Journal of Animal and Feed Sciences, 15, Suppl. 1, 2006, 121–124

Effects of different silage additives on the microbial population and aerobic stability of maize silage*

K. Stryszewska[1] and J.B. Pyś

Agricultural University of Cracow,
Department of Animal Nutrition
Al. Mickiewicza 24/28 Kraków, Poland

ABSTRACT

The effects of bacterial or chemical additives on the microbial population and aerobic stability of silage were examined. Whole-plant maize silages were made: MO, control group without additives; MB, with bacterial inoculant; MBC, with bacterial inoculant and chemical additive; and MC, with chemical additive. The microbial population was determined in silages after opening the silo and exposing the contents to air for 7 days. It was found that the MBC bacterial-chemical additive considerably inhibited the development of mold fungi after air exposition and improved the aerobic stability of maize silage.

KEY WORDS: maize silage, bacterial, additive, aerobic stability

INTRODUCTION

Maize silage is particularly susceptible to aerobic deterioration and secondary fermentation due to the activity of lactate-assimilating yeasts (Woolford, 1990). Degradation of lactic acid raises the pH of silage, leading to the development of opportunistic bacteria (e.g., *Clostridium, Listeria, Eschericha coli)* and molds (Scudamore and Livesy, 1998). The degradation products can affect palability and cause feed refusal by livestock (Wardynski et al., 1993). The growth of molds in silage is undesirable since they not only change the chemical composition of forage, but also produce mycotoxins, which can be potentially lethal (Scudamore and Livesy, 1998). Maize silage can be made resistant to aerobic decomposition and secondary fermentation, and a desirable microbiological profile can be obtained by ensiling forage with bacterial, enzymatic, or chemical additives.

The aim of the study was to determine the effect of adding microbial and chemical additives to whole-plant maize silage, its fermentation quality, microbial population, and aerobic stability.

* Supported by the Ministry of Education and Science, Grant No. 2 P06Z 015 28
[1] Corresponding author: e-mail: kstryszewska@o2.pl

MATERIAL AND METHODS

Whole-plant maize forage (*Zea mays* L.) cv. Eurostar FAO 240 with a dry matter content of 315.2 $g \cdot kg^{-1}$ was cut into 10-15 mm particles. Polyethylene silos with a 15 l capacity were used to make the following maize silages: no additive, MO; bacterial additive (*Lactobacillus plantarum, Propionibacterium acidipropionici* 1.5×10^5 $cfu \cdot g^{-1}$ of forage), MB; bacterial-chemical additive (*L. plantarum, E. faecium, Pediococcus acidilactici* 1×10^6 $cfu \cdot g^{-1}$ of forage, and potassium sorbate), MBC; chemical additive, %: formic acid 59, propionic acid 20, ammonium formate 4.3 and potassium sorbate 2.55, MC. Silages were stored at 13±2°C, for 60 days.

Silages were assayed for dry matter (AOAC, 2000), $N\text{-}NH_3$ (Skumulowski, 1974), pH potentiometrically, and organic acid contents (by gas chromatography, Varian Star 3400 CX and a DB-FFAP capillary column with argon as the carrier gas). Microbial analyses were performed according to Polish Standard PN-R-64791 (1994). Total counts were determined for mesophilic bacteria, after 24 h incubation at 37°C on MPA agar (BTL, P-0012), lactic acid bacteria, after 48 h incubation at 28°C on China blue lactose broth agar (CRhodia), and yeast and molds, after 72 h incubation at 28°C on Czapek-Dox agar (Fluka, 70185). Aerobic stability was determined by incubating silage samples in an air-conditioned room at 20±1°C according to the method of Honig (1985). Stability was defined as the time needed to raise the temperature to 3°C above ambient temperature. The results were subjected to statistical analysis by one-way analysis of variance and the Student-Newman-Keuls test, using the SAS software package (1996).

RESULTS

With the exception of the chemical additive (MC), all of the remaining additives significantly ($P<0.05$) reduced silage pH. The chemical additive was particularly effective in significantly ($P<0.05$) limiting the degradation of protein to ammonia. The highest rate of lactic fermentation was found in the silages with the bacterial-chemical and bacterial additives, the lowest, with the chemical additive. The rate of acetic fermentation increased in the silages with the bacterial-chemical additive, and decreased significantly ($P<0.05$) in the silages with bacterial and chemical additives. Propionic acid was present in the silages with bacterial and chemical additives, but formic acid was found only in the silages with the chemical additive. No butyric acid was detected in the silages. The chemical and bacterial-chemical additives significantly ($P<0.05$) inhibited the aerobic decomposition of silages (Table 1).

