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Assessment of the Effect of Browse Plants' Feed Formulation on Goat (*Capra aegagrus hircus*) Rumen Microbial Activities and *in vitro* Gas Production

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Abstract

This study assessed the effect of browse Plants feed formulation on Goat (Capra aegagrus hircus) rumen microbial activities and in vitro gas production in Red Sokoto goat (Capra aegagrus hircus) over a period of fourteen (14) weeks. Four selected browse plants- Afzelia africana (TAa) Detarium microcarpum (TDm), Daniellia oliveri (TDo) and Khaya senegalensis (TKs) -were used to formulate diets for animal feeding. Three groups of growing Indigenous Red Sokoto goats were assigned to each of the browse plant diets (BPD), while the control group was placed on a basal diet only. Rumen fluids were collected from the goats intermittently for three (3) consecutive periods, and analyzed for pH, methylene blue-reduction time (MBRT), nitrate reduction, cellulose digestion, glucose fermentation and sedimentation activity rate. The results showed that browse plant feed formulation did not have a significant ($p > 0.05$) effect on the pH value of the rumen contents. Significant ($p < 0.05$) reductions in MBRT, nitrate reduction, and glucose fermentation were observed in all treatments except the control diets. However, the period for cellulose digestion significantly increased in all the browse plant-supplemented diet treatments. Furthermore, the volume of gas produced was significantly reduced by 64% (TAa), 62% (TDm), 71% (TDo), and 74% (TKs) in goats fed with browse plants as dietary supplements compared to 7% in the control (Tc). Overall, this study demonstrated that browse plant feed has the potential to significantly reduce the volume of methane produced and released by ruminant animals into the environment.

Keywords: Browse Plants, Gas Production, Indigenous Red Sokoto Goats, Methane gas, Methylene Blue Reduction, Rumen Microbial Ecolgy,

Introduction

The rumen's microbial communities which include bacteria, fungi, viruses, protozoa, and archaea have the potential to transform fibrous, low-quality plant materials into nutrients that are made available to ruminants. The ruminant's special capability to transform plant fodder into high-quality food products has helped to ensure the sustainability of food and agricultural systems. They are of great value to the production of food of animal origin by using crop residues and byproducts as feed sources (Hinsu *et al.,* 2021).

Ruminant animals are extremely valuable due to the demand for their meat and fibre products. The transformation of feed into final products that have an impact on both the animal and the

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environment is greatly influenced by the microorganisms in the digestive tracts. Understanding these processes is essential, as the need and productivity of livestock are on the increase, especially in developing nations. Management and use of feed and other natural resources are essential to the development of sustainable ruminant production (Ellison *et al.,* 2017).

Research has shown that the type and makeup of the feed that ruminants consume may have an impact on the ecology of the rumen microbes and, as a result, the quantity of gases that are produced and released (Malmuthuge *et al.,* 2012; Paone and Cani, 2020). Without the ability of the rumen microbial flora to ferment, it is anticipated that the majority of the feed components for ruminants cellulose and lignin that serve as their primary source of nutrients could not be digested (Mizrahi *et al., 2021*).

One crucial step in the acquisition and digestion of feed for the ruminants is the fermentation process carried out by the gut flora and the methanogenesis activities of the methane-producing organisms. It has been reported that all of these gases are released into the environment as ruminant waste products (Rinninella *et al.,* 2019; Cui *et al.,* 2020).

The rumen gut flora consists of obligate and facultative anaerobic communities that can break down feeds that contain cellulose and lignin. There is a global concern about the impact of accumulated greenhouse gases on the environment and the resultant loss of quality and productivity of livestock. Microbial rumen fermentation processes have been implicated in increased protein losses, like nitrogen with its resultant restriction in the animal's ability to produce at its peak, and energy losses, like methane that add to the environment's greenhouse gas pollution.

Over time it is observed that the manipulation of the rumen microbial population using a Number of chemical feed additives, antibiotics, ionophores, methane inhibitors, and defaunating agents has led to probable toxicity problems for the host animals, the risk of residues in food of animal origin, and the emergence of multi-drug-resistant microbes that may threaten human health (Olafadehan and Okunade 2016).

Additionally, consumer advocacy groups have criticized these supplements for their lack of quality and safety. The search for natural feed additive(s) as substitutes. has been sparked by consumer demands. One such initiative from recent years is the addition of browse plants, which numerous studies have shown to be extremely beneficial for these ruminants as feed. They have year-round availability and sufficient nutritional value to support the animals (Olafadehan and Okunade, 2016).

The secondary plant metabolites, such as saponins, tannins, and essential plant oils, are responsible for the antimicrobial activity of browse plants (Olafadehan and Okunade, 2016). The potential of many of these advantageous browse foliages to control the rumen microbial population and their environment for efficient ruminant animal production and the decrease of pollutants (enteric methane) that contribute to climate change has not been thoroughly investigated. These findings revealed that selected species of browse plants have the ability to manipulate the rumen microbial ecology. Therefore, this study assesses the effect of selected browse plants on rumen microbial activities and *in vitro* gas production in red Sokoto goats *(Capra aegagrus hircus*).

Materials and Methods

Experimental Site

The research was conducted at the teaching and research farm of the Federal College of Wildlife Management, New- Bussa, Niger State. The experimental station (New Bussa) sits at 9° 53'N,9.883°N and 4° 31'E, 4.517°E (NIPOST Archives, 2009).

Sample Collection.

