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Study of Different Priming Treatments on Germination Traits of Soybean Seed Lots

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Abstract

Oilseeds are more susceptible to deterioration due to membrane disruption, high free fatty acid level in seeds and free radical production. These factors are tended to less vigorous seed. Priming treatments have been used to accelerate the germination and seedling growth in most of the crops under normal and stress conditions. For susceptible and low vigor soybean seed, this technique would be a promising method. At first, in separate experiment, effects of hydropriming for (12, 24, 36 and 48 h) with control (none prime) were evaluated on germination traits of soybean seed lots cv. 'Sari' (include 2 drying method and 3 harvest moisture). Then, next experiment was conducted to determination the best combination of osmopriming in soybean seed lots, hence 3 osmotic potential level (-8, -10 and -12 bar) at 4 time (12, 24, 36 and 48 h) were compared. Analysis of variance showed that, except for seedling dry weight, the other traits include standard germination, germination rate, seedling length and vigor index were influenced by osmopriming. Hydropriming had no effect on these traits and decreased rate of germination. Finally the best combination of osmopriming were osmotic potential -12 bar at 12 hours for time, that submitted acceptable result in all conditions and recommended for soybean seed lots cv. 'Sari'.

Keywords: hydropriming, osmopriming, soybean seed

Introduction

Soybean [*Glycine max* (L.) Merr.] is one of the most important legume, rich in protein and oil, which can be used in agriculture and oil extraction industry. It contains up to 40% protein and 20% oil along with calcium, iron, carotene, thiamine, and ascorbic acid (Probst and Judd, 1973). In Iran, the soybean acreage is 95000 ha, producing 160000 metric tones, with an average yield of 1.7 t ha^{-1} (Abbasi *et al.,* 2009). The development plans of the Iranian Government and Ministry of Agriculture envisage oilseed cultivation as much as possible, specially, for soybean. To achieve this target, it is essential to provide high quality seeds to farmers (Abbasi *et al.,* 2009).

Soybean harvesting with minimum seeds losses, drying and storing them in a way that maintain quality until the soybeans are marketed is quite important (Sumner and Williams, 2006). Soybeans should be harvested promptly when they are mature to reduce field losses and minimize the chances of damage from bad weather. However, at this stage, seeds contain too much moisture for safe storage (Abbasi *et al.,* 2009). Soybean seeds can be harvested at moisture content as high as 30-45% if safe drying conditions are provided (Abbasi *et al.,* 2009). In general, soybean seeds are field dried to safe storage moisture content before harvesting. When weather conditions are favorable, this is a satisfactory system for safe storage, however, if conditions are unfavorable as frequently in main center of soybean seed production area in Iran, the soybean seed will deteriorate and harvest losses will increase. Now the contributions of artificial drying were became highlighted. Drying permits harvesting the grain as soon as it matures to avoid field losses and it places the seeds in a condition for safe storage and protecting them from heat damage and molds. In the other hand, priming technique can be a good option in order to improve soybean seed germination after drying.

The general purpose of seed priming is to hydrate the seed partially to a point where germination processes are begun but not completed (Ashraf and Foolad, 2005). Treated seeds are usually dehydrated before use, but they would exhibit rapid germination when re-imbibed under normal or stress conditions. Various seed priming techniques have been developed, but in this study it has been used hydropriming (soaking in water), and osmopriming (soaking in polyethylene glycol solution). Priming can increase the germination and growth of seedling under stressed conditions in oilseeds such as sunflower seeds (Kaya *et al.,* 2006). Similarly, in pigeon pea, hydropriming was determined to be very effective in the mobilization of compounds such as proteins, free amino acids, and soluble

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sugars from storage organs to growing embryonic tissues under salt stress (Jyotsna and Srivastava, 1998). Helsel *et al.* (1986) reported that, priming with PEG promoted rapid and uniform emergence in early plantings.

The aim of this research was to determine the effect of priming on germination traits of dried soybean seeds (natural and artificial) and to find the best combination of PEG concentration and time regime in osmopriming and/or time in hydropriming method.

Materials and methods

Sample preparation

This study was carried out at the Department of Agronomy, Faculty of Agriculture, University of Tehran, Iran. Seed samples of soybean cv. 'Sari' obtained from the University of Tehran Farm, were sealed in the plastic bags and stored in a cold place. Equilibration with ambient air was performed for at least 1 hour prior to every drying treatment. Seeds were hand harvested at three initial moisture content 45, 30 and 15% dry weight basis (d.b) in 2007. Germination and early seedling growth (8 days) of the cultivar were studied using distilled water for control and hydropriming and under osmotic potentials of -8, -10, -12 bar, for polyethylene glycol (PEG 6000) (Michel and Kaufmann, 1973).

