ASSESSMENT OF IMPROVEMENT OF CHEMICALLY AND BIOLOGICALLY TREATED MAIZE STOVER AND SUBSQUENT UTILIZATION OF TREATED MATERIALS FOR BEEF PRODUCTION

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ABSTRACT

Two studies were conducted to evaluate effectiveness of CaO, enzymes, microbes and $Ca(OH)_2$ in improving the feeding value of maize stover (MS) for beef animals. Study I involved five treatments; untreated MS (T_1), CaO treated MS (T_2), CaO + enzymes treated MS (T_3), CaO + microbes treated MS (T_4), CaO + enzymes + microbes treated MS (T_5). Study II involved untreated MS (T_6) and MS treated with Ca $(OH)_2$ (T_7). The treatments were evaluated for chemical composition, degradability, gas production, digestibility and animal performance. Differently treated MS were fed to 80 steers and 32 steers in study I and II respectively. There was increase in CP, CF, ash and calcium while NDF was reduced in treated MS. Disappearance values at 48hrs ranged 40 to 51% units for dry matter (DM) and 40 to 47% units for organic matter (OM). The `**a**' values for DM and OM degradability were 3.17% and 9.44%, 1.02% and 4.71% in untreated and treated MS respectively while the `**c**' value was 0.012%/h and 0.02%/h, for treated and for untreated MS. Gas production values at 24hrs ranged 22 to 35 ml/g DM. The `**b**' values were 36.2% in untreated and 42.54% for treated MS. T_3 had the highest *In vitro* organic matter digestibility (OMD) (55%) while T_5 had the lowest (48.2%). T_1 had the highest Metabolizable Energy (ME) (7.2 MJ/kg DM) while T_4 had lowest (5 MJ/kg DM). Dry matter intake, final body weight and average daily gain were higher for T_1 and T_6 than T_2, T_3, T_4, T_5 and T_7. The steers on T_7 had higher DM digestibility (70.2%) than those on T_6 (55.7%). It is concluded that the lime treatment was effective in solubilizing the cell wall constituents and improve the digestibility of MS but had a negative effect on intake and body weight gain.

DECLARATION

I, PANTALEO VINCENTI MUSHI, do hereby declare to the Senate of Sokoine University of Agriculture that this dissertation is my own original work done within the period of registration and that it has neither been submitted nor being concurrently submitted in any other institution.

________________________________ ____________________________

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Prof A. E. Kimambo Date

(Supervisor)

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LIST OF ABBREVIATIONS AND SYMBOLS

a	Fitted constant for the rapidly degradable feed fraction
ADF	Acid detergent fiber
ADG	Average daily gain
AIA	Acid insoluble ash
ANOVA	Analysis of variance
b	Slowly degradable DM or OM fraction
C	Rate constant at which b is degraded
CF	Crude Fiber
Cm	Centimeter
CP	Crude Protein
DASP	Department of Animal Science and Production
DED	District Council Executive Director
DM	Dry Matter
DML	Dry Matter Loss
ED	Effective degradability
EE	Ether extract
FCR	Feed conversion ratio
FE	Feed efficiency
FWT	Final weight
g	Gram
h	Hour
IVDMD	*In vitro* dry matter digestibility
IVGP	*In vitro* gas production

IVOMD	*In vitro* organic matter digestibility
IWT	Initial weight
Kg	Kilogram
ME (expressed in MJ)	Metabolizable energy (expressed in Mega Joules)
MS	Maize stover
N	Nitrogen
NARCO	National Ranching Company
NDF	Neutral Detergent Fiber
NFE	Nitrogen free extract
NRC	National Research Council
OM	Organic Matter
OMD	Organic matter digestibility
OML	Organic Matter Loss
SUA	Sokoine University of Agriculture
T_1	Untreated maize stover harvested 2012
T_2	Maize stover treated with Calcium Oxide
T_3	Maize stover treated with Calcium Oxide + enzymes
T_4	Maize stover treated with Calcium oxide + microbes
T_5	Maize stover treated with Calcium Oxide + enzymes + microbes
T_6	Untreated maize stover harvested 2013
T_7	Maize stover treated with Calcium hydroxide
VFAs	Volatile fatty acids
W	Week
$W^{0.75}$	Metabolic body weight
WT	Weight

CHAPTER ONE

1.0 INTRODUCTION

1.1 Background Information

The demand for quantity and quality ruminant feed resources has increased steadily over the past 50 years and it is projected to continue to increase, albeit at a diminishing rate compared with the previous century. In Tanzania, ruminant animals have access to quality natural pasture only for a short period (wet season) implying that the large proportion of animal feeds over eight months per year comes from low quality hay and crop residue such as cereal straws and maize stover (Kilongozi, 1992).

Crop residues have been reported to be a vital dry season feed resource for dairy and beef ruminants (Shem, 1993). These residues are produced in large quantities in many countries. In mixed crop – livestock farming system such systems are limited, native grasses are seasonally available and ruminants graze on marginal land/ or on road sides to obtain feeds during the rainy season. During the dry season when pastures are scarce, crop residues and farm wastes represent an important source of feeds for ruminants (Aredo, 2006). Maize stover which is abundant after crop harvest have extensively been used in ruminant feeding to substitute natural grass especially during the dry season. The common practice of using the residues from the maize and other cereals after grain harvest is to allow ruminant animals to graze freely in the field (*in situ*).

In some areas, the residues are harvested and taken to the homesteads. The cereal stover which are produced have poor feeding quality especially when harvest is late in the season. In this period, maize stover are highly indigestible due to high levels of cell wall materials especially lignin which inhibits microbial digestion of cellulose and hemicelluloses (Klopfenstein and Owen, 1981). The low content of nitrogen and deficiency of readily available carbohydrates in the stover also inhibit the rumen microbial activity (Kimbi, 1997). The barrier can be removed through chemicals or biological treatment that, increase digestibility by decreasing strength of bonds between lignin and polysaccharides (Van Soest *et al.,* 1991; Kilongozi, 1992; Lee, 1997; Kilasi, 1998; Russell *et al*., 2011; Peterson, 2014).

Several attempts have been made in various parts of the world to improve utilization of the stover, and the most successful experiments involve the use of chemical treatment. The use of sodium hydroxide for treatment of crop residues results in physical redistribution of lignin and hemicelluloses within the plant material and changes the cellulose crystallinity (Banerjee *et al*., 2012). The use of sodium hydroxide results in increased hemicelluloses solubility and an increased rate of cellulose and hemicelluloses digestion by rumen microbes (Klopfenstein and Owen, 1981). The resulting combination of increased rate and extent of digestion of cellulose and hemicelluloses in the rumen enhances the energy value of the crop residue.

Chemical treatment of stover using NaOH in Tanzania is not economically feasible as chemicals are expensive and unavailable to livestock keepers. Similarly the use of urea to improve feeding values of stover was practiced in Tanzania in late 80`s and early 90`s. Adoption of the method has been limited due to rise in cost of urea. The use of ash to improve feeding values of maize stover was also used in Tanzania but due to labour intensive required in collection and preparation has made it to be one of the expensive chemical and the crude protein of the treated roughage was slightly reduced (Kimario, 2003). So the use of calcium oxide is an alternative chemical reagent for treating stover and can be substituted for sodium hydroxide. Calcium Oxide (CaO) powder also known as quicklime is one of the more cost effective sources of alkali (Peterson, 2014). When combined with water, CaO is hydrated to $Ca(OH)_2$, which is an exothermic reaction resulting in heat generation. Effectiveness of treatment of maize stover with calcium oxide is influenced by treatment time, temperature, calcium hydroxide loading, water loading, and biomass particle size (Shreck, 2013).

Treatment of stover with 7% CaO and addition of water to 50% DM resulted in nine times greater release of glucose and xylose after incubation with cellulase (Kaar and Holtzapple, 2000). Short term studies indicate that maize stover when treated with 5% CaO and added water to 50% dry matter can replace maize silage in diets for lactating dairy cows to at least 25% of the total ration (Donkin *et al*., 2012). CaO treatment swells the cellulose, which makes it easier for the enzymes to work, such that the hydrogen bonds between lignin and hemicellulose are hydrolyzed. The solution loosens the chemical bonds between the stover's lignin and its more digestible components which enables natural enzymes in the cattle's rumen to effectively digest the stover. In other countries like United States of America CaO has been used in treating maize stover for beef production where the steers could gain net 1705.5 g/day (Russell *et al.,* 2011).

Parallel to chemical treatment, biological treatment of maize stover based on the use of enzyme or microbes have shown to improve palatability and degradability potential and keeping quality of the feed material (Milligan *et al*., 1995). Studies by Lui and Ørskov (2000) demonstrated that enzyme treatment hydrolyse cellulose into monosaccharides thus; improve nutritional value of straw. Maize stover treated with fungi was observed to improve degradation of lignin contained in the maize stover that consequently released nutrients for animal utilization (Lee, 1997). Improvement of the feeding value due to various treatments have been evaluated using changes in chemical composition (Shreck *et al*., 2013), Dry matter (DM) and Organic matter (OM) degradability (Ørskov *et al.,* 1980), *In vitro* and *In vivo* digestibility (Menke and Steingass, 1988), feed intake and live weight gain (Russell *et al*., 2011).

Despite the importance of CaO, enzymes, microbial material and Calcium hydroxide in treating maize stover for ruminant feeding, very limited researches have addressed improvement of nutritional value of maize stover for beef cattle in Tanzania.

1.2 Overall Objective

The main objective is to evaluate the effect of different treatments with Calcium Oxide, enzymes, microbes and Calcium hydroxide on the improvement on feeding value of maize stover.

1.2.1 Specific objectives

(i) To determine the effect of treatments of maize Stover with Calcium Oxide, enzymes, microbes and Calcium hydroxide on chemical composition of the treated material.

(ii) To evaluate the effect of treatments of maize stover with Calcium Oxide, enzyme, microbes and Calcium hydroxide on rumen degradability and *in vitro* gas production.

(iii) To evaluate the effect of treatments of maize stover with Calcium hydroxide on *in vivo* digestibility.

(iv) To assess production performance of beef cattle fed rations based on differently treated maize stover.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 General View

The purpose of this review is to collect information on the utilization of maize stover and methods used in the improvement of its feeding value. The review covers factors that affect the use of maize stover as ruminants feed, techniques used to improve the nutritive value of maize stover and different methods used in the treatment of stover. It also covers the use of different chemicals (sodium hydroxide, urea and calcium hydroxide). The chemical composition of the differently treated maize stover, degradability*, in vitro* gas production, digestibility, intake and performance of animals' fed on differently treated maize stover are also covered in the review.

2.2 Role of Maize Stover as Potential Source of Feed for Ruminant Animals

Maize stover is produced in large quantities in many countries after maize grain harvesting. This material is utilized as a source of feed for ruminant animals in various parts of the world (Kilongozi, 1992). In Tanzania maize is the most important cereal crop grown for human food. Its production averaged 4.5 million metric tons in year 2011/12 season (MOAFSC, 2011). Tanzania is endowed with more than 3.3 million hectares of land with suitable climate for the production of maize. The current average yield per hectare is between 1.2 - 2 tones (MOAFSC, 2011) (Table 1). Maize production has been increasing from year to year due to increase in human population.

Using the ratio of 1:2 between grain and amount of crop residues produced, suggest that about 9 million tons of maize stover and maize cobs are produced annually (Kategile *et al*., 1981).

Table 1: Maize Production and yield trends for Tanzania from year 2003/04 to 2009/10

Year	2003/04	2004/05	2005/06	2006/07	2007/08	2008/09	2009/10
Prod. Tons	3 232 400	3 218 540	3 423 025	3 302 058	3 555 833	3 326 200	4 475 416
Export tons	0	0	0	50 000	50 000	150 000	44 690

Source: (MoAFSC, 2011)

This biomass could be utilized as animal feeds if properly harvested and treated to improve its utilization. Among the crop residues which are estimated to contribute about 25% of the total feed energy suitable for ruminant livestock in both developed and developing countries, maize stover has been considered the leading crop residues (Kossila, 1985). Due to the realisation that land development and expansion of crop farming will continue to reduce the available grazing land, efforts are being made to use the crop residues that become available after harvest usually at the onset of dry season. Studies by Kategile *et al.* (1981) indicated that maize stover are potentially valuable feeds for ruminants whose efficient utilization can result in appreciable production improvement in various classes of livestock.

2.3 Factors Affecting the Utilization of Maize Stover as Ruminant Feed

2.3.1 Physical characteristic of maize stover

Maize stover like other cereal crop residue is produced as by product of grain production. It is a coarser material than straws from small grain cereals. It is made up of stalk 46.2%, leaf sheath 11.7%, ear bracts or husks 9.6%, tassels 0.6%, leaves 18.5% and cobs 13.4% (Candice *et al.,* 2010). The stem is coarse tough structure which hold the plant. The leaf sheath is a structure which closely envelops stem. Husks or ear bracts are the outer covering of maize cobs. Tassels are the tuft head of maize stover. Leaves are flattened structure of the stover and typically are blade like. The physical nature of maize stover which is dry and coarse may affect both chewing and grinding energy (Benerjee, 1982). The stalk require more chewing and grinding energy than other parts (leaves, leaf sheath, husks and tassels). The leaf blades have high organic matter digestibility compared to leaf sheath and stalk because of higher fiber digestibility (Tolera and Sundstøl, 1999). The stalk and leaves of the growing plants gradually decrease in digestibility and content of the desirable constituent's protein as plant nutrients.

2.3.2 Chemical characteristic of maize stover

Like other fibrous materials, maize stover is composed largely of polysaccharides, lignin, protein and other organic and inorganic materials. Quantitatively hemicelluloses and cellulose are the most abundant structural carbohydrates in mature tissues. It has high content of cell walls (Sundstøl, 1988). Cell walls contain relatively large amounts of lignin up to 10% of the dry matter. The protein content of maize stover is slightly higher than that of most grain straw (4.7 – 5.4%) (Juma *et al.*, 2006) (Table 2). Crude protein content of maize stover is below threshold of 7% required for rumen microbial activity (Devendra, 1988). Several studies the results of which are summarized in Table 2, shows that generally crude protein content of maize stover is lower but with high lignocellulose content. High acid detergent fiber (ADF). Low levels of minerals especially Calcium and Phosphorus and their imbalance in nature for maize stover reported by Preston and Leng, (1987).

Maize stover has low energy value compared to other common roughages like maize silage (7.49MJ/kg DM) (NRC, 1996). The essential minerals such as calcium and phosphorus values are relatively low in maize stover (Devendra, 1988). The maturity of maize plant at time of harvest affects the feeding value of forages. As plant mature, the feeding value decreases and the fiber concentration increase with maturity of maize plant

(Rusell, 1986). This is due to 5% increased portion of lignin in the NDF component of the mature maize plant compared to less mature maize plant like maize silage (NRC, 1996). As maize plant matures not only does the NDF portion of total DM increase, but also the cellulose and lignin increase while the hemicelluloses proportion decreases causing the greater part of NDF to be even less digestible. According to NRC (1996) maize silage contains just 41% NDF with 7% lignin, where as maize stover contains 65% NDF with 10% lignin. The maize stover comprise of cellulose in a range of 35 - 50%, hemicelluloses 20 - 35% and lignin 5 - 30% (Lynd *et al.,* 2002).

Table 2: Chemical composition of maize stover (%DM) as reported by different authors

	Source														
	a	b	c	d	e	f	g	h	I	J	k	l	m	n	o
DM	93.4	90.6	-	-	-	-	93	90.2	-	93.4	-	93.9	64.2	94.3	92.6
OM	-	-	-	-	-	-	-	-	-	-	-	82.8	93.6	-	90.4
Ash	12.1	6.0	-	-	-	8.1	-	-	-	6	-	17.2	6.4	10.2	9.7
CP	2.3	2.4	6.29	2.59	5.43	3.5	6.0	2.45	-	4	-	5.9	5.2	7.64	7.4
CF	45.3	-	-	-	-	39.4	-	-	-	-	-	46.1	-	26	-
EE	1.8	-	-	-	-	0.6	-	-	-	-	-	-	-	-	-
NFE	38.4	-	-	-	-	48.4	-	-	-	-	-	-	-	-	-
ADF	-	-	-	60.3	-	-	41.4	-	-	54.1	-	49.9	48.1	26.1	44.9
NDF	-	-	-	69.6	-	-	-	-	-	84.5	-	76.1	78.1	-	72.5
ADL	-	-	-	9.92	-	-	-	-	-	-	-	-	-	-	-
Ca	-	-	-	-	-	-	-	-	-	-	-	-	2	-	-
P	-	-	-	-	-	-	-	-	-	-	-	-	0.2	-	-
ME	-	-	-	-	4.56	-	-	9.2	9	-	6.28	-	-	-	-

a = (Musimba, 1980), b= (Urio, 1981), c = (Tubei,1981), d = (Thairu and Tassema, 1985), e = (Dzowela, 1985), f = (Biwi, 1986), g = (Muthali, 1986), h= (Kimambo *et al*., 1990), i= (McDonald *et al*., 1995), j = (Bengaly,1996), k= (NRC, 1996), l = (Bongoro, 2006), m = (Juma *et al*., 2006), n = (Onaleye, 2012) and o= (Assefa *et al*., 2013).

2.3.3 Digestibility of maize stover

The digestibility of feed is defined as the difference in value between the feed eaten and materials voided by animals, expressed as a percentage of the feed eaten. This is referred to as the “Apparent Digestibility” of the feed, because faeces contain also metabolic excretions composing microbial matter in particular and endogenous secretions. True digestibility, on the other hand refers to the balance between the end of the feed residues in the faeces exclusive of metabolic products (Van Soest, 1994). Thus the overall digestibility of maize stover by ruminants is defined as the summation of the digestibility of the component tissues as affected by the morphology, anatomy and chemical composition of the maize stover.

Maize stover has very low digestibility due to high lignin content which inhibit microbial digestion of cellulose and hemicelluloses. Lignin content increase correlated directly with decrease in cellulose digestibility (Shem, 1993). Lignin had an inhibitory effect on cellulose digestion and therefore, fiber digestibility (Kimbi, 1997). The author concludes that lignin had inhibitory effect on cellulose digestion and therefore, fiber digestibility. The low CP level of maize stover leads to low rumen microbial populations, hence poor intake and digestibility of maize stover (Ørskov, 1986). *In vivo* DM digestibility of maize stover is low ranging from 51 – 59% (Aredo and Musimba, 2003, Ndlovu and Manyame 1989) and organic matter digestibility of 59% (Aredo and Musimba, 2003).

The dry matter (DM) degradability is higher in leaf blade and in leaf sheaths and lower in stem than in other morphological fractions. The washing loss is highest in stem and lowest in leaf blade and husk (Tolera and Sundstøl, 1999). On the other hand, leaf blades have higher degradability of water soluble fraction than leaf sheath, stalk and whole stover.

The lag time is highest in the stalk and lowest in the leaf blades. The morphological fractions differed in the volume of gas produced in the following order: husk > whole stover > stalk > leaf sheath > leaf blade > tassel (Tolera and Sundstøl, 1999).

The digestibility of maize stover is affected by how it is harvested and subsequently handled. Leaves are more digestible than stalk, and thus the height of cutting the maize stover influence digestibility (Massawe and Mrutu, 2005). The authors found that maize stover picked up from the field immediately after grain harvest had higher digestibility by about 2 units than stover that was left in the field for some time. This is presumably due to leaf loss and possibly leaching losses when rain falls on the stover before it is picked up.

Due to its poor digestibility maize stover has traditionally been used as bedding for animals or left in the field to decompose and has not been extensively used as livestock feed (Glassner *et al.*, 1998). With rising maize prices, inclusion of maize stover in beef cattle diets may be an economically feasible option for beef producers. There is critical need to improve the feeding value on maize stover. One approach to increase feeding value is to improve digestibility of poor quality forage by chemical treatment.

