American-Eurasian J. Agric. & Environ. Sci., 9 (2): 169-175, 2010
ISSN 1818-6769

Correlation Between Chemical Composition and *In Vitro* Dry Matter Digestibility of Leaves of Semi-Arid Browses of North-eastern Nigeria

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Abstract: The potential nutritive values of eight browse forages namely: *Olea hochstteteri, Ziziphus mauritiana, Zhiziphus spinzchristi, Pterocarpus erinceus, Sterculia setigera, Balanites aegyptiaca, Ficus sycomorus and Adansonia digitata* of North eastern Nigeria were evaluated by chemical composition and *in vitro* dry matter digestibility. The samples were collected and analysed 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 fibre 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 phenolics. An *in vitro* dry matter digestibility (IVDMD, %) study was conducted using rumen liquor as source of inoculum. 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 was highest (78.20%) in *Balanites aegyptiaca* and had the highest CP content while *Olea hochstteteri* 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. There is a positive correlation between CP and phenolics. The result for methane production shows that *Ziziphus spinachisti* had the highest methane value (24ml/200 mg DM) while *Ziziphus mauritiana* had the lowest methane production (4ml/200mg DM). Based on chemical composition and *in vitro* dry matter digestibility, the browse species forages have high potential nutritive value especially as protein supplements to poor quality forages for ruminant animals in the tropics and in terms of rumen and whole tract digestibility.

Key words: *In vitro* · Digestibility · Semi-arid · Browse · Tannin

INTRODUCTION

In semi-arid regions with a severe dry season, shrubby vegetation represents an integral component of the silvo-pastoral systems and sometimes their grazeable material is considered important for the nutrition of range animals [1]. However, quiet often these feed 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 [2, 3].

The *in vitro* gas production technique as modified by Menke and Steingass [4] 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 [5], agro industrial by-products [6], compound feeds [7] and various types of tropical feeds [8]. 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 [4]. The main objective of the present study was to investigate changes in chemical composition and *in vitro* digestibility of leaves of semi-arid browses.

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MATERIALS AND METHODS

Forage Samples: Eight indigenous browse samples (leaves) commonly consumed by ruminants animals were used in this study. The species were: *Olea hochstteteri, Ziziphus mauritiana, Zhiziphus spinzchristi, Pterocarpus erinceus, Sterculia setigera, 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 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. The browse forages were harvested from at least 10 trees per each species selected at random in four locations with the study area at the end of the season. The harvested sample were then pooled for each individual tree species and then oven dried at 105°C for 24h to 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 [9]. 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.*[10]. Total condensed tannin was [11].

***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.* [12] using 120 ml calibrated 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 ($NaHCO_3$ + 3 Na_2HPO_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, 18, 21, 24, 30, 36, 42 and 48h

***In Vitro* Dry Matter Digestibility (IVDMD):** After 48h digestion, the samples were transferred into test tubes and centrifuge 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 as follows:

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

Methane Production: In order to estimate methane production by the substrate and immediately after evacuation from the incubator, 4 ml of NaOH (10 M) was introduced using 5 ml capacity syringe as reported by Fievez *et al.* [12]. The content was inserted into the silicon tube, which was fastened to the 120 ml capacity syringe. The 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. Where significant differences occurred, the means were separated using Duncan multiple range F-test of the SAS [13] options.

RESULTS

Chemical Composition of Browse Forages: The chemical composition of the browse forages is presented in Table 1. Dry matter, crude protein and crude fibre ranged from 94.00 in (*Sterculia setigera*) to 96.00% DM (*Ziziphus mauritiana*), 13.23 (*Olea hochstteteri*) to 18.31% DM) in (*Balanites aegytiaca*); 14.50 in (*Balanites aegytiaca*) to 37.00 in (*Olea hochstteteri*), respectively. *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 (41.39% DM) in *Balanites aegyptiaca* than the other browse forages while the fibre fractions (NDF and ADF) was significantly higher (54.80 and 33.40g/100g DM) in *Ficus sycomorus*. The ADL was significantly higher (66.91g/100g DM) in *Ziziphus mauritiana* and lowest (9.70) in *Adansonia digitata*. The total condensed tannins and phenolics were observed to be higher 0.41 in *Ziziphus spinachisti* and 0.81mg/g DM in *Ficus sycomorus*.

