Chemical additives for silages: When to use it and what are the options?

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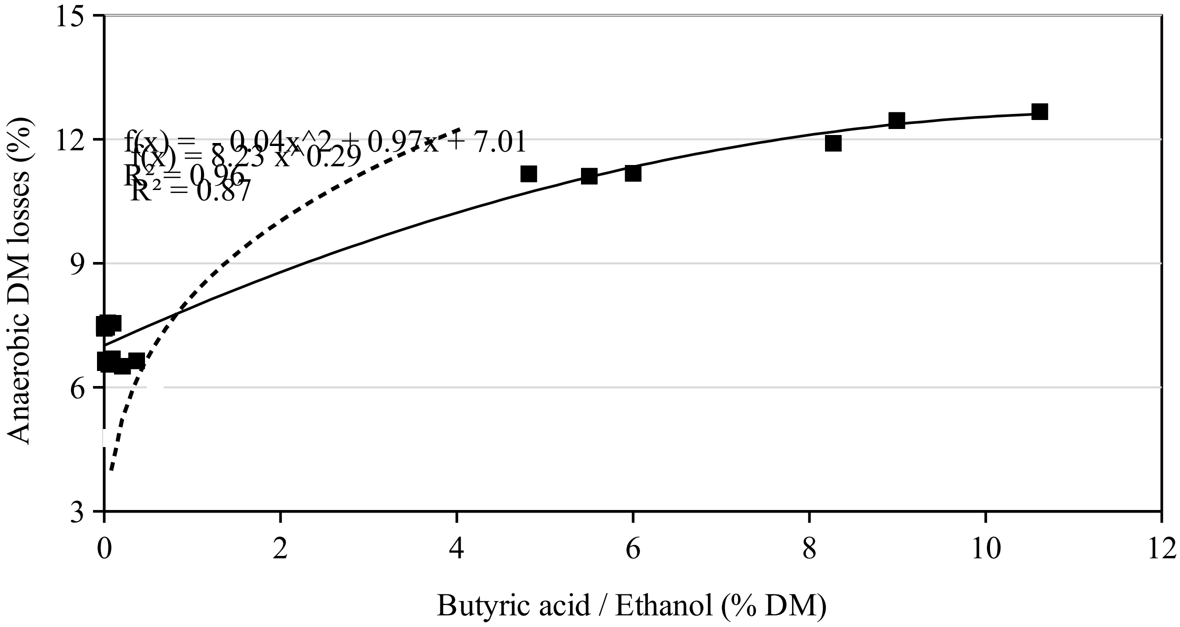
In memory of Professor Dr. Friedrich Weissbach

Introduction

Maintaining the highest possible quality of silage from field to trough poses one of the biggest challenges for dairy and beef producers worldwide as it requires thorough management to reduce dry matter (DM) losses to a minimum from field to feed-out. This is necessary to keep high animal performance and to ensure farm profitability. In addition to the well-known general principles of silage making, the use of additives has become a strategic management tool in silage production. Biological additives, mainly using different genera and species of lactic acid bacteria, have become the dominant additive type but chemical additives still play a major role in certain regions, especially in Europe. Published research provides evidence that there also has been an increasing interest in other areas of the world. This paper aims at providing guidance on which of the available active ingredients that can be used to solve a specific problem in silage production, and under which conditions those should be used.

Chemical additives to overcome the principal challenges for silage quality

Silage quality can be compromised by the activity of a range of undesired microorganisms, which grow and thrive under different conditions. In silages made from crops of high protein content and buffering capacity, especially if ensiled at low DM levels, the risk for secondary fermentation during the storage phase caused mainly by clostridia is most prominent. Their activities result in high DM losses, proteolysis as reflected by high concentrations of ammonia-N, and the formation of biogenic amines (Scherer et al., 2015; Auerbach et al., 2016; König et al., 2016; Borreani et al., 2018; König et al., 2018). However, sometimes high anaerobic DM losses can be associated primarily with the development of yeasts forming ethanol in excess of 10 g/kg DM, which can be observed mainly in high DM grass, whole-crop cereals and corn (Driehuis and van Wikselaar, 1996; Nadeau et al., 2013; Weiss et al., 2016; Auerbach et al., 2018,), although the contribution of other ethanol producers, e.g. heterofermentative lactic acid bacteria (LAB), should not be neglected (Rooke and Hatfield, 2003). Data depicted in Figure 1 show the relationship between butyric acid or ethanol concentrations and anaerobic DM losses (Auerbach et al., 2016; Auerbach et al., 2018). Additional DM losses by heating processes may be incurred during feed-out, which is usually initiated by yeasts. At later stages, bacilli, other aerobic bacteria and molds contribute to deterioration, thereby increasing temperature further and leading to reduced nutritive value and performance (Wilkinson and Davies, 2013; Auerbach and Nadeau, 2018a; Borreani et al., 2018), and mycotoxin formation (Auerbach and Theobald, 2018; Ferrero et al., 2019).



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igure 1. Relationships between the concentrations of butyric acid in silage from alfalfa and orchardgrass (■, solid regression line, n=24, Auerbach et al., 2016) or ethanol in silage from whole-crop rye (□, dotted regression line, n=18, Auerbach et al., 2018) and anaerobic DM losses.

There is a variety of chemicals available that are used as silage additives. These are usually classified according to their inhibitory effects on target microorganisms to improve fermentation or increase aerobic stability (ASTA) (Table 1). This classification has some drawbacks and should be looked at from a practical perspective as already Paracelus (1538) stated that “All things are poisons, and nothing is without poison; only the dosage is it that makes a thing not a poison”. This means that we will see effects of typical fungal inhibitors on undesired bacteria and of typical bacteria inhibitors on fungi given that the application rate is high enough (Table 2). However, cost per kg of active ingredient in combination with high dosage will ultimately increase treatment costs, rendering the use as economically unfeasible.

Table 1. Active ingredients in chemical silage additives (adapted from Auerbach et al., 2012).

Aim / type	Ingredient/species	Effect
Improving the fermentation process		
acids	formic	direct acidification, suppression of undesired spoilage bacteria
salts	calcium formate, sodium formate, ammonium formate, sodium nitrite, hexamethylene tetramine	suppression of undesired spoilage bacteria
Improving aerobic stability		
acids	sorbic, benzoic, propionic, acetic	inhibition of yeasts and molds
salts	sodium benzoate, potassium sorbate, ammonium propionate, calcium propionate, sodium propionate, sodium acetate	inhibition of yeasts and molds after release of respective acid during fermentation

Table 2. Proportion of silages free of butyric acid or with low DM losses as affected by silage additive use (10 trials with 3 replicates per treatment, adapted from Weissbach, 2010b).

Treatment	Proportion (%)	
	Silages free of butyric acid (< 0.3% of DM)[1)]	Low fermentation losses (DM loss < 8%)[2]
Control	33.3[a]	20.0[a]
Formic acid (85%, 4 l/t)	60.0[a]	82.8[b]
Sodium nitrite/Hexamine (3 l/t)[3]	100.0[b]	82.8[b]
Sodium benzoate (2 kg/t)	100.0[b]	100.0[c]
Sodium benzoate (2 kg/t) + Sodium nitrite (0.6 kg/t)	100.0[b]	100.0[c]
Significance[4]	$P < 0.001$	$P < 0.001$

[1]dry matter corrected for the loss of volatiles during drying; [2]n=29 for DM loss, [3]mixture of sodium nitrite (300 g/L) and hexamine (200 g/L); [4]Likelihood ratio Chi-Square test, frequency values with different superscripts differ (Fisher´s Exact Test).

