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Effect of Storage Period on Seed Germination of Different Maize Parental Lines

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Abstract
Duration of seed storage can be one of the factors that cause seed deterioration due to natural seed aging event takes placed. The study was carried out with the objective to determine the effect of storage period on seed germination, emergence and seedling traits of different maize parental lines. Four parental lines of seeds were produced in three different cropping seasons and stored for their respective years. Each parental line seed was tested and necessary data were collected on germination characteristic, emergence and seedling traits. Result of analysis variance showed significant ($p<0.001$) difference within and among parental lines that produced and stored in different years for germination percentage, mean time to germination, germination index, emergence percentage, fresh weight of shoot and root. The highest germination (99.00 %) as well maximum germination curve was recorded for the CML-395 that produced and stored in the early of 2016B whereas the CML-202 produced and stored during 2012A showed the lowest germination (21.00 %), indicating that germination was decreased as the seed stored longer. The lower mean time germination (4.21 days) means the earlier to reach 50 % germination as well the highest germination index (6.01) was recorded for A-7033 which produced and stored in 2015A, indicating that the more energy to germinate earlier and likely had more vigour. Phenotypic correlation observed among germination parameters and seedling traits. Germination revealed a highly significant strong positive correlation with germination index ($r=0.99$) and emergence ($r=0.87$). Germination index showed significant positive correlation with emergence ($r =0.89$) and fresh weight of shoot ($r =0.39$), means that the higher germination index it might be had high rate and vigour. But, MTG showed significant negative associations with germination ($r= -0.72$), emergence ($r= -0.74$) and germination index ($r= -0.78$), indicating that the lower MTG lead to the fastest germination and emergence. The present study revealed that paretnal lines seeds which produced and stored less than in one year were showed better seeds germianted as well as emerged and followed by second years but it varied among maize parental lines and seed storage period. Our result showed that as seed stored longer duration resulted in delayed onset of germination, decreased the germination rate as well as germination index, slow seedling emergence and low weight of seedling traits. Thus directly influence the agronomic seed quality traits, since farmers need high quality of seed that enable to germinate high percentage and uniformity of seedlings under required conditions.
Keywords: maize parental line, seed quality, germination, duration of seed storage

Introduction

Maize (*Zea mays L.*) is one of the most important cereal crops in Ethiopia, ranking second in area coverage and first in total production. In 2013 about 6,674,048 tonnes of maize grain was produced in Ethiopia (http://faostat.fao.org). Such production depend various factors. Among these; seed quality and post- harvest management are the most important. Seed quality is in terms of genetically and physically pure whereas post-harvest managements are used to increase seed quality through seed processing while required storage condition maintain its quality. These factors influence on the success of seed germination, normal seedling and final seed production.

Seed quality is judged by different end users such as farmers and industries. For instance, farmers expect to obtain high quality seeds that are able to germinate and produce normal seedlings under field conditions (Khan *et al*., 2012). Germination is defined as the process in which seeds begin to uptake water, followed by elongation of the embryo and penetration of the radical through the endosperm and seed coat (Bewley and Black, 1994). Seed quality of maize has the capacity to high germinate and might be contributed to high initial downward growth as well fast growing subsequently good seedling establishment. To attend such high germination rate, seed should be stored at required storage period to maintain its quality.

Seed can be maintained through the management of storage conditions and optimum storage period in order to optimize physiological aging and to control insect-pest. The term ‘storage period’ suggests that natural seed aging events take place over prolonged period. If storage period is prolonged, it may accelerate the natural seed aging process and chemical changes thus causes seed deterioration. Seed deterioration is expressed the loss viability, quality and vigour due to natural aging or adverse of environmental factors such as high temperature, high humidity moisture and others (Sisman and Delibas, 2004; Azadi and Younesi, 2013). Storing seed beyond of optimum storage period might be resulted in reduces germination potential, seedling establishment and final seed production (Sisman, 2005).

