



Proximate analysis and *in vitro* gas production of diets containing sweet potato peel supplemented with urea-sulphur mix

*M.T. Ayankoso^{1,2}, A.J. Owolabi¹, O.O. Ogunfowora¹, O. A. Adesunloye¹, V. Olajire¹, O.A. Ganiyu³ and A.O. Yusuf¹

¹Department of Animal Production and Health, Federal University of Agriculture, Abeokuta, Nigeria

²Department of Animal Science, Adekunle Ajasin University, Akungba-Akoko, Nigeria

³Department of Wildlife and Ecotourism, Forestry Research Institute of Nigeria, Ibadan, Nigeria

Abstract

This research was carried out to investigate proximate and *in-vitro* gas production of sweet potato (*Ipomoea batatas*) peels supplemented with urea-sulphur mix fed to sheep. Four iso-nitrogenous and iso-caloric diets (T1, T2, T3 and T4) were formulated which contained sweet potato peels with varying levels of urea and sulphur; T1 (control), T2 (0.2% Sulphur), T3 (0.3% urea), T4 (0.1% sulphur + 0.15% urea). The rumen content was collected from the experimental sheep for the *in vitro* gas production and fermentation kinetics, which were estimated on 3-hourly bases for 48 hours. Data were collected on post incubation gas production, fractional rate of gas production, lag time, dry matter digestibility and proximate composition of the diets. Data collected were subjected to one-way Analysis of Variance at 5% level of probability. Chemical composition of the diets and *in vitro* gas production were not influenced ($P>0.05$) by urea-sulphur mix supplementation. The dry matter digestibility of the diets was significantly ($P<0.05$) highest in the control while least was from diet with 0.1% sulphur and 0.15% urea. The volumes of carbon dioxide gas obtained were significantly ($P<0.05$) different with the highest and least volumes in control and diet with 0.1% sulphur and 0.15% urea mixture, respectively. The study showed that sole supplementation of urea and sulphur improved dry matter digestibility while urea-sulphur combination reduced carbon dioxide production.

Article Information

Keywords: Sweet potato peel, *In vitro* gas production, methane, lag time

*Corresponding author

M.T. Ayankoso
mtayankoso@gmail.com

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INTRODUCTION

Animal protein consumption has been described as awfully low in Nigeria because of the high price of animal products which is the outcome of the high cost of feed ingredients (Sobayo *et al.*, 2013). This necessitates the use of various alternatives to improve the ruminants' diets by diversifying into the use of non-conventional feed ingredients (Taiwo *et al.*, 2005; Ekenyem and Madubuike, 2006; Aye and Adegun, 2010). In Nigeria, ruminants gradually increase weight in the wet period and speedily lose it in the dry season (Babayemi and Bamikole, 2006). This is because grasses and roughages make up above 75% ration for ruminants, which are periodical and of little nutritional values (Babayemi, 2006). Fluctuation in the quantity and quality of forages and other feed components is the main restriction in livestock management. Also, due to the increase in human population, foraging areas are declining rapidly to cater for the immediate necessities of man in terms of food and/or commercial crops (Anbarasu *et al.*, 2004).

Sweet potato (*Ipomoea batatas*) peels are alternative feed sources that can be used to feed

cattle, sheep and goat due to their abundance and low price (Akoetey *et al.*, 2017). However, the peels contain high sugar level, which is an important energy source in ruminant diets (Malik *et al.*, 2011). Research into the use of cheaper agro-industrial by-products and waste materials such as Irish potato peels, cassava peels, corn cobs, maize offal, poultry droppings and abattoir waste at various levels of inclusion for small ruminants has been strengthened to determine their effectiveness in terms of growth and production (Fasae *et al.*, 2012; Kalio *et al.*, 2013).

However, sweet potato peels are deficient in nitrogen and low in crude protein (4.6% DM) (Kohn *et al.*, 1976). Urea has been extensively used in ruminant nutrition as a source of non-protein nitrogen. So, supplementation of urea which is non-protein nitrogen that can be rapidly hydrolyzed to ammonia in the rumen to improve the nutrition of low quality agricultural by-products, will be of immense benefits (Papadopoulos *et al.*, 2001). Also, sulphur is a macro element for ruminants in dietary protein supplementation, which is a useful method to meet their dietary necessities (Qi *et al.*, 1994a).

