

## ORIGINAL ARTICLE

## Comparative assessment on chemical compositions and feeding values of leaves of *Moringa stenopetala* and *Moringa oleifera* using in vitro gas production method

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### ABSTRACT

Evaluating the nutritional value of indigenous shrubs, trees and browse plants is important in Ethiopian situation where availability and quality of forages severely limited during long and dry seasons. A comparative assessment was thus conducted to investigate the nutrient compositions and feeding values of *M. stenopetala* and *M. oleifera* leaves. Feed samples were analyzed for proximate nutrients, minerals and amino acid profiles using official methods. In addition, the metabolizable energy (ME), organic matter digestibility (OMD) and short chain fatty acids (SCFA) were predicated using the Hohenheim *in vitro* gas test method. *M. stenopetala* had 26.6% crude protein (CP), 3.36% fat, 17.9 KJ/kg DM gross energy, 45% nitrogen free extract (NFE), and 38.4% non fiber carbohydrate (NFC). In *M. oleifera*, the contents of CP, fat, NFE, NFC and gross energy were 28.9%, 6.73%, 45%, 38.4% and 17.9 MJ/kg DM, respectively. *M. stenopetala* leaves contained significantly higher crude fiber, acid detergent fiber (ADF) and cellulose than those of *M. oleifera*. However, the acid detergent lignin (ADL) and hemicelluloses contents of *M. oleifera* were significantly higher than those of *M. stenopetala*. The contents of calcium, phosphorous, magnesium, potassium and sodium in *M. stenopetala* were 2.47%, 0.57%, 0.76%, 2.45% and 0.11%, respectively. The values of the corresponding minerals in *M. oleifera* were 2.62%, 0.43%, 0.56%, 2.0% and 0.03%. The concentrations of essential amino acids were significantly higher in *M. oleifera* than those of *M. stenopetala* and were generally comparable with the contents of soybean meal. The highest *in vitro* gas production of 47.9 ml was recorded for *M. stenopetala*, being significantly higher than for *M. oleifera* (40.6 ml). Similarly, values of ME (9.83 MJ/kg DM), OMD (76.4%) and SCFA (101 mmol) in *M. stenopetala* were significantly higher than those of *M. oleifera*. Although not significant, organic matter, CP, fat, ADF and cellulose contents were positively correlated with *in vitro* gas production while DM, NFE, neutral detergent fiber, ADL and hemicelluloses contents were negatively correlated with gas production. The chemical compositions showed the potential of leaves of both *Moringa* species to be used as a protein supplement in ruminant and non-ruminant feeding during the dry season. Moreover, the enhanced values of ME, OMD and SCFA in leaf suggest its ability to meet the energy requirements of tropical livestock.

**Keywords:** *Moringa* leaves, chemical compositions, *in vitro* gas production

## INTRODUCTION

Inadequate nutrition is one of the factors that generally affect livestock productivity in the tropical countries. Total weight gained by ruminants during the rainy season is lost in the dry season due to feed scarcity. The use of tree leaves as fodder for ruminant livestock has been increasingly important in many parts of the tropics particularly during the dry period. Leaves from browse and fodder trees form major parts of livestock feed in the tropical countries (Woods *et al.*, 1994) and play a major role in improving dietary protein (Kaitho *et al.*, 1998). Tree leaves can be rich in crude protein (CP), minerals and digestible nutrients when compared to grasses. *Moringa stenopetala* (*M. stenopetala*) known as African Moringa has a wide range of adaptation from the arid to humid climates and can be grown in a various land use patterns. It grows in the lowlands of West of the Great Rift Valley Lakes from arid to semi-humid areas altitudinal ranging from 390 m to about 2200 m a.s.l. It is a strategic multi-purpose tree plant in being a unique food tree in drought prone areas and has recently been distributed to other regions of Ethiopia, beyond its place of origin. Leaves are used for human consumption and animal feed (Aberra *et al.*, 2011). A study conducted by Aberra *et al.* (2009) indicated that the leaves of *M. stenopetala* are rich in crude protein (28.2%) and contain reasonable amounts of essential amino acids.

