

**INTAKE, DIGESTIBILITY, AND NITROGEN BALANCE OF SHEEP FED BAMBARA
GROUNDNUT HAULM AS SUPPLEMENT TO A MAIZE STOVER BASAL DIET**

BY
KNUST

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DECLARATION

I, **Preston Anderson**, do hereby declare that the work herein submitted as a thesis for Master of Philosophy (Animal Nutrition) Degree has neither been presented nor is being concurrently submitted for any other degree elsewhere. However, work of other researchers and authors which served as sources of information are duly acknowledged.

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DEDICATION

This piece of work is dedicated to my late mother, who was always immersed in imaginary thought about my success though death came ahead of her celebrations of her son. I also dedicate

this work

to my dear son Gastonal John Anderson.

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I am very grateful to the Almighty God for providing me good health, wisdom and strength in my

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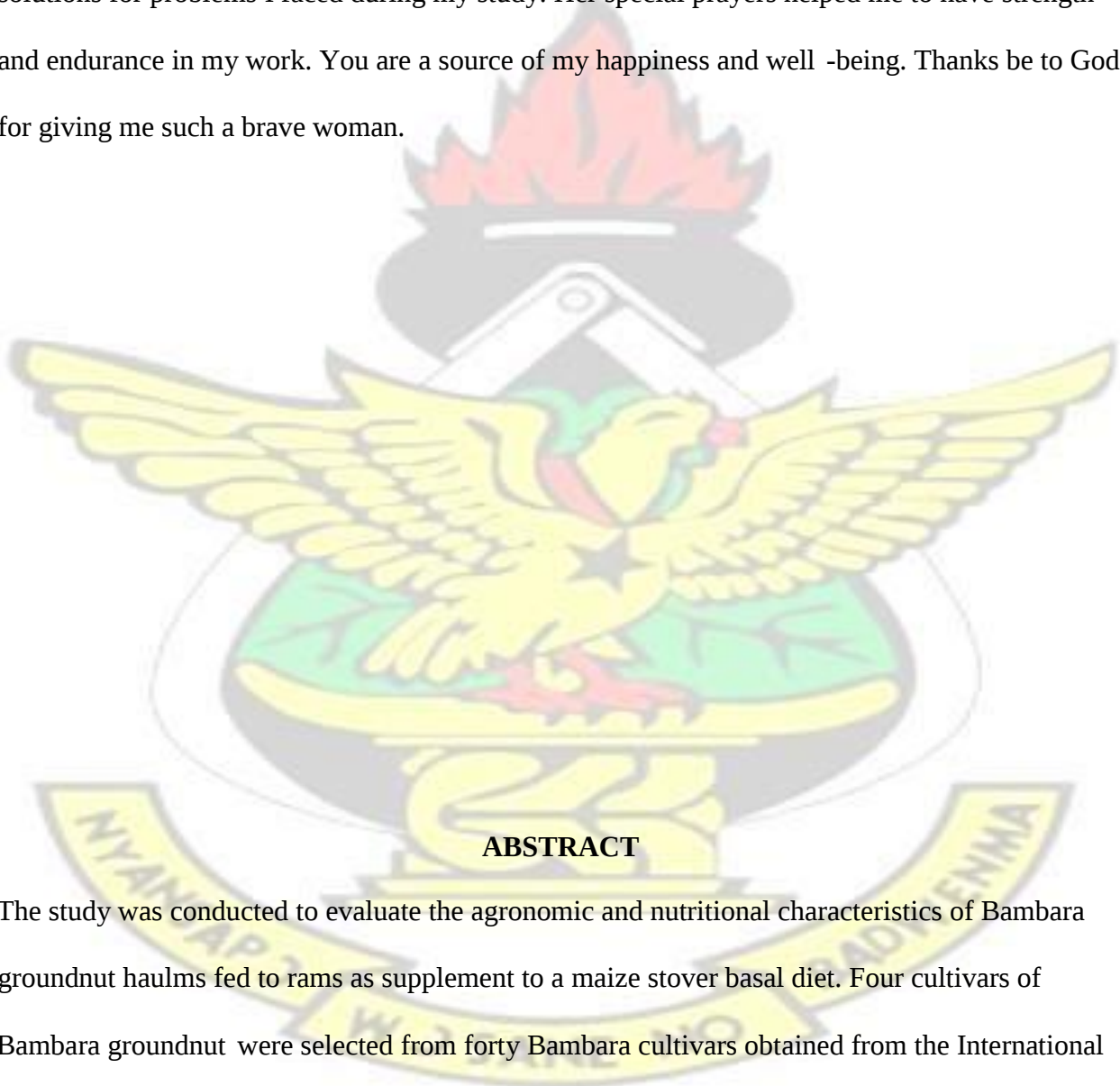
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The logo of KNUST (Kwame Nkrumah University of Science and Technology) is centered in the background. It features a yellow eagle with spread wings perched on a green shield. Above the eagle is a black mortar and pestle with a red flame. Below the eagle is a yellow banner with the text 'NYANAPAKA WADJENMA' in black. The word 'KNUST' is written in large, light grey letters at the top of the page.

ABSTRACT

The study was conducted to evaluate the agronomic and nutritional characteristics of Bambara groundnut haulms fed to rams as supplement to a maize stover basal diet. Four cultivars of Bambara groundnut were selected from forty Bambara cultivars obtained from the International Institute of Tropical Agriculture (IITA) based on their agronomic and nutritional characteristics for metabolism studies using four Djanllonke rams with an average weight of 15.0 ± 0.5 kg. Four

selected cultivars of Bambara groundnut haulm, TVSU (138), TVSU (879), TVSU (690), and TVSU (1446) were evaluated using the *in vitro* gas production technique. The animals were assigned randomly in a 4x4 Latin square design to one of the four test diets namely, T1 (Maize stover only, the basal diet), T2 (Maize stover + 150 g Bambara groundnut haulm), T3 (Maize stover + 300 g Bambara groundnut haulm) and T4 (Maize stover + 450 g Bambara groundnut haulm). The grain yield estimates of all the cultivars ranged from 0.21-4.9t/ha with cultivars TVSU1446 (0.54 t/ha), TVSU138 (0.73 t/ha), TVSU690 (2.3 t/ha) with TVSU879 recording the highest yield of (3.3 t/ha). The haulms yield also ranged from 2-24 haulms per stand at harvest. Whereas TVSU879 recorded the highest (15 haulms per stand), both TVSU690 and TVSU1446 recorded 9 haulms per stand and TVSU138 had the lowest haulms yield of 3 per stand. The chemical analysis indicated a dry matter range of 90.24 (TVSU138) to 90.80% (TVSU1446), crude protein ranged from 14.32 (TVSU690) to 16.15% (TVSU138), neutral detergent fibre range 47.34% (TVSU1446) to 68.45% (TVSU879). While that of acid detergent fibre ranged from 33.46% (TVSU690) to 43.65% (TVSU138). Percentage ash contents recorded were 8 for TVSU690, 9 for TVSU1446, 9.5 for (TVSU879) and 11 for (TVSU138). Dry matter intake of the basal diets increased as the supplement level increased ($P<0.05$). Cumulative gas production at 48 and 72hr was highest for TVSU (879). The highest fermentative gas production was recorded by TVSU690 (34.61) followed by, TVSU1446 (30.90), TVSU879 (23.84) and TVSU138 (21.47). There were significant differences ($P<0.0001$) among the four cultivars in terms of the gas production parameters. The rates of gas production ranged from 0.04 to 0.07 h^{-1} . Cultivar TVSU879 (0.07 h^{-1}) was significantly ($P<0.0001$) highest compared with the other cultivars which recorded the same rate of gas production of 0.04 h^{-1} . The total feed intake did not show any significant ($P>0.05$) difference at all levels of supplementation. Digestibility co-efficient was significantly lower ($P<0.05$) for T1 which had no Bambara groundnut haulm supplementation

compared to the other treatment groups supplemented with BGH which were also statistically ($P>0.05$) similar. Generally, faecal, urine and nitrogen balance values amongst the treatment groups did not differ ($P>0.05$) significantly. From the current study, it can be concluded that, Bambara groundnut haulm seems to be underutilized though could be used to supplement poor quality roughages to increase productivity of ruminant livestock in tropical regions and possibly replace the conventional feed supplements. Furthermore, the four cultivars of the Bambara groundnut haulm under study revealed that they could be valuable alternative animal feed sources in ruminant feeding.

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LIST OF ABBREVIATIONS

ADF	Acid Detergent Fibre
AP	Absorbed Protein
ANOVA	Analysis of Variance
BGH	Bambara Groundnut Haulm
CP	Crude Protein
CT	Condensed Tannins
DM	Dry Matter
DOM	Digestible Organic Matter
DMI	Dry Matter Intake
DE	Digestible Energy

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DAS	Department of Animal Science
EV	<i>Erythrina Variegata</i> et al
g	Gramme
GLM	Generalized Linear Model
GS	<i>Gliricidia Sepium</i>
HT	Hydrolysable Tannins
IITA	International Institute of Tropical Agriculture
IVGPT	<i>In Vitro</i> Gas Production Technique
IVDMD	<i>In Vitro</i> Dry Matter Digestibility
KNUST	Kwame Nkrumah University of Science and Technology
Kg	Kilogram
LL	<i>Leucaena Leucophala</i>
ME	Metabolizable Energy
MS	Maize Stover
MoFA	Ministry of Food and Agriculture, Ghana
N	Nitrogen
NPN	Non-Protein Nitrogen
NDF	Neutral Detergent Fibre
NRC	National Research Council
OM	Organic Matter
RDP	Ruminal Degraded Protein
RUP	Ruminal Un-degraded Protein
SAS	Statistical Analysis System
VFA	Volatile Fatty Acids
VSD	Veterinary Services Directorate
WALL	West African Long-Legged
WAD	West African Dwarf

And Others

FAO Food and Agriculture Organization

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CHAPTER ONE

INTRODUCTION

Ruminant livestock production, according to Nurfeta (2010) is hindered by inadequacy and low quality of feed. High level of productivity cannot be obtained since the tropical grasses which are usually given to these livestock are low or deficient in protein (Kosgey and Okeyo, 2007). It has however been reported that, when these tropical grasses are supplemented with concentrates, their intake and digestibility are improved (Nurfeta, 2010). However, such strategies are rarely adopted by smallholder livestock farmers because these farmers consider concentrates to be scarce and expensive to use. Consequently, there is limited prospect for using cereal grains and by-products as livestock feed by smallholder farmers. In order to mitigate the problems associated with the lack of protein supplement, there is a need to look for alternative protein sources that farmers can produce at their own farm without incurring additional cost.

Protein can often be a limiting nutrient for the growth of young livestock and for milk production in ruminant production systems (Minson, 1990). Legume forages have a large potential to overcome this limitation as they have higher protein concentrations than that of grasses or other forages such as maize (Goodchild, 1990). Ruminant production generally does require forages with crude protein of 100 to 170 g kg⁻¹ DM and legume forages tend to be either at the upper end of, or above this range (Minson, 1990; Groff and Wu, 2005; Dewhurst *et al.*, 2009). For instance, across a wide range of species and regions, the crude protein of forage legumes was found to be approximately 170 g kg⁻¹ DM compared to 115 g kg⁻¹DM for grasses (Minson, 1990). Bambara groundnut (*Vigna subterranean*) is an annual herbaceous, intermediate plant with creeping stems belonging to the *fabaceae* family. Amarteifio and Moholo (1998) reported that it is an underutilised crop and could make a well-balanced feed with a caloric value equivalent to that of a high quality

cereal grain. The grain of Bambara groundnut contains 11.4 % protein, 53.1 % carbohydrate, 6.1 % fat, 6.1 % fibre, 4.4 % ash, 0.097 % calcium, 0.007 % iron, 1.2 % potassium and 0.003 % sodium (Onwubiko *et al.*, 2011).

The haulm of Bambara groundnut is one of the several leguminous plant residues which could be used as a feed supplement in sheep diets to reduce cost of production and improve digestion and nitrogen balance. However, its use as a supplement has not been extensively researched on. It is therefore crucial to determine how they can contribute to small ruminants' productivity through its use as nitrogenous supplement to cereal crop residues.

