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Correlation between chemical composition and *in vitro* Dry Matter Digestibility of Leaves of Semi-arid Browses of North-East Nigeria

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Abstract
The potential nutritive values of eight browse forages namely: *Olea hochstteteri*, *Ziziphus mauritiana*, *Ziziphus spinzchristi*, *Pterocarpus erinceus*, *Sterculia setigera*, *Balanites aegyptiaca*, *Ficus sycomorus* and *Adansonia digitata* of North-east Nigeria were evaluated by chemical composition and *in vitro* dry matter digestibility. The samples were collected and analyzed in triplicates. There were significant differences among species in terms of chemical composition. A range of 2.00 to 6.00% and 12.00 to 18.00% DM were recorded for EE and Ash values for the eight browses. Their fiber parameters showed a range of 34.40 to 54.80, 16.55 to 33.40, 9.70 to 67.17, g/100g DM for NDF, ADF and ADL, respectively. The values reported for anti-nutritive factors range from 0.12 to 0.41 mg/g MD for TCT and 0.24 to 0.81 mg/g MD for phenolic. Dry samples (leaves) of eight semi-arid browses were used as substrates. Crude protein (CP) in leaf dry matter ranged from 13.23 in *Olea hochstteteri* to 18.31% DM in *balanites aegyptiaca.* The IVDMD had the lowest IVDMD (45.00% DM) and lowest CP content (13.23%). The result also reveals a negative correlation of IVDMD with cell wall contents (NDF, ADF and ADL) and a positive correlation with TCT, and a positive correlation between CP and phenolic. The result for methane production shows that *Ziziphus spinachisti* had the highest methane value (4ml/200mg DM). Based on chemical composition and *in vitro* dry matter digestibility, the browse species forage have high potential nutritive value, especially as protein supplements to poor quality forage for ruminant animals in the tropics and in terms of rumen and whole tract digestibility.

Keywords: In vitro, Digestibility, Semi-arid, Browse, Tannin

Introduction

In semi – arid regions with a serve dry season, shrubby vegetation represents an integral component of the silvo – pastoral systems and sometimes their graze able material is considered important for the nutrition of range animals (Papachristou *et al*., 1999). However, quiet often, these resources have been disregarded or undervalued mainly because of insufficient knowledge about their potential feeding value. The nutritive value of forage is, in principle, affected by its chemical composition and digestibility which depend largely on plant species, botanical fraction of the plant, seasonally and maturity (Lambert *et al*., 1989; Papachristou and Papanastasis, 1994).

The *in vitro* gas production technique as modified by Menke and stteingass (1988) is widely used to evaluate the nutritive value of feeds resources consumed by ruminants, especially tree and shrub legume forages, particularly to estimate energy value of straws (Makkar *et al*., 1999), agro industrial by-products (Krishna and Gunther, 1987), compound feeds (Aiple *et al*., 1996) and various types of tropical feeds (Krishnamoorty *et al*., 1995). The use of *in vitro* gas method to estimate the digestion of feed is based on measured relationships between the *in vitro* digestibility of feeds and *in vitro* gas production, in combination with the feeds chemical composition (Menke and stteingass, 1988). The main objective of the present study was to investigate changes in chemical composition and *in vitro* digestibility of leaves of semi-arid browses.

Materials and Methods

Location of the study

***Forage Samples*:** Eight indigenous browse samples (leaves) commonly consumed by ruminants animals were used in the study. The species were: *Olea hochstterteri*, *Ziziphus mauritiana*, *Zhiziphus spinzchristi*, *Pterocarpus erinceus*, *Sterculia setegera*, *Balanites aegyptiaca*, *Ficus sycomorus and Adansonia digitata.* All 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-East 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.

***Experimental procedure*:** The browse forages were harvested from at least 10 trees per each species selected at random in our locations within the study area at the end of the season. The harvested samples were the pooled for each individual tree species and oven dried at 105°C for 24h to constant weight and ground to pass through a 1.0mm, sieve. The samples were then sub-sampled 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 extracts (EE), crude fiber (CF) and ash according to AOAC (2005). The leaves samples were analyzed for neutral detergent fiber (NDF), acid detergent fiber (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 fed, 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 120ml calibrate syringes in three batch incubation at 39°C. Into 200 mg sample (*n = 8*) in the syringe was introduced 30 ml inoculums containing cheese cloth strained rumen liquor and buffer ($NaHCO_3$ + $3Na_2 HPO_4$ + KCL + NaCL + $MgSO_4$. $7H_2O$ + $CaCl_2$. $2H_2O$) (1:4, v/v) under continuous flushing with CO_2. The gas production was measured at 3, 6, 9, 12, 15, 28, 21, 24, 30, 36, 42 and 48h.