The *Moringa oleifera* (*M. oleifera*) is native to the sub-Himalayan tracts of north-west India, Pakistan, Bangladesh and Afghanistan (Makkar and Becker, 1997). This multipurpose tree has been introduced to Ethiopia over the last few years and is grown on nursery sites parallel to *M. stenopetala* in southern parts of the country. Both *M. stenopetala* and *M. oleifera* trees are the most

commonly cultivated Moringa species in the tropics and subtropics which have the potential as alternative animal feed resources during dry periods of the tropics. However, the suitability and digestibility of leaves of both Moringa species in feeding ruminants and non-ruminants under Ethiopian conditions is hardly documented. The objectives of this study were thus to investigate nutrient compositions and feeding values of leaves from *M. stenopetala* and *M. oleifera* using *in vitro* gas production techniques.

## MATERIALS AND METHODS

### Sample collection

Leaf samples of *M. stenopetala* and *M. oleifera* were collected at Chano Mille nursery site of Southern Agricultural Research located at Arbaminch district of Gamogofa administrative zone, Ethiopia. The altitude is 1100 m a.s.l with annual rainfall of 750-900 mm. The samples were collected from both Moringa trees aged 6 years old in December 2009. Each sample was collected randomly from six different Moringa trees. After removing the petioles, leaf samples were dried at 65 °C for 48 h and ground to pass 1 mm sieve size. Ground feed samples were labeled and kept in air-tight plastic containers until analysis.

### Chemical analysis

Analyses of proximate nutrients and fiber fractions were performed as outlined by Verband Deutscher Landwirtschaftlicher Untersuchungs- und Forschungsanstalten (VDLUF, 2006). The samples were analyzed for DM (method 3.1), ash (method 8.1), crude protein (CP, method 4.1.1, N multiplied by 6.25), petroleum ether extract (EE, method 5.1.1), and crude fiber (CF, method 6.1.1, all VDLUF, 2006). Neutral detergent fiber (NDF) assayed with a heat stable amylase and

acid detergent fiber (ADF) were analyzed according to VDLUFA (2006; methods 6.5.1 and 6.5.2) and were expressed inclusive of residual ash. Cellulose and hemicellulose were computed as ADF minus acid detergent lignin (ADL) and NDF minus ADF, respectively. Non-fiber carbohydrate (NFC) content was calculated as 100-(NDF + CP + crude fat + ash) according to NRC (2001). Nitrogen free extract (NFE) was computed by difference of organic matter and the sum of CF, EE and CP. Amino acid contents were measured by ion-change chromatography using an amino acid analyzer L8900 (VWR/Hitachi). After a performic acid oxidation step protein was hydrolyzed with 6 M HCl for 24 h at 113 °C. The amino acids were separated on a cation exchange resin and detected after post column derivatization with ninhydrin reagent using VIS-detection at 570 nm (440 nm for proline). For mineral analysis, samples were incinerated at 550 °C, and the remaining ash was treated with 6 mol per L HCl. Minerals were determined from filtered ash solutions using an Inductively Coupled Plasma spectrometer (ICP-OES) (Rodehutsord and Dieckmann, 2005). All chemical analyses were conducted in duplicate on each individual sample.

### ***In vitro* studies**

Gas production was determined according to the procedure of VDLUFA official method (VDLUFA 2006, method No. 25.1), Menke, and Steingass (1988). About 200 mg of feed sample was weighed in two replicates and transferred into 100 ml calibrated glass syringes, fitted with Vaseline lubricated pistons. To prepare the inoculum, rumen fluid was collected before the morning feeding from two rumen-cannulated, non-pregnant, non-lactating Holstein Friesian cows, fed on medium quality diet and a concentrate. Details

about feeding are described by Steingass and Menke (1986). The rumen fluid was placed directly into pre-warmed thermo flasks and taken immediately to the laboratory. It was then filtered through two layers of cheesecloth and diluted with buffered mineral solution, which was maintained in a water bath at 39 °C under continuous flushing with CO<sub>2</sub>. A total of 30 ml incubation medium consisting of 10 ml rumen fluid, 5 ml of bicarbonate buffer, 5 ml of macro-mineral solution and 10 ml of distilled water was transferred into a pre-warmed glass syringes containing the samples (200 mg) and blank syringes.

