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A New Oxygen Barrier Film Reduces Aerobic Deterioration in Farm-Scale Corn Silage

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ABSTRACT

Recently, many studies have focused on the aerobic deterioration of corn silage at the farm level, because a large part of the product stored in horizontal silos is exposed to air and is more prone to spoilage. The most important factor influencing the preservation of forage ensiling is the degree of anaerobiosis that is usually achieved with sheets of polyethylene. A new black-on-white (125-μm) coextruded oxygen barrier (OB) film has been developed for silage sealing and was tested in the present experiment to assess the effects on fermentation quality, dry matter losses, and yeast and mold counts at opening of whole-crop corn bunker silos compared with conventional polyethylene (ST) film. Two trials were carried out on 2 commercial farms. The bunkers were divided into 2 parts along the length so that half of the feedout face would be covered with ST film and the other half with OB film. Eight plastic net bags with well-mixed fresh material were weighed and buried in the upper layer of the bunker, and 4 bags were buried in the central part. The silos were opened for summer consumption and were fed out at different rates (19 vs. 33 cm/d). The bags were unloaded, weighed, and subsampled to analyze the DM content, pH, lactic and monocarboxylic acids, ammonia, yeast and mold counts, and aerobic stability. The pH of the peripheral silage was different under the 2 films, with a lower value in the OB treatment. The OB film on farm 1 affected the silage dry matter losses, which were reduced 3.7 times in comparison with the ST film sealing. On farm 2, although the dry matter losses were numerically higher in the silage sealed with the ST film compared with OB film (9.0 vs. 5.9%, respectively), the difference was not statistically significant. However, the corn silage sealed with the ST film was less stable than the silage sealed with the OB film. The results indicate that the new OB film is a promising tool to constrain spoilage and dry matter losses under critical farm conditions, when inadequate amounts of silage are removed daily. The OB film further improved the stability of the corn silage in the peripheral areas of the silos even when a proper harvest-to-feedout management was implemented.
Key words: corn silage, oxygen barrier film, dry matter loss, aerobic deterioration

INTRODUCTION

The most important single factor influencing the preservation efficiency of forage ensiling is the degree of anaerobiosis reached in the completed silo (Woolford, 1990). A large part of the silage stored in horizontal silos is exposed to air and is prone to spoilage, especially in the upper part near the walls, which are difficult to seal properly (Ashbell and Lisker, 1988). If the airtight sealing of the silo is not appropriate, air penetrates the silage and aerobic microorganisms multiply, resulting in aerobic deterioration. Among the silages, corn silage is particularly susceptible to aerobic deterioration when it is exposed to oxygen or in the feed bunk (Ashbell and Weinberg, 1992; Kung et al., 1998). This results in losses of highly digestible DM (Bolsen et al., 1993), the possible production of mycotoxins (Borreani et al., 2005; Garon et al., 2006), and the growth of pathogenic species (Ivanek et al., 2006), which make silage less palatable and produce metabolic disorders in dairy cows (Wilkinson, 1999; Trevisi et al., 2003). Many factors can affect silage DM and nutritional losses during conservation and feedout, such as the daily feedout rate (Mahanna and Chase, 2003; Kleinschmit et al. 2005), the fermentation profile and use of silage additives (Weinberg and Muck, 1996), the type of plastic sealing (Savoie, 1988), the DM content at ensiling, the particle size, the filling rate, and the pack density in the silo (Johnson et al., 2002). Yeasts that metabolize lactic acid are the primary spoilage microorganisms in corn silage, although acetic acid bacteria and molds can also cause spoilage (Pahlow et al., 2003). The growth of lactate-utilizing yeasts causes a rise in pH, an increase in temperature, and a loss of DM (Woolford, 1990). In an experiment by Lindgren et al. (1985), oxygen diffusion during storage seemed to be important for the

