

**UTILISATION OF ENSILED MAIZE STOVER AND CONCENTRATE
SUPPLEMENTS BY WEST AFRICAN DWARF SHEEP**

BY

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Matric No : 75496

A Thesis in the Department of Animal Science

Submitted to the Faculty of Agriculture and Forestry

in partial fulfilment of the requirement for the Degree of

DOCTOR OF PHILOSOPHY

of the

UNIVERSITY OF IBADAN

MARCH, 2013.

ABSTRACT

Maize Stover (MS), an abundant crop residue is a potential feed for ruminants. The nutritive value is reduced when left on the field without processing and preservation due to further lignification. The ensiling of MS with additives for preservation and use as feed for ruminant have not been adequately documented. Therefore, utilisation of ensiled MS and concentrate supplements by West African Dwarf Sheep (WADS) was investigated.

Two tonnes of MS were ensiled for 30 days using different additives (molasses, honey and sugar) at the rate of 50g kg^{-1} to obtain the following silages: MS and Molasses (MSM), MS and Honey (MSH), MS and Sugar (MSS) and MS only (control) which served as treatments. The pH, colour, odour and texture silage characteristics were determined using standard procedures. Dry Matter (DM), Crude Protein (CP), ash, Neutral Detergent Fibre (NDF), Acid Detergent Fibre (ADF) and Acid Detergent Lignin (ADL) of the ensiled MS were determined using standard techniques. Eight WADS were used to assess the silage acceptability using Coefficient of Preference (CoP) procedure. Nutritive value of the silages was determined using *in vitro* fermentation technique to obtain Total Gas Volume (TGV), methane and Dry Matter Degradability (DMD). Further, a mixture of the control (C) with Concentrate Supplements (CS) was fed to 20 WADS for 105 days in a completely randomised design as diets: A (75% C + 25% CS), B (50% C + 50% CS), C (25% C + 75% CS), D (100% C) and E (100% CS) to assess Dry Matter Intake (DMI) and Daily Weight Gain (DWG). Blood was collected to determine Packed Cell Volume (PCV), Red Blood Cell (RBC), White Blood Cell (WBC), Total Blood Protein (TBP) and Blood Urea (BU). Data were analysed using descriptive statistics, correlation and ANOVA at $P=0.05$.

The green colour, pleasant and fruity odour, firm texture and pH (3.5-3.7) were similar among the silages. The DM (31.6-35.3%), CP (7.9-8.9 %), NDF (68.6-69.9%), ADF (56.1-63.2%) and ADL (14.0-16.8%) were not significantly affected by the additives. Ash composition was significantly different such that it was lowest in MSH (6.3%). The silages were equally accepted by WADS with feed intake ranging from 517.1 to 558.2gDM/day and CoP between 0.95 and 1.03. The TGV: 31.7ml and 41.0ml; methane: 16.09 % and 25.20 % respectively for MS (control) and MSS silages varied significantly. The DMI of control (242.26 g/day) without supplement was significantly lower than that of control with supplements while DWG (-19.1-81.9g/day) varied significantly among the treatments. An inverse relationship was observed between the inclusion level of control and DMI ($r = -0.85$). However, direct relationship was noticed between DMI and DWG ($r = 0.95$). The RBC

($2.39-3.51 \times 10^6 \mu\text{l}$), WBC ($8.58-43.01 \times 10^3 \mu\text{l}$), BU (23.63-47.27mg/dl) and TBP (9.40-14.26g/dl) decreased significantly with increased inclusion of control but PCV (22.50-30.33%) was not significantly affected.

Ensiled maize stover with concentrate supplements in a ratio of 3:1 respectively enhanced sheep performance and is therefore recommended for West African Dwarf sheep.

Keywords: Maize stover, Silage quality, Concentrate supplementation, Sheep performance

Word count: 485

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CERTIFICATION

I certify that this project was carried out by Ademola Joseph **AMUDA** in the Department of Animal Science, University of Ibadan, Nigeria under my supervision.

.....
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DEDICATION

This work is dedicated to God the Father, the Son and the Holy Ghost

And my beloved and darling wife, Rachael Olawumi **Ademola** who single handedly financed the programme

My little kids: John Adewumi Ademola and Priscilla Aderonke Ademola

And my parents: Pa. Julius S. Amuda and late Mrs. Adejoju Mary Amuda (Nee Adefisayo).

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ACKNOWLEDGEMENTS

At last, I give thanks and praises to God Almighty that made this programme a huge success. If it had not been the LORD who is on my side, now what will I say. Once again, thanks to Jesus Christ my Redeemer, Saviour, the Alpha and Omega of my life. To HIM, I give adoration, honour and glory forever.

My profound gratitude goes to my able, frank, thorough, hard-working and God sent supervisor, Dr. O. J. Babayemi who led me out of wilderness of this programme and brought me to the promised land. Indeed you are the Moses of my time. May Almighty God continue to guide and guard you and your family in Jesus name -Amen. I will ever remain grateful to you.

Furthermore, I wish to express my sincere and deep appreciation to my darling wife. Through thick and thin, she has been supportive; morally, spiritually and financially. Darling, I acknowledge your dogged determination toward my success and achievement, may God Almighty reward you accordingly. Again, I acknowledge moral and financial support of my mother-in-law towards the success of this programme. May God bless you abundantly.

My heartfelt gratitude goes to my spiritual father, Prophet S. O. Olaoye (a.k.a. Baba Land) who used his prophetic office, divine connection and fire packed prayers to part the red sea and fall down the Goliath that stood against the success of this programme. Thank you so much, may God continue to uphold you with His right hand of righteousness -Amen.

Special thanks to Biola Adedosu for his immense contribution to the success of the programme particularly during silage making. May God Almighty reward you with great success in all your endeavours in life.

Special thanks goes to Mr. Ugbodaga Izuagie Alex (a.k.a. Baba Ope) who rendered a unique assistance from day one of feeding trial to the end. May God Almighty reward you accordingly. Once again, I say a big thanks to you and your family.

Special thanks also goes to Prof. A.D. Ologhobo current Head of Department of Animal Science. My deep and sincere appreciation goes to Dr. (Pastor) A. E. Salako for his timely advice, may God uphold you till the end. My unreserved gratitude also goes to Dr. O. A. Abufor his moral supports and words of encouragement.

I want to acknowledge the following members of academic staff in the Department: Prof. E.A. Iyayi, Dr. A. B. Omojola, Dr. M. K. Adewumi, Dr. Tolu Ososanya, Dr. Femi Adebisi, Dr. Bisi Ewuola, Dr. (Mrs) Bisi Agboola (Favour), Dr. Olorunnisomo, Dr. Sokunbi, Dr.

Gbenga Adeyemo, Dr. Gbenga Ogunwole (Post Graduate Coordinator), Dr. (Mrs) Lara Mabel Akinyemi and Mr Henry Osaiyuwu Osamede.

I also appreciate the following non-academic staff: Mrs. Udoh, Mrs Lara Omole, Mrs Olaoluwa, Mrs Lawal (a.k.a Alhaja), Mr Fabowale, Mr Omotoso, Alfa Taofeek Salau and Pastor Ayo Odufoye

The help obtained from the following colleagues Ajayi David, David Okunlola, Dayo Okunlola, Mrs Toyin Ajayi, (a.k.a. Ty), Funmilayo Bamigboye (Nee Familade; a.k.a Mummy Joy), Akinsola Saheed (a.k.a. Nigeria), Olusimbo Kenneth Obosi (a.k.a. Mummy Mercy), Mrs Alasa (a.k.a. Iya Oyo), Tope Ayano, Akinyemi Priscilla, Falola (IAR&T), Taiwo Adesokan, Gladys Mbaze, James Igbekoyi, Akinleye Suleiman Bamidele, Lukeman Saliu, Dr. (DVM) Aminu (Lautech), Dr. Mrs. Olagunju (a.k.a. Iya elepo), Mr Olowu (PYTP Director), Rasheed Iyiola (Faculty driver), Mrs Wojuola (project partner), Alaba (Ph.D Coordinator), Daniel Shittu (Lautech), Dr. (DVM) Abiola (Pastor) is greatly acknowledged.

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TABLE OF CONTENTS

	Page
Title of Project	i
Abstract	ii
Certification	iii
Dedication	iv
Acknowledgements	v
Table of Contents	viii
List of Tables	xiv
List of Figures.. .. .	xv
List of Platesxvi
1.1 CHAPTER ONE : INTRODUCTION	1
1.2 Justification	4
1.3 Objectives of the Study.. .. .	5
CHAPTER TWO : LITERATURE REVIEW	6
2.1 Small ruminant Population	6
2.2 Role of Livestock Farming systems	7
2.3 Crop residue	8
2.3.1 Utilisation of Crop residue	10
2.3.2 Constraints of Crop residue utilisation.. .. .	12
2.3.3 Characteristics of crop residue.. .. .	14
2.4 Improvement of nutritive value of crop residue.. .. .	15
2.4.1 Physical treatments	15
2.4.1.1 Gamma-irradiation	16
2.4.2 Chemical treatments	16
2.4.3 Physico – chemical treatments.. .. .	16
2.4.4 Biological treatments	17
2.4.5 Strategic Supplementation	19
2.4.6 Mineral Supplementation	19
2.5 Fermentation	20
2.5.1 Solid State fermentation	20

2.6	Introduction to Silage Making	22
2.6.1	Silage	24
2.6.2	Why make Silage	25
2.6.3	Silage storage systems	26
2.6.4	Materials to ensile and principle of obtaining good silage					27
2.6.5	Quantity of silage for farm	27
2.6.6	Fermentation dynamics	28
2.6.6.1	Phase I – Aerobic phase	28
2.6.6.2	Fermentation phase	29
2.6.6.3	Stable phase	31
2.6.6.4	Feed out phase..	32
2.6.5	Silage quality evaluation	33
2.6.6	Silage acids	36
2.6.7	Silage microbiology	37
2.6.8	Classification of silages	39
2.6.8.1	Lactate Silages	39
2.6.8.2	Acetate Silages	40
2.6.8.3	Butyrate Silages	40
2.6.8.4	Wilted silages	40
2.6.8.5	Deteriorated silages	41
2.6.8.6	Overheated silages	42
2.6.9	Silage additives	42
2.6.10	Silage temperature	44
2.6.11	Moisture content control during the ensiling process..					47
2.6.12	Silage fermentation in tropical silages	47
2.6.13	Ensiling technology for the tropics	48
2.6.14	The influence of silage fermentation on dry matter intake					50
2.6.14.1	Fermentation acids and silage intake	50
2.6.14.2	Silage pH and silage intake	50
2.6.14.3	Ammonia-N and silage intake	51
2.6.14.4	Clostridia and silage intake	51
2.7	Importance of silage in animal production systems	51
2.8	Rumen Ecology	52

2.9	Ecology of ruminal microbes	54
2.10	Rumen microorganisms	54
2.11	Rumen microbial interactions	56
2.12	Ammonia in rumen fermentation	58
2.13	Protein utilisation in ruminants	59
2.14	Degradation of protein in ruminants	60
2.15	Volatile fatty acids	61
2.16	Dietary fibre	62
2.17	Methane production from ruminants	65
2.18	In vitro gas production technique	67
2.19	The origin of In vitro gas	70
2.20	The application of in vitro gas method in predicting voluntary intake, digestibility of organic matter and metabolisable energy	72
3.0	CHAPTER THREE – EXPERIMENT ONE : Quality characteristics, chemical composition and acceptability of ensiled maize stover with or without additive.	74
3.1	Introduction	74
3.2	Materials and methods	75
3.2.1	Experimental site	75
3.2.2	Harvesting and silage making.. .. .	76
3.2.3	Experimental silos	76
3.2.4	Determination of temperature of the silages	76
3.2.5	Evaluation of the physical characteristics of the silages	77
3.2.6	pH determination of the silages	77
3.2.7	Chemical analysis	77
3.2.8	Acceptability study	77
3.2.9	Experimental Diets	77
3.2.10	Experimental Animals	77
3.2.11	Feeding of Animals	78
3.3	Statistical analysis	78
3.4	Results.. .. .	82
3.5	Discussion	91

3.5.1	Temperature range of silage	91
3.5.2	Physical Characteristics of silage	91
3.5.3	pH of silages	92
3.5.4	Chemical composition of silages	92
3.5.5	Fibre fractions	95
3.6.0	Acceptability	96

CHAPTER FOUR : EXPERIMENT TWO

4.0	In vitro fermentation characteristics and dry matter degradability of ensiled maize stover	98
4.1	Introduction	98
4.2	Materials and Methods	99
4.2.1	Experimental design	99
4.2.2	Experimental site	99
4.2.3	Gas production apparatus	99
4.2.4	Preparation and weighing of the feed samples	100
4.2.5	Collection and preparation of the rumen fluid	100
4.2.6	Preparation of the buffer solution	100
4.2.7	Preparation of the rumen liquor-buffer solution	101
4.2.8	Preparation of the syringes for incubation	101
4.2.9	Duration of incubation	102
4.2.10	Methane Gas Determination	102
4.2.11	Calculations	102
4.3	Statistical Analysis	104
4.4	Results	104
4.4.1	In vitro gas production parameters of ensiled maize stover at 24hrs of Incubation period	104
4.4.2	In vitro gas fermentation characteristics of ensiled maize stover at 24hrs of incubation period	106
4.5	Discussion	108
4.5.1	In vitro gas production parameters of ensiled maize stover at 24hrs of incubation period	108
4.5.2	In vitro fermentation characteristics of ensiled maize stover at 24hrs	108

incubation period	111
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CHAPTER FIVE: EXPERIMENT THREE

5.0 Performance, Digestibility and Nitrogen Balance of WAD Sheep Fed Ensiled Maize stover and Concentrate supplements ...	114
5.1 Introduction	114
5.2 Materials and Methods.. .. .	116
5.2.1 Experimental site	116
5.2.2 Preparation of pens	116
5.2.3 Experimental diets	116
5.2.4 Experimental animals and management	118
5.3 Digestibility study and nitrogen balance	118
5.3.1 Experimental diets	118
5.3.2 Experimental animals	119
5.3.3 Experimental design	119
5.3.4 Animal feeding	119
5.3.5 Chemical analysis	120
5.4 Statistical analysis	120
5.5 Results.. .. .	120
5.5.1 Composition of experimental diets.. .. .	120
5.5.2 Chemical composition of ensiled maize stover and concentrate supplements.	122
5.5.3 Feed intake and performance characteristics of WAD sheep fed ensiled maize stover and concentrate supplements	124
5.6 Apparent digestibility of WAD sheep fed ensiled maize stover ..	128
5.6.1 Nitrogen utilisation by WAD sheep fed ensiled maizestover and concentrate supplements	130
5.6.2 Total digestible nutrients in WAD sheep fed ensiled maize stover and concentrate supplements	134
5.7 Discussion	136
5.7.1 Dry matter intake and growth performance	136
5.7.2 Digestibility and nitrogen balance	139

CHAPTER SIX: EXPERIMENT FOUR

6.0	Haematological parameters and blood chemistry of West African Dwarf sheep fed ensiled maize stover and concentrate supplements ..	144
6.1	Introduction	144
6.2	Materials and methods	146
6.2.1	Collection and evaluation of blood samples	146
6.2.2	Haematology	146
6.2.3	Serum Biochemistry	146
6.2.4	Statistical analysis	146
6.3	Results.. .. .	147
6.3.1	Haematological response of West African Dwarf Sheep fed maize stover and concentrate supplements	147
6.3.2	Serum biochemistry of West African Dwarf Sheep fed ensiled maize stover and concentrate supplements	149
6.4	Discussion	151
6.4.1	Haematological responses of experimental animals	151
6.4.2	Serum biochemical response of experimental animals.. .. .	152

CHAPTER SEVEN

7.0	Summary, Conclusion and Recommendations.. .. .	154
7.1	Summary of findings	154
7.2	Conclusion	156
7.3	Recommendations	157
	References	158

LIST OF TABLES

Table	Page
1 Average normal temperature of silage	46
2 Moisture content and physical characteristics of the ensiled maize stover	84
3 Proximate composition of ensiled maize stover.. .. .	85
4 Fiber fractions (%) of ensiled maize stover	88
5 Dry matter intake and coefficient of preference of WAD sheep fed .. ensiled maize stover	90
6 <i>In vitro</i> fermentation parameters of maize stover at 24hrs incubation period	105
7 <i>In vitro</i> fermentation characteristics of ensiled maize Stover at 24hrs incubation period	107
8 Ingredient and crude protein composition (%) of the concentrate supplements fed to WAD sheep	117
9 Ingredient composition (%) of the experimental diets fed to WAD sheep	121
10 Chemical composition (%) of the ensiled maize stover and concentrate	123
11 Feed intake and performance of WAD sheep fed ensiled maize stover and concentrate supplements	125
12 Polynomial regression of the performance of WAD sheep fed ensiled Maize stover and concentrate supplements	127
13 Apparent digestibility (%) by West African Dwarf sheep fed ensiled maize stover and concentrate supplements	129
14 Nitrogen utilisation by WAD sheep fed ensiled maize Stover and ConcentrateSupplements.. .. .	131
15 Polynomial regression of nitrogen balance of WAD sheep fed ensile maize stover and concentrate supplements.	133
16 Total digestible nutrient intake (%) by West African Dwarf sheep fed ensiled maize stover and concentrate supplements.. .. .	135
17 Haematological parameters of WAD sheep fed ensiled maize stover and concentrate supplements	148
18 Serum biochemistry constituents of WAD sheep fed ensiled maize stover and concentrate supplements	150

LIST OF FIGURES

Figures		Page
1	pH of ensiled maize stover	86
2	The temperature of ensiled maize stover	87

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LIST OF PLATES

Plate		Page
1	Maize plant	79
2	Chopping exercise	80
3	Chopping maize stover.. .. .	81

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CHAPTER ONE

1.1 INTRODUCTION

The livestock sector plays a significant economic role in most developing countries and is essential for the food security of their rural population (Kayouli and Lee, 1999). In Nigeria, livestock rearing plays an important role in the livelihood of Small-Scale farmers and contributes to the regional and national economic development. It contributes to poverty alleviation and provides elements that are essential to the national economy, such as: traction, power, transport, manure as fertilizer and fuel, food, fibre, leather, saving bank, and by generating significant household cash income through sales of live animals or livestock products. In recent years, the human population has increased rapidly and the demand for food in particular livestock products is expected to increase in all developed and developing countries (Phengvilaysouk and Wanapat, 2008, Kayouli and Lee, 1999).

Animal proteins are very important in human nutrition because they contain essential amino acids which are more balanced and readily available to meet human dietary requirements than proteins of plants origin. Unfortunately, animal protein is in limited supply in Nigeria due to low productivity of livestock animals. Several factors are responsible for this low productivity in the livestock sector such as low genetic potential of indigenous animals, poor feeding, susceptibility to diseases and parasites, infections, etc. However, among the major constraints limiting the development of livestock production in many developing countries, inadequacy of animal feed resources is most often the crucial factor. Feed shortages, both quantitatively and qualitatively, are limiting livestock productivity (Kayouli and Lee, 1998). Feeding i.e. nutrition is the most important limitation to livestock production in Nigeria. In many tropical countries, such as Nigeria, the bulk of livestock feed resources comes from grazing mainly poor quality annual and perennial grasses from natural pastures often at a late stage of maturity. In other part of the country, livestock farming is increasingly being limited by the restriction of grazing lands. During the long dry season (at least 6 – 7 months per year) animals are on poor quality feeds characterized by low palatability, low intake and low nitrogen concentration (Kayouli and Lee, 1999).

Mako (2009) also described the major constraint to livestock production in developing countries as inadequacy of feed in terms of quality and quantity all year round. Most ruminants' livestock, especially cattle, sheep and goats obtain most of their nutrients from herbage growing on poor soils. Majority of livestock in Nigeria are reared on very low planes of Nutrition. These animals survive on grazing of the savannah land during the rainy season which is sometimes limited to four or five months of the year. Another problem is that these herbage often grow fibrous and are of low digestibility. These animals gain weight slowly in the rainy season and loose it rapidly in the dry season (Babayemi and Bamikole, 2006) due to all-year round feed inadequacies.

There is usually limited and often inadequate supply of forages especially during the dry season (Babayemi *et al.*, 2003). Smith *et al.* (1995) observed that dry season feeding of ruminant has always been a critical problem to livestock farmers as feed supplies are limited both in quantity and quality. This scarcity of forage also results in decreased feed supply to ruminant, high prices of conventional animal feedstuffs and an overall reduction in the productivity of ruminants in terms of meat and milk thus leading to an inadequacy of their utilisation by man. The natural pastures and crop residues available to animals during the dry season after crop harvest are usually fibrous and devoid of most essential nutrients which are required for improved microbial fermentation and improved performance of the animal (Dixon and Egan, 1987).

The major hindrances to the abundance of all year round high pasture are in two folds. First is the obvious fast rate of infrastructural development and constant change in government policies. Another factor is the constraint rise in population in Nigeria. There is an uncontrollable rise in human head count. The net effect is the negative impact on the available space for herbage production and limited green area for grazing animals (Makkar, 1994). Feeding, alone accounts for approximately 60 – 80% of the total cost of animal production (Aregheore, 2000). The conventional feed resources are also in limited supply, very scarce and expensive. This stemmed from the perpetual competition between man and his livestock. Such feedstuff is maize, a source of dietary energy for livestock farmers. It thus becomes cost ineffective for livestock farmers. All these factors prohibit the use of such feed ingredient for ruminants that may require a large quantity to satisfy energy requirements. Smallholders (Stur *et al.*, 2002) own 95% of all livestock and most of the households produce food mainly for subsistence (Chantalakhana, 2001).

Unfortunately, in most towns and villages, free-range system of animal rearing has been the order of the day. Animals are left to roam the streets as scavengers. These animals face the hazardous conditions of being beaten or knocked down by vehicles or at times being poisoned by angry farmers whose crops have been damaged.

There is an increased demand for food supply especially animal protein to meet the nutritional requirements of the world's ever growing population of about 6.3 billion people (Makkar, 2004). With increasing demand for livestock products as a result of rapid growth in the world economy and shrinking land area, there is a need to increase livestock production (Anandan *et al.*, 1996). Future hopes of feeding the millions of people and safeguarding their food security will depend on the enhanced and efficient utilisation of unconventional resources that cannot be used as food for humans but as feed for livestock (Makkar, 2004).

Crop residues (straw and stover from sorghum, millet, wheat, barley, rice, maize) play a key role in animal feeding mainly in Africa. Researchers in the last three decades realized the need for a pressing integration with the use of less popular feed alternative such that ruminant production could be sustained all year round. Thus efforts have been geared to improve supplemental feeding with the use of grasses, crop residues and agro-industrial by products (AIBPs) over the years. They are readily available in their local area from crop cultivation and industrial processing. They are easily afforded by the farmers at almost zero cost and are important sources of roughages for ruminants. The AIBPs and crop residues have been described as low-quality roughages due to high levels of cellulose, hemicellulose, lignin and low fermentable carbohydrates (Phengvilaysouk and Wanapat, 2008). However, ruminant livestock have the advantage of their unique ability to synthesis high quality protein from non-protein nitrogenous compound (NPN) through the action of micro-organisms present in their digestive tract (Adeleye, 1991).

Various authors (Adebiyi, 2006; Mako, 2009) have reported fermentation as an important tool of upgrading AIBPs and crop residues, particularly the protein level for adequate utilisation by the microbes in the rumen. It also aids in the breakdown of the fibrous cell wall thereby making the feed more susceptible to microbial attack. Achinewhu *et al.* (1998) reported fermentation as

being responsible for product stability, flavour development, fibre breakdown and enhanced nutrient content of feed through the biosynthesis of vitamin, microbial proteins and fibre digestibility. It is recognised in developed countries that the production of silage of high quality cultivated forage can be a valuable component for the development of a high performing and low-cost system of animal production, using a relatively low level of purchase concentrates. In a bid to actualize the mandate of better utilisation of unconventional feed sources, the potential of ensiled maize stover as possible source of feed for ruminants especially during the dry season is being considered.

In Nigeria, maize is one of the arable crops grown abundantly in all over the region. Maize is used in a variety of ways whether mature or not. The grain yield amounts to 1.37 million tones per year (FMAWR, 1988). Maize stovers are in abundance especially in the early season. The residues are estimated to be about 4.11 million tones per year (Adebowale, 1989). The residue are often burnt off or ploughed back into the soil. Maize is an excellent fodder plant and is often grown as silage. Feed energy suitable for ruminants may be obtained from the maize stover. Ruminants have the potential to utilize the stover, but the stover dries up quickly, thus there is a need for preservation. Ensiling has been reported to effectively conserve forages and fodder crops (Babayemi, 2009). Research has shown that the ensiling of crop residues, forages and agricultural by-products is the most suitable method of conservation for long period (Bouque and Fiems 1988; Kayouli 1989; Kayouli *et al.*, 1993; Hadjipanayiotou 1993; 1994; Lien, *et al.*, 1994; Kayouli and Lee 1998). The research is aimed at examining the utilisation of ensiled maize stover and concentrate supplements by West African dwarf sheep.

1.3 Justification of the Study

- Maize is one of the arable crops and being a staple food as fresh maize especially in the early season, the stovers are abundantly available.
- Ruminants are capable of converting roughages and crop residues to human edible foods, assuring a well utilized maize stover.
- Maize stover is fresh, high in moisture content and thus prone to spoilage over a short period, indicating the need for conservation process.

- Hay as forage preservation is cheaper but is made while sunshines, thus the necessity to adopt ensiling method of conservation.
- Maize stover is low in crude protein to meet the requirement of small ruminants, showing the need for supplementation and microbial fermentation to increase the crude protein content.

1.2 Objectives of the Study

The objectives of the study were:

- (1) To ensile maize stover and maize stover with different additives
- (2) To determine the effects of additives on quality characteristics and chemical composition of ensiled maize stover.
- (3) To assess the dry matter intake and coefficient of preference of ensiled maize stover.
- (4) To evaluate the *in vitro* fermentation characteristics and dry matter degradability of ensiled maize stover.
- (5) To assess the performance characteristics, digestibility and nitrogen balance of growing WAD sheep fed with ensiled maize stover and concentrate supplements.
- (6) To determine the haematological and serum biochemistry of growing WAD sheep fed with ensiled maize stover and concentrate supplements

CHAPTER TWO

LITERATURE REVIEW

2.1 SMALL RUMINANTS POPULATION

Nigeria has a wide diversity of climatic zones from the humid forest zone of the south to the mountainous cool belt of the plateaus in the Middle Belt and to dry Sahel region of the north. Vast areas of the forest region lie under the challenge of blood sucking insects; hence, the production of ruminant animal species is somehow limited. It has been estimated that over 90% of the cattle 70% of the sheep and 75% of the goats are found in the “Cattle territory” which lies within the extensive dry savanna land of northern Nigeria. Within the Southern forest zone and the Guinea Savannah are found the remaining livestock which are made up of the indigenous West African dwarf breeds of sheep and goats. FAO (1988) reported that the West African dwarf sheep contributed four million heads to the total livestock resources of Nigeria while Adu and Ngere (1979) reported that sheep contributed 11% of the meat supplied from slaughter houses in Nigeria. In 1979, sheep and goats contributed about 33% of the nation’s total meat supply (Brinkman and Adu, 1977). Most of the sheep and goats are reared under traditional management systems where animals are left to roam with little or no supplements. Small ruminant rearing is an age long traditional production system where animal are managed under extensive system but in the last decade, they are been reared along side with cultivation (Ajala *et al.*, 2008).

Sheep and goat population is higher than that of cattle in Nigeria connoting great potential for productivity. Sheep was reported to be 33,000,000, goats; 52,000,000 and cattle 16,000,000 (FMA, 2008). Low yield accustom this type of management system. The low productivity of the indigenous livestock may be attributed to diseases and parasites, infections problem, heat and humidity of the climate, low genetic potential of the indigenous animals, poor feeding and management, lack of training and experience of the local people in animal husbandry and absence of infrastructure necessary to supply the needed inputs for animal production, processing and distribution. Out of all these, poor feeding and nutrition are the most critical factors responsible for low production of indigenous livestock. However, performance records of the indigenous livestock from the organised system of production such as our research stations; university farms etc. suggested a consistent better performance. Such results indicate promising

prospects of increased production that may accrue from the application of improved production system. Therefore, no effort should be spared to make sure that high plane of nutrition coupled with efficient management are delivered to the existing livestock. Although livestock number in Nigeria increased over the years, the trend in total meat production has been smaller than would be expected to meet requirements of a large population as that of Nigeria. Growth in food demand presently exceeds growth rate in food production. Coupled with high rate of population, growth, the scenario has resulted in a wide gap between supply and demand. Consequently there is a high rate of increase in prices of livestock product.

2.2 ROLE OF LIVESTOCK IN FARMING SYSTEMS

Livestock production is an integral component of agricultural sector of the economy in a developing country. The context for productivity enhancement and increased socio-economic contribution relates to large population size, wide distribution across various agro-ecological zones and production systems as well as diversity of breeds. They are important in supporting the livelihoods of poor resources farmers throughout developing world. Livestock contributes to the protein-calorie intake of people in both developed and developing countries. Livestock is a source of income for small scale farmers in developing countries; which they use for purchasing food, agricultural inputs such as seed, fertilizers etc. Cash can also be generated from the sale of livestock products such as milk, eggs and meat (Kayouli and Lee, 1999).

Ruminants such as sheep, goats and cattle are used to reduce forest under growth, in some countries to reduce risk of forest fires. Bovines, equines, cammellias and elephants are used as sources of drought power for a variety of purposes such as pulling agricultural implements, pumping irrigation and skidding in forests. In many countries, cow dung is used as fuel for cooking, plaster and other building materials. The ash from this dry dung is used as fertilizers. Biogas production from manure is an excellent substitute for fossil fuel or fuel wood for farmers in tropical countries. Manure used for these is obtained from pigs, horses, camels, cattle and poultry (Kumar and Biswas, 1982). By-products from the slaughter house can be used as a good source of protein (offal and visceral) and mineral supplements (bones) in animal feeds. Crop residues such as straws are more efficiently utilised through ruminant feeding instead of burning them and causing air pollution which contributes to global warming. They ultimately find their

way back into the soil to improve its structure and water retention/holding capacity. In some societies, their main reasons for keeping animals is to be sold during social events and religious festivities when they are usually of high demand which thereby increases the sales price.

2.3 CROP RESIDUES

Crop residues are materials which are generated after the crops have been harvested (Dixon and Egan, 1987). They are the parts of plants left in the field after the crops have been harvested and thrashed. The term “crop residue” gives a false impression of the value of the straws, stovers, and other vegetative parts of crops that remain after harvest since it implies something left over that nobody wants (Latham, 1997). This is especially more so since many farmers burn them or otherwise disposing of them. Crop residues are good sources of plants nutrients, are primary sources of organic materials added to the soil, and are important components for the stability of agricultural ecosystems. Crop residue is not a waste rather a tremendous natural resources. About 25% of Nitrogen (N) and phosphorus (P), 50% of sulphur (S), and 75% of potassium (K) uptake by cereal crops are retained in crop residues, making them valuable nutrient (Samra *et al.*, 2003). Crop residues have been estimated to account for about 25 percent of the total feed energy suitable for ruminant livestock in both developed and developing countries (Biwi, 1986). Two third of these crop residues are from cereals. They are characterised with low digestibility, metabolisable energy, nitrogen and contents of available minerals. Crop residues are usually fibrous, low in nutrient and are not directly edible but could be fed to animal. Babayemi and Bamikole (2006) described crop residues as bulky with high fibre, low protein and poorly degradable, while Dixon and Egan (1987) further explained that they are derived from the processing of a particular crop or animal products usually in an agricultural firm. They are available in each locality of production. The nutrient composition and nature of crop residue depends on the amount and types of crop grown in that area.

Within tropical systems, Africa is second to Asia in crop residue production of 2.2 tonnes of dry matter per livestock unit of herbivores (Kossila, 1985). In Ethiopia, it is estimated that a total of 13 million tones of crop residue are annually produced (Seyoum and Zinash, 1998). In Nigeria, like other crop producing countries, crop residues are usually available after the cropping season has ended. Crop residues outputs tend to increase with rural population density and farming

system in place. Crop residues are distinct from by-products of processing such as brans, oil cakes, etc. Usually crop residues are grouped by crop type such as cereals, grain legumes, roots and tubers, etc. (Nordblom and Shomo, 1995).

In developing countries like Nigeria, a great proportion of food crops are grown by small scale farmers on their small plots. Nigeria produces a wide variety of cereals and other food crops. After these crops have been harvested and processed, various residues and by-products remain for example, stovers, husks, cobs and brans from cereal crops, and shells, pulps, peels and tops from other crops. Usually farmers in Nigeria remove legume and cereal grains from seed hulls and leave the crop residue at the threshing grounds. A very small fraction of total crop residues produced is used as feed for local sheep and goat during the harvest season while a greater part of the seed hulls is left to decay. Crop residues occur fairly generally in the rural areas of Nigeria, but their potential for animal feeding has not been fully exploited. It is however possible to increase the nutritive value of some of these residues, thus improving livestock productivity. Although research work has been in top gear to evaluate the use of these crop residues as feed, a large amount of these agricultural wastes produced on both private and government could be wasted. In such findings (Ayoola and Ayoade, 1994) established that in some eastern parts of Nigeria, more than 60% of livestock farmers are not aware of the value of crop residues.

Research to date is geared towards the determination of biological value of residues as they occur, rather than on methods of increasing this value. In an attempt to improve the nutritional quality of fibrous residues, it has been confined mainly to physical treatments, such as grinding. There is the need to explore more and where need be, fully apply other methods of treatments if their potential value as animal feed is to be realised. Presently, there is no competition between man and farm animals for crop residues. Agishi (1986) described the Nigerian livestock industry as “a sleeping giant that will one day awake up to assume a major role in the country’s economy. This awakening may come only by developing applied research techniques to improve the production of livestock in rural areas and by encouraging large scale animal producers to use non-conventional feedstuffs such as crop residues in feed formulation. Hence, the concept of matching ruminant livestock production with available feed resources (Preston and Leng, 1987;

Dargie,1994), has therefore intensified research into more use of crop residues and agro-industrial by products in most countries in the tropics and sub- tropics.

2.3.1 UTILISATION OF CROP RESIDUES

In feeding livestock, the aim is to provide the animals with a balanced ration throughout the year with sufficient nutrients to meet the animal's nutrient requirements for maintenance and production. In Nigeria, forage and fodder supply during the dry season and drought periods is a very important factor limiting livestock productivity. Shortage of feed resources often imposed major constraints on the development of animal production in the tropics and sub-tropics. Considerable quantities of crop residues are generated every year in most developing countries in the tropics and sub-tropics. These are suitable for feeding livestock, however because of lack of technical know-how, they are lost or under-utilised. An intensive feeding system based on locally available agro-industrial by-products and crop residues is an alternative promising feeding system to rear ruminants economically. Ruminants because of their rumen physiological adaptation can utilise inexpensive agro-industrial by-products and crop residues to meet their feed requirements for maintenance, growth, and production. Agro industrial by- products and crop residues in the diets of ruminants support growth and lactation and result in the production of human edible food. Although most have low nitrogen content ,more fibre as well as low nutrient density ,effective processing can raise their nutritive value (Reddy and Reddy, 1992; Areghore, 1994)

Fruit, vegetables and root crops are increasingly integrated in the farming system and play a key role as staples in the human diet in most developing countries. Consequently, there is a wide range of valuable by-products and residues resulting from food crops. Cropping systems and food processing are often inefficiently or totally under-utilised and wasted in developing countries (Kayouli and Lee, 1998). Little information is available on the extent to which small holder farmer in Nigeria use crop residues as livestock feed. It is very likely, however, that these resources are under-utilised. Even when they are utilised, farmers may not be able to incorporate them efficiently into their year-round livestock feeding programmes because they lack suitable storage facilities and technical know-how on treatment and processing methods and on formulating feed ration. The importance of crop residues as animal feeds is increasing

significantly as rangelands are increasingly becoming smaller and smaller making way for human settlements for the increasing human population. On the other hand, the increasing world demand for both cereal and legume grains for both human and animal food is producing abundant supplies of crop residues (Tsopito, 2003).

The importance of crop residues as potential animal feed varies with the type of crops grown, also with the yields of the plant parts. In many parts of the world (particularly in areas with low yield potential), crop residues are essential for feeding animals. As the grazing fails, accepted practice is to redress the deficit with crop residues (Smith, 2002). Animals are left to graze freely on harvested fields or straw is collected off the land and taken to the pen to feed livestock and prepared bedding (Samra *et al.*, 2003).

Crop residues are among the most widely available, low-cost feeds for ruminants in the majority of developing countries (Smith, 1993). In industrialised countries, the contribution of straws rarely exceeds 20 to 40 percent of the diet, the rest of the ration being cereal grains, highly-fertilised grasses and legumes and oil seed cakes (Preston, 1986). In mixed crop livestock farming systems, livestock provide important inputs to cropping, especially manure and traction whereas, crops in turn provide livestock with feed in the form of residues and by-products (ILCA, 1992). Where crop and livestock production are segregated, most crop residues are wasted or used for non-feed purposes like bedding, mulching, firewood and building material (Kossila, 1988). Second to cattle feeding, wheat and barley straws are used as a filling material in making local mattresses in Ethiopia.

