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EFFECT OF INCLUSION OF *Mucuna pruriens* AND *Dolichos lablab* FORAGE IN NAPIER GRASS SILAGE ON SILAGE QUALITY AND ON VOLUNTARY INTAKE AND DIGESTIBILITY IN SHEEP

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SUMMARY

Two high protein legume forages were added to Napier grass silage to determine their effect on silage quality. Each silage was also assessed for its intake by and digestibility in sheep. Napier grass regrowth was harvested at 8 weeks, wilted and chopped. The two legumes, *Dolichos lablab* and *Mucuna pruriens,* were harvested at 12 and 17 weeks, respectively, and were wilted and chopped. Napier grass was ensiled alone or mixed with either legume (at 20% of total weight) to make three treatments: Napier ensiled alone (NG), Napier ensiled with *D. lablab* (NGD), and Napier ensiled with *M. pruriens* (NGM). During ensiling, molasses was mixed at 3% w/w to each of the 3 silages. To determine intake and digestibility, the 3 diets were fed to 6 male Dorper sheep housed in metabolic crates in a double Latin-square design.

The crude protein content of the Napier silage alone was 97g kg^{-1} DM, which was improved with the addition of legumes to 112 and 113 g kg^{-1} DM for NGD and NGM, respectively. Silage pH was 4.6 for NG alone, compared to 4.5 and 4.3 for NGD and NGM, respectively. There was a trend towards improved dry matter intake (DMI, P=0.229) and organic matter intake (OMI, P=0.157) of the silages containing legumes. Inclusion of legumes had no effect on dry matter (DM) digestibility (P=0.069) but organic matter (OM) digestibility tended to be higher in NGD than in NG and lower in NGM than in NG. The OM digestibility of NGD was significantly higher ($P\leq0.05$) than NGM. Metabolizable energy (ME) content (estimated via both gas production and digestible nutrients) was not altered by inclusion of legumes (7.4 MJ kg^{-1}) but nitrogen (N) balance was improved by the legumes (5.5, 9.2 and 8.3 g d^{-1} for NG, NGD and NGM, respectively). It was concluded that inclusion of the legumes in Napier grass silage increased the protein content of the silage and improved the nitrogen balance in sheep.

Key words: legume forage, *Mucuna*, napier, *Dolichos.*

INTRODUCTION

In Kenya, the smallholder dairy systems contribute 80% of the marketed milk (Muriuki, 2001). These systems are typically found in areas with high agricultural potential where the most common feed resource is Napier grass (*Pennisetum purpureum*) under the zero grazing system (Anindo and Potter, 1994). In the rainy season, Napier grass grows rapidly to maturity, its quality deteriorating quickly if it is not harvested. During the dry season, farmers use Napier grass that has been left as a standing reserve, or make use of crop residues and roadside pastures. Napier grass has a low crude protein content ranging from 45 to 85 g kg^{-1} DM (Abdulrazak *et al*., 1996; Kariuki *et al*., 1999; Muia *et al*., 1999) and metabolizable energy (ME) content averaging 8.6 MJ kg^{-1} DM (NRC, 1988). Consequently, diets based only on Napier grass do not meet the nutritional requirements of a lactating dairy cow, thus requiring supplementation with sources of fermentable energy and nitrogen. However, due to the high cost of commercial concentrate feeds, most smallholder dairy farmers do not supplement Napier grass diets. This leads to low exploitation of the genetic potential of the animals for milk production.

The quality of Napier grass fodder can be improved through the incorporation of legumes to supply extra protein (Kariuki *et al.*, 1999) or the addition of both protein and energy sources (Muinga *et al*., 1995). Forage legumes can be grown on farms to supplement Napier grass, which offers an affordable means of improving milk and meat production as well as sustaining soil fertility. A number of studies have been reported on the use of multipurpose trees and forage legumes to supplement Napier-based basal rations for ruminants (Abdulrazak *et al*., 1996; Jones *et al*., 1989; Kariuki *et al*., 1999).

