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A study on seed water pre-treatment effects on, oil and protein percentage and some of agronomic traits in soybean in Iran

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Abstract

To study the Seed Water Pre-Treatment effects on Oil and Protein percentage and some of agronomic traits in Soybean, a research was carried out in Ardebil Islamic Azad University research farm, in 2008. This research was conducted in factorial based on complete block randomized design. One of the Water Pre-Treatment factors was 8, 12, 16 and 20 hours which were soaked in tap water and dried to 30percent moisture. A seed sample was also considered as an observation sample (without pretreatment). The second cultivar factor was Williams and LV (17). Results indicated that there is a significant difference at 1percent level between Water Pre-Treatment durations on Germination percentage, Seedling Weight, grain yield, oil percentage, weight of sub-stems and number of sub-stem. In most traits other than sub-stems weight and number of sub-stems, 8-hour Water Pre-Treatment provided the best yield. The results to the average comparison table indicated that Williams's cultivar had the most height with 8-hour Water Pre-Treatment. Also, there was a significant difference among cultivars on number of sub-stem and weight of sub-stems at 1percent and in most traits LV (17) had a better yields comparing to the Williams cultivar. Considering the results, see Water Pre-Treatment due to the short growth period and to increase the yield and better green in farm seems to be of significance. Also, 8-hour Seed Water Pre-Treatment is suggested for soybeans.

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Introduction

Huge efforts have been made to improve germination condition, seeds and seedlings growth strength for planting in certain environments, during recent years. Using enhancing treatments could improve seed strength, quick germination, uniform appearance and strong seedling deployment. Priming is among the main methods in enhancing germination strength. Seed priming is a treatment which is carried out before seed germination and a controlled amount of water is absorbed by the seed during this treatment (moosavi, 2012). Seed pretreatment is defined as various seed enhancement methods in which controlled seed irrigation is carried out (Faruq *et al.,* 2006 b). The main objective in seed priming is partial irrigation of seeds so that seeds pass the first (physical water absorption) and second (initiating biochemical and processes and sugar hydrolysis) stages of germination, while the third (sugars consumption by the embryo and seed growth) stage is avoided. (Bradford, 1995) Seed priming has various forms based on priming solution. The main common point between all solutions is related to determining the optimal concentration and proper storage duration. Also, seeds should not be immersed in the solution. Placing the seeds in the solution is recommended for 50% for oxygen exchange (Hardegree and Van Vactor 2000, Hardegree *et al*., 2002). Seven *et al* (2000) have reported water pretreatment as the best method in improving zucchini seed germination. During their studies on wheat seeds, Basra *et al* (2005) came to this conclusion that water pretreatment for 24hours has a great effect on germination speed. The effect of water pretreatment in increasing the wet weight of sunflower seedlings is reported to be more sensible comparing to osmotic pretreatment in osmotic stress conditions (Demir Kaya, 2006). Pill (1986) reported that by pre-germinating parsley seeds, shoot period decreases and shoot dry weight increases. Berg *et al* (1989) reported increase in crop due to soy seeds pretreatment. Rashed *et al.* (2006) claimed that barley seeds pretreatment could increase seed and biologic yields. In their researches, Moosavi *et al* (2011) claimed that applying water pretreatment seems to be vital due to short growth period, increase in yield and shoot.

Priming by water affects DNA and RNA synthesis, alpha-amylase activity and better development in embryo. Improvement in germination speed, uniformity in shoots, seedlings strengths and deployment could lead into plant growth (Basra *et al*., 2005; Ruan *et al*., 2002; Harris *et al*., 1999). It is reported that hydro-priming leads into improvement in germination in cotton seeds under stress and non-stress conditions (Casenave and Toselli, 2007). Mehra and Raaj (2002) reported improvement in shooting and seedling deployment in canola under stress conditions. Kaya *et al* (2006) reported a better germination and seedling growth in hydro-primed sunflower seeds under drought and salinity stresses. In addition, Ghassemi and Golezani (2008) reached a higher yield in pea seeds under 16hour hydro-priming treatment. Ghassemi and Golezani (2008) indicated that hydro-priming leads to increase seedling shoot speed and percentage, yield and yield components. They also reported that hydro-priming has a better effect on lentil seeds shoot speed and percentage on field, comparing to osmo priming. Berg *et al* (1989) reported increase in crop due to the pretreatment in soy seeds. In their research on wheat seeds, Basra *et al.* (2005) came to this conclusion that seeds water pretreatment for 24hours has a high effect on germination speed. Water pretreatment effect in increasing sunflower seedlings wet weight is more sensible comparing to osmotic pretreatment in osmotic stress (Demir Kaya, 2006). Pill (1986) reported that pre-germinating parsley seeds could decreases the shooting period and increases the shoot dry weight in the meantime. In a research on soybean, it was reported that 8hours of water pretreatment had the highest germination percent while 20-hour water pretreatment had the lowest germination percentage. In 20hour water pretreatments increased seed cotyledons and deterioration. Accordingly, the germination percentage was decreased (Moosavi *et al*., 2011).