Seed treatments

Before seed treatment with priming technique, seeds were dried in 3 harvest moisture with two methods including: natural and artificial methods (Tab. 1). For osmopriming, soybean seeds were immersed in osmotic solution at 25°C for 12, 24, 36 and 48 hours under dark conditions. Thereafter, the seeds were rinsed with distilled water three times. The treated seeds were surface-dried and dried back to their original moisture content at room temperature (about 25°C) for 24 hours. Hydropriming treatment was similar to osmopriming in priming period but it has been used, just distilled water instead of polyethylene glycol. Study was carried out only for 12 hours, because the germination was over 50% in 24, 36 and 48 h for hydropriming treatment.

Germination tests

Germination tests were performed with three 50-seed samples from each lot, in rolled papers (sandwich). Germination proceeded in a germinator at 25±1°C, in dark condition for 8 days (ISTA, 1999). Germination was considered to have occurred when the radicles were 2 mm long. Germination percentage was recorded every 12 h for 8 days. Germination rate was calculated as described in following formulae (ISTA, 1996):

$$GR = \frac{\text{No. of germinated seed at first count}}{\text{days of first count}} + \ldots + \frac{\text{No. of germinated seed at first count}}{\text{days of final count}}$$

Seedling length, seedling dry weight and vigor index were measured after the 8th day.

$$VI = \frac{\text{Germination percentage (\%)} \times \text{Mean seedling length (cm)}}{100}$$

Tab. 1. Characteristics of seed lots were used in this research

Number of seed lot	Characteristics
1	Harvest moisture 15%, natural drying in 25°C
2	Harvest moisture 30%, natural drying in 25°C
3	Harvest moisture 45%, natural drying in 25°C
4	Harvest moisture 15%, artificial drying with drier in 45°C
5	Harvest moisture 30%, artificial drying with drier in 45°C
6	Harvest moisture 45%, artificial drying with drier in 45°C

Statistical analysis

The statistical analysis was based on a randomized completely block design (RCBD); with three replications and 50 seeds per replicate. Data for germination percentage were subjected to arcsine transformation before analysis of variance was carried out with SAS software. Mean comparison was performed with Duncan's test if F-test was significant at ($P < 0.05$).

Results and discussion

Variance analysis of first experiment was investigated in order to studying hydropriming effect on soybean seed in different time (12, 24, 36 and 48 h). Since seeds germinated in more than 12 h hydropriming, times of 24, 36 and 48 h were eliminated. Analysis of variance for hydropriming in 12 h showed that, hydropriming of soybean seeds in this treatment in comparison with control group, decrease the speed of germination (data not shown). Kaur *et al.* (2005) showed that, produced plant from osmoprimed and hydroprimed seeds of pea possess better emergence, better emergence rate, better biomass, branches, flower numbers, sheath as well as better yield in comparison with control group seeds. They showed that, priming can increase these traits via changing the enzymes

Tab. 2. The ANOVA table showing the osmopriming treatment in comparison with control on germination traits of soybean seed (cv. 'Sari')

		Mean of Squares (MS)				
S.O.V	df	FGP (%)	GR (1/day)	SDW (gr)	SL (cm)	VI
Block	2	17.64 ns	0.0001 ns	0.003 ns	0.018 ns	0.28 ns
Treatment	77	724.4**	0.001**	0.004 ns	10.14**	13.73**
Error	154	11.81	0.0001	0.003	0.032	0.05
Coefficient of variation (CV)	-	4.52	4.39	7.35	2.34	3.11

ns,**,* Respectively non significant and significant of 1 and 5 percent of probability; FGP-final germination percentage; GR-germination rate; SDW: seedling dry weight; SL-seedling length; VI-vigor index of sucrose metabolism. In pigeon pea (*Cajanus cajan* L.), hydropriming was determined to be very effective in the mobilization of compounds such as proteins, free amino acids, and soluble sugars from storage organs to growing embryonic tissues under salt stress (Jyotsna and Srivastava, 1998). In contrast, lipid composition of Korean black soybean (*Glycine max*) seed, including percentages of neutral fats, glycolipids, and phospholipids, was determined to remain unchanged after soaking in water (Oh *et al.,* 1992). Probably, uncontrolled water uptake, causes seed coat weakening and vital electrolyte may leak which results, in germination interference (Jet *et al.,* 1996).

