In vitro fermentation and chemical constituents of urea-molasses feed - blocks made with different binders for ruminants.

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Abstract

This study was carried out to assess the chemical contents, in vitro break down, volatile fatty acids, NH3¬-N and methane concentration of urea- molasses feed - block (UMFB) using different binders. Four feed - blocks were formulated as: UMFB A (Cement only), UMFB B (cement + clay), UMFB C (cement + cassava starch), and UMFB D (cassava starch + clay). The feed - blocks were incubated in vitro for 48 hours to assess gas production and other in vitro- ecology parameters. Results of chemical analysis revealed that UMFB C (21.70%), and B (21.65%) had highest ($p< 0.05$) crude protein content while UMFB D (18.62%) had lowest value. Intermediate crude protein content was recorded for UMFB A (19.58%). However, highest ($p<0.05$) values of ADF (16.8%) and ADL (15.5%) were recorded for UMFB D. Similar value of ADF (16.70%) and ADL (15.3%) were recorded also for UMFB B. The NDF value of feed - block A (34.6%) was highest ($p<0.05$) while lowest value of NDF was recorded for UMFB D (32.1%). Production of volatile fatty acids indicated that propionic acid (30.5%), oleic acid (11.3%,) and lactic acid (36.5%) were highest ($p<0.05$) for UMFB D. Concentration of NH_3-N was highest in UMFB B while significant difference ($p>0.05$) was not observed in the pH and methane values for various UMFB. Highest in vitro digestibility of organic matter was observed in UMFB D. The study showed that the various feed - blocks could serve as a sustainable supplement during dry season and period of scarcity for the ruminants.

Keywords: *In vitro* degradation, urea- molasses feed - block

Introduction

One of the major constraints to increasing livestock productivity in developing countries is the scarcity and fluctuation in quantity and quality of the year-round supply of conventional ruminant feeds. With increase in demand for livestock products as a result of rapid growth in the world human population, expanding economies and shrinking land area, future hopes of feeding the millions of people and safeguarding their food security will depend on the better utilization of feed resources, which cannot be used as food for humans. Urea-molasses multi-nutrient blocks (UMFB) have been found to be highly beneficial and its use has been shown to be practical and effective in overcoming nutritional challenges in sheep, goats, beef and dairy cattle industry (Makkar, 2006). The *in-vitro* gas production method is an accurate and reliable tool for the evaluation of fermentation potentials of feeds and it predicts feed intake, digestibility, microbial nitrogen supply and animal performance (Blümmel and Ørskov, 1993). It is less expensive and less time-consuming. The *in-vitro* fermentation technique is capable of quantifying the amount of methane (energy loss) production (Fievez *et al.,* 2005), and determine the amount of acetate, propionate and butyrate (Babayemi *et al,.* 2004), carbon dioxide and metabolizable energy of feed for ruminants (Blümmel *et al.,* 1997). The objective of this study therefore is to assess the *in vitro* and chemical composition of urea-molasses feed - blocks as feed for ruminants.

Materials and Methods

Preparation of Urea Molasses Feed - Block

Locally available feed ingredients and agro-industrial by-products were sourced from commercial feed - miller in Abeokuta, Ogun State, which is located in the tropical rainforest zone in Nigeria within $7^0 10'$N and $3^0$2'E. The area has an average rainfall of 1100mm, a mean ambient temperature of about 34^0C and an average relative humidity of 82%. Four feed - blocks were formulated (Table 1) as: UMFB A (Cement only), UMFB B (cement + clay), UMFB C (cement + cassava starch), and UMFB D (cassava starch + clay). Production of feed - blocks was done using a "cold process" as described by Brar et al., (2006) but modified for local production (Table I). Urea and molasses were mixed together and left for 24 hours. Common - salt was mixed with corn - cob before mixing with other dry ingredients, to give uniform distribution of the premix. The urea-molasses mixture was poured into the premix of the dry ingredients, prepared on a big plastic bowl of 50 litre capacity, and mixed thoroughly. One kg of the semi-solid mixture was then put in small plastic cases (12.5 x 10 x 5.5cm), covered with a wooden sheet (that fitted well into the case) and compressed within two minutes. The plastic case was later pulled out leaving the formed UMFB on the polythene sheet. The UMFBs were left at room temperature for one week to dry so as to be hard enough for handling, transportation and feeding.