In areas where smallholder dairy farming predominates, forage availability is dependent on seasonal rainfall distribution. During periods of fodder scarcity, milk production and weight gain are severely affected. Ensiling of Napier grass during the rainy season when it is abundantly available could alleviate these fluctuations in the feed supply. Tropical grasses, including Napier, are low in water soluble carbohydrates (≤50 g kg^{-1}) compared to forage maize (>100 g kg^{-1}) and do not ferment well when ensiled without additives. Although previous studies have reported on Napier grass silage with and without additives (Snijders and Woulters, 1990), there is a lack of information on the feeding value of Napier grass silages which incorporate protein rich forages.

This study reports on the use of *Dolichos lablab* and *Mucuna pruriens* forages to improve Napier grass silage quality and *in vivo* digestibility in sheep.

MATERIALS AND METHODS

Forage production

Pre-existing Napier grass fields at the Kenya Agricultural Research Institute (KARI) station in Naivasha, Kenya, were cut to ground level, weeded, and irrigated. In other fields, *Dolichos lablab* and *Mucuna pruriens* seeds were sown after ploughing and harrowing. All the fields were irrigated once a week unless it had rained during the previous week.

Forage preservation

Napier grass was harvested at 8 weeks of regrowth at average height of 1m, while the complete foliage of *Dolichos* was harvested at 12 and that of *Mucuna* at 17 weeks post germination. Napier grass was wilted in the field for one day and legumes for half a day. The forages were chopped to a length of 2.5cm using a motorized chaff cutter. Wilted and chopped Napier grass and legume forage were weighed and mixed at 80:20 ratio. Molasses was added at 3% weight of the Napier-legume mixtures. For ease of application, the molasses was diluted with water at a ratio of 3:1 and mixed with the Napier only and the Napier-legume mixture prior to ensiling.

The silage was preserved in pit silos measuring 6 m^3 (3x2x1 m) lined with polythene sheets. It was compacted by trampling with feet and covered with polythene sheets. Soil was heaped on the pit to maintain anaerobic conditions during storage.

Experimental animals

Six male Dorper sheep (averaging 60.3±4.6 kg) were obtained from the Sheep and Goat Research Station in Naivasha. The sheep were dewormed, sprayed with acaricide and placed in metabolic crates with facilities for feeding, watering and separate collection of urine and faecal material. After an initial period of adaptation, the animals were randomly allocated to the three dietary treatments in a replicated 3 by 3 Latin Square Design: Napier ensiled alone (NG), Napier ensiled with *D. lablab* (NGD), Napier ensiled with *M. pruriens* (NGM). The feeding was done over 3 periods, each consisting of 21 days, 14 days of adaptation period and 7 days of data collection. The animals were fed at 800 and 1500hrs and the rations adjusted daily to ensure at least 10% refusal. Water and mineral lick were available *ad libitum*.

Voluntary intake and *in vivo* digestibility

The feed offered was weighed daily at feeding and refusal was weighed in the morning prior to giving fresh feed. After the adaptation period (14 days), samples of feed offered were collected during the last 7 consecutive days of each period, stored frozen in airtight bags and pooled at the end of period. Dry matter content of the feed offered was determined by drying samples at 105^{o}C to a constant weight, and was used to calculate the daily dry matter intake. A sub-sample was dried at 60^{o}C for 48h, and was milled and stored for chemical analysis.

Daily faecal output was determined by weighing the fresh faecal material every morning, using the method described by Osuji *et al*. (1993). Each day, 10% of the daily fecal output was sampled and kept frozen until the end of the 7-day collection periodThe samples were thawed, pooled for each animal and sub-sampled. Dry matter content was determined by drying the samples at 105^{o}C to a constant weight. A sub-sample was oven dried at 60^{o}C for 48 hours, and was milled and stored for chemical analysis. The apparent dry matter digestibility and organic matter digestibility were calculated as described by Osuji *et al.* (1993).

