GROWTH PERFORMANCE OF WEST AFRICAN DWARF SHEEP FED DIETS SUPPLEMENTED WITH FOSSIL SHELL FLOUR

BY

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ABSTRACT

Inorganic feed additives are commonly used in small ruminant production to promote growth. However, the problem of bioaccumulation of chemical residues in animals fed inorganic feed additives has necessitated the search for organic alternatives such as Fossil Shell Flour (FSF). There is dearth of information on the use of fossil shell flour as a feed additive in the diet of small ruminants. Hence, growth performance of West African Dwarf (WAD) sheep fed diets supplemented with FSF were investigated.

Sixteen WAD rams (18.5±1.05 kg) were allotted to four treatments: Tl (0% FSF), T2 (2% FSF), T3 (4% FSF) and T4 (6% FSF) in a twelve-week growth study. Daily Weight Gain (DWG, kg), Feed Intake (FI, kg/day) and Feed Conversion Ratio (FCR) were measured. Nutrient digestibility was determined using standard procedures. In vitro gas production characteristics, Organic Matter Digestibility (OMD), metabolisable energy (MJ/kg) and Short Chain Fatty Acid (SCFA, µmol/mL) of the feed were determined at three hours interval for ninety-six hours. Synchronized sixteen primiparous WAD ewes (25.75±1.60 kg) mated with proven rams were randomly allotted to the experimental diets. Dams and their lambs were weighed weekly for 13 weeks. Gestation Length (GL, days), Dry Matter Intake (DMI, g/day), Total Weight Change During Pregnancy (TWCDP, kg), Weight Change During Lactation (WCDL, kg) as well as Lamb Weaning Weight (LWW, kg), Pre-Weaning Mortality (PWM, %) and Twin Survival Rate (TSR, %) were monitored. Data were analysed using descriptive statistics and ANOVA at α0.05.

The DWG for rams on T3 (0.20¬±0.04) was significantly higher than T4 (0.11±0.01) but similar to T1 (0.15±0.01) and T2 (0.19±0.03), while FCR was higher for those on T4 (9.90±1.03) than those on T3 (4.50 ±0.69). The FI ranged from 0.90±0.16 (T3) to 1.09±0.11 (T4). Crude protein digestibility was higher in sheep on T3 (82.8±1.6%) than T2 (77.8±1.2%) but similar to T1 (81.7±1.4%) and T4 (81.9±1.5%), while ash digestibility ranged from 63.6±1.2% (T1) to 72.5±1.6% (T2). The OMD ranged from 65±1.6% (T4) to 53.4±1.8% (T1) at 72 hours and from 62.2±1.4% (T2) to 54.5±1.1% (T1) at 96 hours however, no significant difference was observed among the dietary treatments for metabolisable energy and SCFA. The GL ranged from 148.50±0.13 (T1) to 152.72±0.21 (T4), while the DMI for the ewes on T2 (893.29±0.14) was significantly

higher than those on T3 (860.74±0.17) however, T3 (860.74±0.17) and T4 (866.63±0.12) were not significantly different. The TWCDP ranged from 10.25±0.41 (T3) to 15.68±0.56 (T1). The WCDL significantly decreased for sheep on T4 (-2.75±0.16) than those on T1 (-1.09±0.13), T2 (-0.75±0.12) and T3 (-1.50±0.15) however, T1 (-1.09±0.13) and T2 (-0.75±0.12) were not significantly different. The LWW ranged from 8.10±0.12 (T4) to 9.23±0.18 (T3), PWM from 20±0.1 (T4) to 33.3±0.2 (T1) and TSR from 50±0.18 (T4) to 100±1.05 (T2 and T3).

Inclusion of 2.0% fossil shell flour in the diet of West African dwarf sheep improved dry matter intake and reduced weight loss during lactation, while the inclusion at 4.0% enhanced the daily weight gain.

Keywords: Fossil shell flour, Sheep feed intake, Sheep growth rate, Sheep gestation length
Word count: 487

CERTIFICATION

I certify that this work was carried out by **Chibuisi Henry EMERUWA** in the Department of Animal Science, University of Ibadan, under my supervision.

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DEDICATION

This work is dedicated to:

God Almighty and to my wonderful parents Mr Sunday Emeruwa and late Mrs Glory Emeruwa.

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I am grateful to God almighty for His faithfulness, provision, protection and for seeing me through all my endeavours.

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health and long life to live and eat the fruits of all your labour and to see all your children's children in Jesus name - Amen.

To my Jewel of an inestimable worth-Mrs Amarachi Faith Emeruwa, my friend, encourager, motivator and the mother of my three wonderful and lovely children- Joshua Emeruwa, Joan Emeruwa and Joel Emeruwa, I say thank you for all your love, support, care, prayers and for being there always for me and our children. Words may not be enough to express my love and gratitude; you are simply the BEST. Thank you, may God bless you, keep you, prosper you and decorate your life with all goodness, mercy and favour beyond all you can think or imagine.

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CHAPTER ONE

1.0 General introduction

The increasing human population has placed a great demand on food production and forced most countries to focus attention on livestock improvement programmes. These livestock improvement programmes are to put the livestock industry on a strong footing with the aims of reducing cost of inputs, competition between man and animals for food and having large turnover or output from the livestock industry in terms of meat, egg and milk production. In developing countries, the contribution of livestock to food supplies is increasing at a higher rate than that of cereals. Developing countries now account for almost half of the total world meat production (FAO, 2004). The demand for livestock products will continue to rise in developing countries as a result of rapidly expanding population and the trend towards higher income in most of these countries. Livestock not only play a significant role in the socio-cultural aspects of the people but also, help to balance human nutrition (Adam *et al.,* 2010). They occupy about 30% of the planet's ice-free terrestrial surface area and this sector is increasingly organized in long market chains employing approximately 1.3 billion people globally and directly supporting the livelihoods of 600 million small holding farmers in the developing countries (Thornton, 2010).

Livestock products provide essential amino acids, minerals and vitamins in a concentrated form and their fat can also supply much needed calories. Foods of animal origin, mainly meat and milk are judged to be of high quality and essential in the nutrition and development of growing children. In the tropics, cattle, sheep and goats are the primary suppliers of meat and milk. Meat is important in the diet of young children and pregnant women because of its high protein and iron contents. Sheep and goats are raised by marginalized and landless small holders not only for meat (market) but also for other important non-market co-products (Dossa *et al.,* 2008). Small ruminants also play an insurance role to overcome unforeseen necessity of rural households, including settling of medical bills and school fees (Oluwatayo and Oluwatayo, 2012). Ruminants have greater digestive capacity to convert cellulose and other fibrous materials into useful products than their monogastric counter parts. They are widely distributed throughout the world.

Livestock production is faced with a lot of constraints. One of these constraints is the high cost of feed, which leads to high cost of production. Factors that have the most significant and frequently limiting effects on livestock production include Climate, Diseases and Parasites, and Nutrition (Sejian *et al.*, 2012).

Diseases and parasites are among the most severe factors that impact livestock production and productivity. Animal diseases have great impact on food supplies, trade and commerce, and human health globally. The last few decades have seen a general reduction in the burden of livestock diseases. Such reduction is the direct result of the availability and effectiveness of drugs and vaccines, as well as improvements in diagnostic technologies (Pearson, 2006; Thornton, 2010). Ruminant diseases caused by gastrointestinal nematode parasite infections are the diseases with the greatest impact on animal health and productivity (Sejian *et al.*, 2012).

Diatomaceous Earth (DE) or fossil shell flour (FSF) is fossilized deposits of microscopic shells that were created by simple one-celled plant called diatoms. It is so pure that the Food and Drug Administration has given it a "food-grade" designation. Fossil shell flour is safe for human consumption and can be used as an organic feed additive for improving the performance and production of livestock (Nuti *et al.*, 2002; Jean, 2004). The health improvements observed in animals fed diets supplemented with fossil shell flour appear to be as a result of three primary actions of fossil shell flour: Eliminating parasites, reducing physiological stress and increasing assimilation of nutrients from food; it is a rich source of minerals not abundantly available in today's feed crops and it may bind to toxic metal build-up and help rid it from the body (Adebiyi *et al.*, 2009). It eliminates parasites by puncturing the insect's exoskeleton and dehydrating the parasite. As a feed additive for livestock, fossil shell flour increases herd appetite and production. It absorbs destructive and poisonous sediments in animals. It is an effective digestive aid and contains 14 trace minerals which are important in improving animals' performance (Robert, 2002; Jean, 2004).

1.1 Justification

- The use of inorganic feed additives in small ruminant production to promote growth has become undesirable because of the residuals in meat products and development of antibiotic-resistant bacterial populations in humans.
- The use of organic feed additives such as fossil shell flour is of global interest as compared to inorganic feed additives.
- Little information is available on the use of fossil shell flour as a feed additive in the diets of West African Dwarf sheep to improve performance.

1.2 General Objective

To evaluate the effect of fossil shell flour as a feed additive on growth performance of West African Dwarf sheep.

1.2.1 Specific objectives

The specific objectives of the research are to:

- To determine the effect of fossil shell flour on the growth rate of West African Dwarf rams and to evaluate its effect on the dry matter intake, nutrient digestibility and nitrogen utilization of West African Dwarf sheep.
- To determine the effect of fossil shell flour on the blood parameters of West African Dwarf Sheep.
- To evaluate the *in-vitro* gas production of diets supplemented with fossil shell flour at varying levels.
- To determine the effect of fossil shell flour on the reproductive performance of West African Dwarf ewes.

CHAPTER TWO

LITERATURE REVIEW

2.0 Sheep

Livestock systems occupy about 30% of the planet's ice-free terrestrial surface area and this sector is increasingly organized in long market chains employing approximately 1.3 billion people globally and directly supporting the livelihoods of 600 million smallholding farmers in the developing countries (Thornton, 2010). Livestock production is therefore a key component of world agriculture. In fact, throughout the world, human populations largely depend on domestic animals for a multitude of purposes, essentially the production of meat, fat, milk, and other dairy products, eggs and fibers like wool or cashmere as well as other purposes such as transport, draft, and provision of fertilizers, especially in developing countries (Sejian *et al.*, 2012). Livestock production in the tropics and subtropics is mostly influenced by the seasonal scarcity and low quality of feed resources; livestock makes a substantial contribution to the well-being of the people. Small ruminants, such as sheep, are important in feeding the rapidly expanding population of the developing world under typical harsh environmental conditions, due to their low feed and area requirement, short generation interval, faster growth rate and higher environmental adaptability compared to the large ruminants (Tibbo *et al.*, 2006).

All domesticated sheep are *Ovis aries*. They are quadrupedal, ruminant mammals typically kept as livestock. Sheep are members of the Artiodactyla, the even-toed ungulates. The name sheep applied to many species in the genus *Ovis*, in everyday usage it almost refers to *Ovis aries* (Melinda, 2004). Numbering a little over one billion globally, domestic sheep are the most numerous species of sheep. An adult female sheep is referred to as *ewe*, an intact male as a *ram* or occasionally a *tup*, a castrated male as a *wether* and a young sheep as a *lamb* (Schoenian, 2007). Sheep contributes to food production, rural employment and gross national product by converting roughages into meat, wool and skin. It is generally known that raising young animals on high concentrate diets result in higher daily gains, dressing percentage and carcass quality than on a forage system (Johnson *et al*., 2005). Genetic differences among breeds of sheep offer important opportunities for improving efficiency and quality of market ram production through management systems which exploit breed differences. In order to improve sheep production,

evaluation of all available sheep genetic resources is essential to understand their potential roles in fattening.

2.1.0. Indigenous breeds of sheep

In many tropical countries, the keeping of indigenous breeds of sheep is part of the traditional culture. Farm families depend on the sheep for meat, wool, manure and some income. Indigenous sheep have, however, consistently been neglected by researchers, extensionists and government officials, and this has contributed to a very low productivity (Bernardo, 2013). Developing countries including those in Africa have attempted to introduce improved breeds of both sheep and goats to bring about genetic improvement without even adequately investigating the merits of local breeds. This has resulted not only in the reduction of the population of the indigenous breeds but also in endangering the existence of the local genetic material (Ponzoni, 1992). Indigenous animal breeds are extremely valuable for millions of small-scale farmers worldwide because they are very sturdy and resilient. They are adapted to climatic extremes because they possess a good thermoregulation capacity. Indigenous sheep have a good reproductive capacity, partly because females come into heat independently of the season. The females also have a well-developed maternal instinct and are easily milked and the animals' lifespan is long. These excellent physical characteristics form the basis for their economic significance for the farmers (Bernardo, 2013). They make good use of harvest residues such as stubble, straw and dead leaves.

One of the major factors limiting the productivity of small ruminants in developing countries is over dependence on low digestibility feeds which during the dry season cannot meet even the maintenance requirements of these animals (Schoenian, 2011). Of all the factors influencing livestock production, climate, and location are undoubtedly the most significant. In fact, climatology characteristics such as ambient temperature and rainfall patterns have great influence on pasture and food resources availability cycle throughout the year, and types of disease and parasite outbreaks among animal populations (Sejian *et al.*, 2012). In recent years there has been a growing interest in many tropical countries to identify potentially important feeds sources among shrubs and trees for inclusion in the ruminant diet to provide browse (shoots, fruits and pods) that is high in protein to supplement the available low protein forage. This has been recognized as one of the most effective means of improving animal performance

in small holder livestock production (Gworgwor *et al.*, 2006). The importance of environmental components such as improved management practices and nutrition in enhancing higher productivity of indigenous breeds should not be overlooked.

2.1.1 African Sheep

The African sheep can be described as thin-tailed, fat-tailed or fat ramped (Wilson, 1991). The thin tailed sheep are the commonest in the northern dry topics where they are usually of large size or in the western humid areas where they are smaller and often referred to as dwarf or forest sheep. Fat-tailed types predominates Eastern Africa while fat-ramped types are the commonest in North-east African but they have spread to Zimbabwe and other countries of the southern region. However, in Nigeria there are four breeds: The West African Dwarf, Yankasa, Uda and Balami.

2.1.2. The West African Dwarf Sheep

The West African dwarf is the predominant breed of the humid tropics from Southern West Africa through central Africa. They are generally small and short-legged and measure between 40 to 60cm in length. Adult male weighs 30 – 37kg while the matured female weighs between 20 – 25kg (Wilson, 1991). The females are usually polled, when horns appear in female they are usually fine and short. They can be bred at the age of 7 – 8 months and are trypanotolerant. They present a great potential to mitigate the problem of protein malnutrition in the country as well as supply other products (Bitto and Egbunike, 2006).

2.1.3 Yankasa

Yankasa is a meat breed found in North and North central Nigeria. The Yankasa is a medium sized breed of sheep. The tail is long and thin, the ears moderately long and somewhat droopy. Rams have curved horns and a hairy white mane and ewes are polled. They have white coat colour with black patches around the eyes, ears and muzzle. Yankasa rams stand 70 to 80 cm at the withers at maturity. Mature females could weigh 25 to 40kg while male weighs between 35 and 50kg. The milk yield (kg) per lactation is between 30 and 56kg and has a lactation length of 91 days. The peak milk yield per day is 960 grammes (Mason, 1996).

2.1.4 Uda

The Uda is also known as Bororo and North Nigerian Fulani. The Uda is found in Northern Nigeria, Southern Niger, central Chad, Northern Cameroon and Western Sudan. It is one of the hair sheep breeds of the Sahel type. It is also a meat breed. It is a long legged breed of sheep with distinctive coat colour of brown or black anterior and white posterior. They are large with straight and long face. The rams of the Uda are horned and the ewes are usually polled. The Uda is slightly smaller bodied than the Balami, although their size ranges overlap. The weight of mature females could be 30 to 40kg while mature rams weigh 30 to 60kg. Milk yield per lactation lies between 32 and 36kg for an average lactation length of 91 days (Mason, 1996).

2.1.5 Balami

Balami is the largest bodied native sheep in Nigeria. As a pastoral animal, it is confined to the semi-arid north but it is favoured as a stall-fed breed by Muslims throughout the Nigerian middle belt. It is white and hairy with pendulous ears, long leg and a long thin tail. Rams are horned but ewes are normally polled. Another feature that makes the Balami distinctly recognisable is its Roman, bulbous nose that distinguishes it from the Yankasa (Mason, 1996). It has good potential as a meat producer.

Table 1: Global sheep stock in 2008

COUNTRY	POPULATION (MILLION)
China	136.4
Ausralia	79.0
India	65.0
Iran	53.8
Sudan	51.1
New Zealand	34.1
Nigeria	33.9
United kingdom	33.1
WORLD TOTAL	1,078.2

Source: FAO (2008)

2.1.6 Sheep health management

Another serious constraint for small ruminant production has been high prevalence of disease and parasites. This causes high mortality amongst goats and sheep, diminishing the benefits of their high reproductive performance (Markos, 2006). Disease and health problems of sheep are closely associated with management and nutrition. Poor management is creating a favourable environment for disease incidences. Early mortalities, cold stress, starvation and non mothering (as high as 50% in lambs) are among the most important losses associated with poor managements (Tibbo, 2006). Medication cannot cure results of poor management and poor nutrition. However, sheep may fall victim to poisons, infectious diseases and physical injuries. As a prey species, a sheep's system is adapted to hide the obvious signs of illness, to prevent being targeted by predators (Simmons and Carol, 2001). Some signs of ill health are obvious, with sick sheep eating little, vocalizing excessively and being generally listless (Wooster, 2005).

According to Marie 2015, a flock health plan, tailored to the specific needs of a large producer, should include:

- Good mineral/nutritional supplementation program (after determining any deficiencies).
- Control of external and internal parasites.
- Prevention of diseases for which cost-effective vaccines are available.
- Methods to prevent introduction of contagious diseases, such as sore mouth/scab, caseous lymphadenitis, footrot, epididymitis/brucellosis, and Johne disease.
- Improvement of the number of lambs weaned per ewe bred, with enhanced ewe and ram fertility and reduced lamb mortalities (may be related to nutritional program).

The need for traditional anti-parasite drugs and antibiotics is widespread, and is the main impediment to certified organic farming with sheep (Wooster, 2005). Many breeders take variety of preventive measures to problems such as good nutrition and reduction of stress. Simmons and Carol 2001, reported that both external and internal parasites are the most prevalent malady in sheep, and are either fatal, or reduce the productivity of flocks. They are ingested during grazing, incubate within the sheep, and are expelled through the digestive

system. Diatomaceaus earth has been shown to be effective, non-chemical treatment for worm control in sheep (Barbara *et al.*, 2011).

2.2 Improving the performance of indigenous sheep breeds

There is a great, yet untapped, genetic production potential for indigenous sheep in the country, which is expressed through the high variation in milk capacity, live weight, and fertility observed in the country's sheep herds. Animal breeding system will help to overcome the high level of close blood relationships within the flocks, which depresses productivity and increases the number of genetic defects (Bernardo, 2013). Breeding through selection has led to an improvement of several production and fertility traits of indigenous sheep, an increase in the productivity of the flocks, and an improved income from selling slaughtered animals or animals for breeding purpose. One of the first steps in designing sustainable community-based breeding programs in developing countries in the tropics is to understand the socio-economic factors that influence small ruminant (i.e., goats and sheep) breeding (Kosgey, 2004). In the past, a number of livestock improvement programs have been implemented, with varying degrees of success. Reasons for failure of breeding programs designed by development agencies included not adequately understanding the needs and aspirations of the farmers. When farmers are not sufficiently involved in the design and implementation of a breeding program and when the breeding objectives of the breeding organizations are not in line with the farmers, the breeding program will often not be successful (Kosgey *et al.*, 2006a).

2.3.0 Factors affecting sheep production

Numerous factors affect livestock productivity. However, the factors that have the most significant and frequently limiting effects on livestock production include Climate, Diseases and Parasites, and Nutrition (Sejian *et al.*, 2012).

2.3.1 Climate Influence on Livestock Productivity

Of all the factors influencing livestock production, climate, and location are undoubtedly the most significant. In fact, climatology characteristics such as ambient temperature and rainfall patterns have great influence on pasture and food resources availability cycle throughout the year, and types of disease and parasite outbreaks among animal populations (Sejian *et al.*, 2012).

Large areas in Africa have climate which is characterized by rain seasonality and the existence of periods of prolonged droughts. In these vast areas, sheep production relies heavily on indigenous breeds with distinct anatomy namely fat tail and fat rump (Lundie, 2011). Fat tailed sheep anatomy and morphological characteristics have been thoroughly reviewed (Pourlis, 2011). Fat tailed sheep are characterized by a deposition of fat at the level of the hind quarters. The shape and size of the fat tail varies considerably among breeds and populations. Some of the fat tailed sheep breeds have fat accumulations in other parts of the body also, such as the rump or, less frequently, the back of the neck. Fat tailed sheep usually shed their hair, characterized as being sheep with a thicker coat in the cooler months and of varied color (Lundie, 2011).

The adults are tall and slender and tend to have long legs. Fat tailed sheep are particularly adapted to dry climates with long and persistent dry seasons. In fact, adipose tissues accumulating in the tail fat are readily mobilized in case of prolonged periods of food scarcity and correspondingly tend to decrease its size during seasonal periods of weight loss. The tall and slender ‘‘low volume’’ types of bodies of fat tailed sheep are also important adaptations to periods of heat stress characteristic of dry climates (Sejian *et al.*, 2012). Climate change and global warming are the major concern that will define livestock production systems and livestock productivity globally and will have even greater influence on selection of livestock types and breeds in the coming decades (Miraglia *et al.*, 2009), particularly on disease trends in tropical regions of Africa (Van den Bossche and Coetzer, 2008).

