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Combined Effect of Hermetic Bag and Insect Resistant Variety for the Control of Larger Grain Borer and Maize Weevil in Stored Maize

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Abstract: ***Combined effect of hermetic bag and varietal resistance was studied for control of Prostephanus truncatus and Sitophilus zeamais. Two maize varieties: resistant (CKPH08028) and susceptible (PH3253) were used in combination with SuperGrain bag II™, PICS bag, Smartbag -1, Polypropylene bag and Actellic super dust. A mean of 5.2% Carbon dioxide for both PICS and SuperGrain II™ bags and 2.6% for Smartbag -1 were recorded. PICS and SuperGrain II™ bags suppressed insect population, prevented grain loss and cross - infestation of insects from the surrounding environment. Grain weight losses were 0.3% in the PICS and 0.9% in the SuperGrain IV-R™ bags compared to 23.9% in the polypropylene bags, 180 days after storage. No grain protection benefits were gained when either insect resistant (CKPH08028) or susceptible (PH5253) maize grains were stored in PICS or SuperGrain II™ bags (<5% damage and <1% weight loss). Synergistic benefits in protection were gained when the weight loss of CKPH08028 grains stored in either PICS or SuperGrain II™ bags were compared to that of the same variety stored in the polypropylene bags. Admixture of maize grain with Actellic super dust and storage in polypropylene bag did not prevent infliction of damage (45.6%) and weight loss (13.6%) due to insect pests. The novelty of the work is demonstrated in the potential use of hermetic bags in combination with insect resistant maize technologies to significantly reduce weight loss from 30% to less than 1% without use of pesticide. This would improve food security at household level. The findings of this study would support agricultural policy formulation and monitoring of loss reduction activities.***

Keywords: Hermetic bag, insect resistant maize, *Prostephanus truncatus*, *Sitophilus zeamais*, weight loss

1. Introduction

Maize is an important staple food crop grown widely in sub-Saharan Africa and a major source of feed, biofuel and industrial raw material (Purseglove 1992). The crop is important socially, politically and economically (McCann *et al*. 2006). Despite increased production, postharvest losses due to insect and mould infestation remain a big challenge (FAO 2009). During storage maize is prone to attack by field - to -store insect pests among which the majors ones are the maize weevil *Sitophilus zeamais* (Motsch.) and Angoumois grain moth *Sitotroga cerealella* (Oliv.) and the introduced invasive larger grain borer *Prostephanus truncatus* (Horn) (Chebet *et al.* 2013). Adults of these insects feed on undamaged grains generating flour (dust) rendering the grain unfit for human consumption (Ofuya *et al*. 2008). Grain damage leads to loss of weight, poor quality and low germination rate (Enobakhare and Law -Ogbomo 2002). In its area of origin in Meso-America and Mexico, *P. truncatus* was found not to infest maize during storage (Wong - Corral *et al*. 2001). The infestation of maize grain starts in the field just before harvest and the insects are carried into the store where the population builds up fast (Adedire and Lajide 2003, Hodges *et al*. 2011).

Traditional storage structures confer unsatisfactory protection against these insect pests in stored grain (Ngamo *et al.* 2007). The main method for the control of insect pests of stored grain still remains the application of synthetic insecticides. Although insecticides are very effective, there are concerns on their impact on the environment, human health and development of insect resistance (Braga *et al*. 2011; Ogendo 2004, Pereira *et al*. 2009). The limited choice of commercially available insecticides, unreliable supply and cost has contributed to low adoption by resource - poor smallholder farmers (Ogendo *et al*. 2004).

It is estimated that 14 -50% of the maize produced in developing countries is lost during storage (Ojo and Omoloye 2012). To avoid the risk of low quality grain and high grain losses during storage forces smallholder farmers to sell surplus grain immediately after harvest at low prices (Gitonga *et al*. 2013; Yigesu *et al*. 2010). The optimal time for selling grain is determined, among other factors, by grain damage and weight losses (Yigesu *et al*. 2010). While Kenya has for long pursued the goal of self -sufficiency in maize and other crops, majority of rural households are net buyers of maize (Brooks *et al*. 2009). Food shortage is, therefore, a challenge encountered by resource poor smallholder farmers. Affordable, effective, chemical -free and environmentally friendly control methods are urgently required.

