Pak. J. Bot., 40(3): 1121-1127, 2008.

GERMINATION BEHAVIOR OF WHEAT (*TRITICUM AESTIVUM*) VARIETIES TO ARTIFICIAL AGEING UNDER VARYING TEMPERATURE AND HUMIDITY

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Abstract

Seed viability of the conserved plant genetic resources is decreased even under the optimal storage conditions. Seeds of some plant species loose viability faster than others. Artificial seed ageing was used with the objective to determine its effect on seed viability, while outlining the contribution of different factors like temperature and moisture towards seed deterioration in two wheat varieties viz., Wheat Var. Margala-99 and Wafaq-2001; seeds having two types of initial moisture contents (low and high), were experimental materials. Three temperature regimes (40, 50 and 60°C); four incubation durations (24, 48, 72 and 96 h), under two types of relative humidity (low or high) were the treatments compared with control at 25°C. Seed viability after artificial ageing was determined by germination at 25°C (±2) under light conditions. Germination behavior of artificially aged seeds of both wheat varieties indicated that, the viability decreases with the increase in ageing incubation temperature. Incubation of seeds under high relative humidity has more pronounced effect in decreasing the seed viability than low relative humidity. Seeds incubated under high relative humidity, the viability decreases with increase in incubation period. Variation in germination behavior among varieties warrants study on a larger varietal group.

Introduction

Pakistan is 10th largest wheat (*Triticum aestivum* L.) producing country, where it is cultivated over some 8,358,000 hectares (Anon., 2006). Wheat is adapted to a wide range of soil and climatic conditions, consisting of diverse genotypic landraces that have developed over time.

Genebank of National Agricultural Research Centre (NARC), Islamabad maintains more than 24,000 accessions of different species. Seed conservation is faced with problem in view of conservation, as the viability is decreased even under the optimal storage conditions in the genebanks (Anon., 2005; Rao *et al.,* 2006). Seed moisture content (MC) and storage temperature are two of the most important factors affecting seed storage life (Harrington, 1973). The Handbook of Vigor Test Methods (Anon., 1996) lists the seed artificial ageing (AA) test as a recommended test for soybean and as a suggested test for several other species.

Ali *et al.,* (2006) showed that viability of wheat var. Chakwal-97 decreased with the increase in AA incubation temperature; while, Chhabra *et al*., (1992) suggested that seed quality as well as genotype might be responsible for reducing final germination of cultivars. Screening of wheat genotypes for seed ageing under real storage and artificially accelerated ageing conditions would provide guide lines to ensure appropriate storage of germplasm. Therefore, AA was used to check seed viability *via* germinability for two wheat varieties [Margala-99 and Wafaq-2001 (commonly cultivated in rainfed/arid area of Rawalpindi, Pakistan)]; to determine its effect on seed viability as a consequence of seed deterioration, outlining the contribution of different factors like temperature and moisture in seed ageing. Research work was done at NARC, Islamabad, according to Ali *et al*., (2006).

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Materials and Methods

Three temperature regimes (40, 50 and 60°C), four incubation durations (24, 48, 72 and 96h), two types of relative humidity (low or high) were the treatments compared with control (25°C). Seed viability, after AA, was determined by germination at 25°C (±2) under light conditions, using the paper method (Anon., 1996). Seeds of all treatments were soaked in 50ml distilled water at 25°C for 24h before taking EC value ms/m (Tekrony, 2003).

Data was recorded after incubation for seed weight (g) as indicator of seed MC, electrical conductivity (ms/m), germination percentage (%) for 7 days and shoot development [on weight basis (g)] as an indicator of seed vigor as on 7th day. The statistical analysis was applied on data using MS Excel package. Simple percentage and average computations were applied on the pooled data (Mario, 1994). Factorial design of MSTATC software was used to analyze germination data where varieties were kept separate to avoid complexity in analysis design.

Results and Discussion

Table 1 summarizes the basic data statistics, when seeds were treated at different temperatures and either incubated at low RH or high RH for ageing treatment. Significantly higher values of seed weight and EC values and lower germination percentage values are notable when incubated at high RH incubation condition.

