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Evaluation of Chemical Composition and *in vitro* Fermentation Parameters of *Moringa oleifera* Leaf Meal Based Diets as Feed for Ruminants in Nigeria

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Abstract: This study was carried out to investigate the chemical composition and the 24 h *in vitro* fermentation characteristics of various levels of *Moringa oleifera* leaf meal supplementation, sole or combined with other feed ingredients for ruminants. *Moringa oleifera* Leaf meal (MOL), dried cassava peels, palm kernel cake, bone meal and salt were milled and stored separately and then mixed together at different levels. The levels of inclusions were; D1 (0% MOL), D2 (5% MOL), D3 (10% MOL), D4 (15% MOL) and D5 (100% MOL = test ingredient). The potential gas production ranged from 26.00-37.00 mL/200 mg DM. Highest $p<0.05$ potential gas production were obtained with both treatments D1 and D3 while D5 recorded the lowest value. The ME was highest in D1 (8.05 MJ kg^{-1} DM) and lowest for both D2 (6.90 MJ kg^{-1} DM) and D5 (6.82 MJ kg^{-1} DM) which were similar. The percentage OMD was highest ($p<0.05$) in D1 (61.42%) and lowest in both D2 (46.22%) and D5 (46.81%) which did not differ ($p>0.05$). The values for SCFA were highest ($p<0.05$) for both D1 and D3 (0.82 μmol) and lowest for D5 (0.56 μmol). The diet (D1-0% MOL) without the inclusion of *Moringa oleifera* leaf meal had the highest values of *in vitro* gas production parameter measurements, including the ME, OMD and SCFA as compared to the other diets with *Moringa oleifera* leaf meal inclusions. The best level of *Moringa oleifera* leaf meal inclusion in the diets was 10% (D3-10% MOL) while the sole *Moringa oleifera* leaf meal diet (D5-100% MOL) recorded the lowest *in vitro* gas production parameter values. This study, thus demonstrated that *Moringa oleifera* leaf meal when fed in combination with conventional concentrate ingredients could enhance its utilization as feed for ruminants.

Key words: Ruminants, diets, *in vitro* fermentation, gas production, *Moringa oleifera*, Nigeria

INTRODUCTION

There has been increasing demand and consequently, high cost of the conventional animal feed ingredients by livestock farmers in many tropical countries, such as Nigeria. This has created the search for alternative feed resources; such as tropical browse plants and multipurpose trees as sources of nutrients for ruminants as well as non ruminants (Aletor and Omodara, 1994). Aganga and Tshwenyane (2003) reported the use of leaves and twigs of trees as supplements to a wide range of forages and agricultural by-products in the diets of ruminants in Botswana. These browse foliages have the ability to remain green and to maintain a relatively high crude protein during the dry season. Browses could provide above 35% of digestible crude protein requirement for ruminants and also serve as source of vitamins and mineral elements which are usually deficient in grassland pastures (Isah and Gazaly, 2010). The nutritional constraints in the feeding of crop residues to ruminants are low Nitrogen (N) content, poor digestion and low intake, such that productive performance of tropical animals is usually low (Nouala *et al*., 2006). These researchers further explained that improvement in the nutritive value, removal of nutritional limitations to rumen fermentation and a balanced supply of nutrients to host animals would result in an improvement in animal productivity. Also, the supplementation of ruminants' diets with concentrate mixtures such as cereal grains, cereal brans or oil seed meals have resulted in increased intakes under intensive production systems but such feeds were not found to be economical due to their unavailability and high cost (Nouala *et al*., 2006). Thus, other sources of supplementation needed to be investigated.

The use of forage legumes as supplements had been suggested as an alternative to the use of concentrates (Roothaert and Paterson, 1997). However, many tropical

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fodder legumes when fed as sole diets have been found to contain anti-nutritional factors which may have negative influence on growth and bioavailability of other nutrients in humans and livestock (Soetan and Fafunso, 2010). Thus, there is the need to investigate the feeding of concentrate diets in combination with browse foliage. *Morimga oleifera* is a typical non-leguminous multipurpose tree with high crude protein in the leaves (usually >20%), though may also contain negligible contents of anti-nutritional factors, like tannins and saponins when used as food for humans and ruminants (Makkar and Becker, 1996; Nouala *et al*., 2006; Asaolu *et al*., 2011). The *in vitro* gas production method is a laboratory estimation of degraded feeds which allow quick assessment of nutritional value (Babayemi and Bamikole, 2006). The method was also used to estimate organic matter digestibility, metabolizable energy (Menke and Steingass, 1988) and short chain fatty acid (Getachew *et al*., 1999) contents of feedstuffs.