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Numerous researches have shown that sulphur inclusion advances the synthesis of ruminal microbial protein, and cellulose digestion (Reis and Schinckel, 1963; Bray and Hemsley, 1969; Qi *et al.*, 1994b).

Basal feeds deficient in major nutrients can be supplemented with either grain concentrate or forage legumes in ruminant animals, but this is unaffordable to many smallholder farmers and ruminant rearers. Also, Crop by-products available in many rural villages of Nigeria are not efficiently utilized by livestock farmers as potential feed resources for feeding livestock (Ayuk *et al.*, 2011). So, the need to supplement protein and energy with other essential nutrients to improve the productivity of small ruminants becomes imperatives (Mendietta-Araica *et al.*, 2011).

The *in vitro* gas production method is a comparatively simple process of assessing feed ingredients because various samples can be incubated and examined simultaneously. This method has been applied successfully in feed assessment, comprising determination of organic matter digestibility, metabolizable energy of feeds and kinetics of their fermentation, anti-nutritive factors and rumen modifiers by monitoring microbial change in the rumen. Above 50% of the ruminant feed intake pass through gastrointestinal tracts undigested and unutilized, and are excreted as faeces, urine and gases. The *in vitro* gas formation technique can be used to scrutinize animal waste constituents that influence the surroundings and develop appropriate modifications (Getachew *et al.*, 2001). Therefore, there is a need to investigate the proximate and *in vitro* gas production of sweet potato peels diet supplemented with the urea-sulphur mix so as to determine its utilization.

MATERIALS AND METHODS

Experimental site

This experiment was conducted in the laboratory of the Department of Pasture and Range Management, College of Animal Science and Livestock Production, Federal University of Agriculture, Abeokuta, located on Latitude 7° 15' N, Longitude 3° 26' E with an altitude of 76 m above sea level (Google Earth Map, 2019). It is situated in the derived savannah zone of South-Western Nigeria, which has a prevailing tropical climate, average annual rainfall and temperature of 1 037 mm and a temperature of 34.7°C respectively (AccuWeather, 2019) and a yearly

average humidity of 83 % (Meteorological Station, FUNAAB).

Experimental materials

Fresh Sweet potato (*I. batatas*) peels were acquired from some processing firms in Abeokuta and its environment. The peels were air-dried, milled and bulked for analysis. The urea was obtained from the Ministry of Agriculture, Asero, Abeokuta, while sublime sulphur was purchased at the local market around Abeokuta.

Experimental Animals and Management

Twenty West African Dwarf sheep weighing between 10-12kg were purchased from a local market around Ogun state. The animals on arrival were confined in the quarantine pen which had been previously washed and disinfected using Iodophor (Azintol) solution. The animals were treated against internal and external parasites using Ivomec® at 1ml to 25kg body weight and pen strep (a broad spectrum antibiotics) at 2ml to 25kg body weight for three days. They were allocated into individual pens having a raised slatted floor. Clean water was given *ad libitum* throughout the period of the experiment.

Experimental diet

The bulked sweet potato peels were mixed with other ingredients- palm kernel cake (PKC), wheat offal, oyster shell, bone meal and table salt to

Table 1: Ingredient compositions of experimental diets

Ingredients (%)	T1	T2	T3	T4
Maize	15.00	15.00	15.00	15.00
PKC	20.00	20.00	20.00	20.00
Sweet potato peel	30.00	30.00	30.00	30.00
Wheat offal	30.00	30.00	30.00	30.00
Oyster shell	2.00	2.00	2.00	2.00
Bone meal	2.00	2.00	2.00	2.00
Salt	1.00	1.00	1.00	1.00
Urea	-	-	0.30	0.15
Sulphur	-	0.20	-	0.10
Total	100	100	100	100
Calculated nutrients				
Crude Protein (%)	11.80	11.80	11.80	11.80
Metabolizable Energy (MJ/kg)	13.42	13.42	13.42	13.42

T1: sweet potato peel based concentrate (SPPBC), control

T2: SPPBC + 0.2% Sulphur

T3: SPPBC + 0.3% urea

T4: SPPBC + 0.1% sulphur + 0.15% urea

compound concentrate diets (Table 1). The concentrated diets were formulated to be iso-nitrogenous and iso-caloric with varying levels of urea-sulphur mixture designated as treatment 1 (T1) which is the control diet: sweet potato peel based concentrate (SPPBC), Treatment 2 (T2): SPPBC + 0.2% Sulphur, Treatment 3 (T3): SPPBC + 0.3% urea and Treatment 4 (T4): SPPBC + 0.1% sulphur + 0.15% urea. The supplements (sole and mixture) were added to already mill diets and thereafter mixed thoroughly.