After filling the glass syringes with incubation medium, they were immediately placed in a temperature-controlled incubator preset at 39°C. Incubation was completed in duplicate within each run and runs were replicated yielding four observations per sample. Three blanks containing 30 ml of medium as well as triplicate samples of reference hay and concentrate feed of known gas production were included. The gas volume was recorded at 2, 4, 6, 8, 10, 12, 14, 18, 24, 30, 36 and 48 hours of incubation according to the time described by Blümmel and Becker (1997). The volume of gas produced (means of two runs) was plotted against the incubation time and the fermentation kinetics and estimated parameters were expressed according to the equation of Beuvinck and Kogut (1993) described in detail by Boguhn *et al.* (2008). The gas produced by test substrates was corrected for that by the blank syringes (containing no substrate), and 24 h gas production was corrected by the standards for the estimation of OMD, ME and SCFA. The Metabolizable energy (ME, MJ/kg DM) and organic matter digestibility (OMD, %) were computed as established by Menke *et al.* (1979) and Menke and Steingass (1988) and short chain fatty acids (SCFA,

mmol) were calculated as reported by Blümmel *et al.* (1999) with the following equations:

$$\text{ME (MJ/kg DM)} = 2.20 + (0.136 \cdot \text{Gv}) + (0.0057 \cdot \text{CP}) + (0.00029 \cdot \text{EE});$$

$$\text{OMD (\%)} = 14.88 + (0.889 \cdot \text{Gv}) + (0.45 \cdot \text{CP}) + (0.651 \cdot \text{XA});$$

$$\text{SCFA} = 0.0239 \cdot \text{Gv} - 0.0601;$$

Where: Gv, CP, EE and XA are corrected 24 h gas volume (ml/200 mg DM), crude protein, ether extract and ash (g/kg DM) of the incubated samples, respectively.

### Statistical methods

Results on chemical compositions and calculated gas production parameters were subjected to one-way ANOVA analysis by using SAS GLM procedures (SAS, 2004) and differences of means were separated by Duncan multiple range test. Pearson correlation procedures were also performed with SAS software. Time series measurements of gas volumes from 2-48 hrs of *in vitro* incubation were used for the curve fitting to mathematically express gas production over incubation time by using the software GraphPad Prism 4.02 for Windows (GraphPad Software Inc. 2004, La Jolla, CA, USA).

## RESULTS AND DISCUSSION

### Chemical and mineral compositions

The proximate nutrients and fiber fractions of Moringa leaves are presented in Table 1. The contents of crude protein (CP) and fat in *M. oleifera* was significantly ( $p < 0.05$ ) higher than those of *M. stenopetala*. The average CP content in *M. oleifera* was comparable with that of Oduro *et al.* (2008) for the same Moringa specie. Sanchez *et al.* (2006) reported CP contents of 22.8% and 23.3 % for *M. oleifera* leaves, respectively, which are lower than those of the current study. Soliva *et al.* (2005) reported 32.1% of CP for *M. oleifera* leaves whereas Aberra *et al.* (2011)

found 30.6% CP for *M. stenopetala* leaves, which are higher than those of both Moringa species in the current study. The variations in CP contents of the reported values may be due to differences in agro-climatic conditions or to different ages of trees, and possibly due to different stages of maturity. Studies conducted by Yang *et al.* (2006) indicated that mature leaves contained more CP than young shoots. In general, the CP content of Moringa leaves is higher than those of most tropical forage legumes reported by Babayemi (2007).

The fat and ash contents reported by Gupta *et al.* (1989) and Makkar and Becker (1996, 1997) for *M. oleifera* leaves are in good agreement with the current results. However, the fat contents in *M. oleifera* leaves reported by Oduro *et al.* (2008) are lower than those obtained from the current study. The gross energy values (17.8 MJ/kg DM) reported for *M. oleifera* leaves by Aregheore (2002) agrees with those of *M. stenopetala* but lower than those of *M. oleifera*. The gross energy contents of 18.7 and 19.4 MJ/kg DM reported by Makkar and Becker (1996, 1997) for *M. oleifera* leaves are slightly higher than those of *M. stenopetala*. The *M. oleifera* leaves contained high acid detergent lignin (ADL) and hemicelluloses contents compared with *M. stenopetala*. However, the contents of fiber fractions (CF, NDF and ADF) and cellulose for *M. stenopetala* were significantly ( $p < 0.05$ ) higher than those of *M. oleifera*. The NDF values in *M. oleifera* are consistent with those of Makkar and Becker (1997), Aregheore (2002) and Sanchez *et al.* (2006). The ADF contents of *M. oleifera* leaves reported by Makkar and Becker (1996) are in agreement with the present findings. However, ADF and NDF contents in *M. oleifera* leaves in the current study are higher than those of Gupta *et al.* (1989).

