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Analysis of Seed Size Effect on Seedling Characteristics of Different Types of Wheat (*Triticum aestivum* L.) Cultivars

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Abstract: In order to study the effect of seed size on seed vigor index of wheat, a factorial experiment based on a completely randomized design was arranged with four replications at the laboratory. The experiment had two factors. The first factor consisted of 13 wheat genotypes and the second factor included seed size at four levels (1.5, 2, 2.5 and 3 mm in diameter). According to the results, germination percentage, germination rate, root and shoot length, shoot/root length ratio were affected by seed size and genotypes ($p \leq 0.01$). Also it revealed, that there were significant correlation ($R = 0.85$; $p \leq 0.01$) between seedling growth rates and seed size treatments. Stepwise regression method was indicated that, seed size was the first (x_1) and shoot length was the second (x_2) independent variable that could be used to describe the trend of variation in seedling growth rate as dependent variable (Y) ($R^2 = 0.85$). It was observed that by increasing seed size and shoot length, seedling growth rate was increased. As a result, it was concluded that seed size and shoot length are two factors that strongly affect seedling growth rate and could be used to evaluate and assess wheat genotypes and its improvement programs.

Key words: Wheat genotypes, seed vigor, seedling growth rate, correlation, regression

INTRODUCTION

A successful crop production requires the use of high quality seeds to achieve better stand establishment and higher crop yield. This high level of seed quality is demanded by the end user to assure maximum seedling emergence and stand establishment in the field (Tekrony, 2006). A positive relationship between seed weight or size and germination percentage has been found within seed lots in a number of studies (Larsen and Andreasen, 2004; Turk and Tawaha, 2002; Kalakanavar and Shashidhara, 1989). Previous studies have been showed clearly success in seed emergence was obtained when seed could be prevailed the adverse condition and it has shown appropriate reaction (Guillen-Portal *et al.*, 2006; Waldron *et al.*, 2006). Certainly this reaction according to environment condition and genotype will be varied (Hakizimana *et al.*, 2000; Hall and Wiesner, 1990). Seed size could be affect by environment during the seed filling (Egil, 1998). Therefore, stand establishment and primary growth were considered as a positive factor on crop yield (Acevedo *et al.*, 1991; Ludlow and Muchow, 1990). Different factors could reduce stand population and in many environmental conditions, more attention has been paid into initial growth of the seedlings (Ludlow and Muchow, 1990). This preference particularly under

drought stress and low temperature is more effective than other conditions (Willenborg *et al.*, 2005; Cisse and Ejeta, 2003; Turk and Tawaha, 2002). And also some of studies have been shown that seedlings from large seeded species have higher rates of survival than seedlings from small-seeded species under natural conditions (Moles and Westoby, 2004; Petron *et al.*, 1989).

Aprico *et al.* (2002) indicated that there were significant correlation between seed size of wheat and pre-ripening growth and development. According to this result, growth rate and the first leaf area could be affected by seed size. Other studies about the role of seedlings vigor on other plant's characteristic showed that seed size was effected the stand establishment and total yield of wheat (Guillen-Portal *et al.*, 2006; Stougaard and Xue, 2004; Lafond and Baker, 1986). In some studies, medium seeds size had higher germination percentage than both smaller and larger seeds (Mian and Nafziger, 1994; McKersie *et al.*, 1981). In contrast, Willenborg *et al.* (2005) indicated that planting large oat seed improved oat germination characteristics, particularly where low spring soil moisture limited and Turk and Tawaha (2002) also observed increased germination percentage as well as greater speed for two barley cultivars under moisture stress. Moles and Westoby (2004) reported that there was a strong positive relationship between seed mass and the

percentage of seedlings surviving through the first week of establishment across 113 species from around the world. Thus, seedlings from large-seeded species had higher survival rates than seedlings from small-seeded species (Moles and Westoby, 2006).

