





Original Article

Effect of Energy, Protein and Microbial Inoculants Additives on Chemical Composition and Fermentation Characteristics of Corn Stover Silage

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ABSTRACT

Corn stover was taken after harvesting the ears immediately, chopped using a harvester chopper machine to 1.5-2.0 cm of length, and supplemented with energy additive (corn grains and molasses), protein additive (soybean, urea, and optegin) and microbial inoculants (effective microorganisms EM1) and their interactions and ensiled in plastic bags for 45 days. After the ensiling period, representative samples were taken for determination of chemical composition and silage quality traits. Adding ground corn grains to corn stover silage led to a significant (P<0.05) increase in DM content. Moreover, NFE content of corn stover silage increased significantly (P<0.05), however, ash content decreased significantly (P<0.05) with molasses and ground corn grains supplementation. Soybean meal supplementation increased significantly (P<0.05) DM content than those of urea and optigen supplementation

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with insignificant differences with control. The crude protein content of corn stover silage increased significantly (P<0.05) with soybean meal, urea, and optigen supplementation. Ash content decreased significantly (P<0.05) with soybean meal supplementation. Effective microbes supplementation led to a significant (P < 0.05) increase in the contents of CP and NFE of corn stover silage and a significant (P<0.05) decrease in the contents of CF and ash. The interaction between energy and protein supplementation showed a significant difference (P<0.05) in DM content only. Interactions among energy, protein, and effective microbes supplementation showed a significant differences (P<0.05) in DM content only. The pH value of the silage decreased significantly (P<0.05), however lactic acid concentration increased significantly (P<0.05) with molasses and ground corn grains supplementation. The pH value of silage was higher significantly (P<0.05) with urea and optigen compared to soybean supplementation. Urea-supplemented silage recorded control and significantly (P<0.05) the NH₃-N concentration followed by optigen, then soybean meal, whoever control had the lowest concentration. Moreover, soybean mealsupplemented silage showed significantly (P<0.05) higher concentrations of TVFA's and lactic acid compared to urea-supplemented silage. The concentrations of NH₃-N and lactic acid increased significantly (P<0.05) with effective microbes supplement to corn stover silage. Energy and protein supplementation interaction revealed that urea with molasses or ground corn grains recorded significantly (P<0.05) higher pH values and NH₃-H concentration of silage compared to soybean with molasses or ground corn grains. However, soybean meal with molasses or ground corn grains recorded significantly (P<0.05) higher concentrations of TVFA's and lactic acid in silage compared to urea with molasses or ground corn grains. The interaction between protein and effective microbes showed that urea with EM1 supplemented silage had significantly (P<0.05) higher pH value and NH₃-N concentration and lower TVFA's and lactic acid concentrations than those of soybean meal with EM1. The interaction among energy, protein, and effective microbes showed that molasses or ground corn grains with urea plus EM1 supplemented silages had significantly (P<0.05) higher pH value and NH₃-N concentration and lower TVFA's and lactic acid concentrations than those of molasses or ground corn grains with soybean meal plus EM1.

Keywords: corn stover silage, feed additives, composition, fermentation.

INTRODUCTION

Considering the real climate conditions, silage is the best method for preserving fresh forage with minimal losses. Silage quality and nutritional value are influenced by numerous biological and technological factors, when the proper ensilage techniques are used, silage will have a high nutritive value and hygienic quality (Sariçiçek and Kiliç, 2009). However, the results in practice indicate that the quality of silage is often poor or even unsatisfactory. These results are usually achieved when the fermentation

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condition is difficult (Lattemae et al., 2006). Factors that influence fermentation include the degree of green fodder wilting, length of the cut, ensiling technology type, and amount of an additive used (Haigh, 1988). Silage additives are natural or industrial products added in rather large quantities to the forage or grain mass. The purpose of silage additives is to control the preservation process so that by the time of feeding it has retained as many of the nutrients present in the original fresh forage as possible and to ensure that the growth of lactic bacteria predominates during the fermentation process, producing lactic acid in quantities high enough to ensure good silage (Oliveira, 1995). Additives are used to improve the nutrient composition of silage, to reduce storage losses by promoting rapid fermentation, to reduce fermentation losses by limiting the extent of fermentation, and to improve the bunk life of the silage (increase aerobic stability). It is widely accepted that silage additives can increase animal intake and animal performance through their effect on silage quality (Merry et al., 1993). However, the exact nutrient status of the silage will depend on many factors that can only be controlled via management. It is important to remember that silage additives will not make poor-quality forage into good silage but they can help make top-quality forage into excellent-quality silage (Kenilworth and Warwickshire, 2012). Many different silage additives are available and are used for different reasons. It includes fermentation stimulants, fermentation inhibitors, aerobic deterioration inhibitors, nutrients, and absorbents (McDonald et al., 1991). Their main functions are to either increase the nutritional value of silage or improve fermentation so that storage losses are reduced.