A fresh plant sample was harvested in the wild from the bushes surrounding the premises of the Federal College of Wildlife Management in New Bussa, Niger State. The identity of the plant was confirmed and the voucher was assigned to the Herbarium Department, Forestry Research Institute of Nigeria (FRIN), Ibadan, Oyo State, with the Ascension numbers FHI.114086, FHI.114087, FHI.1140868, and FHI.114089, respectively. Fifteen red-growing Sokoto goat breeds weighing between 7 and 10 grams were purchased from the Wawa and Dongogari markets, all in Borgu local government. area, Niger State, Nigeria. They were allowed to acclimatise to the environmental conditions of the experimental site and feed and water were supplied *ad libitum*. One hundred 100 mL of representative rumen content was collected intermittently for three consecutive periods before morning feeding on each day of sample collection from each buck with the aid of a suction tube, as described by Okunade *et al*. (2014). The rumen liquor was collected into the thermoflask that had been pre-warmed to a temperature of 39 °C.

Ethical Clearance

Ethical clearance for the study was obtained from the Federal College of Wildlife Management Ethical Committee with approval number CWM/RERC/2023/667.

Screening of Tanniferrous plant fodders

Samples were analyzed chemically according to the official methods of analysis described by the Association of Official Analytical Chemists (AOAC, 2005). All analyses were carried out in duplicate. Some selected tanniferrous plant fodder were (Ac = *Acacia alibido,* Dm = *Detarium microcarpum,* Ks = *khaya senegalesis*, Pt = *Piliostigma thonnigi*, Do = *Damella oliven,* Tj = *Teminalia jigosona*, Aa = *Afzellia Africana,* Vp = *Vitellaria paradoxa*). pre - screened and analyzed for its proximate and phytochemical composition (crude protein, crude fibre, ether extract and ash). The fibre fractions; neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were determined according to AOAC official method 988.05. Cellulose and hemicellulose were calculated as the differences between ADF and ADL, NDF and ADF respectively in order to know and select the suitable fodder based on concentration or amount of condensed tannins after which the basal diet comprised of (Maize, offal 25kg, cowpea husk 45,50kg, Groundnut cake (GNC) 20kg, Brewers dried grain 5kg, Vitamim – mineral Premix 1kg, Dicalcium phosphate 2kg, sulphur powder 1kg and Table Salt 1kg = 100kg) supplemented with suitable selected Samples of browse foliage were equally analyzed according to the standard methods of (AOAC, 2005).

Chemicals used

All the reagents and chemicals used in this study were of analytical grade and procured from the Central Research Laboratory, Ilorin, Kwara State and the Department of Animal Science, University of Ibadan, Nigeria.

Experimental design and management

Fifteen growing Red Sokoto bucks, 7–9 months old, with an average initial weight of 9.00±0.25 kg, were used for the study. Each goat was housed in an individual pen (1.20 m × 0.80 m × 0.70 m) furnished with drinking and feeding facilities. The goats were treated against

endoparasites and ectoparasites prior to the commencement of the experiment. They were randomly allocated to five dietary treatments in a completely randomized design. Each treatment was replicated with three animals. The experimental diets were formulated to meet the nutritional requirements of growing goats. The animals in the last treatment (control) were served the concentrate (as shown in Figure 1) formulated with a basal diet of threshed sorghum top only, while the remaining four treatments were supplemented with *Afzelia aaricana, Detarium microcarpum, Daniellia oliveri,* and *Khaya senegalensis.* The experimental diets were offered as a complete ration mix (forage and concentrate) in two equal meals. The experiment lasted for 14 weeks. Browsed foliage was served after the basal feed. Provision was made for a daily feed allowance of 10% above that of the previous day`s intake. Clean water was provided *ad libitum* daily.

Physicochemical characteristics of the rumen fluid

Immediately after sample collection, the pH was measured using a pH meter, and the fluid was allowed to sit in a test tube and determine the time (in minutes) for complete sedimentation and flotation of solid particles in order to test for sedimentation activity time (SAT). Smaller particles sink, and larger particles float on the bubbles of fermentation (Ismael *et al.,* 2014). In another portion of the rumen fluid, the methylene blue reduction time (MBRT) was measured: 20 ml of rumen fluid was mixed with 1 ml of 0.03% methylene blue in a test tube and allowed to stand at room temperature, and a timer was set. The time needed for the color of the mixture to change was measured and recorded (Inyang and Ososanya, 2017).

Cellulose digestion test.

Ten (10) ml of rumen fluid was mixed with 0.3 ml of 16% glucose. A thread of pure cellulose was immersed, and the lower end was weighted by a glass bead. The tubes were incubated at 39°C for 72 hours and the time for the bead to be dropped free at the bottom of the tube was recorded.

Nitrate Reduction test

Ten (10) ml of sieved rumen fluid was placed into each of the three test tubes, and 0.2, 0.5, and 0.7 ml of 0.025% potassium nitrate solution were added to the three tubes. The three tubes were put in a water bath at 39 °C. Every five minutes, one drop from each tube was placed on a small ceramic plate. 2 drops of reagent I (2 ml of sulphanilic acid in 30% acetic acid to make 200 ml) and Two **(**2) drops of reagent II (0.6 ml alpha-naphthylamine + 16 ml conc. acetic acid) were added to each drop, and 140 ml of distilled water was also added. The change in color was observed.

Glucose Fermentation test

A glucose solution of 0.5 ml of 16 % was added to 10 ml of rumen fluid. The mixture was placed in a fermentation saccharometer and kept at 39^{o}c for 72 hours. The results was read after 30 and 60 min.

In Vitro Gas Production

All laboratory handling of rumen fluid was carried out under a continuous flow of carbon (IV) oxide. Two hundred milligrams (200 mg) of the dry and milled leaves of each experimental diet were accurately weighed, packed in asbestos cloth sealed in both ends and put into a calibrated transparent 100ml glass syringe fitted with plungers. In vitro, incubation of the samples was conducted in triplicate. Syringes were filled with 30 ml of medium consisting of 10 ml of strained rumen fluid and 20 ml of buffer solution (g/liter of 1.985 (Na_2) HPO_4 + 1.302 KH_2PO_4 + 0.105 $MgCl_2.6H_2O$ + 1.407 NH_2HCO_3 + 5.418 $NaHCO_3$ + 0.390 Cysteine HCl + 0.100

NaOH) and three blank samples containing 30 ml of medium (inoculums and buffer) were incubated at the same time. The syringes were placed in a rotor inside the incubator (39 °C) with about one rotation per minute. The gas production was recorded at 3, 6, 9, 12, 18, 24, 36, and 48 hours. During the post-incubation period, 4 ml of (10 M) sodium hydroxide (NaOH) was dispensed into each incubated sample. Sodium hydroxide was added to absorb carbon dioxide that was produced during the fermentation process, and the remaining volumes of gas were recorded as methane, according to the report by Isah *et al*., (2014). The average volume of gas produced from the blanks was deducted from the volume of gas produced from the samples.