To meet increasing demand of food in Ethiopia, national maize research centre has released different maize technologies (Hybrids and OPV's). Their parental lines are under multiplication by research centre and seed companies. Before distribution, seed can be stored temporally and even some times seed can be not sold from seed lots for one or two years due to leftover and improper annual plan. The remained seed lot is one of the important an economic aspect since storage period could be impact on market values while natural seed aging event take place which resulted in seed deteriorate. Thus, decline viability, abnormal and poor seedling establishment would happen as an impact. Studies indicate that seed aging reduce germination and emergence (Rice and Dyre, 2001). Seed germination, emergence rate and seedling establishment are decreased with increased in seed storage was accompanied with the increase mean germination time (Verma and Tomer, 2003; Basra *et al*., 2003). After one year storage, seed germination declined significantly (Mrda *et al*., 2010). However, it varies among genotypes/maize parental lines. So far little information is available and / or limited studies are conducted on the impact of storage period on the seed germination and seedling characteristic of released hybrids of parental lines. Therefore, the present study was conducted to determine the effect of storage period on germination, emergence and seedling traits of different maize parental lines.

Materials and Methods

Source of maize parental lines

The study was carried on seeds of parental lines of released hybrids. These includes CML-395, CML-202, 142-1-e and A-7033 were obtained from remnant seed lot of different production years at Bako National maize research centre (Table 1). All parental lines of seeds were stored under the same environment condition such as average temperature (22 ^{0}C) and relative humidity (60 %).

Table 1. Name of parental line, it's production year and storage period

Seed of parental lines	Production year	Respective storage period
CML-395	2012A,2014A,2016B	3 y and 6 m, 1 y and 3 m, 1 m
CML-202	2012A, 2014B, 2016B	3 y and 6 m, 2 y, 1 m
142-1-e	2012A, 2014A, 2015A	3 y and 6 m, 1 y and 3 m, 3 m
A-7033	2011A, 2014A, 2015A	4 y and 1 m, 1 y and 3 m, 3 m

Key: A= seed produced during main season, B= seed produced during off-season or by using irrigation, y= storage period in years and m= storage period in months.

Experimental setup

The experiment was conducted at Bako National Maize Research centre during May 2016. Bako is situated an altitude of 1650 m above sea level which represent mid altitude sub-humid agro-ecology zone of Ethiopia. Four parental lines were produced for three years and each of them stored for their respective year. Seed sample of each parental line was taken from remained seed lot of different production year and their respective storage period. The experimental design was complete randomized design (CRD) and replicated four times. For germination test, 120 mm diameter of petri -dish was arranged randomly and one layer of white filter paper was laid on each petri-dish. 25 seeds were placed on each Peter dish in order to germinate and moisture level checked daily and top up with required amount of water as if necessary. For emergence test, pot was filled with 1:1 ratio of sand and clay soils, each treatment was replicated three times and five seeds planted.

Data collection

Germination assay: Seed germination was counted and scored when radical protrusion was observed. Germination was counted daily for eight consecutive days during the germination.

Germination parameters such as Germination index (GI) and Mean germination time (MGT) were calculated as described in the association of official seed analysis (AOSA,1990) using formula:-

Germination Index (GI) = $\sum(\frac{GT}{Tt})$, where Gt is the number of seeds germinated on day t and Tt is the number of days.

Mean germination time (MGT) = $\sum Ti\ Ni/\sum Ni$, where Ni is the number of newly germinated seeds at time Ti.

Emergence percentage and seedling traits: seed emergence was counted daily and converted into percentage. Based on visual observation, three healthy seedlings were taken and weighted for fresh weight of shoot as well as root.

Statistical analysis

Germination percentages, germination index, mean germination time, fresh weight of shoot and root were analysed by using PROC GLM of SAS software (SAS, 2004). Mean separation was performed to compare treatment means using LSD-test at 5% level of significance. Germination curve was calculated based on daily germinated seed. The correlation among germination parameters and seedling traits were analyzed by using

Pearson correlation coefficient analysis following PROC CORR procedure of the SAS software (SAS, 2004).