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
Olea hochstteteri	94.8^{b}	13.23^{d}	6.00^{a}	14.00^{e}	40.52^{b}	30.42^{c}	13.12^{b}	0.12^{g}	0.24^{f}	45.00^{g}
Ziziphus mauritania	96.0^{a}	15.86^{bc}	2.00^{e}	12.00^{d}	38.67^{bc}	16.55^{f}	66.91^{a}	0.21^{d}	0.52^{d}	55.34^{f}
Ziziphus 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}
Pterocarpus erinceus	95.0^{a}	17.96^{a}	2.00^{e}	11.00^{a}	36.40^{d}	26.30^{d}	13.80^{b}	0.23^{c}	0.61^{c}	70.60^{b}
Sterculia setigera	94.0^{ab}	15.77^{bc}	2.00^{e}	15.00^{b}	34.40^{e}	32.10^{b}	12.60^{b}	0.34^{b}	0.48^{e}	58.00^{e}
Balanites aegyptiaca	94.2^{ab}	18.31^{a}	2.00^{e}	18.00^{a}	36.430^{d}	25.74^{de}	13.75^{b}	0.23^{c}	0.68^{b}	78.20^{a}
Ficus sycomorus	95.6^{a}	14.90^{c}	3.00^{b}	18.00^{a}	54.80^{a}	33.40^{a}	12.60^{b}	0.17^{e}	0.81^{a}	68.00^{c}
Adansonia digitata	95.6^{a}	16.12^{b}	3.00^{b}	14.00^{e}	38.50^{bc}	27.20^{d}	9.70^{e}	0.13^{f}	0.50^{d}	63.00^{d}
MEANS	95.05	16.02	2.88	14.63	39.91	26.15	26.19	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). DM = Dry matter; CP = Crude Protein; EE = Ether Extract; NDF = Neutral detergent fibre; ADF = Acid detergent fibre; ADL=Acid detergent lignin; TCT=Total Condensed Tannins; PHE:Phenolics; IVDMD=*In vitro* Dry Matter Digestibility.

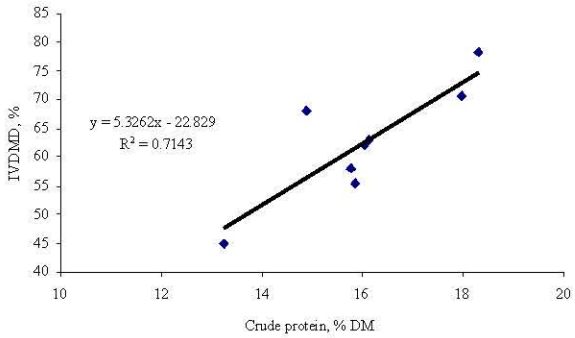

Fig. 1: Relationship between IVDMD and CP of semi-arid browses

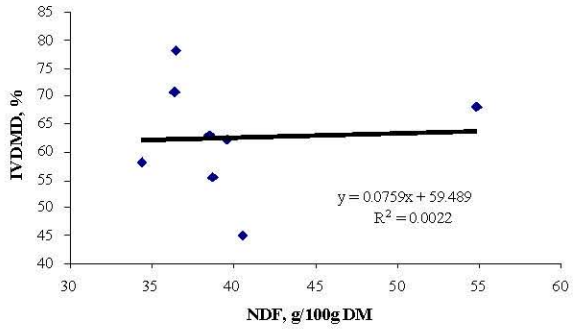

Fig. 2: Relationship between IVDMD and NDF of semi-arid browses

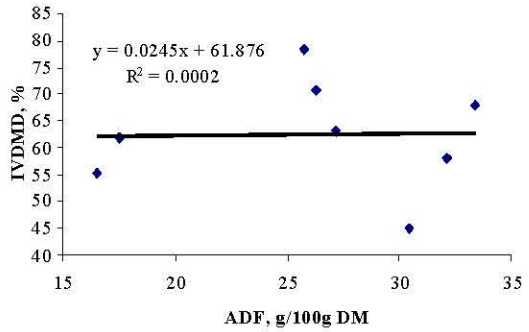

Fig. 3: Relationship between IVDMD and ADF of semi-arid browses

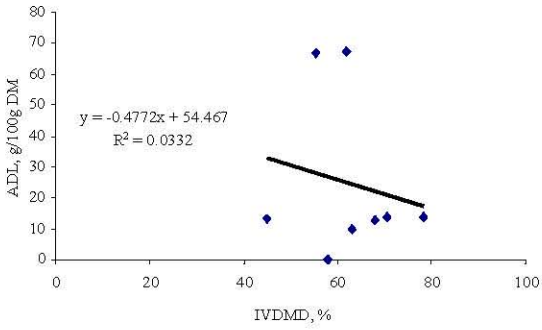

Fig. 4: Relationship between IVDMD and ADL of semi-arid browses

IVDMD and it Relationship to Chemical Composition: Figures 1, 2, 3 and 4 shows *in vitro* DMD and it's relationship to various indicators of nutritive value. The relationship between CP and IVDMD was positive (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) (Fig 2, 3 and 4), respectively.

The relationship between IVDMD and TCT (r=0.98, n=8); CP and Phenolic (r=0.84, n=8) (Fig 5 and 6) was highly significant (P<0.001).

