

***In-vitro* gas production of two cultivars of *Cajanus cajan* (Brown Ogbomoso Indigenous and Ex Kaduna) with cassava (*Manihot specie*) peel as affected by different inclusion levels**

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Abstract

The efficiency in rumen fermentation of leguminous plants are yet to be fully established. An experiment was conducted to determine the influence of two varieties of pigeon pea (ex-kaduna and Brown Ogbomoso Indigenous: BOI) with cassava peel at different inclusion levels (0%, 25%, 50%, 75% and 100%) on *in vitro* gas production characteristics, organic matter digestibility, metabolisable energy and short chain fatty acids at 24, 48 and 72 hours incubation. Standard dose of 20mg/200mg of each of the treatment meals to that of standard maize substrates sample was used and investigated in a completely randomised design as follows: 100% *Cajanus cajan* (ex-kaduna) + 0% *Manihot specie* (T1); 75% *Cajanus cajan* (ex-kaduna) + 25% *Manihot specie* (T2); 50% *Cajanus cajan* (ex-kaduna) + 50% *Manihot specie* (T3); 25% *Cajanus cajan* (ex-kaduna) + 75% *Manihot specie* (T4); 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie* (T5); 100% *Cajanus cajan* (BOI) + 0% *Manihot specie* (T6); 75% *Cajanus cajan* (BOI) + 25% *Manihot specie* (T7); 50% *Cajanus cajan* (BOI) + 50% *Manihot specie* (T8) and 25% *Cajanus cajan* (BOI) + 75% *Manihot specie* (T9). At 24 hours of incubation, 25% *Cajanus cajan* (BOI) with 75% *Manihot specie* produced the highest gas (37.50). At 48 hours and 72 hours, 0% *Cajanus cajan* (BOI) with 100% *Manihot specie* produced the highest gas (42.50 and 50.50 respectively) while 100% *Cajanus cajan* (BOI) produced the lowest gas at 24 hours, 48 hours and at 72 hours. At 24 hours of incubation, T4 and T5 consistently increased the potential gas produced (*a+b*), gas produced from the potentially degradable fraction (*b*) and volume of gas produced at time (*Y*) but decreased incubation time (*t'*). Treatment 6 reduced *a+b*, *b*, *Y* but increased *c* and *t*. The *t'* was lower in T1 compared to T6. Treatment 4 was higher than T9 with respect to *'b'*. At 48 hours of incubation, T5 increased *a+b*, *b*, *c* and *Y*. T6 had reduced *a+b*, *b*, *Y*. The *t'* was reduced in T6 compared to T1. T3 was higher in terms of *a+b* and *b* compared to T8. At 72 - hour, T5 increased *a+b*, *b*, *c* and *Y*. Treatment 6 reduced *a+b*, *b*, *c* and *Y*. However, the differences obtained were within the same time of incubation. The *a+b*, *b* and *Y* were higher in T1 than T6. The *'b'* was higher in T4 compared to T9. Metabolisable energy (ME), organic matter digestibility (OMD) and short chain fatty acids (SCFA) were significantly ($P < 0.05$) affected across the treatments. At 24 hours incubation, T4 consistently had higher ME, and SCFA compared to other treatments. This was not sustained with further incubation to 48 hour. However, at 72 hours of incubation higher ME (6.21), OMD (52.81) and SCFA (0.62) was obtained in T5. It was concluded that rumen microbes degraded *Cajanus cajan* (Ex-Kaduna) and *Manihot specie* more efficiently when incubated at the ratio of 25:75 for 24 hours compared to Brown Ogbomoso Indigenous. However, a 50:50 combination of Brown Ogbomoso Indigenous with *Manihot specie* showed promising results.

Keywords: Rumen fermentation, *in vitro* gas production, pigeon pea, cassava peel, rumen microbes.

