

Vigor evaluation of fifteen months stored delinted *Gossypium hirsutum* L. varieties picked at different intervals

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Abstract: Seed ageing is influenced by two factors moisture and temperature. The deterioration of the stored seed is a natural phenomenon and the seeds tend to lose viability even under ideal storage conditions. The present study was carried out in the Department of Seed Science and Technology Section, CCS Haryana Agricultural University, Hisar, India. This study aimed at evaluating efficiency of different pickings of cotton seed vigor potential during storage. Thereby, three American cotton varieties H-1098 (I), H-1117 and H-1236 were taken into account, were picked at different intervals and after ginning and delinting, initial seed vigor evaluation was done and seeds were stored under controlled condition at 20⁰ C with 6 per cent moisture content for fifteen months. Evaluations of germination per cent, seedling length, seedling weight, vigor indices were performed after five, ten and fifteen months of storage (5, 10, 15 months). It was observed that seed samples of second pick stored seed showed higher vigor followed by first pick stored sample whereas minimum was recorded under third pick stored sample.

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Introduction

A key component of the performance of crop seeds is the complex trait of seed vigor. Crop yield and resource use efficiency depend on successful plant establishment in the field and it is the vigor of seeds that defines their ability to germinate and establish seedlings rapidly, uniformly across diverse environmental conditions. Our knowledge of the regulation of seed vigor has developed in recent times, but understanding the basis of variation and therefore seed performance during the establishment of crops remains limited.

The longevity of a seed lot is the length of time the seeds remain viable after reaching physiological maturity (9). For seed storage purposes, longevity is used synonymously with storability. To preserve the initial seed quality, seeds must be properly stored between the time of harvest and the planting of a subsequent crop. Delouche (9) defined the total seed storage period as comprising segments of bulk storage, which is the period from harvest through packaging including conditioning. Packaged storage was defined as the period between packaging and distribution; and distribution storage covered the period between sales to farmers, including time at wholesalers and retail outlets.

Seed ageing leads to reduction in seed quality, performance and stand establishment. In spite of countless investigations and speculations, we do not

know the cause of loss in viability during storage (22), however, it is clear that most functional macromolecules, proteins, nucleic acids and lipids undergo changes during seed deterioration which ultimately precede death. Lipid auto oxidation has also been suggested to be one of the causes of seed ageing (29) which involve the production of free radicals. Once seed deterioration has happened, this catabolic process cannot be reversed. It is a sequence of events beginning with a chain of biochemical events, predominantly membrane damage and impairment of biosynthetic reactions, and then the resulting losses of various seed performance attributes, starting with reduced germination rate, reduced field emergence, increased numbers of abnormal seedlings and finally seed death.

Seed deteriorates through normal physiological reactions and changes that occur within the seed over time. These changes result in the accumulation of deleterious byproducts that increase the seed's vulnerability to external challenges and decrease the ability of the seed to survive (13). This process is inevitable and irreversible and only its rate can be controlled (9). The process of deterioration, therefore, involves several physiological and structural changes within seed. Structural changes involve membrane permeability, proteins, sugars, nucleic acids, fatty acids and volatile substances, while physiological processes involve enzyme activity, respiratory

competence, lipid peroxidation and physiological repair mechanisms (23,27). According to (26) optimum protocols for seed storage must take into account the chemical composition of the seed, the physiological status of the seed, and the physical status of water within the seed. Two models of seed deterioration have been proposed (27).

This study was set to investigate the influence of various pickings on storability and vigor potential of American cotton varieties. The influence of different picked materials under controlled storage condition on seed vigor and germination over time. The experiment was laid in a Complete randomized design with three replicates.

Material and Methods

Two year study to evaluate the effect of pickings with fifteen days intervals was conducted on three *Gossypium hirsutum* L. varieties. Seeds were collected from the Cotton Section, Department of Genetics and Plant Breeding, CCS Haryana Agricultural University. Different cotton varieties were sown in Randomized Block Design (RBD) with three replications during the month of April 2012-13 and 2013-14 in the cotton section field research area. The non experimental rows were also maintained to avoid border effects. Recommended agronomic package and practices were followed to grow good and healthy crop. The observations were recorded on seeds collected from various pickings. Fifteen plants were tagged for picking treatments and three bulk samples of each treatment /variety/replication were ginned and delinted separately. The pickings were started after 50 percent boll opening and all the three pickings in each variety were taken with 15 days interval and evaluated for seed quality in laboratory of Department of Seed Science and Technology in completely randomized design with following observations.