Soybean meal is a popular source of nitrogen for farmers globally. However, with increasing costs associated with its purchase and an understanding of the environmental impact that the production and transport of soybeans can have, some farmers are looking to move away from soybean as a primary source of nitrogen for their livestock. Soybean meal is generally sourced from areas that have undergone significant land use changes to meet the demand for feeding livestock (Caro *et al.*, 2018). Areas such as the Amazon have experienced significant deforestation to clear room for grazing or crop production (FAO, 2012). Land use change emissions from soybean agriculture are estimated to cover 3.2% of global emissions from livestock (Gerber *et al.*, 2013). There are also several associated negative environmental impacts, including habitat loss and decreased biodiversity (Dalgaard *et al.*, 2008). Although attributing land use change emissions is a complicated process, there is a consensus that soybean meal has a particularly high footprint. Therefore reducing the levels of soybean meal in the diet of a farmer's cattle is likely to reduce the lifecycle emissions of their finished product (Lehuger *et al.*, 2009).

Optigen is a slow-release urea product that is intended to supply nitrogen to ruminants. The product is a non-protein nitrogen source, which concentrates nitrogen supply to the animal to improve ruminant efficiency. In this report, we set out the key

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issues, provide an opinion regarding the addition of Optigen into feeds, and summarise the evidence base used to form that opinion. We focus our opinion on productivity and nutrition rather and also refer to the potential for reduced CH4 emissions that come with a more efficient rumen (FAO, 2017).

Optigen is significantly more concentrated than soybean meal, allowing for increased dry matter space in the rumen. When Optigen replaces soybean meal as a source of nitrogen, soybean meal is effectively replaced at 1:5.5 when crude protein content is taken into account. Cattle require sources of nitrogen to build amino acids which support tissue growth and milk production. This is mainly achieved through sources of protein such as soybean meal which can have a negative environmental impact, both in terms of greenhouse gas emissions and wider environmental concerns. Sources of nitrogen are generally the most expensive component of feed and therefore seeking alternatives to products such as soybean meal could be both lucrative to farmers and also reduce environmental impact (Kazer and Madoc-Jones, 2019).

The objective of the present study was to investigate the effect of energy, protein, and microbial inoculants additives on the chemical composition and fermentation characteristics of corn stover silage.

MATERIALS AND METHODS

The current work was carried out at Sakha Animal Production Research Laboratories, belonging to Animal Production Research Institute. Agricultural Research Center, Ministry of Agriculture in cooperation with Department of Animal Production, Faculty of Agriculture, Kafr El-Sheikh University.

Additives Treatments

Twenty-four supplemental additives treatments of chopped corn stover were done as follows:

- 1- Without additive (control).
- 2-3% molasses.
- 3-3% ground corn grains.
- 4-3% soybean meal.
- 5-0.5% urea.
- 6-0.5% Optigen.
- 7-1 ml effective microbes (EM1)/ kg corn stover.
- 8-3% molasses + 3% soybean meal.
- 9- 3% molasses + 0.5% urea.
- 10- 3% molasses + 0.5% optigen.
- 11- 3% ground corn grains + 3% soybean meal.
- 12-3% ground corn grains +0.5% urea.
- 13-3% ground corn grains + 0.5% Optigen.

14-3% molasses + 1 ml effective microbes (EM1)/kg corn stover.

15-3% ground corn grains + 1 ml effective microbes (EM1)/kg corn stover.

16-3% soybean meal + 1 ml effective microbes (EM1)/kg corn stover.

17-0.5% urea + 1 ml effective microbes (EM1)/kg corn stover.

18-0.5% optigen + 1 ml effective microbes (EM1)/kg corn stover.

19-3% molasses + 3% soybean meal + 1 ml effective microbes (EM1)/kg corn stover.

20- 3% molasses + 0.5% urea + 1 ml effective microbes (EM1)/kg corn stover.

21-3% molasses + 0.5% optigen + 1 ml effective microbes (EM1)/kg corn stover.

22- 3% ground corn grains + 3% soybean meal + 1 ml effective microbes (EM1)/kg corn stover.

23- 3% ground corn grains + 0.5% urea + 1 ml effective microbes (EM1)/kg corn stover.

24- 3% ground corn grains + 0.5% optigen + 1 ml effective microbes (EM1)/kg corn stover.

Silage Making

Corn stover was taken after harvesting the ears immediately, chopped using a harvester chopper machine to 1.5-2.0 cm of length, and supplemented with additives. Then ensiled in plastic bags with about 0.5 kg of weight capacity and pressed by hand to exclude the air from the silos. Treatments were run in triplicates (three of each) and ensiled for 45 days. After the ensiling period, representative samples were taken for chemical analysis and to determine the silage quality.

Chemical Analysis

Samples were dried in a forced air oven at 60 °C for 48 hours, ground, and chemically analyzed for determination of DM, CP, CF, EE, and ash according to AOAC (1990).

