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Effect of ageing and priming on vigour parameters of wheat (*Triticum aestivum*)

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ABSTRACT

A study was conducted during 2006-10, seed of six varieties of wheat (*Triticum aestivum* L.) viz. C 306, PBW 502, WH 542,WH 711, WH 283 and RAJ 3765 were subjected to natural vis-à-vis accelerated ageing conditions and evaluated for relative storage potential of the seeds of respective varieties and then accelerated and natural aged seed lot were hydro and osmoprimed to find out the ability of priming treatment in the repair of deterioration sustained during natural and accelerated ageing. Seed quality parameters like germination percent, seedling length, seedling dry weight, vigour index-I and vigour index-II, and electrical conductivity, measured in, natural as well as accelerated aged seed lots. Overall, natural and accelerated ageing reduced germination, seedling length, seedling dry weight, vigour index-I and vigour index-II in all the varieties. However, priming reduced the deleterious effects of ageing and improved all vigour parameters in all the varieties. Hydration with Thiram @ 0.25%. Hydration with GA_3 (50 ppm for 16-18 hr), hydration with polyethylene glycol (PEG) for 16-18 hr, hydration with KNO_3 0.5% (16-18 hr) and hydration with $CaCl_2$ 2% for 16-18 hr) were proved beneficial for enhancing all vigour parameters significantly. Among all these priming treatments PEG and GA_3 were proved better/superior for increasing all vigour parameters in all the six varieties of wheat.

Key words: GA_3, Osmopriming, Polyethylene glycol, Thiram, Vigour index, Wheat

All seeds undergo aging process during long-term storage which leads to deterioration in seed quality, especially in the humid tropical regions. However, the rate of seed deterioration can vary among various plant species (Merritt *et al.* 2003). Aged seeds show decreased vigour and produce weak seedlings that are unable to survive once reintroduced into a habitat (Atıcı *et al.* 2007). Many of the processes implicated in seed aging during storage appear to be free-radical mediated, and lipid peroxidation is suggested to be a primary cause of deterioration in stored seeds (Wilson and McDonald 1986, McDonald 1999). Some protective mechanisms involving free radical and peroxide scavenging enzymes, such as catalase (CAT), peroxidase (POD) and superoxide dismutase (SOD) have been evaluated within the mechanism of seed aging (Hsu *et al.* 2003, Goel *et al.* 2003, Pukacka and Ratajczak, 2007). Biochemical and physiological deterioration during seed aging has been studied mostly under accelerated aging conditions using high temperature and high seed water content (McDonald 1999, Hsu *et al.* 2003). Although these studies allowed important progress towards the understanding of seed aging mechanisms, a major question has been raised whether mechanisms of seed aging are similar under conditions of accelerated aging and natural aging.