Statistical analysis

Data obtained were subjected to statistical analysis using Analysis of Variance (ANOVA) and the significance mean value was identified using Dunnet posthoc analysis at $P < 0.05$ on SPSS version 21 software**.**

RESULTS

The results for the chemical composition of tanniferrous plant samples pre-screened for selection are presented in Table 1. The amount of crude protein obtained in the selected tanniniferous plant foliage used in this experiment was within the range that had been previously reported by Okunade *et al*., (2014) for tropical tanniniferous plant foliage. Non-fibre carbohydrates and NDF were within the normal levels required for growing goats (Isah *et al*., 2014; Jha *et al*., 2019). Condensed tannin and saponin concentrations in all the plant foliage studied in this work are moderate, except for *A. olibido*, for the level ruminant animals can tolerate without any detrimental effect (Okunade *et al*., 2014). Baker *et al*., (2021) also reported that chemical composition is subjected to wide fluctuations depending on soil and climate characteristics. The nutrient density of this selected tanniferous plant foliage in terms of CP, NFC, and NDF is adequate to meet the nutrient requirements of growing goats (Salah *et al*., 2014; Rinninella *et al*., 2019). In addition, the moderate CT and saponin contents reported in this study, which can increase rumen undegraded proteins and help in enteric methane (CH_4) and carbon dioxide (CO_2) mitigation, indicate that all the selected browse plants have potential feeding value as dietary plant supplement for ruminants in areas where one of the most important factors limiting productivity is feed supply.

Table 1: Chemical composition of tanniferous browse plant sample

Plant Sam	Parameters						
	Crude Protein (%)	Crude Fibre (%)	Dry Matter (%)	Tannin (%)	Saponin (%)	Volatile Oil (%)	Phytic acid (%)
Ac	$14.0^{a} \pm 0.2$	$22.6^{a} \pm 0.1$	$92.6^{a} \pm 0.1$	$10.5^{a} \pm 6.2$	$5.9^{a} \pm 1.9$	$4.9^{b} \pm 4.9$	$16.3^{a} \pm 10.8$
Dm	$5.6^{c} \pm 0.4$	$13.5^{c} \pm 0.1$	$93.0^{a} \pm 0.0$	$3.4^{b} \pm 0.2$	$3.9^{b} \pm 0.3$	$10.8^{a} \pm 0.5$	$4.9^{b} \pm 1.9$
Ks	$4.9^{c} \pm 0.2$	$22.0^{a} \pm 0.2$	$93.3^{a} \pm 0.2$	$3.6^{b} \pm 0.1$	$4.5^{b} \pm 0.4$	$12.5^{a} \pm 1.7$	$12.6^{a} \pm 8.8$
Pt	$9.6^{b} \pm 0.3$	$22.7^{a} \pm 0.0$	$93.6^{a} \pm 0.5$	$3.7^{b} \pm 0.4$	$4.6^{b} \pm 0.5$	$14.2^{a} \pm 1.1$	$7.9^{b} \pm 0.1$
Do	$2.9^{d} \pm 0.4$	$18.8^{b} \pm 0.1$	$92.7^{a} \pm 0.2$	$4.0^{b} \pm 0.2$	$3.9^{b} \pm 0.1$	$12.3^{a} \pm 1.2$	$7.7^{b} \pm 0.6$
Tj	$7.9^{b} \pm 0.5$	$15.2^{b} \pm 0.5$	$93.2^{a} \pm 0.3$	3.8b ± 0.1	$4.1^{b} \pm 0.4$	$11.4^{a} \pm 1.6$	$6.8^{b} \pm 0.3$
Aa	$9.6^{b} \pm 0.1$	$10.9^{c} \pm 0.1$	$94.5^{a} \pm 0.2$	$3.9^{b} \pm 0.1$	$4.4^{b} \pm 0.4$	$13.6^{a} \pm 0.4$	$6.5^{b} \pm 0.0$
Vp	$8.5^{b} \pm 0.2$	$17.3^{b} \pm 0.2$	$94.2^{a} \pm 0.3$	$4.1^{b} \pm 0.3$	$4.2^{b} \pm 0.5$	$14.5^{a} \pm 0.5$	$7.1^{b} \pm 0.9$

Key: Ac = *Acacia alibido,* Dm = *Detarium microcarpum,* Ks = *khaya senegalesis*, Pt = *Piliostigma thonnigi*, Do = *Damella oliven,* Tj = Teminalia jigosona, Aa = *Afzellia Africana,* Vp = *Vitellaria paradoxa.* Tannin values within the range of "3 – 4 %" are within the standard range that could support healthy rumen flora.