Silage Quality

For the determination of silage quality, 20 gm of wet silage was extracted in a blender with 100 ml of distilled water. The extracts were filtered through Whatman No. 40 filter papers and the pH values were determined directly in the filtrate solution using a Bechman pH meter. Total volatile fatty acids (TVFA's) were determined according to Warner (1964), NH₃-N concentrations (AOAC, 1990), and lactic acid was estimated by titration with 0.1 sodium hydroxide solution using 2-3 drops of phenolphthalein indicator according to the methods of Analytical Chemistry of Foods (1995) the following equation:-

Lactic acid % of DM = ml of NaOH x 0.09/ sample weight.

Statistical Analysis

The data were statistically analyzed using the general linear models (GLM) procedure adapted by IBM SPSS Statistics (2020) for user guide with one-way ANOVA. Significant differences in the mean values among dietary treatments were analyzed by Duncan's tests within SPSS program set at the level of significance P<0.05.

RESULTS AND DISCUSSIONS

Chemical composition

The chemical composition of corn stover silage as affected by energy, protein, and microbial additives is shown in Table (1). Adding ground corn grains to corn stover silage led to a significant (P<0.05) increase in DM content. Moreover, NFE content of corn stover silage increased significantly (P<0.05), however, ash content decreased significantly (P<0.05) with molasses and ground corn grains supplementation. Whereas, the contents of OM, CP, CF, and EE were not affected significantly by molasses and ground corn grains supplementation.

Soybean meal supplementation increased significantly (P<0.05) DM content of corn stover silage than those of urea and optigen supplementation with insignificant differences with control (without additives). The crude protein content of corn stover silage increased significantly (P<0.05) with soybean meal, urea, and optigen supplementation. While ash content decreased significantly (P<0.05) with soybean meal supplementation. However, the contents of OM, CF, EE, and NFE were not significantly affected by soybean meal, urea, and optigen supplementation.

Effective microbes supplementation led to a significant (P<0.05) increase in the contents of CP and NFE of corn stover silage and a significant (P<0.05) decrease in the contents of CF and ash. Whereas, the contents of DM, OM, and EE were not significantly affected by effective microbes supplementation.

The interaction between energy and protein supplementation showed a significant difference (P<0.05) in DM content only. Ground corn grains and soybean meal supplementation showed significantly (P<0.05) higher DM content (35.86%) than that molasses and optigen supplementation (34.38%) without significant differences with the other treatments. Whereas, the contents of OM, CP, CF, EE, NFE, and ash were not significantly affected by the interaction between energy and protein supplementation.

Interactions between energy and effective microbes or between protein and effective microbes not revealed any significant differences in the composition of all nutrients among treatments. Whereas, interactions among energy, protein, and effective microbes supplementation showed significant differences (P<0.05) in DM content only.

	DM %	DM Composition, %						
Item		OM	<u> </u>	CF	EE	NFE	Ash	
Energy		0111	01	01	LL		11011	
Control	34.47 ^b	90.85	7.29	29.45	2.38	51.73 ^b	9.15 ^a	
Molasses	34.40 ^b	91.45	7.40	28.57	2.40	53.08ª	8.55 ^b	
Ground corn grain	35.72 ^a	91.78	7.51	28.86	2.39	53.02 ^a	8.22 ^b	
SEM	0.27	0.54	0.05	0.21	0.01	0.38	0.14	
Protein				-			-	
Control	34.47 ^{ab}	90.85	7.29 ^b	29.45	2.38	51.73	9.15 ^a	
Soybean meal	35.56a	91.65	8.54 ^a	28.71	2.41	51.99	8.35 ^b	
Urea	33.68 ^b	91.10	8.53 ^a	28.80	2.43	51.34	8.90 ^a	
Optigen	34.15 ^b	91.05	8.47 ^a	28.71	2.44	51.43	8.95ª	
SEM	0.27	0.46	0.16	0.17	0.01	0.26	0.10	
Effective microbes								
Control	34.47	90.85	7.29 ^b	29.45 ^a	2.38	51.73 ^b	9.15 ^a	
EM1	33.66	91.15	7.74 ^a	28.27 ^b	2.37	52.77 ^a	8.85 ^b	
SEM	0.29	0.67	0.11	0.34	0.02	0.45	0.09	
Interactions								
Energy×protein								
Molasses + soybean	35.27 ^{ab}	91.50	8.64	28.60	2.41	51.85	8.50	
Molasses + urea	34.52 ^{ab}	91.40	8.63	28.65	2.42	51.70	8.60	
Molasses + optigen	34.38 ^b	91.38	8.58	28.68	2.39	51.73	8.62	
Corn + soybean	35.86 ^a	91.80	8.75	28.68	2.44	51.93	8.20	
Corn + urea	35.06 ^{ab}	91.70	8.75	28.71	2.43	51.81	8.30	
Corn + optigen	34.98 ^{ab}	91.60	8.69	28.74	2.38	51.79	8.40	
SEM	0.18	0.36	0.04	0.11	0.01	0.21	0.01	
Energy×effective microbes								
Molasses + EM1	33.62	91.58	7.85	28.12	2.40	53.21	8.42	
Corn + EM1	34.62	91.75	7.97	28.07	2.36	53.35	8.25	
SEM	0.33	0.67	0.06	0.21	0.02	0.39	0.67	
Protein x effective microbes								
Soybean + EM1	35.10	91.40	8.96	28.18	2.37	51.89	8.60	
Urea + EM1	33.98	91.20	8.97	28.07	2.40	51.76	8.80	
Optigen + EM1	33.92	91.14	8.93	28.15	2.42	51.64	8.86	
SEM	0.28	0.53	0.05	0.16	0.02	0.30	0.53	
Energy×protein×effective microbes								
Molasses + soybean + EM1	35.32 ^{ab}	91.35	9.07	27.92	2.43	51.93	8.65	
Molasses + urea + EM1	34.47^{ab}	91.12	9.09	27.98	2.41	51.64	8.88	
Molasses + optigen + EM1	34.32°	91.06	9.03	28.01	2.39	51.63	8.94	
Corn + soybean + EM1	35.75 ^a	91.72	9.18	27.83	2.47	52.24	8.28	
Corn + urea + EM1	34.98 ^{ab}	91.56	9.21	27.86	2.46	52.03	8.44	
Corn + optigen + EM1	34.90 ^{ab}	91.40	9.14	27.89	2.43	51.98	8.60	
SEM	0.18	0.37	0.04	0.11	0.01	0.21	0.37	