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establishment of lactate-utilizing yeast, and the use of a 100-μm polyethylene cover was insufficient to prevent the diffusion of oxygen in bunker silages. The most common material used to seal bunker silos and drive-over piles is polyethylene film. In the early 1990s, Daponte (1992) proposed the use of coextruded barrier films to seal silage, but at that time, plastic manufacturers had no commercial interest in these more expensive films. The situation is now rapidly changing, and recently new developments in sealing strategies have been reported, involving the use of an improved 45-μm-thick coextruded film with reduced oxygen permeability as an alternative to standard polyethylene (Degano, 1999). Studies on the 45-μm non-UV-stabilized translucent barrier film, in combination with a protective tarpaulin, on corn and grass silages have shown both positive (Wilkinson and Rimini, 2002; Berger and Bolsen, 2006) and negligible (O'Kiely and Forristal, 2003; G. Borreani and E. Tabacco, unpublished data) effects on DM losses and visible top surface mold. As the need for a thicker, colored, and UV-stabilized oxygen barrier film at the farm level emerged from these studies, the authors asked a plastics manufacturer to develop a new film with such characteristics. A new black-on-white (125 μm) coextruded oxygen barrier (**OB**) film for silage sealing was recently developed by Industria Plastica Monregalese (Mondovi, Italy) and was tested in the current experiment.

The aim of the study was to assess the effect of the new OB film on the fermentation quality, DM losses, and the yeast and mold counts at opening of whole-crop corn bunker silos on 2 commercial farms with very different potential silage spoilage risks.

MATERIALS AND METHODS

Crop and Ensiling

Two trials were carried out on 2 commercial farms at Saluzzo (Cuneo, Italy; 44°40′ latitude, 7°32′ longitude, 325 m above sea level; farm 1) and at Frossasco (Torino, Italy; 44°75′ latitude, 7°23′ longitude, 290 m above sea level; farm 2) in 2005 and 2006 on corn silage in bunker silos. Whole-corn crops were harvested at around the 50% milk-line stage and chopped with a conventional forage harvester to a 10-mm theoretical length of cut and ensiled within 1 d in bunker silos. The farm 2 corn silage was inoculated with Pioneer 11C33 inoculant (dried *Lactobacillus buchneri* fermentation products, dried *Lactobacillus plantarum* fermentation products, and dried *Enterococcus faecium* fermentation products, Pioneer Hi-Bred Int. Inc., Johnston, IA) at the theoretical rate of 1.1×10^5 cfu of bacteria/g of fresh forage. The effect of 2 types of plastic sheeting used for sealing the silos (standard polyethylene vs. the new OB film) was studied. The 2 sealing treatments were 1) a single sheet of 180-μm-thick (6 and 8 m width for farm 1 and farm 2, respectively) black-on-white polyethylene (**ST**); and 2) a single sheet of 125-μm-thick (6 and 8 m width for farm 1 and farm 2, respectively) black-on-white coextruded polyethylene-polyamide film, with an enhanced OB. The 2 plastic sheets differed according to oxygen permeability, specific gravity, and weight per square meter (Table 1). The bunkers were divided into 2 parts along the length; half was covered with ST film and half with OB film to allow silage sampling of the 2 treatments at the same time. Approximately 2 meters of the plastic sheeting was placed on the side wall and turned on the top surface of the silos at the end of filling, to have an overlap of approximately 40 cm of the 2 sheets in the middle of the silos. The 2 plastic sheets were held to the silage with tires (2 to 3/m^2) and gravel bags near the side walls. Another polyethylene sheet similar to mosquito netting (approximately 2-mm mesh size) was put over the film before the tires to protect the 2 experimental sheets from insect or bird damage. During the filling of the silos, 8 plastic net bags (4 for each treatment) with well-mixed fresh material (approximately 7 kg of fresh weight/bag) were subsampled for preensiling analyses, weighed, and buried in the upper layer of the bunker in 2 sections 10 m apart, as previously described by Ashbell and Lisker (1988). The bags were placed so as to be representative of the peripheral 40 cm of the stored silage. Four more bags were weighed and buried in the central part of each bunker (1.5 m from the top). The silos were opened for summer consumption after 261 d and 325 d on farm 1 and 2, respectively. When the feed-out face reached a distance of 0.5 m from the bags, the bags were removed from the silos for analysis. Each bag was immediately weighed and subsampled to determine the DM concentration (3 replicates), fermentation profile (2 replicates), and microbiological counts (2 replicates). The remaining silage was used to determine aerobic stability. Approximately 3 kg of each replicate from each treatment was placed loosely in duplicate 20-L polystyrene boxes and allowed to deteriorate aerobically at room temperature (20°C). These silages were not disturbed throughout the recording of temperatures. A single layer

