Proximate and *In vitro* Gas Production of Toasted *Enterolobium cyclocarpum* Based Multi-Nutrient Feed Block

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ABSTRACT

This study was conducted to determine the proximate composition (dry matter, crude protein, ether extract, organic matter, ash) of *Enterolobium cyclocarpum* based feed blocks and *in vitro* gas production, *in vitro* gas characteristics including organic matter digestibility (%), metabolizable energy (ME, MJ/Kg DM), short chain fatty acid (μmol/200mgDM), and dry matter digestibility, was investigated.

Dried and milled samples of feed blocks were incubated using 200mg/30ml inoculum from West African Dwarf Rams for 3, 6, 9, 12, 15, 18, 21, 24, 36, 48, and 72 hours. Five feed blocks (FB) were produced containing toasted *Enterolobium cyclocarpum* seed meal (TECSM) and *E. cyclocarpum* foliage meal (ECFM), respectively, at the ratio of 0:0% (Control: FB1), 0:30% (FB2), 10:20% (FB3), 20:10% (FB4), and 30:0% (FB5) inclusion levels. FB2, FB3, FB4 and FB5 had higher dry matter and crude protein content (90.67, 83.33, 67.67, 81.00% DM) and (15.58, 21.36, 18.71, 20.28% DM), respectively, compared to FB1 (64.50% DM) and (12.05 % DM). There was an increase in the volume of gas production as incubation period extended from 3 to 72 hours (Table 3). The highest volume of 42.75 ml/200mgDM was recorded for 30% TECSM + 0% ECFM (FB 5) at the end of 72 hours incubation.

To this effect, ruminant animals could be fed feed blocks (FB) containing toasted *Enterolobium cyclocarpum* seed meal (TECSM) and *E. cyclocarpum* foliage meal (ECFM) up to 30% inclusion level.

(Keywords: toasted *Enterolobium cyclocarpum* seed meal, TECSM, *E. cyclocarpum* foliage meal, ECFM, feed block)

INTRODUCTION

The use of fodder trees and shrubs to enhance productivity of small ruminants in Nigeria has been given some considerable attention over five decades (Makkar and Becker, 1999). Scarcity of conventional feed supply coupled with decline in pasture quality in the dry season of the year has been a major cause of low productivity of livestock in Nigeria (Akinfala and Tewe, 2002; Aye, 2007).

Recent studies (Arigbede, *et al.,* 2008) revealed that some of the indigenous multipurpose tree species (MPTS) such as *Enterolobium cyclocarpum*, which are evergreen (produce seeds) with high protein content and low secondary metabolites do exist. Preliminary analysis of the seeds indicated that they were edible and contained substantial amount of nutrients and low anti-nutritional factor which suggested that they could support high productivity of animals (Arigbede, *et al.,* 2008).

Multi-nutrient feed blocks (MNB) have been used mainly to supply nitrogen to improve rumen function, but have also been the means of delivering other nutrients, minerals or therapeutic substances to improve animal performance (FAO, 2007).

Even though various researches had been done on the use and benefits of feed-blocks as feed supplements, additional information is still needed to fully exploit the benefits of incorporating various forages, nutrients, minerals and additives in feed blocks to serve as whole ration for feeding of livestock (FAO, 2007). This study was therefore targeted towards the production of *Enterolobium cyclocarpum* based multi-nutrient feed blocks thereby using it to preserve large biomass of *E. cyclocarpum* forage and its seed produced during the early raining season. This could then be used as feed supplement to ruminants grazing on poor quality roughages during the dry season and reduce communal clashes between crop farmers and nomadic cattle rearers.

MATERIALS AND METHODS

The feed block production, proximate composition and *in vitro* analysis of the feed block produced was carried out at the laboratory of the Department of Pasture and Range Management, College of Animal Science and Livestock Production, Federal University of Agriculture, Abeokuta, Ogun State.

Experimental Materials

The materials used for the feed block production were *E. cyclocarpum* seed and foliage. The mature *E. cyclocarpum* pods were picked up after falling off from multipurpose tree arboretum established in the intensive feed garden of Directorate of University Farms, Federal University of Agriculture, Abeokuta. They were sun-cured for three days and later de-hulled to obtain the seeds. Molasses was sourced from Dangote Sugar Refinery, Apapa, Lagos State. Other materials such as wood ash, stirrer, common salt, groundnut haulm, cassava peels and water were sourced locally. Items such as plastic molds, nylon, plastic bowls, 200 liters plastic drum, stirrer, were purchased from local market.

Toasting of *Enterolobium cyclocarpum* Seeds

The *Enterolobium cyclocarpum* seeds were toasted in the oven for about 15 minutes at 170^{o}C until the testa started breaking. The foliage was harvested from the same source and air dried for 7 days to 15% moisture content.

