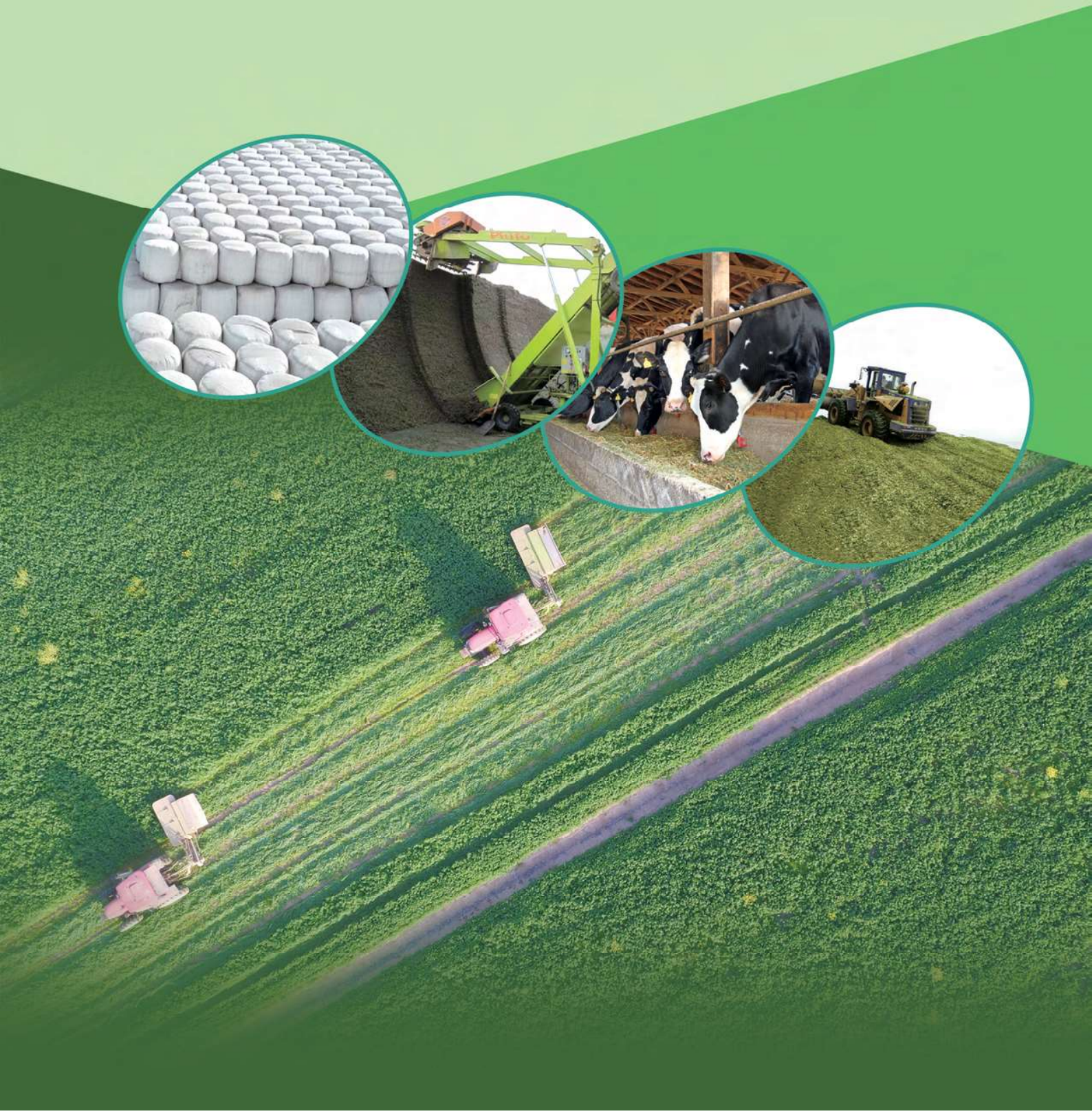




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Microbial community during fermentation and aerobic deterioration of high moisture corn treated with homofermentative and heterofermentative lactic acid bacteria

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Keywords

Aerobic deterioration, bacteria community, fungi, LAB inocula

Introduction

Lactic acid bacteria inoculants have been selected to improve the fermentation efficiency through a homolactic pathway and to improve the aerobic stability through a heterolactic pathway (Muck et al., 2018). Metagenomics allows to improve the understanding of microbial community and can be used as a tool to verify the role of LAB inocula (McAllister et al., 2018). The aim of this work was to analyze the microbial community during fermentation and aerobic deterioration of high moisture corn treated with homofermentative and heterofermentative lactic acid bacteria.

Material and Methods

High moisture corn (whole ear, cob, and grain) was harvested at DM content of 62%. Fresh material was untreated (C) or treated with *Lactobacillus plantarum* (100,000 cfu/g FM, commercial inoculum) (LP) or a mixture of *L. buchneri* and *L. hilgardii* (150,000 + 150,000 cfu/g FM, Lallemand SAS) (COMBO). The fresh forages were sampled prior to ensiling (n=4). The untreated and treated forages (n=4) were then ensiled in 20-L plastic silos and opened after 250 d of conservation. Silages were subjected to an aerobic stability test and sampled at 7 and 14 days of air exposure. Silages were analyzed for fermentative profile and microbial count. DNA was extracted from each sample and microbiota was studied by amplifying the V3 and V4 region of the 16S rRNA and the internal transcribed spacer (ITS) -1 region for the fungi. Data were analyzed by analysis of variance with Bonferroni post-hoc test (SPSS v. 28).

