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Proximate analysis and *in-vitro* gas production of predominant forages in Afe Babalola University rangeland as feed resources for ruminant production

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***Abstract*—*In Nigeria, the major feed resource for ruminant production is the natural grazeland. However, most forage found on such lands cannot absolutely support ruminant production. Therefore, there is need to ascertain the nutritive values of predominant forages in Afe Babalola University before setting up a ruminant farm. Wet season forages: grasses, legumes, forbes and tresses were sampled and analysed for proximate composition and in-vitro gas production using standard techniques. It was found that crude protein ranged between 12.2 and 27.3% in Terminalia catappa and Leucaena leucocephala respectively. The ash content varied from 6.0-22.0% in Andropogon gayanus and Asclepias syriaca respectively. Crude fibre of the forages was between 12.5 and 28.0% in L. leucocephala and Centrosema pubescens respectively. Gas production was measured for 24hrs at 3hr intervals. At mid-fermentation, gas production ranged between 4.0 and 13.3ml/200mgDM (T. catappa and A. syriaca respectively). While at termination, it was from 9.0 - 22.67ml/200mgDM in T. catappa and A. syriaca respectively. Significant differences (P<0.05) existed among tested forages. Organic matter digestibility was from 37.7-58.54% in Tridax procumbens and A. syriaca respectively. Short chain fatty acid ranged from 0.27 - 0.6µmol in T. catappa and A. syriaca respectively. The methane gas ranged from 3.33-5.67mmol in Terminalia catappa and Calopogonium mucunoides respectively. In conclusion, most of the forages were found to be adequate for ruminant production in crude protein component. They were all noted to be very low in methane gas production which connotes energy loss in ruminant production. A good mixture of examined forages therefore, might serve as adequate feed resources for ruminant production in the area.*

***Keywords*— Forages, methane gas, crude protein, ruminants, native grassland.**

I. INTRODUCTION

Low productivity of ruminant livestock is mainly hinged on poor quality forage and its unavailability in quantity. The major resources for ruminants are cereal crop residues and pastures from rangelands. Livestock graze on about 26% of the world's land area. Tchinda *et al.* (1993) reported that native pastures are the most widely available low cost feeds for ruminants in the tropics. Native rangelands offer the cheapest source of nutrients for the ruminants. It is however an accepted fact that for a greater part of the year, grasslands in the tropics do not supply sufficient nutrients to stocks for actualizing enhanced productivity. Forage digestibility is related to intake rate, and affects animal performance positively. In addition, leafy swards provide more suitable intake conditions in view of the characteristics of animal ingestive behavior (Benvenutti *et al*., 2008).

In Nigeria, ruminants slowly gain weight in the rainy season and rapidly loose it in the dry season, yet in the traditional animal husbandry, ruminants are mainly fed with grasses, so that improved livestock production is not likely attainable and sustainable by forage grasses alone (Babayemi and Bamikole, 2006). Babayemi *et al*., (2003) had earlier reported that the forages are unimproved and low in nutritive values during the wet season, while during the dry season proper, they are fibrous, lignified with low protein values and even in short supply. Lamidi *et al*. (2010) agreed with this by reporting that available forages for most part of the year are low in protein content which leads to marked decrease in voluntary intake and digestibility, and subsequently leads to substantial weight loss of the animals during this period. The success of the livestock industry anywhere in the world depends greatly on feed quantity and quality. However, the expensive nature of conventional feed as a result of competition between man

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and livestock (Ogunbosoye and Babayemi, 2010), makes this combination difficult. Afe Babalola University, Ado-Ekiti has a very large expanse of rangeland which might be suitable as feed resource for ruminant production. However, the nutritive value of predominant forages in the area is yet to be determined and documented.

II. MATERIALS AND METHOD

Description of the study area

Afe Babalola University is located in Ado Ekiti, Nigeria. Ado Ekiti is a city in southwest Nigeria, the state capital and headquarters of the Ekiti State. It is also known as Ado. The people of Ado Ekiti are mainly of the Ekiti sub-ethnic group of the Yoruba (Wikipedia, 2016). Ado-Ekiti is mainly an upland zone, rising over 250meters above sea level. Ekiti State lies between longitude 5°13'17 East of Greenwich meridian and latitude 70 37'16 North of the Equator. The weather condition of the study area is tropical climate with temperature 26ºC, humidity 74%, Rainfall 300-1100mm.

