

**ORIGINAL RESEARCH ARTICLE****Comparative evaluation of nutritive value of ensiled *Alternanthera brasiliana* leaf and stem as dry season feed for ruminants in the tropics*****Mako, A. A, Olonade, A. I and Babatunde, W. O**Department of Agricultural Science,
Tai Solarin University of Education, Ijagun.
PMB 2118. Ijebu-Ode, Ogun State, Nigeria*Corresponding author: MAKOAA@tasued.edu.ng
jokemako2006@gmail.com.; +2348023292736**ABSTRACT**

Silage is a method of preserving and sustaining nutritious pasture for ruminants to be consumed during the dry season. Alternanthera brasiliana (AB) is a perennial herb that is available all year round. In this study, AB was ensiled with Palm kernel cake, Molasses and Wheat offal. Experiment I determined silage characteristics and nutrient composition of ensiled AB, Coefficient of preference (CoP) of ensiled AB by WAD sheep were determined in experiment II. The nutritive value of ensiled AB was assessed using in vitro gas production technique to predict metabolizable energy (ME), organic matter digestibility (OMD), short chain fatty acid (SCFA), methane (CH₄) and Total gas produced (TGP) in experiment III. Results revealed that ensiling imparted in the AB plant as the temperature, colour, aroma, texture and pH of all silages were within the recommended range. Ensiling influenced the chemical composition significantly (P<0.05). The dry matter (DM) ranged from 50.36 to 75.52 % in AB ensiled with molasses (ABMOL) and wheat offal (ABWHO) respectively. The Crude protein values for ABMOL, ABWHO and ABPKC are 13.23%, 29.30% and 30.40% respectively. It was observed that ensiling improved the DM. Neutral detergent fibre ranged from 28.77 to 35.23% in ABMOL and ABWHO respectively. Macro and micro minerals analyzed were not significantly affected by ensiling, but within recommended level for consumption. The CoP ranged from 1.00 – 1.25 in ABWHO and ABMOL respectively. Judging by the CoP values, all ensiled AB were acceptable to the animals. However the ABMOL was most preferred. TGP, ME, OMD and CH₄ differed significantly (p<0.05). However, ABMOL recorded the highest values for all parameters analyzed. It can be concluded that ensiling improved the nutrient composition, daily intake and in vitro gas production parameters of AB leaf and stem. Hence, ensiled AB has potential as off season feed for ruminants in the tropics.

Keywords: *Alternanthera brasiliana*, in vitro gas production, nutrient composition, ruminant, silage.

INTRODUCTION

Feed shortage during the dry season has been identified as a critical factor especially in the tropics and sub-tropics (Kolan *et al.*, 2012). This stems from, among other reasons, high population pressure leading to less land allocated to improved pasture production which can be conserved for dry season. As a result farmers rely on harvesting forages from land not suitable for cultivation which are natural communal grasslands. However, rapid clearance of natural communal grasslands for crop cultivation as well as bushfires during dry season reduces the forage supply. In addition, natural forages precariously tend to be of low feeding value especially in the dry season. This

is further compounded by some variation in rainfall pattern that negatively affects pasture production (Kasulo *et al.*, 2012). However these feed resources are not well characterized. This necessitates screening studies to evaluate feeding value of these naturally growing forages utilized by farmers. This will complement earlier natural forage evaluation studies (Munthali and Dzwela, 2005) since the choice of natural forages evaluated could not have been exhaustive. Evaluation of the nutritive value of natural forages is essential to provide a basis for development of adequate diets for ruminants including the need for supplementation (Chumpawadee *et al.*, 2012).

There are various multipurpose plant leaves and fruits which still remain relatively unexplored as feed supplement for ruminant production. Joy weed is one of such multipurpose trees which grow widely as shade plant in Nigeria and other high rainfall areas of West Africa (Arikwe *et al.*, 2012). Joy weed, is a browse plant that is common in many parts of West Africa and it is available throughout the year. Physical and chemical procedures such as grinding and ensiling would enhance the availability of protein, carbohydrate, fibre, fat, mineral etc and subsequently result in increased feed intake (Preston and Leng, 1984)

This study therefore, aimed at determining the nutritive value of ensiled Joy weed leaf and stem for potential supply of required nutrients to ruminants during the dry season period.

