Effect of Mixed Cereal-Legume Silages on Milk Production from Lactating Holstein Dairy Cows (R7010)

Basil Z.Mugweni[1], Marion Titterton[2], Barbara V. Maasdorp[3] And Faith Gandiya[2]

[1] *Agritex, P.O. Box 143, Mutare, Zimbabwe*

[2] *Department Of Animal Science, University Of Zimbabwe, P.O. Box MP167, Harare, Zimbabwe*

[3] *Department Of Crop Science, University Of Zimbabwe, P.O. Box MP167, Harare, Zimbabwe.*

Abstract

Mixed silages of good quality can be produced to partially replace commercial feed supplements without loss in milk yield or quality. However, there is need to ascertain from the responses in low yielding dairy cows, especially cross breds, whether there is potential to replace commercial feeds with mixed forage tree legumes and increase profits. The rate of substitution will depend on the relative costs of ensilage and commercial feeds. Judging from the performance of the mixed silages in this experiment, it is possible to recommend their use for replacing dairy feeds in the diets of heifers and dry cows. Further work into the types of phenolic compounds and their quantities, as affected by ensilage, are needed in order to improve the management of tannins, thereby enhancing protein supply from tanniniferous forages for milk production.

Introduction

In the smallholder dairy sector of Zimbabwe commercial feeds account for over 60% of total production costs (ARDA, 1999). Dairy producers would benefit if the amounts of commercial feeds used could be reduced without a decline in yield and quality of milk.

Traditionally silage has been made from cereals and grasses although legume silages have been produced (Dunn, 1991; Beauchemin *et al.*, 1994; Okine *et al.*, 1993; Belibasakis *et al.*, 1997). Cereal silages are rich in energy but low in crude protein (CP) (± 7%) whilst the converse is true for legume silages (Catchpole and Henzell, 1971). Titterton *et al.*, (1997) found that the CP of mixed maize and legume silage (40 - 60% maize with 60 - 40% legume) was greater than maize silage alone. Titterton *et al.*, (2000) successfully ensiled mixed forage tree legumes (FTLs) with maize and the CP of the mixed silages was reported to be comparable with that of commercial feeds, being 17.2% for maize-leucaena and 18.7% for maize-acacia silages.

Although FTLS are protein-rich forages, they often contain significant levels of anti-nutritional substances such as tannins and toxic chemicals (mimosine) that interfere with digestion and utilisation of protein, minerals and carbohydrates in ruminants (Rittner and Reed 1992). However, some anti-nutritional factors can be inactivated or removed by ensilage. James and Gangadev (1990) reported that the mimosine content of *Leucaena leucocephala* decreased significantly due to ensilage. In the light of these findings, a hypothesis was put forward that ensilage reduces the amount and effect of active tannins.

In this study the quality of mixed silages and the effect of partial substitution of a commercial dairy meal and maize silage with the mixed silages on dry matter intake, milk yield and milk quality were assessed. Economic implications of such substitutions are discussed in terms of savings on costs of supplementary protein.

Materials and Methods

Crops and harvesting. The FTLs used in this experiment were *Acacia boliviana* (Acacia) and L. *leucocephala* (Leucaena) from coppices harvested in 1999. The coppices were cut when more than 25% were at the flowering stage and at a height of about 0.7 m. The leaves were stripped by hand from the branches and twigs. A long-season white maize variety, SC709, was used. The crop was managed as a commercial maize crop, in terms of fertilizer application, weeding and pest and disease control. Harvesting was by hand and a motorised chaff-cutter was used to chop the maize into lengths of about 15cm.

Ensilage process. The crop was ensiled in 50kg plastic bags. Five kg of freshly chopped maize was thoroughly hand mixed with 5 kg of the respective freshly cut legume. The mixed forages were then packed in the plastic bags and compacted by hand, to exclude as much air as possible, and then tied by a string to ensure air-tightness. The material was left to incubate in a room for seven weeks before samples were taken for laboratory analyses. Concurrently, maize from the same crop was ensiled in a bunker. This silage provided the basal diet for the trial animals.