In vitro degradation analysis of the urea-molasses feed - block

Inoculum was obtained through sunction tube from fistulated West African dwarf (WAD) rams before morning feeding. The animals were previously fed with a mixed diet of fresh *Pennisetum purpureum* (60% DM) and concentrates (40% DM). The concentrate feed consisted of (as fed basis) 4% corn, 10% wheat offal, 10% palm kernel cake, 20% groundnut cake, 5% soyabean meal, 10% dried brewers' grain, 1% common - salt, 3.75% oyster shell and 0.25% fish - meal (Babayemi and Bamikole, 2009). The fluid was strained through four layers of cheese cloth into a pre-warmed, insulated bottle. All laboratory handling of rumen fluid was carried out under a continuous flow of CO_2. The incubation procedure was as reported by Menke and Steingass, (1988). The gas

Table 1: Ingredient composition of urea-molasses feed - blocks

Ingredients (%)	Feed-block Treatments			
	A	B	C	D
Molasses	35	35	35	35
Urea	10	10	10	10
Clay	-	5	-	5
Cement	10	5	5	-
Cassava starch	-	-	5	5
GNC	5	5	5	5
Wheat offal	20	20	20	20
Corn cob	10	10	10	10
Rice Bran	8	8	8	8
Common Salt	1.5	1.5	1.5	1.5
Vit/Min Premix	0.5	0.5	0.5	0.5

UMFB A (Cement only), UMFB B (cement + clay), UMFB C (cement + cassava starch), and UMFB D (cassava starch + clay).

production was measured on the calibration of the syringe for 48 hour.

Chemical analysis

Known weight of the UMFB from each feed – blocks was sampled air drying.The samples were oven-dried to constant weight at 65^{0}C. The dried samples were hammer-milled through a 1 mm sieve. Crude protein (CP), crude fibre (CF), ether extract (EE) and ash contents of samples were carried out in triplicates as described by AOAC (1990). The amount of CP was calculated (N×6.25). The neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were determined according to Van Soest et al (1991). The pH was measured using a pH meter (pH meter : CT-6020 Kedid waterproof pH meter XB858 SN: 85886948). The NH_3-N concentration was measured by spectrophotometry according to Chaney and Marbach (1962). The analysis of volatile fatty acids (VFA) concentration was carried out using Gas Liquid Chromatography (Samuel *et al.,* 1997). The molar percentages of propionic, acetic, lactic, butyric acid and oleic acids were determined. The volume of methane gas produced by each sample was determined by addition of 4 ml of 10N sodium hydroxide (Fievez et al., 2005).

Statistical analysis

In vitro Organic matter digestibility (IVOMD%) was assessed as IVOMD =14.88 + 0.889GV +0.45 CP +0.651XA according to Menke and Steingass, (1988). Data obtained were subjected to analysis of variance at p=0.05. Where significant differences occurred, the means were separated using Duncan's multiple range test of the SAS, (1990) options.

Results

Chemical composition of the urea-molasses feed - block

Table 2 shows the result of the chemical composition of the various feed - block. The UMFB D had the highest (p<0.05) DM content. The DM contents of the various UMFB ranged from 88.33 to 91.16%. Highest but similar values were recorded for the crude protein contents of UMFB B (21.65%) and C (21.70%). The UMFB D (18.62%) had least value of CP while intermediate value was recorded for UMFB A (19.58%). The crude fibre contents ranged from 3.70 to 3.19% in UMMB B and A, respectively and ether extract value from 2.73 (UMFB C) to 2.37% (UMFB A). The ash content values of 2.34% (UMFB B) and 2.22% (UMFB C) were noted. Result of fibre fraction revealed that UMFB A contained 34.6% NDF, 15.5% ADF and 14.6% ADL while similar values of ADF and ADL were recorded for UMFB B (16.7% and 15.3 %) and D (16.8% 15.50%), respectively.

Table 2: Proximate composition (g/100gDM) of urea-molasses feed-blocks.

Parameters	Feed-block Treatments				SEM
	A	B	C	D	
Dry matter	89.33^c	88.33^d	90.52^b	91.16^a	0.41
Crude protein	19.58^b	21.65^a	21.70^a	18.62^c	0.50
Crude fibre	31.9^c	37.0^a	35.0^b	35.0^b	0.70
Ether extract	2.37^b	2.40^b	2.73^a	2.70^a	0.06
Ash	2.30^a	2.34^a	2.22^b	2.31^a	0.02

^{a,b,c} Mean values in the same row with different superscripts differ significantly (P< 0.05), **SEM** – Standard Error of Mean. UMFB A (Cement only), UMFB B (cement + clay), UMFB C (cement + cassava starch), and UMFB D (cassava starch + clay).

Table 3: Fibre Composition (g/100gDM) of urea-molasses feed-blocks.