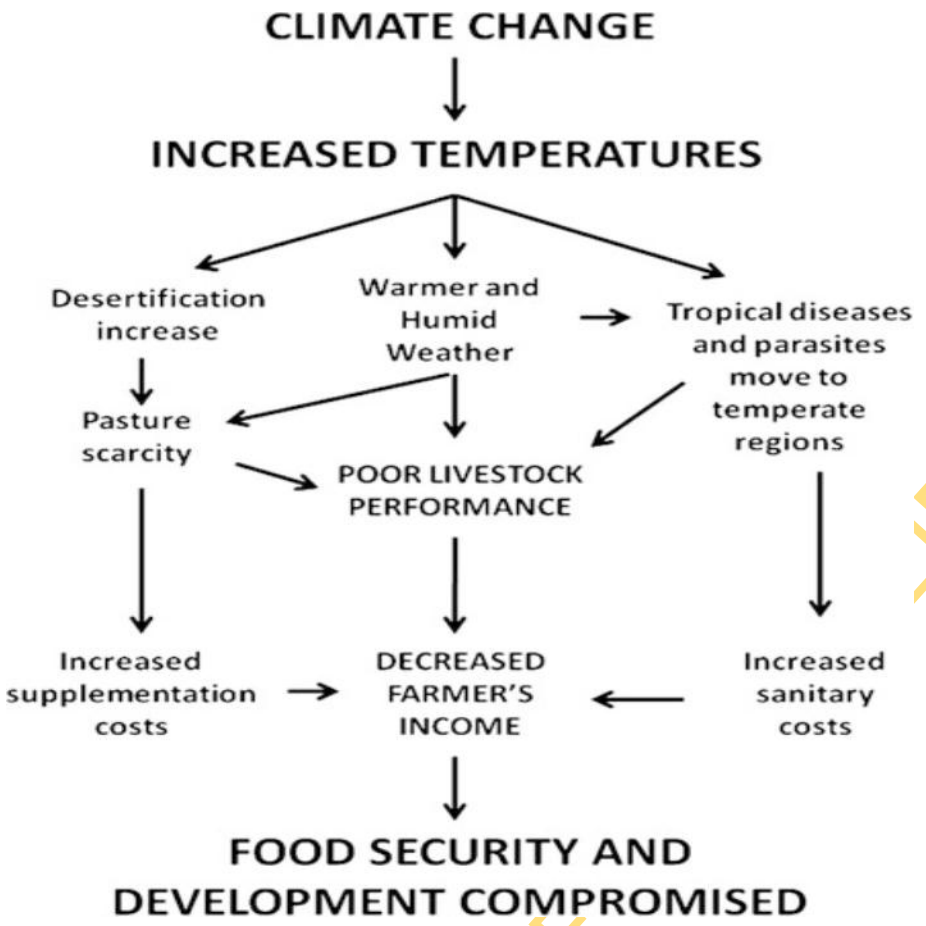

Fig. 1: *Schematic view of the expected outcome of climate change as a consequence of global warming on farm animal productivity and food Security.*

Source: Sejian *et al*., 2012.

2.3.2 Diseases and Parasites: Influence on Livestock Productivity

Diseases and parasites are among the most severe factors that impact livestock production and productivity. Animal diseases have great impact on food supplies, trade and commerce, and human health globally. The last few decades have seen a general reduction in the burden of livestock diseases. Such reduction is the direct result of the availability and effectiveness of drugs and vaccines, as well as improvements in diagnostic technologies (Pearson, 2006; Thornton 2010).

Livestock diseases are essentially an economic problem. Diseases that reduce production, productivity, and profitability are associated with the cost of their treatment, disruption of local markets, international trade, and exacerbate poverty on rural, local, and regional communities. At the biological level, pathogens compete for the productive potential of animals and reduce the share that can be captured for human purposes (FAO, 2009; Rushton, 2009). Livestock diseases can cause direct losses (deaths, stunting, reduced fertility, and changes in herd structure) and indirect losses (additional costs for drugs and vaccines, added labor costs and profit losses due to denied access to better markets and use of suboptimal production technology) in revenue (Rushton, 2009). Ruminant diseases caused by gastrointestinal nematode parasite infections are the diseases with the greatest impact on animal health and productivity (Sejian *et al.,* 2012). Ovine hemonchosis is an endemic helminth disease of considerable economic importance in tropical and sub-tropical regions of the world. Production losses result from depression in food intake, increase in the loss of endogenous proteins, reduced efficiency of use of food energy for tissue deposition, and impairment of bone growth in sheep (Bishop and Morris, 2007). Due to a growing antihelmintic resistance, there is a need to find alternative and sustainable control strategies (Magona *et al.,* 2011).

2.3.3 Nutritional Influences on Livestock Productivity

Lack of adequate year-round feed resources is probably the most important factor contributing to low animal production in arid and semiarid regions in the world (Ben Salem and Smith, 2008; Kawas *et al.,* 2010). Feeding contributes more than 60% of the total cost of production. One of the problems of feeding ruminants in Nigeria is the seasonal variation in the availability and nutritional value of native pasture. This problem can be solved through preservation of feedstuffs

(Obua, 2005). During rainy season pastures are available in higher quantities and show good nutritional quality whereas dry season's pastures have poor nutritional quality with high fiber and low protein contents. A schematic representation of the effects of Nutrition on animal productivity is presented in below.

Fig. 2. Schematic representation of the effects of poor nutrition on farm animal Productivity.

Source: Sejian *et al.*, 2012.

2.4 The Rumen Function

Sheep have rumen which allow them to make use of feeds that would otherwise be wasted if consumed, through the micro- organisms (Microbes) living in the rumen. The digestive system of ruminant is well adapted to forage based diet. Ruminants have a highly efficient anaerobic fermenting vat located at the beginning of their digestive tract, allows them to digest fibrous feed (Eryavuz *et al.*, 2003). The rumen is the largest compartment of the adult ruminant stomach. Its internal surface is covered with tiny projections, papillae, which increase the surface area of the rumen and allow better absorption of digested nutrients. When microbes breakdown and digest plant fibre, they produce volatile fatty acids which are absorbed into rumen, supplying about 60 to 80% of the ruminant's energy. The rumen and the reticulum are sometimes called reticulo-rumen because of their intimate connection. The reticulum is separated from the rumen by a ridge of tissue. Its lining has a raised honeycomb-like pattern, also covered with small papillae. The temperature inside the rumen remains stable at around 39^0C (range 38 – 42^0C) which is suitable for the growth of microbes. The pH of the content of the rumen and reticulum is maintained in the range of 6 – 7 (Kamra, 2005).

The rumen is an anaerobic system with an atmosphere of methane (30 – 40%), carbon dioxide (40%), hydrogen (5%) and a little nitrogen and low oxygen tension (Eryavuz *et al.*, 2003). Digestion in the rumen is by microbial enzymes. Saliva makes chewing and swallowing easier, but primarily, it contains sodium (Na) and Potassium (K) salt that act as buffering agents against acidity. Bacteria are the most numerous of these microorganisms and play a major role in the biological degradation of dietary fiber. *Fibrobacter succinogenes, Ruminococcus albus* and *Ruminococcus flavefaciens* recognized as the major cellulolytic bacterial species found in the rumen (Koike and Kobayashi, 2001; Koike *et al.*, 2003). Increased knowledge concerning the rumen cellulolytic bacterial population will allow insight into the fiber-digestion capabilities of ruminant animals. A number of dietary factors influence rumen fermentation and microbial population dynamics: Basal roughage sources, physical form of feed and fermentation end-products (Wanapat *et al.*, 2008). The omasum lies between the reticulum and abomasum. The materials entering the omasum are mainly up to 90 – 95% water (Eryavuz *et al.*, 2003). The primary function of the omasum is to remove some water and further grind and breakdown feed. Large plate-like folds, known as laminae extend from the walls of the omasum. The laminae are

covered in papillae which direct the flow of feed particles to the abomasum. The abomasum connects the omasum to the small intestine. Acid digestion, rather than microbial fermentation takes place in the abomasum. The lining of the abomasum is folded into ridges, which produce gastric juices containing hydrochloric acid and enzymes (pepsin). The pH of these gastric juices varies from 1 – 1.3 making the abomasum very acidic with pH of about 2 (Kamra, 2005). The acidity in the abomasum kills the rumen microbes. The pepsins carry out the initial digestion of microbial and dietary protein in the abomasum.

Rumen microbes and the host ruminant animal require many macro and micro minerals for normal growth and development. Among these minerals, sulphur is a necessary component of the amino acids; cystine and methionine are building blocks of proteins (Olafadehan *et al.*, 2014). Morrison *et al.,* (1990) found that sulphur supplementation increased the concentration of all three microbial groups but the most dramatic increase was observed with the number of sporangial forms of rumen anaerobic fungi which helps in initial fibre degradation.

2.5.0 Feed additives in ruminant nutrition

Feed additives are products used in animal nutrition to improve the quality of feed and the quality of food from animal origin, or to improve the animals' performance and health (Poppy, 2014). Chemical-based feed additives for ruminant are efficient in stimulating rumen digestion and consequently increasing sheep and goat performances, however, recent concern of consumers about the risk on consuming meat and milk of animals receiving these additives encouraged scientists to look for simple, cost-effective and healthy alternatives (Ben Salem, 2010). Animal scientists aim at replacing antibiotics with natural promoters such as probiotics, prebiotics, feed enzymes, organic acids and herbal extracts in order to achieve their production goals (Ruiz Garcia *et al.*, 2011).

Feed additives like probiotics have been reported to improve FCR in ruminants (Robinson, 2002). Probiotics are non-pathogenic microbes which occur in nature and in the gastrointestinal tract of ruminants, where they exert a positive influence on the host physiology (Dunne *et al.*, 1999). Probiotics supplementation improves feed efficiency (Abdelrahman and Hunaiti, 2008), microbial ecology (Musa *et al.,* 2009), nutrient synthesis and their bio-availability resulting in

better weight gain in farm animals (Oyetayo and Oyetayo, 2005). They improve bio-availability of nutrients and resulting in better growth performance in farm animals (Khalid *et al*., 2011). A positive impact of probiotic supplementation on nutrient intake, weight gain and feed conversion ratio (FCR) in ruminants has been reported (Antunovic *et al*., 2006).

In addition, probiotics also improve nutrient absorption, reduce the incidence of intestinal infection (Casas and Dobrogosz, 2000) and restore the gut microflora in case of diarrhoea (Musa *et al.,* 2009). It is also considered that they compete with other pathogenic micro-organisms for the provision of nutrients and other growth factors (Rolfe, 2000). They enhance immunity (Aattouri *et al*., 2001) by promoting the antibodies, IgA and cytokines production (Trebichavsky and Splichal, 2006). It is further stated that probiotics can stimulate specific groups of beneficial bacteria in the rumen, and has provided mechanistic models that can explain their effects on animal performance (Dutta *et al.,* 2009). Besides the health benefits of probiotics, it improves growth rate and feed conversion efficiency in calves (Ramaswami *et al.,* 2005), microbial protein flow (El-Hassan *et al.,* 1996) and DM intake (Putnam *et al.,* 1997) particularly in poor managemental conditions.

2.5.1 Mechanism of action of probiotics

Probiotics bacteria create different substances that are inhibitory to gram-positive and gram-negative bacteria. These inhibitory materials consist of organic acid, hydrogen peroxide and bacteriocins which are able to decrease the number of living cells in addition to influencing metabolism of bacteria or toxin yields (Rolfe, 2000). Another mechanism of action for probiotics is competitive prevention of bacterial attachment parts on intestinal epithelial surface (Dunne *et al.,* 1999). Therefore certain probiotics strains have been considered according to their capability to epithelial cells. Nutrients competition is another example of mechanism for probiotics in which probiotics utilise nutrients otherwise they can be used by harmful micro-organisms (Dunne *et al.,* 1999). Immunity can also be stimulated in the mechanism of protecting the host from intestinal disease (Antunovic *et al.,* 2006). The concept of mechanism of immune stimulation seems to be complex. However the structure and compounds of the cell wall are believed to increase the immune responses.

According to Poppy (2014), feed additives have potential to deliver the following improvements in ruminant nutrition:

- Increase feed conversion efficiency (FCE) and productivity
- Stabilise rumen pH to reduce risk of acidosis
- Increase dry matter intake (DMI)
- Reduce methanogenesis
- Enhance rumen development
- Reduce pathogen load and shedding
- Improve meat quality
- Enhance rumen stability during dietary transitions
- Buffer against dietary health risks.

Inorganic feed additives were used in some Mediterranean countries to reduce methane. Because their use had been banned by the EC, other alternatives are used such as inclusion of oils in diets, feeding diets rich in unsaturated fatty acids, and modifying feeding practices and adding supplement to roughage-based diets that are deficient nutrients (Robinson, 2002). Dietary manipulations result in methane reduction by decreasing fermentation of organic matter in the rumen and shifting the site of digestion from the rumen to the intestines, diverting hydrogen away from CH4 production during ruminal fermentation, inhibiting methanogenesis by ruminal bacteria or by optimizing the rumen fermentation and thereby decreasing methane emission per unit of organic matter digested (Ben Salem, 2010).

2.5.2 Prebiotics

Prebiotic are known as non-digestible food ingredients that can beneficially influence the host by selectively stimulating the growth or /and activity of a certain number of bacteria in the colon. In fact, they are the food for the friendly bacteria (Schrezenmeir, 2001). They may be added to the diet to provide the situation for effective bacteria to grow and survive in the digestive mechanism. Basically prebiotic foods have certain absorption, fibre contribution, gut integrity, immune function and cholesterol control (Roberfroid, 2000). They can protect against some intestinal pathogens and may be helpful in some inflammatory bowel disease. They can have some anticarcinogenic influences. In some cases, prebiotics are able to facilitate mineral

absorption and help protect against osteoporosis (Schrezenmeir, 2001). Fructo-oligosaccharides or FOS are prebiotics in association with short-chain oligosaccharides comprising D-fructose and D-glucose with 3 or 5 monosaccharides. FOS are also known as neosugar. Short-chain FOS are created on a commercial scale from sucrose utilising a fungal fructosyltransferase enzyme (Delzenne *et al.* 1995). They are digestion resistant in the upper gastrointestinal area. Their activity can stimulate the growth of Bifidobacterium species in the large intestine.

Inulins are prebiotics known as a classification of naturally-occurring oligosaccharides which contain fructose. They are originated from the roots of chicory (*Cichorium intybus*) and Jerusalem artichokes. They can stimulate the growth of *Bifidobacterium species* in the large intestine (Younes *et al.* 1996). According to Delzenne *et al.* 1995, Isomalto-oligosaccharides are composed of a combination of alpha-D- linked glucose oligomers, consisting of isomaltose, panose, isomaltotetraose, isomaltopentanose, nigrose, kojibiose, isopanose and higher branched oligo-saccharides. Isomalto-oligosaccharides develop by various enzymatic reactions. They can stimulate the growth of *Bifidobacterium spp*. and Lactobacillus spp. in the large intestine. Lactilol is a disaccharide analogue of lactulose. Pharmaceutically it is used as medication for constipation and hepatic encephalopathy. It is impervious to digestion in the upper gastrointestinal tract and it can be fermented by a certain level of colonic bacteria, which can intensify the biomass of *bifidobacteria* and *lactobacilli* in the colon (Younes *et al.* 1996).

Lactosucrose can be used as a prebiotic and it is a trisaccharide made up of D-galactose, D-glucose and D-fructose. Lactosucrose occurs enzymatically in which enzymatic transfer of the galactosyl residue in lactose to sucrose occurs. Lactosucrose is unaffected by digestion in the stomach and small intestine. It is selectively used by intestinal *Bifidobacterium spp*. stimulating distinctive growth of these bacteria in the colon. It works on the intestinal microflora as a growth element for *Bifidobacterium species* (Schrezenmeir, 2001). Lactulose is a semisynthetic disaccharide composed of sugars D-Lactose and D-fructose. The sugars are connected by a beta-glycosidic bond which makes it impervious to hydrolysis by the enzymes of human digestive. It is fermented by a certain amount of colonic bacteria, which can result in changes in the colonic ecosystem in favour of bacteria, e.g. *bactobacilli* and *bifidobacteria* that may have health advantages (Younes *et al.* 1996). Pyrodextrins is a prebiotic consisting of a combination of

glucose-containing oligosaccharides which is extracted from the hydrolysis of starch. They are able to enhance the proliferation of *Bifidobacterium species* in the large intestine. Pyrodextrins are produced for the purpose of nutritional supplement (Delzenne *et al.* 1995).

2.5.3 The Mode of action of prebiotics

The action of prebiotics may be anti-carcinogenic, antimicrobial, hypolipidemic and glucose-modulatory. They can contribute to mineral absorption and balance, and have anti-osteoporosis function. The prebiotic oligosaccharides may help increasing the concentration of calcium and magnesium in the colon. The increased concentrations of these cations in the colon are said to aid controlling the extent of cell turnover (Roberfroid, 2000). Prebiotics may encourage the growth of *bifidobacteria* and *lactobacilli* in the large intestines. They are capable of inhibiting the growth of tumors and suppress the bacteria that are likely to convert precarcinogens into carcinogens. The potential antimicrobial action of the prebiotics can be explained by their influence of growth-increasing on *bifidobacteria* and *lactobacilli* (Przemyslaw and Piotr, 2001). These bacteria are able to strengthen the barrier part of the intestinal mucosa, assisting in suppressing the connection of pathogenic bacteria mainly by crowding them out. They can also yield antimicrobial materials and encourage antigen specific and non-specific immune responses.

According to Schrezenmeir (2001), the possible influence of the prebiotics on blood glucose can be explained in different ways; the oligosaccharides may postpone gastric emptying and /or decrease the transit time in small intestinal area. This might be by the short-chain fatty acids created from the oligosaccharides in the colon. Short-chain fatty acids can affect the so-called “ileocolonic brake” that is attributed to the prevention of gastric emptying by nutrients reaching the ileo-colonic junction. Propionate can discourage gluconeogenesis by its metabolic conversion to methylmalonyl-CoA and succinyl-CoA (Przemyslaw and Piotr, 2001). In addition, propinate can decrease plasma contents of free fatty acids. Free fatty acids can reduce glucose usage and allow for insulin resistance. Propionate may increase glycolysis by depletion of citrate in hepatocytes. Citrate is basically an allosteric inhibitor of phosphofructokinase. The short-chain fatty acids produced by the bacterial fermentation of the oligosaccharides can help the colonic absorption of calcium and also magnesium ions. This can be useful in inhibiting osteoporosis and osteopenia.

2.6 Fossil shell flour as an alternative to antihelminitics / prebiotics

In two studies carried out by Barbara *et al.*, (2011) using cattle and sheep to assess the efficacy of fossil shell flour (FSF) as an alternative to antihelminitics in grazing animals, it was discovered that animals treated with fossil shell flour had low faecal egg counts (FEC) for the duration of the experimental period, similar to animals in the antihelminitics groups. Inclusion of fossil shell flour in the diet of grazing ruminants may offer some benefits in controlling internal parasites.

However, Osoweiler and Carson, (1997) in evaluation of fossil shell flour (FSF) as an adjunct to sheep parasite control on organic farming concluded that

- Fossil shell flour can be readily and conveniently incorporated into a pelleted lamb diet with good acceptance and no evidence of dust or related respiratory problems.
- Fossil shell flour used as a feed supplement for grazing lambs did not significantly reduce parasite loads as measured by feacal egg counts and abomasal gastro-intestinal larva counts.
- Fossil shell flour had no significant effect on weight gain in lambs consuming supplement containing up to 10 percent FSF during grazing trial that lasted up to 117 days.
- Fossil shell flour did not improve red cell indices (hemoglobin, packed cell volume) in lambs consuming supplement containing up to 10 percent FSF during grazing trials lasting up to 117 days.

These findings and conclusion of Osweiler and Carson, (1997) quite contradict that of Barbara *et al.*, 2011 and Nuth *et al.*, 1999. In an experiment carried out by Nuth *et al.*, (1999) to test whether there was any effect by dietary fossil shell flour in the control of gastro intestinal nematodes using a pasture trial, with 79 pregnant lactating goats of four different breeds. They concluded that the survival rate, on goats was higher than other breeds across treatments and that the feeding of FSF at 2.5% of the feed was equal to a marginally effective anthelmintics in controlling parasitic disease. It should however be noted that the pasture had been grazed by the combined flock prior to the beginning of the trial and all of the does were treated with oral

Ivermectin at a dose averaging 0.4mg/kg. During the pre-parturient phase of gestation, 12% protein concentrates in addition to access to the pasture.

The treatment groups were:

(a) No treatment (as the control)

(b) 0.4mg/kg oral ivermectin at week 1,4and 7,

(c) 0.4mg/kg oral ivermectin at week 1 plus FSF in the concentrate and

(d) FSF in the concentrate.

Their weekly feacal examinations and haematocrits were taken from each doe. There was no difference detected among the treatment groups based on egg counts or packed cell volumes. However, the estimated survival of individuals (maintained PCV>20 or EPG<4000) in any of the treatment group (2,3and 4) was statistically greater than for the control group, which shows that FSF at appropriate inclusion rate could be used to control helminthes effectively.

