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Research Article

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# SEED QUALITY OF SOYBEAN CULTIVARS AFFECTED BY POD POSITION AND WATER STRESS AT REPRODUCTIVE STAGES

Kazem Ghassemi-Golezani\*, Ramin Lotfi and Majid Norouzi

University of Tabriz, Faculty of Agriculture, Department of Plant Eco-physiology, Tabriz, *Iran* \*Corresponding author, e-mail: *golezani@gmail.com* 

**ABSTRACT**: A field experiment was carried out to investigate the effects of different irrigation treatments (I<sub>1</sub>, I<sub>2</sub>, I<sub>3</sub> and I<sub>4</sub>: well-watering on the bases of 70 mm evaporation from class A pan and irrigation disruptions at flowering, seed filling and during flowering and seed filling stages, respectively) and pod positions (upper, lower and middle parts of the canopy) on seed quality of three soybean cultivars (Clark, Williams and L<sub>17</sub>). Seed quality as determined by seed weight, electrical conductivity of seed leachates, viability and germination percentages, germination rate and seedling dry weight significantly reduced for seeds produced under I<sub>3</sub> and I<sub>4</sub>. Significant differences in mean seed weight, viability, electrical conductivity and germination rate among soybean cultivars under different irrigation treatments were attributed to variation in genetic constitution. Seeds of upper parts of the canopies were larger and exhibited higher quality, compared with those of middle and lower parts. The advantage of the upper seeds was more evident under  $I_3$  and  $I_4$  treatments. Seed weight had the highest positive correlation with seedling dry weight. Thus, sufficient water supply and production of large and uniform seeds could be a practical way to improving seed lot quality in soybean cultivars.

**Keywords**: Pod position, reproductive stages, seed quality, soybean, water stress

## INTRODUCTION

Soybean seed is a major source of high-quality protein and oil for human consumption [1]. Growth, development and yield of soybean are the result of genetic potential interacting with environment. In arid and semi-arid environments, successful crop establishment depends not only on the rapid and uniform germination of the seed, but also on the ability of the seed to germinate under low water availability [2].

Rapid and synchronized seed germination and seedling emergence are crucial for achieving an optimal crop stand and high productivity. By a good seedling establishment, plant life cycle would be guaranteed a strong plant, with a good plant density and uniformity [3, 4, 5, 6]. Low quality of seeds can potentially decrease the rate and percentage of seedling emergence, leading to poor stand establishment in the field and consequently yield loss in many crops such as maize [7, 8, 9] wheat [10, 3], cotton [11], barley [12, 13] and oilseed rape [4, 5].

Factors other than environment can also alter seed size and quality. Seed position on the plant is one of the components of within-plant variation that may account for part of the variation in physical (such as weight, shape) or physiological (such as germination and vigor) seed attributes [14]. In chickpea plants, seeds located within the lower canopy had a higher germination rate and percentage and a greater seedling growth rate than seeds from the middle and upper canopy under well and limited irrigation conditions [15]. Thus, seed position on mother plant may be an important factor influencing seed physiological quality. This research was undertaken to examine the effects of water stress at reproductive stages and pod position on soybean seed germination and vigor.

## **MATERIALS AND METHODS**

A split-split plot experiment (using RCB design) with four replications was conducted in 2011 at the Research Farm of the Faculty of Agriculture, University of Tabriz, Iran (latitude 38.05°N, longitude 46.17°E, altitude 1360 m sea level), in order to determine the effects of water stress and pod position on seed quality of soybean cultivars. The climate is characterized by mean annual precipitation of 245.75 mm per year, mean annual temperature of 10°C.

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Irrigation treatments ( $I_1$ ,  $I_2$ ,  $I_3$  and  $I_4$ : well-watering on the bases of 70 mm evaporation from class A pan and irrigation disruptions at flowering stage, seed filling stage and during flowering and seed filling stages, respectively) were located in main plots and cultivars ('Clark', 'Williams' and ' $L_{17}$ ') were allocated to sub plots. Pod positions were considered as sub-sub plots.