However, studies of the relationship between seed weight or size and germination speed have also provided various conclusions but, the trend of different studies have revealed that there was a positive effects between large seed size, germination and emergence (Moles and Westoby, 2004; Singh and Singh, 2003; Aprico *et al.*, 2002; Fenner and Kitajima, 2000).

Therefore, it was discussed that some of the seedling characteristics such as percentage of seedling emergence, emergence rate index, fresh weight and dry weight of root and shoot and their lengths were heritable (Khan *et al.*, 2002).

The objective of this research was to determine the relationship between seed size and seed vigor index in various type of wheat cultivars. Furthermore, response surface analysis and linear regressions have been undertaken to analysis the data.

MATERIALS AND METHODS

Experimental design: This experiment was conducted to evaluate the effects of seed size of wheat cultivars on seed vigor indicators. It was carried out at the Khorasan laboratories of Agriculture and Natural Resources Research Center of Iran in 2005. The experimental design was completely randomized factorial with four replications. The first factor was 13 wheat cultivars in three types including winter, facultative and spring (Table 1) and the second factor was seed dimensions including 1.5, 2, 2.5 and 3 mm in diameter.

Seed material and sampling: One kilogram of primary seed sample had been taken randomize from each container and then 250 g of seeds as a second sample was

separated from initial sample by electrical divider equipment. Samples of each cultivar had been classified by a digital screening system (OCTAGON model). Finally, different seed size for each cultivar was obtained.

Determination of vigor index: Seed vigor indicators such as percentage of germination and germination rate, maximum root and shoot length, shoot root ratio and seedling growth rate were measured. Germination test was conducted on the basis of the international protocol for seed testing (ISTA, 1995). Ellis and Roberts (1980) equation was used to measure of germination rate. Normal seedlings were used to measure the maximum root and shoot length. After the final counting in the standard germination test, normal seedlings were placed in convection oven then seedling dry weights were measured. The Eq. 1 was used for calculating the seedling growth rates.

$$\text{Seeding growth rate} = \frac{Y}{X - (a + b)} \text{ (mg dry weight)/germinal seedlings} \quad (1)$$

X = No. of seed germination

a = No. of abnormal seedling

b = No. of dead seed

Y = Total dry weight of normal seedling

Statistical analysis: In agronomy appropriate statistical analysis procedures is used to compare the means (Peterson, 1977; Evans *et al.*, 1982) but when quantitative experiment was divided to equal levels, regression techniques and response surface are better than Duncan multiple range test (Peterson, 1977). Therefore, response surface analysis with multiple regression techniques were used to identify independent variables that could describe most of the variations in dependent variable. And also simple correlation coefficient was used to describe the relationships between different characters. Linear multiple-regression was used to select the best surface

Table 1: Descriptions and pedigree of wheat cultivars

Name	Abbreviation	Pedigree/Cross	Growth type cultivar	Year of released
Siossons	Syson	Soissons	Winter	1996
Sabalan	Sabal	(908*FnA12)*1-32-4382	Winter-rain fed	1971
Azar 2	Azar	Kvz/Ym71/3/Maya"s//Bb/Inia/4/Sefid	Winter-rain fed	2000
Sardari	Sarda	Kurdistan landrace	Winter-rain fed	1965
Backcross Roshan	Backc	Rsh2-10120	Facultative	1995
Roshan	Rosha	Esfahan landrace	Facultative	1958
Mahdavi	Mahda	Ti/pch/5/Mt48/3/ Wt*// Nar59/Tota63/4/Mus	Facultative	1995
Toos	Toos	"Spn/Mcd/cama/3/Nzt"	Facultative	2002
Alvand	Alvan	1-27-6275-/CF1770	Facultative	1995
Ghods	Ghods	Rsh/5/Wt/4/Nor10/K54*2//Fn/3/Ptr/6/Omid//Kal/B6	Facultative	1989
Chamran	Chamr	Attila,(CM85836-50Y-OM-OY-3M-OY)	Spring	1997
Pishtaz	Pisht	Alvand//Aldan/Ias58	Spring	2002
Marvdasht	Marvd	HD2172/Bloudan//Azadi	Spring	1999

equation. The final equation to predict the dependent variable is given by Eq. 2:

$$Y = a_0 + (a_1 x_1) + (a_2 x_2) + (a_n x_n) \quad (2)$$

Y = Dependent variable
 a₀ = Intercept and a₁ through a_n partial regression coefficients
 x₁ through x_n = Independent variable

Cluster analysis was carried out using the minimum variance method based on Euclidean distance group (Ward, 1963) and SAS software was used to analysis the data.