This study was carried out to evaluate the chemical composition and *in vitro* fermentation characteristics of *Moringa oleifera* leaf meal when fed solely and at graded levels in combinations with other feed ingredients to growing West African Dwarf goats.

MATERIALS AND METHODS

Collection of diet ingredients: The *Moringa oleifera* leaves were harvested from existing stands within the LAUTECH Teaching and Research Farm. Other feed ingredients used were dried cassava peels, palm kernel cake, bone meal and salt.

Diets preparation and chemical analysis: Sub samples of the feed ingredients were oven dried at 65°C to a constant weight, milled and mixed at the various inclusion levels of *Moringa oleifera* Leaf meal (MOL) as follows: D1 (0% MOL), D2 (5% MOL), D3 (10% MOL), D4 (15% MOL), D5 (100% MOL). Sub samples of the diets D1-D5 were also taken and oven dried at 105°C until constant weight was attained for dry matter determination (AOAC, 2005). The samples that had been oven dried at 65°C to a constant weight were ground through a Thomas Willey laboratory mill (1 mm screen) for analysis. Experimental diets were analysed for crude protein, crude fibre, ether extract, ash (AOAC, 2005). Neutral detergent fibre, acid detergent fibre and acid detergent lignin were determined according to the methods of Van Soest *et al*. (1991).

***In vitro* gas production:** The *in vitro* gas production was determined according to the method of Menke and Steingass (1988). Rumen liquor was obtained from three female West African Dwarf goats. While 200 mg of diets samples D1-D5 were placed in 120 mL calibrated syringes in triplicates, 30 mL inoculums containing (cheese-cloth) strained rumen liquor and buffer solutions (1:4 v/v) under constant flushing with CO_2 were incubated (Babayemi *et al*., 2006). The gas production was measured at 6, 9, 12, 18, 21 and 24 h. After 24 h of incubation, the amount of methane gas produced were estimated (Babayemi *et al*., 2006). Metabolizable Energy (ME, MJ/kg DM) and organic matter digestibility (OMD, %) were estimated as established (Menke and Steingass, 1988) and Short Chain Fatty Acids (SCFA, µmol) were calculated as described by Getachew *et al*. (1999).

Statistical analysis: Data collected were subjected to one-way Analysis of Variance (ANOVA) procedure of SAS (2002). Significant means were ranked using the Duncan's multiple range test of the same package.

RESULTS AND DISCUSSION

Chemical composition: The ingredients and chemical composition of experimental diets are presented in Table 1. The CP content of (7.26%) for D1 (0% MOL) was

Table 1: Ingredients and chemical composition of experimental diets at various levels of inclusions of *Moringa oleifera* Leaf meal (MOL)

Parameters (%)	D1 (0% MOL)	D2 (5% MOL)	D3 (10% MOL)	D4 (15% MOL)	D5 (100% MOL)
Ingredients composition					
Dried cassava peels	35.00	35.00	35.00	35.00	-
MOL	-	5.00	10.00	15.00	100.00
Palm kernel cake	61.00	56.00	51.00	46.00	-
Bone meal	3.00	3.00	3.00	3.00	-
Salt	1.00	1.00	1.00	1.00	-
Total	100.00	100.00	100.00	100.00	100.00
Chemical composition					
Dry matter	92.47	92.42	86.76	90.38	90.46
Crude protein	7.26	7.88	9.63	7.88	18.38
Crude fibre	14.03	13.68	10.14	13.73	14.04
Ether extract	15.85	16.23	13.07	14.23	14.58
Organic matter	94.11	96.42	95.36	92.36	91.62
Neutral detergent fibre	33.58	34.12	26.27	26.27	25.68
Acid detergent fibre	24.11	15.38	15.42	16.59	14.78
Acid detergent lignin	9.48	8.35	8.11	7.98	8.11