Sample collection for analysis

Predominant forages were collected at Afe Babalola University Ado Ekiti. Common forages (trees, legumes, Forbs and grasses) in the study area were collected in the wet season (June peak of rainy season). Two forages per type were harvested for the analysis (tree: *Terminalia catappa* and *Leucaena leucocephala*, legumes: *Centrosema pubescens* and *Calopogonium mucunoides*, forbs: *Tridax procumbens* and *Asclepias syriaca*, grasses: *Panicum maximum* and *Andropogon gayanus*). The fresh samples were weighed and air dried for 48 hours and oven dried to a constant weight at 105ºC. Oven dried samples were milled (2mm sieve) and kept for analysis.

Proximate analysis

Crude protein, crude fiber, ether extract and ash contents of forages were determined according to AOAC (2000). Kjeldahl procedure was used to determine the total nitrogen present in forage samples. It was effected through the breaking down of 2g sample in 25 ml concentrated H_2SO_4 acid plus selenium, using Gerhardt Kjeldahtherm until an opaque colour was obtained. The digested sample was rested for 12hours, diluted with distilled water and made up to the mark in a 250 volumetric flask. 5ml of digest was pipette and distilled with 40% NaOH solution and the ionized ammonium was trapped by boric acid. The distillate was immediately titrated (n = 3) with 0.01N hydrogen chloride. The crude protein was obtained by multiplying the nitrogen with factor: 6.25.

***In-vitro* fermentation procedures**

Preparation of the buffer and rumen liquor was carried out as described by Menke and Steingass (1988). The substrate was placed in calibrated gas tight plastic syringes fitted with a piston. The syringes were put in an incubator at 39±1^{O}C. Rumen liquor was collected from three female West African Dwarf (WAD) goats, sieved with a four layered cheese cloth and mixed with a sodium buffer (9.8g $NaHCO_3$ + 2.77g $(Na)2HPO_4$ + 0.57g KCl + 0.47gNaCl + 0.12g$MgSO_4$ 7H_2O + $CaCl_2$. H_2O per 1000ml) in a ratio1:2 v/v. 200mg DM of each sample with 30ml of rumen liquor and buffer were placed in each syringe and incubated in triplicate under continuous flushing with CO_2. A blank (rumen liquor + buffer) without substrate was incubated at the same time. The reading of the blank was subtracted from that of the other syringes. Gas production was recorded at 3, 6, 9, 12, 15, 18, 21 and 24h. After 24h of incubation, 4ml of NaOH (10M) was introduced into inoculums as reported by Fievez *et al*. (2005) to estimate the amount of methane produced. The value of gas produced at intervals was plotted against the using the equation Y= a + b (1-ect) (Ørskov and Mc Donald, 1979), where Y= volume of gas produced at time t, a= initial gas produced, b= gas produced from insoluble but degradable fraction, c = the rate constant for the degradation of 'b' and t= incubation time.

Statistical Analysis

Data collected were subjected to analysis of variance at p=0.05.

III. RESULTS AND DISCUSSION

Proximate composition of predominant forages in Afe Babalola University rangeland

Shown in Table 1 is the proximate composition of predominant forages in Afe Babalola University**.** The crude protein for the present study varied from 12.2-27.3% in *T. catappa* and *L. leucocephala* respectively. The ash content also varied from 6-22% in *A. gayanus* and *A. syriaca* respectively. Crude fibre of the forages was between 12.5 and 28.0% in *L. leucocephala* and *C. pubescens* respectively. The ether extract ranged from 2.0 - 9.5% (*A. gayanus* and *T. catappa* respectively*)*, while the Nitrogen free extract of the forages was observed to vary from 30.5 - 53.7% in *A. syriaca* and *A. gayanus* respectively. It was noted that there were significant differences (P< 0.05) in all the measured parameters among the forages.

