

## Determining Dry Matter Degradability of Some Semi-Arid Browse Species of North-Eastern Nigeria Using the *In Vitro* Technique

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**ABSTRACT:** The *in vitro* gas production of some semi-arid browse species were evaluated. The relationship between *in vitro* gas measured on incubation of tannin-containing browses in buffered rumen fluid and calculated from short chain fatty acid (SCFA) production was investigated. Crude protein (CP) contents in the browses ranged from 13.85 to 16.65% dry matter (DM). The NDF, ADF and ADL were 37.30 to 51.20, 16.20 to 41.90 and 4.90 to 12.70 g/100g DM respectively. Total condensed tannin (TCT) ranged from 0.15 to 0.39 mg/g DM. The TCT was significantly correlated with gas production ( $r = 0.99$ ;  $P < 0.05$ ). A good relationship ( $R^2 = 0.99$ ;  $P < 0.05$ ) was observed between the measured *in vitro* gas production and that calculated from SCFA. The relationship between *in vitro* gas measured on incubation of browse leaves and that calculated from SCFA allows the prediction of SCFA from gas production. The study showed that the leaves of the browse forages had reasonable nutritive value and thus, may serve as potential supplement for ruminants in Nigeria.

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### INTRODUCTION

Shrub and tree leaves are an important component of diets for goats, cattle, deer, game, and sheep (Papachristou and Nastis, 1996) and play an important role in the nutrition of grazing animals in areas where few or no alternatives are available (Meuret *et al.*, 1990). The presence of tannins and other phenolic compounds in the large number of nutritionally important shrubs and tree leaves hamper their utilization as animal feed (Tolera *et al.*, 1997). High levels of tannins in leaves restrict the nutrient utilization and decrease voluntary food intake, nutrient digestibility and N retention (Silanikove *et al.*, 2001).

The leaves of the evergreen tree and shrub are used as emergency food by goat and sheep in the semi-arid region of North-eastern Nigeria. However, there is little information on their nutritive values. Chemical composition, in combination with *in vitro* digestibility and ME content can be considered useful indicators for preliminary evaluation of the potential nutritive value of previously uninvestigated shrub and tree leaves (Ammar *et al.*, 2005). Current chemical analytical techniques do not reflect the biological effects of tannin therefore the use of *in vitro* techniques has been proposed to supplement the chemical analysis (Nsahlai *et al.*, 1994). The gas production technique has

proved to be efficient in determining the nutritive value of feeds containing anti-nutritive factors (Siaw *et al.*, 1993). The present study was undertaken to determine the nutritive value of some semi-arid browse of North-eastern Nigeria using the *in vitro* gas production technique.

### MATERIALS AND METHODS

#### Experimental location and forage samples:

All the forages were harvested from Gwoza Local Government Area of Borno State Nigeria. The area is located at 11.05° North and 30.05° East and at an elevation of about 364 above sea level in the North Eastern part of Nigeria. The ambient temperature ranges between 30°C and 42°C being the hottest period (March to June) while its cold between November to February with temperatures ranging between 19 - 25°C (Ijere and Daura, 2000).

Eight indigenous browse plants (leaves) commonly consumed by ruminants animals were sampled and used in this study. The species were: *Ficus polita*, *Ficus thonningii*, *Batryospermum paradoxum*, *Kigalia africana*, *Celtis integrifolia*, *Khaya senegalensis*, *Leptadenia lancifolia* and *Ziziphus abyssinica*. The browses were harvested from at least 10 trees per each specie selected at random in

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four locations with the study area at the end of the rainy season. The harvested samples were then pooled for each individual tree species and then oven dried at 105°C for 24 hours to a constant weight and ground to pass through a 1.0mm, sieve. The samples were then sub-sample to obtain three samples for each tree species and used for the laboratory analysis.

**Chemical Analysis:** Browse species were analyzed for dry matter (DM), crude protein (CP), ether extract (EE), crude fibre (CF) and ash according to AOAC (2005). Crude protein was calculated as  $N \times 6.25$ . The leaves samples were analyzed for neutral detergent fibre (NDF), acid detergent fibre (ADF), acid detergent lignin (ADL) and cellulose according to Van Soest *et al* (1991). Total condensed tannin was (Polshettiwar *et al.*, 2007).