What type of technologies that would reduce grain weight loss from 30% to less than 3% is a research question addressed by this study. The target was based on a study by Mutambuki *et al.* (2011) that reported a 30% grain weight loss in stored maize where no insect pest control intervention was applied. Hermetic bagging and insect resistant maize variety is just such an extension of the 'choice' of technologies available to farmers to explore to lever food security and sustain maize- dependent livelihoods. Hermetic bags are airtight containers which prevent oxygen and water movement between the outside atmosphere and the stored grain or product. The respiration by the grain and insect inside the bags changes inter- granular atmosphere consuming oxygen and producing carbon dioxide (Baoua *et al*. 2013). Depending on the type of the container and insect population, oxygen levels is reduced (hypoxia) from 21 to below 10% and carbon dioxide levels increased (hypercarbia) within a short period of time (Mutungi *et al*. 2014). However, the attainment of high enough carbon dioxide level to kill all the insects in hermetic bags is a challenge. Whereas several studies have examined grain storage infestation under hermetic conditions, little work, however, has been reported on the combined effect of hermetic bagging and insect resistant maize variety. Therefore, the objective of this study was to evaluate the combined effect of hermetic bagging and insect resistant maize against the larger grain borer and maize weevil infestation.

2. Materials and Method

2.1 Study Site

The studies were carried out at Kenya Agricultural and Livestock Research Organisation (KALRO) - Kiboko in Makueni County. Kiboko lies within 37.7234^{o}E and 2.2172^{o}S at 975m above sea level (CIMMYT 2013). The region is hot and dry with mean annual minimum and maximum temperatures of 16.5^{o}C and 28.6^{o}C, respectively. Maize is among the main crops grown and marketed in the region.

2.2 Hermetic Bags

The PICS® and SuperGrain II™ bags (74cm wide and 64cm length) of 90kg holding capacity were purchased from the manufacturers' agents in Nairobi. The open end of these bags was cut to fit the height dimension of the conventional farmer bags. As recommended, the hermetic bags were placed inside polypropylene bags to provide support and handling convenience. PICS® and SuperGrain II™ bags are type of multi-layer co-extruded tougher plastics with low permeability in which oxygen is depleted fast.

A newly developed hermetic bag, Smartbag-1, was provided by MashAgrik company of South Africa. It is composed of an outer layer of standard woven polypropylene and inner liner of high density polyethylene (HDPE), 80 microns thick. The hermetic bag had grain - holding capacity of 40kg.

The polypropylene bags (farmers' traditional storage) of holding capacity of 50kg were bought from a local market in Nairobi. Polypropylene bags are made from woven synthetic fibre that is similar to plastic but is more degradable when exposed to sun rays (ACDI/VOCA-Kenya, 2007). The bags somewhat prevent free circulation of air and are difficult to fumigate.

2.3 Insects

The insects used in the study were reared at the postharvest laboratory, KALRO, Kiboko. The *Prostephanus truncatus* (Coleoptera: Bostrichidae) and *Sitophilus zeamais* (Coleoptera: Curculionidae) cultures were maintained on whole susceptible hybrid H513 maize variety at 27^{o}C and 65% relative humidity.

2.4 Sources of resistant and susceptible maize and preparation prior to storage

Seed for resistance (Maize hybrid CKPH08028) to *P. truncatus* and *S. zeamais* was obtained from the International Centre for Maize and Wheat Improvement Centre (CIMMYT) while the susceptible Hybrid maize variety PH3253 was purchased from the Agrovet at the local market. CKPH08028 maize grain texture is semi - flint with yield estimated at 8.03 t/ha while PH3253 grain is dent with 7.32 t/ha yield (Mwololo *et al*. 2013). The two Hybrids were planted at Kiboko research farm for grain increase in mid - April 2013 during the long rains season. Two seeds were planted per hill in a row of 5m length spaced 75cm apart and plant -to -plant distance of 25cm. Following germination, the plants were thinned to one plant per hill after two weeks to attain a population density of 53,000 plants per hectare. Fertilizer was applied at the recommended rate of 60kg N and 60kg P_2O_5 per hectare for Kiboko area. Nitrogen fertilizer was applied twice. The field were kept free of weeds by hand weeding. Water stress was avoided by irrigating the fields. The hybrids were harvested in late - August 2013, the cobs sun -dried for seven days then shelled. The moisture content of the maize grain at the start of the storage period was 12.4 ± 0.0% for resistant hybrid maize CKPH08028 and 12.3 ± 0.0 (wet basis) for the susceptible hybrid PH3253 maize variety while the grain damage for both varieties was 0.6 ± 0.2%. The grains were not disinfested to kill any insect or eggs present due to random natural infestation in the field.

