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Full Length Research Paper

In vitro fermentation characteristics and acceptability by West African dwarf goats of some dry season forages

Babayemi Olaniyi Jacob

Department of Animal Science, University of Ibadan, Ibadan, Nigeria E-mail: oj.babayemi@mail.ui.edu.ng.

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Total weight gained by small ruminants in the rainy season is lost in the dry season due to feed scarcity. Feed supplementation with dry season browse trees, legumes and grasses for ruminants is prudent. Proximate composition, acceptability using twenty four West African dwarf (WAD) goats and *in vitro* gas production of dry season forages were determined. Crude protein content ranged from 7.9% in *Panicum maximum* to 25.7% in *Leucaena leucocephala*. Crude fibre composition was generally high (range 56.8 - 74.1%) among the forages. *Pa. maximum*, *Gliricidia sepium, L. leucocephala* and *Terminalia catappa* were acceptable as coefficient of preference was above unity. Net gas production (NGP) and potential gas (PG) ranged from 41 - 59.7 ml/24 h and 98.5 - 4,545 ml, respectively. Highest (P < 0.05) and lowest (P > 0.05) gas (production were obtained in *Gliricidia* and *Leucaena*, respectively. The value for the metabolizable energy (ME) (MJ/kg DM), organic matter digestibility (OMD) (%) and short chain fatty acids (SCFA) (µml) ranged from 8.31 - 1.88, 57.68 - 81.09 and 0.90 - 1.35, respectively. The ME, OMD and SCFA were highest (P < 0.05) in *G. sepium*. Methane (ml/200 mg DM) production ranged from 18 – 24, the highest being from *G. sepium* and the least from *Pennisetum purpureum*.

Key words: Grass, legumes, browse trees, preference, gas production, goats.

INTRODUCTION

Inadequate nutrition is one of the factors that generally affect livestock productivity. Despite the naturally endowed vegetations, there are still inadequate feeds and feedstuffs for livestock in Nigeria. Period of dry season is always a stressful circumstance for livestock, as the environment is characterised by insufficient feed, occasioned by scarce forage and fibrous standing hays. The negative effect of the period is obvious in the lost weight, reduced milk production and high mortality of the animals. Incidence of disease outbreak is rampant as a result of low immunity arising from malnutrition. Ruminants relish guinea grass (Bamikole and Babayemi, 2004) but such forages become very scarce in the dry season. This situation causes the nomads to travel long distances in search of greener pastures for their livestock (Iyayi et al., 2003) and in the process, loosing their animals to snake bite and exposure to inclement weather. In the Southern part of Nigeria, livestock farmers make supplementary use of the common legume trees including *Leucaena leucocephala* and *Gliricidia sepium* (Babayemi et al., 2006b). Due to the scarcity of other known forages, there is always high pressure on these tree legumes and therefore become inadequate to sustain the usual prolonged dry season periods. The search for and revalidation of other known forages is necessary if livestock industry in Nigeria must grow and be sustained. There are a number of underused forages and browse plants that are well adapted and may be sustainable for Nigerian economy. Such forages have long been ignored and almost forgotten by the livestock keepers as research updates on their nutritive value is scanty.

Centrosema pubescens and *Pueraria phaseoloides* are creeping legumes, high in crude protein and well adapted to varying weather. Apart from being relished by ruminants, farmers often use these legumes for soil reclamation. The legumes are not only going into extinction, their utilisation is limited to research stations. *Pennisetum purpureum* is available throughout the year in the growing areas. Ruminants prefer *Panicum maximum* to *Pe. Purpureum* as the latter grows tall and has sharp edges. However, *Pe. purpureum* is advantageous, as it thrives well in both water and upland places. These arrays of forages need to be reassessed for their nutritive value. Free choice intake (Babayemi et al., 2006b) and *in vitro* fermentation techniques (Babayemi and Bamikole, 2006a) are quick means of evaluating and revalidating nutritive value of feedstuffs.