Production de gaz *in vitro* de deux cultivars de *Cajanus cajan* (Brown Ogbomoso Indigeneus et Ex Kaduna) avec peau de manioc (espèce Manihot) affectée par différents niveaux d'inclusion



Résumé

*L'efficacité de la fermentation ruminale des légumineuses n'est pas encore totalement établie. Une expérimentation a été menée pour déterminer l'influence de deux variétés de pois cajan (ex-kaduna et Brown Ogbomoso Indigeneus : BOI) avec des écorces de manioc à différents niveaux d'inclusion (0 %, 25 %, 50 %, 75 % et 100 %) sur caractéristiques de production de gaz in vitro, digestibilité de la matière organique, énergie métabolisable et acides gras à chaîne courte à 24, 48 et 72 heures d'incubation. Une dose standard de 20 mg/200 mg de chacun des repas de traitement par rapport à celle de l'échantillon de substrats de maïs standard a été utilisée et étudiée dans un plan complètement randomisé comme suit : 100 % *Cajanuscajan* (ex-kaduna) + 0 % espèce *Manihot* (T1) ; 75% *Cajanuscajan* (ex-kaduna) + 25% espèce *Manihot* (T2) ; 50 % *Cajanuscajan* (ex-kaduna) + 50 % espèce *Manihot* (T3) ; 25% *Cajanuscajan* (ex-kaduna) + 75% espèce *Manihot* (T4) ; 0% *Cajanus cajan* (ex-kaduna) + 100% espèce *Manihot* (T5) ; 100 % *cajanuscajan* (BOI) + 0 % espèce *Manihot* (T6) ; 75 % *Cajanus cajan* (BOI) + 25 % espèce *Manihot* (T7) ; 50% *Cajanuscajan* (BOI) + 50% espèce *Manihot* (T8) et 25% *Cajanuscajan*(BOI) + 75% espèce *Manihot* (T9). À 24 heures d'incubation, 25 % de *Cajanuscajan* (BOI) avec 75 % d'espèce *Manihot* ont produit le gaz le plus élevé (37,50). À 48 heures et 72 heures, 0% *Cajanuscajan* (BOI) avec 100% d'espèce *Manihot* a produit le gaz le plus élevé (42,50 et 50,50 respectivement) tandis que 100% *Cajanuscajan* (BOI) a produit le gaz le plus bas à 24 heures, 48 heures et à 72 heures. À 24 heures d'incubation, T4 et T5 ont systématiquement augmenté le gaz potentiel produit (a+b), le gaz produit à partir de la fraction potentiellement dégradable (b) et le volume de gaz produit au temps (Y) mais ont diminué le temps d'incubation ('t') . Le traitement 6 a réduit a+b, b, Y mais a augmenté c et t. Le « t » était inférieur en T1 par rapport à T6. Le traitement 4 était supérieur au T9 en ce qui concerne « b ». À 48 heures d'incubation, T5 a augmenté a+b, b, c et Y. T6 avait réduit a+b, b, Y. Le « t » était réduit dans T6 par rapport à T1. T3 était plus élevé en termes de a+b et b par rapport à T8. A 72 heures, T5 a augmenté a+b, b, c et Y. Le traitement 6 a réduit a+b, b, c et Y. Cependant, les différences obtenues étaient dans le même temps d'incubation. Les a+b, b et Y étaient plus élevés en T1 qu'en T6. Le « b » était plus élevé dans le T4 que dans le T9. L'énergie métabolisable (EM), la digestibilité de la matière organique (OMD) et les acides gras à chaîne courte (AGCC) ont été significativement ($P < 0,05$) affectés à travers les traitements. À 24 heures d'incubation, le T4 présentait systématiquement un ME et un SCFA plus élevés que les autres traitements. Ceci n'a pas été maintenu avec une incubation supplémentaire jusqu'à 48 heures. Cependant, à 72 heures d'incubation, une ME (6,21) plus élevée, une OMD (52,81) et une SCFA (0,62) ont été obtenues en T5. Il a été conclu que les microbes du rumen dégradent les espèces *Cajanuscajan* (Ex-Kaduna) et *Manihot* plus efficacement lorsqu'ils étaient incubés dans un rapport de 25:75 pendant 24 heures par rapport à l'indigène Brown Ogbomoso. Cependant, une combinaison 50:50 de Brown Ogbomoso Indigeneus avec l'espèce *Manihot* a montré des résultats prometteurs.*

Mots-clés : Fermentation du rumen, production de gaz *in vitro*, pois cajan, écorce de manioc, microbes du rumen.

