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EFFECT OF SEED PRE-TREATMENT WITH GROWTH REGULATORS ON SEED YIELD AND YIELD COMPONENTS OF COMMON BEANS (*Phaseolus vulgaris* L.)

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ABSTRACT

Seed priming is employed for better crop stand and higher yield in a range of crops. In order to evaluate the effect of seed treatments with growth regulators on the yield and yield components of common bean (*Phaseolus vulgaris* L.) lines, a factorial experiment was carried out based on a randomized complete block design with three replications at two years. In this research, pre-treatment (priming) of two red bean (D81083 and KS31169) seeds with growth regulators including salicylic acid (SA) and naphthalene acetic acid (NAA) at four levels (P0:control, distilled water; P1: 0.5 m mol L^{-1} NAA; P2: 0.7 m mol L^{-1} SA; P3: combination of SA and NAA hormones at rates of 0.5 and 0.7 m mol L^{-1}) were studied. The results showed that the effect of line in all of the characteristics and seed priming with growth regulators in all of the characteristics except to harvest index were significant. The highest number of grains per pod and biological yield (14602 kg ha^{-1}) due to growth regulator application was obtained from line D81083 in 0.5 m mol L^{-1} NAA. Generally, the use of growth regulators as a seed pre-treatment increased the yield and yield components of red beans.

Keywords: common bean, naphthalene acetic acid, *Phaseolus vulgaris*, priming, salicylic acid

INTRODUCTION

Fabaceae family is one of the most important sources of protein and energy. Food legumes, after maize, wheat and rice, are the most important crops used for feeding the world's people, especially in developing countries (Bagheri et al., 2001). By having 17-40% protein, legumes play an important role in the production of protein and calories needed by humans (Beebe et al., 2015). Common bean (*Phaseolus vulgaris* L.) is a tropical food legume that was derived from temperate legumes in the Tertiary period (van Schoonhoven and Voysest 1991; Ghanbari et al., 2015). Although many varieties of beans have high yield potential in a wide range of environments, yield potential results from the plant adaptation to the conditions in the growing season, photoperiod and crop management practices (Bagheri et al., 2001).

Germination as the first stage of plant development is one of the critical stages in the life cycle of plants and is a key process in the emergence of seedling (De-Villiers et al., 1994). One way to increase germination and seed emergence, especially under stress conditions, is the use of priming. Seed priming is practicing water treatments (sometimes other materials are associated with water) on the seeds before planting to enhance germination, initial establishment, early maturity, and improvement of plant growth, which consequently results in the acceleration and uniformity of germination and increase of crop quality and quantity (Toselli and Casenave, 2007). Seed priming techniques are used to improve germination, to reduce the time between sowing and emergence, and for uniform emergence in the field, especially under adverse environments (Gupta et al., 2008).Several studies have been conducted on physiological and biochemical effects of priming on various species of legumes such as cowpea, pea and lentil which have shown that seed priming can improve germination (Ghasemi Golezani, 2008). In environments that are faced with adverse conditions, the use of seed priming can significantly reduce product loss (Abolrahmani et al., 2010). Plant growth regulators are important factors affecting plant growth and development. Salicylic acid (SA) is a phenolic compound that is produced in plants by root cells. This substance exists in plants in small amounts (mg g^{-1} fresh weight or even less). Salicylic acid plays a central role in the regulation of various physiological processes during plant growth and development such as ion adsorption, photosynthesis, and germination, depending on the concentration used, species, growth period and environmental conditions (Raskin 1992; Iqbal et al., 2006).

Naphthalene acetic acid (NAA) is another plant growth regulator and an important synthetic auxin used in plants. Auxins have a wide variety of impacts on plants and their of other growth regulators. Different effects of the application of NAA in the acceleration of rooting, control of flowering, prevention of fruit drop and increase of fruit formation have been observed in different plants (Prakash and Ganesan, 2001).

Roohi et al. (1991) reported in a study on seedling establishment that priming the seeds of pasture grass with auxin increased its establishment under drought stress conditions because auxin decreased the time interval between germination and adventitious root formation. Da Silva et al. (2005) found that NAA could affect some growth indices in sesame. Moreover, growth stimulators such as NAA can be useful in reducing the flower loss and enhancing the transition into the reservoir (Prakash et al., 2003). Given the positive role of growth regulators in crops and since little research has been conducted on the application of growth regulators as pretreatment for legumes such as beans, the present research was conducted to determine the best hormonal pretreatment of seeds for the optimal use of environmental resources by plant and for the achievement of higher yield and yield components in two lines of red bean.

