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Reducing grain storage losses in developing countries

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RESEARCH ARTICLE

Abstract

We investigated the use of insecticide-treated material and modified atmosphere storage for reducing insect damage in stored maize. Results showed that insecticide-treated netting and insecticide-treated seed bags protected grain from insect damage, with the treated seed bags protecting grain for up to nine months if the grain was free from insects before storing. Covering the opening of grain storage containers with treated netting provided good control of stored-grain insects and may offer additional protection for grain silos being promoted in developing countries. Neither treated bags nor netting provided control if the grain contained insects before storage, thus emphasising the need to provide some means of controlling initial infestations at the time of storage when using bags or netting. We also showed that good control of insects can be achieved if the grain is stored in air-tight rigid containers that cannot be penetrated by insects or rodents. Hermetic plastic bags were readily penetrated by insects and rodents, thus negating their ability to protect grain from insects. We also showed that reducing oxygen levels through the use of a candle in stored environments does little to protect grain from insects. Modifying the atmosphere by reducing oxygen levels or raising carbon dioxide levels also provided good control of stored-grain pests. The solutions outlined in this paper offer farmers in developing countries methods to protect grain from stored-grain insects, thus reducing losses and improving food security.

Keywords: food security, subsistence farmers, hermetic, grains, storage

1. Introduction

Stored grain is susceptible to damage and consumption by insects such as the lesser grain borer (*Rhyzopertha dominica*), maize weevil (*Sitophilus zeamais*), and red flour beetle (*Tribolium castaneum*). Damage by insects in stored grain can be reduced or controlled using techniques such as pre-storage sanitation, applying insecticides, aerating to reduce grain temperatures, adding diatomaceous earth or ash, creating modified atmosphere conditions, and storing in insecticide-treated seed bags (De Groote *et al.*, 2013; Harien and Sbramanyam, 1990; Murdock *et al.*, 2003; Stathers *et al.*, 2007; Tefera *et al.*, 2011a). These intensive management practices can reduce grain losses to less than 1% for one year of storage or longer. If the insects are not controlled, then losses can be 20-80% within a few months after harvest (APHLIS, 2011; Cao *et al.*, 2002). Applying pesticides is practiced in many areas, but adulterated products or misapplications are common (Litsinger, 2005). Subsistence farmers store 50-75% of grain in developing countries on-farm, but often lack the resources to reduce losses caused by insects (Baloch, 1999). This decreases food security and contributes to malnutrition in these countries that struggle to provide adequate food for their nutritional needs.

In an attempt to develop solutions for subsistence farmers in developing countries, various researchers have studied means of reducing insect losses in stored grain using materials that may be sourced locally. Modified atmosphere environments can be created by using common containers that are relatively impermeable to air, such as metal or plastic barrels, or thick plastic bags. If the respiration rate of the grain and insects exceeds the permeability rate of the container, then insects will eventually stop feeding and die (Quezada *et al.*, 2006). Metal or thick plastic containers are an effective means of creating a hermetic storage environment for stored grain (De Groote *et al.*, 2013). Using thick plastic bags, or multiple bags, has proved effective for controlling various stored-grain pests in various commodities (Murdock *et al.*, 2003; Sanon *et al.*, 2011).

Modified atmospheres can be created by introducing gases such as carbon dioxide (CO_2) or reducing oxygen (O_2) levels. Navarro *et al.* (1985) showed that O_2 levels below 10% and CO_2 levels above 30% killed the grain mite *Acarus siro* L. White *et al.* (1995) showed that CO_2 levels above about 17% resulted in high mortality of *Tribolium castaneum* (Herbst), *Cryptolestes pusillus* (Schonherr) and *Cryptolestes ferrugineus* (Stephens) while CO_2 levels above 7.5% resulted in reduced insect activity. Chi *et al.* (2011) showed that feeding activity of cowpea bruchids (*Callosobruchus maculatus*) ceased at about 5% O_2 and 16% CO_2. Bailey (1954) showed that O_2 of 2% or CO_2 of 40% resulted in 100% mortality of *Calandra granaria* L. Since CO_2 can readily be obtained from composting materials, modifying storage atmosphere by introducing CO_2 may be a viable solution to controlling insects in storage in developing countries.