The chemical composition of the feed supplements is presented in Table 2. The lowest value of CP (12.70 g/100 g DM) for *Khaya senegalensis* is well above the range of 7.0–8.0 g/100g DM suggested as the critical limit below which intake of forages by ruminants and rumen

microbial activity would be adversely affected (Isah *et al*., 2014). The optimal concentration of NFC is important in ruminants' diets to avoid acidosis and other metabolic problems. Diets with excess NFC can cause ruminant upsets and health problems. The fibre fraction contents of the plant species were generally moderate and within the limits established by the NRC for ruminant animals to ensure proper digestion and rumination. The mean NDF values of 50.70 and 43.80 g/100 g DM were low to moderate when compared with low-quality roughages, which ruminants can effectively degrade (Salah *et al*., 2014). The low to moderate fiber contents of the browse fodders suggest their high nutritive value since fiber plays a significant role in voluntary intake and digestibility. The range of cellulose concentrations shows that the fodders have the potential to support intestinal movement, promote proper rumen function, and promote dietary efficiency. Kittleman *et al*., 2016, opined that, the higher the hemicellulose fraction, the higher the feed value. The levels of CTs recorded for TAa TDm,TDo, and TKs in this study are much below the range of 60 to 100 g/kg DM, considered to depress feed intake, growth, and cidal effects (Mbomi *et al*., 2011; MaAllister *et al*., 2015). Therefore, the plant species contained CTs at levels beneficial to ruminants because CTs at low levels produce a mild or low protein binding effect (Olafadehan, 2013). Similarly, CT-containing forage, in addition to other benefits, minimizes methane emission by ruminants (methane mitigation), when not included in a high proportion of the diet (Bodas *et al*., 2012; Cieslak *et al*., 2012). Saponin levels in all the samples were lower than the tolerable level of 15-20g/kg DM reported for goats, which suggested that the levels reported herein are not likely to affect the nutritional potential of the plants in ruminants. therefore, feed containing tannin and saponin has been shown to act as defaunating agents, and is capable of reducing methane production (Olafadehan 2013).

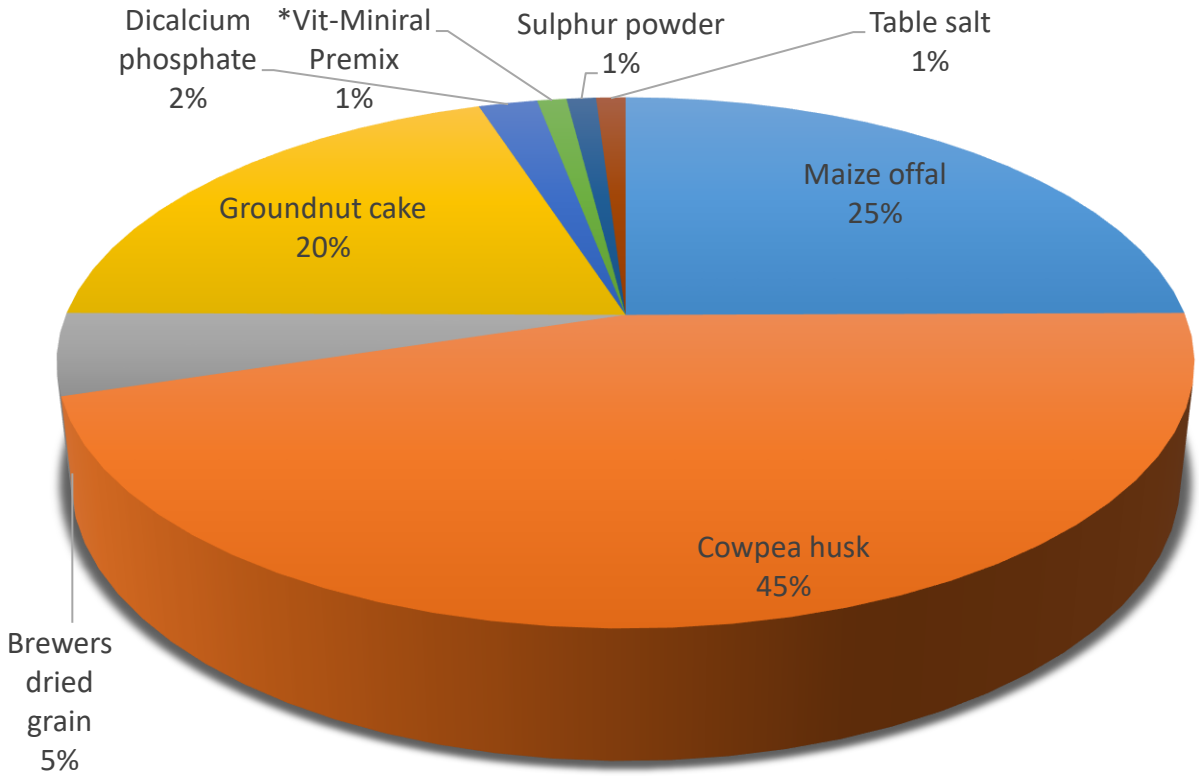

Figure 1: Nutrient content of formulated concentrate

Table 2: Chemical composition of controlled diet and supplemented plants

Feed Formulation	Parameters						
	DM (%)	CP (%)	FAT (%)	CF (%)	NDF (%)	CT (%)	Sap (%)
T1	93.1^{a} ± 0.1	14.1^{a} ± 0.2	3.6^{a} ± 0.1	12.6^{a} ± 0.1	49.0^{a} ± 0.1	3.4^{a} ± 0.2	3.3^{a} ± 0.1
T2	93.4^{a} ± 0.2	16.1^{a} ± 0.0	4.3^{a} ± 0.2	11.7^{a} ± 0.2	44.3^{b} ± 0.1	3.2^{a} ± 0.2	4.2^{a} ± 0.1
T3	92.9^{a} ± 0.1	13.5^{a} ± 0.4	3.2^{a} ± 0.1	14.2^{a} ± 0.2	50.7^{a} ± 0.3	3.3^{a} ± 0.3	3.4^{a} ± 0.2
T4	92.7^{a} ± 0.1	12.7^{a} ± 0.3	3.6^{a} ± 0.1	13.7^{a} ± 0.1	47.8^{a} ± 0.3	3.0^{a} ± 0.4	4.5^{a} ± 0.1
T5	93.2^{a} ± 0.2	15.7^{a} ± 0.4	4.6^{a} ± 0.1	9.1^{b} ± 0.3	43.8^{b} ± 0.1	0.0^{b} ± 0.0	0.0^{b} ± 0.0