2.5 Treatments and Design

Three hermetic bags and the standard woven polypropylene bag in combination with insect resistant or susceptible hybrid maize were tested for their effectiveness against storage insect pests attack. The experiment comprised of nine treatments: (1) SuperGrain II™ bag+ resistant maize (SGB + R); (2) SuperGrain II™ bag + susceptible maize (SGB + S); (3) PICS bag + resistant maize (PICS + R); (4) PICS bag + susceptible maize (PICS + S); (5) Smartbag-I + resistant maize (SMTB + R); (6) Smartbag-I + susceptible maize (SMTB + S); (7) Polypropylene bag + resistant maize (PPB + R); (8) Polypropylene bag + susceptible maize (PPB + S) and (9) Polypropylene bag + susceptible maize + Actellic super dust (PPB + S+A). A completely randomised design was used where the level of factors were 8 treatments and two storage periods. The containers were placed in a room described by DeGroote *et al*. (2013) at Kiboko in four rows, with 0.5m distance between the rows and 0.3m within the rows. All the treatments were held under ambient conditions. The temperature was recorded on a daily basis.

2.5.1 Grain Bagging and Storage

The hermetic (PICS and SuperGrain II™ bag) and polypropylene bags were filled with 30kg grains each of Hybrid maize variety CKPH08028. Similar preparation was made for susceptible Hybrid PH3253. Polypropylene bag filled with susceptible Hybrid PH3253 maize served as a control. The bags were artificially infested with adult larger grain borer and maize weevil at the rate of 1 adult beetles/kg grains (15 *P. truncatus* +15 *S. zeamais*) at the top centre of the grains. The bags were stored at ambient conditions in a shaded barn for six months' period. The entrapped air was squeezed out and then the bags secured tightly with rubber straps. according to manufacturer's instructions. The treatments were arranged in a completely randomised design with four replications. The bags were held on wooden pallets in four rows in the barn with open wire mesh and concrete walls at ambient conditions. The pallets were placed about 0.3m between and 0.2m within the rows.

Oxygen, carbon dioxide and temperature levels inside PICS bag, Smartbag -1 and SuperGrain II™ bag were measured at 10 - day intervals using Mocon Pack Check® Model 325 portable oxygen/carbon dioxide headspace analyser (MOCON Inc., Minneapolis, USA). The analyser is fitted with sensors and automatic internal air sampling pump. To facilitate taking of measurements, the inner HDPE liner was pierced with the analyser needle near the top to draw 5 mL of air for determining oxygen, carbon dioxide and temperature. Oxygen and carbon dioxide levels determined by electrochemical cell and non - dispersive infrared methods, respectively. The needle holes were then plugged with circular adhesive pads (10mm diameter) immediately after taking the readings. The mean of the three 10-day readings in a month was then recorded as 30-day reading. Subsequent readings were taken from the same spot by unsealing and re-sealing with the adhesive pad and re-enforced by wind tape.

2.5.2 Grain Sampling and Damage Analysis

Samples were drawn non - destructively at 90 and 180 days after the setup, respectively, using a vertical sampling 6 -slot spear (probe) with special care taken not to pierce the bags.. Repeated sampling from the same storage structure reflects farmer practices of opening the structures at regular interval to draw grain for use as household food. At each sampling time, the bags are unsealed, grains drawn from the centre and two peripheral points then the bags resealed again. The grains from these three points were bulked and the whole lot (about 450g) per replicate of each treatment used as a working sample. Each sample was put in a clean labelled zip-lock (8 x 6 cm) plastic bag for subsequent insect damage analysis in the laboratory. In the laboratory, a set of sieves (4.75 and 1.0 mm aperture size) were used to separate the insects from the grains. After sieving the samples, a half of each grain sample was obtained by use of riffle divider and sorted out into undamaged and damaged grain fractions. A grain was regarded damaged when its surface showed the presence of holes or tunnels made by insect's emergence or feeding activities while undamaged showed none. The number and weights of each fraction were recorded. Grain damage and weight loss was calculated as follows:

$$\text{Grain damage (\%)} = \frac{\text{Number of insect damaged grain}}{\text{Total number of grain}} \times 100$$

$$\text{Weight loss (\%)} = 100 \times \frac{(W_u \times N_d) - (W_d \times N_u)}{W_u \times (N_u + N_d)} \text{ (Boxall, 1986)}$$

Where W_u and W_d is the weight of undamaged and damaged grain, respectively; N_u and N_d is the number of undamaged and damaged grain, respectively.

The hermetic bag and polypropylene bags were inspected for the presence of holes made by *P. truncatus* during storage at termination time.