Chemical additives to improve fermentation

Fermentability of the crop to be ensiled is one of the most important factors affecting the choice of additive type. Results of an evaluation adapted from Honig and Thaysen (2002) including 673 comparisons between treated and untreated silage are presented in figure 2. The underlying official German silage additive evaluation scheme (Pauly and Wyss, 2019) distinguishes between three classes within the aim-of-action (AoA) 1 – Improving the fermentation process – based on the crop ´s fermentability coefficient (FC). This parameter is calculated using the following equation: FC = DM (%) x 8 WSC/BC, where WSC is water-soluble carbohydrate concentration and BC is buffering capacity expressed as g lactic acid/kg DM required to acidify the silage to pH 4.0: 1) difficult to ensile, crops with insufficient WSC content and/or low DM, FC < 35; 2) moderately difficult to easy to ensile in the lower DM range, FC ≥ 35, DM < 35%; 3) moderately difficult to easy to ensile in the upper DM range, FC ≥ 35, DM ≥35%. It can clearly be seen that chemical additives, including acids or salts solely or mixtures of acids and salts, outperformed homofermentative LAB inoculants. The scoring system back in 2002 considered the parameters pH, acetic acid, butyric acid (sum of n- and iso-butyric acid, n- and iso-valeric acids and n-caproic acid all expressed in % of DM) and ammonia-N (% of total N), with 100 points being the maximum score.

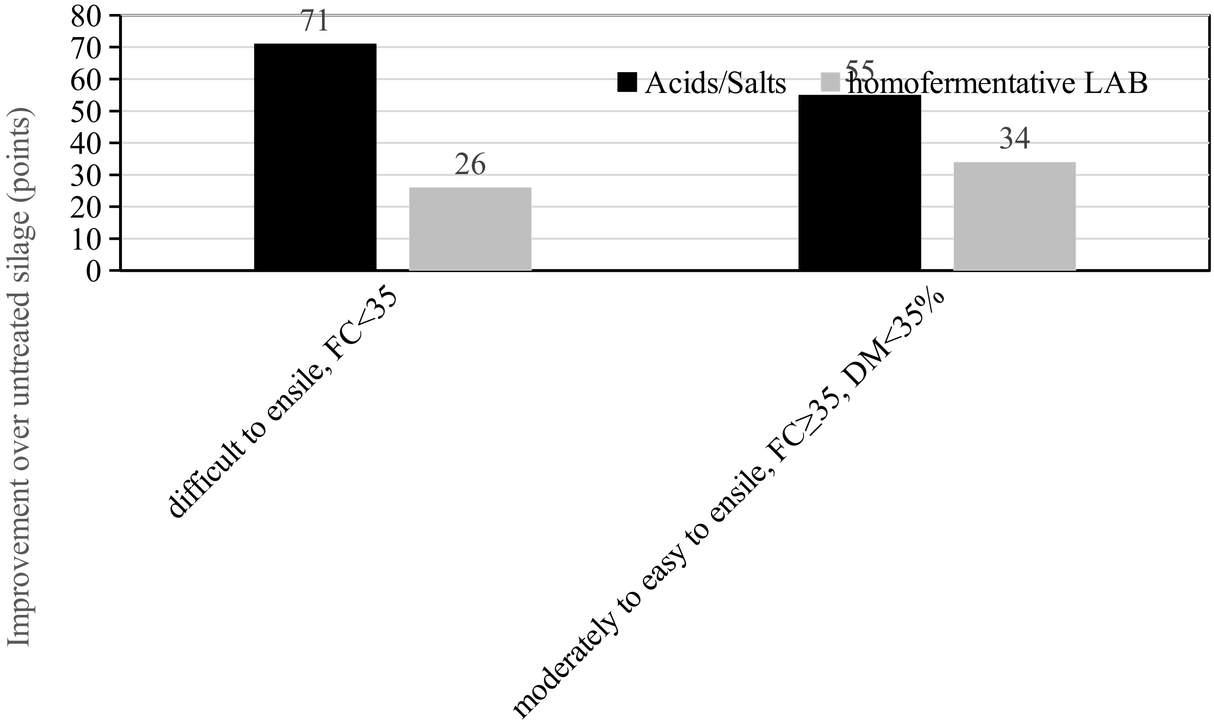

Figure 2. Effects of silage additives on fermentation quality as affected by fermentability based on the silage evaluation scheme of 2002 by The German Agricultural Society (DLG) (max. score 100 points) (Honig and Thaysen, 2002).

However, studies such as this one, or meta-analyses (Kleinschmit and Kung, 2006; Morais et al., 2017; Oliveira et al., 2017) although being very helpful regarding the assessment of general effects and their consistency, have serious limitations as they always use chemical and biological additives of different compositions applied to different crops stored under different conditions, which certainly has a significant effect on the results. Thus, this type of investigation is useful to characterize the potential of additive types, but not of given products. To evaluate commercially available products in terms of consistency of their effect on silage quality, a rather large number of trials (we advise to carry out at least 10 but the more the better) have to be conducted, covering a range of crops for which the additive is intended to be used, and DM concentrations.

From the historical point of view, formic acid has been the most important chemical additive to improve fermentation and numerous studies have been published on the positive effects on fermentation, and animal performance (Haigh and Parker, 1985; Steen, 1990; Nadeau et al., 2000a; Nadeau et al., 2000b; Broderick et al., 2007), which is still the main additive used in northern

Europe. However, due to its corrosive nature on metal and skin/eyes, partial buffering with ammonia or sodium has become very popular, and frequently it is blended with other chemicals, e. g. propionic and benzoic acids. However, improving handling properties comes at the expense of efficacy so that higher application rates are required when compared with pure formic acid (Randby, 2000).

Despite these developments, certain risks remain such as anectodal reports of machinery operators complaining about eye and lung irritation due to pungent formic acid smell when applied on the chopper, or while compacting. Usually, applications of acid-based products are done at the chute`s rear end in the top-deflector of self-propelled harvesters in order to avoid corrosion of the expensive machinery. However, this application location resulted in the largest variation of additive application rate and increased the frequency of samples that had received less than the intended dosage of 5 L/t, although the average dosage did not differ between application locations (Nysand and Suokannas, 2012). Moreover, effluent production posing an environmental risk is increased by pure and buffered formic acid-based products (O`Kiely, 1993; Jones and Jones, 1995,). Heavy compaction may stimulate effluent production and seepage during the early stages of fermentation. This is supported by empirical observations from the field where effluent is produced already during the filling period, which can last several days on large farms, and at DM contents at which seepage is not to be expected.

The aforementioned disadvantages of using formic-acid based products have been overcome by the use of combinations of sodium nitrite, one of the most potent clostridia inhibitors, and hexamethylene tetramine (hexamine), which was first introduced in the Eastern German market in the mid 1980ies after very thorough research under the brand name CEKAFUSIL, containing 300 g/L of sodium nitrite and 200 g/L of hexamine with a maximum application rate of 3 L/t of fresh matter (FM), depending on the crop DM at ensiling (Reuter and Weissbach, 1991). As demonstrated in table 3, this combination applied at 3 L/ t FM was as efficient as pure formic acid (85%, 4 L/t FM), but has no corrosive properties and does not stimulate effluent production.

Table 3. Effects of chemical additives on DM losses during fermentation, butyric acid production and clostridia counts (Weissbach, 2010a).