Results

Lactobacillus genus dominated the fermentation in LP and COMBO (Figure 1, $P < 0.001$). After 6 d of air exposure *Acetobacter* was detected in C silages. *Kazachstania* and *Pichia* were the main yeast genus detected at silo opening and during aerobic exposure (Figure 2). Both species are lactate utilizers, *Pichia* was more active in C and LP ($P < 0.001$). In the advanced state of air exposure *Aspergillus* genus was detected in C. During fermentation LP determined higher lactic-to-acetic acid ratio compared to C and COMBO (Figure 3a, $P = 0.003$). The acetic acid was higher in COMBO than other silages and determined lower yeast count and higher aerobic stability (Table 1, Figure 3, $P < 0.001$). When silages are aerobically unstable an increase of yeast, mold, acetic acid bacteria, and anaerobic sporeformer counts were detected.

Table 1. pH and microbial count at harvest, opening, and after air exposure of high-moisture corn treated with homolactic or heterolactic bacteria.

	C	LP	COMBO	P-value
			<i>pH</i>	
Harvest	5.99	5.99	5.93	ns
Opening	3.80 ^a	3.76 ^b	3.79 ^{ab}	***
7 days air exposure	4.10 ^a	3.96 ^b	3.89 ^c	***
14 days air exposure	5.63 ^a	5.84 ^a	4.07 ^b	**
			<i>Yeast (log cfu/g)</i>	
Harvest	6.30	6.25	6.71	ns
Opening	3.36	3.54	2.92	ns
7 days air exposure	8.41 ^a	8.32 ^a	5.78 ^b	*
14 days air exposure	8.38	8.69	7.36	ns
			<i>Mold (log cfu/g)</i>	
Harvest	6.18	6.14	6.17	ns
Opening	2.59	3.25	2.60	ns
7 days air exposure	< 1.00	< 1.00	< 1.00	-
14 days air exposure	8.76 ^a	8.47 ^a	4.44 ^b	*
			<i>Acetic acid bacteria (log cfu/g)</i>	

(continued)

	C	LP	COMBO	P-value
Harvest	5.83	6.12	5.64	ns
Opening	< 2.00	< 2.00	< 2.00	—
7 days air exposure	5.78 ^a	5.66 ^a	3.02 ^b	***
14 days air exposure	8.08	9.30	8.32	ns
		<i>Anaerobic sporeformer (log spore/g)</i>		
Harvest	3.74	2.88	3.07	ns
Opening	2.45	2.80	2.68	ns
7 days air exposure	2.36	2.41	3.19	ns
14 days air exposure	5.54 ^a	5.20 ^b	2.84 ^c	*

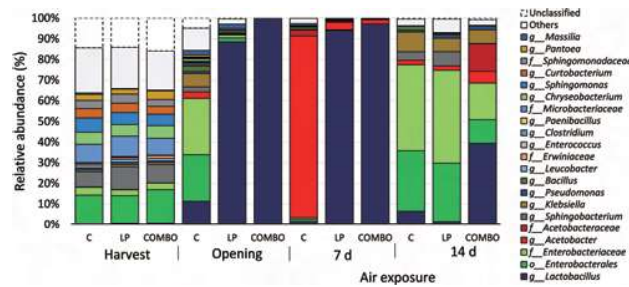


Figure 1. Relative abundance of bacteria at harvest, opening, and after air exposure of high-moisture corn treated with homolactic or heterolactic bacteria.

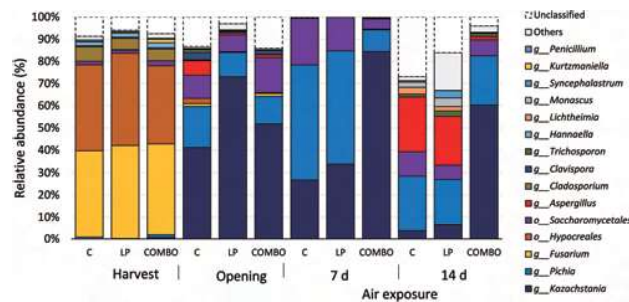


Figure 2. Relative abundance of yeast and mold at harvest, opening, and after air exposure of high-moisture corn treated with homolactic or heterolactic bacteria.

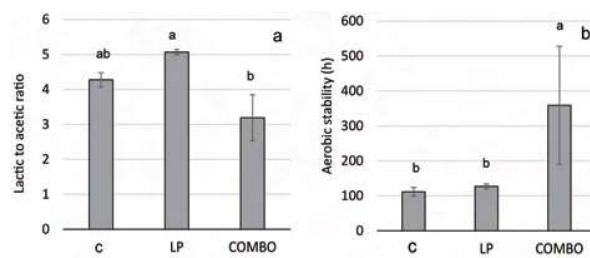


Figure 3. Lactic to acetic ratio (a) and aerobic stability (b) at opening of high-moisture corn treated with homolactic or heterolactic bacteria.

Conclusions

The analysis of the metagenome allows to better understand the role and efficiency of inoculant in silage quality. This work was partially funded by Lallemand SAS.

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