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The effect of traditional storage methods on germination and vigour of maize (Zea mays L.) from northern KwaZulu-Natal and southern Mozambique

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Abstract

In sub-Saharan Africa, maize (Zea mays L.) is one of the most nutritional crops and proper storage of seeds continues to be a challenge for subsistence farmers. Storage fungi, which reduce seed quality, become active in seeds when moisture levels are 14% or higher and this is influenced by the way seeds are stored. The aim of the present study was to test germination and vigour of maize seeds that were subjected to traditional storage during 2005 and to test germination of the maize seeds after storage for one year under conditions of fluctuating temperature. A preliminary survey was conducted and maize samples (white and yellow) were collected from small-scale subsistence farmers in northern KwaZulu-Natal (South Africa) and Mozambique. Seeds that were left in the field to dry and seeds that were commercially treated with Celest® XL served as controls. Germination was measured according to the International Seed Testing Association (ISTA) rules. The maize that was left in the field (NHS) to dry gave 100% germination in 2005. The treated control had a germination of 94.0%. Seeds that were imbibed for 40 h had the highest percentage weight increase following rapid imbibition but four of the six samples maintained germination above 70% following slow imbibition. The conductivity of the solute was read following imbibition. Field stored maize had the lowest solute leakage (1181 µS) and this correlated with the high percentage seeds with living tissue as indicated by the tetrazolium staining following rapid (94.4%) and slow (95.8%) imbibition. The number of fungi isolated from the samples reflected the initial condition of the samples with the fungicide treated control having the lowest percentage infection (11%), NHS had 33% and yellow maize that was stored on the cob and had with insect damage (SIH) had the highest, namely 71%. After the first set of experiments, samples were stored at 26-28 °C to simulate the fluctuating original storage conditions. A year later the samples were subjected to the standard germination test. The decline in seed viability during the storage period was exhibited by results of the standard germination test. Maize that was left in the field had a 74.7% decrease in germination while the sample stored in potato bag (PHEL) and the treated control maintained germination above 80%. Two of the six samples failed to germinate. This study also showed that fungicide seed treatment is a viable option to maintain viability of the seeds, especially when the maize is to be stored until the next season. © 2007 SAAB. Published by Elsevier B.V. All rights reserved.

Keywords: Germination; Maize; Seed treatment; Traditional storage

1. Introduction

Maize (Zea mays L.) is important as a source of energy and protein in the human diet throughout the world (Rehman, 2006). Proper crop storage plays an integral part in ensuring domestic food supply (Thamaga-Chitja et al., 2004) and that seed quality and vigour is maintained (Joao Abba and Lovato, 1999). Fluctuations in temperature, humidity and prolonged storage result in considerable nutrient losses (Shah et al., 2002). Despite significant

advances in food storage methods, many African and South African communities still rely on traditional storage methods for seed to be used as food and fodder (Olakojo and Akinlosotu, 2004; Thamaga-Chitja et al., 2004). Storage facilities not only offer the opportunity to provide a supply of food between staple crop harvests but farmers are able to improve farm incomes by storing crops and selling at premium prices when demand outstrips supply later in the post-harvest period (Florkowski and Xi-Ling, 1990). The most important factors that influence storage are temperature, moisture, carbon dioxide (CO₂), oxygen (O₂), grain characteristics, micro-organisms, insects, mites, rodents, birds, geographical location and storage facility structure (Jayas and White, 2003).

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Insect pests are one of the major organisms that are responsible for the decline in quantity, quality and germination potential of maize seeds in storage (Jayas and White, 2003). A common strategy in many African countries is to sell maize grains immediately after harvest, to avoid losses to insect pests (Olakojo and Akinlosotu, 2004). Farmers in sub-Saharan Africa generally store their un-husked maize on wooden posts (Thamaga-Chitja et al., 2004). In most situations, maize is traditionally left to dry in the fields prior to harvesting. Other storage structures include a traditional silo that is made of mud and twigs. This structure is relatively inexpensive but it is not airtight and often exposes the stored maize to harsh environmental conditions such as sun and rain (Olakojo and Akinlosotu, 2004). Other storage facilities include the use of iron tanks, re-used maize-meal sacks to store maize on the cob and in addition, polyethylene, polypropylene and cotton sacks are frequently used (Thamaga-Chitja et al., 2004).