Table 1. Dry matter content, fermentation parameters and aerobic stability of maize silage

Items	MO	MB	MC	MBC
Dry matter, g·kg^{-1}	298.5^{a}	292.2^{a}	300.7^{a}	300.2^{a}
pH	3.97^{b}	3.87^{c}	4.07^{a}	3.80^{c}
NH_3-N, g·kg^{-1} of total-N	37.1^{a}	30.9^{b}	28.6^{b}	29.9^{b}
Lactic acid, g·kg^{-1} DM	24.6^{c}	35.2^{b}	15.4^{d}	43.5^{a}
Acetic acid, g·kg^{-1} DM	12.3^{a}	8.6^{b}	7.8^{b}	13.1^{a}
Propionic acid, g·kg^{-1} DM	0.0^{b}	2.3^{a}	3.2^{a}	3.0^{a}
Formic acid, g·kg^{-1} DM	0.0^{b}	0.0^{b}	5.2^{a}	0.0^{b}
Butyric acid, g·kg^{-1} DM	0.0	0.0	0.0	0.0
Aerobic stability, h	28^{d}	48^{c}	67^{b}	145^{a}

^{a,b,c,d} means in rows with different letters differ significantly at P<0.05

No significant (P>0.05) effects of the additives used were found on the number of lactic bacteria and yeast in maize silages. The largest number of molds was found in the silages with no additive. No mold fungi were found in the silages with the bacterial-chemical additive (Table 2).

Table 2. Microorganisms (log_{10} CFU·g^{-1} of fresh matter) recovered from maize silage

Time	Item	MO	MB	MC	MBC
After opening	Total bacteria	8.01^{a}	5.05^{b}	8.06^{a}	6.20^{ab}
	LAB	7.62^{a}	7.65^{a}	6.04^{b}	7.09^{ab}
	Yeasts	4.84^{a}	5.57^{a}	5.66^{a}	5.09^{a}
	Molds	2.85^{a}	1.05^{a}	1.2^{a}	0.00^{a}
After 7 d of air exposition	Total bacteria	7.29^{a}	7.51^{a}	8.32^{a}	7.82^{a}
	LAB	6.90^{ab}	7.18^{ab}	6.0^{a}	5.58^{b}
	Yeasts	8.43^{a}	8.41^{a}	8.42^{a}	6.31^{b}
	Molds	7.88^{a}	7.83^{a}	7.12^{a}	5.12^{b}

^{a,b} means in rows with different letters differ significantly at P<0.05. LAB - lactic acid bacteria

Aerobic exposure of silages with no additive and with the bacterial additive led to an increase in the number of lactic bacteria. In all of the silages, the period of aerobic stability was followed by a marked increase in the number of yeast and mold fungi compared with silages before the stability test. The bacterial-chemical additive had the greatest effect on limiting the growth of yeasts and mold fungi in the silages exposed to oxygen (Table 2).

DISCUSSION

The lowest degradation of protein to ammonia in the silages with the chemical and bacterial-chemical additives resulted from their higher rate of lactic and acetic

fermentation and greater amount of propionic acid, which inhibits the growth of proteolytic bacteria (Kung and Ranjit, 2001). The highest aerobic stability of the silages with bacterial-chemical and chemical additives was due to the marked limiting of microorganisms causing secondary fermentation by acetic, propionic and formic acids, which were the most abundant in these silages (Wardynski et al., 1993). The lowest number of yeast and mold fungi in the silages with the bacterial-chemical additive also contributed to their highest aerobic stability. It has been shown that the bacterial additive containing *Lactobacillus plantarum* produced several antifungal substances that reduce the growth of molds and limit mycotoxin production (Ström et al., 2002). Sorbic acid and its salts are also effective inhibitors of mold fungi isolated from silages (Uriarte and Bolsen, 2001).

CONCLUSIONS

Ensiling maize with homofermentative bacteria and potassium sorbate has a marked effect on reducing the number of undesirable microorganisms and delays aerobic decomposition and time to heat damage. Contrary to expectations, the chemical additive used was not the most effective in limiting aerobic decomposition and the growth of undesirable microflora in maize silages.

REFERENCES

AOAC, 2000. Official Methods of Analysis, Association of Official Analytical Chemists. 17th Edition. Washington, DC

Honig H., 1985. Determination of aerobic stability-System. Landbauforsch. Völkenrode, SH 2,3

Kung Jr. L., Ranjit N.K., 2001. The effect of *Lactobacillus buchneri* and other additives on the fermentation and aerobic stability of barley silage. J. Dairy Sci. 84, 1149-1155

PN-R-64791, 1994. Polish Standard. Feedstuffs. Requirements and Microbiological Evaluation (in Polish)

SAS, 1996. User's Guide: Statistics. Release 6.12. SAS Instit. Inc., SAS Campus Drive. Cary, NC

Scudamore K.A., Livesy C.T., 1998. Occurrence and significance of mycotoxins in forage crops and silage: a review. J. Sci. Food Agr. 77, 1-17

Ström K., Sjögren J., Broberg A., Schnürer J., 2002. *Lactobacillus plantarum* MiLAB 393 producers the antifungal cyclic dipeptides cyclo ($_{L}$-phe-$_{L}$-pro) and cyclo ($_{L}$- phe-*trans*-4-OH-$_{L}$-pro) and 3-phenyllactic acid. Appl. Environ. Microbiol. 68, 4322-4327

Uriarte M.E., Bolsen K.K., 2001. Aerobic deterioration of silage: processes and prevention. In: Proceedings of Altech's 17th Annual Symposium, Trumpton Nottingham (UK), pp. 127-144

Woolford M.K., 1990. The detrimental effect of air on silage. J. Appl. Bacteriol. 68, 101-116

Wardynski F.A., Rust S.R., Yokyama M.T., 1993. Effect of microbial inoculation of high-moisture corn on fermentation characteristic, aerobic stability and cattle performance. J. Anim. Sci. 71, 2246-2252