Treated seed bags can be made from insecticide-treated bed nets that are used to control mosquitoes in malaria endemic areas (Dowell and Dowell, 2012), or bags created by impregnating chemicals into the bag such as those produced by Vestergaard-Frandsen (http://www.vestergaard.com/our-products/zerofly). Since treated bed nets are widely distributed in many developing countries, these may offer a means of reducing grain losses during storage in those areas.

The objective of this research was to compare the effectiveness of modifying the storage atmosphere and using insecticide-treated materials as a means for reducing insect damage in stored grain in developing counties.

2. Materials and methods

We studied several different methods for reducing damage to stored maize (*Zea mays)* that may be appropriate for developing countries. Specifically, we studied different methods of modifying the storage atmosphere to reduce insect activity and methods of using treated materials to kill insects in storage.

Hermetic storage

We tested different means of creating hermetic conditions where the respiration of the insects and grain would exceed the permeability rate of the storage medium and thus reduce oxygen to levels where insects could not feed or live. We tested metal, plastic, and glass containers that are relatively impermeable to oxygen flow and commercial storage bags, such as the three-layer plastic bag produced by GrainPro (0.078 mm total thickness, Concord, MA, USA), common plastic freezer bags manufactured by Ziploc (0.07 mm, Racine, WI, USA), and common trash bags manufactured by Glad (0.03 mm, Oakland, CA, USA). The permeability rates for the common plastic bags and bottles are given by Massey (2003) and GrainPro lists the permeability rate of their bag (Villers *et al.*, 2010). All other permeability rates for metal, glass, and ridged plastic containers were assumed to be zero. We attempted to include the Purdue Improved Crop Storage (PICS) triple bags (Murdock *et al.*, 2003), but they were not available in the USA at the time of these tests. The permeability rates for all methods tested are listed in Table 1. Controls consisted of samples exposed to ambient air. Insect-damaged kernels and insect survival were recorded at the conclusion of the storage period. Results were analysed using the Fisher's least-significant-difference (LSD) test.

All treatments contained about 400 g of maize that was free from insects and damaged kernels, except for the multilayer bags which contained 2,000 g of maize. About 50 adult lesser grain borers were added to each treatment, except that about 250 insects were added to the multilayer bags. The lesser grain borer was selected for all tests since it is common pest in the USA and since it is similar to the larger grain borer (*Prostephanus truncatus*) which is one of the most devastating stored-grain pests in many developing countries (Tefera *et al.*, 2011b). Each treatment was replicated three times. Samples were maintained at ~26 °C and 65% relative humidity for about three weeks.

For the clear bags and glass jars, we used a NeoFox Measurement System (Ocean Optics, Dunedin, FL, USA) so that O_2 levels could be observed over time. The NeoFox system uses LED excitation and a photodiode detector that measure O_2 levels based on the interaction with a fluorescing phase detection sensor placed inside the clear bag. The sensor is scanned through bag using a fibre-optic probe.

Table 1. Storage methods used in modified atmosphere tests.

Storage method	Permeability rate (cc/m^2/day)
Glass jars	0[a]
Metal	0[a]
Plastic bottles	4[b]
GrainPro bags	3[c]
Ziploc bags	1,340[b]
Trash bags	2,300[b]

[a] Assumed zero permeability rate.
[b] Massey (2003).
[c] Villers *et al.* (2010).

Modified carbon dioxide and oxygen levels

Since some research shows that increasing CO_2 can reduce insect activities, we studied the effect of increasing CO_2 levels in stored samples by using CO_2 from composting material. CO_2 was produced from about 1000 g of mouldy, high-moisture maize, extracted with a syringe, and inserted into a container with ~100 g maize samples with about 10 adult maize weevils and 50 adult lesser grain borers. Mouldy samples were used since CO_2 could readily be produced in developing countries from decomposing organic material. The four ~100 g samples were prepared by flushing samples with CO_2 multiple times to achieve the desired CO_2 levels. A fifth sample was prepared but left in ambient conditions. After flushing, the final O_2 levels in the containers ranged from 3.7 to 21%, and CO_2 levels ranged from 1 to 29%. The O_2 and CO_2 levels were measured five times over ten days using a Quantek Instruments (Grafton, MA, USA) Model 902D Dual Track O_2/CO_2 sensor by inserting the sampling probe into each container using a hypodermic needle attached to the sampling line. Containers were sampled through an air-tight orifice. The number of dead insects, live insects, and damaged kernels were counted at the end of the tests. The presence of moving insects was noted at each sampling.