Since there is not enough comprehensive data on

Seed Water Pre-Treatment application and effects on soybean cultivars primary growth and yield, the following research tries to study Seed Water Pre-Treatment effects on Oil and Protein percentage and some of agronomic traits

Materials and methods

Materials

To study the water pretreatment effects on soybeans vegetative and reproductive traits and the correlation between these traits and seeds yield, a research was conducted in Ardabil IAU research farm based on a factorial randomized complete block design. The first factor was the seed water pretreatment including control, 8, 12, 16 and 20hours, the second factor were the cultivars of Williams and LV_{17}. The Williams and LV_{17}soybean seeds were provided from Moqan Agricultural Research Center and divided into five equal parts and a sample with a most of around 10percent as the control group was kept in a plastic pack in a refrigerator at 3 to 5° C. the other four samples were watered with distilled water for 8, 12, 16 and 20hoiurs in and incubator at 17.3° C. subsequently, pretreated seeds were spread on a table at the laboratory environment at 20 to 22° C to reach the moist of 30oercent. To determine the seeds moist two 5gr replications from each treatment were separately pounded in porcelain containers to turn into granules. Beaten samples were weighed again and put in oven for at 130° C, for an hour. Subsequently, samples were taken out of the oven and weighed. The seeds moist percentage was calculated through the following formula:

Seeds Moist Percentage = Samples Wet Weight- Samples Dry Weight/ Samples Wet Weight × 100

Methods

Three 20-seed replications were randomly separated from each sample. Seeds for each replication were put on a wet filter paper and another wet filter paper was put on them. Papers where folded 2 to 3cm to the paper end and tubed. The tubed papers for each sample was kept in a plastic pack and put in a beaker at 45degree angle to be kept in an incubator for at 10° C. after 24hours, seeds for all treatments were separately taken out of germinator. After counting germinated seeds on the same day, they were folded in filter papers and kept in the incubator. Emergences of roots with 2mm were counted as a criterion for seeds germination. Germinated seeds were counted daily, for 10days in a row. At the end of the research, the normal and abnormal buds number was determined. After germination test, seeds normal seedling was separated. Roots and shoots were cut from the seeds junctions and dried in an oven at 70° C for 24hours. Seedlings were weighed and the seedlings dry weight average was recorded.

The field research was conducted at Ardabil Research Filed in 05.29.2008. Seeds density was considered to be 20seeds in square meters. During crop ripening, to determine the sub-stem weight and number of sub-stems, 10plants from each treatment were randomly harvested. The sub-stem weight and number of sub-stems were measured. The final harvests of each research unit were finished when the moist reached to 17percent. In this stage, the plants available in 1square meter in each plot were harvested. Seeds were separated from the pod. And the seeds yield was recorded.

Data analysis

All statistical analyses and means comparison were done by SPSS software. Diagrams were drawn by Excel software.

Results and discussion

Germination Percentage

Results to the pretreatment effect on germination percentage variance analysis was significant at 1percent. (Table 1) Results to the mean comparison suggested that controlled and 8, 12 and 16hour pretreatments had the highest germination percentage in the same level and the 20hour pretreatment had the lowest germination percentage. (Table 3) In 20hour pretreatment, most seeds cotyledon exhaustion happened and as a result, germination percentage was decreased. Bosra *et al* (2006) reported that water pretreatment increase low-quality rice seeds germination, so that, water

pretreatment lead to an increase from 38percent of controlled group germination to 77percent in pretreated seeds. Gary *et al* (1991) claimed that the germination improvement influenced by seeds pretreatment is due to the activation of repair mechanisms and metabolic process which occur during water absorption. Basra *et al* (2005) on a study on wheat seeds came to this conclusion that seeds water pretreatment for 24hours had a high effect in accelerating germination. Also, Casiro *et al* (2004) came to this conclusion that water pretreatment is the most effective method for improving germination in onions. In wheat, water and metric pretreatments on germination rate and percentage were higher than NaCl Osmotic pretreatment. (Basra *et al*, 2005).