2.3.4 Voluntary feed intake (VFI)

Voluntary feed intake is the amount of the feed eaten by animal during a given period of time when an excess of the feed is available. The voluntary feed intake (about1.2 kg DM/100kg live weight) of maize stover is low and as such do not adequately support nutrient intake for maintenance and production. Feed intake is important in defining feed conversion efficiency (FCE). Efficient feed conversion, however will be achieved only if animal is able to obtain from the feed a substantial margin of nutrients over maintenance requirements. In many animal production systems, maximum intake may not be sufficient to ensure maximum production, or may be critical to the system (Aredo, 2006). With ruminants “diet dilution” by undegradable fiber is an important reason for intake being depressed below animal’s potential. Intake of maize stover by animal is low due to animal and feed factors. The low intake of maize stover, despite being more digestible than the other residues, could be due to physical difference of the residue. The stems of maize stover are likely to cause relatively more problems in intake than the other plant parts (Tolera and Sandsøl, 1999).

2.3.4.1 Animal factors affecting intake of maize stover

Intake capacity of animal differs with species, age, sex and physiological status. Cattle consume more barley straw treated with urea ($50g/kgW^{-0.75}$) compared with sheep ($30g/kgW^{-0.75}$) (Silva *et al*., 1989). Intake is related to fasting heat production, which is related to metabolic body weight and is smaller in sheep than in cattle (McDonald *et al.,* 1995). Animal age play an important role in maize stover intake where young animals intake is limited by the coarseness and bulkiness of the stover (McDonald *et al*., 1995). The reason is that young animals have small rumen capacity to accommodate large amount of maize stover and also the young animals rumen is underdeveloped and have low population of microbes which are important for maize stover digestion.

Sex also has necessary role on intake of maize stover. Female animals have lower intake than male animals (McDonald *et al*., 1995). The reason is that male animals have more cell number, are heavier and demand more nutrients than female animals and they have higher capacity to chew (Kimario, 2003). Physiological state of animal affect feed intake in a situation where animal is pregnant intake is affected by two factors which are developing foetus causes increased intake, due to increased need of nutrients requirement (McDonald *et al*., 1995). The effective volume of abdominal cavity is reduced as foetus increase in size in late pregnancy therefore the space for rumen capacity for more feed is minimized however animals adapt by increasing rate of passage. Intake increase with onset of lactation due to high demand of nutrients for maintenance and milk production. Also intake increase due to physical effect resulting from reduction of fat mobilized from the abdominal cavity (McDonald *et al*., 1995).

2.3.4.2 Feed factors affecting intake

Feed factors which affect intake of maize stover are physical form of the maize stover. When maize stover are fed without chopping, more time is required for animal to break down as compared to less time on maize stover chopped in small pieces. Reduction of particle size increases the surface area and enhance break down of the feed by the rumen microbes (McDonald *et al*., 1995). Intake of straw was increased when fed moist than in dry form (Ibrahim, 1989). Soaking for 1 to 2 hours increased voluntary feed intake of straw (Mtamakaya, 2002). Soaking causes swelling of cell wall structure and thus should make them more accessible to the cellulolytic microbes, in addition it reduces the dryness and dustiness of the feed. Dryness increases time spent chewing per bolus and thus lower total intake. The intake of maize stover can be affected by the stage of growth. Maturity increases cell wall constituent fractions and decreases protein content both lower maize stover intake.

2.4 Improvement on the Nutritive Value of Maize Stover

Maize stover is widely used to feed ruminants during dry season as animals consume the edible parts of stover and huge mass of stalks remain unutilized in the farms. To improve nutritive value of maize stover, different methods have been used and are grouped as physical, biological and chemical methods.

2.4.1 Physical treatment of maize stover

Maize stover particle size is reduced in order to ease biological and chemical treatment because increase the surface area of the cellulose and therefore improves the enzymatic hydrolysis (Shi, 2007). Physical treatment method refers to mechanical and non mechanical methods. Mechanical treatment includes chopping, grinding, soaking, pressing, cooking, steaming and separating head from stem. Non mechanical treatment includes methods like irradiation where by gamma rays are applied, to make biomass more accessible as to increase hydrolysis rate (Walker, 1984).

The methods include, pressure, and radiation (gamma rays, electron beam and ultrasounds and microwaves) (Walker, 1984). Physical treatment of maize stover increases the accessible surface area to rumen microbes and the size of the pores of cellulose and decrease its crystallinity and polymerization degree (Kimbi, 1997). These physical methods are not very satisfactory if used individually, and many times are employed in combination with chemical treatment methods in order to improve the process efficiency.

Grinding, milling and chopping are the common methods adopted which aim at increasing the surface area available to enzymatic digestion of the cellulose by rumen microorganisms and to increase the animal's voluntary intake. Reduction in particle size facilitates better storage, reduces selective eating by animals, increase ease of handling, reduce wastage and improve feed intake and digestion as relatively larger surface area becomes available for microbial activity (Van Soest, 1982). Grinding of maize stover to small particle size increase digestibility, but reducing size further decrease digestibility. Mechanical grinding of maize stover particularly destroys the structural organization of the cell walls thereby accelerating their breakdown in the rumen and increase feed intake as articulated by McDonald *et al*. (1995). Soaking in water gives inconsistent benefits in terms of improved digestibility, but with ground materials it reduces dustiness and it provides an easy and safe way to feed urea with minimal risks of poisoning.

Stripping done to remove the palatable leaves and top portion of the stem for the use as feed, leaving the discarded stems to be used for fuel or left in the field as the source of organic matter as reported by Massawe and Mrutu (2005) is yet another physical method of improving intake and digestibility and reduction of transport cost. Defoliation of lower leaves of maize stover before crop maturity is another physical method of improving utilization of maize stover. It was shown by Abate *et al*. (1985) that it was possible to obtain up to 1 tone DM/ ha over the three month period of defoliation without adversely reducing grain yield.

2.4.2 Biological treatment of maize stover

The biological treatment of maize stover is based on the use of enzyme/microbes that degrade lignin and increase digestibility of lignocellulolisic material. The advantage of biological treatment with either cell free enzymes or fungi is improved palatability and degradability potential of feedstuff and improve storage characteristics of the feed material as reported by Milligan *et al*. (1995). The enzyme treatment hydrolize cellulose fraction of maize stover into monosaccharides and improve the nutritional value of maize stover (Lui and Ørskov, 2000). Enzymes treatments being added in a controlled manner, could potentially be managed to create optimal conditions for rapid lignocelluloses degradation which may be achieved in a free living systems (Geib *et al.,* 2008). Compared to microbial strategies enzyme treatment allows effective preservation of dry matter. There have been very few reports on lignin decomposition by lignin degrading enzymes, but the possibility of utilizing these enzymes to deconstruct lignin for improving bioconversion efficiency show greater potential (Geib *et al*., 2008).

The two main reasons for using cell wall degrading enzymes as stover additives first to degrade the cell walls into fermentable water soluble carbohydrates which promote lactic acid bacteria fermentation in case where fermentable sugars are scarce and second to decrease fiber content of stover or improved digestibility. Most previous interest in enzymes for the stover focused on cellulose and hemicelluloses generally from the fungi *Trichoderma longibrachiatum, Asperigillus niger, A.oryzae, A.avmor* and occasionally from bacterial sources (Kung *et al.,* 2003).

Enzymes additive such as cellulase and hemicellulase that have been introduced in the process of ensiling maize stover degrade the lignocellulosic cell wall components to fermentable sugars (Richard *et al*., 2002). This can provide structural support for growth of lactic acid bacteria and lower the pH of the ensiling biomass. Cellulase is defined as group of enzymes that catalyse the degradation of cellulose to soluble sugars and glucose (Bhat and Bhat, 1997). Example endoglucannase. Hemicellulase refers to an enzyme complex that catalizes the degradation of hemicelluloses to sugars include endo -1, 4D- xylanase and exoxylosidases (Kung *et al*., 2003). Lignin degradation by white rot fungi known as *phanerochaete chrysosporium* is oxidative process that is catalysed by lignin peroxidases (LiP) (Lee, 1997). Lignin degrading enzymes are produced in nature by flamentous fungi that are able to degrade lignin (Kirk and Farrell 1987). White rot fungi are the best known microorganisms that can efficiently degrade lignin by producing lignolytic enzymes, peroxidase and laccases which use low - molecular -weight mediators to conduct lignin degradation.

There are 4 different lignin degrading enzymes namely:

Lignin peroxidases, Manganese dependent peroxidase, Versatile peroxidase and Laccases (Pe'rez *et al.,* 2002). Several plant consuming animals have gut flora that can be used to degrade lignocelluloses. For example, the termite gut possesses a highly efficient microbial community that can convert 95% cellulose into simple sugars within 24 hours (DOE, 2007). It has been reported that the Asian long horn beetle and Pacific damp wood termite successfully degraded wood lignin within several hours (Geib *et al*., 2008). This discovery may contribute to identify new, highly effective lignin degrading enzymes from the gut of insects instead of fungi.

2.4.3 Chemical method for treating maize stover

2.4.3.1 General information

Research has shown that lignin acts as a natural barrier to microbial attack, enclosing the largely crystalline cellulose micro fibrils and amorphous hemicelluloses polymers, and preventing microbial access for enzymatic hydrolysis of the cellulose (Mansfield *et al.,* 1999). Chemical treatment is thought to remove the lignin crust, allowing for dissolution of hydrogen bonds and swelling of cellulose micro fibrils, which serves to partially break off and solubilize the hemicelluloses polymers. This increases surface area of cellulose and the accessibility to hemicelluloses for increased hydrolysis by rumen microbial cellulases (Kahar, 2013). Lignin is reported to inhibit cellulose hydrolysis by irreversibly adsorbing the cellulose enzymes (Lee *et al.,* 1994). Separation of the lignin from the hemicelluloses / cellulose complex not only increases substrate availability, but also increases cellulase enzymatic activity (Lee *et al*., 1994). It has been reported that chemical treatment may remove acetyl groups on lignin that may have a structurally inhibitory effect on hemicelluloses digestion and therefore may increase the potential rate or extent of digestion of the hemicelluloses components (Bacon and Gordon, 1980). The use of chemical methods for maize stover treatment has been carried out in different parts of the world. The strength of maize stover bond between lignin and polysaccharide which act as barrier for microbes degradation are reduced through chemical treatment and hence increases digestibility. Alkali treatment make structural fiber swollen and hence enabling rumen microbes to attack the structural carbohydrates more easily so palatability and digestibility of the maize stover increases (Van Soest, 1982). Alkali increases the internal surface area of maize stover (biomass), decreases the degree of crystallinity of cellulose and does not degrade hemicelluloses (Kimbi, 1997). Carbohydrates in the presence of alkali and oxygen undergo both oxidation and alkaline degradation to produce a complex mixture of products (Klinke *et al.*, 2002).

2.4.3.2 Methods used

The two methods of alkali treatment are wet and dry methods. The wet method use large amount of water, in the way that the materials to be treated are soaked in the alkali solution for a pre determined time. Dry method uses less water that is small amount of concentrated alkali solution, which is normally sprayed on maize stover and allowed to react for a pre-determined time. The wet methods include Beckmann method, Torgimsby, dip and drip.

2.4.3.3 Wet method

(i) Beckmann method

The method was developed in German in 18th century. The procedure involved soaking straw in large quantity of soda solution, washing out alkali after treatment and the wet straw was fed to the animals. Approximately 12 kg of caustic soda solution was used for the treatment of 100kg of straw with amount of water 8 to 15 times the quantity of straw. The treatment or soaking time was 12 to 24 hours. The traditional Beckmann system was attractively simple, required only a vessel in which straws/ maize stover were soaked in sodium hydroxide solution. The Beckmann gave the end product of high digestibility. Homb (1984) observed an increase in organic matter digestibility of about 25% compared to untreated roughages. Also the increase in organic matter (OM) digestibility (45.75% in untreated to 71.3% treated with 1.5% NaOH). Limitation of Beckmann method was losses of nutrients in treated roughages due to washing after roughages are treated to remove excess sodium hydroxide before feeding. Estimates of 20% dry matter losses are observed to occur due to leaching. The problem of pollution due to disposition of the effluent solution. The method was reported to be labour intensive, difficult to industrialize and difficult in storage of treated materials (Homb, 1984). A large quantity of water was required (up to 50 liters of water per kg of stover/straw) for treatment of material and washing and sodium hydroxide was also required in large amount (12kg/100kg of straw /stover). All these limit the wide application of the method.

(ii) Torgimsby method

Due to the pollution problem caused by Beckmann method. A closed system was recommended in which the amount of water added to the system was equal to the amount of water removed by the treated stover/straws (Homb, 1984). The material treated by this method had about 20% dry matter content and *in vitro* DMD was about 70%. The method required less water and sodium hydroxide compared to Beckmann method and DM loss of treated material was minimized. There was no existence of pollution because the method is close system. Although it is more advantageous than Beckmann method, it is difficult to apply in developing countries due to high cost and un-availability of sodium hydroxide.

(iii) Dip and drip method

Some attempts have been made to overcome disadvantages of Beckmann method. Development of dip and drip method which discourages washing of the treated straw/stover and the solution is used in several times in order to minimize nutrient loss (Biwi, 1986). The straw/stover was dipped in the solution for pre-determined time, lifted and left to drip for 12 hour. The treated straw/stover was sun dried stored until time for feeding. The dip and drip method has been used for sodium hydroxide (Biwi, 1986; Urio, 1981) and wood ash extract (Katambala, 1997; Mtamakaya, 2002; Kimario, 2003). The method improve dry matter digestibility by about 7.5 to 23% depending on the chemical used. Limitation large capital is required to purchase large vessels in which roughages have to be soaked and availability of large volume of water required for soaking is problem in developing countries.

2.4.3.4 Dry method

Dry method involves spraying a pre-determined amount of alkali solution in a provided quantity of roughages and left to react for a predetermined time. With a way of eliminating disadvantages of Beckmann method. A process which straw is treated with small volume of concentrated solution of sodium hydroxide was developed by Wilson and Pigden (1964). The roughages are sprinkled or sprayed with the sodium hydroxide while being mixed. The method recommend use of 4 to 6 kg of sodium hydroxide dissolved in 200 liters of water which is enough to spray 100 kg of roughages (Wilson and Pigden, 1964). The amount of solution required is less (100 - 120) liters if pressure sprayer is used. In a small scale operation for feeding a few animals one adult person could apply the sodium hydroxide solution while another one mix the roughages by using fork.

When using watering can the efficiency of roughages treatment would be less, due to poor wetting of roughages. In treating large batch of roughages, a screw auger with spray nozzles inside it could be used effectively. Another means is a horizontal mixer with an overhead-spraying device. The machinery for roughages mixing could be used either manually or with farm power. Roughages treated in this way has a yellow colour and pleasant odour. Treated roughages have pH ranging from 10 and 11. Ruminant animals eat treated roughages 20 to 30% more than untreated roughages. Digestibility increase by 10 to

15% units (Wilson and Pigden, 1964). For every kilogram of sodium hydroxide per 100kg roughage added sodium content increases by approximately 0.6 percent units.

2.4.4 Chemicals that have been used for treating maize straw/stover

2.4.4.1 Sodium hydroxide (NaOH)

Sodium hydroxide has been reported to have high potential for improving the chemical content of straw. This is because NaOH treatment hydrolyses most of ester bond leading to release of acetyl- group. The chemical (NaOH) cleaves lignin carbohydrates ester bonds and lignin internal linkages (Theander, 1981). Furthermore, Theander (1981) observed that, NaOH hydrolyses hemicelluloses leading to decrease in NDF content of treated roughages and swelling of cellulose fibers within the cell wall matrix. These processes require water; therefore the level of moisture in the roughages used or amount of chemical solution used is likely to have great influence on the effectiveness of the treatment. On the other hand, it increases digestibility, feed intake and the overall growth performance of the animals fed on NaOH treated maize stover (Sundstøl, 1981).

(a) Effect of NaOH treatment on chemical composition of maize stover

Treatment of maize stover with NaOH by soaking method reduced the CP of the treated material due to leaching (Musimba, 1981). The amount of CP content loss increased with increasing concentration of NaOH (Musimba, 1981) and treatment time (Sundstøl, 1981). An increase in ash content following NaOH treatment of maize stover was observed by Musimba (1981). Similar results are shown in Table 3 where ash content of maize stover is increased after NaOH treatment. Higher ash content of maize stover which resulted into lower OM content of the treated maize stover were reported by Meseke *et al.* (1993). Absorption of minerals from the solution resulted to increase in ash content of the treated materials. Contrary results were observed by (Musimba, 1981) that rice straw treated using

NaOH had lower ash content than untreated rice straw. The reasons for low ash content of treated rice straw with NaOH could be due to solubilization of biogenic silica from rice straw which form sodium silicate. As concentration of NaOH increases from 0 to 8%, ash content of treated maize stover increase from 12.5 to 14.9% (Musimba 1981). Meseke *et al*. (1993) reported low NDF content 59.6% of NaOH treated straw compared to untreated straw 80.2%. The breaking down of fiber complex cause cell wall reduction of the treated straw due to solubilization of hemicelluloses during alkali treatment of straw/stover.

Table 3: Chemical composition (%DM) of stover treated with different chemicals

Roughage	DM	ASH	NDF	CP	Source
Untreated MS	93.4	12.5	-	2.31	Musimba (1981)
8%NaOH treated MS	91.3	14.9	-	1.57	Musimba (1981)
Untreated MS	91.7	7.99	80.9	3.14	Biwi (1986)
NaOH treated MS	88.8	9.01	83.7	3.18	Biwi (1986)
MS untreated	-	-	76.2	6.1	Mao *et al*. (1990)
Urea treated MS	-	-	68.9	12.5	Mao *et al*. (1990)
MS untreated	90.2	7.8	79.2	4	Kilongozi (1992)
Urea treated MS	72.5	7.4	71.5	8.9	Kilongozi (1992)
3%Urea treated MS	-	-	84	7.9	Shem *et al*. (1995)
MS untreated	-	-	88.1	4.1	Shem *et al*. (1995)
MS untreated	74.7	5.8	77.1	2.9	Kimbi (1997)
Urea treated MS	77.9	6.8	78.5	5.3	Kimbi (1997)
MS untreated	91.06	7.25	-	2.55	Katambala (1997)
Ash treated MS	92.65	34.6	-	2.01	Katambala (1997)
NaOH treated MS	90.61	18.11	-	2.59	Katambala (1997)
MS untreated	86.15	-	77.2	3.25	Kilasi (1998)
Ash treated MS	82.15	-	73.13	2.51	Kilasi (1998)
5%CaO treated MS	-	-	-	15.8	Johson *et al*. (2013)
Untreated MS	-	-	-	15.4	Johson *et al*. (2013)
4% Urea treated MS	35.9	-	-	14.4	Munthali *et al*. (1990)
Untreated MS	91.7	-	-	2.8	Munthali *et al* .(1990)
Untreated MS	94.3	10.2	70.9	7.64	Onaleye *et al*. (2012)

Key: MS =Maize stover

(b) Effect of NaOH treatment on degradability/digestibility

NaOH treatment of maize stover have higher potential for improving the digestibility components of maize stover as reported for wheat straw by Meseke *et al*. (1993). The *in vitro* digestibility coefficient of dry matter for NaOH treated and untreated maize stover were 67.03 and 44.23% respectively (Biwi, 1986). The increase in digestibility has been attributed to a solubilisation of the total phenolics, arabinoxylans and cellulose arising from cleavage of alkali labile lignin carbohydrate linkage (Vadiveloo, 2000). A significant improvement in *in vivo* digestibility of diet based on maize cobs treated with 5% NaOH but only a slightly further increase when concentration of NaOH was increased from 5 to 7% was observed by Urio (1981). Treatment of maize stover using NaOH at high level (10%) tended to reduce the *in vivo* digestibility. This was due to intake of large amount of salt from NaOH treated materials which led to increase in water intake and rapid passage rate of ingesta through the alimentary canal (Biwi, 1986). High osmolarity due to NaOH intake may create unfavorable environment condition for microbes in the rumen leading to reduced digestibility.