**Table 1.** Comparative chemical compositions of leaves of *M. stenopetala* and *M. oleifera* (in % on DM basis)

Nutrients	<i>M. stenopetala</i>	<i>M. oleifera</i>	Overall mean	Pooled S.E.M	P
Ash	14.8 <sup>a</sup>	13.2 <sup>b</sup>	14.0	0.269	<0.0001
Crude protein	26.6 <sup>b</sup>	28.9 <sup>a</sup>	27.7	0.380	<0.0001
Crude fat	3.36 <sup>b</sup>	6.73 <sup>a</sup>	5.05	0.392	<0.0001
Crude fiber	10.2 <sup>a</sup>	8.51 <sup>b</sup>	9.37	0.600	0.0004
Nitrogen free extract	45.0 <sup>a</sup>	42.6 <sup>b</sup>	43.8	0.701	<0.0001
Neutral detergent fiber	16.8 <sup>a</sup>	16.7 <sup>a</sup>	16.8	0.708	ns
Acid detergent fiber	14.2 <sup>a</sup>	12.1 <sup>b</sup>	13.2	0.861	<0.0001
Acid detergent lignin	5.52 <sup>b</sup>	6.49 <sup>a</sup>	6.00	0.516	0.0154
GE (MJ/kg DM)*	17.9 <sup>a</sup>	16.8 <sup>b</sup>	17.4	0.137	<0.0001
Cellulose	8.73 <sup>a</sup>	5.59 <sup>b</sup>	7.16	1.072	<0.0001
Hemicelluloses	2.55 <sup>b</sup>	4.66 <sup>a</sup>	3.61	0.374	0.0004
NFC	38.4 <sup>a</sup>	34.4 <sup>b</sup>	3.64	0.836	<0.0001

<sup>a,b</sup>Means between Moringa species having different letters are significantly ( $p < 0.05$ ) different \*computed from  $0.0239 \times \text{CP (g)} + 0.0398 \times \text{EE (g)} + 0.0201 \times \text{CF (g)} + 0.0175 \times \text{NFE (g)}$   
GE= gross energy; ns= not significant

As presented in Table 2, leaves of *M. stenopetala* contained significantly ( $p < 0.05$ ) higher phosphorous (P), magnesium (Mg), potassium (K) and sodium (Na) than those of *M. oleifera*. However, the contents of calcium (Ca) and ratio of Ca to P were significantly ( $p < 0.05$ ) higher in *M. stenopetala* leaves than those found in *M. oleifera*. The contents of P, K, Mg and Na in *M. stenopetala* are generally higher than those reported by Aberra *et al.* (2009) for the same species cultivated in the mid-altitude of Hawassa district. However, the Ca content reported by the same

authors for *M. stenopetala* leaves is comparable with the present findings. In *M. oleifera* leaves, Oduro *et al.* (2008) reported 2.01% of Ca, which is slightly lower than obtained from the present study. The contents of P and especially of Ca and Mg in Moringa leaves are higher than those of native pasture reported by Gizachew *et al.* (2002). Accordingly, the nutritive value of both Moringa leaves in terms of macro minerals can play a considerable role for supplementing ruminant and non-ruminant feeds in the tropics.

**Table 2.** Comparative compositions of common major minerals in leaves of *M. stenopetala* and *M. oleifera* (in % on DM basis)

Nutrients	<i>M. stenopetala</i>	<i>M. oleifera</i>	Overall mean	Pooled S.E.M	P
Calcium (Ca)	2.47 <sup>b</sup>	2.62 <sup>a</sup>	2.55	0.026	0.0493
Phosphorous (P)	0.57 <sup>a</sup>	0.43 <sup>b</sup>	0.50	0.019	<0.0001
Ca:P	0.43 <sup>b</sup>	0.61 <sup>a</sup>	0.52	0.018	<0.0001
Magnesium (Mg)	0.76 <sup>a</sup>	0.56 <sup>b</sup>	0.66	0.022	<0.0001
Potassium (K)	2.45 <sup>a</sup>	2.00 <sup>b</sup>	2.22	0.101	<0.0001
Sodium (Na)	0.11 <sup>a</sup>	0.03 <sup>b</sup>	0.07	0.078	<0.0001