Introduction

Human population growth rate in the developed countries has somewhat been established but populations are still increasing rapidly in virtually all developing countries of Africa, Asia and Latin America (Allen, 1983). Most of the World's population is fed by the output of small farms in the developing countries (Galladio, 1984). The farms continue to get smaller with increase in human population pressure. To maximize food production in developing countries, all reasonable options must be considered. Ruminants in the tropics especially cattle, goats and sheep obtain most of their nutrients from herbage growing on unimproved native pasture which often grow fibrous and are of low nutritive value particularly, in the dry season. Unfortunately, these native pastures are the most widely available low cost feeds for ruminants in the tropics (Tchinda *et al.*, 1993). Browse species, because of their resistance to drought, extreme heat, salinity, alkalinity, drifting sand, grazing and repeated cutting are the major feed (forage) resources during the dry season (Fagg and Stewart, 1994) in Ajibola (2000). Legumes are important protein source in developing countries coupled with the relative high cost of conventional ingredients, it became increasingly necessary that the under-utilized protein sources be sought so as to widen the choice of available protein sources for animal feed. Among the suggested alternative is pigeon pea (*Cajanus cajan*). Pigeon pea (*Cajanus cajan*) is a pulse perennial legume and a shrub that could be rain-fed to be available all year round as a principal source of protein in animal diet. Ajayi and Babayemi (2008) reported that legumes are high in methane production which is an indication of inefficient fermentation and so require energy supplement for better utilization in order to sustain livestock production. Cassava peel could be a necessary and

economical energy source to complement the nutrient in pigeon. Therefore, there is need to determine the influence of two varieties of pigeon pea (ex-kaduna and Brown Ogbomoso Indigenous) with cassava peel as affected by different inclusion levels (0%, 25%, 50%, 75% and 100%) on rumen fermentation characteristics using *in vitro* gas production technique at 24, 48 and 72 hours incubation as carried out in this study.

Materials and methods

Site selection and seed collection

The selected Indigenous varieties of *Cajanus cajan* which include ex-Kaduna and Brown Ogbomoso Indigenous were planted at a spacing of 50cm X 75cm on the experimental plot of Ladoke Akintola University of Technology teaching and research farm, Ladoke Akintola University Of Technology. The *in-vitro* digestibility experiment was carried out at the Ruminant Laboratory, Department of Animal Science, University of Ibadan. Seed germination test was carried out using Petri-dishes, litmus paper and distilled water for germination test. The two selected varieties were further tested by picking out the rough coated and seeds that are unsuitable for germination. The selected seeds were poured in water and the seeds that floated were discarded while those that sank were planted.

Sample preparation

The foliage of *Cajanus cajan* was harvested at 8 weeks and cassava peel was collected. Both were oven-dried to a constant weight. The samples were milled to pass through 1mm mesh sieve and stored at room temperature for subsequent laboratory analysis.

Incubation was carried out with rumen liquor obtained from eight matured West African dwarf goats and Sheep offered standard diet.

Experimental design and treatments

A dose of (20mg/200mg substrates

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samples) of each of the meals of the treatments was added to the standard maize substrate samples and investigated in a completely randomised design as follows:

- T1: 100% *Cajanus cajan* (ex-kaduna) + 0% *Manihot specie*
T2: 75% *Cajanus cajan* (ex-kaduna) + 25% *Manihot specie*
T3: 50% *Cajanus cajan* (ex-kaduna) + 50% *Manihot specie*
T4: 25% *Cajanus cajan* (ex-kaduna) + 75% *Manihot specie*
T5: 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie*
T6: 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie*
T7: 75% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 25% *Manihot specie*
T8: 50% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 50% *Manihot specie*
T9: 25% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 75% *Manihot specie*

In-vitro gas production

Rumen liquor was taken by suction method before morning feeding from four West African Dwarf goat, one red Sokoto breed and three West African dwarf sheep fed with standard layers concentrate and *Panicum maximum* (Ntchisi) at a ratio of 40:60 respectively. Rumen liquor was collected into a pre-warmed thermo flask, strained, using four layers of cheese cloth into a plastic container. All handling was carried out with continuous flushing of CO₂. The well mixed CO₂ flushed rumen liquor was added to the buffered solution (NaHCO₃ + Na₂HPO₄ + KCl + NaCl + MgSO₄ .7H₂O + CaCl₂ .2H₂O) which was maintained at a temperature of 35°C-39°C at a ratio of 1:2. Buffered rumen liquor mixture (30ml) was dispensed into each syringe containing the treatments and standard maize substrate. The syringes were immediately placed in an incubator at 39°C and starting time for incubation recorded. Two syringes

containing 30ml inoculum serves as blanks. Syringes were turned carefully in order to ensure complete mixing of incubated contents. Rate of gas production was determined every 3 hours until 72 hours post inoculum to predict the total gas production. At the end of 72 hours termination of incubation, final gas volumes were recorded.