2.6 Data Analysis

The data were subjected to analysis of variance (ANOVA). Insect count data were transformed to $\log_{10}$ (count +1) scale while percentage oxygen, carbon dioxide, , grain damage and weight loss data were square root transformed prior to statistical analysis. The general linear model (GLM) procedure of GenStat software release 12.1 for windows (VSN International Ltd, 2009) was used for all the analysis. The treatments and storage period were the main effects. The associated interactions of the main effects were included in the analysis. Significant differences were separated using Bonferroni test at 0.05 probability.

3. Results

The moisture content of the maize grain at the start of the storage period was 12.4 ± 0.0% for resistant hybrid maize CKPH08028 and 12.3 ± 0.0 (wet basis) susceptible hybrid PH3253 while the grain damage for both varieties was 0.6 ± 0.2%.

3.1. Temperature, oxygen and carbon dioxide levels

Temperature levels changed significantly with storage period ($F_{6,123}$ =40.49; P<0.001) but not with treatment (P = 0.318) (Figure 1). There were no significant interaction differences between hermetic treatments and storage period was detected (P = 0.767). Overall, the temperature levels changed over the course of the storage duration but remained relatively high at 28 - 32°C. This corresponds to the typical hot weather that prevails at Kiboko before the onset of short rains which starts in October and ends in February.

There were significant differences between treatment, storage time and treatment by storage interaction in affecting oxygen and carbon dioxide levels (Table 1). No significant differences between treatments in level of oxygen and carbon dioxide at the onset of the treatments (zero days) was observed (Table 2); however, significant differences were observed 30 days after treatment application. The level of oxygen drastically dropped in PICS bag followed by SuperGrain II™ bag; however, there was a gradual decline in oxygen in Smartbag-I during the storage period (Table 2). There was high concentration of carbon dioxide in SuperGrain II™ bag between 30 and 120 days after treatment application followed by PICS bag; nevertheless, the interaction showed that the highest concentration of carbon dioxide was recorded from PICS bag 150 days after treatment application (Table 2). Smartbag-I sustained the least concentration of carbon dioxide throughout the storage period.

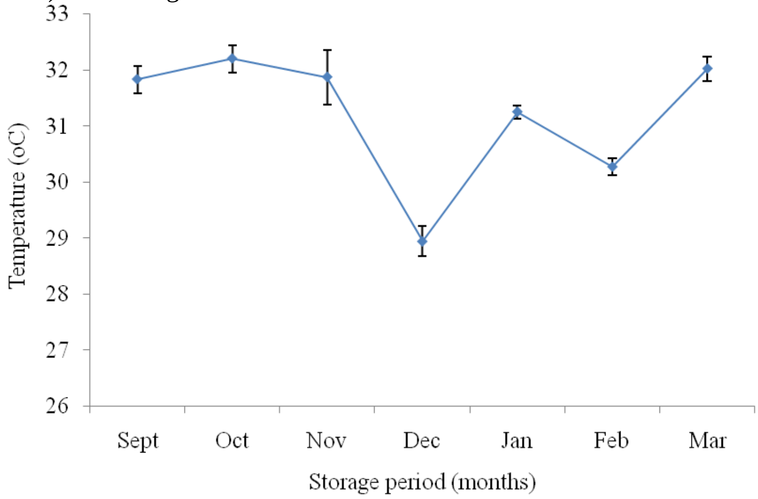

Figure 1: Mean temperature of the hybrid maize grains stored in hermetic bags over six months storage duration.

Table 1: Factorial analyses of the effect of treatment and storage period on changes in oxygen and carbon dioxide levels

Factor	*Mean Square*	*F*	*d.f*	*P-value*
Oxygen level				
Treatment	4.357	587.38	5	<0.001
Storage period	2.236	301.38	6	<0.001
Treatment x Storage period	0.186	25.06	30	<0.001
Residual	0.007		123	
Carbon dioxide level				
Treatment	2.958	136.90	5	<0.001
Storage period	6.493	300.50	6	<0.001
Treatment x Storage period	0.372	17.22	30	<0.001
Residual	0.022		123	

Table 2: Effect of the treatment and storage period interactions on changes in oxygen and carbon dioxide levels in the hermetic bags (mean[1] ± SE)