Result

Analysis of variance showed highly significant difference (*p*<0.001) for germination and emergence percentage within parental lines and /or among parental lines seeds that produced and stored in different years (Figure 1 and 2). As seed stored longer (from recent; 2016B to seed aged; 2011A), the germination and emergence percentage decreased and significantly increased. The highest germination (99.00 %) was recorded for the seed of CML-395 that produced and stored during early of 2016B, this implies short term seed storage can maintain the biological value of seed. Whereas seed of CML-202 produced and stored during 2012A showed the lowest germination (21.00 %), it perhaps due to prolonged storage period it declined seed viability. Similarly, significant difference (*p*<0.001) observed within parental lines in different storage period for mean time germination and germination index (data not shown). The lower mean time germination (4.21 days) was recorded for the parental line seed of A-7033 that produced and stored in 2015A, indicating that the earlier to reach 50 % germination whereas higher mean time germination (5.86 days) recorded for 142-1-e which produced and stored in 2012A. Also, the highest germination index (6.01) was recorded for the seed of A-7033 that produced and stored in 2015A, it might be the more energy to germinate earlier and had more vigour. Whereas the lower germination index (1.01) was observed for the seed of CML-202 which stored and produced in 2012A, indicating that longer storage period had an effect on germination speed and germination index.

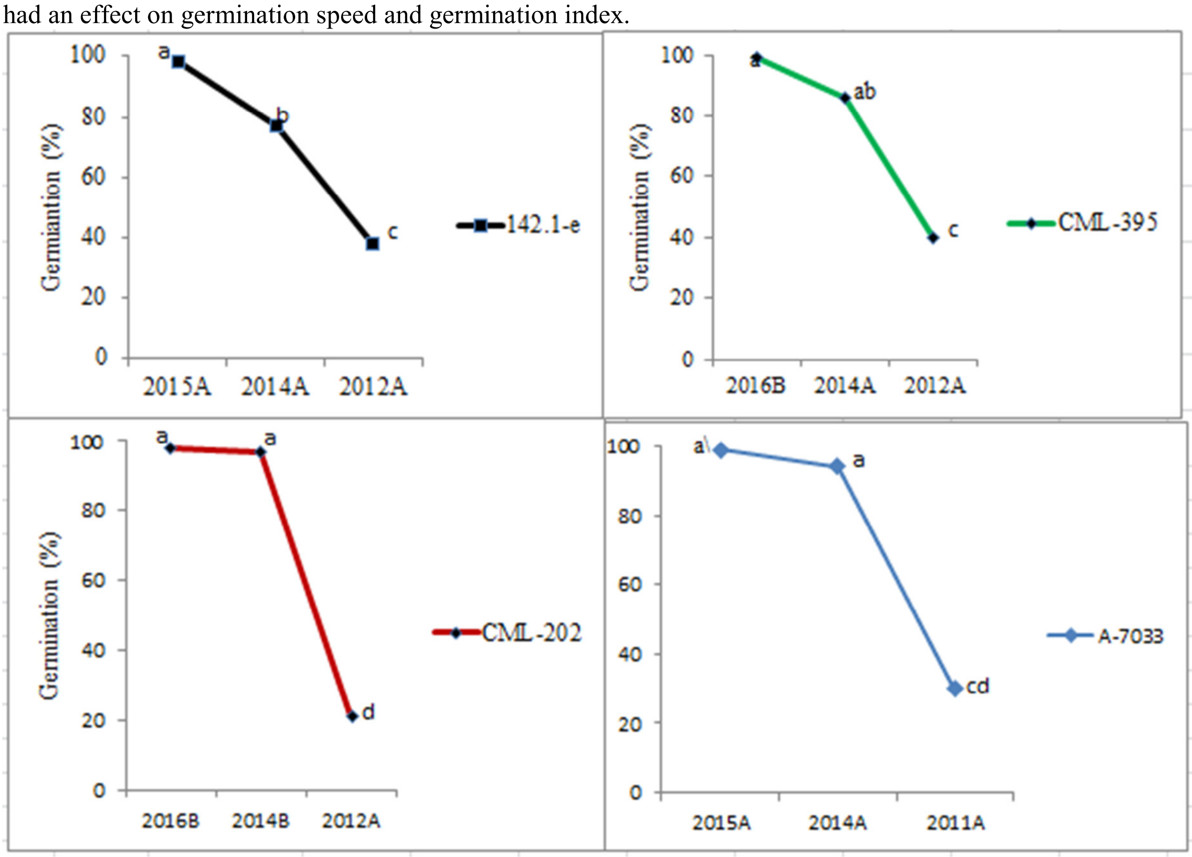

Figure 1. Effect of storage period on seed germination for four maize parental lines that produced and stored in different years. Points that followed by different small letter differ significantly (P < 0.05) at (LSD 0.05=13.91).