Feed Block Production Procedure

The procedure of Asaolu (2012) was used to produce the experimental multi-nutrient feed block.

Proximate Composition of the Feed Blocks

Dried and milled samples of each feed block were analyzed for proximate composition (Dry matter, Crude protein, Ether extract, Ash and Organic matter) according to A.O.A.C. (2000).

In vitro Analysis of Forage Based Multi-Nutrient Feed Blocks

In vitro gas production was run as described Menke and Steingass (1988). 200mg of each of the milled feed blocks in triplicates were placed in 100ml graduated glass syringe. Rumen fluid (inoculum) was collected from six West African Dwarf rams and filtered through three layers of cheese cloth in a pre-warmed, insulated bottle and taken to the laboratory.

The rumen liquor was mixed with sodium and ammonia bicarbonate buffer solution containing 35g $NaHCO_3$ plus 4g NH_4HCO_3 per liter at a ratio 1:2(v/v) to avoid lowering the pH of the rumen fluid which could decrease microbial activities. 200 mg of each feed blocks was accurately measured into the graduated glass syringes. The syringes were placed in a water bath at 39^{0}C in an upright position filled with 30ml of incubation medium consisting of 10ml of rumen fluid and 20ml of buffer solution. The gas released was read off directly on the graduated syringe. Triplicate blank syringes containing 30 ml of the buffered inoculum was only included as control. Gas production was recorded at 0, 3, 6, 9, 12, 15, 18, and up to 72 hours of incubation.

The data obtained were fitted into a non- linear equation:

$$V\ (ml/\ 200mgDM) = b\ (1-e^{-ct})$$

Where V= potential gas production at time t,
b= the volume of gas that evolved with time
c= the fractional rate of gas production.

Initial gas (Abs_g) was calculated as the product of b and c (Larbi *et al*., 1996)

Metabolizable energy was calculated as: $ME = 2.20 + 0.1357GV + 0.0057\ CP + 0.0002859\ EE^2$ (Menke and Steingass, 1988).

Table 1: Composition (%) of Feed Blocks Containing Graded Levels of Toasted *Enterolobium cyclocarpum* (TECSM) Seed and Foliage Meals.

Ingredients	FB 1	FB 2	FB 3	FB 4	FB 5
Cassava peels	39.50	19.50	19.50	19.50	19.50
Groundnut haulms	35.00	25.00	25.00	25.00	25.00
TECSM	0.00	0.00	10.00	20.00	30.00
ECFM	0.00	30.00	20.00	10.00	0.00
Starch	10.00	10.00	10.00	10.00	10.00
Molasses	11.50	11.50	11.50	11.50	11.50
Wood ash	3.00	3.00	3.00	3.00	3.00
Salt	1.00	1.00	1.00	1.00	1.00
Total	100. 00	100. 00	100. 00	100. 00	100. 00

FB = feed block, TECSM = toasted *Enterolobium cyclocarpum* seed meal, ECFM= *Enterolobium cyclocarpum* foliage meal, FB 1 = 0 % TECSM + 0 % ECFM, FB 2 = 0 % TECSM + 30% ECFM, FB 3 = 10% TECSM + 20% ECFM, FB 4 = 20% TECSM + 10% ECFM, FB 5 = 30% TECSM + 0 % ECFM.

Table 2: Proximate and Fiber Composition (%) of Feed Blocks Containing Graded Levels of Toasted *Enterolobium cyclocarpum* (TECSM) Seed and Foliage Meals.

Ingredients	FB 1	FB 2	FB 3	FB 4	FB 5
Dry matter	64.50	90.67	83.33	67.67	81.00
Crude protein	12.05	15.58	21.36	18.71	20.28
Ether extract	1.68	1.76	1.72	1.67	1.69
Organic Matter	97.18	96.50	95.04	95.04	95.64
Ash	2.88	3.50	4.96	4.96	4.36

FB 1 = 0 % TECSM + 0 % ECFM, FB 2 = 0 % TECSM + 30% ECFM, FB 3 = 10% TECSM + 20% ECFM, FB 4 = 20% TECSM + 10% ECFM, FB 5 = 30% TECSM + 0 % ECFM.

Organic matter digestibility (OMD) was calculated as: OMD = 14.88 + 0.889GV + 0.45CP + 0.651 Ash (Menke and Steingass, 1988).

Short chain fatty acids (SCFA) as: SCFA= 0.0239GV – 0.0601 (Getachew *et al*., 1999);

where GV is the total volume of gas produced at 24 hours, CP is the crude protein, EE is the ether extract.

Statistical Analysis

All results obtained were subjected to analysis of variance in a completely randomized design using SAS program general linear model procedure (SAS, 2002). Significant means were compared using the Duncan's Multiple Range Test (Duncan, 1955).