Table 1: Composition of corn stover silage

a, b: means with different superscripts differ significantly at P < 0.05.

Ground corn grains, soybean meal, and effective microbes supplementation showed significantly (P<0.05) higher DM content (35.75%) than that molasses, optigen, and effective microbes supplementation (34.32%) without significant differences with the other treatments. However, the other nutrients were not significantly affected by the interactions among energy, protein, and effective microbes supplementation.

According to the report of (Weiss and Underwood, 2009), added grain may also make wet silage easier to unload from the silo. Recommended application rate for grain is 100 - 200 lbs/wet ton. This rate will increase the dry matter content of silage by about 5 percentage units. The grain should be cracked or rolled before ensiling for maximum benefit.

Four levels (0%, 4%, 8%, and 12%) of dried molasses (97% DM) were applied to chopped Bermuda grass (32.4% DM, 70.2% NDF) pretreated with 1174 Pioneer silage inoculants (1.7 l/t of forage) and packed in 19liter plastic containers. The increasing molasses levels lowered ADF, and NDF percentages in Bermuda grass silages (Nayigihugu *et al.*, 1995). Dry matter and crude protein contents increased to some extent in silage with 3% molasses and corn additives (Bilal, 2009).

Such additives as particularly urea when added to high dry matter, and low buffering forages (maize or sorghum grain) increase crude protein content (Glewen and Young, 1982). Urea addition increased silage crude protein (Nursoy *et al.*, 2003).

Silage Quality

Quality characteristics of corn stover silage as affected by energy, protein, and effective microbes supplementation are shown in Table (3). The pH value of silage decreased significantly (P<0.05), however lactic acid concentration increased significantly (P<0.05) with molasses and ground corn grains supplementation. While the concentrations of ammonia nitrogen (NH₃-N) and total volatile fatty acids (TVFA's) were not affected significantly by molasses and ground corn grains supplementation.

The pH value of silage was higher significantly (P<0.05) with urea and optigen compared to control and soybean supplementation. Urea-supplemented silage recorded significantly (P<0.05) the NH₃-N concentration followed by optigen, then soybean meal, whoever control had the lowest concentration. Moreover, soybean meal-supplemented silage showed significantly (P<0.05) higher concentrations of TVFA's and lactic acid compared to urea-supplemented silage, whereas control and optigen-supplemented silage were insignificantly different.

Effective microbes supplementation didn't have any significant effect on pH value and TVFA's concentration of corn stover silage. Whereas, the concentrations of NH3-N and lactic acid increased significantly (P<0.05) with effective microbes supplement to corn stover silage.

Energy and protein supplementation interaction revealed that urea with molasses or ground corn grains recorded significantly (P<0.05) higher pH values and NH₃-H concentration of silage compared to soybean with molasses or ground corn grains, while optigen with molasses or ground corn grains didn't differ significantly. However, soybean meal with molasses or ground corn grains recorded significantly (P<0.05) higher concentrations of TVFA's and lactic acid in silage compared to urea with molasses or ground corn grains, while optigen with molasses or ground corn grains didn't differ significantly (P<0.05) higher concentrations of TVFA's and lactic acid in silage compared to urea with molasses or ground corn grains, while optigen with molasses or ground corn grains didn't differ significantly.

Interaction between energy and effective microbes didn't show any significant effect on the quality of corn stover silage. While, the interaction between protein and effective microbes showed that urea with EM1 supplemented silage had significantly (P<0.05) higher pH value and NH₃-N concentration and lower TVFA's and lactic acid concentrations than those of soybean meal with EM1, whereas optigen with EM1 didn't differ significantly.

The interaction among energy, protein, and effective microbes showed that molasses or ground corn grains with urea plus EM1 supplemented silages had significantly (P<0.05) higher pH value and NH₃-N concentration and lower TVFA's and lactic acid concentrations than those of molasses or ground corn grains with soybean meal plus EM1, whereas molasses or ground corn grains with optigen plus EM1 didn't differ significantly.