In 1979, the Association of Official Seed Analyst's vigour committee defined seed vigour as "those seed properties, which determine the potential for rapid, uniform emergence and development of normal seedlings under a wide range of field conditions" (Copeland and McDonald, 2001). Seed vigour is defined by the International Seed Testing Association (ISTA) as "the sum total of those properties of the seed that determine the level of activity and performance of the seed during germination and seedling emergence" (ISTA, 2006). Vigour testing involves direct tests (e.g. cold test) where an environmental stress is reproduced in the laboratory and the percentage and or rate of seedling emergence are recorded. In indirect tests (e.g. conductivity) other characteristics of the seed are measured, that have proved to be associated with some aspect of seedling performance (ISTA, 2006).

When dry seeds are plunged into water, they imbibe water rapidly in the first few minutes, followed by a slower phase of imbibition until they become fully hydrated (Copeland and McDonald, 2001). It is concluded that ultra-dry seed storage is beneficial for maintaining seed vigour and that starchy mobilization proceeds regularly during germination (Wang et al., 2005). During the early stages of imbibition the seeds leak solutes such as organic and inorganic ions, sugars, amino acids and even proteins into the surrounding medium. Depending on the condition of the seed this loss means the loss of intracellular constituents and often results in extensive embryo damage and even its death (Duke and Kakefuda, 1981; Copeland and McDonald, 2001). Conductivity of the soak water of the sample gives an estimate of seed vigour. Seed lots that have high electrolyte leakage that is, having high leachate conductivity are considered as having low vigour (Barton, 1961; Coolbear, 1995).

Proper and safe storage conditions are defined as those that maintain seed quality without loss of vigour for three years (Joao Abba and Lovato, 1999). The loss of quality of maize seeds is not only visually observed by the poor condition of the seeds (Hell et al., 2000) but also by the poor performance of this seed when it is planted for the next season (Bellon, 2001). Seeds cannot retain their viability indefinitely and after a period of time, the seeds deteriorate (Pascual et al., 2006). In a study conducted on wheat (*Triticum aestivum* L.), by Gilbert et al. (1997), it was shown that germination after storage at tem-

peratures -10, 2.5 and 10 °C decreased with length of storage. This occurred because most of the stored seeds were infected with Fusarium graminearum Schwabe and although they were stored at an acceptable temperature (10 °C) there were lowered germination percentages (Gilbert et al., 1997). Tekrony et al. (2005) studied the effects of storage of maize on germination and vigour in an "uncontrolled" warehouse and in a controlled environment, where the temperature and humidity were monitored. Their results showed that all seed lots had 87-99% germination prior to storage but a range in seed vigour as measured by the accelerated ageing test (ISTA, 2006). After eight months storage in the "uncontrolled" warehouse, the germination declined to 50-80% (Tekrony et al., 2005). Germination and vigour tests information can be used to make informed decisions regarding the value of different seed lots (Copeland and McDonald, 2001: Tekrony, 2003; ISTA, 2006).

The aim of the present study was to test germination and vigour of maize seeds that were subjected to traditional storage during 2005 and to evaluate the vigour of fungicide treated maize seed when stored for one year under conditions of fluctuating temperature.

2. Materials and methods

2.1. Collection of samples

Maize samples were obtained from small-scale subsistence farmers in Pongola and Kosi Bay area (northern KwaZulu-Natal, South Africa) and Ponto Molangane (southern Mozambique) in 2005 (Table 1). The quantity of the maize seeds that were stored by these farmers was enough to sustain those households that they were obtained from and most gave a small sample of their supply for this study. Of the seed that was kept for food, a small percentage was kept for planting the next season.