T1 = Concentrate diet + *Afzelia Africana*, T2= Concentrate diet + *Detarium macrocarpum,* T3 = Concentrate diet + *Danniellia oliveri,* T4 = Concentrate diet + *Khaya senegalensis,* T5 = Concentrate (Control) diet

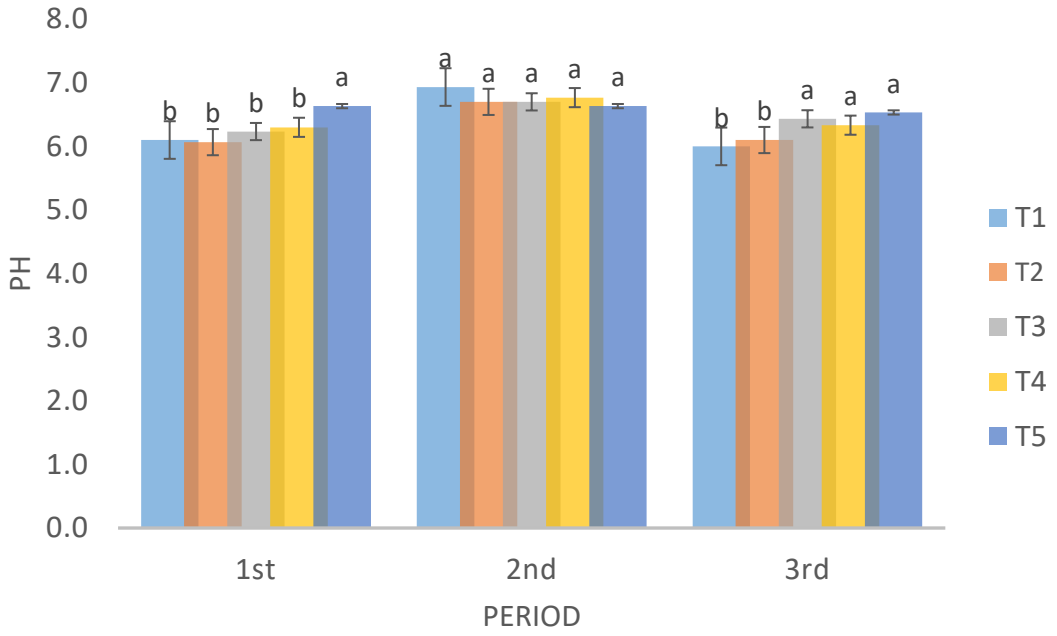

Figure 1: **The values of pH of the rumen Contents**

T1 = Concentrate diet + *Afzelia Africana*, T2= Concentrate diet + *Detarium macrocarpum,* T3 = Concentrate diet + *Danniellia oliveri,* T4 = Concentrate diet + *Khaya senegalensis,* T5 = Concentrate (Control) diet ; The difference in the alphabet on the charts indicate significant difference at P< 0.05

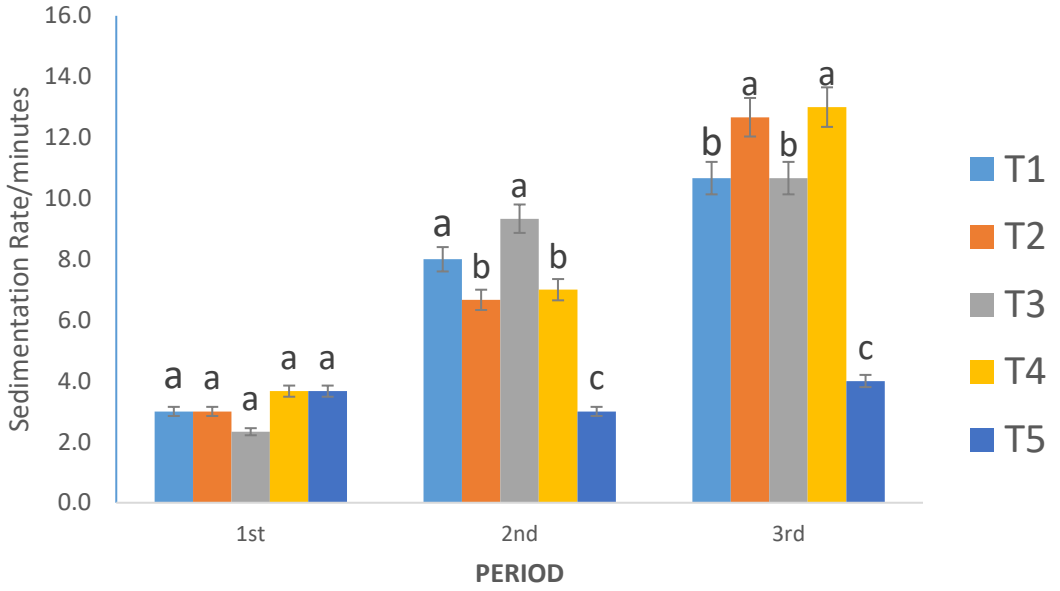

Figure 3: Sedimentation rate per minute of rumen contents
Key: T1 = Concentrate diet + *Afzelia Africana*, T2= Concentrate diet + *Detarium macrocarpum,* T3 = Concentrate diet + *Danniellia oliveri,* T4 = Concentrate diet + *Khaya senegalensis,* T5 = Concentrate (Control) diet ; The difference in the alphabet on the charts indicate significant difference at P< 0.05