Treated netting

For initial tests, 80×80 cm square sections were cut from treated bed nets obtained from Olyset Net (Sumitomo Chemical, London, U.K., permethrin, 1000 mg/m^2), Netprotect (Castelnau-le-Lez, France, deltamethrin, 65 mg/m^2), and PermaNet (Vestergaard-Frandsen, Lausanne, Switzerland, 55 mg/m^2 deltamethrin). About 0.7 kg of uninfested, undamaged whole maize was placed on each cut section. Then the bed net was folded around the grain and fastened with a rubber band (Figure 1). This was replicated three times and placed at about 27 °C and ambient humidity for a total of nine treated bed net samples. Six replicates of untreated netting (Siamdutch Mosquito Netting Co., Ltd., Bangkok, Thailand) were also prepared. Several hundred adult lesser grain borers were placed in the vicinity of the treatments and given free choice to select bags to infest. The number of live insects and damaged kernels were determined after 30 days.

In subsequent tests, 14 round plastic containers with a capacity of about 2,500 cc were prepared by removing the tops and bottoms, and replacing with treated or untreated netting (Figure 1). In the netting tests above, samples were placed inside the bags made from netting, and thus the sample size is limited by the strength of the netting. The purpose of this test using containers was to see if larger containers could be used to protect grain from insects if only the openings to the container were covered with treated netting. Six containers were covered with deltamethrin-treated netting (Netprotect or PermaNet), and eight containers were covered with untreated netting. Each container was filled with about 1 kg of uninfested, undamaged whole maize (Figure 1). A treated and untreated container was placed in a larger plastic tub along with several hundred adult lesser grain borers that were given free choice to infest bags. This was repeated for a total of six treated and six untreated containers. Two untreated containers were placed together in a seventh tub with lesser grain borers to serve as an untreated control. Samples were

Figure 1. Grain storage bag fabricated from treated bednets and plastic containers with tops and bottoms replaced with insecticide-treated or untreated netting.

placed in an environmental chamber at 23 °C and ambient humidity for one month. The number of live insects and damaged kernels was evaluated at that time.

Treated bags

Based on positive results from tests using insecticide-treated netting, polyethylene grain bags with deltamethrin incorporated into the fabric were obtained from Vestergaard-Frandsen. For these tests, nine small bags were constructed by cutting ~80×80 cm squares from the larger treated bags, folding, and sewing 2 sides to create a sample bag (Figure 2). Similarly, 11 additional bags were fabricated from similar untreated material. Each bag was filled with 500 g of uninfested, undamaged maize. Six treated and eight untreated bag replicates contained only uninfested, undamaged maize. To determine if the treated bags can provide control of infested maize, about ten each of adult maize weevils, lesser grain borers, and red flour beetles were placed inside each of the remaining three treated bags and three untreated bags. All bags were placed in chambers at 27.5 °C and 65% relative humidity. Then about 100 each of adult lesser grain borers, rice weevils, and red flour beetles were placed in the vicinity of the bags and given free choice to infest treated or untreated bags. The number of damaged kernels and presence of live insects was recorded after about 1, 3, 5, 9, 13 and 16 months of storage. The recording intervals were selected so that we could track insect population growth and to represent typical storage periods. Damaged kernels are reported as the number of kernels per 100 g.