***In vitro* gas production:** Rumen fluid was obtained from 3 WAD female sheep through suction tube before morning feed, normally fed with 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. Incubation was as reported by Fievez *et al.* (2005), using 120 ml calibrated syringes in three batch incubation at 39°C. Into 200 mg sample ( $n = 7$ ) in the syringe was introduced 30 ml inoculums containing cheese cloth strained rumen liquor and buffer ( $\text{NaHCO}_3 + 3 \text{Na}_2 \text{HPO}_4 + \text{KCl} + \text{NaCl} + \text{MgSO}_4 \cdot 7\text{H}_2\text{O} + \text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ ) (1:4, v/v) under continuous flushing with  $\text{CO}_2$ . The gas production was measured at 3, 6, 9, 12, 15, 18, 21 and 24h. After 24 h post incubation, 4 ml of sodium hydroxide (10 M) was introduced to estimate methane production. The average of the volume of gas produced from the blanks was deducted from the volume of gas produced per sample.

***In vitro* Dry Matter Digestibility (IVDMD):** After 24h digestion, the samples were transferred into test tubes and centrifuge for 1 hour in order to obtain the residues which was then filtered using Whatman No 4 filter paper by gravity and the residues placed in crucible for drying at 65°C for 24h. The dry residues were weighed and digestibility calculated using the equation as follows:

$$\text{IVDMD}(\%) = \frac{\text{Initial DM Input} - \text{DM residue} - \text{Blank}}{\text{Initial DM Input}} * 100$$

**Statistical analysis:** Metabolisable Energy (ME) was calculated as  $\text{ME} = 2.20 + 0.136\text{GV} + 0.057 \text{CP} + 0.0029 \text{CF}$  (Menke and Steingass, 1988). Organic matter digestibility (OMD%) was assess as  $\text{OMD} = 14.88 + 0.889 \text{GV} + 0.45 \text{CP} + 0.651 \text{XA}$  (Menke and Steingass, 1988). Short Chain Fatty Acids (SCFA) as  $0.0239 \text{GV} - 0.0601$  (Getachew *et al.*, 1999), where GV, CP, CF and XA are the totals of 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 the Duncan Multiple Range Test of the (SAS 1988) options.

### RESULTS AND DISCUSSION

The result of the detailed proximate composition of the leaves of the browse forages is presented in Table 1.

The crude protein (CP) contents of the browses studied fall within the range reported for West African browse species (Rittner and Reed 1992). All the browses used in the current study had a CP content of above 13% of DM. The results of the current study, those of Rittner and Reed (1992) and Makkar and Becker (1998) indicate that most tropical browse species are high in CP and can be used to supplement poor quality roughages which increase productivity of ruminant livestock in tropical regions. The tannin content of the browse was low 0.08 to 0.38 mg/g DM. The tannin values in the browse reported could be higher than the values obtained in this study, since a considerable amount of tannins are bound to either fibre and /or proteins and remain unextracted (Jackson, *et al.*, 1996) The beneficial effect of tannins when forages containing low levels of tannins are fed could be due to the protection of protein from microbial degradation by tannins, thus increasing the amount of undegraded protein

entering the small intestine (Barry *et al.* 1986). In addition, a higher flow of microbial protein to the intestine as a result of higher efficiency of microbial protein synthesis (Getachew *et al.*, 2000) has been observed. However, higher

concentration of tannins in the diet is associated with reduction in organic matter digestibility (Silanikove *et al.*, 1997; Waghorn and Shelton, 1997).