Parameter	Control	Nitrite/Hexamine[1] (3 l/t)	Formic acid (85%) (4 l/t)
DM loss[2] (%)	11.0	7.1*	6.2*
Frequency of silages with low			

DM losses[3] (%)	28	70*	73*
Butyric acid content (% FM)[2]	1.17	0.35*	0.45*
Proportion of butyric acid-free-silages[4](%)	27	74*	67*
Proportion of silages with low spore counts[5] (%)	44	71*	43

[1]containing sodium nitrite (300 g/L) and hexamine (200 g/L); [2]n=363, [3]DM loss ≤ 8%, n=291; [4]butyric acid < 0.2% of fresh matter (FM);[5]≤ 1,000 clostridia (MPN)/g, n=75; *denotes significant differences of treatment *vs.* control at $P< 0.05$.

Both formic acid/propionic acid-based additives and nitrite-containing additives in various blends with hexamine, benzoate, sorbate and propionate have been shown to be efficient to eliminate butyric acid production and decrease ammonia-N concentration in grass-clover silage contaminated with soil containing clostridia (Nadeau and Auerbach, 2014). Frequently, nitrite/hexamine-containing additives have been shown to be superior to formic acid-based products in terms of reducing clostridia contamination and butyric acid formation (Lättemäe and Lingvall, 1996; Lingvall and Lättemäe, 1999; Knicky and Lingvall, 2004; Knicky and Spörndly, 2009; König et al., 2016; König et al., 2018). The overall effect of the combination of nitrite and hexamine is caused by eradicating clostridia and their spores by nitrite and its decomposition products, nitric oxides, during the early phases and, at later stages, by the release of formaldehyde from hexamine, which is caused by a drop in pH. The findings by Weissbach (2010a) that hexamine addition to nitrite improved silage quality over that of untreated and silage that received nitrite only was recently opposed by König et al. (2018), who could not find an additional effect. However, this statement warrants caution as it was based on one experiment, where only one crop was ensiled at two different DM contents and the untreated silages contained very low butyric acid concentrations (<0.5 g/kg DM). Unpublished results by the University of Maringa, Paraná, Brazil on tropical grass, which is usually low in sugar and low in DM, showed that also in this crop chemical additives have the potential to improve the efficiency of the fermentation process as reflected by lower DM losses during fermentation by up to 60% when compared with untreated silage, and that the combination of sodium nitrite and hexamine was superior to the sole use of sodium nitrite (Table 4).

Table 4. Effects of additives on fermentation and DM losses of tropical grass (*Panicum maximum cv.* Mombaça) (University of Maringá, Paraná, Brazil, unpublished).

Item	Control	Soybean hulls[1]	Sodium nitrite[2]	Sodium nitrite + Hexamine[3]	Formic acid[4]	SEM	*P* value
pH	4.60^{bc}	4.89^{a}	4.66^{b}	4.79^{ab}	4.44^{c}	0.053	< 0.01
DM loss, %	15.8^{a}	12.9^{b}	9.2^{c}	6.5^{d}	7.1^{d}	0.07	< 0.01
Lactic acid (% of DM)	0.20^{c}	0.92^{bc}	1.72^{b}	2.99^{a}	3.71^{a}	0.323	< 0.01
NH_3-N (% of DM)	0.26^{a}	0.24^{a}	0.21^{a}	0.22^{a}	0.13^{b}	0.014	< 0.01
NH_3-N (% of DM)[5]	0.26^{a}	0.24^{a}	0.17^{b}	0.09^{c}	0.13^{bc}	0.014	< 0.01

[1]100 kg/t; [2]1 kg/t; [3]1 kg/t sodium nitrite + 0.65 kg/t hexamine; [4]85%, 4 L/t; [5]corrected for addition of nitrogen by additives.

Hexamine, which provides a long-lasting effect against clostridia, which may not have been eradicated by sodium nitrite. According to Auerbach et al. (2016), it can likely be replaced by other components, which remain in the silage. In their studies on alfalfa and orchardgrass ensiled at low DM (20-23%) with high ash contents (13-18% of DM), the sodium nitrite concentration was kept identical in all three different additives tested, but hexamine was replaced in two treatments by either sodium formate/sodium benzoate or ammonium formate/potassium sorbate with no difference in DM loss, fermentation pattern and biogenic amine formation. As shown by Nadeau et al. (2016) and Nadeau et al. (2019), both nitrite-containing and formic/propionic-based additives decrease proteolysis during ensiling of grass and grass-legume forages, as indicated by decreased concentrations of nonprotein N and ammonia-N content but increased proportion of cell-wall bound protein compared to untreated silage. Because additives containing sodium nitrite and/or hexamine and ammoniated acid mixtures directly add ammonia, for instance hexamine decomposes under acidic conditions into formaldehyde and ammonia, and nitrite is converted into nitric oxides and ammonia, the correction of the ammonia-N concentrations must be carried out in order to avoid false results because ammonia production originating from these aforementioned reactions is not caused by proteolysis (Table 5). It is obvious that the magnitude of the effect of NH_3-correction is bigger in low-nitrogen crops (Auerbach et al., 2012).

Table 5. Ammonia-N concentrations in silages as affected by correction for the ammonia applied with the additive (Auerbach et al., 2012).

Crop	Crude protein of fresh crop (% DM)	NH_3-N (% total N)		
		Untreated	Nitrite/Hexamine[1]	
			uncorrected	corrected[2]
Green rye, ear emergence	15.2	18.5	13.0	6.8
Grass, not fertilized	12.1	9.0	10.9	5.7
Grass, not fertilized	9.6	9.4	12.8	5.7
Grass/clover	15.3	9.2	11.6	8.6
Grass, not fertilized	9.5	10.7	6.9	1.0
Grass/clover	15.0	6.7	9.4	6.6
Alfalfa	15.5	13.4	12.2	9.5
Grass, late cut	8.9	7.7	10.5	4.9
Whole-crop barley	4.8	10.5	14.0	4.1

Grass, late cut	6.0	8.1	10.8	3.8
Whole-crop barley	4.9	10.1	11.5	3.6
Whole-crop barley	4.6	8.0	10.6	3.2
Grass, late cut	7.1	9.4	9.4	4.7
Grass, late cut	7.9	8.3	8.8	4.1
Alfalfa	22.9	8.3	6.8	5.2
Grass, fertilized	20.5	14.1	10.8	8.4
Grass, fertilized	15.1	17.1	14.2	9.8
Mean	12.1	10.5	10.1	5.3

[1]containing sodium nitrite (300 g/L) and hexamine (200 g//L) applied at 3 L/t; [2]corrected for the addition of NH_3-forming chemicals.

When additives are used to improve the fermentation process, also their effects on ASTA cannot be ignored as well-fermented silages tend to be particularly prone to aerobic instability. Particularly under challenging farm conditions of low feed-out rate, it is important to maintain the nutritive value until the silage is ingested by the animal (Auerbach and Nadeau, 2018a; Auerbach and Nadeau, 2018b). In series of 20 trials by Bader (1997), the use of nitrite/hexamine was shown to be superior to the application of a homofermentative LAB inoculant in terms of DM loss and clostridia contamination (Table 6). However, although the mean ASTA was not affected by treatment, inoculation increased the frequency of silages with low ASTA (≤ 3 days) from 24% in untreated silage to 42% ($P < 0.05$), whereas only 16% of the silages that received nitrite/hexamine had low ASTA ($P < 0.05$), and no difference to untreated silage was found. These data were confirmed by Honig and Thaysen (2002), who also did not observe an effect of acid/salt use on ASTA use but reported a significant decline by 1 day by treatment with homofermentative LAB.