RESULTS AND DISCUSSION

The crude protein contents of the feed blocks ranges from 12.05 in 0% TECSM + 0% ECFM (FB 1) to 21.36% in 10% TECSM + 20% ECFM which is similar to the values (23.20 to31.52 %) reported by Isah, *et al*. (2012), when some selected browse species were analyzed for proximate composition.

There was an increase in the volume of gas production as incubation period extended from 3 to 72 hours (Table 3). The highest volume of 42.75 ml/200mgDM was recorded for 30% TECSM + 0% ECFM (FB 5) at the end of 72nd hour incubation. The values of gas production for the feed blocks from the 12th hour were significantly ($p<0.05$) higher than that of control feed block and 20% TECSM + 10% ECFM. 20% TECSM + 10% ECFM gave the least gas production after 24th hour till 60th hour.

Table 3: *In vitro* Gas Production of Feed Blocks Containing Graded Levels of Toasted *Enterolobium cyclocarpum* (TECSM) Seed and Foliage Meals.

Time (hr)	FB 1	FB 2	FB 3	FB 4	FB 5	SEM
3	3.00ab	2.75ab	3.00ab	1.75b	5.00a	0.40
6	6.75a	5.50ab	7.25a	2.50b	9.50a	0.75
12	15.75a	12.50a	13.25a	4.00b	17.25a	1.47
15	20.00a	15.25a	14.75a	4.75b	20.75a	1.75
18	22.25a	17.50a	16.00a	6.00b	23.50a	1.90
24	27.00a	21.00a	20.00a	8.00b	28.00a	2.26
30	29.00a	23.50ab	22.75ab	11.00b	31.00a	2.34
36	32.25ab	25.00ab	27.50ab	17.50b	34.00a	2.32
48	34.50a	29.50a	33.50a	25.75a	38.25a	2.13
60	36.50	31.75	36.75	35.75	40.50	1.82
72	37.50	33.50	40.75	41.50	42.75	1.67

ab : Means with different superscripts along the row are significant (P<0.05), FB feed block, hr= hour, FB 1-FB 5 –See Table 1 footnote, SEM = Standard errors of mean.

Table 4: Post incubation Parameters, Metabolizable Energy (ME), Organic Matter Digestibility (OMD), Short Chain Fatty Acid (SCFA), Dry Matter Digestibility (DMD), of Feed Blocks Containing Graded Levels of Toasted *Enterolobium cyclocarpum* (TECSM) Seed and Foliage Meals.

Parameter	FB 1	FB 2	FB 3	FB 4	FB 5	SEM
Bg(ml/200mg) DM	15.32d	38.68c	44.63b	55.75a	14.71d	3.78
Cg (ml/hr)	0.79ab	0.04b	0.49ab	1.20a	0.75ab	0.15
Lag time (hr)	1.10a	0.39b	0.18b	1.23a	1.12a	0.11
DMD (%)	78.23c	85.19ab	84.64b	78.01c	88.12a	0.95
ME (MJKg^{-1} DM)	5.94a	5.14a	5.04a	3.39b	6.12a	0.31
OMD (%)	44.50a	40.40ab	42.26a	30.41b	48.87a	1.97
SCFA (μmol)	0.59a	0.44ab	0.42ab	0.26b	0.61a	0.55

abcd : Means with different superscripts along the same row are significant (P<0.05), FB feed block, Bg = Volume of gas produced in time (t), Cg = Fractional rate of gas production, DMD= Dry matter digestibility, ME= Metabolisable energy, SCFA= short chain fatty acid, FB 1-FB 5 –See Table 1 footnote , SEM = Standard errors of mean.

The pattern of the gas production in of in all the experimental feed blocks generally increases from 3rd hour to 72nd hour of incubation, but specifically FB 5 (30% TECSM + 0% ECFM) gas production in this study increases steadily and had the highest gas volume at 24th hour, 48th hour and 72nd hours which is similar to the *in vitro* gas production reported by Babayemi (2009) when wild cocoyam was treated with cold water at 24th hour of gas production and also similar to the report of Babayemi (2009) when some selected tropical seeds were incubated but slightly lower to the report of Babayemi (2006) when *Enterolobium cyclocarpum* seeds were incubated and this could be as a result of toasting method employed in treating the *Enterolobium cyclocarpum* seeds as a component of the experimental feed block.

The gas production increased as time of incubation increases, the greatest proportion of the production occurred in the first 24 hours which is related to the findings of Al-Masri, (2007) who reported that the greatest gas volume obtained was produced in the first 24 hours of incubation when a study was carried out on mixture of different forages.