Maize that was left in the field to dry (NHS) [10 km from Nsalamanga High School — Kosi Bay area] prior to harvesting served as one of the controls. These seeds were in a good condition. The other control was seeds commercially treated with Celest® XL ([fludioxonil (25 g ai/L)+mefenoxam (10 g ai/L)] obtained from Syngenta (SA) Pty. Ltd, Midrand).

After collection, all samples were stored in plastic bags and brown paper bags (depending on the original storage condition), under cool conditions and transported back to the Department of Microbiology and Plant Pathology laboratories (University of Pretoria, South Africa) for tests. The moisture content of the seeds (11%) that were collected in 2005 was within the percentage acceptable for maize (10–14%) (ISTA, 2006). After the tests that were conducted in 2005, all seed samples were stored in the laboratory in brown paper bags at temperatures ranging from 26–28 °C to simulate the conventional storage conditions that the seeds originally came from.

2.2. Standard germination tests

Standard germination tests were conducted on all samples according to the between-paper (BP) method of the International Seed Testing Association (ISTA, 2006). Due to the quantity

Table 1
Storage conditions and characteristics of the treated control and the maize seeds collected in 2005 from the small-scale subsistence farmers

Sample ^a	Colour of maize seeds	Storage structure	Storage container ^b	Other characteristics
Treated Control	Yellow	Commercial store	Commercially packed in plastic sack	
NHS .	White with variegated kernels	Left in field to dry	Not applicable	damage and the kernels were whole) Good condition (seeds were healthy, free of insect damage and the kernels were whole)
BHEK	White	Cement floors and walls	Maize-meal bags	Visually in good condition
JOZ	White	Commercial store	Commercially packed in plastic bags (originally obtained from Vryheid), available in general store in Jozini	Seeds had a small degree of insect damage
MOL	Yellow		Maize-meal bag	Good condition (seeds were healthy, free of insect
R22	Yellow with variegated kernels	Hut with clay walls and thatch roof	Potato bags	damage and the kernels were whole) Maize was stored on the cob, fair condition (seeds were healthy, had a bit of insect damage and some
SIH	Yellow	Cement and stone walls with a plastic sheet serving as a roof	Maize-meal bag	of the kernels were not whole) Maize stored on the cob, severe insect damage
PHEL	Yellow	Cement room	Potato bag	Good condition (seeds were healthy, free of insect damage and the kernels were whole)

^a Treated control: maize that was treated with Celest® XL [Fludioxonil (25 g ai/L)+Mefenoxam (10 g ai/L)] NHS: Maize that was left in the field to dry [10 km from Nsalamanga High School — Kosi Bay], BHEK: Bhekamangwane [Pongola], JOZ: Jozini [Jozini], MOL: Molongane [Mozambique], R22: Area 4 km from Jozini [Jozini], SIH: Sihadla west gate [Pongola], PHEL: Phelandaba [Pongola]. The samples were stored to be either planted in the next season or sustain the household.

^b The maize-meal bags originally contained a powdered form of the maize used for making porridge. The potato bags were thick brown paper bags that originally contained potatoes.

of the samples collected, only two hundred maize seeds could be randomly chosen from each sample. Four replicates of fifty seeds were placed equidistance apart on moist germination paper (containing one sheet of paper towel and four sheets of germination paper) {Anchor Paper 54×30 cm, [Agricol (Pty) Ltd, (South Africa)]} at 25 seeds per paper towel. Paper towels were rolled up and placed individually in polythene bags. Bags were sealed with an elastic band and incubated in an upright position at 25 ± 1 °C. Percentage germination was determined after seven days and rated for normal/abnormal seedlings at 11 days. Seeds were visually assessed according to the ISTA rules (ISTA, 2006). Results were presented as the percentage of seedlings that had germinated by the end of the test period.