Table 6. Effects of sodium nitrite/hexamine and homofermentative LAB on DM losses during fermentation, clostridia contamination and aerobic stability from 20 trials (Bader, 1997).

Parameter	Control	Nitrite/Hexamine[1] (3 l/t)	LAB_{ho}[2]
DM loss[3] (%)	6.1	4.6*	5.8*
Frequency of silages with DM losses < 5% (%)[3]	17	65*	23*
Frequency of silages with low spore counts (%)[4]	70	90*	60*

[1]containing sodium nitrite (300 g/L) and hexamine (200 g/L); [2]combination of two *Lactobacillus plantarum* strains applied at a total inoculation rate of 100,000 cfu/g herbage; [3]n=60, [4]≤ 10,000 clostridia (MPN)/g; *denotes significant differences between nitrite/hexamine or homofermentative LAB *vs.* control ($P < 0.05$).

Our own results from a total of six trials on grasses (three trials, 23.5 to 31.2% DM) and early-cut rye harvested before ear emergence (three trials, 25.0 to 38.8% DM) are presented in figure 3. They prove that inoculation with homofermentative LAB across trials reduced ASTA by 76 hours when compared with untreated silage ($P < 0.001$), and the use of nitrite/hexamine had no effect. Inoculation with homofermentative LAB, whose aim is to dominate the fermentation, results in a shift in metabolic end-products towards more lactic acid at the expense of butyric and acetic acids (Nadeau et al., 2018), which have an antifungal effect, but lactic acid can be utilized by a variety of yeasts as carbon and energy source (Jonsson and Pahlow, 1984; Santos et al., 2016). On the contrary, chemical additives including nitrite and hexamine do not selectively stimulate homofermentative LAB but allow the whole epiphytic LAB flora, which is never exclusively composed of homofermentative species, to develop and produce acetic acid/ethanol, in addition to lactic acid, given that the application rate is not too high to generally inhibit LAB.

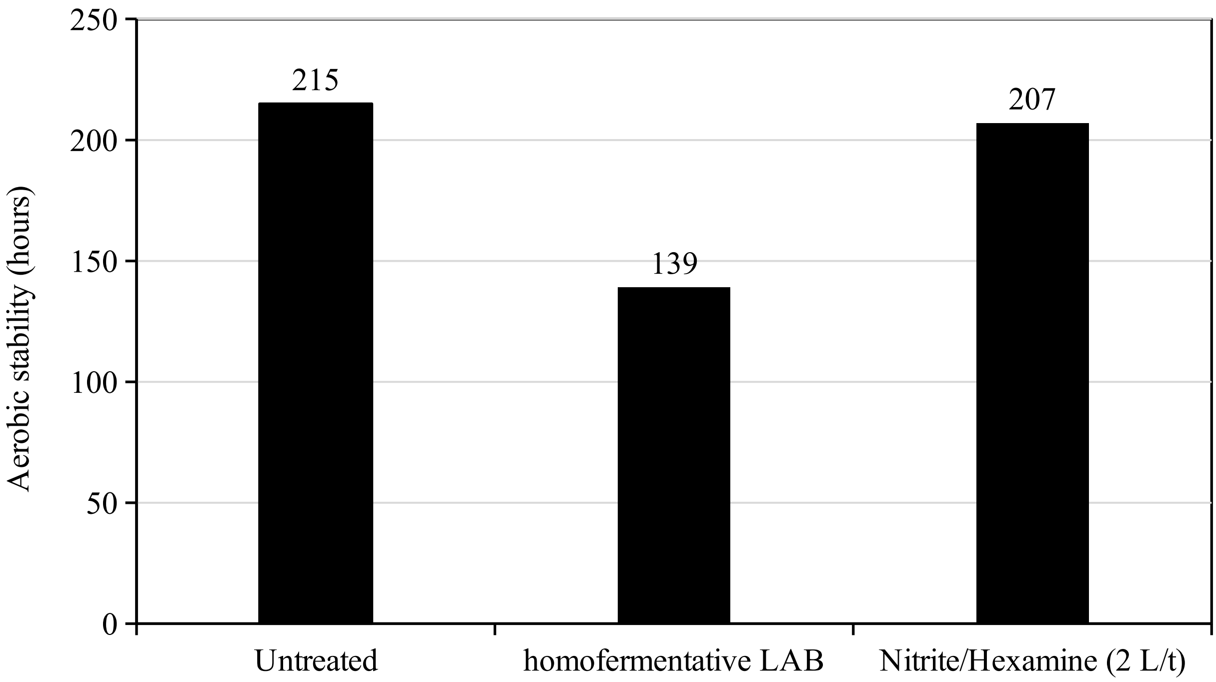

Figure 3. Effects of a liquid blend of sodium nitrite (300 g/L) and hexamine (200 g/L) applied at 2 L/t and a homofermentative LAB inoculant, applied at 1 g/t forage supplying 100,000 cfu/g forage of *Lactobacillus plantarum* DSM 16627 and 50,000 cfu/g forage of *Lactobacillus paracasei* NCIMB 30151, on the aerobic stability of silages from grasses (three trials) and early-cut rye (three trials) stored for > 90 days; $^{a\text{-}b}$bars bearing unlike superscripts differ at $P < 0.05$ (Tukey`s test) (Auerbach et al., unpublished).

The magnitude of the effect of chemical additives not only depends on the composition but also on the application rate. Any approach taken by the farmer "to purposely applying less than the recommended rates of application to save money is a dubious practice because it only increases the probability that the additive will not be successful" (Kung, 2009). It has been well known that application rate is crucial regarding the magnitude of the effect. Randby (2002) showed on grass silage that increasing application rates of a liquid unbuffered acid blend containing 640 g/kg formic acid, 93 g/kg propionic acid and 19 g/kg benzoic acid applied at 2, 3, 4 and 5 L/t forage increased residual water-soluble carbohydrate concentrations and ASTA. Similar dose effects on butyric acid concentrations were obtained by Custodio et al. (2016) on sugarcane silage, which was treated with a combination of lime ($CaCO_3$ at 15 kg/t) and graded doses of sodium nitrite (0.5, 1.0, 1.5 kg/t forage). This is in line with observations on a wide range of crops used in 21 trials by Weissbach (2010a) using a combination of sodium nitrite (300 g/L) and hexamine (200 g/L) applied at either 2 or 3 L/t (Table 7).

Table 7. Effects of application rate of a chemical additive on butyric acid formation in silages made from a variety of crops (n=63, Weissbach, 2010a).

Parameter	Control	Sodium nitrite / Hexamine[1] 2 L/t		3 L/t	
Butyric acid (% FM)					
Mean	1.07	0.36		0.15	
SD[2]	0.90	0.60		0.37	
Difference to control		-0.71	*	-0.92	*
Difference to lower dosage				-0.21	
Frequency of butyric acid-free (< 0.2% of FM) silage					
Mean	30	67		81	
Difference to control		37	*	51	*
Difference to lower dosage				14	

[1]containing sodium nitrite (300 g/L) and hexamine (200 g/L); *denotes significant difference at $P < 0.05$; [2]standard deviation.