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Controlled deterioration treatments (accelerated ageing) involve exposing seeds to adverse storage conditions namely, increased temperature and relative humidity for specific period of time. It is assumed that the process of deterioration which occurs under these artificial ageing conditions to be similar to those which occur during natural ageing (Delouche and Baskin 1973). The main difference between artificial ageing and actual deterioration in poor storage conditions being the speed at which the changes occur in case of accelerated ageing. This can be used to investigate the factors responsible for seed deterioration in storage and the efficacy of various pre-sowing treatments that can improve the performance of a given seed lot. Improved seed invigoration techniques are being used to reduce the germination time, to get synchronized germination, improve germination rate and better seedling stand in many horticultural crops (Rudrapal and Nakamura 1998, Basra *et al*. 2004, Ashraf *et al.* 2008) and rice (Lee and Kim 2000, Basra *et al.* 2004). These invigoration techniques include hydropriming, osmoconditioning, osmohardening, hardening, hormonal and vitamin priming. Seed priming is an important method associated with the process of seed germination and is widely used to synchronize the germination of individual seeds with improved germination or seedling growth (Heydecker *et al*. 1973, Taylor and Harman 1990). Seed priming enhances seed performance by rapid and uniform germination, normal and vigorous seedlings in different crops which have practical agronomic implications, notably under adverse germination conditions (McDonald 2000, Cantliffe 2003). It permits seedling development in a wide range of agro-climatic conditions and decreases sensitivity to external factors (Ashraf and Foolad 2005, Welbaum *et al*. 1998). Seeds performance of various crops can be improved by inclusion of plant growth regulators and hormones during priming and other pre-sowing treatments (Lee *et al*. 1998). Hormone like salicylic acid has also proved for alleviating salinity stress in wheat (Afzal *et al*. 2005). During priming, the germination-related processes are initiated but the emergence of the radicle is prevented since hydration is followed by drying of seeds. The beneficial effects of seed priming with different priming agents have already been successfully expressed in many crop plants, for instance mustard (Srinivasan *et al*. 1999), chickpea (Kaur *et al*. 2002), maize (Nawaz and Ashraf 2010), sunflower (Kaya *et al*. 2006), wheat (Iqbal and Ashraf 2007, Perveen *et al*. 2010), cotton (Casenave and Toselli 2007), rice (Habib *et al*. 2010) and sugarcane (Patade *et al*. 2009).Therefore the present study was conducted to explore the role of chemical and hormonal seed priming in improving emergence,early stand establishment and ion uptake in wheat (*Triticum aestivum* L.) seedlings.This paper reports the improvement of seed quality obtained in artificial and natural aged Indian wheat cultivars by osmo and hydropriming treatments.

MATERIALS AND METHODS

The present research work was carried out in the laboratories of Department of Seed Science and Technology, CCS Haryana Agricultural University, Hisar from 2006 to 2009. Seed material comprised of six varieties of wheat, viz. C306, PBW 502, WH 542, WH 711, WH 283 and RAJ 3765 having germination above minimum seed certification standard (MSCS) was collected at the time of sowing of crop and stored in ambient conditions. For defining the variables for artificial ageing, seed of all six varieties were artificially aged (40±1ºC/72 hr) and observation was recorded after ageing. In case of natural ageing, observation was recorded quarterly on the stored wheat seed in cotton bags in ambient conditions up to one year till germination fall below as compared to fresh seed lot and in case of natural ageing data on seed germination and seed vigour was recorded at quarterly intervals till germination fell below minimum seed certification standards which is 85 per cent for wheat. Standard germination test was conducted as per ISTA seed testing rules (ISTA 2001) and vigour index was calculated (Abdul-Baki and Anderson 1973).

Natural and artificial aged seeds of these varieties were osmotically primed on one layer of filter paper wetted with distilled water then treat dry seeds with 0.25% thiram, hydration with GA_3 (50 ppm for 16-18 h), hydration with polyethylene glycol (PEG) for 16-18 hr, hydration with KNO_3 0.5% (16-18 hr) and hydration with $CaCl_2$ 2% for 16-18 hr. Seeds were kept in a single layer in Petridishes (9 cm dia.) and replicated 3 times for each cultivar. All these seeds then dried in shade at room temperature for overnight on filter paper for one day. After priming, these seeds were subjected to germination testing, which was estimated following ISTA procedure (ISTA 2001) in paper towels soaked in water with 100 seeds in each replicated three times. After 8 days, 5 seedlings from each replication were randomly removed and the seedling length measured. The dry weight of 5 seedlings together was also obtained. The seed vigour index-I was calculated as the product of seedling length and germination whereas the seed vigour index-II was calculated the product of seedling dry weight (Abdul-Baki and Anderson 1973). The data was subjected to analysis of variance to get the treatment and cultivar effect and their interactions.