2.3. Imbibition test

The imbibition tests were conducted according to the rules outlined by ISTA (2006). For rapid imbibition, sterile distilled water (4 mL) was placed into each well of a 24-well plastic icecube tray. These trays were chosen so that each tray represented an experimental unit. Seeds were imbibed for the following time intervals: 6, 24 and 40 h, with one seed per well. Seeds were weighed individually prior to imbibition. At the end of the time intervals, seeds were removed from the wells and left on paper towels and once air-dried were weighed again and planted onto germination paper as described for the standard germination test. Ratings were done after seven and 11 days as described for the standard germination test. In contrast, for slow imbibition, seeds were initially weighed individually and were planted onto germination paper as described for the standard germination test. After 6, 24 and 40 h imbibition, the paper towels were unrolled and seeds weighed and replaced onto the germination

sheets, left to germinate and rated at seven and 11 days as described for the standard germination test. For each time interval a different sample of seeds was used so that at the end of the incubation times the germination of the seeds could be compared following 6, 24 or 40 h imbibition.

The percentage weight increase was calculated as:

% Weight increase = (Weight after imbibition - Initial weight) ÷ Initial weight × 100

2.4. Conductivity and tetrazolium test

The conductivity of the solution, after seeds were subjected to rapid and slow imbibition, was read using an E215 Conductivity meter (Hanna Instruments, South Africa). With rapid imbibition, the seeds were placed in wells of a 24-well plastic ice-cube tray containing 4 mL sterile distilled water for 24 h. Thereafter the conductivity was measured. For slow imbibition, seeds were planted onto germination paper, as described for the standard germination test, for 40 h, and then placed in plastic ice-cube trays containing sterile distilled water for 6 h. Thereafter the conductivity was read.

Seeds from the conductivity test were used for the tetrazolium staining test. A 1% solution of 2,3,5-triphenyl tetrazolium chloride (TTC) (Labretoria, Pretoria) (10 g of TTC dissolved in a small quantity of hot water in a beaker) was transferred to a 1 L flask and tap water was added to make it up to 1 L. The seed coats of the seeds were removed and an incision was made longitudinally through the embryo and 3/4 of the endosperm as outlined in the ISTA rules (ISTA, 2006). Each seed was placed in

Table 2
Percentage germination and vigour of maize seed following storage under stress conditions

Germination (%)			Aspects of vigour measured in 2005				
Sample*	2005	2006	Conductivity (µS)	Tetrazolium staining ^t (% seeds with living tissue)			
	•			Slow imbibition	Rapid imbibition		
Treated control	94.0*de**y	86.0ex	2536 [@] cd	79.2 ^s c^x	75.0ex		
NHS	100gy	25.3bx	1181a	95.8ex	94,4fx		
BHEK	92.0dy	56.0dx	3518e	74.4bcv	62.3bx		
JOZ	88.7cy	0.0ax	2626cd	76.4bcx	75.0bx		
MOL	82.0b	33.3cx	3006de	78.0bcx	74.7dex		
R22	96.7efy	24.7bx	2030bc	89.0dy	72.2ex		
SIH	18.7ay	0.0ax	6164f	49.6ay	28.3ax		
PHEL	99.3fgy	92.0ex	1727ab	76.4bcx	73.1dex		

** Treated control: maize that was treated with Celest® XL [Fludioxonil (25 g ai/L)+Mefenoxam (10 g ai/L)] NHS: Maize that was left in the field to dry [10 km from Nsalamanga High School — Kosi Bay], BHEK: Bhekamangwane [Pongola], JOZ: Jozini [Jozini], MOL: Molongane [Mozambique], R22: Area 4km from Jozini [Jozini], SIH: Sihadla west gate [Pongola], PHEL: Phelandaba [Pongola].

¹ Triphenyl tetrazolium chloride test, a mean of 24 seeds expressed as percentage cotyledons with living tissue.

* Each value is a mean percentage of four replicates of 50 seeds. Means within a COLUMN for percentage germination not followed by the same letters are significantly different from each other (P=0.05).

** Each value is a mean percentage of four replicates of 50 seeds. Means within a ROW for the percentage germination not followed by the same letters are significantly different from each other (P=0.05).