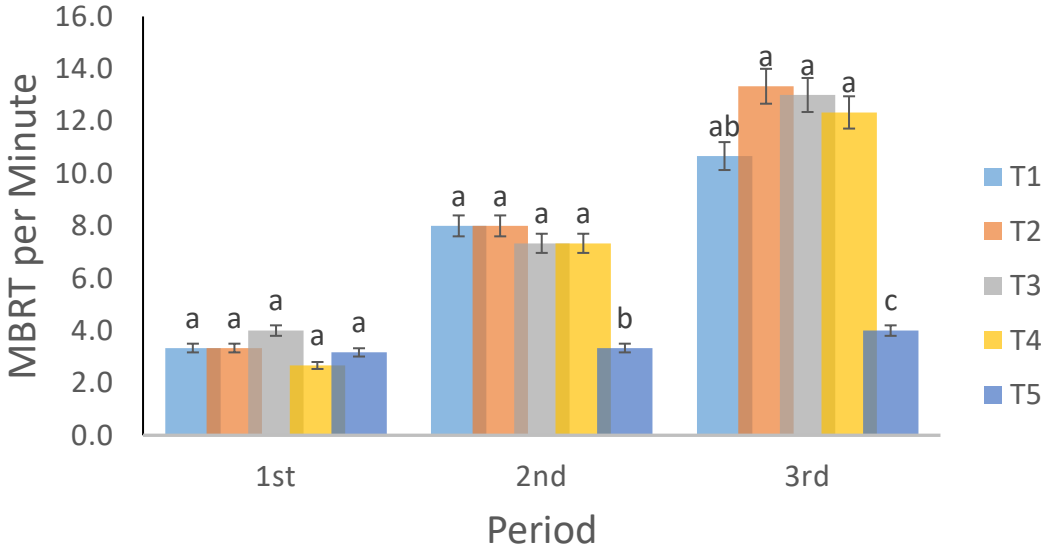

Figure 4: Methylene Blue Reduction Test per minutes of rumen contents
T1 = Concentrate diet + *Afzelia Africana*, T2= Concentrate diet + *Detarium macrocarpum,* T3 = Concentrate diet + *Danniellia oliveri,* T4 = Concentrate diet + *Khaya senegalensis,* T5 = Concentrate (Control) diet ; The difference in the alphabet on the charts indicate significant difference at P< 0.05

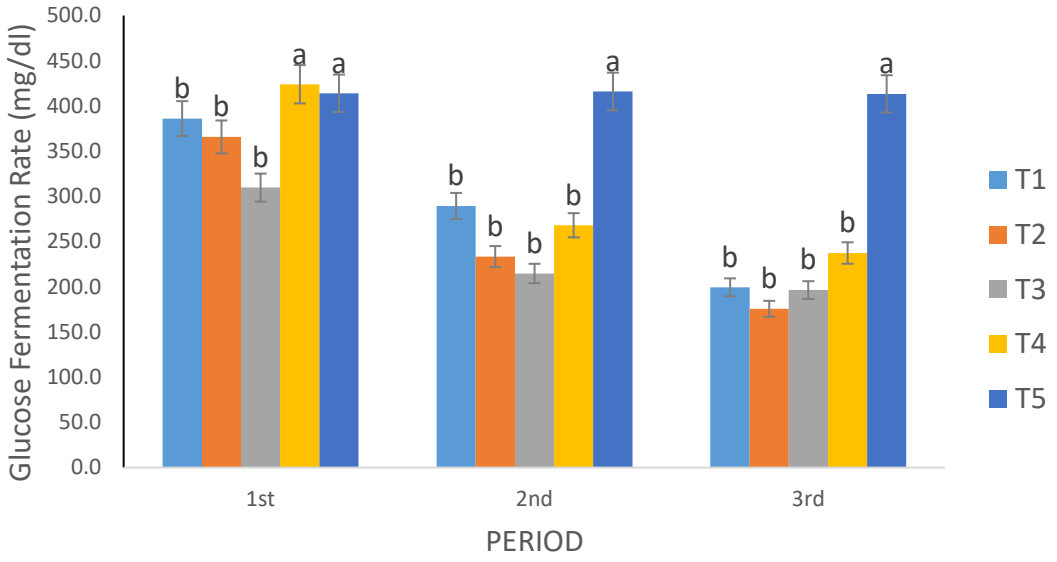

Figure 5: Glucose fermentation test per minutes of rumen contents

T1 = Concentrate diet + *Afzelia Africana*, T2= Concentrate diet + *Detarium macrocarpum,* T3 = Concentrate diet + *Danniellia oliveri,* T4 = Concentrate diet + *Khaya senegalensis,* T5 = Concentrate (Control) diet ; The difference in the alphabet on the charts indicate significant difference at P< 0.05