Seeds of soybean cultivars were treated with 2 g kg<sup>-1</sup> Benomyl and then were sown by hand on 11 May 2011 in 5 cm depth of a sandy loam soil. Seeding density was 60 seeds m<sup>-2</sup>. Each plot consisted of 6 rows of 5 m length, spaced 25 cm apart. All plots were irrigated immediately after sowing and after seedling establishment, plants were thinned to 45 plants m<sup>-2</sup>. Subsequent irrigations were carried out on the bases of 70 mm evaporation from class A pan up to flowering. Thereafter, irrigation disruptions were applied according to the treatments. Hand weeding of the experimental area was performed as required.

At the maturity, when seed moisture content was 18-20%, seeds of the plants at 1 m² of each plot were separately detached from the upper, middle and lower parts of the canopies. Seed moisture content was determined in accordance with ISTA rules [16]. Subsequently, seeds were air dried at 20-25°C and moisture content declined to 14-15°C. Hundred seed weight of each sample was determined in four replicates. Seed samples within separate sealed bags were then placed in a refrigerator at 3-5°C.

Four replicates of 25 seeds from each sample were treated with 2 g kg<sup>-1</sup> Benomyl, before testing. Seeds of each replicate were placed between two 30 × 30 cm wetted and rolled filter papers, which were then placed in plastic bags to prevent water loss. These bags were incubated at 20±1°C and germinated seeds (protrusion of radicle by 2 mm) were counted every day up to 8 days. Rate of seed germination was calculated according to Ellis and Roberts [17]. At the end of each test, normal and abnormal seedlings were counted [16] and germination percentage was calculated. Normal seedlings were then dried in an oven at 80°C for 24 hours [18] and mean seedling dry weight for each treatment at each replicate was determined.

For each seed sample, two containers with 250 ml of distilled water were kept in an incubator at  $20 \pm 1$  °C for 24 hours. Thereafter, two replicates of 50 seeds from each sample were weighed (SW<sub>1</sub> and SW<sub>2</sub>) and then separately immersed in distilled water of the containers for another 24 hours at the same temperature. The seed-steep water was then gently decanted and electrical conductivity (EC) was measured, using an EC meter (EC1 and EC2). The following equation was applied to calculate conductivity per gram of seed weight for each sample [19]:

 $EC (\mu S/cm/g) = [(EC1/SW1) + (EC2/SW2)]/2$ 

All the data were analyzed on the basis of experimental design, using MSTATC and SPSS software. The means of each trait were compared according to Duncan multiple range test at  $P \le 0.05$ . Excel software was used to draw figures.

#### RESULTS

Analysis of variance of the data showed significant effects of irrigation on 100 seed weight, viability, electrical conductivity (EC), germination percentage, germination rate and seedling dry weight. Irrigation  $\times$  cultivar interactions were significant for electrical conductivity of seed leachates, germination percentage and germination rate. Interaction of irrigation  $\times$  pod position was only significant for germination percentage (Table 1).

Mean 100 seed weight, viability, germination percentage, germination rate and seedling dry weight were decreased for seeds produced under  $I_3$  and  $I_4$  (irrigation disruption at seed filling and during flowering and seed filling stages, respectively), but the highest reduction of these traits was observed for seeds obtained from plants under  $I_3$ . The highest electrical conductivity of seed leachates was also recorded for seeds produced under  $I_3$ . Mean seed weight, viability and germination rate of Williams were greater than the other cultivars. However, the lowest electrical conductivity was observed in Clark (Table 2).

Mean electrical conductivity of seed leachates for Williams and  $L_{17}$  under  $I_1$ ,  $I_2$  and  $I_3$  was statistically similar, but EC of  $L_{17}$  under  $I_4$  was significantly higher than that of Williams. Clark had the lowest electrical conductivity under  $I_1$ ,  $I_2$ , and  $I_3$ , but under  $I_4$  it was statistically similar with  $L_{17}$ .

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Mean germination percentage of seeds produced under I<sub>1</sub>, I<sub>2</sub> and I<sub>3</sub> and Mean germination rate of those produced under I1 and I2 were statistically similar for all cultivars. The highest germination percentage of I4 seeds was recorded for L<sub>17</sub>, which was not significantly different from Clark. Mean germination rate of Williams under I<sub>3</sub> and  $L_{17}$  under  $I_4$  was greater than that of other cultivars (Table 3).