Table 1. Characteristics of the films used in the trials

Item	Plastic film[1]	
	OB	ST
Thickness, μm	125	180
Specific gravity, g/cm^3	0.963	0.937
Weight, g/m^2	120	169
Oxygen permeability, cm^3/m^2 per 24 h at 1 bar		
At 23°C, 0% relative humidity	70	990
At 23°C, 85% relative humidity	100	990

[1]OB = oxygen barrier film; ST = standard polyethylene film.

of aluminum cooking foil was placed over each container to prevent drying and contamination, but also to allow air penetration. The room temperature and the temperature of each silage were measured each hour by a data logger. Aerobic stability was defined as the number of hours the silage remained stable before rising more than 2°C above the ambient temperature (Ranjit and Kung, 2000). Eight core samples (45 mm diameter and 500 mm length) were taken from the feedout face of each silo, weighed, and oven-dried to determine the wet bulk and DM silage densities.

Sample Preparation and Analyses

The preensiled material and the silage were subsampled and immediately analyzed for DM content by oven-drying at 80°C for 24 h and for total nitrogen, according to the Dumas method, with a MicroN nitrogen analyzer (Elementar, Hanau, Germany). A second subsample of the preensiled material was dried for qualitative analyses in a forced-draft oven to constant weight at 60°C, air-equilibrated, weighed, and ground in a Cyclotec mill (Tecator, Herndon, VA) to pass a 1-mm screen. The dried samples were analyzed for ash by ignition to 550°C, NDF and ADF as described by Robertson and Van Soest (1981), and ether extract and starch as determined according to AOAC (1995) methods. Wet samples stored at –30°C were homogenized and extracted for 4 min in a Stomacher blender (Seward Ltd., Worthing, UK) in water or in 0.1 *N* H_2SO_4. The ammonia nitrogen contents, determined by using a specific electrode, were quantified in the acid extract. The lactic and monocarboxylic acids (acetic, propionic, and butyric acids) were determined by HPLC (Canale et al., 1984). Ethanol was determined by an HPLC instrument, coupled to a refractive index detector, on a Aminex HPX-87H column (Bio-Rad Laboratories, Richmond, CA). The analyses were performed isocratically under the following conditions: mobile phase 0.0025 *M* H_2SO_4, flow rate 0.5 mL/min, column temperature 37°C, injection volume 100 μL. Duplicate analyses were performed for all the determined parameters. The duplicates were averaged and the 4 means (replicated silage bags) were considered as 4 observations in the statistical analysis. The DM losses were calculated as the difference between the amount of DM placed in each bag at ensiling and the DM removed at the end of conservation.

The colony-forming units of yeasts and molds were counted by using the pour plate technique with 40.0 g/L of yeast extract glucose chloramphenicol agar (YGC agar, Difco, West Molesey, Surrey, UK) after incubation at 25°C for 3 and 5 d for yeasts and molds, respectively.

Characteristics of the 2 plastic films were measured as follows: thickness with a digital electronic micrometer (Series 422, Mitutoyo, Aurora, IL); specific gravity and weight per square meter, by weighing 10 m^2 of sheeting (3 replicates); and oxygen permeability following ASTM standard method D 3985-81 (ASTM, 1980).