***In vitro dry Matter Digestibility (IVDMD)*:** After 48h digestion, the sample were transferred into test tubes and centrifuged for 1h in order to obtain the residues which was then filtered using whatman No 4 filter paper by gravity and the residues placed in for drying at 65°C for 24h. The dry residues were weighed and digestibility calculated using the equation thus:

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

***Methane Production*:** In order to estimate methane production by the substrate and immediately after evaluation from the incubator, 4 ml of NaOH (10 M) was introduced using 5 ml capacity syringe as reported by Fievez *et al.* (2005). The 120 ml capacity syringe; clip was then opened, while the NaOH was gradually released. The content was agitated, while the plunger began to shift position to occupy the vacuum created by the absorption of CO_2. The volume of methane was read on the calibration.

***Statistical Analysis*:** Data obtained were subjected to analysis of variance (ANOVA) using SAS (1988). Where significant differences occurred, the means were separated using Duncan multiple range test (Duncan, 1955)

Results and Discussion

Results

***Chemical Composition of Browse Forages*:** The chemical composition of browse forages is presented in Table 1. Dry matter, crude protein and fiber ranged from 94.00 in *Sterculia setigera* to 96.00% DM (*Ziziphus mauritiana*), 13.23 (*Olea hochstteteri*) to 18.31% DM in *Balanites aegytiaca*. *Olea hochstteteri* had the highest level of ether extract, while, *Balanites aegytiaca* and *Ficus sycomorus* were higher in ash content. The NFE was significantly higher (14.39% DM) in *Balanites aegytiaca* than the other browse forages, while, the fiber fraction (NDF and ADF) was significantly higher (54.80 and 33.40g/100g DM) in *Ficus sycomorus.* The ADL was significantly highest (66.91g/100g DM) in *Ziziphus mauritiana* and lowest (9.70) in *Adansonia digitata.* The total condensed tannins and phenolic were observed to be higher (0.41) in *Ziziphus spinachisti* and 0.81mg/g DM in *Ficus sycomorus.*

***IVDMD and its Relationship to Chemical Composition*:** Figures 1a, 1b, 1c, 1d, 1e, and 1f shows *in vitro* DMD and its relationship to various indicators of nutritive value. The relationship between CP and IVDMD was positive and significant ($R^2 = 0.71$, *n*=8), indicating that an increase in CP of the browse leads to an improvement in IVDMD. There were significant negative correlation between IVDMD and cell wall content (NDF, ADF and ADL) (Figs. 1b, 1c, and 1d,). Figs. 1e and 1f shows the relationship between IVDMD and TCT ($r = 0.98$, $n = 8$); CP and phenolic ($r = 0.84$, $n = 8$) was highly significant ($p<0.001$) but negatively correlated. Methane production (ml/200 mg DM) was highest in *Ziziphus spinachisti* (21 ml/200 mg DM) and *Ziziphus mauritiana* (4 ml/200 mg DM) being the least (Fig. 2).

Discussion

The crude protein (CP) contents of the browses studied had a similar range as those from West Africa (Rittner and Reed, 1992). All the browse used in the current study had a CP content above 13% DM. the results of the current study, and others (Rittner and Reed, 1992; Makkar and Becker, 1998) indicated that most tropical browse species are high in CP and can be used to supplement poor quality roughages; it increase productivity of ruminant livestock tropical regions. The *in vitro* DMD of leaves of all the browses under study were generally higher, though, there were significant differences ($p<0.05$) among the browse forages, the higher values may be due to low tannin content of the leaves as shown in Table 1. This result is similar to the findings of Njidda and Ikhimioya (2010). There was a positive correlation between *in vitro* DMD and TCT (r = 0.45, n= 8). The result is consistent with findings of Frutos *et al.* (2002) and Seresinhe and Iben (2003). The beneficial effect of tannins when forages containing low levels of tannins (Barry *et al*., 1986) 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. In addition, a higher flow of microbial protein to the intestine as a result of higher efficiency of microbial protein synthesis has been observed by Getachew *et al.* (2000). However, higher concentration of tannins in the diet is associated with reproduction in organic matter digestibility (Silanikove *et al*., 1997; Waghorn and Shelton, 1997). The tannin values in browses could be even higher than the values obtained in this study, since a considerable amount of tannins are bound to either fiber and/or proteins and remain unextracted (Gambel *et al*., 1996; Jackson *et al*., 1996).