(c) NaOH treatment on DM intake

Roughage treatment with NaOH improves DMI of treated roughage by ruminant animal (Saadulah *et al.,* 1981). Feeding wheat straws treated with 4% NaOH and supplemented with urea molasses mixture improved DMI (Meseke *et al.,*1993).The intake of NaOH treated roughage was improved due to increased palatability and digestibility (Bod'a 1990). Urio, (1981) reported an intake of 50, 61.1 and 63.5g/kg$W^{0.75}$ for 0, 3 and 6% NaOH, treated maize cobs respectively. Treatment of soya bean straw with NaOH reduced organic matter intake (OMI) due to increase in mineral content of the treated bean straw as reported by Ikem and Felix (1992).

(d) NaOH treatment on animal performance

Different workers have reported improvement on daily gain and growth rate of animal fed NaOH treated straw/stover (Saadulah *et al*., 1981; Urio, 1981). Animal fed on NaOH treated stover improved weight gain due to increased in intake of digestible nutrients from the treated maize stover (Urio, 1981).

Although sodium hydroxide is effective in improving the digestibility of several crop residues, it is expensive, corrosive, poses human health risks in handling, and the resulting feeds may provide excess dietary sodium relative to animal requirements.

2.4.4.2 Urea

Like dry treatment with NaOH, urea is applied to 50% moisture maize stover and ensiled. Urea treated material must be stored in a manner to prevent ammonia evaporation and stored for 14 - 21 days before feeding (Fahmy and Klopfenstein, 1992). Although, urea treatment does not improve the digestibility of the treated stover to the same extent of the other chemical methods, urea is safer to handle and accessible. It also increases the Nitrogen content of the treated material (Kilongozi, 1992). Ammonia, which is produced from the treated straws/ stover during ammoniation inhibit growth of mould.

(a) Effect of Urea treatment on chemical composition of the treated material

Different workers have reported on increase in CP content of urea treated stover (Kilongozi, 1992; Shem, 1993; Salem *et al*., 1994; Kimbi, 1997; Mgheni *et al*., 2001). Research conducted by Salem *et al*. (1994) whereby stover was treated using 5.3% urea and ensiled for 8 weeks resulted in an increase in CP content of the treated stover from 5.8% (untreated) to 11.8% for urea treated stover. Increased CP content of treated stover was due to binding of ammonia released from urea to stover which is important in N fixation (Salem *et al*., 1994). Treatment time and increasing concentration of urea increase the CP content of treated straw by a fraction of 2 to 2.5 (Nguyen *et al.,*1998; Dias-da-Silva *et al*., 1988; Ojala *et al*., 1988). When urea is used in the treatment of maize stover, treatment ratio of liquid to stover is important (Nguyen *et al.,* 1998). This is due to the fact that hydrolysis of urea to NH_3 can only occur if sufficient water is available. Moisture help in the compaction of stover which assist in removing out air hence conserving ammonia. Nguyen *et al.* (1998) reported that the acceptable treatment ratio of stover to liquid is 1:1.

Ash content of straw/stover treated with urea is variable. Different authors observed an increase in ash content of treated stover/straw from 7.3 to 15.1% for maize stover treated with urea and from 16.2 to 19.02% for rice straw treated with urea (Katambala 1997; Mtamakaya 2002). Low variation of ash content was reported by Joy *et al*. (1992) where ash content ranged from 4.8% for untreated to 5.6% for barley straw treated with 5.5% urea. The change in ash content was not constant when the concentration of urea was increased from 3 to 6%. When concentration of urea changed from 3 to 6% the ash content varied from 8.9% for untreated to 7.9% for urea treated materials. When Moisture content of urea treated stover/straws was increase from 20% to 30% ash content increased from 8.7 to 9.1%. When moisture content was increase further up to 40% the ash content was decrease to 8.7% (Joy *et al*., 1992).

Effect of urea treatment on cell wall component of straws/ stover have been studied by Nguyen *et al.* (1998); Salem *et al*. (1994); Joy *et al.* (1992) who reported on small decrease on NDF content. Treatment of sorghum stover with 5.3% (DM bases) urea slightly decreased NDF content from 68.7% for untreated stover to 67.2% for urea treated stover (Salem *et al.,* 1994). Similar results were observed by Joy *et al*. (1992) whereby NDF content was reduced from 86.1% for untreated barley straw to 78.7% for urea treated barley straw. A decrease in NDF content from 72.9% for untreated rice straw to 70.9% for rice straw treated with 5% urea was reported by Nguyen *et al*. (1998). Urea treatment of maize stover has reduced NDF content of the stover by 6% units (Alemu *et al*., 2005).

(b) Effect of urea treatment on degradability/digestibility

Urea treatment releases free phenolic group from the cell wall matrix which upgrade straw digestibility in the rumen (Shen *et al*., 1998). Maize stover treatment with urea affect lignin and delignification of stover. When urea concentration was increased from 3 to 5% higher CF and CP degradability were observed (Shen *et al*., 1998). When maize stalk was treated with urea and incubated at 45^0C for 1-14 days, higher IVOMD was observed for those incubated for 6 to 14 days when compared to 0 incubation period (Fahmy and Klopfenstein, 1992). The values for *in vitro* digestibility changed from 49.6% for 0 hour incubation period to 54.37 and 54.57% for 6 to 14 days incubation period, respectively.

The workers concluded that level of 6.6% urea and incubation period of 6 days and above 45^0C could be used for urea treatment. Sorghum stover was treated with 5.3% urea and ensiled for 8 weeks increased OM and NDF digestibility by 3.7 and 1.4% units respectively (Salem *et al*., 1994). Apparent DM digestibility of 5% urea treated maize stover was increased by 27% units that of rice straws by 18.1% and that of wheat straw by 13.3 units when measured in growing steers and lactating buffaloes (Saadullah *et al*., 1982).

(c) Effect of urea treatment on DM intake

Compared with the untreated straw urea treatment brought an improvement of 32% in dry matter intake (Saadullah *et al*., 1981). Also treatment of straw with urea was found to improve dry matter intake over untreated straw (Khan *et al*., 1999). The use of 4% urea for wheat and rice straws treatment increased dry matter intake by 20.2 and 17.5% over untreated wheat and rice straws respectively (Khan *et al*., 1999). Improved intake of urea treated rice straw could be due to improved degradation of the treated straw compared to untreated. Positive effect on intake due to addition of N associated with urea treatment of straw that facilitated microbial growth, which is important in digestibility and intake of feeds. Animals fed on rice straw treated with 5% urea level had higher intake than 3% urea level (Saadullah *et al*., 1982). This was due to increased in CP content for 5% urea treated than for 3% urea treated rice straw.

(d) Effect of urea treatment on growth performance

Animals fed on urea treated maize stover have shown higher rate of gain than those fed untreated stover (Kilongozi, 1992). Male calves fed on 10% urea treated maize stover gained higher weight (237g/day) compared to those fed untreated stover that gained 96g/day (Kilongozi, 1992). Goats fed on urea treated rice straw had higher weight gain (49.1g/day) compared with 13.0g for those fed on untreated rice straw observed by Mgheni *et al*. (1993). When grazing crossbred calves were given urea treated straws with molasses urea blocks they nearly doubled their live weight gain (Vu *et al*., 1999). Higher weight gain for animals fed urea treated straw/stover could be due to increased N content of the treated stover/straw, higher intake and digestibility. Use of urea in treating roughages to improve feeding values of stover and straw was practiced in Tanzania in late 80`s and early 90`s. Adoption of the method has been limited due to rise in cost of urea. The efficiency of utilization of urea during treatment of stover/straws is poor, only 30% of the nitrogen from urea applied is recovered in the treated stover/straws while the rest pollutes the atmosphere as ammonia.

2.4.4.3 Calcium oxide

Calcium oxide, commonly called *quick lime* or *burnt lime*, is a white crystalline solid with a melting point of 2572°C, is produced by heating limestone, coral, sea shells, or chalk, which are mainly $CaCO_3$, to drive off carbon dioxide.

$$CaCO_3(s) \xrightarrow{500 - 600°C} CaO(s) + CO_2(g)$$

The reaction is driven to the right by flushing carbon dioxide from the mixture as it is released. The production of calcium oxide from limestone is one of the oldest chemical transformations produced by man. Calcium Hydroxide, commonly called hydrated lime *or* slaked lime, is produced by adding water to calcium oxide in a controlled reaction. Moreover, the heat energy CaO produced when forming the hydrated $Ca(OH)_2$ in water is favorable for reaction acceleration. $Ca(OH)_2$ has attracted much attention in treating stover/straw because they can easily be recycled after treatment with established lime kiln technology and through evaporation, respectively (Siroh and Rai, 1998). Lime is weakly soluble and non volatile, so that a large amount of lime is needed to get it properly distributed and act as a reserve which gradually dissolves and maintains the concentration of hydroxide (OH^-) in the water. As (OH^-) is taken up by the straw, more $Ca(OH)_2$ comes from the reserved into solution (Zaman *et al.*, 1994).

One of the alkaline treatment that has not been extensively researched but is gaining popularity is Calcium Oxide (CaO). Lime works by increasing digestibility of the fiber. The treatment swells the cellulose, which makes it easier for the enzymes to work. Lime also improves acetyl groups that have been shown to affect hydrolysis rates. Lime removes lignin and improves cellulose digestion by enzymes through opening up the structure and reducing non- productive cellulose absorption. Additionally, the hydrogen bonds between lignin and hemicelluloses are hydrolyzed. The effects of CaO, NaOH and alkaline hydrogen peroxide (AHP) treatments on the rate and extent of disappearance of wheat straw in the rumen of sheep compared by Chaudhry (1999) showed that AHP was the most effective treatment for improving ruminal degradation of wheat straw, it was the most expensive and least practical. CaO was effective in modifying NDF composition of wheat straw to improve ruminal digestion which was also observed by Chaudhry, (1999). While CaO was slightly less effective in improving ruminal digestion of wheat straw than NaOH, CaO was more desirable treatment option due to low cost, ease of handling, relative ease of storage and application as well as fewer safety concerns when compared to NaOH (Chaudhry, 1999).

CaO or quicklime, is an efficient means of alkaline treatment in terms of cost and labour. It is cheaper and safer than NaOH or $Ca(OH)_2$ since it can be added in dry form to moistened maize stover and does not require a special apparatus for application. Calcium Oxide has high potential in the improvement of chemical composition of roughages compared to sodium hydroxide. Calcium Oxide and Calcium hydroxide are both alkalis of high pH levels derived from heating limestone. Because the least expensive alkali is lime, that is available as either quick lime (CaO) or slake lime $Ca(OH)_2$, treatment with this chemical provides a low cost alternative for lignin removal at higher pH values. Calcium ion, Ca^{2+}react with carbon dioxide to form Calcium Carbonate which gradually deposits in the lignocellulosic matrix. Carbon dioxide may be generated from delignification and degradation of cellulose and hemicelluloses or it can be present in the air getting in through the biomass (Lōpez, *et al.,* 2000). Calcium Oxide is normally dusted to the residues that have 50% moisture and ensiled for 5 to 7 days and at a reasonable concentration. The optimal moisture for CaO to react properly on maize stover or straws ranges between 50 - 60 percent (Russell *et al*., 2011). Furthermore, CaO is environmentally friendly to crop land and future crop production, it is not corrosive to equipments (Russell *et al*., 2011). Other merits of CaO is that it assist in growth of young animals and improve nervous and heart functions, and it also promote increase in milk production in dairy animals (Sirohi and Rai, 1998). The chemical also improve digestibility of crop residues thus, increasing the overall digestibility of DM of crop residues (Sirohi and Rai, 1998) and boost intake in ruminants (McDonald *et al.,* 1995).

(a) Effect of CaO/Ca $(OH)_2$ on Chemical composition

Chemical composition of different feeds can be changed by chemical treatment. CaO/ $Ca(OH)_2$ treatment decreases the NDF content of the stover/straw. A decrease in NDF content from 80.6 (untreated) to 78.3% and 76.3% of straw treated using 2% and 5% lime respectively was reported by Sirohi and Rai (1998). The moisture content found to be appropriate for the treatment of roughages was 50% and the NDF content of roughages was reduced linearly with increasing moisture from 20 to 60% (Sirohi and Rai, 1998). The NDF content was found to decrease with increasing treatment time. Calcium hydroxide do not have significant effect on CP content of straw even on increased concentration of $Ca(OH)_2$. Both moisture and concentration level did not affect CP content of straw (Sirohi and Rai, 1998). Calcium oxide treated maize stover show higher %CP (15.8) than untreated maize stover (15.4) (Johnson *et al.*, 2013). The ash content of rice straw treated with lime was increased (Nguyen *et al*., 2001).

(b) Effect of CaO/Ca $(OH)_2$ on degradability/digestibility

Alkaline treatments increase the digestibility of rice straw and wheat straw by 40% - 60% (Chaudhry, 1999).Treatment of straw using CaO/$Ca(OH)_2$ improve digestibility when compared to untreated straws (Pradhan *et al.,* 1997). An increase in dry matter digestibility from 46.2 (2% lime) to 59.7 when lime was increased to 5% was reported by Sirohi and Rai (1998). Moisture level has more influence on *in vitro* DM digestibility of the treated than untreated (Sirohi and Rai, 1998). Moisture level of 50 to 60% improved *in vitro* DM digestibility from 52.4 to 55.7% respectively, when wheat straw was treated with lime (Sirohi and Rai, 1998).

A chemical solution to roughages ratio of 2:1 increased *in vitro* DM digestibility as concentration of CaO was increased, while the ratio of 4:1 had no further effect on *in vitro* digestibility. When treatment time is increased the digestibility of CaO treated roughages increases (Sirohi and Rai, 1998). Wheat straw treated with 2 - 5% CaO and ensiled for 1 - 3 weeks increased *in vitro* DM digestibility from 52.4 to 56% (Sirohi and Rai, 1998). The effects of feeding CaO treated maize cobs, maize stover and wheat straw at 25% of the diet DM, to replace maize grain, on ruminal metabolism and digestibility was studied by Shreck (2013). They reported that there were no differences in DM or OM digestibility between cattle fed the treated maize stover diets or those fed on maize grain based control diet.

Feeding of CaO treated maize stover increased NDF digestibility by cattle when compared to feeding maize grain. The digestibility of dry matter of straw increased significantly with lime treatment from 38 to 49% (Saadullah *et al*., 1981). The *in vivo* digestibility of CaO treated maize stover was 83.2% higher than untreated maize stover (75.5%) (Russell *et al*., 2011).

Although 6% lime was more effective than 3% lime in cell wall solubilization, improving gas production and degradability of the straw as a substrate, a level of 3% lime seemed to be better for rumen ecosystem (Nguyen *et al*., 2001). Even though treatment at 6% lime is more effective in improving digestibility of the treated material, this high concentration is toxic to microbes in the rumen ecosystem. On the other hand contradicting findings have been reported where *in vitro* digestibility was reduced from 55.7 to 52.4% for untreated and CaO treated maize stover (Russell, 2013). The digestibility of straw treated with lime gave poor results due to Calcium (Ca) content that can produce an imbalance in the Calcium: Phosphorus (Ca:P) ratio (Verma, 1981). The energy and protein digestibility was reduced when cattle were fed a diet containing 4.4% Ca.

(c) Effect of CaO/Ca$(OH)_2$ on DM intake

Improved intake of Ca$(OH)_2$ treated wheat straw when fed to sheep was reported by Djajanegara *et al.* (1985) and Sahoo *et al.* (2002). While a reduction in intake of lime treated straws due to poor palatability was reported by Pradhan *et al*. (1997). In the study of Saadullah *et al*. (1981) where rice straw treated with lime was fed to sheep intake of the treated materials was lower than untreated straw.

(d) Effect of CaO/Ca$(OH)_2$ on growth performance

Feeding cattle 5% CaO treated maize stover at 20% of the diet DM improved intake (Shreck *et al.,* 2011). A follow up study compared 5% CaO treatment of maize stover to that of 5% CaO treated maize cobs and wheat straw (Shreck *et al*., 2012a). They observed that feeding CaO treated maize stover and straw to cattle resulted in increased final body weight, average daily gain (ADG) and feed efficiency when compared to feeding untreated maize stover and straw, while treating maize cobs was not effective. In a follow up study, (Shreck *et al.* 2012b) observed that reducing particle size of ground maize stover from 7.62 to 2.54 cm prior to feeding increased feed efficiency. One possible mechanism to explain the aforementioned improvement in feed efficiency may be increased microbial attachment, due to greater surface area, and increase digestibility. Growth performance improvement was observed by Pacho *et al*. (1977) as cited by Kimario (2003) who reported an increase in weight gain from 520g/day (untreated) to 720 g/day in cattle fed rice straw treated with Ca$(OH)_2$. Also Saadullah *et al.* (1981) reported an increase in weight gain from 35g per day for calves fed untreated to 75g/day for calves fed on $Ca(OH)_2$ treated rice straw.

Limitations on use of CaO/ $Ca(OH)_2$

$Ca(OH)_2$ is weaker alkaline than NaOH and needs long time to react on the maize stover depending on the ambient temperature. Furthermore poor solubility of $Ca(OH)_2$ represents a considerable disadvantage to its use and render it less effective. Lime is comparatively cheaper but one problem associated with lime treated stover/straws is growing of mould when not properly covered (Zaman *et al.,* 1994). Also limitation of using $Ca(OH)_2$ is that controlling the treatment conditions such as temperature, moisture and time interactions, to ensure maximum reactivity and hence influence overall digestibility and palatability of the product is difficult. It may also affect Ca:P ratio of the feed and could result into imbalance intake of these materials by the animals. Too high a level of lime (6%) may create unfavorable rumen conditions for microbial activity and limit stover palatability (Nguyen *et al*., 2001).

2.5 Methods of Evaluating Improvement of Differently Treated Maize Stover

2.5.1 *In sacco* degradability

The nylon – bag (*in sacco*) technique describes the procedure for feed evaluation where small porous nylon bags containing feed samples for test are incubated in the rumen of fistulated animals for specified time. The nylon bag technique makes use of porous nylon mesh which can vary in surface areas including 6.5 x 14 cm; 5 x 5cm; 5 x 10 cm or 10 x 20 cm. Livestock feed offered to ruminants in third world countries are generally of low quality (Jayasuriya, 2002), their degradability become important and needs to be established if they are to be utilized efficiently (Olaisen *et al*., 2003). Attempt to evaluate the nutritive value of feeds by measuring disappearance of feed material from small bags incubated in the rumen of fistulated cattle and thus expose to digestive process began as early as 1700s. In initially, the *in sacco* technique was developed for assessing the degradation of protein (Ørskov and McDonald, 1979). There exists a relationship between the feeding value of feed and their degradability patterns, the faster the degradation, the higher the feeding value.

In sacco digestion can predict the potential degradability, intake, feeding value of feeds and compliment other feed evaluation methods such as proximate analysis in ranking feeds (Thadei *et al*., 2001). The *in sacco* approach is useful in that it exposes feeds to digestive conditions thought to be similar to those of *in vivo*, thus allowing for inferences made to be of greater acceptance (Broderick and Cochran, 2000). The feed intake potential, which is the ability of feeds to provide a given level of digestible nutrient intake, can also be predicted with aid of feed dry matter (DM) degradation characteristics. This method can detect the improvement of the feeding value due to treatment. The limitations of this method is washing loss, animal effects, drying procedures, bag types and pore size. Washing loss of dry matter have usually been identified as the water soluble, readily fermentable fraction of roughages in ruminant degradation studies (Ørskov and Shand, 1997).