Methane production (ml/200 mg DM) was highest in *Ziziphus spinachisti* and lowest in *Ziziphus mauritiana* with *Ziziphus spinachisti* having 21 ml/ 200 mg DM and *Ziziphus mauritiana* having 4 ml/200 mg DM.

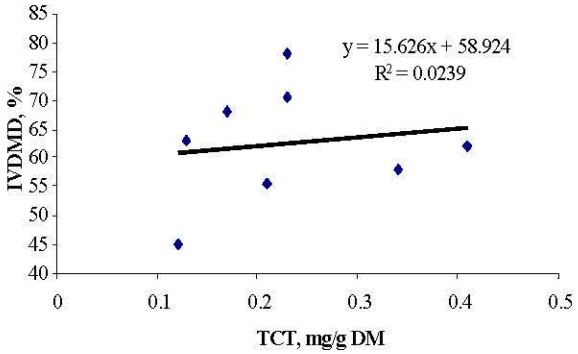

Fig. 5: Relationship between IVDMD and TCT of semi-arid browses

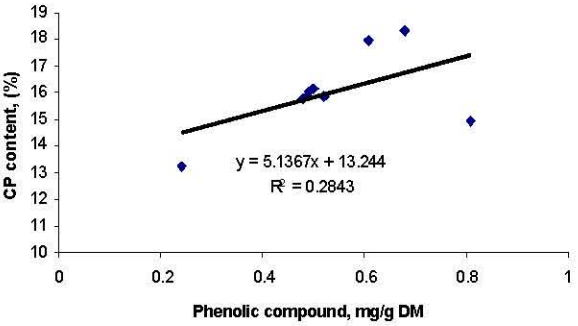

Fig. 6: Relationship between CP and Phenolic compounds of semi-arid browses

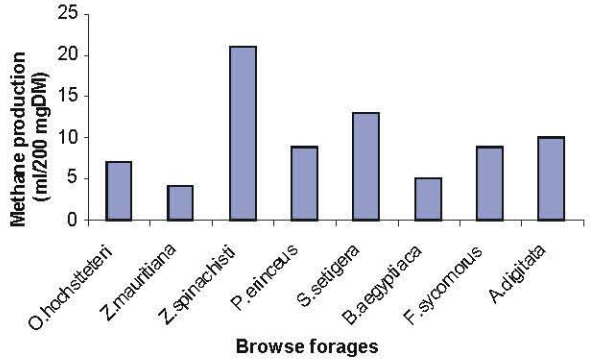

Fig. 7: Methane production of semi-arid browse forages of North-eastern Nigeria

DISCUSSION

The crude protein (CP) contents of the browses studied had a similar range as those from West Africa [14]. All the browses used in the current study had a CP content of above 13% DM. The results of the current study, those of Rittner and Reed [14] and Makkar and Becker [15] indicated that most tropical browse species are high in CP and can be used to supplement poor quality roughages to increase productivity of ruminant livestock in 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 [16].There was a positive correlation between *in vitro* DMD and TCT ($r = 0.99$, $n = 8$). The result is consistent with findings of Frutos *et al.*[17] and Seresinhe and Iben [18]. The beneficial effect of tannins when forages containing low levels of tannins [19] 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.* [20]. However, higher concentration of tannins in the diet is associated with reduction in organic matter digestibility [21, 22]. 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 fibre and/or proteins and remain unextracted [23 , 24] .

Crude protein was positively correlated to IVDMD ($r = 0.53$, $n = 8$) and phenolic ($r = 0.85$, $n = 8$). CP in the present study is in the level permissible fore optimal feed intake and rumen 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. 6) 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 [18] and Ammar *et al.* [25], indicate that the cell wall indices in the present group of samples were relatively poor predictors of IVDMD. Other studies , Madibela and Modiakgotla [26] and Njidda and Nasiru [27] reported that ADF has a negative effective on energy content of forages and this was consistent with a highly negative 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.* [2]. It is well accepted that forage degradation in the rumen is mainly affected by the cell wall content and its lignification, 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 [28]. 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 [28]. 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 [29,30]. Similar negative relationships occurred between NDF, lignin(sa) and phenolics with *in vitro* digestibility in some perennial grasses [31] found similar impacts of proanthocyanidins in some browse legumes. Our results are similar to Rubanza *et al.* [32] 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 being from *Ziziphus mauritiana* and *Ziziphus spinachristi*, 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 through methane production can range from 0.02 to 0.12 of gross energy intake varying with the type of diet fed [33] and on the other hand, when accumulates in the rumen, it results in bloat [34]. 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 compete in such conditions [35]. Methane production is also an integrated part of carbohydrate metabolism [36]. 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 will not be appropriate as the grazing 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 plant compounds responsible for reducing methane production.

CONCLUSION

The chemical composition of these species should not be only point to assess their relative importance as ruminant feeds and other parameters, in particular palatability, need to be considered. Anyway, the foliage of both legume shrubs showed high CP contents, sufficient 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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