Seed weight: The 50 seed weight (g) was recorded to know the amount of moisture content in seeds as indicated by increase in weight over control. The weight in control was 1.85g. (Fig. 1). High seed weight was recorded in seeds incubated under high RH either with initial HMC or LMC as compared to that at low RH or control (Fig. 1, Table 1) for both varieties. Seed weight was different within the varieties at different temperature treated seeds as well as between varieties as evident from data range and mean values (Table 1). The seeds MC of Margala-99 incubated at different temperatures did not differ significantly among themselves when incubated at low RH, and remained at par to control, whereas, when incubated at high RH their MCs were increased significantly.

The elevated moisture intake at 50°C was indicator of lost viability, while the slight increase at 40°C incubation was indicator of produced ageing effect during storage that was revealed on germination test. In Wafaq-2001 similar increase in seed MC was observed after treatment. It may be suggested that high RH should be induced while carrying the artificial accelerated ageing of wheat seed for all varieties.

Electrical conductivity (EC): EC value of seeds was different at three temperatures (Table 1). It ranged from 4.05 to 18.15 ms/m. Only the seeds incubated under high RH showed significant differences in EC. The EC value of control in Margala-99 was 4.76 ms/m; and that of Wafaq-2001 was 4.69 ms/m (Fig. 1). EC value followed the same trend as in seed weight behavior with respect to RH during incubation (Table 1). EC values were comparable to control for all temperatures and incubation duration regimes when incubated at low RH. For variety Margala-99 under high RH at 40°C the EC of the seeds with HMC were lower than that of respective EC of LMC seeds. Almost same trend was followed by EC for seeds at 60°C and high RH (Fig. 1). This particular trend was not found in Wafaq-2001. The temperature during soaking and/or the evaluation will influence the results of the conductivity test for several species (Loeffler *et al.,* 1988; Hampton *et al.,* 1992). Though varietal response was observed, use of 50°C as incubation

temperature under high RH could produce significant changes in EC values. The elevated EC values can be used as an indicator of aged seed (Fig. 1). Our results suggest that EC values are indicative of seed deterioration only when the seeds were incubated at high RH therefore soaking the seed to enhance initial seed MC is not necessary. However, this behavior needs to be studied across a wide range of genotypes. Similarly the incubation temperature for AA should be more than 40°C to achieve signified difference to control, but less than 50°C for wheat.

Seed germination percentage: Seed germination percentage in Margala-99 ranged from 0 to 95% for treated seeds (Table 1) and up to 96.5% for control. In case the RH was increased, germination dropped to zero at both high temperatures (50 and 60°C). Seed germination percentage in Wafaq-2001 ranged from 0 to 93%; and higher temperature's negatively affected on germination was noticed at high RH. Even the seeds at 40°C showed gradual decline in viability at high RH, dropping to 5.5% with incubation time (Fig. 2). High temperature and moisture accelerate loss of viability in most species (Barton, 1961). The germination response to AA was most influenced by the RH during the incubation period particularly at higher temperatures of 50°C and above (Fig. 2). The initial MC of seeds resisted ageing when being low. Banks *et al*., (1998) have stated that moisture content of 10% or below is essential for safe cotton seed storage, even for a short time period. Wheat varieties may behave differently but our result point towards high seed MC as well as RH, as being detrimental to seed viability.

In case of Margala-99 the germination response was quite different under the different RH conditions (Table 1). The germination was higher for corresponding LMC seeds under low RH and lower for the same under high RH. The germination declined with the increase in temperature and the effect was pronounced if the seed initial MC was high when incubated at low RH. Maurel (1997) has stated that the heat treatment (40°C) affected the germination after the seed MC had reached a threshold level. Hence, when incubating seeds at high RH the germination was lost at higher temperatures i.e., 50°C or above and even at 40°C the germination declined with the increase in duration of incubation (Fig. 2). Temperature and moisture play a significant role in determining the storage longevity of seeds. The pattern of seed ageing is, in general, described in terms of its water content during storage (Walters, 1998).

Genotypic variation for seed ageing among wheat genotypes exists (Madan *et al.,* 1989). It may be stated that Wafaq-2001 is less susceptible to storage especially at high RH. The results of EC value and germination data indicate that even one-day incubation at 50°C and high RH could deteriorate the seed. Loosing the germinability will render this method ineffective for evaluating the vigor. Thus the present study suggests that incubation of seed at a range of temperatures 40-50°C under high RH for at least 4 days would be suitable for comparing genotype behavior.