Wheat straw are used relatively less for livestock feeding because of the health problem that it causes. Ninety five percent of the households growing wheat reported that the straw is poor in feeding value and causes health problem to cattle. However, scientific reasoning for this is not available, except that McDonald *et al.* (1995) found wheat straw to be so poor in nutritional value (unless alkali treated) that its usage as a feed for farm animals is not recommended. Most crop residues are fibrous and therefore of only low to moderate nutritive value. Some of them have special physical and chemical characteristics that make them difficult to incorporate into the diets of animals. Cereal crop residues are low in nitrogen, minerals, vitamins and available

energy. They have low digestibility coefficients. They also have a dry matter intake (DMI) of 1.5 – 1.8% and thus sometimes cannot even support maintenance needs of the animal (Tsopito, 2003). However, in the absence of quality feeds, they still can provide the staple energy feed for ruminants. On the other hand, leguminous crop residues may contain higher levels of nitrogen adequate to meet maintenance and production requirements of some classes of animals (Tsopito, 2003).

2.3.2 CONSTRAINTS TO CROP RESIDUE UTILISATION

In spite of the fact that large quantities of fibrous crop residues are used as animal feeds in many developing countries (Kossila, 1988), there are certain constraints to their efficient utilisation. Kossila (1985) stated that a much lower level of crop residue utilisation is possible because of problems of collection, transportation, storage, processing, alternative use, seasonal availability and an apparently poor nutritional value. The bulky nature of straws and stovers make it difficult and costly to transport them, thus inhibiting their greater and efficient utilisation for livestock feeding. Owen and Aboud (1988) cited the risk of loss due to fire and reduction in nutritive value due to moulding and damage by vermin and insects as the major problems associated with the storage of crop residues. The consequences of lack of transportation and improper storage were reported to have been the major causes of crop residues wastage. With regard to crop residue wastage, Thairu and Tessema (1987) stated that small parts of the thousands of tonnes of crop residues available in the crop land of Kenya are used as a feed due to difficulties of collection, transportation and storage. Sibanda (1986) reported that farmers who do not collect their stover, but leave it in the field, could possibly lose half of its value through trampling by animals.

Tesfaye and Nashon (1999) reported that problems associated with feeding of crop residues result mainly from improper feeding practices or are caused by the physical nature of the residues. Both *in situ* grazing and feeding from threshed residue upon the ground are regarded as improper feeding practices in the sense that they result in inefficient utilisation of the residues as a result of the trampling effect of animals, and the spoilage by their dung and urine. In investigating the role of crop residues in intensive smallholder system in the tropics, Smith (1993) reported that when left on the field, crop residues rapidly deteriorate, and a larger amount is usually trampled upon and wasted.

Tesfaye (1999) observed that physical nature of residues as a feeding problem is evident mostly in maize and sorghum stovers. These residues are hard and stemy so that, animals prefer the finer parts thus causing significant losses of the residues. El-hag and Kurdi (1986) identified bulkiness, location of the residue in areas with low animal population density, poor nutritive value and unsuitability for direct animal use as the major problems of crop residue. It has been observed (Owen, 1985; Owen and Jayasuriya, 1989; Devendra, 1991) that despite impressive animal performance response on experimental stations to crop residues utilisation, farmer's uptakes of research findings have been minimal. This trend has been partly as a result of much less effort being put into identifying the socio-economic factors limiting greater utilisation of crop residues and adoption of new feeding systems by farmers.

Various livestock production systems can be found in developing countries and the feed resources they use also varies. An appropriate feeding strategy is required because of the many nutritional interactions that exist within different crop residues and between these feeds and the tropical/sub-tropical regions. The existence of such interactions makes it difficult to use classical feed composition and nutrient requirement standards for ration formulation. An attempt to do this usually results in the actual levels of animal performance achieved being much less than those predicted from the feeding standards (Tsopito, 2003).

The availability of crop residues at the farm level depends on production levels and also on a variety of social and economic factors. Land, crop and animal ownership patterns, cultural practices, the use of modern crop varieties for market and non market exchanges, all influence a farmer's access to the residues that are locally produced. Although the crop residues are abundant, there generally seems to be low utilisation of crop residues by small holder farmers. Lack of utilisation of crop residues could probably be due to lack of knowledge of the importance of these residues as livestock supplement feeds and financial constraints involved in harvesting, storage and conservation. These factors should be considered in national assessments of crop residues production as they may limit improved utilisation (Tsopito, 2003).

2.3.3 CHARACTERISTICS OF CROP RESIDUES

Only a part of agricultural products can be utilised by man himself. The amount of crop residues for feeding farm animal can be considerable. There is a considerable variation in quantity and quality of residues among the crops, influenced by varieties, climate, season and stage of harvest. The most important parts of roughage are the aerial parts (stems, leaves). These can be utilised fresh, dry, ensiled, cut or grazed, in the field or in the stable barn.

Human do not consume crop residues. These residues contain high amount of fibrous material, and are not easy for disposal due to threat on environmental pollution. Statistics on production and utilisation of fibrous residues in Nigeria is inadequate. However, the production of residues could be fairly estimated accurately from crop production, if reliable data are available.

Low quality roughage is found in poor grazing rangelands. It also include huge amount of cereal crop residues such as rice straw, bean straw, maize stover, sorghum stover, corn cobs and rice hulls. Analysis of roughages by detergent procedures (Goering and Van Soest, 1970) showed that they are high in lignin, cellulose and hemicellulose.

Most cereal crop residues are characterised as highly fibrous, low crude protein, low available energy, high biomass and deficient in certain minerals. These low quality roughages are inefficiently utilised by ruminants. This is due to low digestibility and poor nutritive value associated particularly with cereal straw. Their utilisation is also limited because of low voluntary intake of the animals due to their bulkiness which makes transportation more costly.

The chemical composition of residues varies with the variety of plant (Kharat, 1974; Saleem and Jackson, 1975), location (Van Soest and McQueen, 1973) and agricultural practices employed in the growing of the crop and handling of the residue from which they are obtained. Chahal (1985), referred those differences between crop residues and wood residues to be due to their chemical composition. He found that crop residues contain 30 – 40% cellulose; 16 – 25% hemicelluloses, 3 – 13% lignin and 3.6 – 7.2% crude protein, while the wood residues contain 45 – 56% cellulose, 10 – 25% hemicelluloses and 18 – 30% lignin.

2.4.0 IMPROVEMENT OF NUTRITIVE VALUE OF CROP RESIDUES

Approximately 2 billion tones of cereal grains and 140 million tones of legume and oil seeds are produced throughout the world each year, which yield an estimated 230 million tones of fibrous material as part of a variety of crop residues (Choct, 1998). In legumes, non starch polysaccharides (NSP) also play a role as an energy storage material. Longe (1988) and Dierick, (1989) advocated the increased utilisation of non-conventional feed resources in non-ruminant feed. They suggested processing techniques, which are simple and inexpensive, and do not significantly increase costs but still make it worthwhile in term of nutrient availability. With regard to crop residue treatment, Smith (1993) listed chopping, grinding, ensiling with urea or animal manure, and ammoniation using urea as the most appropriate methods of improving the feed value of crop residues at the small holder level. Processing techniques widely documented in literature could be grouped into physical, chemical and biological treatments.

2.4.1 Physical treatments

The limiting factors in the utilisation of poor quality roughages as a production feed are related both to their low voluntary intake and poor digestibility. The grinding of roughages which reduces the particle size is a simple physical treatment. It results in an increased voluntary intake but the digestibility is either not affected or reduced. Ground poor quality roughages (straws, stovers) have been successfully used in Canada, USA, etc. to replace the major portion of the forage fraction of the ruminant ration. High pressure – high temperature saturated steam has been used as a physical treatment for improving the nutritive value of poor quality roughages by many workers (Fyriier, 1973; Rexen *etal.*, 1976; Ranjhan, 1981; Smith, 1993; Ranjhan, 2003).

In small holder livestock systems, most physical treatments of crop residues are either too expensive, the equipment is not available or too labour intensive. However, there are benefits in reducing particle size (not necessarily grinding), for ensiling and in stall-feeding. Reduction of particle size can be achieved by using a power driven chopper, a hand operated chaft cutter a guillotine blade or a good sharp cutlass (Ranjhan, 2003). There are other advantages, in that the surface area of non-lignin material exposed to microbial attack in the rumen is increased, thus increasing the rate of digestion, thereby reducing a possible limitation to intake (Van Soest,

1982). The smaller the particle size the less scope there is for selection. Osafo *et al.* (1977) found that chopping increased intake in sheep but not in cattle.

2.4.1.1 Gamma-irradiation

Gamma-irradiation according to Doyle *et al.* (1986) may reduce resistance of fibrous residues to physical degradation without the necessity for fine grinding. McManus *et al.* (1972) indeed noted that irradiated rice straws had a shorter mean retention time in sheep than non-irradiated straw, suggesting that irradiation rendered the straw more susceptible to physical breakdown. Walker (1984) also reported that irradiation solubilises cellulose, hemicellulose and lignin in the cell wall. *In vivo* results, however, do not confirm these apparently beneficial effects, as the procedure has in general been shown to depress dry matter digestibility, and to have no effect on voluntary intake (McManus *et al.*, 1972). The process obviously has no practical application for farmers in humid Africa.

2.4.2 Chemical treatment

The nutritive value of the poor quality roughages can be improved by the alkali treatment. The potential for increasing digestibility and intake of fibrous residues through treatment with alkali has been widely researched and comprehensively reviewed (Sundstol and Owen, 1984). The cell wall constituents of fibrous crop residues are fermented more slowly than the cell contents of the same plant material. The rate of fermentation of such crop residues can be increased by processing techniques that employ chemical treatment. This treatment has been known to increase both the digestibility and rate of intake of most of these crop residues (Jackson, 1977). The treatment procedure will vary according to circumstances. Smith *et al.* (1989) found that 5% urea, in solution, added at a rate of at least 20%, weight for weight, solution to dry stover, followed by an incubation period of five weeks gave the greatest improvement. The stover had been rotor slashed before treatment. Although successful upgrading of whole stover is possible, reduction of particle size aids ensiling (Smith *et al.*, 1989). Urea treatment is relatively easy to apply and is effective.

2.4.3 Physico-Chemical Treatments

There is physical evidence that combining physical treatments such as milling, grinding and steaming, which decrease particle size, with chemical treatments, increased the effectiveness of

the chemicals (Thiruchittampalam and Jayarisuya 1978), although the effects may not always be additive (Coombe *et al.* 1979). In any case, such severe physico-chemical treatments may be out of reach of village farmers in humid West Africa.

2.4.4 Biological treatments

Of all the available biological methods, ensiling appears to be the only feasible and available method for small holder farmers. Composting causes substantial losses of organic matter leading to an increase in ash and lignin content (Doyle *et al.*, 1986), with a resultant decrease in nutritive value. Fungal and purified enzyme inoculations have also been used, and have been shown to be useful only under ideal conditions of temperature, pH, aeration and moisture content. Ensilage of crop residues with forages or animal wastes appears to be a simple effective means of improving crop residue quality.

Reports from Asia indicate that silages made from rice straw and (i) water hyacinth (Chhibbar and Singh, 1971), (ii) potato vines (Krishna, 1982) (iii) poultry litter (Neog and Pathak, 1976) supply enough nutrients for maintenance. In Cameroon Fomunyan and Meffeja (1986) fed goats maize stover ensiled with brewers grains, and obtained digestibility values similar to those reported for sodium hydroxide and ammonia treated maize stovers. Simple processing such as chopping, ensiling with urea or animal manure or chemical treatment with crop residue ash solution are the more practical and economical means of improving the nutritive value of crop residues under the small scale farming context.

2.4.5 Strategic Supplementation

According to Devendra (1985) the characteristics of a maintenance feed for adult ruminants are: a crude protein level of 6 – 7%, a dry matter digestibility of 50 – 55%, and a dry matter intake of about 1.7 percent of body weight. Chemical or other treatments reviewed earlier may improve intake and digestibility, but unless adequate supplementation of deficient nutrients is made, much of the additional energy released will be inefficiently used. Adequate supplementation is therefore required for efficient utilisation of crop residues.

Preston and Leng (1984) have suggested that to optimize the utilisation of crop residues, nutritional supplements should provide the following (i) fermentable energy, (ii) fermentable nitrogen (iii) micronutrients e.g. sulphur, phosphorus and B vitamins (iv) roughage (v) by-pass protein (vi) by-pass energy. The first four ensure an adequate rumen ecosystem, while the last two complement the needs of the animal as a whole. In other words, adequate supplementation is essential for proper utilisation of crop residues. Alhassan and Akorfur (1982) reported a significant increase in the digestibility of rice straw fed to West African Dwarf sheep, when the straw diet was supplemented with oil palm slurry. More impressive result was obtained by Sudana and Leng (1985) when they supplemented sheep fed a basal straw diet with urea – molasses (fermentable nitrogen and energy) and cotton seed cake (by-pass protein). Winugroho and Chaniago (1984) reported an increase in the nitrogen content (1.0 to 2.4 percent) and *in vitro* dry matter digestibility (35 to 47 percent) of pelleted rice straw with cassava leaf. Acceptable dry matter intakes and growth rates were obtained in goats fed diets consisting entirely of crop residues by Soedomo-Reksohadiprodjo (1985). The combination of crop residues was such that the basal diets (corn stover, sorghum stover and sugarcane tops) were well complemented. The extent and rate of digestion of fibrous feeds are increased by a nitrogen supplement, resulting in a greater dry matter intake (Preston and Leng, 1987). This is reflected in the extent of live weight change.

Evidently, supplementation along the lines suggested earlier will improve crop residue utilisation by goats and sheep. The choice of supplement must tilt towards the more readily available and less costly alternatives. As availability and cost of the various supplements may vary from one area to another, no blanket recommendation can be made. Nevertheless, locally available protein sources such as legume residues, pods, green fodder or forages (e.g. leucaena, gliricidia, cassava tops) poultry manure, urea-molasses mixture in liquid or block form, peels of tubers, and oilseed residues (e.g. from the extraction of cottonseed, soyabean and sunflower oil for household use) may play increasingly significant roles as supplements to crop residues feeding (Sansoucy, 1986, Preston and Leng, 1987; Smith *et al.*, 1990; Manyuchi *et al.*, 1992a and 1992b; Ncube and Mubaiwa, 1994; Mupeta and Makombo, 1995). By-pass protein sources such as oil seed cake and fish meal may be too expensive for use in the small scale farming system.

2.4.6 Mineral Supplementation

At the level of productivity obtained under unimproved feeding systems in the small scale farming setting, goats and sheep do not often show symptoms of mineral deficiencies or respond to mineral supplementation. Responses to mineral supplementation only occur after the major nutrients imbalances have been corrected. A feeding strategy based on treated and supplemented crop residues may correct these major nutrient deficiencies and improve productivity to such an extent that mineral requirements increase. Mineral supplementation may then become important not only to avoid deficiency problems but also improve performance further.

According to Little (1985) crop residue based diets are more likely to be deficient in sodium, copper, and phosphorus. These are the same minerals found to be marginal or deficient in tropical grasses (Kabaija, 1985). Preston and Leng (1986) reported that most straws are deficient in the same three minerals in addition to sulphur, cobalt and calcium. The high concentrations of oxalates and silicates in some of the straws, such as rice straw, may further reduce the availability of calcium and magnesium, which are lost as silicates and oxalates in the urine and faeces. It is possible therefore that goat and sheep fed mainly on well supplemented crop residues may become deficient in other minerals. Meanwhile, as suggested by Little (1985), routine provision of salt supplements to animals fed crop residues is necessary.

2.5.0 FERMENTATION

The scientific meaning of fermentation is the energy-yielding anaerobic metabolism of nutrients, such as sugars (Motarjemi, 2002). Fermentation converts these nutrients mainly into lactic acid, acetic acid and ethanol. In a broader meaning fermentation can be described as the change in state of the physical and chemical condition and make up of a substance or substrate (feed) to a more stable form. This process changes the total make up of the substrate, by breaking down the complex formation of the feed through natural or artificial introduction of microorganism (fungal, bacteria or yeast) which breaks down the complex cell wall into a more simple form. Fermentation will gradually change the characteristics of the food by the action of enzymes, produced by bacteria, moulds and yeast (Motarjemi, 2002).

Fermentation is a process of undergoing an effervescent change as by the action of yeast; in a wider sense, the transformation of an organic substance into new compounds by action of fermentation. Fermentation could be aerobic or anaerobic. Fermentation is one of the oldest applied biotechnologies, having been used in food processing and presentation as well as beverage production for over 6,000 years (Motarjemi, 2002). The fermentation process of staples serve as a means of providing a major source of nourishment for large rural populations, and contributes significantly to food security by increasing the range of raw materials, which can be used in the production of edible products (Adewusi *et al.*, 1999). Fermentation enhances the nutrient content of feeds through the biosynthesis of vitamins, essential amino acids and proteins, by improving protein quality and fibre digestibility. It also enhances micronutrient bioavailability and aids in degrading antinutritional factors (Achinewu *et al.*, 1998). This process of substrate break down is advantageous in many ways. It increases the surface area of the feed particles for the host's microorganisms for adequate utilisation.

- (a) Fermentation brings about a more stable product thereby increasing the shelf-life of the product.
- (b) It helps to improve the nutritive value of the substrate through protein enhancement.
- (d) It enhances acceptability of the feed.
- (d) The aroma of the end substrate is improved.

2.5.1 SOLID STATE FERMENTATION

Aerobic microbial transformation of solid materials or “Solid Substrate Fermentation” (SSF) can be defined as the application of living organisms and their component to industrial products and the process not industrial in itself, but an improvement technology that will have a large impact on many different sector (Hamlyn, 1998). Aderolu (2000) considered SSF as a process in which solid substrate are decomposed by known mono or mixed cultures of micro-organisms under controlled environmental conditions, with the aim of producing high quality product. The substrate is characterised by relatively low water content (Zadrazil *et al.*, 1990).

Many authors (Chahal, 1992; Haltrich *et al.*, 1996; Jecu, 2000) have reported that solid state fermentation (SSF) is an attractive alternative process to produce fungal microbial enzymes using lignocellulosic materials from agricultural wastes due to its lower capital investment and

lower operating cost. For the reasons stated, SSF process will be ideal for developing countries. Solid state fermentations are characterized by the complete or almost complete absence of free liquid. Water which is essential for microbial activities is present in an absorbed or in complexed form within the solid matrix of the substrate (Cannel and Moo-Young, 1980). These cultivation conditions are especially suitable for the growth of fungi, known to grow at relatively low water activities. As the microorganisms in SSF grow under conditions closer to their natural habitats, they are more capable of producing enzymes and metabolites which will not be produced or will be produced only in low yield in submerge conditions (Jecu, 2000). The SSFs are practical for complex substrates including agricultural, forestry and food-processing residues and wastes which are used as carbon sources for the production of lignocellulolytic enzymes (Haltrich *et al.*, 1996). Compared with the two-stage hydrolysis-fermentation process during ethanol production from lignocellulosics, Sun and Cheng (2002), reported that SSF has the following advantages:

- (1) Increase in hydrolysis rate by conversion of sugars that inhibit the enzyme (Cellulase) activity
- (2) Lower enzyme requirement
- (3) Higher product yield
- (4) Lower requirement for sterile conditions since glucose is removed immediately and ethanol is produced
- (5) Shorter process time
- (6) Less reactor volume.

Malherbe and Cloete (2003), reiterated that the primary objective of lignocellulose treatment by the various industries is to access the potential of the cellulose encrusted by lignin within the lignocelluloses matrix. They expressed the opinion that a combination of SSF technology with the ability of an appropriate fungus to selectively degrade lignin will make possible industrial-scale implementation of lignocellulose-base biotechnologies. New applications of SSF have been suggested for the production of antibiotics (Barrios *et al.*, 1994), secondary metabolites (Trejo-Hernandez *et al.*, 1992, 1993) or enriched foodstuffs (Senez *et al.*, 1980). The SSF is a batch process using natural heterogenous materials (Raimbault, 1998 and Tengerdy, 1985), containing complex polymers like lignin (Agosin *et al.*, 1989), pectin (Kumar, 1987, Oriol, *et al.*, 1988a)

and lignocellulose (Roussos, 1985). SSF has been focused mainly to the production of feed, hydrolytic enzymes, organic acids, gibberellins, flavours and biopesticides.

Bacteria, yeast and fungi can grow on solid substrates, and find application in SSF processes. Bacteria are mainly involved in compositing, ensiling and some other food processes (Doelle *et al.*, 1992). Yeasts can be used for ethanol and food or feed production (Saucedo Castaneda *et al.*, 1992a, 1992b). Filamentous fungi are the most important group of microorganisms used in SSF process owing to their physiological enzymological and biochemical properties. The hyphae mode of fungal growth and their good tolerance to low water activity and high osmotic pressure conditions make fungi efficient and competitive in natural microflora for bioconversion of solid substrates (Raimbault, 1998). Microorganisms are currently the primary source of industrial enzymes; 50% originates from fungi and yeast, 35% from bacteria, while the remaining 15% is either from plant and animal origin (Bonpathy, 1994). Microbial enzymes are either produced through submerged fermentation (SMF), or Solid State Fermentation (SSF) techniques. According to the Central Food Technological Research Institute (CFTRI) in India, enzymes production by SSF accomplishes high productivity per unit volume of fermentor space than SMF technique. Processing waste such as soybean hulls and cassava peels (Ofuya and Nwajuba, 1990) has been upgraded through production of enzymes by SSF technique. The work of authors like Yang *et al.*, (1993), Onilude (1996), Balagopalan (1996), Belewu and Banjo, (1999), Iyayi and Losel (2001), Iyayi and Aderolu (2004), among others clearly showed the use of microorganism for upgrading lignocelluloses into animal feeds. Like all technologies, SSF has its disadvantages and these have received the attention by Mudgett (1986). Problems commonly associated with SSF are heat build-up, bacteria contamination, scale-up, biomass growth estimation and control of substrate content.

2.6.0 Introduction to Silage-Making

Silage-making is a management tool that allows producers to match feed resources (forages, crop residues, agro-industrial by-products etc) with feed demand for livestock production. The basic function of silage-making is to store and preserve feed for later use with minimal loss of nutritional qualities. Ensilage is the process of preserving fresh cut forages in a silo under completely anaerobic conditions (McDonald *et al.*, 1991). Some crops are purposely grown for

silage making while others are ensiled when surplus, after fulfilling the immediate feeding requirements of the livestock. The need to store feed for use by livestock is not a novelty. The Naples museum in Italy displays Egyptian paintings dated 1000 – 1500BC showing storage of fodder in what look like stone silos (Vanbelle, 1985).

In modern animal agriculture, hay-making of excess pasture preceded silage-making as the primary method of preservation on the farm; however, silage-making has progressively replaced hay making as the technique of choice in some parts of the world. Silage making is less dependent than hay-making on good weather conditions and can be extended to a great variety of forage crops (corn, sorghum, immature cereal grains, etc). Actually, the practice of silage making evolved in parallel with the success of corn as a high yielding crop that is preserved extremely easily in a silo. Difficulties arose when silage making was extended to other forage crops that are less easily preserved as silage, in particular legumes (Wattiaux, 1999).

Silage making can be considered the most effective way of preserving green forages over hay-making, if all essential steps of silage making are followed. Silage making is less dependent on weather. Moreover, earlier cutting of forages for silage had also showed higher dry matter (DM) or organic matter digestibilities. Chauhan, (1985) compared the nutritive values of hay and silage made from maize stovers. The hay was found to be higher in DM content, but lower in crude protein (CP) content, DM intake and DM of digestibility. Because of these advantages, silage making is gaining popularity in many parts of the world (Altaf-ur-Rahman and Ancela,2004).

Silage making has become an important tool for producers to manage crop production and dairy herd feeding programs in many production systems around the world. However, silage making requires considerable capital and labour investments on the farm; it also demands a fairly high level of technical expertise. The understanding of how ensiling works to preserve a crop by fermentation is important. This knowledge is key to making the best management decisions for minimizing the inevitable losses that occur when fresh feed resources are ensiled and preserved for long period of time in a silo (Michel, 1999).

2.6.1 SILAGE

Excess forages can be conserved as hay or silage. However, ensiling generally produces better quality roughage than hay because less time is required to wilt the feed. Hay making requires a longer period of rain-free days, which are often rare in the tropics during the wet season when excess forage feed generally occur (Moran, 2005).

Hay making is difficult in many tropical regions because at the time when the forage is of acceptable quality for forage conservation to be worthwhile, which is normally early in the wet season. The weather is likely to be too unreliable for sun drying. Artificial drying is expensive and facilities are not widely available (FAO, 1999, t' Mannetje, 1999 and Babayemi, 2009). Silage on the other hand can be made using fresh or preferably wilted vegetable materials. Fresh forage crops, such as maize, grasses, legumes, and crop residues can be preserved by ensiling. Ensiling, is simple and low-cost option, which can preserve feeds that are seasonally abundant for later feeding during periods of feed shortage. Ensiling can also render some previously unpalatable products useful to livestock by changing the chemical nature of the feed (Kayouli and Lee, 1999). In many countries, ensiled forages are highly valued as animal feed. In European countries such as the Netherlands, Germany and Denmark, more than 90% of the forages locally produced are stored as silage. Even in countries with generally good weather conditions for hay making such as France and Italy, about half of the forages are ensiled (Wilkinson *et al.*, 1996).

Silage, therefore, can be defined as forage, crop residues or agricultural and industrial by-products preserved by acids, either artificially added or produced by natural preservation, in the absence of oxygen (Moran, 2005 and Mannetje, 1999). McDonald (1995) described silage as the product of controlled fermentation of green fodder retaining high moisture content; the process of making silage is called ensiling and the container (if used) is called silo. Ensiling is based on lactic acid (ideally) fermentation under anaerobic (no oxygen) conditions. The lactic acid bacteria ferment the plant sugars (water soluble carbohydrates) in the crop to lactic acid, and to a lesser extent to acetic acid. The production of these acids reduces the pH (acidity) of the ensiled forage which inhibits spoilage microorganisms (due to their reduced activities). However, ensiled under incorrect conditions different and poorer quality fermentations can occur

producing other acids such as butyric acids resulting in unpalatable and lower quality silage (Moran, 2005). It is therefore essential to have a good microbial fermentation process to produce high quality silage. A good fermentation process is not only dependent on the type and quality of the forage crop, but also on the harvesting and ensiling technique. Quality silage is achieved when lactic acid is the predominant acid produced, as it is the most efficient fermentation acid and will drop the pH of the silage the fastest. The faster the fermentation is completed, the more nutrients will be retained in the silage.

2.6.2 Why make silage

The principle of silage making is more relevant to the temperate region, with its distinct seasons than to the tropical region with its relativity more or less evergreen environs. Nonetheless, silage production has become relevant to fulfill the storage needs of livestock farmers. The reasons for the major interest in silage production in the tropics are many and are as follows:

- All the major forages (grasses, forage legumes, tree legumes, by-products and crop residues) can be store as silage (Moran, 2005; t' Mannetje, 1999).
- Silage making can solve the problem of weight loss by animals that had been gained during the wet season (Montemayor *et al.*, 2000 and Babayemi, 2009).
- During the wet season, the tropical forage species grow very fast, with forage yields often exceeding animal requirement, if not cut and fed, it will continue to grow, producing very long and fibrous material, low in energy and protein (Moran, 2005).
- As the countries of the tropics become more developed the aspirations of the farmers also become more sophisticated.
- Silage making offers the option of securing feeds during seasons of high vegetative production for conservation and storage for a later use in a period of scarcity (Cowan, 2000, Babayemi 2009, and Mannetje, 1999).
- As nations become more developed, the accessibility of animals to roadside pastures/grazing becomes limited for reasons of safety to motorists (Cowan, 2000).
- Efficient fermentation ensures more palatable and digestible feed which encourages optimal dry matter intake that translates into improved animal performance (Wasaya *et al.*, 2008).

- Silage making produces 30 – 50% forage and nutrients per unit area than the same crop converted into grain and crop residue increasing the carrying capacity of a farm (Wasaya *et al.*, 2008).
- Stable composition of the feed (silage) for a longer period (up to 5 years).
- More economical use of plants with high yield of green mass.
- The fermentation in silage reduces harmful nitrates accumulated in plants during droughts and in over-fertilized crops (Moran, 2005).
- Allow crop residues and agricultural products (from sugar beat processing, maize stover, straw, sorghum stover e.t.c.) to be optimally utilised.
- Silage can provide a major diet source as basal ration as well as a feed supplement for grazing animals.
- It improves palatability, reduces significantly toxic substances present in some fresh vegetables to safe level concentrations (such as cyanogenic glucosides in fresh cassava leaves) and destroys harmful micro-organisms possibly present in poultry litter or fish wastes (Kayouli and Lee, 1999).

2.6.3 Silage Storage Systems

Silos for silage are the facilities in which the crops ferment and where they are stored until feeding. There are various types of silos which are selected according to operator preference and feeding circumstances. Silo capacity should be determined according to feeding needs (herd size), and its dimensions should be calculated according to feed-out rate, in order to minimize silage exposure to air. The most abundant types include stack (clamp) without retaining walls, tower silo, bunker silo, horizontal plastic sleeves and big bale (McDonald *et al.*, 1991).

Bunker silos are constructed of concrete of floor and walls. The silage parts adjacent to the walls and the top parts of the silage are the most susceptible to air penetration and to spoilage (Ashbell and Kashanchi, 1987, Ashbell and Weinberg, 1992). Tower silos are made of metal or concrete and can be top or bottom unloaded. The metal silos are less air permeable than concrete silos. Bale silage (0.5 – 1.0t) is prepared from forage crops which are not chopped at harvest and are wrapped by plastic sheating. Baling enables flexible use of the silage in grazing sites.

Recently Ashbell *et al.*, (2001) showed that it is possible to ensile forage in small plastic bags (10 – 20kg) which can be used by small holder cattle owners in Africa. It was hypothesized that although such bags are not air tight, the volatile fatty acids (e.g. acetic acid) that result from the fermentation accumulate in the bags and so inhibit aerobic deterioration. Lane (2000) earlier reported on the use of small plastic bags containing 6kg of silage and Pariyar (2005) more recently used plastic bags holding 6 and 12kg of silage.

2.6.4 Materials to ensile and principles of obtaining good silage

Anything that has a feed value can be ensiled, the major factors being availability and quality (t' Mannetje, 1999). Examples are: grasses, legumes, banana skin, plantain skin, crop residues, fodder crops, oil-palm fronds, peels of tuber crops, poultry litter, banana stem, plantain stem and citrus pulp, e.t.c. Irrespective of the amount of silage to be made, the following principles for good silage apply (FAO, 2000).

- The material to be ensiled must have high nutritive value
- The forage must be free of soil contamination.
- The forage should be chopped into pieces not longer than 1 – 3cm for stems and 3 – 8cm for leaves to facilitate compaction and reduce air retention.
- It is necessary to expel the maximum amount of air within the forage before closing the storage system preventing air and water penetration.
- The accumulation of the forage and sealing should be done in the shortest possible time.
- During the feeding of the silage, the area exposed to air should be as small as possible and the time of opening and finishing the storage as short as possible. Silage storage system or silo types could be: pit, tower, bunker, trench, plastic bag, plastic drum, plastic container, and steel drum or cement box silos (FAO, 2000).

2.6.5 Quantity of silage for farm

The quantity of silage to store depends on several factors such as

How many animals are to be fed?

For how long to be fed?

How much to be fed?

The storage space available

The amount of excess feed to conserve

Forage dry matter content

Available labour and total costs

Type of livestock receiving silage (Moran, 2005 and Wooderd *et al*, 2002).

2.6.6 Fermentation Dynamics

Aerobic and anaerobic bacteria are involved in silage fermentation (Harrison *et al.*, 1991, McDonald, 1991 and Woolford, 1984). Aerobic activity occurs while the silos is being filled and at feedout. Good silo management minimizes aerobic activity, thus reducing dry matter losses (Seglar, 2003). Oxidation of energy rich sugars produces excess heat, which can damage forage protein. Good silo maximizes the anaerobic conversion of water-soluble carbohydrate to silage acids thus reducing pH to a range that is inhospitable to spoilage organisms. This conversion results from anaerobic hetero-and homofermentation.

Heterofermentative anaerobic converts water-soluble carbohydrate into various fermentation end products at the expense of energy (because dry matter is decreased). Homofermentative bacteria (Lactic-acid bacteria) convert water-soluble carbohydrate to lactic acid; little energy is consumed, and dry-matter loss is diminished. Heterofermentation and homofermentative anaerobes are both essential to silage fermentation; however efficient fermentation minimizes heterofermentation and maximizes homofermentation. Silage fermentation can be divided into four phases (McCullough, 1984).

2.6.6.1 Phase 1 Aerobic phase

This phase begins from the time of harvesting the crop to the time oxygen is depleted from the sealed silo. Due to continued plant respiration, plant enzymes and epiphytic (indigenous; i.e. those microorganisms naturally present on the plant) aerobic microorganisms cause nutrient losses by degrading plant proteins and converting water-soluble carbohydrate (sugar) to carbon dioxide and water, and generating heat. The heat released by respiration raises the temperature of the forage. Temperatures greater than 26 – 32⁰C may cause significant loss of nutrients. Excessive heat production (i.e. temperatures above 42 – 44⁰) can result in Maillard reactions which reduce the digestibility of both protein and fibre constituents (Bolsen *et al.*, 1996). Researcher indicates that the rise in temperature is not as high in well packed silos as it is in

poorly packed silos (Pitt, 1990). The rapid expulsion of oxygen is desirable because it decreases both the length of the respiration phase and the associated nutrient losses. Also, the plant's own enzymes promote the hydrolysis of starch and hemicelluloses to monosaccharides. This hydrolysis provides additional sugars for later lactic acid fermentation. The neutral detergent fibre (NDF) content increases slightly after ensiling, mainly because the water soluble carbohydrate (WSC) content is reduced. Normally, respiration continues for one to two days, but takes place only as in the silage. Thus, compacting silage to remove as much air as possible as rapidly as possible will curtail respiration losses. The aerobic phase results in nutrient loss. However, it helps create anaerobic conditions and produces certain antimycotic compounds that may serve to extend aerobic stability of the silage during feed out (Seglar, 2003).

Practical aspects of the aerobic phase:

- Fill the storage site quickly (1 – 2 days).
- Chop the material as short as possible (1 – 3cm)
- Compact the storage container as well as possible, as fingers should not be able to be inserted into the compacted forage.
- Seal the storage container and make it air tight
- Weight the top of the stack to maintain an airtight seal between the cover and compacted forage
- Seal as soon as possible after harvesting is completed (Moran, 2005) .

2.6.6.2 Fermentation phase

This stage begins once the oxygen is gone and the storage becomes anaerobic. Once anaerobic conditions are reached, anaerobes (anaerobic microorganisms) begin to grow. The lactic acid bacteria are the most important microflora, because forages are preserved by lactic acid. The other microorganisms of importance are clostridia, yeast and moulds which have negative impacts on silage. They compete with the lactic acid bacteria for fermentable carbohydrates (water soluble carbohydrate) and many of their end products have no preservative action (Seglar, 2003). Depending on the properties (i.e buffering capacities, moisture content and maturity) of the ensiled crop and the ensiling condition, this phase may last from 7 – 21 days. However, the use of high quality inoculants may reduce fermentation time down to 3 – 10 days from time of filling (Bolsen *et al.*, 1996).

Buffering capacity is the crop's resistance to drops in pH. It is due to the organic acids (e.g. citrate, malate, and quinate), orthophosphates, sulphates, nitrates, chlorides and non-protein nitrogen in the material (McDonald, 1991). Buffering capacity is measured as the amount of lactic acid required to reduce the pH of 1g of dry matter from pH 6 to 4. A successful fermentation will see the number of lactic acid producing bacteria dominate, reducing the pH to 3.5 – 4.5 (Sheperd and Kung, 1996). The fermentation of plant sugars into organic acids by anaerobic bacteria lower silage pH from above 6 to about 5. If silage pH drops slowly and the moisture concentration is high due to harvesting too early, clostridia bacteria may grow. These bacteria degrade sugars and convert lactic acid to butyric acid, releasing strong offensive odours. They also break down protein to nonprotein nitrogen and undesirable end products like amines. These changes lead to increased dry matter loss and reduced palatability and quality of the silage (Adesogan and Newman, 2010). However the rate and end-point of the final pH drop in the ensiled crop depend largely on the type and moisture of forage being ensiled. Corn silage terminates at or below pH 4. Legumes, which have less water soluble carbohydrates content and a higher buffering capacity, generally reach a terminal pH of about 4.5. When terminal pH is reached, the forage is in a preserved state. Measurement of pH show whether the crop is preserved, within the silo but not an indication of rate or quality of the resulting fermentation. At this point (i.e. low pH), fermentation of sugar by lactic acid bacteria has ceased, either because the low pH inhibited their growth or there was a lack of sugar for fermentation (Seglar, 2003).

The lactic acid bacteria ferment sugars (water soluble carbohydrates) to primarily lactic acid, but also produce some acetic acid, ethanol, carbon dioxide and other minor products (Bolsen *et al.*, 1996). The lactic acid bacteria are a large group of bacteria comprising organisms in six genera namely: *Lactobacillus* spp., *Pedicoccus* spp., *Enterococcus* spp., *Lactococcus* spp., *Leuconostoc* spp. and *Streptococcus* spp (McDonald *et al.*, 1991 cited by Bolsen *et al.*, 1996).

They are divided into two categories, the homofermentative and heterofermentative. The homofermentative (the first four genera) produce only lactic acid from fermenting glucose and other hexoses, while the heterofermentative (the last two genera) produce lactic acid, acetic acid, ethanol and carbon dioxide. In the fermentation phase, competitions between strains of lactic

acid bacteria determine how the ensiling process will progress. Practical aspects of the fermentation phase

- Mix additives such as molasses (at 3 – 5% on wet basis), a substrate source for the bacteria to encourage lactic acid fermentation.
- If possible, wilt forage to preferably about 30% Dry Matter (Moran, 2005).