3. Results and discussion

Hermetic storage

This type of storage attempts to reduce O_2 levels or raise CO_2 levels to a point where insects are not active. Since grain and insects respire, the respiration rate of the grain and insects must be greater than the permeability rate of the storage container. For metal and plastic containers, or multi-layer plastic bags, the permeability rate is about zero. Insects can feed and remain active at oxygen levels as low as about 5% (Emekci *et al.*, 2001; Margam, 2009). Thus, oxygen levels must be reduced to very low levels to prevent damage to stored crops. For our tests using metal, plastic, and glass containers, which all had a ~zero permeability rate, there were no live insects after about 3 weeks. However, some of these containers had up to 5% damaged kernels (Figure 3). Thus, some damage occurred before oxygen levels were depleted to levels low enough to kill insects or stop their feeding activity. The metal, plastic and glass containers, and the GrainPro and plastic trash bags all had damage levels that were significantly (P<0.05) less than the infested controls. The metal, plastic and glass containers, and the GrainPro plastic bag all had damaged kernels that were not significantly larger than the uninfested control.

The Ziploc and trash bags all had live insects and as many as 9% damaged kernels. Thus, permeability rate of the bags was not low enough to restrict oxygen levels to a point of controlling insects. The GrainPro bags had 4% damaged kernels and most of the insects were alive. Observations in other tests showed that insects and rodents could penetrate these GrainPro bags (Figure 4). De Groote *et al.* (2013) reported that all GrainPro bags tested in their study in Kenya were perforated, probably by *P. truncatus*. We also experienced holes in bags where rodents had destroyed bags in field tests in Zimbabwe (data not shown). All insects in the controls were alive, with an average of 11.7% damaged kernels.

Bern *et al.* (2013) summarise hermetic storage systems and their costs and useful life. The cost of GrainPro bags is about $ 0.05/kg, and the metal silo, which is conceptually similar to the metal container we tested, costs about $ 0.10/kg. The useful life of the bags is 2-5 years, whereas the metal

Figure 2. Grain storage bags fabricated from insecticide-treated or untreated material.

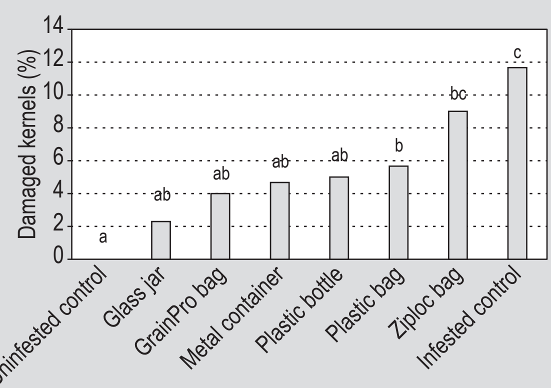

Figure 3. Insect-damaged maize kernels in various types of storage containers. Bars with the same letter are not statistically different at *P*<0.05.

Figure 4. Holes in SuperGrain (GrainPro, Inc.) hermetic storage bags created by insects. Note the insects inside the bag feeding on the grain.

silo can last over 20 years. Thus, the storage cost per kg/yr for the metal silos is much lower than the bag alternatives.

Although the plastic bags had some insect damage, if multiple plastic bags are used then the permeability rate could be reduced to levels where insects cannot survive. This is the concept used in the PICS bags (Murdock *et al.*, 2003) that have been used in several developing countries. While these may provide a low-cost alternative to other storage methods, they are still susceptible to damage by rodents and would be most effective if placed inside rodent-proof containers such as 200 l metal drums or polypropylene tanks.

Bern *et al.* (2013) also reported the use of recycled plastic containers such as those used to store edible oil, which is conceptually similar to the plastic container we tested. These are readily available in developing countries at minimal cost. They showed 100% control of weevils when using these recycled containers when they were properly sealed.

In tests where we monitored O_2 levels in infested and uninfested grain, we showed that in the uninfested hermetic control the O_2 level dropped from 21.9 down to 10% due to the respiration rate of the maize. Thus, the maize consumed the O_2 in the GrainPro bag faster than the air permeated into the bag. The O_2 level in the infested grain decreased to 1.7% in about 3 days causing insects to die. However, after the insects died the O_2 increased to about 5% due to the permeability of the bag. Thus, since insects can penetrate the bag, reinfestation can occur.