Shoot development: The statistical description of shoot development (weight basis) of both varieties is presented in Table 1 as averaged for each incubation temperature. As the seeds were grown between paper towels and supplied only with distilled water and light condition, it was only the seed potential vigor to support the growth. For all the germinated seeds, shoot developed (Fig. 2), however, the variability of data for this character makes it irrelevant for inferring any seed vigor relationship. Studying shoot weight at low RH for longer durations or at high RH at other temperatures in the range of 40 to 50°C may reveal inferable data.

Table 1. Summary of descriptive analysis (MS Excel) for seed weight (g), EC (ms/m), shoot weight (g) and germination percentage after artificial ageing of wheat seeds for varieties Wafaq-2001 and Margalla-99. Under each variety first 5 rows show data statistics for low RH and next 5 rows for high RH data analysis result.

Varieties		Seed weight (g)			Electrical conductivity (ms/m)			Shoot weight (g)			Germination percentage		
		60°C	50°C	40°C	60°C	50°C	40°C	60°C	50°C	40°C	60°C	50°C	40°C
Wafaq-2001													
(Low RH)	Mean	1.818	1.825	1.869	5.512	6.558	6.649	0.828	0.832	0.584	70.813	86.188	84.313
	Std. Error	0.025	0.028	0.017	0.085	0.101	0.578	0.025	0.027	0.033	5.369	1.674	0.801
	Std. Dev	0.071	0.078	0.049	0.241	0.285	1.635	0.070	0.077	0.093	15.184	4.735	2.267
	Minimum	1.730	1.680	1.813	5.208	6.238	4.310	0.738	0.740	0.470	48.000	81.000	81.000
	Maximum	1.948	1.925	1.950	5.863	7.058	8.135	0.930	0.968	0.698	88.500	93.500	87.000
(High RH)	Mean	2.538	2.401	2.309	12.000	10.453	6.881	-	0.398	0.594	0.000	5.813	58.500
	Std. Error	0.097	0.109	0.057	0.563	1.325	0.850	-	0.161	0.041	0.000	2.283	10.492
	Std. Dev	0.275	0.308	0.162	1.591	3.747	2.404	-	0.456	0.117	0.000	6.458	29.677
	Minimum	2.184	2.215	2.110	10.133	6.963	4.138	-	0.000	0.469	0.000	0.000	5.500
	Maximum	2.926	3.125	2.565	15.555	18.148	10.568	-	0.994	0.778	0.000	15.000	87.000
Margalla-99													
(Low RH)	Mean	1.681	1.699	1.752	4.861	5.248	4.607	0.657	0.730	0.630	79.938	86.500	90.313
	Std. Error	0.018	0.016	0.016	0.142	0.135	0.130	0.021	0.026	0.044	4.353	2.151	1.172
	Std. Dev	0.051	0.047	0.046	0.403	0.382	0.368	0.059	0.074	0.125	12.313	6.083	3.316
	Minimum	1.610	1.623	1.660	4.228	4.720	4.023	0.583	0.638	0.523	65.000	76.000	84.000
	Maximum	1.778	1.760	1.813	5.445	5.805	5.020	0.755	0.858	0.835	95.000	92.500	94.500
(High RH)	Mean	2.134	2.812	2.154	9.155	14.408	5.395	-	-	0.688	0.000	0.000	67.750
	Std. Error	0.057	0.133	0.037	0.229	0.788	0.443	-	-	0.041	0.000	0.000	5.849
	Std. Dev	0.162	0.376	0.105	0.648	2.230	1.253	-	-	0.116	0.000	0.000	16.544
	Minimum	1.893	2.245	2.030	8.535	11.365	4.053	-	-	0.580	0.000	0.000	43.500
	Maximum	2.320	3.350	2.368	10.623	18.005	7.785	-	-	0.928	0.000	0.000	84.500