After termination, 10 molar solution of NaOH was introduced through the silicon. Cumulative *in vitro* gas production data were fitted to the exponential equation $Y=a+b(1-e^{-ct})$ described by Ørskov and McDonald (1979) as adopted by Menke and Steingass (1988) where Y=volume of gas produced at time t, a=intercept (gas produced from the soluble fraction), b=gas produced from the potentially degradable fraction, (a+b) = potential gas produced, c=rate of degradation of b, t=incubation time. Metabolizable energy (ME, MJ/Kg DM) and organic matter digestibility (OMD g/100g) were estimated as established (Menke and Steingass 1988) and short chain fatty acids (SCFA) was calculated as reported by Getachew *et al.* (1999).

$$ME = 2.20 + 0.136GV + 0.057CP + 0.0029CF$$

$$OMD = 14.88 + 0.88GV + 0.45CP + 0.651XA$$

$$SCFA=0.0239GV+0.0601$$

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

Chemical analysis

The proximate analysis was carried out according to method of AOAC (1995). Neutral detergent fibre (NDF), Acid detergent fibre (ADF) and Acid detergent lignin (ADL) was determined using the method described by Van Soest *et al.* (1991). Hemicellulose was estimated as the difference between NDF and ADF, and cellulose estimated as the difference between ADF and ADL.

Statistical analysis

Data obtained were subjected to one-way analysis of variance (ANOVA) of SAS (2008), the means were separated using Duncan multiple range test at $p < 0.05$. Data were obtained from three replicate determinations.

Results and discussion

The result of chemical composition of samples used (%as fed) are shown in Table 1 below. The crude protein content obtained

for both ex-Kaduna and Brown Ogbomoso Indigenous falls within the range of 10-18% reported by Akinola *et al.* (1994) but lower than the 7.1% reported by Duke (1981). More so, it was above the 8-10% recommended by Gatenby (2002) for growing lambs and minimum of 7% requirements recommended for ruminants in the tropics (Minson, 1980). Dry matter content of ex-Kaduna and Brown Ogbomoso Indigenous was 33.08 and 30.58 respectively and higher than the 29.6% reported by Duke (1981).

Table 1: Chemical composition of samples used (%as fed)

Samples	Protein	Fat	Ash	Crude Fibre	Moisture	NFE	NDF	ADF	Lignin
<i>Manihot specie</i>	4.68	2.43	16.34	21.87	69.88	43.76	52.88	40.84	0.46
<i>Cajanus cajan</i> (Brown Ogbomoso Indigenous)	17.42	2.81	16.15	14.34	69.42	49.34	52.08	40.65	0.61
<i>Cajanus cajan</i> (ex-Kaduna)	16.30	2.35	16.52	14.98	66.92	49.96	50.59	37.11	0.40

Manihot spp incubated at 24hrs is presented in Table 2. Treatment with 25% *Cajanus cajan* (ex kaduna) + 75% *Manihot specie* and 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie* consistently increased the potential gas produced, gas produced from the insoluble fraction and total volume of gas produced but decreased the time of incubation. The increase in gas production

and reduced incubation time could be attributed to the increase in fermentable carbohydrate proportion in the feed. Fermentable carbohydrate increases gas production while degradable nitrogen compound decreases gas production to some extent due to binding of carbohydrate with ammonia (Menke and Steingass, 1988).

Table 2: Fermentation characteristics of graded level of *Cajanus cajan* (Pigeon pea) and cassava peels incubated at 24H

