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Effect of irrigation cutoff at seed formation stage on seed yield and germination indices of sugar beet seed

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ABSTRACT

One of the most important goals of sugar beet cultivation is to achieve high seedling emergence and establishment under water deficit conditions. In this experiment irrigation of the seed bearing plants was cutoff in four periods of 750, 1000, 1250 and 1500 GDD after schtechlings were planted and the seeds were formed and grown under water stress. The experiment was carried out as a randomized complete block design with four replications. All treatments were harvested simultaneously and seed yield and seed physical quality characters were determined. In the second step, germination qualities of treatments were determined under 0 and -12 bar osmotic potential in four replicates. Results showed that seed yield of the last irrigation cutoff (1500 GDD) was 7 times more than that yield of the first irrigation cutoff (750 GDD). Standard seed size and germination were increased by postponing irrigation cutoff. Root length was affected more than stem length by the irrigation cutoff time so that, postponing irrigation cutoff up to days before harvest root and stem length 13 and 50% , respectively.

Keywords: Drought, Germination, Moisture deficit conditions, Root length, Seed vigor, Stem length, Sugar beet seeds

INTRODUCTION

Water is regarded as the most crucial input in agriculture. Most farmers avoid irrigating sugar beet farms at germination and plant establishment stages due to sugar beet high water requirement at these stages on the one hand and the water limitation on the other hand, and devote the irrigation water to other crops hoping the sugar beet seeds to germinate under the rainfall. Although relatively acceptable results are realized in some rainy seasons, particularly when precipitation occurs at early-season, the sugar beet seeds germinate and the plants establish under moisture deficit conditions in most cases resulting in the severe loss of plant establishment. Therefore, the increase in germination potential and plant establishment of sugar beet under water deficit conditions is one of the most important techniques for increasing sugar yield per unit area. Though improving genetic capabilities is the best method for increasing drought tolerance, numerous parameters increase seed germination vigor and establishment under water deficit conditions of soil including seed formation conditions, parental plant nutrition, seed treatment with plant growth promoting rhizobacteria (PGPR) and seed washing and priming (Janda et al. 2007). Studies of Fenner (1991) and Benech-Arnold et al. (1991) showed that although drought stress during seed formation stage reduced seed yield in some crops, seed germination percentage was increased under moisture deficit conditions.

Owing to indeterminate growth of sugar beet, its flowering period usually lasts for 35-50 days. Podlaski and Chrobak (1980) indicated that 456-612 GDDs over the base temperature of 7.2°C was required after the flowering initiation until seed

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maturity. Also, Grimwade et al. (1987) reported that the fruits of sugar beet can germination about 20 days after pollination, but the highest germination belonged to fruits that received 500 GDDs after about 55 days after blossoming. On the other hand, fruit weight and seed protein and starch reserves of sugar beet are increased until about 55 days after blossoming and so, the seeds reach to their physiological maturity about 55 days after inoculation and/or after receiving 500 GDDs. Sroller (1984) reported that dry matter content was increased during seed maturity period and in total, seeds with 55-60% dry matter content had the highest germination percentage. In relation to the increasing trend of the number of viable seeds of sugar beet, Longden and Johnson (1977) stated that the number of viable seeds increased until about 14-42 days after flowering reaching to the peak 56-84 days after flowering.

Battle and Whittington (1969) reported that irrigation during fruit formation and growth retarded maturity, increased the wooden precursors and germination inhibitor in fruits and decelerated germination. Given the possibility of the occurrence of drought stress at different growth stages of seed-bearing sugar beet plants, it is very useful to know the quality of seed germination (viability and vigor). The present study was aimed at answering whether the formation and development of sugar beet seeds under drought stress conditions results in their higher vigor and germination than their formation and development under optimum moisture conditions, i.e. whether a sort of drought resistance is induced in the seeds that are formed and grown under drought stress conditions.