2.6.6.3 Stable phase

The third stage of fermentation, the stable phase, lasts throughout storage. Following the active growth of lactic acid bacteria, the ensiled material enters the stable phase. This phase is not static because various changes can occur, depending on environmental conditions and the number and types of aerobic organisms (yeasts, molds and aerobes) on the crop at harvest. If the silo is properly sealed and the pH has been reduced to a low level, little biological activity occurs in this phase. Once the pH level has dropped, and air and water is not permitted to enter the storage, most microorganisms of phase 2 slowly decrease in numbers, resulting in silage which is relatively stable. The amount of fermentation substrate remaining and the level and types of fermentation acids present in the silage are important in stable phase. However, very slow rate of chemical breakdown of hemicelluloses can occur, releasing some sugars. If active fermentation ceases because of a lack of water soluble carbohydrates, lactic acid bacteria might ferment the sugars released by hemicelluloses breakdown, causing a further slow rate of pH decline (Seglar, 2003).

Another major factor affecting silage quality during the stable phase is the permeability of the silo to air (oxygen). Oxygen entering the silo is used by aerobic microorganisms (via microbial respiration) causing increases in yeast and mould populations, losses of silage dry matter, and heating of the ensiled mass. The amount of aerobic loss in this phase is related not only to the permeability of the silo but also to the mass density of the silage. If the silage is left unsealed, substantial dry matter losses can occur at the exposed surface (Bolsen *et al.*, 1996). These losses can be reduced by covering the surface of the ensiled material with polythene sheeting. Cracks in the silo wall or holes in the polythene seal increase the rate at which oxygen can penetrate the silage mass (Moran, 2005).

How to conduct the stable phase:

- Maintain an air tight seal around the silage
- Repair holes as soon as they are noticed.

2.6.6.4 Feed out phase

Phase 4, which is the last fermentation phase, occurs when silage is fed from the storage structure. This phase is as important as the others but is often neglected. Up to 50% of dry matter losses result from secondary aerobic spoilage on the surface of the silage in storage and in the feedbunk (Cai *et al.*, 1997; N.F.I.A., 1998).

When the silo is opened, oxygen usually has unrestricted access to the silage at the phase. Aerobic microbial activity is stimulated because oxygen is re-introduced into the silo. The aerobic activity produces heat and reduces the palatability and nutrient availability of silage. Silage is predisposed to aerobic instability if high epiphytic populations of yeast, mould, or aerobic bacteria are present or if the silage contains excess water soluble carbohydrate (Seglar, 2003).

During this phase, the largest losses of dry matter and nutrients can occur because of aerobic microorganisms consuming sugars; fermentation products (i.e lactic acid and acetic acids); and other soluble nutrients in the silage. These soluble nutrients are respired to carbon dioxide and water producing heat. Yeast and moulds are the most common micro organisms involved in the aerobic deterioration of the silage, but bacteria, such as *Enterobacteriaceae* and *Bacillus* spp., also have been shown to be important (Muck and Pitt, 1993).

Besides the loss of highly digestible nutrients in the silage, some species of moulds can produce mycotoxins and other toxic compounds that can affect livestock and human health. The microbial activity in the feed out phase is the same as that occurring because of oxygen infiltration during the stable phase. The major difference is the amount of oxygen available to the microorganisms. At the feed out, the microorganisms on the surface of silage have unlimited quantities of oxygen, allowing them to grow rapidly. Once yeasts or bacteria reach a certain population or colony, the silage will begin to heat and digestible components like sugars and

fermentation products will be lost quickly. The time required for heating to occur is dependent on several factors:

- Number of aerobic microbes in the silage
- Time exposed to oxygen prior to feeding
- Silage fermentation traits
- Ambient temperature (Cai *et al.*, 1999).

It should be noted that this feed out phase or aerobic spoilage phase also begins when holes are made in storage sites by mice, birds or other agents or it becomes uncovered for feeding out. The rate of spoilage is highly dependent on the numbers and activity of the spoilage organisms in the silage and may be in the range of 1.5 to 4.5% DM loss/day (Filya, 2004; Moran, 2005). Aerobic stability of silage is more of a problem if the crops had been exposed to environmental stresses.

Some practical aspects of the aerobic spoilage phase:

- Maintain an airtight seal,
- feeding out to ensure about 20 to 30cm removal from the entire silage face each day.
- If the silage gets hot, feed it out at a faster rate.
- If silage heating occurs, consider a smaller stack face next harvest (Moran, 2005).

2.6.5 Silage quality evaluation

Seglar (1999) reported that fermentation quality can be assessed at the silo by making visual observations and with the help of several simple analytical tools. Jianxin (2002) also reported that silage quality can be accurately evaluated by subjective and chemical methods. The subjective method involves the use of criteria such as colour, smell, texture (structure) and pH values.

Moisture: Proper moisture at harvest is critical for compaction of the silage mass, air exclusion, and to provide sufficient moisture to promote lactic acid fermentation. Stefanie *et al.* (1999) and t'Mannetje (1999) reported that lactic acid concentration usually falls when the forage moisture content and the amount of oxygen trapped in the ensiled forage during filling of the silo are high. This results in the growth of facultative aerobic microbes such as coliforms. Jianxin (2002)

reported that ensiling materials that are too low in moisture content (< 50%) can result in prolonged or restricted fermentation, aerobically unstable silage, high pH, with evidence of yeasts, mould and bacillus growth and spontaneous heating. Low moisture silages often have high levels of heat damaged proteins and are monitored by acid detergent insoluble nitrogen (ADIN) as % of total crude protein (Ward, 1996).

Colour: Good silage quality usually preserves well the original colour of the standing plant (t'Mannetje, 1999; Jianxin, 2002). Properly fermented silage is light green to yellow in colour. When green raw materials produce silage with light green or yellow colour, it can be considered as good quality silage. Temperature is one of the important factors affecting silage colour. The lower the temperature, during ensilage, the less will be the colour change. Above 30⁰C, grass silage becomes dark yellow. Above 45 – 60⁰C, the colour become closer to brown (Maillard or browning reactions). Maillard reactions reduces digestibility (Muck, 1996). Beyond 60⁰C, the colour darkens towards black due to caramelization of sugars in the forage (Mc Donald *et al*, 1995; t' Mannetje, 1999, Jianxin, 2002; Oduguwa *et al*, 2007).

In addition, the colour of the silage juice may also be a useful indicator of silage quality. However, silage quality can be misjudged by the colour. Jianxin (2002) reported that silage from red clover or Chinese-milk vetch is often dark brown instead of light brown known and may be considered failed silage despite its excellent quality due to colour.

Smell: Properly fermented silage has a pleasant Vinegar aroma. (Meneses *et al*. 2007). The smell of the silage is a good indicator of the quality. According to Jianxin (2002), good silage has a mild, slightly acidic and fruity smell resembling that of cut bread and of tobacco due to the presence of lactic acid. A rancid and nauseous smell denotes the presence of butyric acid and signifies failed silage. A musty smell is a sign of deficient compaction and presence of oxygen. A distinctive unpleasant smell of sow's urine and faecal matter signifies marked protein degradation during ensilage.

Seglar (2003) reported that some yeast will produce objectionable odours and tastes from the production of various end-products. He however, stated that alcohol mixed with the vinegar

odour of acetic acid produces a stinging smell and sometimes causes cows to refuse silages. Other yeast end-products are methyl-and ethyl-acetates, which resemble the smell of fingernail polish remover. Seglar (2003) however, concluded that it is the combination of high acetic acid levels, along with the presence of alcohol, and methyl-acetates that probably causes feed refusal by cow.

Structure: Plant structures (stems and leaves) should be completely firm and recognisable in the silage (i.e. firm tissues). A destroyed structure is a sign of severe putrefaction. Mc Donald, *et al.*, (1995) and Jianxin (2002) reported that a viscous, slimy appearance reveals the activity of proteolytic or sporulating microorganisms.

pH: The pH of an ensiled sample is a measure of its acidity, but is also affected by the buffering capacity of the crop Kung 2010. pH is a key criterion to evaluate silage fermentation. The pH may be determined on the farm using wide range-pH paper or meter. Moreover; it is a simple method to predict silage quality. Wilkinson *et al.* (1976), Etman *et al.* (1994), McDonald *et al.* (1995), Sheperd and Kung (1996) and EL-Shinnawy (2003) found that;(i) a good quality silage have a pH value of 4.2 or less and (ii) the pH value increased dry matter (DM) content.

Generally, the lower the pH, the better preserved and more stable is the silage (Seglar, 2003).

The pH is influenced by the moisture content and the buffering capacity of the original materials. However, pH alone is not a totally accurate monitor of silage fermentation, determination of silage acid levels that contribute to lowering pH is also important (Seglar, 2003). A sharp acid taste is indicative of pH < 4.5 (Harris, 2004). Two samples may have the same pH, but different concentrations of acids. In general, legume silages have a higher pH than corn or other grass silages and take longer to ensile because of their higher buffering capacity (Kung, 2010).

Temperature: The optimum temperature of fermenting forage varies from 32 – 41°C. Temperature outside this range results in poorer quality silage even though palatability may remain good. Overheated silage gives a drab green colour, strong aroma, slimy soft tissue, insipid taste, and a pH of about 5.0. Overheated silage frequently referred to as “heat damaged”,

range in colour from brown to dark brown and has a charred hay or tobacco aroma. Heat damage resulting from high temperatures in fermenting forages has been reported (Adesogan and Newman, 2010). The digestibility of protein has been found to be reduced in the presence of oxygen and high temperatures (Muck, 1996). The longer the heat, the more protein damage. Also, the rate of damage increase with temperature. Heating appears to not only decrease the availability of protein to the animal but also reduce the availability of carbohydrates. While excessively heated silages remain very palatable and are readily consumed by animals, a considerable proportion of their food value is lost (Nagel and Broderick, 1992). However, temperatures of fermenting forages varying from 27⁰C to 38⁰C should produce excellent silages.

Buffering Capacity: Buffering capacity measures to what degree a forage sample will resist a change in pH. All forages have different buffering capacities. Fresh forage with a high buffering capacity will require more acid to reduce its pH than forage with a low buffering capacity. In general, fresh legumes have a higher buffering capacity than do fresh grasses or corn (Seglar, 2003).

2.6.6 Silage acids

Lactic acids: Lactic acid is the primary fermentation acid resulting from desirable homofermentation. Ideal silage will usually (but not always) have three times more lactic acid than what comprises volatile fatty acids. Lactic acid is the strongest of all silage acids and its presence will drop pH more effectively than the other volatile fatty acids (VFAs) (Seglar, 2003). Generally, the presence of high lactic acid levels indicates efficient fermentation. Fermentations that produce lactic acid result in the minimal losses of dry matter and energy from the crop during storage. Lactic acid should be at least 65 to 70% of the total silage acids in good silage (Kung, 2010).

Volatile Fatty Acids: These acids give silages their characteristic smell, because they evaporate quite easily when introduced to air. Lactic acid has a bland odour and does not volatilize upon exposure to air. The VFAs provides aerobic stability properties.

(i) Acetic acid: It is the principal acid produced during fermentation for maintaining aerobic stability. Usually found at less than 3% in silages. Anything over 3% suggests inefficient heterofermentative fermentation (Seglar, 2003). Extremely wet silages (< 25 – 30%), prolonged fermentations (due to high buffering capacity), loose packing, or slow silo filling can result in silages with high concentrations of acetic acid (> 3 to 4% of DM). In such silages, energy and DM recovery are probably less than ideal. Silages treated with ammonia also tend to have higher concentrations of acetic acid than untreated silage, because the fermentation is prolonged by the addition of the ammonia that raises pH (Kung, 2010).

(ii) Propionic acid: Produces a sharp sweet smell and taste and usually, lower level of this acid are produced during fermentation for maintaining aerobic stability. Most silage contains very low concentrations of propionic acid (< 0.1 to 0.2%) unless the silage is very wet (< 25% DM). In silages with more typical concentrations of DM (35 to 45% DM), concentrations of propionic acid may be undetectable. Concentrations of propionic acid that is higher than 0.3 to 0.5% are usually associated with poor fermentations.

(iii) Butyric acid: Produces a rancid butter smell and taste. Quality silage should be less than 0.1% in butyric acid. High concentration of butyric acid (> 0.5% of DM) indicates that the silage has undergone clostridial fermentation, which is one of the poorest silage fermentations. Silages high in butyric acid are usually low in nutritive value and have high ADF and NDF levels because many of the soluble nutrients have been degraded. Such silages may also be high in concentrations of soluble proteins and may contain small protein compounds called amines that have sometimes shown to adversely affect animal performance due to significant reduction in dry matter intake and energy level of the forage. High concentrations of butyric acid have sometimes induced ketosis in lactating cows and because the energy value of silage is low, intake and production can suffer (Seglar, 2003).

2.6.7 Silage microbiology

Aerobic fungi and bacteria are the dominant microorganisms on fresh herbage, but as anaerobic conditions develop in the silo they are replaced by bacteria able to grow in the absence of oxygen, such as species of *Escherichia*, *Klebsiella*, *Bacillus*, *Clostridium*, *Streptococcus*,

Leuconostoc, *Lactobacillus* and *Pediococcus*. Yeast are also present and being facultative anaerobes (i.e. able to grow both aerobically and anaerobically), can survive and proliferate in silage (McDonald *et al.*, 1987).

The lactic acid bacteria, which are also anaerobes, are normally present on growing crops in small numbers but usually multiply rapidly after harvesting, particularly if the crop is chopped or lacerated. They can be divided into two categories, the homofermentative and the heterofermentative lactic acid bacteria. When the crop is ensiled, the lactic acid bacteria continue to increase, fermenting the water soluble carbohydrates in the crop to organic acids, mainly lactic, which reduce the pH value (McDonald *et al.*, 1987). At a certain critical pH level, which varies with moisture content, the acids inhibit the growth of other bacteria and at about pH 3.8 to 4.0 microbial activities virtually ceases and the material remain stable for as long as anaerobic conditions are maintained.

If a stable pH has not been achieved then the saccharolytic clostridia, which are present on the original crop as spores, will multiply; they ferment lactic acid and residual water soluble carbohydrates to butyric acid causing the pH to rise (McDonald *et al.*, 1987). The less acid tolerant proteolytic clostridia then usually become active, leading to a further increase in pH caused by the production of ammonia. Clostridia are particularly sensitive to water availability and they require very wet conditions for active growth. With very wet crops, even the achievement of a pH value as low as 4 may not inhibit their activity (McDonald *et al.*, 1987).

Another group of organisms, which are normally found on harvested crops and which can multiply in the silo, is the coliform bacteria. They are members of the family *Enterobacteriaceae* and include *Escherichia* and *Klebsiella* spp. These organisms have been referred to as the 'acetic acid bacteria' since acetic acid is a major product when they ferment sugars. The optimum pH for the growth of the coliform bacteria is about 7.0 and they are usually only active in the early stages of fermentation when the pH is favourable for their growth (McDonald *et al.*, 1987).

2.6.8 Classification of silages

The composition of the ensiled forage and the subsequent fermentation will determine the type of silage produce. Silage produced can be broadly classified into six main types (McDonald *et al.*, 1987).

2.6.8.1 Lactate silages

In this, the fermentation is dominated by lactic acid bacteria (LAB). Lactate silages are characterized by having a low pH (3.7 – 4.2) and a high concentration of lactic acid. Lactate silages have a pleasant, acidic and sometimes sweet smells. It is the commonest type of silage produced from unwilted grasses and whole cereal crops. The lactic acid contents of maize silages are usually much lower than those of well-preserved grass silages because of the higher dry matter and lower buffering properties of the maize crop (McDonald *et al.*, 1987).

Lactate silages usually contain small amounts of acetic acid and may also contain traces of propionic and butyric acid. Variable amounts of ethanol and mannitol derived from the activities of heterofermentative lactic acid bacteria and yeasts are present. The buffering capacities of these silages are high and the water soluble carbohydrates low. The nitrogenous components are mainly in a non-protein, soluble form and, because very little deamination of amino acids will have occurred; the free ammonia N values will be low. However, the high soluble non-protein N content of these silages, coupled with the low levels of soluble carbohydrate, can result in high ammonia concentrations in the rumen, which lead to poor utilisation of the silage N (McDonald *et al.*, 1987).

Because of the extensive changes to the soluble carbohydrates, resulting in the formation of high energy compounds such as ethanol, the gross energy concentrations of these silages higher than those of the parent material. The digestibility of lactate silages is similar to that of the original crop. Methane production in the rumen is the same as that arising from animals on fresh grass diets, although the urinary energy losses might be slightly higher from animals consuming lactate silage than from those consuming grass, reflecting the poor utilisation of silage nitrogen referred to earlier. One further disadvantage of lactate silages is that, when given ad libitum to ruminants,

they promote lower dry matter intakes than comparable fresh or dried herbage (McDonald *et al.*, 1987).

2.6.8.2 Acetate silages

Under certain ill-defined conditions acetic acid bacteria (enterobacteria) may dominate the fermentation. Acetate silages are common in the tropics than temperate countries. The pH values of these silages are higher than those of lactate silage at the same dry matter content and characterised by a sour, and vinegar smell. More likely to occur when unwilted or lightly wilted, low dry matter forage is ensiled. The water soluble carbohydrates are primarily converted to acetic acid. Acetate silages contain high levels of acetic acid and relatively low levels of lactic acid. Deamination of amino acids is usually extensive, and consequently ammonia levels in these silages are higher than those found in lactate silages. Because of the negative correlation of acetic acid content with dry matter intake, it is reasonable to assume that the latter will be low in animals given these silages *ad libitum*. Dry matter and energy losses can be significant in these silages (McDonald *et al.*, 1987).

2.6.8.3 Butyrate silages

These silages have undergone clostridia fermentation. They usually have pH values within the range of 5 to 6, and contain low concentrations of lactic acid and water soluble carbohydrates. Butyric acid is usually the dominant fermentation product, although acetic acids contents are also frequently high. Because of the extensive breakdown of amino acids caused by clostridia, silages of this type will contain high concentrations of ammonia-N. Decarboxylation of amino acids to amines occurs, thus silages are unpalatable to livestock. As a result of these changes, the subsequent utilization by ruminants of the nitrogenous compounds in butyrate is likely to be low or poor. The dry matter intake of ruminants given these silages is low (McDonald *et al.*, 1987).

2.6.8.4 Wilted silages

Wilting a crop prior to ensiling restricts fermentation increasingly as dry matter content increases. In such wilted silages, clostridia activity is minimal although some growth of lactic acid bacteria occurs. The fermentation is restricted because of the high dry matter content (> 30%). Less water soluble carbohydrates are converted to lactic acid and pH values are higher

than those of lactate silages. Total fermentation acids and buffering capacity values are lower than in unwilted silages and there are usually some residual water soluble carbohydrates. Wilting does not prevent proteolysis occurring, although deamination of amino acids is considerably reduced. Both gross energy and metabolisable energy concentrations of wilted silages are normally similar to those of the parent material (McDonald *et al.*, 1987).

However, very dry forages are harder to compact, especially if chop length is long; there is a greater risk of yeast and mould growth because oxygen levels in the pit or bale are high in poorly compacted silages. Higher residual water soluble carbohydrates, poor compaction and carry-over yeast and mould spores can make these silages more aerobically unstable. Although wilting usually results in an increase in silage dry matter intake, any beneficial effects in terms of improved animal performance, compared with well-preserved, unwilted, silage diets are difficult to demonstrate. The main advantages of wilting are that the risk of obtaining butyrate silage is decreased and the production of effluent is reduced (McDonald *et al.*, 1987).

2.6.8.5 Deteriorated silages

The continuous infiltration of air during the storage period in the silo results in the growth of aerobic micro organisms which break down the organic matter to form composted material unfit for animals. Such wasted material is commonly found on the surface and sides of silage made in bunker and stack silos. This deterioration process will also occur during the feeding period when silage is exposed to air for varying periods of time. Initially the soluble components in the silage, such as organic acids, alcohols and sugars will be oxidised, but continuous exposure to air eventually leads to the destruction of the more stable components such as cell wall polysaccharides. The organisms responsible are initially yeasts and bacteria which are followed by moulds. The factors governing the rate of deterioration are unknown but the extent of dry matter breakdown in silage exposed to air over a 10-day period may range from virtually nil to over 30%. Silages in which fermentation has been restricted by wilting or by the use of chemical additives, are more prone to aerobic deterioration than those in which either lactic acid bacteria or clostridia have been active. The presence of propionic, butyric and caproic acids in silages improves their stability in air (McDonald *et al.*, 1987).

2.6.8.6 Overheated silages

These silages are produced from overwilted material ensiled in bunker-type or stack silos without adequate consolidation. If the temperature in the mass exceeds 55°C, protein digestibility may be reduced. Overheated silages, which are dark brown or even black in colour may be palatable to animals, but are of low nutritional value because of excessive oxidation of soluble nutrients (McDonald *et al.*, 1987).

2.6.9 Silage additives

Fermentation in the silo can be a very uncontrolled process leading to less than optimal preservation of nutrients. Silage additives have been used to improve the ensiling process (better energy and dry matter recovery) with subsequent improvements in animal performance. Additives are used to improve nutrients composition of silage, to reduce storage losses by promoting rapid fermentation, to reduce fermentation losses by limiting extent of fermentation, and to improve bunk life of silage (increase aerobic stability) (McDonald *et al.*, 1987).

Silage additives can be used in order to enhance silage fermentation and their nutritional quality. Many different silage additives are available and are used for different reasons. They are classified according to their function: fermentation stimulants, fermentation inhibitors, aerobic deterioration inhibitors, nutrients and absorbents (McDonald *et al.*, 1991). Additives should be used according to needs and silage properties. However, it should be emphasised that additives can improve silage quality and minimize losses, but cannot compensate for poor silage making and management. There is a long list of available additives (Bolsen and Heidker, 1985) which come in a variety forms: liquid, powders or suspensions. Additives can be applied during the harvesting chopping operation or during filling of the silo (Ashbell and Weinberg, 2006).

Bacterial inoculants are used in order to enhance the ensiling fermentation. They are safe, easy to use, and non-corrosive to machinery and regarded as natural products. Most commercial inoculants for silage include homofermentative lactic acid bacteria (e.g. *Lactobacillus plantarum*, *Enterococcus faecium* and *Pediococcus* spp. They are used because they are fast producers of lactic acid (Ashbell and Weinberg, 2006). However, in whole crop cereal silages such inoculants resulted in spoilage upon aerobic exposure because their fermentation did not

produce enough volatile fatty acids such as acetic acid, that inhibit fungi. The new types of inoculants are tailored according to silage properties, and for whole crop cereal silage heterofermentative lactic acid bacteria (LAB) is included, such as *Lactobacillus buchneri* (Weinberg and Muck, 1996)

Chemical additives include various organic and mineral acids that lower the pH artificially and inhibit specific microbial populations. For very moist crops formaldehyde can be used at 2-4g^l⁻¹. Other chemicals are available as well e.g. sulphur based chemical that inhibit fungi (Ashbell *et al.*, 2001). Absorbents are used in crops with a low dry matter content to prevent excessive effluent losses. Certain crops are deficient in dietary components essential for ruminants. The nutritional quality of these crops can be improved by supplementation with specific additives at the time of ensiling. Additives that have been used in this respect are ammonia and urea to increase the crude and true protein content of the silage, and limestone and MgSO₄ to increase the calcium and magnesium contents. The above mentioned additives generally have no beneficial effect on silage fermentation, but urea and ammonia can improve the aerobic stability of the silage (McDonald) *et al.*, 1991).

Most commercial additives contain more than one active ingredient in order to enhance efficacy and have a broad range of applicability such additives referred to as combined additives. Very popular are for example, combinations of inoculants stimulating homofermentative lactic acid fermentation together with sugar releasing enzymes, or combinations of fermentation and aerobic deterioration inhibiting chemicals such as formic acid, sulphite salts and propionic acid (Rider, 1997, Anon, 1999). Promising results have been obtained by combining homofermentative or facultative heterofermentative lactic acid bacteria with chemicals such as ammonium formate and sodium benzoate (Kalzendorf, 1992; Bader, 1997), or by combining facultative heterofermentative lactic acid bacteria with the obligate heterofermentative *L. buchneri*.

Enzyme additives usually degrade plant cell walls and sometimes starch. In theory, the degradation of plant cell walls should reduce the concentrations of neutral and acid detergent fibre in the silage and at the same time, release additional sugar, which is the primary substrate for lactic acid-producing bacteria. Substrate sources are primarily sugars, such as molasses,

glucose, sucrose, dextrose, cracked maize and their likes. They provide additional substrate for lactic acid producing bacteria. Silage inhibitors are products like formic, propionic, hydrochloric and sulphuric acids which are primarily organic that effectively sterilize the silage. They are used in extremely wet silages.

However, it should be emphasised that the efficacy of any additive will ultimately be assessed by animal performance and by DM recovery from the silo, which are parameters not commonly determined. Most of the experiments are restricted to measurements of traditional fermentation patterns under controlled laboratory conditions, where even untreated silages made from thick-stemmed *Pennisetum* species may show acceptable preservation (Woodard *et al.*, 1991; Spitaleri *et al.*, 1995). In contrast, bad fermentation products, such as biogenic amines that cause intake depression in ruminants (Phuntsok *et al.*, 1998) are not detected by conventional silage analysis. It has been suggested that the current parameters used to predict silage fermentation and quality may need some re-evaluation (Jones, 1995).

In some countries there are voluntary schemes by which new silage additives are tested by independent institutes. The approval categories of the additives are according to their declared functions (e.g., fermentation improvement, reducing dry matter losses, improving aerobic stability and enhancing animal intake, weight gain and milk production) (Ashbell and Weinberg, 2006).

2.6.10 Silage temperatures

The production of heat is a normal occurrence during silage fermentation. If silage is well packed and sealed immediately, the average temperature of the mass should not rise to more than – 12.22 to – 6.67⁰C above the ambient temperature at filling (Kung, 2008). The optimal internal temperature during fermentation is below 37.78⁰C. Higher temperatures often result in poorer – quality silage (Adesogan and Newman, 2010). However, it is common to measure temperatures as high as 43.33 – 54.44⁰C in the upper most layers of silages during silo filling. These high temperatures are a result of excessive amounts of air trapped in the top layers of forage. The key is that these temperatures should decrease quickly as further packing removes air from the mass. Prolonged high temperature above 46.11 – 48.89⁰C can lead to heat damaged protein.

Temperatures in this range can also be detrimental to many lactic acid bacteria that are needed to achieve a successful fermentation. Temperatures above 37.78⁰C could reduce the fermentation quality, enhance protein degradation and reduce the rapid pH decline necessary for an efficient fermentation. Excessively heated or heat-damaged silages have a brown to dark brown colour with a tobacco-type smell. Part of the protein in “heat-damaged” silages is complexed with carbohydrates and is less digestible. The concentration of heat-damaged protein depends on both the temperature and the length of time the temperature is elevated. “Heat damaged” silage may be palatable, but part of the protein and some of the energy it contains will be unavailable to livestock (Adesogan and Newman, 2010). Thus, chop forage adequately, pack quickly and tightly and seal as soon as possible to keep the air out of the forage mass.

When the active phase of fermentation is complete, temperatures in the core of the silo often fall to 21.11 – 29.44⁰C. However, a second wave of heat can be produced in silos because of aerobic deterioration. Penetration of air into the silage mass allows spoilage yeast to metabolise lactic acid. As a result of this, the mass reheats and silage pH increases. Moulds and opportunistic bacteria that thrive on oxygen cause more heating and spoilage. In some cases, we have measured temperatures in silage phases in excess of 62.78⁰C (Kung, 2008). Signs that silage is aerobically spoiling include measuring temperatures in excess of 37.78⁰C ten to twenty centimeters (10 – 20cm) in back of the silo face at feed out, reheating in the bunk, visible mould, lack of a sharp or sweet smell to the silage and/or a flat or mouldy/musty smell. If a pH meter is available, a mouldy smell coupled with a high pH may also be a good indicator that a feed has undergone aerobic deterioration. Aerobic deterioration of silages is of course more common during warmer weather (Kung, 2008). During cool weather, steam is often released during feed out from the face of large silos because of the difference between retained heat and the ambient temperature. The presence of steam does not always mean that silage is spoiling. In fact, large silos can retain significant amounts of heat for prolonged periods of time. Retained heat should seldom register above 35⁰C especially after 2 – 3 months of storage (Kung, 2008).

Table 1: Average normal temperatures of Silage

Stage	Normal Temperature Range
Early ensiling, core temperatures	29.44 - 40.56 ⁰ C
Early ensiling; shallow surfaces, Loosely packed	29.44 – 57.22 ⁰ C
During storage – large silos, deep Core temperature	21.11 – 35 ⁰ C
Active, aerobically spoiling silages	49.33 – 48.89 ⁰ C

Source: Kung (2008).

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2.6.11 Moisture content control during the ensiling process

One of the major factors affecting the fermentation process is the moisture content of the forage. Generally, the optimum moisture content for precision-chopped silage is about 65 – 70% (t'Mannetje, 1999; Coblenz, 2000; and Cowan, 2000). This degree of hydration will facilitate the fermentations process and help to eliminate oxygen from the silage mass during packing.

Ensiling forages at moisture contents greater than 70% is not often recommended. High forage moisture levels at ensiling may cause silage effluent and favour undesirable (clostridial) fermentations. Such silages are less acidic, have a high concentration of butyric acid and ammonia nitrogen. Silage of this type has a strong offensive and rancid odour, and is poorly consumed by ruminants. This is an important consideration when the silage crop also has high nitrogen/crude protein content, high buffering capacity (i.e. resistance of forage to pH change) and/or low sugar content. Unwilted alfalfa of the temperate climates is an example of a forage crop that best meets these conditions and is most likely to undergo clostridial fermentation. In case of the tropics, most species are difficult to ensile because of their low dry matter (i.e their resistance to change in pH). To enable them to undergo a more satisfactory fermentation, wilting is required. Wilting involves laying the cut forage on rocks, on polythene sheets or against walls to allow air or the sun's heat to evaporate some moisture from the plants (Moran, 2005).

On the other hand, ensiling forages when the moisture content is low (less than 50%) can result in restricted fermentation (t'Mannetje, 1999). This will produce less stable silages that have lower lactic acid concentrations and are less acidic (higher pH). It is also more difficult to exclude oxygen from the silage mass during the filling and packing processes. It is to be noted that maintaining the integrity of the silo walls is absolutely critical to the long-term preservation of the silage. Moulds and spontaneous heating are more common in these silages (Cowan, 2000).

2.6.12 Silage fermentation in tropical silages

Ensiling of forage crops, residues and agro-industrial by-products could make an important contribution to the optimisation of tropical and sub-tropical animal production systems but thus far, it has not yet been widely applied (Wilkins *et al.*, 1999). This is due not only to the low prices for animal products, the low levels of mechanization and the high costs of silo sealing

materials, but also to a lack of ensiling experience. The low dry matter (DM) and water soluble carbohydrate (WSC) content of tropical (C₄) grasses during adverse climatic conditions wilting would need to be prolonged, which might lead to poor fermentation due to proteolysis by endogenous enzymes, which is reflected by a lower “true protein” proportion in the forage and, consequently, a higher ammonia-N proportion in the silage. Use of certain additives may be an alternative to wilting, particularly with thick-stemmed, erect fodder crop grasses (*Pennisetum*, *Panicum*, etc.) that produce a large mass of plant material, where pre-conditioning and handling is difficult to mechanize and labour-consuming. Tropical forage grasses (*Cynodon*, *Brachiaria*, *digitaria*, *Setaria*, *Chloris*, etc.) can be wilted more easily but, when wilted excessively it affects compression in the silo and thus fermentation quality (Catchpole and Henzell, 1971). Even under controlled wilting conditions, additives are being recommended to improve fermentation and nutritive value of conventional as well as round bale silages (Bates, *et al.*, 1989, Staples, 1995). More research is needed to address the specific problems associated with tropical silages. Tropical grasses and legumes have, for example, a relatively high concentration of cell wall components and a low level of fermentable carbohydrates compared to temperate forage crops (Catchpole and Henzell, 1971; Jarrige *et al.*, 1982). Furthermore, on average, storage temperature in tropical climates are higher than in temperate climates, which might give bacilli a competitive advantage over lactic acid bacteria (Gibson *et al.*, 1958). In addition, it has to be taken into account that some silo sealing material cannot withstand intense sunlight and this might impair the aerobic stability of the silage. Nevertheless, it seems likely that ensiling technologies from temperate climates can be modified for tropical conditions.

2.6.13 Ensiling technology for the tropics

In many tropical countries, such as in Africa, most of the farmers are small holder cattle owners who own two to five dairy cattle. Usually the cows rely on natural pasture which is abundant during the rainy season. However, in the dry season that can last for 6 months, the animals survive only on remaining dry pasture and on body reserves. Since lactating animals need extra feed for milk production, the lack of forage during the dry season is an obstacle to the development of dairy husbandry in these areas. Under such conditions the weak animals are unable to survive and those with better body conditions do not produce any milk (Dube, 1995; Smith, 1995). Therefore, preservation of forage crops would enable animals to be fed throughout

the year, increase milk yields considerably and this might have a great economical and social impact in this region.

Preservation of forage crops in the tropics might be problematic. If the crops are harvested at the end of the rainy season, it is impossible to dry them to hay, because of rain interruption. If the harvest is postponed to the beginning of the dry season, then the nutritional value of the crops decreases considerably, and they are much less digestible (Maclaurin and Wood, 1987). Ensiling is an alternative preservation method which can be practiced during the entire growing season and may yield higher quality and quantities of preserved forage. In farm situations, silage making often faces drawbacks which compromise the basic principles of silage making, especially where technology is limiting, such as with small-scale producers in the tropics and subtropics (Bayer and Waters-Bayer, 1998). Additives can never be substitute for good ensiling management. For example, additives will not make up for the negative effects on fermentation quality of tropical forages caused by practices such as the use of low quality, oxygen-permeable plastic covers, or extended storage under temperatures in excess of 30⁰C (Tjandraatmadja *et al.*, 1991). However, conventional ensiling technology requires large capital investments in silos and machinery, which the small holder cattle owner could not afford. Therefore, special ensiling technology should be developed for such farms, to meet the needs and to be economically feasible. In any case, the technology should be adapted to the local farmers through extension programmes.

In some areas farmers are reluctant to use ensiling and prefer to use low quality hay or even straw during the dry season. Ensiling experiments using small plastic bags have indicated that it is possible to ensile various crops (grass, wheat, maize, sorghum) in such units (Lane, 2000; Shariffah *et al.*, 2000; Ashbell *et al.*, 2001). There are many advantages for this technology: the bags are relatively inexpensive, the ensiling can be done manually by a few workers and the bag units can be used individually according to feeding requirements. Experimental results indicate that the quality of the silage obtained within the bags is quite good. It is hypothesised that the bags should not be completely air impermeable and that the volatile fatty acids which are produced during the fermentation are retained within the bags and inhibit spoilage yeasts and moulds (Ashbell *et al.*, 2001). Another possibility is to ensile in a pit dug in the soil which is lined with plastic sheeting and covered with plastic sheeting and soil (Kebe, 2004).

2.6.14 The Influence of silage fermentation on dry matter intake

It is generally recognized that voluntary intake of silage is less than that of the same forage that has not undergone fermentation. When same forage ensiled for varying lengths of time (0, 21, or 63 days) was fed to dairy cows, intakes of such silages was found to decline as the concentration of silage ammonia and butyric acid increase (Cushnahan *et al.*, 1995; Cushnahan and Gordon, 1995; Charmley, 2000).

2.6.14.1 Fermentation acids and silage intake

Butyric acid was first implicated as being responsible for reducing silage intake in 1963 (Harris and Raymond, 1963). Since then other volatile fatty acids (VFAs) have been identified (Brown and Radcliffe, 1971, Wilkins *et al.*, 1971; Wilkins *et al.*, 1978) as well as lactic acid (Choung and Chamberlain, 1993b). In studies to predicting voluntary intake from silage fermentation characteristics, Dawson and Mayne (1998) found no relationship between organic acid concentration and dry matter intake, while these authors (Rook and Gill, 1990; Rook *et al.*, 1990; Steen *et al.*, 1998; Wright *et al.*, 2000; Seglar, 2003) found only moderate correlation between fermentation acids and voluntary intake. Seglar (2003) reported that, it is the combination of high acetic acid levels, along with the presence of alcohol, and methyl-acetates that probably is really causing feed refusal.

2.6.14.2 Silage pH and silage intake

Low pH in silages is often associated with poor intakes because low pH in the rumen reduces cellulolytic activity and depresses intake. However there is no relationship between silage pH and rumen pH (Rooke, 1995). Silage is neutralized by saliva upon consumption. Low pH is typically associated with grain-based not forage based diets. Rooke (1995), suggest that lactic acid may have a direct effect on palatability, since sour taste is associated with reduced palatability. Any direct effects of lactic acid on silage dry matter intake may be more important in the short term than the long term and related to a negative feed back (Offer, 1997; 1998).

2.6.14.3 Ammonia -N and Silage Intake

Ammonia-N in silages has long been associated with reduced silage intake. It is a product of clostridia fermentation of amino acids. Clostridia silages become unpalatable primarily from nitrogenous proteolysis, which results in production of unpalatable amines, amides and nitrogenous end-products, which can reduce intake (Buchanan, 1990, Seglar, 2003). Silage ammonia concentration itself may not be important. Nevertheless, ammonia still ranks as the most frequently implicated factor for reduced silage dry matter intake (Cushnahan and Gordon, 1995; Patterson *et al.*, 1996; Steen *et al.*, 1998).