Samples and sample preparation. Two kg samples were taken from each batch of the respective legume and chopped maize. All the batch samples were then thoroughly hand mixed before three 2 kg samples were taken for laboratory analyses. Samples of freshly mixed maize-legume material were also taken. Three 0.5kg samples of the dairy meal were also taken for laboratory analysis.

Two of the three fresh 2 kg samples of the silages were stored in sealed plastic bags in the freezer. One of the samples of each was immediately used to determine dry matter (DM) content. The oven-dried samples were then ground through a 1.5mm screen and stored at room temperature until subsamples for laboratory analysis were taken after thorough mixing by stirring with a glass rod.

Laboratory analyses. The analyses were made on samples of the mixed silages, bagged maize silage, bunker maize silage and the dairy meal. The parameters analysed on the fresh material and the silages included neutral detergent fibre (NDF) modified acid detergent fibre (MADF), CP and ash. All analyses were done in duplicate. Dry matter was also estimated: fresh forages and silages were dried in a forced air oven at 60°C for 48 hours; and dairy meal in an oven at 105°C for 24 hours. Crude protein content was determined by the Kjeldahl method (MAFF, 1985). Fermentation characteristics of silages, pH, ammonia nitrogen (NH_3-N), volatile fatty acids (VFAs), and lactic acid (LA) were also determined (MAFF, 1985).

Ration formulation. Rations were formulated to give an overall CP content of 13% and a metabolizable energy (ME) concentration of 10MJ/kg (estimated from forage analysis and the AFRC, 1993). The bunker silage was the basal diet for the experimental animals. A commercial dairy meal was used to balance the rations for CP and energy content and it also ensured that mineral requirements were met. The diets consisted of 10kg treatment silage, 20+kg of basal maize silage (from the bunker) and 6.5 to 10.5kg of the dairy meal (19.6% CP and 13MJ/kg ME). Table 1 gives a summary of the rations used in the study.

***Table 1: Requirements (dry matter intake,* DMI*; crude protein,* CP*) and amounts of feed offered, to the three groups of cows, of diets containing maize silage* (MZ) *(basal diet and control) and the treatment silages, maize-leucaena* (ML) *and maize-acacia* (MA)*, together with dairy meal* (D).**

Silage	Requirements			Feeds used (kg DM/d)			Nutrients offered			Concentration	
Type	DMI (kg)	ME (MJ)	CP (kg)	Trt silages	Basal MZ	D	DM (kg)	ME (MJ/kg)	CP (kg)	ME (MJ/kg)	% CP
MZ	18.2	167.5	2.0	2.7	6.2	7.3	16.2	183.9	2.1	11.6	13.1
ML	18.9	159.0	1.8	2.8	6.2	6.3	15.3	171.4	2.0	11.6	12.9
MA	18.6	1723.0	2.1	3.4	6.2	7.2	16.8	189.3	2.2	11.6	13.0
SD	1.95	16.91	0.30	0.32		1.46	1.52	20.34	0.20	0.10	0.15

Animals and treatment allocation. Twelve Holstein dairy cows, with an initial live-weight of 610 kg ($\pm$ 71), and all in mid-lactation (days in milk 166 ($\pm$ 27)) were used. The animals were arranged into four groups of three animals each according to parity. The three cows in each group were randomly allocated to one of the three treatment silages i.e. maize (control), maize-leucaena and maize-acacia. All the experimental animals were then randomly allocated to individual feeding troughs in the feeding shed.