The *in vitro* gas production method is accurate and predicts feed intake, digestibility, microbial nitrogen supply and animal performance (Blummel and Ørskov, 1993). For the past two decades, the technique had been used in advanced countries as an instrument to determine the amount of short chain fatty acids, carbon dioxide and metabolizable energy of feed for ruminants (Blummel and Becker, 1997; Getachew et al., 1999). Methane is an important gas among gases produced by ruminants at fermentation, and has been reported (Baba-yemi and Bamikole, 2006a) to be an energy loss to the animals and when emitted, it contributes to the des-truction of ozone layer. The *in vitro* fermentation techni-que is capable of quantifying the amount of methane (energy loss) production (Fievez et al., 2005). The pre-sent study was designed to determine the proximate composition, free choice intake and *in vitro* gas production kinetics of some grasses, legumes and browse trees in Southwest Nigeria.

MATERIALS AND METHODS

Collection of forages

The legumes (*C. pubescens* and *Pu. phaseoloides*), grass (*Pa. maximum* and *Pe. purpureum*), browse trees (*L. leucocephala*, *G. sepium* and *Terminalia catappa*) were collected at the pick of dry season, between the months of February and March, 2006 from the Teaching and Research Farm, University of Ibadan, Ibadan, Nigeria. The ages of legumes were not known. The browse trees and grasses were strategically cut back to obtain a regrowth of 12 and 6 weeks, respectively. The location is 7º27'N and 3º45'E at altitude 200 - 300 m above sea level; mean temperature of 25 - 29°C and the average annual rainfall of about 1250 mm. Dry matter composition of the forages and browse plants were carried out immediately at 105ºC.

Proximate composition

Crude protein, crude fibre, ether extract and ash contents of the grasses, legumes and browse plants were determined (AOAC, 1990) and the amount of crude protein was calculated (N x 6.25).

Acceptability study

Twenty four West African dwarf female goats weighing 10 – 12 kg and about two years old were used to evaluate the free choice intake of *Pa. maximum*, *Pe. purpureum*, *C. pubescens*, *Pu. phaseoloides*, *G. sepium*, *L. leucocephala* and *Terminalia catappa*. The goats were purchased from the surrounding villages of the University, Ibadan, Nigeria. The animals were immediately placed on prophylactic treatment through the administration of antibiotics (long acting). Animals were also treated against endoparasites and ectoparasites using 10% levamisol and diasuntol, respectively. During the adaptation period, which lasted for four weeks, the goats were fed only with the feedstuffs they were served from where they were purchased, including wheat bran, corn glutton, cassava peels and corn offals. An open pen that had been designed to accommodate 15 – 25 matured goats was used. The pen wall was 4 m high and raised by strong wood to 10 m height. The floor was made of concrete and the top covered with corrugated iron. The floor of the pen was covered with wood shavings to a depth 3 cm for the absorption of urine and faeces. In triplicates, 25 kg each of the fresh forages were placed in strategic locations in feeder troughs measuring 2 m x 5 m. The goats were allowed to feed from 10:00 to 18:00 h daily and for upward of 14 days. Consumption was measured by deduction of remnants from the amount of feed served. The forage preferred was assessed from the coefficient of preference (COP) value, calculated from the ratio between the intakes for the individual forages, divided by the average intake of the forages (Babayemi et al., 2006a). Therefore, forage was inferred to be relatively acceptable provided the COP was greater than unity.

