Durability of Packing Systems for Cost Effective Conservation of Germplasm

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Germplasm is generally conserved in genebanks wherein temperature and relative humidity are adjusted according to term of storage. At Directorate of Wheat Research, Karnal, more than 10,000 wheat accessions are conserved under medium-term storage facility. However, it proved to be a costly affair. As an alternative to cut costs, 7,691 accessions were stored in a room under cold dry conditions at 3,000 m above sea level at Dalang Maidan, Lahaul and Spiti, Himachal Pradesh (HP), India. Three types of packing, namely, cloth bags (A), water proof paper bags (B) and water proof aluminium bags (C) were used for storing. In order to observe the effects and durability of the packing system on viability of seeds stored under these conditions, germinability of a sample comprising of 29 accessions was recorded every year. The difference in germination under two conditions after eight years of storage was non-significant indicating that conservation costs can be reduced by storing material under natural cold dry conditions. Besides, the stability parameters revealed that genotypes C 306, CPAN 3004, GW 173, HD 2009, HD 2285 and HP 1744 were stable in germination and therefore, these genotypes can be stored in any of the packing systems, the most economical and convenient being packing A.

Key Words: Conservation, Cost effective, Genebank, Germplasm, Packing systems, Stability, *Triticum aestivum*

Introduction

The commercialization of agriculture, area development projects and other related activities have led to shrinking of diversity in genetic resources. To save the biodiversity from unforeseen disasters and extinction and to ensure its availability for future, it is conserved using both *in situ* and *ex situ* means. *Ex situ* conservation of plants involves three methods, namely, field genebanks, seed banks and *in vivo* storage. Of these, seed banks are the most efficient and effective method of conservation for orthodox seed. It is an effective and compact method of storage. The seeds are placed in packets and stored in medium-term storage facilities (maintained at 0 to 5°C temperature and 15-20% relative humidity) as active collections. Most of the material is also kept in long-term storage facilities (held at colder temperatures, –20°C to –18 °C). The seed samples are expected to remain viable for 20-30 years in medium-term storage and for up to 100 years in long-term storage depending upon the species, the initial seed quality and specificity of storage environment and general state of infrastructure (such as electricity supplies), etc. (Koo *et al.*, 2002).

However, conserving germplasm in genebanks at low temperature and at low moisture regime is a reasonably costly affair, which includes labour, buildings, equipments, electricity supply, power back up, their maintenance and other operational costs (Pardey *et al.*, 2001). According to a study (Koo *et al.*, 2003), the cost for conserving and distributing the genetic material held in CGIAR genebanks is US $5.7 million per year while to provide these genebank services to all future generations may cost US $149 million. With heavy competition for funding among the world's research institutions, genebanks generally have not fared well (Duvick, 1995; McFerson *et al.*, 1996). Due to lack of funds, equipment malfunction and unreliable power supply, seed genebanks in developing countries often have a hard time fulfilling their mandate to conserve plant genetic resources for future use. Therefore, in order to cut cost, alternative conservation strategy has to be explored.

At Directorate of Wheat Research (DWR), Karnal, wheat germplasm is conserved in medium-term storage facilities where temperature is maintained at 4 ± 1 °C temperature and 23% relative humidity in water proof aluminium packets. Before storage, the seed moisture content is reduced to 9–10%. One set of the germplasm is stored under cold dry natural conditions at DWR Regional Station, Dalang Maidan, Lahaul and Spiti (HP). The station is situated at an altitude of 3,000 m above sea level, where the average maximum temperature remains around 15°C during summers and up to –22 °C during winters. At this station, the germplasm is stored in a room using three types of packings; (i) cloth bags (A), (ii) water proof paper bags (B) and (iii) water proof aluminium bags (C). Presently, about 10,339 accesssions comprising of *T. aestivum, T. dicoccum, T. durum* and *Triticale* are conserved in the module at DWR. Of these, 7,691 accessions are also stored at alternative place,

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Dalang Maidan. In order to determine the economical efficiency of conservation under natural conditions and durability of packing system for storage over eight years period under cold dry conditions, these accessions were evaluated for their germination percentage from 1999 to 2006 and the results are discussed in this paper.