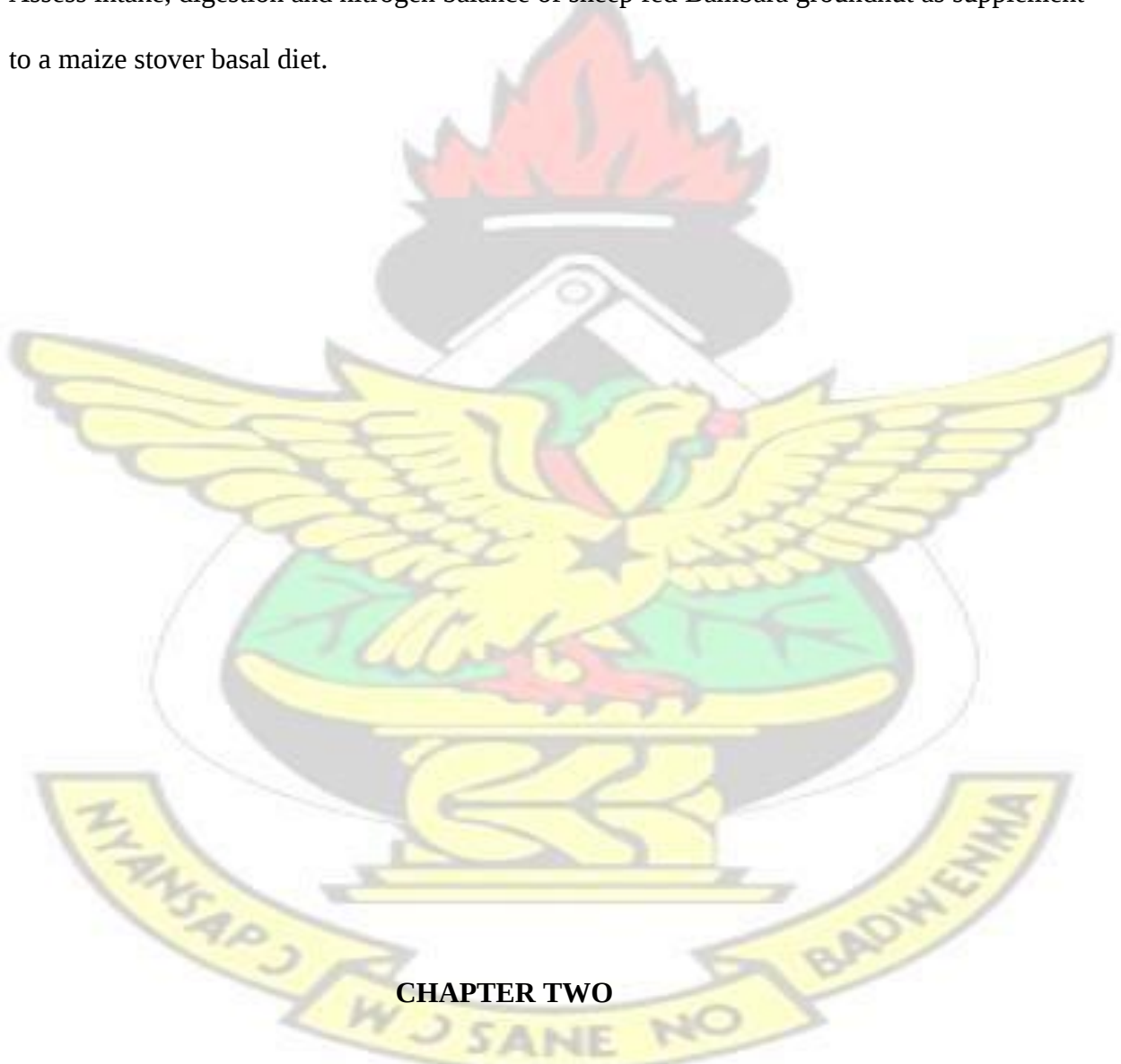
In vitro estimations of feed degradation are imperative tools for ruminant nutritionists. These methods measure either substrate loss or fermentation products (Blümmelet *al.*, 1997). It has been suggested that the gas production technique is more dependable than the nylon bag method for determining nutritive value of feeds containing anti-nutritive factors (Khazaal and Ørskov, 1993). Therefore, the study sought to evaluate the nutritive and gas production characteristics of the haulm of Bambara groundnut as well as the intake, digestion and nitrogen balance of sheep fed Bambara groundnut as supplement to a maize stover basal diet.

1.1 OBJECTIVE(S)

1.1.1 SPECIFIC OBJECTIVES

The specific objectives were to:

1. Evaluate the nutritive value of Bambara groundnut using the *in vitro* gas technique.
2. Assess intake, digestion and nitrogen balance of sheep fed Bambara groundnut as supplement to a maize stover basal diet.



CHAPTER TWO

LITERATURE REVIEW

2.1 CROP RESIDUES

Crop residues are the ingredients that are left after the main crop has been harvested. Crop residues are unconventional feed resources that are obtainable locally from crop production, which can be used as feed for ruminant livestock. In this backdrop, crop residues, (4 billion metric tons globally) comprising mainly of straw from fine grains such as wheat, oat, rice etc. and stover from coarse grains such as maize, sorghum, millets etc. obtained after harvesting the crops, constitute some of the ruminant feed sources. Jutzi (1993) disclosed that ruminants fed on the remains of crop after harvesting. By-products and arable weeds are basically misused in the absence of a ruminant component in the system. Roughage and cereal crop remains are excellent feed for ruminant livestock production (Wora-Anu *et al.*, 2000). Crop residues have been used as ruminant feed since millennia (Schiere, 2010). For the most part, cereal crop residues are of low nutritive value (Sundstol and Owen, 1984) in light of their moderately low digestibility (<500 g digestible organic matter (DOM) per kg dry matter (DM), low crude protein (<50 g/kg DM) as well as low available minerals and vitamins content. These deficiencies combine to make crop residues unpalatable, thus their intake is also low (usually less than 15 g DM/kg live weight daily). Some straws such as legume crops have largely better nutritive value, forage value and thus are nutritionally higher than cereal straws. Stover have better nutritional quality than straws with respect to intake and organic matter digestibility (>50% vs. <50%) (Walli, 2004).

Walli (2004) had reported that straws and stover are generally used to feed low producing animals or can likewise serve as a source of energy in the ration of the high producers' ration to satisfy their desire for food, which assists effectively in reducing the fibre deficiency for milk fat synthesis in high concentrate feeding systems and may give extra heat increase during cold stress conditions.

Even though improvements in digestibility and nitrogen level have been noticeable, inherent dry matter losses (up to 40%) cannot be eradicated and the know-how is not reasonable (Mahesh and Mohini, 2013). Conversely, a sizable variation in the quality of straws in terms of digestibility as well as metabolizable energy exists among different varieties of rice, wheat, barley and stovers of sorghum, pearl millet as well as groundnut haulms (Krishnamoorthy *et al.*, 1995; Blümmel *et al.*, 2010). Therefore, to have varieties with superior stover value without affecting grain yield (i.e., general ratio of straw: grain is 1:1 to 1:3), studies must be spread beneath food-feed crops research which would also have the added advantage of lowering methane emission by ruminant enteric fermentation similar to catalytic supplements (Walli, 2004).

Yuangklang *et al.* (2005) reported that low fibre-low protein feeds are acquired from renewable energy crops such as sugarcane by-products and root crops. In addition, low fibre-low protein feeds are mostly rich in energy and low in protein. Some examples are oil palm slurry and molasses waste material arising from the fruit processing industry (citrus pulp, pineapple waste and tomato pomace) and cassava root crop (Ngamsaeng, 2005). Low digestibility is a common characteristic of such feeds, including low protein content and a low mineral content. The available amounts tend to be site definite. Most countries rely on the availability of these feed resources, whose quantity and quality are most likely to be variable and limiting.

In Ghana, feed resources available for ruminant livestock during critical period of feed scarcity can be described as high fibre-low protein feeds and include residues arising from cereal crops, such as rice, millet, sorghum and maize (Alhassan *et al.*, 1999). These residues are characterised by high fibre content (>700 g of cell wall material/kg DM), low metabolisable energy (<7.5 MJ/kg DM), low levels of crude protein (20–60 g of crude protein/kg DM) and low to moderate digestibility (<30–45% organic matter digestibility) (Konlan *et al.*, 2016). The daily intake of the small

ruminant is often limited to less than 20 g dry matter/kg live weight and most of the residues are also deficient in fermentable carbohydrates, reflected by the relatively low organic matter digestibility (Alhassan *et al.*, 1999). Another category of available crop residue is less fibre-high protein feeds such as residues derived from legume crops (tops and haulms from groundnut, cowpea, and soya beans) and agro-industrial by-products such as bran from rice and maize and brewer's spent grain. They are generally less fibrous (below 700 g but above 400 g of cell wall material/kg DM) than those in the first category but have relatively high amounts of crude protein (> 60 g/kg DM) (Alhassan *et al.*, 1999). The leaves from leguminous trees and browse plants such as *Glyricidia*, *Leucaena*, *Ficus sp* and *Erythrina* have been reported to contain around 250–350 g of crude protein/kg DM, and can also be considered in this category (Alhassan *et al.*, 1999).

Maize stover is low in protein that varies from 2.3% (Kabatange and Shayo, 1991) to 7.1% (Woyengo *et al.*, 2004) and, thus, requires supplementation. Treatment of the stover before feeding may be essential to improve stover utilization by animals (Woyengo *et al.*, 2004). Four mature Anglo-Nubian×Fiji local goats with pre-experimental body weight of 25.0 ± 0.6 kg, and 22–24 months old, were used to study the effect of supplementation of a basal diet of maize stover with *Erythrina variegata* (EV), *Gliricidia sepium* (GS) and *Leucaena leucocephala* (LL) on dry matter intake (DMI) and nutrient digestibility (Aregheore and Perera, 2004). The dry matter intake (DMI) of the urea treated stover diet was significantly lower ($P < 0.05$) than that of the diets of untreated stover supplemented with forage legumes. The DMI was significantly lower ($P < 0.05$) in the GS diet than in the EV or LL diets. Significant ($P < 0.05$) differences existed between the urea-treated stover and the diets of stover supplemented with forage legumes in the digestibility of dry matter (DM), crude protein (CP), neutral detergent fibre (NDF), organic matter (OM) and energy. Butterworth and Mosi (1986) and Alayu (1987) also reported increased digestibility of low quality

roughages after protein supplementation. This increased digestibility could lead to higher productivity in terms of weight gain and wool growth as observed by Cronje and Weites (1990) when they supplemented wheat straw with cotton seed cake. Besides, Oosting (1993) fed sheep on wheat straw (with high lignin content of 75 g/kg) and found that nitrogen (N) supplementation did not increase the DM digestibility of the straw (372 vs. 360 g/kg).

2.1.1 Techniques of improving nutritive value of crop residues

Ruminants fed on crop residues, by-products and arable weeds increase the value of resources which are to a great extent wasted without a ruminant component in the system (Jutzi, 1993). The intervening limitation on feed supply in essential tropical ruminant livestock zones is the seasonality in the availability and quality of animal feed supplies harmonizing with dry and wet seasons (Awad and Elhadi, 2010). Large-scale crop residue usage is unusual in southern Nigeria because of low level of livestock farming yet a considerable measure of residue are produced from cropping and from households (Jutzi, 1993). In the northern grassland environment, the opposite is the situation. Pastoralists use significant amounts of crop residue while their stock returns manure to the soil. Little scale ammonification treatment of maize and sorghum strains has been initiated in Northern Nigeria (Kano) to enhance the feeding quality of the straw. Crop by-products have in like manner been utilized to decrease the feed expense of dairy goats (Reed and Brown, 1988). Much attention has been put on straw upgrading procedures utilizing treatment with chemicals (Owen and Jayasuriya, 1989) sodium hydroxide or ammonia in temperate nations and urea-ammonia in tropical nations. Treatment with these chemicals increases digestibility by 100 to 150 g DOM/kg DM and expands intake by around 4 g DM/kg live weight/day (25%). The three alkaline treatments are portrayed briefly as:

- a. ***Sodium Hydroxide (NaOH) treatment:*** Several Sodium Hydroxide treatment strategies to enhance the utilization of crop residues for ruminant feeding have been developed as

studied by Jackson (1977), Berger et al. (1994) and Arieli (1997). The key points of interest of the diverse NaOH treatment strategies are increased degradability and palatability of treated straw, compared with untreated straw (Chaudhry and Miller, 1996; Vadiveloo, 2000). Though, Sodium Hydroxide is not generally available as a resource for small scale farmers and might be excessively costly, making it impossible to utilize. Likewise, the utilization of NaOH can be a reason for ecological contamination, bringing about a high substance of sodium in the environment (Sundstøl and Coxworth, 1984).