RESULTS AND DISCUSSION

Natural and accelerated ageing of wheat seeds showed significant deleterious effect on germination and other quality traits. Overall reduction in germination, seedling length, seedling dry weight, vigour index-I and vigour index-II . In the present study, all the six varieties of wheat showed a considerable variability in respect to viability. As indicated in Table 1 germination percentage decreased after natural and accelerated ageing in all the six varieties in spite of this in natural aged seed lot

The significant reduction in germination was observed in variety RAJ 3765 (71.33%) than C 306 (71.33%) and in accelerated aged seed lot significant reduction was recorded in variety RAJ 3765 (44.00%) followed by C 306 (47.00%). Similar results were reported in okra (Narwal 1995). Incase of seedling length was recorded significantly lower after accelerated ageing in all the six varieties (Table 2). The results indicated that in natural aged seed lot the PBW 502 recorded higher seedling length (13.57) where as RAJ 3765 recorded significantly lower (12.50). In accelerated aged lot

Table 1 Effect of natural and artificial ageing on germination of wheat

Varieties	Ageing			
	Fresh	Natural	Accelerated	Mean
C 306	98.67	71.33	47.00	75.33
PBW 502	97.67	79.00	55.00	77.22
WH 542	98.00	74.33	49.33	73.88
WH 711	97.33	77.67	50.00	75.00
WH 283	95.33	74.00	52.67	74.00
RAJ 3765	98.00	71.33	44.00	68.11
Mean	97.50	74.61	49.67	

Factors	*CD*	*SE(d)*	*SE(m)*
Treatment (A)	1.361	0.668	0.472
Varieties (B)	1.924	0.945	0.668
Factor (A × B)	3.333	1.637	1.157

Table 2 Effect of natural and artificial ageing on seedling length of wheat

Varieties	Ageing			
	Fresh	Natural	Accelerated	Mean
C 306	30.00	13.17	13.03	18.73
PBW 502	28.07	13.57	13.50	18.38
WH 542	27.33	13.50	13.37	18.06
WH 711	26.33	12.60	12.73	17.22
WH 283	27.67	13.20	12.83	17.56
RAJ 3765	25.33	12.50	11.93	16.58
Mean	27.46	13.09	12.91	

Factors	*CD*	*SE(d)*	*SE(m)*
Treatment (A)	0.686	0.337	0.238
Varieties (B)	0.970	0.476	0.337
Factor(A × B)	1.680	0.825	0.584

significantly higher seedling length was recorded in PBW 502 (13.50) and lower seedling length recorded in RAJ 3765 (11.93) and dry weight per seedling was decreased after artificial ageing in all the six varieties of wheat (Table 3). In natural aged seed lot it was recorded maximum in variety WH 542 (12.43) and minimum in variety WH 283 (11.13) which is significantly lower. In case of accelerated aged seed lot maximum dry weight recorded in PBW 502 (13.60) and minimum in variety C 306 (11.13).

The decreased seedling length and dry weight after accelerated ageing were recorded by Nagrajan *et al.* (2004) and Doijode (1999) in okra. In the present study, the variability in the seeds of all the six varieties of wheat were evaluated for seed vigour based on seedling length and seedling dry weight after accelerated ageing. Seed vigour also decreased after natural and accelerated ageing but seed vigor index–I expressed as a product function of standard germination percent and seedling length is depicted in Table 4. The vigor index-I decreased after accelerated ageing in all the six varieties. In case of natural aged seed lot the maximum vigor index-I was found in PBW 502 and minimum vigor index was recorded in RAJ 3765, in accelerated aged seed lot maximum vigor index was recorded in PBW 502 and minimum vigor index was recorded in variety RAJ 3765. The data for seed vigor index-II, calculated by multiplying germination percent with seedling dry weight presented in Table 5. Vigor index-II was decreased after accelerated ageing in all the six varieties of wheat. In case of natural aged seed lot maximum value was recorded in PBW 502 and minimum vigor index-II was recorded in WH 283. In accelerated aged seed lot maximum vigor index-II was recorded in variety WH 542 and minimum was recorded in variety C 306 .The reduction in seed vigour after accelerated ageing were also

Table 3 Effect of natural and artificial ageing on seedling dry wt of wheat

Varieties	Ageing			
	Fresh	Natural	Accelerated	Mean
C306	12.60	11.87	11.13	11.87
PBW502	13.20	11.87	13.60	12.89
WH542	13.10	12.43	12.73	12.76
WH711	11.50	11.23	12.00	11.58
WH283	11.70	11.13	11.50	11.44
RAJ3765	12.50	12.07	11.50	12.02
Mean	12.43	11.77	12.08	