Table 1. Analysis of variance for the evaluated traits at different Seed Water Pre-Treatment levels in Williams and Lv_{17} cultivars in Laboratory in 2008.

Source of Variations	df	Mean Square	
		Germination percentage	Seedling Weight
Cultivar	1	414.408 ^{ns}	0.003 ^{ns}
Seed Water Pre-Treatment	4	673.346 *	0.217 *
C*SWPT	4	160.129 ^{ns}	0.016 ^{ns}
Error	20	129.908	0.016
CV (%)		13.07	11.24

* and ** Significantly at $p < 0.05$ and < 0.01, respectively.

Seedling Weight

Considering the variance analysis table for studied factors, except seed pretreatment, all factors and their interactions showed no significant differences on seedling weight. (Table 1) Considering mean comparison table (Table 3) controlled and 8 and 12hour water pretreatments had the highest seedling weight in the same level. In controlled and 8hour water pretreatments, germination time, healthy seed and its high strength could lead into the increase in seedling weight. Alvord and Bradford (1988) came to this conclusion that tomato seeds pretreatment increase the seedling growth and this supremacy is preserved during growth period for a long time. This increase in plants strength in pretreated seeds is related to their germination and uniformity. Basra *et al* (2006) believe that the increase in seedlings weight in pretreated wheat seeds is due to the high germination rate and uniformity which lead to the quick deployment of seedlings. The main seed pretreatment effect on seedling's growth is due to the quicker germination which leads the seedling to have longer growth period. Faruq *et al* (2006) reported that rice pretreated seeds seedlings dry weight is significantly higher than the control group seedlings. They came to this conclusion that the highest seedling weight was due to the 48hour water pretreatment. Other findings on have been reported on the relation between seedlings weight increase due to the water pretreatment. (Basra *et al*, 2002; 2005) Water treatment effect in sunflower seedlings wet weight is more considerable, comparing to the osmotic pretreatment in osmotic conditions. (Demirkaya, 2006).

Number of Sub-Stems

There was no significant relation found between replication effects and treatment interaction on cultivar on number of sub-stems. However, considering the variance analysis table (Table2) cultivar and treatment effects on this trait were significant. Among the cultivars, $LV_{(17)}$ cultivar had the most number of sub-stems and among the treatments, the 20-hour seed aqueous pretreatment had the most number of sub-stems. It should be mentioned that control treatments had no significant difference in 8 and 12-hour treatments. The reason to this result could be the plant strength in control treatments of 8 and 12-hour aqueous pretreatment and low plant density in area unit in 20-hour

pretreatment. Kaur *et al* (2002) have reported increase in amylase enzymes and sucrose synthase in shoot and root of treated seedlings. They claimed that priming leads to the increase in amylase enzymes activity and converting the savings substances into transitional substances and as a result increase in plant growth.

Table 2. Analysis of variance for the evaluated traits at different Seed Water Pre-Treatment levels in Williams and Lv_{17} cultivars in 2008.

Source of Variations	df	Mean Square				
		Number of Sub-Stems	Weight of Sub-Stems	Oil percentage	Protein percentage	Grain yield
Replication	2	1.213^{ns}	0.245^{ns}	0.196^{ns}	2.359^{ns}	49409.085^{**}
Cultivar	1	5.208^{**}	7.792^{**}	0.768^{ns}	0.481^{ns}	17079.941^{**}
Hydro-Priming	4	3.737^{**}	2.917^{**}	3.644^{**}	6.686^{ns}	19956.800^{**}
C * H-p	4	0.047^{ns}	0.161^{ns}	0.197^{ns}	3.354^{ns}	1456.316^{ns}
Error	18	0.368	0.429	0.357	76.022	2975.028
CV (%)		13.84	10.11	3.99	4.70	16.66

* and ** Significantly at $p < 0.05$ and < 0.01, respectively.

Weight of Sub-Stems

There was no significant relation found between replication effects and treatment interaction on cultivar on weight of sub-stems (Table 2). Since this trait is dependent on weight and number of stems, they have more correlation so their results are similar to number of sub-stems. Data mean comparison shows that among the cultivars, LV $_{(17)}$ has the highest weight of sub-stems and among the treatments, the highest weight is related to 16 and 20-hour aqueous pretreatment.