While some small particles can pass through the pores. Pore size and porosity of the bag is a variable that may influence results. These factors determine the ease of traffic of microbes and of degraded products in and out of the bag. Pore size of less than 3µm restrict penetration of microbes into the bag and may completely exclude entry of some large rumen micro organism such as protozoa and fungi. This may result in underestimation of the extent of potential degradability of the sample. On the other hand pore size of more than 50µm may exaggerate the rate and extent of degradation. Most authors have suggested a pore size of between 40 – 50 µm (Michalet- Doreau and Ould – Bah, 1992). This size is believed to be optimal for microbial entry and escape of the degradation products. Bag materials are preferably polyster.

2.5.*2* *In vitro* gas production

The basic principle of gas production is that the *in vitro* fermentation of feeds incubated with buffered rumen fluid is accompanied by the production of gas (Awati *et al*., 2006). The gas is formed directly by microbial fermentation of the substrate as well as indirectly by the release of carbon dioxide caused by the production of Volatile Fatty Acids (VFA) from biochemical buffer (Van Soest, 1994). However, it is very difficult to separate direct and indirect gas production. Based on the fact that both are directly related to the fermentation of a substrate, the gas production measured at each time point can be considered as an index for fermentation activity (Groot *et al*., 1996).

Gas production is the result of fermentation of carbohydrates to VFA (Blümmel and Ørskov, 1993). The three end products of carbohydrate fermentation in rumen were summarized by Hungate (1966) as follows:

1mol Hexose $+2H_2O \rightarrow 2$ acetate $+2CO_2 +4H_2$

1mol Hexose $+2H_2 \rightarrow 2$ Propionate $+2H_2O$

1mol Hexose $\rightarrow 1$ Butyrate $+2 CO_2 + 2H_2O$

The reaction equation above show that the total amount of gas produced differs depending on both amount of substrate fermented and the amount of molar proportions of the VFA end-products formed (Davies *et al*., 2000). Rapidly fermentable carbohydrates yield relatively higher amount of propionate as compared to acetate, and the reverse takes place when slowly fermentable carbohydrates are incubated (Van Soest, 1994). More propionate and lower acetate ratios in the rumen fluid of cows fed high grain diet were reported by

Gatachew *et al*. (1998). The gas produced from VFA production and bicarbonate buffer in the rumen are eructed to the atmosphere. It is this gas that is being measured *in vitro*. Advantages of *in vitro* gas production methods are that they are less time consuming and less costly. Samples of feed components can be studied in isolation, and smaller quantities of feed are required. A limitation of general use of *in vitro* methods is that they require donor animals.

2.5.3 *In vivo* digestibility/marker technique

(a) Total collection

Evaluation of ruminants' feeds has been made predominantly through *in vivo* trials. Detailed procedures are provided by, Schneider and Flatt (1975) as cited by Chenyambuga (1995). The standard procedures involve measuring the quantity of a nutrient consumed and the quantity voided during research trial. From the amounts and the chemical compositions of the feed consumed and the faeces excreted, the digestibility of the feed and its various fractions are computed. The arithmetic difference in the quantity of the nutrients between the feed consumed and the faeces excreted by animal is the apparent digestibility of nutrient. Male animals are more often used due to convenience in sample collection. Some of the basic requirements for the procedure are the need for a preliminary period, accurate estimate of intake and faeces excreted and duration of sample collection that ensures true presentation of digestibility.

Feeds that exhibit wide variation in voluntary intake such as crop residues will require longer preliminary duration in order to standardize the level of intake. A number of factors are known to influence the digestibility value obtained through *in vivo* measurements. Age of the animal, level of intake, health and breed (Schneider and Flatt, 1975) as cited by Chenyambuga (1995). The plane of nutrition is one of the primary factors that affect digestibility of feed. Research has shown that livestock usually, digest a larger percentage of nutrients in their feed when fed restrictedly than when they receive full feed. Most data show some depression in apparent digestibility as level of intake is increased. This may be due to a more rapid movement of the feed through the tract allowing short time for digestion and absorption.

(b) Markers technique

Several internal and external markers have been used to estimate digestibility and passage rate. Markers have also been used to measure digestion where direct measurements are inconvenient. The acid insoluble ash a natural marker method of ration digestibility studies offers some distinct advantages over other marker methods and the traditional total faecal collection method. The use of marker to determine nutrient digestibility of feeds overcome the need to make exact measurement of feed intake and faecal output in the traditional faecal collection method. Acid insoluble ash (AIA) is the indigestible mineral components mainly silica, left after treatment in hydrochloric acid (HCL) and ashing. Silica has been used for many years as digestibility indicator but is not widely used at present due to its possible absorption in the gut and risk, especially with grazing animals of contamination of feed with soil (Rymer, 2000). Acid insoluble ash method in accordance to the use of the marker technique to determine the digestibility has the advantage that grab sample could be taken from animals, resulting in obtaining samples from free living animals before any chemical changes in faeces occur. Furthermore animals that have a social structure could be kept in groups for nutrient digestibility studies. Special care should be taken when applying this method to grazing animals where ingestion of soil could occur. Soil and dust in feeds and laboratories should be avoided. The AIA as marker in nutrient digestibility studies is easy and simple analysis used but difficult in that a high degree of precision is needed.

2.5.4 Feed intake

Feed intake is one method of evaluating the quality of feeds and therefore can be used to evaluate improvement in feeding value due to physical chemical or biological treatment. Most of crop residues are deficient in protein, essential minerals like sodium, phosphorous and calcium and are rather fibrous (40 to 45% crude fiber). The increased intake of urea treated straw in a range of 15 to 50% has been reported by Chenost and Kayouli (1997). Sheep fed wheat straw treated with urea was also reported to have increased by 13% dry matter intake (Singh and Klopfenitein, 2001). Sheep fed on urea treated sorghum stover had an increase of 34% dry matter intake (Reddy and Reddy, 2002).

Urea treatment increases the acceptability and voluntary intake of the treated straw as compared to untreated straw when fed ad libitum. The depressed intake of lime treated sesame straw (Aregawi *et al.,* 2014). Similar observations on 4% lime treated rice straw were reported by Saadullah *et al*. (1981). The intake of lime treated rice straw decreased due to reduced acceptability of treated straws by ruminants (Sarnklong, 2010). To the contrary increased OM intake (Djajanegara *et al*., 1985) and increased DM intake (Sahoo *et al.,* 2002) were observed in Calcium hydroxide treated wheat straw. The dry matter intake for 6.6% Calcium hydroxide treated maize stover was 12.5kg and 11.9kg for untreated (Peterson, 2014). Also the dry matter intake of 5% Calcium Oxide treated maize stover was 10.1kg and 9.4kg for untreated reported by the same author.

2.5.5 Growth performance

Growth performance is another method of evaluating improvement of feeds as a result of chemical treatment. The treated diet to be evaluated is given to a group of animals for a specific period of time. Ruminants fed on either CaO, enzymes, microbes and $Ca(OH)_2$ treated maize stover have been mainly assessed through increased average daily gain

(ADG). The ADG (kg/day) is normally calculated from final live weight (kg) of the beef animal minus initial live weight (kg) divide by the duration of gain (days). Improved nutritive value of lime treated maize stover was confirmed through increased ADG (Shreck, 2013). Beef cattle finished on forage typically take longer to reach market weight and often have depressed average daily gain (ADG). $Ca(OH)_2$ treated rice straw by soaking method was found to increase the *in vitro* organic matter digestibility and when fed to growing cattle grew between 38 and 90g/day (Doyle *et al.,*1986). The ADG of beef fed maize stover treated with $Ca(OH)_2$ was 1.50kg /day and 1.35kg/day for untreated (Peterson, 2014). Also ADG of CaO treated maize stover was 1.36kg/day and 1.25 for untreated reported by the same author. Feed conversion ratio (FCR) is also one of the parameters used to measure performance of animal. Feed conversion ratio is a measure of animal`s efficiency in converting feed mass into increased body mass. Feed conversion ratio or feed conversion efficiency is measured in terms of amount of feed required to produce a unit of gain (McDonald *et al*., 1995). The FCR of maize stover treated with 5% CaO and untreated were 6.84 and 7.17 kg stover per kg gain (Shreck *et al*., 2011). The FCR is said to range from 5 to 8 for least cost feeds, 9-10 for moderate cost and 11-20 for the poor and costly feeds. This means that the lower the FCR the lower the cost of production and the higher the growth rate of the animals. This is estimated by calculating the ratio of amount of feed consumed per total weight gained.

2.6 Conclusion from the Literature Review

From the review of the literature, it appears crop residues namely maize stover, are in abundance. Chemical and biological treatment can be used to treat crop residues, but many factors such as chemical, substrate, temperature and moisture affect the utility of treatment. Chemical and biological treated maize stover lead into improvement on chemical characteristics of the different treated maize stover, especially reduced cell wall content.

There are also reports on improvement on degradability, *in vitro* gas production (IVGP), *in vivo* digestibility of differently treated maize stover and performance of animals fed on differently treated maize stover. These positive results were reported when chemicals such as NaOH and Urea were used.

In Tanzania the use of such chemicals is limited because they are expensive, both chemicals have potential hazards for animals and human, environment, however uniform adoption of urea treatment technology under field conditions across the country is not possible under such conditions one has to look into the possibility of employing cheaper source of chemical with adequate efficiency such as CaO/ $Ca(OH)_2$. Studies on the improvement of maize stover utilization for beef cattle production through chemical and biological treatment showed promising results.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Overview

Two studies were conducted in 2013 in Morogoro, Tanzania to evaluate the effectiveness of lime, enzymes, microbes and Calcium hydroxide treatments in improving the quality of maize stover.

Study I involved five treatments which were:

(a) Untreated maize stover harvested 2012(T_1)
(b) Maize stover treated with Calcium Oxide (T_2)
(c) Maize stover treated with Calcium Oxide + enzymes (T_3)
(d) Maize stover treated with Calcium Oxide + microbes (T_4)
(e) Maize stover treated with Calcium Oxide + enzymes + microbes (T_5).

The effectiveness of the treatment were evaluated by different methods that included chemical analysis, degradability, *In vitro* gas production and animal performance of beef cattle fed the differently treated maize stover.

Study II involved two treatments which were:

(a) Untreated maize stover harvested 2013 (T_6)
(b) Maize stover treated with $Ca(OH)_2$ (T_7).

The effectiveness of the treatments were evaluated by different methods that included chemical analysis, degradability, *In vitro* gas production, *In vivo* digestibility using marker technique and animal performance of the animal fed untreated and $Ca(OH)_2$ treated maize stover.

3.2 Source of Maize Stover

The maize stovers used in this study were TMV_1 and Staha improved varieties that were manually harvested from Sokoine University of Agriculture farm in Morogoro region, Tanzania during July/August, 2012 and June/July 2013. The stover was chopped using pulverizing chopper to 1 - 2 cm length. A portion of stover was set aside as untreated stover to act as control diet.

3.2.1 Study I: Treatment of Maize Stover

Calcium Oxide was purchased from Simba lime factory in Tanga Tanzania. It was used to treat 12 tons of chopped maize stover harvested in July/August 2012. The DM of the stover was determined and amount of water required to bring the moisture content of stover to 50% was calculated. Similarly the amount of CaO to be applied at a rate of 5% DM of maize stover was calculated. A manageable weight of stover was weighed and spread on concrete floor using forks. Similarly, the required amount of water was sprayed on the stover using watering can. The required amount of Calcium Oxide was weighed and dusted on the wetted stover followed by thorough mixing. The treated material was moved to the silo for ensiling.

Similar procedure was followed until all stover were treated. The treated stover was compacted and covered with double layer of polythene sheet and ensiled for 5 days, a recommended time for lime treatment (Russell *et al*., 2011). The CaO treated and untreated maize stovers (Treatment 1) were transported to Dodoma region (Kongwa National Ranch Company) for enzyme and microbial treatment. The limed maize stover was divided into four portions and treated differently as follows:

(a) Limed maize stover + water

The first portion of limed maize stover was set as Treatment 2. The DM of the stover was determined and amount of water required to bring the moisture content of stover to 50% was calculated. A manageable weight of stover was weighed and spread on the concrete floor using forks. Similarly the required amount of water was sprayed on the stover using watering can followed by thorough mixing and the material was moved to the ensiling place. Similar procedure was followed until all the stover was finished. The stover was compacted and covered with double layer of polythene sheet and ensiled for three weeks, a recommended time (Russell *et al.*, 2011).

(b) Limed maize stover + enzymes

The second portion of limed maize stover was treated with enzymes (NS2202) obtained from Novozymes, in Denmark and set as Treatment 3. The enzyme was in a solution form filled in plastic container and kept at room temperature. The enzyme to be applied at the rate of 0.3% of DM of the stover was weighed and dissolved in the calculated volume of water mixed and sprayed on the maize stover. A manageable weight of stover was weighed and spread on the concrete floor using forks. Similarly the required amount of water with the enzyme was sprayed on the stover using watering can followed by thorough mixing and the material was moved to the ensiling place. Similar procedure was followed until all the stover was finished. The stover was compacted and covered with double layer of polythene sheet and ensiled for three weeks.

(c) Limed maize stover +microbial material

The third portion of limed maize stover was treated with microbes (NZS086) obtained from Novozymes, in Denmark and set as Treatment 4.The microbes was in a form of powder packed in metal container, was kept at room temperature. The microbial mixture at a rate of 0.0025% of DM stover was weighed and mixed with the calculated volume of water. A manageable weight of stover was weighed and spread on the concrete floor using forks. Similarly the required amount of water and microbes were mixed and sprayed on the stover using watering can followed by thorough mixing and the material was moved to the ensiling place. Similar procedure was followed until all the stover was finished. The stover was compacted and covered with double layer of polythene sheet and ensiled for three weeks. Maize stover as one of low degradation rate requires, times as long as three weeks for a significant change in the structure of maize stover.

(d) Limed maize stover + enzymes+ microbial material

The fourth portion of limed maize stover was set as Treatment 5. The Enzyme and Microbes to be applied at the rate of 0.3% and 0.00256% of DM stover respectively was calculated. A manageable weight of stover was weighed and spread on the concrete floor using forks. Similarly the amount of water to bring the moisture content to 50% was calculated, enzymes and microbes were mixed with water and sprayed on the stover using watering can and mixed with forks. The treated material was moved to ensiling place. Similar procedure was followed until all the stover was finished. The stover was compacted and covered with double layer of polythene sheet and ensiled for three weeks.

3.3 Supplementary Concentrate

A concentrate diet was formulated from 30% hominy meal, 37% molasses, 30% cotton seed cake,1% urea, 1% mineral mix, 0.5% salt and 0.5% limestone to contain 14% CP and 13 ME MJ/kg DM. The dry component were mixed together in a commercial feed mixer and the molasses component was added during feeding and offered to treatment animals at a rate of 2kg per day per animal as a supplement.

3.4 Evaluation of Differently Treated Maize Stover

3.4.1 Sample preparation

At the end of the study collected samples (feed, concentrates and concentrate ingredients) were dried in oven at 60^0C to constant weight. All samples were milled using a Christy and Norris hummer to pass through 2mm screen for degradability and 1mm screen for chemical, mineral and gas production measurements.

3.4.2 Chemical analysis

Samples of differently treated maize stover, refusals, faecal and supplementary feed and its ingredients were analysed for dry matter (DM), crude protein (CP), crude fiber (CF), ether extract (EE) and ash using AOAC (1995) procedures where as neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined according to Van Soest *et al.* (1991). Metabolizable energy (ME) was obtained using a prediction equation derived from Menke and Steingass (1988).

3.4.3 Mineral analysis

Five grams of the ground differently treated maize stover samples were weighed into labeled crucibles and ashed using furnaces at 550^0 C for 3 hours (AOAC, 1995). After which the crucibles were removed and cooled down in desiccators for 30 minutes. Then 20mls of dilute hydrochloric acid (50%) 1:1 was added to each crucibles containing ash sample and covered with lids and allowed to digest for 24 hours. The digested ash was then filtered separately into labeled graduated bottles using filter papers, after which the volume was made to 100mls with distilled water. The clear filtered solution was used in Calcium and Phosphorus determination. Calcium was analyzed using atomic absorption spectrometers (AAS) while P was determined using UV/ Vis Spectrophotometer.

3.4.4 Degradability

Degradability values for DM and OM were determined using the nylon bag technique as described by Ørskov and McDonald (1979) procedure, in which four fistulated animals (steers) were used for the incubation of samples. Two grams (2g) of each samples from differently treated and untreated maize stover (T_1 –T_7) ground/ milled to pass through a 2 mm screen was weighed in duplicate in labeled nylon bags of known weight. The bags were tied and inserted in the rumens of each of the four fistulated animals and incubated for 6, 12, 24, 48, 72, 96 and 120 hours. After each incubation period the bags were removed, washed and dried in oven set at 60^0C to constant weight for determination of DM of the residue. The contents from the bags were emptied into weighed dried crucibles and ashed at 550^0C for 3 hour, Degradability of %Dry Matter Loss (%DML) and %Organic Matter Loss (%OML) of the samples were calculated from disappearance of DM and OM from the bags after rumen incubation and washing.

$$\%DML = \frac{\text{Wt. DM incubated} - \text{Wt.DM residue}}{\text{Wt.DM incubated}} \times 100 \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(1)$$

$$\%OML = \frac{\text{Wt. OM incubated} - \text{Wt. OM residue}}{\text{Wt. OM incubated}} \times 100 \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(2)$$

An exponential description of degradation which fits most situations was used. The expression of the type: $P = a + b\,(1\text{-}e^{-ct})$ (Ørskov and McDonald, 1979) was used.

3.4.5 *In vitro* gas production

About 200 mg dry weight of the samples were weighed in triplicates into calibrated glass syringes of 100 mls and were incubated in rumen fluid following the procedures of Menke and Staingas (1988). Gas volume reading was recorded before incubation (0 h) and 3, 6, 12, 24, 48, 72, 96 and 120 h after incubation. The results (means of three runs) were fitted to the exponential equation of the form $G = b(1\text{-}e^{-ct})$ suggested by Menke and Steingass (1988) G = volume of gas production (ml)/ 200mg DM with time t, b = asymptote exponential b $(-e^{-ct})$, c = rate of gas production. These degradation constants were used to estimate effective degradability (ED) following the model of (Ørskov and McDonald, 1979); ED=a+[bc/ck+c] where k = percentage outflow rate from the rumen estimated at 3%.

3.4.6 Performance of animals (study I)

Study I was done at Kongwa ranch which is located between latitude 5 ^{o} 36, and 6^{o} 10, S and longitude 36 ^{o} 15’ and 36^{o} 46, East. The climate of Kongwa is semi arid with a long dry season with a mean temperature ranges from 20 - 26^{0}C and average annual rainfall ranging between 500 to 800mm.

3.4.6.1 Experimental animals and design

A total of 80 beef cattle of 2 years of age with mean weight of 181.7 ± 5.52 kg were randomly allocated to 5 dietary treatments in a completely randomized design. Each treatment consisted of 16 beef cattle which were confined in pens, two animals per pen.

3.4.6.2 Feeds and feeding

The animals were fed on differently treated maize stover on *ad lib* bases allowing 20% refusal rate and supplemented with 2 kg of concentrate indicated in section 3.3 per animal per day. All animals had access to clean drinking water all the time and feeding lasted for 35 days.