Parameters (%)	Feed-block Treatments				SEM
	A	B	C	D	
NDF	34.6[a]	34.1[b]	34.1[b]	32.1[c]	0.36
ADF	15.5[b]	16.7[a]	15.5[b]	16.8[a]	0.24
ADL	14.6[b]	15.3[a]	11.5[c]	15.5[a]	0.61

[a,b,c] Mean values in the same row with different superscripts differ significantly (P< 0.05), **SEM** – Standard Error of Mean, NDF - Neutral Detergent Fibre, ADF - Acid Detergent Fibre, ADL - Acid Detergent lignin. UMFB A (Cement only), UMFB B (cement + clay), UMFB C (cement + cassava starch), and UMFB D (cassava starch + clay).

Concentration of methane, NH_3-N and pH of urea-molasses feed - block.

In Table 3, UMFB B had the highest (p< 0.05) acetic, oleic and lactic acid productions (48.00, 11.13 and 36.00%, respectively), while similar values (p>0.05) of oleic and lactic acids were recorded for UMFB C (11.3 and 36.5%) and D (11.3 and 0.0364) respectively. The UMFB A had the least propionic acid (19.50%). UMFB B had highest butyric acid concentration (20.2%) and highest ratio of acetic and propionic acids (1.89). The value of methane production ranged from 13.50 in UMFB A to 17.00 ml/200mgDM in UMFB B. Low pH (p>0.05) was recorded for all the UMFB. The pH values ranged from 5.8 to 6.3. Concentration of NH_3-N and methane production observed in this study were significant (p<0.05) with UMFB B having the highest values of NH_3-N (6.1 $mgdL^{-1}$) and methane (17.00 ml/200mgDM) while UMFB A had least values of NH_3-N (4.6 $mgdL^{-1}$) methane (13.50 ml/200mgDM).

Discussion

Chemical composition of the urea-molasses feed - block

Fertilizer grade - urea was used to prepare the multi nutrient feed - blocks for the supply of non-protein nitrogen for rumen microbes while molasses supplied readily fermentable carbohydrate for energy. The crude protein obtained in this study was lower than the report of Wanapat and Khampa (2006), who reported lower NDF but higher ADF values. However, higher NDF values were reported by Boukila et al. (2006). This might be because higher quantity of coarse rice bran and lower quantity of urea was used in their study. Comparable results of ether extract, ash and crude fibre were reported by Onwuka (1999). The reason for the high DM content of UMFB C and D containing cassava

Table 4: Volatile fatty acid production of urea-molasses feed-blocks.

Parameters (%)	Feed-block Treatments				SEM
	A	B	C	D	
Propionic acid	19.50[c]	25.50[b]	24.50[b]	30.50[a]	0.15
Acetic acid	18.00[c]	48.00[a]	24.00[b]	23.00[b]	0.44
Oleic acid	8.50[b]	11.30[a]	11.30[a]	11.30[a]	0.45
Lactic acid	27.00[b]	36.00[a]	36.00[a]	36.50[a]	0.15
Butyric acid	15.80[c]	20.20[a]	19.60[b]	19.50[b]	0.66
Acetic:Propionic	0.78[c]	1.89[a]	0.98[b]	0.75[c]	0.18

[a,b,c] Mean values in the same row with different superscripts differ significantly (P< 0.05), **SEM** – Standard Error of Mean. UMFB A (Cement only), UMFB B (cement + clay), UMFB C (cement + cassava starch), and UMFB D (cassava starch + clay).

Table 5: Concentration of methane, NH_3-N and pH in urea-molasses feed-blocks.

Parameters	Feed-block				SEM
	A	B	C	D	
Methane(ml/200mgDM)	13.50^{c}	17.00^{a}	14.00^{c}	16.00^{b}	0.30
NH_3-N($mgdL^{-1}$)	4.60^{d}	6.10^{a}	4.80^{c}	5.10^{b}	0.20
pH	5.80	5.86	6.30	5.90	0.12

^{a,b,c}Mean values in the same row with different superscripts differ significantly ($P< 0.05$), **SEM** – Standard Error of Mean

NH3-N = ammonia nitrogen. UMFB A (Cement only), UMFB B (cement + clay), UMFB C (cement + cassava starch), and UMFB D (cassava starch + clay)

starch as binder might probably be as a result of the fact that cassava starch might not be able to confer high water retention ability on the final mixture of the feed - block mixtures when compared with cement and clay binders (Mahir, et al., 2009). The significantly high value observed in CP content of UMFB A could not be explained because cement is not a protein feedstuff. It might be that the cement inclusion triggered proliferation of micro-organisms, leading to the rise in protein. However, the CP content of the various UMFBs was higher than the 7% minimum CP requirement recommended for ruminants in the tropics (Minson, 1980). Feed-block C having 0% clay, 5% cassava starch and 5% cement in its component had the least ash content.