Chemical analyses

For determination of silage pH, an extract was prepared by blending 100g of fresh silage, sampled at the beginning of each period, with 1 litre of distilled water for 30 seconds. The pH was immediately measured using a glass electrode pH meter (Fisher Instruments, USA) at KARI station, Naivasha. All other chemical analyses were conducted at the University of Bonn, Germany. Ash content was determined according to the method by AOAC (1990).

The crude protein was determined by the nitrogen combustion method of Dumas (AOAC 1990) using a Leco analyser (Leco Instrumente, Monchengladbach, Germany). Neutral detergent and acid detergent fibre were determined according to Van Soest and Robertson (1995) with the modification that fibre bags were used to hold the samples instead of the Fibertec crucibles. The gross energy contents of feed and faecal material were determined by bomb calorimetry (Adiabatic bomb calorimeter, IKA, Germany). Water soluble carbohydrates were determined according VDLUFA (1997).

Metabolizable energy estimation from gas production

Oven-dried (60^{o}C) silage samples milled to pass 1mm mesh were incubated with buffered rumen fluid in the standardised Hohenheimer Futterwertstest (HFT), also known as the Menke gas production technique (Menke and Steingass, 1988). The incubation apparatus included incubation syringes (model Fortuna, Häberle Labortechnik, Germany) having 32mm internal diameter and 100ml volume and an incubator equipped with a slow rotating rotor and maintained at 39^{o}C. Gas volume was read after 8 and 24 hours of incubation. After corrections for blank incubation and gas production from standard substrates (hay and concentrate), the gas volume was used to calculate the metabolizable energy content of the silages according to the following formulae:

$$ME(MJ.kg\,DM^{-1}) = \frac{136GP + 5.7CP + 0.286EE^{2} + 2200}{1000} \quad \text{(GfE 2001)}$$

$$ME(MJ.kg\,DM^{-1}) = 0.0312*gDEE + 0.0136*gDCF + 0.0147*g(DOM-DEE-DCF) + 0.00234*gCP$$

(Menke and Steingass, 1988)

Where :
DEE is digestible ether extract,
DCF is digestible crude fibre,
DOM is digestible organic matter, and
GP is gas production in ml.

Statistical analyses

Data from the feeding trial were analysed as a 3x3 Latin Square Design. Since the effect of period was not significant, only the dietary effects were considered. Duncan's multiple range test was used to determine differences between means (Snedecor and Cochran, 1980). The statistical package used was SPSS (SPSS Inc., 2001).

RESULTS AND DISCUSSION

The estimated dry matter yields of the two legumes (12 weeks for *Dolichos* and 17 weeks for *Mucuna*) are shown in Table 1. The yield of these two legumes has previously been reported to be 2-4 t DM ha^{-1} yr^{-1} (Njarui *et al.*, 1996) in a semi-arid environment and a peak biomass yield of 4 t DM ha^{-1} at 16 weeks for *Mucuna* (Maasdorp and Titterton, 1997). Under irrigation, annual yields per hectare can be much higher as several crops can be obtained in a year.

The nutrient composition of the Napier grass and legumes at time of ensiling and that of the silage are shown in Table 2. Two Napier grasses (NG1 and NG2) harvested at different times but of the same age were used. The crude protein (CP) content of Napier ranged from 95-107 g kg^{-1} DM at 8 weeks. This result was slightly lower than the 11.7% content reported by Kariuki *et al.* (1999) at the same age, which may be due to the fact that the CP content of Napier is highly dependent on soil fertility (Kariuki, 1998). The NDF (define) and ADF (define) content for the Napier grass varied from 579 to 613 and from 303 to 318g kg^{-1} DM, respectively, and were within the range reported by Kariuki *et al.* (1999).

Table 1. Dry matter yield of *Mucuna pruriens* and *Dolichos lablab.*

Legume	Yield (kg DM ha^{-1})	Comments
Mucuna pruriens	4795 (17 weeks)	Seeds treated by scarification and overnight soaking in water. Germination after 2 weeks.
Dolichos lablab	3753 (12 weeks)	Germination after 1 week.