Experimental Design

The twenty West African Dwarf sheep were divided into four groups of five animals per group where each animal was an experimental animal. The animals were randomly assigned into four experimental diets in a completely randomized design. The animals were fed at 4% of their body weight which was adjusted weekly for twelve weeks and the ruminal fluid was collected from the animals for *in vitro* gas production.

Proximate analysis

The crude protein content of the diets was determined using the automated Kjeldahl method (AOAC, 2000). Dry matter was calculated by drying the samples at 65 °C for 48 hours in an oven, while ash was measured by burning further at 500 °C for 24 hours in a muffle furnace. Dried Samples were also analyzed for ether extract as described by (AOAC, 2000). Dry matter digestibility was determined through two consecutive digestion stages. During the first stage, plant materials were incubated in an anaerobic condition with rumen microbes for 48 hours at 39 °C while in the second stage, acid pepsin digestion was carried out anaerobically at 39 °C for 24 hours which was followed by 72 hours' incubation, the residual plant materials were collected and oven dried at 106 °C for 12 hours.

In vitro gas production

Ruminal fluid was obtained from the West African Dwarf sheep with a suction pipe before morning feed and put into a thermo flask that had been previously warmed to a temperature of 39 °C as described by Babayemi *et al.* (2009). The incubation was conducted following standard procedures using 120 ml calibrated syringes in three replicates maintained at a temperature of 39 to 40 °C (Menke and Steingass, 1988). About 200 mg of the feed samples were then weighed into the syringe and added to 30 ml of inoculums

containing cheesecloth strained rumen liquor and buffer (comprising 9.8 g NaHCO₃ + 2.77 g, Na₂HPO₄ + 0.57 g KCl + 0.47 g NaCl + 0.12 g MgSO₄·7H₂O + 16 g CaCl₂·2H₂O) per litre. Ruminal fluid and buffer solution were mixed in the ratio 1 to 4 (v/v) under continuous flushing with CO₂. Using 50 ml calibrated plastic syringe, 30 ml of inoculum was dispensed into the substrate through the silicon tube. The plunger was pushed upward by pushing the inoculums to the syringe's tip for the total elimination of air. A metal clip fastened the silicon tube in the syringe to avert leakage of gas. The quantity of gas produced was determined on 3-hourly bases (3, 6, 9, 12, 15, 18, 21, 24, 27, 30, 33, 36, 39, 42, 45, and 48 h). The volume of gas formed was then determined by calculating the interval between the top of the piston and the liquid in the syringe.

Measurement of methane gas produced

After 24 hours of incubation, 4 ml of 10M NaOH was introduced into the syringes through the silicon tube and properly shaken for proper mixing and absorption of CO₂ gas by addition of NaOH solution according to Fievez *et al.* (2005) procedure. After all the CO₂ gas might have been absorbed, the volume of methane left in the syringes was recorded in millilitre (ml). The average volume of gas formed from the gaps was subtracted from the volume of gas per sample to estimate the net gas produced for each sample.

Statistical analysis

Data obtained for proximate and *in vitro* studies were analyzed using Analysis of Variance (ANOVA) as described by SAS (1999), while the treatment means were separated using the Tukey procedure as contained in the software package.

RESULTS

The proximate analysis of diets containing sweet potato peel supplemented with the urea-sulphur mix is shown in Table 2. The results showed that all proximate parameters were not significantly ($P > 0.05$) different across the diets. However, diet with 0.3% urea had numerical higher value of crude protein, crude fibre, fat, ash and dry matter with 17.96, 4.7, 9.51, 6.50 and 92.03%, respectively. Moisture content ranged from 8.22 to 10.53% while nitrogen free extract ranged from 61.34 to 66.75%.