Crude protein was positively correlated to IVDMD (r = 0.34, n = *8*) and phenolic (r = 0.84, n = 8). CP in the present study is in the level function considering the ranges of IVDMD (51.33 to 78.20% DM). A positive correlation between IVDMD and CP indicate that as the crude protein increase, there was an improvement in IVDMD. The inverse relationship between CP and phenolic compounds (Fig. 1f) indicated that considerable attention should be given in germplasm evaluation programmes to avoid selection against materials of high CP content. Further studies are required to understand the physiological mechanisms of plants that lead to the inverse relationship between contents of CP and phenolic compounds and hence to make decisions in plant selection and screening programmes.

The negative correlation between IVDMD and cell wall content observed in this study is consistent with the findings of Seresinhe and Iben (2003) and Ammar *et al.*(2004), indicates that the cell wall in the present group of samples were relatively poor predictors of IVDMD. Other studies; Madibela and Modiakgotla (2004) and Njidda and Nasiru (2010) reported that ADF has a negative effective on energy content of forages, and this was consistent with a high generating correlation observed between ADF and IVDMD. Irrespective of the maturity stage, leaves were always more digestible than stems, in agreement with those obtained by Lambert *et al.* (1989). It is well accepted that forage degradation in the rumen is mainly affected by the cell wall content and its signification, as lignin is indigestible fraction and acts as a barrier limiting the access of microbial enzymes to the structural polysaccharides of the cell wall. NDF, ADF and lignin were significant and negatively correlated with *in vitro* digestibility as reported by Van Soest (1994). It is well established that a low content of poorly digestible cell wall components (ADF and ADL) and a high CP content are indicators of a good forage quality (Van Soest, 1994). Therefore, at the light of our results, leaves have a higher nutritive value than stems. A negative correlation between secondary compounds and *in vitro* gas production and DM degradability of some browse tree species has been reported (Salem, 2005; Salem *et al*., 2007). In some perennial grasses (Casler and Jung, 2006) similar impacts of proanthocyanidins in some browse legumes were reported. Our results are similar to Rubanza *et al.* (2003) who reported a negative relationship between chemical composition and phenolic compounds, with in vitro degradability of legumes of *in vitro* incubation.

Methane (ml/200 mg DM) production (Figure 2) ranged from 4 to 21 among the browse forages, the least and the highest from *Ziziphus mauritiana* and *Ziziphus spinachisti*. 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 ranging from 0.02 to 0.12 of gross energy intake varying with the type of diet fed (Benchaar, *et al*., 2001), and on the other hand, when accumulates in rumen, it results in boat (Ellis, 2008). A reduction in methane production is expected when the residence time of feed in the rumen is reduced since ruminal digestion decreases and methanogenic bacteria are less able to decrease and methanogenic bacteria are less able to compete in such conditions (Moss *et al*., 2000). Methane production is also an integrated part of carbohydrate metabolism (Boadi *et al*., 2004). All the browse forages showed characteristics of low methane production which yielded less than 10 ml/200 mg DM methane of microbial fermentation of the browse forages. These plants in monoculture as they also inhibited overall fermentation when constituting the sole microbial substrate, but they may still be suitable as part of mixed diet and may be particularly useful for identifying specific plants compounds responsible for reducing methane production.

Conclusion

The chemical composition of these species should not be only to access their relative importance as ruminant feeds and other parameters; in particular, palatability need to be considered as high protein forages that can be used as supplements for low quality roughage's. All browse species could have a satisfactory energy value, considering their high DM digestibility.

