photosynthesis and membrane properties. *Acta Physiologiae Plantarum*. 31:937-945.
- Ghiyasi M., A. Abbasi S. Tajbakhsh M., Amirmia M. and Salehzade H. 2008. Effect of osmopriming with polyethylene glycol 8000 (PEG8000) on germination and seedling growth of wheat (*Triticum aestivum* L.) seeds under salt stress. *Research Journal of Biological Science*. 3(10):1249-1251.
- Hoagland, D.R. and D.I. Arnon. 1950. The water-culture method for growing plants without soil. *California Agricultural Experiment Station Circular*. 347:1-32.
- Iqbal M, Ashraf M, Jamil A, Ur-Rehman S. 2006. Does seed priming induce changes in the levels of some endogenous plant hormones in hexaploid wheat plants under salt stress. *Journal of Integrative Plant Biology*. 48(2):181-189.
- Jie L., Gong She O., Dong Mei L., Fang Fang W. 2002. Effects of PEG on germination and active oxygen metabolism in wildrye (*Leymus chinensis*) seeds. *Acta Prataculturae Sinica*. 11:59-65.
- Kaur S. Gupta, A.K, and Kaur N. 2005. Seed priming increases crop yield possibly by modulating Enzymes of sucrose metabolism in chickpea. *Journal of Agronomy and Crop Sciences*. 191:81-87.
- Khalil S., Mexal J., Ortiz M. 1997. Osmotic priming hastens germination and improves seedling size of *Pinus brutia* var. *eldarica*. *Tree Planters Notes*. 48:24-27.
- Khan H., Ayub C., Pervez M., Bilal R., Wahid M., Ziaf K. 2009. Effect of seed priming with NaCl on salinity tolerance of hot pepper (*Capsicum annuum* L.) at seedling stage, *Soil & Environment*. 28(1):81-87.
- Mattioni C., Lacenza N. G., Troccoli A., De Leonardis A. M. and Fonzo D. 1997. Water and salt stress-induced alterations in proline metabolism of *Triticum durum* seedlings. *Physiologia Plantarum*. 101:787-792.
- Mehmet Y.M and Kaydan. D.D. 2008. Alleviation of osmotic stress of water and salt in germination and seedling growth of triticale with seed priming treatments. *African Journal of Biotechnology*. 7(13):2156-2162.
- Midaoui M., Serieys H., Griveau Y., Benbella M., Talouizte A., Berville A. and Kaan F. 2003. Effects of osmotic and water stresses on root and shoot morphology and seed yield in sunflower (*Helianthus annuus* L.) genotypes bred for Morocco or issued from introgression with *H. argophyllus* T.&G. and *H. debilis* Nutt. *Helia*. 26:1-16.
- Mohammadi N, Nassiry B. 2015. The effect of osmopriming on morphological, physiological characteristics and biochemical changes of *Ocimum basilicum* under drought stress, *International Journal of Farming and Allied Sciences*. 4(2):102-110.
- Mohammadi J.M., Dezfuli. P.M. and Sharifzadeh. F. 2008. Seed invigoration techniques to improve germination and early growth of inbred line of maize under salinity and drought stress. *General and Applied Plant Physiology*. 34(3-4):215-226.
- Munns R.A., Gardiner M.L., Rawson H.M. 1988. Growth and development in NaCl treated plants. II. Do Na⁺ or Cl⁻ concentrations in dividing or expanding tissue determine growth in barley. *Australian Journal of Plant Physiology*. 15:529-540.
- Murungu F., Nyamugafata P., Chiduza C., Clark L., Whalley W. 2003. Effect of seed priming, aggregate size and soil matric potential on emergence of cotton (*Gossypium hirsutum* L.) and maize (*Zea mays* L.). *Soil and Tillage Research*. 74:161-168.
- Neamatollahi E., Bannayan M., Souhani Darban A. and Ghanbari. A. 2009. Hydropriming and osmopriming effects on cumin (*Cuminum Cuminum* L.) seeds germination. *International Journal of Biological, Biomolecular, Agricultural, Food and Biotechnological Engineering*. 57:526-529.
- Otroshy M., Zamani A., Khodambashi M., Ebrahimi M. and Struik P., C. 2009. Effect of Exogenous Hormones and Chilling on Dormancy Breaking of Seeds of Asafoetida (*Ferula asafoetida* L.). *Research Journal of Seed Science*. 2(1):9-15.
- Rashid A., Hollington P.A., Harris D. and Khan P. 2006. On-farm seed priming for barley on normal, saline and saline sodic soils in NWFP, Pakistan. *European journal of agronomy*. 24 (3):276-281.
- Tavili Ali; Zare Salman; Moosavi Seyed Amir; Enayati Abdolghader. 2010. Effects of Priming Techniques on Seed Germination and Early Growth Characteristics of *Bromus tomentellus* L. and *Bromus inermis* L. *Notulae Scientia Biologicae*. 2 (1):104-108.
- Vajanti M., Pahoja S., Siddiqui S., Narejo H., Umrani. 2013. Response of hydropriming and osmopriming on germination and seedling growth of sunflower (*Helianthus Annus* L.) under salt stress, *International Journal of Agricultural Science and research*. 3(2):71-80.



Van't Hoff, J. W. 1887. Die Rolle des osmotischen Druckes in der Analogie zwischen Lösungen und Gasen. *Z Physik Chemie*.1:481-493.

Yu Lei MK, Shaozheng L. 2000. Research on salt tolerance of some tree species on muddy sea shore of north China. In: International Seminar on Prospects for Saline Agriculture. April 10-12, Islamabad, Pakistan.

Zangoie M., Parsa S., Mahmoodi M., Jami Al-Ahmadi M. 2012. Evaluation of cardinal temperature for germination of asafetida (*Ferula assafoetida* L.) seeds. *Journal of Plant Production*. 19(3):193-202.