2.7.0 Organic livestock farming

Organic livestock farming systems are rising in many countries including developing countries due to increased consumers' demand for organic products and environmental concerns. However, organic farmers face challenges of prevention and control of diseases in farm animals and enhanced production because of ban on use of chemical drugs and feed additives (Patra, 2007). Nutritional technologies are valuable to combat some of the diseases and disorders and for improved health and welfare of animals. In recent years, organic farming is of growing importance over conventional farming in several countries worldwide (Thamsborg *et al.*, 1999) including developing countries possibly due to increased demand from consumer's for organic products. Concerns about risk of chemical drug residues, transfer of antibiotic resistance from animal to human through animal derived foods, animal welfare associated with conventional farming system, environmental effects and improved food quality in pasture based organic livestock farming have perhaps led consumers to organic food (Sundrum, 2001).

However, the process of conversion from conventional to organic farming faces several problems mainly due to inadequate technical knowledge and value-added activities at farm or regional level with poorly organized marketing (Nardone *et al.*, 2004). Breed selection, sound

animal husbandry practices and nutritional management appropriate to a particular environment are the essential keys to improve animal welfare and health and therefore successful livestock organic farming (Patra, 2007). Farming animals under an organic system requires an even greater standard of management than under conventional systems. Therefore, a central task for the future is to identify and to transfer knowledge for good manufacturing organic practices because many health problems can be resolved with good management and nutrition (Schumucher, 2004).

2.7.1 The role of organic farming in improving animal's health

Organic farming in livestock production plays a great role in internal parasite control and in the improvement of the immune system. Gastrointestinal parasitic infection is probably one of the most economic and production losses in livestock worldwide (Coop and Holmes, 1996; Waller, 2006). Nematode infections decrease feed intake, utilization of feed, body weight gain, milk production and reproductive performance. Grazing management combined with nutritional supplementation with concentrates and/or forage was the most frequently reported anti-parasite strategy in organic livestock farming (Svensson *et al.*, 2000). The sustainable nematode control strategies should entail a greater variety of control measures in combination (Waller, 2006). Coop and Kyriazakis 2001, reported that protein supplementation either in the form of by-pass protein or higher dietary protein improves resilience and expression of immunity to gastrointestinal parasites.

Minerals deficiency in animals could be more prevalent in organic animal farming because of minerals deficiency in pastures in home grown feeds and forages (Koshi and Scott, 2003). Zinc plays an important role to build up a successful immune response against gastrointestinal nematodes. Iron had presumably no direct effect on parasitic control; however, iron supplementation improves host performance because it restores iron status in the body which is lost through blood during gastrointestinal parasitic infections (Koski and Scott, 2003). Deficiency of vitamin A, B12 (or cobalt), E (or selenium) have shown to delay the adult worm expulsion, more parasitic eggs in faeces and increased fecundity due to changes in host intestinal physiology that promote host protection. Vellema *et. al.,* (1996) noted that vitamin B12 deficient lambs had higher faecal egg counts than vitamin B12 supplemented one after natural infection with gastrointestinal nematodes. The immune system is designed to resist the infectious bacteria,

virus, fungi, and protozoa. This utilizes a diverse cell populations and acts through highly developed mechanism to ward off infectious diseases. Several trace minerals, vitamins and other nutrients have been recognized as essential factors for proper functioning of immune system (Patra, 2007).

2.7.2 Benefits of using organic trace minerals for livestock production

Organic minerals have been shown to have beneficial effects under a wide range of applications in ruminants. These include higher production, increase in quality of milk, and higher reproductive efficiency. Trace mineral absorption and tissue deposition from some organic sources appear to be higher than from inorganic sources (Pal and Gowda, 2015). A relative bioavailability value of Cu and Zn from Cu-methionine and Zn-methionine was found to be 33% and 52% higher than from inorganic copper and zinc sulphate, respectively in ewes (Pal *et al.*, 2010). Many studies (Nocek *et al.*, 2006; Griffiths *et al.*, 2007; Siciliano-jones *et al.*, 2008; Hackbart *et al.*, 2010) have shown a positive effect on milk yield by supplementing cows with organic trace minerals in place of inorganic minerals.

Cows fed organic trace minerals also have greater milk fat level than those fed inorganic trace minerals. With the same level of supplementation to cows: Zn (15 mg/kg), Mn (20 mg/kg) and Cu (10 mg/kg) from chelated sources resulted higher milk yield (11%), milk fat and protein percentages (both approx.7%) when compared with inorganic sources (El Ashry *et al.*, 2012). Similarly in ewes supplemented with 40% less Zn from chelated sources (Zn-methionine) increased milk yield by 12% along with 26% and 31% increase in protein and fat production in comparison with inorganic zinc sulphate source (Hassan *et al.*, 2011). The supplementation of 40% less minerals from chelated sources resulted in 4% increase in milk yield as compared with 100% inorganic minerals supplementation in cows at late lactation stage (Somkuwar *et al.*, 2011). Several studies have suggested that organic trace minerals improve various indices of reproduction in cows, including an increase in pregnancy percentage (Nocek *et al.*, 2006; Defrain *et al.*, 2009), a reduction in open days and services per conception (Kellogg et al., 2003), and a decrease in days to first postpartum oestrus (Campbell *et al.*, 1999). Supplementation of minerals through organic sources may be more effective in ameliorating the reproductive problems in ruminants (Pal and Gowda, 2015).

2.8 Minerals

Trace minerals are essential nutrients for livestock and they affect many aspects of an animal's health and performance. These elements play critical roles in the proper functioning of enzymes, hormones and cells. Deficiencies can result in performance issues such as compromised rates of gain, milk production and reproduction. Trace mineral intake and absorption help livestock achieve optimal performance and their genetic potential, making trace mineral absorption integral to a successful herd management program (Sims and Garreth, 2010). There are many minerals that are required in the diet of sheep. Macro minerals are required in larger amounts, with that requirement expressed as a percentage of the diet or as gains per head per day. They include potassium, calcium, phosphorus, magnesium, sulphur and sodium. Micro minerals are minerals needed in very small quantities. They are also called trace minerals. The requirement by animals for these minerals are expressed in milligrams per head per day or in parts per million. They include Iron, copper, manganese, zinc and selenium (Galyean *et al.*, 1999).

Minerals deficiencies may be more prone in organic farming as farmers are to feed mostly home grown pasture and feeds where soil may be deficient in minerals. There is need to ensure that organically produced feeds are sufficient in any nutrients to keep the animals in full health and vigor at all stages of their productive life. Trace minerals that have been identified as important for normal immune function and disease resistance are zinc, manganese, selenium and copper in many field conditions (Galyean *et al*., 1999). Zn and Mn are essential for the integrity of the epithelial tissue such as gastrointestinal, urinary, reproductory and respiratory tract thereby reduce infiltration of pathogen through these epithelial lining and protect. Copper and zinc are important for both cell and humoral-mediated immunity through many molecular functions such as production of antibodies, neutrophils and lymphocytes replication, and antioxidant enzyme production (Patra, 2007). They enhance recovery rate in bacterial and viral diseases including mammary gland infection during lactation. Chirase *et al.*, (1991) noted that Zn supplementation in the form of zinc-methionine (89 ppm) or zinc oxide (90 ppm) enhanced the recovery rate from challenged infection of bovine rhinotracaeitis virus as compared to control (31-35 ppm) in cattle.

Organic Zn supplementation (Zn-proteinate) also decreased infections of the mammary gland during lactation in dairy cows (Spain 1993). Cu supplementation (20 ppm) reduces severity of udder infection challenged with *Escherechia coli* than control (6.5 ppm) in dairy heifer (Scaletti *et al.*, 2003). There is positive relationship between selenium status in conjunction with vitamin E and resistance against infections. Burton *et al.*, (1993) noted greater humoral and cell mediated immunity in cattle supplemented with 0.5 ppm chelated Cr than in unsupplemented cattle during peripaturient and early lactation when animals appeared to be in stress. Cr supplementation could be beneficial to resist important production diseases such as mastitis in dairy cows and bovine respiratory disease complex in feeder cattle (Burton *et al.*, 1993). Trace minerals provide the essential nutrients animals need for metabolic functions such as growth and development, immunity and reproduction (Scaletti *et al.*, 2003).

Underwood and Suttle (1999), stated that trace mineral functions can be described by four broad categories: structural, physiological, catalytic and regulatory. Structural function refers to minerals forming structural components of body organs and tissue. An example is the contribution of zinc to molecular and membrane stability. Physiological function occurs when minerals in body fluids and tissues act as electrolytes to maintain osmotic pressure, acid base balance, and membrane permeability. Catalytic function is probably the largest category for trace minerals as it refers to catalytic role of metalloenzymes in enzyme and hormone systems. Trace elements serve as structural components of metalloenzymes. Upon removal of the trace element or lack 7of adequate trace mineral levels the enzyme activity is lost.

There are numerous metalloenzymes that are required for a wide range of metabolic activities such as energy production, protein digestion, cell replication, antioxidant activity and wound healing (Scaletti *et al.*, 2003). Regulatory function is exemplified by the role of zinc to influence transcription and iodine serving as a constituent of thyroxine, a hormone associated with thyroid function and energy metabolism. Trace minerals bound to organic molecules have been shown to have greater bioavailability and/or tissue retention versus inorganic forms. Greater trace mineral bioavailability may support improved immune function during times of stress as well as faster growth (Bruce, 2016).

2.9 Vitamins

Sheep with their ruminant digestive system can make vitamins from the raw materials consumed in their diet. They do this very well with all of the B-vitamins. Vitamin A and E are made from compounds found in green – forage. Vitamin A can be stored in the liver for 2 to 3 months after the sheep have been eating green forage for several months (Galyean *et al.*, 1999). Consequently, when eating fresh pasture or well – made hay no supplemental vitamins are needed. However, when sheep are eating forage that is old, mature or low in vitamin A precursor, then this vitamin should be added to the mineral mixture. Vitamin D is made from exposure to sunshine. For sheep housed indoors for more than 2 to 4 weeks, such as lambs being kept in confinement, vitamin D should be included in their diets (Bendich 1993). Most commercial minerals for sheep will contain added vitamins A, D, and E. When making a total mixed ration, vitamin premixes can be added to the formulation if a free – choice mineral is not going to be fed. Carotenoids (beta-carotene and lycopene), vitamin A, E and C are naturally-occurring antioxidant nutrients that scavenge detrimental free radicals produced through normal cellular activity and from various stressors (Bendich, 1993) and appear to be important for animal health. The antioxidant function of these micronutrients could enhance immunity by maintaining the functional and structural integrity of important immune cells. Both *in vitro* and *in vivo* studies showed that these antioxidant vitamins generally enhance different aspects of cell mediated and humoral immunity. A compromised immune system will result in increased susceptibility to diseases, thereby leading to increased animal morbidity and mortality, and reduced animal production efficiency. Galyean *et al.*, 1999, in a review suggested that vitamin E supplementation greater than 400 IU could be beneficial to decrease morbidity to infectious diseases such as bovine rhinotraceitis and weight gain. These vitamins like trace minerals may be required in greater amount for normal immunity than recommended by different agencies for normal growth and milk production.

2.10.0 Nutrition in pregnant Ewe

The main goal of sheep husbandry for meat production is to wean, within reasonable amount of time, healthy lambs that have been able to fill their inherent capacity for growth and

development from ewes that are healthy and in acceptable condition to start their next production cycle (Ingvartsen, 2006). The transition time, i.e. the late gestational period that is characterized by rapid foetal growth and preparation of the mammary gland and the first days postpartum when growth potential of the lamb is highest is extremely important in order to achieve that goal. 80% of the foetal growth takes place in the last trimester of the pregnancy (Robinson *et al.*, 1999) nutritional requirements of the ewe increase to a great extent during this period. During the last six weeks of pregnancy energy requirements of twin bearing ewe are raised by more than 80 % (Robinson *et al.*, 1999) and protein requirements by 100% (Ólafsson, 1995). At the same time, pregnancy can reduce eating capacity but increase passage rate through the digestive tract. Those combined changes in late pregnancy all make it difficult to fill energy requirements of the pregnant and lactating ewe without using significant amounts of concentrates along with high quality roughage (Banchero *et al.*, 2006).

Nutrition of the pregnant ewe affects the growth of the foetus. Both directly by providing glucose, amino acids and other chemical elements necessary for development and indirectly by influencing the endocrine mechanisms that control the uptake and partitioning of nutrients to the gravid uterus and foetus (Robinson *et al.*, 1999). The nutrition of ewes in this critical prepartum period not only affects the growth of the developing foetus, but also the ability of the ewe to supply the lamb with adequate amount of colostrum and milk postpartum.

2.10.1 Energy nutrition

The main energetic substrates for foetal development and colostrum/milk production are glucose and amino acids (Ingvartsen, 2006). There is only small amount of glucose absorbed from the rumen and therefore glucose requiring tissues rely on gluconeogenesis, mainly in the liver (Banchero *et al.*, 2006). Of the fermentation products mentioned above, propionate is the main precursor for gluconeogenesis. Therefore, supplementing ewes with high starch diet such as maize and barley can result in increased foetal weight and colostrum production (Banchero *et al.*, 2007). The use of highly fermentable concentrates (rich in starch) to supply the pregnant or lactating ewe with adequate metabolizable energy (ME) has its limitations. It can induce some

metabolic disorders such as rumen acidosis and reduce degradability of other nutrients fed to the animal due to alterations of the microbial fauna. The severity of these effects depend on the type of concentrates as well as the roughage quality, particularly with regard to fibre type and content (Kaur *et al.,* 2008).

The body fat of the ewe is an important source of energy in late pregnancy and early lactation. Ruminants have excellent ability to mobilize fat and protein from their body reserves during undernutrition (Chilliard *et al.,* 2000) and negative energy balance at parturition is in fact quite normal condition (Ingvartsen, 2006). Up to 80% of initial body fat, can be mobilized when needed (Chilliard *et al.,* 2000) and research by Bell *et al.,* 2000, showed up to 34% of casein and 24% of lactose to be derived from body tissue protein. Mobilization of body tissues for energy and protein supplying is a process of high energetic cost and furthermore, use of amino acids as energy source is not very efficient. Therefore use of maternal body protein reserves is not a very profitable way for energy utilization (Thorsteinsson *et al.,* 1993).

2.10.2 Protein nutrition

Insufficient dietary protein supply as well as high level of rumen undegradable protein therefore reduces ammonia concentration in the rumen, hence the growth of the microbial population and subsequently carbohydrate digestion is reduced (Bell *et al.* 2000). Immature forages that are very rapidly fermented can yield twice as much microbial protein as diet containing high level of nutrients that are slowly fermented in the rumen (McDonald *et al.,* 2002). Excessive rumen degradable protein along with deficiency of the fermentable organic matter needed to sustain microbial growth however raises ammonia level in the rumen which causes ammonia to be absorbed into the blood and converted to urea in the liver (Nottle *et al.,* 1998). Even though some of the urea is conserved and reused by returning it to the rumen substantial part of it is excreted from the body and therefore wasted. Undegradable protein is the protein content that escapes the rumen undigested and is not utilized in any way until in the small intestines. A well-known source of undegradable protein is fish meal and other products derived from animals as well as some specially treated plant proteins (McDonald *et al.,* 2002).

Undegradable protein has for the last decades been considered one of the most important factors in nutrition of the pregnant and lactating ewe (Bell *et al.,* 2000). The use of the undegradable protein is generally thought to make it easier for the ewe to use energy from her body reserves for foetal growth and preparation of lactation (Frutos *et al.,* 1998). It is quite clear that the efficiency of protein supplementing differs to some extent between studies and the outcome is affected by several factors (McDonald *et al.,* 2002). Bell *et al.,* (2000) suggested that high feeding level postpartum could conceal the effect of prepartum nutrition. Among other benefits, some sources of undegradable protein, such as fish-meal, have especially high concentration of certain essential amino acids. McNeill *et al*., (1997) and Frutos *et al.*, (1998) found up to 7% and 6% respectively of carcass protein to be mobilized the last 30 days of pregnancy, but McNeill (1997) stated that carcass protein was the most important reserve for mobilization of carcass nitrogen. However, it has to be kept in mind that extensive protein deficiency and subsequent mobilization can be detrimental for some parts of normal protein metabolism in the body such as sustaining of the bone matrix and cause bone erosion (Frutos *et al.,* 1998, Robinson *et al.,* 1999).

2.10.3 Weight and body condition

Using live weight and weight change as an indicator of adequacy of the ewe's nutrition in late pregnancy has its limits, unless the number of foetuses is known and ewes can be compared only with ewes carrying the same litter size (Ocak *et al.,* 2005). It has been suggested that some assessment of body condition is better indicator than live weight on the fatness of the animal (Frutos *et al.,* 1998). Body condition scoring (BCS) can be used independently of litter size. Based on this knowledge BCS can be considered a useful estimator of mobilization of energy within the pregnant ewe (Frutos *et al.,* 1998) and ewe live weight and BCS together can serve as an estimator of the sufficiency of nutrition in the last week's prepartum. Both ewe weight and BCS can be affected by supplementation of the ewe in late pregnancy (Ocak *et al.,* 2005). Restrictively fed ewes lose weight earlier than body condition (Nørgaard *et al.,* 2008). When acquiring the optimal BCS it has to be kept in mind that body fatness of the ewe can affect feed intake negatively, for example through changes in the efficiency of energy utilization (Tolkamp

et al., 2007) and reduced space available for expansion of the digestive tract with increased fat deposit in the abdomen (McDonald *et al*., 2002).

2.10.4 Eating capacity

Food quality, for example with regard to fibre content and, for silage, concentrations of fermentary products, is known to affect roughage intake (Roy *et al.*, 1999). Decreased intake with lower energy content of diet has, among other factors, been related to negative effect on VFA uptake in rumen epithelium. As a result VFA can accumulate in the rumen and cause decrease in rumen pH (Ingvartsen, 2006). Furthermore, low energy diet often has high content of undegradable fibre which reduces rumen passage rate and subsequently eating capacity (Ingvartsen, 2006). Concentrate supplementing decreases roughage intake, especially with high availability and quality of roughage, a phenomenon known as substitution. Physiological factors, such as body fatness can affect feed intake to some extent (Tolkamp *et al.*, 2007). Eating capacity decreases as pregnancy proceeds and in dairy cattle pregnancy lowers eating capacity by 20-25% (Ingvartsen, 2006).

2.10.5 Blood parameters

During late gestation several metabolic changes and adaptations take place in the pregnant animals body (Ingvartsen, 2006). These changes are to some extent reflected in concentrations of certain metabolites in plasma which can provide information regarding adequacy of nutrition at each time. The advantage of measuring blood parameters to estimate nutritional status is that it gives more immediate information compared to ewe weight and body condition scoring (BCS), lamb birth weight and growth rate, the latter only presenting adequacy of nutrition on a longer term basis (Ocak *et al.*, 2005). Glucose is the main energy source for foetal growth and the undernourished ewe is able to make several alterations to make sure that the foetal needs for glucose are fulfilled (Robinson *et al.*, 1999). Elevated glucose need is reflected in the fact that glucose turnover is increased by 45% during late pregnancy and by 119% during lactation (Schlumbohm & Harmeyer, 2004). Positive relationship has been reported between energy intake and plasma glucose that is significantly elevated in the end of pregnancy (O´Doherty and

Crosby, 1997). Nevertheless, even though gluconeogenesis is increased in liver and the use of glucose in body tissue (except for mammary) is reduced, glucose level in plasma usually drops postpartum (Ingvartsen, 2006).

Non-esterified fatty acids (NEFA) are released into the blood circulation as a result of the lipolysis in adipose tissue that occurs when substrates for foetal growth and preparation of lactation are limited (Chilliard *et al.,* 2000) and serve as an important source of energy. Therefore NEFA concentration in plasma can be used as an indicator of adipose tissue mobilization (Ingvartsen, 2006). NEFA is useful in supplying the growing uterus and udder with energy and it also serves as one of the central elements in the development of the metabolic disorder "fatty liver" and can subsequently be a sign of increased risk of displaced abomasum (Chilliard *et al.,* 2000). NEFA concentration in plasma has been negatively correlated with dry matter intake (DMI), i.e. dip in DMI of late pregnancy cows causes elevated levels of NEFA in plasma (Ingvartsen, 2006).

High level of ketone bodies such as beta-hydroxy-butyrate (BHB) in plasma, along with low to normal level of glucose can be signs of the metabolic disorder ketosis. Ketosis is developed when the ewe unsuccessfully attempts to meet the increasing energy demands of her growing foetus (Harmeyer and Schlumbohm, 2006) and can develop from few circumstances. One example is energy deficiency in feed since the undernutrition stimulates lipid transfer from body reserves (Ingvartsen, 2006). According to Banchero *et al*., (2006), great loss of body condition scoring (BCS) in late pregnancy is reflected in high BHB concentration in plasma. High butyrate level in diet as well as health problems that for example cause depression in feed intake and consequently lowered glucose level are other risk factors for ketosis (Ingvartsen, 2006). Physiological status of the animal affects plasma BHB level which is significantly higher in late gestation and early lactation compared to the dry period. Furthermore, twin bearing ewes in late pregnancy and lactation have elevated BHB levels compared to single bearing ewes (Harmeyer and Schlumbohm, 2006). Based on the factors listed above it can be suggested that BHB levels

are raised as a result of increased ketogenesis in the liver when difference between required and available ME is increased (Ingvartsen, 2006).