- b. **Ammonia (NH₃) treatment:** Treatment of straw with anhydrous and aqueous ammonia, urea or other ammonia-releasing compounds has been generally explored to enhance degradability (Abou-EL-Enin *et al.*, 1999; Selim *et al.*, 2002; Fadel-Elseed *et al.*, 2003). The principle of ammonia treatment should be like that of NaOH treatment. Ammonia treatment expands the degradability of the straw, as well as includes nitrogen (AbouELEnin *et al.*, 1999) and preserves the straw by hindering mold development (Calzado and Rolz, 1990). Also, improvement in degradability of structural carbohydrates, ammonia treatment is a successful method for reducing the amount of supplemental nitrogen, decreasing the expenses of acquiring protein-rich feedstuffs, and improving acceptability and voluntary intake of the treated straw by ruminants. Though relative studies in enhancing the energy value of straw have demonstrated that ammonia treatment is less effective than NaOH (Liu *et al.*, 2002), its utilization might be more profitable for farmers as the additional ammonia serves as a source of nitrogen. In a past study using sheep, Selim *et al.* (2004) treated rice straw gathered in polyethylene bags for 4 weeks with gaseous ammonia (3 g NH₃ per 100 g dry matter). The excess ammonia was expelled before offering the straw to animals. The ammonia treatment increased the nitrogen content in the

rice straw from 8.16 to 18.4 g kg⁻¹ (crude protein content increased from 51 to 115 g kg⁻¹).

The ammonia treatment little bit reduced the NDF content from 571 to 551 g kg⁻¹, as a result of dilution with the extra N, however increased the ADF content from 303 to 327 g kg⁻¹, showing that the cell wall properties were changed.

- c. **Urea treatment:** Rice straw can likewise be treated with urea, which releases ammonia after dissolving in water. For practical use by farmers, urea is more secure than utilizing anhydrous or aqueous ammonia and furthermore gives a source of nitrogen (unrefined protein) in which straw is lacking (Schiere and Ibrahim, 1989. Since urea is a solid chemical, it is likewise simple to handle and transport (Sundstøl and Coxworth, 1984) and urea can be acquired effortlessly in numerous developing nations. Additionally, urea is impressively less expensive than NaOH or NH₃. Vadiveloo (2003) reported that rice varieties with a low degradability responded better to urea treatments over higher quality straw, increasing the *in vitro* dry matter degradability from 45 to 55-62%. Urea treatment may in this way be most appropriate for small-scale farmers to enhance the quality of straws, especially assortments demonstrating a low degradability.

2.1.2 Microbial activity on crop residues

For appreciable microbial digestion of plant materials to happen in the rumen, a nearby physical affiliation is crucial between the plant tissue and the organisms in charge of the digestion (Cheng *et al.*, 1983/84; Orpin, 1983 /84). It has been known that enzymic activity is liable to be relative to the mass of cellulolytic microorganisms. Yates (1984) has discovered for cotton thread, that the rate of cellulose digestion is connected with the mass of attached colonizing microbes, in supporting this hypothesis. Smallholder farmers in developing nations have mostly acknowledged the benefits to dairy cattle of including a little measure of fresh green herbage to straw-based diets

(Leng, 1990). There are various helpful impacts in these practices, for example, the supply of vitamin A and key minerals and of ammonia and peptides/amino acids.

In recent times, the role of supplements on the digestibility of a low quality basal diet has been examined (Ndlovu and Buchanan-Smith, 1985; Silva and Ørskov, 1988). These studies showed that where the supplemental forage in a straw-based diet given to sheep was of high digestibility, an expansion to digestibility of the basal diet happened even at moderately little levels of supplementation. According to Cheng *et al.* (1990), which reported that the rate of digestibility of straw rests on the rate and degree of colonization of fibre and the biomass of adherent organisms. It has always been expected that colonization of fibre entering the rumen is from the free-floating pool of microbes in the rumen. Krebs *et al.* (1989) recommended that colonization of microorganisms happens from fibre to fibre without going through the free-floating pool. Then again, clarification given by Leng (1990) to this proposal was that the valuable impacts of the incorporation of high digestibility forage in an otherwise low-digestibility forage diet could be that this applies a vast impact on digestibility by giving an exceptionally colonized fibre source to "seed" microorganisms onto the less digestible fibre.

Supplementation with legume crop residues adds fermentable energy to the rumen in the form of available cellulose and hemicellulose which stimulate fibre digestion (Silva and Ørskov, 1985).

Bauchop (1981) reported that it is likely that offering such material before the daily feeding of straw may instigate a more prominent level of colonization of straw by rumen bacteria and by rumen fungi, which have been included in the breakdown of fibre. Extra factors might be included. For instance, Ørskov and Dolberg (1984) expressed that if animals fed on untreated straws or low quality roughages are supplemented with substrates which increase the fermentation rate of

cellulose, the rumen environment becomes related to that of animals getting ammonia treated straws.

Ndlovu, (1991) expressed that, one of the greatest encounters in feeding low quality fodders to ruminants is to enhance their intake. Organic treatments, while successful, present various practical issues for smallholder Farmers. The addition of higher value feeds, for example, legume forages to a low quality basal diet is more practicable. It is known that the degree of substitution of the basal diet by the forage legume, resulting in reduced intake of the basal diet, relies on the level of addition of the supplement (Ndlovu, 1991).

2.2 FORAGE LEGUMES

Legume forages are considered more imperative for use as high quality forage for livestock, both in cultivated pastures and in naturally occurring associations. Tropical fodder legumes are important in the sustenance of small ruminants (goats and sheep). Nutritionally, they are 2-3 times richer in protein than cereal grains. There is a surging concern in the use of leguminous trees as a source of high quality feed for grazing and as a supplement to improve the output of ruminants fed poor quality roughages. Leguminous trees are usually long-lived and have low maintenance requirements and, hence, augment the sustainability of farming systems (Gutteridge and Shelton, 1994). While legumes serve good protein supplements to low quality fibers, they are constrained by their high content of anti-nutritional factors, arising predominantly from secondary metabolism in plants. Tree legumes have wide range of plant structures: fodder from leaves, twigs and pods, as well as shade, live fences, timber and firewood. Sunken pods from the rain tree are highly palatable to livestock (Kathaperumal *et al.*, 1988).

Anti-nutritional factors such as condensed tannins (CT) inhibit plant protein degradation and decrease sulphur availability in the rumen, which in turn affect the digestibility of total tract nitrogen (Animut

et al., 2008) and plant cell walls (Aganga and Tshwenyane, 2003). Duttan *et al.* (1999) also observed a depression in the value of CP and OM degradability in goats under a basal diet of rice straw supplemented with *Prosopis cineraria* because of the high contents of tannin in the leaves. Hence, understanding of concentration of limiting factors in forage legumes and identifying potential inclusion levels in ruminant diets would be a good approach of utilizing such resource efficiently by decreasing nitrogen excretion (McMahon *et al.*, 1999). Also, antinutritional factors affect ruminant animal production by decreasing feed intake and by producing derivatives which reduce rumen microbial activity and decreases growth (D'Mello and Devendra, 1995). However, the seed of *Mucuna* has been used without any adverse health (Buckles, 1995), though Topps and Oliver (1993) reported that the pods have an unduly laxative effect if fed at more than 2 kg per day to cattle. Gutteridge and Shelton (1994) observed that tannins in fodder legumes reduce the digestibility of herbage and protein, and phylloides (expanded and flattened leaf petioles) of some species, instead of pinnate or binnate leaves, are very high in fibre have the ability to lower digestibility.

2.2.1 Underutilised forage legumes

In numerous African nations ruminant productivity is low as far as milk and carcass yield compared to that obtained in developed nations. The unlimited feed resource that offer the bulk of ruminant feed are biomass from grassland and crop residues which cannot sustain the animals' output especially throughout the dry season. This poor nutrition leads to economic losses to the farmers because of loss in weight and condition of animals, poor reproductive performance and increase rates of mortality mainly the young animals (Iheshiulor *et al.*, 2011). Better feeding systems based on supplementation of grass with forage legume (Babayemi *et al.*, 2006) will expand milk yield and meat production.

There is the need to supplement legumes in the diets of livestock species in order to boost their productivity. Under-utilized grain legume foliage like African Yam beans (*Sphenostylis stenocarpa* Hochst ex A. Rich) Harms, Lima beans (*Phaseolus lunatus*) sword beans (*Canavalia gladiata*), pigeon pea (*Cajanus cajan*), Bambara groundnut (*Vigna subterranean*), Jack beans (*Canavalia ensiformis*), Dolichos Lablab (*Lablab purpureus*) could be explored to ascertain their effect in livestock feeding (Ajayi, *et al.*, 2009). These legumes are classified as minor grain legumes because they are under exploited and are native legumes usually cultivated in mixed farming with arable crops like cassava, yam, etc. (Adeparusi, 2001; Fasoyiro *et al.*, 2004).

These legumes are widely grown in Nigeria and in other West African countries like Ghana, Cameroon, Ivory Coast and Togo for the seeds and at the end of the growing season, the foliage are burnt (Klu *et al.*, 2001).

In addition, the main problem facing livestock farmers in tropical areas is the right nutrition for their animals during the dry season when pastures, cereal residues and maize stover are limiting in nutritional quality. Usually, it is during this season that problems such as disease and weight loss due to a poor dietary profile ascend. One way of improving the utilization of such crop residues is by proper supplementation with leguminous forages (Poppi and McLennan, 1995).

Harricharan *et al.* (1988) disclosed that forage legumes can be grazed, harvested and fed fresh or stored as hay or silage. A sustainable way of enhancing the feeding value of poor quality crop residues and pastures, particularly for resource poor small holders, is through supplementation with forage legumes. Despite the fact that there are a few forage plants that have the ability to produce exceptional returns of dry matter, they contribute less to the much needed improvement of livestock production, since information on their nutritive qualities are uncommon (Barro and Ribeiro, 1983).

2.2.2 Nutritive values of forage legumes

Tree forages that have a low neutral detergent fibre (NDF) content (200 – 350 g/kg) usually have high digestibility (Norton, 1994) with high crude protein content (140 – 300 g/kg) compared to that of crop residues (30 – 100 g/kg) (Teferedegne, 2000). Legume forages for the most part prompt higher intake and animal productivity than grass of equal digestibility. This is whether they are silages (Dewhurst *et al.*, 2003) or grazed herbage (Fraser *et al.*, 2004). Harris *et al.* (1998), affirmed that the value of incorporating white clover in zero-grazed herbage was lost when feed intake was restricted. Grazing of forage legumes led to increase growth rates and a significant reduction in time to slaughter of sheep (Speijers *et al.*, 2004). These grains were accomplished without negative impact on carcass characteristics; to be sure, carcass weight and slaughtering out percentage were increased significantly for sheep grazing red clover. Ruminal nitrogen and digestibility were enhanced when dairy cow were fed lucerne (Lee *et al.*, 2002). Scalbert (1991) reported that plant species which contain some tannin give both degradable and undegraded rumen nitrogen (N) and are more active sources of supplemental nitrogen for ruminants.

Forage protein serves as a source of absorbed protein (AP) to the ruminant by giving both ruminal degraded protein (RDP) for microbial protein synthesis in addition to ruminal undegraded protein (RUP) that escapes microbial breakdown. Rapid and extensive ruminal degradation of proteins in legume and grass forages for the most part lead decreased protein efficiency. Forage NPN is available as both protein and non-protein nitrogen (NPN). High quality silages frequently contain excessive measures of NPN (Muck, 1987). Broderick (1994) described that the mean chemical values for alfalfa silage fed in 19 lactation trials were 54 (10%) NPN (total N basis) and 20.4 (1.41%) CP and 43 (51%) NDF (DM basis) for alfalfa harvested at an average 43 (91%) dry matter (DM). In an research to determine the nutritional potential of two leafy vegetables (*Moringa*

oleifera and *Ipomoea batatas*), Oduro *et al.* (2008) reported that *Moringa oleifera* leaves contained 27.51% crude protein, 19.25% crude fibre, 2.23% crude fat, 7.13% ash, 76.53% moisture, 43.88% carbohydrates, and caloric value of 1296.00 kJ/g (305.62 cal/g). Calcium and Iron content in mg/100 g (DM) were 20.09 and 28.29, respectively. They resolved that *Moringa oleifera* leaves could support to the nutrient requirements of livestock. Studies have revealed that supplementation of teff straw with either *Sesbania* or *Leucaena* sp. increases the fractional rate of passage of particulate matter by 23-53% and of liquid phase by 9-43% (Bonsi *et al.*, 1994; Umunna *et al.*, 1995) and also, supplementation of teff straw with graded levels of cowpea or *Lablab* significantly increased microbial N supply in calves. The number of microbial protein formed in the rumen is correlated to dietary DE rather than protein (Broderick and Merchen, 1992). Though, there is a limit to the amount of DE that can be fed to dairy cows since of problems (e.g., low ruminal pH) that may result from high ration fermentability. There is a trend toward increased grazing of legume and grass pastures in the United States; N consumption by grazing ruminants is unusually ineffective (Beever, 1982). Poor usage of dietary N in ruminants, particularly dairy cattle, has led to increased concern about the support of excreta to N pollution of the surroundings (Tamminga, 1992).