Silage fermentation is a dynamic process that is affected by a variety of factors. Research on silage and silage additives has been conducted for many years to improve the nutritive value of silages and to reduce some of the risks during the ensiling process (Henderson, 1993). A silage additive should be safe to handle and reduce DM losses, silage additives are added to the forage or crop at ensiling, may improve the ensiling (fermentation) process, reduce losses, reduce aerobic deterioration at feedout, improve the hygienic quality of the silage, limit secondary fermentation, improve aerobic stability, increase the nutritive value of the silage, as the result increase animal production and give the farmer a return greater than the cost of the additive (Merensalmi and Virkki, 1991). Some silage additives may also reduce unavoidable losses, particularly those associated with the plant enzymes and microorganism or field losses. Examples of the five main classes of silage additives are fermentation stimulants (bacteria culture and carbohydrate sources), fermentation inhibitors (acids, formaldehyde, etc.), aerobic deterioration inhibitors (lactic acid bacteria, propionic acid, etc.), nutrients (urea, ammonia, etc.) and absorbents (barley, straw, etc.) (McDonald et al., 1991).

According to the report of (Weiss and Underwood, 2009), the addition of grain to corn silage is not useful, but adding it to hay crop silage has two benefits. First, adding grain to hay crop silage increases the energy content of the silage. This will reduce the amount of supplemental grain that has to be fed. If silage will be the main or only feed

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offered, then adding some grain to the forage at ensiling will make it a more complete feed. However, grain mixed with silage before ensiling or at feeding is nutritionally equal; therefore, if supplemental grain must be fed anyway, no true benefit is realized. Secondly, adding grain to forage will increase the dry matter content of the silage. Hay crops that are not wilted sufficiently before ensiling can cause seepage and result in undesirable fermentation. Added grain may also make wet silage easier to unload from the silo. Recommended application rate for grain is 100 - 200 lbs/wet ton. The grain should be cracked or rolled before ensiling for maximum benefit. In general, adding grain does not improve fermentation because starch (the main carbohydrate in grain) is not readily fermented in the silo.

Cane molasses (75% DM) has been widely used and added up to 10% w/w to provide fast fermentable carbohydrates for the ensilage of tropical herbages. Due to its viscosity, it is difficult to apply and should be diluted preferably with a reduced volume of warm water to minimize seepage losses. When applied to tropical grasses molasses should be used in relatively high concentrations (4% to 5%) and with crops of very low DM content, a considerable proportion of the additive may be lost in the effluent during the first days of ensilage (Henderson, 1993). However, according to Woolford (1984), the provision of extraneous sugar alone is not sufficient to permit the lactic acid bacteria to compete with other components of the silage microflora and thus ensure preservation. So, under high moisture conditions, molasses can also induce clostridial spoilage, especially with forages contaminated with soil.

Molasses in numerous silage experiments have been proven to be an effective silage additive in terms of promoting lactic fermentation, reducing silage pH, discouraging clostridial fermentation and proteolysis, and generally decreasing organic matter losses. It is of particular benefit when applied to forage crops low in fermentable carbohydrates for lactobacilli. Recently, Keady (1996) reviewed the published literature on molasses as silage additives and concluded that molasses treatment improved silage preservation.

Sugarcane molasses added at the rate of 3% (w/w, fresh basis) to Napier grass (12.9% DM, 6.6% WSC) produced silages of reasonably good fermentation quality, however, the nutrient recovery from the silo was reduced as compared to formic acid treated silage (Boin, 1975).

Dwarf elephant grass cut at 72 days of growth (14.4% DM, 7.1% WSC) with a high buffering capacity was treated with 4% molasses and ensiled in 4 kg polythene bags with the resulting silage having lower pH and ammonia-N than the control silage (Tosi *et al.*, 1995).

Four levels (0%, 4%, 8%, and 12%) of dried molasses (97% DM) were applied to chopped Bermuda grass (32.4% DM, 70.2% NDF) pretreated with 1174 Pioneer silage inoculants (1.7 l/t of forage) and packed in 19liter plastic containers. The increasing molasses levels lowered pH in Bermuda grass silages (Nayigihugu *et al.*, 1995). The

pH decreased and lactic acid increased in silage with 3% molasses and corn additives (Bilal, 2009).

In a report by Glewen and Young (1982) on the use of urea as a silage additive for elephant grass it was concluded that with low DM forage and in the absence of additives rich in WSC such type of product should not be recommended when aiming an improvement of fermentation. Generally, pH value, ammonia-N, and acetic and butyric acid contents are increased.

Singh *et al.*, (1996) registered the highest pH values and ammonia-N levels associated with higher anaerobic proteolytic bacterial populations in *Sorghum bicolor* silages (34% DM) made with 0.5% urea. Other NPN sources as ammonium sulfate and biuret, either alone or associated with urea, calcium carbonate, or starch sources have also been tested on their effects on silage fermentation, digestibility, and intake. The results as reported by Vilela (1984) do not favour their use as silage additives either. According to Bolsen (1999) NPN always acts as a buffer during fermentation, requiring extra lactic acid to be produced to lower the pH enough for preservation, thus increasing DM loss.