[®] Each value is a mean percentage of four replicates of 24 seeds. Means within a COLUMN for conductivity values not followed by the same letters are significantly different from each other (P=0.05).

Seach value is a mean percentage of four replicates of 24 seeds. Means within a COLUMN for slow imbibition not followed by the same letters are significantly different from each other (P=0.05).

^ Each value is a mean percentage of four replicates of 24 seeds. Means within a ROW for the percentage germination not followed by the same letters are significantly different from each other (P=0.05).

an individual well of a 24-well plastic ice-cube tray and covered with the TTC solution. The trays were covered and incubated at 30 °C for 2 h in the dark. The seeds were then removed from the stain, cut into two halves and the cut surface was examined under a stereo-microscope (Nikon/SMZ-1, Japan). The seeds were rated as 1 — entire embryo was stained (seeds containing living tissue), 2 — part of the seed not stained and 3 — seed totally unstained (e.g. hard seed). Results were expressed as the percentage seeds containing living tissue.

2.5. Isolation of fungi

A hundred seeds from each batch of the samples and the controls were surface sterilized in 1% sodium hypochlorite (NaOCl) for 1 min. Thereafter they were rinsed three times in sterile distilled water. Seeds were directly plated onto potato dextrose agar (PDA) (Merck, Johannesburg) and malt extract agar (MEA) (Merck, Johannesburg) supplemented with rifampicin (Calbiochem®, Johannesburg). Five seeds were plated onto one plate with one seed placed in the centre and one in each quadrant. Plates were incubated at 25 °C for seven days with light/dark

cycles. Plates were examined for fungal growth. Fungi were isolated and cultured onto PDA for identification purposes. Identification of some genera was done according to Nelson et al. (1983). The fungi that occurred in most of the samples were noted and recorded at genus level.

2.6. Statistical analysis

Two-way analysis of variance (ANOVA) was performed on all data and least significant differences (P=0.05) were determined according to the student's t test.

3. Results and discussion

The sample descriptions for the abbreviations SIH, PHEL, NHS, JOZ, R22, BHEK and MOL are outlined in Table 1. In 2005, with the exception of SIH (18.7%), all the samples had percentage germination above 80% (Table 2). NHS, which gave 100% germination, differed significantly from all of the samples except PHEL (99.3%). However, after storage under suboptimum conditions for one year the percentage germination of all the samples decreased significantly. Two samples, JOZ and SIH failed to germinate in 2006 (Table 2).

In this study the treated control had a germination of 94.0%, which decreased to 86.0% due to storage under sub-optimum conditions (Table 2). This was still an acceptable decrease in germination, compared to most of the other samples, as the acceptable percentage germination of maize is 70% according to the Plant Protection Act (1976). Storing these treated seeds had little effect on the germination. This was also confirmed by the results of the vigour tests. Following rapid imbibition this sample had a 55.7% weight increase and maintained germination above 85% after 40 h rapid imbibition (Table 3). Likewise, following 40 h slow imbibition there was a 46.0% weight increase of these treated seeds, but a percentage germination of 95.8% (Table 3). Slow imbibition is the natural way that seeds imbibe water (Copeland and McDonald, 2001; ISTA, 2006) and the increase in weight did not have an effect on the percentage germination. This sample had a leachate conductivity value of $2536\,\mu\text{S}$ and was lower than some of the samples (Table 2). This result was reflected by the percentage seeds with living tissue in the tetrazolium staining test. Results for rapid (75%) and slow (79%) imbibitions did not differ significantly and is an indication of the good condition of the seeds, relative to some of the other samples (Table 2). The treated control had the lowest percentage of storage fungi (11%) with the genus Rhizopus (5%) predominating (Table 4).

Researchers from Centro Internacional de Mejoramiento de Maíz y Trigo (CIMMYT) have been helping small-scale farmers in Mexico with training in proper storage of maize since 1997 (Bellon, 2001). The training helped farmers to rather make use of a storage pesticide (fostoxin) and silos for their grain so that the stored seed could be kept for a longer period of time (Bellon, 2001). In this current study, results confirmed that treating seeds prior to storage was advisable, as the treated control maintained acceptable germination percentages even after storage under sub-optimum conditions.