MATERIALS AND METHODS

The research was conducted in the Bean Research Station of Khomein, Khomein, Iran. The station is in the village of Khorramdasht 8 km from Khomein-QoorchiBashi Road, (49°57' E,33°2' E and 1930 m above the sea level) in the west of Khomein Town, Iran.

A factorial experiment was carried out as Randomized Complete Block Design(RCBD) with three replications at two years (2013-2014 and 2014-2015 cropping seasons). The research consisted of two common beans (*Phaseolus vulgaris L.*) having red seeds (KS31169 and D81083) and seed pre-treatment with growth regulators containing salicylic acid (SA) and naphthalene acetic acid (NAA) in four levels including P0: control, pretreated with distilled water; P1: pretreated with 0.5 m mol L^{-1} NAA; P2: pretreated with 0.7 m mol L^{-1} SA; P3: pretreated with combination of 0.5 and 0.7 m mol L^{-1} SA and NAA hormones. For priming, after the preparation of different doses of SA and NAA, bean seeds were submerged under different treatments for six hours at a temperature of 4°C. Then the seeds were dried at the room temperature and planting operations began in the field as direct cultivation.

The distance between the rows was 50 centimeter and the distance between plants in each row was 5 cm. The length of each plot was 6 meter and its width was 3 m and 6 rows were planted in each treatment. To weeds control, Trifluraline was used by two liters per hectare before planting. During the vegetative growth stage, 50 kg ha^{-1} of urea fertilizer and 1 kg ha^{-1} of iron fertilizer were used with irrigation water before flowering. Weed control in different stages of growth and development of beans was done by manual weeding. Pest and diseases were fought during the growing season in accordance with the technical recommendations.

At the end of the growing season, after physiological maturity of grains 2 m^2 of each experimental unit was harvested after the removal of the margin from both sides and biological yield was measured in kg as well as yield components including the number of pods per plant, number of grains per pod, and 100-grains weight.

MSTATC and Excel software were used in this study for statistical analysis. In order to ensure the accuracy of tests and data, variance homogeneity test was performed. The means were compared using Duncan Multiple Range Test at 5% and 1% levels of probability.

RESULTS AND DISCUSSION

Number of pods per plant

The ANOVA results showed that the effect of lines and seed priming with growth regulators on the number of pods per plant were statistically significant (P1%) in Table 1. KS31169 had higher number of pods per plant than that of D81083. Mean comparison of hormonal priming levels showed that the highest number of pods per plant belonged to priming with (NAA+SA) and the lowest number belonged to the control (Table 2).

The exact mechanism of salicylic acid action is not clear yet, but it is likely that like auxin, salicylic acid is involved in the elongation and division of cells (Singh, 1980). Given that salicylic acid is known as a pseudo hormonal substance, it seems that it increases the number of pods per plant by affecting vegetative and reproductive meristems. The results of the research are consistent with the findings of some researchers such as Aldesuquy and Ibrahim (2000). They showed that the increase of yield and yield components in bean resulted from the seed priming and the increase of number of pods per plant, pod length, and the number of grains per pod.

Table 1. Analysis of variance of hormonal treatments and lines effects on seed yield and yield components of red beans.