2.6.14.4 Clostridia and silage intake

Often times, producers make claims that cows are refusing silages from mycotoxins, when in reality it is the combination of high acetic acid levels along with the presence of other fermentation end-products. Clostridia silages that produce only butyric acid do not always impede appetites as much as when proteolytic end products are present (Seglar, 2003). Yeast organisms that cause aerobically unstable and hot silages do not always deter appetites. Other yeast strains however, have been implicated to produce objectionable odours that sometimes cause cows to refuse silages. However, the likelihood of increasing clostridia silage acceptance by dairy cows can be accomplished by removing a 12 – 24 hour supply of the silage to be fed and letting it “air out” to let the volatiles evaporate (Seglar, 2003).

2.7 Importance of Silage in Animal Production Systems

Overall, the role of silage as feed reserve is not important in the traditional village system leading to a general lack of adoption by smallholder farmers under the system. This is because, in many parts of the tropical region, these smallholder farmers are still able to feed browses, crop residues and agricultural by-products during dry season. It is noted that during the dry period, when green forages are scarce, crop by-products and farm wastes represent the important sources of feed for ruminant livestock (Khajarern and Khajarern, 1985). Similarly, during dry season, farmers have enough agriculture by-products such as rice straw, corn stover and pineapple waste for their animals (Poathong and Phakaew, 2001). In smallholder farms where livestock fodder such as crop residues and weeds abound and can be used conveniently without cost involved, silage production/utilisation has not found a place; but in some areas, where green corn is the

major product and farmers raise two or more ruminants or in small-scale dairying, silage may find a niche on smallholder farms ('t Mannetje, 1999). The latter is proven in the case of smallholder, farmers in Malaysia undertaking sweet corn stover ensilage activities in the state.

Interest in silage as feed reserve is limited to government ranches or institutions as well as progressive farmers undertaking specialised, commercial dairying or beef fattening in feedlots who see the importance of and need for readily available feed resources which can be efficiently utilised and can ensure feed security throughout the year. Silage as a routine feed to increase productivity of animals has gained in importance in the region, although actual cases are still small (Chin, 2002). These cases found in the tropics, indicate that farmers now realise silage can play a significant role as part of the routine diet of ruminants. In developed and Asian countries, the benefits of silage cannot be overemphasized particularly for commercial cattle operations and in dairying, where it has great economic benefits ('t Mannetje 1999). The importance of storing potentially unstable material to enable their extended use is well recognized by the most specialise commercial farmers but very few small older farmers are taking advantage of it, preferring to utilise forage resources in the form available to them (Chin, 2002). The importance of ensilaging as a means to utilise excess growth of pasture for better management and utilisation is also not fully realised on pasture based ranches in the region and in many instances pastures are seen to be allowed to become overly matured, thus deteriorating into poor quality swards which are either useful only as roughage or mowed down and wasted (Chin, 2002). Presently, silage has not become important under the livestock tree crop integration system. It is obvious that feeding forage resources fresh or in the form produced/obtained, is more important than feeding these after conservation. This is similar to the situation in India, where preference by most farmers to feed fresh hampers adoption of silage (Rangnekar, 1999).

2.8 Rumen Ecology

The stomach of the ruminant is divided into four compartments namely rumen, reticulum, omasum and abomasum. The rumen also known as a paunch, form the largest part of the reticulumen, which is the first chamber in the alimentary canal of ruminants. It serves as the primary site for microbial fermentation of ingested feed. The lining of the rumen wall is covered with small finger like projection called papillae which are flattened, approximately 5mm in

length and 3mm wide in cattle. The rumen has been well organized as an essential fermentation site that is capable of preparing end products particularly volatile fatty acids and microbial protein as major energy and protein for the ruminant host. The more efficient rumen is, the better the fermentation end product being synthesized (McDonald *et al.*, 1987).

In recent years, there has been increasing research directed at rumen ecology and rumen manipulation (Orskor and Flint, 1989). The rumen environment is a function of the type and quantity of feed eaten by the animal at a particular time. The periodic mixing through contraction of the rumen; salivation and rumination, diffusion or secretion into the rumen, absorption of nutrients from the rumen and passage of materials down the digestive tract (Preston and Leng, 1987) are immense factors. Under an abnormal situation, the rumen environment may be disorganized. For instance, a sudden introduction of a feed (diet) not normally included into the feed offered (like grain) could result in lactic acidemia, an ailment that causes the accumulation of lactic acid in the rumen. This could be due to a drop in ruminal pH, growth of *Streptococcus bovis* and the accumulation of lactic acid. The saliva helps in maintaining the pH of the rumen as a buffer (McDonald *et al.*, 1987).

Saliva also helps to maintain the fluid state of the rumen environment and so facilitates access of microorganisms to plant materials. The quantity of saliva secreted by ruminants depends on the diet. The presence of protozoa population affects the salivary flow and may be reduced by its presence. The protozoa rapidly assimilate starch and sugar and remove the need for copious salivation to maintain rumen pH (Preston and Leng, 1987). Saliva is a buffered solution of about pH 8. It contains high concentration of bicarbonate and phosphate ions which act as buffers. Both the saliva and bicarbonate movement across the rumen epithelium maintain the pH within narrow limits. The buffered rumen liquor favours the growth of the anaerobic bacteria, protozoa and fungi with the accumulation of volatile fatty acids (VFAs) in the fluid (up to 0.2 molar). For continuous fermentation, however, the ruminal pH must be constantly maintained at 5.5 – 6.5 (McDonald *et al.*, 1988) to ensure VFAs absorption through rumen wall. The biomass of microbes in the rumen is also maintained at a constant level by the passage of microbes down the digestive tract and by the death and lyses of the microorganisms within the rumen. The microbial cells, together with undegraded food components such as bypass protein pass to the abomasum

and intestines; there they are digested by enzymes secreted by the host animal and the products of digestion absorbed. Methane and carbon dioxide are produced as the end products of fermentation. At low rumen pH, carbon dioxide comes out of solution and accumulates in a pocket of the dorsal sac. Methane and carbon dioxide are largely eliminated by belching or eructation (Dougherty *et al.*, 1964). At high pH most of the carbon dioxide produced by fermentation or entering the saliva is absorbed and excreted via the lungs.

2.9 Ecology of Ruminal Microbes

The rumen is, to all intents and purposes, an open ecosystem; virtually all microbial species have the opportunity to grow there. The rumen microbial ecosystem is complex and highly dependent on the diet (Mako, 2009). The vast majority of ruminants consume a mixture of carbohydrates of which cellulose and hemicelluloses are the highest components. The diet can contain large amounts of soluble carbohydrates or starch (e.g. Molasses or grain). Plants have developed molecular structure in their cell wall specially to stop invasion by microorganisms. In the rumen, the main agents that break down carbohydrates are anaerobic bacteria, protozoa and fungi. The anaerobic bacteria are the principal agents for fermenting plant cell-wall carbohydrates but the anaerobic phycomycetous fungi, may at times be extremely important (Bauchop, 1981). There is a close relationship between fungi and other microbes in the rumen since the fungi appear to be the first organism to invade plant cell wall, which allows bacterial fermentation to start and to continue. Some bacteria in the rumen assumed a syntropic association, where one organism uses the products of fermentation of another and the removal of the end product allows further fermentation of the primary feeds source by the first organism (Preston and Leng, 1987).

2.10 Rumen Microorganisms

Microbes in the rumen include bacteria, protozoa and fungi. Bacteria along with protozoa are the predominant microbes and by mass account for 40 – 60% of total microbial matter in the rumen. The bacteria number is in the region of 10^9 to 10^{10} per ml of rumen content (McDonald *et al.*, 1988). Over 60 species have been identified. Most are non-spore-forming anaerobes. A number of the most important species are *Bacteriodes*, *succinogene*, *B. ruminicola*, *Ruminicoccus flavefaciens*, *R.albus*, *Streptococcus bovis* and *Megasphaera elsdenii* (McDonald *et al*, 1988). They are categorised into several functional groups, such as fibrolytic, amylolytic

and proteolytic type, which preferentially digest structural carbohydrate and protein respectively. The activities of a given species of bacteria may vary from one strain of that species to another. The total numbers of bacteria and the relative population of individual species, vary with the animal's diet; for example diets rich in concentrate foods, promote high total counts and encourage the proliferation of Lactobacilli. The anaerobic bacteria are the principal agents fermenting cell-wall carbohydrates (McDonald *et al.*, 1988).

Protozoa are present in much smaller numbers (10^6 per ml) than bacteria but, being larger, may equal the latter in total mass. In adult animals, most of the protozoa are ciliates belonging to two families. The Isotrichidae, commonly called the holotrichs, are ovoid organisms covered with cilia, they include the genera *Isotricha* and *Dasytricha*. The *Ophryoscolecidae*, or *oligotrichs*, include many species that vary considerably in size, shape and appearance; they include the genera *Entodinium*, *Diplodinium*, *Epidinium* and *Ophryoscolex* (McDonald *et al.*, 1988). The oligotrichs can ingest food particles and can utilise both simple and complex carbohydrates, including cellulose. The holotrichs, on the other hand, do not generally ingest food particles and cannot utilize cellulose.

A normal rumen flora (bacteria) and fauna (protozoa) is established quite early in life as early as six weeks of age in calves. However, if ruminants are born into, and reared in a special germ free environment (so-called gnotobiotic animals) they may be inoculated with one or more selected species of bacteria. Animals reared in a normal environment, but kept away from other ruminants, do not develop a protozoa population. Protozoa are easily killed by a low rumen pH, and are generally absent from animals fed on diets which promote- even a transiently- a low pH; all- concentrate diets, which are fermented rapidly, come into this category. Ruminants without rumen protozoa are apparently normal and healthy (McDonald *et al.*, 1988).

The fungi of the rumen have been studied for less than 20 years, and their place in the rumen ecosystem has yet to be fully characterized (McDonald *et al.*, 1988). They are regarded as part of the rumen population especially the anaerobic phycomycetous fungi (Bauchop, 1981). There is close relationship between fungi and other microbes in the rumen since the fungi appear to be the first organisms to invade plant cell walls, which allow bacterial fermentation to be initiated.

Rumen fungi are strictly anaerobic, and their life cycle includes a motile phase (as a zoospore) and a vegetative phase (sporangium). During the latter phase they become attached to food particles by rhizoids, which can penetrate cell wall. Several species or strains have been identified typically those belonging to the genus *Neocallimastix* (McDonald *et al.*, 1995). The rumen fungi are capable of utilising most polysaccharides and many soluble sugars; some carbohydrates not used by these fungi are pectin, polygalacturonic acid, arabinose, fructose, mannose and galactose (McDonald *et al.*, 1995). The contribution of the rumen fungi to the fermentation of food has not yet been quantified, but it is known that they are most numerous (constituting 10% of the microbial biomass) when diets are rich in fibre (i.e. not cereal diets or young pasture herbage) (McDonald *et al.*, 1995).

2.11 Rumen Microbial Interactions

The rumen microorganisms can be envisaged as operating together, as so called consortia, to attack and break down foods. A myriad of microorganisms are found throughout the digestive tract of the ruminant, but it is only the microbiota in the rumen that have a true symbiotic relationship with the host (Idahor, 2006). The rumen contains varied and dense microbial population predominantly anaerobic bacteria, protozoa and fungi. Microorganisms depend on the ruminant to provide the physiological conditions necessary for their existence. They in turn are essential for digestion and fermentation of the large amounts of fibrous feeds which the host cannot efficiently utilise (Czerkawski, 1986; Yokoyama and Johnson, 1993). Since the rumen naturally utilises the end products of microbial fermentation and biosynthetic activities to meet its own nutritional requirements. Interestingly, there is no indication of host specificity of these microorganisms in ruminants. While many species are unique in the rumen, others closely resemble those found in the digestive tracts of other ruminants.

Rumen microbial population varies within an animal with time after feeding between days in the same animal or in animals in different countries on similar feed (Hungate, 1975). However, the end products of fermentation are virtually the same. Bacteria associate with related organisms and function as a couple, one organism growing on the end-products of metabolism of another. The sequential fermentation process involving different species of organisms converting

cellulose to volatile fatty acids is well recognised. Within the rumen, there are often very close associations of bacterial species, dependent on simple materials liberated by each to mutual benefit of both (syntrophic association). The interrelationships between levels are high above a certain optimum, in which ammonia is incorporated into amino acids without using ATP. These interactions of rumen bacteria appear to be highly beneficial and there appears to be little that can be done to manipulate the associations, other than inhibition of methanogenesis (Wolin, 1979). There are interactions between bacteria and protozoa. Protozoa ingest and digest bacteria and can reduce the bacteria biomass floating free in solution in the rumen (Coleman, 1979). This may reduce the rate at which bacteria colonize ingested food particles. Protozoa effectively compete with bacteria for soluble sugars and starch, storing these carbohydrates within their cells. In this way the protozoa can reduce severity of acidosis in some diets.

On sugar-based diets (e.g. sugarcane) the protozoa biomass is probably larger than the bacteria biomass (Preston and Leng, 1987). It has been observed that elimination of protozoa in the rumen lead to an increase in the number of bacteria in the liquid pool. In a study with sheep using total faecal collection procedures, the apparent digestibility of dry matter was increased by 18% when protozoa were not present (Soetano, 1986). This was possibly because of the effects of protozoa on the relative proportion of the rumen volatile fatty acids.

Some microorganisms, like the fungi, are capable of invading and colonizing plant tissues; others follow up to ferment the spoils of the invasion. As the microbial mass synthesized in the rumen provides about 20% of the nutrients absorbed by the host animal, the composition of microorganisms is important (McDonald *et al.*, 1988). Interactions among microorganisms in the rumen are complex and not often to the advantage of the host. Large protozoa population in the rumen has been shown to reduce animal productivity possibly lowering the amino acid to energy ratio in the absorbed products of digestion. It appears that protozoa reduce the biomass of bacteria and of fungi in the rumen of animal on diets high in fibre and this may reduce the rate of digestion of fibre feeds (Preston and Leng, 1987).

2.12 Ammonia in Rumen Fermentation

Approximately 40 – 60% of the dry matter of the microbial cells is protein. The synthesis of amino acid and proteins are the reactions that require ATP. The pathways of synthesis of amino acids in rumen microbes are not clearly defined. It is however; abundantly clear that ammonia is highly important for the efficient synthesis of amino acids and therefore microbial protein (Satter and Slyter, 1974). Ammonia is intensively used by many species of rumen microorganism as a source of nitrogen for synthesis of their nitrogenous constituent. Some species of organism commonly found in the rumen require pre-formed peptide or amino acids. If these are not provide in the diet and are low in concentration in rumen fluid, some micro-organism may arise and others may disappear from the rumen, changing the balance of species. At low ammonia level in rumen fluid, reactions that fix ammonia into acids require ATP, whereas when ammonia level is high above a certain optimum, the ammonia is incorporated into amino acids without using ATP (Satter and Slyter, 1974).

It has been suggested (Satter and Sylyter 1974) that maximum microbial synthesis rate occurs at ammonia concentrations between 5 and 8mg N/100ml. Different options have been reported, suggesting that diet influences optimum level of ammonia. Another study Schaefer *et al.* (1980) suggests the value may be as high as 14mg N/100ml depending on diet. The high ammonia concentration needed for maximum cell growth suggests that the rumen micro organisms probably have similar mechanism for incorporation of ammonia via glutamate dehydrogenase.

Food protein of ruminant are hydrolysed to peptides and amino acids by rumen microbes. Some amino acids are degraded further to organic acids, ammonia and carbon dioxide. The ammonia produced, together with some small particles and free amino acids are utilised by the rumen organisms to synthesise microbial protein. When the organisms are carried through to the abomasum and small intestine, their cell proteins are digested and absorbed. The rumen bacteria are capable of synthesising both essential and non-essential amino acids, thus they render their host independent of dietary supplies of the former. With most diets, the greater part of the protein reaching the ruminant's small intestine will be microbial protein of fairly constant composition. To a lesser extent, the nitrogen of food protein entering the rumen may leave it in the same form of the protein escapes degradation, this is called by-pass protein and it is subsequently digested

in the small intestine which will vary in amino acid composition according to the nature of the diet (Schaefer *et al.*, 1980).

Ammonia in rumen liquor is the key intermediate in microbial degradation synthesis of protein. When a diet is protein-deficient or if the protein resists degradation, the concentration of rumen ammonia will be low and the growth of rumen microorganisms will be slow, consequently, the breakdown of carbohydrates will be retarded. However, in grazing ruminants, this situation occurs less frequently than might be expected because sheep and cattle show ability to select material of high nitrogen content from poor quality pasture (Loosli and McDonald, 1968). On the other hand, if protein degradation proceeds more rapidly than synthesis, ammonia will accumulate in rumen liquor and optimum concentration will be exceeded. When this happens, ammonia is absorbed into the blood, carried to the liver and converted to urea. Some of the urea may be returned to the rumen via the saliva and also directly through the rumen wall, but the greater part is excreted in the urine and thus wasted (McDonald, *et al.*, 1988).

It has been found that although lactating cow can be maintained on protein free diet (Virtanen, 1996), for maximum milk production 20% of the dietary nitrogen has to be supplied as protein.

2.13 Protein Utilisation in Ruminants

The protein content of forage legume leaves (12 – 30%) is high compared with that of mature grasses (3 – 10%), (Norton, 2010). Proteins are digested in the rumen to provide ammonia and amino acids for microbial protein synthesis. Later, microbial cells pass to the small intestine, producing a major source of absorbed amino acids for the ruminant. Sometimes, feed proteins escape digestion (by-pass proteins) in the rumen and provide additional protein for absorption in the small intestine. The microbial population in the rumen requires a minimum level of ammonia (70mg N/L) to support optimum activity (Norton, 2010). Lower values are associated with decreased microbial activity (digestion) and indicative of nitrogen deficiency. Feeds containing less than 8% CP are considered deficient as they cannot provide the minimum ammonia level required. However, ARC (1980) reported a value of 7% as the minimum requirement for ruminant function while NRC (1981) considered the value of 6% as being the least required crude protein content and less than that will decrease the voluntary feed dry matter intake. As the

voluntary dry matter intake decreases, the rumen efficiency decreases due to reduced energy and protein availability (NRC, 1981) and consequently, growth is affected.

2.14 Degradation of Protein in Ruminants

Solubility of protein differs from diet to diet. Solubility of protein expresses the rate at which the protein is hydrolysed in the rumen (Orskov and Ryle, 1990). They are incorporated into microorganisms with an efficiency that is low (McAllister *et al.*, 1994). Some part of the formed microbial protein will equally be lost as nucleic acid (which is not usable by the host ruminant). However, when the solubility is low (e.g. fish meal) amount hydrolysed in the rumen will be relatively low (McAllister *et al.*, 1994). The rest remains as by-pass protein and enters into the abomasum to undergo an enzymatic digestion which makes the resulting amino acid to be available to the host ruminant directly and less of it is lost (Orskov and Ryle, 1990).

Degradation of protein in the rumen involves a sequence of microbial enzymes starting with proteases and continuing with peptidase and deaminases, until eventual ammonia is liberated (Smith, 1989). Chalupa, (1980), reported that when supplementary diets with antibiotic (or other agents such as monensin etc.) are fed to ruminants, protein degradation is altered or reduced. Bacteria are generally regarded with good reason, as being mainly responsible for degrading dietary protein, but it appears that protozoa degrade the protein occurring in bacteria and in some feed particles that they are able to engulf. Protozoa could sometimes contribute appreciably to overall protein degradation in the rumen (Hino and Russel, 1987).

It is apparent that ammonia is a common and important intermediate in both protein and non-protein nitrogen (NPN) digesting in ruminants (Perry, 1980 and Chaudhry, *et al.*, 1997). It is important therefore, to maximize efficiency of conversion of ammonia in the rumen. Some ammonia is inevitably absorbed from rumen and carried by the blood to the liver, where it is converted to urea (Chaudhry *et al.*, 1997). Perry, (1980) and McAllister *et al.*, (1994) reported that, the efficiency of utilisation of NPN decreases rapidly as the amount of NPN fed increases, as the protein level in the supplemented diet increases and as the total digestible nutrient (TDN) content decreases.

2.15 Volatile Fatty Acids

The diet of the ruminant contains considerable quantities of cellulose, hemicelluloses, starch and water-soluble carbohydrates. All the carbohydrates, but not lignin, are attacked by rumen microorganisms. The main end products of the metabolism of carbohydrates by rumen microorganisms are volatile fatty acids (VFAs). The predominant volatile fatty acids in the rumen fluid are acetic, propionic and butyric acids with isobutyric, isovaleric, valeric and other acids generally present in small amounts. The rate and volume of the end products produced is directly proportional to the microbial activity in the rumen (Mako, 2009). Also, Bergman, (1990 and Tortuero *et al.* (1994) stated that concentration and relative proportion of volatile fatty acids are related to the nature of the feed and dietary fibre. In a similar report (Firkins *et al.*, 1986 and Robinson *et al.*, 1986) the volatile fatty acids produced depend on the extent of digestibility (effective degradability) of the feed ingested by the animals which subsequently determines the amount of substrate available for fermentation. Ruminal fermentation of glucose, fructose and sucrose results in the production of lactic, acetic, propionic and butyric acids. Maltose and galactose are fermented more slowly (Perry, 1980).

Furthermore, the total concentration of volatile fatty acids in the rumen liquor varies according to the animal's diet and time that has elapsed since the previous meal. The relative proportions of acids also vary. The predominant acid is acetic, and roughage diets high in cellulose give rise to acid mixture particularly high in acetic acid rises. As the proportion of concentrates in the diet is increased, the proportion of acetic acid falls and that of propionic acids. With all concentrate diets the proportion of propionic acid may exceed that of acetic. In a similar report (Perry 1980), diets heavy in starch or sucrose favour propionic acid production. The production of propionic acid in the rumen does not result in energy losses from gas production during rumen fermentation (Perry, 1980). However, certain nutrient could at times be highly inefficient, i.e. acetate (with a high heat increment) when given in a diet (Leng, 1989).

Volatile fatty acids produced in large amount through ruminal fermentation and are of paramount importance, in that they provide greater than 70% of the ruminant energy supply. Virtually all of acetic, propionic and butyric acids formed in the rumen are absorbed across the ruminal epithelium from which they are carried by ruminal vein to the portal vein and hence to the

liver. Continuous removal of volatile fatty acids from the rumen is very important not only for distribution but to prevent excessive and damaging drop in pH of rumen fluid (Cronje *et al.*, 2000). Immediately after a meal in the ruminant, there is a rapid rate of gas production. Typical composition of rumen gas is: carbon dioxide, 40%; methane, 30 – 40%; hydrogen, 5%; together with small varying proportions of oxygen and nitrogen from ingested air (McDonald *et al.*, 1988).

2.16 Dietary Fibre

Advances in our understanding of fibre digestion, analytical methods for useful fibre analysis and computer models for their practical application are transforming the practice of ration formulation for ruminants. Today, the profitability of ruminant production systems depends largely on maximizing productivity through the efficient use of available resources (Johnston and Tricarico 2007). Grasses, one of the most important sources of roughage used in ruminant nutrition, contain substantial amounts of cell wall carbohydrates. Cell wall carbohydrates can be quantified by determination of neutral detergent fibre (NDF), which includes cellulose, hemicellulose and lignin as the major components (Van Soest *et al.*, 1991). Due to the variability of NDF in rumen degradation and its influence on animal performance, the knowledge of NDF digestibility in forage is critical for effective ruminant feeding (Oba and Allen, 1999). The importance of fibre in ruminant diets is undisputed and neutral detergent fibre (NDF) is widely used to characterise the fibre fraction of ruminant diets. The role of NDF in ensuring proper rumen function and the influence of NDF digestibility on productivity are widely recognised. To put the latter in perspective, Oba and Allen (1999) estimated that a one percent (1%) increase in ruminal NDF digestibility, estimated *in vitro* or *in situ*, will lead to a 0.25kg increase in 4.0% fat corrected milk production.

Dietary carbohydrates can be divided into two basic fractions; fibre and non-fibre carbohydrates (NFC). Fibre plays a fundamental role in ruminant and dairy cattle nutrition. It has been widely demonstrated that both the amount and physical form of dietary fibre are important in lactating dairy cows ration in order to maintain proper ruminal function, animal health status and milk composition (Afshar and Naser, 2011). Some nutritionists define fibre as the any component in a feed that is not digested by mammalian enzymes. Some of these components are soluble under

mild extraction procedures and thus result in “soluble” and “insoluble” fibre. Most constituents of soluble fibre (pectin, fructans, and beta-glucans) are readily fermented in the rumen and may even be readily fermented in the large intestine of monogastric animals (Kearney, 2005, Righi *et al.*, 2008 and Mertens 1997). Nutritionally, fibre has both physical and chemical attributes that are related to the mechanical processes of digestion (chewing and passage) and to enzymatic degradation associated with fermentation (Mertens, 1997).

Mertens (1997) stressed that chemical definition of dietary fibre such as neutral detergent fibre (NDF) or acid detergent fibre (ADF) content was an inadequate description of the fibre content of a diet. The ADF fraction of feedstuffs includes cellulose and lignin as the primary components. Concentrations of ADF and lignin are correlated more with digestibility than with intake. Many factors influence the relationship between ADF and digestibility, including forage variety, maturity at harvest, and storage conditions (Varga *et al.*, 1998, Van Soest *et al.*, 1991). It is an indicator of digestibility, as the ADF increases, digestibility decreases. NDF is a measure of cellulose, hemicelluloses, and ADF and lignin fractions of feeds. NDF is more highly correlated with feed volume and chewing activity than ADF or crude fibre (CF) (Varga *et al.*, 1998 and Coppock, 1987). Some of the NDF is highly digestible. Forage NDF is the best indicator of voluntary feed intake (VFI) of an animal. As the NDF content of forage increases, the VFI decreases. Also it has been shown severally that the digestibility of a plant material in the rumen is related to the proportion and lignifications of plant cell walls (NDF). Forage with high lignin contents are often of low digestibility (Norton, 2010).

The content of indigestible NDF (INDF), which needs long *in sacco* incubation periods (Fonseca *et al.*, 1998), presents an important indicator of the quality of grass cell wall carbohydrates and can be a good predictor of *in vivo* digestibility of roughages (Nousiainen *et al.*, 2003). Forage digestibility in ruminants is constrained by the extent of cell wall (NDF) digestion (Van Soest, 1994). Indigestible neutral detergent fibre (INDF) is the most important factor affecting the total diet organic matter digestibility (Nousiainen *et al.*, 2004). A part of the forage cell wall, i.e. INDF, is unavailable to microbial digestion in ruminants, even if the total tract residence time of fibre could be extended to an infinite time (Huhtanen *et al.*, 2006).

The influence of maturity at harvest on the chemical composition and digestibility of grasses is more pronounced than other management factors such as particle size, dry matter (DM) at harvest or harvesting system (Harrison *et al.*, 2003). Incomplete degradation of cell walls is a major factor limiting the value of forages and straws for animals (Ahmad and Wilman, 2001). Grenet and Besle (1991) and Nagadi *et al.* (2000) postulated that the cell wall carbohydrates are little degraded in the rumen due to a high extent of lignification. Lignin is generally accepted as the primary component responsible for limiting the digestion of forages (Van Soest, 1994; Traxler *et al.*, 1998; Agbagla-Dohnani *et al.*, 2001).

The National Research Council (NRC, 2001) recommends NDF to be maintained at 25% of dietary DM with at least 75% from forage for the NDF requirement. Therefore, there is room for up to 25% of the NDF from non-forage fibre sources (NFFS) to meet the NDF requirement (Shane, 2010). Mertens (1997) proposed definitions for both effective NDF (eNDF) and physically effective NDF (peNDF). The peNDF of a feed is related to the physical properties of its fibre (primarily particle size) that stimulates chewing activity and establishes the biphasic stratification of ruminal contents (floating mat of large particles on a pool of liquid and small particle, (Mertens, 1997). The peNDF content of the diet can be determined by multiplying the NDF concentration by the proportion of particles retained on a 1.18mm sieve or by its physical effectiveness factor (Shane, 2010). The eNDF is related to the sum total ability of a feed to replace roughage so that the percentage of fat in milk is effectively maintained. Because peNDF relates only to the physical properties of fibre, it is a more restricted term and concept than eNDF. The peNDF will always be less than NDF, whereas eNDF can be less than or greater than the NDF concentration in a feed (Mertens, 2000).

Effective NDF (eNDF) is required by dairy cows to stimulate chewing, maintain optimal rumen environment and prevent milk fat depression (Mertens, 1997). Several parameters, including chewing, ruminal pH, acetate; propionate ratio, and milk fat percentage, have been used as animal responses assess the effectiveness of NDF in dairy ration (Kononoff *et al.*, 2000). The peNDF value of non-forage fibre sources is considerably lower than long-stem forages, but may be higher than some forms of concentrates, grains and ground forages. Increased amounts of fibre in dairy rations stimulate chewing activity and reduce acid production. The cascade of

events leading to a decrease in animal performance when too little effective fibre is fed includes decreased chewing activity, leading to less salivary buffer secretion, which leads to lower ruminal pH and results in altered ruminal fermentation patterns and the low ratios of acetate to propionate (A: P) that ultimately result in modified animal metabolism and reduced milk fat synthesis.

Methods for NDF determination are time consuming and expensive. Prediction equations, based on basic parameters of chemical analysis, are cheaper and faster for institutions without availability of experimental animals (Jancik *et al.*, 2008). Today, the accurate knowledge of NDF digestibility has become extremely important particularly due to the rising costs of most feed ingredients. Under these circumstances, improved NDF digestibility becomes imperative to take full advantage of available resources in ruminant feeding programme (Johnston *et al.*, 2007).

2.17 Methane Production from Ruminants

Ruminants depend on microorganisms to digest and ferment plant cell walls and polysaccharides into energy sources such as volatile fatty acids and other organic acids. Microbial fermentation in the rumen also produces waste products such as carbon dioxide (CO₂) and methane (CH₄). Methane production in the rumen is a loss of energy, since the proportion of animal feed which is converted to CH₄ is eructed as gas into the atmosphere. Carbon dioxide is produced partly as a by-product of fermentation and partly by the reaction of organic acids with the saliva. The basic reaction by which methane is formed is the reduction of carbon dioxide by hydrogen. Methanogenesis is a complex process which involves folic acid and vitamin B₁₂. Approximately 6 – 7% of dietary gross energy intake is lost to the atmosphere as methane CH₄ (Holter and Young, 1992; DeRamus *et al.*, 2003). About 4.5g of methane is formed for every 100g of carbohydrate digested (McDonald *et al.*, 1988).

Recently, emission of methane and other volatile organic compounds from ruminants and their effect on air quality has attracted the attention of air regulatory agencies in many parts of the world. Methane contributes to climate change in form of global warming (Johnson and Johnson, 1995). Methane is a green house gas whose atmosphere concentration has increased dramatically over the last century. Methane released to the atmosphere by domestic ruminant livestock is

considered to be one of three largest sources on a global scale. Methane is the largest potential contributor to the global warming phenomenon (Moss *et al.*, 2000). Fermentation of feeds in the rumen is the largest source of methane from enteric fermentation. This influences the production of different volatile fatty acids which has a marked effect on production of methane in the rumen. Acetate and butyrate promote methane production while propionate formation can be considered as a competitive pathway for hydrogen use in the rumen.

Methane contributes to the atmospheric greenhouse effect by trapping outgoing terrestrial infrared radiation 20 times more effectively than CO₂ which leads to increased surface temperature and directly affects atmospheric oxidation reactions that produces CO₂ in animal agriculture. The National Institute of Water and Atmosphere, New Zealand (NIWA, 2005) predict that as a result of global warming temperature will increase by 0.3 to 1.4⁰C by 2030 and projected increases for 2070 of between 0.6 and 2.8 degrees. This will create a rise in sea levels up to 35cm over the next 30 – 70 years. Current trend suggests there may be need to reduce the extent of methane production by manipulating diet through management practices that may influence ruminal microbial fermentation (Johnson and Johnson, 1995). Environmental pollution and menace from dairy farms could be caused by overfeeding and/or poor synchronisation of release of nutrients in the rumen. Consequently, attempts have been made to manipulate rumen fermentation using ration manipulation strategies which may include the addition of ionophores, fats and yeast cultures. For example, addition of monensin to dairy cattle rations decreased CH₄ production, decreased feed intake and increased milk yield (Sauer *et al.*, 1998), suggesting that reduction, in methane production per unit of ingested feed is associated with improvement of feed utilisation efficiency.

Getachew, *et al.*, (2001); Dohme *et al.*, (2001) and Fievez *et al.*, (2003) have observed that fat composition and its total amount can exert biologically important influences on rumen fermentation. Ability of fat to suppress methanogenesis is largely due to the reduction in protozoa population in the rumen. Protozoa produce a considerable amount of hydrogen which can only be transformed to methane by the action of associated methanogenic bacteria and without protozoa, these bacteria lack substrate.

Apart from the above ways of methanogenesis reduction, saponin and tannins are also involved. They may be effective in reducing emission of methane into the atmosphere. Hess *et al.* (2003) observed that supplementation of saponin-rich fruits of the tropical multipurpose tree *Sapindus saponaris* reduced methane emission without negatively affecting organic matter degradation. Also, Babayemi *et al.* (2004) found the seeds of *Albizia lebbek* and *albizia rhizonse* to depress methanogenesis is due to the presence of saponin in the seeds. Saponins suppress protozoa, the main butyrate producers in the rumen. Saponins act as potential defaunating agent (Teferedegne, 2000).

The effect of tannins in the suppression of methane production has been reported. Hess *et al.* (2004) observed *in-vitro* that inclusion of the tropical legume *Calliandra calothyrsus* (270g of condensed tannins/kg dry matter) in a grass-based diet suppressed methane production relative to dry matter degraded by over 30% and that this was probably due to tannins in the legume. Carulla *et al.* (2005) also reported a depressed methane production in lambs supplemented with approximately 25g condensed tannins/g dry matter which was probably due to the suppression effect of fibre degradation by tannins. Furthermore, Field *et al.* (1989) observed that a direct effect of condensed tannins on ruminal methanogens cannot be excluded. Similarly, Fievez *et al.* (1999) reported that some undegraded nutrients in the rumen due to tannins are degraded in the hindgut. There is therefore the possibility of lowering the methane emission because fermentation in the hindgut differs from that in the rumen by a lower methane production per unit of ferment nutrient. Whichever method adopted, methanogenesis reduction can be mediated by diet manipulation through the intervention of ionophores, fats, yeast cultures, saponins and tannins.

2.18 In Vitro Gas Production Technique

The in vitro gas production technique was developed by Menke *et al.* (1979) and has been used by Blummel and Ørskov (1993) to determine gas production at several incubation times. Gas production reflects all nutrients fermented, soluble as well as insoluble, the fractions that are not fermentable do not contribute to gas production. Although, gases produced during rumen fermentation are waste products and no nutritive value for the ruminant; gas production tests are used routinely in feedstuff research as gas volumes are related to both extent and rate of substrate

degradation (Blummel *et al.*, 1997). The gas method has been used successfully to predict the metabolizable energy (ME) content of feeds.

Recently, seven laboratories around the world that use a gas method carried out a comparative test to assess the repeatability of *in vitro* technique in predicting the energy value of feeds, and found that the gas method was repeatable among laboratories (Getachew *et al.*, 2002). *In vitro* methods for laboratory estimations of graded feeds are important for ruminant nutritionist. *In vitro* methods have the advantage not only for being less expensive and less time consuming, but they maintain experimental conditions more precisely than *in vivo* trials. Three major biological digestion techniques are currently available to determine the nutritive value of ruminant feeds.

- Digestion with rumen microorganisms as in Tilley and Terry (1963).
- *In situ* incubation of samples in nylon bags in the rumen (Mehrez and Ørskov, 1977).
- Gas method (Menke *et al.*, 1979).

The Tilley and Terry (1963) technique is convenient and largely used when large-scale testing of feedstuffs is required. This method is employed in many forage evaluation laboratories and involves two stages in which the forages are subjected to 48 hours fermentation in a buffered solution containing rumen fluid, followed by 48 hours digestion with pepsin in acid solution. The residue after 48 hours incubation is treated with neutral detergent solution to estimate true dry matter digestibility. Despite that Tilley and Terry (1963) method has been justified with *in vivo* values (Van Soest, 1994) it appears to have several disadvantages. The method gives only one observation and unless lengthy and labour-intensive time course studies are made, the technique does not provide information on the kinetics of forage digestion. The residue determination destroys the sample and therefore a large number of replicates are needed. This method is therefore difficult to apply to containers /materials such as tissue culture samples or cell wall fractions.

The nylon bag technique (*in sacco* technique) is probably the most used method for feed studies. The nylon bag technique has been used for many years to provide estimates of both the rate and content of disappearance and potential ruminal degradability of feedstuffs. However, some draw

backs have been pointed out (Michalet-Dorean and Ould-Bah, 1992). In this technique however, only a small amount of forage samples can be assessed at any time and also requires at least three fistulated animals to account for variations due to animals. Hence it is of limited value in laboratories undertaking routine screening of a large number of data samples, very laborious, time consuming and provides a limited number of data points (Cone, 1991). Therefore it requires a large number of samples; large error could result in values obtained at early stages of digestion due to a reduced weight loss and adherence of microbes to poor quality roughages at early stages. This can lead to higher weights and thus distortion of results.