Seed Lots (P) Three of each pickings in three American cotton varieties

Gossypium hirsutum: H-1098-(I), H-1117, H-1236,

Seed moisture content (%) of above fresh seeds at each picking stage were estimated by hot air oven method ($80 \pm 1^\circ\text{C}$ for 24 h) and expressed in percentage.

Seed weight (g) of 100 seeds (replicated thrice) of each variety and each picking was recorded and expressed in g.

Seed density (g/cc) from each sample were taken after weighing seeds on electronic balance than after dipping it in toluene solution.

Seed density (g/cc) = Weight of 100 seeds / Volume of displaced seed (CC)

Standard Germination (%) as per ISTA, 2011

Final count was recorded on 12 th day (ISTA,

2011). Normal seedlings were expressed as percent germination. ☐

Seedling length (cm) of ten randomly selected normal seedlings was recorded and average seedling length was expressed in centimeters (cm). ☐

Dry weight (mg) ten randomly taken normal seedlings whose length was measured were dried in a hot air oven for 24 h at $103 \pm 1^\circ\text{C}$.

Vigour index –I & II

Vigour Index–I = Standard germination (%) × average seedling length (cm)

Vigour Index–II=Standard germination (%) × average seedling dry weight (mg)

Electrical conductivity test ($\mu\text{S cm}^{-1}\text{g}^{-1}$) as per ISTA, 1999.

Three replicates of 50 normal seeds were soaked in a 100 ml beaker containing 75 ml of distilled water and kept at 25°C . The leachetes were measured after 24 h with systronic conductivity meter 306 and it was expressed as $\mu\text{S cm}^{-1}\text{g}^{-1}$.

Result and Discussion

The experiment result showed significant difference between different picked sored seeds and varieties on seed germination percent and vigor. Germination and seed vigor percentage was found to decrease with increase in storage duration. The mean germination and vigor irrespective of various pick stored seeds samples, was found to decrease continuously from initial 75.52 to 64.3 % after 15 months of storage respectively. In the present study, second pick stored samples seeds recorded higher seed germination (81.11 to 68.56%) throughout the storage period of fifteen months. The second pick stored sample in sealed plastic containers had the highest seed germination per cent 81.11% (after five months) which declined to 68.56 (after fifteen months) % whereas minimum germination percent found in third picked stored seeds which was 67.11 % after five months and declined to 58.44% after fifteen months of storage. During deterioration, vigour is the first component of seed quality, which is lost, this is followed by a loss of germination capacity and viability as reported by (25). Second picking seeds though decrease in vigour showed better performance during storage than both first and third picking stored seeds. Germination of the seeds observed decreasing progressively with ageing. Decreasing of germination per cent in aged seeds can be due to reduction of α -amylase activity and carbohydrate contents (4) or denaturation of proteins (19) or it may be related to chromosomal aberrations that occur under long storage conditions (1). The main mechanisms for the reduced germination as a result of seed deterioration are not known, but some studies showed that lipid peroxidation caused by oxidative damage can lead to

inactivation and/or depletion of key enzymes of recipient protein transport or ion channels, as well as

impairment of RNA and DNA synthesis (5,16,18).

Table: 1 Effect of storage on seed quality parameters after five, ten and fifteen months of storage