Chemical additives to improve aerobic stability

Aerobic deterioration of silage has become a major problem in silage production on many farms in the world, with corn silage being considered the most vulnerable silage type but also silages from grasses with high sugar concentrations or from whole-crop cereals can undergo severe spoilage process during feed-out. Since its introduction to the markets in the late 20th century, the heterofermentative *Lactobacillus buchneri* either applied solely or in combination with homofermentative LAB have become very popular globally due to the improvements in ASTA (Kung et al., 2003; Kleinschmit and Kung, 2006; Muck et al., 2018). In addition to acetic acid production from sugar, which is facilitated by all obligate heterofermentative LAB species, *Lactobacillus buchneri* also anaerobically converts lactic acid to antifungal acetic acid, 1,2-propanediol and ethanol (Oude-Elferink et al., 2001). This metabolic pathway increases DM losses during the anaerobic fermentation phase and requires a minimum of 6-8 weeks of storage before silo opening in order to have sufficiently high acetic acid concentrations to inhibit fungi consistently. In a meta-analysis by Kleinschmit and Kung (2006), an overall decrease in DM recovery by *Lactobacillus buchneri* inoculation was observed in silages from corn or grass/small grain silages, which depended on the inoculation rate. Usually though, the increased DM losses during storage offset those incurred by fungi under aerobic conditions. Honig and Thaysen (2002) found a significant increase in DM losses by *Lactobacillus buchneri* treatment by 10% (n=173) compared with untreated, whereas the application of chemical additives of unspecified composition did not have an effect. No difference in the magnitude of the effect on ASTA were found, ranging between +2.4 days for heterofermentative LAB and +2.2 days for chemicals when compared with untreated silages.

Under certain circumstances, such as DM concentrations below 30% (Auerbach and Weiss, 2012; Gomes et al., 2018) excessive acetic acid concentrations can be produced. Occasionally, also at typical DM contents ranging between 30 and 40%, very high concentrations of this organic acid can be detected, leading to much higher DM losses than usually seen compared with untreated (Driehuis et al., 1999; Kleinshmitt et al., 2013; Auerbach and Nadeau, 2018b). On the contrary, lower DM losses were found when untreated silages underwent ethanolic fermentations, which can be prevented by *Lactobacillus buchneri*, as reported for sugarcane silage by Rabelo et al. (2018) and for whole-crop rye by Auerbach et al. (2018). When tested in the same trial, however, chemical additives outperformed *Lactobacillus buchneri*-type products regarding anaerobic DM losses (Auerbach and Weiss, 2012; Auerbach and Nadeau, 2018a; Auerbach and Nadeau, 2018b).

The question as to which chemical is best to improve silage ASTA is not quite easy to answer. It has been well documented that the minimum inhibitory concentration (MIC) of organic acids and their salts depend on the microorganism tested, and the pH of the medium (Woolford, 1975a; Woolford, 1975b; Auerbach, 1996; Stanojevic et al., 2009). Reducing pH from 5 to 4 decreased MIC (mmol/L) against yeasts and molds for each chemical investigated, but the following order remained unchanged: potassium sorbate < sodium benzoate < propionic acid < acetic acid < formic acid (Woolford, 1975a, Woolford, 1975b). Although *in vitro* screening tests are very useful to determine the relative differences between chemicals, the MIC values derived from those tests should be treated with caution, and due to the complexity of the silage ecosystem they can very likely not be directly extrapolated to a silage environment. Moreover, the tested microorganisms may not even be found in silage. Driehuis and van Wikselaar (1996) conducted one trial on grass and maize silage using equimolar concentrations of formic (3.3 kg/t), acetic acid (4.3 kg/t) or propionic acid (5.3 kg/t) and concluded that formic acid improved ASTA in maize silage but not in grass silage, in which propionic acid had the best effect. Acetic acid failed to increase ASTA in both trials. The lack of response to propionic acid in maize silage, despite the lowest yeast count, was explained by the fact that acetic acid bacteria and not yeasts caused silage to deteriorate, whereas in grass silage acetic acid bacteria were below the detection limit in all treatments. However, only testing the organic acids at the same application (kg/t) would have made a direct comparison possible. Our own data presented in Figure 4 on corn ensiled at 41% DM and stored for 59 days with air ingress for 24 hours on day 28 and 52 show that already the lowest concentration of potassium sorbate (500 g/t) improved ASTA, which substantiates recent findings by Huenting et al. (2018) using potassium sorbate at 400 g/t. On the contrary, 1000 g/t of sodium benzoate were required to show an effect on ASTA, supporting data by Kleinschmit et al. (2005) who detected a similar effect for potassium sorbate at 500 g/t and sodium benzoate at 1000 g/t.

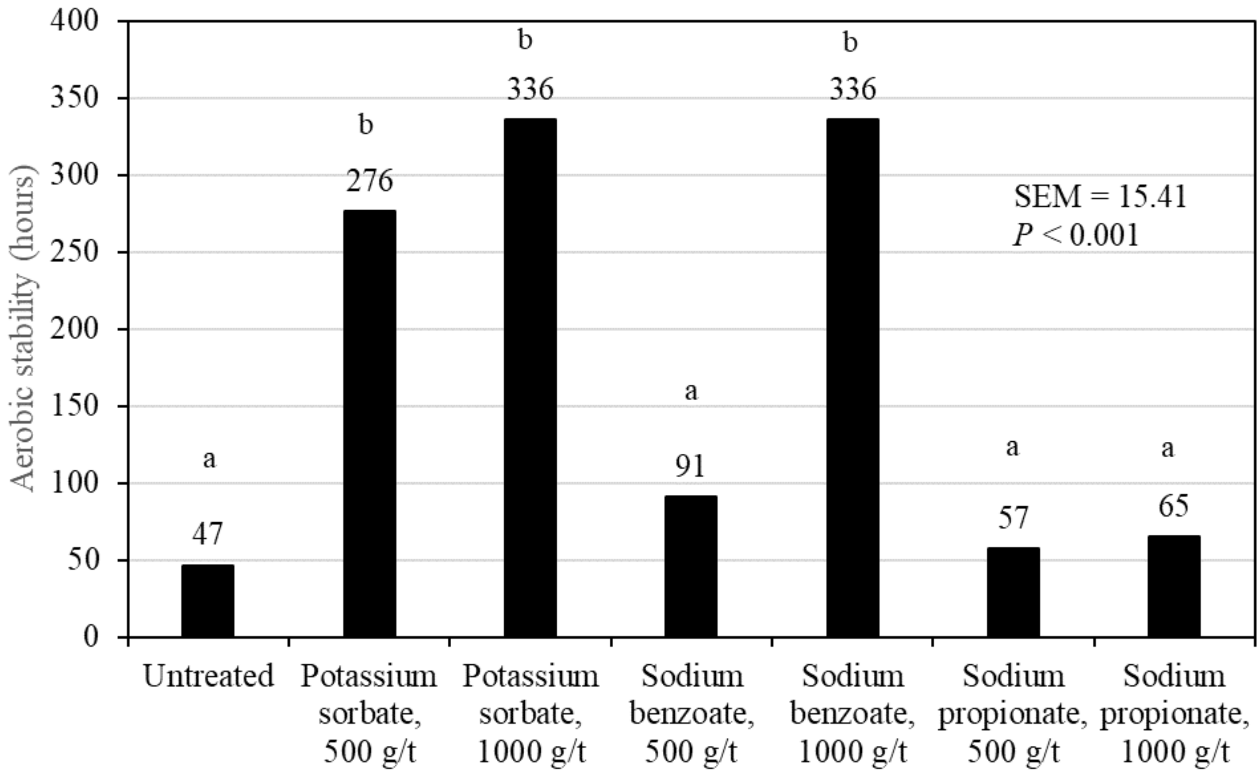

Figure 4. Effects of antimycotic chemical additives applied at 500 or 1000 g/t on the ASTA of corn silage ensiled at 41% DM and stored for 59 days with air ingress for 24 hours in day 28 and 52 during subsequent air exposure for 336 hours after silo opening. [a-b]bars with unlike superscripts differ at $P < 0.001$ (Tukey´s test, n = 3)) (Auerbach et al., unpublished).