Ørskov and Ryle (1990) showed the possibility of underestimation of dry matter loss from the nylon bag technique at an early stage of incubation which could be due to adherence of microbes. Tilley and Terry (1963) established that the nylon bag technique overestimated fermentation. This extent was due to the carbohydrate composition of feeds, especially at short incubation times and this infers that it could be caused by a rapidly fermentable fraction which was lost from bags before it was fermented. Gas measuring technique (*in vitro* gas method): The gas measuring technique has been widely used for evaluation of feeds. Gas measurement provides a useful data on the digestion kinetics of both soluble and insoluble fractions of feedstuffs. Several gas measuring techniques and *in vitro* gas methods are in use by several groups. The *in vitro* gas method based on syringes (Menke *et al.*, 1979, Blummel *et al.*, 1997) appears to be the most suitable for use in developing countries.

Gas measurement focuses on the appearances of fermentation products (soluble but not fermentable products do not contribute to gas production). In the gas method, kinetics of fermentation can be studied on a single sample and therefore a relatively small amount of sample is required or a large number of samples can be evaluated at a time. The *in vitro* gas method is more efficient than the *in sacco* method in evaluating the effects of tannins or other anti-nutritive factors. However, the *in vitro* rumen fermentation method as described by (Getachew, 2002) has the following advantages.

- I. It has the potential for screening a large number of feed resources, for example in breeding programmes for the development of varieties and cultivars of good nutritional value.

- II. It could also be of great value in the development of supplementation strategies using locally available conventional and non-conventional feed constituents to achieving maximum microbial efficacy in the rumen.
- III. It provides better insight into nutrient interactions.
- IV. The method is also being used increasingly to screen plant derived rumen modulators.
- V. It is less expensive and allows maintaining experimental conditions more precisely than do *in vivo* trials.

Moreover, the use of *in vitro* fermentation will help to alleviate the problems posed by the disposal of various agro-industrial by-products, thereby improving the quantity and quality of the year-round feed supply for livestock production especially in the developing nations of the world.

2.19 The Origin of *In Vitro* Gas

The relationship between rumen fermentation and gas production has long been established (Getachew *et al.*, 1998). The genesis however of rumen gas fermentation, technique began in the early 1940s (Quin, 1943). The idea of this method became a routine method of feed evaluation after the works of Menke *et al.*, (1979) where a high correlation between gas production and *in vitro* apparent digestibility was reported. Menke *et al.*, (1979) first described a method where the gas, evolved during fermentation by rumen microbes was collected and used as a measure of extent of fermentation. Essentially similar to the first stage of the Tilley and Terry method incubation were however conducted in large glass syringes to trap the gas evolved, whereas in the Tilley and Terry method, gases are allowed to escape to the atmosphere. Theodorou *et al.* (1994) described a gas production method which used a pressure transducer to monitor the production of gas. The method is essentially the same in principle as the Menke method, the key difference being that instead of using glass syringes, sealed bottle were used. The gas pressure inside the bottles is measured using the pressure transducer and the gas produced is removed, the volume measured and the gas discarded.

During the incubation of feedstuffs with buffered rumen fluid *in vitro*, carbohydrates are fermented to short chain fatty acids (SCFA), gases (mainly CO₂ and CH₄) and microbial cells. Fermentation of carbohydrates to acetate, propionate and butyrate result in gas production

(Blummel and Ørskov, 1993; Beuvink and Spoelstra, 1992). When protein is fermented, gas produced is relatively small compared to carbohydrates fermentation (Getachew *et al.*, 1998). Gas production from fat fermentation is negligible (Menke and Steingass, 1988, Getachew *et al.*, 1997). Getachew *et al.* (1997) observed that incubating 200 mg of coconut oil, palm kernel oil and/or soybean oil, 2.0 to 2.8ml of gas were produced while 200 mg of Casein and Cellulose produced about 23.4ml and 80ml of gas respectively. In the technique, gas produced is the direct gas produced as a result of fermentation (CO_2 and CH_4) and the indirect gas produced from the buffering of SCFA (CO_2 released from the bicarbonate buffer). Incubation of roughages with bicarbonate buffers produced about 50% of the total gas from buffering of the SCFAs and rest is generated from fermentation (Blummel and Ørskov, 1993). At very high molar propionate levels, the amount of SCFA is about 60% of total gas production. SCFA produced from fermentation releases 0.8-1.0 mmol of CO_2 from the buffered rumen fluid solution depending on the amount of phosphate buffer present. A highly significant correlation has been observed between SCFA and gas production (Blummel and Ørskov, 1993).

When a substrate is fermented to acetate and butyrate, gas is produced. A conclusion was drawn from Van Soest, (1994) that substrate fermentation to propionate yields gas only from buffering of the acids. This suggested that relatively lower gas production is associated with propionate production. It is also important to note that the type of substrate fermented influences the major proportions of different SCFA (acetate, propionate and butyrate) (Beuvink and Spoelstra, 1992, Blummel and Ørskov, 1993). Hence, the molar ratio of acetate to propionate was used to substantiate substrate related differences. Rapidly fermentable /degradable carbohydrates yield relatively higher propionate as compared to acetate and the reverse takes place when slowly fermentable carbohydrates are incubated (Getachew *et al.*, 1998).

There are a number of factors that affect rate of fermentation of feeds by rumen microorganisms and hence gas production. Among these are intrinsic characteristics of the carbohydrate fractions, such as the proportion of starch or cellulose and the extent of lignifications of the cell wall. The extrinsic factor however is the supply of fermentable nitrogen required by microorganisms to help them to synthesise cellular constituents such as proteins and nucleic acids required for growth. Ammonia is the simplest form of fermentable nitrogen required by microorganisms

(Hungate, 1966). Many research efforts have looked at the effects of the ruminal ammonia concentration on feed degradation, microbial outflow. Hume (1970) in an *in vivo* study reported that the microbial protein was maximal when ammonia was 88mg N/l but microbial protein flow was highest with an ammonia concentration of 133mgN/l. Allen and Miller (1976) however, found that the greatest flow of non-ammonia concentration in the rumen was between 160 and 220mgN/l. However, Menke *et al.*, (1979) using an *in sacco* method, observed that an ammonia concentration of 200mg N/l was necessary to acquire the maximum rate of disappearance of barley DM in Sheep. Similarly, Wallace (1979) perceived increased *in situ* DM and CP degradation rates of barley grain accomplished by increased bacterial growth when rumen ammonia concentration was increased from 97 to 214mg N/l.

2.20 The Application of *In Vitro* Gas Method in Predicting Voluntary Intake, digestibility of Organic Matter and Metabolizable Energy

The main constraints to the utilization of roughages by ruminants is voluntary feed intake, therefore prediction of voluntary feed intake, particularly of fibrous roughage, is one of the important factor in ruminant nutrition. The *in vivo* determination in quantifying intake and digestibility of feedstuff has been found to be time consuming (Coelho *et al.*, 1988, Carro *et al.*, 1994) laborious, expensive and also require large quantity of feed. This makes it better suitable for large scale evaluation. In recent times, major attempts were made in predicting intake and digestibility using laboratory procedures. *In vitro* gas production has been used to predict dry matter (DM) intake.

Various workers have reported significant correlation between *in vitro* gas production and DM intake. Forage cells walls have considerable influence on voluntary feed intake through rumen fill mechanism (Van Soest, 1994). Gas production from extracted neutral detergent fibre was shown to be better correlated to voluntary intake than the values obtained from the incubation of whole roughages. The use of various models for intake prediction was investigated and its currently appears that combination of gas volume measurements (4 – 8hrs) with concomitant determination of the amount of substrate degraded (> 24hrs) is superior to the models based on kinetics of gas production only. The *in vitro* gas production from NDF explained more (82% V

75%) of the variation in DM intake than gas production from whole roughages (Getachew, *et al.*, 1998).

In vitro techniques had been widely used for evaluation of nutritive value of feeds. Fievez *et al.*, (2005) reported that *in vitro* fermentation techniques have been employed to predict the nutritive value of tropical browse seeds. Although gases produced during rumen fermentation are colossal waste products and of no nutritive value to the ruminants but gas production tests are used routinely in feed research as gas volumes are related to both the extent and rate of substrate degradation (Blummel *et al.*, 1997). *In vitro* gas method for evaluation of feeds are an important tool as they allow guide assessment of nutritional value and potential deleterious activity of any anti-nutritional compound present in the material. Ruminal fermentation results in redistribution of feed organic matter (OM) into microbial mass, volatile fatty acid (VFA) and gas in variable proportions. An imbalance of protein and carbohydrate availability to rumen microbes can be a reason for low efficiency of microbia protein production, (Russel, 1998). The proportional redistribution of organic matter among individual VFA is varied as it depends on diet composition, feeding regime and other factors that affect microbial metabolism.

Fermentation of carbohydrate to acetate result in a higher supply of ATP to microbial metabolism, but lower supply of ATP in the form of VFA energy to the animal than butyrate and especially, propionate production per unit of hexose fermented (Beever, 1993). One might expect that rumen microbes use fermentation pathways that maximize the amount of ATP available for microbial metabolism per unit of hexose. Therefore, acetate is the most abundant VFA in the rumen, where fermentable carbohydrates are normally limiting. Where fermentable carbohydrate are abundant, bacterial growth can increase due to higher ATP production per unit time rather than per unit substrate, in such cases, fast growing lactate and propionate producing microbes outgrow the slower growing acetatge producers (Russel and Wallace, 1988). Sveinbjornsson *et al.* (2006b) reported that proportional incorporation of substrates into microbial matter and fermentation end products were not only affected by the total availability of different substrates but also by their degradation rate.

CHAPTER THREE

3.0 QUALITY CHARACTERISTICS, CHEMICAL COMPOSITION AND ACCEPTABILITY OF ENSILED MAIZE STOVER WITH OR WITHOUT ADDITIVES

3.1 Introduction

One of the challenges facing ruminant livestock farmers in the tropics especially in Nigeria is poor nutrition of their animals occasioned by the dry season feed scarcity. There is decline in supply and quality of herbage for livestock during the dry season. The concern of Animal Scientists is feed production and utilisation in the dry season to stem the cyclic pattern of weight gain and loss between seasons (Sowande, *et al.*, 2008). Livestock production activities among small scale farmers in the high and medium areas of Africa are integrated with crop production activities (Thairu and Tessema, 1987). The degree of integration varies, but generally intensifies with increasing human population density. Crop production benefits from animals' draught power for tillage, animal manure for fertilisation of crops, while crop residues constitute an important feed resource for animals especially in the dry season (Preston and Leng, 1987). With increasing human population, cropping land is expanding leading to increase production of crop residues. However, this is associated with decreasing land availability for fodder production, thus forcing crop residues to contribute significantly to the livestock feed resources pool. In Nigeria, large quantities of crop residues such as cereal straw and stover, legume crops, straw and hulls, sugar cane tops, cassava leaves and sweet potato vines are left in the field and/or harvested for livestock feeding. However, these crop residues are generally poorly utilised as animal feed each year because small-scale farmers lack the technical knowledge on how best to use them (Methu, 2003).

Farmers generally utilise these crop residues for livestock feeding without considering the use of any of the existing improvement technologies. This situation may be reversed by adapting known technologies that have been developed for local conditions, such as urea treatment, legume supplementation and ensiling process or method. In the presence of a dynamic market system, livestock production could thus be intensified and made profitable for small-scale farmers (Preston and Leng, 1987).

Field observations show that maize stover is the most abundant residue in small holder crop production systems, but poorly handled and stored (Syomiti, *et al.*, 2009). The most commonly observed methods of handling the maize stover are harvesting and either stacking in the field for gradual collection as required for feeding, storing under trees or in the home compound usually in the open and very rarely in roofed barns. This loss of considerable amounts and nutrients is due to weathering and leaf shattering. Improper management and storage methods drastically reduce the proportions of maize stover available as feed as well as the efficiency of utilisation (Promma *et al.*, 1994).

Ensiling has been reported to effectively conserve forages and fodder crops (Babayemi, 2009). The ensiling of crop residues and by-products is a simple and appropriate method of conservation. It is the most-effective way to improve animal feed resources through the national use of locally available agricultural and industrial by-products likely to be available to small scale farmers at village level.

A concrete way of addressing the problem of feeding ruminant livestock in the dry season is using silage or hay. Silage is a sustainable means of supplementing poor quality feed for ruminants in the dry season (Ajayi *et al.*, 2012). Silage making can be considered the most effective way of preserving green forages over hay making, if all essential steps of silage making are followed. Silage making is less dependent on weather. This study was undertaken to document effects of different additives on silage quality characteristics and chemical composition of ensiled maize stover.

3.2 MATERIALS AND METHODS

3.2.1 Experimental Site

The experiment was carried out at the Teaching and Research Farm of the University of Ibadan, Nigeria in August 2010. It is situated in the derived savanna vegetation belt (Latitude $7^{\circ}27'1''N$ and $3^{\circ}45'1''E$) at an altitude between 200m and 300m above sea level; mean temperature of $25 - 29^{\circ}C$ with an average annual rainfall of about 1250 mm. the soils are much drained and belong to the afisol (Rhodic Kandustalf) (Babayemi *et al.*, 2003).

3.2.2 Harvesting and Silage Making

Freshly harvested green maize stovers were collected from Practical Year Training Programme (PYTP) Farm University of Ibadan. Harvesting was done in the month of July 2010 and the samples were collected in batches. Harvested maize stovers were chopped into 3 – 5cm pieces size (for easy compaction) according to t'Mannetje (1999). Cutlass was used for the chopping exercise. Thereafter, the chopped materials were wilted under shade for 24 hours on concrete floor. The chopped maize stover was then weighed, mixed and divided into equal portions (1kg) for the application of four experimental treatments. Each additive was added at 5% level treatment. Treatment A = Maize Stover + Molasses (MSM); Treatment B = Maize Stover + Honey (MSM); Treatment C = Maize stover + Sugar (MSS); Treatment D = Maize Stover only (MS). The control forage was ensiled without additive. All were replicated five times in a completely randomised design. Fermentation period was 30 days as reported by Babayemi (2009).

3.2.3 Experimental Silos

Each of the treatment was ensiled in polythene bags, each capable of holding a 30kg of wilted maize stover were used as silos. Each polythene bag was placed inside a 65 litres capacity plastic basing for reinforcement and ease of fermentation. Ensiling was done by rapid compaction of the material (to eliminate air) into the silos. Sealing of the silos was done by placing a 25kg sandbag on top of the polythene bags after tying carefully and firmly.

3.2.4 Determination of Temperature of the Silages

Silages were opened after 30 days for silage quality assessment. The temperature of the silages was taken by dipping a thermometer inside the silage mass and kept in place for 5 minutes before taken the reading.

Thereafter, 100g of samples were taken from different depths of the silos and mixed to ensure homogeneity of samples. Another 200g was sampled, mixed up thoroughly and kept in the freezer at -4°C for subsequent laboratory analysis.

3.2.5 Evaluation of the Physical Characteristics of Silages

Silage colour was assessed by visual observation and colour chart. The odour/smell and texture of the silage were assessed by a 5-man panel. This step was taken to guide against a bias attitude of a one-man assessor since the involved physical attributes are on the nominal scale rather than ordinal.

3.2.6 pH Determination of the silages

The pH of the silages were determined by taking about 25g of sample from each treatment, mixed with 100ml of distilled water in a beaker for 1 hour and agitated for 2 minutes (Babayemi, 2009). Following this, a pH meter glass electrode was then inserted into the supernatant for 1-2 seconds and pH determined following a standard procedure (AOAC, 1995).

3.2.7 Chemical Analysis

The samples were ground in the laboratory with hammer mill of 1mm sieve and subjected to chemical analysis for determination of dry matter, organic matter, crude protein and nitrogen free extract as described by AOAC (1995). Neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were assayed by the method of Van Soest *et al.* (1991). Hemicellulose was calculated as the difference between NDF and ADF and cellulose as the difference between ADL and ADF (Rinne *et al.*, 1997).

3.2.8 Acceptability study

3.2.9 Experimental Diets

The experimental diets used were ensiled maize stover with or without additives and are indicated as follows:

Diet A - MS + Molasses (MSM)

Diet B - MS + Honey (MSH)

Diet C - MS + Sugar (MSS)

Diet D - MS only (Control) (MS)

3.2.10 Experimental Animal

A total of eight growing West African dwarf (WAD) sheep weighing between (10 – 12kg) were used for the study. They aged between 7 – 8 months as dentition was used to estimate the age.

The animals were purchased from local market at Iwo in Osun state. Animals were subjected to free choice feeding to evaluate acceptability of the ensiled maize stover (MS) prepared with different additives (Diets: A, B, C and D) in a cafeteria feed preference study (Babayemi *et al.*, 2006). The animals were housed together in the group pen in the sheep unit of the University of Ibadan with adequate ventilation. The floor of the house was made of concrete and there were wood shavings on the floor to serve as bedding and also for easy cleaning.

3.2.11 Feeding of Animals

The four silages were introduced on a cafeteria basis to the animals (sheep) in four different wooden feed troughs according to Babayemi *et al.*, (2009), so that each animal had free access to each of the silages in the trough. The plastic feeder (150cm x 60cm) was used to enable the sheep feed simultaneously in a convenient situation. The positioning of silage in a trough was changed daily to prevent bias by an animal from sticking permanently to a trough. The feeding was allowed from 0800 to 1600 hours daily. The intake was measured by deducting the orts or remnants from the amount of feed offered. Daily sample of silage was taken during the fourteen (14) days trial for DM content determination. Silage preference was determined from the coefficient of preference (CoP) value; calculated from the ratio between intake of each individual silage divided by the average intake of the four silage types (Bamikole *et al.*, 2004, Babayemi *et al.*, 2009).

$$\text{Coefficient of preference (CoP)} = \frac{\text{Intake of individual silage}}{\text{Mean intake of the four silage types}}$$

If CoP is < 1, the material is poorly accepted and when > 1, the material is well accepted. (Karbo *et al.*, 1993, Bamikole *et al.*, 2004).

3.3 Statistical Analysis

The experimental design used was Completely Randomised designed (CRD) and data collected were subjected to analysis of variance (ANOVA) using procedure of SAS (2003). The significant means were separated by the use of Duncan (1995) Multiple Range F-test.



Plate 1: Maize crop (Magnification x 1)

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Plate 2: Chopping Exercise of green maize stover (Magnfication x 1)



Plate 3: Chopped Maize Stover (Magnification x 1)

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3.4 RESULTS

Figure 1 shows temperature development in prepared silages. The temperature ranged from 26⁰C in treatment B (Maize Stover + Honey) to 26.3⁰C in treatment C (Maize Stover + Sugar). There were no significant ($P > 0.05$) differences among the treatments. However, no particular trend was established between the no “additive” silage (Control) and the three others with different additives: MSM, MSH, and MSS. The physical qualities of the ensiled maize stover as reflected in terms of colour, texture and odour are shown in Table 2. The colour of MS silage (control) and MS silages with additives varied from greenish-brown to greengage (greenish yellow). In terms of texture, all the silages were firm. Also the odours were similar among the four treatments as the silages were characterised with fruity, alcoholic and pleasant odour. Figure 2 shows the pH of the silages. The pH ranged from 3.5 in treatment B (MSH) to 3.7 in treatment C (MSS) silages.

Dry matter, Crude Protein (CP), Crude Fibre (CF), ash, Ether Extract (EE) and Nitrogen Free Extract (NFE) of the ensiled maize stover (MS) and unensiled are presented in Table 3. The dry matter ranged from 31.1 in MS unensiled to 35.3% in MSS silage. Crude protein ranged from 7.9% in (MSH) to 9.3% (MS-fresh/unensiled). Crude fibre (CF) ranged from 30.0% in (MSH) to 32.3% (fresh MS). Similarly, ash ranged from 6.3% in (MSH) to 7.4% (fresh MS). Also, EE ranged from 1.4 in MS silage (control) to 1.8 in (MSS) silage and MS fresh (unensiled). All the CP, CF and EE were similar across the treatments, however ash was significantly ($P < 0.05$) different such that it was lowest in MSH (6.3%) silage and highest in fresh/unensiled MS (7.4%). NFE ranged from 51.0% (MS fresh) to 54.3% in (MSH) silage. Nitrogen free extract (NFE) of MSH was significantly ($P < 0.05$) different from other treatments.

The fibre detergent fractions of ensiled maize stover were presented in Table 4. All the fractions were similar among the treatments. The NDF (total cell walls) ranged from 68.6% (MSS) to 69.9% (MSM). While the ADF (lignocelluloses) ranged from 56.1% (MSS) to 63.2% (MSH), the ADL ranged from 14.0% (MSM) to 16.8% (MSS). All the three silages with additives and control were somehow similar in contents of NDF, ADF and ADL but MSM was highest in NDF while MSH was highest in ADF. Also MSS was highest in ADL. Comparing the MS (fresh) and MS (control), the latter was higher in NDF and ADF while the former was higher in ADL.

Hemicellulose and cellulose values ranged from 10.81 -12.49% and 39.30-44.81% respectively. The hemicellulose composition of fresh/un-ensiled and silages were similar across the treatments. However, the cellulose composition was significantly ($P < 0.05$) different across the treatments such that it was highest in MSM and lowest in MSS.

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Table 2: Moisture content (%) and physical characteristics of the ensiled maize stover.

Silage treatments	Quality indicators			
	Moisture	Colour	Texture	Odour/smell
MS only (Control)	68.2	Greenish brown	Firm	Fruity
MS + Molasses	68.7	Greengage	Firm	Pleasant
MS + Honey	67.3	Greengage	Firm	Alcoholic
MS + Sugar	64.7	Greengage	Firm	Alcoholic

MS – Maize Stover

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Table 3: The proximate composition of ensiled maize stover

Nutrient	Unensiled	Control	MSM	MSH	MSS	SEM
Dry matter	31.1 ^b	31.6 ^b	31.3 ^b	32.7 ^{ab}	35.3 ^a	0.50
Crude protein	9.3 ^a	8.4 ^a	8.3 ^a	7.9 ^a	8.6 ^a	0.46
Crude fibre	32.3 ^a	31.9 ^a	30.0 ^a	30.0 ^a	31.4 ^a	0.55
Ash	7.4 ^a	7.1 ^a	7.2 ^a	6.3 ^b	6.9 ^a	0.11
Ether extract	1.8 ^a	1.4 ^a	1.7 ^a	1.5 ^a	1.8 ^a	0.69
Nitrogen free extract	49.4 ^b	51.2 ^{ab}	52.9 ^a	53.7 ^a	51.5 ^{ab}	0.58

a,b = Means on the same row with different superscripts are significantly ($P < 0.05$) different

Control = Ensiled Maize stover without additives.

MSM = Ensiled Maize stover + Molasses

MSH = Ensiled Maize stover + Honey

MSS = Ensiled Maize stover + Sugar

SEM = Standard Error of Means

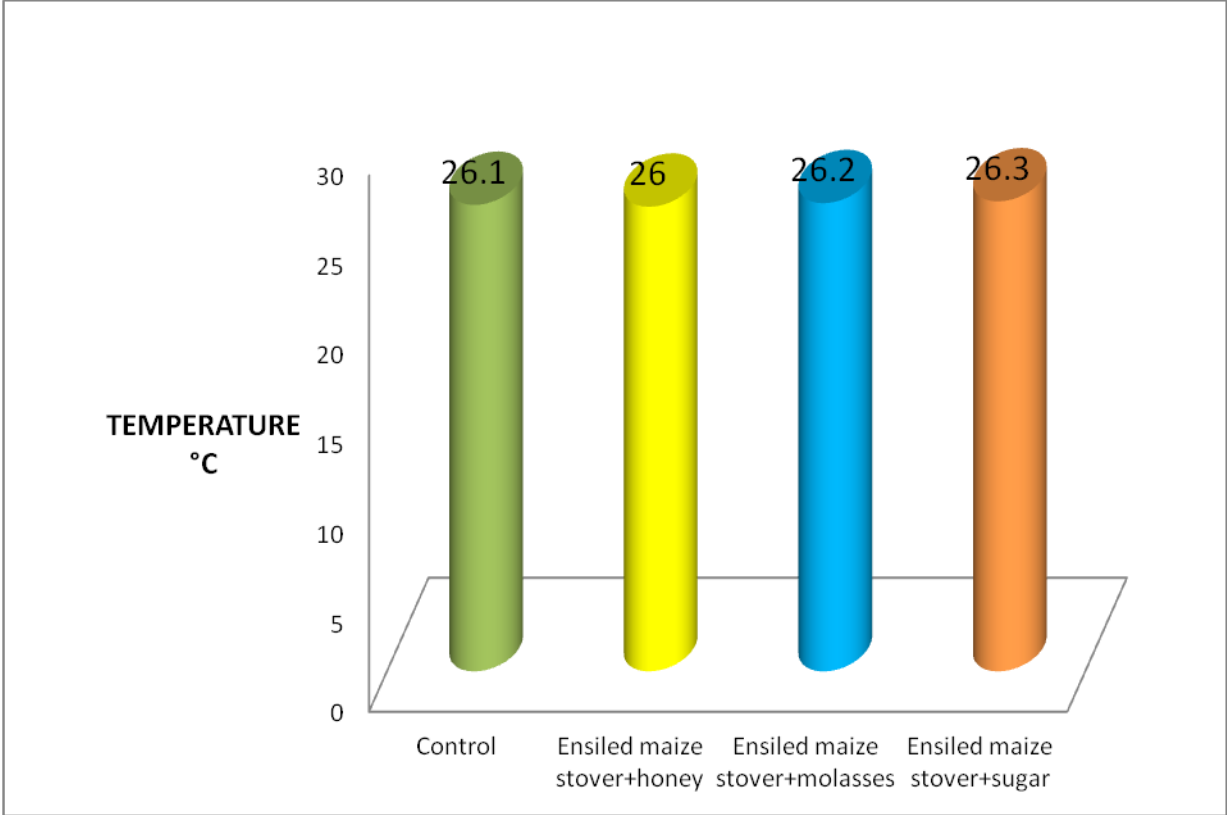


Figure 1: The temperature of ensiled maize stover

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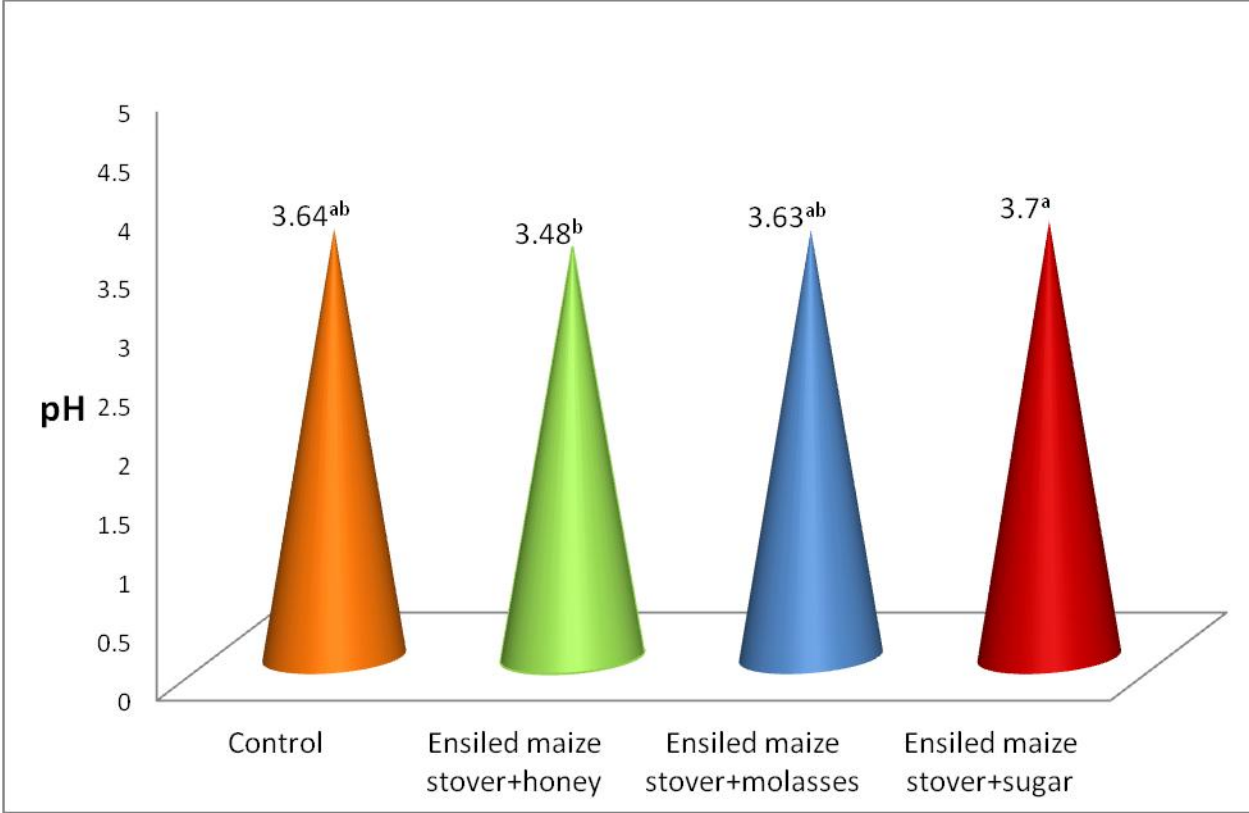


Figure 2: pH of ensiled maize stover

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Table 4: Fibre fractions (%) of ensiled maize stover

Nutrient	Unensiled MS	MS only (control)	MSM	MSH	MSS	SEM
NDF	69.20 ^a	69.30 ^a	69.90 ^a	69.60 ^a	68.60 ^a	0.63
ADF	57.50 ^a	58.50 ^a	59.50 ^a	63.20 ^a	56.10 ^a	1.66
ADL	16.50 ^a	15.60 ^a	14.00 ^a	14.80 ^a	16.80 ^a	0.11
Hemicellulose	11.73 ^a	10.81 ^a	10.92 ^a	11.39 ^a	12.49 ^a	0.58
Cellulose	41.00 ^{ab}	42.97 ^{ab}	44.81 ^a	43.44 ^{ab}	39.30 ^b	0.92

a, b = Means on the same row with different superscripts are significantly ($P < 0.05$) different

MS = Maize stover

Control = Ensiled sole maize stover

MSM = Maize stover + Molasses

MSH = Maize stover +Honey

MSS = Maize stover + Sugar

SEM = Standard Error of Means

The coefficient of preference of ensiled maize stover with or without additives fed to WAD sheep is shown in table 5. In this study, MSS recorded the highest CoP value of 1.03 compared to other silages followed by MS (control) 1.02, while the remaining two silages had values of 0.998 for MSM and 0.95 for MSH, which was less than unity.

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Table 5: Dry Matter Intake and Coefficient Preference of WAD Sheep fed ensiled Maize Stover

Silage type	Mean daily consumption of animals (g/DM)	Coefficient of Preference
MSM	541.32	0.998
MSH	517.13	0.95
MSS	558.21	1.03
MS (control)	553.35	1.02

MSM - Maize stover + Molasses

MSH - Maize stover + Honey

MSS - Maize stover + Sugar

MS - Maize stover

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3.5 DISCUSSION

3.5.1 Temperature Range of Silages

The temperature of fermenting forage varying from 27 – 38⁰C was presumed to produce excellent silage (Muck, 1996). The temperature range of 26.0 to 26.3⁰C of MS silages was within 25 – 27.5⁰C obtained by Babayemi (2009) in silage of Guinea grass fed to WAD sheep. This temperature range would appear to be the operating temperature for normal silage fermentation. As observed by Bolsen *et al.* (1996), any excessive heat production can result in Maillard or browning reactions which can reduce the digestibility of both protein and fibre constituents. The useful proteins form complexes with carbohydrates and thereby making them less digestible. Although “heat damaged“silage may be palatable, but part of the protein and some of the energy it contains will be unavailable to livestock (Adesogan and Newman, 2010). Furthermore, temperature is one of the essential factors affecting silage colour. The lower the temperature during ensilage, probably the less will be the colour change (Adesogan and Newman, 2010). If the temperature obtained for the present silage was above 30⁰C or 45 – 60⁰C, they would have become yellowish brown to brown or dark brown colour with tobacco-type smell due to caramelisation of sugars in the silages (McDonald *et al.*, 1995; Jianxin, 2002, Babayemi, 2009). However, in this study, the range (26.0-26.3⁰C) obtained during fermentation period (30 days) was in agreement with the temperature of 26.5⁰C reported by Menesses *et al.* (2007) for artichoke by-product ensiled for 50 days.

3.5.2 Physical Characteristics of Silages

The greenish brown and greengage colour, firm texture, alcoholic, pleasant and fruity odour given by ensiled MS without additives (control) and those with additives are characteristics of a good fermented silage. Chemical changes during ensilage process can affect the colour of forage and the fermentation acids can convert chlorophyll into the brown magnesium-free pigment phacophytin. Consequently, the greenish brown colour developed in the silages was probably caused by the action of organic acids on chlorophyll pigment of maize stover which was converted to phacophytins, as a magnesium free derivative of chlorophyll. Good silage usually preserves the original colour of the forage used to produce it (t'Mannetje, 1999). Consequently the brightly coloured expected. The greenish brown and greenage colour obtained in this study were in order. The greenish brown and greengage colour were closed to the original colour of the

maize stover which was an indication of good quality silage that was well preserved. The firm texture, observed in this study expected to be the best texture and pleasant and fruity smell is accepted for good silage as reported by Kung and Shaver cited by Babayemi (2009). The use of additive, especially soluble carbohydrate additives (e.g molasses, maize grain) in making maize stover base silage may not be necessary since the MS (control) without additive possessed good quality characteristics similar to those with additives.

3.5.3 pH of Silages

Generally, pH is one of the simplest and quickest ways of evaluating silage quality. The lower the pH, the better preserved and more stable is the silage. However pH may be influenced by the moisture content and the buffering capacity of the original materials. Silage that has been properly fermented will have a much lower pH (be more acidic) than the original forage. The pH value of good silage below 4.4 has been considered as an index of good silage (Harrison *et al.*, 1995). Similar results reported by Eweedah (2005) indicated that good quality silage made from corn stover has pH value of 4.1. The pH value of 3.5 to 3.7 of silages was slightly lower than 4.1 reported by Eweedah (2005), 4.5 – 5.5 by Menesses *et al.*, (2007), 4.2 – 5.0 by Babayemi (2009) and 4.3 – 4.7 by Kung and Shaver (2002). However the pH values for both control and silages with additives in the present study were in agreement with 3.2 – 3.8 reported by Oduguwa *et al.*, (2007) and 3.11 – 3.62 by Fasina (2012) for well preserved and good quality silage. The low pH range in silages reported in this study is an indication of proper fermentation and good qualities. The observation made here is further evidence that maize stover can be ensiled without additives and produce good silage. This is because the stem is rich in water soluble carbohydrates (natural sugars) and ferment more easily, but majority of other forages will require one additive or combination of additives for easy fermentation (Bolsen *et al.*, 1996).

3.5.4 Chemical Composition of Silages

Silage prepared with additives had dry matter, (DM) of 31.3 – 35.3% as compared to that without additive having 31.6% and unensiled maize stover having 31.1%. Dry matter of silages with and without additives was consistent with 30 – 35% reported by FAO (2010) and lower than 37.02% obtained by Elkholy *et al.* (2009). However, it was higher than a range of 21.2 – 28.46% obtained by Moran (2005), Ashbell and Weinberg (2002) and Altaf un-Rahman and Ancela (2004). The differences could be as a result of difference in additives used. No obvious change

was observed between dry matter of fresh/unensiled maize stover (31.10%) and control (31.60%) which indicates that the silage was well preserved with no loss of dry matter. It was observed that the dry matter content of the silages had been drastically increased from 31.10% in the fresh/unensiled maize stover or from 31.60% in the maize stover without additive (control) to 35.30% in the maize stover and sugar (MSS). Chemical composition of ensiled maize stover showed the decreased in crude protein content of ensiled maize stover with and without additive but the difference was minimal. This is similar to a report made by Idris *et al.*, (1999) that corn stover after harvest of matured cobs at 75 days of growth contained 9.60% CP and after ensiling the value decreased to 8.20%.

Crude protein (CP) content (9.30%) obtained for fresh/unensiled is consistent with the 9.6% established by Idris *et al.*, (1999). The crude protein in the silages ranged from 7.9 in the MSH to 8.60% in MSS. The crude protein of ensiled maize stover with or without additives is relatively low. This is in agreement with the report of Adegbola (1998), who observed that cereal stovers and straws, which form the bulk of crop residues, are inherently low in crude protein. The decreased in crude protein in this study could also be due to dilution effects as all additives are carbohydrates. Ajayi *et al.*, (2011), Moran (2005), Elkoly *et al.*, (2009), Altaf-ur-Rahman and Ancela (2004), Nour *et al.* (1987), Idris *et al.* (1999), Fomunyan and Meffeja (1986) reported CP of 5.12, 5.70, 8.99, 8.76, 8.20, 5.40 and 2 – 80% respectively. The crude protein and dry matter obtained in this study compare well with those of some tropical grasses and could be used to replace them in feeding animals. Levels of crude protein are within the critical value of 7.0% or 70g/kg recommended for small ruminants by NRC (1981) but lower than minimum protein requirement of 10 – 12% recommended by ARC (1985) and Gatenby (2002) for ruminants. Since maize stover is available throughout the entire regions in Nigeria and some part of the World, it is expected to have a great potential towards the alleviation of feeding problem usually encountered during the dry season and therefore maize stover silage could bridge the gap between forage availability in the rainy season and unavailability during the dry season. Furthermore, it is important to note that fermentation had no effect on boosting the crude protein level of fermented silage since the objective of the present study was not to increase the crude protein but to make forage available at all time. However, this result is in conflict with the result obtained by Altaf-ur-Rahman and Ancela (2004) where CP content of ensiled maize stover

increased from 7.70 (fresh) to 8.76%. This was attributed to microbial protein synthesis during ensilage. Phipps and Wilkinson (1985) also found increased in CP content and decreased in DM and sugar content in maize stover silage. The objective of making the silages was not to alter chemical composition but to make forage available at all times.