In the second experiment, which were done in order to evaluation of the best osmotic potential level as well as osmopriming duration, variance analysis of data showed that all of the germination traits of seed lots except seedling weight were effected by osmopriming treatments (Tab. 2). Mean comparison showed that seed lot features such as drying manner and seed harvest moisture affect germination traits. Totally, natural dried seeds and seeds with low harvest moisture possess better standard germination, better germination rate, better seedling length as well as more vigor index (Fig. 1 to 5).

Final germination percentage (FGP)

Osmopriming treatments improved final germination percentage, especially in artificial dried seeds with high moisture (Fig. 1). Regarding the germination percentage,

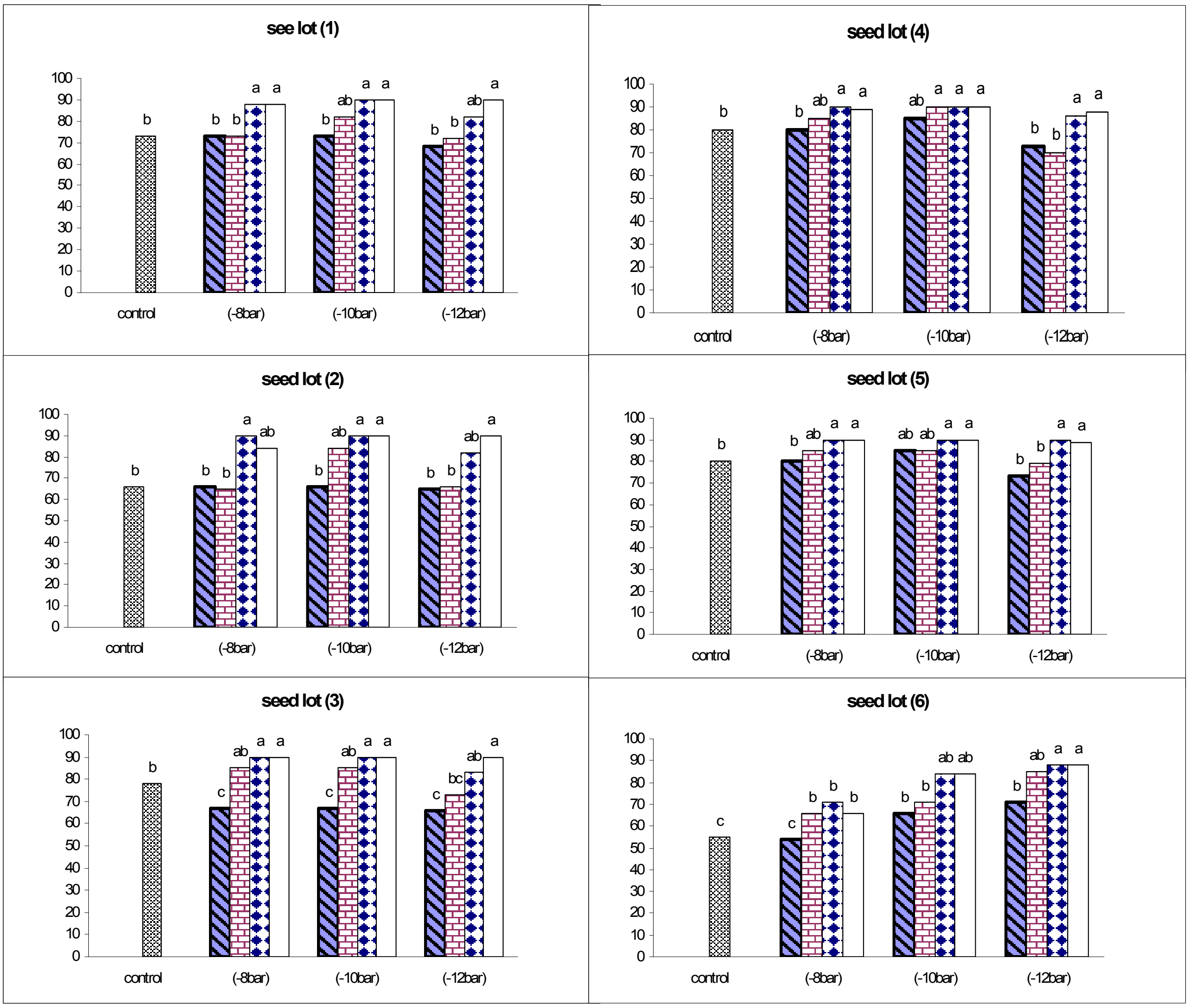

Fig. 1. Influence of osmotic potential levels of osmopriming treatments on the final germination percentage (FGP) in soybean seed lots (cv. 'Sari') under 12, 24, 36 and 48 hours durations. Horizontal axe shows priming with -8 bar; -10 bar; -12 bar; polyethylene glycol and 0 bar as a control group. Vertical axe is germination percentage (%)