lowest, followed by CP content of 7.88% in both D2 and D4 then 9.63% CP in D3 while D5 contained 18.38% CP. The CP content of 7.26% for D1 (0% MOL) was slightly below the minimum level for maintenance of 7.7% for goats (NRC, 1981). However in D2-D4, CP content range of 7.88-9.63% was within this minimum level. The CP content in sole MOL was highest (18.38%). The inclusions of MOL in D2-D4 (5-15% MOL) could be responsible for the increases in the CP contents of the diets D2-D4. These CP contents of 7.88 and 9.63% were just within minimum of 8% necessary to provide the minimum ammonia levels required by rumen micro-organisms to support optimum rumen activity (Norton, 2003). The value of 18.38% CP obtained for sole *Moringa oleifera* leaf meal diet D5 was <20% CP usually reported and was also lower than the values of 23.27% CP reported by Nouala *et al.* (2006) and 22.2% CP by Asaolu *et al.* (2010). This lower value could be due to variation in the level of stalks in the *Moringa oleifera* leaf meal used in the current study.

The CF, NDF, ADF and ADL levels of the experimental diets ranged from 10.14-14.04%; 25.68-34.12%; 14.78-24.11% and 7.98-9.48%, respectively. These values were lower than the ranges of 16.67-21.50% CF; 48.63-52.49% NDF; 20.88-42.51% ADF and 9.73-11.26% ADL reported for similar diets inclusion of *Zizphus mauritiana* browse plant fed to goats of mixed breed (Bornu white and sokoto red) (Njidda *et al.*, 2010). Nouala *et al.* (2006) outlined that the environmental conditions of the rumen are normally in favour of the fibrolytic micro-organisms which aid the degradation of high fibre diets in contrast to the negative effects of concentrates high in carbohydrates.

***In vitro* fermentation parameters:** Table 2 and Fig. 1 show the *in vitro* fermentation parameters of diets D1-D5. At the 24 h incubation period, the highest ($p<0.05$) potential gas volume of 37.00 mL/200 mg DM was obtained for both diets D1 (0% MOL) and D3 (10% MOL). This was probably due to the high content of degradable carbohydrate source (dried cassava peels) and protein source (palm kernel cake) present in the diets.

Blummel and Becker (1997) explained that generally gas production is a function and a mirror of the degradable carbohydrate in the diet and thus, the amount of gas produced depends on the nature of the carbohydrate. Also, this might be attributed to the fact that there might have been more microbial access of the degrading fungi (in the rumen liquor) in these diets which encouraged their degradation after inoculation, thereby increasing gas production rate with the decreased particle sizes of the diet samples incubated as explained by Menke and Steingass (1988).

The highest ($p<0.05$) potential gas production volume of 37.00 mL/200 mg/DM was obtained for both treatments D1 (0% MOL) and D3 (10% MOL) while the lowest value of 26.00 mL/200 mg DM was recorded for D5 (100% MOL), the test ingredient (Table 2, Fig. 1). This was probably due to the higher CP content of 18.38% in diet D5 as compared to the CP content of 7.26% in D1 and 9.63% in D3. Babayemi and Bamikole (2006) explained that there is relatively lower gas production from diets having higher protein content. The volume of gas produced from the experimental diets D1-D5 were consistently highest for D1 (0% MOL) as shown in Fig. 1. This probably could be attributed to the differences in composition and the nature of the fibre in the diets.