The *in vitro* total gas production from the degradation of substrates containing sweet potato peel supplemented with urea-sulphur is presented in Table 3. Results obtained across treatments

Table 2: Proximate compositions of diets containing sweet potato peel supplemented with urea-sulphur combination (%)

Parameter	T1	T2	T3	T4	SEM*	P. value
Moisture	8.22	8.62	7.98	10.53	0.33	0.21
Dry matter	91.78	91.39	92.03	89.47	0.33	0.11
Crude protein	16.40	15.08	17.96	14.29	0.47	0.23
Ash	6.24	6.17	6.50	5.89	0.08	0.30
Fat	9.39	9.16	9.51	8.85	0.12	0.17
Crude fibre	4.59	4.41	4.70	4.23	0.06	0.21
Nitrogen free extract	63.40	65.20	61.34	66.75	0.70	0.34

*SEM: Standard error of mean

T1: sweet potato peel based concentrate (SPPBC), control

T2: SPPBC + 0.2% Sulphur

T3: SPPBC + 0.3% urea

T4: SPPBC + 0.1% sulphur + 0.15% urea

Table 3: *In vitro* gas production (ml) of diets containing sweet potato peel supplemented with urea-sulphur mix at 3-hour interval

Parameter (hrs)	T1	T2	T3	T4	SEM*	P – Value
3	2.00	1.00	2.50	2.50	0.42	0.47
6	6.00	3.00	7.00	8.50	0.85	0.17
9	11.00	8.00	12.50	13.50	1.30	0.42
12	17.00	11.00	19.00	19.00	1.54	0.19
15	21.00	15.00	25.00	23.50	1.90	0.15
18	25.50	20.00	28.50	29.50	1.98	0.21
21	29.50	23.00	32.00	33.00	2.05	0.25
24	31.50	27.50	35.00	36.50	2.12	0.43
27	35.00	30.00	43.50	39.50	2.40	0.32
30	37.00	32.00	40.00	42.50	2.48	0.49
33	38.50	34.00	42.00	45.50	2.53	0.55
36	40.00	35.00	42.00	45.50	2.60	0.62
39	41.00	37.50	44.00	46.50	2.63	0.69
42	41.00	38.50	45.00	48.00	2.71	0.69
45	42.00	41.00	45.00	48.00	2.65	0.83
48	43.00	41.50	46.00	48.00	2.73	0.83

*SEM: Standard error of mean

T1: sweet potato peel based concentrate (SPPBC), control

T2: SPPBC + 0.2% Sulphur

T3: SPPBC + 0.3% urea

T4: SPPBC + 0.1% sulphur + 0.15% urea

were not significant ($P>0.05$), and there was a direct relationship between gas production and incubation time.

The *in vitro* gas characteristics (volume of carbondioxide gas produced in time (t), fractional rate of gas production, lag time, methane

production and dry matter digestibility) of diets containing sweet potato peel supplemented with urea-sulphur mix after 48 hours of incubation are presented in Table 4. Volume of carbondioxide gas produced with incubation time was highest ($P<0.05$) in diet with 0.2% urea (46.53 ml), followed by diet containing 0.3% sulphur (27.65

ml) and the least (19.16 ml) in diet with 0.1% sulphur and 0.15% urea. There was significant ($P < 0.05$) difference in the dry matter digestibility with highest (38.50%) and least (31.00%) values in control and diet with 0.1% sulphur and 0.15% urea, respectively. Fractional rate of gas production, lag time and methane production were similar ($P > 0.05$) across the treatments.

for all the diets, this agrees with Adeyosoye *et al.* (2010) where in vitro gas degradation of sweet potato (*Ipomea batatas*) and wild cocoyam (*Colocasia esculenta*) peels was carried out. High volume of gas observed for all the diets, with the highest numerical value obtained in urea-sulphur mixture at the end of 48 hours of incubation might be attributed to high dry matter and fibre

Table 4: Post incubation parameters of diets containing sweet potato peels supplemented with urea-sulphur mix at 48 hours incubation period

Parameter	T1	T2	T3	T4	SEM*	P-value
Volume of CO ₂ produced in time (t)	24.28 ^c	46.53 ^a	27.65 ^b	19.16 ^d	3.65	0.02
Fractional rate of gas production (ml/hr)	0.36	0.17	0.36	0.50	0.07	0.56
Lag time (hr)	2.53	3.84	2.43	1.48	0.37	0.23
Methane (ml)	22.00	18.50	17.00	22.50	1.09	0.10
Dry matter digestibility (%)	38.50 ^a	37.50 ^b	37.00 ^b	31.00 ^c	0.24	0.01