Table 3. Comparison of Means of traits at different Seed Water Pre-Treatment levels in Williams and Lv_{17} cultivars in Laboratory.

Seed Water Pre-Treatment levels	Characters	
	Germination percentage	Seedling Weight (gr)
Without pretreatment	87.37 ab	0.6202 ab
8 hours	97.82 a	0.7902 a
12 hours	93.02 a	0.7030 ab
16 hours	88.05 ab	0.5773 b
20 hours	69.83 b	0.2882 c

Differences between averages of each column which have common characters are not significant at probability level of 5%.

Hydro-priming affects DNA and RNA synthesis, ATP availability, alpha-amylase activity and embryo's better growth. Hence, germination better rate, growth consistency, seeding vigor and deployment leads to better plant growth. (Basra *et al.*, 2005; Ruan *et al.*, 2002; Harris *et al.*, 1999).

Grain Yield

According to the variance analysis table (Table 2) there is a significant difference between economic yield in replications, cultivars and treatments. The interaction between cultivar and treatment is not significant in this trait. Average comparison results show that among the cultivars, LV $_{(17)}$ has the highest

economic yield and among the treatments, 8-hour aqueous pretreatment has the highest yield. It should be mentioned that there was no significant difference found between 8-hour and 12-hour treatments. 20-hour aqueous treatment had the lowest economic yield. Pod number, plant height and grain dry weight are among the factors which could affect the yield. In aforementioned traits also, 8-hour aqueous pretreatment had the highest yield. Rashed *et al* (2006) reported that barley seed pretreatment could increase the yield up to 53percent. Increase in seed yield has been observed in corn and rice seeds due to the seed pretreatment. Faruq *et al* (2006) and Harris *et al* (1999) believe that the rice seed yield due to the pretreatment is a result of growth percentage improvement and yield execution such as seed weight. Ghassemi- Golezani *et al* (2008) showed that hydro-priming could lead into seedling growth rate and percentage and also yield and yield components. Kahlon *et al* (1992) Hussain *et al* (2006) reported higher seed yield in hydro-primed seeds of sunflower and wheat, respectively. Moreover, Ghassemi-Golezani *et al* (2008) obtained a higher pea seed yield in 16-hour hydro-priming.

Table 4. Comparison of Means of traits at different Seed Water Pre-Treatment levels in Williams and Lv_{17} cultivars.

Hydro-Priming levels	Characters			
	Number of Sub-Stems	Weight of Sub-Stems (gr)	Grain yield(gr/m2)	Oil percentage
Without pretreatment	3.86 BC	5.88 C	326.7 ABC	15.48 AB
8 hours	3.56 C	5.88 C	409.6 A	15.83 A
12 hours	4.15 BC	6.24 BC	348.3 AB	15.08 AB
16 hours	4.83 AB	7.10 AB	297.3 BC	14.53 BC
20 hours	5.52 A	7.33 A	255.3 C	13.87 C

Differences between averages of each column which have common characters are not significant at probability level of 5%.

Harris *et al* (1999) reported that hydro-priming results in corn, pea and Upland rice better seedling deployment and vigor which increases the growth, flowering maturity and yield.

Oil and Protein Percentage

The data variance analysis related to the oil percentage (Table 2) indicates that all effects except the treatments effects are not significant. Data mean comparison show that among treatments, 8-hour aqueous pretreatment, control treatment and 12-hours aqueous pretreatment had the highest oil percentage and 20-hour aqueous pretreatment had the lowest oil percentage. According to the variance table (Table 4) the protein percentage in replication, cultivar, treatment and cultivar treatment interaction was not significant. This shows that treatments are not effective in protein yield. Ashrafi and Razmju (2009) in a study on safflower claimed that 6 hours of hydro-priming could improve the hydro-primed seeds physiologic and biochemical characteristics and this leads to increase in oil and protein in seeds. Hydro-priming results in better growth a plant system protection against tension and increase in oil and protein amount. Seeds priming affects DNA and RNA synthesis and also improves the embryo's growth. (McDonald, 2000) Results from this research show conformity with previous studies in oil percentage while the protein percentage was not in accordance with previous studies.

Conclusion

According to the results in this research, due to the growth short period and using seed aqueous pretreatment in increasing the yield and improvement in growth, the activities before seed

aqueous pretreatment seem to be of essence and the 8-hour aqueous pretreatment is recommended for soybeans.

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