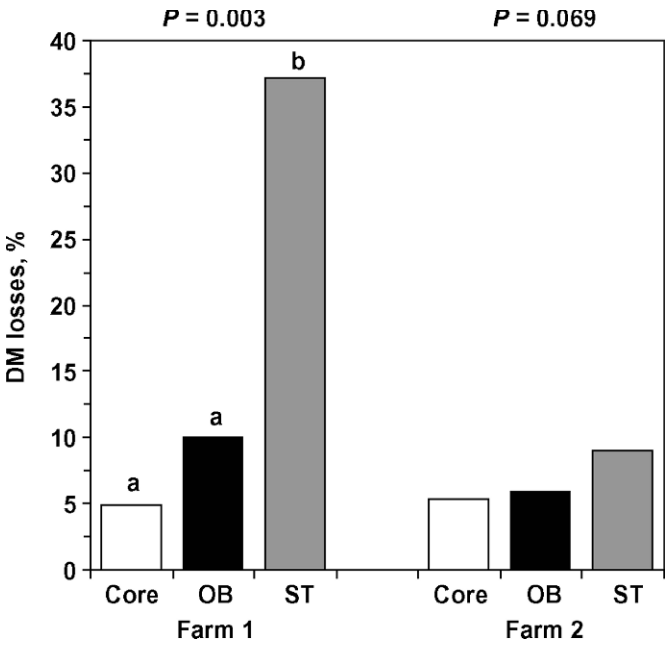

Figure 1. Effect of plastic film on DM losses in different zones of bunker corn silage in 2 farms over summer consumption. CORE = silage in the core of the bunker; OB = silage under the oxygen barrier film; ST = silage under the polyethylene film; farm 1 = untreated silage, 19 cm/d removal rate; farm 2 = *Lactobacillus buchneri*-treated silage, 33 cm/d removal rate. [a,b]Different letters indicate a significant difference.

Statistical Analysis

The chemical compositional data and microbial counts were analyzed for their statistical significance via ANOVA, with significance reported at the 0.05 probability level, by using the GLM of the Statistical Package for the Social Sciences (version 11.5, SPSS Inc., Chicago, IL). All microbial counts were $\log_{10}$ transformed to obtain log-normal distributed data. The DM losses were analyzed by performing ANOVA on angular transformed values (arcsine transformation). Significant differences between means were identified by the *P*-values of ANOVA, and the effects were considered significant at $P < 0.05$. In Figure 1 the DM losses of peripheral areas were compared with the DM losses measured in the core of the silos, and analyzed via ANOVA. When the calculated values of *F* were significant, Duncan's multiple range test ($P < 0.05$) was used to interpret any significant differences among the mean values.

Table 2. Characteristics of the bunker silos used in the 2 trials

Item	Farm 1	Farm 2
Period of consumption	Summer	Summer
Consumption, t of fresh matter/d	1.86	6.26
Silo size, m		
Width	8	12
Height	2.6	2.9
Length	19	58
Conservation, d	261	325
Additive	Untreated	Inoculated[1]
Feedout, cm/d	19	33
Wet bulk density, kg/m^3	541	568
DM density, kg/m^3	192	217

[1]Inoculated with dried *Lactobacillus buchneri*, *Lactobacillus plantarum*, and *Enterococcus faecium*.

RESULTS AND DISCUSSION

The characteristics of the bunker silos used in the trials are reported in Table 2. The 2 farms were selected because of their different silage management techniques, which can influence the proneness of silage to aerobic deterioration. The silos were both conserved for more than 8 mo and opened in summer, when silage is more prone to aerobic deterioration (Mahanna and Chase, 2003). The daily feedout rate differed greatly on the 2 farms because of the different silo sizes and daily consumption. Furthermore, the corn silage on farm 2 was treated with an *L. buchneri*-based inoculant, which is known to improve the aerobic stability of silages (Kleinschmit and Kung, 2006). The wet and DM densities were similar and were typical of well-managed bunker corn silage (Savoie et al., 2004). The chemical and fermentation characteristics of the silage cores from the 2 studied silos are given in Table 3. All the silages were well preserved, and the DM content was in the range of that for corn harvested at the dough stage (around one-third to one-half kernel milk line). The main fermentation acids found were lactic and acetic acids, and no butyric or propionic acids were found. Ammonia nitrogen and ethanol were lower than 10% total nitrogen and 1% DM, respectively. The values were within the normal range for well-fermented corn silage harvested at comparable DM levels (Muck and Pitt, 1994). The silages contained an average of 1.35 and 1.16 log cfu of molds/g of silage and 3.05 and 1.61 log cfu of yeasts/g for farm 1 and 2, respectively. The yeast counts in both silos were less than 3.1 log cfu/g, which corresponds to a greater than 50-h aerobic stability (Muck, 2004).