^{a,b} Means on the same row having different superscripts are significantly ($P < 0.05$) different

*SEM: Standard error of mean

T1: sweet potato peel based concentrate (SPPBC), control

T2: SPPBC + 0.2% Sulphur

T3: SPPBC + 0.3% urea

T4: SPPBC + 0.1% sulphur + 0.15% urea

DISCUSSION

The proportion of crude protein obtained in this study falls within the values reported by Ibrahim and Olaniyi (2018) where sweet potato peel was used to replace maize in diets. Therefore, the protein content of the diets is capable of supporting the performance and metabolic activities in animals which agrees with the report of Johnson-Delancy (2006). The crude protein obtained in this study when supplemented with mixture of sulphur and urea is within the range proposed by Pugh (2020) that it is sufficient of meeting the nutritional requirement of sheep. The dry matter obtained across the diets was higher compared to the results reported by Adeyosoye *et al.* (2010) and Okoruwa and Bamigboye (2015). The variation could be as a result of supplementation of urea and sulphur in this present study.

The gas produced by different diets increased with increasing hours of incubation, but gradually stabilized at 48 hours of incubation. The gas production trends of the diets are in consonance with the observation of Akinfemi *et al.* (2009). The production of biogas (CO₂ and CH₄) at the third hour of incubation suggests that degradation of the substrates have commenced

contents, which is similar to the study carried out by Adeyosoye *et al.* (2010) and Onaleye *et al.* (2018) where fibres supplemented with additives produced higher volume of gas compare to those without. It is also similar to the findings of Okoruwa and Bamigboye (2015), who included sweet potato peel and cashew nut shell with *Ocimum gratissimum* leaves in the diet of West African Dwarf goat. The study also confirmed that, sweet potato peels supplemented with sulphur and urea mixture recorded the highest gas volumes after 48 hours of incubation.

Similarity in the content of methane production could be as a result of iso-nitrogenous nature of the diets which is in line with the report of Onaleye *et al.* (2018) whereby low methane production was reported, which might be due to the presence of low soluble protein in the diets used. This may also be attributed to the fact that, the diet mixtures have fermentable and readily degradable cell wall components, which would increase the substrates available to cellulolytic microbes with a subsequent increase in the population of microorganisms (Van Soest, 1982; Adebawale, 1994). Quantum of methane produced within the shortest lag time, coupled with the fractional rate of gas production, in the diet supplemented with combination of urea and

sulphur connotes that, at long term, more methane would be produced which can be of high advantage when used in energy generation firms. Also, the least production of carbon dioxide generated in diet with urea-sulphur mixture supplementation predicts that animals fed with it are in good state of well-being since low level of carbon dioxide will be retained in their blood components and efficient transport system of dissolved metabolic materials will be enhanced by the help of oxy-haemoglobin which invariably stimulate better soundness of animals through effective circulatory system (Donohoe et al., 2021).

This study showed that methanogenesis reduced carbon dioxide production in urea-sulphur mix diet which is suggested to have inhibited dry matter digestibility during rumination which is in line with the report of Ellis et al. (2008) who proposed that methane reduces carbon dioxide production by reducing fermentation of plant materials (mostly carbohydrates) into volatile fatty acid (VFA). This study agrees with Allard (2009) who reported an inverse relationship between digestibility of diets and methane production in ruminants.

This study concluded that for efficient performance in sheep production, there is no need for urea-sulphur supplementation since the results are similar. In addition, the extent of the gas produced within the incubation period suggests that the use of urea-sulphur mix supplementation in sweet potato peels based diets improves rumen microbial interaction with a better gas production characteristic than the use of only cassava peels. By implication, this study confirmed that fermentation of sweet potato peels when supplemented with urea and sulphur combination is an inexpensive technique to generate methane gas as energy source for home and industrial manufacturing usage, and invariably cleanse surroundings from pollution or contamination and the excess of methane produced can be used in livestock feed mill to generate energy which can be used to power gas turbine engines. Diet supplemented with combination of urea and sulphur better enhanced methane production. Further *in vitro* and *in vivo* study should be carried out using higher levels of inclusion to determine the effects of combination of urea and sulphur on dry matter digestibility and methane production in ruminants.

Conflicts of interest: The authors declare no

conflicts of interest.

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