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Table.1: Proximate composition (% DM) of predominant forages in Afe Babalola University

Forages	Ash	CP	CF	EE	NFE
C. pubescens	7.0^{g}	18.7^{d}	27.0^{c}	7.0^{b}	40.3^{c}
T. catappa	14.0^{d}	12.2^{f}	17.7^{e}	9.5^{a}	46.6^{b}
C. mucunoides	10.0^{e}	23.8^{b}	28.0^{b}	8.0^{b}	30.2^{e}
A. gayanus	6.0^{h}	13.7e	24.6^{d}	2.0^{f}	53.7^{a}
T. procumbens	20.0^{b}	18.9^{d}	18.5^{e}	7.5^{b}	35.1^{d}
L. leucocephala	16.0^{c}	27.3^{a}	12.5^{f}	8.0^{b}	36.2^{d}
P. maximum	8.0^{f}	13.6e	31.9^{a}	4.5^{e}	42.0^{c}
A. syriaca	22.0^{a}	20.4^{c}	18.1^{e}	9. 0^{a}	30.0^{e}
S.E.M	0.88	1.24	1.0	0.045	1.58

^{a,b,c,d,e,f}= Means on the same column with similar superscript letters are not significantly different($P< 0.05$).

Where, CP = Crude protein, CF = Crude fibre, EE = Ether extract, NFE = Nitrogen free extract. SEM = Standard error of means

Proximate composition is usually the basic and most common form of forages evaluation by animal nutritionist. There are many factors affecting proximate composition and mineral content of forages such as stage of growth maturity, species or variety (Agbagla-Dohnani *et al*., 2001; Promkot and Wanapat, 2004). The Crude protein (CP) content of *Asclepias syriaca, Calopogonium mucunoides* and *L. leucocephala* were higher than the other species. The CP of the forage species ranged from 12.2 to 27.3%, which is above the 7% CP requirement for ruminants which will provide ammonia required by rumen microorganisms to support optimum microbial activity. *Andropogom gayanus* had a higher CP of 13.7% compared to Odedire and Babayemi (2008) CP: 9.36% the ether extract and ash had lower values to that reported by the same authors. The ash content (8%) of *Panicum maximum* noted in this study was different and lower to the findings of Odedire and Babayemi (2008) however, the CP (13.6) was higher than that reported by the same authors.

Amata and Lebari, (2011) reported crude protein of 11.70% in *Terminalia catappa* as against 12.2% noted in the present study. The workers however reported lower level of crude fibre and ash than reported in the present study. The values obtained for proximate composition of *Calopogonium mucunoides* were lower in terms of crude protein: 23.8% and Nitrogen free extract: 30.2% but higher in ash: 9.80%, crude fibre: 28.0% and ether extract:8.0% compared to what Mecha and Adegbola (1980) reported; ash (9.80%), crude fibre (21.60%), ether extract(3.10%) crude protein (24.08) and Nitrogen free extract (41.42).

The nutrient composition of *Centrosema pubescens* as obtained in this present study differs from that of Nworgu and Egbunike, 2013 who obtained 9.14% (ash) which is considered higher than the present study ash: 7.0%, crude protein 23.4% was also found higher than crude protein 18.7% of the present study. However, lower values for crude fibre: 8.80 and ether extract: 3.32% compared to that of the present study: crude fibre: 17.7% and ether extract: 9.5% were observed.

Generally, the variation that existed between the present study and the past works on the forages considered may be traced to time and seasons of harvest, age of plant, leaf to petiole ratio, ecological location and edaphic (soil) (Makkar and Beacker, 1997; Babayemi and Bamikole, 2006).

***In-vitro* gas production of forages collected from the rangeland in Afe Babalola University.**

Presented in Figure 1 is the *in-vitro* gas production of forages from the rangeland in Afe Babalola University. At the onset of fermentation, gas production ranged from 0.00ml/200mg DM in C. *mucunoides, P. maximum, T. procumbens, C. pubescens, A. gayanus and T. catappa* to 2.33ml/200mg DM in L. *leucocephala*. However at mid fermentation, it ranged from 4.00ml/200mg DM (*T. catappa*) to 13.33ml/200mg DM (*A. syriaca*), while at termination, the gas production ranged from 9.00ml/200mg DM to 22.67ml/200mg DM in *T. catappa* and *A. syriaca* respectively. Gas production varied significantly ($P< 0.05$) in the forages from the 3rd hour to termination at 24th hour in the present study.