<sup>a,b</sup>Means between Moringa species having different letters are significantly ( $p < 0.05$ ) different

### Concentrations of amino acids

The concentration of essential amino acids in leaves of *M. stenopetala* and *M. oleifera* has been presented in Table 3. Except for cysteine, the concentration of all essential amino acids in *M. oleifera* was significantly ( $p < 0.05$ ) higher than those of *M. stenopetala*. Most of the essential amino acids in *M. oleifera* are slightly higher than those reported by Makkar and Becker (1996) for leaves of the same Moringa specie. The

concentrations of leucine, lysine and threonine, reported by Booth and Wickens (1988) for *M. oleifera* leaves are comparable with the current findings. However, the contents of arginine, isoleucine, methionine, phenylalanine and valine reported by the same author were relatively lower than those of the present study. These variations might be possibly attributed to age of the tree, agro-climatic conditions (including altitude) and soil type.

**Table 3.** Concentrations of essential amino acids in leaves of *M. stenopetala* and *M. oleifera* (g/kg DM)

Amino acids	<i>M. stenopetala</i>	<i>M. oleifera</i>	Overall mean	Pooled S.E.M	P
Arginine	13.1 <sup>b</sup>	15.4 <sup>a</sup>	14.3	0.366	<0.0001
Cysteine	3.91 <sup>a</sup>	3.55 <sup>b</sup>	3.73	0.072	<0.0001
Isoleucine	9.41 <sup>b</sup>	10.9 <sup>a</sup>	10.1	0.317	0.0132
Leucine	18.6 <sup>b</sup>	21.4 <sup>a</sup>	20.0	0.598	<0.0001
Lysine	12.2 <sup>b</sup>	13.2 <sup>a</sup>	12.7	0.220	0.0004
Methionine	3.65 <sup>b</sup>	4.24 <sup>a</sup>	3.94	0.136	<0.0001
Phenylalanine	13.7 <sup>b</sup>	16.4 <sup>a</sup>	15.1	0.476	<0.0001
Threonine	11.4 <sup>b</sup>	13.0 <sup>a</sup>	12.2	0.376	<0.0001
Valine	12.0 <sup>b</sup>	14.0 <sup>a</sup>	13.0	0.458	0.0048

<sup>a,b</sup>Means between Moringa species having different letters are significantly ( $p < 0.05$ ) different

Recent studies conducted by Aberra *et al.* (2011) indicated that Moringa leaf meal was effectively used to replace soybean meal in the growth performance of growing Rhode Island Red chickens. Moreover, Moringa leaf extracts exhibited anti-microbial activity including inhibition of the growth of *Staphylococcus aureus* strains isolated from food and animal intestines. Moringa added to fodder can be thus used as a potential bioceutical agent to substitute for antibiotics in livestock production (Yang *et al.*, 2006).

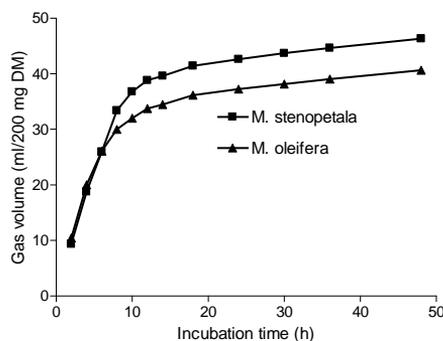
### *In vitro* gas production and calculated parameters

The pattern of *in vitro* gas production in leaves of *M. stenopetala* and *M. oleifera* is presented in Figure 1. Gas production is

basically the result of fermentation of carbohydrates to acetate, propionate and butyrate. Gas production parameters suggest differences in nutritional values that were generally closely related to chemical composition (Cerrillo and Juarez, 2004). Leaves of both Moringa species showed rapid gas production in the early stage of *in vitro* fermentation which indicates a higher content of rapidly fermentable soluble components.

However, the *in vitro* gas volume from *M. oleifera* significantly decreased thereafter indicating it may have had more slowly fermentable carbohydrate contents than *M. stenopetala* in the later incubation periods. The high *in vitro* gas production observed in *M. stenopetala*

after the initial phase of incubation may suggest a higher extent of fermentation throughout the incubation periods.