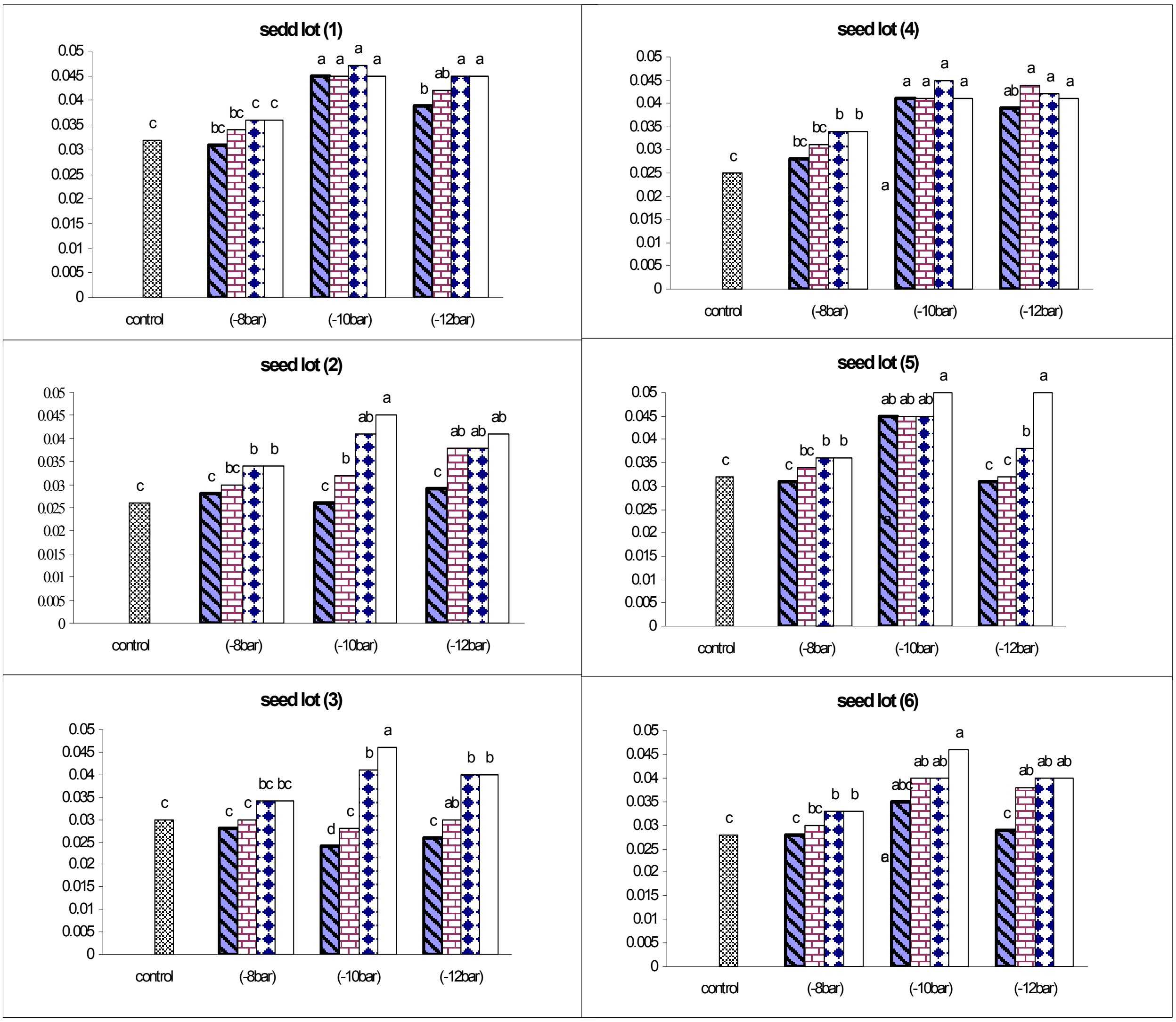

Fig. 2. Influence of osmotic potential levels of osmopriming treatments on the germination rate (GR) in soybean seed lots (cv. 'Sari') under 12, 24, 36 and 48 hours durations. Horizontal axe shows priming with -8 bar; -10 bar; -12 bar; polyethylene glycol and 0 bar as a control group. Vertical axe is germination rate