In sheep, the level of ketone bodies in blood and consequently the risk of developing clinical ketosis or pregnancy toxaemia seem to be highest in late pregnancy. That is inconsistent with cows that are in the highest risk of pregnancy toxaemia in early lactation which is, in sheep as well as cows, the period when the gap between required and available energy is highest and the greatest mobilization of body reserves should occur (Harmeyer and Schlumbohm, 2006). Specific nutrition factors are known to affect BHB level. Soy bean protein supplementation resulted in decreased level of plasma BHB the last two weeks of pregnancy (O´Doherty and Crosby, 1997). So did supplementing concentrates to ewes grazing fields of various heights (Kerslake *et al.*, 2008). On the other hand, ewes only fed silage have shown elevated BHB levels even though no signs of pregnancy toxaemia or ketosis were found with lowered energy intake (O´Doherty and Crosby, 1997). Individual difference is however substantial, some animals showing symptoms of pregnancy toxaemia at BHB levels others can achieve without expressing any signs of discomfort (Banchero *et al.*, 2006).

Relationship between ME and BHB seems to be linear up to certain limit, afterwards BHB increases with extra energy indicating an optimum level of nutrition (O´Doherty and Crosby, 1998). Ketosis in sheep does not seem to be as affected by level of nutrition as in many other species (Harmeyer and Schlumbohm, 2006). Hyperketonaemia inhibits hepatic glucose production as well as glucose uptake and utilization in peripheral tissues and as a result of that it increases negative energy balance. Ketone bodies are not suitable as fuel for brain function and foetal growth. Therefore, the risk of pregnancy toxaemia is increased prepartum since the animal cannot use the ketone bodies to meet increasing demands of the foetus (Banchero *et al.*, 2006). When amino acids in diet exceed what is needed for maintenance and production at each time, and energy is limited, extra amino acids can be used as an energy source. Efficiency of amino acids as an energy source is however limited and disposing of extra nitrogen requires energy (Thorsteinsson *et al.*, 1993). With protein being one of the most expensive nutrients, feeding

dietary protein above requirements can be considered highly unfavourable. Acquiring the efficiency of protein feeding, detection of plasma urea level can be useful since urea is the animal's way to transport excess nitrogen without risking toxic effect (Withers, 1998). Plasma urea increases with undegradable protein supplementing but not, with "non-protein supplements" even though they give higher total protein level (O´Doherty and Crosby 1997). Plasma urea reflects adequacy of protein availability (dietary and maternal protein) compared to needs (Banchero *et al.,* 2006). Plasma urea as well as uric acid also serves as transporters of the nucleic acids derived from the small intestine digestion of the rumen microbes (Kerslake *et al.,* 2008).

Uric acid is derived from the breakdown of nucleic acids, especially purines, and its concentration is commonly related to level of microbial growth in the rumen and hence microbes digested and absorbed in the small intestines. Therefore uric acid in plasma can be used to evaluate the amount of microbial protein available for absorption in the small intestines (Pulido *et al.,* 2010). Since a great portion of nutrient partitioning and utilization takes place in the liver, concentration of some liver enzymes in plasma can indicate the level of metabolism. As an example glutamate dehydrogenase (GLDH) and aspartate aminotransferase (AST) reflect the rate of utilization of ammonia in the liver indicating active protein catabolism and metabolism and gamma-glutamyl transferase (GGT) plays an important role in peptide formation (McDonald *et al.,* 2002). Isocitrate-dehydrogenase (ICDH) however takes part in lipogenesis and is one of the most active enzymes in that matter (Suagee *et al.,* 2010). Based on this, those enzymes levels can be related with fat accumulation in the liver and the subsequent metabolic disorder fatty liver (McDonald *et al.,* 2002 and Suagee *et al.,* 2010).

Requirements for calcium are severely extended the last days prepartum and the first days postpartum as a result of high calcium level in colostrum and milk. With sufficient supply of vitamin – D the ewe has good ability to mobilize calcium from her skeleton for use in lactation but yet, calcium level of the diet at this critical time should be elevated (Suagee *et al.,* 2010) . In that respect it has to be kept in mind that reduced feed intake, for example caused by insufficient roughage quality, reduces absorption of calcium from the intestines resulting in lowered level of

plasma calcium (Harmeyer and Schlumbohm, 2006). Furthermore, both roughage and supplements are of great diversity respecting calcium supply.

2.10.6 Lamb birth weight

Insufficient nutrition in pregnancy has been shown to have effect on growth of the foetus (Frutos *et al.,* 1998 and Husted *et al.,* 2007), the effect ranging from gradually slowing down to complete cessation of growth with more sudden and severe undernutrition. Some compensation can occur in foetal growth when nutrition is increased after short period of undernutrition but longer periods result in the foetuses lacking the ability to return to normal growth (Husted *et al.,* 2007). Furthermore, late gestational undernutrition can affect the lamb's metabolic control postpartum, up to 19 weeks of age and in their adult lives under metabolic stress such as in late pregnancy (Husted *et al.,* 2008).

Ocak *et al.,* (2005) suggested that increased litter weight from ewes fed excess protein could be related to elevated production of propionate compared to other volatile fatty acids (VFA). The propionate being important source for glucose, its availability to the foetus would be higher with increased protein in diet. Birth weight is also affected by maternal live weight at parturition, measured at start of lambing. High priority of foetal growth above maternal tissues can be seen where increased level of nutrition in late gestation results in higher birth weight rather than Body condition scoring (BCS) and therefore it can be stated that birth weight is influenced by some combination of the condition of the ewe in the latter part of mid pregnancy – as defined by previous feeding and late pregnancy nutrition (Kerslake *et al.,* 2008). In addition to this, body condition of the ewe (along with its size and age) also influences the growth of the foetus by taking part in the partitioning of nutrients between the conceptus and other mammary tissues and the maternal body reserves (Robinson *et al.,* 1999).

There is a positive effect of lamb birth weight on lamb survival, especially around parturition (Nottle *et al.,* 1998). Partially this is an effect of heavier lambs being more developed and fatter at parturition, especially with regard to the amount of the brown adipose tissue that is essential

for the thermoregulation and energy supply of lambs during their first hours postpartum (Robinson *et al.,* 1999). Bigger body surface relative to body weight in lighter lambs compared to those heavier also increase their risk of hypothermia. Moreover, heavier lambs are in less risk of trauma or death because of respiratory or digestive problems (Husted *et al.,* 2007). That can to some extent be caused by the fact that heavier lambs receive more colostrum than the lighter ones and therefore absorb greater amount of immunoglobulin making them better prepared for infections. Extremes in birth weight however can result in increased lamb mortality, especially due to lambing difficulties.

2.10.7 Lamb growth rate

Although condition of the ewe and birth weight of the lamb are to some extent good indicators of nutrition during pregnancy the main goal of the prepartum feeding must be to secure sufficient rearing ability of the ewe i.e. its ability to supply enough milk to meet with the lambs capacity to grow fast (Ocak et al., 2005). Sormunen-Cristiana and Jauhiainen (2001), found positive effect of elevated energy and protein levels in late pregnancy upon growth rate of lambs during the first six weeks of their life. Furthermore, colostrum production in restrictedly fed ewes was only half of that produced by *ad-libitum* fed ewes (Nørgaard *et al.,* 2008).

Banchero *et al*., (2006) found that feeding 70% of requirement was insufficient to sustain optimal colostrum production. Moreover, Banchero *et al*. (2007) found that supplementing restrictedly fed ewes with energy rich concentrates, supplying extra ME but not undegradable protein resulted in increased colostrum production. That is probably because glucose is the main precursor for lactose synthesis and subsequently milk production and easily fermentable supplements providing high ME content are likely to elevate glucose level rapidly. Even though, Nørgaard *et al*., (2008) detected as much decrease in colostrum output with feed restriction as described previously, milk production in his restricted experimental ewes had as soon as five days postpartum reached the level found in the *ad-libitum* fed ewes. Furthermore, for the whole lactation no significant effect were found on milk production measured as lamb live weight though lambs from restricted ewes were on average little lighter than others. Undernourished

ewes have, as a result of extensive tissue mobilization in the critical period around parturition, less body reserves to rely on postpartum. Negative effect of restricted pregnancy feeding however can be overridden by successful nutrition during lactation but that requires greater quality and quantity of diet than for better nourished ewes (Nørgaard *et al.*, 2008).

Litter size, i.e. number of lambs reared by one dam, affects growth rate, probably to some extent because of lower birth weight of lambs (Sormunen-Cristiana and Jauhiainen, 2001). Lighter lambs have lower growth rate the first weeks. Lambs reared by undernourished ewes also have restricted growth rate. Since lamb growth rate is highest the first six to seven weeks of their life, the effects of ewe undernourishment as well as other factors affecting lamb birth weight cease with age, except that difference between sexes increases. As the growth period proceeds, bigger portion of the lambs' nutrition is derived from herbage allowance compared to the mothers' supply of milk (Nørgaard *et al.*, 2008).

2.11.0 Fossil shell flour (DIATOMACEOUS EARTH)

Introduction

Although it is mined from the ground, fossil shell flour (FSF) is not an earth, but fossilized deposits of microscopic shells that were created by simple one-celled plant called diatoms. Millions of years ago, in all waters of the earth these microscopic one-celled diatoms took the minerals from the waters and created protective shell for themselves (Jean, 2004). Once they died their shells drifted to the bottom of the sea beds as tiny pieces of porous "sand". In this manner, vast deposits of diatom shells were laid down where geological changes put these deposits on dry land making them accessible to mankind. Diatoms reproduce at a phenomenal rate each capable of dividing every 18 to 36 hours (Barbara, 2011). Billions of these aquatic creatures died and their shells formed mammoth deposits, some up to 700 feet thick. Layer upon layer of sediment was laid down, the primal waters came and went, but the tiny shells remained. It is this process of fossilization which partially determines the key feature of fossil shell flour namely its nature crystalline silica contents (Jean, 2004). There is probably more available

organic matter in other words, food contained in fossil shell flour than any other living thing (Osweiler and Carson, 1997).

According to Nuth *et al.*, (1999), fossil shell flour is a geological deposit made up of the fossilized skeletons of diatoms which are unicellular algae that live in ponds, streams lakes and seas. This material is made up of tiny silicon shells left by trillions of microscopic one celled algae. It is a naturally occurring soft, chalk-like sedimentary rock that is easily crumbled into a fine white to off-white powders. This powder has an abrasive feel, similar to pumice powder and is very light, due to its high porosity. Diatoms have one property that sets them apart from other micro-organisms: they have microscopic shells which they use for their protection and locomotion. These shells are covered with a pattern of tiny holes so regular that even the slightest change in their design usually signifies a different species (Robert, 2002). As some of the ancient species died, their shells survived slowly piling up in deposits at the bottom of geological lakes and lagoons. When these lakes dried up, what remained were huge deposits of fossil shell. These deposits are then mined from underwater beds or from ancient dried lake bottoms. Once fossil shell is mined, it can be milled or processed until it is almost completely amorphous for it to be used in agriculture.

Fossil shell flour has several benefits to man and to the livestock industry (Jean, 2004). Fossil shell flour is safe for human consumption and can be used as an organic feed additive for improving the performance and production of livestock (Nuti *et al.*, 2002; Jean, 2004). The health improvements observed in animals fed diets supplemented with fossil shell flour appear to be as a result of three primary actions of fossil shell flour: Eliminating parasites, reduces physiological stress and increases assimilation of nutrients from food; it is a rich source of minerals not abundantly available in today's feed crops and it may bind to toxic metal build-up and help rid it from the body (Adebiyi *et al.*, 2009). It eliminates parasites by puncturing the insect's exoskeleton and by dehydrating the parasite. As a feed additive for livestock, fossil shell flour increases herd appetite and production. It absorbs destructive and poisonous sediments in animals. It is an effective digestive aid and contains 14 trace minerals which are important in improving animals' performance (Robert, 2002; Jean, 2004).

2.11.1 Fossil shell flour as an anti-caking agent

As an anticaking agent, fossil shell flour prevents clumping of feed particles by keeping them separate so there is improved flowability, mixability and handling of the animal feed (Decrusta, 2005). It mixes well with all feeds while guarding against insect damage. It prevents worms and virus epidermis from developing. It also improves the health and growth of thriving young animals by exterminating parasites both ecto- and endo-parasites (Barbara, 2011). It works by adhering to and absorbing the waxy coating on insects causing their death by dehydration. It has been used to control pests in stored products (Barbara, 2011).

2.11.2 Fossil shell flour as a toxin binder

According to Schiable, 2003, fossil shell flour absorbs viruses, fungi and bacteria and their toxins and takes them out of the body and the animal is protected from unnecessary stress and possible death. Passing through the digestive system, fossil shell flour rubs against parasites and, being very abrasive, causes serious damage, causing the parasite to die and pass out of the animals with no negative side effects. Fossil shell flour mixes well with all feeds while guarding against insect damage. Prevents worms and keeps virus epidermis from developing, saves albumin, destroys harmful acids safeguards the stomach, improves health and growth of young animals, causes better digestion, allowing animals to absorb a higher percentage of protein from its regular diet (Schiable, 2003 and Barbara, 2011).

2.11.3 The Silica Connection

Next to oxygen silicon is the most abundant mineral on our planet making up more than 25% of the earth crust (Decrusta, 2005). On the land, the basic food for all animals is grass. Those animals that do not eat grass eat the animals that do eat grass. The silica content of all living organism is linked with the diet, silica is highest for the pure plant eaters and lowest for the pure meat eaters (Robert, 2002). Silicosis refers to lung contamination and imitation by crystalline or free silica (SiO_2). “Crystalline” describes the orientation of the SiO_2 molecule which occurs in a

fixed pattern in contrast to the non-periodic random molecular arrangement defined as amorphous (Nuti *et al*., 2002).

Silica is necessary for the formation of collagen for bones, and connective tissue, for healthy nails and hair and for calcium absorption. Silica is essential to animal health. Absence of silica results in bone and collagen abnormalities. Further research on humans shows that silica increases the thickness and strength of skin, diminishes wrinkles and gives hair and nails a healthier appearance (Philip, 2005).

Other beneficial effect of amorphous silica:

- Stimulate cell metabolism and cell formations.
- Inhibits the aging process in tissues.
- Necessary for the formation and functioning of connective tissues.
- Strengthens and stimulates the immune system.
- Silica is important for the development of healthy nails and hair and regular intake can stop unnecessary hair loss.
- Strengthens and stimulates the vascular system. Lowers blood pressure and improves the condition called arteriosclerosis.
- Increases elasticity and firmness of the blood vessels.
- Silica is indispensable for the elasticity of lung tissue and therefore is a basic therapy for lung and respiratory disorders.
- Have anti-inflammatory, disinfecting, absorbing and odor binding effects.

2.12.0 The Benefit of fossil shell flour (FSF) to Livestock Industry

According to Decrusta (2005), once fossil shell is mined, it can be milled or processed into a myriad of types for an even greater variety of uses. Filtering and filter are the two main uses but FSF also ends up in paints, cosmetics, drugs, chemical insecticides etc. Because the milling produces different sized and shaped particles, it is important not to use the filtering types for agricultural purposes. Filter-grind has long crystalline structures which will puncture tissue and

injure animals which inhale or ingest it. Fossil shell flour used in agriculture must be milled until it is almost completely amorphous (Nuti *et al.*, 2002). This means, it has no crystalline form left to cause damage to larger organisms instead, it has sharp edges, which can damage tiny particle, larvae, on stored grains.

2.12.1 Mineralization

Fossil shell flour (FSF) reduced the parasite population which in the process decreases stress on the animal and increases food assimilation and results in effective efficiency of feed conversion ratio (Barbara, 2011). Also provide a broad spectrum of naturally occurring chelated minerals. These include calcium, magnesium, iron, phosphate, sodium, titanium, potassium, and others (Philip, 2005). An Alabama study on hogs showed complete stopping of wood feeder chewing when FSF was added to the feed. It contains 14 trace minerals which are important in improving animals' performance (Robert, 2002; Jean, 2004). Robert, 2002 in his report, stated that feeding FSF at 2% can take care of both deworming and mineralization.

2.12.2 Deodorization/Absorption

Jean (2004), reported that deodorization and absorption are natural functions of fossil shell flour (FSF) and these will continue to happen as undigested FSF passes through with manure. Reduced fly hatching is usually observed in manure from livestock fed FSF. Some dairy and hog farmers are also spreading it in bedding (for odour and moisture control) in addition to that coming through the manure.

2.12.3 Fly Control/Insecticide

Fly control is a major problem with livestock operation. Fossil shell flour (FSF) can be placed in tightly woven burlap bags and hung in doorways. Livestock will be attracted to it and work the bag until their heads are covered with powder which repels flies. In closed areas FSF can be fogged with hand cranked or electric foggers to wipe out flies (Decrusta, 2005). Since FSF "kill" is always mechanical in nature, it is important that the materials come in direct contact with the insect. Mixing FSF with flies' attractants around the farm may cause them to ingest FSF in their

attempt to eat the attractant. Besides fly control, FSF can be used as an insecticide on most crops. FSF has been found to be effective in controlling aphids, brown mites, red spider mites, twig borers, oriental fruit moths and coddling moths in orchards (Decrusta, 2005).

2.12.4 Grain Protection

Fossil shell flour (FSF) offers the only easy answer to chemical contamination of stored grain wheat, oats, and spelt were kept in open bins for two years or more with no insect damage by applying FSF at approximately 7 pounds per tones (Philip, 2005). In this experiment, Philip discovered that after 12 months storage, the FSF treated material had 15 insects compared to 4884 for Malathion (a chemical insecticide) and 16,994 for untreated grains. This finding strongly supports the potency of FSF as an insecticide which can be used to store grains.

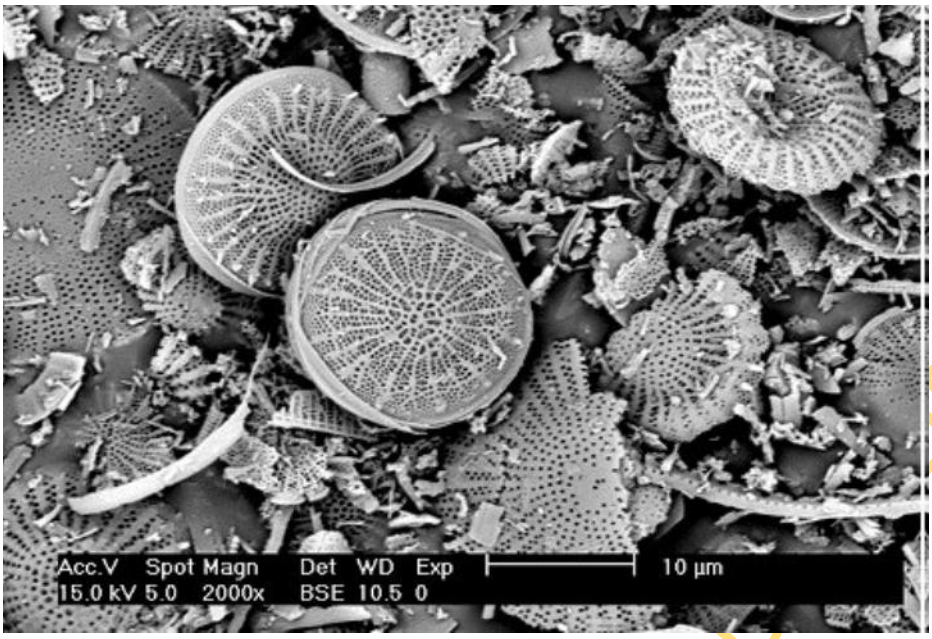

Fig. 3: Diatomaceous earth/ Fossil shell flour

Source: Barbara *et al.*, (2011)

Table 2: Some chemical composition of fossils shell flour.

Minerals	Percentage (%)
Aluminum(Al)	0.65
Boron (B)	0.0023
Phosphorus (P)	0.037
Calcium (Ca)	0.40
Potassium (k)	0.61
Sodium (Na)	0.26
Strontium (Sr)	0.0059
Copper (Cu)	0.0019
Sulphur (S)	0.062
Iron (Fe)	0.72
Titanium (Ti)	0.042
Magnesium (Mg)	0.21
Vanadium (V)	0.0044
Zinc (Zn)	0.074
Manganese (Mn)	0.0052
Silica (as SiO_2)	79.90

Source: *www.fresh waterorganics.com (accessed on 18/11/12)*

Table 3: Various functions of trace mineral and their deficiencies.

Mineral	Trace Mineral Function	Trace Mineral Deficiency
Zinc	• Protein synthesis • Vitamin A utilization • Epithelial tissue integrity • Immune System • Reproduction	• Abnormal skin and hooves • Bone and joint problems • Poor wound healing • Fertility problems
Manganese	• Bone and Cartilage Synthesis • Enzyme Systems • Reproduction • Immune Response	• Abnormal bones and joint development • Impaired ability to make or repair joint cartilage • Abnormalities in skin, hair and hooves • Reproduction challenges
Copper	• Collagen synthesis and maintenance • Enzyme function • Red blood cell maturation • Reproduction • Immune response	• Bone and joint disease • Tendon and ligament problems • Poor coat color • Early embryonic losses
Cobalt	• Required by ruminants for synthesis of Vitamin B12 by bacteria in the gut • Fiber fermentation by bacteria	• Low Vitamin B12 levels • Poor Growth • Low body condition
Iron	• Oxygen transport in hemoglobin	• Anemia is the final stage of iron deficiency • Can be caused by blood loss
Selenium	• Component of Glutathione Peroxidase • Thyroid hormone metabolism • Immune response	• Muscular cramping • Poor stress tolerance • Impaired immunity • Subpar performance
Iodine	• Thyroid hormone synthesis • Thermoregulation	• Enlarged thyroid gland; goiter • Hair loss and dry scaly skin

Source: www.zinpro.com (accessed on 07/12/16)

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1.0 STUDY ONE:

Performance and nutrients digestibility of WAD rams fed diets supplemented with varying levels of fossil shell flour

3.1.1 Experimental site

The experiment was conducted on a farmland located at Olororo along Oyo road, Ibadan, Oyo state. The location is 7.45^0 N and 3.91^0E at altitude 200-300m above sea level. The climate is humid tropical with mean temperature of 25-29^0C and the average annual rainfall of about 1250mm.