Source of variation	d.f.	Mean squares					
		Number of pods per plant	Number of grains per pod	100-grain weight	Biological yield	Grain yield	Harvest index
Year	1	$0.15^{n.s}$	1.76^{**}	$1.98^{n.s}$	$80296.52^{n.s}$	$551857.05^{n.s}$	$35.46^{n.s}$
Replication (year)	4	0.65	0.03	0.5	27252.76	438725.56	31.48
Line	1	1743.15^{**}	2.25^{**}	811.47^{**}	6280472.02^{**}	12817384.32^{**}	539.75^{**}
Priming	3	79.243^{**}	0.77^{**}	26.99^{**}	28105665.69^{**}	1961850.33^{**}	$4.38^{n.s}$
Line×Priming	3	$1.91^{n.s}$	$0.03^{n.s}$	8.51^{**}	3187048.9^{**}	$17794.63^{n.s}$	$12.56^{n.s}$
Year×Line	1	$0.86^{n.s}$	$0.001^{n.s}$	$0.38^{n.s}$	$60379.87^{n.s}$	$128038.28^{n.s}$	$8.67^{n.s}$
Year×Priming	3	$0.08^{n.s}$	$0.051^{n.s}$	$0.22^{n.s}$	$243786.35^{n.s}$	$131003.55^{n.s}$	$13.82^{n.s}$
Year×Priming×Line	3	$0.25^{n.s}$	$0.01^{n.s}$	$0.57^{n.s}$	$252634.7^{n.s}$	$43794.57^{n.s}$	$3.31^{n.s}$
Error	28	1.28	0.05	0.51	261958.7	65665.31	5.29
CV%		6.71	6.14	2.07	4.16	7.44	8.21

ns, * and ** are non-significant, significant at the 5% and 1% levels of probability, respectively.

Table 2. Mean comparisons of lines**,** hormonal treatments and their interaction in seed yield and its components.

Treatment		Number of pods per plant	Number of grains per pod	100-grain weight (g)	Biological yield (kg ha^{-1})	Grain yield (kg ha^{-1})	Harvest index (%)
Line	KS31169	22.94 a	3.8 a	30.37 b	12644.4 a	3960.43 a	31.35 a
	D81083	10.89 b	3.44 b	38.6 a	11920.99 b	2926.94 b	24.65 b
Hormonal priming	Distilled water (DW)	13.73 b	3.25 b	33.86 bc	12028.55 b	3282.78 b	
	NAA	15.95 ab	3.47 ab	36.20 a	14491.96 a	4000.24 a	
	SA	18.91 a	4.02 a	35.12 ab	11468.94 b	3360.4 ab	
	NAA+SA	19.08 a	3.59 ab	32.75 c	11013.74 b	3031.31 b	

Different letters within each column indicate significant difference using DMRT test at p≤0.05.

Number of grains per pod

The ANOVA results showed that the effect of years, lines and hormonal priming levels and their interactive effect on the number of grains per pod were significant (P 1%) (Table 1). KS31169 had higher number of grains per pod. Mean comparison of hormonal priming levels showed that was not significantly different from another pretreated. Salicylic acid had the highest number of grains per pod and other hormonal pretreatments were not significantly different from each other and the lowest number of grains per pod belonged to seed pretreatment with control treatment (Table 2).

It is possible that application of salicylic acid increase available carbohydrates to be transferred into storage organs so that the rate of abortion decreases and the number of grains per pod increases (Gunes et al., 2005). The number of grains per pod determines the plant storage capacity. In other words, the higher number of grains as the plant reservoir is larger for assimilate production and whatever factor increases this feature will in fact increase the yield (Hanson et al., 2001).

Subedi and Ma (2005) showed that the number of seeds and plant yield in primed seeds was more in comparison to unprimed seeds. Harris et al. (2005) reported that seed priming increased the number of grains. Rashid et al. (2004) reported the increase of the number of grains in pod resulting from the primed seeds.

100-grain weight

The ANOVA results showed that the effect of different red bean lines and hormonal priming levels and their interactive effect on 100-grain weight were significant (P 1%) as shown in Table 1. D81083 had a higher weight than that of KS31169. In examining the interactive effect of lines and priming levels, the highest weight of 100-grain belonged to D81083×NAA and the lowest belonged to KS31169×compound priming (NAA+SA) (Table 3). Application of naphthalene acetic acid might increase grain filling rate or period and the reservoir power plays the key role in this regard. Kaur et al. (2005) have referred to the increase of reservoir power due to the increasing activity of invertase and sucrose synthase enzymes in pod walls of plant resulting from primed seeds of pea which improves grain filling.

Farooq et al. (2006) believed that the increase of grain yield in rice due to the seed priming results from the effect of this treatment on the increase of the number of pods per fertile tiller and 1000-grain weight. Harris et al. (2005) have also reported the increase of 1000-grain weight due to hormonal priming of the seed.

Table 3. Mean comparisons of lines × hormonal priming interaction in 100-grain weight and biological yield.