MATERIALS AND METHODS

The present study was conducted at Elite Sugar Beet Seed Research Station of Firouzkouh, Iran on the basis of a Randomized Complete Block Design with four treatments of irrigation withdrawal at late-growth season and four replications in 2008 and 2009. Irrigation withdrawal treatments included irrigation withdrawal of seed-bearing plants after receiving 750, 1000, 1250 and 1500 growth degree-days (GDDs) since the planting of the roots. Each plot was composed of four sowing rows 6 meters long with the spacing of 65 cm among which the middle four rows were devoted to male sterile (maternal) and two marginal rows were devoted to male fertile (paternal) parents. The plants were spaced 45 cm apart. Two unsown rows were left between the two border plots, and the blocks were spaced 2 meters apart.

The number of days from sowing to irrigation withdrawal was 81, 102, 119 and 131 days after sowing of roots for these treatments, respectively; and the number of days from irrigation withdrawal to seeds harvest was 57, 36, 19 and 7 days for these treatments, respectively. It should be noted that the seeds had been formed on maternal plant before the first treatment of irrigation withdrawal.

Until the imposition of the treatments of irrigation withdrawal, all treatments were irrigated after the depletion of 50% of soil available moisture.

At harvest time (138 days after sowing of roots), all treatments were simultaneously harvested and spread on a tent. After drying, they were pounded and the seeds were polished (their wastes were removed). The soil, leaf residues, stalks, branches, florets, and tiny seeds were removed by separator and weighed. At the next step, the seeds of the treatments were separated to 3.5, 4.5 and 5.5-mm sizes by a sieve and their weight percentage was measured. Then, the seeds with the size of 3.5-4.5 mm were classified into 2.25 and 3.25-mm sizes by a sieve. The seeds with the size of 2.25-3.25 mm were regarded as standard seeds. The seeds with the size of <3.5 mm and >5.5 mm were regarded as undersized and oversized seeds, respectively, and they were weighed and their percentages were determined. The seeds were then graded with a full-automatic machine.

At the next step, the viability of >4.5-mm seeds was determined in accordance with ISTA guidelines. Final viability percentage was the accumulative sum of the number of germinated seeds (healthy roots with the length of 5 mm) in each counting (3, 7 and 14 days after sowing). Abnormal shoots were not included in viability count. At the final counting, the ungerminated seeds were counted and the number of empty and filled seeds was recorded. Mechanical viability was the sum of the number of normal germinated seeds, abnormal germinated seeds and ungerminated filled seeds.

Afterwards, germination (vigor) percentage and root and stalk length were determined under osmotic pressures of -2 and -12 bars. Osmotic pressure of the solutions was determined by Michel and Kaufmann (1973)'s equation where *Ψ* is the osmotic pressure of the solution (MPa), *C* is the concentration (g kg^{-1} H_2O) and *T* is the perimeter temperature (°C):

Fig. 1. Method of seed cultivation between two germination papers and putting them in culture tube

$$\Psi_s = -(1.18\times10^{-2})C - (1.8\times10^{-4})C^2 + (2.67\times10^{-4})CT + (8.39\times10^{-7})C^2T$$

Vigor was determined by sowing the seeds between two germination papers and putting it in tube. In this method, 25 seeds are disposed on a 50×15 cm^2 germination paper 2 cm away from the edge of the paper. Then, another filter paper is put on them and is sprayed with 30 ml solution (with osmotic pressure of -2 and/or -12 bars). Then, culture paper was rolled over and vertically put in a culture tube (with the height of 30 cm and the diameter of 5 cm) inside a culture container (Fig. 1). The culture container was filled with the intended solution up to the height of 3 cm. The container lid was completely closed to prevent moisture exchange. Then, the culture container was put in a dark germinator at 20°C and after 10 days, the samples were taken out and the number of germinated seeds, abnormal shoots, rootlet length and hypocotyl length were measured by a ruler. Seed vigor was measured as the product of the shoot length by vigor. All data were statistically analyzed by SAS and MS-Excel software.