MATERIALS AND METHODS

Sample Collection and processing of *Althernanthera brasiliiana* (AB) plants for ensiling

Thirty (30) *Althernanthera brasiliiana* (AB) plants within the premises of Tai Solarin University of Education, Ijebu-Ode were marked randomly for collection of samples. 5kg of leaf and stem was harvested from each plant. Ensiling was carried out at the Agricultural Science Laboratory where wilting and storage were done. After harvesting, fresh plant shoots were separated from the roots, chopped and then wilted under shade for 24 hours on polythene sheets.

The material was then weighed and mixed in turn with the following additives: (i). Palm kernel cake (PKC), (ii) Wheat offal (WHO) and (iii). Molasses (MOL) at ratio 4: 1. (4 parts of AB plant to be ensiled: 1 part of additive). This was replicated three times in a completely randomized design for each silage treatment. Fermentation period was 21days.

Experimental silos

Polythene bags each capable of holding at least 30 kg of chopped wilted AB were used as silos. Each bag was placed inside a 65litre capacity plastic basin for reinforcement and ease of fermentation. Ensiling was by rapid compaction

of the material into the silos to displace the air until the polythene bags were filled. Sealing of the silo was done by placing a 25 kg sand-bag on top of the polythene bags after tying carefully and firmly.

Silage characteristics determination

Temperature: Immediately the silage was opened, thermometer was dipped into each silage bag to measure the temperature.

pH: 2g of each silage sample was measured into a 50 ml beaker, then 15 ml of distilled water was added, the beaker and the content was placed on bunsen burner and allowed to boil for 5 minutes, the liquid content was decanted and allow to cool. The pH meter was standardized using buffer solution, after which it was dipped into the cooled decanted liquid to determine the pH (OECD, 2013).

Aroma: The smell was observed as pleasant or unpleasant

Texture: Some samples were taken by hand to feel the texture to be either wet or firm

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 method (1995).

Analysis of minerals

A total of ten minerals were analysed. Plant parts were digested with HNO₃ / HClO₃ mixtures (nitric acid and perchloric acid) (20:5 v/v). The digest was made up to 100 ml in standard volumetric flask with deionized 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 (AOAC 2000).

***In vitro* gas production**

Rumen fluid was obtained from three female West African dwarf 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 °C. 30 ml inoculums was introduced into 200 mg samples in the syringes containing cheese cloth strained rumen liquor and buffer (NaHCO₃ + Na₂HPO₄ + KCl + NaCl + MgSO₄.7H₂O + CaCl₂. 2H₂O) (1:2, v/v) under continuous flushing with CO₂.

The gas production was measured at 3, 6, 9, 12, 15, 18, 21 and 24hrs after which 4 ml of NaOH (10 M) was introduced into syringes 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, μ 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

Acceptability study

The acceptability study was carried out at the sheep and goat unit of the Teaching and

Research farm, Department of Agricultural Science, Tai Solarin University of Education, Ijagun. Ijebu-Ode. Ogun State. The ensiled AB plants were used for the study. Eight adult West African Dwarf Sheep housed in group pen were used in the cafeteria feed preference study that lasted for two weeks, including one week of adaptation. 4kg each of AB plant ensiled with molasses, palm kernel cake and wheat offal were introduced on cafeteria basis to the animals in three different containers. The positioning of the ensiled plant was changed daily to prevent bias by the animals taking a particular part of the pen as the position for a particular silage. The amount consumed was monitored for eight hours daily and the quantity consumed for each silage was recorded. The animals were then released for grazing. Feed preference was determined from coefficient of preference (CoP) value calculated from the ratio between the intakes of each individual feed sample divided by the average intake of three feed samples (Mako, 2009). On this basis, a feed was taken to be relatively preferred if the CoP value is greater than unity.

$$CoP = \frac{\text{Intake of individual forage offered}}{\text{average intake of all forage samples}}$$

Statistical analysis

Data obtained were analyzed and subjected to analysis of variance procedure (ANOVA) of SAS (2012). Significant treatment means were separated by Duncan's multiple range test of the same package.