Table 3

Percentages weight increase and germination following fast and slow imbibitions of maize seeds that were subjected to stress conditions

		Samples#							
		Treated control	NHS	BHEK	JOZ	MOL	R22	SIH	PHEL
% Weight increase		,							FIREL
Slow imbibition Rapid imbibition	6 24 40 6 24 40	13.9*ab**x 31.8by 46.0cz 23.7cx 40.9cy 55.7dz	20.7ex 35.5cdy 38.1by 17.7bx 32.4by 47.5cbz	11.2ax 36.1dy 51.9dz 21.2cbx 41.4cy 44.2by	43.0ex 52.7fy 51.1dy 42.9dx 51.2dy 57.0dy	11.3ax 27.1ay 33.3ayz 13.5ax 32.4by 30.0ay	17.6bex 33.4cby 39.4by 23.0ex 35.7by 52.1edz	37.7dx 42.8ex 60.2ey 40.0dx 51.0dy 64.2ez	13.1abx 31.9by 45.7ez 19.0bx 24.2ax 42.0by
% Germination Slow imbibition Rapid imbibition	40 40	95.8dey 88.9ex	100ey 93.1efx	91.7dx 95.8fx	41.6by 23.6bx	84.8cy 73.0cx	97.2dex 97.2fx	2.8ay 0.0ax	100ey 79.3dx

^{*}Treated control: maize that was treated with Celest® XL [Fludioxonil (25 g ai/L)+Mefenoxam (10 g ai/L)], NHS: Maize that was left in the field to dry [10 km from Nsalamanga High School — Kosi Bay], BHEK: Bhekamangwane [Pongola], JOZ: Jozini [Jozini], MOL: Molongane [Mozambique], R22: Area 4 km from Jozini [Jozini], SIH: Sihadla west gate [Pongola], PHEL: Phelandaba [Pongola].

The field sample (NHS) that was used as a second control represented freshly harvested seed and gave 100% germination in 2005. As soon as these seeds were stored in brown paper bags at between 26 and 28 °C, they were placed under the same stress conditions as the other samples, and germination dropped to 25.3% in 2006 (Table 2). Generally maize left in the field has more time to dry so that it has a lowered moisture content percentage (Appert, 1987). Results from the vigour test confirmed that the seeds had high vigour in 2005 with a low leachate conductivity value of 1181 μS and high percentage seeds with living tissue (95.8% following slow imbibition and 94.4% following rapid imbibition) (Table 2). The weight increase following slow and rapid imbibitions was lower when compared to some of the other samples (Table 3). The percentage weight increase following slow and rapid imbibitions for 40 h was 38.1 and 47.5%, respectively and percentage germination was 100 and 93.1%, respectively (Table 3). These results mirrored the germination in 2005 (Table 2). This seed sample was infected with predominantly Cladosporium spp. (16%) and Fusarium spp. (10%) (Table 4). Although this control had 100% germination when initially tested, storage under sub-optimum conditions would allow for storage fungi to become a major threat to the quality of the grain. In a study by Qasem and Christensen (1958), the storage fungi most often involved in deterioration of field stored maize were typically found after maize had been stored under warm conditions, when the moisture content was between 14 and 18%.

With the samples that still had maize on the cob, R22 (96.7%) did not differ significantly from PHEL (99.3%) in the standard germination test. In 2005, R22 did not differ significantly from the treated control (94.0%). However, in 2006 the germination of this sample (R22), decreased by 72%. R22 had a 39.4% weight increase and 97.2% germination following slow imbibition for 40 h and had a conductivity value of 2030 μ S. This was reflected by the percentage seeds with living tissue (89 and 72.2%), following slow and rapid imbibitions (Table 2). In contrast, PHEL

had a 7.3% decrease in germination and did not differ significantly from the treated control in 2006. This sample had a conductivity value of 1727 μS and had percentage seeds with living tissue above 70% following both slow and rapid imbibitions (Table 2). Most subsistence farmers prefer to store maize on the cob over the fire and the smoke helps to prevent seeds from spoiling and from pest infestation but most of the time quality of the maize was found to be inferior, leading to a low germination rate and lower yields (Modi, 2004). Sparg et al. (2005) found that in many other crops the application of smoke stimulated seed germination.

