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Studies on nutritional potentials of *Anacardium occidentale* (Cashew) leaf as off season feed for ruminants in the humid tropics

[*]Mako, A. A., Ettu, R. O and Olaleye, D. J.
Department of Agricultural Science, Tai Solarin University of Education, Ijagun. Ijebu-Ode. Ogun State. Nigeria

[*]***Corresponding Author:*** *jokemako2006@gmail.com* ***Telephone Number:*** *08023292736 & 08057085436*

Abstract
This study was designed to investigate the nutritional potential of Anarcardium occidentale leaf (AOL) as off season feed for ruminants. Two studies were conducted; the first study involves evaluation of the nutrient, mineral and anti-nutrient content of green, yellow and brown AOL while the second study involves the in vitro gas production (IVGP) of AOL which was carried out over a period of 24 hr incubation to predict Metabolizable energy (ME), organic matter digestibility (OMD) and short chain fatty acids (SCFA). Methane (CH_4) was measured by introducing 10 M NaOH into the inoculums after incubation. Results revealed significant ($p<0.05$) differences in the values of the nutrient composition. The dry matter ranged between 89.68% and 90.91% in green and brown leaf respectively. The highest crude protein (17.44%) was obtained in green leaf, while the lowest (16.06%) was recorded for brown leaf. The mineral and anti-nutrient content of AOL were not significantly different from each other, but within acceptable limit for optimum metabolism. The total IVGP varied significantly ($p< 0.05$) with the green leaf recording the highest (16.00ml) value, while the lowest (7.00ml) value was obtained in brown leaf. Same trend was observed for IVGP parameter and characteristics, with green leaf recording the highest values, while the lowest values were obtain from brown leaf for all parameters measured). It can be concluded that AOL has potential in livestock nutrition. Also the problem associated with disposal of falling brown leaf can be conveniently reduced by incorporating them into ruminant feed.

Key words: *Anacardium occidentale leaf, nutritional potential, in vitro gas production, ruminants,*

Introduction

It is an established fact that feed accounts for 60 -70% cost of production (Thirumalaisamy *et al.,* 2016). Grasses are the bulk of feed for ruminants and are available in abundance with high nutrients during the lush season, animals are fattened during this period with less cost. These grasses soon become scarce, lignified and low in nutrients during the dry season. Feed shortage during the dry season has been identified as a critical factor, especially in the humid and sub-humid regions (Baur *et al.,* 2016). The animals are left with crop residues and agricultural wastes which could not meet their nutrient requirements nor sustain them (Ogunbosoye and Babayemi, 2008).

The implication of this is poor performance of animals as a result of malnutrition. Hence the supply of animal protein for human consumption becomes inadequate. Malnutrition and hunger – related diseases cause over 60% of deaths (UNICEF, 2007). Eliminating hunger and malnutrition is one of the most fundamental challenges facing humanity (Lomborg, 2004). Adequate food and optimal nutritional status are the foundation blocks for the building of healthy, secure lives and thus form the basis for development in any nation (NPFN, 2016). Nigeria's per capita daily protein intake is estimated to be 45.3g as against FAO minimum 53.8g for optimal performance (Iyangbe and Orewa, 2009), this is inadequate. A nation that demand optimum performance from her citizen must provide food at affordable cost. In order to ameliorate this challenge, alternative feed resources that is underutilized as fodder must be researched into. There are a number of browse plants yet to be exploited as ruminant feed. The forages are important because they are evergreen especially during the off season when grasses are almost not available.

This study therefore, aimed at investigating the nutritional potentials of *Anacardium occidentale*
(Cashew) leaf as feed supplements for ruminants in Nigeria.

Key words: *Anacardium occidentalea* leaf, *in vitro* gas production, nutritional potential, ruminant feed

Material and methods

Collection of samples

Green and yellow leaves are plucked from *Anacardium occidentale* tree while the brown leaves were picked from the floor around the root of the plant. Reasonable numbers of each leaf type were collected and taken to the laboratory for analysis.

Chemical analysis

Crude protein, crude fibre, ether extract and total ash of samples were analyzed in triplicates using standard procedure of A.O.A.C (2012). The crude protein was determined with the micro kjeldahl distillation apparatus, while the NDF, ADF and ADL were determined by Van Soest (1995) method (1995).