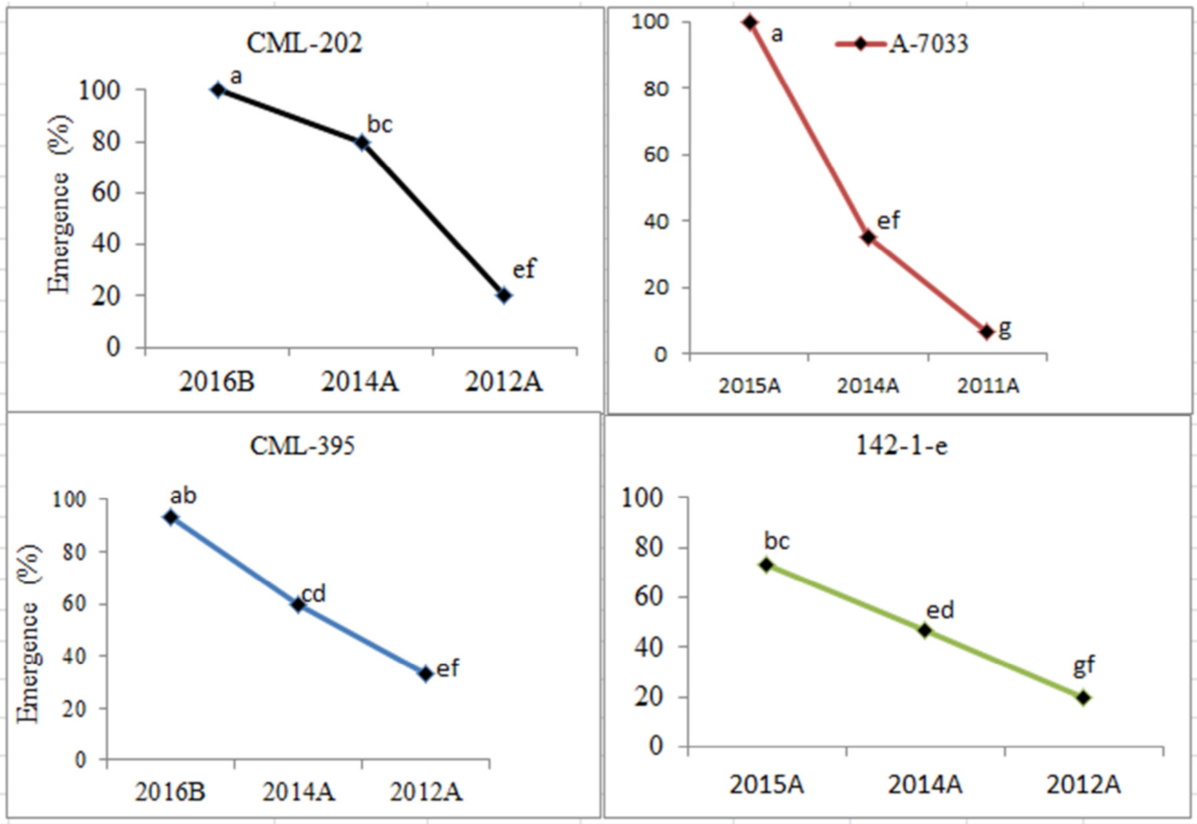

Figure 2. Effect of storage period on seed emergence for four maize parental lines which produced and stored in different years. Points that followed by different small letter differ significantly (P < 0.05) at (LSD 0.05=20.81).

Germination curve

Germination progress curve verses time also estimate time taken for cumulative daily germination to reach 50 % and maximum germination percentage. There were clear differences between duration of storage period with respect to germination curve. Within parental line of different production year and storage period, seeds of recent produced and stored quickly achieved maximum germination percentage than seeds stored more than three years. For instance, the germination curve graph of seeds CML-395 which produced and stored in 2016B shown very rapid increase until achieved maximum germination (99%) whereas CML-395 that produced during 2021A was very slow and achieved below 50% germination (Figure 3). Among the three germination curve, low seed germination progress curve observed for parental line of 142-1-e across years (Figure 4). Overall, the highest (99 %) and lowest (21%) germination were recorded for CML-395 and CML-202 which was produced and stored in 2016B and 2012A respectively (Figure 3 vs 5). This showed that seeds stored longer period had impact on cumulative maximum germination percentage.