Table 4
Percentage of fungi found in the maize seed controls and samples that were subjected to stress conditions

Fungal genera	Samples ^{a b}								
	Treated control	NHS	BHEK	JOZ	MOL	R22	SIH	PHEL	
Aspergillus spp.	2		14	10	12	6	14	2	
Cladosporium spp.	_	16	25	-	20	16	10	12	
Fusarium spp	-	10	3	-	-	~	30	5	
Penicillium spp.			-	-	-	_	2	-	
Rhizopus spp.	5	_	5	20	_		15	2	
Stenocarpella spp.	-		-	2	***	-		_	
Other	4	7	2	4	8	20	_	~	
Total fungi	11	33	49	36	40	42	71	21	

a Treated control: maize that was treated with Celest® XL [Fludioxonil (25 g ai/L)+Mefenoxam (10 g ai/L)], NHS: Maize that was left in the field to dry [10 km from Nsalamanga High School — Kosi Bay], BHEK: Bhekamangwane [Pongola], JOZ: Jozini [Jozini], MOL: Molongane [Mozambique], R22: Area 4 km from Jozini [Jozini], SIH: Sihadla west gate [Pongola], PHEL: Phelandaba [Pongola].

b For the percentage of fungi for each sample, four replicates of 25 seeds were used per sample.

^{*} Each value is a mean percentage of four replicates of 24 seeds. Means within a ROW for the weight increase not followed by the same letters are significantly different from each other (P=0.05).

^{**} Each value is a mean percentage of four replicates of 24 seeds. Means within a COLUMN for percentage germination not followed by the same letters are significantly different from each other (P=0.05).

The germination of BHEK was 92.0% and did not differ significantly from the treated control in 2005. The germination decreased to 56% in 2006 (Table 2). This sample had a high leachate conductivity value (3518 µS) and percentage seeds with living tissue below 75% following slow (74.4%) and rapid (62.3%) imbibitions (Table 2). In contrast to all the trends noticed with the other samples, the high conductivity value was not an indication of the condition of the seed lot as percentage germination following slow and rapid imbibitions was 91.7 and 95.8% respectively (Table 3). In this sample, the high conductivity did not necessarily indicate a low vigour seed lot. Decreased membrane integrity could be as a result of storage deterioration (most of the samples in this study) and mechanical injury (Copeland and McDonald, 2001), however, other factors could play a role in increased leachate conductivity, such as initial seed moisture and seed size (Tao, 1978; Gras et al., 1990).

JOZ and MOL had percentages germination of 88.7 and 82.0%, respectively and differed significantly from each other and from the other samples, as most of the other samples had percentages above 90% except SIH (18%) (Table 2). JOZ decreased from 88.7% (2005) to 0.0% (2006). The vigour tests in 2005 showed that this sample had percentage weight increase above 50% for both slow and rapid imbibitions after 40 h. The percentage germination was lower following slow (41.6%) and rapid (23.6%) imbibitions compared to the other samples (Table 3). JOZ had a conductivity value of 2626 μS (Table 2). The deterioration in the JOZ sample could be explained by the fact that these seeds had mild insect damage. Even though they were commercially packaged in plastic bags, they were in a general store without air-conditioning and the temperature during summer (when the seed was collected) reached 35-37 °C during midday. This was not conducive to maintaining the quality of this sample. Combined with the temperature, insect damage and storage stress, these seeds failed to germinate in 2006.