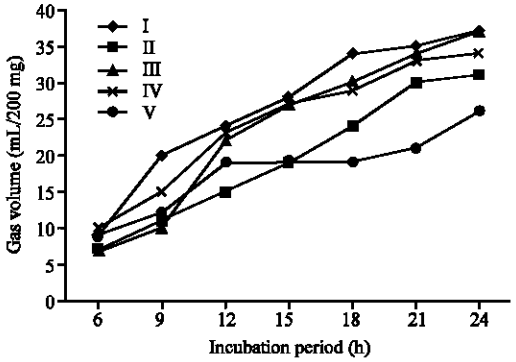

Fig. 1: *In vitro* gas production of the experimental diets and test ingredient at various levels of inclusions of *Moringa oleifera* Leaf meal (MOL); I = D1-0% MOL; II = D2-5% MOL; III = D3-10% MOL; IV = D4-15% MOL; V = D5-100% MOL

Table 2: *In vitro* gas production characteristics of experimental diets

Diets	a	a+b	b	c	t	y
D1 (0% MOL)	6.50^{a}	37.00^{a}	30.50^{a}	0.096^{a}	12.00^{b}	25.00^{a}
D2 (5% MOL)	7.00^{a}	31.00^{c}	24.00^{c}	0.053^{c}	13.50^{a}	21.00^{b}
D3 (10% MOL)	5.00^{b}	37.00^{a}	32.00^{a}	0.065^{b}	12.00^{b}	22.00^{a}
D4 (15% MOL)	7.00^{a}	34.00^{b}	27.00^{b}	0.078^{a}	12.00^{b}	23.00^{a}
D5 (100% MOL)	7.00^{a}	26.00^{d}	19.00^{d}	0.068^{b}	10.50^{c}	17.00^{c}
SEM	0.97	2.62	2.76	0.020	1.45	3.18

$^{a-d}$Means on the same column with different subscripts are significantly different ($p<0.05$); y = Volume of gas produced (mL/200 mg DM) at time, t; a = Gas production (mL) from the soluble fraction; b = Gas production from an insoluble fraction; c = Gas production rate constant from insoluble fraction, b; a+b = Potential gas production (mL), t = Incubation time

Table 3: Metabolizable energy (MJ/kg DM), organic matter digestibility (%) and short chain fatty acid (μmol) of the experimental diets

Diets	ME	OMD	SCFA
D1 (0% MOL)	8.05[a]	61.42[a]	0.82[a]
D2 (5% MOL)	6.90[c]	46.22[d]	0.68[c]
D3 (10% MOL)	7.81[a]	52.41[b]	0.82[a]
D4 (15% MOL)	7.31[b]	49.15[c]	0.75[b]
D5 (100% MOL)	6.82[d]	46.81[d]	0.56[d]
SEM	0.55	1.70	0.03

[a-d]Means on the same column with different subscripts are significantly different ($p<0.05$); ME = Metabolizable Energy; OMD = Organic Matter Digestibility; SCFA = Short Chain Fatty Acid

The nature of fibre contained in D1 (0% MOL) probably yielded slightly more gas during the fermentation process. This was explained by Odenyo *et al.* (1999) who reported that many factors determine the amount of gas produced during fermentation and this may depend on the nature and level of fibre, the presence of secondary metabolites and the potency of the rumen liquor used for incubation.

Metabolizable energy, organic matter digestibility and short chain fatty acid levels: As shown in Table 3, the values of ME, OMD and SCFA ranged between 6.82 and 8.05 MJ kg^{-1} DM; 46.22 and 61.42% and 0.56 and 0.82 μmol, respectively. Significant differences were observed for all the parameters measured and treatment diet D1 recorded the highest ($p<0.05$) values consistently. These values of ME, OMD and SCFA levels compared well with the range of values of 6.66-7.11 MJ kg^{-1} DM for ME; 52.94-58.18% for OMD and 0.62-0.77 μmol for SCFA, for ensiled cassava tops, guinea grass mixtures plus energy additives as reported by Binuomote *et al.* (2010).

CONCLUSION

The diet (D1-0% MOL) without the inclusion of *Moringa oleifera* leaf meal had the highest values of *in vitro* gas production parameter measurements, including the ME, OMD and SCFA, as compared to the other diets with *Moringa oleifera* leaf meal inclusions. The best level of *Moringa oleifera* leaf meal inclusion in the diets was 10% (D3-10% MOL) while the sole *Moringa oleifera* leaf meal diet (D5-100% MOL) recorded the lowest *in vitro* gas production parameter values. This study, thus demonstrated that *Moringa oleifera* leaf meal when fed in combination with conventional concentrate ingredients could enhance its utilization as feed for ruminants.

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