The crude fibre content of maize stover decreased from 32.30% (fresh) to 30.00%, after ensiling. This may be due to the degradation action of microbes especially fungi on fibre component of the ensiled materials during fermentation process. The crude fibre value of 30.00 to 32.30% in this study is slightly lower than 36.30 – 36.65% reported by Ajayi *et al.* (2011) and ANL (1999) 35.81% but higher than values reported by Moran (2005) 26.4% and Elkholy *et al.* (2009) 25.03%. However, the value obtained in this study was within the range value of 28 – 46% reported by Fomunyan and Meffeja (1986) and similar to 32.1% what was obtained by Nour *et al.* (1987). Generally, the crude fibre level obtained in this study is within the range of that of grasses and sorghum stover.

Mean ash represents inorganic matter which mainly includes plant minerals. The ash content of the fresh/unensiled maize stover was 7.4% while the level in the silages ranged from 6.3 – 7.2%. The level of ash in the fresh plant is within the range 3.05 – 7.70 obtained by Ajayi *et al.* 2011, Altaf-un-Rahaman, and Ancela (2004). As for the silages, Elkholy *et al.* (2009) reported 6.99%, Altaf-un-Rahaman (2004), Nour *et al.* (1987). Fomunyan and Meffeja (1986) reported 8.69%, 8.51% and 9 to 15% respectively. Whatever variation reported in the literature as values might be connected with the age of the plants, period of the year of collection, the nutrient level of each maize stover, fermentation length of the silage and the different additives used for fermentation. Maize stover may differ in chemical constituents from one period to another and from one place to another in a year due to variation in soil nutrient composition which has a direct correlation or relationship with the plant nutrient composition. The ash obtained in this study compared favourably with normal ash of legume grass forage of 7 – 9% and sorghum stover.

The ether extract in maize stover ranged from 1.4 to 1.8% in the silages. The level in the fresh maize stover was 1.8. Analysing samples of maize stover, Elkholy *et al.* (2009), Ajayi *et al.* (2011), Altaf-ur-Rahaman and Ancela (2004), Fomunyan and Meffeja (1986), and Nour *et al.*

(1987) reported 2.4, 0.82, 1.57, 1.0 – 2.0 and 1.43 % respectively. The range value obtained in this study generally agreed with those of these authors. The low level of ether extract may be due to fibrous nature of plant material being a crop residue, is characterised with low level of nutrients and ether extract is among the nutrients and is therefore affected.

For nitrogen free extract (NFE) of maize stover silages, Fomunyan and Meffeja (1986) obtained a ranged value of 35 – 53% which is comparable to the range of 51.20 – 53.70% obtained in the silages or the 49.40% obtained in the fresh materials in this study. Furthermore, in the ensiled material, Elkholy *et al.*, (2009), Ajayi *et al.* (2011), Nour *et al.* (1987), Altaf-un-Rahman and Ancela (2004) obtained 56.58, 60.09, 51.7 and 55.26% respectively. The nitrogen free extract level of the silages in this study is within the range of common grasses and sorghum stover. It has been shown that corn-stover can be successfully ensiled without any significant change in its nutritive value (Soliman *et al.*, 1975 and 1977).

3.5.5 Fibre Fractions

The fibre fractions (NDF, ADF, and ADL) have implication on digestibility. The neutral detergent fibre (NDF), which is a measure of the plants cell wall contents, is the chemical component of feed that determines its rate of digestion. Neutral detergent fibre (NDF) is inversely related to the plants digestibility (McDonald *et al.*, 1995, Gillespie, 1998). The higher the NDF the lower the plants digestible energy. NDF is correlated with the level of dry matter intake by cows; the lower the NDF, the higher the level of intake. The neutral detergent fiber (NDF) level (68.6 – 69.2%) obtained is higher than 48% reported by Elkholy *et al.* (2009). High NDF could result in low intake while high ADF may engender low digestibility (Babayemi *et al.*, 2010). The acid detergent fibre (ADF) consist mainly lignin and cellulose. Acid detergent fibre (ADF) is correlated with the digestibility. Acid detergent fibre (ADF) level of 56.1 – 63.2% obtained was higher than 29.0% reported by Elkholy *et al.* (2009) and 38% and 39%, reported for Guinea grass and *Andropogon gayanus* by Odedire and Babayemi (2008) respectively. Acid detergent lignin (ADL) of a plant is the most indigestible component of the fibre fraction (Gillespie, 1998), and its amount will also influence the plant digestibility. Lignin is generally accepted as the primary component responsible for limiting the digestion of forages (Van Soest, 1994; Traxler et

al., 1998; Agbagla-Dohnani et al., 2001). The ADL level of 14.0 – 16.80 % obtained in this study was within the values of 14 and 17 % reported for Guinea grass/lablab mixture by Alasa *et al.*, (2010) but higher than 7.60 and 9.87 % reported for Guinea grass ensiled for 47 days by Babayemi (2008). Since fibre fractions (i.e. NDF, ADF and ADL) content of the silages were relatively high, the intake and potential digestibility will be low when feed alone to ruminants without concentrate supplements. According to Van Soest (1994), forage digestibility in ruminants is constrained by the extent of cell wall (NDF) digestion. Furthermore, it is the most important factor affecting the total diet organic matter digestibility (Nousiainen et al., 2004) and it has influence on animal performance (Oba and Allen, 1999). The high level of fibre fractions observed in this study may be attributed to the age or maturity at harvest and leaf to stem- ratio of the ensiled maize stover.

The hemicellulose and cellulose are cell wall constituents and polysaccharides. They are very indigestible in monogastrics but digestible in ruminants through fermentation by rumen microbes. Hemicellulose values obtained for silages ranged from 10.81 – 12.49% while cellulose ranged from 39.30 – 44.81%. These values are not too high for ruminants due to the nature of their stomach and the presence cellulolytic bacteria and fibrolytic fungi in the rumen. According to McDonald et al. (1995), ruminants can be fed sole on feed that contained 40% cellulose and 20% hemicellulose. McDonald et al. (1995), further stated that in mature herbage, and in hay and straw, the proportion of cellulose and hemicellulose is much higher and that of water soluble carbohydrates is much lower, and that all the carbohydrates but not lignin are attacked by the rumen microorganisms. This is suggesting that ruminants can cope with feed that contain relatively high level of cellulose and hemicellulose with the aid of rumen microbes.

3.6.0 Acceptability

Acceptability or free choice intake attributes of a feed connotes the actual response of an animal to a particular feed and the possible visual effects of the feed to the animal. This conversely depicts the efficiency of the feed in the rumen (Van Soest, 1993). There are many ways of assessing the nutritive value of feeds for ruminants; the direct intake by the animals is the best method. Free choice intake or acceptability study of a feed is a quick assessment of the physical quality of the feed by the animal. It is one of the *in vivo* trials that reveal the actual reaction of

animals to a feed. Coefficient of Preference (CoP) is a direct measure of acceptability and nutritional capabilities of a feedstuff. In recent times, cafeteria techniques have been used to assess the acceptability of some forage (Bamikole *et al.*, 2004, Babayemi *et al.*, 2006; and Babayemi, 2007).

In this study, the mean dry matter and coefficient of preference (CoP) by sheep placed on ensiled maize stover (MS) with and without additives are indicated. The CoP varied from 0.95 to 1.03 DM/day with the order of preference as; MSS > MS (control) > MSM > MSH. In other words, MSS silage and MS (Control) silage with CoP ranging above unity were accepted or preferred. Conversely, MSM and MSH i.e. silages with molasses and honey additives with CoP values of 0.998 and 0.95 each were not accepted. There are number of factors that may influence acceptability of feed by small ruminants. Plant physical structure and chemical composition are the most important factors that influence preference (Babayemi *et al.*, 2009 and VanSoest, 1994). Generally, acceptability, seasonal variation and availability are some factors that influenced feed intake by an animal.

In studies to predict voluntary intake from silage fermentation characteristics following authors (Rook and Gill, 1990; Rook *et al.*, 1990; Steen *et al.*, 1998; Wright *et al.*, 2000; and Seglar 2003) found moderate correlation between fermentation acids and voluntary intake. Seglar, (2003) reported that, it is the combination of high acetic acid levels, along with the presence of alcohol, and methyl-acetates that probably is really causing feed refusal. Therefore, the alcoholic odour imposed on the MSM and MSH feed by fermentation due to conversion of sugars to alcohol by yeast probably caused a shift in acceptability by sheep from these silages (Seglar, 2003 and Bolsen *et al.*, 1996). It is therefore concluded that silage of good quality characteristics can be produced from maize stovers without additives by wilting under shade for 24hrs. On dry matter basis, maize stovers as any other stovers contain protein, ether extract and mineral matter as many conventional forages. Fibre values are usually relatively higher than for common forages.

CHAPTER FOUR

EXPERIMENT THREE

4.0 *IN VITRO* FERMENTATION CHARACTERISTICS AND DRY MATTER DEGRADABILITY OF ENSILED MAIZE STOVER

4.1 Introduction

A major constraint to livestock production in developing countries is the scarcity and fluctuating quantity and quality of the year round feed supply. Providing adequate good quality feed to Livestock to raise and maintain their productivity is and will be a major challenge to agricultural scientists and policy makers all over the world. Increase in population and rapid growth in the world will lead to increase in demand for animal products; an increase of approximately 30% in both meat and milk production is expected in the coming 20 years (Makkar, 2004). At the same time, the demand for food crops will also increase. Future hopes of feeding the millions and safeguarding their food security will depend on the enhanced and efficient utilisation of unconventional resources, which can not be used as food for humans, as feed for livestock (Makkar, 2004).

In developing countries, livestock are fed mainly on crop residues and agro-industrial by-products containing a larger proportion of ligno-cellulosic feeds like cereal straws, stovers, sugarcane by-products and similar other feeds. These feed are poor in protein, energy, minerals and vitamins. Addition of foliage from tree or supplementation with seed meals, ensiling or even urea can improve the utilisation of low quality roughages mainly through the supply of nitrogen to rumen microbes. The use of simple techniques for evaluation of the nutritional quality of these feed resources will contribute to their efficient utilisation.

Both growth and milk yield of ruminants are largely limited by forage quality which is mainly reflected in low voluntary intake and digestibility. The importance of these parameters in animal nutrition has long been recognised. The determination of intake and digestibility of feedstuffs *in vivo* is time-consuming, laborious, expensive, requires large quantities of feed and unsuitable for large scale feed evaluation. Therefore many attempts have been made to predict intake and digestibility using laboratory techniques. Much efforts has been directed towards the development of regression equations to predict digestibility from forage chemical composition,

but a regression equation that satisfactorily predicts a wide range of forages has not yet been derived. Gas production reflects all nutrients fermented, soluble as well as insoluble; and fractions that are not fermentable which do not contribute to gas production. Furthermore, the kinetics of fermentation can be obtained from a single incubation, allowing the rate of fermentation to be calculated. Gas measurement is a direct measure of microbial activity and can be a better index of forage ME content than an indirect *in vitro* measured based on nutrients (Makkar, 2004).

The objective of this study was to evaluate the *in vitro* fermentation characteristics and dry matter degradability of ensiled maize stover with and without additives.

4.2 MATERIALS AND METHODS

This experiment was carried out as described by Ørskov and McDonald (1979).

4.2.1 Experimental Design

The experimental design used was Completely Randomised Design (CRD).

4.2.2 Experimental Site

The experiment was carried out at the Department of Animal Science, Ruminant Laboratory, Faculty of Agriculture and Forestry, University of Ibadan, Ibadan,

4.2.3 Gas Production Apparatus

The following apparatus were used during the *in vitro* fermentation of the samples.

50ml long plastic syringes (piston pipettes) calibrated to 100ml into capillary attachment.

- Silicon rubber tube about 4.5cm long steel clips.
- Analytical balance.
- Incubator
- Rubber tube/hose
- Carbon dioxide cylinder with regulator
- Thermometer
- Stirrer
- Beakers
- Thermos flask
- 100ml plastic syringe

- Plastic bucket and cheese cloth

4.2.4 Preparation and Weighing of the Feed Samples

Samples of each silages (from A, B, C and D) were oven dried at 105⁰C until constant temperature was attained. The dry samples were milled using hammer mill of 1mm sieve. 200mg each of the dried samples were weighed carefully into the cylinder of the syringe. Each syringe was marked with numbers e.g. 1, 2, 3 etc. Each of the samples was replicated 3 times and the blank which contained no substrate. The piston was greased with Vaseline to ensure easy movement and precise fitting was then pushed down the cylinder gently to avoid thrusting out of the sample through the cylinder tube. The silicon rubber tube attached to the capillary attachment (needle) of the syringe was then closed with a plastic clip. Fermentation was carried out in this plastic syringe.

4.2.5 Collection and Preparation of the Rumen Fluid

The rumen fluid was collected prior to the early morning feeding. The rumen fluid was collected through the suction method by means of the hose from three West African Dwarf (WAD) goats under the same feeding regime. The animals were fed with 40% concentrate feed (40% corn, 10% what offal, 10% palm kernel cake, 20% groundnut cake, 5% soybean meal, 10% dried brewers grain, 1% common salt, 3.75% oyster shell and 0.25% fish meal) and 60% Guinea grass. The fluid was collected into a thermos flask and taken to the laboratory. It was filtered through a four-layered cheese cloth into a warm flask, flushed with carbon dioxide (CO₂) gas, and stirred using an automatic stirrer.

4.2.6 Preparation of the Buffer Solution

The buffer solution prepared was the McDougall's buffer which consisted of Sodium bicarbonate (NaHCO₃) Sodium phosphate dibasic (NaHPO₄) Potassium chloride (KCl), Sodium chloride (NaCl). Magnesium sulphate (MgSO₄.7H₂O) and Calcium chloride (CaCl₂. 2H₂O). The buffer solution was freshly prepared and stored in a dark bottle.

The following were the quantity of reagents used in preparing the buffer.

McDougall's Buffer

	Compound	gm/litre
Sodium bicarbonate	(NaHCO ₃)	9.8
Sodium phosphate dibasic	(Na ₂ HPO ₄)	2.77
Potassium chloride	(KCl)	0.57
Sodium chloride	(NaCl)	0.47
Magnesium sulphate	(MgSO ₄ .7H ₂ O)	0.12
Calcium chloride	(CaCl ₂ .7H ₂ O)	0.16

The reagents were dissolved in distilled water. The calcium chloride was added only after the other reagents were completely in solution. Prior to use, a volume of buffer was warmed at 30°C and reduced with a stream of CO₂. During the warming and reducing step, urea was added to the McDougall's buffer at the rate of 1.0 gm/litre.

4.2.7 Preparation of the Rumen Liquor-Buffer Solution

The rumen liquor-buffer solution was mixed in the ratio 1: 4 (v/v) for this incubation.

4.2.8 Preparation of the Syringes for Incubation

The plastic syringes containing the substrates were placed inside the incubator at 38 – 39°C an hour before incubation started. The steel clips on the syringes were loosened gently to allow for inoculation of the syringes; 30ml of the rumen fluid-buffer, mixture was pipetted with a syringe into each of the pre-warmed plastics syringes.

The zero time (i.e the time when injection of the rumen: buffer mixture was introduced into the syringes) was stated and the incubation time was recorded. The rumen liquor buffer mixture is called an inoculum. The level of the pistons of the syringes and the time when the filling of the syringes with the rumen liquor buffer mixture was finished were recorded. The syringes were then incubated at 39 ± 1°C in an incubator. The blanks contained only 30ml of the rumen-buffer solution.

4.2.9 Duration of Incubation

The incubation lasted for 24 hours and gas production was recorded at 3-hours intervals. The gas produced was read by measuring the head space formed between the top of the piston and the liquid in the syringe.

The gas produced was recorded as the gas produced (in ml) at 24 hours of incubation. After every reading (i.e 3 hours interval), the content in the syringes was shaken properly to allow mixing of the substrate and the liquid.

4.2.10 Methane Gas Determination

10 Molar Sodium hydroxide was prepared and introduced into the syringe at the end of the 24 hours incubation. 4ml of the 10MNaOH solution was introduced through the silicon tube fitted to the tips of the syringe after opening the steel clip. The NaOH introduced absorbed CO₂ gas contained in the syringe leaving methane gas. When the NaOH solution was introduced into each of the syringe; the clip was immediately tightened back to prevent escape of CO₂ gas. They were shaken to allow proper mixing and absorption of the CO₂ gas. After all the CO₂ gas was absorbed, the volume of methane gas left in each of the syringes was recorded in ml.

4.2.11 Calculations

The average volume of gas produced from the blanks was subtracted from the volume of gas produced from each sample. This gave the net gas produced (GP) for each sample.

Graphs of the volume of gas produced every 3-hour interval of the 3 replicates of each sample was plotted against the incubation time.

From the graph, the degradation characteristics were estimated as defined in the equation:

$Y = a + b(1 - e^{-ct})$ (Ørskov and McDonald, 1979) where

Where Y = gas volume production at time (t)

a = gas produced from the soluble fraction

b = gas produced from insoluble but degradable fraction

c = rate of gas production

t = incubation time

The asymptote (a+b) represents the potential gas production. The intercept of the curve is represented by “a” and given the gas production value at zero hour. The “b” value was calculated as the difference between the asymptotic (a+b) and the intercept “a” i.e. (a + b) – a.

To calculate the rate of degradation the above equation was transformed

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

$$Y = \left[\begin{array}{c} a + b \\ - b \end{array} \right] = e^{-ct}$$

Note $Y^t = e^{-ct}$

Take the natural logarithmic

Derivative of both sides

In $Y^t = -ct$

Hence $C = \frac{\ln Y^t}{-t}$

To get good estimates of (c), Y was selected (i.e. DMD% at time) when the curve changed rapidly. The gas produced on incubation of 200mg feed DM after 24 hours of incubation together with the levels of other chemical constituents was used to predict digestibility of organic matter determined *in vivo* and metabolisable energy as proposed by (Menke et al., 1979)

$$ME \text{ (M1/kg DM)} = 2.20 + 0.136 \text{ GP} + 0.057 \text{ GP} + 0.0029 \text{ CF}$$

$$\text{OMD (\%)} = 14.88 + 0.889 \text{ GP} + 0.45\text{CP} + 0.0651\text{XA}$$

$$\text{SCFA} = 0.0239\text{GV} + 0.0601$$

Where ME is the metabolisable energy;

OMD = Organic matter digestibility

CP = Crude protein in %

CF = Crude fibre in %

XA = Ash in % and

GP = The net gas production in ml from 200mg dried sample after 24 hours of incubation and after correction; for the day to day variation in the activity of rumen liquor using the Hohenheim standard. The short chain fatty acids were also calculated. The relationship between SCFA production (mmol) and gas volume (ml) after 24 hours of incubation was (Getachew *et al.*, 2000) in the absence of PEG.

- SCFA - Short chain fatty acids
PEG - Polyethylene glycol
GAS - Net gas production in ml from 200 mg dry sample after 24 hours of incubation.

4.3 Statistical Analysis

Data collected were subjected to analysis of variance (ANOVA) procedure using SAS package of (1999). Significant means were separated by Duncan multiple range test of the same package (Duncan, 1955).

4.4 RESULTS

4.4.1 *In vitro* gas Fermentation parameters of ensiled maize stover at 24hrs of incubation

The *in vitro* gas fermentation parameters of ensiled maize stover with or without additives at 24hrs are presented in Table 6 and significant ($P < 0.05$) differences were obtained among the silages in all the parameters observed at 24hrs. The total gas volume (TGV) ranged between 31.67 and 41.00 ml. However, TGV value (44.68ml^3) of T_1 (unensiled/fresh maize stover) was significantly higher than T_3 and T_4 but similar to T_5 and T_2 (control) silage. For methane (CH_4), the range value was 16.09 – 28.36%. T_1 was significantly higher but similar to T_5 while T_2 , T_3 and T_4 were not significantly different. Organic Matter Digestibility (OMD %) of silages ranged from 51.44 to 59.66% while the fresh (T_1) was 63.55%. Treatments 1, 2 and 5 were similar statistically while treatments 3 and 4 were significantly lower than T_1 . A similar trend was observed for metabolisable energy (ME). The Metabolisable Energy (ME) ranged between 7.07 and 8.90 (MJ/kg DM). The short chain fatty acid (SCFA) mean values were similar across the treatments except treatment 4. Furthermore the SCFA mean values were similar among the silages. The mean values of SCFA ranged from 0.70 to $1.01\mu\text{ml}$. The Dry Matter Degradability (DMD %) of treatments 1, 2 and 3 were similar statistically while 4 and 5 were significantly different ($P < 0.05$). The ranged value for DMD was 40.00 – 70 %. The highest value (70%) was recorded for T_2 (control) while the least value (40%) was recorded for T_4 as shown in Table 6.

Table 6: *In vitro* fermentation parameters of maize stover at 24hrs incubation period

Parameters	Treatments					SEM
	T1	T2	T3	T4	T5	
TGV (ml)	44.68 ^a	39.33 ^{ab}	31.67 ^c	36.00 ^{bc}	41.00 ^{ab}	1.75
CH ₄ (%)	28.36 ^a	16.09 ^c	17.90 ^c	22.22 ^b	25.20 ^{ab}	0.43
OMD (%)	63.55 ^a	58.18 ^{ab}	51.44 ^c	54.83 ^{bc}	59.66 ^{ab}	3.2
ME (MJ/Kg DM)	8.90 ^a	8.11 ^{ab}	7.07 ^c	7.64 ^{bc}	8.36 ^{ab}	0.18
SCFA (μml)	1.01 ^a	0.88 ^{ab}	0.88 ^{ab}	0.70 ^{bc}	0.92 ^{ab}	0.03
DMD (%)	53.33 ^{ab}	70.00 ^a	60.00 ^{ab}	40.00 ^c	46.67 ^{bc}	3.29

a, b, c Means along the same row with different superscripts are significantly ($P < 0.05$) different

TGV = Total Volume Gas, CH₄ = Methane, ME = Metabolisable Energy, OMD = Organic Matter Digestibility, SCFA = Short Fatty Acid, DMD = Dry Matter Degradability.

T₁ = Unensiled Maize Stover

T₂ = Ensiled Maize Stover Only (Control)

T₃ = Ensiled Maize Stover and Molasses (MSM)

T₄ = Ensiled Maize Stover and Honey (MSH)

T₅ = Ensiled Maize Stover and Sugar (MSS)

SEM = Standard Error of Means

4.4.2 *In vitro* gas fermentation characteristics of ensiled maize stover at 24hrs incubation period

The potential gas production, gas production from the insoluble fraction, extent and rate of gas production, volume of gas produced and time of production at 24hrs incubation period is presented in Table 7. The *In vitro* gas production characteristics varied significantly ($P < 0.05$) among the fermented silages. The intercept value (a) for all the silages including the unensiled/fresh maize stover (T_1) ranged from 8.67 to 12.67 at 24hrs. The extent of gas production 'b' values were similar across the treatments except treatment 3 which was significantly ($P < 0.05$) different from other treatments such that, it was the lowest among the treatments. Potential gas production (a+ b) was significantly ($P < 0.05$) different. T_1 was significantly higher than treatments 3 and 4 but similar to 2 and 5.

There were no significant ($P > 0.05$) differences in gas production rate ('c') and 'y' of the incubated samples. Incubation time ('t') followed similar trend. The rate of gas production 'c' ranged from 0.028 to 0.047ml h⁻¹ for all the treatments while the volume of gas 'y' produced at time ('t') ranged from 18.00 to 23.33 for all the treatments. Time of most rapid increase in gas produced 't' ranged from 11.00 for T_2 , T_5 and to 14.0hrs for T_1 .

Table 7: *In vitro* fermentation characteristics of ensiled maize stover at 24hrs incubation period.

Fermentation characteristics	Treatments					SEM
	T1	T2	T3	T4	T5	
a(ml ³)	12.67 ^a	10.67 ^{ab}	8.67 ^b	8.00 ^b	10.67 ^{ab}	0.62
b (ml ³)	32.00 ^a	28.67 ^a	23.00 ^b	28.00 ^a	30.33 ^a	0.71
a+b(ml ³)	44.67 ^a	39.33 ^{ab}	31.67 ^c	36.00 ^{bc}	41.00 ^{ab}	1.07
c(mlh ⁻¹)	0.046 ^a	0.036 ^a	0.047 ^a	0.037 ^a	0.028 ^a	0.01
t (hrs)	14.00 ^a	11.00 ^a	13.00 ^a	12.00 ^a	11.00 ^a	0.82
Y (hrs)	23.33 ^a	20.00 ^a	18.67 ^a	18.00 ^a	18.67 ^a	0.99

a b c=Means on the same row with different superscripts differ significantly (P < 0.05)

a=zero time which ideally reflects the fermentation of soluble fraction

b=extent of gas production from insoluble but degradable fraction

a+b=potential extent of gas production

c=rate of gas production at time (t)

Y=volume of gas produce at time (t)

T₁=Unensiled/Fresh Maize Stover

T₂=Maize Stover Only (Control)

T₃=Maize Stover and Molasses (MSM)

T₄=Maize Stover and Honey (MSH)

T₅=Maize Stover and Sugar (MSS)

SEM = Standard Error of Means

4.5 DISCUSSION

4.5.1 *In vitro* gas production parameters of ensiled maize stover at 24hrs incubation period.

Proximate composition is usually the basic and the most common form of feed evaluation by animal nutritionists. A more reliable technique of estimating livestock feed is *in vitro* gas fermentation (Menke and Steingass, 1988). Although the two methods are independent of each other however, they are interrelated. Gas production is an indication of microbial degradability of samples (Babayemi *et al.*, 2004b, Fievez *et al.*, 2005). All the parameters, observed in this study indicating that the treatments had significant effects on the nutritive value of maize stover silage with or without additives. The lowest and highest gas production and CH₄ production was obtained in maize stover silage (T₂) control and unensiled/fresh MS (T₁). However, the total gas volume (TGV) of control (T₂) silage compared well with fresh MS. In most cases, feedstuffs that showed high capacity for gas production were also observed to be synonymous for high methane production. Methane (CH₄) production in the rumen is an energetically wasteful process, since the portion of the animal's feed, which is converted to CH₄, is eructated as gas. Generally, gas production is a function and a mirror of degradable carbohydrate and therefore, the amounts depends on the nature of the carbohydrates (Demeyer and Van Nevel, 1975; Blummel and Becker, 1997). The presence of fatty acid (FA) in silages may also affect gas volume measurements in bicarbonate buffered *in vitro* measures, where about half of the gas volume is accounted for by CO₂ released upon buffering SCFA (Blummel and Orskov, 1993) and inhibitory effect of saturated fatty acid on rumen microbial ecosystem (Palizdar *et al.* 2011).

Incubation of roughages with bicarbonate buffers produced about 50% of the total gas from buffering of the SCFAs and the rest is generated from fermentation (Blummel and Ørskov, 1993). The organic matter digestibility (OMD) which could be said to be a measure of degradability (potentials) of the microbes on the substrates especially in the presence of sufficient ammonia nitrogen (NH₃-N) which has influence on bacterial fermentation was highest in treatment 1, followed by 5 (1>5>2>4>3). This suggest that T₂ (Control) was the best followed by T₃ in terms of gradual release of gas because quick release of gas from fermentation processes can quickly accumulate and cause distention of the rumen which subsequently cause the

diaphragm to be under pressure which may lead to suffocation as a result of difficulty in breathing especially when the animal has difficulty in expelling the gas (Bloat).

The mean value for methane production was lowest in treatments (Control) silage. Although values obtained from treatments 2, 3 and 4 were not significantly different ($P > 0.05$) but were significantly different ($P < 0.05$) from treatments 1 (unensiled/fresh MS) and 5 (maize stover and sugar) silage. Research on rumen methanogenesis and its inhibition was initiated with aim of increasing feed efficiency (Czerkaszski, 1969). This means that reduced methane production will lead to greater efficiency in feed utilisation. Depending on the level of feed, composition of the diet and digestibility, 2.15% of the gross energy in the feed is lost through methane production (Johnson *et al.*, 1991; Holter and Young, 1992). This also implied that there will be more energy for the animals on treatments 2 (control) and 3 (maize stover and molasses) silages. However, for economical reason, treatment 2 (control silage) will be recommended for the farmers since molasses is costly and not readily available.

Furthermore, gas production is a nutritionally wasted product (Mauricio *et al.* 1999) but provides a useful basis from which metabolisable energy (ME), organic matter digestibility (OMD) and short chain fatty acids (SCFA) could be estimated. More importantly, gas production helps to measure digestion rate of soluble and insoluble fractions of feedstuff (Menke and Steingass, 1988. Pell and Schofield, 1993). The gas produced is directly proportional to the rate at which substrate are degraded (Doano *et al.*, 1997). Somart *et al.* (2000) reported that gas volume is a good parameter to predict digestibility, fermentation and its product and microbial protein synthesis of the substrate by microbes in the *in vitro* system. Gas volumes also have shown a close relationship with feed intake (Blummel and Becker, 1997) and growth rate in cattle (Blummel and Ørskov, 1993).

Metabolisable energy (ME), short chain fatty acid (SCFA) and dry matter degradability (DMD) production all differed significantly for the five treatments. The value for the ME, SCFA and DMD ranged from 7.07 to 8.90, 0.70 to 1.01 and 40.00 to 70.00 respectively. A correlation between ME values measured *in vivo* and predicted from 24hr *in vitro* gas production and chemical composition of feed was reported by Menke and Steingass (1988). The *in vitro* gas

production method has been widely used to evaluate the energy value of several classes of feed (Getachew *et al.*, 1998; 2000). The result obtained in this study is in order with that reported for forage legumes and crop residues by Babayemi *et al.*, (2009), Hriston *et al.* (2009), Alasa *et al.* (2010), Binuomote *et al.* (2010) and Babayemi (2007). Further more, the ME values of the silage (7.07-8.90 MJ/Kg) were within the ranges reported by Menke and Steingass (1988), where the ME values of various European feeds ranged from 4.5 to 15 MJkg⁻¹ DM. Metabolisable energy (ME) values are very useful and important for purposes of ration formulation and to set the economic value of feeds for trading purposes.

When feedstuffs are incubated with buffered rumen fluid (inoculums) *in vitro*, gas production is basically the result of microbial degradation of carbohydrates under anaerobic condition to acetic, propionic and butyric acids (Wolin, 1960, Steingass and Menke 1988, Getachew *et al.* 1998; Khazaal *et al.* 1995 and France and Siddon 1993). Gas production from protein fermentation is relatively small compared to carbohydrate fermentation. The contribution of fat to gas production is negligible (Wolin, 1960). Beuvink and Spoelstra (1992) further stated that gas is produced mainly when feedstuff carbohydrates are fermented to acetate and butyrate with fermentation to propionate yielding gas only from buffering of the acid, therefore forage which produce high amount of propionate should produce low gas volume. Acetate and butyrate are lipogenic which leads to synthesis of butter fat in milk while propionate is glucogenic which leads to production of lean meat. The non significant result obtained for unensiled/fresh MS (T₁), control (T₂) and MSS (T₅) silages on SCFA levels was in line with other reports (Babayemi 2007; Binuomote *et al.* 2010). Gas production was directly proportional to SCFA (Beuvink and Spoelstra, 1992), the higher the gas produced, the higher the short chain fatty acids. Short chain fatty level indicates that the energy is available to the animal and it contributes up to 80% of animal daily energy requirement (Fellner, 2004). Short chain fatty acid (SCFA) is directly proportional to metabolisable energy (ME) (Menke *et al.*, 1979) in this study.

Furthermore, variation of *in vitro* gas production parameters among the silages could also be partly due to differences in sources of rumen fluid (i.e animal and (or) physiological state of the animal). Bonsi *et al.* (1995) studied the influence of donor animal diet on *in vitro* gas production and reported that rumen fluid from animals on different diets resulted in different gas values at

different times of incubation. For example, rumen fluid from animals fed *teff* straw resulted in lower gas values compared to those fed either *Sesbania* or *Leucaenia*. This could be due to the low level of N content of *teff* straw resulting in low rumen ammonia N concentrations (Bonsi *et al.*, 1995), which reduced microbial growth. These authors also reported an interaction between the diet of the donor animal and the type of feed incubated, and Trei *et al.* (1970) also reported differences in gas production between rumen fluid from two animals fed the same diet. The relative proportion of concentrate and forage in the diet will have a considerable influence on *in vitro* gas production. Nagadi *et al.* (2000) reported that differences in the diet of the donor animal influenced gas production from different substrates differently, thereby indicating that there is an interaction of diet of the donor animal and type of feed incubated. The diet of the donor animal exerted considerable influence on bacterial concentrations and so influenced *in vitro* gas production. Since different feeds can affect the relative proportion of microbes in the rumen, this may influence the extent of fermentation of feeds. The magnitude of the diet effect can vary with the type of feed incubated (Bonsi *et al.*, 1995; Nagadi *et al.*, 2000). The major microbial species involved in cellulose degradation adhere closely to plant cell wall surfaces to digest cell wall (Cheng *et al.*, 1983), and use of hay-based diets to donor animals may promote growth of such bacteria thereby increasing rate and extent of fibre digestion.

Moreover, short chain fatty-acids (SCFA) is very important for relating feed composition to production parameters and to net energy value of the forages, therefore production of SCFA from *in vitro* gas measurement will be increasingly important in a developing country. Dry matter degradability (DMD), value of 70.00% obtained for T₂ (control) was the highest while the least value of 40.00% was recorded for T₄ (MSH) silages. The DMD value is a good measure of the amount of dry matter in the feed that can be degraded by microbes in the rumen of ruminants. The result obtained indicates that control silage (T₁) will do better compared to other silages with additives.

4.5.2 *In vitro* Fermentation Characteristics of ensiled maize stover at 24hrs incubation period

The potential gas production from the insoluble fraction, extent and rate of gas production, volume produced and time of production at 24hrs incubation period are presented in Table 7.

The *in vitro* gas production characteristics of the substrate in the liquors from the animals showed that there were significant differences in the 'a', 'b' and 'a + b' values. This may be due to the different additives used to prepare the silages. The values for the nitrogen free extract (NFE) that represents the soluble carbohydrate fraction of the silages had values that were significantly different ($P < 0.05$) for all the treatments. Therefore the treatments behaved similarly in term of 'a' 'b' and 'a + b'. Getachew *et al.* (1998) reported that gas production can be attributed to the nature of carbohydrate fractions contained in the substrates. The intercept value 'a' for all the silages (treatments) at 24hrs ranged from 8.00 in T₄ (maize stover and Honey) silage to 12.67 in T₁ (unensiled/fresh MS). The T₁ (Fresh MS) was similar to T₂ (control) and T₅ (MSS), meaning there was minimal loss of water soluble carbohydrate during fermentation and storage from the original material. The value for absolute 'a' used ideally reflects the fermentation of soluble fraction in this study. The soluble fraction makes the attachment by rumen microorganisms to be done easily and leads to much gas production. Therefore, more ruminant microorganism worked on Fresh MS (T₁) and T₂ (Control) and this leads to higher gas production.

The extent of gas production 'b' described fermentation of the insoluble but degradable fraction in fresh MS (T₁) and MS and Honey (T₅) which recorded high values of 32 and 30.33 respectively could be attributed to the relatively high amount of crude protein in the stover. This facilitated high rate of microbial activities by supplying the required nitrogen for their cellular protein synthesis as established by Roger *et al.* (1977). A linear relationship has been established between high crude protein in forages and *in vitro* degradability (Njidda *et al.*, 2010). The values of 'b' obtained in this study (23.00 – 32.00) were consistent with those reported for dry matter (DM) degradation of some tropical legumes and grasses (Ajayi *et al.*, 2007 and the values of 9.5 – 32.0ml/200mg DM reported for some crop residues (Babayemi *et al.*, 2009). The potential degradability 'a+b' of a diet depicts the level at which the diet could be degraded if it were in the actual rumen of the animal (*in vivo*). This largely depends on how much of the fibre fractions (NDF and ADF) have been broken down for easy access of the microbes to the nutrients available in the diet. At 24hrs, there were significant variations among the treatments such that it was highest for the T₁ (Fresh MS) and lowest for the T₃ (MSM) respectively. However, T₁ values for 'a + b' was similar to T₂ (control) and T₅ (MSS) silages. The high value of the potential

extent of gas production recorded for T₁, T₂ and T₅ was due to abundant of carbohydrate fraction embedded in Fresh MS and MS only (control) and MS and sugar. Getachew *et al.*, (1998) stated that it is well known that gas production is basically the result of fermentation of carbohydrate to volatile fatty acid (acetate, butyrate and propionate). Menke and Steingass (1988) also reported that fermentable carbohydrate increase gas production while degradable nitrogen compound decrease gas production to some extent because of their binding of carbohydrate with ammonia.

The volume of gas 'y' at time 't' is the peak of gas production for each sample at 24hrs incubation period. Since rate 'c' of gas production at time 't' and volume of gas 'y' of the incubated samples were similar across the treatments, it means that additives had no effect on MS silage regarding the 'c' and 'y' characteristics of the gas. However, there are many factors that may determine the amount of gas to be produced during fermentation, depend on the nature and level of fibre, the presence of secondary metabolites (Babayemi *et al.*, 2004) and potency of the rumen liquor for incubation. It is possible to attain potential gas production of a feedstuff if the donor animal from which rumen liquor for incubation was collected got the nutrient requirement met. The utilisation of roughages is largely dependent on microbial degradation therefore the rate and potential extent of gas production would provide a useful basis for the evaluation of the MS silage as potential feed resources. Since gas production is dependent on the relative proportion of soluble, an insoluble but degradable and undegradable particle of feed; mathematical description of gas production profiles allows evaluation of substrate and fermentability of soluble and slowly fermentable component of feeds (Getachew *et al.*, 1998).