Table 2. Average composition of the Napier grass, *Dolichos*, and *Mucuna*, as well as the silages made from their mixtures (g kg^{-1} except where stated).

	Forage				Silage		
Variable	NG1	NG2	*Dolichos*	*Mucuna*	NG1	NG1 + *Dolichos*	NG2 + *Mucuna*
DM	160	160	170	170	302	287	284
OM	809	791	865	870	785	799	798
CP	95	107	174	166	97	112	113
NDF	613	579	390	497	520	478	539
ADF	303	318	253	389	308	295	333
WSC	39	40	55	50	7	9	2
GE (MJ kg^{-1} DM)	n.d..	n.d.	n.d.	n.d.	16	16	15
pH	n.d.	n.d.	n.d.	n.d.	4.6	4.5	4.3

OM= organic matter, CP=crude protein, NDF= neutral detergent fibre, ADF=acid detergent fibre, GE=gross energy, NG= Napier grass, WSC = water soluble carbohydrates, n.d. = not determined.

The CP and fibre levels of the legume are highly dependent on age at harvest. The nutrient contents of *Mucuna* (Table 2) are within the range of those reported by Ravidran and Ravindran (1988). However, CP (at 166 g kg^{-1}) is lower than 20.4% reported by Njarui *et al.* (2000). In this study, the variation in *Mucuna* foliage with age was determined by sampling the forage at 52, 84, 110 and 121 days after emergence. The CP content at these four times was 252, 233, 193 and 166 gkg^{-1} DM, respectively. The CP level of *Dolichos* (17.4%) was comparable with 16.09 % reported by Njarui *et al.* (2000) but was lower than 22.1% reported by Muinga *et al.* (2000). Both NDF and ADF content of *Dolichos* were lower than those of *Mucuna*, probably due to maturity difference at harvesting.

Inclusion of the legumes in the silage significantly increased the CP content of the silage. The original aim was to improve the CP of the silage to 130g kg^{-1} DM but this effect was not achieved because the CP content of legumes was lower than expected. The *Dolichos* CP was similar to one study). The fibre content of the silages reflected those of their constituents. The water soluble carbohydrate (WSC) content was higher in the legumes than in Napier grass, but both were below 10% content required for sufficient fermentation capacity (Kellems and Church, 1998). As expected, WSC decreased during ensiling as it supplied energy to the microbes during the fermentation process. The pH of the silages was within expected range of well-fermented silages.

The dry matter intake and digestibility coefficients in sheep and the ME content of the 3 silages are shown in Table 3. The voluntary dry matter intake of silage tended to be higher (p=0.229) for the legume-enriched Napier silage than for the control. Lack of significant effect could be attributed to the high variation in intakes (good, high variation in intake should also be stated in the methods section or in a results section showing the actual variation in intakes). Inclusion of legumes in Napier grass-based diets has previously been shown to increase dry matter intake (DMI) (Abdulrazak *et al.*, 1996), which has been attributed to improved palatability and digestibility. This observation has also been shown for wheat-pea silage fed to sheep and cattle (Adesogan *et al.*, 2002; Salawu *et al.*, 2001) and for maize stover-*Leucaena diversifolia* fed to sheep (Hindrichsen *et al.*, 2002). However, in the present study, digestibility of organic matter was slightly higher (p=0.069) for the NGD silage compared to NG silage. NGM had the lowest digestibility. The digestibility of silage was probably related to its fibre content. The ME content, calculated from gas production and digestible nutrients, was highest for NGD and lowest for NDM and can be attributed to the difference in OM digestibility. Adesogan *et al*. (2002) also recorded only marginal increases in the digestibility of legume-supplemented wheat silage (wheat-pea). Thus, the advantage of legume inclusion in grass-based diets (fresh forage or silage) is the increased DMI, which ensures higher digestible nutrient uptake.