Feeding system and measurements. The cows were given three meals per day, at 06:00, 12:00 and 17:00 for a period of 21 days of which 14 days were for adaptation followed by 7 days of data collection. Animals were allowed at least 2 hours feeding time after which the refusals were removed and weighed. The treatment silage (10 kg) was fed at 06:00. At 12:00 and 17:00 the cows were given 10 kg of the basal bunker silage. The dairy meal allocation was given in three equal portions, one at each meal time. Refusals were removed after each feed and daily DM intake (DMI) calculated. Water was available between meals. Milk yields were recorded at each (am and pm) milking.

Milk samples and laboratory analyses. Milk sampling was done twice per week, during morning and afternoon milking. Twenty ml samples were stored in bottles containing a Bromopol (2-bromo, 2-nitropraine, 1,3 Diol + Natamycine) preservative tablet to prevent any spoilage before chemical analysis. The milk samples were analysed for butter fat (BF), lactose, protein, and total solids using a Bentley 2000 infrared milk analyser.

Statistical analysis. Fermentation characteristics and nutritive composition was analysed using SAS (1994) procedures for a completely randomised design as represented by the model below.

$R = m + T_i + E_{ij}$ Where R = response variable (e.g. dry matter, crude protein, pH etc),

m = overall mean, T_i = treatment effect (i = 1, 2, 3),

E_{ij} = random error.

For the feeding trial, SAS (1994), general linear models (GLM) procedures for a completely randomised block design were used for the analyses of DMI, milk yield and milk composition data. The following model was used for data analysis:

$R = m + P_i + T_j + E_{ijk}$ Where: R = response variable (DMI, milk yield, protein, butterfat, lactose and total solids),

m = overall mean, P_i = effect due to parity (i = 1, 2, 3,4),

T_j = treatment silage effect (j = 1, 2 or 3), E_{ijk} = random error.

The differences among the means were assessed by Tukeys method.

Results

Silage fermentation characteristics. Silage fermentation quality was assessed by DM content, pH, lactic acid (LA), VFAs and NH_3-N content (Table 2). The DM content of the silages was similar (P>0.05). Bagged maize silage had the lowest pH followed by maize-acacia and bunker maize silages, with similar values, and maize-leucaena silage had the highest pH value (P< 0.05). NH_3-N per cent in relation to total nitrogen in the silage was similar (P>0.05) for all silages.

Bagged maize silage had significantly higher LA concentration than the two mixed maize FTL silages (Table 2). The volatile fatty acids that could be identified by the GLC method were acetic acid (AA), propionic acid (PA), n- and iso-butyric acids (BA) and iso-valeric acid. Acetic acid and PA amounts did not vary between silages (P>0.05). The amount of acetic acid in the mixed maize-FTL silages was double that of the maize silage when expressed as a percentage of the total organic acids in the silage. The n- and iso-butyric acid levels across the silages varied (P<0.05), with both being highest in the maize silage followed by mixed maize-leucaena silage and mixed maize-Acacia silage. Iso-valeric acid could not be detected in the maize silage but was in appreciable amounts in the mixed maize-FTL silages.

Nutritional composition of the silages and the meal. The NDF content of the silages were not different but they all differed from that of the meal (P < 0.05) (Table 2). Bagged maize silage and mixed maize-Acacia silage had similar MADF whilst bunker maize silage and mixed maize-leucaena had higher MADF. The dairy meal had the highest D value followed by the bagged maize silage, mixed maize-acacia silage, bunker maize silage and the mixed maize-leucaena silage. The estimated D value of the bagged maize silage was different from that of the maize-leucaena and the bunker maize silage (P<0.05) but similar to that of the maize-Acacia silage. The D value of maize-acacia silage did not differ (P > 0.05) from that of the bunker silage and the mixed maize-leucaena silage. The same trend was found with estimated metabolizable energy values.

The CP content of maize-acacia silage was the highest whilst the bunker maize silage was the lowest. The CP content of the dairy meal was similar to that of mixed silages although maize-Acacia had a greater CP content, significantly higher than that of maize-leucaena silage (P<0.05) (Table 3). The ash content was highest (P<0.05) in the mixed maize-leucaena silage followed by the bagged maize silage and then the dairy meal with similar levels to those of the bunker and the mixed maize-Acacia silages.