RESULTS AND DISCUSSION

Effect of genotypes and seed size was significant (p<0.01) for all measured traits, but the interaction of cultivar and seed size for germination percentage and germination rate was not significant (Table 2).

The effect of Seed size of 1.5 mm was significant for germination percentage, germination rate, root length,

shoot root length ratio and seedling growth rate, but these traits were not significant for the other seed sizes (Table 3).

By increasing seed size, germination rate index increased and the highest rates of germination was achieved in seed size of 3 mm (Table 4).

Effect of seed size on seedling growth rate was significant and by increasing seed size, seedling growth rate was increased (Table 5).

These results revealed that germination percentage and germination rate were significant for cultivars of facultative types. In contrast these two traits were not significant in other genotypes (Table 6).

Correlation of seed size and seedling growth rate was positive and significant (Table 7) and the maximum coefficient of correlation for seedling growth rate calculated for seed size (R = 0.85, p<0.01). These results agree with formers studies (Larsen and Andearsen, 2004; Moles and Westoby, 2004; Turk and Tawaha, 2002).

To determine the best equation of surface analysis, stepwise regression method was used. As the basis of results, seed size was the first independent variable (x₁) and shoot length was the second (x₂). These two traits

Table 2: F value from analysis of variance for measured characteristics

Source of variation	df	Germination	Germination rate	Root length	Shoot length	Shoot/root length	Seedling growth rate
Genotype (G)	12	3.25**	4.37**	71.02**	129.45**	230.12**	25.84**
Seed size (SS)	3	5.70**	5.65**	20.78**	4.94**	17.11**	597.22**
SS×G	36	1.20ns	1.39 ^{ns}	4.68**	2.84**	7.30**	7.10**
Error	156						

**Significant at p<0.01 level, ^{ns}: Not significant

Table 3: Mean of measured characteristics for seed size effect

Seed size (mm)	Germination (%)	Germination rate	Root length (cm)	Shoot length (cm)	Shoot/root length ratio	Seedling growth rate (mg dry weight)/germinal seedling
3	95.23a [†]	7.84a	11.86b	7.57b	0.64b	15.85a
2.5	95.69a	7.9a	12.22a	7.65ab	0.63c	14.48b
2	94.76a	7.83a	12.09ab	7.82a	0.65b	11.50c
1.5	92.34b	7.63b	11.09c	7.44b	0.68a	8.23d

[†]: Means with the same letter(s) within each column are not significantly different (p<0.05)

Table 4: Interaction of seed size and genotype on germination, germination rate and root length