2.3 BAMBARA GROUNDNUT

Bambara groundnut (*Vigna subterranean*, L. Verdc) is an indigenous legume that is widely grown by subsistence and small-scale farmers in sub-Saharan Africa. Bambara groundnut belongs to the family Fabaceae, sub-family Papilionoideae, and it is the third most important legume after groundnut (*Arachis hypogaea*) and cowpea (*Vigna unguiculata*) in semi-arid Africa (Hocking, 1994). It bears protein-rich and nutritious seeds, capable of growing in poor soils and tolerant to drought stress (Heller *et al.*, 1997), allowing Bambara groundnut to become a potential crop in

easing future global food security issues. The centre of origin of Bambara groundnut has been suggested to be the region between north eastern Nigeria and northern Cameroon, where the wild form of Bambara groundnut were found (Begemann, 1988). The domestication is believed to have occurred within Jos plateau and Yola regions, towards Garoua in Cameroon and probably even Central African Republic (Begemann, 1988).

Bambara groundnut has been widely cultivated in tropical regions since the 17th century. In addition to Nigeria, Ghana, Burkina Faso as well as Eastern Africa and Madagascar. Bambara groundnut is also grown in South America, Oceania and Asia such as Indonesia, Malaysia, Philippines, India and Sri Lanka (Linnemann and Azam-Ali, 1993).

2.3.1 Agronomic characteristics of Bambara

Bambara groundnut is an annual, herbaceous, intermediate legume of up to 30 cm-35 cm in height with well-developed tap root and lateral roots under the soil (Heller *et al.*, 1997). The roots form nodules in association with Rhizobia for nitrogen fixation (Heller *et al.*, 1997). General appearance of the crop is trifoliate leaves with erect petiole grown from short, creeping, multibranched lateral stems on the ground level (Heller *et al.*, 1997). Each lateral stem has numerous nodes and the distance (or the length of branch) from the base of the plant to the nearest node is always shorter than the more distant ones (Heller *et al.*, 1997). Due to the length of internodes, Bambara groundnut landraces differ from each other in terms of growth habit, ranging from spreading, semibunched to bunch types (Chijioke *et al.*, 2010). The petioles that are borne from the nodes are long, stiff and grooved, with a base of a range of colour such as green, purple and brown (Swaminathan *et al.*, 2012). In contrast, wild forms of Bambara groundnut exhibit a slightly

different appearance in which they have a spreading growth habit, limited numbers of elongated lateral stems and no distinct tap root with penta-foliolate leaves (Basu *et al.*, 2007).

According to Brough and Azam-Ali (1992), that Bambara groundnut seed makes a balance food as it contains adequate amounts of carbohydrate (63%), protein (16.25%) and fats (6.3%) with relatively high extents of lysine and methionine as percentage of the protein (6.6 and 1.3% respectively). The essential amino acid content of Bambara groundnut, for example, lysine (6.82g/16gN), methionine (1.85g/16gN) and cysteine (1.24g/16gN) is comparable to that of soya bean (6.24g/16gN) lysine (1.14g/16gN), methionine and (1.80g/16gN) cysteine (Fetuga *et al.*, 1975).

2.3.2 Nutritional characteristics of the haulm of legumes

One of the significant difficulties when feeding low quality forages to ruminant is to increase their intake (Ndlovu, and Hove, 1995). The impacts of incremental levels of groundnut haulm as supplements to a low quality basal diet have been explored (Ehoche, 2002; Aregheore, 2009). Alhassan (1985) in a relative investigation of maize residues with other crop residues fed 1-1.5 year old red Sokoto goats with different cereal or legume residues observed that dry matter intake range from 0.7% of body weight for maize stover to 2% for sorghum leaves (5.5% CP), while leguminous crop residues consumption range from 0.8% body weight for cowpea vines (5.9% CP) to 3.4% body weight for groundnut haulms (16.7%CP). Adebawale (1988) recognized the advantageous impacts of the groundnut haulms, to incorporate increment in metabolisable energy and nitrogen consumption, enhanced palatability, expanded available minerals and vitamins, better rumen function and a laxative impact on the alimentary canal. Groundnut haulms and shells are by products which are generally used to fatten animals in northern Nigeria. Adu and Lakpini (1983) fed chopped groundnut haulms solely, to growing Yankasa lambs and recorded live weight gains

of 90.2g /day for unchopped haulms. Supplementary feeding of groundnut haulms to cows improved growth rates of suckling calves compared to the non-supplemented control group which was attributed to increased milk consumed by calves arising from the increased milk output when their dams were supplemented (Ehoche *et al.*, 2001). Nicholson (1989) observed that in partial milking system, approximately 60% of the milk produced is consumed by the calf. When feeding maize residues was compared with other cereal or legume crops, it was found that live weight gain compared favorably.

Higher feed consumption was recorded for maize residue, although this was not significantly higher than the sugar cane tops. However, the high consumption did not produce better live weight gain, except with sorghum stalks when fed to Red Sokoto goats with groundnut haulms (Alhassan, 1985). Topps (1995) demonstrated that the enhanced intake due to supplementation of groundnut haulms in animals to some extent reduce weight losses.

For considerable microbial digestion of plant materials to happen in the rumen, a close physical relationship is key between the plant tissue and the microbes in charge of the digestion (Orpin, 1984). Reports have revealed that where the supplemental forage in a straw-based diet was given to sheep, enhancement in the digestibility of the basal diet happened even at moderately small levels of supplementation possibly as a result of the rate and degree of colonization of fibre and the biomass of adherent organisms (Cheng *et al.*, 1990). As it had been reported by Kreb *et al.* (1989), that colonization of bacteria occurs from fibre to fibre without passing through the freefloating pool and concluded that fibres are colonized by free- floating pool of bacteria in the rumen.

Leng (1990) clarified that the advantageous impacts of the inclusion of high digestibility forage in an otherwise low digestibility forage diet could be that it applies a vast impact on digestibility by

given an extremely colonized fibre source to 'seed' bacteria onto the less digestible fibre. Supplementation with groundnut haulm contributes fermentable energy to the rumen in the form of available cellulose and hemi-cellulose which stimulate fibre digestion (Silva and Orskov, 1988).

Bauchop (1981) opined that it is conceivable that offering such material before the daily feeding of straw may stimulate a more significant level of colonization of basal diet by rumen bacteria and fungi, which have been involved in the breakdown of fiber. Manyuchi *et al.* (1994) reported that groundnut hay did not modify the in Sacco degradation of low quality grass hay. It is likely that any adjustment in the degradation of the basal diet as a result of an expansion in microbial activity rely upon the quantity of available sites for microbial connection as described by Cheng *et al.* (1990).

2.3.3 Factors of plants that affect intake

The main differences in nutritional systems of grazing and housed ruminants have been defined by McDonald (1968) and Osuji (1974). The type of food eaten will differ chemically and physically, for instance, in water content, proportions of leaf to stem, type and concentration of carbohydrates, and protein constituents. It has been demonstrated that energy outflow and the requirement for nutrients is distinctly affected by the grazing animal's environment (Osuji, 1974). Moreover, Makkar, (1991) reported that anti-nutritional factors in food are substances which either by themselves or through metabolic products in the system, interfere with food consumption or interrupt the health and production of animals. Among the few separated anti-nutritional factors which cause losses in the animals business are: tannins, mycotoxin, mimosine, cyanogen and nitrates. According to Mehanson *et al.* (1987) tannins are water soluble phenolic compounds of plants with a molecular weight equal to or greater than 500 dalton and with the capability to precipitate gelatin and other proteins in aqueous solution. Hydrolysable tannins (HTs) and

condensed tannins (CTs) are the two sorts of these compounds which might be separated by their structure and reactivity towards hydrolytic reagents. The main anti-nutritional effects of tannins present in forage, tree and shrub legumes are: decrease in voluntary feed intake, reduced digestibilities of nutrients and opposing effects upon rumen metabolism.

The identification of the above range of factors is the main forage intake assertion whether the animals will actually eat the diet. Though, during evolution plants have developed survival approaches to prevent them being eaten by voracious herbivores or in some instances also making use of them to spread their roots or seeds. During some growth stages, the animals are discouraged from eating them while in others they may be encouraged. Diverse herbivores have additionally developed survival plans, similar to the ability to choose certain parts of the plants or to develop microbial population fit for minimizing anti-nutritive factors, for example, the microbial destruction of mimosine (Jones, 1981) and a few tannins from tanniferous plants (Brooker *et al.*, 1993). Subsequently, Khazaal and Ørskov (1993) utilized the simple yet effective gas evolution procedure of Menke and Steingass (1988) to recognize anti-nutritive factors. The difference in gas evaluation with and without a compound which compressed anti-nutritive tannins provided a measure of the extent to which fermentation was withdrawn.

2.3.4 Degradation of forages

The low protein content of tropical grasses and crop residues has been cited as a major constraint to animal production (Egan, 1997; Minson, 1990). Ruminant diets need to contain adequate amounts of protein that would be degraded in the rumen to energise the rumen microbes to the fullest extent for fermentation, to ensure a sufficient amount microbial protein reach the intestine to fulfil amino acids requirement of the animal (Siddons *et al.*, 1985; Hvelplund and Madsen, 1990). The composition of diets depends on the real measurement of crude protein level in feed

materials which degrade in the rumen (Broderick *et al.*, 1988; Tamminga *et al.*, 1991). High producing ruminants supplied with some individual amino acids under some situations meet their needs (Schwab *et al.*, 2007). Degradation of proteins in the rumen by bacterial enzymes (protozoa lapidate) produces peptides and amino acids and ammonia, which is one of the main sources of nitrogen. Rumen bacteria are essential in this process (Orskov, 1982; Stern *et al.*, 1994). The bacteria also have the ability to decompose cellulose (Lindberg, 1985; Wallace and Cotta, 1988). On the other hand, protozoa has the capacity to degrade the protein also (Russell and Hespell, 1981). Type and number of microbes affect the rate of degradation of protein in the rumen. It is important to provide a sufficient amount of protein to be degraded in the rumen to meet the needs of the bacteria to produce the largest amount of microbial protein with essential amino acids (Stern *et al.*, 1994; Klopfenstein *et al.*, 2001). Crude protein in the feed is important as a nitrogen source in the rumen (Orskov, 1982) and in the feed which suffers from a lack of protein degraded in the rumen, like most grains, will limit the fermentation of microbes, which has a negative impact on digestion of fiber in the rumen (Martin-Orou *et al.*, 2000). There is little benefit to raise the level of protein degradable or un-degradable when formulating diets with higher levels of protein desired (Sloan *et al.*, 1988). Foods containing low degradable proteins in the rumen are particularly important for ruminants that need high protein level in their diet (Broderick *et al.*, 1991).

In addition, feed crude protein (CP) can be divided into an undegradable fraction, a potentially (slowly) degradable fraction and a rapidly degradable fraction (NRC, 2001). Degradability of a feed CP is determined by the fraction that is undegradable, while degradation or disappearance of CP is determined by the relationship of rate of degradation and rate of passage out of the rumen (Van Straalen and Tamminga, 1990; Broderick *et al.*, 1991), with the latter mainly influenced by dry matter intake (DMI). The National Research Council (NRC) applies a discount factor to

decrease energy value and ruminal CP degradability at high intakes (NRC, 2001). For example, when DMI increases from 25 to 30 kg/day, the metabolisable energy (ME) value of the feed is discounted by 4% and the diet CP degradability is decreased by about 3% (NRC, 2001). Factors affecting the amount of CP degraded in the rumen include the amount of CP ingested, solubility of CP in rumen fluid and the time the CP is retained in the rumen (Netemeyer *et al.*, 1980). Differences in degradability can be caused by differences in the rumen environment and by differences in resistance to proteolytic enzymes (Ørskov, 1992). Degradability of CP varies among feeds, within feeds and due to chemical or physical treatments of the feed (Lindberg, 1985; Madsen and Hvelplund, 1985; Ørskov, 1992).