all of the seeds showed positive response to osmotic potential of -8 bar in 12 h except 2 and 6 seed lots. But in 2 and 6 seed lots which had the low control germination in comparison with the others, best responses were shown in treatments of -8 bar in 24 h and -10 bar in 12 h respectively (Fig. 1). Demir and Van de Vanter (1999) reported that, the osmopriming of watermelon seeds caused to decrease mean germination time and increase of its percentage. Most of researchers reported that priming can improve seed germination percentage (Misra and Dwivedi, 1980; Chiu *et al*., 2002).

Germination rate (GR)

The osmopriming levels and its duration effects on germination rate were significant (Tab. 2). Germination rate in seed lots were different significantly regarding to harvest moisture rate and drying manner (Fig. 1 to 5). Germination rate reaction to osmopriming treatment was similar in all of the seed lots regarding to osmotic potential and it was observed that seed lots have the maximum germination rate in the potential of -10 bar and 12 h (Fig. 2). Since germination rate possess an important role in the seedling establishment and in the growth improvement as well as crop yield, especially in the farm unfavorable conditions. It is recommended to use the potential of -10 bar in the time of 12 h in these cases. Basra *et al.* (2003) showed that in canola, germination percentage as well as germination rate were increased in response to priming. The suitable conditions of the beginning of germination ensure to the seeds quickly emerge. Subedi and Ma (2005) noted that one of the most important conditions for crop potential performance is fast and uniform germination which is a resultant of priming in the farm.