RESULTS AND DISCUSSION

Table1 presents the silage quality of the different silage diets. A well fermented silage should exhibit colour similar to the actual forage ensiled (Babayemi *et al.*, 2009). AB plant is known to be purplish brown in colour. The texture of a good forage should be firm (Akinwande, 2011). All silages properly fermented come out with pleasant aroma confirming the fact that additives might have contributed positively to making good silages. This agrees with the report of Abegunde *et al.*, (2017). The temperature ranged between 29 °C and 30 °C, this is consistent with the values reported by Akinwande *et al.*, (2011). This indicated a well preserved silage as

temperature is one factor that could affect silage colour. High temperature above 30 °C could lead to caramelization of sugar in the forage. pH values ranged between 4.2 and 4.5 which may be regarded as pH of good silages. Kung (2010) classified a good silage to be below a pH of 5.5.

The pH range obtained in this study was lower and better than the pH range (4.45 – 5.40) reported by Abegunde *et al.*, (2017) for water hyacinth ensiled with breadfruit as feed for WAD goats.

Table 1: Quality of ensiled *Alternanthera brasiliana* leaf and stem

Ensiled plant	Temp (°C)	pH	Aroma	Colour	Texture
ABWHO	29.0	4.2	Pleasant	Brown	Firm
ABPKC	30.0	4.5	Pleasant	Dark brown	Firm
ABMOL	29.0	4.5	Pleasant	Dark brown	Firm

ABWHO= *Alternanthera brasiliana* leaf and stem ensiled with wheat offal; ABPKC= *Alternanthera brasiliana* leaf and stem ensiled with palm kernel cake; ABMOL= *Alternanthera brasiliana* leaf and stem ensiled with molasses

Presented in Table 2 is the chemical composition of ensiled AB plant. All parameters analyzed differed significantly ($p < 0.05$). Dry matter ranged from 50.36 to 75.52% in ABMOL and ABWHO respectively. This result is similar to those reported by Amata and Lebari (2011). The CP ranged from 13.23 - 30.40% in ABMOL and ABPKC respectively. These values are higher than values reported by Akinwande *et al.*, (2011) for water hyacinth ensiled with different additives. The CP content of all ensiled AB plant were higher than the minimum requirement for growth (113g CP/Kg DM) and lactation (120 CP/Kg DM) in ruminants (NRC, 2001). It was observed that ensiling increased the CP content of AB plant, this might be due to relatively higher crude protein content of additives and a rapid drop in pH that inhibited proteolytic activity and ammonia nitrogen produced helped in getting the aerobic stability because of its fungicidal properties (Abegunde *et al.*, 2017). The neutral detergent fibre ranged from 28.77 to 35.23% in ABMOL and ABWHO respectively. Same trend was observed for acid detergent fibre and lignin, they ranged from 15.61 to 19.19 and 7.60 to 10.35% respectively. Fibre is very essential in livestock feed as it provides roughages which aids digestion (Neelam *et al.*, 2015). However, the values obtained here are below the recommended limit (60%) below which intake might be depressed (Messiner, 1999).