3.4.6.3 Data that were recorded

(a) Body weights

Initial animal body weights were taken for three consecutive days and the mean weight was taken as the initial weights. Thereafter, each animal was weighed weekly in the morning before feeding. At the end of experiment animals were weighed for three consecutive days and the mean weights were taken as final weights. Growth was determined as a difference between initial weight and final weights.

(b) Feed intake

The animals were fed twice per day at 07.30 and 03.00 hours. The amount of feed offered to the two animals per pen was weighted daily and refusals collected and weighted on the next day and dried in an oven to get DM content of the refusal. The difference between feed DM offered and refused DM was considered as intake for the two animals in a pen, the value was divided by 2 to obtain individual intake. Feed conversion ratio was obtained by the relationship of feed intake (DMI) to weight gain during the fattening period.

3.5 Statistical analysis (study I)

3.5.1 Chemical composition, degradability and *in vitro* gas production

The values obtained at the various incubation periods for both the *in sacco* dry matter degradability (DMD) and the *in vitro* gas production of the differently treated maize stover were subjected to statistical analysis of variance using General Linear Model (GLM) procedure of the statistical analysis system (SAS, 2000). Where means were generated for variables studied, when means were significantly different by ANOVA at $P<0.05$, they were separated by Least significant difference test.

Statistical model adopted:

$Y_{ij} = \mu + T_i + e_{ij}$ ……………………………………………………………..…..(3)

Where $Y_{ij} =$ record of j^{th} sample belongs to i^{th} stover treatment, μ = Overall mean, $T_i =$ effect due to i^{th} stover treatment and $e_{ij} =$ random error.

Animal performance (study I), the independent variables assessed were the dietary treatments. The data obtained for the dependent variables weight gain, feed intake and feed conversion efficiency were subjected to statistical analysis of variance using GLM procedure of SAS (2000). Differences in weight at the beginning of the study were adjusted by analysis of covariance. Where means were generated for variables studied, when means were significantly different by ANOVA at $P<0.05$, they were separated by Least significant difference test.

The model used.

$Y_{ij} = \mu + Ti + b(x_{ij} - \overline{X}) + eij$ …………………………………………………...…..(4)

Where; $Y_{ij} =$ record of j^{th} animal belongs to i^{th} dietary treatment, μ = Overall mean, $T_i =$ effect due to i^{th} dietary treatment, $x_{ij} =$ Initial body weight of an individual animal, b = regression of Y_{ij} on x_{ij}, $\overline{X}$ = Overall mean of initial body weight, $e_{ij} =$ random error.

3.6 Study II: Treatment of Maize Stover

The chopped maize stover harvested in 2013 was divided into two portions. One portion of the maize stover was not treated with $Ca(OH)_2$ and was set aside to act as control diet during the second feeding trial and was considered as Treatment 6. The second portion of maize stover (3250 kg) was treated with Calcium hydroxide ($Ca(OH)_2$) that was imported from Denmark and set aside as Treatment 7. The DM of the stover was determined and amount of water required to bring the moisture content of stover to 50% was calculated.

$Ca(OH)_2$ to be applied at a rate of 6.6% of DM stover was calculated. The stover was weighed and spread on the concrete floor using forks. A required amount of water and $Ca(OH)_2$ were measured and solution was sprayed on the stover using watering can followed by thorough mixing. The treated material was moved to the ensiling place, compacted and covered with double layer of polythene sheet and ensiled for 7 days, as recommended period for $Ca(OH)_2$ treatment by Peterson (2014).

3.7 Supplementary Concentrate

Supplementary concentrate as explained in the section 3.3 was provided to the animals that is 2 kg / day / animal.

3.8 Evaluation of Differently Treated Maize Stover

Evaluation of differently treated maize stover for chemical and mineral analyses followed procedures described in section 3.4.1, 3.4.2 and 3.4.3

3.8.1 *In vivo* digestibility using marker technique

(a) Experimental design and treatment

In vivo digestibility using marker technique was carried out using a total of 32 beef cattle aged 2 years and mean initial weight of 195 ± 3.18kg that were randomly allocated to 2 dietary treatments (untreated and hydrated lime treated maize stover harvested in 2013) in a complete randomized design. Each treatment consisted of 16 beef cattle which were confined in pens, two (2) animals per pen.

(b) Feed and feeding

Animals were fed twice on either untreated or $Ca(OH)_2$ treated maize stover at an *ad libitum* allowing 20% refusal rate and supplemented with 2 kg per animal per day of concentrate indicated in section 3.3. The animals were fed at 07.30 and 03.30 hour. All animals had access to fresh clean drinking water all the time. Feed and feed refusals samples were collected every morning for the subsequent analysis.

(c) Faecal sample collection

Faecal samples were obtained from each animal under study by grab method at 4 hour interval for 12 hour period which made a total of three samples per animal per day. The faecal samples were weighed and 20 – 30% of each collected faecal sample was sub-sampled weighed, sun dried, partially ground and stored in plastic bags until the end of the collection period of 10 days.

(d) Sample preparation and chemical analysis

At the end of the collection period, the collected stored samples, (feed, feed refusal and faeces) were dried to a constant weight in a forced air oven set at 60^0C. The daily dried faecal samples collected from each animal for ten days were mixed together to obtain one sample per animal. The samples were ground, and packed in labeled bottles for subsequent analysis. Feed and refusal samples were also ground, and then four (4) representative samples for 10 days were obtained.

(e) Measurement of dry matter and organic matter digestibility

The 2N HCL Acid Insoluble Ash analytical procedure of Van Keulen and Young (1977) was used for determination of AIA content of feed and faeces samples. Acid-insoluble ash which is a natural constituent of the feed was used as internal marker to measure digestibility coefficients of the ration. A duplicate of 5 g sample of the dried and ground faeces were weighed into 50 ml crucible and dried in a forced air oven (135^0C) for 2 hour. The samples were cooled in a desiccators to room temperature, re-weighed (Ws) and then ashed for 3 hours at 450^0 C. The ash was weighed and transferred to a 600 ml Berzelius beaker and 100 ml of 2N HCL was added. The mixture was boiled for 5 minute in a hotplate. The hot hydrolysate was filtered using ash free filter paper of known weight and washed free of acid with hot distilled water (85 and 100^0C).

The ash and filter paper were transferred into a crucible of known weight and was dried weighed and then ashed for 3 hours at 450^0C. The crucible and the content were cooled in a desiccator to room temperature, weighed while containing ash (Wf) and re-weighed immediately after emptying (We). Percentage AIA was calculated from the equation: (Wf – We)/Ws *100, where Wf = weight of crucible with insoluble ash, We = weight of empty crucible and Ws = weight of sample dry matter. The 2N HCL procedure involved an initial oven drying and ashing step of the feed and faeces which allowed determination of the dry matter and organic matter content of the actual analyzed samples.

The digestibility coefficients was calculated as follows:

$$\text{Apparent Digestibility (\%)} = 100 - \frac{\text{percentage indicator in feed}}{\text{percentage indicator in faeces}} \text{x } 100 \ldots\ldots.\ldots.(5)$$

Further more degradability, *in vitro* gas production, were done using procedures described in section 3.4.4 and 3.4.5. In this study a total of 32 steers were divided in two groups group one was fed untreated maize stover while group 2 was fed $Ca(OH)_2$ treated maize stover for a total of 42 days each.

The study used same animals that were used in section 3.8.1 (a). Likewise feed and feeding was as described in section 3.8.1 (b), untreated and treated maize stover were used.

3.8.2 Data that were recorded

Body weights and feed intake was recorded as indicated in section 3.4.6.4 (a) and 3.4.6.4 (b).

3.9 Statistical analysis (study II)

Data for digestibility and animal performance were analyzed using t - test procedure.

The model adopted was:

$$t = \frac{\bar{x}_1 - \bar{x}_2}{S_{x_1 \ x_2} * \sqrt{\frac{1}{n}}} \quad \text{…………………………………………………………………..…..(6)}$$

Where t = t - test, $\bar{x}_1$ = mean for treatment 6 (T_6), $\bar{x}_2$ = mean for treatment 7(T_7) S_{x1x2} = Pooled standard deviation of the two treatments, $\sqrt{\frac{1}{n}}$ = square root inverse of n and n= number of observations.

While data for chemical composition, degradability and *in vitro* gas Production in Study II were analyzed together with data from study I as indicated in section 3.5.1

CHAPTER FOUR

4.0 RESULTS

4.1 Quality of Differently Treated Maize Stover

4.1.1 Chemical composition (study I and II)

The chemical composition of basal diets, concentrates and concentrates ingredients used in study I and II are presented in Table 4. Dry matter content of all diets, concentrates and concentrates ingredients used were fairly similar. Crude protein ranged from 2.55 to 4.33% for basal diets and for concentrates and ingredients ranged from 14.1 to 34.5% DM. There was a slight increase in CP of the treated maize stover. Treatment of maize stover with CaO reduced NDF content by 10.1 units while treatment with $Ca(OH)_2$ reduced it by 16 units. Maize stover treated with lime + enzymes with microbes had slightly higher NDF values than those treated with lime only. In the current study the ADF of CaO, enzymes, microbes and $Ca(OH)_2$ treated maize stover were higher compared to untreated maize stover shown in Table 4.

The treated maize stover had higher ash and Ca^{2+} content than untreated maize stover. The chemical components of the concentrate ingredients were within the expected range.

4.1.2 Dry matter degradability (study I and II)

The degradation characteristics of DM for the differently treated maize stover differed significantly ($P < 0.05$) as shown in Table 5. The results showed that the chemical treatments applied in the current study altered the degradation coefficients of the maize stover. The immediately soluble **'a'** fraction ranged from 3.17% in (T_1) to 15.8% in (T_2). (T_7) was observed to contain the highest amount of potentially degradable DM of 86.4%.

Table 4: Chemical composition of different feeds and feed ingredients that were fed to the experimental animals

	Chemical composition %DM										
Feedstuffs	DM	OM	CP	EE	CF	NFE	Ash	NDF	ADF	Ca	P	MEMJ/kg DM
(T_1)	93.6	92.6	2.71	0.28	36.9	46.3	7.44	78.3	44.5	0.03	0.15	7.20
(T_2)	76.2	85.6	4.16	0.25	43.8	13.6	14.4	68.2	43.3	0.96	0.10	6.90
(T_3)	83.7	86.1	3.15	0.35	46.7	19.6	13.9	73.6	51.1	0.92	0.18	7.00
(T_4)	80.7	85.9	2.55	0.38	42.6	30.1	14.1	68.3	44.2	0.80	0.10	5.50
(T_5)	77.8	88.1	2.99	0.29	39.2	23.4	11.9	75.0	52.4	0.66	0.07	6.20
(T_6)	92.7	89.1	3.39	0.47	30.0	47.9	10.9	79.7	48.7	0.15	0.11	6.40
(T_7)	73.9	85.2	4.33	0.29	39.3	15.2	14.8	63.7	44.1	0.80	0.07	6.60
Conc.	72.3	90.1	14.1	5.66	6.30	36.3	9.91	29.8	9.66	0.32	0.62	12.4
CSC	84.3	93.8	34.5	7.13	14.0	21.4	6.23	36.0	23.1	0.12	0.88	13.2
HM	87.5	95.0	15.3	2.08	2.64	62.5	4.99	39.6	8.25	0.03	0.86	12.8
Molasses	45.9	88.1	3.86	0.16	0.00	30.0	11.9	1.34	0.59	0.24	12.8	12.6

In this and subsequent tables % DM = Dry matter percentage, OM = Organic matter, CP = Crude protein, EE = Ether extracts, CF = Crude fiber, NFE = Nitrogen free extract, NDF = Neutral detergent fiber, ADF = Acid detergent fiber, ME = Metabolizable energy (Mega Joules/kgDM), Ca = Calcium, P= Phosphorus, T_1 = Untreated maize stover 2012, T_2 = Treated maize stover with CaO 2012, T_3 = Treated maize stover with CaO and enzymes, T_4 = Treated maize stover with CaO and micro organisms, T_5 = Treated maize stover with CaO, enzymes and micro organisms, T_6 = Untreated maize stover 2013, T_7 = Treated maize stover with slaked lime ($Ca(OH)_2$) 2013, Conc. = Concentrates, CSC = Cotton seed cake, HM = Hominy meal.

Effective degradability (ED) of DM calculated at 3% outflow rates from the rumen showed that (T_2) consistence had significantly highest values while the least value was recorded in (T_6).The disappearance of the DM contents in the differently treated maize stover by the end of 48 hrs of incubation is shown in Table 5. At 48 hrs incubation the estimates for DM disappearance from the (T_7) was higher than that of other treatments.

Table 5: Dry matter degradability (DMD) constants of untreated and treated maize stover

Diet types	a	b	c	48 h D.	PD	ED (3%)
(T_1)	3.17^{g}	62.5^{e}	0.02^{a}	46.2^{b}	65.7^{e}	30.3^{b}
(T_2)	15.8^{a}	67.1^{b}	0.01^{b}	43.0f	82.9^{b}	31.0^{a}
(T_3)	6.87^{d}	64.7^{d}	0.01^{b}	40.9^{g}	71.6^{c}	28.3^{d}
(T_4)	7.53^{c}	47.1^{g}	0.02^{a}	45.3^{c}	54.7^{g}	26.6^{e}
(T_5)	4.78^{f}	65.7^{c}	0.02^{a}	42.3^{e}	70.4d	29.8^{c}
(T_6)	6.02^{e}	56.7^{f}	0.01^{b}	43.3^{d}	62.7f	23.6^{g}
(T_7)	12.2^{b}	74.2^{a}	0.02^{c}	51.2^{a}	86.4^{a}	26.4^{f}
SEM	0.07	0.11	0.003	0.07	0.07	0.06
P-value	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001

$^{a\ b\ c\ d\ e\ f\ g}$ Column means with same superscripts do not differ (P >0.05). a = Intercept, b = Slowly degradable component (Potentially degradable fraction), c = Rate of degradability constant, 48 h D= 48 hours degradability, PD =Potential degradability, ED = Effective degradability (3%) and SEM = Overall standard error of mean

Degradation of dry matter is graphically illustrated in Figure 1 and 2. Fig.1 shows the trend of dry matter degradability from study I. There was an increase in dry matter degradability with time in all treatments from 12 hour to hour 72 h although the magnitude of increase varied between treatments. After 72 h of incubation (T_1) and (T_5) had reached asymptote while the other treatments were still increasing.

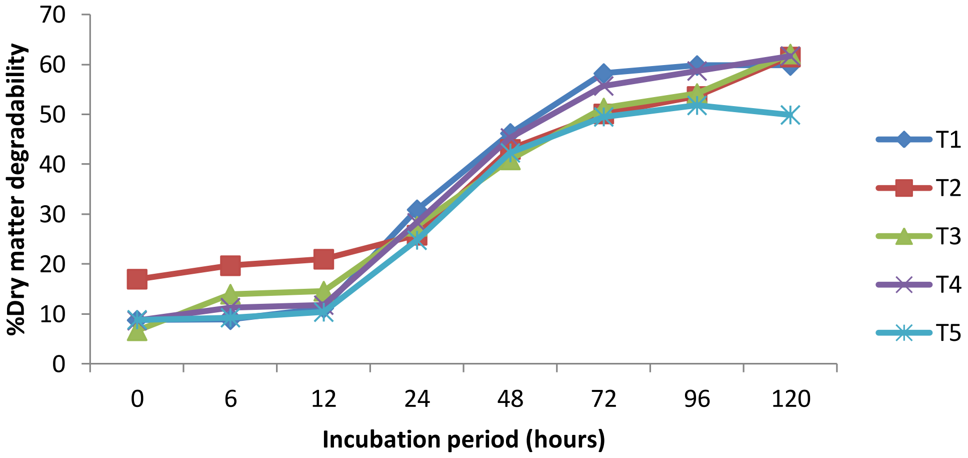

Figure 1: Effect of Treating maize stover with CaO, enzymes and microbes on Dry Matter degradability

Fig. 2 Shows the trend for dry matter degradability of untreated and maize stover treated with $Ca(OH)_2$. There was increase in dry matter degradability from 6 hour to hour 120 for the $Ca(OH)_2$ treated maize stover while for the untreated maize stover there was a minimal increase after 72h of incubation.

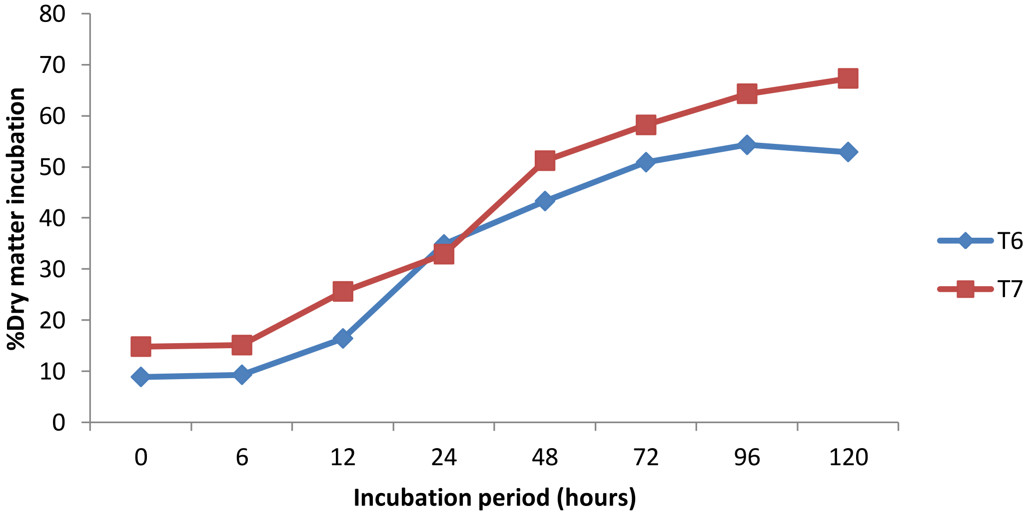

Figure 2: Effect of treating maize stover with $Ca(OH)_2$ on Dry Matter degradability

4.1.3 Organic matter degradability (study I and II)

Estimates for OM degradation characteristic of differently treated maize stover studied are shown in Table 6. Variations in all the parameters measured were significant.

The readily soluble OM fractions **`a'** was significantly higher in (T_2) with a value of 10.8% as compared to least value of (-0.34%) in (T_6). (T_3) had the highest estimate for the insoluble but rumen degradable fraction '**b**' with value of (85.9%) as compared to the least value of 53.9% in (T_4). The estimated degradation rate '**c**' differed significantly for differently treated maize stover. The rate of degradation '**c**' of OM in the differently treated maize stover was fastest in (T_1), (T_4), (T_5) and (T_6) (0.02/hr). Potentially degradable OM was higher in the (T_3) with values of 91.2% than other treatments. Effective degradability as calculated at 3% outflow rate from the rumen showed that (T_1) had significantly higher ($p<0.05$) values while the least values were recorded in (T_6). The disappearance of the OM in all treatments (differently treated maize stover) from the *in situ* bags increased with increasing incubation time. At 48hrs incubation time, the estimate for OM disappearance from the (T_6) was higher than that of other treatments.