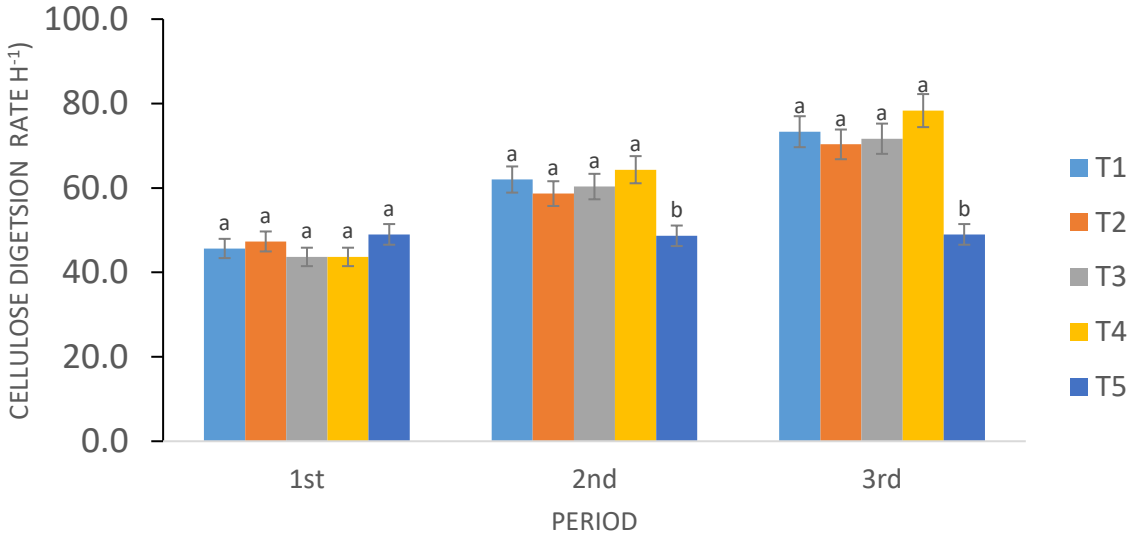

Figure 6: Cellulose digestion rate of rumen contents

T1 = Concentrate diet + *Afzelia Africana*, T2= Concentrate diet + *Detarium macrocarpum,* T3 = Concentrate diet + *Danniellia oliveri,* T4 = Concentrate diet + *Khaya senegalensis,* T5 = Concentrate (Control) diet ; The difference in the alphabet on the charts indicate significant difference at P< 0.05.

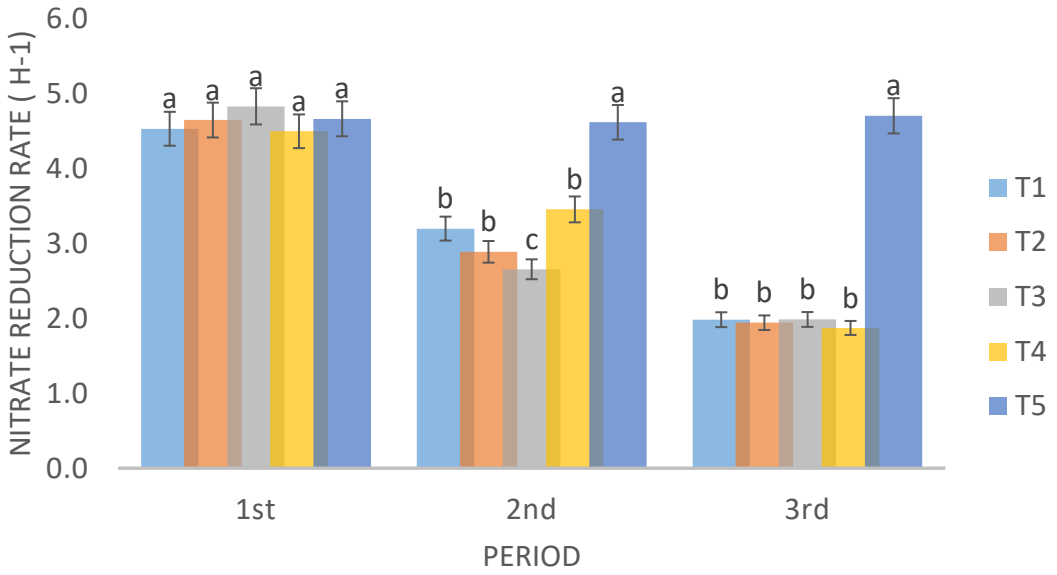

Figure 7: Nitrate reduction rate of rumen contents

T1 = Concentrate diet + *Afzelia Africana*, T2= Concentrate diet + *Detarium macrocarpum,* T3 = Concentrate diet + *Danniellia oliveri,* T4 = Concentrate diet + *Khaya senegalensis,* T5 = Concentrate (Control) diet ; The difference in the alphabet on the charts indicate significant difference at P< 0.05

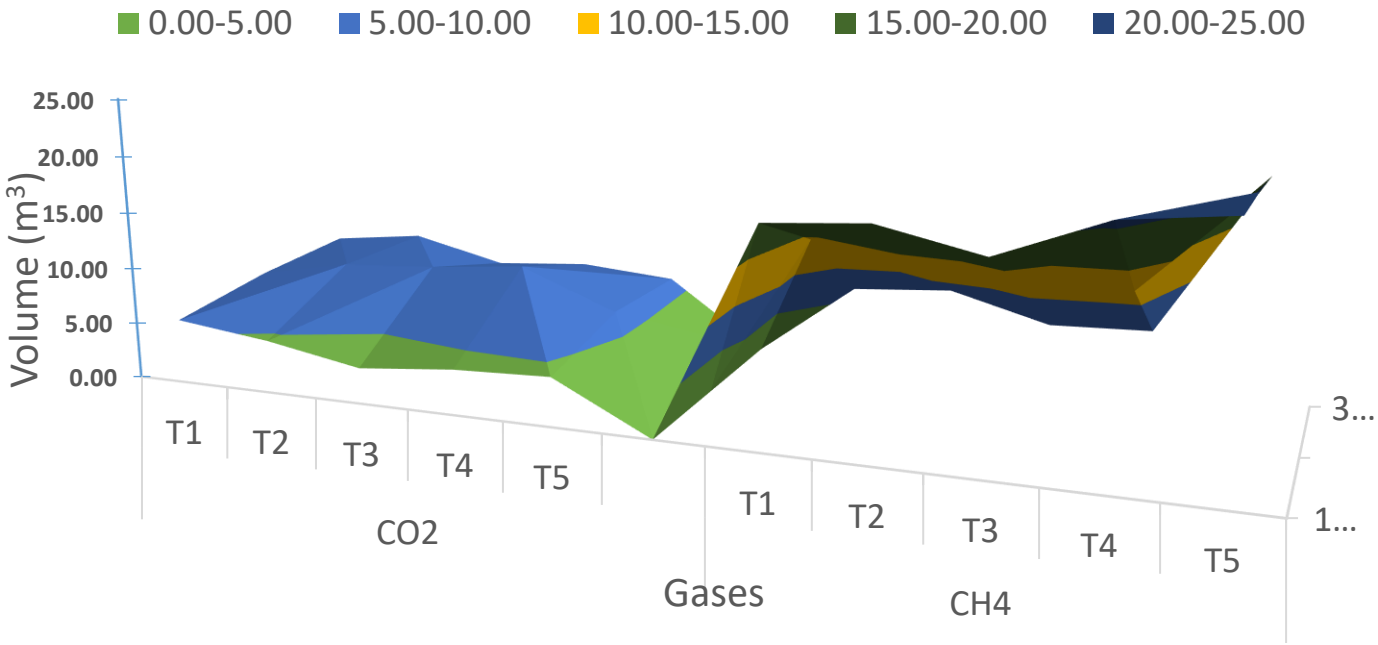

Figure 8: Average gas production of experimental animals fed with Tanniferrous foliage