In vitro gas production study

Rumen fluid was obtained from three West African dwarf female goats. The method for collection was as previously described (Babayemi and Bamikole, 2006a) using suction tube from goats were previously fed with 40% concentrate feed (40% corn, 10% wheat offal, 10% palm kernel cake, 20% groundnut cake, 5% soybean meal, 10% dried brewers grain, 1% common salt, 3.75% oyster shell and 0.25% fish meal) and 60% *Pa. maximum* at 5% body weight. The rumen liquor was collected into the thermo flask that had been pre-warmed to a temperature of 39ºC from the goats before they were offered the morning feed. Incubation procedure was as reported by Menke and Steingass (1988) using 120 ml calibrated transparent plastic syringes with fitted silicon tube. The sample weighing 200 mg (n = 3) was carefully dropped into the syringes and thereafter, 30 ml inoculums containing cheese cloth strained rumen liquor and buffer (g/liter) of 9.8 $NaHCO_3$ + 2.77 Na_2HPO_4 + 0.57 KCl + 0.47 NaCl + 2.16 $MgSO_4.7H_2O$ + 0.16 $CaCl_2.2H_2O$) (1:4 v/v) under continuous flushing with CO_2 was dispensed using another 50 ml plastic calibrated syringe. The syringe was tapped and pushed upward by the piston in order to completely eliminate air in the inoculums. The silicon tube in the syringe was then tightened by a metal clip so as to prevent escape of gas. Incubation was carried out at 39 ± 1ºC and the volume of gas production was measured at 6, 12, 18, 24 and 30 h. At post incubation period, 4 ml of NaOH (10 M) was introduced to estimate methane production as reported by Fievez et al. 2005). The post incubation parameters such as metabolisable energy, organic matter digestibility and short chain fatty acids were estimated at 24 h post gas collection according to Menke and Steingass (1988). The average of the volume of gas produced from the blanks was deducted from the volume of gas produced per sample.

Calculation and statistical analysis

The volume of gas produced at intervals was plotted against the incubation time, and from the graph, the gas production characteristics were estimated using the equation $y = a + b (1 – e^{-ct})$ as described by Ørskov and McDonald (1979), where y = volume of gas produced at time 't', a = intercept (gas produced from the soluble fraction), b = gas production from the insoluble fraction, c = gas production rate constant for the insoluble fraction (b), t = incubation time. Metabolisable energy (ME) was calculated as ME = 2.20 + 0.136GV + 0.057 CP + 0.0029 CF (Menke and Steingass, 1988). Organic matter digestibility (OMD %) was assessed as OMD = 14.88 + 0.889 GV + 0.45 CP + 0.651 XA (Menke and Steingass, 1988). Short chain fatty acids (SCFA) as 0.0239 GV – 0.0601 (Getachew et al., 1999) was also obtained, where GV, CP, CF and

Table 1. Proximate compositions (g/100 g DM) of some dry season grasses, legumes and browse plants.

Forage sample	DM	CP	CF	EE	Ash
Panicum maximum	35.13	7.9	74.1	9.8	10.1
Pennisetum purpureum	37.7	11.4	69.3	12.5	10.8
Centrosema pubescens	40.4	16.0	61.4	2.6	4.1
Pueraria phaseoloides	17.9	22.0	58.9	2.1	3.6
Gliricidia sepium	27.02	25.7	66.3	2.3	3.6
Leucaena leucocephala	34.43	27.6	67.5	3.5	3.7
Termilania catappa	20.98	8.9	56.8	4.7	5.3

DM = Dry matter, CP = Crude protein, CF = Crude fibre, and EE = Ether extract.

Table 2. Mean dry matter intake (MDI) (kg DM) and coefficient of preference (COP) by WAD goats of some dry season grasses, legumes and browse plants.

Forage sample	Acceptability day									
	2		4		6		8		10	
	MDI	COP	MDI	COP	MDI	COP	MDI	COP	MDI	COP
Panicum maximum	3.3	1.52	3.8	1.31	2.8	1.04	1.10	0.4	2.7	1.0
Pennisetum purpureum	1.4	0.65	3.3	1.14	1.8	0.67	1.4	0.6	1.5	0.54
Centrosema pubescens	0.4	0.18	0.9	0.31	1.5	0.55	2.7	1.07	1.6	0.58
Pueraria phaseoloides	0.9	0.41	1.0	0.35	1.4	0.52	2.6	1.03	1.4	0.51
Gliricidia sepium	4.3	1.98	4.8	1.66	5.6	2.07	3.0	1.19	4.7	1.70
Leucaena leucocephala	2.0	0.92	2.4	0.80	2.3	0.82	3.1	1.23	3.9	1.41
Termilania catappa	2.9	1.34	4.0	1.38	3.5	1.30	3.8	1.50	3.6	1.30