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

Browse species	DM	CP	EE	Ash	NDF	ADF	ADL	TCT	PHE	IVDMD
O. hochstteteri	94.8^{b}	13.23^{d}	6.00^{a}	14.00^{c}	40.52^{b}	30.42^{c}	13.12^{b}	0.12^{g}	0.24^{f}	45.00^{g}
Z. Mauritania	96.0^{a}	15.86^{be}	2.00^{c}	12.00^{d}	38.67^{be}	16.55^{f}	66.91^{a}	0.21^{d}	0.54^{d}	55.34^{f}
Z .spinachisti	95.2^{a}	16.04^{b}	3.00^{b}	15.00^{b}	39.59^{b}	17.54	67.17^{a}	0.41^{a}	0.49^{e}	62.00^{d}
P. erinceus	95.0^{a}	17.96^{a}	2.00^{c}	11.00^{a}	36.40^{d}	26.30^{d}	13.80^{b}	0.23^{c}	0.61^{c}	70.60^{b}
S. setigera	94.0^{ab}	15.77^{be}	2.00^{c}	15.00^{b}	34.40^{a}	32.40^{d}	12.60^{b}	0.34^{b}	0.48^{e}	58.00^{e}
B. aegytiaca	95.2^{ab}	18.31^{a}	2.00^{c}	18.00^{a}	36.430^{d}	12.60^{b}	13.80^{b}	0.23^{c}	0.68^{b}	78.20^{a}
F. sycomorus	95.6^{a}	14.90^{c}	3.00^{b}	18.00^{a}	54.80^{a}	13.80^{b}	09.70^{c}	0.17^{e}	0.81^{a}	68.00^{c}
A. digitata	95.6^{a}	16.12^{b}	3.00^{b}	14.00^{c}	38.50^{be}	9.7^{c}	26.15^{c}	0.13^{f}	0.50^{d}	63.00^{d}
MEANS	**95.05**	**16.02**	**2.88**	**14.00**	**39.91**	**26.15**	**11.2**	**0.23**	**0.54**	**61.87**
SEM	**0.09**	**0.39**	**0.42**	**0.73**	**0.74**	**0.90**	**2.89**	**0.02**	**0.05**	**0.03**

a, b, c, means in the same column with different superscript differ significantly (p<0.05), SEM =Standard error means; * = significant