DISCUSSION

Condensed tannin fortification did not affect the study's pH, as seen in Figure 2, as there was no statistically significant difference between the treatments. Inyang and Ososanya (2017) also reported this claim, stating that after eight weeks, the pH levels of probiotics, fibrinolytic enzymes, and control-supplemented diets remained unchanged. They explained this by

saying that probiotics do not affect the pH level of the rumen, preventing lactic acid from building up there and creating a stable environment for rumen fermentation. The pH result obtained in this study is in line with earlier findings and also agrees with the report of Ismael *et al.,* (2014), which stated that condensed tannin did not affect goats' rumen pH value with any significance. However, it stabilized pH in a range that is compatible with the optimal ruminal ecological dominance (Ismael *et al.,* 2014).

The current study's sedimentation activity time (SAT) (Figure 3) was susceptible to the effect of condensed tannin concentration, with higher sedimentation times compared to control treatments in the second and third periods across treatments 1–4. This outcome differs from that of Inyang and Ososanya (2017). where the probiotic-supplemented diet showed the lowest value in the eighth week, differing by 3.25 minutes from the control (4.75 minutes). The present result showed increased SAT due to an alteration in microbe activity, which in turn affected substrate degradation. The normal range for SAT is 4–8 minutes. Inactive fluids show slow sedimentation with little to no flotation due to the effect of condensed tannins, which are bound to the membranes of the microbes (Inyang and Ososanya, 2017).

The activity of the anaerobic rumen flora is measured using the methylene blue reduction time (MBRT). According to Ismael *et al.,* (2014), the normal range for MBRT is 3–6 minutes, but prolonged discolouration, longer than 10–15 minutes, indicates insufficient anaerobic bacterial population, rumen acidosis, or indigestible roughage. The study's findings are consistent with Ismael *et al.,* (2014). Methylene blue reduction time (an increased rate) (Figure 4) observed in the second and third periods between T1 and T4 in comparison to the control diets in the present work indicated inactive ruminal microflora. This claim holds true for animals fed on probiotics or condensed tannin diets, which significantly reduced the ruminal microflora population and, in turn, affected their activity (Inyang and Ososanya, 2017).

According to Abdul-Majeed *et al.,* (2011), nitrate acts as an alternative hydrogen sink and thereby lowers enteric methane production. This is supported by the nitrate reduction test period obtained in the current study (Figure 5) in treatments 1-4 for the 2nd and 3rd periods, which indicates nitrogen balance in the diets as compared to the control diets. Growth rates and nitrogen retention tend to be higher for goats with fermentable nitrogen in their diets. which is likewise in line with what Inyang and Ososanya (2017) reported.

Figure 6 of the current study illustrates the slower rate of cellulose digestion, which was also noted in the earlier study and is consistent with the findings of Han *et al.,* (2008) and Inyang and Ososanya (2017). Hence, the higher time rate recorded in this present study was attributed to inactive microbial activity when medium- or low-grain-containing rations were used (Cui *et al.,* 2020).

The glucose concentration as presented in Figure 7 in the present study showed a reduced glucose level across all the treatments with browse plants compared to the control in the second and third periods, which indicates reduced gas production (Abdul-Majeed *et al., 2011*). The present study agrees with the report of Okunade *et al.*. (2014), who opined that higher molecular weight tannins reduce glucose levels and influences the rate and quantity of gas production.

This study also evaluate the amount of *in vitro* gas (CO_2 and methane) production from dietary treatments (control and browse plant-supplemented diets). The main factors affecting total *in vitro* gas production are the condensed tannin and saponin contents since all the diets

understudied had adequate CP, NFC, and moderate NDF that are below the level that can lower or hinder digestibility. The volume of gas produced was significantly reduced by 64% (TAa), 62% (TDm), 71% (TDo), and 74% (TKs) in goats fed with browse plants as dietary supplements compared to 7% in the control (Tc). This observation agrees with the reports of Isah *et al*., (2014) and Patra *et al., (*2017). Okunade *et al.,* (2014) also opined that the molecular weight of tannins influences the rate and quantity of gas production *in vitro*. In the present study, tannin showed a depressing effect on the fermentability and digestibility of plants. The control diet (Tc), which is without condensed tannin plant supplementation, was the most fermentable and digestible dietary treatment that could be associated with the non-detectable level of phenolic compounds (tannins and saponin), which probably allowed the faster degradation of high CP, NCF, and NDF, which led to higher gas production compared to other diets (Elghandour *et al*., 2017). On the other hand, browse plant-supplemented diets (TAa, TDm, TDo, and TKs) recorded lower total in vitro gas production in that chronological order compared to control diets, which could be due to the negative influence of high CT. and this is in agreement with earlier works (Gunnu *et al*., 2016; Elghandour *et al*., 2017).

CONCLUSION

Results obtained showed that the concentration of condensed tannins contained in the browse plants formulated feed has led to reduced activities of rumen microbes in breaking down feedstuff and reduction in total gas production and its accompanying enteric pollutants (CO_2 and CH_4) thereby reduced the GHG emission, improved feed efficiency and total animal productivity.

Plant secondary metabolites such as tannins and saponins contained in browse plants can be used strategically in rumen microbes for reduction of CO_2 and enteric methane. This will eventually improve ruminant performance and minimize greenhouse gas emission thereby contributing to addressing climate change challenges.

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