Treatment	Storage period (days)							
	0	30	60	90	120	150	180	Mean[2]
Oxygen level (%)								
SGB + R	20.3 ± 0.0mn	13.4 ± 0.1fg	15.1 ± 0.5g-i	16.0 ± 0.5h-j	16.0 ± 0.0h-j	15.9 ± 0.2h-j	15.5 ± 0.2hi	16.0d
SGB + S	20.4 ± 0.1mn	12.1 ± 0.2ef	15.5 ± 0.1hi	14.6 ± 0.6gh	14.5 ± 0.3gh	16.0 ± 0.3h-j	15.1 ± 0.2g-i	15.5c
PICS + R	20.3 ± 0.1mn	9.4 ± 0.0a-c	11.1 ± 0.1de	8.9 ± 0.7ab	8.2 ± 0.3ab	12.0 ± 0.5ef	11.4 ± 0.2de	11.6a
PICS + S	20.4 ± 0.0mn	9.0 ± 0.3ab	12.4 ± 0.3ef	9.2 ± 0.7ab	10.9 ± 0.4c-e	13.3 ± 0.2fg	10.1 ± 0.2b-d	12.2b
SMTB + R	20.4 ± 0.1n	18.4 ± 0.2k-n	18.3 ± 0.1k-n	17.6 ± 0.2j-l	18.2 ± 0.1k-m	18.9 ± 0.1l-n	19.3 ± 0.3l-n	18.7f
SMTB + S	20.5 ± 0.0n	16.7 ± 0.2i-k	16.4 ± 0.2h-k	16.7 ± 0.4i-k	16.5 ± 0.2h-k	19.0 ± 0.3l-n	19.6 ± 0.1l-n	17.9e
Mean[2]	20.4e	13.2a	14.8b	13.8b	14.0c	15.9d	15.2d	
Carbon dioxide (%)								
SGB + R	0.8 ± 0.1ab	6.7 ± 0.5m-p	5.3 ± 0.3i-o	5.2 ± 0.5i-o	5.1 ± 0.1i-n	5.6 ± 0.3k-o	5.4 ± 0.2j-o	4.9c
SGB + S	0.7 ± 0.1a	6.9 ± 0.1n-p	5.4 ± 0.2j-o	6.6 ± 0.7l-p	6.6 ± 0.3l-p	6.4± 0.8l-p	6.5 ± 0.4l-p	5.6d
PICS + R	0.8 ± 0.0a	5.1 ± 0.1i-n	3.5 ± 0.2e-i	5.3 ± 0.4j-o	6.4 ± 0.7l-p	7.4 ± 0.7o-q	8.6 ± 0.1pq	5.3cd
PICS + S	0.7 ± 0.0a	5.3 ± 0.0j-o	3.1 ± 0.3e-h	5.0 ± 0.4i-n	4.8 ± 0.3h-m	6.5 ± 0.1l-p	9.7 ± 0.4q	5.0c
SMTB + R	0.6 ± 0.0a	2.9 ± 0.4d-f	2.7 ± 0.1c-f	3.6 ± 0.2e-j	2.9 ± 0.1d-g	2.2 ± 0.1c-e	1.6 ± 0.3bc	2.4a
SMTB + S	0.6 ± 0.0a	2.9 ± 0.1d-f	3.9 ± 0.2f-k	4.6 ± 0.3g-l	4.6 ± 0.2h-l	2.7 ± 0.3c-f	1.7 ± 0.2bc	3.0b
Mean[2]	0.7a	5.0c	4.0b	5.1c	5.1c	5.1c	5.6c	

Where SGB + R=SuperGrain II™ bag + resistant maize, SGB + S=SuperGrain II™ bag + susceptible maize, PICS + R =PICS bag + resistant maize, PICS + S =PICS bag + susceptible maize, SMTB + R= Smartbag -1 + resistant maize and SMTB + S = Smartbag -1 + susceptible maize.

[1]Means within the same column and row followed by the same letter are not significantly different at P = 0.05 level.
[2]Means of the main treatment effects followed by the same letter are not significantly different at P = 0.05 level.

3.2 Number of adult P. truncatus and S. zeamais

There were significant differences between treatment, storage time and treatment by storage interaction in affecting the number of insects (Table 3). The least number of both insects was recorded when the susceptible (PH3253) or resistant (CKP08028) maize hybrids were stored in PICS bags (Table 4). Grains stored in Smartbag-I harboured large number of *P. truncatus* than SuperGrain II™ bag. The susceptible hybrid (PH3253) stored in polypropylene bag treated with Actellic dust had the highest number of *P. truncatus* than *S. zeamais*. Both susceptible (PH3253) and resistant (CKP08028) maize hybrids had the highest number of *S. zeamais* when stored in polypropylene bag.

Table 3: Factorial analyses of the effect of treatment and storage duration on the number of *Prostephanus truncatus* and *Sitophilus zeamais* in grain samples.