Moreover, our data confirm those by Teller et al. (2012) who found improved ASTA in corn silage by adding potassium sorbate applied at 1000 g/t, but the magnitude of the effect was lower in their trial (+121 hours higher ASTA compared with untreated). Da Silva et al. (2014) reported higher ASTA of corn silage treated with sodium benzoate applied at 2000 g/t when compared with untreated silage, or those that were inoculated with *Lactobacillus buchneri*. Furthermore, Bernardes at al. (2014) tested 1000 g/t and 2000 g/t of either potassium sorbate or sodium benzoate in corn silage ensiled at 37% DM and found a dose-response regarding the onset of aerobic instability (silage temperature +2 °C above ambient), but no difference between the chemicals within application rate. However, yeast development differed. At the end of aerobiosis, silage treated with potassium sorbate had the lowest yeast counts indicating that growth was retarded. Sodium propionate did not improve ASTA in our study regardless of the application rate. In a trial by Kung et al. (2000) on corn, a dose-response to the application of a buffered propionic acid-based product (1000 to 3000 g/t) was observed but only the highest dosage had a significant effect over that of untreated silage.

Single-component chemical additives to minimize aerobic deterioration are rarely used in practical farming. It has been much more common to combine different active ingredients, which makes it very difficult, if not impossible, to directly compare the results. Blending different ingredients aims at optimizing costs due to large differences in price between raw materials, at extending the range of microorganisms to be inhibited due to potential differences between substances, and sometimes at creating synergistic effects, but these have only been reported for a mixture of sodium nitrite and sodium benzoate on *Candida albicans* and not for other important silage yeasts or mold species (Stanojevic et al., 2009).

The meta-analysis on the effects of chemical additives carried out by Morais et al. (2017) on high-moisture corn (HMC) or high-moisture winter cereal grains from wheat, barley and rye (HMWCG) revealed improvements in ASTA by a range of additives of different compositions and application rates by 131 hours in HMC ($P < 0.01$) and by 116 hours in HMWCG ($P = 0.10$) but this study also does not allow to draw conclusions on the superiority of any tested product. Combinations of sodium benzoate and potassium sorbate alone (Auerbach and Weiss, 2012; Auerbach and Nadeau, 2013; Weiss et al., 2016), or with sodium nitrite (Knicky and Spörndly, 2009, Knicky and Spörndly, 2011, Auerbach and Nadeau, 2013; Knicky and Spörndly, 2015; da Silva et al., 2015; Kung et al., 2018,); or ammonium propionate (Auerbach et al. 2015; Nadeau et al., 2015; Schneider et al., 2018) have consistently been shown to improve ASTA, and a dose-response was usually observed, highlighting the importance of the added application regarding the magnitude of the effect. In a series of corn silage trials on the effects of storage conditions and the addition of a mixture of potassium sorbate (134 g/L), sodium benzoate (257 g/L) and ammonium propionate (57 g/L) applied at 1 and 2 L/t by Auerbach et al. (unpublished) (Figure 5), it was found that under challenging conditions (four trials, air ingress for 24 hours on day 28 and one week before silo opening, max. storage length 63 days) only the dosage of 2 L/t was successful to improve ASTA. On the contrary, if corn silage was stored strictly anaerobically for > 90 days (two trials), already the lower application rate increased ASTA, highlighting the interaction between storage conditions and required dosage (Weber et al., 2006). Despite positive effects of formic acid-based additives on ASTA of corn silage, their use should be scrutinized because of frequently detected stimulation of ethanol formation, leading to higher anaerobic DM losses (Auerbach et al., 2012; Weiss and Auerbach, 2012; Weiss et al., 2016).

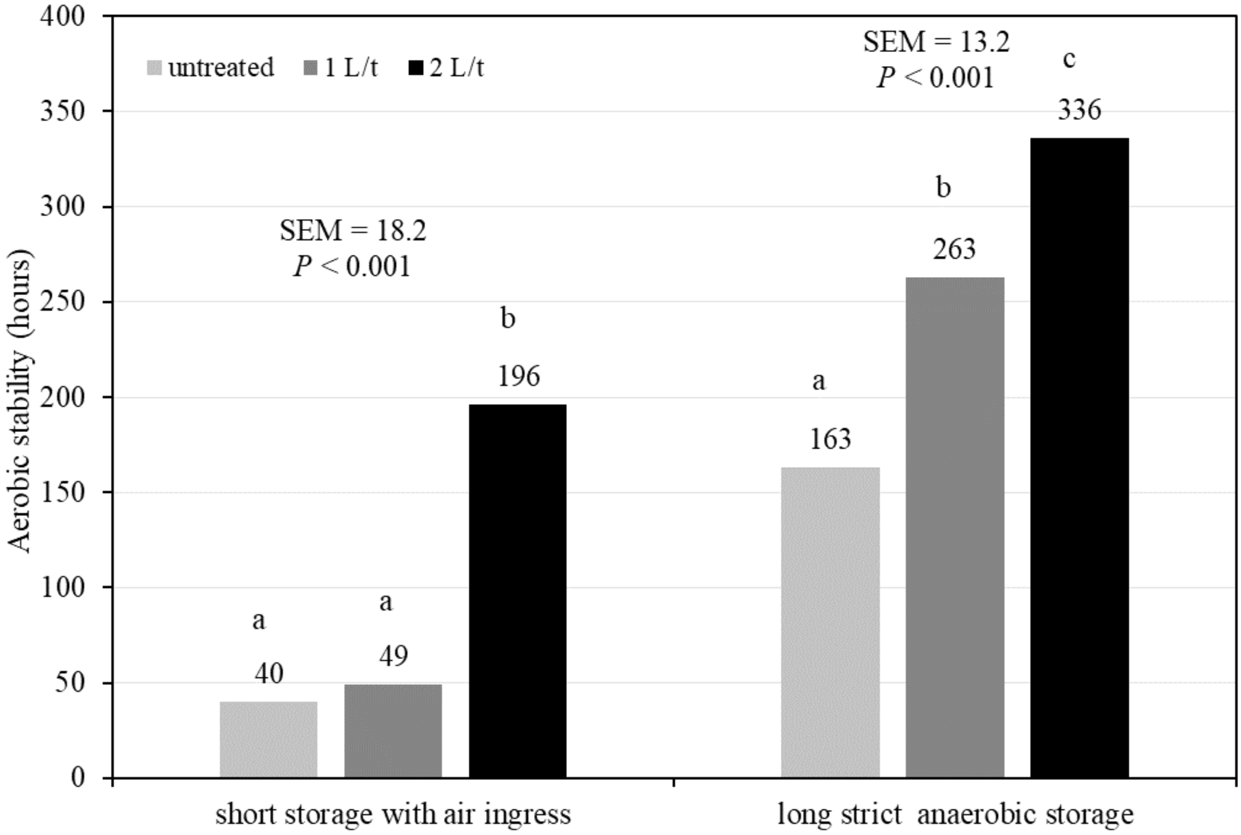

Figure 5. Effects of the application rate of an antimycotic chemical silage additive composed of 257 g/L sodium benzoate, 134 g/L potassium sorbate and 57 g/L ammonium propionate on the aerobic stability of corn silages stored under different conditions (short storage of max. 63 days with air ingress for 24 hours on day 28 and one week prior to silo opening (n = 12) or strict anaerobic storage for > 90 days (n = 6)). [a-c]bars with unlike superscripts differ at $P < 0.05$ (Tukey´s test) (Auerbach et al., unpublished).