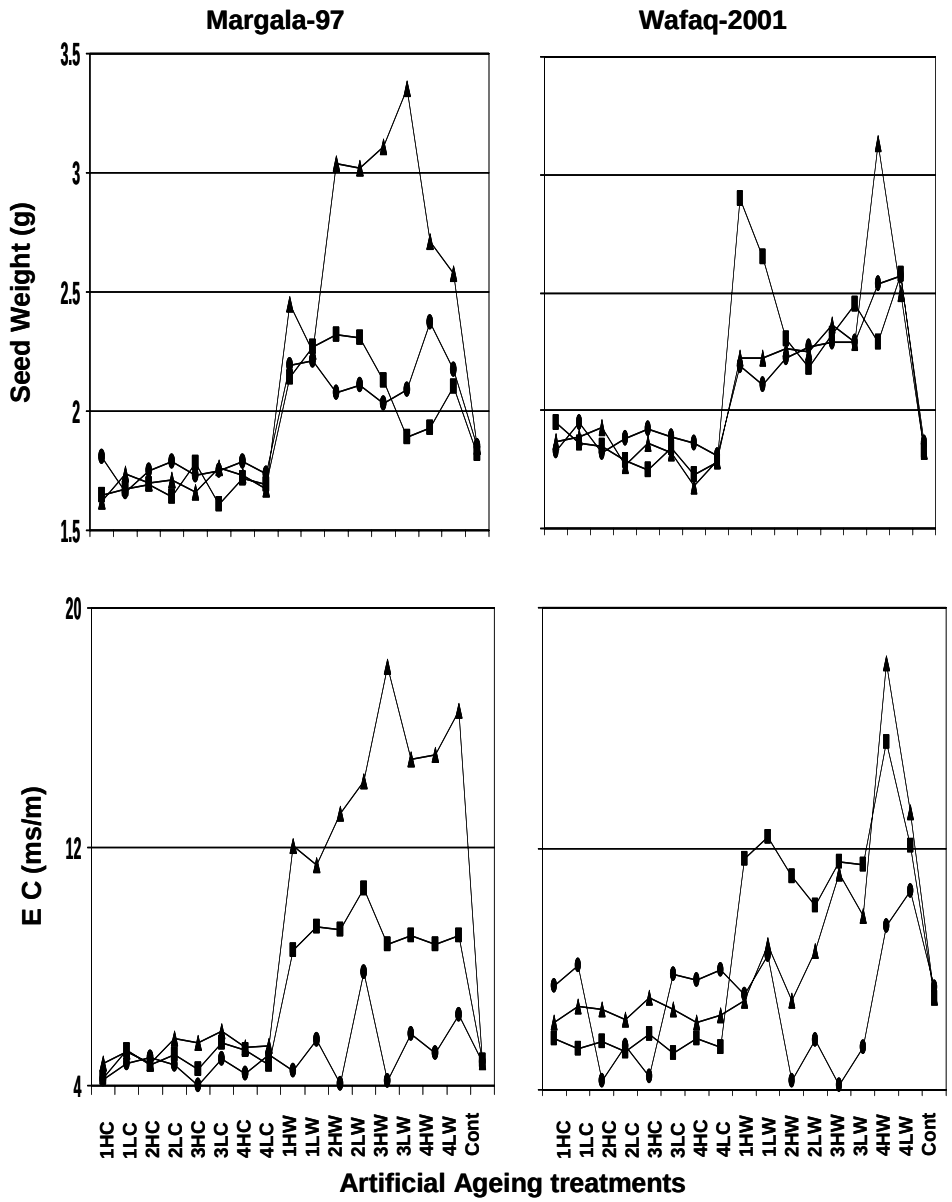

Fig. 1. Effect of artificial ageing on seed weight g (above) and EC value ms/m (below) of wheat seeds (varieties Margala-97 and Wafaq-2001, respectively).
In legends; ■ 60°C, ▲50°C, ● 40°C are the ageing incubation temperatures. At X-axis, 1-4 represent days of incubation (equivalent to 24~96 h), H=high seed moisture content, L=low seed moisture content, C=ambient RH and W= high RH up to 100% inside vessel of seed incubation.