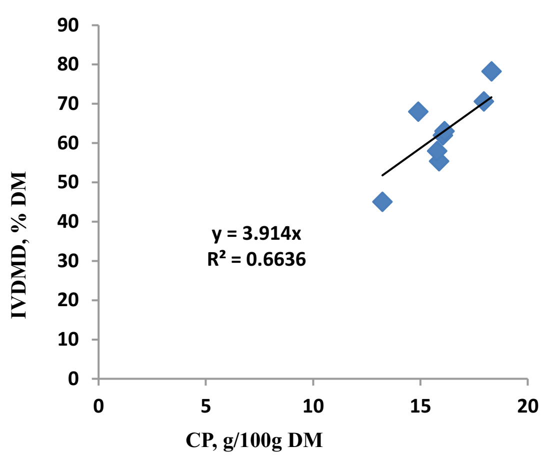

Fig. 1a: Relationship between IVDMD and CP of semi-arid browses

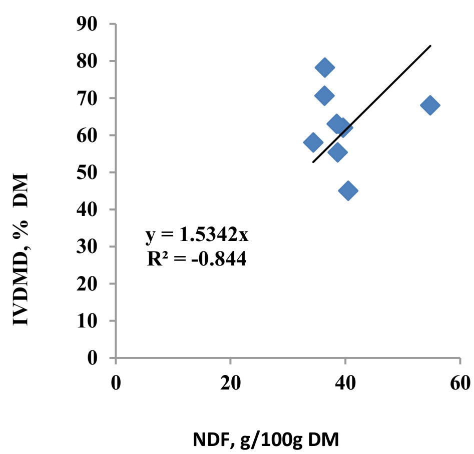

Fig. 1b: Relationship between IVDMD and NDF of semi-arid browses

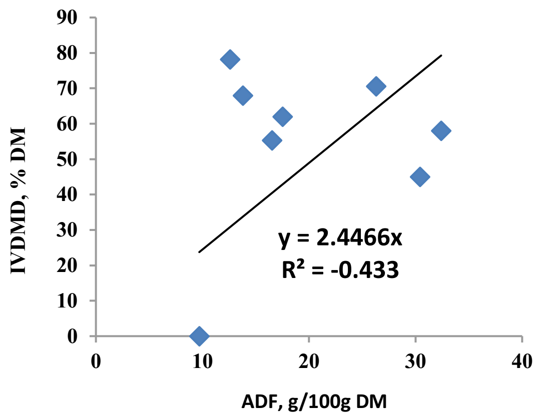

Figure 1c: Relationship between IVDMD and ADF of semi-arid browses

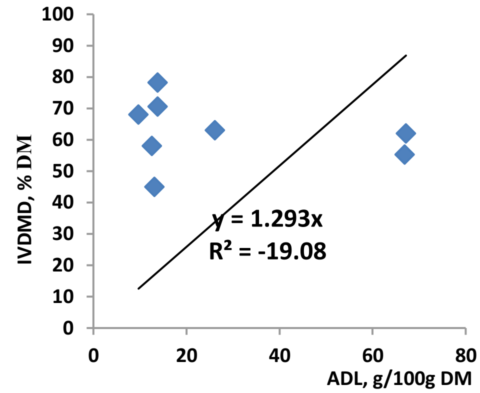

Figure 1d: Relationship between IVDMD and ADL of semi-arid browses

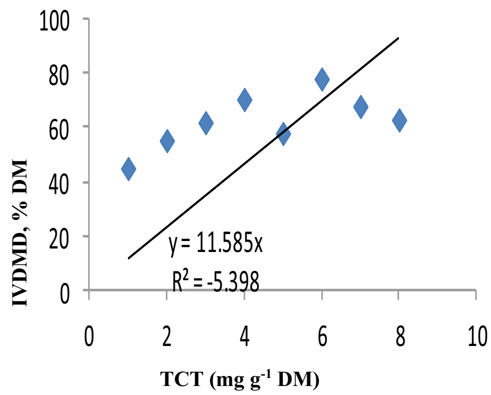

Figure 1e: Relationship between IVDMD and TCT of semi-arid browses

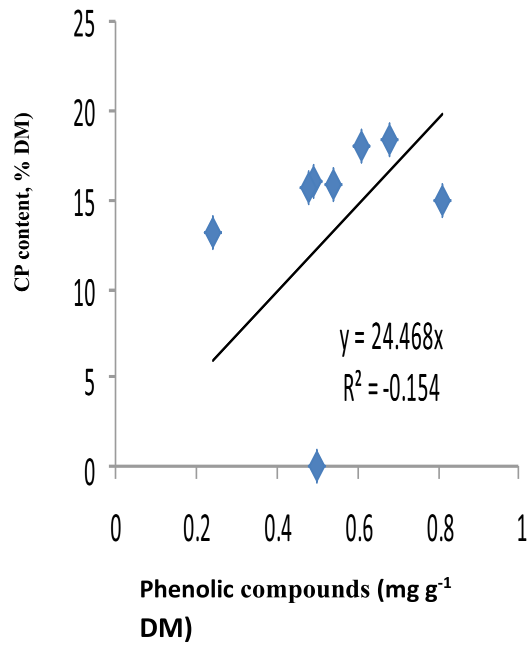

Figure 1f: Relationship between CP and Phenolics of semi-arid browses

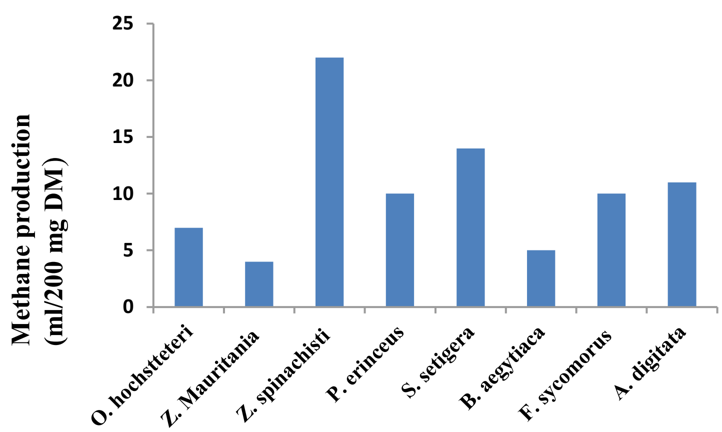

Figure 2: Methane production of semi-arid browses