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EVALUATION OF SILO BAGS FOR TEMPORARY STORAGE OF WHEAT

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ABSTRACT

Trials were conducted to evaluate the effectiveness of silo bags for temporary storage of hard red winter wheat under field conditions in Kansas, USA. Newly-harvested wheat with moisture content of 11.6 ± 0.2% was stored in four silo bags, each of 50 metric tonne capacity for a period of four months starting August 24 to December 14, 2010. The dimensions of the silo bags were approximately 20.0 m long and 3.0 m in diameter. The average temperature of the top and bottom layers of grain in bags was 32.3 and 26.7°C, respectively, in August. In December, average temperature of the top layer was -3.1°C and the temperature of the bottom layer was 13.0°C. During the study period, relative humidity of the top and bottom layers of grain fluctuated between 46.7 to 55.9%. The carbon dioxide concentration ranged from 0.53 ± 0.05% to 1.45 ± 0.25% during the study period. The analysis of wheat quality at the beginning and at end of the storage period clearly indicated that the parameters such as moisture content, test weight, falling number, and kernel weight were unaffected during storage in silo bags. The fungal concentration and mycotoxin levels did not change significantly during storage. The 30 adults of lesser grain borer, *Rhyzopertha dominica* (F.) (Coleoptera: Bostrichidae), confined in PVC cylinders and placed in silo bags in August increased by approximately 10 fold at the end of the storage period. The population growth of *R. dominica* in PVC cylinders in silo bags was 14 to 19 times lower when compared with growth in PVC cylinders placed in a laboratory growth chamber at 28°C and 65% r.h. Despite low carbon dioxide levels, silo bags can be used for temporary wheat storage when there is limited storage space during years of over production.

Keywords: Silo bags, modified atmospheres, wheat, fungi, mycotoxins, wheat quality, *Rhyzopertha dominica*

INTRODUCTION

Wheat in the United States is typically stored for six to nine months on farms (Martin et al., 1977). In years of bumper harvest, wheat storage on farm and at grain elevators is a huge challenge and this has forced farmers to sell their crops at a lower price which usually prevails

at the harvest time. Under such situations the low wheat prices have caused an increased demand for more storage capacity. Producers and elevator managers will overfill their bins/silos and are forced to set up temporary storage structures. Overfilling bins/silos makes grain quality management difficult. There is an urgent need to conduct research on alternative grain storage systems and evaluate their effectiveness in maintaining stored-grain quality, and their efficacy in minimizing fungal and stored-product insect problems. The use of silo bags for storing grain has been adopted successfully in recent years in Latin American countries, especially Argentina (Abalone et al., 2011a). In these relatively airtight silo bags, the elevated carbon dioxide (CO_2) and decreased oxygen (O_2) may create modified atmospheres detrimental to insects and fungi (Fleurat-Lessard, 1990; Adler et al., 2000; Navarro, 2006). More recently, silo bags are being used in the USA and Canada, primarily for temporary storage of grain. In 2010 we cooperated with a Kansas elevator operator using silo bags for storage of wheat. Our objectives were to determine environmental conditions inside silo bags holding wheat, and to evaluate impact of short-term storage of wheat (4 months) on wheat quality parameters, fungal infection, and effects on the lesser grain borer, *Rhyzopertha dominica* (F.) (Bostrichidae: Coleoptera) populations.

MATERIALS AND METHODS

Hard red winter wheat (11.6 ± 0.2% moisture content) harvested in the 2010 crop year was stored in four silo bags, each of 50 tonne capacity, for a period of four months starting August 24 to December 14, 2010 in Manhattan, Kansas. The dimensions of the silo bags were 20 m long and 3 m in diameter. Grain temperature and relative humidity inside the silo bags were recorded using HOBO® data logging units (Onset Computer Corp., Bourne, Massachusetts, USA). Two data loggers were used at each of three sampling points within each bag. At each point a logger was placed at the top (dorsal) side of the bag and at the bottom (1 to 1.5 m depth) of the silo bag. Levels of CO_2 in each bag were monitored monthly using a CO_2 detection device (SilCheck Company, Buenos Aires, Argentina) at each of the three sampling points.

Grain samples were collected at the beginning of storage (August 24), on October 23 and December 14, 2010. Grain samples were collected at each of three sample points using a 1.52 m long grain probe at the top (0 to 0.5 m), middle (0.5 to 1.0 m) and bottom (1.0 to 1.5 m) grain layers. At the top, middle, or bottom locations, the probe sample collected 160 g. In each bag, all top samples were pooled, as were middle and bottom samples. Wheat quality parameters measured included test weight, protein content, falling number, kernel weight, kernel diameter, and kernel moisture. Kernel weight, diameter, and moisture from 300 kernels were determined using Perten's single kernel characterization system, SKCS 4100 (Perten Instruments, Hägersten, Sweden). All wheat quality evaluations (except mycotoxins) were performed at the Wheat Quality Laboratory in the Department of Grain Science and Industry, Kansas State University, using official approved methods.