XA are total gas volume, crude protein, crude fibre and ash, respectively. Data obtained were subjected to analysis of variance. Where significant differences occurred, the means were separated using Duncan multiple range F-test of the SAS (1988) options.

RESULTS AND DISCUSSION

Proximate composition

In Table 1, the dry matter and proximate composition of the forages are shown. The least (7.9%) and the highest (25.7%) CP was in *Pa. maximum* and *L. leucocephala*, respectively. Crude fibre was generally high (range 56.8 - 74.1%) among the forages. The value of protein among the grasses, legumes and browse plants was within the range reported (Bamikole and Babayemi, 2004). Since the amount of crude protein for the guinea grass and other grasses in the present study might not be adequate to meet the requirement for ruminants, proper supplementation with *L. leucocephala* had been reported to enhance livestock performance (Babayemi et al., 2006b).

Acceptability of forage

Presented in Table 2 is the acceptability of the forages. Among the grasses, *Pa. maximum* was highly preferred to *Pe. purpureum*. The reason for the less preference for elephant grass was probably because of its coarse and tough nature. In a free grazing trial, elephant grass was not consumed by West African dwarf goats (Babayemi and Bamikole, 2006b). The two legumes seemed to have been preferred equally but were not initially consumed in the first two days of offering to the animals. The animals generally showed willingness to consume *G. sepium* and *T. catappa* from day 1 to day 10 than *L. leucocephala*. Goats were observed to be nonchalant to the initial consumption of *Gliricidia* but about 2 hours later, they now crowded on the browse plant. Poor receptivity of goats to *L. leucocephala* although with a high protein, could be due to the presence of antinutrtional factor in it (Babayemi et al., 2006b). In a rangeland grazing studies, unlike *Leucaena leucocephala*, *G. sepium* was not browsed by goats (Babayemi and Bamikole, 2006b) suggesting that goats likely prefer browsing on leucaena in the range rather than that of the cut plant.

In vitro gas production

Presented in Table 3 are net gas productions (NGP), immediately soluble gas (ISG), potential gas (PG), rate of gas production (RGP), potential gas production (PGP) and effective gas production (EGP) of the forages. The

Table 3. *In vitro* gas production characteristics of some dry season forages.

Forage sample	Gas production characteristics					
	Net gas (ml/24 h)	Soluble (ml)	Potential (ml)	Rate (ml/h^{-1})	PGP (ml)	EGP (ml)
Panicum maximum	45d	6.61b	1228^{d}	0.00083^{d}	1237^{c}	37.55^{d}
Pennisetum purpureum	45.7d	9.80a	744.1^{e}	0.0012^{c}	739.1^{d}	35.77^{d}
Centrosema pubescens	49.7c	4.38c	1998^{b}	0.00068^{de}	2050^{b}	47.41^{b}
Pueraria phaseoloides	54.3b	2.49d	4545.1^{a}	0.0004^{e}	4602.5^{a}	56.76^{a}
Gliricidia sepium	59.7a	9.44a	670.1^{e}	0.0019^{b}	680.1^{d}	48.29^{b}
Leucaena leucocephala	41e	2.81d	98.5^{f}	0.0253^{a}	97.5^{e}	42.77^{c}
Termilania catappa	43.3e	6.58b	1310.1c	0.0007^{de}	1328.5^{c}	37.07^{d}
SEM	0.667	0.20	12.9	0.000048	26.70	0.50

PGP = potential gas production and EGP = effective gas production.