Items	T1	T2	T3	T4	T5	T6	T7	T8	T9	SEM
a+b	16.00 ^{de}	19.00 ^{de}	25.50 ^{bc}	37.50 ^a	36.00 ^a	14.00 ^c	20.00 ^d	27.50 ^{bc}	32.00 ^{ab}	1.05
B	10.00 ^c	13.00 ^{de}	16.50 ^{cd}	25.00 ^a	26.00 ^a	9.00 ^c	12.00 ^c	17.00 ^c	21.00 ^b	0.72
C	0.104 ^{ab}	0.100 ^{abc}	0.091 ^{abc}	0.054 ^c	0.066 ^{abc}	0.106 ^a	0.085 ^{abc}	0.089 ^{abc}	0.058 ^c	0.008
T	7.00 ^b	7.00 ^b	6.00 ^b	6.00 ^b	6.00 ^b	11.00 ^a	7.00 ^b	6.00 ^b	6.00 ^b	0.51
Y	10.83 ^d	12.50 ^{cd}	16.00 ^{abc}	19.00 ^a	18.50 ^a	10.83 ^d	13.33 ^{bcd}	17.50 ^{ab}	17.00 ^{abc}	0.81
ME	4.58 ^c	4.65 ^d	4.94 ^b	5.17 ^a	4.95 ^b	4.65 ^d	4.80 ^c	5.17 ^a	4.93 ^b	0.04
OMD	42.61 ^g	42.80 ^f	44.60 ^c	45.93 ^b	44.16 ^d	42.89 ^f	43.70 ^c	46.17 ^a	44.23 ^d	0.24
SCFA	0.20 ^h	0.24 ^g	0.32 ^e	0.40 ^a	0.38 ^b	0.20 ^h	0.26 ^f	0.36 ^c	0.35 ^d	0.01

a,b,c,d,e,f,g,h,i: Means on the same column with different superscripts are significantly different ($P < 0.05$); T1: 100% *Cajanus cajan* (ex-kaduna) + 0% *Manihot specie*; T2: 75% *Cajanus cajan* (ex-kaduna) + 25% *Manihot specie*; T3: 50% *Cajanus cajan* (ex-kaduna) + 50% *Manihot specie*; T4: 25% *Cajanus cajan* (ex-kaduna) + 75% *Manihot specie*; T5: 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie*; T6: 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie*; T7: 75% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 25% *Manihot specie*; T8: 50% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 50% *Manihot specie*; T9: 25% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 75% *Manihot specie*; b (Gas production from potentially degradable but insoluble fraction); c (Rate of gas production); t (Time of most rapid increase in gas production); Y (Total volume of gas produced); ME (Metabolisable energy); OMD (Organic matter digestibility); SCFA (Short chain fatty acid); SEM: standard error of mean

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Treatment with 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie* reduced potential gas produced, gas produced from the insoluble fraction, total volume of gas produced but increased gas production rate for the insoluble fraction and incubation time. The low gas production could be as a result of increased protein, fiber and lignin content of the feedstuff and this agrees with Ososanya and Adeniji (2009) who reported that gas production from protein fermentation is small compared to carbohydrate and that the contribution of fat to gas production is negligible. The reduced time of incubation of 100% *Cajanus cajan* (ex kaduna) + 0% *Manihot specie* compared to 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie* could be attributed to the nature of fibre in the *Cajanus cajan*. This could also have been responsible for the treatment with 25% *Cajanus cajan* (ex-kaduna) + 75% *Manihot specie* having higher gas produced from the insoluble fraction than 25% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 75% *Manihot specie*. Treatment with 25% *Cajanus cajan* (ex-kaduna) + 75% *Manihot specie* and 50% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 50% *Manihot specie* had higher ME compared to others. The poor performance of T1 (100% ex-Kaduna) and T6 (100% Brown Ogbomoso Indigenous) probably suggest that legumes should not be fed solely to ruminants but should be used in combination with energy diet which is in accordance with Ajayi and Babayemi (2008) who reported that legumes are high in methane production and so require energy supplement for better utilization in order to sustain livestock

production. The reduced ME observed in this study could be an indication of inefficient fermentation due to increased energy loss in the form of gas production.

The Result of *in vitro* gas production characteristics of graded levels of two strains of *Cajanus cajan* and *Manihot spp* incubated at 48hrs is presented in Table 3. At 48 hr, treatment with 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie* increased potential gas produced, gas produced from the insoluble fraction, gas production rate for the insoluble fraction and total volume of gas produced. Treatment with 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie* had reduced potential gas produced, gas produced from the insoluble fraction, total volume of gas produced. The reduced gas production could be attributed to inadequate efficiency in protein degradation since energy is required for optimum activities of rumen microbes and this energy could be supplied by *Manihot specie*. Time of incubation was reduced in treatment with 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie* compared to 100% *Cajanus cajan* (ex-kaduna) + 0% *Manihot specie*. This could be due to the nature of the feedstuff as feedstuff with higher fiber component may take longer time to be fermented compared to the one with lower fiber and with fermentable or soluble carbohydrate. Treatment with 50% *Cajanus cajan* (ex-kaduna) + 50% *Manihot specie* was higher in terms of potential gas produced and gas produced from the insoluble fraction compared to 50% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 50% *Manihot specie*.