Therefore, degradation of crude protein in a mixed diet can be manipulated by selecting ingredients with high or low degradability (Tamminga, 1979; Van Straalen and Tamminga, 1990). When diets are often balanced for rumen degradable crude protein (RDP) and rumen undegradable crude protein (RUP), average or book values for these fractions of individual feeds are used (Stallings *et al.*, 1991; Aldrich *et al.*, 1996). The degradability of the diet can be predicted using CP degradability values of individual ingredients (Stallings *et al.*, 1991; Aldrich *et al.*, 1996).

2.4 IN VITRO GAS PRODUCTION TECHNIQUE AS A TOOL TO EVALUATE FEEDSTUFFS

In the rumen, feed fermentation is associated with the evolution of gas, principally carbon dioxide and methane. On the assumption that the quality of gas produced from in vitro incubation of feedstuffs with rumen fluid is closely correlated to digestibility, and therefore the energy value of feed to ruminants, recommended the production system to evaluate feedstuffs (Menke *et al.*, 1979). In this system, the substrate is incubated (the incubation media are rumen fluid and a buffer) in a calibrated gas tight glass syringe fitted with a plunger to allow gases evolved (CH₄ and CO₂) to be

retained and recorded manually over a selected time depending on the type of substrate being incubated. Based on the volume of gas gathered over time, different empirical equations were established to predict *in vivo* digestibility from chemical composition and *in vitro* gas production technique (Menke and Steingass, 1988).

Different approach adopted the pressure transducer procedure to measure accumulated head-space gases resulting from microbial fermentation (Wilkins, 1974). Theodorou *et al.* (1994) further exploited the pressure transducer technique and recognized the potential of this technology to offer the advantage of studying the fermentation kinetics of soluble and insoluble fractions of feed.

Consequently, the procedure provides an estimate of rate and extent of feed digestibility. Fermentation of feed in this system results in the production of short chain fatty acid, microbial biomass and gases (CO_2 and CH_4). Gas produced arises directly from substrate degradation by rumen microbes and indirectly from the reaction of volatile fatty acid (VFA) end products with the bicarbonate fraction of the buffer used in media preparation (Beuvink and Spoelstra, 1992). The gas produced gives indications the digestibility of the feeds.

McBee (1953) at first built up the *in vitro* gas production technique (IVGPT) and it was enhanced by (Hungate, 1966). Later on, Tilley and Terry (1963) prescribed the two phase strategy to appraise *in vitro* digestibility and to reduce the amount of feed required to assess feedstuffs. The *in vitro* gas production technique was developed to predict fermentation of ruminant feedstuffs (Rymer *et al.*, 2005). Researchers from various parts of the world and from various fields have been using *in vitro* gas procedure due to the possibility to study on the effect of livestock production on the environs (Krishanmoorthy *et al.*, 2005). As indicated by Dahnoa *et al.* (2004), feed assessment and studies including ruminal fermentation have utilized the *in vitro* fermentation technique. This technique yields dependable measurements of rates of fermentation of fiber that can be utilized to

decide energy availability of feeds. The accuracy of the outcomes is a product of the precision and reproducibility of the approach (Tilley and Terry, 1963). Thus, in a technique such as this a mistake toward the beginning of the procedure can change the effect of the investigation. Getachew *et al.* (2004) have conveyed that, ruminant efficiency is connected with the precision of investigation of the quality and composition of forage and feeds. While traditional *in vitro* methods measure the digestibility of one substrate part, IVGPT measures the fermentation of soluble and insoluble substrates (Tilley and Terry, 1963).

Tilley and Terry (1963), working with grasses, reported a decent relationship of digestibility between anticipated data (*in vivo*) and observed data (*in vitro*) with a linear regression equation ($Y = 0.99 \times X - 1.01$, $SE = \pm 2.31$). The digestibility approximation by the IVGPT was highly related with that anticipated by *in vivo* method (Marten and Barnes, 1980). Monson *et al.* (1968) reported a high relationship ($r = 0.92$) between digestibility *in vivo* and *in vitro* of Coastal Bermuda grass. A significant relationship ($r = 0.79$) between *in vitro* and *in vivo* digestibility was discovered (Sileshi *et al.*, 1996). Blümmen *et al.* (1997), working with roughages observed a high relationship amongst IVGPT and apparent and real digestibility ($r = 0.96$ and 0.95 ; respectively). Then again, a difference in lag time, shorter for *in vivo*; faster digestion rate for *in vivo*; and greater level of digestion for the *in vivo* technique (Varel and Kreikemeier, 1995). Lund *et al.* (2007) found a difference in digestibility and suggested that the difference was due to an incorrect sampling of the forages.

2.5 SUMMARY OF LITERATURE REVIEW

Low quality and inadequacy of feeds all year round are considered to be the major constraints hampering productivity of ruminants (Nurfeta, 2010). Tropical grass is known to improve intake and digestibility of roughages when supplemented with concentrates (Nurfeta, 2010). Conversely,

the uses of such supplements are limited under smallholder livestock production systems due to the scarcity and high cost of concentrates.

Legumes have a large potential to overcome this limitation as they have higher protein concentrations than that of grasses or other forages such as maize. Ruminant production generally does require forages with CP contents of 100 to 170 g kg⁻¹ DM and forage legumes tend to be either at the upper end of, or above this range (Minson, 1990; Groff and Wu, 2005; Dewhurst *et al.*, 2009). For instance, across a wide range of species and regions, the CP content of forage legumes was found to be approximately 170 g kg⁻¹ DM compared to 115 g kg⁻¹ DM for grasses (Minson, 1990).

Studies by Marley *et al.* (2007) showed that forage legumes, such as red clover (*Trifolium pratense*), offered fresh or ensiled increased the growth rates in ruminants due to higher nitrogen utilization efficiency and dry matter intakes. More so, feeding a diet with canola meal as the main source of protein has been shown to increase wool growth in pregnant and weaner sheep (Masters *et al.*, 1996). Protein can often be a limiting nutrient for the growth of young livestock and for milk production in ruminant production systems (Minson, 1990). Besides, the leaves from leguminous trees and browses plants such as *Glyricidia*, *Leucaena*, *Ficus sp* and *Erythrina* have been reported to contain around 250–350 g of crude protein/kg DM, and can also be considered in this category (Alhassan *et al.*, 1999).

Bambara groundnut (*Vigna subterranean*) is an annual herbaceous, intermediate plant with creeping stems belonging to the *fabaceae* family. Amarteifio and Moholo (1998) reported that the

crop could make a well-balanced feed with a caloric value equivalent to that of a high quality cereal grain. Its potential, however, as a feed supplement to sheep has not been extensively studied. Nevertheless, other forages such as cowpea haulm have been researched into as a supplement to poorer quality hay and maize stover. Cowpea haulm addition improves nutrient supply and growth of livestock over the use of low quality forages alone but degree of weight change varies relative to total nutrient supply (Ngwa and Tawah 2002; Baloyi *et al.*, 2006; Antwi *et al.*, 2014).

Maize stover, on the other hand which is available in large quantities is of low nutritive value due to its low digestibility and its failure to provide the rumen microorganisms and the host animal with all the nutrients required (Undi *et al.*, 2001). The nutritive value of crop residues such as maize stover, has been increased by alkali treatment and/or nitrogen (N) supplementation (Preston, 1995; Chandrasekharaiah *et al.*, 1996).

CHAPTER THREE

MATERIALS AND METHODS

3.1 DESCRIPTION OF THE EXPERIMENTAL SITES

The study was conducted at the Department of Animal Science (DAS), Kwame Nkrumah

University of Science and Technology, Kumasi, Ghana, between June and December 2015.

The area is located at altitude 285m on latitude 06° 40' N and on longitude 001° 33' W). It is within the semi-deciduous forest zone of Ghana. Temperatures are relatively high throughout the year with the highest average temperature of 35.2 °C recorded in March and the lowest of about 20.2 °C recorded in January. The average yearly rainfall is 1510.0 mm (Meteorological Service Department, 2007). The major rainy season occurs from April to July and the minor rainy season from September to October. The dry season is from November to March.

3.2. AGRONOMY EXPERIMENTS

3.2.1 Source of experimental material and Bambara establishment

A total of forty (40) improved lines of Bambara groundnut seeds were obtained from the International Institute of Tropical Agriculture (IITA), situated in Ibadan, South-Western region of Nigeria. They were established on the arable field of the Department of Animal Science, KNUST.

3.2.2 Land preparation and field layout

A 0.22-acre field was mechanically slashed of weeds, ploughed, harrowed, demarcated, ridged and pegged. A randomized complete block design was used with four replicates in blocking the field into 40 plots. Each plot size was 18 m x 2 m. Individual plots within a block were separated from each other by 1m, while the blocks were separated from each other by a distance of 2 m. Inter-row

spacing was 50 cm and intra-row spacing was 20 cm. Two seeds per stand were sown with a cutlass to a depth of about 3-5 cm.

3.3.3 Cultural practices

The field weeds were controlled by hand hoeing during the fourth and sixth week of sowing while thinning was done 21 days after planting. The forages were harvested at maturity (three months after planting), weighed and sundried. Four varieties from the forty samples (TVSU9A-138, TVSU879, TVSU690, and TVSU1446) were selected based on the agronomic characteristics and used for the digestibility and balance trials.

3.4 INTAKE AND DIGESTION TRIAL

3.4.1. Housing, Medication and Feed Intake Measurements

Four Djallonke rams of about 12 months of age and average body weight of 15.5 ± 0.5 kg were used in this trial. Internal parasites medication was administered on the first day of the arrival of the animals. The sheep were housed in 60 cm x 100 cm metabolism cages. The four rams were accustomed to the metabolism crates for a three day period (adaptation period) prior to intake and digestion trials. Feed intake was measured and recorded daily. The quantities of feed offered and refused were also recorded daily and the difference was calculated as feed intake. Total feed offered, refused and feces voided were bulked for each ram for determination of digestibility. Total fecal output was sun and oven dried at 100°C for 24 h. They were ground through a 1 mm screen using a Wiley mill grinder (Arthur H. Thomas, Philadelphia, USA). Two grams of fecal samples were further taken for chemical analysis. Urine samples were also collected and frozen immediately pending chemical analysis.

3.4.2 Dietary treatments

The dietary treatments comprised treatment 1 (maize stover only), treatment 2 (maize stover + 150g Bambara groundnut haulm), treatment 3 (maize stover + 300g Bambara groundnut haulm) and treatment 4 (maize stover + 450g Bambara groundnut haulm) respectively.

The animals were adjusted to the diets for 14 days after which they were weighed and assigned randomly to the four test diets. The four selected haulms and maize stover were chopped into 4-5 cm lengths and fed to the sheep. The supplement was offered at 08:30 h while half of the basal diet was offered at 10:30 h. The other half was offered at 15.00 h. Instances where the haulm was not completely eaten, it was withdrawn (reserved in a separate plastic container) before feeding the maize stover. Clean water was available *ad libitum*.

3.5 CHEMICAL ANALYSES

3.5.1 Sample preparation

Dried samples of Bambara groundnut haulms, maize stover and the fecal dry matter were finely ground to pass through a 1 mm mesh sieve and stored in separate plastic bags until required for analysis. The chemical composition determined included DM, CP, and Ash according to the procedures of the Association of Official Analytical Chemists (AOAC, 1990).

3.5.2 Fibre Analysis

The Ankom Daisy technique was used to determine neuter detergent fibre (NDF) and acid detergent fibre (ADF) contents of the selected samples, using the Ankom²⁰⁰ Fibre Analyzer.

$$\text{NDF/ADF (as-is basis)} = \frac{[W_3 - (W_1 \times C_1)]}{W_2} \times 100\%$$

Where

Where W_1 = weight of bag

W_2 = weight of sample that was placed in the bag

W_3 = final weight of bag+sample after digestion

3.5.3 *In vitro* gas production method

Rumen content was collected from 3 slaughtered cattle and then strained through a cheese cloth (with continues flushing with CO_2 gas) to obtain the rumen fluid. McDoughal's buffer was prepared using 9.8 NaHCO₃, 2.77 NaH₂PO₄, 0.57 KCl, 0.47 3 2 4 NaCl, 2.16 MgSO₄ 7HO, 16 CaCl₂ 2HO). The rumen fluid was mixed with the prepared McDoughal's buffer in a ratio of 1:2 and transferred into a Winchester bottle. Three hundred milliliters (300 mls) of the resultant Rumen: Buffer solution was displaced into 16 calibrated glass syringes containing 0.5g of the 4 selected feed samples, with 3 replicates each and a blank for each sample. The syringes were then incubated in a water bath of set temperature of 37 °C and the volume of gas produced was measured at 3, 6, 9, 12, 15, 18, 21, 24, 48, 72 and 96h. The volume of gas produced from the blanks was deducted from the volume of gas produced per sample against the incubation time and from the graph, the gas production characteristics were estimated using the equation:

$$Y = a + b(1 - e^{-ct})$$

Where:

Y = volume of gas produced at time t, c, = intercept (gas

produced from the insoluble fraction (b), t= incubation time.