We had included using a candle to consume oxygen in closed containers based on recommendations by groups such as CIMMYT (www.cimmyt.org, The Metal Silo) and other research (De Groote *et al.*, 2013; Tefera *et al.*, 2011a). That literature states that a lighted candle inside a sealed container will consume all oxygen and thus created a hermetic storage environment. However, our results using the NeoFox measurement system did not show reductions in O_2 content below a few percentage points when a candle was used in a closed container (data not shown). A further review of literature shows that a flame cannot be sustained at oxygen levels below about 15% (Belcher and McElwain, 2008; Wildman *et al.*, 2004). Thus, addition of a candle to a closed container does little to create conditions that affect insect activity. Unfortunately, organisations and individuals continue to promote the false idea that using a candle in closed containers reduces insect activity.

Modified carbon dioxide and oxygen levels

We used CO_2 produced from fermenting maize to raise CO_2 levels in stored maize infested with insects. Results showed that damaged kernels decreased as O_2 levels decreased, and CO_2 levels increased from ambient levels of 20.9% O_2 and 0.039% CO_2 (Figure 5). We had very few damaged kernels at levels above 16% CO_2 or below 4% O_2. Thus reduced O_2 levels and increased CO_2 levels from ambient is effective at reducing insect-damaged kernels in storage. Our results agree with literature where White *et al.* (1995) showed that CO_2 levels above about 17% will cause high mortality in stored-grain pests, while CO_2 levels above 7.5% reduced insect activity. Emekci *et al.* (2001) showed that insect activity doesn't change much until O_2 levels reduce to less than about 5%. Thus, much of the control in our study may be due to the increase in CO_2 levels. Margam (2009) varied CO_2 and O_2 inversely so that the percentage always equalled 20% and showed a linear decrease in feeding activity as CO_2 increased and O_2 decreased, and that feeding stopped at about 6% O_2 and 14% CO_2, further confirming our results.

While the effect of CO_2 and O_2 levels in storage have been reported by others, this is the first report of using CO_2 obtained from composting material as a source for CO_2 to modify the grain storage atmosphere. Since composting is a recommended method (www.foundationsforfarming.org) for providing soil nutrients for subsistence farmers that are the target of these grain storage concepts, harvesting CO_2 from composting materials may fit in well with recommended farming practices. One could conceive composting material in a closed large plastic container, such as a large polypropylene water tank and transferring CO_2 to a closed grain storage container using a modified bicycle pump that can transfer air from one container to the other.

Treated netting

In initial tests to evaluate the concept of storing grain in insecticide-treated bed nets, no insects and only two damaged kernels were found in any of the replicates that utilised the treated netting after 30 days of storage. Many

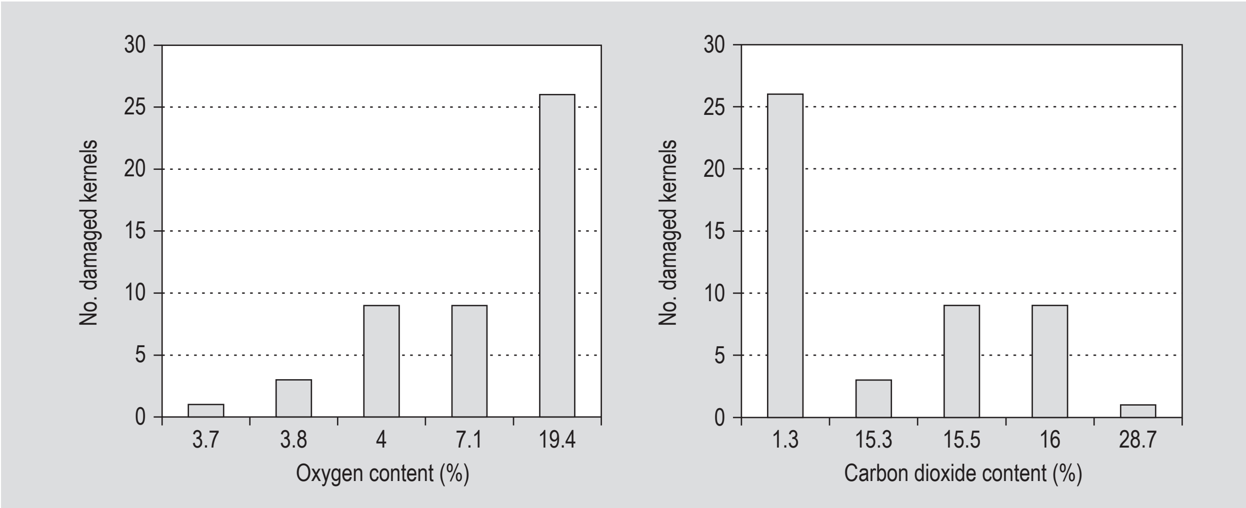

Figure 5. Insect-damaged kernels versus oxygen or carbon dioxide content after 10 days of storage.