Treatment		100-grain weight (g)	Biological yield (kg ha^{-1})
Line × Priming	KS31169 × DW	29.08 e	12179.14 b
	KS31169 × NAA	31.42 d	14381.88 a
	KS31169 × SA	31.27 d	12684.77 b
	KS31169 ×(NAA+SA)	29.72 de	11331.97 bc
	D81083 × DW	38.64 b	11877.95 bc
	D81083 × NAA	40.98 a	14602.05 a
	D81083 × SA	38.98 ab	10508.47 c
	D81083 × (NAA+SA)	35.78 c	10695.51 c

Different letters within each column indicate significant difference using DMRT test at $p \leq 0.05$.

Biological yield

The ANOVA results showed that red bean lines and the effect of hormonal pretreatment and the interactive effect of line × hormonal pretreatment on biological yield were significant P1% (Table 1). Mean comparison of the interactive effect of lines and hormonal pretreatments showed that the highest biological yield belonged to line D81083 with NAA which was not significantly different from hormonal pretreatments with NAA to line KS31169; the lowest belonged to D81083 with SA pretreatment (Table 3). Among the effects of application of auxin and its role in photosynthesis process can be referred to which affects biological yield and the rate of assimilates. Among the indirect effects of auxin the delay in leaf aging can be referred to which increases the rate of assimilate mobilization and net production of photosynthesis during the growing season by increasing leaf area durability and consequently the growth rate will be higher due to high photosynthesis (Prakash et al., 2003).

In primed seeds of mung bean 80% of biomass increase was observed in comparison with the control seeds (Rashid et al., 2004). In tissues of plant meristem areas resulting from the primed seeds of bean the increased activity of invertase acid was reported which increased plant growth and biomass (Kaur et al., 2005). Farooq et al. (2006) referred to the effect of seed priming on the increase of grain yield and straw of wheat after rice. In studies conducted by Ali et al. (2008), seed priming increased grain yield and biological yield.

Grain yield

The ANOVA results showed that the effect of different red bean lines and hormonal pretreatments on grain yield was significant (P 1%) (Table 1). D81083 had the highest yield and NAA produced the highest yield which was not significantly different from hormonal pretreatments with SA (Table 2).

It was found that on-farm priming using hormonal regulators is a successful method in enhancing the seed yeild and biomass of different crops (Rehman et al. 2011). It is believed that auxins such as NAA increase duration of vegetative growth and maturity, so might have increased the transfer of assimilates into grains and ultimately might have increased the grain weight. On the other hand, the factors that control assimilates transfer into destination, also control assimilate distribution and by influencing enzyme activity and flexibility of target cells, hormones have a significant effect on the distribution of assimilates (Meyyappan et al., 1991). Foliar application of auxin and gibberellins in medicinal pumpkin plants showed that auxin increased the number of female flowers and the number of fruits per plant (Sedghi et al., 2008). This can lead to the increase of plant yield due to the increase of the number of fruits. . Ghodrat (2000) observed that the effects of auxin and gibberellin increased photosynthesis, accelerate flowering, and delayed tissues aging, and resulted in increasing of the yield. Some reports have been published about the effect of salicylic acid on the increase of yield in some crops such as soybean, cowpea and peas (Prakash et al., 2003).

Harvest index

The ANOVA results showed that the effect of different red was significant P 1% (Table 1). Harvest index value was higher in D81083 than that KS31169 (Table 2). Harvest index is an important physiological index reflecting the percentage of assimilates mobilization from vegetative organs of plant into grains. It was reported that seed priming in sunflower improved dry matter partitioning to the grain and increased harvest index and grain yield (Hussain et al., 2006). The increase of harvest index due to seed priming could result from stimulation of further transmission of assimilates into the pods and consequently the increase of grain yield. Researchers have shown in different experiments that priming leads to the increase of harvest index and grain yield (Farooq et al., 2006).

In conclusion, the results of this study showed that priming technique with the use of hormonal substances can increase yield and yield components in beans. Reservoir size (number of grain endospermic cells) and reservoir activity are the major determinant of final grain weight. On the other hand, hormones could be one of the factors that regulate the reservoir activity. Therefore, understanding the relationships and analyzing them may be effective in interpreting the changes of grain yield of beans through external use of hormones.

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