Dry matter intake. The cows given mixed maize-Acacia and maize silage (control) ate more than those fed the mixed maize-leucaena silage (P<0.05) (Table 4).

Milk yield and quality. Milk yield (Table 4) was higher ($P < 0.05$) in cows fed mixed maize-Acacia and maize silages than in animals on mixed maize-leucaena silage. However, milk composition in terms of BF, lactose, protein and total solids was similar ($P>0.05$) across treatments.

Table 2: The Fermentation characteristics of the silages

Silage Type	Bunker maize	Bagged maize	Maize-leucaenae	Maize-acacia	Standard Error of Means
Dry matter (DM, g/kg)	309[a]	271[a]	276[a]	339[a]	12.3
PH	4.5[b]	3.7[c]	4.8[a]	4.5[b]	0.02
NH_3-N (g/kg DM)	9.59[a]	7.46[a]	10.09[a]	8.09[a]	0.756
Organic acids (g/kg DM):					
Acetic acid	Nd	9.56[a]	9.91[a]	7.76[a]	0.485
Propionic acid	Nd	1.00[a]	0.99[a]	0.72[a]	0.076
iso-butyric acid	Nd	3.10[a]	2.24[ab]	1.75[b]	0.149
n-butyric acid	Nd	0.96[a]	0.68[ab]	0.47[b]	0.067
iso-valeric acid	Nd	—	2.34[a]	1.59[a]	0.386
Total organic acids	—	87.87	49.46	41.29	

Note: [1] Nd = Not determined

All values are least square means except for total organic acids.

Values with different superscripts across the rows are significantly different ($P<0.05$).

Table 3: Dry matter (DM, g/kg fresh) and nutritive composition (g/kg DM) of the forage tree legume silages in relation to maize silage and a commercial dairy meal.

Feed Type	Bunker maize	Bagged maize	Maize-leucaena	Maize-acacia	Dairy meal	Standard error of means
DM	309[a]	271[a]	276[a]	339[a]	865§	12.34
CP	65.0[c]	71.2[c]	176.0[b]	208.7[a]	196.9[ab]	0.45
NDF	665.0[a]	608.2[a]	658.4[a]	602.6[a]	420.0[b]	17.52
MADF	353.5[a]	304.4[b]	357.4[a]	318.6[b]	98.9[c]	4.43
ME	9.3[c]	10.2[b]	9.2[c]	10.0[bc]	13.0[a]	0.13
Ash	5.6[b]	6.6[ab]	7.4[a]	5.6[b]	5.6[b]	0.23
*Digestibility (%)	57.9[c]	63.8[b]	57.6[c]	62.2[bc]	90.7[a]	1.50

Note: *Values with different superscripts across the rows are significantly different* ($P<0.05$)

§ *The value was not included in the separation of the means.*

**The values were calculated using* MADF (*Givens et al*, 1989).

Table 4: *Dry matter intake* (DMI/100 *kg live-weight*), *milk yield* (*kg/d*) *and milk composition* (%) *in cows fed mixed cereal-legume silages.*

	Maize silage (control)	Maize-leucaena silage	Maize-acacia silage	Standard error of means
DMI	3.3[a]	3.1[b]	3.3[a]	0.03
Milk yield	7.0[a]	14.1[b]	15.7[a]	0.69
Butterfat	3.6[a]	3.7[a]	3.6[a]	0.11
Protein	4.0[a]	3.4[a]	3.5[a]	0.05
Lactose	4.6[a]	4.67[a]	4.4[a]	0.04
Total solids	12.5[a]	12.7[a]	12.4[a]	0.16

Values with different superscripts within rows differ significantly ($P<0.05$).