Table 6: Organic matter degradability (OMD) constants of untreated and treated maize stover

Diet types	a	b	c	48 h D	PD	ED (3%)
($T_{1`}$)	1.02^{f}	67.1^{e}	0.02^{a}	46.8^{a}	68.1^{f}	30.3^{a}
(T_2)	10.8^{a}	71.6^{c}	0.01^{b}	45.2^{b}	82.4^{b}	28.2^{d}
(T_3)	5.29^{b}	85.9^{a}	0.014^{c}	41.8^{d}	91.2^{a}	28.9^{c}
(T_4)	2.38^{d}	53.9^{g}	0.02^{a}	40.3^{f}	56.3^{e}	25.7^{e}
(T_5)	1.97^{e}	70.9^{d}	0.02^{a}	46.7^{a}	80.0^{c}	29.8^{b}
(T_6)	-0.34^{g}	60.7^{f}	0.02^{a}	47.0^{a}	60.3^{d}	20.6^{g}
(T_7)	3.13^{c}	79.2^{b}	0.01^{b}	43.2^{c}	82.3^{b}	22.6^{f}
SEM	0.03	0.09	0.002	0.12	0.09	0.09
P-value	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001

$^{a\ b\ c\ d\ e\ f\ g}$ Column means with same superscripts do not differ ($P > 0.05$). a = Intercept, b = Slowly degradable component (Potential extent of degradability), c = Rate of degradability constant, 48 h D = 48 hours degradability, PD =Potential degradability, ED = Effective degradability (3%) and SEM = Overall Standard Error of Mean

Degradation of organic matter (OM) of differently treated maize stover is graphically illustrated in Figure 3 and 4. Fig. 3 shows the trend of organic matter degradability of maize stover used in study I. There was an increase in organic matter degradability in all treatments from 0 hour to120 hours except for (T_4) and (T_1) although the magnitude of increase varied between treatments. The graphs shows that the OM degradability of (T_4) reached asymptote after 72 h while that of (T_2) reached asymptote after 96 h while others were still rising even after 120 h.

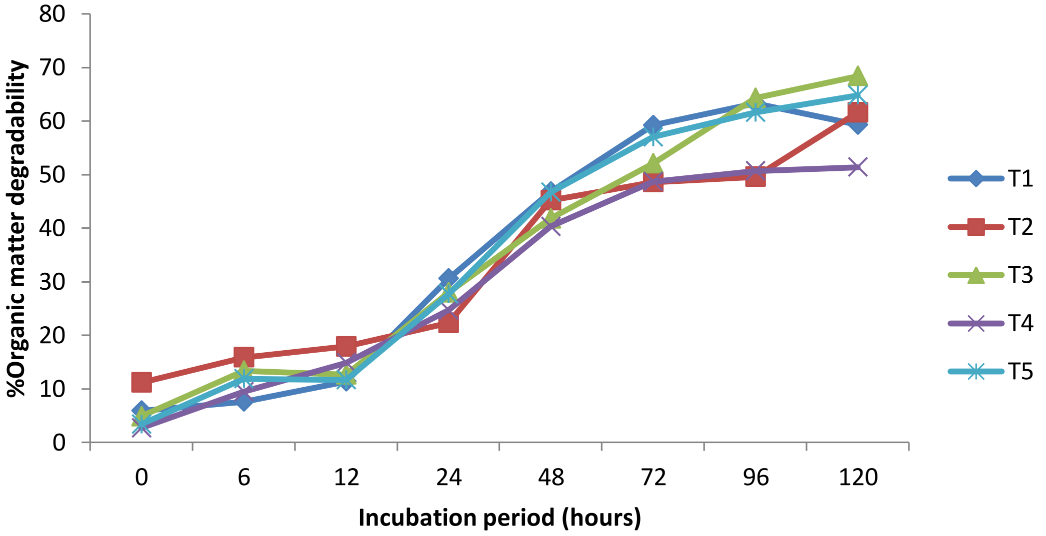

Figure 3: **Effect of Treating maize stover with CaO, enzymes and microbes on Organic Matter degradability**

Fig. 4 Shows the trend on organic matter degradability of untreated and $Ca(OH)_2$ treated maize stover used in study II. There was an increase in organic matter degradability in the two treatments from 0 h to 96 h although the magnitude of increase was different between treatments. The increase in OM degradability with time showed an increase with decreasing rate after 72 h of incubation for (T_6) and both reached asymptote after 96 h of incubation.

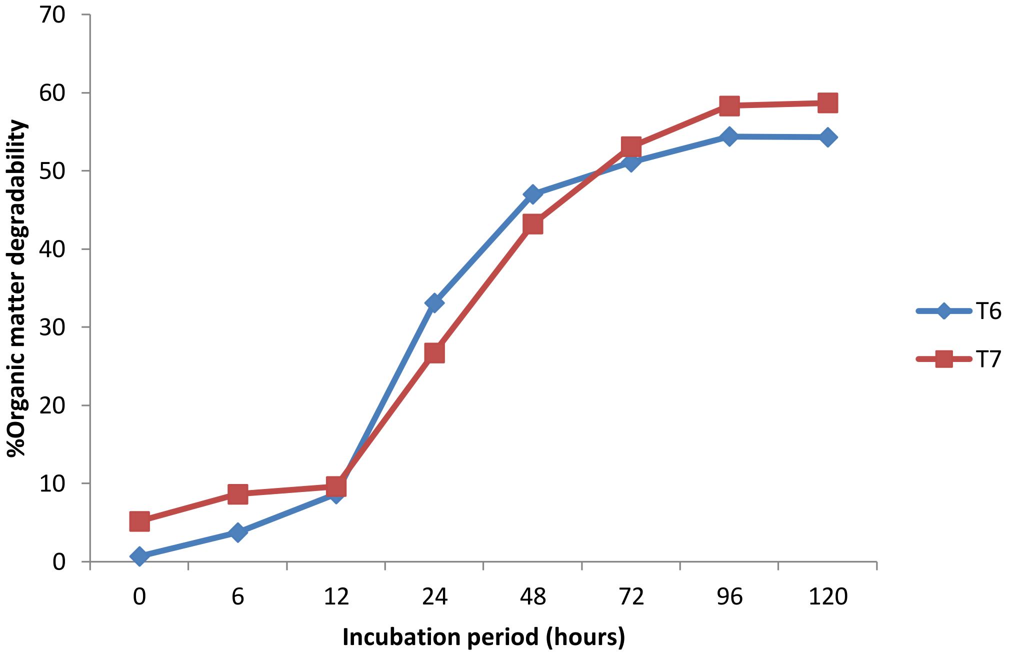

Figure 4: **Effect of Treating maize stover with $Ca(OH)_2$ on Organic Matter degradability**

4.1.4 *In vitro* gas production

In vitro gas production parameters (b and c) and OMD% values of the experimental samples are shown in Table 7. The **'b'** fraction of gas production was significantly higher in (T_5) and (T_2) than other treatments. While there was an increase in '**b**' fraction of gas production for differently treated maize stover than untreated.

Fig. 5 Shows the trend of *in vitro* gas production of differently treated maize stover. There was an increase in gas production in all treatments from 12 hour to 96 hours although the magnitudes of increase varied between the treatments. There was lower gas production in (T_5) followed by (T_4). While (T_1) had higher gas production at all incubation times. Asymptote gas production was attained after 96 h of incubation.

Table 7: *In vitro* gas production and OMD of untreated and treated maize stover (ml/200mg DM)

Treatments	Gas Production Parameters ml/200mg DM			
	b	c	EGP	%OMD
(T_1)	36.2^{b}	0.02	9.46^{b}	52.1^{d}
(T_2)	44.6^{a}	0.02	13.5^{a}	54.3^{b}
(T_3)	43.0^{ab}	0.02	13.0^{a}	55.0^{a}
(T_4)	40.2^{ab}	0.02	12.1^{a}	44.0^{g}
(T5)	46.1^{a}	0.02	10.7^{b}	48.2^{f}
(T_6)	33.4^{c}	0.02	11.0^{ab}	49.3^{e}
(T_7)	38.8^{b}	0.02	11.4^{ab}	53.1^{c}
SEM	0.21	0.003	0.26	0.25
P-value	0.0001	0.98	0.001	0.0001

$^{a\ b\ c}$ Column means with same superscripts do not differ ($P > 0.05$).p, b and c are described by equation $p = b(1-e^{-ct})$, where p = volume gas produced with time (h), c = rate constant, b = Potential extent of gas production rate (%h).The intercept is not included in the model as there will be no gas production from unfermented feed. EGP = Effective gas production, %OMD = Percentage organic matter digestibility

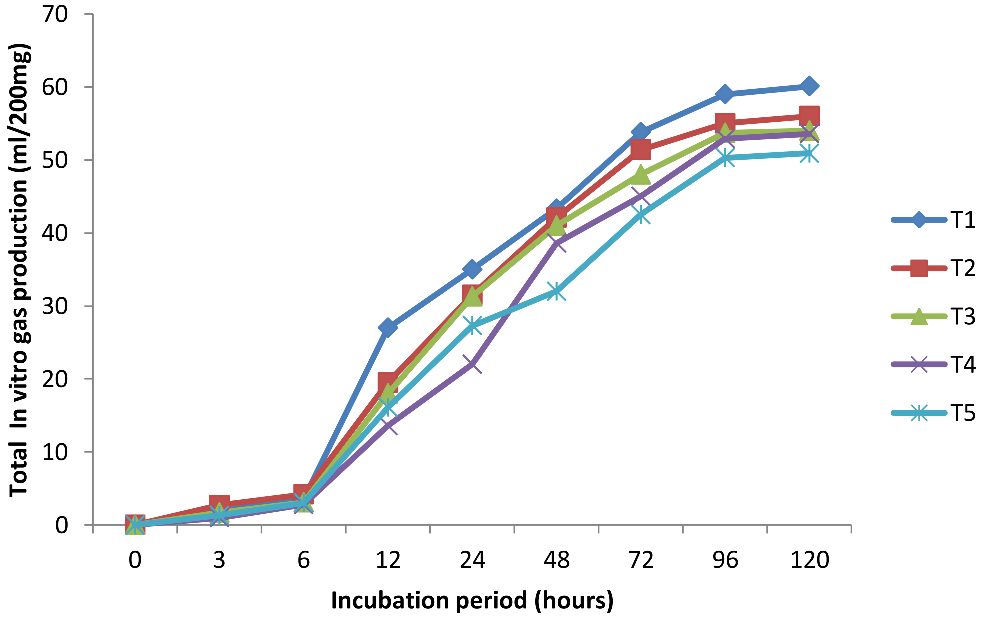

Figure 5: Effect of treating maize stover with CaO, enzymes and microbes on *in vitro* gas production

Fig. 6 Shows the trend of gas production from untreated and $Ca(OH)_2$ treated maize stover. There was an increase *in vitro* gas production from 6 hour to 96 hours for the untreated and maize stover treated with $Ca(OH)_2$. The *in vitro* gas production of the two treatments (untreated and treated maize stover) (T_6) and (T_7) started at the same level at 0 hour but followed different patterns thereafter.

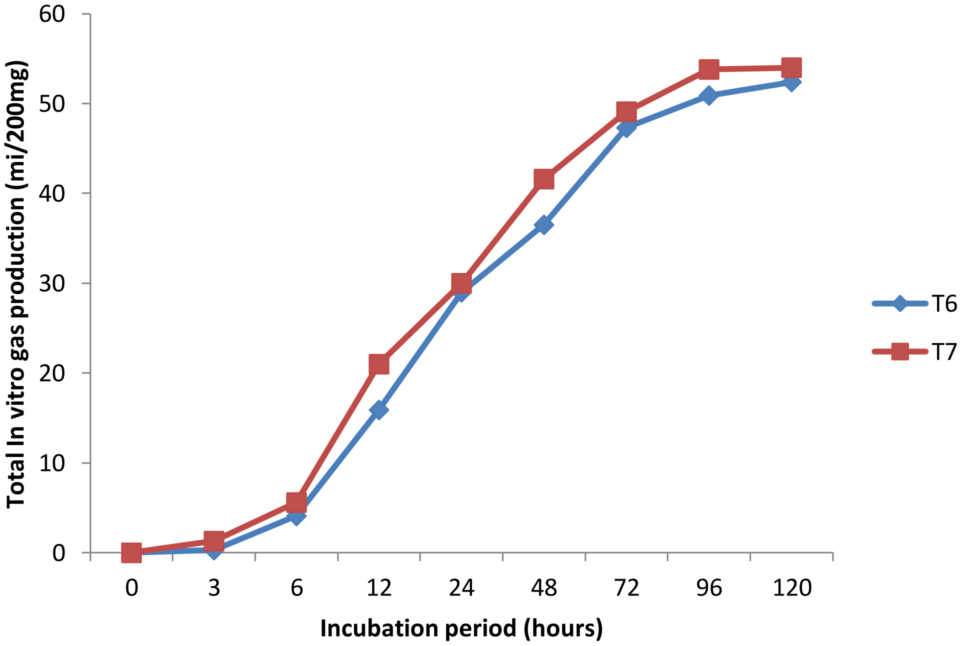

Figure 6: Effect of treating maize stover with $Ca(OH)_2$ on *in vitro* gas production

4.1.5 Animal performance in study I

Total daily dry matter intake of the animals fed differently treated maize stover (study I) was not significantly different (P>0.05) between the five treatments. Least square means for feed DM intake, are shown in Table 8. Intake levels on dry matter (DM) basis ranged from 4.83 ± 0.11 (T_2) to 5.03 ± 0.11 kg DM/day for (T_1) respectively. The lowest DM intake was observed in animals that consumed CaO treated maize stover

(4.83 ± 0.11). Whilst total energy intake (obtained by multiplying energy concentration and DM intake per day) ranged from 43.8 to 48.9 ME/day respectively. Feed conversion ratio expressed as kg feed/kg gain was found to be better for animals that consumed untreated maize stover (T_1) (3.89) followed by animals that consumed treated stover (T_2) (4.67) followed by animals that consumed treated stover (T_3) (4.98) where (T_5) (7.47) and the least were (T_4) (7.52).

The average daily gain was not significantly different (P>0.05) among the five treatments, although average daily weight gain was slightly higher for animals under (T_1) than other treatments.

Table 8: LS means ± SEM for performance of steers fed untreated and treated maize stover (study 1)

	Treatments/diet types						
Parameters	T_1	T_2	T_3	T_4	T_5	SEM	P-value
Initial wt.(kg)	173	179	178	190.35	188	5.52	-
Weekly body weights							
W1	201	198	202	197	198	3.18	0.6585
W2	207	203	205	203	203	3.07	0.8818
W3	211	208	207	206	206	3.02	0.7034
W4	214	212	212	209	209	3.21	0.4589
W5(Final weight) (kg)	218	216	213	213	211	3.08	0.4586
Wt. gain for 35days(kg)	45.0	37.0	35.0	22.7	23.0	3.07	0.4586
ADG (kg)	1.29	1.06	1.00	0.65	0.66	0.09	0.4572
Conc. DMI (kg)	85.3	85.3	85.3	84.1	85.3	0.01	0.1251
Stover DMI (kg)	91.2	84.1	88.9	87.3	87.1	1.04	0.1422
TDMI (kg)	176	169	174	171	172	3.68	0.1882
ADFI (kg/day)	5.03	4.83	4.97	4.89	4.93	0.11	0.1902
FCR (feed/gain)	3.89	4.67	4.98	7.52	7.47	1.33	0.2604
ME Intake	48.9	46.7	47.9	43.7	45.6	2.51	0.1241

W1=Week 1, W2=Week2, W3=Week3, W4=Week4, Wt. = Weight, ADG= Average day gain, Conc.DMI = Concentrate dry matter intake, Stover DMI = Stover dry matter intake, TDMI= Total Dry Matter intake, ADFI= Average dry matter intake per day, FCR = Feed conversion ratio and ME =Metabolizable Energy

4.1.6 *In vivo* digestibility in study II

Table 9: *In vivo* dry matter digestibility of untreated and maize stover treated with Calcium hydroxide

Diet types	% Dry matter Digestibility
(T_6)	55.7^b
(T_7)	70.2^a
SEM	0.72
t value	14.04
Pr>[t]	<.0001**

(T_6)= Untreated maize stover 2013, (T_7)= Maize stover 2013 Treated with hydrated lime and t=T- test. Maize stover treated with Ca $(OH)_2$ had higher ($P<0.0001$) *in vivo* dry matter digestibility than untreated maize stover.

4.1.7 Animal performance in study II

The weight gain between the two treatments was not significantly different ($P>0.05$) where animals that consumed untreated maize stover had slightly higher weight gain. Total daily dry matter intake of the animals in study II was not significantly different ($P>0.05$) between the two treatments. Least square means for feed DM intake, proportion of intake relative to body weight are shown in Table 10. Animals that consumed treated maize stover had lower dry matter intake than those that consumed untreated maize stover.

Intake levels on dry matter (DM) basis was from 4.11± 0.38kgDM/day for treated maize stover and 4.38 ± 0.38 kgDM/day for untreated maize stover. The lowest DM intake was observed in animals that consumed treated maize stover (4.11± 0.38). Where the total energy intake (obtained by multiplying energy concentration and DM intake per day) ranged from 35.6 to 36.8 ME/kg DM respectively, where the animals under (T_6) had the highest energy intake and the lowest was those animals under (T_7). Feed conversion ratio expressed as kg feed/kg gain was found to be better for animals that consumed treated maize stover (3.80) followed by animals that consumed untreated stover (4.11).

Table 10: LS means ± SEM for the performance of steers fed untreated and treated maize stover (study II)

	Treatments				
Parameters	T_6	T_7	SEM	t value	Pr >[t]
Initial weight (kg)	196	194	11.8	-0.43	0.6736
Weekly body weights					
W1	233	234	13.5	0.17	0.8668
W2	239	236	12.9	-0.64	0.5261
W3	240	237	11.8	-0.70	0.4888
W4	242	240	12.5	-0.44	0.6628
W5	244	242	12.9	-0.42	0.6744
Final Body weight (kg)	247	244	13.6	-0.61	0.5468
Wt. gain for 42days (kg)	51.0	50.0	6.17	-	0.3718
ADG (kg)	1.21	1.19	0.37	-0.22	0.8283
Conc. DMI (kg)	60.7	60.7	0.09	-9.51	0.7238
Stover DMI (kg)	123.6	112.4	15.9	-1.77	0.0870
TDMI (kg)	184	173	15.8	-1.77	0.0869
ADMI (kg)	4.38	4.11	0.11	-1.77	0.0591
FCR (Feed/gain)	4.11	3.80	1.58	0.56	0.5782
ME Intake	36.8	35.6	5.17	-	0.3581

Means within the same row with different superscript letters are significantly different ($p < 0.05$).
W1=week1, W2= week2, W3= week3, W4 = week4 and W5 = week 5,ADG = Average Daily Gain, TDMI = Total Dry matter Intake, ADFI = Average Day Feed Intake, FCR = Feed Conversion Ratio and ME = Metabolizable Energy

CHAPTER FIVE

5.0 DISCUSSION

5.1 Chemical Composition

The observed Crude Protein (CP) of the differently treated maize stover in the present study was slightly higher for T_2 and T_7 than those of untreated stover. The possible cause could be increase in microbial biomass growth in the form of single cell. Even though fungal growth could not be observed by eyes but on use of microscope could have revealed their growth. Similar increase in CP content in maize stover treated with 5% CaO and 6.6% $Ca(OH)_2$ were reported by Russell *et al.* (2011); Shreck *et al.* (2011); Johnson *et al.* (2013); Russell (2013). The lower CP values for maize stover treated with lime + microbes and those treated with lime +Enzymes + microbes could be due to uptake of some N from the maize stover by the microbes. These results were in agreement with those observed by Koc *et al.* (2008) who found a decrease in CP content of maize silage inoculated with microbes. The increased ash content observed in the current study might be caused by rapid absorption of minerals from the solution of Calcium Oxide and Calcium hydroxide. The absorption of minerals from the solution is related to the differences in minerals concentration between the solution and the maize stover. Since maize stover has low mineral content the minerals moved from the solution to the stover. Similar observations were reported by Mtamakaya (2002) who found that the mineral content of rice straw treated with wood ash extract was increased by 40.5% of the original content.