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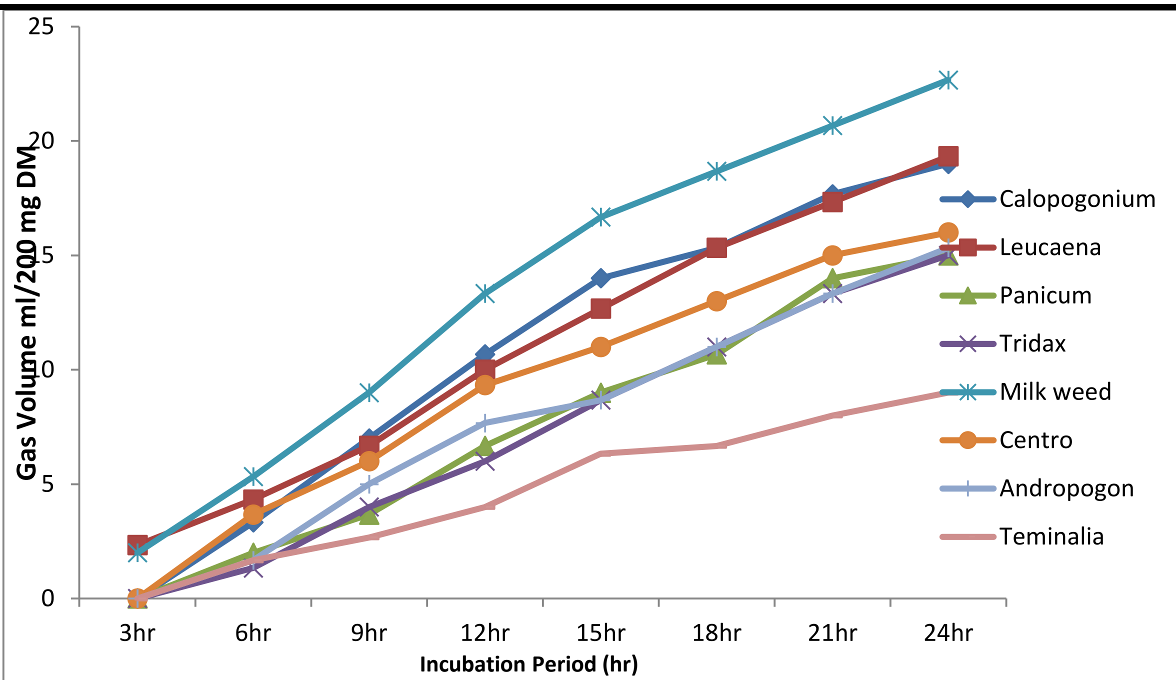

Fig.1:In vitro gas production of predominant forages in Afe Babalola University

Key

Terminalia catappa (Terminalia) *Leucaena* leucocephala (Leucaena), Centrosema *pubescens* (Centro), *Calopogonium mucunoides* (Calopogonium), *Tridax procumbens* (Tridax) *Asclepias syriaca* (milk weed), *Panicum maximum* (panicum) and *Andropogon gayanus* (Andropogon).

Presented in Table 2 is the Metabolisable energy MJ/Kg DM, organic matter digestibility (%) and short chain fatty acid (µmol) of predominant forages in Afe Babalola University. ME is an indication of energy and it ranged between 5.00 and 6.50 MJ/Kg DM (*Tridax procumbens* and *Asclepias syriaca* respectively). It was found to be significantly different among forages. Organic matter digestibility ranged from 37.7-58.54% in *Tridax procumbens* and *Asclepias syriaca* respectively with significant difference among forages. Short chain fatty acid which is an indication of energy made available to the host animal ranged from 0.27-0.60 µmol (*Terminalia catappa* and *Asclepias syriaca* respectively).

Table.2: Metabolisable energy (MJ/Kg DM), organic matter digestibility (%) and short chain fatty acid (µmol) of predominant forages in Afe Babalola University

Forages	ME	OMD	SCFA
C. pubescens	5.51^{bc}	50.63^{b}	0.44^{bc}
C. mucunoides	5.92^{b}	44.72^{c}	0.52^{b}
L. leucocephala	5.57^{bc}	46.67^{c}	0.52^{b}
A. syriaca	6.50^{a}	58.54^{a}	060^{a}
P. maximum	5.68^{b}	45.44^{c}	0.42^{c}
T. procumbens	5.00^{d}	37.70^{d}	0.40^{c}
A. gayanus	5.15^{cd}	39.85^{d}	0.43^{c}
T. catappa	5.02^{d}	45.48^{c}	0.27^{d}
SEM	0.07	2.90	0.0021

^{a,b,c}= Means on the same column with similar superscript letters are not significantly different($P< 0.05$).