Factors	*CD*	*SE(d)*	*SE(m)*
Treatment (A)	0.420	0.206	0.146
Varieties (B)	0.593	0.291	0.206
Factor (A × B)	NS	0.505	0.357

Table 4 Effect of natural and artificial ageing on vigor index-I of wheat

Varieties	Ageing			
	Fresh	Natural	Accelerated	Mean
C 306	2960.00	939.41	612.41	1403.94
PBW 502	2740.87	1072.03	742.50	1518.46
WH 542	2678.67	1003.45	659.54	1447.22
WH 711	2563.33	978.64	636.50	1392.82
WH 283	2637.67	976.80	675.75	1430.07
RAJ 3765	2482.67	891.62	524.92	1299.73
Mean	2668.86	976.99	550.27	

Factors	*CD*	*SE(d)*	*SE(m)*
Treatment (A)	62.960	30.918	21.862
Varieties (B)	89.039	43.725	30.918
Factor (A × B)	154.219	75.733	53.551

Table 5 Effect of natural and artificial ageing on vigor index-II of wheat

Varieties	Ageing			
	Fresh	Natural	Accelerated	Mean
C 306	1243.13	847.20	489.40	859.91
PBW 502	1289.40	938.80	670.93	966.38
WH 542	1283.60	911.87	701.73	965.73
WH 711	1119.13	872.53	600.53	864.07
WH 283	1115.50	812.93	605.03	844.49
RAJ 3765	1225.00	860.87	539.73	875.20
Mean	1212.63	874.03	601.23	

Factors	*CD*	*SE(d)*	*SE(m)*
Treatment (A)	34.373	16.879	11.936
Varieties (B)	48.610	23.871	16.879
Factor (A × B)	84.195	41.346	29.236

reported earlier by Basra *et al.* (2003) in cotton and Khan *et al.* (2005) in turnip.

The seed germination process is initiated when seeds begin to imbibe water and it is common practice in many laboratories to submerge seeds under water for one to several hours before planting to hasten the germination (Kay *et al.* 1977). The present investigation revealed that various priming treatments enhanced the standard germination in natural and accelerated aged seed lots in all the varieties of wheat. The increased seed germination by hydration-dehydration can be due to quenching effect on the propagation of free radicals (Basu 1976). The beneficial effect of priming is known to occur due to higher mitochondrial activity, formation of more high energy compounds and vital bio-molecules (Henckel 1961). In present study effect of various priming treatments was observed on standard germination of natural aged seed lot of all six cultivars, viz. C 306, PBW 502, WH 542, WH 711, WH 283 and RAJ 3765.It was observed that natural aged seed lot was deteriorated and standard germination was observed below minimum seed certification standards (MSCS).

Priming treatment to seed lot of all the cultivars showed significant improvement in germination. Within treatment all treatments also showed improvement up to significant level. The improvement in natural aged seed lot of all six cultivars was recorded significantly higher in GA_3 primed seed lots which improve the seed above minimum seed certification standard. Among the varieties WH 283 showed maximum (11.33%) improvement and minimum improvement was recorded in C 306 (9.34%) when primed with GA_3 (Table 6), which indicated that this cultivar is able to recover after stress conditions by priming. Similar results were reported by Pallavi *et al.* (2003) in sunflower, Pandita and Nagrajan (2004) in bitter gourd, Tajbakhsh *et al.* (2004) reported that GA_3 alone was able to improve the seed germination in onion. Similar results were observed by Khan *et al.* (2003) in sunflower.

In Table 7 effect of various priming treatments was observed on standard germination of accelerated aged seed lot of all six cultivars, viz. C 306, PBW 502, WH 542, WH 711, WH 283 and RAJ 3765.It was observed that accelerated aged seed lot was deteriorated and standard germination was observed below minimum seed certification standards (MSCS).