**Table 1: Proximate composition (% DM) of semi arid browses NDF, ADF, ADL (g/100g DM); TCT (mg/g DM)**

Browse Forages	DM	CP	EE	Ash	NDF	ADF	ADL	TCT	IVDMD
<i>Ficus polita</i>	95.20	16.21 <sup>a</sup>	3.00 <sup>c</sup>	10.00 <sup>d</sup>	37.30 <sup>g</sup>	27.20 <sup>d</sup>	4.90 <sup>d</sup>	0.20 <sup>b</sup>	63 <sup>c</sup>
<i>Ficus thonningii</i>	95.20	16.47 <sup>a</sup>	2.00 <sup>d</sup>	18.00 <sup>a</sup>	51.20 <sup>a</sup>	41.20 <sup>a</sup>	10.00 <sup>c</sup>	0.24 <sup>b</sup>	63 <sup>c</sup>
<i>B. paradoxum</i>	95.80	14.63 <sup>b</sup>	5.00 <sup>a</sup>	8.00 <sup>c</sup>	47.60 <sup>b</sup>	32.10 <sup>b</sup>	11.60 <sup>b</sup>	0.19 <sup>c</sup>	64 <sup>c</sup>
<i>Kigalia Africana</i>	96.40	13.85 <sup>c</sup>	3.00 <sup>c</sup>	18.00 <sup>a</sup>	38.40 <sup>f</sup>	29.60 <sup>c</sup>	12.10 <sup>a</sup>	0.08 <sup>d</sup>	64 <sup>c</sup>
<i>Celtis integrifolia</i>	96.20	15.89 <sup>a</sup>	3.00 <sup>c</sup>	16.00 <sup>b</sup>	42.30 <sup>d</sup>	31.20 <sup>b</sup>	12.50 <sup>a</sup>	0.39 <sup>a</sup>	67 <sup>b</sup>
<i>Khaya senegalensis</i>	97.00	14.11 <sup>b</sup>	3.00 <sup>c</sup>	10.00 <sup>d</sup>	44.60 <sup>c</sup>	32.10 <sup>b</sup>	11.30 <sup>b</sup>	0.21 <sup>b</sup>	68 <sup>b</sup>
<i>L. lancifolia</i>	95.80	16.65 <sup>a</sup>	4.00 <sup>b</sup>	18.00 <sup>a</sup>	41.20 <sup>d</sup>	31.70 <sup>b</sup>	12.70 <sup>a</sup>	0.15 <sup>c</sup>	64 <sup>c</sup>
<i>Ziziphus abyssinica</i>	97.00	14.37 <sup>b</sup>	2.00 <sup>d</sup>	14.00 <sup>c</sup>	39.60 <sup>e</sup>	16.20 <sup>e</sup>	5.90 <sup>d</sup>	0.38 <sup>a</sup>	70 <sup>a</sup>
<b>Means</b>	<b>96.07</b>	<b>15.27</b>	<b>3.13</b>	<b>14.00</b>	<b>42.77</b>	<b>30.41</b>	<b>10.13</b>	<b>0.23</b>	<b>65.38</b>
<b>SEM</b>	<b>0.14</b>	<b>0.18</b>	<b>0.12</b>	<b>0.69</b>	<b>0.88</b>	<b>0.82</b>	<b>0.74</b>	<b>0.01</b>	<b>0.42</b>

a, b, c, means in the same column with different superscript differ significantly (P<0.05). DM = Dry matter; CP = Crude Protein; EE = Ether Extract; NDF = Neutral detergent fibre; ADF = Acid detergent fibre; TCT=Total condensed tannin; IVDMD=*In vitro* dry matter degradability

**Table 2: Net Gas Volume, Metabolizable Energy, Organic matter digestibility, Short Chain Fatty Acid of semi-arid browse forages**