In order to overcome the effects on ASTA by qualitative and quantitative differences in composition between commercial chemical additives, and to explain why certain products perform better than others, Auerbach and Nadeau (2013) have introduced the concept of "sodium benzoate equivalents" (SBE). This concept is based on results by Auerbach (1996) who studied the effects of sorbate, benzoate and propionate at pH 4 on the growth of the most important silage mold in temperature climate, *Penicillium roqueforti*. It assumes a relative effect size of 0.5:1:2 for potassium sorbate, sodium benzoate and sodium propionate. The relationship between SBE and ASTA was first described by Auerbach and Nadeau (2013) using data from a corn silage trial. The power of the curvilinear relationship ($R^2 = 0.85$, $P < 0.01$) supported the author´s assumptions. More recently, we applied this approach to another corn silage trial (Auerbach et al., 2017), in which two sodium benzoate/potassium sorbate containing additives supplemented with either sodium nitrite or ammonium propionate were applied at different dosages and confirmed previous findings (Figure 6). With increasing application rate of SBE, the count of yeast was reduced and, concurrently, the ASTA was improved. However, further studies are warranted to substantiate the general validity of this interesting approach.

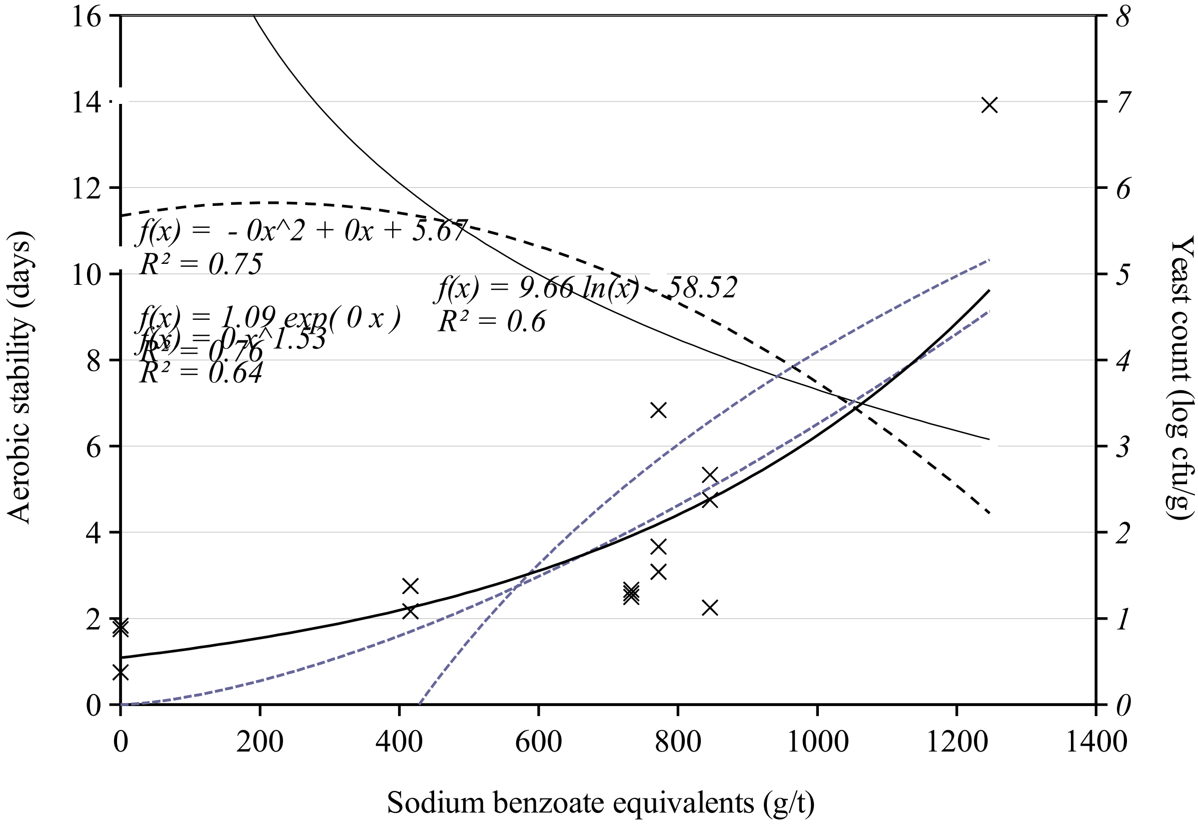

Figure 6. Relationship between the concentration of sodium benzoate equivalents and the counts of yeasts (□, dotted regression line, n = 18) and the aerobic stability (■, solid regression line, n = 18) in corn silage ensiled at 30% DM and stored for 63 days with air ingress for 24 hours on day 28 and 57 followed by air exposure for 14 days after silo opening (adapted from Auerbach et al., 2017).

Under conditions of short storage length, the use of antimycotic chemical additives seems warranted due to the high risk of aerobic spoilage during feed-out. Data by Kung et al. (2018) from corn silage and da Silva et al. (2015) from HMC using a chemical blend of potassium sorbate, sodium benzoate and sodium nitrite support our own observations on corn silage ensiled at 27% DM and stored for up to 142 days before aeration (Figure 7). The ASTA was improved over untreated silage by the use of a liquid mixture of potassium sorbate, sodium benzoate and ammonium propionate already after 7 days and this effect persisted until day 34. With increasing storage length ASTA increased ($P < 0.001$), but an additive-by-fermentation length interaction was observed ($P < 0.01$). Delayed sealing, however, had a pronounced effect on ASTA, highlighting the importance of fast sealing of silos.