The total fractional rate of gas production of feed blocks studied showed that 20% TECSM + 10% ECFM had the highest (P<0.05) rate of gas production with the value of 1.20 ml/hr while 0 % TECSM + 30% ECFM had the lowest rate (0.04 ml/hr) (Table 3). The volume of gas produced during incubation of 20% TECSM + 10% ECFM (55.75 ml/200mgDM) was also significantly (P<0.05) higher than volume produced from 0% TECSM + 0% ECFM, 0% TECSM + 30% ECFM, 10% TECSM + 20% ECFM and 30% TECSM + 0% ECFM.

The highest (P<0.05) lag time was recorded at 20% TECSM + 10% ECFM (1.23 hr) while the least lag time was recorded at 10% TECSM + 20% ECFM (0.18 hr).

The post- incubation parameters metabolizable energy (ME), organic matter digestibility (OMD), short chain fatty acid (SCFA) of the experimental feed block are presented in Table 4. The values for ME ranged from 3.39 $MJKg^{-1}DM$ in 20% TECSM + 10% ECFM to 6.12 $MJKg^{-1}DM$ in 30% TECSM + 0% ECFM.

The digestibility of organic matter in feed block was highest in 30% TECSM + 0% ECFM (48.87%) and least in 20% TECSM + 10% ECFM (30.41%).The dry matter digestibility in feed block was highest in 30% TECSM + 0% ECFM (88.12%) and least in 20% TECSM + 10% ECFM (78.01%).

The organic matter digestibility recorded in this study was significantly different ($P< 0.05$). This is in line with the observation of Babayemi (2009) who reported an organic matter digestibility range of 32.26% to 56.60% when wild cocoyam was treated with cold water. The differences noted could be as a result of different feed resources used.

Gas production is a reflection of the generation of short chain fatty acids (SCFA) and microbial mass (Getachew, *et al*., 1994). The short chain fatty acids (SCFA) estimated from the gas production in this study were lower than the range observed by Babayemi (2009) who reported a range of 0.65 µmol to 0.97 µmol when wild cocoyam was treated with cold water and 0.75 µmol to 1.185 µmol when some tropical seeds were analyzed for ME, OMD, and SCFA by Babayemi (2009).

The values of short chain fatty acid (SCFA) ranged from 0.26 µmol in 20% TECSM + 10% ECFM to 0.61 µmol in 30% TECSM + 0% ECFM.

Metabolizable energy is a good index for measuring the quality of feeds particularly forages. The metabolizable energy was highest ($p<0.05$) in the FB 5 (30% TECSM + 0% ECFM) (6.12 KJ/Kg DM) and significantly lowest ($p>0.05$) in FB 4 (20% TECSM + 10% ECFM) (3.39 KJ/Kg DM) but the range observed in this study was lower than the range of 7.5 to 10.4 KJ/Kg DM observed by Babayemi, *et.al*. (2009), when some tropical seeds were analyzed for their nutritive quality. Similar trends were observed for OMD and SCFA productions. These show that there was a mutual relationship between ME, OMD and SCFA (Aganga and Mosase, 2001).

The fermentation of the insoluble but degradable fraction (b) which was measured by the volume of gas produced in time (t) value is higher than what was reported by Isah *et al*. (2012) for browse plant.

The value obtained for the fractional rate of gas production (c) of the feed blocks was higher than values of 0.01-0.95 $mlhr^{-1}$ reported elsewhere (Isah, *et al*., 2012). The relative higher value obtained in this study may be attributed to higher nutrient composition of the feed blocks as influenced by different constituent it was compounded with.

The rate at which different chemical constituents were fermented is a reflection of microbial growth and accessibility of the feed to microbial enzymes (Getachew, *et al.,* 2004). Similarly, Khazaal, *et al.* (1996) suggested that the intake of a feed is mostly explained by the fractional rate of gas production (c) which affects the rate of passage of the feed through the rumen. Thus the high value obtained for the (c), indicates a better nutrient availability for rumen microorganisms in animal fed with these feed blocks.

Dry matter digestibility ranged from 78.01 to 88.12% which was similar to the values of 68.66 – 86.79% reported by (Aye and Adegun, 2010). Osakwe *et al.* (2007) also recorded similar values of 69.6 – 84.9% dry matter digestibility. Results (Oladotun, 2003; and Taiwo *et al.*, 1995) showed that the digestibility of feeds increased as protein sources increased. This observation was consistent with Giri, *et al.* (2000) and Aregheore (2000) who affirmed that digestibility of nutrients varied with nutrient composition.

CONCLUSION

In conclusion, the results of this study shows that high crude protein content in all the feed blocks indicates that the feed block have the potential of being used as protein supplement for ruminants fed low quality roughages during the dry season. Though 30% TECSM + 0% ECFM (FB5) gave the highest positive responses in terms of gas volume, dry matter digestibility, organic matter digestibility, short chain fatty acid and substantial crude protein coupled with dry matter content, it could also be worthy of investigation for on-farm experiment and adoption.

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