In the present study the CaO treated maize stover had higher Ca content than untreated maize stover, an increase of about 0.93% units. This was due to absorption of Ca mineral from the solution used in sprayed maize stover. Similar observation were reported by

Johnson *et al.* (2013) that Ca content was increased by about 0.07% for CaO treated maize stover when compared with untreated.

The Neutral Detergent Fiber (NDF) of differently treated maize stover was reduced relative to the values for the untreated stover. This could be due to dilution caused by increasing ash content of the differently treated maize stover. Similar observations were reported by Russell (2013) where maize stover treated with 5% CaO had NDF reduced by about 8.6% units. Similarly reduction of NDF content as a result of lime treatment was reported for rice straw (Nguyen *et al*., 2001). Urea treatment of rice straw was found to delignify hemicellulose thus reducing its NDF content (Nguyen *et al.,* 2001). The extent of release of aromatic compound and cell wall sugars depends on the strength of the alkali (Doyle *et al*., 1986). In the current study the ADF content of differently treated maize stover with CaO, enzymes, microbes and $Ca(OH)_2$ were higher compared to untreated stover. The increased ADF is probably due to proportionate decrease in NDF due to dilution caused by increase ash and solubilization of hemicelluloses during $CaO/Ca(OH)_2$ treatment and increase in lignin. Similar observation was reported by Russell *et al.* (2011).

5.2 Dry Matter and Organic Matter Degradability of Differently Treated Maize Stover

Degradability of maize stover treated with CaO only and $Ca(OH)_2$ were higher than those of untreated. The results are in line with the previous results by Kilongozi (1992); Shem (1993); Kimbi (1997), who treated maize stover with urea. DM and OM degradability for differently treated maize stover showed increase in degradability than untreated maize stover.

The observed '**a**' values for DM degradability (4.60) and OM degradability (0.34) for untreated maize stover in the current study were lower than those reported by Kimbi (1997) and Shem (1993) for DM degradability for untreated maize stover. This discrepancy could be due to different maize stover variety respond different to chemicals (Shem, 1986). The observed '**a**' values for DM degradability (9.44) and OM degradability (4.71) for treated maize stover in the current study were lower than those reported by Kimbi (1997) and Shem (1993) for DM degradability for 3% urea treated maize stover. The possible reason could be maize stover variety respond differently to chemical treatment. The '**a**' values for DM degradability of rice straw treated with 3%, 6% lime and 2%, 4% urea reported by Nguyen *et al.* (2001) were higher than one observed in the current study.

The readily soluble fraction of organic matter in the current study for differently treated maize stover was observed to be highest 10.8% in (T_2). This may be due to the presence of readily degradable nutrients, particularly non – structural ones like, protein and fat, and soluble carbohydrates components that may make organic matter readily degradable *in situ* in the rumen. The range of **'a'** value observed in the current study were lower than the range for roughage (2.33 to 16.12%) reported by Ariel *et al*. (1998). This implies that crop residues respond differently to chemical treatment.

The observed '**b'** values for DM degradability (59.6) and OM degradability (63.9) for untreated maize stover in the current study were lower than those reported by Kimbi (1997) for untreated maize stover. Whereas lower **'b'** values than those observed in the current study for DM degradability of untreated maize stover and untreated rice straw were reported by Shem (1993) and Nguyen *et al.* (2001) respectively. The observed '**b'** values for DM degradability (63.8) and OM degradability (72.2) for treated maize stover in the current study were lower than those reported by Kimbi (1997) for 3% urea treated maize stover and higher than those reported by Shem (1993) for 3% urea treated maize stover and for rice straw treated with 3% and 6% lime (Nguyen *et al*., 2001). The possible reason could be due to different crop residue respond differently to different chemical treatment.

The observed **'c'** values for DM degradability (0.02) and OM degradability (0.04) for untreated maize stover in the current study was similar for DM and higher for OM than those reported by Kimbi (1997) for untreated maize stover. Whereas higher **'c'** values for DM degradability for untreated maize stover than in the current study was reported by Shem (1993) and also same **'c'** values as in the current study for DM degradability of untreated rice straw was reported by Nguyen *et al.* (2001). The observed **'c'** values DM degradability (0.02) and OM degradability (0.04) for treated maize stover in the current study was lower for DM and higher for OM than those reported by Kimbi (1997) for 3% urea treated maize stover. The possible cause could be different varieties of maize stover and different crop residues respond differently to chemical treatment.

Whereas higher **'c'** value DM degradability for maize stover treated with 3% and rice straw treated with 6% lime were reported by Shem (1993) and Nguyen *et al*. (2001) respectively. The observed 48 h DM degradability values (44.8) and OM degradability (46.9) for untreated maize stover in the current study were lower than those reported by Kimbi (1997) for untreated maize stover. The observed 48 h DM degradability values (44.5) and OM degradability (43.9) for treated maize stover in the current study were lower than those reported by the same author for 3% urea treated maize stover. The possible cause could be maize stover variety respond differently to chemical treatment. The observed potential degradability values for DM degradability (64.2) and OM degradability (64) for untreated maize stover in the current study were lower than those reported by the same author for untreated maize stover. Whereas lower potential DM degradability than in the current study for untreated maize stover and untreated rice straw were reported by Shem (1993) and Nguyen *et al.* (2001) respectively.

The observed potential degradability values for DM (73.2) and OM (78.4) for treated maize stover in the current study were lower than those reported by Kimbi (1997) for 3% urea treated maize stover. Whereas lower potential DM degradability than in the current study for 6% lime treated rice straws was reported by Nguyen *et al*. (2001). The possible reason could be due to different crop residue respond differently to different chemicals treatment as reported by Mgheni *et al*. (2001) who observed higher potential degradability for rice straw than maize stover when they were treated with 3% urea.

The observed amounts of low soluble fraction, higher insoluble degradable fraction and low rate of degradation for DM are common for maize stover (Mgheni *et al*., 1993). The evaluation of the degradability characteristics of two tropical crop residues (maize stover and rice straw) done by Mgheni *et al.* (2001) revealed that these crop residues had low soluble component '**a**' but quite high insoluble but slowly fermentable component **'b'** that were associated with a low rate of degradation **`c'.**

Alkalis affect the cuticle wax to enhance digestibility of parenchyma tissue through swelling of the wall of parenchyma cells and cracking the wall of vascular tubes. Therefore alkali caused both physical and chemical changes in fibrous feed tissues and cell walls facilitating degradation by rumen bacteria and increase in degradability. The observed DM effective degradability values (27) and OM (25.5) for untreated maize stover in the current study were lower than those reported for untreated maize stover by

Onaleye *et al*. (2012). The possible cause could be different varieties of maize stover respond differently to different chemical treatment. Rumen retention time of stover/straw is reported to vary from 36 to 60 hours (Ørskov, 1986), therefore the potential degradability which will be reached after this period has no nutritional importance to the animals, beyond this period nutrients had depleted in the particular basal diet. This may show that the higher degradability of differently treated maize stover at 24 and 48hour was an indication of the improvement that can be realized by chemical and microbes used in the treated materials.

From the curve (Fig 1 and 2) and when considering 48 h degradability values for DM, it can be seen that the differently treated maize stover are highly degradable in the rumen because more than 50% of the potentially degradable DM is degraded within 48 h. Dry matter degradability results showed that (T_7) had a potential degradability value which was lower than the reported potential degradability by Kimbi (1997). The possible cause could be maize stover variety respond differently to different chemical treatment. From the curve (Fig 3 and 4) when considering 48 h degradability values for the OM it can be noted that the differently treated maize stover are readily degradable in the rumen. The value in Table 6 for OM shows that more than half of the potentially degradable crude nutrient of the differently treated maize stover would be degraded in the rumen within 48 h and finally become available for absorption in the rumen wall and post – rumen. The DM and OM degradability values at 48 h of incubation are the most important indicator of quality of roughages as this period is closer to the mean retention time of 48 h for fibrous feeds in the rumen. A high 48 h DM and OM degradation values (Table 5 and 6) indicates that a high percentage of the nutrients would have been degraded and ready for absorption via the rumen wall and post – rumen. So the 48 h degradation values could be considered as signs of *in vivo* digestibility coefficients for the differently treated maize stover. The differently treated maize stover in the current study may be qualified as feedstuff for ruminants on the basis of DM and OM degradability because the feedstuff dry matter and organic matter degradability values are within the range of 40 - 50% recommended by FAO (1986) for feedstuffs for ruminants.

The 48 h organic matter degradability values for untreated (24.5%) and 5% CaO treated corn stover (34.6%) reported by Shreck (2013) were lower than the values observed in the current study. Possible reason could be the variety, since different varieties respond differently to chemical treatment as reported by Shem (1986).

5.3 *In vitro* gas Production

The *in vitro* gas production technique has the potential to reflect the *in vivo* digestibility of feeds for ruminants, the highest predictive value for the *in vivo* digestibility of roughage was obtained after 45 to 52 h of *in vitro* fermentation (Prasad *et al*., 1994). It can be seen that the results obtained in the current study showed that the insoluble fraction '**b**' of the CaO/ $Ca(OH)_2$, enzymes and microbes treated maize stover were higher compared to untreated maize stover. Possible reason is that lignin was reduced through chemical treatment allowing the carbohydrate fraction to become available to the microbial population (Chaji *et al*., 2010). The slightly higher OMD estimated for T_3 and T_2 may be the result of the major carbohydrate being readily fermented by rumen microbes. This implies that the rumen microbes and the host animal have high nutrient uptake.

The amount of gas released when feeds are incubated *in vitro* has been reported to be closely related to digestibility of feed for ruminants (Menke and Steingass, 1988). Gas volume can be considered a good reflection of substrate fermentation to VFA, and estimate of potential digestibility in the rumen. This additionally could be a reflection of a higher proportion of carbohydrate available for fermentation. Similar observation were reported by Liu *et al*. (2002) who found that NaOH treatment improved gas production of rice straw than untreated straw. Also NaOH treated sugarcane pith caused an increase in gas produced parameters (b and c) (Chaji *et al*., 2010). They described that treating sugarcane pith with NaOH had the highest cell wall degradability.

The NaOH treated wheat straw removed some chemical linkages within the hemicelluloses and thus enhanced their solubility in detergent solutions (Chaudhry, 1999). The observed '**b**' and '**c**' values of untreated and CaO/Ca$(OH)_2$ treated maize stover in the current study were lower than those reported by Ouda *et al*. (2006). The possible cause could be that due different crop residue respond differently to chemical treatment. It was reported that an alkali solution solubilizes the inhibitory phenolic compounds and hemicelluloses (Chen *et al.,* 2007). It is proposed that alkali react with lignocelluloses to yield partially delignified products that are highly susceptible to enzymatic and microbial attach. Cell wall content (NDF and ADF) is negatively correlated with gas production at all incubation times. The negative correlation between gas production and the cell wall content might be a result of low microbial activity and long time it takes to degrade the cell wall content. The amount of gas released is closely related to the digestibility of the feed (Osuji *et al*., 1993), and a highly digestible feed will produce more gas than a feed with low digestibility (Ouda *et al*., 2005). However, for those feeds with a similar digestibility, the one with lowest gas production could be regarded to have a higher nutritive value since more of its degraded fraction is likely to be incorporated into the microbial biomass or be absorbed directly by host animal (Rymer, 1999). The gas production is also a better predictor of *in vitro* organic matter digestibility and hence can be used to predict the feeding value of forages (Chenost *et al*., 2001).

The **'b'** values observed in the current study for Calcium Oxide treated maize stover were the same to those of rice straw treated with Sodium hydroxide reported by Liu *et al.* (2002) and slightly lower for maize stover treated with calcium hydroxide and untreated than the same author. Possible reason is that maize stover variety respond differently to $CaO/Ca(OH)_2$ treatment. In the current study fermentation resulted in lower total gas production compared to value reported by DePeters *et al.* (2007) for grain. Also the results observed in current study were lower than values for stover reported by Tolera and Sundsøl (1999). It is well known that feeds rich in non structural carbohydrates produce higher propionate compared to feeds rich in fiber, resulting in lower yields of gas volumes (Beuvink *et al*., 1992). The degradation curves and gas production curves for differently treated maize stover were similar indicating that either method could be used to evaluate improvement due to chemical treatment.

5.4 Growth Performance and Feed Efficiency in study I

5.4.1 Feed intake

The observed mean DMI in the current study was within the range of 2.2 – 6.6 kg DM /day suggested by Kearly (1982) for maintenance and growing steers of body weight between 100 to 250 kg for study I. Similarly, the overall range of Metabolizable Energy intake (43.8 to 48.9MJ /day) in study I was within the range (15.79 – 70MJ/day) reported by Kearly (1982) for maintenance and growing steers of body weight between 100 to 250kg. The research findings reported by Kombe (2000) showed that DMI and MEI of steers with average body weight of 244kg to be 8kg/day and 62.3MJ/day, respectively when the animals were fed low quality hay supplemented with molasses based concentrate. The observed intake of CaO treated maize stover in the current study was lower than those reported by Russell *et al.* (2011) for CaO treated maize stover and Smith *et al*. (1989) for maize stover treated with urea. The possible cause could be lime used by

Russell was better than the lime used in the current study, urea treated maize stover increase CP of the stover which increase microbial activity and increase digestibility thus increase intake of the treated maize stover (Kilongozi, 1992). The observed lack of significant difference in DMI between the steers fed treated and untreated maize stover in the current study (study I) could be due to supplementation effect of concentrates that contained molasses and urea, which improved intake of all feeds.

5.4.2 Average daily gain

Steers fed on CaO, treated maize stover had lower average daily gain than those animals fed an untreated maize stover this may be due to reduced intake of the treated stover. The observed average daily gain for animals fed CaO treated maize stover in the current study was lower than those reported by Russell *et al*. (2011) for CaO treated maize stover and Bui and Le (2001) for urea treated rice straw. The possible cause could be lime used by Russell was better than the one used in the current study which had higher proportion of $CaCO_3$.

5.4.3 Feed conversion ratio (FCR)

The Feed Conversion Ratio (FCR) is a measure of animal`s efficiency in converting feed mass into increased body mass. Cattle and sheep need more than 8kg of feed to put on 1 kg of live weight, their ratio is thus 8:1, FCR for pig 3.5:1 and poultry 2:1. In the current study the values for FCR of differently treated maize stover with CaO, enzymes and microbes were higher than untreated maize stover in study I, indicating that more feed was required per kg gain in the treated stover than untreated. These results are comparable to those reported for lime and/ or urea treated Sesame straw fed to sheep (Aregawi *et al*., 2014). The nutrient mass balance for corn stover treated with 5% CaO and untreated were 6.22 and 7.05 respectively (Johnson *et al*., 2013). The reported FCR for crop residues after chemical treatment and anaerobic treatment indicated that corn stover treated with 5%CaO and untreated were 6.84 and 7.17 kg stover per kg of gain respectively (Shreck *et al*., 2011), indicating that lime grade differs in efficiency. Less feed was required for steers fed treated corn stover than untreated corn stover thus the results differ with the current study, where treated stover had higher FCR. This may imply that lime used in Shreck *et al.* (2011) was more effective in improving the utilization of corn stover than one used in the current study. Steers fed on (T_1), (T_2) and (T_3) treatments in the current study had low ratio (3.9:1, 4.7:1 and 4.9:1 respectively) probably due to the fact that animals were consuming 50% concentrates and 50% stover.

5.5 Digestibility

The observed 14.5% higher dry matter (DM) digestibility of the Calcium hydroxide treated stover than the untreated maize stover was similar to that observed by Russell *et al.* (2011) where as the dry matter digestibility of Calcium oxide treated maize stover was 7.7% units higher than untreated maize stover. These results were however, lower than 27% increase in apparent digestibility obtained by Saadullah *et al*. (1982) for 5% urea treated straw and 16.8% IVDMD of straw treated with CaO reported by Shreck *et al.* (2011). The possible cause could be different crop residue respond differently to chemicals treatment. From the point of view of making efficient use of the low quality and yet the abundantly available feed resources, the small increment in digestibility mean a lot to livestock owners. With this regard Ørskov *et al*. (1990) stated that in areas where straw /stover is the main feed for ruminants a proportional increase of 0.1 in digestibility can have enormous implication for resource availability and thus animal performance.

5.6 Growth performance and Feed Conversional Ratio in Study II

5.6.1 Feed intake

The observed mean DMI in the current study was within the range of 2.2 – 6.6 kg DM /day suggested by Kearly (1982) for maintenance and growing steers of body weight between 100 to 250 kg for study II. Similarly, the overall range of Metabolizable Energy intake (35.6 to 36.8 MJ /day) in study II was within the range (15.79 – 70MJ/day) reported by Kearly (1982) for maintenance and growing steers of body weight between 100 to 250kg. The observed lack of significant difference in DMI between the steers fed treated and untreated maize stover in the current study (study II) could be due to supplementation effect of concentrates that contained molasses and urea, which improved intake of all feeds. The intake of $Ca(OH)_2$ treated maize stover observed in the current study was lower than those reported by Djajanegara *et al*. (1985) for wheat straw treated with $Ca(OH)_2$ but were similar with intake values reported by Sarnklong *et al*. (2010) for rice straws an indication that crop residues respond differently to $Ca(OH)_2$ treatment. (Shem, 1986).

5.6.2 Average daily gain

The observed average daily gain of steers fed $Ca(OH)_2$ treated maize stover (1.19 kg/day) in the current study were lower than those reported by Peterson (2014) for cattle fed maize stover treated with $Ca(OH)_2$ (1.50 kg/day). The possible cause could be $Ca(OH)_2$ used to treat maize stover by Peterson was better than the one used in the current study. The daily gain of steers observed in the current study where steers fed untreated stover had higher gain differ with those reported by Sarnklong *et al.* (2010). The possible cause could be crop residue respond differently to chemical treatment. Also weight gain of steers fed on $Ca(OH)_2$ treated rice straw was observed to be higher than for those fed untreated (Doyle *et al*., 1986). This may be due to increased intake of the treated straws.

Average daily gain for calves fed on $Ca(OH)_2$ treated rice straw reported by Saadullah *et al*. (1981) was higher than results from current study. This may suggest that rice straw respond better to $Ca(OH)_2$ treatment than maize stover.

5.6.3 Feed conversion ratio (FCR)

In the current study the FCR values for study II for maize stover treated with $Ca(OH)_2$ were lower than untreated maize stover. The results are in line with the previously reported values for maize stover and rice straw treated with urea, where FCR values for both treated crop residues were lower than for the untreated maize stover and rice straw. The reported FCR for corn stover treated with $Ca(OH)_2$ (8.33) and untreated (8.81) reported by Peterson (2014) were in agreement with the results obtained in the current study.

CHAPTER SIX

6.0 CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

It is concluded from the study that lime treatment was effective in solubilizing the cell wall constituents and improved the digestibility of calcium hydroxide treated maize stover, but had negative effect on the feed intake and animal growth.

6.2 Recommendations

(i) Repetition of similar studies using different varieties of maize stover and better quality of CaO should be attempted to see whether they will respond differently to the ones used in the current study.

(ii) Extension of the period of feeding to see whether animals will get used to the treated material.

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APPENDICES

Appendix 1: Dry matter degradation of untreated and treated maize stover with CaO, enzymes and microbes

Treatment	Degradation at hour after incubation							
	0	6	12	24	48	72	96	120
(T_1)	8.78	8.88	11.2	30.9	46.2	58.2	59.8	59.8
(T_2)	17	19.7	21	25.8	43	50.1	53.6	61.5
(T_3)	6.67	14	14.6	27.6	40.9	51.3	54.2	62.1
(T_4)	8.68	11.3	11.8	28.3	45.3	55.7	58.7	61.6
(T_5)	8.87	9.29	10.4	24.8	42.3	49.5	51.8	49.9

T_1 = Untreated maize stover 2012, T_2 = Treated maize stover with Calcium Oxide (CaO), T_3 = Treated maize stover with CaO and enzyme, T_4 = Treated maize stover with CaO and microbial, T_5 = Treated maize stover with CaO, enzyme and microbial.