Table 3: Fermentation characteristics of graded level of *Cajanus cajan* (Pigeon pea) and cassava peels incubated at 48hrs

Items	T1	T2	T3	T4	T5	T6	T7	T8	T9	SEM
a+b	23.50 ^{ef}	26.50 ^{de}	36.00 ^{bc}	37.00 ^b	42.50 ^a	21.00 ^f	31.00 ^{cd}	28.00 ^{de}	36.00 ^{bc}	0.94
B	17.50 ^{ef}	21.00 ^{de}	26.00 ^{bc}	30.00 ^a	30.00 ^a	16.00 ^f	24.50 ^{cd}	21.50 ^d	28.50 ^{ab}	0.71
C	0.047 ^{bc}	0.038 ^c	0.038 ^c	0.060 ^{ab}	0.067 ^a	0.047 ^{bc}	0.041 ^c	0.053 ^{abc}	0.048 ^{bc}	0.003
T	12.00 ^a	9.00 ^{ab}	9.00 ^{ab}	9.00 ^{ab}	9.00 ^{ab}	8.00 ^b	9.00 ^{ab}	9.00 ^{ab}	9.00 ^{ab}	0.61
Y	13.33 ^{cd}	11.50 ^d	17.50 ^{bc}	19.50 ^b	26.00 ^a	10.00 ^d	14.00 ^{cd}	14.50 ^{bcd}	17.50 ^{bc}	0.96
ME	4.91 ^e	4.52 ^h	5.14 ^c	5.24 ^b	5.92 ^a	4.55 ^g	4.89 ^e	4.78 ^f	5.00 ^d	0.08
OMD	44.83 ^d	41.91 ^h	45.93 ^c	46.38 ^b	50.83 ^a	42.23 ^g	44.38 ^e	43.53 ^f	44.67 ^d	0.49
SCFA	0.26 ^f	0.22 ^g	0.36 ^c	0.41 ^b	0.56 ^a	0.18 ^h	0.28 ^e	0.29 ^d	0.36 ^c	0.02

a,b,c,d,e,f,g,h: Means on the same column with different superscripts are significantly different ($P < 0.05$); T1: 100% *Cajanus cajan* (ex-kaduna) + 0% *Manihot specie*; T2: 75% *Cajanus cajan* (ex-kaduna) + 25% *Manihot specie*; T3: 50% *Cajanus cajan* (ex-kaduna) + 50% *Manihot specie*; T4: 25% *Cajanus cajan* (ex-kaduna) + 75% *Manihot specie*; T5: 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie*; T6: 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie*; T7: 75% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 25% *Manihot specie*; T8: 50% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 50% *Manihot specie*; T9: 25% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 75% *Manihot specie*; b (Gas production from potentially degradable but insoluble fraction); c (Rate of gas production); t (Time of most rapid increase in gas production); Y (Total volume of gas produced); ME (Metabolisable energy); OMD (Organic matter digestibility); SCFA (Short chain fatty acid); SEM: standard error of mean

The ME, OMD and SCFA of the treatment with 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie* were consistently higher compared to other treatments. This is probably due to the increased energy and lipid content of *manihot specie* as compared to *cajanus cajan*. The Result of *in vitro* gas production characteristics of graded levels of two strains of *Cajanus cajan* and *Manihot spp* incubated at 72hrs is presented in Table 4. At 72-hr, treatment with 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie* increased potential gas

produced, gas produced from the insoluble fraction, gas production rate for the insoluble fraction and total volume of gas produced. Inclusion of 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie* reduced potential gas produced, gas produced from the insoluble fraction, gas production rate for the insoluble fraction and total volume of gas produced. However, the differences obtained were within the same time of incubation.