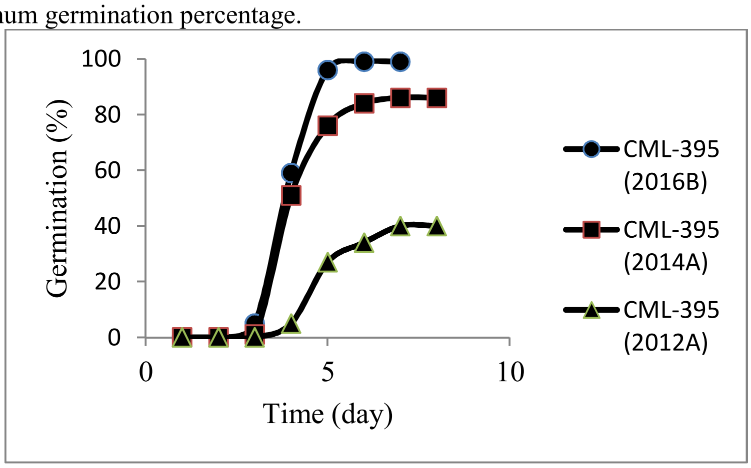

Figure 3. Cumulative germination percentage curve versus time for different storage period of maize parental line CML-395

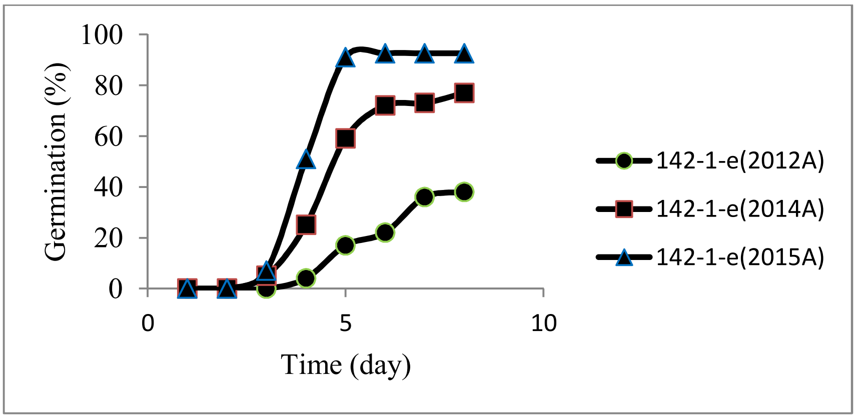

Figure 4. Cumulative germination percentage curve versus time for different storage period of maize parental line 142-1-e.

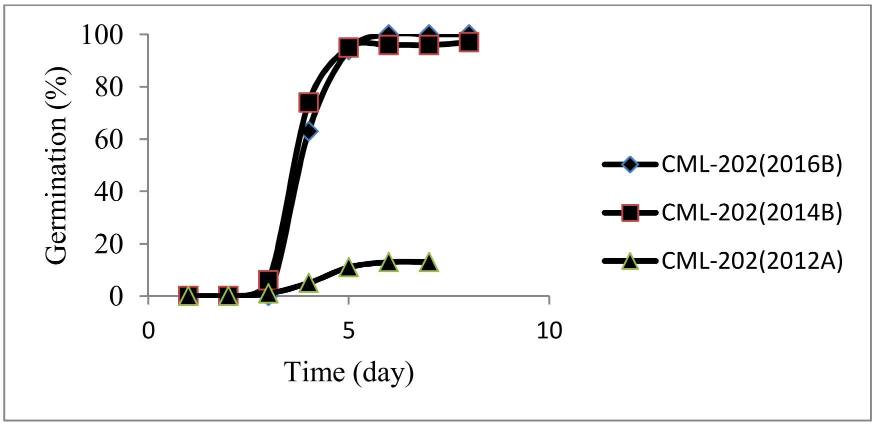

Figure 5. Cumulative germination percentage curve versus time for different storage period of maize parental line CML-202

Correlation analysis

The result of Pearson correlation analysis showed significant and positive as well negative trend association (p<0.001, p<0.05) among germination parameters and seedling traits. Germination revealed a highly significant strong positive correlation with germination index (r = 0.99) and emergence (r = 0.87). Also, germination percentage showed significant positive weak association with TKW (r = 0.39) and fresh weight of shoot (r = 0.35), indicating the higher TKW might be contributed for maximum germination and seed better germinated had likely more fresh weight. But, MTG showed significant negative associations with germination (r = -0.72), emergence (r = -0.74) and germination index (r = -0.78), this implies the higher MTG resulted in the lower germination percentage, emergence and germination index.