Where ME – Methabolisable energy, OMD – Organic matter digestibility, SCFA – Short chain fatty acid, SEM = Standard error of means

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Norton (2003) justifies the use of forages in small quantities in order to supplement poor quality pastures and crop residues. It has been suggested that the gas production technique is more reliable than the nylon bag method for determining nutritive value of feeds containing anti-nutritive factors (Khazaal *et al.,* 1993). Nature and fibre levels, presence of anti-nutrition factor had been reported to influence the amount of gas produced during fermentation (Babayemi 2004). High level of crude fibre reduce digestibility which is synonymous to *in-vitro* gas production. In-vitro technique is a more reliable tool for evaluating ruminant forages. Though the two methods are independent of each other, they are interrelated (Babayemi *et al.,* 2004; Fievez *et al., 2*005).

Gas production is associated with volatile fatty acid production following fermentation of substrate (Blummel and Ørskov 1993). In addition, the application of models permits the fermentation kinetics of the soluble and readily degradable fraction of the feeds, and more slowly degradable fraction to be described (Gatechew *et al*., 1998). Moreover the gas production parameters of trees might demonstrate differences in their nutritional value that may be closely related to their chemical composition (Cerrillo and Juarez 2004). The inconsistency observed in the gas production is as a result of the different rate of different anti nutritional content as well as the forage degradability. *In-vitro* estimations of feed degradation are important tools for ruminant nutritionists. These methods measure either substrate disappearance or fermentation products (Blümmel *et al* 1997). In the present study, forages with high CP produced higher gas volume. Digestibility has been reported to be synonymous to *in vitro* gas production (Fievez *et al.*, 2005) that is, forages with high gas production will exhibit better digestibility.

***In-vitro* gas production characteristics of predominant forages in Afe Babalola University rangeland**

Presented in Table 3 is the *in-vitro* gas production characteristic of predominant forages in Afe Babalola University. It was observed that 'a' which is the initial gas produced ranged between 1.67 and 3.67 ml in *T. procumbens*, *A. gayanus, T. catappa* and *C. pubescens* respectively. Significant differences existed among the considered forages (P< 0.05). The potential gas production from insoluble but degradable fraction 'b' varied from 7.33-20.65 ml *T. catappa* and *Asclepias syriaca* respectively, while the rate of potential gas production (a+b) ranged from 9.00-22.67 ml in *T. catappa* and *Asclepias syriaca* respectively. Rate at which gas is produced 'c' ranged from 0.032-0.059(ml/h) in *Andropogom gayanus* and *Calopogonium mucunoides* respectively with no significant differences among the forages.

Table.3: In-vitro gas production characteristic of predominant forages in Afe Babalola University rangeland

Forages	a	a+b	b	c	T	Y
C. pubescens	3.67^{a}	16.00^{b}	12.33^{d}	0.051	12.00^{ab}	9.33^{ab}
C. mucunoides	3.33^{ab}	19.00^{b}	15.67^{bc}	0.059	12.00^{ab}	11.00^{a}
L. leucocephala	2.33^{ab}	19.00^{b}	17.00^{b}	0.052	13.00^{ab}	11.33^{a}
A. syriaca	2.00^{ab}	22.67^{a}	20.67^{a}	0.043	9.00^{b}	9.67^{ab}
P. maximum	2.00^{ab}	15.00^{c}	13.00^{cd}	0.041	12.00^{ab}	7.33^{ab}
T. procumbens	1.67^{b}	14.33^{c}	12.33^{d}	0.037	11.00^{ab}	6.33^{b}
A. gayanus	1.67^{b}	15.33^{c}	13.67^{cd}	0.032	10.00^{b}	6.00^{b}
T. catappa	1.67^{b}	9.00^{d}	7.33^{e}	0.054	14.00a	5.67^{b}
SEM	0.96	3. 67	3. 00	0. 0022	4.88	4.88

$^{a,b,c,d}{}_{=}$ Means on the same column with similar subscript letters are not significantly different(P< 0.05).

a- initial gas produced/intercept, b- insoluble but degradable fraction, a + b- potential extent of gas production, c-rate at which gas is produced, Y- gas volume, SEM = Standard error of means.