The fermentation and microbial characteristics of the peripheral areas of the silage from farm 1 are summarized in Table 4. A film type effect occurred for the pH, lactic acid, and butyric acid. More lactic acid was produced in the silage sealed with the OB film, which is consistent with the lower pH values. Berger and Bolsen (2006) reported a better fermentation profile in corn silage in the top 0 to 46 cm under a 45-μm OB film compared with corn silage under a 150-μm polyethylene sheet. The pH value of the silage sealed with the ST film was 5.87, which is typical of severely deteriorated whole-crop corn silages, as reported by Ashbell and Weinberg (1992) and Berger and Bolsen (2006), who indicated pH values ranging from 4.8 to 8.5. The lower oxygen permeability of the OB film also affected the microbial counts by reducing molds, whereas the aerobic stability was similar in the 2 treatments. However, it should be pointed out that 2 of the 4 bags from the ST treatment were completely deteriorated at unloading, so an aerobic stability test was not performed on these bags. An average aerobic stability of 72 and 69 h was obtained for the

Table 3. Chemical and fermentative characteristics of the silages in the core of the 2 bunkers

Item	Farm 1	Farm 2
DM, %	34.0 ± 0.39	38.2 ± 1.13
NDF, % of DM	41.2 ± 1.04	52.5 ± 1.84
ADF, % of DM	22.0 ± 0.54	26.8 ± 0.62
CP, % of DM	6.34 ± 0.46	9.69 ± 0.54
Ether extract, % of DM	2.03 ± 0.33	1.66 ± 0.26
Ash, % of DM	3.98 ± 0.35	5.31 ± 0.63
Starch, % of DM	29.4 ± 1.13	23.0 ± 0.70
pH	3.60 ± 0.04	3.81 ± 0.03
Lactic acid, % of DM	4.92 ± 0.67	4.40 ± 0.42
Acetic acid, % of DM	1.71 ± 0.41	3.69 ± 0.65
Butyric acid, % of DM	<0.001	<0.001
Propionic acid, % of DM	<0.001	<0.001
Ethanol, % of DM	0.90 ± 0.11	0.37 ± 0.02
NH_3-N, % of total N	7.34 ± 0.90	5.28 ± 0.66
Molds, $\log_{10}$ cfu/g	1.35 ± 0.51	1.16 ± 0.09
Yeasts, $\log_{10}$ cfu/g	3.05 ± 0.88	1.61 ± 0.17

Table 4. Fermentation characteristics and concentrations of molds and yeasts in the peripheral area of the bunker silo on farm 1, sealed with an oxygen barrier (OB) or standard polyethylene (ST) film

	Plastic film			
Item	OB	ST	SE	*P*-value
DM	32.3	27.4	1.83	0.211
pH	3.99	5.89	0.47	0.037
Lactic acid, % of DM	2.05	0.87	2.73	0.025
Acetic acid, % of DM	3.72	2.58	3.54	0.110
Butyric acid, % of DM	<0.001	0.18	—[1]	—
Propionic acid, % of DM	0.68	0.35	0.61	0.003
Ethanol, % of DM	0.54	0.62	1.00	0.687
NH_3-N, % of total N	10.6	11.0	1.34	0.906
DM losses, %	10.00	37.17	0.08	0.040
Molds, $\log_{10}$ cfu/g	1.55	5.07	0.53	<0.001
Yeasts, $\log_{10}$ cfu/g	<1.00	<1.00	—[1]	—
Aerobic stability,[2] h	72	69[3]	19.4	0.947

[1]Statistical analysis not performed.

[2]Time for a 2°C increase in silage temperature (h).

[3]The measurement was performed on only 2 out of 4 bags, because 2 bags proved to be completely deteriorated at unloading.

Table 5. Fermentation characteristics and concentrations of molds and yeasts in the peripheral area of the bunker silo on farm 2, sealed with an oxygen barrier (OB) or standard plastic (ST) film

	Plastic film			
Item	OB	ST	SE	*P*-value
DM	34.0	31.1	1.05	0.169
pH	3.85	4.05	0.05	0.040
Lactic acid, % of DM	3.96	2.30	0.52	0.116
Acetic acid, % of DM	2.33	3.36	0.42	0.239
Butyric acid, % of DM	<0.001	<0.001	—[1]	—
Propionic acid, % of DM	0.44	0.43	0.07	0.964
Ethanol, % of DM	0.79	0.46	0.12	0.209
NH_3-N, % of total N	6.02	7.95	0.55	0.074
DM losses %	5.91	8.96	0.04	0.175
Molds, $\log_{10}$ cfu/g	1.46	1.39	0.10	0.713
Yeasts, $\log_{10}$ cfu/g	<1.00	1.66	—[1]	—
Aerobic stability,[2] h	355	178	38.0	0.015

[1]Statistical analysis not performed.