dead insects were found around the treated netting, indicating that the treated netting is effective at preventing lesser grain borers from entering stored grain. In the six untreated controls, there were an average of 19 damaged maize kernels per 100 g and 17 live insects in each sample after the one month of storage, which was significantly more ($P<0.01$) than in the samples protected by treated bed nets. Thus, these tests indicate that commercial treated bed nets are effective at protecting grain from lesser grain borers and justified further testing.

Since treated bed nets are not manufactured with sufficient tensile strength to hold grain, we studied the concept of storing grain in larger containers and only covering the openings with bed nets. Results from these tests showed that there were essentially no live insects or damaged kernels in the containers covered by treated netting after one month of storage (Figure 6). However, containers covered with untreated netting had high levels of lesser grain borers and significantly more ($P=0.07$) damaged kernels than grain protected by treated netting. Thus, these tests show that if uninfested grain is placed in a container and all openings are covered with treated netting, then grain should remain insect-free.

An application of these findings could be to cover openings in larger storage containers, such as metal silos with treated netting. These silos have become popular in some developing countries (De Groote *et al.*, 2013). If the openings are not adequately sealed, then insects can enter through the broken seals. Covering the opening with treated netting could help keep reduce re-infestation of grain stored in these systems.

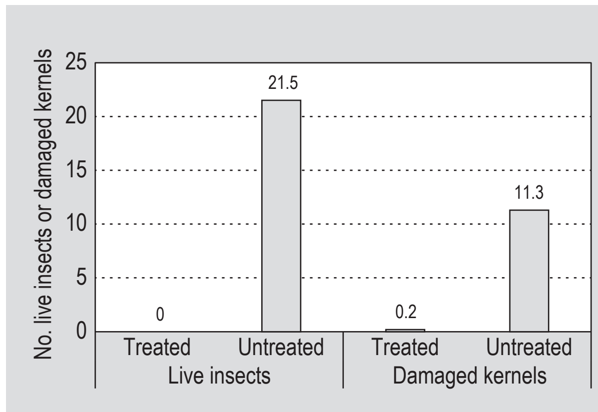

Figure 6. Mean number of live insects and damaged kernels stored for 1 month in containers whose openings were covered with untreated or insecticide-treated bed nets.

Treated bags

Since treated netting was effective at protecting grain against stored-grain pests, we studied the effectiveness of the Vestergaard-Frandsen grain storage bags manufactured using the same chemicals as used with treated bed nets. Figure 7 show that at up to nine months of storage, treated bags that were exposed to external infestations had no damaged kernels and very few live weevils. However, the samples internally infested in untreated bags had over 400 damaged kernels and over 650 live insects after 5 months of storage. Due to the very large number of insects at 5 months, these untreated bags were not sampled in subsequent months. Those samples that were internally infested in treated bags had about 300 damaged kernels and over 500 live maize weevils after 9 months of storage. The number of damaged kernels increased in all samples after 9 months of storage. The number of damaged kernels and

