Moments of action of *Lactobacillus* combination in maize silage

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Introduction: Various bacterial and chemical additives with antifungal characteristics have been used in order to reduce aerobic spoilage of silage. However, the use of microbial inoculants for maize silages is not a consensus among farmers, researchers and technicians. This situation shows potential for increased sales as well as stimulating the development of new products. This study aimed to evaluate fermentation, total dry matter losses and aerobic stability of maize (*Zea mays*) silages inoculated with different commercial additives.

Materials and Methods: The research was performed by Centro de Pesquisas em Forragicultura (CPFOR) at Canguiri Experimental Farm of Federal University of Paraná. The corn plant was harvested in February with dry matter content (DM) of 343 g/kg. At ensiling, forage was divided in three piles for application of the following treatments: Control (10 mL distilled water per ton of fresh forage); Commer (*Lactobacillus buchneri* and *Lactobacillus plantarum* ($1.1x10^{11}$ cfu/g), maltodextrin and sodium aluminosilicate diluted in 10 mL distilled water per ton of fresh forage), and CommerLC (*L. buchneri* and *L. plantarum* ($1.1x10^{11}$ cfu/g), *Lactobacillus casei*, maltodextrin and sodium aluminosilicate diluted in 10 mL distilled water per ton of fresh forage). Additives were sprayed onto the forage and homogenized. Fifteen plastics buckets (5 silos/treatment) were filled up with density of 650 kg fresh matter/m^3. The silos were storage during 66 days in ambient temperature. At the opening, silage was homogenized and sampled for pH and DM. The pH followed the methodology of Kung Jr et al. (2000). The determination of fermentative losses was made according to equations described by Jobim et al. (2007). For assessing aerobic stability, 5 kg of silage were stored in plastic buckets, without compaction, for 240 h in a temperature-controlled room (23 ± 1.7°C). One thermometer datalogger was placed in the center of each bucket to register temperature every 30 min. Aerobic stability (AS) was considered according to Moran et al. (1996). The data were analyzed as a completely randomized design, with three treatments and five replicates. All variables were analyzed for normality. After, ANOVA and Tukey test ($P<0.05$) were applied. The analyses were made by Statgraphics Centurion program.

Results and Discussion: Fermentation variables and losses are presented in Table 1. The average silage DM was 4.4 g/kg higher than the fresh forage. Similarly, the pH values characterize well preserved silage (McDonald et al., 1991).

Table 1. Dry matter content, pH and fermentative losses of treated and untreated maize silages

	Treatment[2]			
Variable[1]	Control	CommerLC	Commer	SEM[3]
DM (g/kg)	355.2^{a}	339.6^{b}	347.4^{ab}	0.21
pH	3.82^{ab}	3.87^{a}	3.70^{b}	0.03
DML (% DM)	-1.26^{b}	2.20^{ab}	2.82^{a}	0.03
GL (% DM)	-2.61	0.51	-0.36	1.12

EP (kg/t FM)	13.45[b]	16.96[ab]	19.81[a]	1.33

[1]DM = dry matter; DML = total dry matter losses; GL = gas losses; EP = effluent production.
[2]Treatment: Commer = *L. buchneri*, *L. plantarum*, maltodextrin, sodium aluminosilicate; CommerLC = *L. buchneri*, *L. plantarum*, *L. casei*, maltodextrin, sodium aluminosilicate.
[3]SEM = standard error of the mean.
Means followed by different letters are significant by Tukey test (P>0.05).

Despite the well conserved silage values (Deminicis et al., 2009), the evaluated additives were not able to reduce losses, resulting in larger losses than untreated silage. The Control treatment not only had the lowest losses but also presented negative loss results. The mean gross weights of the silos have decreased 0.296 g/silo, while the average of DM weight increased 0.04 g. It shows that in general the fermentative losses were still very low.
Negative values for DML are commonly considered experimental errors, even though frequently observed in silage tests. But it is important to note that, especially in the case of DML, the SEM was extremely low, which also relates to the low probability of errors.
Although the reasonable action of additives in the fermentation phase, the opposite was observed during AS (Table 2). The additive Commer maintained AS during study period as well as the lowest maximum silage temperature. The DM losses during air exposure (DMLas) were also lower in additive treatments, on average about 10% lower.

Table 2. Aerobic stability, maximum temperature and DM losses of maize silages

	Treatment[2]			
Variable[1]	Control	CommerLC	Commer	SEM[3]
AS (hours)	99.7[b]	118.1[b]	>240[a]	16.93
Tmax (°C)	28.5[a]	28.2[a]	20.9[b]	1.40
DMLas (% DM)	15.74[a]	8.24[b]	2.84[b]	1.69

[1]AS = aerobic stability; Tmax = maximum temperature; DMLas = silage dry matter losses after 240 h of air exposure.
[2]Treatment: Commer = *L. buchneri*, *L. plantarum*, maltodextrin, sodium aluminosilicate; CommerLC = *L. buchneri*, *L. plantarum*, *L. casei*, maltodextrin, sodium aluminosilicate.
[3]SEM = standard error of the mean.
Means followed by different letters are significant by Tukey test (P>0.05).

The presence of *L. casei* in the CommerLC treatment resulted in lower DM, higher pH, but one of the lowest fermentative losses in relation to the other treatments; similar results were found by Nishino et al. (2004). While the magnitude of the fermentation losses in the Commer treatment did not influenced AS. It is suggested that there was no synergism between *L. casei* and *L. buchneri* + *L. plantarum*, in which the latter dominated the process. The cost of this combination in the inoculant development should be analyzed.

Conclusions: The use of *L. casei* in combination with another *Lactobacillus* should be reviewed. Silages treated with Commer additive are indicated for periods of greater post-opening challenge, but they use during fermentation should be cautious.