3.1.2 Management of Animals

Sixteen (16) WAD rams weighing 18.5 ±1.05kg were randomly assigned into four (4) experimental diets with four replicates per treatment. The animals were adjusted for two (2) weeks before the commencement of the experiment. During this period the animals were dewormed with levamisole and treated with oxytetracycline at a dosage of 1ml/10kg body mass against bacterial infection.

3.1.3 Housing of the Animal

The sheep were housed in metabolic cages. The cages were constructed with wood with a dimension of (2x2x4) m. The cage was covered with a corrugated iron sheet and the floor was made of slated wood to permit the passage of faeces.

3.1.4 Feeding of the Animals

The animals were fed twice daily (8am and 4pm). The animal were fed compounded ration at the rate of 4% of their body weight, fresh-clean water was served *ad-libitum* daily. The animals were weighed weekly for twelve (12) weeks to determine weight gain, Feed intake and Feed conversion ratio (FCR).

3.1.5 Digestibility and Nitrogen determinations

Digestibility of feed was carried out by the total faecal and urine collection method. During the last seven days, total feed refused (ort), faeces and urine collected were weighed and 10% aliquot taken for analysis. Urine samples were frozen until needed while feed and Faecal samples were dried at 65^0C to constant weight, milled and stored in air tight polythene bags.

3.1.6 Analytical procedure

Dried milled samples of feeds and faeces were analysed for their proximate composition according to AOAC (2002) procedure. Nitrogen contents of feed, faeces and urine samples were determined by the Kjeldahl technique using the markham distilation apparatus. Neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were determined according to Van Soest *et al*. (1991). The results obtained were used for the calculation of nutrient digestibilities and nitrogen utilization by the WAD sheep.

3.1.7 Statistical analysis

Data obtained were subjected to analysis of variance using procedure of statistical analysis system (SAS, 1999).

3.1.8 Experimental diets

TABLE 4. COMPOSITION OF EXPERIMENTAL DIETS (%).

Ingredients (%)	**T1 {control} (0%FSF)**	**T2 (2%FSF)**	**T3 (4%FSF)**	**T4 (6% FSF)**
Palm Kernel cake	16.67	14.67	14.67	14.67
Wheat Bran	20.00	20.00	19.00	19.00
Corn Bran	26.00	26.00	25.00	25.00
Breweries dried grain	31.33	31.33	31.33	29.33
Salt	2.00	2.00	2.00	2.00
Oyster shell	3.00	3.00	3.00	3.00
Premix	1.00	1.00	1.00	1.00
Fossil shell flour	0.00	2.00	4.00	6.00
Total	100.00	100.00	100.00	100.00
Calculated Nutrients				
Crude protein (%)	16.5	16.18	15.90	15.54
Metabolizable Energy (Kcal/kg)	2006.91	1963.41	1919.71	1880.11

3.2.0 STUDY TWO:

Haematological and serum biochemical studies of WAD sheep fed diets supplemented with varying levels of fossils shell flour.

3.2.1 Experimental site

The experiment was conducted on a farmland located at Olororo along Oyo road, Ibadan, Oyo state. The location is 7.45^0 N and 3.91^0E at altitude 200-300m above sea level. The climate is humid tropical with mean temperature of 25-29^0C and the average annual rainfall of about 1250mm.

3.2.2 Management of Animals

Sixteen (16) WAD sheep were randomly assigned into four (4) experimental diets with four replicates per treatment. The animals were adjusted for two (2) weeks before the commencement of the experiment. During this period the animals were dewormed with levamisole and treated with oxytetracycline at a dosage of 1ml/10kg body mass against bacterial infection.

3.2.3 Housing of the Animal

The ewes were housed in metabolic cages. The cages were constructed with wood with a dimension of (2x2x4) m. The cage was covered with a corrugated iron sheet and the floor was made of slated wood to permit the passage of faeces.

3.2.4 Feeding of the Animals

The animals were fed twice daily (8am and 4pm). The animal were fed compounded ration at the rate of 4% of their body weight, fresh-clean water was served ad-libitum daily. The animals were weighed weekly for eight (8) weeks.

3.2.5 Haematological Studies

Blood was collected fortnightly from each animal at 0800 hours by jugular vein puncture into bijou bottles. One of the bottles contained ethylenediaminetetraacetic acid (EDTA) at 1.5 mg/ml of blood as anticoagulant for haematological studies and the other was without EDTA for serum biochemical analysis. The first blood sample was collected before placing the animal on their

experimental diets. 10ml of blood was collected from each animal. 5ml of the blood samples was deposited bottles containing EDTA for heamatological studies. The remaining 5ml of the blood samples was deposited in EDTA free bottle and allowed to clot at room temperature within 3hours of collection. The serum samples were stored at -20°C for biochemical studies. Total erythrocyte (RBC) and leucocytes (WBC) counts and packed cell volume (PVC) determination was done on the same day the blood was collected. PVC was determined by microhaematocrit method. Haemoglobin (Hb) values were estimated by the alkali-haematin method according to Schalm *et. al* (1975). Values of RBC were determined by microscopic method in a counting chamber after dilution with Eayemis solution. Estimation of WBC was done in the improved Neubaurer haemocytometer chamber using 2% acetic acid as diluents. The mean corpuscular value (MCV) and mean corpuscular Haemoglobin concentration (MCHC) were calculated mathematically from values of PVC, Hb and RBC as described by (Schalm *et. al.*, 1975). Serum total protein and Albumin were determined by the method described by Peters *et al.*, (1982).

3.2.6 Normal haemathological and serum biochemical range for healthy sheep

Table 5: Normal Haematological Range for Healthy Sheep

WBC ($X10^3mm^3$)	RBC ($X10^6mm^3$)	PCV (%)	Hb (g/100mL)	MCV (μ^3)	MCHC (%)
4-12	8-15	27-45	9-15	23-48	31-34

Source: Kaneko *et al.*, 2008

Table 6: Normal Serum Biochemical Range for Healthy Sheep

Albumin (g/dL)	total protein (g/dL)	Glucose (mg/dL)	Alanine Amino transferase (ALT) (u/L)	Urea (mg/dL)	Cholesterol (mg/dL)
2.4 – 3.0	5.7 -7.9	50 -80	26 – 34	8 - 20	52 -76

Source: Kaneko *et al.*, 2008

3.2.7 The experimental diets

The composition of the experimental diets is same as given in Table 4

3.2.8 Statistical Analysis

The data collected were subjected to analysis of variance using procedure of statistical analysis system (SAS 1999).

3.3.0 STUDY THREE:

In-vitro gas production characteristics of diets containing varying levels of fossil shell flour

3.3.1 Experimental site

This study was conducted at the Ruminants Nutrition laboratory of Department of Animal science, University of Ibadan.

3.3.2 Experimental diets

The composition of the experimental diets is same as given in Table 4

3.3.3 Experimental Animals

Five (5) West African dwarf sheep were fed 40% concentrate and 60% P. maximum at 5% body weight.

3.3.4 In-vitro fermentation

Rumen fluid was obtained from the animals. The method of collection was done through the use of suction tube as described by (Babayemi and Bamikole, 2006). The rumen liquor was collected into a thermo flask that had been pre-warmed to a temperature of 39^{O}C from the sheep before they were fed in the morning. Incubation procedure was as reported by (Menke and Steingass, 1988) using 120 ml calibrated transparent syringe with fitted silicon tube. The feed sample weighing 200mg was carefully dropped into the syringe and thereafter, 30ml rumen liquor and buffer (9.8 $NaHCO_3$ +2.77 Na_2HPO4 + 0.57 KCl +0.47 NaCl +2.16 $CaCl_2 2H_2O$) was injected into the syringe containing the feed sample under continuous flushing of CO_2. The syringe was tapped and pushed upward by the piston in order to completely eliminate air in the inoculums. The silicon tube in the syringe was the tightened by a metal clip to prevent escape of gas. Incubation was carried out at 39^{O}C ± 1^{O}C and volume of gas production was measured. Readings of gas production were recorded from 6 – 96 hours. At post incubation period NaOH (10ml) was introduced to estimate methane production as reported by Fievez *et al.*, (2005). Blank syringes were incubated in each run.

3.3.5 Fermentation characteristics

The gas production characteristics were estimated using the equation

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

where y = volume of gas produced at time 't'

a = intercept (gas produced from the soluble fraction)

b = gas production from the insoluble fraction

c = gas production rate constant for the insoluble fraction (b)

t = incubation time.

Fermentation parameter estimates

- Metabolizable energy=2.20+0.136GV+0.057CP+0.0029CF (Menke and Steingass, 1988)
- Organic matter digestibility=14.88+0.889 GV+0.45CP+0.651 XA (Menke and Steingass, 1988)
- Short chain fatty acids=0.0239GV - 0.0601 (Getachew *et al*., 1999)

Where GV, CP, CF and XA are total gas volume (ml/200 mg DM), crude protein, crude fibre and ash of the incubated samples respectively

3.3.6 Statistical Analysis

The data collected were subjected to analysis of variance using procedure of statistical analysis system (SAS 1999).

3.4.0 STUDY FOUR:

Performance of pregnant WAD ewes fed diets supplemented with varying levels of fossil shell flour

3.4.1 Experimental site

The experiment was conducted on a farmland located at Olororo along Oyo road, Ibadan, Oyo state. The location is 7.45^0 N and 3.91^0E at altitude 200-300m above sea level. The climate is humid tropical with mean temperature of 25-29^0C and the average annual rainfall of about 1250mm.

3.4.2 Management of the animals

Sixteen (16) primiparous ewes weighing 30-33kg were randomly assigned into four (4) treatments with four (4) replicates per treatment. The animals were dewormed with Levamisole and treated with oxytetracycline at a dosage of 1ml/10kg body mass against bacterial infection. The animals were group penned fed twice daily and weighed weekly for 33 weeks. Clean water was constantly available throughout the experimental period.

3.4.3 Oestrus synchronization

Oestrus was artificially synchronized in all the animals with Prostaglandin 2F-alpha. Rams of known fertility and lineage bred at the research center in Olororo, Ojoo Ibadan were introduced once signs of heat were detected. The rams were allowed to mate before 0800 hours and after 1800 hours to minimize heat stress.

3.4.4 Routines observed during trial

All ewes were weighed at mating and weekly until parturition. Total feed intake was taken prior to mating and continued monthly after mating until parturition. At parturition dams were weighed, lambs were weighed and lambs' sex was recorded. Lambs were weighed weekly for 13 weeks.

3.4.5 Experimental diets

The composition of the experimental diets is same as given in Table 4

3.4.6 Analytical procedures

All data obtained were analysed using SAS (1999)

CHAPTER FOUR

RESULTS

4.1.0 STUDY ONE:

Performance and nutrients digestibility of WAD rams fed diets supplemented with varying levels of fossil shell flour

4.1.1 The chemical composition of the experimental diets

The chemical composition of the experimental diets is presented in Table 7. The dry matter content (%) ranged from 94.00 (2% inclusion level of fossil shell flour) to 95.50 (6% inclusion level of fossil shell flour), the crude protein (%) from 15.60 (6% inclusion level of fossil shell flour) to 18.55 (2% inclusion level of fossil shell flour) and the crude fibre (%) from 10.68 (6% inclusion level of fossil shell flour) to 12.37 (2% inclusion level of fossil shell flour). The ether extract (%) values were: 1.50, 1.48, 1.46 and 1.43 for 0%, 2%, 4% and 6% inclusion level of fossil shell flour respectively. However, these values were not significantly different ($p>0.05$). The ash content (%) was significantly higher (18.00) for 6% inclusion level of fossil shell flour than 0% inclusion level of fossil shell flour (5.00) but similar to 2% inclusion level of fossil shell flour (16.00) and 4% inclusion level of fossil shell flour (17.50). The values for the nitrogen free extract, neutral detergent fibre (NDF, %), acid detergent fibre (ADF, %) and acid detergent lignin (ADF, %) were not significantly different.

Table 7: Chemical Composition of Experimental Diet

Constituents (%)	T1(0%FSF)	T2(2%FSF)	T3(4%FSF)	T4(6%FSF)	SEM
Dry matter (DM)	94.50	94.00	95.00	95.50	0.80
Crude Protein (CP)	18.55	16.80	15.75	15.60	1.20
Crude fibre (CF)	12.00	12.37	10.75	10.68	1.09
Ether extract (EE)	1.50	1.48	1.46	1.43	0.57
Ash	5.00 [b]	16.00 [a]	17.5 [a]	18.00 [a]	1.25
Nitrogen free extract	62.95	53.35	54.54	54.29	1.85
Neutral detergent fibre (NDF)	40.50	39.90	40.00	40.60	0.90
Acid detergent fibre (ADF)	26.00	26.05	27.00	26.90	0.65
Acid detergent lignin (ADL)	11.50	12.00	11.90	11.85	0.45

a,b,... means on the same row with different superscripts differ significantly ($p<0.05$)

4.1.2 Performance characteristics

The results for the initial weight, final weight, total weight, daily weight gain, feed intake and feed conversion ratio (FCR) of the rams fed diets supplemented with varying levels of fossil shell flour is presented in Table 8. The Initial weight (kg) and feed intake (kg/day) of the rams were not significantly different. Final weight (kg) was significantly higher (30.50kg) for rams on 4% inclusion level of fossil shell flour than those on 6% inclusion of fossil shell flour but similar to those on 2% inclusion of fossil shell flour, however, 0% inclusion level of fossil shell flour was significantly higher (27.38 kg) than those on 6% inclusion level of fossil shell flour.

Total weight gain (kg) was significantly higher for rams on 4% inclusion level of fossil shell flour (12.00kg) than those on 6% inclusion level of fossil shell flour (5.26kg) but similar to those on 0% and 2% inclusion level of fossil shell flour, however, there was no significant difference ($p>0.05$) between 0% and 6% inclusion level of fossil shell flour. Average daily gain (kg) for rams on 4% inclusion level of fossil shell flour was significantly higher (0.20kg) than 6% inclusion level of fossil shell flour (0.11kg) but similar to 0% and 2% inclusion levels of fossil shell flour. Feed conversion ratio was significantly higher (9.90) for rams 6% inclusion level of fossil shell flour than those 0%, 2% and 4% inclusion level of fossil shell flour, however, no significant difference was observed for rams on 2% and 4% inclusion levels of fossil shell flour which were significantly lower than those on 0% inclusion level of fossil shell flour.

Table 8: Performance of WAD sheep fed diets supplemented with varying levels of fossil shell flour

Parameters	T1(0%FSF)	T2(2%FSF)	T3(4%FSF)	T4(6%FSF)	SEM
Initial weight (kg)	18.25	18.00	18.50	18.00	2.30
Final weight (kg)	27.38^{b}	29.25^{a}	30.50^{a}	23.26^{c}	2.05
Total weight gain(kg)	9.13^{ab}	11.25^{a}	12.00^{a}	5.26^{b}	4.58
Average daily gain(kg)	0.15^{a}	0.19^{a}	0.20^{a}	0.11^{b}	0.03
Feed intake (g/day)	1000	930	900	1009	9.00
Feed conversion ratio	6.67^{b}	4.90^{c}	4.50^{c}	9.90^{a}	0.03

a,b,c... means on the same row with different superscripts differ significantly ($p<0.05$)

4.1.3 Nutrients digestibility of WAD sheep fed diets supplemented with varying levels of fossil shell flour (FSF).

The results for the nutrients digestibility of WAD sheep fed diets supplemented with varying levels of fossil shell flour is presented in table 9. The dry matter (%), crude fibre (%), nitrogen free extract (%), neutral detergent fibre (%), acid detergent fibre (%) and acid detergent linin (%) digestibilities were not significantly different. Crude protein (%) digestibility was significantly higher (82.79) for sheep on 4% inclusion level of fossil shell flour than those on 2% inclusion level of fossil shell flour (77.81) but similar to those on 0% and 6% inclusion levels of fossil shell flour. Ether extract (%) digestibility was significantly ($p<0.05$) higher (77.95) for sheep on 0% inclusion level of fossil shell flour than those on 6% inclusion level of fossil shell flour (70.04), however 2%, 4% and 6% inclusion levels were not significantly different. Ash digestibility (%) was significantly higher (72.52) for sheep on 2% inclusion level of fossil shell flour than those on 0% inclusion level of fossil shell flour (63.60) but similar to those on 4% and 6% inclusion levels of fossil shell flour, however, 0% and 6% inclusion levels of fossil shell flour were not significantly different ($p>0.05$). Nitrogen free extract (%) digestibility was significantly higher (54.95) for sheep on 0% inclusion level of fossil shell flour than those on 6% inclusion level of fossil shell flour (40.24) but similar to those on 2% inclusion level of fossil shell flour while 4% and 6% inclusion levels of fossil shell flour were not significantly different.

TABLE 9: Nutrients digestibility of WAD sheep fed diets supplemented with varying levels of fossil shell flour (FSF).

Constituents (%)	T1(0%FSF)	T2(2%FSF)	T3(4%FSF)	T4(6%FSF)	SEM
Dry matter (DM)	91.30	89.66	91.31	87.90	4.82
Crude Protein (CP)	81.67 [a]	77.81 [b]	82.79 [a]	81.86 [a]	2.56
Crude fibre (CF)	77.54	76.81	79.40	75.64	5.93
Ether extract (EE)	77.95 [a]	71.78 [b]	73.76 [b]	70.04 [b]	3.86
Ash	63.60 [b]	72.52 [a]	71.57 [a]	69.07 [ab]	6 .26
Nitrogen free extract	54.95 [a]	50.05 [a]	44.44 [b]	40.24 [b]	5.40
Neutral detergent fibre (NDF)	49.50	47.00	48.25	48.50	3 .05
Acid detergent fibre (ADF)	27.04	27.80	28.05	27.00	1.98
Acid detergent lignin (ADL)	16.05	15.90	16.50	17.60	1.95

a,b... means on the same row with different superscripts differ significantly ($p<0.05$)

4.1.4 Nitrogen utilization of WAD sheep fed diets supplemented with varying levels of fossil shell flour (FSF)

The nitrogen utilization of WAD sheep fed diets supplemented with varying levels of fossil shell flour is presented in table 10. The nitrogen intake (g/day), faecal nitrogen (g/day) and urinary nitrogen (g/day) were not significantly different among the treatment groups. However, the nitrogen balance (g/day) was significantly higher (7.85) for sheep on 2% inclusion level of fossil shell flour than for those on 4% inclusion level of fossil shell flour (6.15) but similar to those on 0% inclusion level of fossil shell flour while, 4% and 6% inclusion levels of fossil shell flour were not significantly different ($p>0.05$) from each other. Nitrogen retention (%) was significantly higher (72.35) for sheep on 2% inclusion level of fossil shell flour than those on 4% inclusion level of fossil shell flour (63.33) but similar to those on 0% inclusion level of fossil shell flour (71.69) while, 4% and 6% inclusion levels of fossil shell flour were not significantly different from each other.

Table 10: Nitrogen utilization of WAD sheep fed diets supplemented with varying levels of fossil shell flour (FSF)

Parameters	T1(0%FSF)	T2(2%FSF)	T3(4%FSF)	T4(6%FSF)	SEM
N intake (g/day)	10.88	10.85	9.71	10.05	1.25
Faecal N (g/day)	2.27	1.95	2.06	1.90	0.77
Urinary N (g/day)	0.81	1.05	1.51	1.64	0.38
N Balance (g/day)	7.80[a]	7.85[a]	6.15[b]	6.50[b]	0.47
N Retention (%)	71.69 [a]	72.35 [a]	63.33 [b]	64.67 [b]	1.38

a,b,... means on the same row with different superscripts differ significantly ($p<0.05$)

4.2.0 STUDY TWO:

Haematological and serum biochemical performance of WAD sheep fed diets supplemented with varying levels of fossil shell flour.

4.2.1: Haematological parameters of West African Dwarf sheep fed diets supplemented with varying levels of fossil shell flour.

Table 11 shows the Haematological parameters of West African Dwarf sheep fed varying levels of fossil shell flour. The Packed cell volume for sheep on 2% inclusion level of fossil shell flour was significantly ($P<0.05$) higher (36.00%) than those on 0% inclusion level of Fossil flour (34.30%) but similar to those on 4% inclusion level of fossil shell flour (35.70%) and 6% inclusion level of fossil shell flour (35.05). However, the haemoglobin values (g/dL) for sheep on 4% inclusion level of fossil shell flour was significantly ($P<0.05$) higher (11.86 g/dL) than those on 0% inclusion level of Fossil flour (11.39 g/dL) but similar to those on 2% inclusion level of fossil shell flour (11.81 g/dL) and 6% inclusion level of fossil shell flour (11.80 g/dL). The red blood cell values (mm^3 $x10^6$) of sheep on 6% inclusion level of fossil shell flour was significantly ($P<0.05$) higher (13.42) than those on 0% inclusion level of Fossil flour (11.51) while those on 2% and 4% inclusion of fossil shell flour were not significantly different, however, both were significantly higher than those on 0% inclusion of fossil shell flour but lower significantly than those on 6% inclusion of fossil shell flour.