Analysis of minerals

A total of ten minerals were analysed. Plant parts were digested with nitric HNO_3 / $HClO_4$ mixtures (nitric acid and perchloric acid) (20:5 v/v). The digest was made up 100 ml in standard volumetric flask with deionzed water. Ca, Na, K, Fe, Cu, Mn, Zn, Mg and Pb in the digest were determined with the atomic absorption spectrophotometer model 420. (Gallenkemp and Co. Ltd). Phosphorus in the digest was estimated with vanadomolybdate solution. The colour so developed was read with spectrophotometer at 420 m/u.

Quantitative determination of phytochemicals

Tannin, saponin, oxalate and flavonoid contents were determined as described by Ejikeme *et al.*, (2014). Phytate and alkaloid contents was determined as described by Maga (1983) and Harbone (1973) respectively.

In vitro gas production

Rumen fluid was obtained from three West African Dwarf female goats through sunction tube before the morning feed. The animals were fed concentrate consisting of 40% corn bran, 35% wheat offal, 20% palm kernel cake, 4% oyster shell, 0.5% salt and 0.5% growers premix for three days prior to the collection of rumen liquor. Incubation was as reported (Menke and Steingass 1988) using 120 ml calibrated syringes in three batch incubation at 39 ^{0}C, into 200 mg samples in the syringes was introduced 30 ml inoculums containing cheese cloth strained rumen liquor and buffer ($NaHC0_3$ + Na_2HP0_4 + KCl + NaCl + $MgS0_4.7H_20$ + $CaCl_2.2H_20$) (1:2, v/v) under continuous flushing with $C0_2$. The gas production was measured at 3, 6, 9, 12, 15, 18, 21 and 24, after 24h of incubation, 4 ml of NaOH (10 M) was introduced to estimate the amount of methane produced. The average of the volume of gas produced from the blanks was deducted from the volume of gas produced per sample. The volume of gas produced at intervals was plotted against the incubation time, and from the graph, the gas production characteristics were estimated using the equation Y= a+b ($1-e^{-ct}$) described by Orskov and McDonald (1979), where Y= volume of gas produced at time 't', a = intercept (gas produced from insoluble fraction), c = gas production rate constant for the insoluble fraction (b), t = incubation time, metabolizable energy (ME, MJ /Kg DM) and organic matter digestibility (OMD, %) were estimated as established (Menke and Steingass 1988) and short chain fatty acids (SCFA, *u*mol) was calculated as reported (Getachew *et al*, 1999).

$$ME = 2.20 + 0.136^{*}GV + 0.057^{*}CP + 0.0029^{*}CF$$

$$OMD = 14.88 + 0.889GV + 0.45CP + 0.651XA$$

$$SCFA = 0.0239^{*}GV - 0.0601$$

Where GV, CP, CF and XA are net gas productions (ml /200 mg DM), crude protein, crude fibre and ash of the incubated samples respectively.

Statistical analysis

Data obtained were subjected to analysis of variance procedure (ANOVA) of SAS (2012). Significant means were separated by Duncan's (1955) Multiple Range Test.

Results and Discussion

The result of the chemical composition is presented in Table 1. It was observed that crude protein (CP), crude fibre (CF), neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) varied significantly ($p<0.05$), while dry matter (DM), ether extract (EE) and ash were not significantly different among treatment means. DM and CP ranged from 89.67% – 90.91% and 16.06% -17.44 % in green and brown leaf respectively. This result is in agreement with the findings of Omoniyi *et al.*, (2013) who reported 90.52 % (DM) and 17.26% (CP) for Albizia saman green leaf, but lower than the findings of Brenda *et al*., (1997), who reported 39.0% DM and 9.77% CP for cashew green leaf. NDF, ADF and ADL values ranged between 48.67 and 51.41; 36.41 and 39.07; 15. 05 and 18.00% respectively. These values are similar to values (55.99 %) for NDF; (42.4%) for ADF and 15.55% for ADL reported elsewhere for *Chromolena odorata* leaf (Omoniyi *et al.*, 2013). The CP content is in excess of that proposed as the minimum requirement for growth (113g CP/Kg DM) and lactation (120 CP/Kg DM) in ruminants (NRC, 2001). This makes *Anacardium occidentale* a good source of protein supplement to low quality roughage. The value range of NDF (48.67 to 51.41%) obtained here is lower than the recommend limit of 60.00% guaranteed for forage intake by ruminants (Wanapat *et al.*, 2013).

The macro and micro mineral contents of different leaf forms are presented in Table 2. The result revealed that all leaf forms contained all minerals investigated. There were no significant variations among the different leaf forms for all the minerals investigated. However the values are within the recommended safe limit that will enhance optimum functioning of the body system (NRC, 2001).