> Table 1. Analysis of variance of the effects of water disruption at reproductive stages and pod position on seed quality parameters of sovbean cultivars

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Source	df	100 seed	Electrical	viability	Germination	Germination	Seedling dry
		weight	conductivity		percentage	rate	weight
Replication	3	2.139 ns	53.359 ns	18.593 ns	148.222 ns	0.001 ns	0.006 ns
Irrigation (I)	3	83.611**	2328.344**	380.074*	843.111*	0.087**	1.268**
Error	9	1.837	115.131	54.519	184.272	0.002	0.03
Cultivar (C)	2	5.028*	585.069**	145.528**	126.028 ns	0.007*	0.025 ns
I×C	6	1.441 ns	368.715**	46.046 ns	191.472*	0.004*	0.02 ns
Error	24	1.027	38.102	25.065	71.148	0.001	0.013
Position (p)	2	13.795**	20.975 ns	11.861 ns	710.861**	0.001 ns	0.157**
I×P	6	0.265 ns	19.62 ns	13.491 ns	49.306*	0 ns	0.004 ns
C×P	4	0.214 ns	8.963 ns	1.444 ns	6.444 ns	0.002 ns	0.003 ns
$I \times C \times P$	12	0.092 ns	26.638 ns	16.63 ns	8.667 ns	0.001ns	0.004 ns
CV%		4.16	14.04	3.81	5.01	7.020	8.11
Error	72	0.217	27.832	13.361	20.824	0.001	0.003

<sup>\*,\*\*</sup> Significant at p≤0.05 and p≤0.01, respectively

Table 2. Means of the seed quality parameters of soybean affected by water disruption at reproductive stages

Treatment	100 seed	Electrical	Viability	Germination	Germination	Seedling
	weight	conductivity	(%)	(%)	rate	dry weight
	(g)	(µS/cm/g)			(per day)	(g)
Irrigation						
$I_1$	10.84 a	35.12 b	98.00 ab	93.11 a	0.4934 a	0.8008 a
$I_2$	11.52 a	29.01 с	99.39 a	95.78 a	0.4879 a	0.8084 a
$I_3$	8.090 c	48.35 a	92.22 c	84.44 b	0.3894 с	0.4568 b
$I_4$	9.453 b	37.78 b	94.61 bc	90.89 ab	0.4296 b	0.5061 b
Cultivars						
Clark	9.996 ab	33.67 b	94.21 b	89.58 a	0.4361 b	0.625 a
Williams	10.290 a	38.72 a	97.67 a	92.79 a	0.4563 a	0.669 a
$L_{17}$	9.644 b	40.37 a	96.29 ab	90.79 a	0.4578 a	0.634 a
Pod position						
Up	10.38 a	37.71 a	95.5 a	95.33 a	0.451 a	0.7027 a
Middle	10.19 a	38.176 a	96.208 a	89.96 b	0.444 a	0.6379 b
Low	9.367 b	36.872 a	96.458 a	87.88 c	0.454 a	0.5885 c

Different letter in each column indicate significant difference at p≤0.05

The highest 100 seed weight, germination percentage and seedling dry weight were obtain from seeds of upper position, followed by seeds of middle and lower positions, respectively. However, the difference in 100 seed weight of upper and middle parts of the canopy was not statistically significant (Table 2). In all irrigation treatments the seeds produced from upper position had comparatively higher mean germination percentage. Germination of middle seeds produced under I<sub>1</sub> and I<sub>2</sub> was higher than that of lower seeds, but under I<sub>3</sub> and I<sub>4</sub> it was statistically similar for middle and lower seeds (Figure 1).

Table 3. Mean electrical conductivity, germination percentage and germination rate
Of sovbean cultivars under different irrigation treatments

	<u> </u>				
Irrigation	Cultivar	Electrical	Germination	Germination	
		conductivity	percentage	rate	
	'Clark'	31.85 cde	93.17 ab	0.4773 ab	
$I_1$	'Williams'	35.61 bc	94.00 ab	0.5011 a	
	'L <sub>17</sub> '	37.89 b	92.17 abc	0.5017 a	
	'Clark'	28.06 e	94.00 ab	0.4882 ab	
$I_2$	'Williams'	29.96 cde	98.17 a	0.4855 ab	
	'L <sub>17</sub> '	29.28 de	95.17 ab	0.4849 ab	
	'Clark'	35.47 bc	87.67 bcd	0.3825 f	
$I_3$	'Williams'	54.39 a	84.83 cd	0.4112 de	
	'L <sub>17</sub> '	55.19 a	80.83 d	0.3745 f	
	'Clark'	39.29 b	83.50 d	0.3963 ef	
$I_4$	'Williams'	34.91 bcd	94.17 ab	0.4333 cd	
	'L <sub>17</sub> '	39.13 b	95.00 ab	0.4590 bc	

Different letter in each column indicate significant difference at  $p \le 0.05$   $I_1$ ,  $I_2$ ,  $I_3$ ,  $I_4$ : well-watering and irrigation disruption at flowering, grain filling and during flowering and grain filling, respectively.