	Five Months				Ten Months				Fifteen Months				
	P1	P2	P3	MEAN	P1	P2	P3	MEAN	P1	P2	P3	MEAN	
Germination (%)	H-1098(I)	80	82	69.67	77.22	75	77.67	66	72.89	68.33	70.33	61.67	66.78
	H-1117	77	80	64.01	73.67	70	72.33	60.67	67.67	62.67	66	56.33	61.67
	H-1236	78	81.33	67.67	75.67	71.33	73.67	63	69.33	66.67	69.33	57.33	64.44
	MEAN B	78.33	81.11	67.11	75.52	72.11	74.56	63.22	69.96	65.89	68.56	58.44	64.3
	CD	V=0.998, P=0.998, VxP=1.729				V=0.792, P=0.792, VxP=N/A				V=0.859, P=0.859, VxP=1.488			
	SeM	V=0.333, P=0.333, VxP=0.577				V=0.264, P=0.264, VxP=0.458				V=0.287, P=0.287, VxP=0.497			
Seedling Length (cm)	H-1098(I)	35.17	38.05	35.59	36.27	31.64	33.46	30.68	31.93	28.26	31.07	26.62	28.65
	H-1117	31.34	34.85	28	31.4	27.87	30.37	24.8	27.68	21.91	23.94	19.18	21.68
	H-1236	32.9	35.01	29.82	32.57	28.77	30.67	25.85	28.43	21.62	27.26	20.28	23.05
	MEAN B	33.13	35.97	31.14	33.41	29.43	31.5	27.11	29.35	23.93	27.43	22.02	24.46
	CD	V=0.885, P=0.885, VxP=1.533				V=0.613, P=0.613, VxP=1.061				V=0.805, P=0.805, VxP=1.395			
	SeM	V=0.296, P=0.296, VxP=0.512				V=0.205, P=0.205, VxP=0.354				V=0.269, P=0.269, VxP=0.460			
Seedling weight (mg)	H-1098(I)	0.387	0.405	0.372	0.388	0.373	0.398	0.361	0.377	0.323	0.334	0.266	0.308
	H-1117	0.346	0.385	0.308	0.346	0.339	0.37	0.304	0.338	0.256	0.279	0.249	0.262
	H-1236	0.375	0.386	0.334	0.365	0.365	0.379	0.324	0.356	0.28	0.296	0.241	0.272
	MEAN B	0.369	0.392	0.338	0.366	0.359	0.382	0.33	0.357	0.286	0.303	0.252	0.281
	CD	V=0.006, P=0.006, VxP=0.011				V=0.004, P=0.004, VxP=0.006				V=0.003, P=0.003, VxP=0.005			
	SeM	V=.002, P=0.002, VxP=0.004				V=0.001, P=0.001, VxP=0.002				V=.001, P=0.001, VxP=0.002			
VIGOR I	H-1098(I)	2813	3120	2479	2804	2373	2599	2025	2332	1,931	2,185	1,641	1,919
	H-1117	2413	2788	1792	2331	1951	2197	1505	1884	1,373	1,581	1,080	1,345
	H-1236	2566	2847	2017	2477	2053	2259	1628	1980	1,442	1,890	1,163	1,498
	MEAN B	2597	2918	2096	2537	2125	2352	1719	2065	1,582	1,885	1,295	1,587
	CD	V=7.40, P=67.40, VxP=116.73				V=47.13, P=47.13, VxP=N/A				V=5.92, P=55.92, VxP=96.86			
	SeM	V=2.51, P=22.51, VxP=38.99				V=5.74, P=15.74, VxP=27.26				V=8.68, P=18.68, VxP=32.35			
VIGOR II	H-1098(I)	30.93	33.18	25.92	30.01	28	30.91	23.83	27.58	22.05	23.52	16.38	20.65
	H-1117	26.66	30.79	19.71	25.72	23.73	26.79	18.44	22.99	16.07	18.43	14.03	16.18
	H-1236	29.25	31.4	22.63	27.76	26.04	27.92	20.43	24.8	18.66	20.54	13.8	17.67
	MEAN B	28.95	31.79	22.75	27.83	25.92	28.54	20.9	25.12	18.93	20.83	14.74	18.17
	CD	V=.530, P=0.530, VxP=0.918				V=.381, P=0.381, VxP=0.660				V=.353, P=0.353, VxP=0.611			
	SeM	V=.177, P=0.177, VxP=0.307				V=.127, P=0.127, VxP=0.220				V=0.118, P=0.118, VxP=0.204			
EC ($\mu\text{S cm}^{-1}\text{g}^{-1}$)	H-1098(I)	0.332	0.352	0.351	0.345	0.404	0.391	0.443	0.412	0.486	0.456	0.494	0.479
	H-1117	0.321	0.343	0.341	0.335	0.432	0.404	0.479	0.438	0.532	0.509	0.567	0.536
	H-1236	0.401	0.437	0.409	0.416	0.438	0.396	0.471	0.435	0.521	0.486	0.542	0.516
	MEAN B	0.351	0.377	0.367	0.365	0.425	0.397	0.464	0.428	0.513	0.483	0.534	0.51
	CD	V=.002, P=0.002, VxP=0.004				V=.007, P=0.007, VxP=0.012				V=.005, P=0.005, VxP=0.009			
	SeM	V=0.001, P=.001, VxP=0.001				V=.002, P=0.002, VxP=0.004				V=.002, P=0.002, VxP=0.003			

