Impact of storage conditions and packaging materials on seed germination and field emergence of okra (*Abelmoschus callei*) at different seasons

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Abstract

The conservation of okra [*Abelmoschus esculentus (L.)*] seeds in genebanks is essential for success of their use in breeding programmes. This study was conducted to investigate the impact of storage conditions and packaging materials on germination and field emergence of okra seeds. One okra accession produced during the late growing season of 2015 was used for the study. The experiments were set up using 3x3 factorial in completely randomized design (CRD) and randomized complete block design (RCBD) for germination and field emergence experiments respectively with three replications. One hundred seeds per replicate were subjected to standard germination test and immediately followed by field evaluations during four growing seasons. The results of combined ANOVA revealed that storage conditions, packaging materials, year of storage and their interactive effects were highly significant on seed germination and field emergence of okra. Okra seeds stored in plastic container had highest germination value (79.67%) and field emergence value (78.67%) under short term storage conditions while seeds stored in aluminum foil had highest seed germination value (74.33%) and field emergence value (75.33%) under medium term storage conditions. The materials stored inside deep freezer using aluminum cans had highest values for seed germination (67.00%) and field emergence (77.67%) suggesting that plastic containers, aluminum foils and aluminum cans would be suitable for storing okra seeds under short term, medium term and freezer storage conditions respectively. However, comparatively lower germination counts for okra seeds stored in aluminium can under freezer conditions suggests slow release of dormancy due to low temperature.

Keywords: short term, medium term, freezer, conservation

INTRODUCTION

Okra *Abelmoschus callei* (L) Moench is an economically important crop belonging to the family Malvaceae. It is a nutritious vegetable grown in tropical and sub-tropical parts of the world containing 86.1% water, 2.2% protein, 0.2% fat, 9.7% carbohydrate, 1% fiber and 0.8% ash (BARI, 2010). However, in spite of its benefits okra has been considered as a minor crop and no attention has been paid to its improvement in the international research programme in the past (Sanjeet et al*.,* 2010). In order to promote the use of this indigenous vegetable as an important crop to mitigate food insecurity and alleviate malnutrition, improving its genetic potential is very important.

Conservation of okra germplasm is mainly by seed storage in genebanks hence efficient management of seed collections, especially under tropical conditions, depends on, among other factors, conditions of the storage environments and packaging materials used in seed storage. However, most genebanks are vulnerable to financial constraints leading to downsizing, as well as chronic losses in diversity due to storage methods, catastrophic losses from equipment failures, among other things (McGuire and Qualset, 1990). It is therefore essential to conserve of okra seeds in appropriate storage conditions for success of their use in breeding programmes.

In Nigeria, The National Centre for Genetic Resource and Biotechnology (NACGRAB), Ibadan, has the institutional mandate for conservation and maintenance of valuable genetic resources for immediate utilization and posterity. The centre's genebanks currently hold over 400 accessions of okra seeds stored in both short and medium term storage chambers. However, in order to facilitate the conservation of this germplasm, it is essential to investigate laboratory germination and field emergence performance with a view to ascertain most suitable packaging materials for respective storage environments. The objective of this study therefore was to investigate the impact of three packaging materials (aluminum foil bag, aluminum can and plastic container) under three storage environments (short term, medium term and deep freezer) on germination and field emergence of okra seeds.

MATERIALS AND METHODS

Genetic material and experiment location

Okra accession (NGB 00372) with 12% moisture content produced during the late growing season of 2015 was used for the study. The laboratory experiment was carried out at the seed testing laboratory of NACGRAB prior field emergence. The field emergence experiment was carried out at experimental field of NACGRAB, Moor Plantation, Ibadan located on latitude 007^0 48' 11.3'' N, longitude 003^0 50' 52.0''E and altitude of 183m above sea level from 2016 to 2019 growing seasons.

Seed storage and measurement of storage conditions

Five hundred grams of the processed seed of okra accession (NGB 00372) was drawn and divided into three lots. Each lot was further subdivided into three samples and packaged using aluminum foil bags, aluminum cans and plastic containers. Thereafter the samples from each packaging material were stored in short term, medium term and freezer environments. Power supply to the storage environments was ensured for minimum of twelve hours daily. The temperature and relative humidity of the storage environments were monitored in the morning (10:00 am) and afternoon (3:00 pm) daily throughout the entire period of study using digital thermo-hygrometer.

Experimental design

The experiments were set up using 3x3 factorial in completely randomized design (CRD) and randomized complete block design (RCBD) for germination and field emergence experiments respectively with three replications. The factors were: storage environments (SE 1: Short Term, SE 2: Medium Term, SE 3: Freezer) and packaging materials (PM 1: Aluminum foil bag, PM 2: Aluminum can, PM 3: Plastic container).