Several microorganisms that are not hoLAB have been used as silage inoculants specifically to improve aerobic stability. For example, Propionibacteria are able to convert lactic acid and glucose to acetic and propionic acids that are more antifungal than lactic acid. Flores-Galaraza et al. (1985) reported that the addition of P. shermanii prevented the growth of molds and markedly reduced the initial population of yeast in high-moisture corn where the final pH was greater than 4.5. Dawson (1994) reported similar findings in high-moisture corn. Weinberg and Ashbell (1993) saw little benefit from adding Propionibacteria to pearl millet and corn silage (final pH < 4.0) but reported improvements in the aerobic stability of wheat silage when the decline in pH was slow. In studies conducted in laboratory silos, there were no observed beneficial effects of *Propionibacteria* in corn silage (final pH 3.6 to 3.8) (Kung et al., 1989). However, Bolsen et al., (1996) reported more propionic acid, lower yeasts and molds, and greater aerobic stability in corn silage (pH of 3.6) treated with Propionibacteria. Some concerns relative to the use of Propionibacteria that have not been adequately addressed are the loss of DM (from CO₂ production) and the fact that Propionibacteria have proteolytic activity. The primary reasons for the ineffectiveness of these organisms include the fact that they are strict anaerobes, they are slow growing, and they are relatively acid intolerant.

Heterolactic lactobacilli may also be useful as silage inoculants. For example, two new isolated heterolactic strains of *Lactobacillus plantarum* have been shown to improve the aerobic stability of corn silage by an average of 28 hours (Allman and Stern, 1999). These organisms were selected for fast growth, production of lactic and acetic acids, and the ability to suppress the growth of 5 major strains of yeasts that cause spoilage in corn silage.

Table 2. Quality characteristics of corn stover shage.									
Item	рН	NH3-N % of total-N	TVFA's % of DM	Lactic acid % of DM					
Energy									
Control	4.37 ^a	2.98	1.88	4.68 ^b					
Molasses	4.18 ^b	3.02	1.94	5.04 ^a					
Corn	4.20 ^b	3.07	1.93	5.01 ^a					
SEM	0.04	0.02	0.01	0.06					
Protein									
Control	4.37 ^b	2.98°	1.88 ^{ab}	4.68 ^{ab}					
Soybean	4.31 ^b	3.29 ^b	1.90 ^a	4.93 ^a					
Urea	4.73 ^a	3.48 ^a	1.76 ^b	4.57 ^b					
Optigen	4.57 ^a	3.40 ^{ab}	1.81 ^{ab}	4.71 ^{ab}					
SEM	0.05	0.06	0.02	0.05					
Effective microbes									
Control	4.37	2.98 ^b	1.88	4.68 ^b					
EM1	4.31	3.16 ^a	1.90	4.93 ^a					
SEM	0.03	0.05	0.01	0.07					
Interactions									
Energy×protein									
Molasses + soybean	4.36 ^b	3.33 ^b	1.88^{a}	4.89 ^a					
Molasses + urea	4.64 ^a	3.52 ^a	1.79 ^b	4.65 ^b					
Molasses + optigen	4.52 ^{ab}	3.42 ^{ab}	1.83 ^{ab}	4.75 ^{ab}					
Corn + soybean	4.39 ^b	3.37 ^b	1.87^{a}	4.86 ^a					
Corn + urea	4.67 ^a	3.57 ^a	1.77^{b}	4.62 ^b					
Corn + optigen	4.54 ^{ab}	3.45 ^{ab}	1.80^{b}	4.73 ^{ab}					
SEM	0.03	0.03	0.01	0.03					
Energy ×effective microbes									
Molasses + EM1	4.25	3.20	1.91	4.97					
Corn + EM1	4.27	3.25	1.88	4.92					
SEM	0.03	0.03	0.02	0.04					
Protein ×effective microbes									
Soybean + EM1	4.34 ^b	3.60 ^b	1.89 ^a	4.91 ^a					
Urea + EM1	4.62 ^a	3.76 ^a	1.79 ^b	4.66 ^b					
Optigen + EM1	4.51 ^{ab}	3.69 ^{ab}	1.83 ^{ab}	4.76^{ab}					
SEM	0.05	0.03	0.02	0.04					
Energy×protein×effective microbes									
Molasses + soybean + EM1	4.35 ^b	3.63 ^b	1.93 ^a	4.92 ^a					
Molasses + urea + EM1	4.60 ^a	3.78 ^a	1.81 ^b	4.67 ^b					
Molasses + optigen + EM1	4.49^{ab}	3.71 ^{ab}	1.86 ^{ab}	4.78^{ab}					
Corn + soybean + EM1	4.32 ^b	3.65 ^b	1.91 ^a	4.90 ^a					
Corn + urea + EM1	4.58 ^a	3.81 ^a	1.80 ^b	4.71 ^b					
Corn + optigen + EM1	4.46^{ab}	3.73 ^{ab}	1.86 ^{ab}	4.83 ^{ab}					
SEM	0.03	0.02	0.01	0.03					

Table 2: Quality characteristics of corn stover silage.

a, b: means with different superscripts differ significantly at P < 0.05.