Discussion

Silage fermentation quality. The quality of the mixed silages produced appeared satisfactory, in that the DM was within the recommended range, for maize and grass silages of 21 – 32 %. Dry matter indicates the bulkiness and the subsequent feeding value of a feed. In silages a match between high DM (25% to 32%) and high nutrient content is required. For ruminants, DM has a bearing on rumen fill and thus voluntary feed intake. This in turn influences the rate of passage and overall digestibility of a feed. pH values of less than five were achieved in the mixed silages; NH_3–N was also low, being <11% of total N in the silage (Catchpole and Henzell, 1971). The pH values were similar to those found by Titterton *et al.* (1999). The variation in the pH and the NH_3–N values could be explained by seasonal variation of the quality of the material and the harvesting and ensiling techniques followed in each case.

Tjandraatmadja *et al* (1993) also gave the following standards for tropical silages, pH should be less than 4.2, LA levels of 50%+ of total organic acids, BA levels of less than 0.5% of DM and NH_3–N of less than 10%. According to these criteria the quality of the bagged maize silage was good whilst those of the mixed maize-FTL were satisfactory.

It is generally believed that leguminous forages have high buffering capacity, which would result in relatively high pH values in silages made from such material (Uchida and Kitamira, 1987). The pH values achieved in this study seem to suggest that when the legume (FTL) is mixed with maize that has high levels of fermentable carbohydrates the buffering effect is reduced and desirable pH levels are achieved. These findings also confirm the technical feasibility of mixed maize-legume silages. The pH, LA, BA, NH_3-N, levels in this experiment indicate that there was little proteolytic decomposition and putrefaction, even by temperate standards. The plastic bag silo technology may have contributed to the production of good quality silage because it seems to provide better anaerobic conditions than bunker maize silage. This can be confirmed by the fact that the amount of the NH_3-N expressed as a percentage of total N in silage, which gives a reflection of the extent to which the decomposition of nitrogenous compounds has taken place, was low (<11%) in all samples, including the mixed silages, where there was a higher level of proteins. High LA, low pH and low NH_3-N levels found in this experiment suggest that there was little proteolysis of the protein. However, a good fermentation process does not depend on the type and quality of the forage crop only, but also on harvesting and ensiling techniques (Stefanie *et al.* 1999).

Nutritional composition of the silages. The CP of the mixed silages (17 – 21%) is comparable to that of commercial dairy feeds giving them an advantage over maize silage (6.8%), as reported by Titterton *et al.* (1999), although the values in this study were slightly higher. However, the efficiency of utilisation of the CP in mixed silages is not guaranteed due to perceived interference from polyphenolic compounds. Therefore, the feeding value of mixed silage can best be judged from animal performance. The CP content and the availability of protein in any livestock feed is important in that it has a bearing on supplementary requirements for this expensive nutrient.

The NDF levels of the mixed maize-FTLs were within the range for some forage silages in the tropics. For example, guinea 'A' grass silage in Sri Lanka had 69.9-71.9 NDF (Panditharatne *et al* 1986), napier grass silage in Thailand had 64.2 - 70.2 NDF (Shinoda *et al*. 1999). The MADF of forages and silages should be within the 22-50% range as suggested by Slater (1991). The lower the MADF, the higher the energy level in a forage or silage. The levels found in this study are within this range and this indicates that the mixed maize-FTL silages have a potential to replace the silage from such traditional crops maize and sorghum, if other factors are ideal. It is important to note that the NDF and MADF levels are dependent on the maturity stage of any given forage, since they are essentially indicating the levels of cell wall components, mainly the cellulose, hemicellulose and lignin (for NDF), and cellulose and lignin (for MADF).