The intake of a feed is mostly explained by the rate of gas production (*c*) which affects the passage rate of feed through the rumen, whereas the potential gas production (*a +b*), is associated with degradability of feed (Khazaal *et al*., 1995). Therefore the higher values obtained for the potential gas production in the *Calopogonium mucunoides*, *Leucaena leucocephala* and *Asclepias spp* might indicate a better nutrient availability for rumen microorganisms.

Methane gas (mmol) produced at 24 h of incubating predominant forages in Afe Babalola University

Presented in Figure 2 is the methane gas (mmol) produced from incubating predominant forages in Afe Babalola University. The methane gas ranged from 3.33-5.67mmol in *Terminalia catappa* and *Calopogonium mucunoides* respectively.

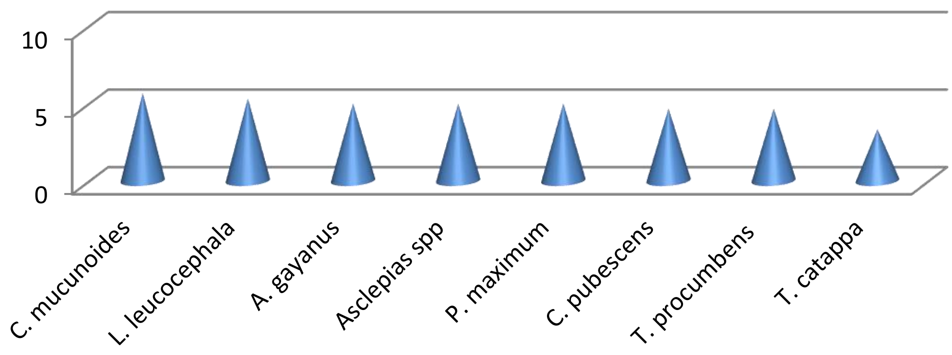

Fig.2: Methane gas (mmol) produced at 24 h of incubating forages

Methane production represent a significant energy loss to ruminants; it also contributes to global warming which is a worrisome phenomenon in the recent time and many tropical feedstuff have been indicated to increase methanogenesis (Babayemi *et al.,* 2004; Babayemi and Bamikole, 2006).

IV. CONCLUSION

The results revealed that *In-vitro* gas production techniques can be used to assess the nutritive value of forages. It unveiled the fact that most of the forages seem to be adequate in crude protein but deficient in ash content and fibre. Methane gas production was generally low and this is an indication that energy loss will be reduced and the forages will be environmentally friendly when fed to ruminants.

REFERENCES

[1] Agbagla-Dohnani, A., P. Noziere, G. Clement and M. Doreau, (2001). *In sacco* degradability chemical and morphological composition of 15 varieties of European rice straw. Anim. Feed Sci. Technol., 94: 15-27.

[2] AOAC (2000). Official Methods of Analysis, 21th edn. (Association of Official Analytical Chemist, Airlington,VA).

[3] Babayemi OJ, Bamikole MA (2004). Feeding goats with Guinea grass – Verano stylo and nitrogen fertilizer grass with energy concentrate. Arch. Zootec. 53:13 – 23.

[4] Babayemi, O.J, Bamikole, MA (2006). Effect of Tephrosia candida DC leaf and its mixtures with Guinea grass on *In-vitro* fermentation changes as feed for ruminants in Nigeria. Pakistan J. Nutr.,5(1): 14-18

[5] Babayemi, O.J., Bamikole, M.A and Odunguwa B.O. (2003). Haematological and Biochemical components of West African dwarf goats fed *Tephrosia bracteolata* – based forage. *Trop. Anim. Prod. Invest*. 6:31-38.

[6] Benvenutti, M.A.; Gordon, I.J.; Poppi, D.P.; Crowther, R.; Spinks, W. 2008. Foraging mechanics and their outcomes for cattle grazing reproductive tropical swards. Applied Animal Behaviour Science 113:15-31.

[7] Blümmel M, Makkar H P S and Becker K (1997) *In-vitro* gas production: a technique revisited. *Journal of Animal Physiology and Animal Nutrition, 77:24-34*

[8] Blummel, M. and E.R. Ørskov, (1993). Comparison of *In-vitro* gas production and nylon bag degradability of roughages in predicting feed intake in cattle. Anim. Feed Sci. Technol., 40: 109-119.