Based on the above assumption, therefore, it could be adduced that among the MS silages studied, MSM (T₃) silage and fresh MS (T₁) would provide minimal proportion of residue that would take up space if utilised in *in vivo* studies also persists as indigestible residue. Ørskov and Ryle (1990) reported that the rate (c) determines digestion time and consequently how long a potentially digestible material would occupy space. Therefore the potential extent of digestion ('b') values obtained for treatments 1, 2, 4 and 5 demonstrated that they possess more potentially degradable carbohydrates than T₃. Also the results presented in Table 7 actually demonstrated that digestion rates ('c') and potential extent ('b') of gas production provided a more meaningful index of nutritional value than ultimate digestibility comparatively. However, the conversion of true fermented organic matter into gas varied with the type of additive used to prepare the silage

CHAPTER FIVE

EXPERIMENT THREE

5.0 PERFORMANCE , DIGESTIBILITY AND NITROGEN BALANCE OF WAD SHEEP FED ENSILED MAIZE STOVER AND CONCENTRATE SUPPLEMENTS

5.1 Introduction

The beneficial roles of livestock in production of meat, milk, fibre and skin in alleviation of poverty, malnutrition and conserving natural resources has not been adequately exploited (Chidebelu and Ngonodjou, 1997, Hugo *et al.*, 2002). Agricultural production in most of the tropics and subtropics are predominantly small-scale crop livestock or mixed farming systems. Part of the solution in Nigeria is an increase in the production of small ruminant animals, mainly sheep and goats which are found in most of the households in Southern Nigeria (Sumberg and Cassaday, 1985).

Traditional ruminant livestock production in Africa is based predominantly on animals grazing natural pastures which are often of low nutritive value especially during the dry season. The nutritive value of the natural pastures varies according to season. Protein content is between 8 and 12% of DM at the start of the rainy season but drops to 2% in the 6 – 7 month dry season (Chimwano, 1978 and Harrison, 1989). The grasses grow rapidly during the wet season, later becoming fibrous, coarse, and highly lignified rendering it indigestible. Their quality declines further during the dry season when they become standing hay. This results in loss of palatability and ineffective utilisation of the pastures by the animals.

Inadequate nutrition is one of the factors that generally affect livestock productivity. Despite the naturally endowed vegetations, there are still inadequate feeds and feedstuffs for livestock in Nigeria (Babayemi, 2007). Ruminants in the tropics are raised predominantly on grasses which are inherently poor in digestibility, nutritive value and unavailable in the off-season (Babayemi *et al.*, 2009). The most important factors determining the profitability of any livestock enterprise is optimal level of feeding. This aim is most problematic to achieve during the dry season when available feed is scarce and of low nutritive quality (Adejumo and Ademosun, 1985; Davies and Onwuka, 1993; Pamo and Pieper 2000). It is therefore a common feature to find well-fed and

robust small ruminants in the rainy season to have appreciably lost weight in the following dry season (McDowell, 1972; Pagot, 1992; Adegbola, 1998).

The concept of ruminant animal production requires feeding animals on rich diet so that they can attain slaughter weight within short time usually 70 – 120 days (Madziga *et al.*, 2012). Good nutrition is a prerequisite for good health, good reproduction, high milk yield, fast growth rate and a successful rearing system (Ochepo *et al.*, 2009). Recent trends in animal nutrition have focused attention on the use of crop residues and agro-industrial by-products but these are low in protein, high in fibre and low in digestibility. Expensive concentrates and milling by-products are forcing farmers to rely more on upon crop by-products as sources of energy. Performance of animals fed crop residues is limited by poor intake, low nitrogen contents and poor digestibility (Peterson *et al.*, 1981). However, sheep and goats can play an active role in converting crop residues of no human dietary value to meat and milk of high nutritive value for man (Fajemisin *et al.*, 2010).

The search for alternative and locally available sources of energy and protein to enhance productivity of sheep and goats during the period of scarcity and dry season has placed attention on the used of post-harvest crop residues (Sodeinde *et al.*, 2007). Although there is scarcity of natural pastures during the dry season, there is usually an abundance of crop residues especially cereal straws and stovers, which have potentials to be used as feed. These can be used to improve the nutrition of ruminant livestock during the dry season through the strategic supplementation of animals with crop residues. In Nigeria, very few crop residues are utilised as ruminants feed by small holder farmers. Maize crop residue (stover) can be an inexpensive source of forage, and it may be grazed, stacked or ensiled. Preservation of maize stover as silage makes it possible to preserve plant nutrients that otherwise would be lost by physiological activity or leaching, offering the possibility of using stover in rations for growing animals.

To achieve efficient utilisation of the crop residues especially ensiled maize stover as a potential feed for the ruminants; it has to be supplemented with concentrate. Sustainability of sheep production could come through the feeding of ensiled maize stover and concentrate supplements.

After the chemical composition, silage quality characteristics, acceptability study and *in vitro* fermentation assessment showed that ensiled Maize Stover (MS) without additives (control) had similar quality characteristics and nutritive value with those MS silages with additives and was accepted by the animals. Therefore, silage for growth performance, digestibility and nitrogen balance study was prepared based on control MS. This study was therefore, undertaken to assess the dietary effect of ensiled maize stover and concentrate supplements on dry matter intake, growth performance, digestibility and nitrogen balance of West African dwarf sheep.

5.2 MATERIALS AND METHODS

5.2.1 Experimental Site

The study was carried out between the period of May to September 2011 at the Sheep and goat unit of the Teaching and Research Farm, University of Ibadan, Ibadan, Nigeria.

5.2.2 Preparation of Pens

The animal pens were made of low walls of 1.90m (Height) and 7.10m by 13.94m in size and each pen was about 1.83m long and 1.54 m wide. The floor of the pen was made of concrete and the roof of the sheep unit which housed the pens was made of corrugated iron sheets. The pens were dusted and washed thoroughly with detergent and were further disinfected with broad spectrum insecticide, acaricides and larvicides (Diazintol). The feeding and drinking troughs were washed and disinfected and the whole house was left to rest for two weeks before usage. Wood shavings were spread on the floor of the pens up to a depth of 5cm as bedding materials to enhance prompt removal of urine and faeces and were replaced fortnightly.

5.2.3 Experimental Diets

Silage fermentation procedure and duration were as described as in experiment 1. Concentrate supplements was prepared (See Table 8)

- Diet A = 75% Control + 25% Concentrate supplements
B = 50% Control + 50% Concentrate supplements
C = 25% Control + 75% Concentrate supplements
D = 100% Control (Silage only)
E = 100% Concentrate supplements

Table 8: Ingredient and crude protein composition (%) of concentrate supplement fed to WAD sheep

Ingredient	Percentage
Wheat bran	60.00
Palm kernel cake	25.00
Corn bran	10.00
Oyster shell	3.00
Salt	1.00
Premix (Ruminants)	1.00
Total	100.00
Crude protein	15.30

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5.2.4 Experimental Animals and Management

Twenty (20) post weaned male West African dwarf sheep with average initial body weight of 14.0 – 16.0kg and 9 – 12 months old were used for the experiment. They were purchased from Iwo in Osun State of Nigeria. On arrival, animals were given a prophylactic treatment which consisted of oxytetracycline long acting antibiotic (1ml/10kg body weight of the animal) and vitamin B complex. They were also drenched with albendazole to control endoparasites and treated for mange and other ectoparasites using Ivomec^(R). They were later vaccinated against *Pestes des petits ruminants* (PPR) using a tissue culture Rinderpest Vaccine. During the adaptation of six weeks, sheep were offered diets they were eaten from where they were purchased, but were introduced to the experimental diets two weeks before the end of adaptation period.

At commencement of the feeding trial, animals were weighed and allotted to the five dietary treatments on weight equalisation basis such that the average initial weights per treatment were not statistically different. Silage and concentrate supplements were fed to the in a mixture. Feed were offered at 0800h and 1500h at 5% body weight. Ration offered was frequently adjusted to ensure that each animal received 10% of feed above its previous week's consumption. For the determination of daily feed intake, the orts were weighed daily before feeding and amounts deducted from total amounts served the previous day. Sample from orts were taken for proximate composition. Fresh water and feed were served *ad libitum* each day for the one hundred and five days (105) duration of the experiment. Salt licks were placed permanently in each pen. Weights of sheep were taken on a weekly basis using scale before the morning feeding.

5.3 Digestibility and Nitrogen Balance Study

5.3.1 Experimental Diets

The experimental diets were the same as in growth performance.

- Diet A = 75% Control + 25% Concentrate supplements
- B = 50% Control + 50% Concentrate supplements
- C = 25% Control + 75% Concentrate supplements
- D = 100% Control (Silage only)
- E = 100% Concentrate supplements

5.3.2 Experimental Animals

The experiment was carried out at the sheep and goat unit of the Teaching and Research Farm of the University of Ibadan, Ibadan, Nigeria in the month of August 2011. Fifteen (15) post weaned female West African dwarf sheep aged 8 – 10 months weighing 16 – 20kg were used for the experiment. They were purchased from Iwo town, in Osun state, on arrival; the sheep were given prophylactic intramuscular treatment of oxytetracycline and vitamin B complex; at the dosage of 1ml/10kg body weight of the animal. They were also drenched with albendazole to control endoparasites and treated for mange and other ectoparasites using Ivomec^(R).

5.3.3 Experimental Design

The animals were allowed two weeks (2 wks) of adjustment/adaptation to their new environment (acclimatisation) and the effect of the administered drugs to wear out. The sheep of similar average body weight were randomly allotted into separate metabolic cages with fitted facilities for separate collection of faeces and urine (Akinsoyinu, 1974). The design of the experiment was a completely randomised design (CRD).

5.3.4 Animal Feeding

The experiment lasted for fourteen days (14 days) in which the first seven (7) days was to adapt the sheep to the new test diets. The silage and concentrate supplements were fed in a mixture to the animals. The sheep were offered the feed during a seven-day adaptation period prior to another seven days collection period. Feed was served at 5% body weight of the animals. Water and salt lick were provided to the animals *ad libitum* throughout the metabolic trial. Animals were weighed at the beginning and end of the digestibility trial. Feed refused was weighed at 0800 hours every morning and deducted from the total offered for intake determination prior to serving new feed daily. During the seven days of collection period, total faeces and urine were collected, weighed daily. A 10% aliquot sample was stored in the freezer at -4°C . Thereafter, the sample from each day's collection was bulked, mixed and dried in the oven at 105°C for chemical analysis. Total urine from each animal was collected and measured daily, using a measuring cylinder and kept separately in labeled containers. Two drops of concentrated sulphuric acid (H_2SO_4) was added to each container daily after collection of each sample to prevent microbial growth, organic matter decay and loss of nitrogen/nitrogen volatilisation.

Approximately, 10% of total urine samples were later pooled for the 7 days period and frozen at -4°C in a freezer till required for nitrogen analysis.

5.3.5 Chemical Analysis

Dry matter, Crude protein, Crude fibre, ash ether extract and nitrogen free extract were determined according to AOAC (1995). All samples were analysed. Neutral detergent fibre, acid detergent fibre and acid detergent lignin were determined according to Van Soest *et al.* (1991). Hemicellulose was calculated as the difference between NDF and ADF and cellulose as the difference between ADL and ADF (Rinne *et al.*, 1997).

5.4 Statistical Analysis

The experimental design was completely randomized design (CRD). The feed intake, growth rate and feed conversion ratio were computed and subjected to a one-way analysis of variance using the procedure of SAS (1999). Significant treatment means were compared using Duncan (1955) option of the same software.

Experimental model: $Y_{ij} = \mu + \alpha_i + e_{ij}$

Where Y_{ij} = individual observation μ = general mean of the population

α_i = treatment effect e_{ij} = composite error effect

and

Polynomial regression model: $Y = \alpha + \beta_1X + \beta_2X^2 + \dots + \beta_nX^n$

Where, Y = dependent variable, X = an independent variable, α = the intercept

β_i (i = 1,.....,n) = Partial regression coefficient associated with the i th degree polynomial

n = number of observation

5.5 RESULTS

5.5.1 Composition of Experimental Diets

Table 9 show the composition of experimental diets and the concentrate supplement fed to WAD sheep.

Table 9: Ingredient composition (%) of experimental diets fed to WAD sheep

Ingredient	Experimental Diets				
	A	B	C	D	E
Silage (C)	75		25	100	-
Concentrate	25		75	-	100
Supplements (CS)					
Total	100	100	100	100	100

C = Control

CS = Concentrate supplements

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5.5.2 Chemical Composition of ensiled maize stover and Concentrate Supplements

Chemical composition of ensiled maize stover and concentrate supplements are presented in Table 10. The amounts of dry matter content (31.6%) in maize stover silage was comparable with amount in diet A (46.1%) but appreciably lower than the contents in concentrate supplements (89.5%), diet B (60.5% and C (75.0%) respectively. The crude protein content of the silage (8.4%) was lower than other silages with concentrate supplements. Ash content in silage (8.8%) was relatively comparable to concentrate supplements (13.9%) and other diets with varied concentrate supplements. Nitrogen free extract content silage was relatively the same with other diets with concentrate supplements. Fibre content and acid detergent lignin contents in maize stover silage were higher than the rest diets with concentrate. The neutral detergent fibre (i.e. cell walls) contents (46.8%) was slightly higher than in concentrate supplements but lower than contents in diets A, B and C. The lignin content in silage was slightly higher than other diets and concentrate supplements but lower in hemicelluloses and cellulose contents than other dietary treatments.

Table 10 : Chemical composition (%) of ensiled maize stover and concentrate supplements

Parameters	Diet composition				
	A	B	C	D	E
Dry matter	46.08	60.54	74.99	31.62	89.45
Organic matter	92.11	91.03	89.21	91.20	86.06
Crude protein	12.60	14.35	16.45	8.40	15.05
Ether extract	8.86	7.84	8.50	6.77	7.88
Ash	7.89	8.97	10.79	8.80	13.94
Nitrogen free extract	45.76	46.09	43.39	46.25	43.30
Crude fibre	24.89	22.75	20.87	29.78	19.83
Neutral detergent fibre	54.60	49.69	52.80	46.83	44.74
Acid detergent fibre	32.76	28.82	27.84	26.86	29.69
Acid detergent lignin	11.92	10.91	10.81	12.81	9.50
Hemicellulose	21.84	20.87	24.96	19.97	15.05
Cellulose	20.84	17.91	17.03	14.05	20.19

Diet A = 75% Control + 25% Concentrate supplements

B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplements

D = 100% Control (Silage only)

E = 100% Concentrate supplements

5.5.3 Feed Intake and performance characteristics of WAD Sheep fed ensiled maize stover and concentrate supplements

Table 11 shows the daily body weight gain (BWG), daily dry matter intake (DMI) and feed conversion ratio (FCR) of WAD sheep fed ensiled maize stover and concentrate supplements. Significant differences ($P < 0.05$) occurred in BWG, DMI, metabolic weight gain, metabolic feed intake and FCR among the treatment means except for initial body weight (IBW). The BWG of animals on diet A (concentrate supplements only) was significantly highest (81.90g/d) among other dietary treatments and similar to animals on diet C (66.55g/d). However, BWG of animals on control diet D (silage alone) was significantly ($P < 0.05$) negative (-2.00g/d). DMI decreased significantly with increased inclusion of control (maize stover silage) whereas the FCR of diet E (100% CS) was not significantly ($P > 0.05$) different from diet C (75% CS and 25C). Similarly, there was no significant ($P > 0.05$) differences between FCR mean of diet A (75% C + 25 CS) and B (50% C + 50% CS). However, FCR of control diet (D) was significantly ($P < 0.05$) negative. The metabolic body weight gain and metabolic daily dry matter intake followed similar trend as BWG and DMI.

Table 11: Feed intake and performance of WAD sheep fed ensiled maize stover and concentrate supplements

Parameters	Experimental diets					SEM
	A	B	C	D	E	
Initial body weight (Kg)	15.00 ^a	16.50 ^a	13.75 ^a	15.75 ^a	15.00 ^a	0.91
Final body weight (Kg)	18.50 ^{bc}	20.75 ^{ab}	21.50 ^{ab}	14.00 ^c	24.50 ^a	0.86
Body weight gain (Kg)	3.50 ^c	4.25 ^{bc}	7.75 ^{ab}	-2.00 ^d	9.50 ^a	0.64
Daily body weight gain (g/d)	33.34 ^c	40.48 ^{bc}	73.81 ^{ab}	-19.05 ^d	90.48 ^a	6.13
Metabolic body weight gain (g/dW ^{0.75})	13.63 ^b	15.28 ^b	25.11 ^{ab}	-7.39 ^c	29.14 ^a	1.92
Total daily dry matter intake (g/d)	454.49 ^b	473.29 ^b	530.93 ^{ab}	242.26 ^c	592.41 ^a	18.21
Metabolic daily dry matter intake (g/dW ^{0.75})	54.96 ^a	52.74 ^a	58.83 ^a	31.67 ^b	64.13 ^a	0.68
Feed conversion ratio	16.15 ^a	18.72 ^a	7.39 ^b	-5.32 ^c	7.06 ^b	2.51

a, b ,c= Means on the same row with different superscripts are significantly (P < 0.05) different

SEM = Standard Error of Means

Diet A = 75% Control + 25% Concentrate supplements

B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplements

D = 100% Control (Silage only)

E = 100% Concentrate supplements

Table 12 shows the polynomial regression of performance of WAD sheep fed ensiled maize stover and concentrate supplements. Under polynomial regression, final body weight (FBW), body weight (BWG), daily body weight gain (DBWG) and metabolic daily body weight gain (MDBWG^{0.75}) were significant cubically and quartically. However, dry matter intake total (DMIT) and dry matter intake total metabolic (DMITM^{0.75}) were significant quadratically, cubically and quartically while feed conversion ratio (FCR) was only significant cubically. The residual values showed that the magnitude of departure of predicted values from the actual value is minimal, indicating that the model used is perfect.

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Table 12: Polynomial Regression of Performance of WAD Sheep fed ensiled maize stover and concentrate supplements.

Var	FBW			BWG			DBWG			DMIT			Ditmm			MDBWG			FCR		
Polyeq	$Y=19.85+19.5X^3+0.47X^4$			$Y=4.60+1.85X^3+0.72X^4$			$Y=43.81+17.62X^3+7.0X^4$			$Y=458.67+22.62X^2+59.9X^3+19.57X^4$			$Y=52.46+2.59X^2+5.13X^3+1.92X^4$			$Y=15.14+6.1X^3+2.31X^4$			$Y=8.80+3.89X^3$		
Trt.	Act	Pre	R	Act	Pred	R	Act	Pre	R	Act	Pred	R	Act	Pred	Re	Act	Pred	R	Act	Pre	Re
A	18.5	18.02	0.48	3.50	3.47	0.03	33.33	33.19	0.14	454.48	463.48	-9.00	54.95	53.25	1.70	13.62	11.35	2.27	16.15	14.91	1.24
B	20.75	21.87	-1.12	4.25	5.42	-1.17	40.47	51.05	-10.58	473.28	477.17	-3.89	52.73	52.45	0.28	15.28	18.10	-2.82	18.72	16.58	2.14
C	21.50	22.67	-1.17	7.75	8.92	-1.17	73.80	85.61	-11.81	530.93	530.85	0.08	58.82	59.98	-1.16	25.11	29.00	-3.89	7.39	8.80	-1.41
D	14.00	14.15	-0.15	-2.20	-1.98	-0.22	-19	-19.4	0.40	242.26	237.77	4.49	31.67	31.93	-0.26	-7.47	-6.46	-1.01	-5.3	1.02	-6.32
E	24.50	22.27	2.23	9.5	7.17	2.33	90.4	68.43	21.97	592.4	583.48	8.92	64.13	63.51	0.62	29.14	23.54	5.60	7.06	12.69	-5.63

Res= residual which can be obtained by subtracting the predicted (pred) from actual(act) values

Variable 'IBW' was excluded because no polynomial term was significant for it

Note: Linear term was not significant for all the regressions for all Performance.

IBW = Initial Body Weight, FBW = Final Body Weight, DBWG =Daily Body Weight Gain, DMIT = Dry Matter Intake Total, MDDMI = Metabolic Daily Dry Matter Intake, MDBWG = Metabolic Daily Body Weight Gain, FCR = Feed Conversion Ratio.

Diet A = 75% Control + 25% Concentrate supplements, B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplements, D = 100% Control (Silage only)

and E =100% Concentrate supplements

Degree of Polynomial

X^1 = Linnear (1st)

X^2 = Quadratic (2nd)

X^3 = Cubic (3rd)

X^4 = Quartic (4th)

5.6 Apparent digestibility of WAD Sheep fed ensiled maize stover and concentrate supplements

The dry matter intake (DMI) values ranged from 437.50 to 694.41gDM/day. It varied significantly across the treatments such that it was highest in diet C and lowest in diet D (100% MS) silage. Apparent digestibilities (%) of dry matter (DM), organic matter (DOM), crude protein (DCP), crude fibre (DCF), ether extract (DEE), nitrogen free extract (DNFE), ash (DA), neutral detergent fibre (DNDF), acid detergent fibre (DADF), acid detergent lignin (DADL), hemicelluloses and cellulose in WAD sheep are presented in Table 13. The DDM ranged from 62.18 to 67.44. There was no significant difference ($P > 0.05$) in the apparent dry matter digestibility (DMD) of diets. Treatment C (25C + 75%CS) gave the highest value while diets D (100%C) and E (100%CS) gave the least. The DOM ranged from 73.00 to 84.50. There was significant difference. While experimental diet D (100%CS) gave the least. For the DCP, the range was 47.82 to 74.15. Dietary treatments A, B, C and E were statistically similar while diet D was significantly different from others. The DCF ranged from 35.09 to 58.56. There was no ($P > 0.05$) significant difference between the treatment means. Diet D (100% Silage) had the highest value while diet C had least value. The DEE ranged from 52.39 to 69.99. There was significant ($P < 0.05$) between the treatments. Dietary treatment A was significantly higher than treatment D but similar to dietary treatments B, C and E. For DA, the range was 19.29 to 32.77. There was no significant ($P > 0.05$) difference. The DNFE followed similar trend. There was no significant ($P > 0.05$) difference between the treatments means. The range was 74.51 to 84.26; Diet B gave the highest value while diet D gave the least. The DNDF, ranged from 46.47 to 57.47. There was significant difference. While diet C (25% C + 75%) gave the highest values, E (100% CS) gave the least. DADF ranged from 30.27 (100% Silage – control) to 45.65 (50% C + 50% CS). There was no significant difference. The ADL ranged between 31.08 in diet C (25% C + 75%CS) and 48.48 in diet B (50%C + 50% CS). For hemicelluloses, the value ranged from 67.23 (Diet E) to 87.95 (Diet C). There was significant ($P < 0.05$) difference. Treatment means of diet C was significantly higher than diet E but similar to diets A, B and D (control) while dietary treatment E was significantly ($P < 0.05$) lower to treatment C but similar to diets A, B and D. The cellulose ranged from 58.27 to 74.47. The dietary treatment B was significantly higher than D but similar to A, C and E. Metabolisable energy (ME) values ranged from 10.95 to 12.68 MJ/KgDM and differed significantly ($P < 0.05$) across the dietary treatment.

Table 13: Apparent digestibility (%) of WAD Sheep fed ensiled maize stover and concentrate supplements

Parameters	Experimental Diets					SEM
	A	B	C	D	E	
Dry Matter intake	437.5 ^b	681.79 ^a	694.41 ^a	321.65 ^c	612.22 ^a	17.13
Dry Matter	62.32 ^a	66.32 ^a	67.44 ^a	62.18 ^a	62.18 ^a	3.29
Organic Matter	83.50 ^a	78.50 ^b	78.50 ^b	84.50 ^a	73.00 ^c	0.52
Crude Protein	66.40 ^a	73.32 ^a	74.15 ^a	47.82 ^b	66.62 ^a	2.82
Crude Fibre	46.43 ^a	36.48 ^a	35.09 ^a	58.56 ^a	43.50 ^a	5.67
Ether Extract	69.99 ^a	66.07 ^{ab}	68.67 ^a	52.39 ^a	65.64 ^{ab}	2.62
Ash	22.05 ^a	19.29 ^a	23.93 ^a	32.77 ^a	29.90 ^a	6.72
Nitrogen Free Extract	76.95 ^a	84.26 ^a	76.87 ^a	74.51 ^a	80.97 ^a	1.87
Neutral Detergent Fibre	56.73 ^a	59.67 ^a	57.47 ^a	48.71 ^{ab}	46.47 ^{ab}	3.60
Acid Detergent Fibre	44.97 ^a	45.65 ^a	34.09 ^a	30.27 ^a	35.94 ^a	5.58
Acid Detergent Lignin	43.96 ^a	48.48 ^a	31.08 ^a	38.11 ^a	44.66 ^a	5.62
Hemicellulose	74.33 ^{ab}	79.04 ^{ab}	87.95 ^a	73.40 ^{ab}	67.23 ^b	1.76
Cellulose	69.79 ^{ab}	74.47 ^a	68.61 ^{ab}	58.27 ^b	67.64 ^{ab}	5.85
Metabolizable Energy	12.53 ^a	11.78 ^b	11.78 ^b	12.68 ^a	10.95 ^c	0.08

abc –Means on the same row differently superscripted are significantly (P<0.05) different

Diet A = 75% Control + 25% Concentrate supplements

B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplements

D = 100% Control (Silage only)

E = 100% Concentrate supplements

SEM = Standard Error of Means

5.6.1 Nitrogen utilisation by WAD Sheep fed ensiled maize stover and concentrate supplements

Table 14 shows the Nitrogen utilisation by the experimental sheep. N-intake, faecal-N, urinary-N, N-total excreta, N-balance, N-balance per metabolic weight and N-retention ranged from 4.32 – 18.28, 2.23 – 4.98, 0.93 – 3.23, 3.16 – 8.21, 1.16 – 11.75, 0.190 – 1.217 and 26.46 – 67.13 respectively. N-intake was significantly ($P < 0.05$) highest (18.28) in animals fed dietary treatment C and lowest (4.32) in animals that received treatment D. No significant ($P > 0.05$) difference was observed for faecal-N in all the treatments. For urinary-N, there was significant ($P < 0.05$) difference. Animals on experimental diets A, C and E were not significantly ($P > 0.05$) different while those in diet B and D were also similar statistically. N-excreted was significantly ($P < 0.05$) higher in animals on diet E than animals on diet D (control) but similar to those in groups A, B and C diets. Positive N-balance was observed for all the five diets. However, there was significant ($P < 0.05$) variation between the treatments means, diet B (50%C + 50%CS) gave the highest value while diet D (100%C) gave the least. N-balance per metabolic weight was significantly ($P < 0.05$) different such that it was highest in diet B and lowest in diet D. Similarly, significant ($P < 0.05$) variation was observed between the dietary treatments for N-retention (%). It was highest (67.15) in diet B and lowest (27.61) in diet D.

Table 14: Nitrogen utilisation by WAD sheep fed ensiled maize stover and concentrate supplements

Parameters	Experimental diets					SEM
	A	B	C	D	E	
Nitrogen intake (g/d)	8.82 ^c	15.65 ^b	18.28 ^a	4.32 ^d	14.74 ^b	0.40
Nitrogen (faeces) (g/d)	2.96 ^a	4.17 ^a	3.34 ^a	2.23 ^a	4.98 ^a	0.70
N – urine (g/d)	2.57 ^a	0.98 ^b	3.19 ^a	0.93 ^b	3.23 ^a	0.04
N – total excreta (g/d)	5.54 ^{ab}	5.14 ^{ab}	6.52 ^{ab}	3.16 ^b	8.21 ^a	0.60
N – balance (g/d)	3.28 ^c	10.51 ^a	11.75 ^a	1.16 ^c	6.54 ^b	0.47
N – balance W ^{0.75} (g/d)	0.436 ^c	1.089 ^{ab}	1.217 ^a	0.190 ^c	0.836 ^b	0.06
N – retention (%)	37.23 ^{bc}	67.13 ^a	64.99 ^{ab}	26.46 ^c	44.23 ^b	2.95

abc = Means on the same row differently superscripted are significantly different (P < 0.05)

Diet A = 75% Control + 25% Concentrate supplements

B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplements

D = 100% Control (Silage only)

E = 100% Concentrate supplements

SEM = Standard Error of Means

All the parameters considered under Nitrogen balance using polynomial regression analysis were significant quadratically, cubically and quartically except for nitrogen total excreta (NTE) that was significant quartically. The residual values, irrespective of the signs whether negative or positive, shows magnitude of departure of predicted value from the actual value. Alternatively it shows the response of predicted value to actual value. Further more, where the response is zero, it indicates that both the actual and predicted value are the same and model is perfect.

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Table 15: Polynomial Regression of Nitrogen Balance of WAD Sheep fed ensiled maize stover and concentrate Supplements.

Var	NI			UN			NTE			NB			NR (%)			NB M ^{0.75}		
Polyn.	Y=12.36-			Y=2.17+0.83X ² +0.25X ⁴			Y=5.71+0.28X ⁴			Y=6.64-			Y=48.01-			Y=0.76-		
equa	0.67X ² +2.85X ³ +0.76X ⁴									1.10X ² +2.2X ³ +0.48X ⁴			4.33X ² +8.83X ³ +1.39X ⁴			0.08X ² +0.22X ³ +0.05X ⁴		
Trt.	Act	Pred	Res	Act	Pred	Res	Act	Pred	Res	Act	Pred	Res	Act	Pred	Res	Act	Pred	Res
A	8.82	8.93	-0.11	2.57	4.00	-1.43	5.53	5.99	-0.46	3.28	2.72	0.56	37.23	31.91	5.32	0.46	0.44	0.02
B	15.65	14.55	1.10	0.97	0.35	0.62	5.14	4.05	1.09	10.51	8.03	2.48	67.13	64.44	2.69	1.09	1.08	0.01
C	18.28	18.26	0.02	3.18	2.01	1.17	6.52	7.39	-0.87	11.70	11.72	-0.02	64.99	65.01	-0.02	1.22	1.22	0
D	4.32	4.95	-0.63	0.93	0.34	0.59	3.16	4.05	-0.89	1.16	1.43	-0.27	26.46	29.12	-2.66	0.19	0.19	0
E	14.74	15.97	-1.23	3.22	4.08	-0.86	8.21	5.99	2.22	6.53	7.12	-0.59	44.23	49.57	-5.34	0.84	0.86	-0.02

Res= residual which can be obtained by subtracting the predicted from actual (act) values

Variable 'N-Fecal' was excluded because no polynomial term was significant for it

Note: Linear term was not significant for all the regressions for all Nitrogen balance.

NI = Nitrogen Intake, UN = Urinary Nitrogen, NTE = Nitrogen Total Excretal, NB = Nitrogen Balance, NR (%) = Nitrogen Retention

NBM^{0.75} = Nitrogen Balance Metabolic

Diet A = 75% Control + 25% Concentrate supplements, B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplement, D = 100% Control (Silage only)

and E = 100% Concentrate supplements

Degree of Polynomial

X¹ = Linear (1st)

X² = Quadratic (2nd)

X³ = Cubic (3rd)

X⁴ = Quartic (4th)

5.6.2 Total digestible nutrients by WAD sheep fed ensiled maize stover and concentrate supplements

The digestible crude protein (DCP), digestible crude fibre (DCF), digestible ether extract (DEE), digestible nitrogen free extract (DNFE) and total digestible nutrients (TDN) presented in Table 16 ranged from 3.99 – 12.20%, 7.32 – 17.23%, 7.96 – 13.95%, 33.35 – 38.85% and 63.65 – 70.92% respectively. Significant ($P < 0.05$) differences were observed for all the parameters.

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Table 16 : Total digestible nutrients intake (%) by West African Dwarf sheep fed ensiled maize stover and concentrate supplements

Nutrient	Experimental diets					SEM
	A	B	C	D	E	
Crude protein	8.36 ^c	10.53 ^b	12.20 ^a	3.99 ^d	10.04 ^b	0.17
Crude fibre	11.34 ^b	9.96 ^b	7.32 ^c	17.23 ^a	8.63 ^b	0.47
Ether extract	13.95 ^a	11.90 ^b	13.13 ^a	7.96 ^c	11.67 ^b	0.19
Nitrogen free extract	35.22 ^b	38.85 ^a	33.35 ^b	34.46 ^b	33.06 ^c	0.31
Total digestible nutrient	68.87 ^{ab}	70.92 ^a	65.70 ^{ab}	63.65 ^b	65.15 ^{ab}	1.15

abc - Means on the same row differently superscripted are significantly different ($P < 0.05$)

SEM = Standard Error of Mean

Diet A = 75% Control + 25% Concentrate supplements

B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplements

D = 100% Control (Silage only)

E = 100% Concentrate supplements

5.7 DISCUSSION

5.7.1 Dry matter intake and growth performance

The dry matter content of ensiled maize stover only (control diet) 31.6% compared well with other forage grasses such as Guinea grass, (*Panicum maximum*) 26.0 – 35.1% Elephant grass (*Pennisetum purpureum* (37.7%) and Gamba grass (*Andropogon gayanus*) 21.7% reported by Babayemi (2007) and Odedire and Babayemi (2008) respectively. Similarly, the organic matter content of ensiled maize stover (91.20%) compared well with diets A, B, C and E with concentrate supplements at varied level. It also compared well with 91.5 and 93.0% reported by Elkholy *et al.* (2009) and Nour *et al.*, (1987) respectively. This observation suggests that the silages could furnish nutrients to small and large ruminants. Dry matter (DM) content obtained in this study was clearly higher than 23% and 21.2% reported in similar MS-silages by Moran (2005) and Ashbell and Weinberg (2002) respectively but lower than 37.02% reported by Elkholy *et al.*(2009). The obvious differences in the various values may be in the differential proportions of morphological parts i.e stem to leaf ratio. The crude protein obtained in silages used in this study (8.4%) exceeds that of Guinea grass (7.9%) and also the 6 – 7%, that can provide the minimum ammonia level for efficient rumen microbial activities (Minson, 1982). Therefore, it thus appears that the ensiled maize stover could supplement or replace poor quality grasses especially during the dry season.

Furthermore, feed type is an important factor that affects sheep growth and performance. Andrews and Ørskov (1970) reported that average daily body weight gain (ADBWG) of growing lambs improved as dietary protein level increased in the diets. The observation on the of CP content on ADBWG in the present study is in agreement with Kanjanapruthipong and Leng (1997), Warly *et al.*, (1994), Hossain *et al.*, (1995) and Thu and Uden (2001) who reported that the level of protein in the diets would improved DMI, digestibility and maximizes efficiency of microbial cell synthesis in the rumen for live-weight gain.

The NDF level of maize stover silage only (control diet) was 46.8% while the other silages with concentrate ranged from 49.7 for diet B (50%C + 50CS) to 54.6% for diet A (75%C + 25% CS). For maize stover silage based diet, Elkholy *et al.* (2009) obtained 48.0% level of NDF for sheep diet. Consequently, the range value 46.8 – 54.6% of NDF obtained in this study for maize stover

silage and concentrate supplements should not be detrimental to the intake and digestibility as reported by Bamikole *et al.* (2004) that value of 55.0 – 60.0% can limit intake of forage. From the dry matter intake (DMI) and growth study trial, the DMI of the diets A (454.5g), B (473.3g) and C (492.1g) increased proportionately as the rate/level of inclusion of concentrate supplements (CS) with silage increased and compared favourably with DMI (523.1g) of diet E (100% CS). However, the DMI (245.8g) of control diet D (100% silage) decreased significantly compared to other silages with concentrate supplements. The metabolic dry matter intake (DMI) followed a similar trend. For the control, (D), A, B, C and E diets translated to – 3.3, 3.5, 4.2, 6.7 and 7.9% respectively dry matter intake per body weight approximately. As such, apart from the control diet, the other four diets met the 3 – 5% DMI per body weight recommended by ARC (1980) and Devendra (1980). The value obtained in this study are lower than the values of 617.2 – 759.1g obtained by Babayemi (2009) while working with a group of WAD sheep fed *Panicum maximum* silage. The DMI is known to be a basic limiting factor in feed utilisation since this determines or controls the quantity of intake of every other nutrient in the feed and therefore invariably the overall performance of the farm animals. The variation observe in the DMI of test diets may be expected. Babayemi *et al.* (2009) reported that plant physical structure and chemical composition are some of the most vital factors governing intake. However, forage quality is best defined in terms of animal output when fed alone (Devendra,1997). It has been shown that the feed value of a forage depends on the biomass produced, voluntary intake, digestibility, and growth rate by an animal; which is the best assessed through sole feeding (Devendra,1997). The artificial flavor impose on the diets during fermentation may have caused a shift in palatability.