Browse Forages	Gas production parameters			
	NGV(ml/200mg)	ME(MJ/Kg)	OMD(%)	SCFA(mmol)
<i>Ficus polita</i>	3.50 <sup>b</sup>	3.72	32.48 <sup>c</sup>	0.03 <sup>c</sup>
<i>Ficus thonningii</i>	4.50 <sup>a</sup>	3.84	37.94 <sup>a</sup>	0.05 <sup>a</sup>
<i>Batryospermum paradoxum</i>	1.50 <sup>d</sup>	3.28	28.44 <sup>e</sup>	-0.02 <sup>e</sup>
<i>Kigalia africana</i>	2.50 <sup>c</sup>	3.39	34.83 <sup>b</sup>	-0.0003 <sup>f</sup>
<i>Celtis integrifolia</i>	4.50 <sup>a</sup>	3.78	36.28 <sup>a</sup>	0.05 <sup>a</sup>
<i>Khaya senegalensis</i>	3.00 <sup>b</sup>	3.49	30.83 <sup>d</sup>	0.01 <sup>d</sup>
<i>Leptadenia lancifolia</i>	4.00 <sup>a</sup>	3.77	37.24 <sup>a</sup>	0.04 <sup>b</sup>
<i>Ziziphus abyssinica</i>	1.50 <sup>d</sup>	3.35	31.62 <sup>c</sup>	-0.02 <sup>e</sup>
<b>MEANS</b>	<b>3.13</b>	<b>3.58</b>	<b>33.70</b>	<b>0.02</b>
<b>SEM</b>	<b>0.16</b>	<b>0.04NS</b>	<b>0.42</b>	<b>0.002</b>

Net Gas Volume (NGV), Metabolizable energy (ME), Organic matter digestibility (OMD), Short Chain Fatty Acids (mmol) of semi-arid browse forages.

Metabolizable energy (ME), organic matter digestibility (OMD) and short chain fatty acids (SCFA) of the browse forages are presented in Table 4. The value for the ME, OMD and SCFA ranged from 3.28 in *Z. abyssinica* to 3.84 MJ/Kg DM in *F. thonningii*, 28.44 in *B. paradoxum* to 37.94% in *F. thonningii*, -0.0003 in *K. africana* to 0.05mmol in *F. thonningii*, respectively. There were significant differences (P<0.05) among the forages in ME, OMD and

SCFA. The values obtained in the present study were similar to those reported for tropical browses (Getachew *et al.*, 2002) but lower to those reported for forages by Babayemi (2007). Feedstuffs that are inherent with certain anti-nutritive factor had been reported to be low in metabolisable energy and organic matter digestibility (Aregheore and Abdulrazak, 2005).

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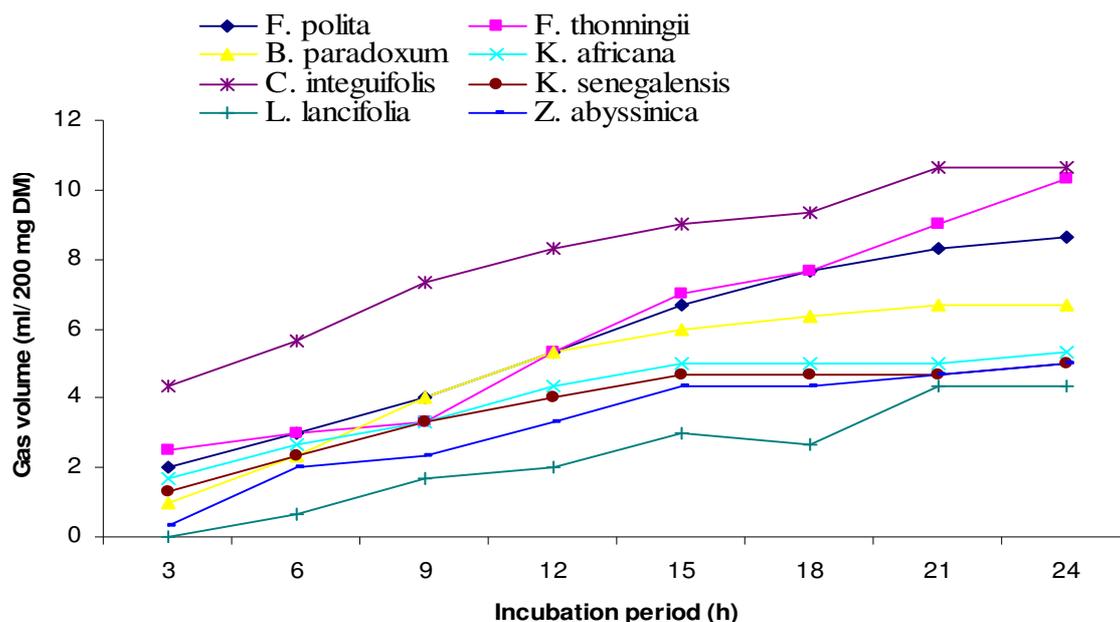