C_1 = blank bag correction

3.6 ANIMALS AND EXPERIMENTAL DESIGN

Four Djallonke rams of about 12 months of age and average body weight of 15.5 ± 0.5 kg were used in this trial. The animals were adjusted to the diet (maize stover *ad libitum* and 200 g Bambara groundnut haulm) for two weeks. The animals were assigned randomly in a 4x4 latin Square Design to one of the four test diets namely, T1 (Maize stover only, the basal diet), T2 (maize stover + 150g Bambara groundnut haulm), T3 (maize stover + 300g Bambara groundnut haulm) and T4 (maize stover + 450g Bambara groundnut haulm). Each period lasted for 3 days. The animals were rested for additional three days after each period and allowed to accustom to new treatments in the subsequent period for another week.

3.6.1 Intake, digestion trials and measurement

The four rams were accustomed to the digestibility crates for three days earlier to intake and digestion trials, during each period following a week adaptation period. Feed intake was measured and recorded daily. The quantities of feed offered and refused were recorded daily; the difference was calculated as feed intake. Total feed offered, refused and feces voided were bulked for each ram for digestibility determination.

3.7 STATISTICAL ANALYSIS

All data collected were analysed using Analysis of Variance (ANOVA) using Genstat version 12.1 and the means separated by Duncan's Multiple Range Test (DMRT) at 5% probability level. The *in vitro* gas production data recorded was analyzed as a replicated 4 x 4 Latin Square using PROC MIXED of SAS (Version 9, 2006) according to the following model.

$$Y = \mu + P_i + \tau_j + A_{ij(k)} + \varepsilon_{ij(k)}$$

Where, Y_{ijkl} = measured dependent variable;

μ = overall mean;

P_i = fixed effect period i ($i =$

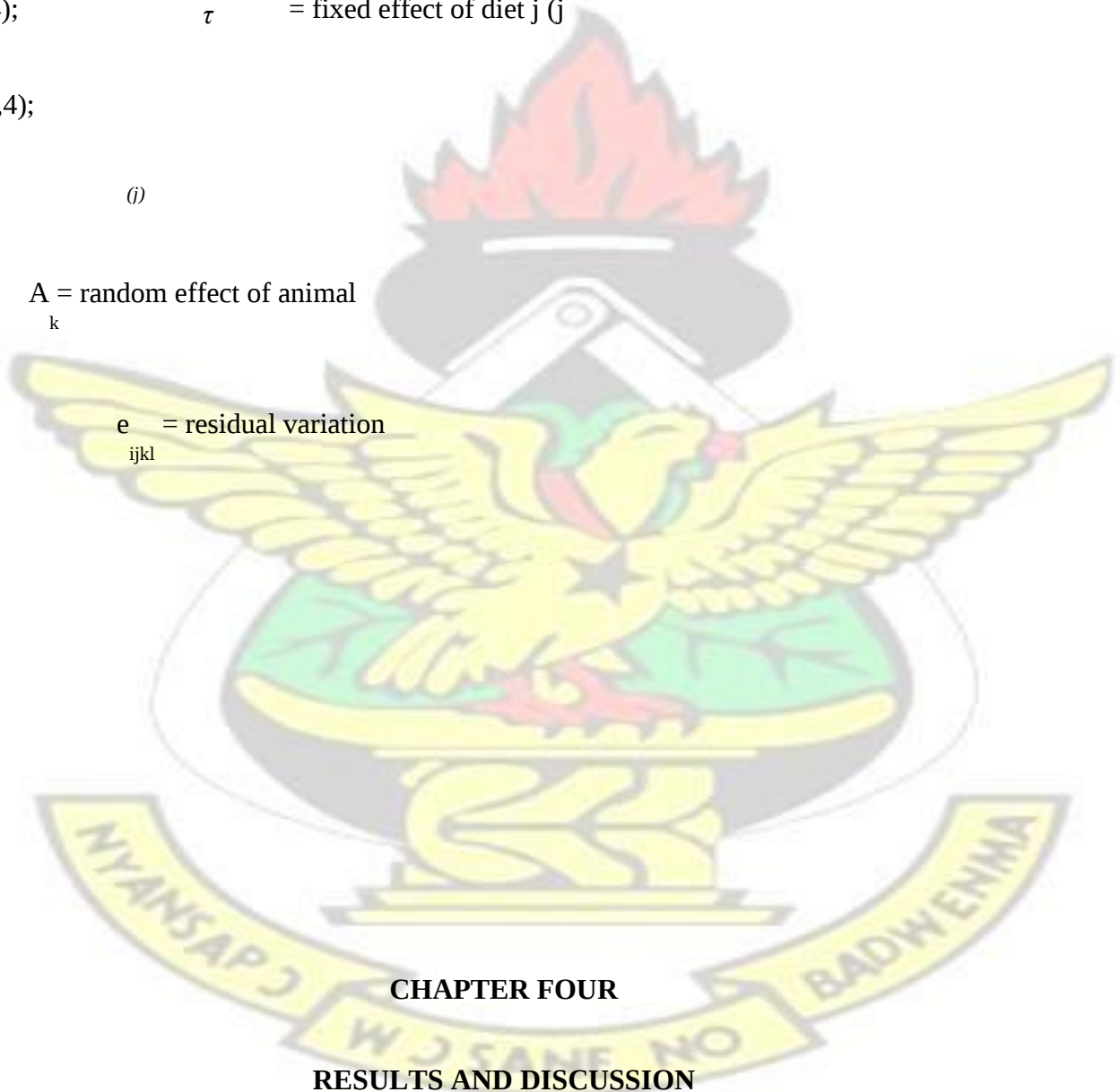
$1, \dots, 4$); τ_j = fixed effect of diet j (j

$= 1, \dots, 4$);

(i)

A_k = random effect of animal

e_{ijkl} = residual variation



CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 OVERVIEW

Forty varieties of Bambara groundnut (see appendix) were established on the arable field of the

TVSU921	10	3,780
TVSU381		3,661
TVSU879		3,280
TVSU750		3,229
TVSU552		3,169
TVSU378		2,950
TVSU383		2,907
TVSU759		2,895
TVSU751		2,878
TVSU385		2,381
TVSU690		2,344
TVSU854		2,335
TVSU1447		2,210
TVSU516		2,100
TVSU337		1,975
TVSU577		1,840
TVSU282		1,508
TVSU1820		1,437
TVSU476		1,328
TVSU1825		1,317
TVSU1252		1,050
TVSU374		800
TVSU138		733
TVSU346		689
TVSU501		650
TVSU323		629
TVSU506		622
TVSU544		611
TVSU1446		544
TVSU482		509
TVSU168		447
TVSU1474		424
TVSU1471		418
TVSU674		319
TVSU1243		288
TVSU1458		286
TVSU513		245
TVSU1482		211

4.2 NUTRIENT ANALYSIS OF THE HAULM

Department of Animal Science, KNUST. The forages were harvested at maturity (three months after planting) weighed and sundried following grain and haulm yield determination. The haulms were assayed for their nutrient composition. Four varieties were selected for the animal

experiment. Four varieties from the forty samples (TVSU138, TVSU879, TVSU690, and

17 4,925 18 15 21 13 24 15 20 18

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9 **Table 4.2: Chemical composition of Bambara groundnut haulm and maize stover**

COMPONENT	MAIZE (%)	BAMBARA	GROUNDNUT	HAULM	VARIETIES
	<i>TVSU(138)</i>	<i>TVSU(879)</i>	<i>TVSU(690)</i>	<i>TVSU (1446)</i>	STOVER
DM	90.24	90.67	90.28	90.80	83.93
CP	16.15	14.64	14.32	14.75	4.18
ASH	11.00	9.50	8.00	9.00	10.00
NDF	57.09	68.45	50.43	47.34	73.52
ADF	43.65	35.84	33.46	33.60	47.95

Where DM= Dry Matter; CP= Crude Protein; NDF=Neutral Detergent Fibre; ADF= Acid Detergent Fibre.

The dry matter values for the various cultivars ranged from 90.24 (TVSU138) to 90.80% (TVSU1446) (Table 4.2). The maize stover recorded a dry matter of 83.93%. The CP values obtained for variety TVSU9A (138) was 1.10 times higher than what was recorded for varieties TVSU14B (879), TVSU15B (690), and TVSU25B (1446), and 3.86 times higher than the basal diet. The crude protein contents of these haulms were similar to other leguminous forages classified as browse plants with CP content ranging from 140-300g/kg (Norton, 1994). Therefore, they can be used to supplement poor quality roughages to increase productivity of ruminant livestock in tropical regions. The importance of protein intake as the determinant of performance in ruminants has been strongly emphasized by Preston and Leng, (1987). Maize stover is known to have low CP content (Kabatange and Shayo, 1991) of which the result indicates. The percentage of CP in maize stover is within the range of 2.3% (Kabatange and Shayo, 1991) to 7.1% (Woyengo *et al.*, 2004). Therefore, nitrogenous supplement to a basal diet of maize stover may be essential to improve feed consumption by animals (Woyengo *et al.*, 2004).

Percentage ash content recorded was 8 for TVSU690, 9 for TVSU (1446), 9.5 for TVSU (879) and 11 for TVSU (138). In this study, the maize stover recorded the highest NDF followed by TVSU14

(879), TVSU9 (138) and TVSU15 (690) whereas TVSU25 (1446) recorded the lowest. Conversely, the NDF values reported by Norton (1994) for forages are lower as compared to the values of these varieties of Bambara groundnut haulm. Moreover, the highest ADF value was recorded by the basal diet which was about 1.2 times higher than the other varieties. It has been reported (Ball *et al.*, 2001) that higher quality forages have an ADF of 25 to 45% and NDF of 35 to 55% while lower quality forages are considered to have an ADF of 35 to 45% and NDF of 55 to 70% (as fed). This implies that TVSU15 (690) and TVSU25 (1446) could be classified as higher quality forages with NDF and ADF ranging from 47.3 to 50.4% and 33.4 to 33.6% respectively whereas the rest of the haulms including the basal diet could be classified as lower quality forages with relatively higher fibre content. This may therefore limit their comparative feeding value as feeds of high fiber contents reduce intake and digestion (Van Soest, 1994).

4.3 IN VITRO GAS PRODUCTION

effect on readily fermentable fraction shown as “a”, potential gas production “b” and the rate of gas production “c” had significant differences ($P = 0.0001$).

Table 4.3: Least square means ($\pm$ s.e) of the effect of four cultivars of Bambara groundnut haulms on cumulative in vitro gas production (ml gas/0.5g DM)

	SE													
Cultivars														
	TVSU(138)	TVSU(879)	TVSU(690)	TVSU(1446)										
GP Parameter														
A	0.01 ^b	-0.34 ^b	2.56 ^a	0.41 ^b	1.129	B	21.47 ^d	23.84 ^c	34.61 ^a	30.90 ^b	1.531	c	(h ⁻¹)	0.04 ^b 0.07 ^a
	0.04 ^b	0.04 ^b	0.024											

Means with the common superscripts (a,b,c) within rows are not significantly different according to the Waller-Duncan k-ratio t-test with $t=100$.

Where a = readily fermentable fraction; c = rate of gas production (GP) from the slowly fermentable fraction, b; SE = standard error

The cumulative gas production as a function of incubation time is showed in Figure 4.1. The lowest gas production was observed in cultivar TVSU (138) followed by TVSU (879), TVSU (1446) and TVSU (690) in ascending order. On the other hand, the rate of gas production was highest for cultivar TVSU (879) whereas the other cultivars had similar values.