The mean corpuscular haemoglobin concentration (MCHC, %) of sheep on 6% inclusion level of fossil shell flour was significantly ($P<0.05$) higher (33.67) than those on 2% inclusion level of Fossil flour (32.81) while those on 0% and 4% inclusion of fossil shell flour were not significantly different, however, both were significantly higher than those on 2% inclusion of fossil shell flour but lower significantly than those on 6% inclusion of fossil shell flour. Mean corpuscular value (µ3) for sheep on 2% inclusion level of fossil shell flour was significantly ($P<0.05$) higher (28.87) than those on 6% inclusion level of Fossil flour (26.12) but similar to those on 0% inclusion of fossil shell flour (29.80) and 4% inclusion level of fossil shell flour (29.05). The White blood cell (mm^3 $x10^3$) for sheep on 0% inclusion level of fossil shell flour was significantly ($P<0.05$) higher (12.95) than those on 2% inclusion level of Fossil flour (8.33), however, those on 2%, 4% and 6% inclusion level of fossil shell flour were not significantly different.

Table 11: Haematological parameters of West African Dwarf sheep fed diets supplemented with varying levels of fossil shell flour

Parameters	T1 (0% FSF)	T2 (2% FSF)	T3 (4% FSF)	T4 (6% FSF)	SEM
Packed cell volume (%)	34.30^{b}	36.00^{a}	35.70^{a}	35.05^{a}	0.97
Haemoglobin (g/dL)	11.39^{b}	11.81^{a}	11.86^{a}	11.80^{a}	0.35
Red blood cell (mm^3 $x10^6$)	11.51^{c}	12.47^{b}	12.29^{b}	13.42^{a}	0.67
MCHC (%)	33.21^{b}	32.81^{c}	33.22^{b}	33.67^{a}	0.42
Mean corpuscular value (μ^3)	29.80^{a}	28.87^{a}	29.05^{a}	26.12^{b}	1.16
White blood cell (mm^3 $x10^3$)	12.95^{a}	8.33^{b}	8.95^{b}	8.52^{b}	0.95

a,b, c; Means along the same row with different superscripts differ significantly (P<0.05).

MCHC = Mean corpuscular haemoglobin concentration

4.2.2: Serum biochemical parameters of West African Dwarf sheep fed diets supplemented with varying levels of fossil shell flour.

The serum biochemical parameters of West African Dwarf sheep fed diets supplemented with varying levels of fossil shell flour is presented in Table 12. The total protein, alanine amino transferase, urea and the cholesterol of sheep fed diets supplemented with varying levels of fossil shell flour were not significantly different among the treatment groups. Albumin (g/dL) for sheep on 0% inclusion level of fossil shell flour was significantly ($P<0.05$) higher (2.79 g/dL) compared to those on 2% (2.55 g/dL), 4% (2.53 g/dL) and 6% (2.50 g/dL) inclusion level of fossil shell flour which were not significantly different from each other. Glucose (mg/dL) was significantly higher (55.35 g/dl) for sheep on 0% inclusion level of fossil shell flour than those 6% (50.52 g/dL) inclusion level of Fossil flour, however, 2% and 4% and 6% inclusion levels of fossil shell flour were not significantly different.

Table 12: Serum biochemical parameters of West African Dwarf sheep fed diets supplemented with varying levels of fossil shell flour

Parameters	T1 (0% FSF)	T2 (2% FSF)	T3 (4% FSF)	T4 (6% FSF)	SEM
Total protein (g/dL)	6.02	5.97	5.89	6.00	0.44
Albumin (g/dL)	2.79[a]	2.55[b]	2.53[b]	2.50[b]	0.12
Glucose (mg/dL)	55.35[a]	50.57[b]	50.84[b]	50.52[b]	1.22
Alanine Amino Transferase (ALT, u/L)	31.23	31.70	30.02	31.54	2.48
Urea (mg/dL)	16.26	15.87	15.84	16.77	3.13
Cholesterol (mg/dL)	63.98	64.82	64.31	63.95	2.06

a,b,… Means along the same row with different superscripts differ significantly (P<0.05).

4.3.0 STUDY THREE: *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour

4.3.1 *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 24 hours

Table 13 shows the *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour for 24 hours. The potential gas production (a+b) ranged from 15.67ml for 0% inclusion level of fossil shell flour to 19.67ml for 6% inclusion level of fossil shell flour but was not significantly different ($p>0.05$). The gas production rate (c), immediate soluble (a), insoluble but fermentable fractions (b), gas production time (t), effective degradability (y), Metabolizable energy and short chain fatty acid showed no significant differences ($p>0.05$). The organic matter digestibility (%) was significantly ($p<0.05$) higher (51.10) for diets with 6% inclusion level of fossil shell flour than those with 0% inclusion level of fossil shell flour (43.67) but similar to those with 2% and 4% inclusion levels of fossil shell flour.

Table 13: *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 24 hours

	Fermentation characteristics								
Treatments	**a**	**a + b**	**b**	**c**	**y**	**t**	**ME (MJ/Kg)**	**OMD (%)**	**SCFA (µmol)**
T1	1.67	15.67	14.00	0.061	9.33	12.00	5.47	43.67^{b}	0.43
T2	2.33	20.33	18.00	0.048	7.33	7.00	5.96	50.93^{a}	0.55
T3	1.67	19.00	17.33	0.078	11.00	11.00	5.71	50.25^{a}	0.51
T4	2.33	19.67	17.34	0.062	10.46	10.56	5.79	51.10^{a}	0.53
SEM	0.30	2.87	1.35	0.012	1.48	1.58	0.17	1.13	0.03

a,b,.. means in the same column with different superscripts differ significantly (p<0.05)
a= immediate soluble; b = insoluble but fermentable fractions; a+b = potential gas production; c = gas production rate; t= time, y= effective degradability, ME = Metabolizable energy; OMD = organic matter digestibility; SCFA = short chain fatty acid; SEM = Standard error of Mean.
T1= 0% FSF, T2 = 2% FSF, T3 = 4% FSF and T4 = 6% FSF

4.3.2 *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 48 hours

Table 14 shows the *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 48 hours. The immediate soluble fraction "a" was higher ($p<0.05$) for diets with 4% inclusion level of fossil shell flour (3.00ml) than those with 0%, 2% and 6% inclusion levels of fossil shell flour however, no significantly difference ($p>0.05$) among treatment groups were observed for those with 0%, 2% and 6% inclusion levels of fossil shell flour. The potential gas production, gas production rate (c), insoluble but fermentable fractions (b), effective degradability (y), gas production time (t) , Metabolizable energy, organic matter digestibility and short chain fatty acid were not significant different.

Table 14: *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 48 hours

Fermentation characteristics									
Treatments	**a**	**a + b**	**b**	**c**	**y**	**t**	**ME (MJ/Kg)**	**OMD (%)**	**SCFA (µmol)**
T1	0.67 [b]	23.00	22.33	0.05	16.33	10.00	6.46	50.19	0.61
T2	0.67 [b]	23.33	22.67	0.03	8.00	15.00	6.37	53.60	0.62
T 3	3.00 [a]	26.33	23.33	0.03	14.00	25.00	6.71	56.77	0.69
T 4	0.33 [b]	22.00	21.00	0.04	6.67	28.00	6.11	53.18	0.59
SEM	0.37	1.98	2.06	0.01	2.07	4.12	0.27	1.76	0.05

a,b,.. means in the same column with different superscripts differ significantly (p<0.05)
a= immediate soluble; b = insoluble but fermentable fractions; a+b = potential gas production; c = gas production rate; t= time, y= effective degradability, ME = Metabolizable energy; OMD = organic matter digestibility; SCFA = short chain fatty acid; SEM = Standard error of Mean.
T1= 0% FSF, T2 = 2% FSF, T3 = 4% FSF and T4 = 6% FSF

4.3.3 *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 72 hours

The results for the *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 72 hours is presented in Table 15. The immediate soluble fraction (a), potential gas production (a+b), gas production rate (c), insoluble but fermentable fractions (b), effective degradability (y), gas production time (t) , Metabolizable energy and short chain fatty acid were not significant different. However, the organic matter digestibility (%) of the diets with 6% inclusion level of fossil shell flour was significantly ($p<0.05$) higher (65.03) than those with 0%, 2% and 4% inclusion levels of fossil shell flour however, diets with 2% and 4% inclusion levels of fossil shell flour were not significantly different ($p>0.05$) from each other but were significantly different from those with 0% inclusion level of fossil shell flour.

Table 15: *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 72 hours

	Fermentation characteristics								
Treatments	**a**	**a + b**	**b**	**c**	**y**	**t**	**ME (MJ/Kg)**	**OMD (%)**	**SCFA (µmol)**
T1	5.33	26.67	21.33	0.03	11.67	10.00	6.96	53.44 ^{c}	0.71
T2	5.33	32.00	26.67	0.03	10.33	7.00	7.54	61.30 ^{b}	0.82
T 3	4.00	29.33	25.33	0.04	11.67	10.00	7.12	59.44 ^{b}	0.82
T 4	5.67	34.34	28.67	0.03	11.33	7.00	7.93	65.03 ^{a}	0.90
SEM	0.30	1.84	1.78	0.02	0.99	0.91	0.25	1.64	0.04

a,b,c... means in the same column with different superscripts differ significantly ($p<0.05$)
a= immediate soluble; b = insoluble but fermentable fractions; a+b = potential gas production; c = gas production rate; t= time, y= effective degradability, ME = Metabolizable energy; OMD = organic matter digestibility; SCFA = short chain fatty acid; SEM = Standard error of Mean.
T1= 0% FSF, T2 = 2% FSF, T3 = 4% FSF and T4 = 6% FSF

4.3.4 *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 96 hours

Table 16 shows the *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 96 hours. The immediate soluble fraction (a), potential gas production (a+b), gas production rate (c), insoluble but fermentable fractions (b), effective degradability (y), gas production time (t) , Metabolizable energy and short chain fatty acid were not significant different. However, organic matter digestibility (%) was significantly ($p<0.05$) higher (62.19) for diets with 2% inclusion level of fossil shell flour than those with 0% inclusion of fossil shell flour (54.53) but similar to those with 4% and 6% inclusion levels of fossil shell flour.

Table 16: *In-vitro* gas production characteristics of diets containing varying levels of fossil shell flour incubated for 96 hours

	Fermentation characteristics								
Treatments	**a**	**a + b**	**b**	**c**	**y**	**t**	**ME (MJ/Kg)**	**OMD (%)**	**SCFA (µmol)**
T1	3.33	28.33	25.00	0.04	10.00	9.00	7.19	54.53 [b]	0.74
T2	3.00	33.00	30.00	0.04	13.67	13.00	7.68	62.19 [a]	0.85
T 3	4.00	32.33	29.33	0.03	8.33	6.00	7.53	62.10 [a]	0.83
T 4	2.67	29.00	26.33	0.04	12.33	13.00	7.53	62.12 [a]	0.83
SEM	0.43	1.29	1.25	0.02	1.42	1.78	0.18	1.14	0.03

a,b,.. means in the same column with different superscripts differ significantly (p<0.05)
a= immediate soluble; b = insoluble but fermentable fractions; a+b = potential gas production; c = gas production rate; t= time, y= effective degradability, ME = Metabolizable energy; OMD = organic matter digestibility; SCFA = short chain fatty acid; SEM = Standard error of Mean.
T1= 0% FSF, T2 = 2% FSF, T3 = 4% FSF and T4 = 6% FSF

4.3.5: *In vitro* gas production values (ml/200mgDM) of diets containing varying levels of fossil shell flour incubated for 96 hours

The *In vitro gas* production values (ml/200mgDM) of diets containing varying levels of fossil shell flour incubated for 96 hours is presented in Figure 4. At the 24th hour, gas production volume ranged from 15.67ml for diets with 0% inclusion level of fossil shell flour to 20.33ml for diets with 2% inclusion level of fossil shell flour while at the 48th hour it ranged from 22.00ml for diet with 6% inclusion level of fossil shell flour to 26.33ml for diets with 4% inclusion level of fossil shell flour. At the 72th hour, the volume of gas produced ranged from 26.67ml for diets with 0% inclusion level of fossil shell flour to 34.34ml for diets with 6% inclusion level of fossil shell flour and from 29.00ml for diets with 6% inclusion level of fossil shell flour to 33.00ml for diets with 2% inclusion level of fossil shell flour at the 96th hour.

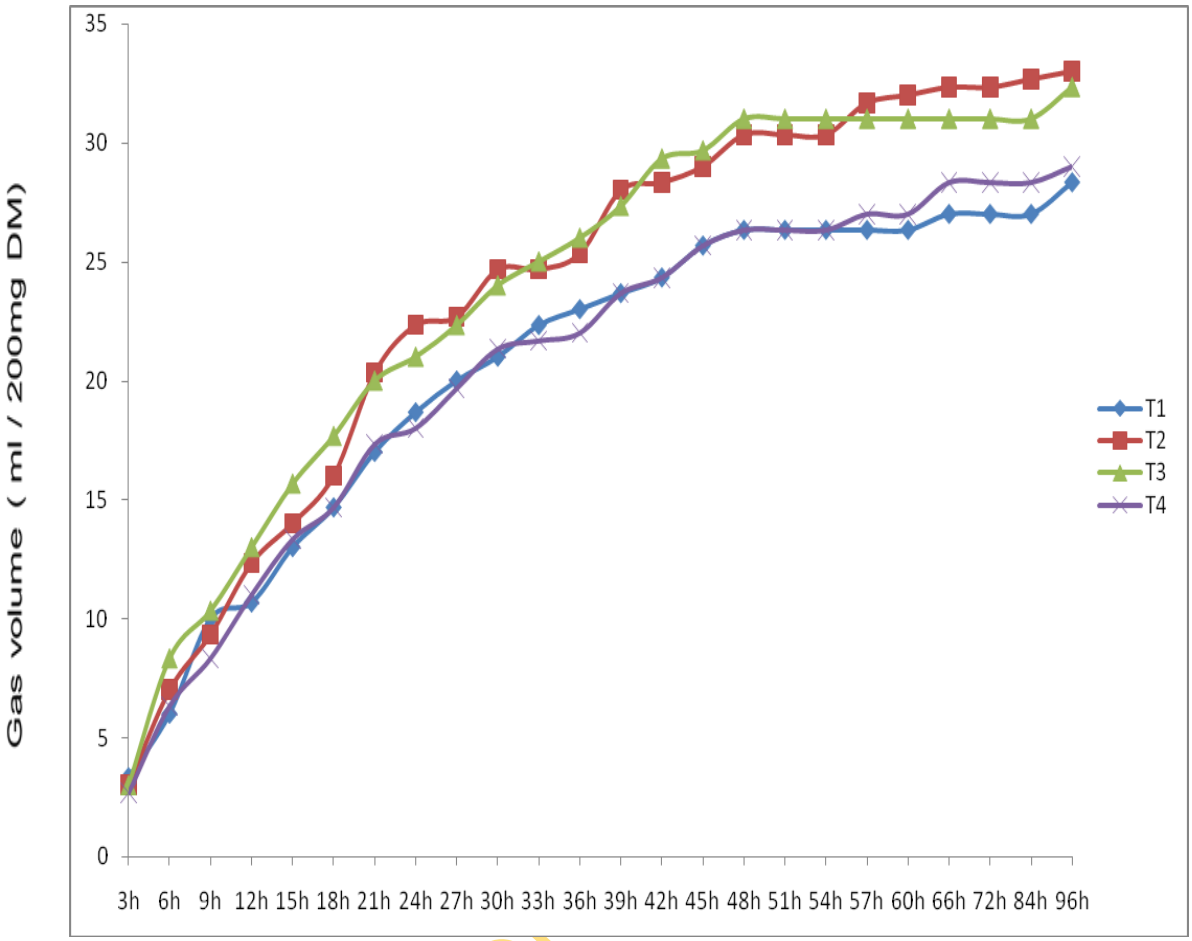

Fig. 4*: *The In vitro gas production values (ml/200mgDM) of diets containing varying levels of fossil shell flour incubated for 96 hours

4.3.6 Methane gas production of diets containing varying levels of fossil shell flour

The methane gas production of diets containing varying levels of fossil shell flour between 24th to 96th hours are presented in figures 5 to 8. At the 24th hour, methane gas production volume ranged from 14.75ml for diets with 0% inclusion level of fossil flour to 15.05ml for those with 4% inclusion level of fossil shell flour while at the 48th hour it ranged from 13.00ml for those with 0% inclusion level of fossil shell flour to 16.00ml for those with 2% and 4% inclusion levels of fossil shell flour. At the 72th hour, the volume of methane gas produced ranged from 17.00ml for diets with 2% inclusion level of fossil shell flour to 21.05ml for those with 6% inclusion level of fossil shell flour and from 18ml for diets with 6% inclusion level of fossil shell flour to 25ml for those with 4% inclusion of fossil shell flour at the 96th hour.

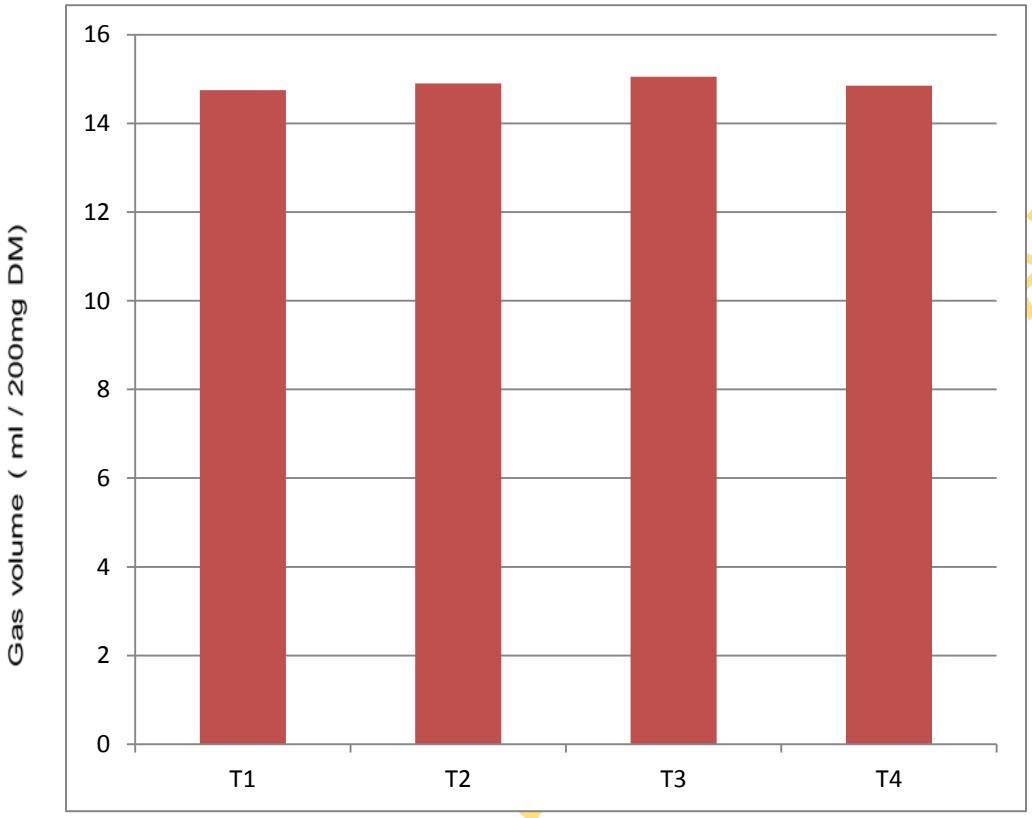

Fig.5: Methane gas production of diets containing varying levels of fossil shell flour at 24 hours