**Figure 1.** Pattern of *in vitro* gas production in leaves of *M. stenopetala* and *M. oleifera* measured over 48 hours of feed samples incubation [plotted according to Beuvinck and Kogut (1993) described in detail by Boguhn *et al.* (2008)].

The *M. oleifera* leaves had the highest CP content and yet they produced lower gas volume than those of *M. stenopetala* (Figure 1). Normally, low gas production would indicate low degradability in the rumen, but feedstuffs high in CP produce less gas during fermentation, even if their extent of degradation is high. This is because protein fermentation produces ammonia, which influences the carbonate buffer equilibrium by neutralizing H<sup>+</sup> ions from volatile fatty acids without release of carbon dioxide (Cone and Van Gelder 1999). According to Makkar and Becker (1996) about 24% of the CP was soluble in phosphate buffer (pH 7, 0.05 M) for *M. oleifera* leaves. Apart from this, the low gas production from *M. oleifera* leaves could be attributed to the low content of NFC compared with those of *M. stenopetala*. Getachew *et al.* (2004) reported that NFC was positively correlated with gas production at 6 h, 24 h and 48 h of incubation (De Boever *et al.* 2005). Furthermore, the lower gas volume of *M. oleifera* leaves might be further explained by the presence of high fat (6.73% vs. 3.36%), which contribute to

negligible gas production as reported by Aberra *et al.* (2009).

As presented in Table 4, *M. stenopetala* leaves contained significantly ( $p < 0.05$ ) higher calculated ME, OMD and SCFA values compared with those of *M. oleifera*. Similarly, values of estimated parameters for *M. stenopetala* leaves were significantly higher than those of *M. oleifera*. The ME and OMD predicted from the extent of fermentation in the *in vitro* incubation reported by Makkar and Becker (1996) for *M. oleifera* leaves was 9.5 MJ/kg DM and 74.1%, respectively, which agrees with those of the current findings for *M. oleifera* (9.3 MJ/kg DM and 72.0%). *In vitro* calculated ME and OMD values reported for *M. stenopetala* leaves by Aberra *et al.* (2009) are consistent with the present findings for *M. stenopetala*. Moreover, consistent with the present findings of both Moringa leaves, Kiran and Krishnamoorthy (2007) reported an average ME value of 10.2 MJ/kg DM for common protein supplements. In agreement with the current findings, Anele *et al.* (2009) reported ME values of 9.56 - 10.6 MJ/kg DM for leaves of tropical multi-purpose trees. The

calculated SCFA for both Moringa leaves was much higher than reported for different forage species by Babayemi (2007). Higher production of gas and

predominance of SCFA in leaves could probably describe an increased proportion of acetate and butyrate but a decrease in propionate production.

**Table 4.** Calculated metabolizable energy, organic matter digestibility, short chain fatty acids and estimated parameters in leaves of *M. stenopetala* and *M. oleifera*

Moringa species	Calculated parameters <sup>1)</sup>			Estimated parameters <sup>2)</sup>		
	ME (MJ/kg DM)	OMD (%)	SCFA (mmol)	b	$\mu$ r	$\mu$ s
<i>M. stenopetala</i>	9.83 <sup>a</sup>	76.4 <sup>a</sup>	101 <sup>a</sup>	47.9 <sup>a</sup>	2.85 <sup>a</sup>	0.197 <sup>a</sup>
<i>M. oleifera</i>	9.30 <sup>b</sup>	72.0 <sup>b</sup>	89.5 <sup>b</sup>	40.6 <sup>b</sup>	1.73 <sup>b</sup>	0.042 <sup>b</sup>
Overall mean	9.56	74.1	95.4	44.3	2.29	0.119
S.E.M	0.15	0.91	2.79	2.415	0.268	0.029

<sup>a,b</sup>Means between Moringa species having different letters are significantly ( $p < 0.05$ ) different

ME= metabolizable energy; OMD= organic matter digestibility; SCFA= short chain fatty acids; S.E.M= standard error of the mean

<sup>1)</sup> Parameters were calculated from 24 hours gas production data.