Table 4: Fermentation characteristics of graded level of *Cajanus cajan* (Pigeon pea) and cassava peels incubated at 72hours

Items	T1	T2	T3	T4	T5	T6	T7	T8	T9	SEM
a+b	31.00 ^d	32.50 ^{cd}	41.00 ^{abc}	43.00 ^{ab}	50.50 ^a	21.00 ^e	29.50 ^{de}	48.50 ^a	38.50 ^{bcd}	1.72
B	26.00 ^{bc}	24.00 ^{cd}	32.50 ^{ab}	33.00 ^{ab}	39.00 ^a	17.50 ^d	22.00 ^{cd}	39.00 ^a	24.50 ^{cd}	1.47
C	0.049 ^b	0.062 ^{ab}	0.067 ^{ab}	0.060 ^{ab}	0.082 ^a	0.056 ^b	0.054 ^b	0.053 ^b	0.061 ^{ab}	0.004
T	7.00 ^a	7.00 ^a	7.00 ^a	6.00 ^a	7.00 ^a	6.00 ^a	7.00 ^a	6.00 ^a	6.00 ^a	2.15
Y	17.17 ^b	16.67 ^{bc}	20.50 ^{ab}	20.00 ^{ab}	28.33 ^a	8.50 ^c	14.33 ^{bc}	20.00 ^{ab}	21.50 ^{ab}	1.51
ME	5.41 ^c	5.18 ^c	5.52 ^b	5.30 ^d	6.21 ^a	4.34 ^g	4.93 ^f	5.50 ^b	5.50 ^b	0.09
OMD	48.24 ^c	46.43 ^c	48.51 ^b	46.76 ^d	52.81 ^a	40.81 ^g	44.59 ^f	48.42 ^b	48.14 ^c	0.60
SCFA	0.35 ^e	0.34 ^f	0.43 ^c	0.42 ^d	0.62 ^a	0.14 ^h	0.28 ^g	0.42 ^d	0.45 ^b	0.02

a,b,c,d,e: Means on the same column with different superscripts are significantly different ($P < 0.05$); T1: 100% *Cajanus cajan* (ex-kaduna) + 0% *Manihot specie*; T2: 75% *Cajanus cajan* (ex-kaduna) + 25% *Manihot specie*; T3: 50% *Cajanus cajan* (ex-kaduna) + 50% *Manihot specie*; T4: 25% *Cajanus cajan* (ex-kaduna) + 75% *Manihot specie*; T5: 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie*; T6: 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie*; T7: 75% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 25% *Manihot specie*; T8: 50% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 50% *Manihot specie*; T9: 25% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 75% *Manihot specie*; b (Gas production from potentially degradable but insoluble fraction); c (Rate of gas production); t (Time of most rapid increase in gas production); Y (Total volume of gas produced); ME (Metabolisable energy); OMD (Organic matter digestibility); SCFA (Short chain fatty acid); SEM: standard error of mean

Potential gas produced, gas produced from the insoluble fraction and total volume of gas produced were higher in treatment with 100% *Cajanus cajan* (ex-kaduna) + 0% *Manihot specie* than 100% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 0% *Manihot specie*. The gas produced from the insoluble fraction was higher with 25% *Cajanus cajan* (ex-kaduna) + 75% *Manihot specie* inclusion compared to 25% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 75% *Manihot specie*. The ME, OMD and SCFA were higher in treatment with 0% *Cajanus cajan* (ex-kaduna) + 100% *Manihot specie* compared to others. Gas production is basically the result of carbohydrate fermentation to volatile fatty acids (Getachew *et al.*, 2002) due to microbial degradation of samples (Babayemi *et al.*, 2004; Fievez *et al.*, 2005). The resultant short chain fatty acid is an indication of the energy availability to the animal, contributes up to 80% of animals' daily energy requirements (Fellner, 2004) and is directly proportional to ME (Menke *et al.*, 1979) and gas production (Beuvink and Spoelstra, 1992)

Conclusion

The study showed that a more efficient rumen fermentation can be achieved when *Cajanus cajan* (Ex-Kaduna) and *Manihot specie* was incubated at the ratio of 25:75 respectively, for 24 hours. Ex-Kaduna performed better and was more fermented or degraded by rumen microbes than Brown Ogbomoso Indigenous. The incubation of 50% *Cajanus cajan* (Brown Ogbomoso Indigenous) + 50% *Manihot specie* was also promising. Higher gas production at reduced incubation time could imply increase feed intake and probably increased performance when such feedstuff or feed combinations are offered to ruminants.

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