Factor	*Mean Square*	*F*	*d.f*	*P-value*
No. of *P. truncatus*				
Treatment	2.359	23.31	8	<0.001
Storage period	14.655	144.84	1	<0.001
Treatment x Storage period	0.861	8.51	8	<0.001
Residual	0.101		51	
No. of *S. zeamais*				
Treatment	9.334	5.19	8	<0.001
Storage period	211.400	117.43	1	<0.001
Treatment x Storage period	3.722	2.07	8	0.057
Residual	1.800		51	

Table 4: The effect of treatment and storage period interaction on the mean[1] number of *Prostephanus truncatus* (± SE) and *Sitophilus zeamais* (± SE) in grain samples

Treatment	No. of *P. truncatus*			No. of *S. zeamais*		
	Storage period (days)			Storage period (days)		
	90	180	Mean[2]	90	180	Mean[2]
SuperGrain II™ bag + CKPH08028	1 ± 1ab	9 ± 4abc	5ab	1 ± 0a	32 ± 8bcde	17ab
SuperGrain II™ bag + PH3253	0 ± 0a	8 ± 5abc	4ab	0 ± 0a	36 ± 14bcde	18ab
PICS bag + CKPH08028	0 ± 0a	1 ± 0ab	1a	0 ± 0a	9 ± 2abc	5a
PICS bag + PH3253	1 ± 1ab	1 ± 1ab	1a	0 ± 0a	10 ± 4abc	5a
Smartbag -1 + CKPH08028	2 ± 1ab	96 ± 31de	49cd	4 ± 1ab	12 ± 5abcd	8a
Smartbag -1 + PH3253	4 ± 3abc	187 ± 48e	95de	4 ±1ab	43 ± 12cde	23bc
Polypropylene bag + CKPH08028	5 ± 3abc	22 ± 7cd	13bcd	2 ± 1a	52 ± 18de	27ab
Polypropylene bag + PH3253	1 ± 1a	17 ± 4cd	9abc	8 ± 2abc	57 ± 17e	32b
Polypropylene bag +Actellic + PH3253	10 ± 2bc	407 ± 41e	209e	0 ± 0a	15 ± 5abcde	7ab
Mean[2]	3a	83b		2a	29b	

[1]Means within the same column and row followed by the same letter are not significantly different at P=0.05 level.
[2]Means of the main treatment effects followed by the same letter are not significantly different at P=0.05 level.

3.3 Grain Damage and Weight Loss

There were significant differences between treatments, storage time and interaction between treatment and storage time in grain damage and weight loss (Tables 5 and 6). SuperGrain II™ bag and PICS bags were the safest storage structures in protecting grains against *P. truncatus* and *S. zeamais* with less than 5% grain damage and less than 1% weight loss 180 days after storage, irrespective; both in susceptible and resistant maize (Table 6). Grains of both the susceptible (PH3253) and resistant (CKP08028) hybrids stored in Smartbag-1 bags had the highest damage (49%) and weight loss (15%) followed by grains stored in polypropylene bags treated with Actellic super dust. Some level of grain protection against the *P. truncatus* and *S. zeamais* was observed when either of Smartbag-1 bag (22% weight loss) or polypropylene bag (17.4% weight loss) was combined with the resistant maize (CKPH08028) after 180 days of storage (Table 6) compared to susceptible PH3253 maize variety. A significant increase in grain damage and loss was observed with an increase in storage time in different treatments but not in PICS bags.

Table 5: Factorial analyses of the effect of treatment and storage duration on damage and weight loss caused by storage insect pests

Factor	*Mean Square*	*F*	*d.f*	*P-value*
Grain damage (%)				
Treatment	38.483	1027.23	8	<0.001
Storage period	243.728	6505.93	1	<0.001
Treatment x Storage period	15.842	422.89	8	<0.001
Residual	0.037		51	
Weight loss (%)				
Treatment	12.350	693.15	8	<0.001
Storage period	94.458	5301.58	1	<0.001
Treatment x Storage period	7.332	411.50	8	<0.001
Residual	0.018		51	

Table 6: The effect of the treatment and storage period interactions on the mean[1] percentage grain damage (± SE) and weight loss (± SE) caused by storage insect pest infestation