Conduct of the experiment

1. **Standard germination tests.**

Standard germination test was carried out on okra seed samples before storage in December 2015 to ascertain the initial quality of seeds. However, effect of the factors on germination was assessed in August 2016, June 2017, September 2018 and July 2019 prior field emergence trials. The germination test was performed using the top paper method. Three replications of one hundred seeds were used per plot which were randomly distributed on the wet filter paper in petri dishes 9cm in diameter and placed inside an incubator set to a constant temperature of 25°C throughout the testing period. Germination percentages were calculated by expressing the number of seedlings in a replicate that germinated 9 days after planting as a percentage of the number of seeds planted according to ISTA rules (ISTA, 1999).

2. Field Emergence trials.

Field emergence trials were carried out immediately after standard germination tests for four seasons: late season of 2016, early season of 2017, late season of 2018 and early season of 2019. One hundred seeds were sown in three replicates on a well prepared seed bed with adequate soil moisture. Field emergence was determined on the 9th day after sowing calculated

by expressing the number of seedlings in a replicate that emerged at 9 days after planting as a percentage of the number of seeds planted.

3. Statistical analysis

Data obtained from standard germination tests and field emergence trials were subjected to analysis of variance (ANOVA), using Statistical Analysis System software (SAS, 1990). Since data on percentages do not conform to normal distribution, the germination data were log-transformed before subjecting them to ANOVA. However, since ANOVA did not detect any significant difference between transformed and untransformed values, undet transformed values were hereby presented. Pertinent means were thereafter separated by use of the least significant difference (LSD).

RESULTS AND DISCUSSION

Conditions of the storage environments

The mean temperature and relative humidity in the three storage environments during the study (2016-2019) were presented in Table 1. The mean temperature under short term environment ranged from 11.9-23.8^{o}C while the relative humidity ranged from 22.0 to 42.3%. The conditions in the medium term storage environment revealed that mean temperature ranged from -1.6 to 2.9^{o}C while the relative humidity ranged from 43.0 to 68.1%. The mean temperature of the freezer environment ranged from -4.2 to 4.0^{o}C while the relative humidity ranged from 44.0 to 67.0%. The variation in the values of temperature and relative humidity of the storage environments suggests the impact of minimum of twelve hours in power supply to the storage environments resulting into fluctuation of temperature and relative humidity of the storage environments (Table 1).

Seed germination and field emergence performance of okra seeds stored under varied conditions.

The results of combined analysis of variance revealed that effects of storage environments, packaging materials and storage year were significant at $P<0.01$ on seed germination and field emergence of Okra (Table 2). There were also significant interactive effects among the treatments on seed germination and field emergence of okra except for storage year by packaging materials interactive effect on seed germination (Table 2). These results implied that germination performance of seed lots depended on the combined effect of storage environments, packaging materials and the time of storage. This is agreement with the report of Omar et al., (2012) that cultivars, storage environments, packaging materials, storage periods as well as their interactions significantly affected seed germination and vigour of wheat.

Table 1. Atmospheric conditions of the storage environments during the study.

Storage		Short term		Medium term		Freezer	
Year	EC	M1	M2	M1	M2	M1	M2
2016	TM (^{o}C)	17.0	20.2	-0.4	1.6	-4.2	4.0
	RH (%)	30.1	31.5	43.0	66.0	44.0	67.0
2017	TM (^{o}C)	18	23.8	-0.3	2.9	0.40	3.5
	RH (%)	33.5	41.1	54.0	59.0	45.0	65.0
2018	TM (^{o}C)	11.9	19.5	-1.6	1.2	-1.50	2.5
	RH (%)	29.0	40.1	53.0	68.1	46.0	66.0
2019	TM (^{o}C)	18.1	23.1	-1.3	2.2	-4.20	4.0
	RH (%)	22.0	42.3	54.0	67.0	44.0	66.0

EC= Environmetal Conditions; TM = Temperature; RH= Relative Humidity; M1= Minimum; M2= Maximum

Table 2: Means squares from the analysis of variance for germination and field emergence of okra seeds as affected by storage year (SY), storage environment (SE), packaging materials (PM) and their interaction at NAGRAB, Ibadan.

Sources of Variation	DF	Germination (%)	Field (%)
Rep	2	36.45ns	6.33ns
Storage Year (SY)	3	1079.74**	504.59**
Packaging Material (PM)	2	2267.95**	2347.44**
Storage Environment (SE)	2	1704.73**	1369.33**
PM*SE	4	928.34**	1839.78**
SY*PM	6	1813ns	121.07**
SY*SE	6	97.27*	209.93**
SY*SE*PM	12	85.44**	63.41**
Error	70	23.84	17.72
Total	107	171.57	189.78
R^2		0.91	0.94
CV		6.12	5.78
Mean		79.82	72.89

*, **, Significant at probability level of 0.05 and 0.01, respectively; ns = Not Significant
PM = Packaging material, SE = Storage environment, SY = Storage year

Effect of storage environments, packaging materials and storage time on germination and field emergence of okra seed