RESULTS

Seed yield

Combined analysis of seed yield revealed that the main effect and the interaction of year and irrigation withdrawal treatment were significant at 1% probability level (Table 1). Although mean seed yield did not show statistically significant differences under different irrigation treatments in the two years, the pattern of the variations was uniform. Early withdrawal of irrigation at seed formation phase had an extremely adverse impact on seed yield, so that the seed yield was seven times higher in no irrigation withdrawal treatment than in early withdrawal. Seed yield was 391 and 2363 kg ha^{-1} in the treatment of irrigation with-

Table 1. Means squares of combined analysis of variance of some important traits in two years (2008 and 2009)

Sources of variation	df	Seed yield	>3.2 long sieve	Round sieve			Vigor	
				3.5-1.5	<3.5	>4.5	Stressed	Standard
Year	1	137812**	201**	1830**	319*	617**	512*	132*
Replication (year)	6	56018	14	225	299	28	243	111
Treatment	3	10229978**	39	1286	2858	334	6703**	2094*
Year × treatment	3	394568**	65**	269*	420*	244**	110**	205**
Error	18	110148	25	344	647	152	168	115
Total	31	10928526	343	3954	4543	1374	7736	2656

When the interaction between year and treatment was significant, the treatments were tested with this interaction.

Table 1. continued.

Sources of variation	df	Long sieve		Losses due to seed polishing	Mean length of			Vigor		
		<2.2	3.2-2.2		Stem	Root	Shoot	Stem	Root	Shoot
Year	1	6.8	971**	3469**	0.26	0.01	0.20	1817	4334	117055
Replication (year)	6	12.4	141	287	0.88	11.5	14.6	24074	247318	223125
Treatment	3	3.6	815**	2303**	3.26	65**	95**	496965**	1191194**	3643283**
Year × treatment	3	8.8	101	225	0.72	3.1	3.5	17733	121995	59904
Error	18	80	205	1112	9.12	27	46	58442	526250	300016
Interaction of year, treatment and error	21	89	308	1336	9.84	30.4	49.9	76175	648245	359920
Total	31	112	22333	7397	14.25	107	160	615388	2091090	4343383

When the interaction between year and treatment was significant, the treatments were tested with this interaction.
* and ** show significance at 5 and 1% probability level.

Table 2. Grouping of two-year means of some important traits in the two consecutive experiments in 2008 and 2009 (Duncan Multiple Range Test)

Irrigation withdrawal (GDD after root planting)	Seed yield (kg.ha^{-1})	Losses due to polish (%)	Seed percentage by round sieve Ø mm			Seed percentage by long sieve # mm		
			<3.5	3.5-4.5	>4.5	<2.2	2.2-3.2	>3.2
750	391 d	37 a	72.6 a	21.6 c	5.8 b	5.0 a	14.4 c	2.2 b
1000	730 c	34 a	62.5 b	29.8 b	7.8 b	4.8 a	21.1 b	3.9 a
1250	1343 b	22 b	53.3 c	34.0 ab	12.7 a	4.9 a	24.7 ab	4.4 a
1500	1862 a	17 c	47.7 c	38.9 a	13.4 a	5.6 a	28.0 a	5.2 a
Mean of the year								
2008	1016 b	17.1 b	5.5 b	38.6 a	55.8 b	6.5 a	27.6 a	4.6 a
2009	1387 a	37.9 a	14.3 a	23.5 b	62.2 a	1.4 b	16.5 b	5.5 a

Figures in each column with similar letter(s) did not show significant differences at 5% probability level.

Table 2. continued.

Irrigation withdrawal (GDD after root planting)	Seed viability		Length (cm) of			Vigor of		
	Standard	Moisture stress	Stem	Root	Shoot	Stem	Root	shooting
750	67 d	49 c	6.8 b	6.6 c	13.4 b	333 c	323 c	657 c
1000	77 c	65 b	7.5 b	8.4 b	15.9 a	488 b	546 b	1034 b
1250	85 b	82 a	7.5 b	10.2 a	17.7 a	615 a	836 a	1451 a
1500	88 a	85 a	7.7 a	9.9 a	17.5 a	655 a	842 a	1488 a
Mean of the year								
2008	77 b	66 b	7.5 a	8.8 a	16.2 a	495 b	581 a	1069 b
2009	81 a	74 a	7.3 a	8.8 a	16 a	540 a	651 a	1184 a

Figures in each column with similar letter(s) did not show significant differences at 5% probability level.

drawal after receiving 750 and 1500 GDDs (Table 2).