The results of the gas production data for the cultivars are presented in Table 4.3. The cultivar's

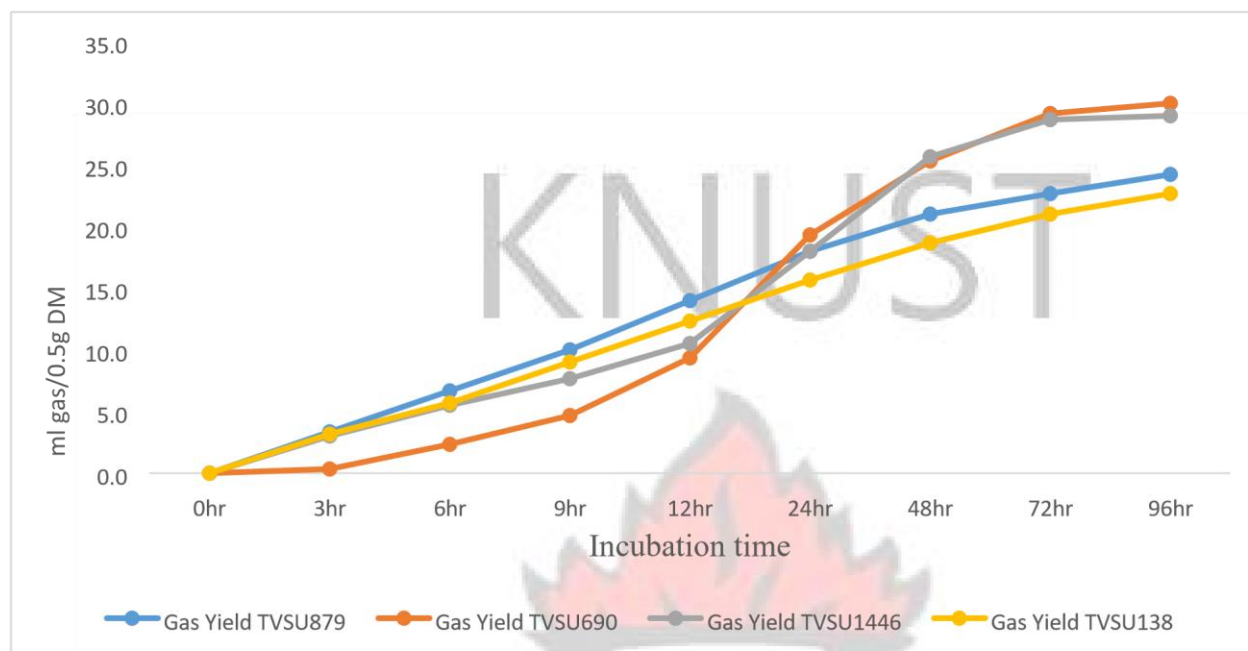


Figure 4.1: Gas production profiles of four cultivars of Bambara groundnut haulm

The highest fermentative gas production was recorded in TVSU (690) and was followed by, TVSU (1446), TVSU (879) and TVSU (138) in descending order (Table 4.3). The rates of gas production ranged from 0.04 to 0.07 h⁻¹. The fastest rate of gas production was observed in TVSU(879), possibly influenced by the soluble carbohydrate fraction readily available to the microbial population. Slower rates were observed in the other cultivars and this indicated that these haulms were less readily available to the microbes in the rumen (Nitipot and Sommart, 2003). More so, the least gas buildup, which was from cultivar TVSU (138), could be attributed to high cell wall content (lignin and crude fibre). Lignin content has been reported (Jung and Deetz, 1993) to be negatively correlated with gas production which in effect affect the functioning of the rumen microbes by limiting fermentation and enzymatic breakdown of forage polysaccharides. Gas production is noted to be positively correlated with feed fermentation, hence, the cultivar TVSU (138) could be designated as having low feeding value due to its low gas production.

4.4 EFFECT OF BAMBARA GROUNDNUT HAULM SUPPLEMENTS ON THE INTAKE

OF MAIZE STOVER

It was observed that as dry matter intake of BGH increased at different levels of supplement, the dry matter intake of MS also decreased. The animals consumed more of the basal diet at zero level of inclusion of the supplements and were partly substituted by the supplement as the levels increased. The intake of the supplements however did not show any significant difference ($P>0.05$) as their levels were increased. Conversely, the control and the supplemented treatment groups intake did showed significant ($P<0.05$) difference at the end of the experimental period (Table 4.4).

Table 4.4: Effect of supplement level on feed intake of sheep

PARAMETERS (g)	DIETARY TREATMENTS				SEM	P-VALUE
	T1 (SL=0)	T2 (SL=150)	T3 (SL=300)	T4 (SL=450)		
MS	410 ^b	350 ^a	340 ^a	320 ^a	0.014	0.024
BGH	0.0 ^a	110 ^b	130 ^b	170 ^b	0.021	0.007
TOTAL FEED INTAKE	410	460	470	490	0.022	0.119

^{a b} Means in the same row with different superscripts are significantly different ($p<0.05$); MS= maize stover; SL= Supplement Level; BGH= Bambara groundnut haulm; SEM= Standard error of means.

It is believed that the extent of replacement of the basal diet by forage legume, resulting in a reduced intake of the basal diet, depends upon the level of addition of the supplement (Ndlovu, 1991). Dixon and Egan (1999) reported a substitution effect of supplementation of mixtures of cereals and/or protein supplement to the grass hay based feeding. The maize stover intake was higher for lambs receiving low level of supplement than those receiving a high level of supplement.

Similarly, voluntary intake of forage was lower ($P<0.05$) in lambs offered barley and oat supplements than those offered the basal diet (Ponnampalam *et al.*, 2004). Nsahliai and Ummuna (1996) reported that the response to supplementation depends on feed and animal factors, the

former including the quality of basal roughage and supplement feed.

In the current study, the sheep accepted the BGH readily but could not consume the total amount of supplement offered.

The supplementation of BGH had significantly ($P < 0.05$) influenced the dry matter intake of the basal diet (MS). This might be due to the bulk volume of maize stover and the Bambara groundnut haulm (Aredo and Musimba, 2003). Tolera and Sundstøl (2000) and Assefa (2007) reported similar observations on the intake of tef straw, maize stover and natural pasture hay supplemented with an increasing level of multipurpose tree leaves and herbaceous legumes to sheep. Eroarome (2002) also reported a decrease in the DM intake of goats on the basal diet *Panicum maximum* as the level of *Leucaena leucocephala* leaf increased, regardless of the form of presentation (fresh or wilted).

4.5 DRY MATTER INTAKE, APPARENT NITROGEN DIGESTIBILITY COEFFICIENT AND NITROGEN BALANCE

There were no significant differences ($P > 0.05$) among the treatments means for the dry matter intake (DMI) but the trend was that, higher feed intake was observed as the levels of the Bambara groundnut haulm supplement increased (Table 4.5).

Table 4.5: Dry matter intake, apparent nitrogen digestibility co-efficient and nitrogen balance of sheep fed maize stover supplemented with Bambara groundnut haulm

PARAMETERS	DIETARY TREATMENTS				SEM	P-VALUE
	T1 (SL=0)	T2 (SL=150)	T3 (SL=300)	T4 (SL=450)		
DMI (%)	34.13	38.27	40.50	42.30	0.020	0.099
Faecal dry matter (%)	91.78	92.46	92.69	92.35	0.478	0.612
49.4 ^b 54.66 ^b	57.16 ^b	8.78	0.017	efficient (%)		
N in Feed (g/kg)	0.27 ^a	0.48 ^b	0.52 ^b	0.61 ^b	0.047	0.012
N in urine (g/kg)	0.00061	0.00082	0.00061	0.00064	0.00012	0.554
N in faeces (g/kg)	0.012	0.011	0.011	0.013	0.0012	0.667
N balance (g/kg)	0.26 ^a	0.47 ^b	0.51 ^b	0.59 ^b	0.047	0.011

^{a b} Means in the same row with different superscripts are significantly different ($p < 0.05$); SEM = Standard error of means

The faecal dry matter for treatment 3 was the highest (92.69%) as compared with treatment 1 which recorded the lowest (91.78%). Faecal and urine amongst the treatment groups did not differ ($p > 0.05$) significantly. The digestibility co-efficient, nitrogen in feed and nitrogen balance were significantly lower ($p < 0.05$) for treatment one which had no Bambara groundnut haulm supplement compared to the other treatment groups supplemented with BGH which were also statistically ($p > 0.05$) similar. In the current study, nitrogen intake and nitrogen balance increased with supplementation of the basal diet. This is in agreement with the observation made by McDonald *et al.* (1996) which reported that dietary nitrogen intake in animal is directly related to the proportion of nitrogen in the diet.

The higher apparent digestibility coefficient for the supplemented groups compared to the sole maize stover treatment could be due to the presence of higher crude protein content in the supplement diet than the sole maize stover treatment which provides more nitrogen for microbial utilization (Yahaya *et al.*, 2000; Abdulrazak *et al.*, 1997). This is also in consonance with a study by McMeniman *et al.* (1988) who reported an increase rate of rice straw degradation when

supplemented with leguminous hay. Likewise, Ndlovu and Buchanan-Smith (1985) found similar observations when Lucerne hay was used as a supplement for barley straw. These results indicate that the maize stover digestibility was lower due to the high fiber concentration. Mostly, the feeding value of forages and the extent of forage degradation in the rumen is constrained by the amount of fiber content (NDF) (Von Keyserlingk *et al.*, 1990; Van Straalen and Tamminga, 1990 and Aregheore, 2007). Forages containing high cell wall content show restricted voluntary intake due to their slow degradability and accumulation of fiber in the rumen (Martin-Orou *et al.*, 2000). Excretion of urinary and fecal nitrogen were not influenced ($P>0.05$) by inclusion of Bambara groundnut haulm in the diet. However, the nitrogen excretion in faeces was higher than in urine. According to Van Soest (1994), the increase in nitrogen intake is associated with the increase of the urea production in the liver and, consequently, of its excretion in urine, while a decreased nitrogen intake leads to a reduction in the excretion of urea in urine for maintenance of the pool of urea in the plasma, which is under homeostatic physiological control.

CHAPTER FIVE

CONCLUSIONS AND RECOMMENDATIONS

5.1 CONCLUSIONS

The results gathered from this study indicate that, the crude protein contents of the selected varieties of Bambara groundnut haulm the range between 14.32% and 16.15% and therefore can be used to supplement poor quality roughages to increase productivity of ruminant livestock in tropical regions. Also, the high crude protein and relatively low ADF and NDF contents of Bambara groundnut haulm compared to nitrogenous supplement indicated the potential of this legume to be used as a supplement diet for poor quality roughage in ruminant diets. In addition, the four cultivars of the Bambara groundnut haulm under study revealed that these cultivars could be valuable alternative animal feed sources in ruminant feeding. The levels of supplementation of Bambara groundnut haulm recorded 47% to 59% Nitrogen balance which was better than the basal diet (26%) which makes it better alternative source of ruminant feed.

5.2 RECOMMENDATIONS

1. Based on the above results, it is recommended that small ruminant farmers, sheep farmers in particular, could feed whole Bambara groundnut haulm as supplement, specifically in the dry season when there is scarcity of roughage and the nutritive value is low.
2. Further research on Bambara groundnut haulm beyond the 300 g/d should be conducted to ascertain the optimum level for sheep production.
3. Further studies should also be carried out to determine the effect of supplementation of Bambara groundnut on blood cellular and biochemical indices of sheep.