Samples were also analyzed for total fungal counts (cfu/g) using Dichloran-Glycerol (DG 18) agar media (Oxoid Limited, Hampshire, United Kingdom). The levels of aflatoxin, fumonisin, and vomitoxin in wheat samples were quantified using the AOAC (Association of Analytical Chemists, Gaithersburg, Maryland, USA) approved method based on an Enzyme Linked Immmunosorbent Assay (ELISA) kit (AgraQuant® mycotoxin ELISA test kits, Romer Labs Inc., Union, Missouri, USA). In order to assess how insect populations would develop inside silo bags, PVC cylinders of 0.3 m long and 9 cm in diameter were used in only two out of the four bags. One end of the cylinder was glued with a 200 µm heavy duty mesh. We used

R. dominica as the test species as it is an economically important pest insect associated with wheat in Kansas. Organic hard red winter wheat (Heartland Mills, Marienthal, Kansas, USA) was used to fill 80% of each cylinder (890 g), and 30 unsexed adults of mixed ages, reared at 28°C and 65% r.h. on the same wheat, were added to each cylinder. After adult introduction, the open end of the cylinder was glued with the mesh. Into each silo bag at each of three sampling points two cylinders with insects (30 adults) were inserted by making an entry point in the bag, which was later sealed with duct tape. There were a total of six cylinders per silo bag. One cylinder was at the top (0 to 0.5 m depth) and the other at the bottom (1.0 to 1.5 m depth) at each of the three sampling points per bag. Three cylinders from each of the two bags were sampled on October 23, 2010, and the remaining three on December 14, 2010. Control treatments included nine cylinders with wheat and insects placed in a growth chamber at 28°C and 65% r.h., and sampled in August, October, and December. Wheat in cylinders was sifted and the number of adult progeny produced enumerated (live and dead for October and December samples).

The percent CO_2 data without transformation (because data were normal) by sampling location on bag and month were subjected to two-way analysis of variance (ANOVA) to determine significant differences ($P = 0.05$). Based on this analysis, data were pooled by sampling point and subjected to one-way ANOVA and means among months were separated by Bonferroni *t*-tests (SAS Institute 2005). The wheat quality parameters data (untransformed) were subjected to two-way ANOVA to determine significant differences by month and sampling depth. If wheat quality parameters were not influenced by sampling depth, data were pooled and differences among months were determined by one-way ANOVA and Bonferroni *t*-tests. Individual fungal counts and mycotoxin data were analyzed using two-way ANOVA to determine differences among months and sampling depth. Additionally at each month, differences among depths for fungal counts and each mycotoxin were determined by one-way ANOVA and Bonferroni *t*-tests. The insect data by month from each of the two bags and the control treatment were analyzed as one-way ANOVA followed by Ryan-Einot-Gabriel-Welsch (REGWQ) test for mean separation after transforming insect count data to logarithmic scale.

RESULTS AND DISCUSSION

The temperature of the top and bottom layers of grain in the bags averaged 32.3°C and 26.7°C, respectively, in August; in December it was -3.1 and 13.0°C, respectively. The average relative humidity of the top and bottom layers was 52.7 and 55.9%, respectively in August. In December the corresponding values were 46.6 and 51.7%. The CO_2 levels were significantly different among storage months ($F = 5.25$; df = 4, 45; $P = 0.0015$), but not among sampling points in the bags ($F = 1.03$; df = 2, 45; $P = 0.3670$). The interaction of months and sampling points was also not significant ($F = 0.72$; df = 8, 45; $P = 0.6744$), indicating that the differences in CO_2 levels among the three sampling points remained consistent across the storage months. Therefore, the sampling location data were pooled and the one-way ANOVA showed significant differences among months ($F = 4.28$; df = 4, 15; $P = 0.0166$). The CO_2 levels were similar between August and October, and dropped significantly in December (Table 1), perhaps due to the onset of cold weather. However, the levels observed (0.53 to 1.45%) were too low to exert any adverse effects on biological organisms. Newton (1991, 1993) reported that levels of CO_2 should be about 60% to effectively control stored product insects and mites. Levels of 10 to 30% of CO_2 can be toxic to insects provided the level of O_2 is 0.5 to 2.6% (Krishnamurthy et al., 1986).

Table 1. Carbon dioxide (CO_2) levels in silo bags

Month	Mean ± SE CO_2 level (%)[a]
Aug	0.98 ± 0.23ab
Sep	1.45 ± 0.25a
Oct	1.38 ± 0.11a
Nov	1.10 ± 0.17ab
Dec	0.53 ± 0.05b

[a] Means (n=4) followed by different letters are significantly different ($P < 0.05$).