In sugar beet plants, flower buds are formed on auxiliary and main branches as the flowering stem elongates. Drought stress at flowering initiation phase – when flower buds are formed and need assimilates for the growth of embryo and the increase in seed perisperm – reduces total shoot weight, bud number per flower and finally, seed yield. Drought stress from flowering termination phase until seed harvest tends to halt bud-bearing and blossoming of the pre-formed buds. Also, drought stress at this phase increases the number of unfilled seeds and decreases seed viability. Furthermore, drought stress can reduce assimilate mobilization to the formed seeds and consequently, can decrease 1000-seed weight and yield by reducing life cycle and the activities of pollens, reducing seed formation duration and early maturity of the plants (Csapody 1980).

Seed shedding

Statistical analysis in 2009 showed that the effect of different times of irrigation withdrawal was significant on seed shedding at 1% probability level (Table 3), so that early withdrawal of irrigation resulted in higher seed shedding. Irrigation withdrawal after receiving 750 GDDs by plants after the planting of the roots resulted in 43% shedding of the produced seed weight. However, even in optimum irrigation treatment 7% of the produced seed was shed (Table 4). It suggests that the lack of irrigation of the seed-bearing plants results in early-maturity of seeds and that delayed harvest

Table 3. Means squares of analysis of variance of seed shedding percentage, viability of harvested and shed seeds of sugar beet in 2009

Sources of variations	df	Viability of shed seeds	Viability of harvested seeds	Seed shedding
Replication	3	276	238	982
Treatment	3	11274**	13187**	3055**
Error	9	1934	740	1210
Total	15	13485	14166	5248

** shows significance at 1% probability level.

Table 4. Grouping of means of the effect of irrigation withdrawal time on the shedding of sugar beet seeds (2009)

Irrigation withdrawal time (GDD)	Seed shedding (%)	Viability of harvested seeds	Viability of shed seeds (%)
750	43.7 a	25 b	31 b
1000	33.5 ab	31 b	33 b
1250	19.3 bc	81 a	84 a
1500	7.3 c	89 a	85 a

Figures with similar letter(s) in each column were not significantly different at 5% probability level.

of seeds results in the shedding of the seeds and decrease in seed yield. In addition, the viability of the shed and harvested seeds were the same on the maternal plants.

Losses caused by seed polishing

Combined analysis of the two experimental years for the losses caused by seed polishing revealed that only the main effects were significant at 1% probability level (Table 1). Therefore, the means of two years were compared (Table 4). Irrigation withdrawal after receiving 750 and 1500 GDDs resulted in the loss of 37 and 17% of seed weight due to polishing, respectively. The potential of the tolerance of polish is a good index of seed setting and maturity. In the treatment of early withdrawal of irrigation (irrigation withdrawal after receiving 750 GDDs), the husks were thin and the embryos were not fully grown. In addition, in this treatment seeds were harvested about 50 days after the final irrigation and thus, the seeds were dry and had lower moisture content. Therefore, these three factors resulted in 37% loss of seed weight in seed processing phase due to polishing (Table 2).

Weight percentage of different seed classes

According to combined analysis for weight percentage of under-sized, standard and over-sized seeds, only the main effects were significant at 1% probability level (Table 1). The weight percentage of under-sized seeds was 78 and 53% under the treatments of irrigation withdrawal after receiving 750 and 1500 GDDs, the weight percentage of standard seeds was 14 and 28% and the weight percentage of over-sized seeds was 8 and 19%, respectively (Table 2). It suggests that the effect of irrigation withdrawal on the weight percentage of the standard seeds decreases over time. Early withdrawal of irrigation at seed formation phase reduces seeds extraction coefficient by interrupting embryo-bearing.

Seed viability

Combined analysis for seed viability revealed that the main effects and the interaction between year and treatment were significant at 1% probability level (Table 1). Although mean viability of the treatments did not exhibit statistically significant differences between the two years, the pattern of its variations was uniform. It was found that early withdrawal of irrigation at seed formation phase significantly reduced seed viability and vigor. The viabilities of seeds harvested from the treatments of irrigation withdrawal after receiving 750 and 1500 GDDs were 67 and 88%, respectively with the difference being significant at 1% probability level. Mean viability of seeds harvested after receiving 1250 and 1500 GDDs was not statistically significant. In addition, this result suggests that retarding irrigation withdrawal was associated with lower seed viability and vigor, so that the difference in seed viability between standard conditions and moisture stress after receiving 750 and 1500 GDDs was 18 and 3%, respectively (Table 2). It shows that although seeds harvested after early irrigation withdrawal (after receiving 750 GDDs) can germinate under ideal conditions, their germination capacity sharply decreases under moisture stress conditions.