Furthermore, feed intake is known to be controlled by crude protein level of diet, gut fill, body fat, palatability and changes in body chemical constituents (Ahamefule, 2005). It thus appeared that diets A, B and C silage supplemented with concentrate at varied level that were combined like the diet E (100% CS) had not much physical and chemical characteristics to preclude its consumption. The converse is true for diet D (control) which is ensiled maize stover only. The nutritive value of crop residues depends not only on their digestibility but also on the amount of voluntary intake by an animal. Crop residues, such as maize and most cereal stovers, are nutritionally deficient in nitrogen, and when fed to ruminants as silage require supplementing

with a protein supplement. Maize stover, as a true of other fibre crop residues, is generally deficient in nutrient content and low in digestibility (Doyle *et al.*, 1986). The extent and rate of digestion of fibrous feeds are increased by a nitrogen supplement resulting in a greater dry matter intake (Preston and Leng, 1987). It is well documented that the utilisation of the energy components of such materials by ruminant animals is highly dependent on the efficiency of the fermentative activity of the microbes. For optimum or maximum fermentation on a given diet, a certain level of ammonia (NH₃) concentration in the rumen is required. Otherwise feed intake may be reduced if NH₃ concentration is limiting the rate of fermentation (Mehrez *et al.*, 1977). Animal fed on such materials (i.e ensiled maize stover only) as their sole diet will show low dry-matter intakes and decline in liveweight. This is reflected in the extent of live-weight change. The daily body weight gains of animals on test diets A, and B (33.34 g/d and 40.48 g/d) were statistically similar to diet C (66.55g/d) while BWG of animals on dietary treatment C was similar to animal on experimental diet E (81.94 g/dl). However, BWG of animals on diet D (-2.00 g/d) showed decline in liveweight compared to other dietary treatments. The metabolic weight gain followed a similar trend. There was no significant ($P > 0.05$) difference shown in the initial weight of the sheep in the five diet groups. However, at the termination of the study (105 days) the final weights of sheep fed diets A (75%C + 25%C), 18.50kg, B (50%C + 50CS) 20.75kg and C (25%C + 75%CS) 20.20kg were statistically similar to diet E (100%CS) except diet D (100%C) 14.00kg was significantly lower than the others and the initial body weight. Variation in daily body weight gains of the sheep could be attributed to variation in nutrient supply in the silage and concentrate supplements (Oddy and Sainz, 2002). The negative value of BWG obtained for the animals on diet D indicated that sheep on ensiled maize stover only lost weight. This implied that nutrient contents of maize stover silage is inadequate to support growth, thus there is need for concentrate supplementations. The result indicated that utilisation of ensiled maize stover using 25% concentrate supplements by growing sheep at 75% dietary level of inclusions of maize stover silage was beneficial.

This observation was similar to previous report of Nour *et al.* (1987), that utilisation of corn stover (maize stover) by sheep was greatly improved by adding 30% concentrates at 70% dietary level of inclusion of maize stover was beneficial and economical. The feed conversion ratio (FCR) differed across the treatment means, meaning that inclusion of ensiled maize stover in

each dietary treatment had effect on the efficient utilisation of the feed by the animals. The higher the feed conversion ratio, the less desirable or efficient is a treatment or diet. Feed conversion ratio (FCR) is a measure of an animal's efficiency in converting feed mass into increased body mass. Animals that have low FCR are considered efficient users of feed. The higher FCR was recorded in diet A (75%C + 25%CS) 13.63 which implies that the animals utilised the supplied feed with least efficiency. However, the best efficiency was obtained in diet E (100%CS) 6.39 and C (25%C + 75%CS) 7.39 in that order. The control diet as a reference with FCR value of - 12.94 was statistically differed from other treatments with negative value. The negative values obtained for BWG and FCR was inclined to - 53g/day live weight change obtained by Sudana and Leng (1985) when they fed lambs wheat straw without concentrate supplements. The BWG and FCR for the sheep on the 100 % ensiled maize stover (control) was negative due to the low level of crude protein (CP). Some data for sheep illustrate variation of FCR. A feed conversion ratio (FCR) (kg feed dry matter intake per kg live mass gain) for lambs is often in the range of about 4 to 5 on high-concentrate rations (Knott, *et al.*, 2003; Brand *et.al.*,1991 and National Research Council, 2007), 5 to 6 on some forages of good quality (Fahmy,et al., 1992), and more than 6 on feeds of lesser quality (Malik *et al.*, 1996). On a diet of straw, which has a low metabolisable energy concentration, FCR of lambs may be as high as 40 (Cronje, 1990). Other things being equal, FCR value tends to be higher for older lambs (e.g 8 months) than younger lambs (e.g 4 months) (National Reseach Council 2007). Common FCR values for cattle and sheep grazing pasture are around seven to ten whereas pigs and poultry on complete grain-based rations can be two or lower. This reflects the digestion systems of pigs and poultry, which are monogastric (one stomach) omnivores compared to ruminants which are herbivores with four stomachs designed to digest fibrous plant material. Faster growing cattle, sheep and goats typically have a better feed conversion ratio than those growing slower. This is because feed used for maintenance is lower overall than for a slow growing animal.

5.7.2 Digestibility and Nitrogen Balance

The level of dry matter intake (DMI) increased as the level of concentrated supplements increased. This indicates that the feeding of protein supplements with silage diets increases intake of silage. The ranged value of 437.50 to 694.41 gDM/day from the current study is relatively close to the ranged values of 617.20 to 759.10 gDM/day reported by Babayemi (2009)

for Guinea grass silage fed to WAD sheep. A possible mechanism for a direct effect of nitrogen (N) on intake is that low N supply reduces digestibility of the fibre component of the diet which in turn leads to decreased intake due to rumen fill effects (Rook and Gill, 1990). Alternatively inadequate N supply to the animal's tissues may create an effective energy surplus leading to decreased intake (Egan, 1965). Furthermore, the high dry matter intake (DMI) of sheep on silages with different levels of concentrate supplements could be due to high CP content. The level of DMI is influenced by several factors, such as body composition of animals (composition of body fat), environmental conditions especially climate, genetic factors, weight of animals, type of management, feed composition and quality (ARC,1980). Dry matter intake was high. However, it has been observed that DMI could be favourably influenced by CP level (Karim *et al.*,2001; Karim Santra, 2003). Overall, DMI of sheep were within 310 to 870g/day values reported by ARC (1980) and McDonald *et al.* (1987) as adequate for sheep with body weight of 20 to 35 kg. However, NRC (1985) reported that DMI could go up to 1000 to 1300g/day for growing sheep. Some factors, e.g low pH (Shaver *et al.*, 1985) as well as high contents of acetic acids (Wilkins *et al.*,1971) and lactic acids (McLeod *et al.*,1970) have been attributed to the reduced intake of silage.

Food and agriculture organisation (1995) classified digestibility of feed as; high (>60%), medium (40-60%) and low (<40%). Apparent digestibility was high for all the nutrients except the medium and low values obtained for CF, ADF, ADL and ash. The apparent dry matter digestibility (DMD) of diets (the four silages and concentrate supplements) were similar statistically. Non-significant values obtained in current study for DMD indicates that silage digestibility in the current study reacts similarly to the concentrate supplements as did the forage in the study of Morris *et al.* (2003). However, the diet C (25%C + 75%CS) had the highest DMD (67.44%) followed by diet B (50%C + 50%CS), 66.32% and least, in diets D (100%C) 62.18% and E (100%CS) 62.18%. These values were in consonance with the work of Nour *et al.*, (1987) and Elkholy *et al.* (2009) who obtained 50.25 – 64.01% and 68.02 – 70.55% respectively. The values were however, greater than the 53% and 57% obtained by Alhassan (1984) and Olayiwole *et al.* (1978) when treated and untreated maize stover and sorghum stover silages were fed to goat, sheep and cattle. More recently, in a Guinea grass silage study with WAD sheep, Babayemi (2009) obtained a digestibility value of 61.80 – 73.50%. Values of DMD obtained in the present

study suggest that dry matter digestibility is not a limiting factor and that when maize stover silage is used in ruminants with concentrate supplements; it could be of benefit to the ruminants. Similar suggestions have been given by Nour *et al.* (1987), Siulapwa and Simukoko (2000), Preston and Leng (1987), Syomiti *et al.* (2009) and Jianxin (2002). This further supports the need for the inclusion of maize stover silage as a potential feed resource in Nigeria and could form an important feed base for smallholder farmers.

The OMD ranged values of 73.00 – 83.50% is consistent with the literature ranged value of 72.86 to 78.41% by Elkholy *et al.* (2009) and el-Shinnawy *et al.* (1999) but greater than ranged value of 51.81 – 65.22% reported by Nour *et al.* (1987). The observed difference in values obtained in this study and in literature might due to variation in levels of concentrate supplements and additives used. The digestion coefficient values for crude protein were significantly higher ($P < 0.05$) in group (A, B, C and E). Moreover the DCP was significantly higher for maize stover silage diets containing 25, 50, 75 and 100% concentrate supplements than maize stover silage without supplements. The high intake resulting in higher protein digestibility may be connected to the nature of the silage and the levels of concentrate supplements. High crude protein in the diet has been considered an important factor that enable high intake of the silage. Crude protein digestibility was higher than 47.20 % reported by Taiwo *et al.*, (1995). Digestibility of CP often increases as CP intake decreases because metabolic faecal N usually makes up a larger part of faecal N at low intake than at high intake (Wheeler *et al.*, 1995). This indicates that concentrate supplements had significant effect on DCP of the maize stover silage.

The crude fibre digestibility, though not significant, but ranged from 35.09 to 58.56 ($D > A > E > B > C$). These values are lower than range values 47.68 – 63.34 and 50.56 – 64.10 reported by Nour *et al.* (1987) and Elkholy *et al.* (2009) respectively. Several factors are known to affect fibre digestion including level of intake (Scholljegerdes *et al.*, 2004) and interactions with supplementation strategy (Hannah *et al.*, 1990; Loy *et al.*, 2007). Decreasing fibre digestibility is generally associated with decreased forage intake (Allen and Oba, 1999). However, the higher DCF of diet D (100%C) might due to proliferation of fibrolytic microorganism or increase in activities of fibre digesting fungi and fibrolytic bacteria in rumen of the sheep on that feed due to

adaptation and nature of diet. The DEE ranged values (52.39 – 69.99) obtained in this study is higher than ranged value (44.79 – 59.61) obtained by Nour *et al.* (1987) but lower than ranged value (81.83 – 85.63%) for maize stover silage treated with yeast and urea, fed to lamb in the work of Elkholy *et al.* (2009) and Sabbah *et al.* (2006).

The digestibility coefficient values of NFE did not show any significant differences for maize stover silages with varied level of concentrates compared to silage without concentrate supplements. However, the value (84.26%) obtained for diet B (50% C + 50% CS) was higher than value (74.51%) observed for diet D (100%C). These results agreed with Sabbah *et al.* (2006) and Elkholy *et al.* (2009) who reported that the digestibility (%) of cross bred Rahmany male lambs fed on rations containing corn stover silage with 5g yeast/head/day increased (80.34) for NFE.

The digestibility coefficient value of neutral detergent fibre (NDF) was significantly similar across the treatments. Similarly, there were no significant ($P > 0.05$) differences observed for DADF and DADL. The fibre fractions were relatively digested by WAD sheep. This is expected since sheep is a ruminant. The hemicellulose and cellulose in the diet were well digested by sheep.

Apart from energy, nitrogen (N) is also one of the most limiting nutrient for ruminant animal production. Therefore, the incorporation of this nutrient in a feeding system based on low-nitrogen fibrous diet is of paramount importance. The metabolic nitrogen balance (N-balance $W^{0.75}$) of diets ranged from 0.190 – 1.217g/day in the order C > B > E > A > D, all positive N-balance. The N-retention followed a similar trend ranging from 26.46 to 67.15%. The levels of N-retention may be as a result of the high digestibility of nutrients in all the diets containing maize stover silage. N-balance and retention values were high and this shows the potentiality of the diets to enhance N-utilisation. The positive N-balance and retention recorded in this study are indicative of animals gaining weight or conserving nitrogen during the period of experimentation. This therefore suggests that the diets A, B and C (i.e. all silage diets) except diet D which is 100% silage with lowest level of N-balance and N-retention could be deemed adequate. As a further support, the total digestible nutrient (TDN) values ranged from 63.65 to

70.92 (Table16) and varied significantly among the treatments. The silage diets with concentrate supplements were consistently superior to the maize stover silage only (diet D). However, the high level of faecal and urine nitrogen recorded for animals on diets B, C, E and A, C, E may be due to poor utilisation of dietary nitrogen as well as absorbed excess NH_3 excreted as urea. Moreover, when animals are not fasted, the excreted nitrogen in faeces are derived from structural nitrogen of dietary sources while those in urine are mostly derived from broken down microbial protein not utilised by the animals as well as absorbed excess NH_3 excreted as urea (Van Soest,1982). The relatively high proportion of nitrogen intake excreted in faeces and urine for diets A, B, C and E is an indication of the poor quality of its protein, since it was not properly digested (those in the faeces) or it was wasteful broken down and excreted in the urine (Fraser *et al.*,2000).

For metabolisable energy (ME) values ranged from 10.95 to 12.68 MJ/Kg DM. It is not the total energy in a feed that is important, but the amount of energy that can be used by the sheep. This is known as metabolisable energy. The values of ME obtained for ensiled maize stover and concentrate supplements is in line with the value of 12MJ/KgDM reported for good-quality grass by Gatenby (2002). According to FAO (1995), the energy value of silage and the efficiency of its utilisation, are largely determined by the relative balances of glucogenic energy, long chain fatty acids and essential amino acids absorbed by the animal. It could then mean that this diet contained a balance of nutrients, which efficiently interacted to give the highest average daily gain. Sainz and Wolff (1990) reported that the rate of fat deposition relate more to the amount of energy available in excess of requirements for maintenance and lean growth.

CHAPTER SIX

EXPERIMENT FOUR

6.0 HAEMATOLOGICAL PARAMETERS AND BLOOD CHEMISTRY OF WEST AFRICAN DWARF SHEEP FED ENSILED MAIZE STOVER AND CONCENTRATE SUPPLEMENTS

6.1 Introduction

To meet the high demand for meat as a source of animal protein in the future, much of the increase in meat production would have to come from short-cycle animals which require a little management practice to rear them. Examples are the domestic goat, sheep and other mini-livestock such as the grasscutter.

Sheep are believed to have evolved in the dry and mountainous regions of Southwest and Central Asia. Present day domesticated sheep were derived from strains of the wild animals in existence in Southwest Asia some 8,000 – 10,000 years ago (FDLPCS, 1991). They can survive in many areas especially in arid tropics where cattle would perform poorly and play a significant role in the economy and nutrition of rural and urban dwellers. In Nigeria where production of small ruminants is largely extensive, and fodder provision erratic throughout the year, survivability of sheep has been low due to poor nutrition. Available reports (Nuru, 1985, Ahamefule *et al.*, 2006) indicate that post-weaning mortality in small ruminants in Nigeria is between 30 – 40%. Among other reasons, poor postnatal weights of kids and lambs, which invariably is linked to poor nourishment has been majorly implicated (Ahamefule *et al.*, 2006).

Furthermore, carcass products and by products are important indicators of type and level of feeding (Usuhor *et al.*, 2009). Nutrition is one of the most important factors in production. So animals on good plane of nutrition regardless of breed are likely to dress out better (Warris, 2000). However, the high cost of conventional feed ingredients which has resulted to high cost of animal production (Taiwo *et al.*, 2003) limits profitable production. Thus the need for search and usage of unconventional feedstuffs which if properly harnessed can constitute potential feedstuffs for livestock feeding. Harvested crop residues constitute an important feed for ruminants. Estimates in Africa alone show that more than 340 million tones of fibrous crop residues are produced annually most of which are unutilised (Kossila, 1984). Maize stover is a residue from maize plant after harvested the cob. The residues are estimated to be about 4.11

million tones per year (Adebowale, 1989). Ruminants have potential to utilise the stover but the stover dries up quickly. There is thus a need for preservation. Ensiling has been reported to effectively conserve forages and fodder crops (Babayemi, 2009). Silage can be an economical source of nutrients for sheep and goats especially on large farms where feeding can be mechanized (Susan, 2009).

Haematological and serum biochemical values have been considered useful for the evaluation of body condition and the nutritional and immune status in animal where other tissue related measurements are not available (Ogunrinde *et al.*, 1981; Bush 1991 and Ogunsanmi *et al.*, 1994). The significance of determining haematological and biochemical indices of domestic animals have been well documented (Oduoye and Otesile, 1977), and changes of these parameters have been studied in cattle (Gheregariu *et al.*, 1984), Sheep (Oduye and Adedevoh 1976) and goat (Ahamefule *et al.*, 2006). When blood is examined, it provides a good opportunity to clinically investigate the presence of several metabolites and other constituents in the body of an animal. This is in line with the recommendation of WHO (2002) on the use of blood biochemical values in medical nutritional assessment. Blood examination is also a good way of assessing the health status of an animal as it plays a vital role in the physiological, nutritional and pathological status of an animal/organism (Onifade, 1993). Haematological and serum biochemical parameters are good indices of the physiological status of animals and changes in the values of these parameters can be used to assess the response of animals to various physiological situations (Esonu *et al.*, 2006). Tewe *et al.* (1981) reported that the importance of investigating blood composition is to have a way of distinguishing normal status from state of stress. Such stress factor can be nutritional, environmental or physical; thus, this study was designed to evaluate the haematological and serum biochemical values of WAD sheep fed ensiled maize stover and concentrate supplements.

6.2 MATERIALS AND METHODS

6.2.1 Collection and Evaluation of Blood Samples

At the end of the experiment, blood samples were collected from the jugular vein of each animal (sheep) for haematology and serum biochemistry analysis. Blood samples for haematology were collected into sterile vacutainer tubes containing EDTA (Ethylene diamine tetra acetic acid) while that of serum separation was without EDTA to allow blood clotting and serum was decanted for the analysis. During the collection, care was taken to avoid contamination with hairs, dirt and micro organism.

6.2.2 Haematology

The packed cell volume (PCV) and haemoglobin (Hb) were determined using micro haematocrit method and cyanmethaemoglobin method as described by Mitruka and Rawnsley (1977). Red Blood Count (RBC) and White Blood Count (WBC) were determined using Neubauer haemocytometer after appropriate dilution (Schalms *et al.*, 1995) and Kelly (1979)..

6.2.3 Serum Biochemistry

Serum total protein was determined using Biuret method as described by Reinhold, (1953) and Kohn and Allen (1995) while albumin was determined using bromocresol green (BCG) method as described by Peter *et al.* (1982). The globulin concentration was obtained by subtracting albumin from the total protein while the albumin/globulin ratio was obtained by dividing the albumin value by the calculated globulin value. Serum urea was determined by urease method and creatine by Folinwu filtrate methods as described by Toro and Ackermann (1975). Also, serum glucose was determined by O-Tuidine method using acetic acids (Cooper and McDaniel, 1970) while serum cholesterol was determined using appropriate laboratory kits (Friedwald *et al.*, 1972, Gowenlock *et al.*, 1988).

6.2.4 Statistical Analysis

The experimental design was completely randomized design (CRD). Data obtained were subjected to analysis of variance (ANOVA) using the procedure of SAS (2003) package to determine the effect of dietary treatments on the various parameters studied. Significant means were separated using Duncan multiple range test of the same software.

6.3 RESULTS

6.3.1 Haematological response of West African Dwarf Sheep fed ensiled maize stover and concentrate supplements

The results of haematological response of the animals are shown in Table 17. The results obtained showed that Packed Cell volume (PVC) and haemoglobin (Hb) concentrations in the blood did not differ significantly ($P > 0.05$) between the dietary treatments. However, there was significant ($P < 0.05$) differences in red blood cells concentration between the dietary treatments such that it was highest in diet A ($3.51 \times 10^6 \mu/\text{ml}$).

The white blood cell (WBC), neutrophil (NEUT), lymphocyte (LYMP) and eosinophil (EOSI) counts of WAD sheep fed the experimental diets were significantly ($P < 0.05$) influenced by the treatments. The mean WBC counts of animals fed diets A and B were significantly higher than those animals that were fed diets C and D but similar to diet E. However, lymphocyte counts of animals fed diet E was significantly ($P < 0.05$) lower to those fed diet A but similar to those fed diets B, and D. The values for EOSI counts were significantly highest in diet E and D while it was lower in diet B. However, values for monocytes counts for animals in experimental diets were similar ($P > 0.05$) across the treatments.

Table 17: Haematological parameters of WAD sheep fed ensiled maize stover and

concentrate supplements

Parameters	Dietary treatment					SEM
	A	B	C	D	E	
PCV (%)	22.50	30.33	27.80	29.50	28.25	3.19
HB (g/dl)	8.30	10.07	9.24	9.80	9.33	1.05
RBC ($\times 10^6 \mu\text{l/ml}$)	3.51 ^a	2.39 ^c	3.19 ^{ab}	2.79 ^b	3.44 ^a	2.09
WBC ($\times 10^3 \mu\text{l/ml}$)	11.51 ^a	11.08 ^a	8.66 ^b	8.58 ^b	10.31 ^{ab}	2.21
NEUT (%)	43.00 ^b	56.33 ^a	46.60 ^b	57.00 ^a	55.25 ^a	4.80
LYMP (%)	51.67 ^a	37.33 ^b	48.00 ^{ab}	36.00 ^b	34.00 ^b	4.91
MONO (%)	1.67	1.67	1.20	1.00	2.00	0.64
EOSI (%)	3.67 ^b	2.00 ^b	4.20 ^b	6.00 ^a	6.50 ^a	1.33

a b c-Means on the same row differently superscripted are significantly ($P < 0.05$) different.

PCV-Park Cell Volume, Hb-Haemoglobin, RBC-Red Blood Cell, WBC- White Blood Cell,

NEUT-Neutrophils, LYMP-Lymphocytes, MONO-Monocytes, EOSI-Eosinophils

WAD-West African Dwarf, SEM-Standard Error of Means.

Diet A = 75% Control + 25% Concentrate supplements

B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplements

D = 100% Control (Silage only)

E = 100% Concentrate supplements

SEM = Standard Error of Means

6.3.2 Serum biochemistry of West African Dwarf Sheep fed ensiled maize stover and concentrate supplements

The total protein of the WAD sheep fed the experimental diet D was significantly ($P < 0.05$) lowest among the treatments means while the serum protein of animals on diet E was significantly ($P < 0.05$) highest between the dietary treatments but similar to animals fed diets A, B and C.

The mean serum albumin, albumin/globulin ratio and creatinine of the animals assessed in this study, were not significantly influenced by the dietary treatments. However, serum urea, cholesterol and glucose were significantly ($P < 0.05$) affected. The mean serum urea of animals fed diet B was significantly ($P < 0.15$) higher than animals fed diets A and D but similar to animals on diet C and E. The mean serum cholesterol of animals fed diets A,C,D and E were similar but significantly ($P < 0.05$) higher than for animals fed diet B. There was significant ($P < 0.05$) difference in the value for serum glucose such that it was highest in diet E and lowest in diet B.

Table 18: Serum Biochemistry Constituents of WAD sheep fed ensiled maize stover and concentrate supplements

Parameters	Dietary treatment					SEM
	A	B	C	D	E	
Total protein (g/dl)	13.44 ^{ab}	12.50 ^{ab}	13.50 ^{ab}	9.40 ^b	14.26 ^a	1.23
Albumin (g/dl)	6.02	5.82	5.56	4.77	5.56	0.67
Globulin (g/dl)	7.42 ^a	6.68 ^{ab}	7.94 ^a	4.63 ^b	8.70 ^a	0.58
Albumin/globulin ratio	0.81 ^a	0.87	0.70	1.03	0.64	0.12
Urea (mg/dl)	26.36 ^b	47.27 ^a	34.54 ^{ab}	23.63 ^b	34.32 ^{ab}	4.22
Creatinine (mg/dl)	1.03	1.05	0.97	1.33	1.24	0.16
Cholesterol (mg/dl)	73.40 ^{ab}	62.12 ^b	71.20 ^{ab}	79.50 ^a	73.65 ^{ab}	4.81
Glucose (mg/dl)	69.03 ^c	67.32 ^c	89.10 ^b	106.62 ^a	114.80 ^a	4.07

a, b, c= Means on the same row with different superscripts are significantly ($P < 0.05$) different

Diet A = 75% Control + 25% Concentrate supplements

B = 50% Control + 50% Concentrate supplements

C = 25% Control + 75% Concentrate supplements

D = 100% Control (Silage only)

E = 100% Concentrate supplements

SEM = Standard Error of Means

6.4 DISCUSSION

6.4.1 Haematological Responses of Experimental Animals

The PCV range of 22.50 – 30.33% obtained in this study were within values (22.00 – 37.00%) reported by Sowande *et al.* (2008) and Fajemisin *et al.* (2010) for normal healthy sheep. These values were also relatively close to 29.9 – 33.6% reported by Mitruka and Rawnsley (1977) for clinically healthy sheep except for animals on diet A that had PCV value of 22.57. The Hb values obtained in this study fell within normal values recorded for healthy sheep (Benjamin, 1981; Fajemisin, *et al.*, 2008), an indication that diets B, C, D and E seemed to be capable of supporting high oxygen carrying capacity in the animals.

For RBC, the range of value of 2.39 to 3.51 obtained for the current study fell relatively within the range value of 2.40 – 4.20 reported by Sowande *et al.* (2008). The values for white blood cells (WBC) neutrophils (NEUT) and eosinophil (EOSI) were above normal range reported for healthy sheep (Mitruka and Rawnsley, 1977) while values for lymphocyte and monocyte were within normal range reported by the same authors for clinical healthy sheep. WBC in animals possesses phagocytic function (Campbell and cole, 1986), differential WBC counts were used as an indicator of stress response, and sensitive biomarkers crucial to immune function (Graczyk *et al.* 2003). The higher WBC and differential counts reported in this study indicated that, the WAD sheep seems to possess protective system, providing a rapid and potent defense against any infectious agent. This probably is the physiological basis for the adaptation of this species to this ecological zone characterised by high prevalence of disease.

Furthermore, non significant values of PCV and Hb of WAD sheep fed ensiled maize stover and concentrate supplements diets relative to the control group is an indication that the animals were not anaemic. The PVC and RBC values reported in this study were within the range of values reported by Eniolorunda *et al.* (2005), Sowande *et al.* (2008) and Jain (1993) respectively for sheep in separate experiments, which indicated that the animals used in this study were not susceptible to anaemia related diseases. The Hb values go against the report of Talebi *et al.* (2005) that nutrition affects the blood profile of the animals and this implies that up to 100% inclusion of maize stover silage had a positive effect on the relative quantity of the blood cell as

well as total volume of blood. No indication of leukemia or leukocytosis was observed which showed that the feed was not toxic to the animals and has no adverse effect on their health status.

6.4.2 Serum Biochemical response of experimental animals

Serum biochemical constituents of WAD sheep fed experimental diets are shown in Table 18. Most studies involving the use of combinations of ensiled maize stover and concentrate supplements did not include haematological and biochemical constituents. Therefore, little comparison could be made. The mean ranged values (9.40 – 14.26g/dl) for serum protein examined in the animal for all dietary treatment were above the normal ranged values (5.70 – 9.10g/dl) reported by Mitruka and Rawnsley (1977) and the ranged value (6.3 – 7.2g/dl) reported by Elkholy *et al.* (2009) and El-Ashry *et al.* (2002) respectively. Information regarding nutritional status and malnutrition is often obtained from the total protein (Allinson, 1995). The increased level of serum protein observed in this study may be due to high protein content of the diets except diet D that was low in protein and the serum protein value of animals in the group was within normal range values. However, serum creatinine, globulin and albumin/globulin ratio mean values fell within the normal range reported for healthy sheep by Mitruka and Rawnsley (1977) and Ahamefule and Ozuzu-Ejimogu (2012). However, the range value (4.63 – 9.13 g/dl) of serum globulin obtained in this study was higher than ranged value (3.6 – 4.4 g/dl) reported by Elkholy *et al.* (2009) for sheep fed ensiled green stover and stalk treated with urea and yeast.

The albumin mean range value (4.77 – 6.02 mg/dl) obtained in this study was slightly higher than normal value (2.70 – 4.55 g/dl) reported by Mitruka and Rawnsley (1977) and ranged value (2.7 – 3 g/dl) reported by Elkholy *et al.* (2009) for healthy sheep. The serum urea level (23.63 – 34.54 mg/dl) of animals on diets A, C, D and E in this study fell within the normal values reported (15.0 – 36.0 mg/dl) by Mitruka and Rawnsley (1977) for healthy sheep except for animals on diet B that had serum urea value of 47.27mg/dl. However, the mean ranged values of urea (23.63 – 47.27 mg/dl) obtained was relatively close to range values (29.7 – 40.50 mg/dl) reported by Sowande *et al.* (2008) for normal healthy WAD sheep. Furthermore, the blood urea-N concentration value obtained from this study was also higher than value (19.5 mg/dl) reported by Taghizadeh *et al.* (2007) for sheep fed corn silage treated with 1% urea.

The high level of serum urea observed in animals on diet B may be attributed to excessive tissues protein catabolism (Oduye and Adadevoh, 1976) and high releasing of ammonia in rumen as a result of high absorption of ammonia from the rumen to the blood (Taghizadeh *et al.* 2007) while decreased levels are most commonly due to inadequate protein intake, malabsorption or liver damage. The urea and creatinine concentrations in the blood were used as kidney and liver function test (David and Berdt, 1994). Kung (2010) reported that blood or milk urea nitrogen could be used as an indicator of excess ruminal-degraded protein (RDP).

For serum cholesterol and glucose levels of 62.12 – 79.50 mg/dl and 67.32 – 114.80 mg/dl obtained in this study were within the normal range of 50.00 – 140.00mg/dl and 55.0 – 131.00 mg/dl reported respectively for normal healthy sheep by Mitruka and Rawnsley (1977). However, the serum cholesterol level observed in this study was lower than values (96 – 117 mg/dl) reported by Elkholy *et al.* (2009) for normal healthy sheep while the value reported for serum glucose level was higher than ranged value (59 – 65 mg/dl) reported by Elkholy *et al.* (2009), Briggs, (1967) and Ayyat *et al.*, (2007) who reported that supplementation of readily available carbohydrates with non-protein nitrogen (NPN) to the basal diet increased the level of blood glucose. Cholesterol is a group of fats vital to cell membranes, nerve fibres and bile salts, and a necessary precursor for the sex hormones. High levels indicate diet high in carbohydrates/sugars while low levels indicate low fat diet, malabsorption, or carbohydrate sensitivity. Glucose is the chief source of energy for all living organism. The levels obtained across the dietary treatments suggest that feed was adequate in energy supply for the animals. Generally, levels of biochemical blood profile of WAD sheep fed ensiled maize stover and concentrate supplements were within reported range values for same species by Oduye and Adadevoh (1976) and Elkholy *et al.* (2009).

CHAPTER SEVEN

7.0 SUMMARY, CONCLUSION AND RECOMMENDATIONS

7.1 Summary of Findings

The potential effect of additives on nutritive value of ensiled maize stover (*Zea mays* L) an annual crop grown in Nigeria and other part of the world was examined. This crop (corn) is grown mainly for grain and it is the general farm practice to harvest the cob in green (fresh) or dry and leave the stovers/straw in the field to dry completely. The dry maize stover (straw) therefore is not for animal feeding and is only used as fuel. However, there is a shortage of feed in the country and the utilisation of ensiled maize stover (crop residue) and concentrate supplements can easily fulfil the perennial shortage of quality and quantity of forage grass being experienced in Nigeria and other developing nations of the world. The incessant increase in prices of conventional feedstuffs are some of the the factors hindering the adequate production of livestock in Nigeria. Recent efforts are therefore, geared towards the use of alternative sources, which are, less expensive, not in competition with man as feedstuff and are readily available in each locality.

Experiment 1 involved the preparation of silages from wilted maize stover with or without three additives (Molasses, Honey and Sugar) giving silages MS (control), MSM MSH and MSS. Thereafter, the physical characteristics (colour, odour and texture), temperature, pH, chemical composition and dry matter intake (DMI) and acceptability of silages prepared were assessed by West African Dwarf Sheep using the coefficient of preference (CoP).

In experiment 2, the nutritive value of the ensiled maize stover with or without additive was investigated using the in vitro technique, with a view to adopting as a forage basal diet or supplements to livestock in the dry and wet season. Experiment 3, the assessment of utilisation of maize stover silage (control) and concentrate supplements (vis-a-vis the intake and growth) by West African Dwarf Sheep at 75% silage and 25% CS, 50% silage and 50% CS, 25% silage (C) and 75% CS, 100% silage (C) and 100% of concentrate supplements (CS) over a 105-day feeding period. It also involved the digestibility and nitrogen balance of ensiled maize stover (control) and concentrate supplements (CS) fed to growing West African Dwarf Sheep.

Investigated in experiment four was the haematological parameters and serum biochemistry of the West African Dwarf sheep fed the ensiled maize stover and concentrates supplements diets. The MS (control) silage (i.e. maize stover silage without additive) had a low pH of 3.6 similar to those with additives, fruity odour greenish brown colour and firm texture, while the other silages with varying additives had a low pH ranging from 3.5 – 3.7, pleasant, fruity and alcoholic odour, greengage colour and firm texture. Chemical composition of fresh/unensiled maize stover gave 31.1% dry matter, 9.3% crude protein, 32.3% crude fibre, 69.2% neutral detergent fibre, 57.5% acid detergent fibre, 16.5% acid detergent lignin, 11.7% hemicelluloses and 41.6% cellulose. The chemical characterisation of maize stover silages were dry matter, 31.6% to 35.3%; crude protein, 7.9 to 8.6%; crude fibre, 30.0 to 31.9%; ash, 6.9 to 7.2% neutral detergent fibre, 68.6 to 69.9%; acid detergent fibre, 56.1 to 63.2% acid detergent lignin 14.0 to 16.8%; hemicelluloses, 10.81 to 12.49% and cellulose, 39.3 to 44.8%. Silage diets MSS and MS (control) were acceptable by WAD sheep with coefficient of preference above unity ranging from 1.03 to 1.02 DM/day.

In vitro fermentation studies showed that ensiled maize stover with sugar (T₅) had the highest TGV, CH₄, OMD and SCFAs. The significant least quantities of these parameters were from molasses ensiled maize stover (T₃), Dry matter degradability (DMD) was significantly influenced by the various additives except for molasses additives which did not differ significantly from the control. Results of feed utilisation in experiment four showed that diet B (50% control silage (C) and 50% concentrate supplements (CS) provoked the best performance in terms of N-retention (67.2%) digestibilities of dry matter (66.3%), neutral detergent fibre (59.7%), acid detergent fibre (45.7%) and acid detergent lignin (48.5%) and total digestible nutrient (70.9%). This treatment was closely followed by diet C (25% Control silage and 75% concentrate supplements) and least in D (100% Control silage).

Feed performance results in experiment five clearly put diets C (25% Control silage and 75% concentrate supplements) at par with diet E (100% concentrate supplements) in terms of daily body weight gain, dry matter intake, metabolic daily body weight gain and feed conversion ratio while performance indices of animals on diets A, B and C were similar. However, diet D (100% Control silage) could not support growth rather animal on that treatment lost weight and dry

matter intake was very low among the dietary treatments. The results of haematological and serum biochemical constituents of WAD sheep fed ensiled maize stover and concentrate supplements showed no adverse effect on the health status of the animals at 100% level of maize stover silages.

7.2 Conclusion

In conclusion, the control silage (i.e. maize stover silage without additives) exhibited good quality characteristics similar to those prepared with additives suggesting that good silage can be prepared from maize stover without additive, since it contained enough soluble/a readily fermentable carbohydrate. To conserve maize stover as silage, pre-wilting is a must which can be carried out under shade and airy atmosphere within 24hrs.

The ensiled maize possessed low crude protein, varied dry matter and ash content and similar NDF and ADF. *In vitro* digestibility results demonstrated that net gas production, organic matter digestibility (OMD), metabolisable energy (ME), short chain fatty acid (SCFA) and dry matter degradability varied based on the type additives and the control silage compared well with fresh/unensiled MS confirming the earlier statement that maize stover can be converted as silage without using additive. The result obtained further demonstrated that from the chemical profile and net gas production, estimates of nutrients digestibility (OMD, NDF and DMD) of maize stover and other crop residues in Nigeria could be readily accomplished by *in vitro* gas production technique.

High values of digestibilities of organic matter and fibre fraction of silages and concentrates supplements diets together with positive nitrogen retention denote the ability of silage with concentrate supplements to meet the nutrients requirement of stock. Optimal performance of feed utilisation was attained with livestock placed on silage and concentrate supplements suggesting efficient feed utilisation. However, the low dry matter intake and decline in weight observed for animals on a sole silage diet (control) denote that maize stover silage without concentrate supplementation cannot support growth let alone production due to inherent low nitrogen level as indicated in chemical composition coupled with its fibrous nature being a crop residue. This study demonstrated that ensiled maize stover and concentrate supplements can be incorporated in

sheep diets as alternative feed source at ratio 3:1 without negative effect on the animal's weight gain and protein metabolism. The inclusion level of maize stover silage is safe and economical in diets for sheep production without any adverse effect on haematological parameters. Finally, this investigation demonstrated that available maize stover in Nigeria have potential as ruminant livestock feedstuffs if properly harnessed.

7.3 Recommendations

Based on the scientific evidence available from this study, the following recommendations are considered necessary:

- Since maize stover is available during the planting season in Nigeria, its harvesting and preservation as silage (for a ready use) will go a long way to curtail its wastage on farmland and improve effective land utilisation.
- The use of non-protein nitrogen (NPN) additive especially to high-dry matter low buffering forages such as maize stover is a pre-requisite to increase the crude protein content and to improve aerobic stability at feed out.
- Silage prepared from maize stover is nutritionally adequate as a forage basal diet for sheep feeding at least up to 75% level of inclusion.
- Evaluation of ensiled maize stover on other physiological states (like pregnancy and lactation) and other ruminants should be studied in future.
- More works should be done on haematology and serum biochemical profile of WAD sheep fed ensiled maize stover.
- There is need to study and compare the effects of feeding fresh maize stover or hay with the ensiled form.
- Comparison of feeding quality of ensiled green and dry maize stover study should be done.

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