Similarly DM and CP of a silage all depend on the type and stage of maturity of the crops at the time of ensiling, method of harvesting and technique of ensiling. Feeds with high fibre content have low digestibility and hence are of poor quality. The MADF of the bagged maize silage and that of the mixed maize-acacia were similar and so were those of the bunker maize silage and that of the mixed maize-leucaena silage but they were all within the 22-50% range suggesting that quality is acceptable. If NDF is considered, the picture is different, with all the four silages having a similar concentration (Table 2). In this regard MADF seems a better indicator of potential digestibility of silage than NDF.

The ash content of the mixed silages was comparable to that and the maize silage of the dairy meal, that contained added minerals for lactating dairy cows. Mixed maize-leucaena silage had a significantly higher level of the ash than the dairy meal and other silages used in this study. This suggests that there may be no need to add commercial mineral supplements if mixed silages are fed to moderately yielding dairy cows.

D*ry matter intake*. There was no difference between the DMI of maize-acacia and the maize silage demonstrating the potential of the mixed maize-acacia silage as a source of protein in dairy cattle feeding. Dry matter intake is an important parameter in assessing the nutritive value of a feed or forage. The CP content of a feed influences DMI because it tends to improve palatability. However, CP content alone can not be responsible for high DMI because the energy content of the feed also plays an important role since animals eat to satisfy their energy requirements (Syed and Leaver, 1999). The DMI reflected the influence of NDF, MADF and digestibility levels in the experimental treatment silages (see Table 2). The low DMI of the maize-leucaena silage could have been due to high fibre levels resulting in the rumen fill effect. In this study it seems that the DMI has been influenced by the fermentation quality of the silages.

M*ilk yield and quality*. Milk yield and quality are influenced within bred, by stage of lactation, parity, and animal size and body condition at calving within bred in addition to the type and level of feeding. Rations that stimulate high milk yield will depress BF% and boost TS. High levels of feeding tend to stimulate high milk yields and lactose but depress BF, protein and minerals. Conversely under-feeding results in high BF, protein and minerals and low milk yield and lactose (Slater, 1991). In this study maize silage resulted in milk yields similar to those of the maize-Acacia silage, indicating that the mixed silage has the potential to replace the maize silage. However, the value of the mixed silages, on-farm, cannot be guaranteed as this depends on the prevailing economic situation. Low DMI seemed to have affected the milk yield from the maize-leucaena silage. Milk yields from animals supplemented with L. *leucocephala* hay were higher than those from animals fed A. *angustissma* and C*alliandra calothyrsus* hay supplements (Hove, 1999). These findings suggest that the processing done prior to feeding influences the performance of forages. Sun, oven or freeze drying have varying effects on tannin levels (Ahn *et al*., 1989), thus affecting DMI and subsequent the milk output. In this experiment ensilage seems to have had varied effects on tannin levels depending on the type of forage.

There were no differences in the quality of milk across the treatments, although Kumagai *et al*. (1993) suggested that milk yield and composition in dairy cows might be influenced by the source of roughage. The present study agrees with the conclusions drawn by Khorasani *et al*. (1996), that dairy cows can maintain similar milk yields, despite marked differences in the type of end products arising from carbohydrate and protein digestion. Chenais *et al*. (1993) carried out similar studies using mixed maize-red clover silage and lucerne silage and found that the mixed silage increased milk yield compared to the maize silage alone (control) but lucerne silage was out-performed by the control. The same authors also reported that the legumes compared to the maize silage lowered milk fat and protein levels.

Bequette *et al.* (1993) reported that protein supplementation resulted in increased milk output although there was a significant proportion of protein channelled to the mammary gland for tissue growth. These varying results indicate that there is need for more research into the subject of mixed silages and their influences on milk yield and composition in given environments. This is important, since the quality of milk has an influence on processing it into milk products. Long-term studies are needed to determine the effects of mixed forages on udder development and the subsequent milk yields.

The advantages of the mixed silage can be expressed in terms of savings in costs, compared to using commercial feeds, while the disadvantages are reduced milk yields. The substitution of commercial feeds by FTLs must allow for the full costs of ensilage and the cost of commercial feeds

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