Table 1: Chemical composition (%) of *Anacardium occidentale* (cashew) leaf

	Different forms of cashew leaf			
Parameters	Green	Yellow	Brown	SEM
Dry matter	89.68	90.33	90.91	1.05
Crude protein	17.44[a]	16.83[b]	16.06[c]	0.33
Crude fibre	15.91[c]	18.01[b]	19.52[a]	0.38
Ether extract	4.14	3.86	3.60	0.49
Ash	13.30	13.15	12.92	0.41
Neutral detergent fibre	48.67[c]	50.01[b]	51.41[a]	0.39
Acid detergent fibre	36.42[c]	37.71[b]	39.07[a]	0.30
Acid detergent lignin	15.05[c]	16.32[b]	18.00[a]	0.20

[a,b,,c=]means on the same row with different super script differed significantly ($p<0.05$).
SEM:Standard error of mean.

Table 2: Macro and micro mineral content of *Anacardium occidentale* (cashew) leaf

	Macro minerals (%)					Micro minerals (mg/kg)				
Leaves	Ca	P	K	Na	Mg	Fe	Zn	Cu	Mn	Pb
Green	0.523	0.358	0.971	0.348	0.440	324.8	38.2	8.7	74.1	15.3
Yellow	0.515	0.376	0.955	0.365	0.461	311.4	36.4	7.6	72.3	14.8
Brown	0.481	0.354	0.832	0.341	0.427	306.6	32.1	7.2	69.7	14.0
SEM	0.21	0.15	0.33	0.15	0.29	10.9	4.5	0.8	5.3	0.9

SEM= Standard Error of mean

Presented in table 3 is the anti–nutrient content of different leaf forms of *Anacardium occidentale*. All the leaf forms were implicated for all the anti-nutrients investigated. No significant variation occurred among the different leaf form. The level of tannin, oxalate, saponin, phytate, alkaloid and flavonoid obtained in this study ranged from 0.33-1.19; 0.401-0.420; 0.328-0.74; 0.55-0.681; 0.50-0.57 and 2.00-2.23 % respectively. The values obtained here are lower than values reported for *Moringa oleifera* leaf (Ezeonu and Ejikeme, 2016). Tannins were reported as one of the most important of the bioactive constituents of plants. Tannins are plant polyphenols, which have the ability to form complexes with metal ions and with macro-molecules such as proteins and polysaccharides thereby protecting them from ruminal degradation (Akubugwo and Ugbogu, 2007). They can reduce feed efficiency and weight gain when value is above threshold (Ogbe and George, 2012). Saponin suppresses methanogenesis which is a major energy loss to animals (Wang *et al*., 2009). Oxalate can form complexes with most essential trace elements, thereby making them unavailable for enzymatic activities and other metabolic processes, consumption of large doses of oxalic acid causes corrosive gastroenteritis, shock, convulsive symptoms and renal damage (Eneobong, 2001). Phytic acid inhibits the absorption and utilization of some mineral elements (Eneobong, 2001). Alkaloids have a wide range of pharmacological activities including antimalarial (quinine), anticancer (e.g. homoharringotonine) (Kittakoop *et al*., 2014), antibacteria (e.g. chelerythrine) (Cushnie *et al.*, 2014) and antihyperglycemic activities (e.g. piperine) (Qiu *et al*., 2014). Flavonoids are known to have antioxidant effects and have been shown to inhibit the initiation, promotion and progression of tumors (Kim *et al.,* 1994), also reduction of coronary heart disease has been reported to be associated with intake of flavonoids (Hertog *et al.,* 1993). Most phytochemicals serve as natural antibiotics, which assist the body in fighting microbial invasion and infections (Sodipo *et al*., 2000). All phytochemical investigated were within the safe limit for consumption (Teferegene, 2000). This makes the presence of these phytochemicals in *Anacardium occidentale* leaf beneficial.

Table 3: Phytochemical content (%) of different forms of *Anacardium occidentale* (cashew) leaf

	Phytochemicals					
Leaf form	Tannin	Oxalate	Saponin	phytate	Alkaloid	Flavonoid
Green	1.19	0.420	0.74	0.681	0.57	2.23
Yellow	0.52	0.413	0.336	0.635	0.53	2.16
Brown	0.33	0.401	0.328	0.559	0.50	2.00
SEM	0.27	0.31	0.31	0.31	0.20	0.50