Physiological attributes of initial bud

Combined analysis for the length of stem and root revealed that only the main effects were significant at 1% probability level (Table 1). Retarding irrigation withdrawal was associated with longer buds which was mainly caused by the increase in root length so that the variations of stem length was very slight under four irrigation withdrawal treatments but root length was greatly varied. Therefore, drought stress at seed formation phase affects root length more than stem length. Additionally, it was found that the variations of bud length was not tangible after receiving 1250 GDDs (Table 2) and thus, it can be concluded that the seeds are physiologically fully matured after receiving 1250 GDDs.

Seed vigor

According to combined analysis of germination vigor, only the main effects were significant at 1% probability level (Table 1). Retarding irrigation withdrawal increased germination vigor significantly. However, the treatments of irrigation withdrawal after receiving 1250 and 1500 GDDs did not show any significant differences in terms of germination vigor. Also, it was found that retarding irrigation withdrawal increased the slope of the increase in root vigor more than the increase in stem vigor (Table 2). Hence, it can be inferred that when embryos are formed, the plants allocate more assimilates for increasing root volume than to the stem and this pattern of assimilate mobilization continues until receiving 1500 GDDs.

Discussion

One of the most important objectives of sugar beet cultivation is to produce high-quality seeds. High-quality seeds are the basis for modern cultivation, are regarded as a critical factor in higher number of plants per unit area and ensure the optimum sugar yield. It has been shown that mean daily water requirement of seed-bearing sugar beet plants is 1.76, 5.00, 7.82 and 4.99 mm during the initial phase (sowing to the initiation of stem-bearing, 20 days), plant development phase (the initiation of stem-bearing until the initiation of flowering, 30 days), intermediate phase (the initiation of flowering until the end of flowering, 35 days), and from the end of flowering to seed maturity (80-85 days after root planting) (Chegini 2006). Therefore, the highest water requirement of sugar beet is from 30 to 85 days after the planting of the roots. Obviously, drought stress at this stage can impose irreversible impacts on the yield and seed quality of sugar beets. Thus, it is recommended to regularly irrigate seed-bearing sugar beet fields from mid-June until two weeks before the harvest of the seeds. Chegini (2006) showed that severe depletion of soil moisture (90% of available water) at early flowering phase resulted in 40% loss of seed yield and severe depletion of soil moisture at late growing season resulted in the production of undersized and less vigorous seeds, but the depletion of 70% of soil moisture at early growing season had no effects on quantitative and qualitative traits of seeds. Sadeghzadeh Hemayati (2007) stated that the share of unfilled seeds in no-irrigation treatment was nearly twice as great as that in no water stress treatment. In addition, the share of >3.5-mm seeds in sugar beet seed yield was 49% in control which was decreased to 46, 50, 43 and 37% under the treatments of stress at the aforementioned phases. Csapody (1980) found that in regions with deficit precipitation, the irrigation of seed-bearing sugar beet plants before and during flowering prolonged growth period and retarded harvesting date by four days. In addition, the share of unfilled seeds in no-irrigation treatments was about twice as great as that in the irrigated plots. On the other hand, irrigation increased sugar beet seed germination rate from 40-60% to 67-70%. Gizbullin (1984) stated that drought stress shortened vegetative growth period, but the germination of the seeds of the plants treated with drought stress had no difference with the germination of the seeds produced under no-stress conditions. It seems that he has only focused on germination rate of standard seeds and has ignored the germination of whole seeds. Early withdrawal of irrigation resulted in greater seed shedding, lower seed yield, lower percentage of standard seeds, and higher percentage of under-standard seeds. On the other hand, drought stress at seed formation phase reduced germination rate of sugar beet seeds both under optimum conditions and under water deficit conditions. Nonetheless, the reduction was greater under water deficit conditions. Thus, the irrigation of seed-bearing sugar beet fields is recommended to continue until 10 days before harvest time.

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