Table 2: Chemical composition (%) of ensiled *Alternanthera brasiliana* leaf and stem

Parameters	ABWHO	ABPKC	ABMOL	SEM
Dry matter	75.52 ^a	53.87 ^b	50.36 ^c	0.51
CP	29.30 ^b	30.40 ^a	13.23 ^c	0.23
EE	4.57 ^b	4.96 ^a	3.29 ^c	0.02
Ash	9.12 ^c	10.26 ^b	14.34 ^a	0.12
NDF	35.23 ^a	32.17 ^b	28.77 ^c	0.35
ADF	19.19 ^a	16.10 ^b	15.61 ^c	0.20
ADL	10.35 ^a	9.71 ^b	7.60 ^c	0.15

a,b,c=means on the same row with different super script differed significantly ($p < 0.05$); ABWHO= *Alternanthera brasiliana* leaf and stem ensiled with wheat offal; ABPKC= *Alternanthera brasiliana* leaf and stem ensiled with palm kernel cake; ABMOL= *Alternanthera brasiliana* leaf and stem ensiled with molasses. SEM= standard error of mean, CP=Crude protein, EE= Ether extract, NDF= Neutral detergent fibre, ADF= Acid detergent fibre, ADL= Acid detergent lignin

The values obtained for micro and macro minerals in this study were not significantly different. (Table 3). However, the values obtained were within the accepted limit for optimal metabolic activities (NRC, 2001).

Data of gas production parameters for AB ensiled with different additives are presented in Table 4. The ME and OMD values for all ensiled AB plants differed significantly ($p < 0.05$). The ME, SCFA and OMD ranged from 5.31 to 6.31 MJ/Kg DM; 43.94 to 38.46% respectively. This shows that the ABP ensiled with molasses contains more metabolizable energy than AB ensiled with PKC and wheal offal.

Table 3: Mineral contents of ensiled AB leaf and stem

Silage	Macro (%)					Micro (Mg/kg)				
	Ca	P	K	Na	Mg	Fe	Zn	Cu	Mn	Pb
ABWHO	0.350	0.480	0.870	0.215	0.440	225.15	38.39	5.10	15.82	0.006
ABPKC	0.395	0.499	0.880	0.228	0.451	221.70	42.10	5.65	17.40	0.008
ABMOL	0.417	0.572	0.962	0.236	0.509	237.00	44.18	5.80	17.92	0.004
SEM	1.02	1.03	1.02	1.03	1.02	10.92	11.11	1.00	9.30	0.01

a,b,c=means on the same column with different super script differed significantly (p<0.05); ABWHO= *Alternanthera brasiliana* leaf and stem ensiled with wheat offal; ABPKC= *Alternanthera brasiliana* leaf and stem ensiled with palm kernel cake; ABMOL= *Alternanthera brasiliana* leaf and stem ensiled with molasses. SEM= standard error of mean

This value is lower than those reported for some tropical browse plants which ranged from 8.31-11.88 MJ/kg DM (Babayemi, 2007). Metabolizable energy is source of energy to animals consuming the feed samples. Feedstuff that are inherent with certain anti-nutritive factor have been reported to be low in metabolizable energy and organic matter digestibility. (Aregheore and Abdulrazak 2005). The high ME and OMD obtained in this study might be an indication of very low anti-nutritional factor present in AB plant. SCFA ranged from 0.453 to 0.334µmol in ABWHO and ABMOL respectively. However, no significant differences was observed among the treatment means (p>0.05) for SCFA. These values are lower than the values range (0.94 - 0.10µmol) recorded for *Gmelina arborea* (Abiola Olagunju *et al.*, 2018), however, the SCFA value obtained in this study compared well with the values reported for some dry season forages (Mako *et al.*, 2012). Short chain fatty acid is an indication that the silages contain energy. The methane production values obtained here indicates that *Alternanthera brasiliana* plant ensiled with molasses yielded more methane gas than the samples ensiled with PKC and wheat offal. This is expected because it is an established fact that feedstuff with high gas production also have high methane (CH₄) production (Mako, 2009). Methane production indicates an energy loss to the ruminant (Babayemi and Bamikole, 2006).

Table 5 presents the Coefficient of preference (CoP) of AB plant ensiled with different additives by WAD sheep. The CoP by the animals showed that all the ensiled AB plant were acceptable to the animals since the CoP for ABWHO, ABPKC and ABMOL in this study was greater than unity (Mako and Babayemi, 2008).