Figure 1: *In vitro* gas production of semi-arid browse forages

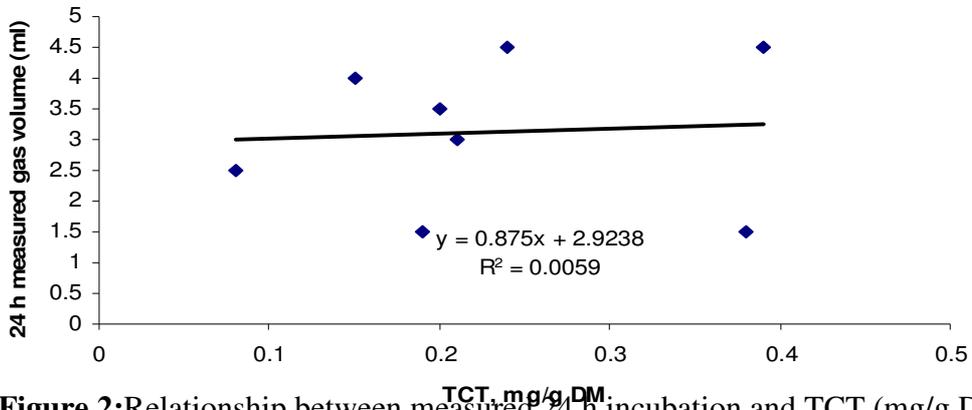
Figure 1 shows the *in vitro* gas production of the browse forages. There was a steady increase in the gas production for over a period of 24h. Significant differences ( $P < 0.05$ ) occurred among the forages in NGV, OMD, ME and SCFA. The highest and lowest gas production were obtained from *Ficus thonningii* and *Ziziphus abyssinica* respectively. 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.*, 2004) and potency of the rumen liquor for incubation. It is possible to attain potential gas production of feedstuff if the donor animal from which rumen liquor for incubation was collected got the nutrient requirement met. Generally, gas production is a function of 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 24 h. The situation here depicted that of typical dry season in Nigeria, when most of

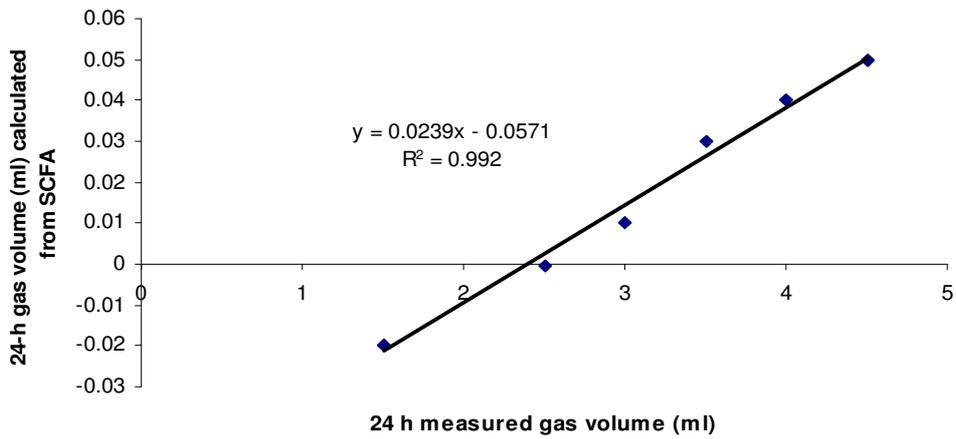
the forages are fibrous and therefore take longer time to degrade in the rumen. The highest gas production was obtained from *Ficus thonningii* and *Ziziphus abyssinica* for reason that was not clear since the secondary metabolites are all within the normal ranges as shown in Table 3 (Paper 1), although high crude protein in feed enhances microbial multiplication in the rumen, which in turn determines the extent of fermentation.