Priming treatment to seed lot of all the cultivars showed significant improvement in germination. Within treatment all treatments also showed improvement up to significant level. The improvement in accelerated aged seed lot of all six cultivars was recorded significantly higher in GA_3 primed seed lots which improve the seed above minimum seed certification standard. Among the varieties PBW 502 showed maximum (14.00%) improvement and minimum improvement was recorded in RAJ 3765 (6.33%) when primed with GA_3.

Effect of various priming treatments was observed on standard germination of natural and accelerated aged seed lot of all six cultivars of wheat. It was observed that priming treatment to natural and accelerated aged seed lot show significant improvement in germination. This type of response has been reported by various workers (Tarquis and Bradford (1992) in lettuce, Goel *et al*. (2003) in cotton, and Benamar *et al*. (2003) in pea seed. Nagarajan and Pandita (2001) reported that osmo priming can delay the onset of deterioration caused by accelerated ageing in tomato. Karssen *et al*. (1989) concluded after studying the effect of priming in tomato, celery, and lettuce seeds that after priming, the seeds contained

Table 6 Effect of priming on seed germination of a natural aged seed in Wheat

Varieties	Treatments						
	T_0	T_1	T_2	T_3	T_4	T_5	Mean
C 306	71.33	78.67	80.67	78.00	77.00	74.33	76.67
PBW 502	79.00	84.33	88.67	85.00	82.67	80.00	83.28
WH 542	74.33	79.67	85.33	81.33	77.67	74.33	78.78
WH 711	77.67	82.33	87.00	83.33	80.33	77.33	81.33
WH 283	74.00	79.00	85.33	80.67	77.00	74.33	78.39
RAJ 3765	71.33	77.33	82.00	78.67	75.33	72.67	76.22
Mean	74.61	80.22	84.83	81.17	78.33	75.50	

Factors	*CD*	*SE(d)*	*SE(m)*
Treatment (A)	1.1653	0.5844	0.4133
Varieties (B)	1.1653	0.5844	0.4133
Factor (A × B)	NS	1.4316	1.0123

T_0 – Control, T_1 – Hydration (16-18 hr) a followed by dry dressing with Thiram @ 0.25% and drying at room temp. below 25ºC, T_2 – Hydration with GA_3 (50 ppm for 16-18 hr), T_3 – Hydration with PEG (6000) (16-18 hr), T_4 – 0.5% KNO_3 hydration (16-18 hr) and drying at room temp, T_5 - 2% $CaCl_2$ at room temperature (16-18 hr) and surfaced drying.

Table 7 Effect of priming on seed germination of artificial aged seed in Wheat

Varieties	Treatments						
	T_0	T_1	T_2	T_3	T_4	T_5	Mean
C 306	44.00	51.33	55.00	52.00	49.33	48.67	50.06
PBW 502	55.00	60.33	69.00	65.33	60.67	63.33	62.28
WH 542	50.00	55.33	62.33	61.33	58.67	57.67	57.56
WH 711	49.33	55.33	61.00	63.00	57.00	56.33	57.00
WH 283	52.67	54.67	63.67	60.67	60.00	60.00	58.61
RAJ 3765	43.67	52.00	50.00	59.00	59.00	55.33	53.17
Mean	49.11	54.83	60.17	60.22	57.44	56.89	

Factors	*CD*	*SE(d)*	*SE(m)*
Treatment (A)	1.501	0.752	0.531
Varieties (B)	1.501	0.752	0.531
Factor (A × B)	3.677	1.841	1.302

increased level of products such as protein (enzymes). Generally, priming of these 3 species permitted rapid and uniform germination by stimulating extensibility of the cell wall in the radicle and by weakening endosperm cell wall in front of the radical tip. Priming reduced the deleterious effects of ageing and improved all vigor parameters in all the varieties. Hydration with Thiram @ 0.25%, hydration with GA_3 (50 ppm for 16-18 hr), hydration with polyethylene glycol (PEG) for 16-18 hr, hydration with KNO_3 0.5% (16-18 hr) and hydration with $CaCl_2$ 2% for 16-18 hr were proved beneficial for enhancing all vigor parameters significantly. Among all these priming treatments PEG and GA_3 were proved better/superior for increasing all vigor parameters in all the six varieties of wheat.

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