Appendix 2: Dry matter degradation of untreated and $Ca(OH)_2$ treated maize stover

Treatment	Degradation at hour after incubation							
	0	6	12	24	48	72	96	120
T_6	8.87	9.29	16.4	34.8	43.3	50.9	54.3.	52.9
T_7	14.8	15.1	25.6	32.9	51.2	58.2	64.3	67.3

T_6 = Untreated maize stover 2013, T_7 = Treated maize stover with Calcium hydroxide.

Appendix 3: *In sacco* degradability ANOVA Tables

Dependent Variable: a

Source	DF	Sum of squares	Mean square	F value	Pr > F
Diet type	6	352.7278286	58.7879714	Infinity	<.0001
Error	14	0.0000000	0.0000000		
Corrected Total	20	352.7278286			

R-Square	Coeff Var	Root MSE	a Mean
1.000000	0	0	.052857

Dependent Variable: b

Source	DF	Sum of square	Mean square	F value	Pr > F
Diet type	6	1331.682857	221.947143	1.2E16	<.0001
Error	14	0.000000	0.000000		
Corrected Total	20	1331.682857			

R-Square	Coeff Var	Root MSE	b Mean
1.000000	2.17736E-7	1.36241E-7	62.57143

Dependent Variable: c

Source	DF	Sum Squares	Mean square	F value	Pr > F
Diet type	6	0.00088457	0.00014743	4.76E15	<.0001
Error	14	0.00000000	0.00000000		
Corrected Total	20	0.00088457			

R-Square	Coeff Var	Root MSE	c Mean
1.000000	1.33916E-6	1.76E-10	0.013143

Dependent Variable: PD

Source	DF	Sum of squares	Mean square	F value	Pr >F
Diet type	6	2223.582857	370.597143	Infinity	<.0001
Error	14	0.000000	0.000000		
Corrected Total	20	2223.582857			

R-Square	Coeff Var	Root MSE	PD Mean
1.000000	0	0	70.62857

Dependent Variable: ED

Source	DF	Sum of squares	Mean square	F value	Pr > F
Diet type	6	124.5000000	20.7500000	Infinity	<.0001
Error	14	0.0000000	0.0000000		
Corrected Total	20	124.5000000			

R-Square	Coeff Var	Root MSE	ED Mean
1.000000	0	0	28.00000

Appendix 4: Organic matter degradation of untreated and treated maize stover with CaO, enzyme and microbes

	Degradation at hour after incubation							
Treatment	0	6	12	24	48	72	96	120
(T_1)	5.93	7.64	11.4	30.6	46.8	59.2	63.3	59.3
(T_2)	11.2	15.9	17.9	22.3	45.2	48.6	49.6	61.6
(T_3)	4.88	13.4	12.7	28	41.8	52.1	64.3	68.4
(T_4)	2.71	9.46	14.9	24.7	40.3	48.7	50.7	51.4
(T_5)	3.41	11.9	11.7	27.7	46.7	57	61.6	64.8

T_1 = Untreated maize stover 2012,T_2 = Treated maize stover with Calcium Oxide (CaO), T_3 = Treated maize stover with CaO and enzyme, T_4 = Treated maize stover with CaO and microbial, T_5 = Treated maize stover with CaO, enzyme and microbial.

Appendix 5: Organic matter degradation of untreated and $Ca(OH)_2$ treated maize stover

	Degradation at hour after incubation							
Treatment	0	6	12	24	48	72	96	120
(T_6)	0.67	3.72	8.65	33.1	47.0	51.1	54.4	54.3
(T_7)	5.15	8.62	9.62	26.7	43.2	53.1	58.3	58.7

UMS2013 = Untreated maize stover 2013, TMS2013= Treated maize stover with Calcium hydroxide.

Appendix 6: Organic matter degradability ANOVA Tables

Dependent Variable: a

Source	DF	Sum squares	Mean square	F value	Pr > F
Diet type	6	243.3401143	40.5566857	Infinity	<.0001
Error	14	0.0000000	0.0000000		
Corrected Total	20	243.3401143			

R-Square	Coeff Var	Root MSE	a Mean
1.000000	0	0	3.464286

Dependent Variable: b

Source	DF	Sum of Squares	Mean square	F value	Pr > F
Diet type	6	2084.580000	347.430000	Infinity	<.0001
Error	14	0.000000	0.000000		
Corrected Total	20	2084.580000			

R-Square	Coeff Var	Root MSE	b Mean
1.000000	0	0	69.90000

Dependent Variable: c

Source	DF	Sum of squares	Mean square	F value	Pr > F
Diet type	6	0.00103114	0.00017186	2.22E16	<.0001
Error	14	0.00000000	0.00000000		
Corrected Total	20	0.00103114			

R-Square	Coeff Var	Root MSE	c Mean
1.000000	6.09913E-7	8.8002E-11	0.014429

Dependent Variable: PD

Source	DF	Sum of Squares	Mean square	F value	Pr > F
Diet type	6	3018.342857	503.057143	Infinity	<.0001
Error	14	0.000000	0.000000		
Corrected Total	20	3018.342857			

R-Square	Coeff Var	Root MSE	PD Mean
1.000000	0	0	74.37143

Dependent Variable: ED

Source	DF	Sum of squares	Mean square	F value	Pr > F
Diet type	6	253.7657143	42.2942857	Infinity	<.0001
Error	14	0.0000000	0.0000000		
Corrected Total	20	253.7657143			

R-Square	Coeff Var	Root MSE	ED Mean
1.000000	0	0	26.58571

Appendix 7: *In vitro* gas production of CaO, enzymes and microbes treated and untreated maize stover

	In vitro gas production at different hour after incubation (ml/g DM)								
Treatments	0	3	6	12	24	48	72	96	120
(T_1)	0	2	3.5	27	35	43.3	53.8	59	60.1
(T_2)	0	2.7	4.2	19.5	31.5	42.1	51.4	55	56
(T_3)	0	1.7	3.1	18	31.3	41	48	53.7	54
(T_4)	0	1	2.8	13.6	22	38.6	45	52.9	53.6
(T_5)	0	1.3	3	16.1	27.3	32	42.5	50.3	50.9

T_1 = Untreated maize stover 2012,T_2 = Treated maize stover with Calcium Oxide (CaO), T_3 = Treated maize stover with CaO and enzyme, T_4 = Treated maize stover with CaO and microbial, T_5 = Treated maize stover with CaO, enzyme and microbial.

Appendix 8: *In vitro* gas production of untreated and Ca(OH)2 treated maize stover

	In vitro gas production at different hour after incubation (ml/g DM)								
Treatment	0	3	6	12	24	48	72	96	120
(T_6)	0	0.3	4.1	15.9	29	36.5	47.3	50.9	52.4
(T_7)	0	1.3	5.6	21	30	41.6	49.1	53.8	54

T_6 = Untreated maize stover 2013, T_7 = Treated maize stover with Calcium hydroxide.

Appendix 9: ANOVA table for in vitro gas production

Dependent Variable: b

SOURCE	DF	SUM OF SQUARES	MEAN SQUARE	F value	Pr > F
Diet type	6	1134.848571	189.141429	Infinity	< .0001
Error	56	0.000000	0.000000		
Corrected Total	62	1134.848571			

R-Square	Coeff Var	Root MSE	b Mean
1.000000	0	0	40.32857

Dependent Variable: EGP:

SOURCE	DF	SUM SQUARES	MEAN SQUARE	F value	Pr > F
Diet types	6	104.4843429	17.4140571	1.7216	<.0001
Error	56	0.0000000	0.0000000		
Corrected Total	62	104.4843429			

R-Square	Coeff Var	Root MSE	EGP Mean
1.000000	2.74791E-7	3.186E-8	11.59429

Appendix 10: ANOVA table for animal performance in experiment 1

Dependent Variable: W1

source	DF	Sum of squires	Mean squire	F value	Pr > F
Treat	4	384.177974	96.044493	0.61	0.6585
IWT	1	6310.807153	6310.807153	39.92	<.0001
Error	74	11698.50535	158.08791		
Corrected Total	79	18950.18750			

R-Square	Coeff Var	Root MSE	W1 Mean
0.382671	6.312294	12.57330	199.1875

Dependent Variable: W2

Source	DF	Sum of squires	Mean squire	F value	Pr >F
Treat	4	172.191432	43.047858	0.29	0.8818
IWT	1	4927.717798	4927.717798	33.51	<.0001
Error	74	10882.53220	147.06125		
Corrected Total	79	16791.20000			

R-Square	Coeff Var	Root MSE	W2 Mean
0.351891	5.941637	12.12688	204.1000

Dependent Variable: W3

Source	DF	Sum of squires	Mean Squire	F value	Pr >F
Treat	4	309.840765	77.460191	0.54	0.7034
IWT	1	3173.554977	3173.554977	22.31	<.0001
Error	74	10524.32002	142.22054		
Corrected Total	79	14697.55000			

R-Square	Coeff Var	Root MSE	W3 Mean
0.283941	5.745213	11.92563	207.5750

Dependent Variable: W4

Source	DF	Sum of squires	Mean Squire	F value	Pr > F
Treat	4	305.598001	76.399500	0.47	0.7544
IWT	1	3061.703985	3061.703985	19.01	<.0001
Error	74	11917.17102	161.04285		
Corrected Total	79	15719.55000			

R-Square	Coeff Var	Root MSE	W4 Mean
0.241889	6.009360	12.69027	211.1750

Dependent Variable: W5

Source	DF	Sum of Squires	Mean Squire	F value	Pr > F
Treat	4	540.772825	135.193206	0.92	0.4589
IWT	1	3471.972762	3471.972762	23.54	<.0001
Error	74	10914.40102	147.49191		
Corrected Total	79	15563.57624			

R-Square	Coeff Var	Root MSE	W5 Mean
0.298722	5.675400	12.14462	213.9871

Dependent Variable: FWG (35 days)

Source	DF	Sum of Squire	Mean Squire	F value	Pr > F
Treat	4	541.11321	135.27830	0.92	0.4586
IWT	1	17494.96301	17494.96301	118.61	< .0001
Error	74	10914.99110	147.49988		
Corrected Total	79	30299.82505			

R-Square	Coeff Var	Root MSE	FWG Mean
0.639767	44.35115	12.14495	27.38363

Dependent Variable: ADG

Source	DF	Sum of squires	Mean squire	F value	Pr > F
Treat	4	0.44374068	0.11093517	0.92	0.4572
IWT	1	14.25409674	14.25409674	118.17	<.0001
Error	74	8.92611576	0.12062319		
Corrected Total	79	24.72619500			

R-Square	Coeff Var	Root MSE	ADG Mean
0.639002	44.39866	0.347308	0.782250

Dependent Variable: TDMI

Source	DF	Sum of Squires	Mean Squire	F value	Pr >F
Treat	4	1360.228453	340.057113	1.58	0.1882
IWT	1	494.637953	494.637953	2.30	0.1336
Error	74	15913.30117	215.04461		
Corrected Total	79	17799.57487			

R-Square	Coeff Var	Root MSE	TDMI Mean
0.105973	8.857955	14.66440	165.5506

Dependent Variable: ADFI

Source	DF	Sum of squires	Mean Square	F value	Pr > F
Treat	4	1.10545009	0.27636252	1.57	0.1902
IWT	1	0.41242799	0.41242799	2.35	0.1297
Error	74	12.99504701	0.17560874		
Corrected Total	79	14.54219500			

R-Square	Coeff Var	Root MSE	ADFI Mean
0.106390	8.860024	0.419057	4.729750

Dependent Variable: FCR

Source	DF	Sum of squares	Mean Square	F value	Pr > F
Treat	4	150.5949813	37.6487453	1.35	0.2604
IWT	1	334.5237364	334.5237364	11.97	0.0009
Error	74	2067.317551	27.936724		
Corrected Total	79	2508.418420			

R-Square	Coeff Var	Root MSE	FCR Mean
0.175848	62.34027	5.285520	8.478500

Dependent Variable: FE

Source	DF	Sum of Squire	Mean Squire	F value	Pr > F
Treat	4	217.193301	54.298325	1.09	0.3669
IWT	1	6962.963842	6962.963842	140.03	<.0001
Error	74	3679.59305	49.72423		
Corrected Total	79	11354.52280			

R-Square	Coeff Var	Root MSE	FE Mean
0.675936	42.37705	7.051541	16.64000

Appendix 11: T-test procedure for Digestibility results using Acid Insoluble Ash markers Dependent Variable: Digestibility

Diets	N	Mean	Std Dev	Std Err	Minimum	Maximum
(T_7)	32	70.1654	4.9237	0.8704	63.3658	79.3772
(T_6)	32	55.6682	3.1441	0.5558	48.9188	59.9910
Diff (1-2)		14.4972	4.1309	1.0327		

Diets	Method	Mean	95% CL Mean		Std Dev	95% CL Std Dev	
(T_7)		70.1654	68.3902	71.9406	4.9237	3.9474	6.5460
(T_6)		55.6682	54.5346	56.8018	3.1441	2.5207	4.1801
Diff (1-2)	Pooled	14.4972	12.4328	16.5615	4.1309	3.5145	5.0115
Diff (1-2)	Satterthwaite	14.4972	12.4255	16.5688			

Method	Variances	DF	t Value	Pr > |t|
Pooled	Equal	62	14.04	<.0001
Satterthwaite	Unequal	52.677	14.04	<.0001

Equality of Variances

Method	Num DF	Den DF	F Value	Pr > F
Folded F	31	31	2.45	0.0148

Appendix 12: T-test tables for animal performance in experiment II

The TTEST Procedure Statistics

Variable	Treat	N	Lower CL Mean	Mean	Upper CL Mean	Lower CL Std Dev	Std Dev	Upper CL Std Dev	Std Err
In. Wt.	T_6	16	189.71	196	202.29	8.7198	11.804	18.269	2.951
In. Wt	T_7	16	187.96	194.23	200.5	8.688	11.761	18.203	2.9403
W_1	T_6	16	226.98	233.19	239.4	8.6114	11.657	18.042	2.9144
W_1	T_7	16	225.87	234	242.13	11.274	15.262	23.621	3.8155
W_2	T_6	16	233.64	239	244.36	7.4361	10.066	15.58	2.5166
W_2	T_7	16	227.6	236	244.4	11.649	15.769	24.406	3.9423
W_3	T_6	16	234.95	240	245.05	6.9976	9.4728	14.661	2.3682
W_3	T_7	16	229.4	237	244.6	10.537	14.264	22.077	3.566
W_4	T_6	16	236.84	242	247.16	7.1518	9.6816	14.984	2.4204
W_4	T_7	16	231.81	240	248.19	11.355	15.371	23.79	3.8427
W_5	T_6	16	238.69	244	249.31	7.3574	9.9599	15.415	2.49
W_5	T_7	16	233.47	242	250.53	11.825	16.008	24.776	4.0021
W_6	T_6	16	240.85	247	253.15	8.5189	11.532	17.848	2.8831
W_6	T_7	16	235.66	244.03	252.4	11.599	15.702	24.302	3.9256
Wt gain	T_6	16	50	51	52	6.34	7.11	8.40	3.4321
Wt gain	T_7	16	49	50	51	5.98	6.87	7.65	4.3212
ADG	T_6	16	1.0042	1.2143	1.4244	0.2913	0.3943	0.6103	0.0986
ADG	T_7	16	1.0043	1.1858	1.3672	0.2516	0.3405	0.5271	0.0851
TConc.DMI	T_6	16	60.747	60.747	60.747	0	0	0	0
TConc.DMI	T_7	16	60.747	60.747	60.747	0	0	0	0
TSt.DMI	T_6	16	110.13	123.26	136.39	18.197	24.634	38.126	6.1586
TSt.DMI	T_7	16	107.99	111.89	115.79	5.4044	7.316	11.323	1.829
TDMI	T_6	16	170.88	184.01	197.13	18.197	24.634	38.126	6.1586
TDMI	T_7	16	168.74	172.64	176.54	5.4044	7.316	11.323	1.829
ADMI	T_6	16	4.0686	4.3811	4.6936	0.4333	0.5865	0.9078	0.1466
ADMI	T_7	16	4.0176	4.1105	4.2033	0.1287	0.1742	0.2696	0.0435
FCR	T_6	16	3.1503	4.1198	5.0893	1.3441	1.8195	2.816	0.4549
FCR	T_7	16	3.0848	3.8018	4.5188	0.994	1.3456	2.0826	0.3364
FE	T_6	16	23.053	27.832	32.61	6.6244	8.9676	13.879	2.2419
FE	T_7	16	24.448	28.934	33.42	6.2187	8.4183	13.029	2.1046

The TTEST Procedure

T-Tests

Variable	Method	Variances	DF	t Value	Pr > |t|
Inwt	Pooled	Equal	30	-0.43	0.6736
Inwt	Satterthwaite	Unequal	30	-0.43	0.6736
W1	Pooled	Equal	30	0.17	0.8668
W1	Satterthwaite	Unequal	28.1	0.17	0.8668
W2	Pooled	Equal	30	-0.64	0.5261
W2	Satterthwaite	Unequal	25.5	-0.64	0.5270
W3	Pooled	Equal	30	-0.70	0.4888
W3	Satterthwaite	Unequal	26.1	-0.70	0.4896
W4	Pooled	Equal	30	-0.44	0.6628
W4	Satterthwaite	Unequal	25.3	-0.44	0.6634
W5	Pooled	Equal	30	-0.42	0.6744
W5	Satterthwaite	Unequal	25.1	-0.42	0.6749
W6	Pooled	Equal	30	-0.61	0.5468
W6	Satterthwaite	Unequal	27.5	-0.61	0.5472
TSTDMI	Pooled	Equal	30	-1.77	0.0870
TSTDMI	Satterthwaite	Unequal	17.6	-1.77	0.0941
TCONCDMI	Pooled	Equal	30	-9.51	<.0001
TCONCDMI	Satterthwaite	Unequal	30	-9.51	<.0001
TDMI	Pooled	Equal	30	-1.77	0.0870
TDMI	Satterthwaite	Unequal	17.6	-1.77	0.0941
ADMI	Pooled	Equal	30	-1.77	0.0870
ADMI	Satterthwaite	Unequal	17.6	-1.77	0.0941
ADG	Pooled	Equal	30	-0.22	0.8283
ADG	Satterthwaite	Unequal	29.4	-0.22	0.82
ADG	Satterthwaite	Unequal	29.4	-0.22	0.8283
FCR	Pooled	Equal	30	-0.56	0.5782
FCR	Satterthwaite	Unequal	27.6	-0.56	0.5786
FE	Pooled	Equal	30	0.36	0.7226
FE	Satterthwaite	Unequal	29.9	0.36	0.7226

Equality of Variances

Variable	Method	Num DF	Den DF	F Value	Pr > F
Inwt	Folded F	15	15	1.01	0.9889
W1	Folded F	15	15	1.71	0.3076
W2	Folded F	15	15	2.45	0.0924
W3	Folded F	15	15	2.27	0.1239
W4	Folded F	15	15	2.52	0.0834
W5	Folded F	15	15	2.58	0.0758
WW6 15	Folded F	15	15	1.85	0.2433
TSTDMI	Folded F	15	15	11.34	<.0001
TCONCDMI	Folded F	15	15	1.00	1.0000
TDMI	Folded F	15	15	11.34	<.0001
ADMI	Folded F	15	15	11.34	<.0001
ADG	Folded F	15	15	1.34	0.5774
FCR	Folded F	15	15	1.83	0.2540
FE	Folded F	15	15	1.13	0.8098