Table 4: *In vitro* Parameters of *Alternanthera brasiliana* plant ensiled with different additives

Ensiled	<i>In vitro</i> gas production Parameters			
	ME (MJ/Kg DM)	SCFA (µmol)	OMD (%)	CH ₄
ABWHO	5.31 ^c	0.334	38.46 ^c	4.00 ^c
ABPKC	5.89 ^b	0.418	41.53 ^b	5.30 ^b
ABMOL	6.31 ^a	0.454	43.94 ^a	6.50 ^a
SEM	1.10	0.15	2.05	1.00

a,b,c=Means on the column with different super script differed significantly (p<0.05); ME = Metabolizable energy; SCFA = Short chain fatty acid; OMD = Organic matter digestibility; ABWHO= *Alternanthera brasiliana* leaf and stem ensiled with wheat offal; ABPKC= *Alternanthera brasiliana* leaf and stem ensiled with palm kernel cake; ABMOL= *Alternanthera brasiliana* leaf and stem ensiled with molasses. SEM = standard error of mean

A number of factors may influence acceptability of feed by ruminants. Nikki (2012) reported that forage aroma, taste, texture and moisture are the most vital factors that influence preference for feed by animals. The aroma and texture of ABMOL was the most pleasant and firm. This might be the reason why it is most preferred.

Table 5: Mean of daily intake of ensiled AB plant and the coefficient of preference by WAD sheep

Leaf forms	Mean daily consumption by all eight animals (kg/DM)	Coefficient of preference
ABWHO	3.00	1.00
ABPKC	3.40	1.10
ABMOL	4.00	1.25

ABWHO= *Alternanthera brasiliana* leaf and stem ensiled with wheat offal; ABPKC= *Alternanthera brasiliana* leaf and stem ensiled with palm kernel cake; ABMOL= *Alternanthera brasiliana* leaf and stem ensiled with molasses.

The rate and extent of gas production (Fig.1) is an indicator of the degree of digestibility and fermentation of feeds and microbial protein synthesis (Elghandour *et al.*, 2015). The result showed that the net volume of gas produced increased progressively with time of incubation. Total gas produced ranged from 1.5 – 2.5ml within the first 3hrs, and after 24hrs the total gas produced increased to 28ml in ABP ensiled with molasses and 17ml in ABP ensiled with wheat offal. Molasses are known to present typical characteristics of a readily available substance with the fastest rate of gas production. It increased the energy sources thus resulting in a greater volume of gas produced (Chuanqi *et al.*, 2018). This may have been the reason for high gas produced by molasses treated *Alternanthera brasiliensis* plant.

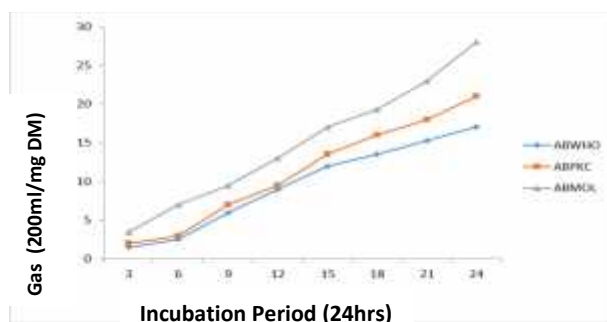


Fig 1: Gas production (200ml/mg DM) of ensiled *Alternanthera brasiliensis* plant

CONCLUSION

The findings obtained for quality of the silages, chemical composition, *in vitro* gas production parameters and acceptability of ensiled *Alternanthera brasiliensis*, confirms the plant as a potential feed resource for ruminants during the off season in the tropics. Any of the additives is suitable for ensiling the plant. Coefficient of preference revealed that AB ensiled with molasses was most preferred.

CONFLICT OF INTERESTS

Author declared that no conflict of interest exists.

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