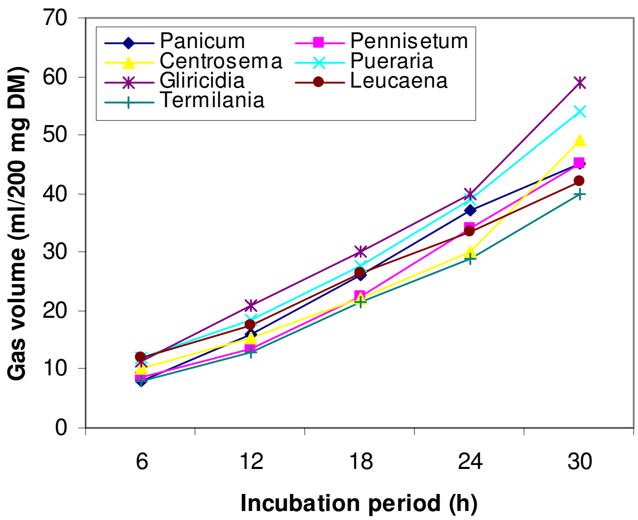

Figure 1. *In vitro* gas production of some grasses, legumes and browse plants.

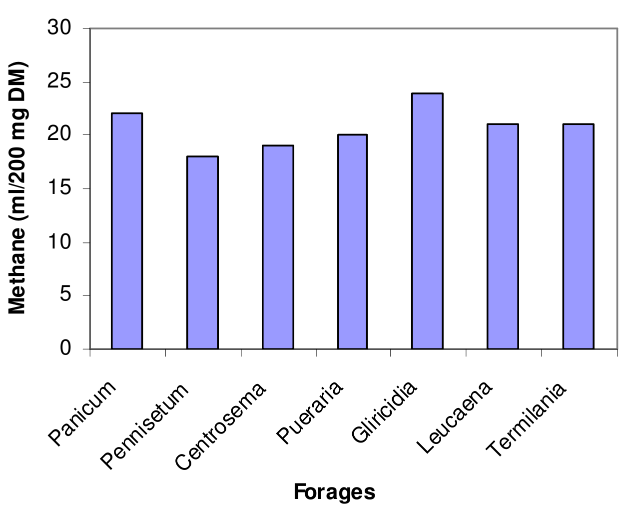

Figure 2. Methane (ml/200 mg DM) production grasses, legumes and browse plants.

NGP, ISG, PG, RGP, PGP and EGP ranged between 41 and 59.7, 2.49 and 9.8, 98.5 and 4,545.1, 0.004 and 0.0253, 97.5 and 4,692.5 and 35.77 and 56.76, respectively. Significant differences ($P < 0.05$) occurred among the forages in NGP, ISG, PG, RGP, PGP and EGP. The highest and the lowest gas production were obtained from *Gliricidia* and *Leucaena* respectively. Gas production between guinea grass and elephant grass was similar possibly due to the comparable possession of crude fibre. This observation was not noticed for *Gliricidia* and *Leucaena*. There are many factors that may determine the amount of gas to be produced during fermentation, depending on the nature and level of fibre, the presence of secondary metabolites (Babayemi et al., 2004a) and potency of the rumen liquor for incubation. It is possible to attain potential gas production of a feedstuff if the donor animal from which rumen liquor for incubation was collected got the nutrient requirement met. Generally, gas production is a function and a mirror of degradable carbohydrate and therefore, the amount depends on the nature of the carbohydrates (Demeyer and Van Nevel, 1975**;** Blummel and Becker, 1997). The *in vitro* gas production pattern of the forages shown in Figure 1 indicated that more degradation of dry matter were still possible beyond 30 h. The situation here depicted that of typical dry season in Nigeria, when most of the forages are fibrous and therefore take longer time to degrade in the rumen. The highest gas production was obtained from *Gliricidia* and the lowest from *Leucaena* and *Termilania* for reason that was not really clear. This study did not determine the presence of secondary metabolites in the forages, although high crude protein in feed enhances microbial multiplication in the rumen, which in turn determines the extent of fermentation. The phenol in Leucaena might had been responsible for its low gas production (Babayemi et al., 2004b). Methane (ml/200 mg DM) production (Figure 2) ranged from 18 to 24 among the forages, the least and the highest being from *Pe. purpureum* and *G. sepium*, respectively. In most cases, feedstuffs that show high capacity for gas production are