[9] Cerrillo M A and Juarez R A S (2004). *In-vitro* gas production parameters in cacti and tree species commonly consumed by grazing goats in a semiarid region of North Mexico. Livestock Research for Rural Development. 16 (4) 2004, http://www.cipav.org.co/lrrd/lrrd16/4/cerr16021.htm

[10] Fievez, V., O.J. Babayemi and D. Demeyer, (2005) Estimation of direct and indirect gas production in syringes: a tool to estimate short chain fatty acid

[11] Getachew G, Blümmel M, Makkar H P S and Becker K (1998) *In-vitro* gas measuring techniques for assessment of nutritional quality of feeds: A review. Animal Feed Science and Technology, 72:261-281.

[12] Khazaal K A, Dentinho M T, Ribeiro R and Ørskov E R (1993). A comparison of gas production during incubation with rumen contents *In-vitro* and nylon bag degradability as predictors of the Apparent digestibility in vivo and voluntary intake of hays. Animal Production, 57: 105-112.

[13] Khazaal, K., M.T. Dentinho, J.M. Ribeiro and E.R. Ørskov, (1995). Prediction of apparent digestibility and voluntary feed intake of hays fed to sheep: Comparison between using fibre component, *in-vitro* digestibility or characteristics of gas production or nylon bag degradation. Anim. Sci., 61: 521-538

[14] Lamidi, A.A., Aina, A.B.J and Sowande, S.O. (2010). Nutrient digestibility and nitrogen balance in West African dwarf goats fed blended diets for dry season. Proc. 35th Conf. Nig. Soc. Anim. Prod. held 14th – 17th March, 2010 at the Univ. of Ibadan, Nigeria. Pp. 499 – 501.

[15] Mecha I. and T.A. Adegbola (1980). Department of Animal Science University of Nigeria Nsukka, Nigeria: Chemical composition of some southern Nigeria forage eaten by goats.

[16] Norton, B.W. (2003). The Nutritive value of tree legumes. http://www.fao.org/ag/AGPC/doc/Publicat/ Gutt-shel/x5556e0j.htm.pp.1-10

[17] Odedire, J. A. and O. J. Babayemi (2008): Comparative studies on yield and chemical composition of *Panicum maximum* and *Andropogon gayanus* as influenced by *Tephrosia candida* and *Leucaena leucocephala*. *Livestock Research for Rural Development* 20(2): 1-8.

[18] Ogunbosoye, D. O. and Babayemi, O. J. (2010). Voluntary intake of Non-legume fodders offered simultaneously to West African Dwarf goats for a period of six hours. Proc. 35th Conf. Nig. Soc. for Anim. Prod. 14 – 17th March, 2010, Univ. of Ibadan, Nigeria. Pp. 518 – 520.

[19] Promkot, C. and M. Wanapat, (2004). Ruminal degradation and intestinal digestion of crude protein of tropical resources using nylon bag and three-step *In-vitro* procedure in dairy Cattle. In: Proceedings of the Agricultural Seminar, Animal Science/Animal Husbandry. Held at Sofitel Raja Orchid Hotel 27-28 January 2004.

[20] Stabile, S.S.; Bodini, A.P.; Jank, L.; Renno, F.P.; Santos, M.V.; Silva, L.F.P. 2012. Expression of genes from the lignin synthesis pathway in guineagrass genotypes differing in cell-wall digestibility. Grass and Forage Science 67:43-54.

[21] Statistical Analysis System Institute Inc. (2000): SASSTAT programme, Cary, NC: SAS Institute Inc.

[22] Tchinda B, D Wegard and R M Njwe (1993): Rumen degradation of elephant grass supplemented with graded levels of perennial peanut by West African dwarf sheep. In: Lebbie S H B, Rey B and Irungu E K (Editors). Small Ruminant Research and Development in Africa. Proceedings of the second biennial conference of the African small ruminants research network AICC, Arusha, Tanzania, 7-11, December 1992. ILCA/CTA. ILCA. Addis Ababa, Ethiopia pp 187-190

http://www.fao.org/Wairdocs/ILRI/x5472B/x5472b11.htm