[2]Time for a 2°C increase in silage temperature (h).

OB and ST treatments, respectively. These values are greater than those reported by Kleinschmit and Kung (2006) for uninoculated corn silage (25 h), and could be due to the relatively high concentration of acetic acid, which is a better inhibitor of yeasts than lactic acid at a given pH (Moon, 1983).

The effect of film type on the peripheral areas of the silage on farm 2 was less evident than on farm 1 (Table 5). However, the pH differed ($P = 0.040$), with a lower value under the OB film. The aerobic stability of corn silage sealed with the ST film was lower ($P = 0.015$) than that of silage sealed with the OB film (178 vs. 355 h, respectively). On the whole, the aerobic stability of the silages from farm 2 was greater than that observed on farm 1, and was consistent with an average aerobic stability of 503 h of corn silage inoculated with *L. buchneri* at $>1 \times 10^5$ cfu/g (Kleinschmit and Kung, 2006). The yeast counts in the OB corn silage were always under the detection limit, whereas yeasts were detectable in the ST film silage, with a mean value of 1.66 $\log_{10}$ cfu/g. It can be speculated that the establishment of yeasts during storage is due to a greater diffusion of oxygen caused by leakage through the plastic cover (Lindgren et al., 1985), which results in a lower aerobic stability after exposure to air. This is consistent with the observations made by Kung et al. (1998) and Muck (2004), who found a negative correlation between the number of yeasts and the hours of aerobic stability of corn silage.

Oxygen permeability of the film also influenced the DM losses in the upper 40 cm of the silage, as reported in Figure 1. The OB film on farm 1 affected the DM losses of the silage ($P = 0.040$), which were reduced by 3.7 times in comparison with the ST film sealing. The DM losses on farm 2 were numerically greater in the silage sealed with the ST film compared with the OB film (9.0 vs. 5.9%, respectively), but the difference was not statistically significant ($P = 0.175$). Berger and Bolsen (2006) reported DM losses in the top 46-cm layer ranging from 36 to 52% in unsealed silage and from 14 to 28% in silage covered with a single sheet of 100 to 150 μm polyethylene in a survey of corn silage from 127 bunker silos and piles in Kansas. We found higher DM losses in the ST treatment on farm 1 compared with the values reported by Berger and Bolsen (2006) because of feedout over summer and a longer conservation period than 260 d. The data are consistent with Ashbell and Lisker (1988), who observed, in a subtropical climate, that DM losses of corn silage in the upper layer and near the walls were between 10.2 and 35.8%. The DM losses of silages under the OB film (Figure 1) were not significantly different from those measured in the cores of the silos on both farms, and were in the range of well-conserved corn silage (Ashbell and Lisker, 1988; Johnson et al., 2002).

The results of the current experiment confirm the relevance of silage management factors, such as daily feedout rate and the use of an *L. buchneri*-based inoculant, on the fermentation quality of the whole mass stored in horizontal bunker silos. The introduction of the OB film on the farm, where management factors to contain aerobic deterioration are critical, resulted in a great improvement in the silage quality and a reduction in the DM losses in the peripheral area of the silo. On the farm with the well-managed silo, the quality of silage was generally good, with DM losses lower than 10%. On this farm, the OB film further improved the silage quality in the peripheral area, in terms of pH and aerobic stability, and contained DM losses to values closer to those observed in the core of the silo.

CONCLUSIONS

The results obtained in the current study indicate that the new OB film is a promising tool to contain spoilage and DM losses, especially under critical farm conditions, when inadequate amounts of silage are removed daily, and under high environmental temperatures during feedout. However, even when proper management from harvest to feedout was implemented (fast filling rates, high packing densities, and sufficient quantities of silage removed between feedings) and effective silage additives (an *L. buchneri*-based inoculant) were used, the OB film showed a further improvement in the stability of corn silage in the peripheral area of the silos.

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