Table 2. Pearson correlation among germination parameters and seedling traits for four parental lines that produced and stored in different year.

	Germination	MTG	GI	EM	FWRT	FWST	TKW
Germination	1						
MTG	-0.72**	1					
GI	0.99**	-0.78**	1				
EM	0.87**	-0.74**	0.89**	1			
FWRT	0.24	-0.31	0.29	0.32	1		
FWST	0.35*	-0.35*	0.39*	0.43*	0.35	1	
TKW	0.39*	-0.26	0.40**	0.37*	0.42*	0.39	1

Germination (%), MTG= Mean time germination, GI= Germination index, EM= Emergence (%), FWRT= Fresh Wight of root, FWST= Fresh weight of shoot, TKW= Thousand kernel Weight.

Discussion

Signifcant varations were observed among and within parental lines for germination and emergence percentage as well seedling traits in different storage period. Germination, emergence and fresh weight (shoot and root) were decreased as the seed stored longer duration. This finding is in agreement with (Verma and Tomer, 2003; Basra *et al*., 2003) demonstrated that seed germination, emergence rate and seedling establishment are decreased with increased in seed storage period. Also, similar finding reported that seed germination declined significantly after one year storage Mrda *et al*., 2010). Some parental line of seed declined viability with a faster rate than other parental line which produced and stored under similar condition. For instance, Seed of CML-202 showed faster rate decreased germination than seed of CML-395 from recent to longer storage duration. Further explanation, the fastest and slowest seed germination were differed by maize parental lines/genotypes, production year and its storage period. Maize parental lines of seeds with storage period less than one year were shown better seed germianion and followed by parental line seeds with storage period of two years whereas parental lines seeds with storage period over three years were shown very low germination and emergence percentages. Also, similar trends were observed for others germination parameters and seedling traits in the present study. This finding is in agreement with Joa-Abba and Lovato, 1999) who reported that reduced germination and subsequently seedling growth due to long storage period. The current study revealed that as seed stored longer duration resulted in delayed onset germination, decreases the germination rate as well germination index and decreases maximum germination, decreased emergence and fresh weight of shoot as well root. Thus directly influence the agronomic seed quality traits, since farmers need high quality of seed that enable to germinate high percentage and uniformity of seedlings under required field conditions.

Understanding the physiological and underlying molecular mechanism how the seeds stored for longer duration that associated with seed aging can delay and fail to seed germination is quite important. As seed stored longer, natural seed aging events would be happened and physiological changes can be occurred (Sisman and Delibas, 2004; Morda Shaba, 2013). The phenomenon was observed in the present study including decreased germination parameters, low weight of shoot and root. We didn't test in this experiment how the biochemical change during seed storage period increased. But research report showed that seed stored for longer period would impact on chromosomal aberration, DNA can be damaged and or protein degradation thus causes the seed deterioration might be resulted in loss seed germination potential and seedling establishment (Gidrol *et al*., 1998; Whittle, 2006). These might be the reason why seeds of a few fail to germinate and emerge under prolonged seed store in the present study.

Concolusion

Maize parental lines of seeds which prodcued and stored less than one year were shown better seed germiantion as well as emergence and followed by parental lines seeds with two years of storage period, but variations were observed among genotypes/ maize parental lines studied. As seed storage period increased, the tested maize parental lines seeds were showed low germiantion, decreased gremination rate and index, emergence and low weight of seedling triats. Thus directly influence the agronomic seed quality traits, since farmers need high quality of seed that enable to germinate high percentage and uniformity of seedlings under required field conditions.

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Conflict of Interests: The authors declare that they have no conflict of interest

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