Genotypes	Germination (%)				Germination rate				Root length (cm)			
	1.5	2	2.5	3	1.5	2	2.5	3	1.5	2	2.5	3
Ghods	3.82±90	1.91±97	1.15±98	1.51±98.0 [†]	0.29±7.65	0.14±8.07	0.08±8.15	0.05±8.10	0.47±13.62	0.27±14.25	0.98±15.02	0.17±13.80
Roshan	2.82±92	2.82±96	1.91±97	1.63±96.0	0.24±7.62	0.19±7.87	0.18±8.02	0.08±7.70	0.24±8.90	0.25±10.20	0.42±10.30	0.43±9.85
Toos	1.63±96	1.00±93	2.30±96	1.63±96.0	0.10±7.87	0.13±7.65	0.17±8.00	0.19±7.77	0.45±12.35	0.10±12.02	0.05±12.15	0.18±12.25
Backros	2.00±90	0.00±88	1.15±94	1.91±93.0	0.17±7.40	0.00±7.30	0.11±7.70	0.16±7.67	0.67±10.97	0.41±11.12	0.47±12.95	0.40±11.80
Mahdavi	1.91±91	1.15±98	1.91±95	1.15±98.0	0.16±7.55	0.10±8.12	7.72±0.20	0.06±8.05	0.60±8.22	0.35±9.35	0.61±9.72	0.39±9.72
Chamran	0.00±96	1.15±98	1.91±97	2.51±95.0	0.00±8.00	0.11±8.07	0.20±7.90	0.20±7.87	0.22±8.72	0.21±11.05	0.14±13.57	0.23±13.57
Sysons	3.86±92	1.15±98	0.00±100	1.91±95.0	0.28±7.65	0.04±8.07	0.05±8.25	0.17±7.87	0.15±10.40	0.43±11.27	0.40±10.20	0.09±9.97
Marvdash	1.15±88	1.63±96	1.91±97	1.91±95.0	0.10±8.07	0.12±7.97	0.13±8.05	0.12±7.80	0.35±11.82	0.23±14.10	0.14±14.22	0.39±14.17
Alvand	4.43±91	3.26±88	2.00±94	1.91±95.0	0.35±7.57	0.24±7.27	7.80±0.17	0.21±7.82	0.22±12.77	0.39±13.75	0.25±13.37	0.32±12.95
Peshtaz	1.91±97	1.15±98	3.82±94	1.00±93.0	0.20±8.00	0.08±8.15	0.30±7.82	0.07±7.77	0.39±10.30	0.42±11.22	0.78±9.27	0.31±8.02
Sardari	1.00±91	2.51±91	3.41±89	2.00±90.0	0.04±7.40	0.16±7.45	0.29±7.37	0.15±7.45	0.42±12.80	0.17±14.37	0.14±13.62	0.33±13.62
Sabalan	1.91±87	2.30±96	1.91±97	1.15±98.0	0.13±7.17	0.16±7.97	0.13±8.02	0.09±8.10	0.37±9.87	0.63±11.00	0.58±11.12	0.23±11.50
Azar±2	1.00±89	3.00±95	2.82±96	2.30±96.0	0.13±7.32	7.90±0.24	0.25±7.92	0.20±7.95	0.29±13.45	0.12±13.52	0.50±13.32	0.49±13.05

[†]Standard Deviation = SD

Table 5: Interaction of seed size and genotype on shoot length, shoot root length ratio and seedling growth rate