Treatment	Grain damage (%)			Weight loss (%)		
	Storage period (days)			Storage period (days)		
	90	180	Mean[2]	90	180	Mean[2]
SuperGrain II™ bag + CKPH08028	1.0 ± 0.1a	7.0 ± 0.4b	4.0b	0.2 ± 0.0a	0.9 ± 0.2cd	0.5bc
SuperGrain II™ bag + PH3253	1.2 ± 0.1a	7.7 ± 0.6b	4.5b	0.4 ± 0.1abc	0.9 ± 0.1d	0.6c
PICS bag + CKPH08028	1.1 ± 0.1a	1.6 ± 0.2a	1.3a	0.1 ± 0.0a	0.3 ± 0.1ab	0.2a
PICS bag + PH3253	1.7 ± 0.1a	1.5 ± 0.1a	1.6a	0.2 ± 0.0a	0.3 ± 0.0ab	0.2a
Smartbag -1 + CKPH08028	6.3 ± 0.3b	77.1 ± 5.1de	41.7de	0.9 ± 0.1d	22.0 ± 1.1f	11.5d
Smartbag -1 + PH3253	8.5 ± 0.3b	89.9 ± 0.7f	49.2f	1.3 ± 0.2d	28.8 ± 0.9h	15.0f
Polypropylene bag + CKPH08028	6.9 ± 0.6b	49.1 ± 1.5c	28.0c	1.1 ± 0.1d	17.4 ± 0.5e	9.2d
Polypropylene bag + PH3253	6.0 ± 0.3b	73.2 ± 0.2d	39.6d	0.6 ± 0.0bcd	23.9 ± 0.4fg	12.3de
Polypropylene bag +Actellic + PH3253	7.5 ± 0.3b	83.8 ± 1.2ef	45.6f	1.0 ± 0.1d	26.3 ± 0.7h	13.6ef
Mean[2]	4.5a	43.4b	24.0	0.6a	13.4b	7.0

[1]Means within the same column and row followed by the same letter are not significantly different at P =0.05 level
[2]Means of the main treatment effects followed by the same letter are not significantly different at P =0.05 level

4. Discussion

The temperature regime of the grain affects to a great extent the rate of metabolism, growth, development and insect population level (Girish 1965). Higher temperature results in greater insect activity. Temperature regime that favours most insect pests of stored grain is reported to be between 25°C and 30°C (Hayma 2003). In this study, the temperature in the bags environment was high enough to favour grain damage by the insects, their growth and reproduction.

It is evident that oxygen depletion and carbon dioxide evolution in the current study did not reach extreme levels. This could be attributed probably to bag perforation made by *P. truncatus* and insufficient respiration and metabolism of insects and grain itself. The decline in oxygen and increase in carbon dioxide levels is dependent on air-tightness of the storage structure, insect population, grain moisture content, grain quality and fungi load. Low grain moisture content and insect population in the absence of fungi lead to low oxygen demand in the bags (Moreno-Martinéz *et al*. 2000). Probably the dry grain in the present study coupled with low insect population and bag perforation prevented creation of depleted oxygen and enriched carbon dioxide environment. The survival of a few insects did seem to increase progeny production in grains stored in hermetic bags. Although there were significant differences in oxygen levels with hermetic treatments, the levels in SuperGrain II™ bag and Smartbag-I were sufficiently high to support the survival of *P. truncatus* and *S. zeamais*. Apart from Smartbag-I and polypropylene bag treated with Actellic dust, the rest of the treatments recorded more *S. zeamais* than *P. truncatus*. It is not clear whether *S. zeamais* has the capacity to switch to anaerobic metabolism as an adaptation to hypoxic environment. Extreme oxygen reduction attainment failure under hermetic conditions has been reported in clean grain stored without pre-storage insect and fungal attack (Moreno-Martinéz *et al*. 2000) and in maize grain infested with or without *P. truncatus* (Njoroge *et al*. 2014). In Njoroge *et al*. (2014) work, the build up of carbon dioxide on average stabilised at 13.8% in grain stored in PICS bag over six months storage. Carbon dioxide averaged 5.2% for both SuperGrain II™ and PICS bags and 2.6% of Smartbag -1 bag in the current study.

Grain damage and weight loss of maize stored in of Smartbag -1 and polypropylene bags was economically significant at 180 days storage period. On average, grain damage and weight loss of 83.5% and 25.4% was recorded in Smartbag -1 whilst 61.1% and 20.7%, was from polypropylene bag. PICS bag maintained maize with no visible grain damage and weight loss over the entire storage duration. The finding of this study is in concordance with 0.3% weight loss reported by Hell et al. (2010) over same storage duration. Although extreme levels of oxygen and carbon dioxide were not attained, the low damage and weight loss levels observed indicated that the insects were inactive. Very low oxygen level has been shown to change fecundity and delay development without causing mortality of the insects while elevated carbon dioxide level in some insects may induce diapauses (Bailey and Banks, 1980). The study demonstrates that even with infested grain before storage, PICS and SuperGrain II™ bags would achieve satisfactory protection against damage and weight loss. On termination of the study, Smartbag -1 and polypropylene bags and maize stored therein were heavily holed/damaged.