The correlation between *in vitro* gas productions measured after 24 h incubation of tropical browses and that calculated from SCFA was similar to that reported for conventional feeds (Blummel *et al.* 1999). About 94% of the variation in the *in vitro* gas production on incubation of browse leaves was explained by SCFA produced, which mainly comes from carbohydrate fermentation. These results suggest that from browses with a wide range of CP contents, the SCFA production from sources other than carbohydrates is negligible.

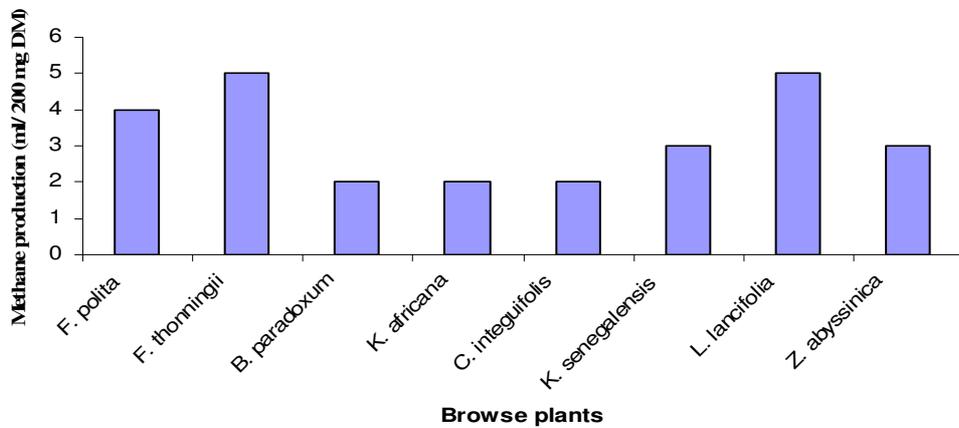
The relatively strong correlation between CT and percent increase in gas production ( $r = 0.99$ ,  $n = 7$ ) observed in the present study was consistent with those of Tolera *et al.* (1997) and Getachew *et al.* (2002).



**Figure 2:** Relationship between measured 24 h incubation and TCT (mg/g DM) browses and SCFA production



**Figure 3:** Relationship between measured gas at 24 h incubation and TCT (mg/g DM)



**Figure 4:** Methane production of semi-arid browse forages of North-eastern Nigeria

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Methane (ml/ 200 mg DM) production (Figure 3) ranged from 2 to 5 among the forages, the least and the highest being from *F. thonningii* and *Z. abyssinica*, respectively. In most cases, feedstuffs that show high capacity for gas production are also observed to be synonymous for high methane production. Methane production indicates an energy loss to ruminant and many tropical feedstuffs have been implicated to increase methanogenesis (Babayemi and Bamikole, 2006b) as an intergrated part of carbohydrate metabolism (Demeyer and Van Nevel, 1975). Similar observation was made by Njidda and Nasiru (2010) for semi-arid browses of North-eastern Nigeria.

The result of the IVDMD is shown in Table 1. IVDMD was higher for all browses with *Z. abyssinica* having the highest (70%). In tree leaves, tannins are present in the NDF and ADF fractions and are tightly bound to the cell wall and cell protein and seem to be involved in decreasing digestibility (Reed *et al.*, 1990). The higher IVDMD observed may be due to the low level of tannin in the browse plants which suggest that it could be a valuable protein supplement in ruminant diets (Aganga and Mosase, 2001). This was further manifested in methane production (Fig. 4) where methane production showed inhibitory features. This suggest that feeds containing high level of CP and low levels of tannin could generate more methane in the rumen.

**Conclusion:** Chemical composition and *in vitro* digestibility can be considered useful indicators for the preliminary evaluation of the likely nutritive value of previously uninvestigated shrubs. Semi-arid browses are forages with high protein concentration and effective *in vitro* DM digestibility. As such, they have potential as a forage for farmers during the long period of dry season when feed is scarce.

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