In a study conducted by Casini (1999), the advantages of storing dry grain in plastic bags were evaluated. Dry grain (maize, soybean and wheat) can be stored in plastic bags for 24 months, if certain guidelines are followed. Ideally grain should be kept with low oxygen content and a high concentration of carbon dioxide (CO₂). This gives control of insects and fungi that are the major causes of increases in the temperature of the grains. In this sample, such guidelines were not followed in contrast to the treated control. However, storage in plastic bags is better than storage in paper bags but the original condition and an optimum temperature needs to be taken into account (Casini, 1999).

For the SIH sample, decline in germination from 18.7% to failure to germinate the following year was expected as this sample had severe insect damage. The standard germination results of this sample differed significantly from the other treatments and the control in 2005 as all the other samples had germination percentages above 80%. With the exception of JOZ (0.0% germination), SIH differed significantly by failing to germinate (0.0%) in 2006 when compared to the other samples. This was comparable to a study by Thamaga-Chitja et al. (2004) who found that storing maize seeds in sacks provided little protection against insects and maize stored in this manner absorbed mois-

ture from the floor (typically mud, sealed with cow dung or cement). Following slow and rapid imbibitions, SIH had a percentage weight increase that was above 60% (Table 3) and had a 2.8 and 0.0% germination, respectively. The weakened initial condition of the SIH seeds was indicated by the high leachate conductivity value (6164 µS) and was mirrored by the low percentage seeds with living tissue following the tetrazolium test (49.6 after slow imbibition and 28.3% after fast imbibition) (Table 2). The standard germination test should ideally provide the seeds with optimum conditions to germinate (Copeland and McDonald, 2001; Tekrony et al., 2005), however even with these optimum conditions SIH did not germinate well and adding stress (vigour tests) in addition to the insect damage, illustrated the extremely low vigour potential of this seed lot. Most of the vigour tests give an indication of the field performance of the seed lot, however, there are other factors to consider as well, such as environmental conditions (Copeland and McDonald, 2001). This sample when exposed to a "controlled stress environment" failed to germinate so the chances of this sample producing any seedlings in an uncontrolled field environment is very low to non-existent. The storage fungi isolated from this sample totalled 71% positive incidence with Aspergillus (14%), Fusarium (30%) and Rhizopus (15%) species predominating (Table 4).

Of the storage conditions that were presented in this study the two samples (R22 and PHEL) that were still on the cob and in potato bags had a high germination, above 95%, in 2005. The difference in the decrease in their germination in 2006 (24.7 and 92.0%, respectively) can be explained by the initial condition of those seeds. R22 was in fair condition in contrast to PHEL, which was in good condition. Field stored maize is useful as fresh seed for immediate use and for planting. Long-term storage as indicated in this study is not feasible as the moisture content of the seed will increase to above 14% and as the subsistence farmers may not have the knowledge and equipment to get those seeds back to an acceptable moisture content, those seeds will deteriorate. Modi (2004) showed the limitations of the conventional storage structures, where structures are made very weak and allow insects to enter and provide an environment for storage fungi to thrive. Bags stored in either very cold temperatures or in cement structures work very well in terms of protecting seeds from most pest and insects. Sealed plastic bags, as was the case in the treated control, are the best as indicated by Gras et al. (1990), but seeds need to be in a good condition (mechanically and insect damaged seed must be removed) and storage temperatures must be kept as low as possible (4-10 °C).

This study reiterated the importance of proper storage techniques and their impact on germination and vigour of maize seeds. Apart from correct storage, the original condition of the seeds needs to be taken into account before they are stored as insect damage could aggravate the problem as shown in this study. Seed treatments have a major role in protecting the seed during storage (Chen and Burris, 1993) and can also play an important role in achieving uniform seedling emergence under certain conditions (Rane and Ruhl, 2002). This study confirmed that using a fungicide such as Celest® XL protected the seed and was effective even after storing seeds at 26–28 °C.

Small-scale farmers that may not have facilities to store their seed at $4-10\,^{\circ}\text{C}$, will benefit from protecting their seed with a fungicide so as to provide undamaged seed for planting the following season.

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