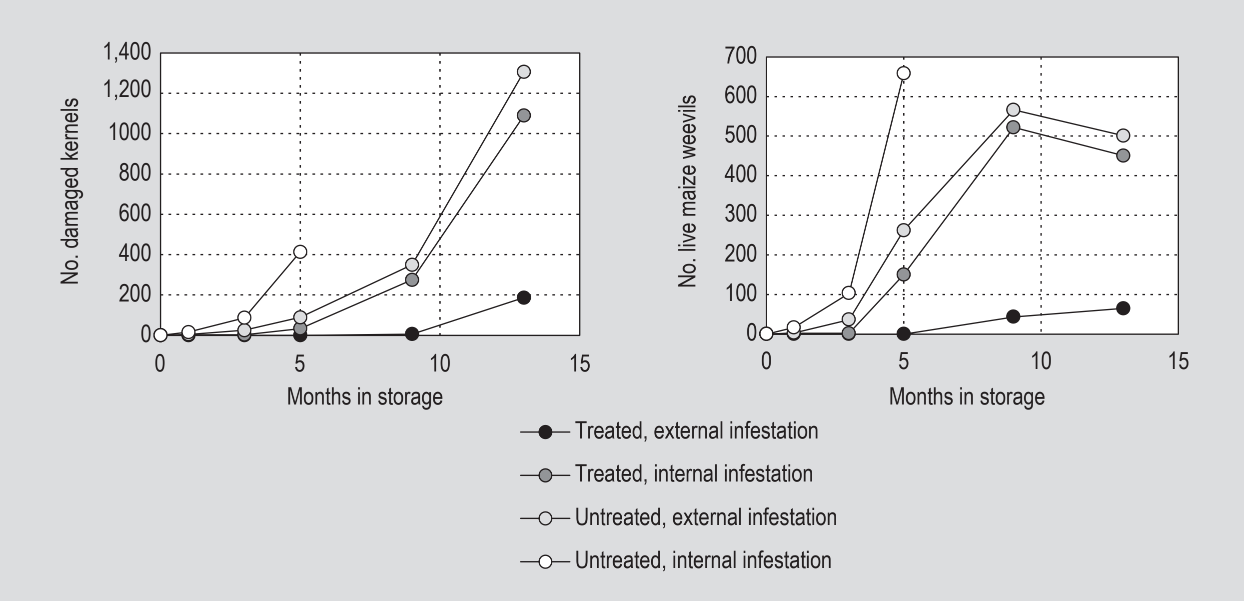

Figure 7. Number of insect-damaged kernels (left) and live maize weevils (right) in 500 g of maize in insecticide-treated and untreated seed bags during storage.

live maize weevils was significantly less ($P<0.05$) in treated bags subject to external insect infestation than untreated bags after 5 months. These results agree with field results reported by Vestergaard-Frandsen where they showed few maize weevils and few damaged kernels after 5 to 12 months of maize storage in Zambia, Ghana, and Senegal (http://www.vestergaard.com/our-products/zerofly).

It was noted that within one day of placing samples inside the chambers that there were many dead insects around the treated bags but not around the untreated bags. Thus, insects attempting to penetrate the treated bags died upon contact with the bags. Untreated bags had many insects crawling on the bags after one day of storage. It may be that if treated bags had a small hole such as at a seam, then insects may eventually entered the bags without contacting the treated surface. Thus, bag integrity must be maintained for this type of storage to provide lasting protection.

Treated bags that were initially internally infested had no significant difference ($P=0.84$) in damaged kernels and weevils when compared to untreated bags (Figure 7). Lesser grain borer and red flour beetle populations were much lower when compared to maize weevils, and were not present in many treated or untreated bags after a few months of storage. Damaged kernels and live insects increased sharply after 5 months of storage in the untreated bags and internally infested bags. Untreated bags that were initially infested internally had many damaged kernels and live insects after only 3 months of storage. Thus, treated bags provided little advantage over untreated bags if there were existing infestations at the time of storage. Thus, the best insect control was when uninfested grain was stored in treated bags. These results show that if treated bags are to be used, then samples should be fumigated at the time of storage if any existing infestation is suspected.

4. Conclusions

Results showed that seed bags protected grain from insect damage for up to nine months if the grain was free from insects before storing. Treated netting protected the grain for the one month storage test. Treated bags or netting did not provide control if the grain contained insects before storage, thus necessitating the need to fumigate grain at the time of storage. Good protection was achieved if the grain was placed in a bag incorporated with insecticide, or in a storage container where any opening is covered with the insecticide-treated material. We also showed that good control of insects can be achieved if grain is stored in air-tight, rigid containers that cannot be penetrated by insects or rodents. Modifying the atmosphere by reducing O_2 levels or raising CO_2 levels also provided good control in stored-grain pests. The solutions outlined in this paper offer farmers in developing countries methods to protect grain from stored-grain insects, thus reducing losses and improving food security.

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Disclaimers: Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture. USDA is an equal opportunity provider and employer.

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