The germination percentages for okra seeds stored in short term environment had highest germination percentage (86.53%) followed by okra seeds stored in medium term (80.17%). These two values were significantly higher than value of okra seeds stored in the freezer environment. These results suggests that relatively lower temperature increases seed hardness in okra seed. It has been reported that okra plants exhibit seed hardness that interferes with seed germination (Mohammadi et al., 2011) and this phenomenon can be increased by environmental (dry) conditions during seed maturation, and seed storage (Baskin & Baskin, 1998). This these results coroborated with findings of Hnin et al., (2019) who reported significantly higher germination percentage (40.41%) under ambient conditions compared with 25.61% under cold storage of sesame seeds attributed to slow release from dormancy due to low temperature under cold storage. Luis Felipe et al., (2010) reported that the occurrence of seed hardness and the low percentage of seed germination are major challenges in growing okra. The field emergence percentages for okra seeds stored in short term (76.11%) and medium term (76.78%) environments were not significantly different but were significantly higher than value of okra seeds stored in the freezer environment (Table 3). Moreover, germination percentages for okra seeds stored in aluminium foil bags (85.31%) and aluminium cans (83.44%) were significantly higher than okra seeds stored in plastic containers (70.72%) (Table 3). Similar results were observed on the field. The field emergence percentages for okra seeds stored in aluminium foil bags (78.33%) and aluminium cans (76.72%) were significantly higher than okra seeds stored in plastic containers (63.61%) implicating that aluminium foil bags and aluminium cans could be recommended as suitable packaging materials for storing okra seeds under cold environments. The germination values of okra seeds evaluated in 2016 (75.96%) and that of 2017 (74.00%) were significantly lower than germination values of 2018 (88.15%) and 2019 (81.19%) suggesting that okra seeds in storage takes some time to release from dormancy. The field emergence values of okra seeds evaluated were 77.70% in 2016, 70.15% in 2017, 75.26% in 2018 and 68.44% in 2019 suggesting that okra seeds field performance also depends on the environmental conditions on the field (Table 3).

Table 3 Germination and field emergence of okra seeds as affected by storage environments, packaging materials and storage year.

Storage environment	Germination (%)	Field emergence (%)
Short term	86.53a	76.11a

Medium term	80.17b	76.78a
Freezer	72.78c	65.78b
LSD	2.3	1.98
Packaging material		
Aluminium foil bag	85.31a	78.33a
Aluminium can	83.44a	76.72a
Plastic container	70.72b	63.61b
LSD	2.3	1.98
Storage Year		
2016	75.96c	77.70a
2017	74.00c	70.15c
2018	88.15a	75.26b
2019	81.19a	68.44c
LSD	2.65	2.29

Means with the same letter are not significantly different at P= 0.05

Interactive effect of storage environments and packaging materials on germination and field emergence of okra seed

The effect of storage environments on seed germination and field emergence of okra highly depended on the packaging materials. Okra seeds stored in plastic container had highest germination (79.67%) and field emergence (78.67%) values under short term storage conditions suggesting that plastic container was most suitable for storing okra seeds in this environment. Okra seeds stored in aluminum foil had highest seed germination (74.33%) and field emergence (75.33%) values under medium term storage conditions suggesting aluminium foil as most suitable packaging materials for storing okra seeds in this environment. Moreover, okra seeds stored in aluminium can inside freezer had germination counts of 67.00% and field emergence values of 77.67% although the values were not significantly different from that of aluminium foil bag in the same environment which had germination values of 64.33% and field emergence values of 69.83%. However, in spite of relatively lower temperature values under medium and freezer environments, the best packaging materials under these environments had relatively lower germination values compared with the best packaging materials under short term storage environment suggesting that increasing storage temperature can alleviate seed dormancy in okra (Figure 1). Bazin et al. (2011) had earlier reported similar results that increasing storage temperatures alleviated seed dormancy in sun flower (*Helianthus annuus*) at seed moisture content between 2.5% and 12%. Hence, in order to avoid reduce germination due to slow release from dormancy which may occur as a result of low temperature in storage, breaking of dormancy in okra should be taking into consideration when evaluating for germination or field emergence especially when seed lots were stored under cold environments.

Figure 1. Germination and field emergence of okra seeds as influenced by the interaction of packaging materials (PM) and storage environments (SE)
PM 1=Aluminium foil bag; PM 2 = Aluminium can; PM 3 = Plastic container
SE 1=Short term; SE 2=Medium term; SE 3=Freezer
SGT= Standard germination; FE= Field emergence

CONCLUSION

The study concludes that seed germination and field emergence of was highly influenced by the interactive effect of storage environments and packaging materials. Plastic container and aluminum foil bag and aluminum can would be suitable for storing okra seeds under short term, medium term and freezer storage conditions respectively. However, relatively lower germination counts especially for seeds stored under freezer conditions irrespective of packaging materials compared to other storage environments suggesting slow release of dormancy in okra seeds due to low temperature hence in order to avoid reduce germination which may occur as a result of low temperature in storage, breaking of dormancy in okra should be taking into consideration when evaluating for germination or field emergence especially when seed lots were stored under cold environments.

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