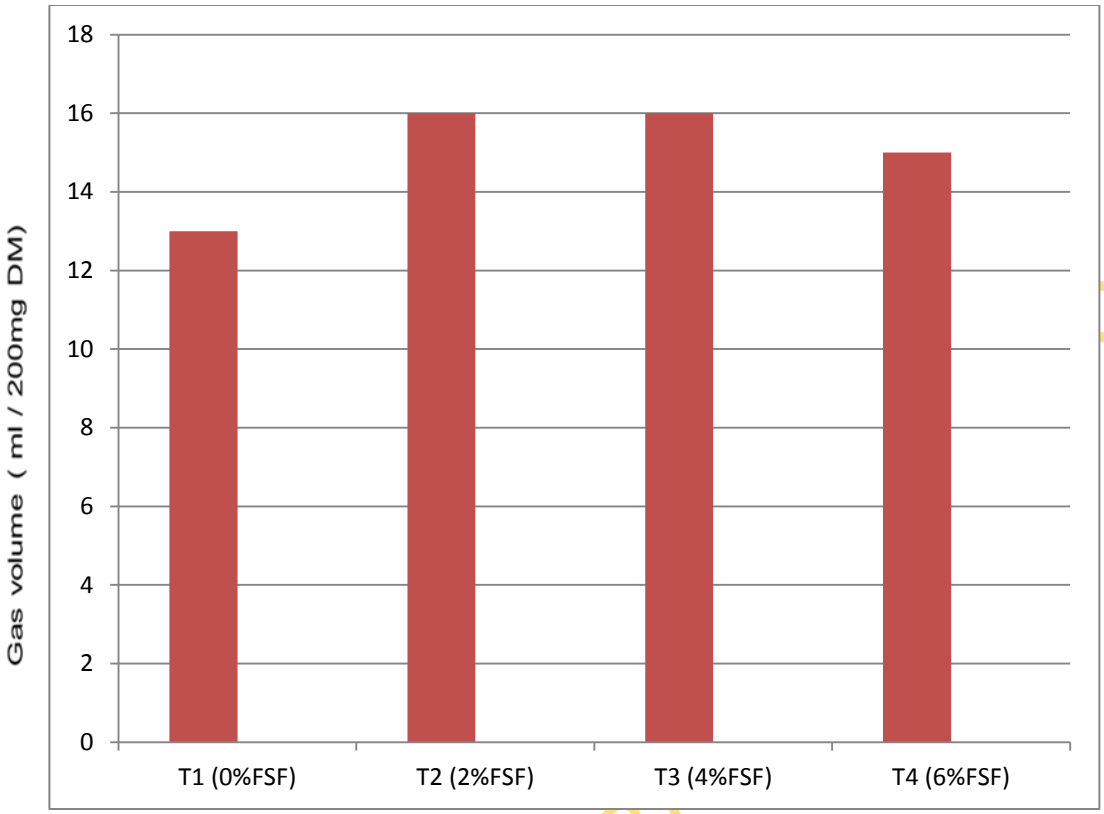

***Fig.6*:** Methane gas production of diets containing varying levels of fossil shell flour at 48 hours

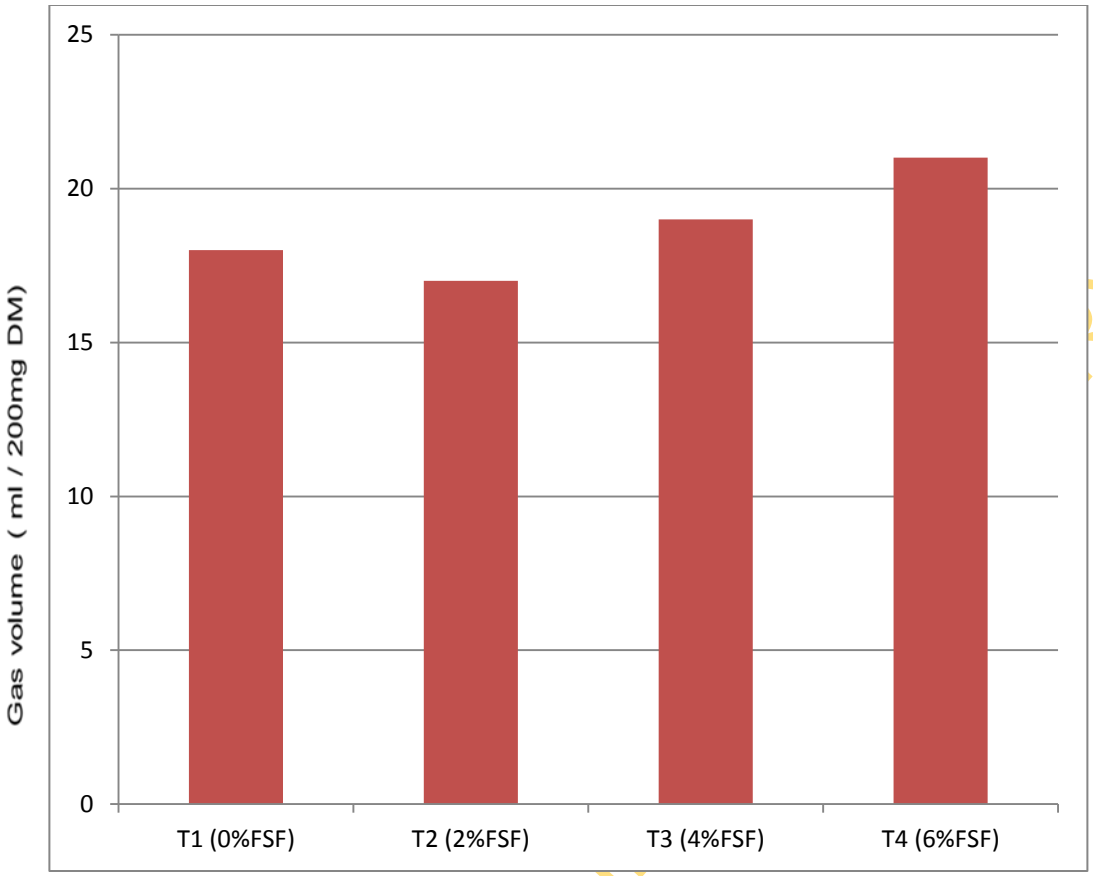

Fig.7: Methane gas production of diets containing varying levels of fossil shell flour at 72 hours

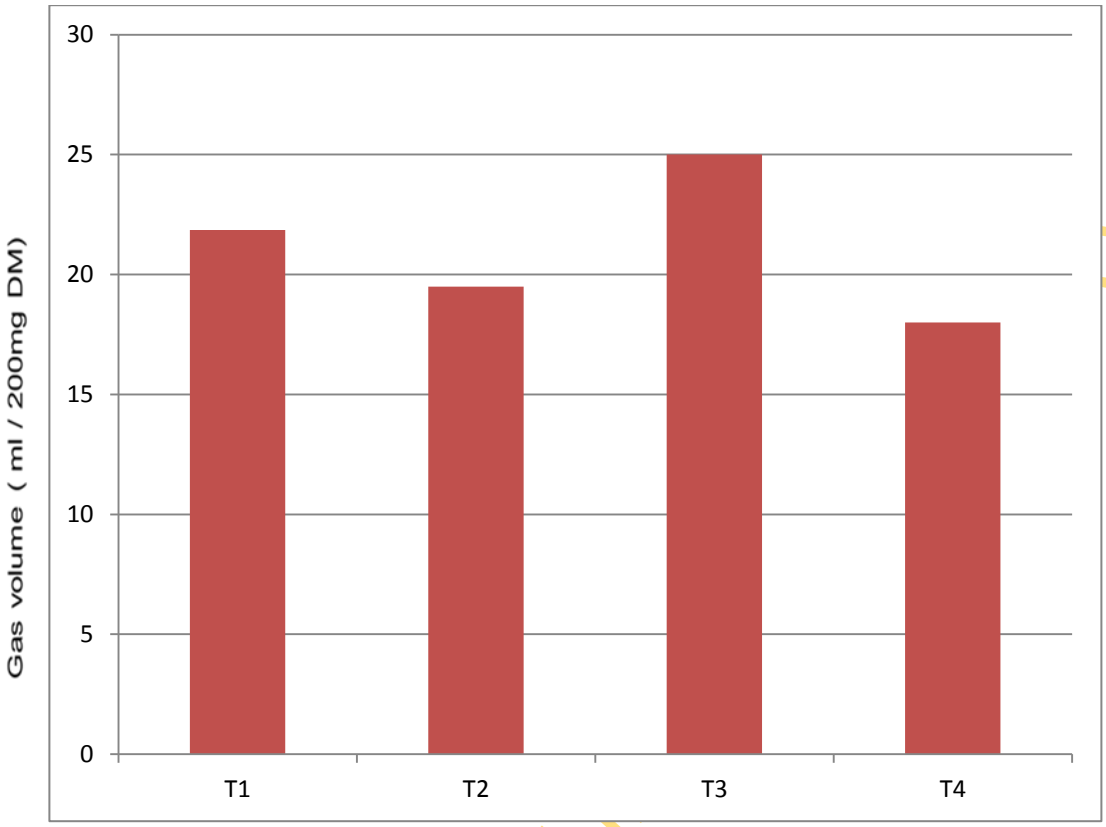

Fig.8: Methane gas production of diets containing varying levels of fossil shell flour at 96 hours

4.4.0 STUDY FOUR:

Performance of pregnant WAD ewes fed diets supplemented with varying levels of fossil shell flour

4.4.1 Reproductive performance

The reproductive performance of WAD ewes fed diets supplemented with varying levels of fossil shell flour were represented in table 17. Weight at mating, gestation length, weight gain during pregnancy, lamb birth weight and lamb weight gain were not significantly different among the treatment groups. Weight before parturition (kg) was significantly higher (48.68kg) for ewes on 0% inclusion level of fossil shell flour than those on 2%, 4% and 6% inclusion levels of fossil shell flour, however, there was no significant difference among the treatment groups for those on 2%, 4% and 6% inclusion levels of fossil shell flour. Weight at parturition (kg) also followed similar trend, ewes on 0% inclusion level of fossil shell flour was significantly higher (43.88kg) than those on 2%, 4% and 6% inclusion levels of fossil shell flour, however, there was no significant difference among the treatment groups for those on 2%, 4% and 6% inclusion levels of fossil shell flour. Dam weight at weaning was significantly higher (42.79 kg) for those on 0% inclusion level of fossil shell flour than those on 6% inclusion level of fossil shell flour but similar to those on 2% inclusion level of fossil shell flour (39.63kg), however, there was no significant difference among the treatment groups for those on 2%, 4% and 6% inclusion levels of fossil shell flour. Daily weight gain was significantly different ($p<0.05$) across the treatments groups with those on 0% inclusion level of fossil shell flour higher (72.53g/day) than those on 4% inclusion level of fossil shell flour (45.00g/day).

The dry matter intake was significantly ($p<0.05$) higher (893.29g/day) for ewes on 2% inclusion level of fossil shell flour than those on 4% inclusion level of fossil shell flour (860.74g/day) however, those on 4% and 6% inclusion levels were not significantly different ($p>0.05$). Feed conversion ratio was significantly higher (19.13) for ewes on 4% inclusion level of fossil shell flour than those on 0% inclusion level of fossil shell flour (12.09) but similar to those on 6% inclusion level of fossil shell flour however, those on 0% and 2% inclusion levels of fossil shell flour were not significantly different. Total weight gain during pregnancy was higher significantly (15.68kg) for ewes on 0% inclusion level of fossil shell flour than those on 4% inclusion level of fossil shell flour (10.25kg), however those on 4% and 6% inclusion level of

fossil shell flour were not significantly different but were significantly lower than those on 2% inclusion level of fossil shell flour. The twin: single ratio were 1:1, 3:1, 1:3 and 1:3 for ewes on 0%, 2%, 4% and 6% inclusion levels of fossil shell flour respectively, while the twin survival rate ranged from 50.0% for those on 6% inclusion level of fossil shell flour to 100.0% for those on 2% and 4% inclusion levels of fossil shell flour.

Lamb weaning weight was significantly higher (9.23 kg) for ewes on 4% inclusion level of fossil shell flour than those on 6% inclusion level of fossil shell flour (8.10kg) but similar to those on 0% and 2% inclusion levels, however, 0%, 2% and 6% inclusion levels were not significantly different among the treatment groups. Weight changes during lactation significantly decreased for ewes on 6% inclusion level of fossil shell flour (-2.75kg) compared to those on than those on 0%, 2% and 4% inclusion levels of fossil shell flour however those on 0% and 2% inclusion levels of fossil shell flour were not significantly different ($p>0.05$). Male: female ratio were 3:3, 2:5, 3:2 and 2:3 for ewes on 0%, 2%, 4% and 6% inclusion levels of fossil shell flour respectively while pre-weaning mortality ranged from 0.00% for ewes of 2% and 4% inclusion levels of fossil shell flour to 33.33% for those on 0% inclusion levels of fossil shell flour.

Table 17: Performance of pregnant WAD ewes fed diets supplemented with varying levels of fossil shell flour

Parameters	Treatments				
	T1(0%FSF)	T2(2%FSF)	T3(4%FSF)	T4(6%FSF)	SEM
Wt. at mating (kg)	33.00	30.25	31.50	30.75	4.55
Wt. before parturition (kg)	48.68^{a}	43.88^{b}	41.75^{b}	42.25^{b}	3.51
Wt. at parturition (kg)	43.88^{a}	40.38^{b}	38.25^{b}	38.88^{b}	2.49
Dam wt. at weaning (kg)	42.79^{a}	39.63^{ab}	36.75^{b}	36.13^{b}	4.49
Gestation length (days)	148.50	148.75	151.00	152.75	5.21
Wt. gain during pregnancy (kg)	10.88	10.13	6.75	8.13	4.61
Daily Wt. gain (g/day)	72.53^{a}	67.53^{b}	45.00^{d}	54.20^{c}	2.13
Dry matter intake (g/day)	876.44^{b}	893.29^{a}	860.74^{c}	866.63^{c}	7.14
Feed conversion ratio	12.09^{c}	13.23^{bc}	19.13^{a}	15.99^{ab}	4.14
Total wt. gain during pregnancy(kg)	15.68^{a}	13.63^{b}	10.25^{c}	11.50^{c}	1.41
Ratio of twin: single (%)	50:50	75:25	25:75	25:75	
Twin survival rate (%)	75^{b}	100^{a}	100^{a}	50^{c}	5.25
Lam b birth weight (kg)	2.32	2.16	2.26	2.24	0.85
Lamb weaning weight (kg)	9.00^{ab}	9.05^{ab}	9.23^{a}	8.10^{b}	0.98
Lamb wt. gain 0-13 weeks (kg)	6.68	6.89	6.97	5.86	1.85
Wt. changes during lactation(kg)	-1.09^{a}	-0.75^{a}	-1.50^{b}	-2.75^{c}	0.38
Ratio Male: female	1:1	2:5	3:2	2:3	
Pre-weaning mortality (%)	33.33	0.00	0.00	20.00	

a,b,c,... means on the same row with different superscripts differ significantly ($p<0.05$)

wt = weight

4.4.2. Average feed intake (g/day) of WAD ewes fed diets supplemented with varying levels of fossil shell flour during pregnancy

The average feed intake (g/day) of WAD ewes fed diets supplemented with varying levels of fossil shell flour during pregnancy is presented in Figure 9. At the 1st month, average feed intake ranged from 764.25g/day for ewes on 6% inclusion level of fossil shell flour to 808.50g/day for ewes on 2% inclusion level of fossil shell flour. While at the 2nd month it ranged from 706.75g/day for ewes on 6% inclusion level of fossil shell flour to 745.75g/day for ewes on 2% inclusion level of fossil shell flour. At the 3rd month, it ranged from 832.25g/day for those on 6% inclusion level of fossil shell flour to 870.75g/day for those on 2% inclusion level of fossil shell flour. At the 4th month it ranged from 908.25g/day for those on 4% inclusion level of fossil shell flour to 980g/day for those on 0% inclusion level of fossil shell flour. At the 5th month of pregnancy, it ranged from 966.75g/day for ewes on 0% inclusion level of fossil shell flour to 994.54g/day for those on 2% inclusion level of fossil shell flour while at the 6th month, feed intake decreased significantly among the treatments compared to the 5th month and ranged from 879.50g/day for ewes on 4% inclusion level of fossil shell flour to 986.75g/day for those on 2% inclusion level of fossil shell flour.

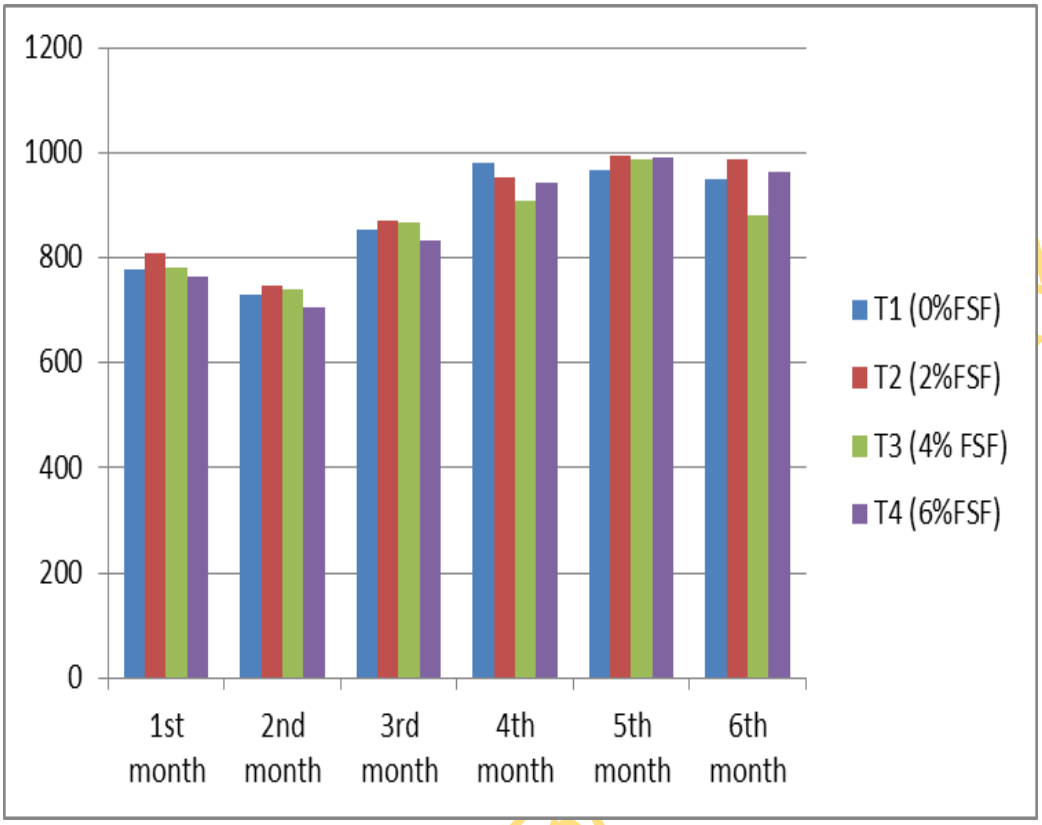

Fig. 9: Average feed intake (g/day) of WAD ewes fed diets supplemented with varying levels of fossil shell flour during pregnancy

4.4.3 Average daily weight gain (g/day) of WAD ewes fed diets supplemented with varying levels of fossil shell flour during pregnancy.

The average daily weight gain (g/day) of WAD ewes fed diets supplemented with varying levels of fossil shell flour during pregnancy is presented in figure 10. At the 1st month, average daily weight gain ranged from 40.00g/day for ewes on 4% inclusion level of fossil shell flour to 67.52g/day for ewes on 0% inclusion level of fossil shell flour. While at the 2nd month it ranged from 41.00g/day for ewes on 4% inclusion level of fossil shell flour to 68.54g/day for ewes on 0% inclusion level of fossil shell flour. At the 3rd month, it ranged from 44.00g/day for those on 4% inclusion level of fossil shell flour to 71.52g/day for those on 0% inclusion level of fossil shell flour. At the 4th month it ranged from 47.00g/day for those on 4% inclusion level of fossil shell flour to 74.54g/day for those on 0% inclusion level of fossil shell flour. At the 5th month of pregnancy, it ranged from 70.52g/day for ewes on 2% inclusion level of fossil shell flour to 75.53g/day for those on 0% inclusion level of fossil shell flour while at the 6th month, it ranged from 50.00g/day for ewes on 4% inclusion level of fossil shell flour to 77.54g/day for those on 0% inclusion level of fossil shell flour.

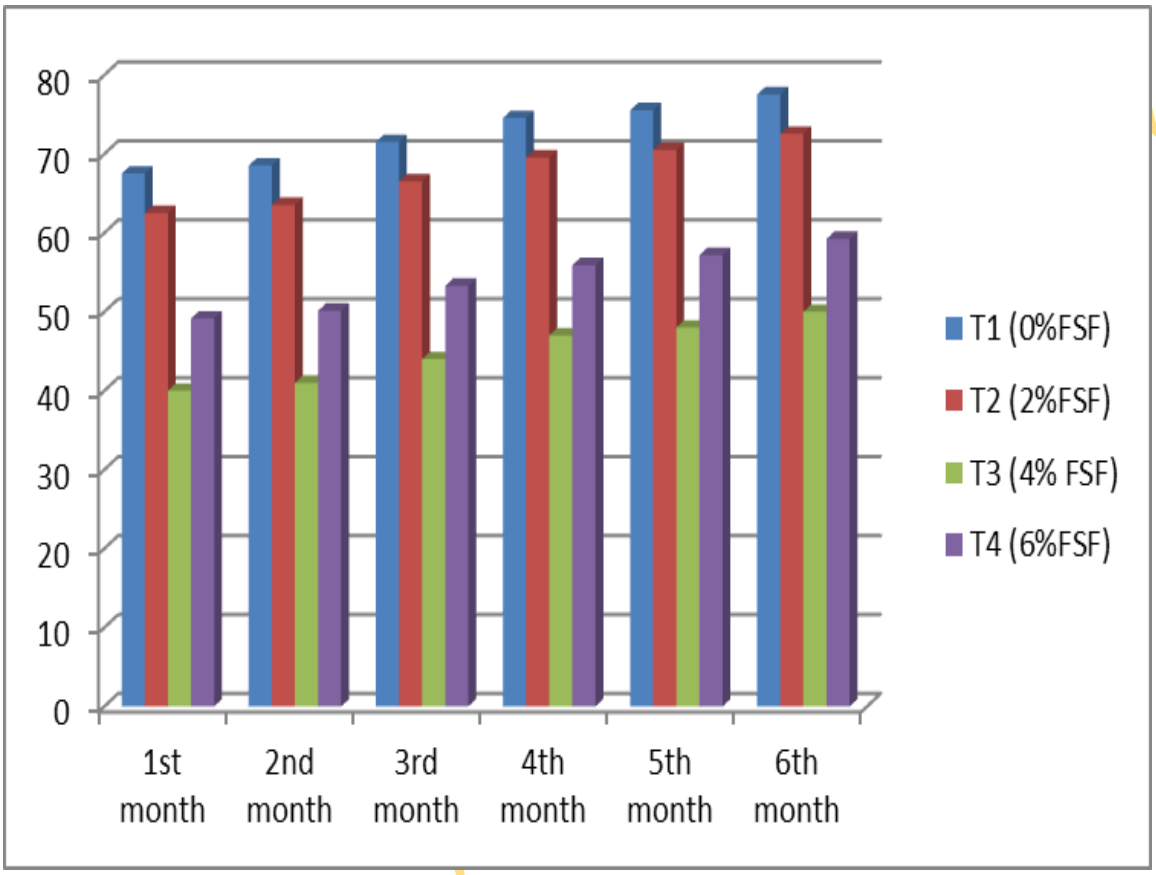

Fig. 10: Average daily weight gain (g/day) of WAD ewes fed diets supplemented with varying levels of fossil shell flour during pregnancy

CHAPTER FIVE

DISCUSSION

5.1.0 STUDY ONE: Performance and nutrients digestibility of WAD rams fed diets supplemented with varying levels of fossil shell flour

5.1.1 Chemical composition of experimental diet

The high crude protein content of the concentrate diet shows that it is high enough to meet the optimum microbial need in the rumen. The values for all the treatment groups are above 7% protein requirement for optimum microbial growth in the rumen. The values are also above the 10 to 12% crude protein requirement for growth performance of sheep and goats (Gatemby, 2002). The ash content of the diet increased across the treatments. This variation may be as a result of fossil shell flour containing some trace minerals which are needed to improve the performance of the animals (Babara *et al.*, 2000). The fibre content decreased at higher inclusion of fossil shell flour i.e 4-6% level of inclusion of fossil shell flour. The fibre fraction ADF, NDF and ADL were not significant different ($p>0.05$) across the treatments. The knowledge of nutrient requirements is therefore important for the estimation of genetic potential of the animals. Nutrient requirements depend on body size or growth rate with the quality of the feed and environmental condition. There are many factors that affect the chemical composition and mineral content of feedstuffs such as stage of growth (maturity), spices or variety (Agbagla-Donhnani *et al*., (2001); Promkot and Wanapat, (2004), drying methods, growth environment (Mupangwa *et al.,* 1997) and soil types (Thu and Preston, 1999). These factors may be applicable in this study.