Another heterolactic acid bacteria having potential to improve the aerobic stability of silages are *Lactobacillus buchneri*. Dreihuis *et al.* (1996) reported that corn silage treated with *L. buchneri* was more stable than untreated silage. They suggest that improved aerobic stability was due to the ability of *L. buchneri* to ferment lactic acid to acetic acid and 1,2 propanediol (Ojeda, 1993). Ridla and Vehida (1998) added *L. buchneri* to corn silage at a rate of 10^3 to 10^6 cfu/g of silage and found decreased numbers of yeasts in silage and increased acetic acid in silage (from 1.8% to 3.6% DM basis).

Aerobic stability was markedly improved by inoculation (control silage heated after 26 hours while treated silages remained cool for more than 400 h in silage). Increases in acetic and propionic acids in silages treated with *L. buchneri* accompanied improvements in the aerobic stability of barley silage. Also, he has observed improved aerobic stability in high-moisture corn treated with *L. buchneri* (Kung *et al.*, 1999).

CONCLUSION

From these results, it could be concluded that feed additives such as energy, protein, and microbial inoculants improved the composition and quality of corn stover silage.

CONFLICT OF INTEREST

The author(s) declare no conflicts of interest regarding the publication of this paper.

REFERENCES

Allman, J.G. and L.A. Stern (1999). BiomaxÒ5 Microbial Inoculant for Corn Silage.

- Analytical Chemistry of foods (1995). Published by Blockie academic and professional, an imprint of chapman & Hall, western cleddens Road, Bishopbriggs, Glasgow G64 2NZ, UK.
- AOAC (1990).Official methods of analysis of the Association of Official Analytical Chemists. 2 vols. 15th ed. Washington, DC, USA.
- Bilal, M.Q. (2009). Effect of molasses and corn as silage additives on the characteristics of mott dwarf elephant grass silage at different fermentation periods. Pakistan Vet. J., 29(1): 19-23.
- Boin, C. (1975). Elephant (Napier) Grass Silage Production Effect of Additives on Chemical Composition, Nutritive Value and Animal Performance. Ph.D. Thesis, Cornell University, Ithaca, p. 215.
- Bolsen, K.K. (1999). Silage Management in North America in the 1990s. In: Lyons, T.P. and Jacques, K.A., Eds., Biotechnology in the Feed Industry, Proceedings of the 15th Annual Symposium, Nottingham University Press, Nottingham, 233-244.
- Bolsen, K.K.; D.R.Bonilla; G.L.Huck; M.A.Young; R.A.Hart-Thakur and A.Joyeaux (1996). Effect of a propionic acid bacterial inoculant on fermentation and aerobic stability of whole-plant corn silage. Journal of Animal Science, 74: 274.

- Caro, D., S.J. Davis; E. Kebreab and F. Mitloehner (2018). Land-use change emissions from soybean feed embodied in Brazilian pork and poultry meat. Journal of cleaner production, 172: 2646-2654.
- Dalgaard, R.; J. Schmidt; N. Halberg; P. Christensen; M. Thrane and W.A. Pengue (2008). LCA of soybean meal. The International Journal of Life Cycle Assessment, 13(3): 240.
- Dawson, T.E. (1994). Propionic acid-producing bacteria as bioinoculants for the preservation of ensiled high-moisture corn. Ph.D. Dissertation, Michigan State University, East Lansing.
- Dreihuis, F.; S.F.Spoelstra; S.C.J. Cole and R.Morgan (1996). Improving Aerobic Stability by Inoculation with Lactobacillusbuchneri. Proceedings of the 11th International Silage Conference, Aberystwyth, 8-11 September 1996, 106-107.
- FAO. (2012). States of the World's Forests. Food and Agriculture Organisation of the United Nations.
- FAO. (2017). Formation of the Technical Advisory Group on feed additive environmental assessment.
- Flores-Galaraza, R.O.; B.A.Glatz; C.J. Bernand L.D. Van Fossen (1985). Preservation of high moisture corn by microbial fermentation. Journal of Food Protection, 48: 407-411.
- Gerber, P.J.; H. Steinfeld; B. Henderson; A. Mottet; C. Opio; J. Dijkman; A. Falcucci and G. Tempio (2013). Tackling climate change through livestock: a global assessment of emissions and mitigation opportunities. Food and Agriculture Organization of the United Nations (FAO).
- Glewen, M.J. and A.W. Young (1982). Effect of Ammoniation on the Refermentation of Corn Silage. Journal of Animal Sciences, 54:713-718.
- Haigh, P.M. (1988). The effect of wilting and silage additives on the fermentation of Autmn made grass silage ensiled in bunkers on commercial farms in South Wales. Grass and Forage Science, 43: 337-345.
- Henderson, N. (1993). Silage Additives. Animal Feed Science and Technology, 45, 35-56.
- IBM SPSS Statistics (2020). Statistical Package for the Social Science. Release 27. SPSS, Inc., Chicago, Illinois, USA.
- Kazer, J. and J. Madoc-Jones (2019). Alltech Optigen® Carbon Trust Validation Report. In confidence and not for external publication. Carbon Trust Advisory Limited.
- Keady, T.W.J. (1996). A Review of the Effects of Molasses Treatment of Unwilted Grass at Ensiling on Silage Fermentation, Digestibility and Intake, and on Animal Performance. Irish Journal of Agricultural and Food Research, 35, 141.
- Kertz, A.F. (2010). Review: Urea Feeding to Dairy Cattle: A Historical Perspective and Review. <u>The Professional Animal Scientist</u>, 26(3).
- Kung, L., W.M. Craig and L.D.Satter (1989). Ammonia-Treated Alfalfa Silage for Lactating Dairy Cows. Journal of Dairy Science, 72: 2565-2572.
- Kung, L.; N.K.Ranjit; J.R.Robinson and R.C.Charley (1999). The Effect of Lactobacillusnbuchneri on the Fermentation and Aerobic Stability of Barley Silage. Proceedings of the 12th International Silage Conference, Uppsala, 5-7 July 1999, p. 272-273.