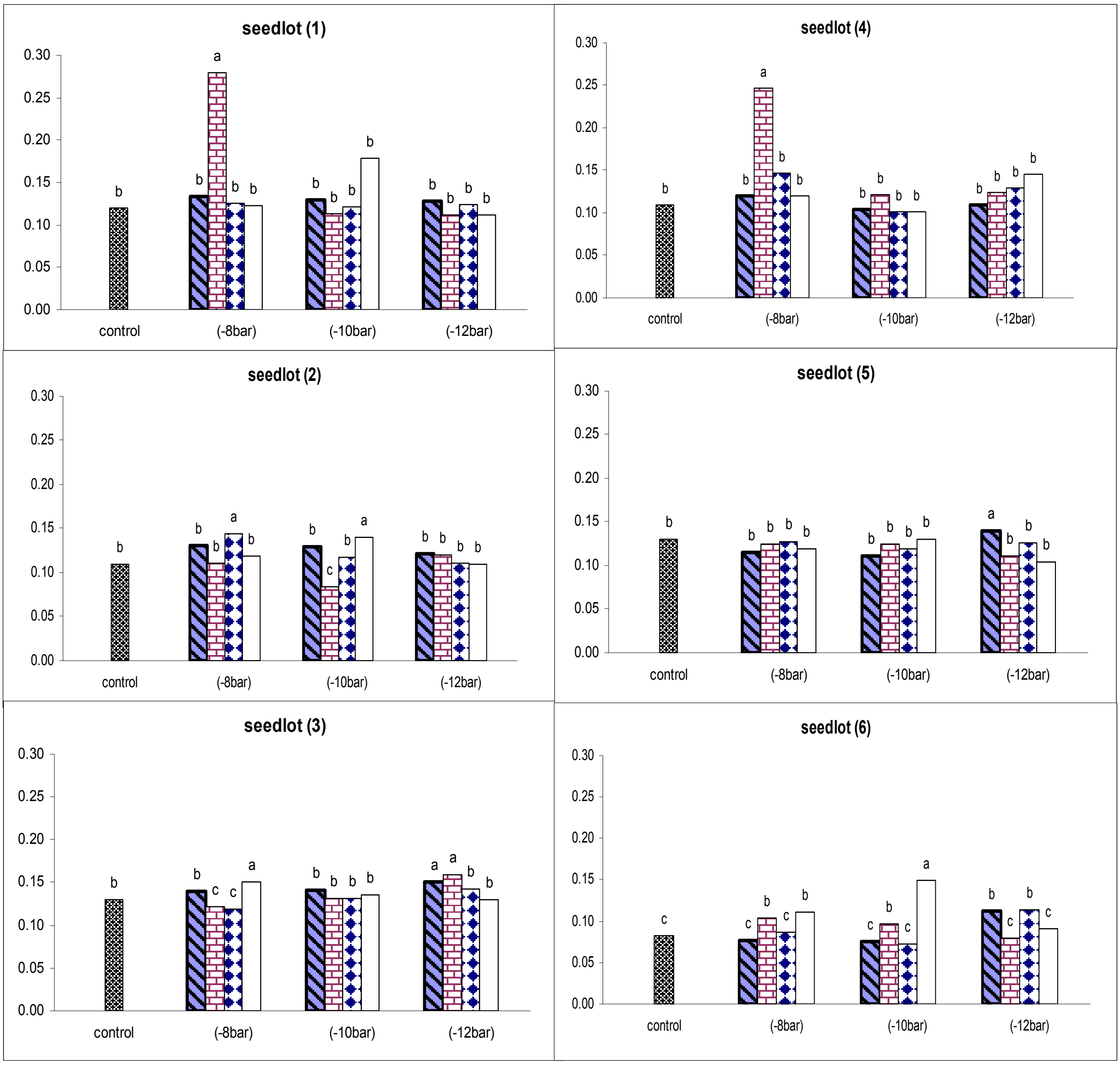

Fig. 3. Influence of osmotic potential levels of osmopriming treatments on the seedling dry weight (SDW) in soybean seed lots (cv. 'Sari') under 12, 24, 36 and 48 hours durations. Horizontal axe shows priming with -8 bar; -10 bar; -12 bar; polyethylene glycol and 0 bar as a control group. Vertical axe is seedling dry weight (gr)

Seedling dry weight (SDW)

Osmopriming effect on seedling dry weight was not significant and did not show any constant trend (Tab. 2). But naturally-dried seeds with the moisture of 15% had more seedling weight (Fig. 3). Khodadadi *et al.* (2003) studied the influence of priming on germination features of onion seeds in the salt stress condition and showed that dry weight of seedling was not affected by osmopriming with NaCl. It seems that because in seed lots with low germination the condition is more suitable for low number of seedlings; it is possible to produce seedlings with higher dry weight. Pretreatment of soybean seed with 0.25 M $CaCl_2$ for 24 h resulted in plants with increased seed number and reduced number of seedless pods, although it did not affect 100-seed weight (Eleiwa, 1989).

Seedling length (SL)

Osmopriming treatment effect on seedling length of seed lots was significant (Tab. 2). Also the comparison of means, showed that in treatment combination of -10 bar and 12 h this trait reached to its maximum rate (Fig. 4). This result was matched with the observation of Jet *et al.* (1996) but opposed with the observation of Afzal *et al.* (2002).