Materials and Methods

Of the 90 accessions stored under natural cold dry conditions in 1998, twenty nine were randomly selected and were evaluated for their germination percentage. With the addition of new accessions every year, the total number of accessions stored at Dalang Maidan has risen to 7,691. However, these twenty nine accessions were evaluated for their germination percentage every year from 1999 to 2006. Four replications of each accession of twenty five seeds from each type of packing were germinated on moist filter paper in petri dishes. The experiment was laid out in completely randomized block design (CRD) with four replications under lab conditions at DWR, Karnal, during November every year. The same accessions from Karnal genebank were evaluated once during 2006 after eight years of conservation. The data on germination percentage was recorded and analyzed following the design of experiment. Packing system in which genotypes' germination remained consistent and did not change over eight years was considered durable. The data were subjected to combined analysis of variance as per Eberhart and Russel (1966) Model considering packing and storage duration as environments, separately.

Results and Discussion

The analysis revealed significant differences over different years and accessions stored under natural conditions at Dalang Maidan, Lahaul and Spiti location. The average germination decreased marginally from 97.7% in 1999 to 88.0% in 2006. However, the germination percentage under medium-term storage facility for 8 years was 100% irrespective of the accessions investigated. The type of packing used for storage also influenced the germination pattern. It was statistically significant for seeds stored in different packings. Germination percentage for cloth bags (packing A) and aluminium packing (C) as well as cloth bags and waterproof paper bags was significantly different for all the years. However, the difference in water proof paper bags and aluminium packing was non-significant initially for 5 years (1999-2003) but was significant thereafter (Table 1). The average germination was recorded the least (90%) in cloth bags followed by that in water proof paper bags (93%) and the maximum in water proof aluminium bags (95%). Over the years, the decline in germination percentage was from 96 to 82% in cloth bags, from 98 to 88% in water proof paper bags and from 99 to 94% in water proof aluminium bags.

The analysis of variance (Table 2a) revealed that genotypes and environments were significant ($p = 0.01$) in all the packing systems indicating differences among the genotypes and environments (storage duration). However, genotype x storage duration interaction was significant ($p = 0.01$) in packing system A and C indicating that germination behaviour of genotypes was inconsistent with duration. Packing system B did not exhibit genotype x storage duration interaction, revealing that genotypic response to germination pattern for eight years period was consistent. The variance due to storage duration (L) and genotype x storage duration (L) when tested against pooled deviation mean square was found significant ($p = 0.01$). Similarly, pooled deviation when tested against pooled error was also found significant ($p = 0.01$) indicating that both linear and non-linear components of variance contributed to genotype and storage duration (environment) interaction.

Considering the packing system as environments, the germination percentage data were separately analysed over different durations of storage. The results of analysis (Table 2b) revealed that genotypes were found to be significant ($p = 0.01$) after one, three, four, six and seven years of storage duration, while environments (packing) showed significance in all storage durations. The genotype x packing interaction component of variance,

Table 1. Mean germination[s] (%) of accessions stored under natural conditions

Year	Duration of storage (years)	Packing A	Packing B	Packing C	CD
1999	1	96.2	97.7	98.8	2.354
2000	2	92.5	95.6	95.3	2.227
2001	3	94.1	96.0	96.7	2.307
2002	4	90.9	94.4	94.8	2.350
2003	5	87.3	92.3	91.4	2.556
2004	6	87.6	91.1	93.8	2.395
2005	7	86.1	89.8	95.0	2.421
2006	8	82.1	87.6	94.3	2.544

Table 2(a). ANOVA for genotype x storage duration interaction for germination of wheat genotypes in three packings

Source	d.f.	Packing A	Packing B	Packing C
Genotype (G)	28	**	**	**
Storage Duration (S)	7	**	**	**
G x S	196	**	ns	**
S + (G* S)	203	ns	ns	ns
S (L)	1	**	**	**
G x S (L)	28	**	**	**
Pooled Deviation	174	**	**	**

** = indicate mean squares significance at 1% level,
NS = mean squares non-significant

Table 2(b). ANOVA for genotype x packing interaction for germination of wheat genotypes in eight storage duration

Source	d.f.	Storage duration (years)@							
		1 (1999)	2 (2000)	3 (2001)	4 (2002)	5 (2003)	6 (2004)	7 (2005)	8 (2006)
Genotype (G)	28	**	*	**	**	*	**	**	*
Packing (P)	2	**	**	**	**	**	**	**	**
G X P	56	**	ns	**	ns	ns	ns	ns	ns
P + (G* P)	58	**	**	**	**	**	**	**	**
P (L)	1	**	ns	ns	*	*	ns	**	ns
G x P (L)	28	ns	ns	ns	ns	ns	ns	ns	ns
Pooled Deviation	29	**	**	**	**	**	**	ns	**