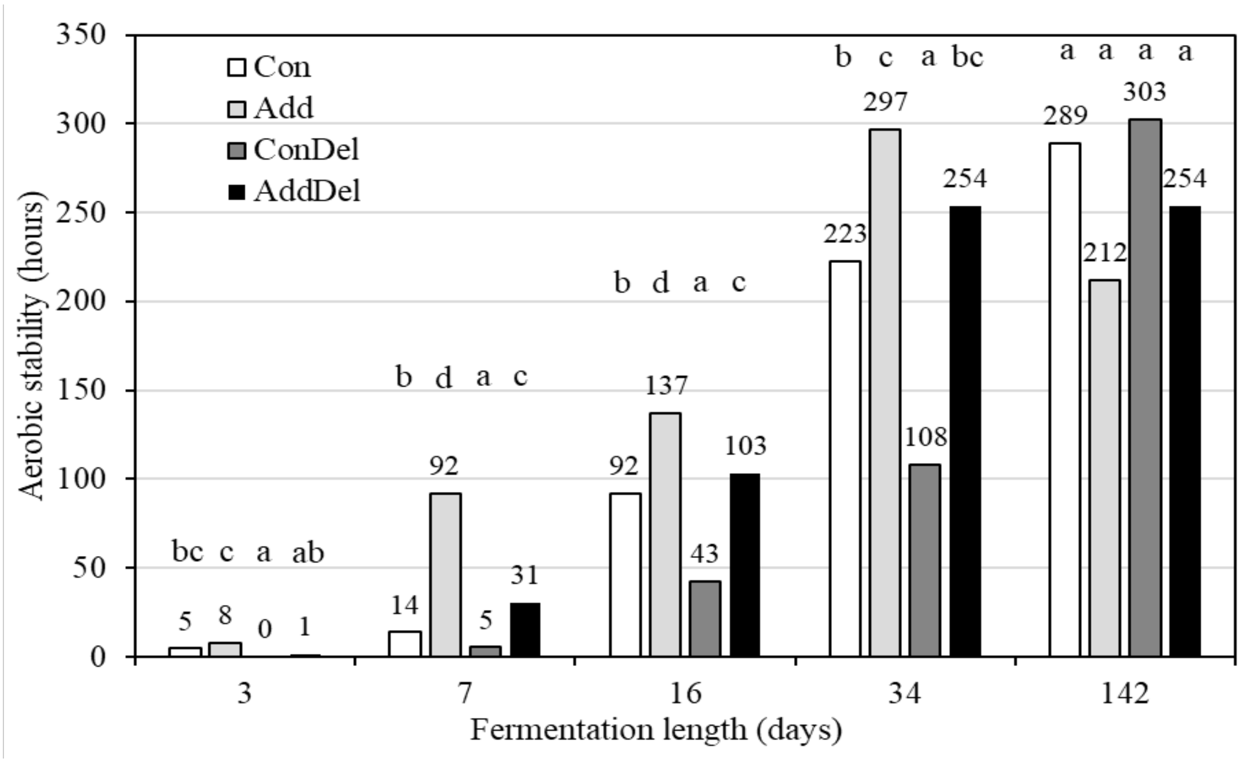

Figure 7. Effect of delayed sealing and the use of a chemical additive composed of 257 g/L sodium benzoate, 134 g/L potassium sorbate and 57 g/L ammonium propionate applied at 2 l/t on aerobic stability of corn silage ensiled at 27% DM and stored for 3, 7, 16, 34 and 142 days. Con = untreated, sealed immediately, Add = chemical additive, sealed immediately, ConDel = untreated, sealed with a delay of 24 hours, AddDel = chemical additive, sealed with a delay of 24 hours; Effects of treatment: day 3, SEM = 0.75, $P < 0.001$; day 7, SEM = 0.94, $P < 0.001$; day 16: SEM = 1.73, P < 0.001; day 34: SEM = 15.73, $P < 0.001$; day 142, SEM = 34.94, $P = 0.326$. [a-d]bars with unlike superscript within fermentation length differ at $P < 0.05$ (Tukey´s test, n = 3) (Auerbach et al., unpublished).

Recent studies on the use of obligate heterofermentative LAB other than *Lactobacillus buchneri*, e.g. *Lactobacillus diolivorans* and *Lactobacillus hilgardii,* to evaluate whether silos could be opened earlier than 6-8 weeks, have shown their potential to improve ASTA (Huenting et al., 2018; Thaysen and Kramer, 2018). However, their effects were highly variable (Ferrero et al., 2018a; Ferrero et al., 2018b) and seemed to be largely affected by crop and DM concentration, and the effect averaged over five corn silage studies was low (<1 day) when compared with untreated silages stored for up to 30 days before aeration.

Chemical additives and animal performance

In addition to decreased DM losses and improved aerobic stability, chemical additives have the potential to improve animal performance. Formic acid restricts fermentation and protein degradation during ensiling, which have shown to increase intake by 1.0 kg/day and daily live-weight gain (LWG) by 270 g of steers when fed direct-cut grass silage treated with 3.3 L/t of formic acid (Winters et al., 2001). Likewise, compared to untreated alfalfa silage, Broderick et al. (2007)

reported decreased proteolysis and, thereby, lower contents of soluble nonprotein N, ammonia N and free amino acid N in alfalfa silage treated with ammonium tetraformate (7 L/t). When fed to dairy cows, the daily DM intake increased by 1.0 kg and the 3.5% fat-corrected milk increased by 2.1 kg. Content and yield of milk true protein and nitrogen efficiency in milk N per unit of N intake were also increased (Broderick et al., 2007). However, this production response was not observed in a second trial. In an experiment by Agnew and Carson (2000), beef steers were fed unwilted grass silage untreated or treated with a blend of ammonium hexamethanoate, ammonium hexapropionate and octanoic acid (6 L/t) ad libitum. The additive increased silage intake, which resulted in an increased carcass gain. Also, carcass conformation and fat grade were higher in steers fed the treated silage (Agnew and Carson, 2000). In a dairy cow experiment, where ammonium propionate at 5 L/t was applied to corn silage, no effect on intake or milk yield was observed (Levital et al., 2009). Furthermore, Diaz et al. (2013) found no improvements in diet digestibility, nitrogen balance, liveweight gain (LWG) or carcass quality of finishing steers fed ammoniated high-moisture ear corn. However, Nadeau and Arnesson (2016) reported increased live weight at birth (6.0 vs. 5.2 kg) and a tendency to increased LWG until weaning (442 vs. 409 g/day) in lambs suckling ewes fed grass-clover silage treated with an additive, containing sodium nitrite, hexamine, sodium benzoate, potassium sorbate and sodium propionate applied at 2 L/t. The same additive (2 L/t) was used on grass-legume silage fed to dairy cows (Nadeau et al., 2014). The additive decreased milk urea content (230 vs. 240 mg/L, $P < 0.001$) and tended to increase the excretions of purine derivatives in urine (115 vs. 95 g/day), suggesting an increase in the microbial protein flow to the duodenum. Furthermore, cows fed the treated silage had lower somatic cell counts in milk (52,000 vs. 92,000 per mL, $P < 0.05$). The improved performance of the cows was attributed to decreased proteolysis and increased sugar content of treated as compared to untreated silage (Nadeau et al., unpublished; Nadeau et al., 2014). In a later experiment (Nadeau et al., 2015b), dairy cows were fed grass-clover silage treated with the same additive or untreated silage in diets differing in rumen undegradable protein (RUP, 4.9 vs. 2.9% of DM at 15% CP of DM). The additive-treated silage produced 3.1 and 3.4 kg higher yields of milk and ECM, respectively, compared to untreated without affecting intake in the low RUP diets. The improved performance of the cows on additive-treated silage when fed a diet with low RUP could partly be related to the additive decreasing proteolysis and increasing sugar content of the silage as compared to untreated silage (Nadeau et al., unpublished; Nadeau et al., 2015b).

Conclusions

Chemical silage additives play an important role in ensuring high silage quality from field to trough. The decision on what additive to use needs to be based on the target microorganism to be inhibited. Formic-acid based products and mixtures containing sodium nitrite and hexamine are recommended to improve the fermentation process by inhibiting clostridia, whereas potassium sorbate, sodium benzoate and salts of propionic acid improve the ASTA of silages by suppressing yeasts and molds. It is crucially important to use a sufficient quantity of active ingredients in order to make the best use of the potential of the additives. The additive that is most suitable to achieve the intended goals will not only depend on its effect and consistency, but also on factors such as storage conditions, handling properties and safety to animal, user and environment, and the cost per tonne of treated forage. If applied properly, chemical additives will minimize qualitative and quantitative losses, thereby improving animal performance and farm profitability.

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