Silage inoculants improves quality and aerobic stability in grass, clover-grass and lucerne silage

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Introduction

The type of silage additive will influence the characteristics of fermentation, dry matter (DM) losses, hygienic quality and aerobic stability of silage. The appropriate use of an additive also depends on the crop to which it is applied. Homofermentative lactic acid bacteria (LAB) strains are selected for rapid growth under under variable temperatures, herbages and dry matter concentrations. The homofermentative LAB are highly competitive and produce largely lactic acid and reduce pH and ammonia-N content compared to untreated silage (Muck 2012). Heterofermentative LAB such as *Lactobacillus buchneri* reduce the growth of yeast and mould and increase aerobic stability. These effects are retained when heterofermentative LAB are added in combination with homofermentative LAB (Kleinschmit *et al.* 2005). An alternative to the combination of homo- and heterofermentative inoculants is to use homofermentative LAB in combination with a chemical component such as sodium benzoate (NaBe). In an experiment by Jaakkola *et al.* (2010) a combination of *Lactobacillus plantarum* and sodium benzoate were more efficient than *Lactobacillus plantarum* and *Lactobacillus buchneri* to prevent heating of silage. The objective of the study was to investigate efficacy of different inoculant types on fermentation characteristics and aerobic stability of Lucerne (L), ryegrass (R), red clover-ryegrass (RCR) and ryegrass-timothy (RT) silages.

Methods

The experiment was conducted at the Institute of Animal Science of Lithuanian University of Health Sciences. The following LAB combinations were tested: *Lactobacillus buchneri* CCM 1819 (HET); *Lactobacillus buchneri* CCM 1819, *Enterococcus faecium* NCIMB 11181 and *Lactobacillus plantarum* DSM 16568 (HOM+HET); and *Enterococcus faecium* NCIMB 11181, *Lactobacillus plantarum* DSM 16568 and *Lactococcus lactis* DSM 11037 supplemented with sodium benzoate at 400 g/ton forage (HOM+Chem). All inoculants were diluted with distilled water and applied at the same rate (4 ml solution/kg of crop) or/and 150 000 cfu/g fresh forage. The untreated control (C) received 4 ml of distilled water/kg of crop.

The 3-liter mini silos were filled with chopped L, R, RCR, RT and RCR with DM contents of: 328, 308, 317, 265 and 266 g/kg, respectively; Water soluble carbohydrate (WSC) contents: 49, 85, 92, 102 and 89 g/kg DM, respectively and crude protein (CP) contents: 229, 153, 199, 172 and 174 g/kg DM, respectively. Each treatment and crop were replicated five times and stored at 20°C for 90 days before determining DM, pH, DM losses, lactic acid (LA), acetic acid (AA), butyric acid (BA), ethanol, ammonia-N, and aerobic stability (AS), defined as a temperature increase of 2^{o}C above the ambient temperature. Data were statistically analyzed as a randomized complete block by using the GLM procedure of SAS.

Results

From a silage fermentation standpoint, ensiling lucerne, grass and clover-grass with three silage inoculants (HET, HOM+HET and HOM+Chem) offers the advantage of better-preserved silages (see Table 1). All inoculants reduced (P<0.05) the pH, increased (P<0.05) lactic acid and acetic acid formation (except RT product HOM+Chem) and decreased (P<0.05) DM losses in R, RCR, RT and RCR, silages compared to the untreated control. On an average, the use of additives reduced fermentation losses, by 2.7 percentage units (P<0.05) (variation from 0.5 to 6.6 percentage units) compared to the untreated control silages.

Lactic acid was the predominant fermentation product detected in RTand RCR silages inoculated with product HOM+Chem. Marked increases (P<0.05) in lactic acid contents in RT and RCR inoculated with HOM+Chem resulted in the highest lactic to acetic acid ratios among treatments. All additives resulted in reduced concentrations of ammonia-N, butyric acid and ethanol (P<0.05). Our results correspond well with results from laboratory experiments by Saarisalo *et al.* (2006) in which inoculation resulted in reduced pH and ammonia-N, and increased lactic acid production. The additives likely reduced respiration and/or proteolysis by plant enzymes, by changing the fermentation pattern and inhibiting the activity of clostridia and aerobic micro-organisms such as yeast and mould (Muck 2012).

The additives had the greatest effects on concentrations of lactic acid and acetic acid. The HOM+Chem resulted in silage with a higher lactic:acetic

Table 1. Effects of silage additives in a range of crops.

Item	L		R		RCR (70:30)		RT (70:30)			RCR (50:50)		
	C	HET	C	HET	C	HET	C	HET	HOM+ Chem	C	HOM+ HET	HOM+ Chem
DM (g/kg)	313	319*	292	301*	294	299*	250	252	253	239	250*	252*
DM loss (%)	6.8	4.6*	7.0	4.1*	10.2	6.7*	6.3	5.8	4.9*	12.3	6.7*	5.7*
pH	5.4	5.0*	5.1	4.3*	4.7	4.3*	4.6	4.2*	3.9*	5.6	4.7*	4.4*
NH3-N (g/kg N)	102	79*	59	45*	57	40*	60	51*	44*	92	52*	52*
LA (g/kg DM)	17	39*	23	40*	27	32*	31	34	72*	14	38*	57*
AA (g/kg DM)	34	49*	22	35*	29	38*	28	46*	19*	18	32*	33*
LA:AA	0.5	0.8	1.0	1.1	0.9	0.8	1.1	0.7	3.8	0.8	1.2	1.7
BA (g/kg DM)	14.0	1.0*	4.7	0.4*	6.5	0.3*	2.5	1.9	0.7*	37	1.9*	0.3*
Ethanol (g/kg DM)	12	7*	9	7*	11	7*	7	6*	5*	14	6*	4*
Yeast (log cfu/g)	-	-	2.9	1.0*	-	-	3.2	1.3*	1.5	-	-	-
Moulds (log cfu/g)	-	-	4.1	1.3*	-	-	3.0	1.3*	1.4	-	-	-
AS (hours)	-	-	63	257*	96	>168*	104	>312*	182*	198	>450*	397*

acid ratio. Inoculation with HET or HOM+HET resulted in lower lactic:acetic acid ratios. The higher amount of acetic acid in HET and HOM+HET silages was expected because the heterofermentative LAB *Lactobacillus buchneri* may result in high levels of acetic acid (Oude Elferink *et al.* 2001). Acetic acid reduces growth of yeast and mold in response to increase daerobic stability of silage. Growth of yeast and mold was reduced ($P<0.05$) by additives when determined.

All additives also resulted in increased aerobic stability ($P<0.05$) when exposed to air, and aerobic stability was increased from 48 to 216 hours compared with untreated silage. Aerobic stability of silage treated with additives containing *Lactobacillus buchneri* (HET and HOM+HET) was greatest among all treatments.

Conclusion

All silage additives resulted in improved fermentation, reduced DM loss and increased aerobic stability compared with untreated silage. Additives containing *Lactobacillus.buchneri* (HET and HOM+HET) improved aerobic stability more than HOM+Chem and had a more heterolactic fermentation pattern whereas the use of HOM+Chem resulted in a more homolactic fermentation pattern.

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