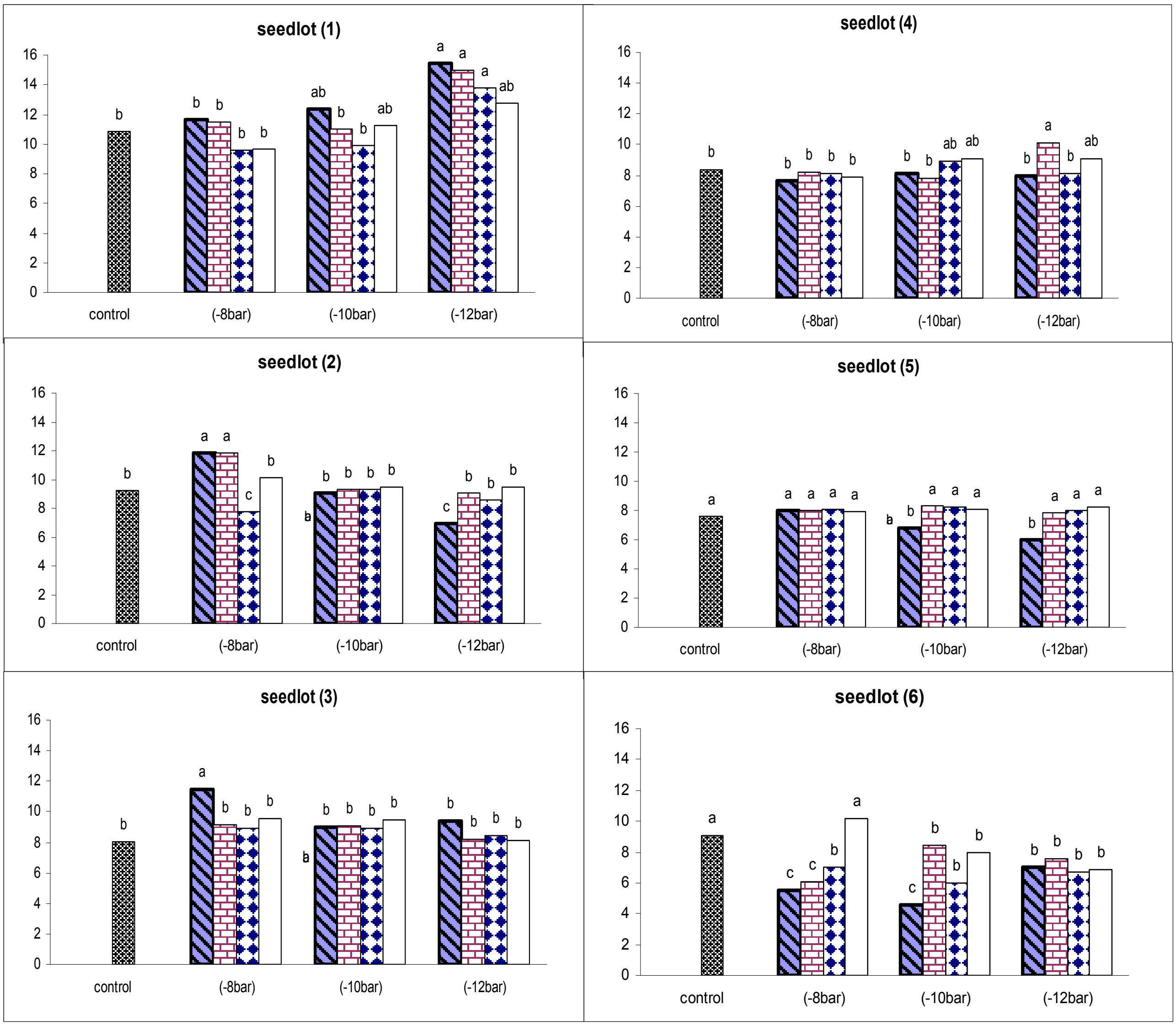

Fig. 4. Influence of osmotic potential levels of osmopriming treatments on the seedling length (SL) in soybean seed lots (cv. 'Sari') under 12, 24, 36 and 48 hours durations. Horizontal axe shows priming with -8 bar; -10 bar; -12 bar; polyethylene glycol and 0 bar as a control group. Vertical axe is seedling length (cm)

Vigour index (VI)

Vigor index amount decreased as a result of artificial drying and increase of initial moisture content. But after osmopriming treatment the difference between initial moisture was minimized. Faster seed germination made them to better use of environmental condition and increase of seedling length and vigor index as a result (Fig. 5). The result of means comparison showed that, in all of the seed lots except number 3, vigor index amount under the effect of osmotic potential -8 bar and time of 12 h was higher so, it is recommended in soybean. In seed lot number 3, also, osmotic potential increase to -10 bar can increase this trait outstandingly (Fig. 5).

Conclusions

Finally, another subject was regarded the study of recovery possibility or efficiency increase of soybean seeds with different quality via seed enhancement treatments. So, seed lots with different qualities from 'Sari' variety were chosen and hydro-and osmoprimed. Results showed that osmopriming treatment of -8 bar potential and time of 12 h was the best for final germination percentage, germination rate, seedling length and vigor index (some of the seed lots which had low primary germination were excepted).

This treatment has acceptable results and is more economic regarding to time human force and expenses in comparison with other treatments with the same results. Since seed water uptake gradually in the shortest period and lower PEG amount. Increase of germination rate is one of the important traits of seed germination which should be regarded in response to osmopriming treatment. So it is recommended to use -10 bar potential when seeds face adverse factors such as low viability or standard germination and/or unfavorable condition, late planting time, drought and stress condition. Probably, priming increase seedling