Table 4. Metabolizable energy (ME) (MJ/Kg DM), organic matter digestibility (OMD) (%) and short chain fatty acids (SCFA) (µmol) of some forages.

Forage samples	*In vitro* gas parameters		
	ME	OMD	SCFA
Panicum maximum	8.90 ± 0.84^{f}	64.68 ± 2.44^{f}	1.02 ± 0.01^{d}
Pennisetum purpureum	9.18 ± 0.59^{e}	66.68 ± 1.29^{e}	1.02 ± 0.03^{d}
Centrosema pubescens	9.95 ± 0.91^{c}	68.14 ± 1.74^{c}	1.11 ± 0.02^{c}
Pueraria phaseoloides	10.97 ± 0.88^{b}	74.98 ± 2.39^{b}	1.23 ± 0.02^{b}
Gliricidia sepium	11.88 ± 0.75^{a}	81.09 ± 1.87^{a}	1.35 ± 0.01^{a}
Leucaena leucocephala	8.31 ± 0.43^{g}	57.68 ± 1.25^{g}	0.90 ± 0.01^{f}
Termilania catappa	9.68 ± 0.62^{d}	66.90 ± 1.82^{d}	0.94 ± 0.01^{e}

Means on the same column with different superscripts are significantly different (P < 0.05).

also observed to be synonymous for high methane production. Methane production indicates an energy loss to the ruminant and many tropical feedstuffs have been implicated to increase methanogenesis (Babayemi et al., 2004a; Babayemi and Bamikole, 2006a; Babayemi and Bamikole, 2006b) as an integrated part of carbohydrate metabolism(Demeyer and Van Nevel, 1975).

Metabolizable energy (ME), organic matter digestibility (OMD) and short chain fatty acids (SCFA) of the forages are presented in Table 4. The value for the ME, OMD and SCFA ranged from 8.31 in *Leucaena* to 11.88 in *Gliricidia,* 57.68 in *Leucaena* to 81.09 in *Gliricidia* and 0.90 in *Leucaena* to 1.35 in *Gliricidia*, respectively. There were significant differences (P < 0.05) among the forages in ME, OMD and SCFA. In all these parameters, *Gliricidia* was observed to be the highest and *Leucaena* the lowest. The values obtained in the present study were higher than those reported for agro-industrial wastes (Aregheore and Abdulrazak, 2005), *Tephrosia candida* and guinea grass mixtures (Babayemi and Bamikole, 2006a) and spent tea leaf (Babayemi et al., 2006c). Feedstuffs that are inherent with certain antinutritive factor had been reported to be low in metabolisable energy and organic matter digestibility (Aregheore and Abdulrazak, 2005).

Conclusion

The study has exhibited that the grass (*Pa. maximum* and *Pe. purpureum*), legumes (*C. pubescens* and *Pu. phaseoloides*) and browse trees (*G. sepium*, *L. leucocephala* and *T. catappa*) are prospective forages for ruminants in the dry season. Proximate composition designated that the selected legumes and browse trees might be practical protein supplements. However, coefficient of preference demonstrated that *Pa. maximum* and the three browse plants were fully accepted by goats while *Pe. purpureum* and the two legumes were rejected. The organic matter digestibility of all the forages was high and culminated in enhanced total gas production, predicted metabolisable energy and short chain fatty acids. *Pa. maximum* and *G. sepium* were highest in methane production, suggesting an inclusion into a feed that may suppress methanogenesis.

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