The test weight of the wheat was not affected by storage time (Type III SS (1 missing value) $F = 1.66$; df = 2, 80; $P = 0.1957$) and sampling depth ($F = 0.03$; df = 8, 80; $P = 1.000$). The storage time and sampling depth interaction also was not significant ($F = 0.03$; df = 16, 80; $P = 1.000$). The protein content, falling number, kernel weight, kernel diameter, and kernel moisture were significantly different among storage times or months (F range = 4.45 to 285.04; df = 2, 81; $P \leq 0.0131$) but not at the sampling depths (F range = 0.14 to 0.81; df = 8, 80; $P \geq 0.5943$). The storage time (df = 8, 81) and sampling depth interaction (df = 16, 81) was also not significant (F range = 0.19 to 0.1.52; $P \geq 0.1635$). Since quality parameters were similar among sampling depths, data were pooled by depth, and differences among storage times for each of quality parameters (except for test weight) were analyzed using one-way ANOVA. However, one-way analysis showed only protein and moisture to be different among August, October, and December (Table 2). The lack of differences among storage times for falling number, kernel weight, and dry matter was perhaps a result of pooling data across sampling depths, which eliminated any variance due to sampling depths.

Table 2. Mean ± SE wheat quality parameters by storage month

Month	Test weight (kg/hl)[a]	Protein (%)[a]	Falling number (sec)[b]	Kernel weight (mg)[a]	Kernel diameter (mm)[a]	Kernel moisture (%)[b]
Aug	79.0 ± 1.8	12.6 ± 0.2	411.1 ± 8.5c	27.8 ± 0.4	2.6 ± 0.01	11.6 ± 0.2a
Oct	79.2 ± 0.3	13.1 ± 0.2	553.1 ± 10.3a	28.2 ± 0.3	2.6 ± 0.02	10.8 ± 0.4b
Dec	77.8 ± 2.1	12.9 ± 0.1	490.0 ± 5.3b	27.7 ± 0.2	2.6 ± 0.02	11.3 ± 0.1a

[a] Means (n=4) among months for each of the variables was not significant (F range = 0.2 to 1.96; df = 2, 9; $P > 0.1966$).
[b]Means (n=4) within months followed by different letters are significantly different ($P < 0.05$).

Fungal counts varied by storage time ($F = 13.76$; df = 2, 27; $P < 0.0001$) and by sampling depth within bags ($F = 119.10$; df = 2, 77; $P < 0.001$), but the interaction of sampling time and depth was not significant ($F = 0.19$; df = 4, 27; $P = 0.9409$). Therefore, differences among sampling depths and each month were determined. Results showed significant differences among the three depths during August, October, and December (F range = 33.0 to 55.5; df = 2, 9; $P < 0.0001$) (Table 3). In general, the fungal counts at 1.0 to 1.5 m depth were significantly higher than those at 0 to 0.5 m and 0.5 to 1.0 m depths.

Table 3. Mold propagules (cfu/g) by storage month and sampling depth within silo bags

Month	Sampling depth (m)	Mean ± SE cfu/g[a,b]
Aug	0-0.5	127.5 ± 4.3b
	0.5-1.0	145.0 ± 8.9b
	1.0-1.5	218.8 ± 10.9a
Oct	0-0.5	138.8 ± 4.3b
	0.5-1.0	162.5 ± 7.5b
	1.0-1.5	242.5 ± 13.8a
Dec	0-0.5	157.5 ± 2.5b
	0.5-1.0	173.8 ± 5.5b
	1.0-1.5	257.5 ± 12.5a

[a]cfu/g, colony forming units per gram.
[b]Means (n = 4) within months followed by different letters are significantly different ($P < 0.05$).

The aflatoxin level did not vary by storage time (F = 1.26; df = 2, 27; P = 0.2989) but varied by sampling depth (F = 11.23; df = 2, 27; P = 0.0003). The sampling time and sampling depth interaction was not significant (F = 0.47; df = 4, 27; P = 0.7590). In the case of fumonisins and vomitoxin, both sampling time and sampling depth were significant (F range = 3.74 to 46.48; df for both variables = 2, 27; $P \leq 0.0369$), but the interaction of sampling time and depth was not significant ($F > 0.36$; df = 4, 27; $P > 0.6952$). One-way ANOVA by storage month showed both the fumonisins and vomitoxin levels to be greater in December compared with August or October (Table 4).