*, ** = indicate mean squares significance at 5% and 1% level,
NS = mean squares non-significant,
@ parenthesis indicate year of germination test.

however, was significant only in two cases *i.e.* after one and three years of storage duration, revealing that genotypes and packing interacted. This indicated that germination of the stored genotypes in different packings remained consistent over different storage durations. The components of environment (packing) (linear) were observed to be significant in all durations of storage. The G x E (linear) interaction, however, was significant for one, four, five and seven year durations. Pooled deviation when tested against pooled error was found to be significant in all storage durations except for seven year duration.

Mean performance of genotypes stored in different packing systems (Table 1) from 1999 to 2006 varied from 82.1% (in A packing 2006) to 98.8% (in C packing 1999). In general, germination percentage was the highest in packing C from 1 to 8 years of storage followed by packing B. But, statistically both were at par. Germination percentage in packing A from 1999 to 2006 was significantly low. Based on overall mean performance of germination and interaction of genotypes with duration of storage, packing system B was found to be superior over A and C packing. Non-significant G x E interaction in case of B packing revealed that germination pattern of all genotypes was consistent and predictable under different durations of storage.

When germination pattern of genotype was analyzed after different durations of storage over three packing systems, it was observed that genotypes interacted with the packing systems only after one and three years of storage duration. In remaining cases, the germination pattern was consistent and predictable. This showed that packing system based on overall mean performance of different genotypes could be adjudged superior or inferior. This further confirmed that packing system B, which did not exhibit genotype x storage duration interaction and showed statistically better germination pattern was superior over the two other systems of packing. The packing system B (water proof paper bag) was also economical and convenient for storage than packing system C (water proof aluminium bags) and hence, was found to be durable.

The germplasm storage in genebanks involves maintenance of low temperature and low relative humidity. At Dalang Maidan, the climatic conditions are cold and dry, which support the survival of germplasm. It is evident from high germination percentage observed during the present study. However, there was reduction in germination over eight years, 15% in case of cloth bags, 10% in water proof paper bags and 5% in water proof aluminium bags. This is attributed to packing type. The non-water resistant packing *i.e.* cloth bags had maximum

reduction whereas the difference in reduction in germination in water proof packings was non-significant.

On examining the individual genotypic response, in different packings and for different storage durations, it was observed that the highest (100%) germination was recorded in ten genotypes in packing A, eight in B and two in C after one year of storage, one genotype each in B and C packing after two years storage. Eight genotypes in packing C and four each in packing A and B recorded 100% germination after three years storage duration. In general, some of the genotypes in packing C recorded up to 98% germination after 4 to 8 years of storage duration.

According to Eberhart and Russel (1966) model, regression coefficient (bi) equal or near to one coupled with zero squared deviation from linear regression (s^2d) indicated average stability. When this is associated with higher genotypic mean value than the population mean, genotypes were categorized having general adaptability and when associated with lower mean value, genotypes were termed poorly adapted to all environments. Based on this, the stability parameters i.e. regression coefficient (bi) and squared deviation from linear regression (s^2d) in packing A and C revealed that genotypes C 306, CPAN 3004, GW 173, HD 2009, HD 2285, HP 1731, HP 1744 and HI 977 were found to be stable in performance. This emphasized that considering the economics of three different packing systems, these genotypes could be stored in cheapest and easiest packing. Genotypic response to different storage durations over three packing systems revealed that GW 173, HD2009 and HD 2285 genotypes recorded consistent germination pattern for all eight durations of storage, while C 306, CPAN 3004 and HP 1744 for seven, GW 190, HD 2329 and HI 977 for six, HP 1731 and HUW 206 for five and HP 1633 and HUW 234 for four durations. These results suggested that the genotypes showing consistent behaviour for 7-8 durations were suitable for medium-term storage in any of the packing systems, possibly the most economical and convenient one *i.e.* packing A, cloth bags than water proof paper bags and water proof aluminium bags. It is further recommended that the longevity of these genotypes could be prolonged under long-term storage and their germination may be tested after 2-3 years period instead of every year.

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