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APPENDIXES

PROCEDURES

Appendix 1: The GLM Procedures on Dry Matter

Table A: Analysis of Variance for Dry Matter

Source of variation	DF	Sum of squares	Mean Squares	F Value	P Value
Animal stratum	3	23.237	7.746	2.83	

Period stratum	3	30.275	10.092	3.68
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Animal Period stratum

Treatment	3	5.331	1.777	0.65	0.612
Residual	6	16.449	2.741	1.20	

Animal Period.*Units* stratum

	32	73.061	2.283
Total	47	148.354	

Appendix 2: The GLM and Mixed Procedures of the chemical composition of four varieties of Bambara groundnut haulm

Table A. Analysis of Variance for crude protein

Source of variation	DF	Sum of Squares	Mean Squares	F Value	Pvalue
Treatment	48	567.7296	11.8277	19.78	0.001
Residual	49	29.3004	0.5980	Total	
97	597.0300				

Table B. Analysis of Variance for NDF

Source of variation	DF	Sum of Square	Mean Squares	F. Value	PValue
Treatment	48	7670.98	159.81	2.16	0.004
Residual	49	3630.34	74.09	Total	
97	11301.32				

Table C. Analysis of Variance for ADF

Source of variation	DF	Sum of Squares	Mean Squares	F. Value	PValue
Treatment	48	3998.230	83.296	15.04	0.001
Residual	49	271.299			
Total	97	4269.529	5.537		

Table D. Analysis of Variance for ASH

Source of variation	DF	Sum of Squares	Mean Squares	F. Value	P-Value
Treatment	48	1195.632	24.909	12.76	0.001
Residual	49	95.630	1.952		
Total	97	1291.262			

Appendix 3: The GLM Procedures for Bambara groundnut haulm on maize stover intake (g/d)

Table A. Analysis of Variance on Bambara groundnut haulm intake

Source of variation	DF	Sum of Squares	Mean Squares	F. Value	P-Value
Animal stratum	3	0.031764	0.010588	1.91	
Period stratum	3	0.008331	0.002777	0.50	

Animal. Period stratum

Treatment	3	0.189406	0.063135	11.41	0.007
Residual	6	0.033186	0.005531	2.73	

Animal. Period.*Units* stratum

	32				
		0.064950	0.002030		
Total	47	0.327637			

Table B. Analysis of Variance on maize stover intake

Source of variation	DF	Sum of Squares	Mean Squares	F. Value	P-Value
Animal stratum	3	0.201217	0.067072	26.74	
Period stratum	3	0.619783	0.206594	82.36	
Animal. Period stratum					
Treatment	3	0.050983	0.016994	6.78	0.024
Residual	6	0.015050	0.002508	0.79	

Animal. Period.*Units* stratum

32

0.101867

0.003183

Total

47

0.988900

Table C. Analysis of Variance on total feed intake

Source of variation	DF	Sum of Squares	Mean Squares	F. Value	P-Value
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Animal stratum

3

0.386722

0.128907

23.17

Period stratum

3

0.564739

0.188246

33.84

Animal. Period stratum

Treatment

3

0.049656

0.016552

2.98

0.119

Residual

6

0.033378

0.005563

0.90

Animal. Period.*Units* stratum

32

0.197417 0.006169

Total

47 1.231912

Appendix 4: The GLM Procedures for concentration of Nitrogen in urine and Feces Table A: Analysis of Variance on urine

Source of variation	DF	Sum of Squares	Mean Squares	F. Value	P. Value
Animal stratum	3	4.4613	1.4871	3.06	
Period stratum	3	4.5416	1.5139	3.12	
Animal Period stratum					
Treatment	3	1.1364	0.3788	0.78	0.547
Residual	6	2.9137	0.4856	1.78	

Animal Period.*Units* stratum

32
8.7510 0.2735

Total 47 21.8041

Table B: Analysis of Variance on Feces

Source of variation	DF	Sum of Squares	Mean Squares	F. Value	P. Value
Animal stratum	3	2.47872	0.82624	2.03	
Period stratum	3	11.19297	3.73099	9.16	
Animal Period stratum					
Treatment	3	0.51755	0.17252	0.42	0.743
Residual	6	2.44489	0.40748	6.54	

Animal Period.*Units* stratum

32

1.99307

0.06228

Total

47

18.62720

Appendix 5: The GLM Procedures on Digestibility Coefficient

Table A: Analysis of Variance for Digestibility Coefficient

Source of variation	DF	Sum of Squares	Mean Squares	F Value	P Value
Period stratum	3	9633.3	3211.1	3.47	
Animal stratum	3	3153.1	1051.0	1.14	
Period Animal stratum					
Treatment	3	21762.3	7254.1	7.84	0.017

Residual	6			
		5554.8	925.8	2.47

Period Animal.*Units* stratum

32	11999.1	375.0
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Total	47	52102.4
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Appendix 6: The GLM Procedures on Initial Weight and Weight Gain

Table A: Analysis of Variance for initial weight

Source of variation	DF	Sum Squares	Mean Squares	F Value	P Value
Animal stratum	3	1.620625	0.540208	24.70	
Period stratum	3	14.455625	4.818542	220.28	
Animal Period stratum	78				
Treatment	3	0.145625	0.048542	2.22	0.187

Residual	6			
		0.131250	0.021875	4.20

Animal Period.*Units* stratum

32	0.166667	0.005208
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Total	47	16.519792
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Table B: Analysis of Variance for weight gain

Source of variation	DF	Sum of Squares	Mean Squares	F Value	P Value
Animal stratum	3	0.395625	0.131875	0.67	
Period stratum	3	0.520625	0.173542	0.88	
Animal Period stratum					
Treatment	3	0.970625	0.323542	1.64	0.278

Residual

6

1.186250 0.197708 37.96

Animal Period.*Units* stratum

32

0.166667 0.005208

Total

47 3.239792

APPENDIX 7: Proximate analysis of the 40 varieties of BGH and maize stover

Varieties/Samples	Parameters			
	CP	ASH	NDF	ADF
TVSU516	10.90	11.00	50.43	40.60
TVSU323	10.14	12.00	60.57	46.87
TVSU385	11.52	8.50	44.09	37.70
TVSU1447	11.90	10.95	52.56	43.71
TVSU544	12.18	12.50	57.48	43.13
TVSU513	8.86	9.50	52.41	44.70
TVSU1243	10.02	14.00	53.40	43.30
TVSU506	8.90	17.50	54.05	36.23
TVSU282	8.81	16.50	37.93	45.95
TVSU1482	14.32	7.50	54.81	39.63
TVSU1252	10.72	15.75	45.52	38.71

TVSU577	12.83	10.50	48.78	46.65
TVSU168	13.17	17.00	51.44	36.00
TVSU482	13.57	13.50	49.64	39.94
TVSU1471	11.45	16.50	58.21	44.85
TVSU1825	9.38	12.00	55.54	49.51
TVSU688	13.38	8.00	47.33	35.72
TVSU378	7.14	15.00	59.96	52.57
TVSU674	10.00	12.00	57.17	42.08
TVSU346	9.60	11.50	58.83	50.52
TVSU750	10.49	12.00	54.64	47.38
TVSU921	10.30	7.00	50.65	39.25
TVSU751	11.20	9.00	52.26	39.64
TVSU138	16.15	11.00	57.09	43.65
TVSU383	10.28	9.00	47.97	42.10
TVSU1458	9.18	20.00	46.14	53.46
TVSU374	10.53	10.00	56.59	46.89
TVSU381	7.11	10.00	56.34	48.49
TVSU879	14.64	9.50	68.45	35.84
TVSU369	9.25	19.00	71.65	63.42

TVSU854	12.67	 9.00	50.88	32.78
TVSU552	11.13	11.00	63.79	42.14
TVSU337	15.21	8.00	91.40	32.80
TVSU759	13.72	8.00	53.17	39.42

TVSU1474	9.29	14.00	49.91	43.40
TVSU1820	10.14	17.00	70.44	42.89
TVSU501	11.15	15.50	58.67	45.58
TVSU476	10.58	11.00	57.38	42.11
TVSU1446	14.75	9.00	47.34	33.60
Maize Stover	4.18	10.00	73.52	47.95
S.E.M	0.77	1.40	8.61	2.35
P-value	<0.001	<0.001	<0.004	<0.001

KNUST

Appendix 8: Ankom Protocol

METHODS

Sample Prep

1. Grind samples using a 40mm screen and air dry.
2. Label filter bags with special Ankom marker or Sharpie (Note: Sharpie will eventually

Day 1:

- fade and will need to be relabeled)
3. Dry bags in oven at 105°C for 30 min. Cool in desiccator for 30 min.
 4. Weigh each bag alone and tare balance.
 5. Weigh out .5 g (0.05 g) of sample into bag and record weight. Include a blank bag in each run (each batch of 24) for a blank bag correction.
 6. Seal the bag closed within 0.5 cm from the open edge using the heat sealer. Be sure to also seal the blank bag.
 7. Spread sample uniformly inside the filter bag by shaking and lightly flicking the bag to eliminate clumping.
 8. A maximum of 24 bags may be placed in the sample tree. All nine trays are used regardless of the number of bags being processed. Place three bags per tray and then stack trays on center post with each level rotated 120 degrees. Each tray will sit in the notches of the tray below it. The weight is placed on top of the empty 9th tray to keep the bag suspender submerged.

NDF Extraction: This method determines Neutral Detergent Fiber, which is the residue remaining after digesting in a detergent solution. The fiber residues are predominantly hemicelluloses, cellulose, and lignin.

1. Place bag suspender into the chamber with weight.
2. When processing 24 sample bags add 1800-1900 ml of ambient Neutral Detergent solution into the chamber. Be sure the valve on the left side of the machine is closed! If you're processing less than 24 samples, load the bag suspender from the bottom up and fill the chamber until the bags are covered. I found that 13 bags are covered by 1500 ml of solution.

3. Turn *Agitate* and *Heat* ON and confirm that the bag suspender is agitating properly. Set the timer for 75 minutes. Close and seal the lid of the chamber.
4. Fill 2-3 teakettles with DI water and start them boiling 10 minutes before the end of the run.
5. After 75 minutes turn *Agitate* and *Heat* OFF, open the drain valve and drain the hot solution BEFORE opening the lid. The neutral detergent can be dumped down the drain.
WARNING: The solution in the vessel is under pressure. The valve should be opened first to remove pressure before the lid can be opened. Ensure that the exhaust hose is securely positioned for safe disposal of effluent.
6. After solution has been exhausted, close the valve and open the lid. Pour in rinse #1 of boiling water to ~2 cm below lip of chamber. Lower lid but do not seal. *Agitate* for 5 minutes, but do NOT *Heat*. Refill kettle.
7. After 5 minutes, drain the rinse water down the sink. Repeat rinse 3 more times for a total of 4 rinses.
8. Remove the tree stand and place in a plastic tub. With acetone gloves, gently press water from bags and place in 3 or 4 L beaker. Add enough acetone to cover bags and soak for 3 minutes, gently agitating.
9. After 3 minutes, pour out acetone into a waste container. With acetone gloves, gently press acetone out of bags. Spread bags out on glass baking dishes in hood and allow to dry for ~ 40 minutes.
10. Place bags in an aluminum cake pan and dry at 105°C overnight.
WARNING: Do not place bags in oven until acetone has completely evaporated

KNUST

Day 2:

ADF Extractions: This method determines Acid Detergent Fiber, which is the residue remaining after digesting with H_2SO_4 and CTAB. The fiber residues are predominantly cellulose and lignin.

1. Remove dried bags from oven and cool in desiccator for 30 minutes. Weigh dried bags and replace bags in the tree.

2. Add 1800-1900 ml of ADF solution into the chamber. Be sure the valve on the left side of the machine is closed!
3. Turn *Agitate* and *Heat* ON and confirm that the bag suspender is agitating properly. Set the timer for 60 minutes. Close and seal the lid of the chamber.
4. Fill 2-3 teakettles with DI water and start then boiling 10 minutes before the end of the run.
5. After 60 minutes turn *Agitate* and *Heat* OFF, open the drain valve and drain the hot solution BEFORE opening the lid. The solution can be dumped down the drain. WARNING: The solution in the vessel is under pressure. The valve should be opened first to remove pressure before the lid can be opened. Ensure that the exhaust hose is securely positioned for safe disposal of effluent.
6. After solution has been exhausted, close the valve and open the lid. Pour in rinse #1 of boiling water to ~2 cm below lip of chamber. Lower lid but do not seal. *Agitate* for 5 minutes, but do NOT *Heat*. Refill kettle.
7. After 5 minutes, drain the rinse water down the sink. Repeat rinse 3 more times for a total of 4 rinses.
8. Remove the tree stand and place in a plastic tub. With acetone gloves, gently press water from bags and place in 3 or 4 L beaker. Add enough acetone to cover bags and soak for 3 minutes, gently agitating.
9. After 3 minutes, pour out acetone into a waste container. With acetone gloves, gently press acetone out of bags. Spread bags out on glass baking dishes in hood and allow to dry for ~ 40 minutes.
10. Place bags in an aluminum cake pan and dry at 105°C overnight.

WARNING: Do not place bags in oven until acetone has completely evaporated

CALCULATIONS

$$\text{NDF/ADF (as-is basis)} = \frac{[W_3 - (W_1 \times C_1)]}{W_2} \times 100\%$$

Where W_1 = weight of bag

W_2 = weight of sample that was placed in the bag

W_3 = final weight of bag+sample after digestion

C_1 = blank bag correction

PLATES OF BAMBARA GROUNDNUT HAULMS, PODS AND SEEDS



Bambara groundnut Haulm



Haulm and pods



Seeds