Genotypes	Shoot length (cm)				Shoot/root length ratio				Seedling growth rate (mg dry weight)/germinal seeding			
	1.5	2	2.5	3	1.5	2	2.5	3	1.5	2	2.5	3
Ghods	0.26±7.020	0.09±7.30 ¹	0.23±7.25	0.16±7.55	0.02±0.49	0.01±0.51	0.02±0.48	0.01±0.54	0.17±7.85	0.52±11.50	1.20±14.60	0.49±16.47
Roshan	0.46±10.27	0.12±10.27	0.40±9.90	0.35±9.65	0.02±1.15	0.01±1.00	0.01±0.95	0.02±0.97	0.27±8.19	0.36±12.04	0.66±16.58	0.85±18.43
Toos	0.27±9.400	0.19±8.75	0.10±8.82	0.32±7.95	0.01±0.76	0.02±0.73	0.01±0.72	0.02±0.64	0.23±9.05	0.25±12.26	0.33±15.53	0.31±15.52
Backcross	0.37±9.850	0.10±9.52	0.19±10.12	0.32±9.40	0.02±0.90	0.03±0.85	0.04±0.78	0.01±0.79	0.21±8.33	0.29±12.78	0.26±17.26	0.37±19.26
Mahdavi	0.21±6.450	0.10±6.72	0.50±5.47	0.13±6.17	0.04±0.79	0.02±0.72	0.01±0.55	0.01±0.63	0.19±6.79	0.12±10.63	0.44±11.86	0.33±13.83
Chamran	0.17±6.470	0.16±6.77	0.13±7.97	0.31±7.52	0.01±0.74	0.01±0.61	0.01±0.58	0.02±0.55	0.16±6.99	0.25±10.30	0.24±17.35	0.54±19.15
Sysons	0.24±5.320	0.16±6.35	0.10±5.65	0.11±5.92	0.02±0.51	0.01±0.56	0.01±0.55	0.01±0.59	0.08±7.05	0.45±9.59	0.36±12.35	0.29±13.36
Marvdash	0.20±5.720	0.13±6.87	0.10±6.40	0.10±6.42	0.01±0.48	0.01±0.48	0.01±0.45	0.01±0.45	0.18±7.60	0.15±12.05	0.25±13.90	0.31±16.02
Alvand	0.10±7.870	0.37±8.12	0.13±8.12	0.17±8.12	0.01±0.61	0.01±0.58	0.01±0.60	0.01±0.62	0.01±9.52	0.49±12.57	0.18±15.28	0.26±16.49
Peshitaz	0.19±5.570	0.23±6.92	0.50±5.90	0.11±5.52	0.01±0.54	0.02±0.61	0.02±0.63	0.01±0.69	1.35±10.26	0.29±10.64	1.00±11.60	0.40±11.48
Sardari	0.17±8.570	0.17±8.92	0.29±9.27	0.08±9.25	0.01±0.67	0.01±0.62	0.02±0.67	0.01±0.68	0.11±9.37	0.08±11.56	0.20±14.90	0.44±16.64
Sabalan	0.20±5.650	0.29±6.54	0.63±6.25	0.22±6.77	0.02±0.57	0.01±0.58	0.03±0.55	0.01±0.58	0.48±7.02	0.43±10.58	0.68±12.77	0.32±13.97
Azar±2	0.22±8.550	0.07±8.70	0.23±8.30	0.33±8.17	0.01±0.63	0.01±0.64	0.01±0.62	0.01±0.62	0.10±9.04	0.26±13.07	1.05±13.95	1.16±15.39

¹Standard Deviation = SD

Table 6: F-value for non-orthogonal contrasts analysis

Contrasts [†]	df	Germination	Germination rate	Root length	Shoot length	Shoot/root length ratio	Seedling growth rate
W	1	1.45 ^{ns}	1.07 ^{ns}	54.02 ^{**}	0.17 ^{ns}	43.25 ^{**}	1.69 ^{ns}
F	1	5.72 [*]	11.60 ^{**}	46.10 ^{**}	124.10 ^{**}	2.95 ^{ns}	1.72 ^{ns}
S	1	1.35 ^{ns}	0.33 ^{ns}	30.68 ^{**}	70.00 ^{**}	262.20 ^{**}	0.03 ^{ns}
WR and W+S	1	0.51 ^{ns}	0.35 ^{ns}	21.09 ^{**}	99.37 ^{**}	196.50 ^{**}	0.39 ^{ns}
W and S	1	0.33 ^{ns}	0.20 ^{ns}	25.13 ^{**}	114.20 ^{**}	265.70 ^{**}	4.56 [*]
S and F	1	0.22 ^{ns}	0.34 ^{ns}	13.85 ^{**}	271.90 ^{**}	423.90 ^{**}	11.49 ^{**}
F and W	1	0.07 ^{ns}	0.22 ^{ns}	16.97 ^{**}	8.60 ^{**}	3.28 ^{ns}	1.99 ^{ns}

*Significant at p≤0.05 level, **Significant at p≤0.01 level, ns: Not significant, [†]: W: Winter type, F: Facultative type, S: Spring type, WR: Winter Rain fed type