<sup>2)</sup> Estimated parameters obtained by fitting gas production data to the equation of Beuvink and Kogut (1993) [where b is the maximum value of gas production;  $\mu$ r (in ml/h) is the rapid gas production rate during early stages of fermentation;  $\mu$ s (in ml/h) is the slower gas production rate during later stages of fermentation].

#### Correlations of chemical compositions with *in vitro* gas productions

Results of correlation analysis of chemical compositions with *in vitro* gas production and estimated parameters are presented in Table 5. Gasmi-Boubaker *et al.* (2005) reported a positive correlation of CP with gas production at 24 h for tropical browse species which agrees with the findings of the current study. It should be noted that although not significant, CP content showed a trend of positive correlation with gas production at 24 h.

Contrary to the present findings, Getachew *et al.* (2004) observed a negative

correlation between the volume of gas produced and the CP content of the feed. In general, although not significant, the ADF and ADL contents were negatively correlated with gas production, which are consistent with those of Gasmi-Boubaker *et al.* (2005). De Boever *et al.* (2005) and Parissi *et al.* (2005) reported a negative association of gas production with NDF, ADF and ADL contents. These feed constituents are known to be less degradable than soluble carbohydrates and therefore reduce gas production.

**Table 5.** Correlation coefficients (r) of relationship of chemical composition and *in vitro* gas production and estimated parameters

Chemical compositions	Gv at various incubation times <sup>1)</sup>			Estimated parameters <sup>2)</sup>		
	Gv 8h	Gv 24h	Gv 48h	B	$\mu_r$	$\mu_s$
Dry matter	-0.186	-0.393	-0.234	0.439	0.164	0.160
Organic matter	0.029	0.282	0.143	0.522	-0.378	-0.342
Crude protein	0.105	0.343	0.204	0.453	-0.290	-0.254
Crude fat	0.602	0.529	0.377	-0.425	0.224	0.217
Crude fiber	0.069	0.029	0.122	0.927**	0.167	0.206
Nitrogen free extract	-0.293	-0.296	-0.321	-0.838*	-0.183	-0.874*
Neutral detergent fiber	-0.593	-0.316	-0.461	-0.017	-0.864*	-0.874*
Acid detergent fiber	0.220	0.508	0.551	0.699	-0.542	-0.504
Acid detergent lignin	-0.203	-0.518	-0.414	-0.581	0.523	-0.480
Cellulose	0.247	0.555	0.531	0.701	-0.523	-0.480
Hemicellulose	-0.687	-0.603	-0.763	-0.423	-0.451	-0.481
Non fiber carbohydrate	0.277	-0.012	0.217	0.151	0.734	0.738
Gross energy	-0.093	0.131	0.000	0.492	-0.271	-0.128

\*P<0.05; \*\*P<0.01; \*\*\*P<0.001; ns= not significant

<sup>1)</sup> Gv8= gas produced at 8 h; GP24= gas produced at 24h; GP48= gas produced at 48 h.

<sup>2)</sup> b (ml) is the maximum value of gas production;  $\mu_r$  (mL/h) is the rapid gas production rate during early stages of fermentation;  $\mu_s$  (mL/h) is the slower gas production rate during later stages of fermentation.

A highly significant ( $p<0.01$ ) positive association was observed between CF and parameter b. However, a significant ( $p<0.05$ ) negative relationship was found between NDF and estimated parameters  $\mu_r$  (rapid gas production rate during early stages of fermentation) and  $\mu_s$  (slower gas production rate during later stages of fermentation). In agreement with the findings of Garcia *et al.* (2005), there were no correlations observed between the maximum values of gas production (parameter b) with some chemical compositions.

Moreover, the positive association values of parameter b with NDF and parameter  $\mu_r$  (the rapid gas production rate during early stages of fermentation) with NDF and ADL contents reported by Parissi *et al.* (2005) are consistent with the current findings. However, the presence of a high negative correlation between the slower gas production rates during later stages of fermentation (parameter  $\mu_s$ ) with NDF content disagrees with the findings of Getachew *et al.* (2004). In conclusion, leaves of both Moringa species with high crude protein and essential amino acid with low structured carbohydrate contents had a higher nutritive value in terms of

energy content and *in vitro* organic matter digestibility. Thus, leaves of both Moringa species have the potential to contribute substantially as feed supplements to ruminants and non ruminants in animal production systems of Ethiopia and other sub-Saharan African countries.

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