Table 4. Mycotoxin (mean ± SE) levels by storage month and sampling depth within silo bags

Month	Sampling depth (m)	Total aflatoxins (ppb)	Fumonisins (ppm)[b,c]	Vomitoxin/DON (ppm)[b,c]
Aug[a]	0-0.5	0.50 ± 0.05	0.07 ± 0.02b	0.02 ± 0.02b
	0.5-1.0	0.20 ± 0.16	0.28 ± 0.06b	0.11 ± 0.05b
	1.0-1.5	0.56 ± 0.27	0.51 ± 0.11a	0.41 ± 0.02a
Oct[a]	0-0.5	0.10 ± 0.10	0.11 ± 0.51b	0.11 ± 0.05b
	0.5-1.0	0.25 ± 0.18	0.40 ± 0.07a	0.25 ± 0.07b
	1.0-1.5	0.73 ± 0.23	0.55 ± 0.09a	0.57 ± 0.06a
Dec	0-0.5	0.12 ± 0.19b	0.16 ± 0.05c	0.17 ± 0.07b
	0.5-1.0	0.30 ± 0.19b	0.47 ± 0.08b	0.31 ± 0.08b
	1.0-1.5	1.10 ± 0.30a	0.73 ± 0.08a	0.73 ± 0.07a

[a]Mean (n = 4) aflatoxin levels within months were not significant ($F > 1.99$; df = 2, 9; $P > 0.0835$; one-way ANOVA).
[b]Fumonisin and vomitoxin levels within months were significant (F range = 8.35 to 32.15; df = 2, 9; $P < 0.0089$; one-way ANOVA).
[c]For each month, means followed by different letters are significantly different ($P < 0.05$).

This trend was observed with afltaoxin only in December samples; samples from August and October had similar aflatoxin level, irrespective of the sampling depth. The increased level of fungal and mycotoxin contamination is difficult to explain because the

moisture of samples taken at all depths was essentially similar as determined in the laboratory from samples collected in the field (see Table 2). Wheat samples from the bottom contained slightly more broken kernels, dust, and infected kernels (visual observation) compared to top samples. Damaged and broken kernels are more likely to be contaminated by fungi and mycotoxins (Shotwell et al., 1985). Further, the samples at the bottom were close to the ground and understanding changes in ground temperature and humidity as they relate to temperature differences and moisture migration patterns within the silo bag may shed light on why the bottom samples had higher fungal counts and mycotoxin levels. During the storage period the silo bags were damaged by pests such as rodents. Additionally, the silo bags were extensively damaged by the cadelle beetle, *Tenebroides mauritanicus* L. Damage by these pests at the ground and silo bag interface may have resulted in moisture seeping into the bottom of the bag resulting in increased fungal and mycotoxin levels

The number of *R. dominica* adults within PVC cylinders showed a significant increase during October and December with each of the two bags and in the control treatment (F = 2006.20; df = 8, 18; $P < 0.001$) (Table 5). The 30 introduced adults in the control treatment at 28°C and 65% r.h. increased to 102 and 140 fold by October and December, respectively. However, PVC cylinders with insects inside the two silo bags showed only a 7 to 10 fold increase in *R. dominica* numbers in October and December. Therefore, *R. dominica* adults in PVC cylinder in silo bags were 14 to 19 times lower than those observed in cylinders placed at 28°C and 65% r.h. The reduced numbers found in silo bags was not due to the CO_2 levels observed but due to the onset of cooler weather during September, October, and November which may have prolonged the insect's development time.

Table 5. Adults of *R. dominica* in PVC cylinders in silo bags and in a growth chamber

Treatment	Month	Mean ± SE[a] numbers of adults/cylinder
Bag C	Aug	30.0 ± 0.0f
	Oct	198.0 ± 7.2e
	Dec	276.0 ± 25.2cd
Bag D	Aug	30.0 ± 0.0f
	Oct	226.7 ± 3.4de
	Dec	308.2 ± 13.6c
Chamber (28°C/65% r.h.)	Aug	30.0 ± 0.0f
	Oct	3066.0 ± 103.2b
	Dec	4206.7 ± 173.0a

[a]Means (n=3) followed by different letters are significantly different ($P < 0.05$).

Levels of CO_2 in interstitial spaces of wheat stored in silo bags are influenced by changes in grain temperature, moisture, and storage time (Abalone et al., 2011a). For wheat at 12 to 13% moisture, the level of interstitial O_2 was more than 12% and that of CO_2 was less than 7% after six months of storage (Abalone et al., 2011b). Our four month study showed that silo bags can be used to store hard red winter wheat without appreciable loss of wheat quality, but the CO_2 levels observed were not high enough to arrest insect and fungal development and prevent mycotoxin contamination. The low CO_2 concentrations observed may be due to gaps in sealing at both ends of the bags or loss of CO_2 due to damage by rodents and the cadelle beetle larvae. Wheat in silo bags should be stored on leveled ground with good drainage and on ground free of stones and gravel. Protecting against rodent damage

for bags placed outdoors on farms or at elevators will be difficult, but protection against insect attack, especially by species capable of penetrating the silo bags, can be prevented by spraying an approved insecticide on the outside of bags.

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