Table 7: Simple correlation coefficient

Parameters	SS [†]	G	GR	RL	SL	SL/RL	SGR
Seed size	1	0.33 ^{**}	0.28 ^{**}	0.14	0.01	-0.10	0.85 ^{**}
Germination		1	0.95 ^{**}	-0.06	-0.37 [*]	-0.17	0.25 [*]
Germination rate			1	-0.05	-0.30 ^{**}	-0.23	0.17
Root length				1	0.22 [*]	-0.47 ^{**}	0.39 ^{**}
Shoot length					1	0.68 ^{**}	0.37 ^{**}
Shoot/root length ratio						1	0.04
Seedling growth rate							1

[†]SS: Seed Size, G: Germination, GR: Germination Rate, RL: Root Length, SL: Shoot Length, SL/RL: Shoot/Root Length Ratio, SGR: Seedling Growth Rate

were the best independent variables to describing the trend of variation in seedling growth rate as dependent variable (Y).

To determine the best equation for surface analysis, different linear multivariate regression models with two independent variables were calculated and the following model was the best.

$$Y = -5.000955 + (5.128078X_1) + (0.784943X_2)$$

$$R^2 = 0.85$$

Response surface of seedling growth rate vs. seed size (x₁) and shoot length (x₂) was showed in Fig. 1. Results indicated that by increasing seed size and shoot length, seedling growth rate was increased and the highest seedling growth rate was obtained in highest seed size and shoot length. As a result, it is suggested that

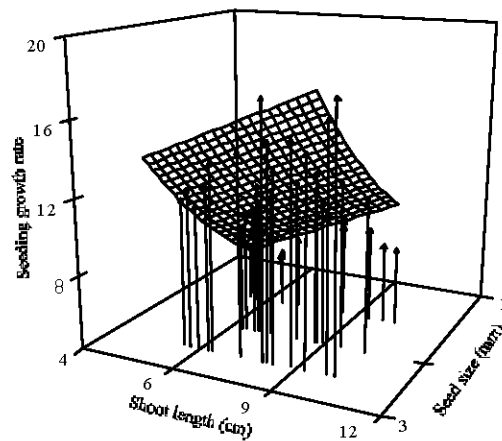


Fig. 1: Response surface of seedling growth rate vs. seed size (x₁) and shoot length (x₂)

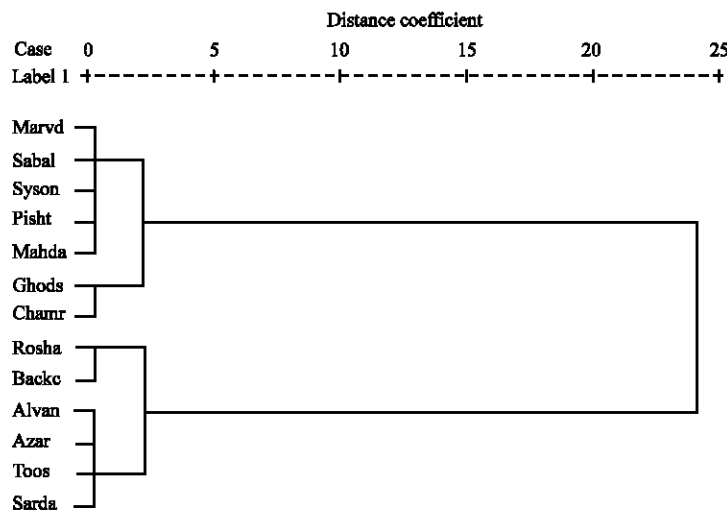


Fig. 2: Cluster analysis of wheat cultivars for shoot and root length by Euclidian distance

seed size and shoot length are two factors that strongly affect on seedling growth rate.

To determine of distance between cultivars, cluster analysis was carried out on the results of orthogonal contrasts for root and shoot length. As the basis of these results, all genotypes in this study was discriminated in two clusters (Fig. 2).

The results of cluster analysis showed that the most of facultative varieties and two rain fed genotypes have sited in one class and the other genotypes have been situated in the other classes.

Thus, in conclusion and according to the results of surface analysis, seed size, shoot length and seedling growth rate plays an important role in modulating and evaluating in wheat genotypes for improvement programs.

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