Admixture of maize grain with Actellic super dust and storage in polypropylene bag did not prevent infliction of damage and weight due to insect pests. Loss of grain protectants' biological activity depends on factors such ambient temperature. Higher temperatures generally lead to greater rates of decay of protectant efficacy (Athanassiou *et al*., 2008). The influence of higher safe level of temperature is shown by reduced efficacy (Samson *et al*., 1988). Whereas the insecticidal potency of Actellic Super dust® has been shown to reduce with time (Denloye *et al*., 2008), the inadequate control of *P. truncatus* and *S. zeamais* could not be attributed to the ambient temperatures. Dilute contact insecticide such as Actellic Super dust is usually admixed with grains as a preventive strategy on the storage day. However, even with proper admixture a portion of grains remains uncovered resulting in partial treatment (Vassilakos and Athanassiou *et al*., 2012). Whereas *S. zeamais* would be controlled when exposed to partially treated grain, *P. truncatus* exhibit some degree of tolerance. This may explain the high number of *P. truncatus* recorded in the current study. Although the two species occur together in storage, it has been predicted that *P. truncatus* dominates the interaction with *S. zeamais* at temperatures greater than 28°C (Howard 1983). *P. truncatus* competitive performance is higher in the presence of flour, which is generated by its feeding activities, mixed with whole grain. Since the average temperature in the hermetic bags was above 30°C (Figure 1), it is likely that *P. truncatus* outcompeted *S. zeamais* at such temperature. This observation confirms the prediction by Howard (1983).

While smallholder famers rely greatly on maize production and marketing as source of food supply and income, use of hermetic storage and insect resistant maize technologies would play an important role in ensuring constant supply of the staple food grain in many households. The current study examined the combined effect of hermetic bag and insect resistant maize to establish if some benefits would be gained in grain protection against *P. truncatus* and *S. zeamais* in stored maize. Grain weight loss is the most important criterion used to classify maize into either resistant or susceptible varieties (Derera *et al.*, 2014; Mwololo *et al.*, 2012). The treatment combination showed that storing either resistant (CKPH08028) or susceptible maize (PH3253) in hermetic PICS or SuperGrain II™ bags under same conditions had similar protection as indicated by the percentage grain weight loss except for Smartbag -1 and polypropylene bags after 180 days of storage. Although the oxygen levels were sufficiently high in both SuperGrain II™ and Smartbag -1 bags to support feeding activities of the test insects that result in weight loss, added protective value differences were only observed when either resistant maize grains (22% weight loss) were stored in the Smartbag -1 bags compared to the susceptibe maize (28.8% weight loss). In the laboratory study, the resistant hybrid maize variety (CKPH08028) recorded 25.4% weight loss under non - hermetic conditions (Mwololo *et al.*, 2012). The current work which simulated field conditions demonstrated that no benefits were gained when either resistant or susceptible maize grains were stored in either PICS or SuperGrain II™ bags. Nevertheless, at 180 days of storage period, CKPH08028 grain stored in PICS bags performed better than SuperGrain II™ bags. Synergistic benefits in protection were gained when the weight loss of CKPH08028 grains stored in either PICS or SuperGrain II™ bags were compared to that of the same variety stored in the polypropylene bags. The same maize variety when stored in Smartbag -1 performed poorly compared to either PICS or SuperGrain II™ bags. This could be attributed to the heavy perforation of Smartbag -1 bags by *P. truncatus* and as a result sufficiently low oxygen and high carbon dioxide was not achieved. The novelty of the work is demonstrated in the potential use of hermetic bags in combination with insect resistant maize technologies to significantly reduce weight loss from 30% to less than 1% without use of pesticide. This would improve food security at household level. The findings of this study would support agricultural policy formulation and monitoring of loss reduction activities.

5. Conclusion

Enriched carbon dioxide environment was not attained in the hermetic bags. Carbon dioxide averaged 5.2% for both PICS and SuperGrain II™ bags and 2.6% of Smartbag -1 bag. PICS and SuperGrain II™ bags maintained low damage and weight loss levels. No grain protection benefits were gained when either insect resistant (CKPH08028) or susceptible (PH5253) maize grains were stored in either PICS or SuperGrain II™ bags. Synergistic benefits in protection were gained when the weight loss of CKPH08028 grains stored in either PICS or SuperGrain II™ bags were compared to that of the same variety stored in the polypropylene bags. Admixture of maize grain with Actellic super dust and storage in polypropylene bag did not prevent infliction of damage and weight due to insect pests. The study demonstrated the potential in the use of hermetic bags in combination with insect resistant maize technologies to significantly reduce weight loss from 30% to less than 1% without use of pesticide.

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