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- Lattemae, P.; A.Laats and U.Tamm (2006). The Technological Factors Affecting the Quality of Big Bale Silage. www.eria.ee/public/files/summary_1.8_2006./link.
- Lehuger, S.; B. Gabrielle and N. Gagnaire (2009). Environmental impact of the substitution of imported soybean meal with locally-produced rapeseed meal in dairy cow feed. Journal of Cleaner Production, 17(6): 616-624.
- McDonald, P.; A.R. Henderson and S.J.E.Heron (1991). The Biochemistry of Silage. 2nd Edition. Chalcombe Publications, Marlow, Bucks.
- Merry, R.J.; R.F.Cussen-MacKenna; A.P. Williamsand J.K.S. Tweed (1993). The Effect of Different Inoculants on Fermentation and Proteolysis in Silages of Differing Perennial Ryegrass and White Clover Content. Proceedings of the 10th International silage Conference, Dublin, 6-8 September 1993, p. 83.
- Nayigihugu, V.; D.W. Kellogg; Z.B. Johnson; M. Scott and K.S. Anschutz (1995). Effects of adding levels of molasses on composition of bermudagrass (*Cynodon dactylon*) silage. J. Animal Sc., 73, Suppl.1: 200.
- Nursoy, H.; S. Deniz; M. Demirel and N. Denek (2003). The Effect of Urea and Molasses Addition into Corn Harvested at the Milk Stage on Silage Quality and Digestible Nutrient Yield. Turk. J. Vet. Anim. Sci., 27: 93-99.
- Ojeda, F. (1993). Conservantesquimicosen la preservacion de ensilajestropicales. Pastos y Forrajes, 16: 193-200.
- Oliveira, A.S. (1995). Rapid pH reductions in silages. Vol. 12, RevistaBrasileirade Saúde e Produção Animal, Salvador, 1-5.
- Ridla, M. and S.Vehida (1998). Effects of combined treatment of lactic acid bacteria and cell wall degrading enzymes on fermentation and composition of rhodes grass (Chloris gayanaKunth). Asian-Australasian Journal of Animal Sciences, 11: 522-529.
- Sariçiçek, B.Z. and U. Kiliç (2009). The Effects of Different Additives on Silage Gas Production, Fermantation Kinetics and Silage Quality. Ozean Journal of Applied Sciences, 2(1).
- Singh, A.; J.C. Edward; S. Mor and K. Singh (1996). Effect of Inoculation of Lactic Acid and Additives on Ensiling M.P. Chari (Sorghum Bicolor). Indian Journal of Animal Sciences, 66: 1159-1165.
- Tosi, H.; L.R. Rodrigues; A. De; C.C. Jobim; R.L. Oliveira; A.A.M. Sampaio and B. Rosa (1995). Ensilagem do Capim-Elefante cv. Mott sob diferentes tratamentos. Reunião da Sociedade Brasileira de Zootecnia, 24: 909-916.
- Vilela, D. (1984). Aditivos na ensilagem. Coronel Pacheco, MG: EMBRAPA-CNPGL, Circular Técnica 21, p. 32.
- Warner, A.C.I. (1964). Production of volatile fatty acids in the rumen, methods of measurements. Nutr.Abst. and Rev., 34:339.
- Weinberg, Z.G. and G.Ashbell (1993). Biological Silage Additives-Summary of Experiments in Israel. Proceedings of the 10th International Silage Conference, 6-8 September 1993, Dublin, 26.
- Weiss, B. and J. Underwood (2009). Silage Additives. Ohio State University Extension Department of Horticulture and Crop Science, Columbus.
- Woolford, M.K. (1984). The Silage Fermentation. Marcel Decker, New York.