SEM = standard error of mean

Table 4 shows the *in vitro* gas production parameters of green, yellow and brown leaf of *Anacardium occidentale* estimated from gas production. Significant ($p<0.05$) differences was observed among the different leaf forms for metabolizable energy (ME), organic matter digestibility (OMD), short chain fatty acid (SCFA). They ranged from 3.64 – 5.50 MJ/Kg DM, 39.83 – 51.27% and 0.17 – 0.50 µmol in brown and green leaf respectively. This result is comparable and in agreement with the value range of 4.90- 6.22 MJ/Kg DM; 38.43- 48.67 % and 0.33-0.50 µmol reported for some non-leguminous browse plants (Ogunbosoye and Babayemi, 2010). Methane (CH_4) production from the different leaf forms of *Anacardium occidentale* ranged from 2.00; 4.50 and 7.00 in brown, yellow and green respectively. This result is lower than the values of 18.17 ml and 20.01 reported for *Spondia mombin* leaf (Ogunbosoye and Babayemi, 2010 and Omoniyi *et al*., 2013) respectively. Feedstuff that show high capacity for gas production are also observed to be synonymous for high methane production (Njidda, 2010), hence the reason for high methane production in green *Anacardium occidentale* leaf than yellow and brown leaf. Methane production is an energy loss to ruminants and also has environmental implication as a major contributor to greenhouse gas emissions (USEPA, 2014). Chemical composition in combination with *in vitro* digestibility and ME content can be considered useful indicators for preliminary evaluation of potential nutritive value of forages (Kafilzadeh and Heidary, 2013).

Table 4: Post-incubation Parameters of different forms of *Anacardium occidentale* (cashew) leaf

	In vitro Parameters			
Leaves	ME (MJ/Kg DM)	SCFA (µmol)	OMD (%)	CH_4 (ml/200mg DM)
Green	5.50^a	0.50^a	51.27^a	7.00
Yellow	4.14^b	0.26^b	42.81^b	4.50
Brown	3.64^c	0.17^c	39.83^c	2.00
SEM	0.36	0.06	2.38	3.5

$^{a,b,c=}$means on the same column with different super script differed significantly ($p<0.05$)
ME = Metabolizable energy; SCFA = Short chain fatty acid; OMD = Organic matter digestibility; SEM = standard error of mean

The *in vitro* gas production characteristics: soluble degradable fraction (a), insoluble degradable fraction (b), potential degradability (a+b) varied significantly, except for rate of degradation (c) that was not significantly different among the leaf forms (Table 5). Fraction 'a' indicates the amount of gas produced from soluble degradable fraction of the samples. This is the fraction that rumen microbes first ferment for them to obtain energy for immediate requirement. The 'a' fraction ranged from 2.50 – 6.00ml brown and green leaf respectively, this result indicate that green leaf consist of more degradable carbohydrates. The 'b' indicates the fraction that is insoluble but degradable, the highest (17.34ml) was obtained in green leaf, while the lowest (7.00ml) value was recorded for brown leaf. These values are lower and at variance with the value range (49.041 and 52.487 ml) reported *Leucaena leucocephala* leaves collected during wet and dry season respectively (Arifuddin *et al.,* 2017). This is the fraction that is fermentable at a slower rate compared to the 'a' fraction. It is the fraction that rumen microbes ferment after the rapidly fermented fraction has been depleted; the 'b' fraction then becomes the major source of gas generated during the course of fermentation. The lower 'b' values obtained in this study are indications of the fibrous nature of the incubated samples (Mako, 2009). The 'a+b' fraction indicates the potential degradability of the green, yellow and brown leaf. It ranged from 9.50 –23.34ml in brown and green leaf respectively. These values are lower than values of 98.1 ml reported for *Leucaena leucocephala* (Babayemi 2007). The green leaf has the highest (1.19 ml/h) rate of degradation 'c' among the leaf forms, while the lowest (0.13ml/h) was reorded for the brown leaf. This result is lower and at variance with the report of Ogunbosoye and Babayemi (2010) for *Spondia mombin* leaf. The values of ME, OMD and SCFA obtained here is an indication that animals will be able to obtain energy from the different leaf forms.