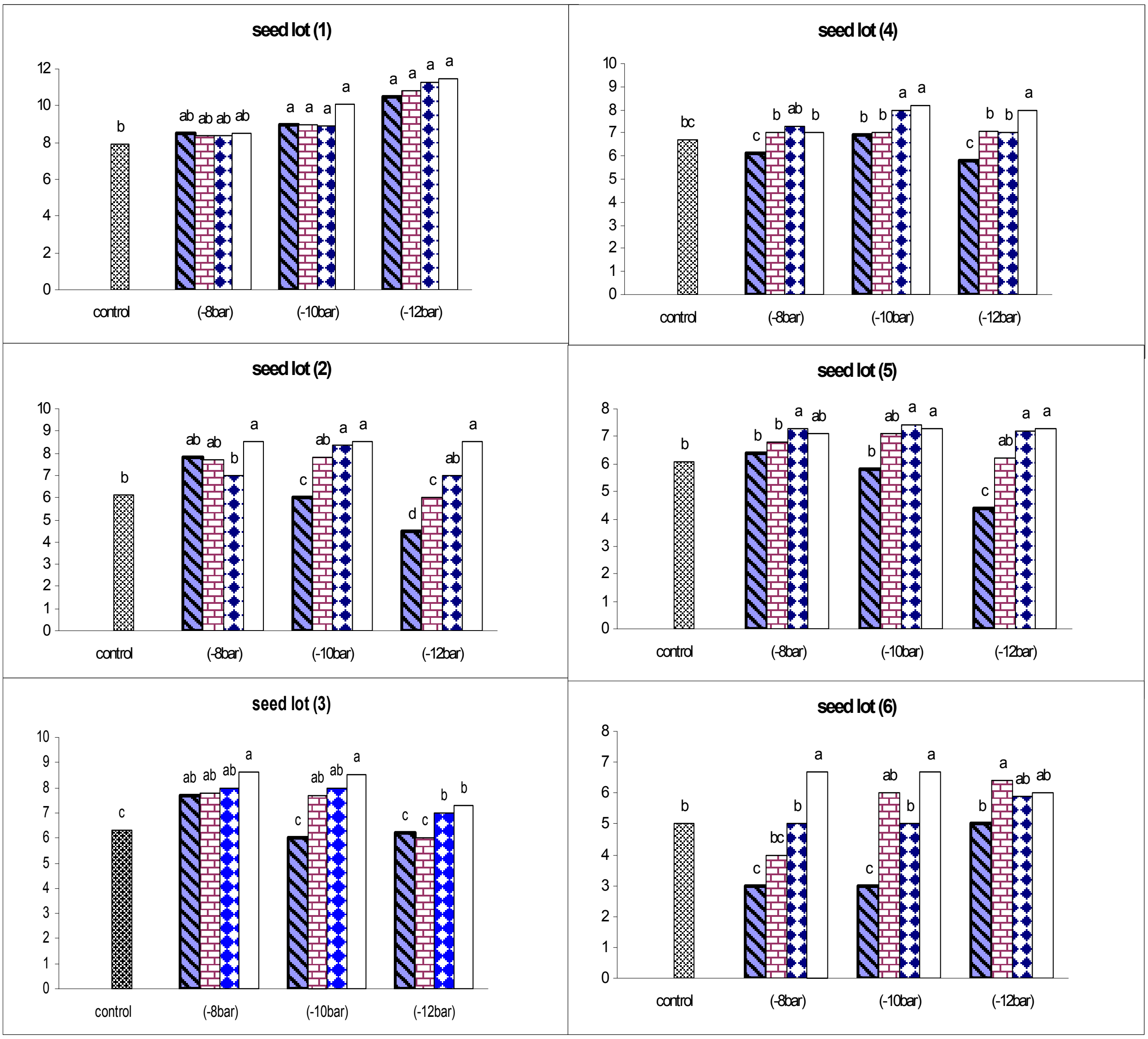

Fig. 5. Influence of osmotic potential levels of osmopriming treatments on the vigor index (VI) in soybean seed lots (cv. 'Sari') under 12, 24, 36 and 48 hours durations. Horizontal axe shows priming with -8 bar; -10 bar; -12 bar; polyethylene glycol and 0 bar as a control group. Vertical axe is vigor index

emergence, plant dry weight, its growth and yield as a result of increasing germination rate and seed lot efficiency with low viability. So, extra expenses of using PEG or other used material can be compensated. Although osmopriming improved germination traits of seeds, hydropriming decreased them. It is possible that water increase as well as uncontrollable water uptake causes seed coat weakening and vital electrolyte leakage which results in germination interference (Jet *et al.*, 1996).

Totally, evidence shows that many physiological mechanisms take part in priming process such as: reconstructing damages of seed cells (Bray, 1995), metabolic event development which happened in the second phase of water uptake that resulting radicle emergence (Dell Aquila and Bwely, 1989). As well as events such as better endosperm and stored materials partitioning which allow embryo to growth (Bourgas and Powel, 1984). Osmopriming in comparison with hydropriming can preserve plasma membrane structure and cause seeds to have better response regarding to germination traits because of controlled long hydration in seed (Jet *et al.*, 1996). Results of investigation in this research showed that the osmopriming of soybean seeds can improve the efficiency of seeds with low vigor and cause quick homogenous establishment of seedling intact.

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