The results of the nitrogen balance measurements (Table 4) showed an increased intake of nitrogen by the NGD- and NGM-fed sheep, which also resulted in higher nitrogen balances. The amount of N retained as a percentage of N intake was highest for NGD and lowest for NGM. This difference could be attributed to possible lower digestibility of protein in the latter silage, which may have been NDF-bound, as the faecal-N excretion in NGM was higher. However, fibre bound nitrogen was not analysed in this study. Efficiency of N-capture at a ruminal level could also help explain the results, as the NGM silage had the highest N:ME ratio.

Table 3. Voluntary DM intake, DM and OM digestibility, *in vitro* gas production and metabolizable energy content of Napier grass, Napier-*Dolichos*, and Napier-*Mucuna* silages.

	Silage type		
Variable	Napier alone	Napier + *Dolichos*	Napier + *Mucuna*
DM ($g\ kg^{-1}$)	302 ± 19[a]	287 ± 13[a]	284 ± 15[a]
DMI ($g\ kg^{0.75}\ day^{-1}$)	79 ± 4.9 [a]	86 ± 5.2 [a]	88 ± 3.2 [a]
OMI ($g\ kg^{0.75}\ day^{-1}$)	63 ± 3.8 [a]	68 ± 4.0 [a]	70 ± 2.4 [a]
DMD (%)	57 ± 1.4 [a]	60 ± 1.3 [a]	56 ± 1.4 [a]
OMD (%)	66 ± 1.4 [ab]	67 ± 1.1 [b]	63 ± 1.3 [a]
Gas production ($ml\ 200\ mg^{-1}$)	33.1 ± 0.9 [abc]	34.4 ± 0.3 [a]	31.2 ± 0.6 [c]
ME^1 ($MJ\ kg^{-1}$ DM)	7.8 ± 0.01 [b]	8.0 ± 0.02[a]	7.53 ± 0.02 [c]
ME^2 ($MJ\ kg^{-1}$ DM)	7.37 ± 0.3 [ab]	7.67 ± 0.03[a]	7.20 ± 0.3 [bc]

ME^1($MJ.kg^{-1}$ DM)= 0.0312*g DEE+0.0136*g DCF+0.0147*g(DOM-DEE-DCF)+0.00234*g CP (GfE 2001)
ME^2 ($MJ.kg^{-1}$ DM)= (136GP+5.7CP+0.286EE^2+2200)/1000 (Menke and Steingass, 1988)
DEE=digestible ether extract, DCF=digestible crude fibre, DOM=digestibly organic matter, and GP=gas production in ml. Means with different superscripts are significantly different (P≤0.05).

Table 4. Nitrogen balance of sheep fed Napier, Napier-*Dolichos* and Napier-*Mucuna* silages.

	Silage type		
Variable	Napier	Napier+*Dolichos*	Napier+*Mucuna*
N intake ($g\ day^{-1}$)	23.5 ± 1.5 [a]	29.1 ± 2.5 [b]	30.2 ± 2.5 [b]
Faecal N loss ($g\ day^{-1}$)	11.1 ± 0.9 [a]	12.3 ± 1.0 [a]	14.1 ± 1.4 [a]
Urinary N loss($g\ day^{-1}$)	7.1 ± 0.4 [a]	8.6 ± 0.8 [a]	9.9 ± 1.6 [a]
N(g):ME(MJ)	2.0 ± 0 [a]	2.3 ± 0.04 [b]	2.4 ± 0.03 [c]
N balance ($g\ day^{-1}$)	5.5 ± 0.8 [a]	9.2 ± 1.2 [b]	8.3 ± 1.4 [b]
N retained (%)	24 [a]	28 [a]	20 [a]

Means with different superscripts are significantly different (P≤0.05).

CONCLUSION

Inclusion of high protein legumes (*Dolichos lablab* and *Mucuna pruriens)* in Napier silage improves dry matter intake and protein content of the silage, thus leading to a higher N retention in the animal.

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