Table 5: *In vitro* gas production characteristics of different forms of *Anacardium occidentale* (cashew) leaf

	In vitro Characteristics			
Leaf forms	A	b	`c	a+b
Green	6.00^{a}	17.34^{a}	0.08	23.34^{a}
Yellow	3.50^{b}	9.76^{b}	0.11	13.26^{b}
Brown	2.50^{c}	7.00^{c}	1.19	9.50^{c}
SEM	0.94	2.17	0.11	2.68

$^{a,b,c=}$means on the same column with different super script differed significantly ($p<0.05$)
a= soluble degradable fraction; b= insoluble degradable fraction; a+b = Potential degradability; c= rate of degradation,
SEM = standard error of mean

Figure 1 presents the gas production pattern of green, yellow and brown leaves of *Anacardium occidentale* incubated for 24 hr. Gas production is a reflection of the effectiveness and extent of degradability of forages because forages that have high ruminal degradability of dry matter tend to have high gas production (Njidda and Nasiru, 2010). High level of lignin influences degradation of fibre and non-fibre negatively (Mizubuti *et al.,* 2011). The content of lignin obtained here is high, hence the low gas production. There was steady increase in the gas production over a period of 24 hr, no significant variation was observed in the gas volume produced at every interval of time. There are many factors that may determine the amount of gas to be produced during fermentation, depending on the nature and level of fibre, presence of secondary metabolites and the potency of rumen liquor (Babayemi *et al.,* 2004). The total gas produced here ranged from 6.00; 8.50 and 16.0ml in brown, yellow and green leaf respectively. The value for green leaf is similar to value (20.0ml) reported for cashew (Brenda *et al*., 1997), but lower and at variance with the range of 43 – 175 ml reported for *Spondia mombin* (Ogunbosoye and Babayemi, 2010). This could be attributed to the age of the plant at harvest, which is an indicator of high lignin content. The low gas production of *Anacardium occidentale* leaf in this study connotes low digestibility of the leaf, which may be due to the presence of anti-nutritional factors and high level of fibre.

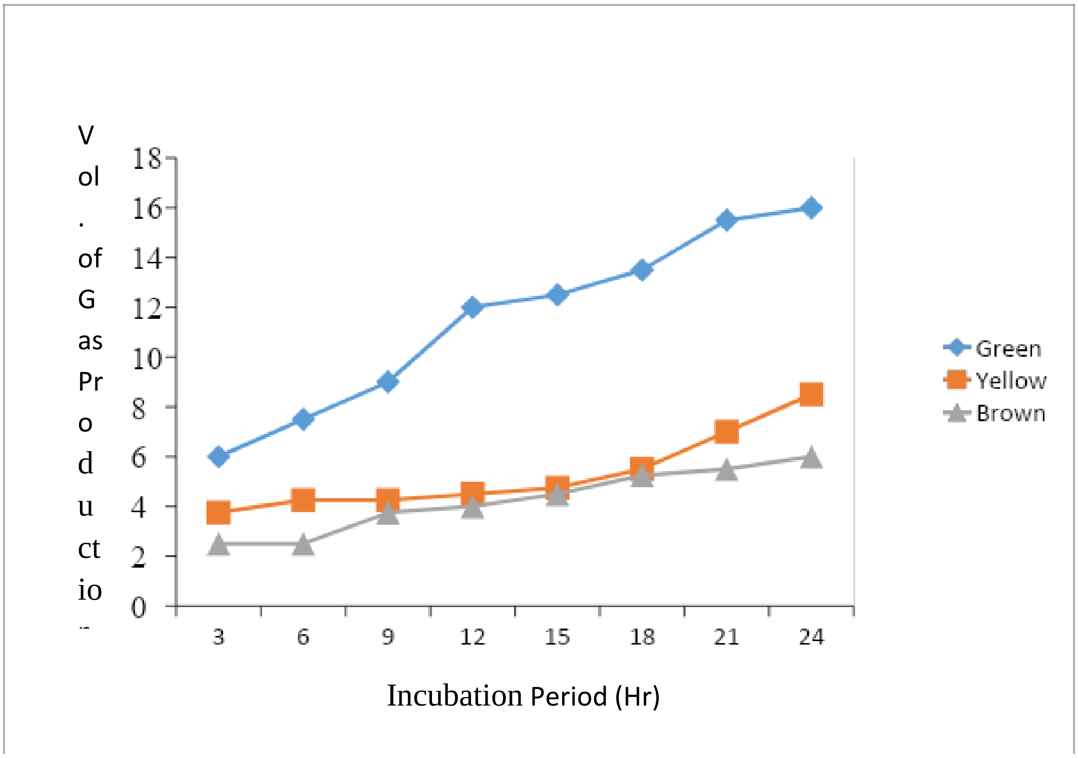

Fig 1: Gas Production (ml/200mg DM) of green, yellow and brown leaf of *Anacardium occidentale*

Conclusion and Recommendations

The results obtained in this study revealed that the different forms of *Anacardium occidentale* leaf has potential as forage for ruminants especially in the dry season.

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