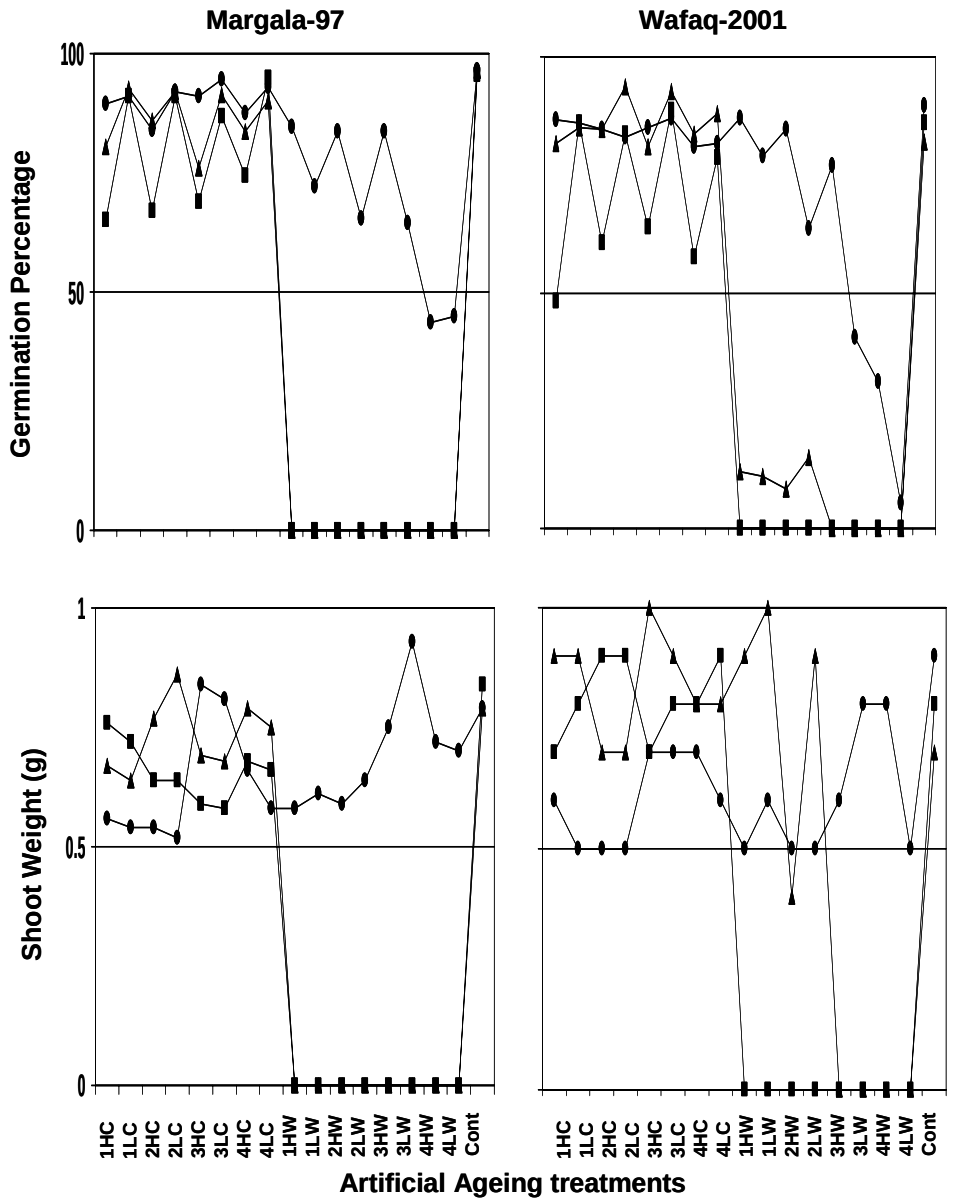

Fig. 2. Effect of artificial ageing on seed germination percentage (above) and seedlings shoot weight (below) of treated wheat seeds (varieties Margala-97 and Wafaq-2001, respectively).
In legends; ■ 60°C, ▲50°C, ● 40°C are the ageing incubation temperatures. At X-axis, 1-4 represent days of incubation, H=high seed moisture content, L=low seed moisture content, C=ambient RH and W= high RH up to 100% (inside vessel of seed incubation). The germination percentage is of 7th day. Ten seedlings were collectively weight (g) as an indicator of seedling vigor.

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(Received of publication 27 March 2008)