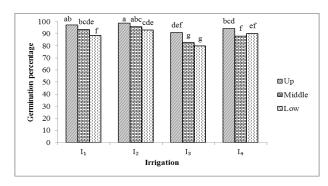


Figure 1. Mean germination percentage of soybean cultivars affected by pod position and irrigation treatment

I<sub>1</sub>, I<sub>2</sub>, I<sub>3</sub>, I<sub>4</sub>: well-watering and irrigation disruption at flowering, grain filling and during flowering and grain filling, respectively.

Mean seeds weight was positively and significantly ( $p \le 0.01$ ) correlated with viability, germination rate, germination percentage and seedling dry weight. However, electrical conductivity of seed leachates negatively and significantly correlated with other parameters. Viability, germination rate, germination percentage and seedling dry weight had also significant and positive correlations with each other (Table 4).

Table 4. Correlation coefficients of various seed quality parameter in soybean

Traits	100 seed	Electrical	Viability	Germination	Germination	Seedling
	weight	conductivit		percentage	rate	dry weight
		y				
'100 seed weight'	1					
'Electrical	-0.598**	1				
conductivity'						
'Viability'	0.515**	-0.373**	1			
'Germination rate'	0.630**	-0.419**	0.592**	1		
'Germination	0.515**	-0.466**	0.741**	0.552**	1	
percentage'						
'Seedling dry weight'	0.819**	-0.572**	0.653**	0.682**	0.740**	1

\*\* significant at p≤0.01

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## **DISCUSSION**

Coinciding water stress with seed filling stage may stimulate seed maturity, leading to decrease seed filling duration and mean seed weight (Table 2), as reported in lentil [20], chickpea [21], wheat [22], and common bean [23].

Reports on maize and sorghum [24], common bean [23] and faba bean [25] indicated that water deficit had no significant effects on seed quality. However, in this research water stress during reproductive stages, particularly during grain filling, significantly reduced seed quality soybean cultivars (Table 2). Water stress during growth and development had similar effects on seed quality of dill [26] and lentil [27].

Significant differences in 100 seed weight, viability, electrical conductivity and germination rate among soybean cultivars under different irrigation treatments (Table 3) can be attributed to variation in genetic constitution, which may strongly influence seed physiological quality [5; 28, 29].

Seeds of upper position of the canopies exhibited higher quality, compared with those of middle and lower plant positions. This could be related to early pod formation and longer duration of pod filling at upper parts of the soybean plants which resulted in the production of larger seeds. Other reports on soybean [30, 31, 32, 33] also suggest that the seeds of the upper half of the main stem are large and vigorous. In contrast, Ghassemi-Golezani *et al.*, [15] reported that large and vigorous seeds of chickpea were obtained from the lower part of the canopy. The advantage of the upper seeds was more evident under  $I_3$  and  $I_4$  treatments (Figure 1).

A significant positive correlation of 100 seed weight with seedling dry weight suggested that the heavier seeds may produce larger seedlings. The advantage of large seeds in enhancing the seedling size lies in their higher reserve content and the ability to provide energy to the growing seedling at a faster rate [3]. A positive relation between seed size and quality was also reported for soybean [34] and chickpea [15]. Correlations of seed weight and germination rate with seedling dry weight was higher than the other seed quality parameters (Table 4). This result clearly showed that the seed weight and germination rate had the most effect on seedling size.

## CONCLUSIONS

Seed quality of soybean cultivars was significantly reduced by water stress during reproductive stages, particularly during seed filling phase. Seeds of upper position of the canopies were large and had high quality under all irrigation treatments. Variation in seed quality of soybean cultivars was attributed to differences in their genetic constitution. Seed weight and germination rate had the most effect on seedling size of soybean. Thus, sufficient water supply and production of large and uniform seeds could be a practical way to improving seed lot quality in soybean cultivars.

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