Concentration of methane, NH_3-N and pH of urea-molasses feed-blocks.

The insignificant result observed in methane production might be due to the fact that the same quantity of urea and molasses were supplied to meet microbial need in the

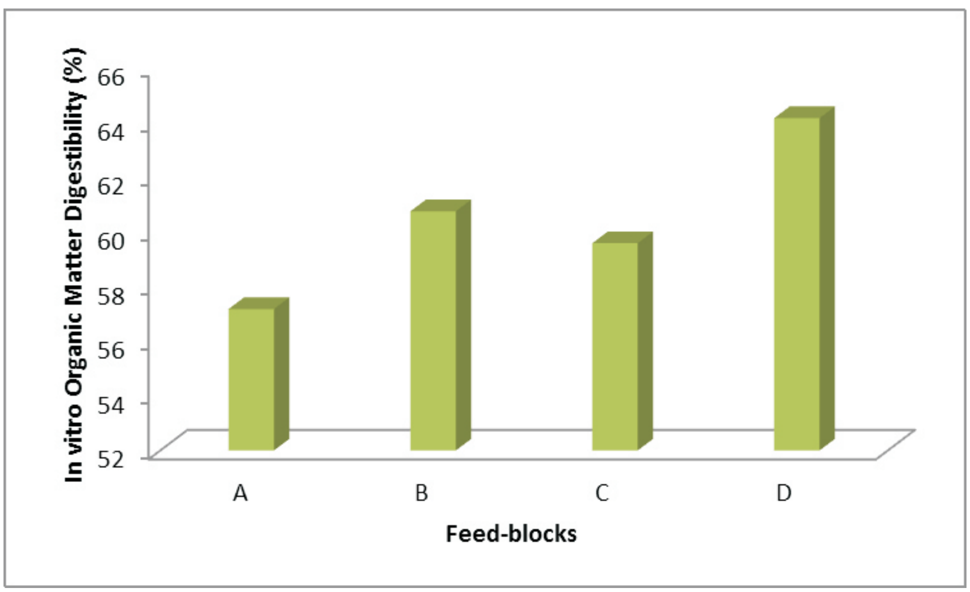

Figure1: In vitro organic matter digestibility (%)

rumen. Urea is a non-protein nitrogen source which plays active role in supplying ammonia to rumen microbes for feed degradation which results in microbial protein and methane formation. The result of methane production in the present study was however higher than the report of Isah (2011) when some tropical grasses were degraded *in vitro*. The high IVOMD result recorded for UMFB D without cement as binder might be as a result of the binding effect of cement on the component of other UMFB containing cement in the other treatments, which might lead to slow release of nutrients for thorough degradation of the organic matter component of the feed. However, when cement was absent in UMFB D, other feed ingredients were not closely bound together to prevent their release for microbial degradation. Also, rapidly fermentable carbohydrates yielded relatively higher propionate as compared to acetate (Makkar, 2000) was the case in UMFB D which had highest propionic acid concentration. UMFB B with cement and clay as binders yeilded highest concentration of the various volatile fatty acids determined except propionic acid while UMFB A with only cement as binder yeilded least quantity of volatile fatty acids. This indicates that combining cement with other binders such as clay and cassava starch could be more effective because microbes use ammonia and energy more effectively if they are released gradually to maintain stable rumen pH for optimum microbial actvity. However, UMFB A had least methane production. The pH results ($p>0.05$) observed in this study is lower than that reported by Foiklang et al. (2001) for rumen fermentation of swamp buffalo. However, the pH values are above 5.5 which cause depression to protozoa population in the rumen due to acid intolerance. NH_3-N concentration recorded in the present study was higher than the report of Molina et al. (2009), but lower than the observation of Thu, (2001) and Koakhunthod et al. (2001). This might be due to variation in the formulation of the feed-blocks.

Conclusion

The study showed that the various feed - blocks can serve as a dependable supplement which may improve rumen fermentation efficiency during dry season and period of scarcity for ruminants and are therefore recommended as feed-blocks for ruminants on a low-quality forages. Supplementation with UMFB can improve the utilization of low quality roughages by satisfying the requirement of the rumen micro-organisms, by creating better fermentation of fibrous material and increasing production of microbial protein and volatile fatty acids. UMFB B having cement and clay as binders yeilded highest concentration of of the various volatile fatty acids determined except propionic acid while UMFB A with only cement as binder yeilded least quantity of Volatile fatty acids. However, UMFB A had least methane production. It is recommended that the various UMFB be fed to animal to assess their performance.

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