V= Variety, P= Picking, VxP = Variety x Picking (interaction)

Maximum seedling length 35.97 cm was observed in second pick stored sample compared to other stored samples. Minimum 31.14 cm was found in third pick stored samples. During storage for fifteen months continuous decline in seedling length was observed. After fifteen months of storage maximum seedling length 27.43 cm was observed in second pick stored samples and minimum 22.04 cm was observed in third pick stored samples whereas 23.93 cm was observed in first pick stored samples after fifteen months of storage. Seedling weight also found decreasing in all pick stored samples and among all cotton varieties. But maximum seedling weight 0.392 mg was observed in second pick stored

samples and minimum 0.338 mg in third pick stored samples after five months of storage. After fifteen months of controlled storage maximum seedling weight 0.303 mg was found in second pick stored sample and minimum 0.252 mg observed in third pick stored samples after fifteen months of storage. Among all pickings second pick showed better seedling weight and seedling dry weight, which indicated less vigour loss in physiological matured seeds during storage. Reduction in seedling length and seedling dry weight may be due to decrease in mobilization of reserve substances during germination of the stored seeds (10). Earlier reports have shown that storage could cause depletion of important nutrient reserves

((18). Ageing led to decrease in seedling length and seedling dry weight which is confirmed with the earlier findings of (30) in okra, (15) in lentil, (24) in cotton and (3) in sorghum.

Varietal differences during storage in the present study were highly significant. Among the American cotton varieties, H-1117 showed poor results than other two varieties H-1098 (I) and H-1236. H-1098 (I) decreased from 3120 vigor indices after five months of storage to 2185 after fifteen months of storage in vigor indices. It was however, observed that all varieties vigor index fell after fifteen months of storage. After five months of storage maximum vigor index I, II was found in second pick stored samples 2918, 31.78 whereas after fifteen months it declined to 1885 and 20.83 in second pick stored samples. Minimum vigor index was observed in third pick 2096, 22.75 after five months of storage which declined to 1295, 14.74 after fifteen months of storage. Results are in conformity with the findings of (14). Reported a reduction in seedling dry mass after different periods of accelerated ageing on seeds of different varieties of lentil (*Lens culinaris* Medikus) (17). Similar findings were reported by (30) in okra, (7) in chickpea and (8).

The Electrical Conductivity of seed leachates increased in all varieties with increase in duration of storage. These results are similar to the results obtained in seeds of cotton by Presley (1958). Minimum leakage of electrolytes $0.351 \mu\text{S cm}^{-1}\text{g}^{-1}$ were observed in first pick stored sample whereas maximum found in second pick stored sample $0.377 \mu\text{S cm}^{-1}\text{g}^{-1}$ after five months of storage. Maximum electrical conductivity $0.534 \mu\text{S cm}^{-1}\text{g}^{-1}$ was observed in third pick stored sample whereas minimum $0.483 \mu\text{S cm}^{-1}\text{g}^{-1}$ observed in second pick stored sample. Membrane disruption is one of the main reasons of seed deterioration as a result, seed cells are not able to retain their normal physical condition and functioning, which in turn resulted in increased leaching of electrolytes. Effect of storage time period was significant on leakage of electrolytes in all varieties. Leakage of electrolytes increased with increase in storage period. It was studied that under storage, enhancement in leakage of electrolyte was observed in all pickings but enhancement of leakage was high in third pick stored sample in comparison to other picking sample in both American cotton varieties. Enhancement of leakage was due to alterations in the membranes of aged seeds which led to electrolyte leakage. Cell expansion was reduced by ageing to a greater extent comparing to cell division as reported by (6). During ageing there was destruction in the membrane which reduces its vigour (28) because of the destruction of the membrane system, many electrolytes flowed out of cells, so the vigor of seed

reduced. Seed deterioration leads to lipid peroxidation that subsequently causes membrane perturbation (10) and increase in free fatty acid which destroy seed (Goel *et al.*, 2003). The membrane integrity lost by damage of phospholipids leads to increased membrane permeability and exit of electrolytes and other substances, such as enzymes, from cells (31). Results were in conformity with earlier findings of (10) in cotton (21) in pea seeds in common bean (20).

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