5.1.2 Performance characteristics

Feed additives like probiotics and prebiotics have been reported to improve feed conversion ratio (FCR) in ruminants (Robinson, 2002). It improves microbial ecology (Musa *et al.*, 2009), nutrient synthesis and their bio-availability resulting in better weight gain in farm animals (Oyetayo and Oyetayo, 2005). Jang *et al.* (2009) found that probiotics supplementation tended to increase weight gain in lambs. Higher weight gain in lambs fed diets containing probiotics could be due to augmented microbial protein synthesis leading to more amino acids supply at

post-ruminal level (Erasmus *et al.*,1992). Probiotics enhanced digestion and FCR and improved weight gain in small ruminants (Robinson, 2002).

Mutassim *et al.*, (2008) found that supplementation of 2 g cyc-methionine/lamb/day showed better FCR compared to control, indicating that feeding sheep yeast and methionine in the form of cyc-methionine with low level of 2g/day improves the efficiency of feed utilization. The inclusion of trace minerals in the diets of small ruminants provide the essential nutrients needed by animals for metabolic functions such as growth and development, immunity and reproduction (Scaletti *et al.*, 2003). Growth rate and feed conversion ratio (FRC) are among the most important factors affecting economical ram meat production (Esenbuga *et al.*, 2009).

In this study, the finding obtained for average daily weight gain was significantly influenced by the inclusion of fossil shell flour in the diets of the rams with the highest value (0.20kg/day) for those on 4% inclusion level of fossil shell flour and the lowest value of (0.11kg/day) for those on 6% inclusion of fossil shell flour. Feed conversion ratio values (FCR) was lowest with rams on 4% inclusion level of fossil shell flour (4.50). The values of FCR approximating 4.90 to 4.50 for rams on 2% and 4% inclusion levels of fossil shell flour could indicate a relatively good feeding level with experimental ration. This is in agreement with work done by kawas *et al.*, 2010 who recorded FRC of 5.70 to 4.10 from rations of higher concentrate inclusion ratio. Much higher values (9.00 to 12.00) were observed by Mahgoub *et al.*, (2000), with Omani rams, whose ration consisted of 60% forage. The efficiency at which rams converted feeds for body weight in this study also compared favourably with the previous study of Gatemby (2002) for sheep. Daily feed intake was also lowest for rams on 4% inclusion level of fossil shell flour (900g/day). The daily feed intake values of 930g/day and 900g/day obtained in this study for rams on 2% and 4% inclusion levels of fossil shell flour were consistent with those of Gokdal *et al.*, (2004) and Esenbuga et al., (2009) who studied different rams.

5.1.3 Nutrients digestibility of WAD sheep fed varying levels of fossil shell flour.

According to Abd El-Ghani, 2004, feed additives like Probiotics improve nutrient digestibility, degradation of fiber (El-Waziry and Ibrahim, 2007) and ruminal digestion (Kamel *et al.*, 2004) more likely fiber degradation (Dawson and Tricarico, 2002) by increasing pH in the rumen (Mohamed *et al.*, 2009; Paryad and Rashidi, 2009), enhancing growth and/or cellulolytic activity

by rumen bacteria (Dawson and Tricarico, 2002) and preventing ruminal acidosis by balancing the VFAs ratios in the rumen (Arcos-Garcia *et al.*, 2000). Krehbiel *et al.*, (2003) reported that feed additives and direct-fed microbes improved digestibility of the diet. Whitley *et al.*, (2009) also reported improved apparent DM, CP, NDF and ADF digestibility in goats fed diet supplemented with commercial probiotics than control group. In this study the dry matter and crude protein were more than 80% digestible in all the treatments except for rams on 2% inclusion level of fossil shell flour where the crude protein digestibility was 77.81%. The values of crude protein obtained in this study were slightly higher than those obtained by Hadjipanayiotu (1990), who obtained crude protein digestibility of 74 – 75% for goats fed concentrate supplement diets.

The CP digestibility obtained in the present study is in conformity with the digestibility values reported by Maigandi and Wasagu (2002) whose values ranged between 75.5% and 81.06% when they fed *Ficus sycomorus* leaves to Yankasa rams. The result also contradicts the CP digestibility values reported by Ahamefule *et al.*, (2002) who reported 43.66 to 57.69% when they fed potatoes peels-yeast slurry diet to West African Dwarf goats. The DM digestibility values in the present study are higher than results obtained by Usman *et al.*, (2008) whose values ranged between 53.02% and 74.87% when they fed fore stomach digesta as replacement for cowpea husk. The result also conformed with the DMD values reported by Aruwayo *et al.*, (2007) whose values ranged between 61.39% and 84.37% when they fed fore stomach digesta and poultry waste to Uda lambs. In this study, ash digestibility was greatly influenced by the inclusion of 2% and 4% inclusion levels of fossil flour compared to that of rams on 0% inclusion level of fossil shell flour. This could be due to the fact that fossil shell flour is mainly made up of organic minerals. Meanwhile, FAO (1995) classified digestibility of feed as high (>60%), medium (40-60%) and low (< 40%). The apparent digestibility of dry matter, crude protein, crude fibre, ether extract and ash were high while that for nitrogen free extract and neutral detergent fibre were medium and low for acid detergent fibre and acid detergent lignin. The high values observed in this study were in agreement with the observation of Roberts (2002), stating fossil shell flour as an effective digestive aid in ruminants.

5.1.4 Nitrogen utilization of WAD sheep fed varying levels of fossil shell flour

The results from this study shows that the inclusion of fossil shell flour up to 6% inclusion level in the diets of WAD rams had no effect on nitrogen intake, faecal nitrogen and urinary nitrogen. This is in agreement with work done by Hernandez *et al*., (2009), who reported no effect of probiotics supplementation on N-intake and fecal and urinary N in lambs fed mature orchard grass. The result on Nitrogen retention from this study is in agreement with the report of Paryad and Rashidi (2009), who stated that feed additives like Probiotics have been reported to enhance N-retention by enhancing microbial peptidolytic and proteolytic activities in the rumen and post-ruminal amino acid flow. The results obtained from this study indicated that all the treatments promoted positive balance with rams on 2% inclusion level of fossil shell flour having the highest value (7.85g/day) followed by those on 0% inclusion level (7.80g/day), 6% inclusion level (6.50g/day) and least for those on 4% inclusion level of fossil shell flour (6.15g/day). The positive nitrogen balance observed in all the sheep suggested that nitrogen absorbed was well utilized by the sheep. This also indicated that the treatment furnished enough nitrogen required for body growth and maintenance.

5.2.0 STUDY TWO:

Haematological and serum biochemical studies of WAD sheep fed varying levels of fossils shell flour.

5.2.1 Haematological parameters

The haemoglobin values obtained in this study fell within normal values recorded for healthy Sheep. This is an indication that diets seemed to be capable of supporting high oxygen carrying capacity in the animals. The Packed cell volume and Red blood cell values reported in this study were within the range of values reported by Eniolorunda *et al.*, (2005) and Jain (1993) respectively for sheep in separate experiments, which indicated that the animals used in this study were not susceptible to anaemia related diseases. The values for the Packed cell volume and haemoglobin were higher in the sheep on the treatment groups i.e for sheep on 2-6% inclusion levels of fossil shell flour than the control (0% inclusion of fossil shell flour). This contradicted the finding of Osweiler and Carson (1997) that did not find any improvement in the haemoglobin and Packed cell volume of lambs fed diets supplemented with fossil shell flour. The white blood cell count was also within the normal range for healthy sheep for all treatments except for those on 0% inclusion level of fossil shell flour. This could be as a result of the fact that the sheep on 0% inclusion of fossil shell flour may be reacting against the external and internal parasites hence producing more white blood cells (WBC) to fight the pathogens or even toxins of the parasites. This is not observed in the treated groups with the inclusion of fossil shell. Fossil shell flour is known to be effective in handling these parasites by effectively killing these parasites by abrasive physical method (Nuti *et al.*, 2002; Schiable, 2003; Jean, 2004). Under a wide range of feeding regimes, an animal maintains its blood picture within ranges normal for that breed in that environment. Once feed is adequate, variation in haematological indices could indirectly be due to reactions to external changes rather than direct response to feeding regime (Uwechue, 2000).

5.2.2 Serum biochemical parameters

Blood urea nitrogen (BUN) concentration is indicative of renal function. Gluconeogenesis is a major source of glucose in lambs as it is responsible for the satisfaction of 75% of the totalglucose needs in ruminants (Donkin and Hammon, 2005). In this study, the variations

observed for serum biochemical parameters were within the normal range for healthy sheep. This indicates that the inclusion of fossil shell flour up to 6% in the diets of WAD sheep may not impair protein digestion, metabolism and utilization and may also not have negative effect on the kidney. This is in agreement with the findings of Robert, 2002 who reported no organ abnormality in dairy cows fed diets supplemented with 2% fossil shell flour.

5.3 STUDY THREE:

In-vitro gas production characteristics of diets containing varying levels of fossil shell flour

5.3.1 In-vitro fermentation characteristics and gas production

Gas production is a function and mirror of degradable carbohydrates and the amount of gas produced depends on the nature of the carbohydrates (Blummel and Becker, 1997). It is nutritionally wasteful but provides useful basis from which metabolizable energy, organic matter digestibility and short chain fatty acid may be predicted. The high potential gas production observed in all the treatments is due to the highly fermentable nature of the diets. However, diets with 2% and 4% fossil shell flour respectively have the highest gas production compared to other treatments this could be attributed to their high OMD (organic matter digestibility) depicting better degradability. Getachew *et al.,* (1998) reported that rapidly soluble carbohydrates produce higher acetate. Fermentable carbohydrates increase gas production while degradable nitrogen compounds decrease gas production to some extent because of the binding of carbondioxide to ammonia (Krishnamoorthy *et al.,* 1995). There are many factors that may determine the amount of gas to be produced during fermentation, depending on the nature and level of fibre and potency of the rumen liguor for incubation. High crude protein in feed enhances microbial multiplication in the rumen, which in turn determines the extent of fermentation (Babayemi, 2007).

Gas production is positively related to microbial protein synthesis (Hillman *et al*., 1993). Therefore, a feedstuff containing degradable protein would also occasion high gas production due to the activities of the microbes. Energy supplement produced higher gas compared to protein supplement (Khazaal *et al.,* 1995). Gas production is a good parameter to predict digestibility, fermentation end product and microbial protein synthesis of the substrate by rumen microbes in the *in vitro* system. However, no particular trend was observed in the fermentation of soluble fraction "a", metabolizable energy (ME), organic matter digestibility (OMD) and short chain volatile fatty acid (SCFA), potential gas production (a+b) and fractional rate of gas production (c).

5.3.2 Methane gas production

Methane gas is an important gas among gases produced by ruminants at fermentation, and has been reported (Babayemi and Bamikole, 2006) to have negative effects on the animal. When methane gas accumulates in the rumen, it results in bloat and it is also an energy loss to the animals and when emitted it contributes to the destruction of the ozone layer. In most cases feedstuffs that show high capacity for gas production are also observed to be synonymous for high methane production. Methane production indicates an energy loss to the ruminant and tropical feedstuffs have been implicated to increase methanogenesis (Babayemi and Bamikole, 2006a). Methane production in the rumen is an energetically wasteful process, since the portion of the animal's feed, which is converted to CH4, is eructated as gas. Results from this study shows that the diets with 4% inclusion level of fossil shell flour had higher methane gas production than other treatments except at the 72 hour and this reflects its high gas production while the diets with 6% inclusion level of fossil shell flour had the lowest methane gas produced at the 96^{th} hour.

5.4 STUDY FIVE:

Performance of pregnant WAD ewes fed diets supplemented with varying levels of fossil shell flour

5.4.1 Weight changes (from mating to weaning) of WAD ewes

Ewes on 0% inclusion of fossil shell flour (T1) were heaviest at mating (33.00kg) while those on 2% inclusion level of fossil shell flour (T2) had the least weight (30.25kg) although no significant differences were observed. At parturition and weaning, ewes on 0% inclusion of fossil of fossil shell flour were heaviest (43.88kg and 42.79kg respectively). There was no significant correlation between live weight at mating and duration of pregnancy. This agrees with the findings of Uwechue, (2000) who found no significant correlation between live weight at breeding and length of pregnancy.

5.4.2 Weight gain during pregnancy

All the ewes gained weight during pregnancy showing that DM intake were sufficient for both maintenance and production. Ewes on 0% inclusion of fossil shell flour gained the highest (10.88kg) while ewes on 4% inclusion level of fossil shell flour gained the lowest (6.72kg). These weight changes were calculated as the difference between weight at parturition and weight at mating and show the increase in dam body weight due to pregnancy. Total weight gain during pregnancy (weight at mating minus maximum weight reached during pregnancy) was higher for ewes on 0% inclusion of fossil shell flour (15.68kg) than those on 6% inclusion level of fossil shell flour (11.50kg). This shows the weight loss during parturition. When compared with the body weight gain at pregnancy, ewes on 0% inclusion level of fossil shell flour with the highest weight gain during pregnancy (10.88kg) had a large weight loss during parturition (4.8kg) while ewes on 6% inclusion level of fossil shell flour with weight gain (8.13kg) lost 3.37kg during parturition.

5.4.3 Weight changes during lactation

This is the period between parturition and weaning of lambs. It lasted for 13 weeks in this study. All animals lost weight significantly ranging from -0.75kg (2% inclusion level of fossil shell flour) to -2.75kg (6% inclusion level of fossil shell flour). Ewes on 2% inclusion level of fossil shell flour with the highest number of twins (75%) lost the least (-0.75kg). The result of the weight changes during lactation observed in this study agrees with work done by other researchers stating that ewes generally lose weight during lactation or gain weight at a very low rate depending on their nutrition (Uwechue, 2000 and Okoli, 2003).

5.4.4 Types of birth

All parturitions took place unassisted. Most parturition took place in the morning hours except for three (one each from ewes on 0%, 4% and 6% inclusion levels of fossil shell flour), which took place in the evening. All foetal membranes were recovered and none were consumed by the ewes. In all the group the types of birth was either single or twins. The twin: single ratios for this study were: 50:50, 75:25, 25:75 and 25:75 respectively for ewes on 0%, 2%, 4% and 6% inclusion levels of fossil shell flour. Out of four ewes per treatment, two on 0% inclusion levels of fossil shell flour, three on 2% inclusion levels of fossil shell flour, one on 4% inclusion levels of fossil shell flour and one on 6% inclusion levels of fossil shell flour gave birth to twins. Values obtained in this study were still within the range reported by Uwechue, (2000).

5.4.5 Lamb sex ratio at birth.

The ratios of male: female lambs for ewes on 0%, 2%, 4% and 6% inclusion levels of fossil shell flour were 50:50, 29:71, 60:40 and 40:60 respectively. Ososanya *et al.*, (2007) revealed sex ratio of 48:52. However, in this study more female lambs were observed for ewes 2% inclusion levels of fossil shell flour. Further observations on larger number of ewes need to be done to ensure that the ratios obtained in this study are actually repeatable.

5.4.6 Lamb birth weight and rate of gain up to weaning

Birth weight of lamb is influenced by age, size, nutrition of the dam, gestation length, sex of the offspring and litter size. Under-nourishment during late pregnancy may cause pregnancy toxaemia, low birth weight of lambs and poor lamb survival. However under good nutrition and management, at least 80% of ewes mated should lamb with about 25% of the ewes producing twins (Ososanya *et al.,* 2007). No significant differences were observed for Lamb birth weight. Lower birth weights were observed for those on 2% inclusion levels of fossil shell flour with the highest number of twins. This agrees with work done by Opera *et al.* (2006) and Ososanya *et al.,* (2007) stating that type of birth affect birth weight. Birth weight is useful in predicting future performance of the lamb as it is related to vigour at birth and determine subsequent rate of gains. Lamb weight gain were: 6.68kg (0% inclusion levels of fossil shell flour), 6.89kg (2% inclusion levels of fossil shell flour), 6.97kg (4% inclusion levels of fossil shell flour) and 5.89kg (6% inclusion levels of fossil shell flour). However, lambs on 4% inclusion levels of fossil shell flour had the heaviest (9.23kg) lamb weaning weight but was not significantly different from those on 2% inclusion levels of fossil shell flour (9.05kg) with the lowest lamb birth weight (2.16kg) and the highest number of twins (75%).

5.4.7 Lamb mortality

Pre-weaning mortality is normally very low in housed, intensively managed well-fed sheep with good preventative medical care and high in grazing sheep especially during rainy season. This is due to a heavy parasitic burden, which affects the lambs directly, as well as indirectly by a reduction in milk production of the dam (Uwechue, 2000). Pre-weaning lamb mortality was only recorded for lambs on 0% inclusion levels of fossil shell flour (33.3%) and those on 6% inclusion levels of fossil shell flour (20.0%). One out of each set of twins on 0% and 6% inclusion levels of fossil shell flour died within five days of birth. This agrees with work done by Uwechue, (2000) and Opera *et al*., (2006) stating that twins die significantly more than singles and also with the work done by Okoli, (2003) who reported that about 21% lamb mortalities occurs during neonatal period (i.e between the first 7 days of birth). However, none of the set of twins on 2% inclusion levels of fossil shell flour died, this agrees with work done by Nuti *et al*., (2002) and Jean, (2004) stating that the inclusion of fossil shell flour eliminates parasites,

reduces stress and mortality in ruminant. No lamb mortality at birth and post-weaning were recorded in this study.

5.4.8 Average feed intake and weight changes of WAD ewes with advancing pregnancy

Feed intake of ewes during pregnancy decreased towards the end of the last trimester of pregnancy for all the treatment groups. This may be due to physical restriction placed on intake by advanced pregnancy. This is in agreement with the report of Uwechue, (2000) who stated that, pregnancy can reduce eating capacity but increase passage rate through the digestive tract. Robinson *et al.,* (1999) reported that since 80% of the foetal growth takes place in the last trimester of the pregnancy, nutritional requirements of the ewe increase to a great extent during this period. During the last six weeks of pregnancy energy requirements of twin bearing ewe are raised by more than 80 % (Robinson *et al.,* 1999) and protein requirements by 100% (Ólafsson, 1995). Ewes on 0% inclusion level of fossil shell flour had the highest daily weight gain (77.54g/day) while those on 4% inclusion level of fossil shell flour had the lowest (50g/day). In this study, the rate of gain was fastest in the last trimester for all the ewes across the treatments. This agrees with the work done by Uwechue, (2000) and Robinson *et al.,* (1999) and this is because 70% of the final foetal weight is estimated to be laid down at this time, since early pregnancy periods deal mainly with foetal organ differentiation.

CHAPTER SIX

6.0 SUMMARY, CONCLUSIONS AND RECOMMENDATION

6.1 Summary of Findings

Performance and nutrients digestibility of WAD sheep fed diets supplemented with fossil shell flour was evaluated. Daily weight gain was highest in rams on 4% inclusion level of fossil shell flour with the lowest Feed conversion ratio. The observed daily weight gain value might be as a result of the ability of the rams to properly utilize the diet for body weight gain when compared with other dietary treatments. The better performance characteristics observed in rams fed 4% fossil shell flour could be attributed to its inclusion in the diets which resulted in elimination of parasites, reducing physiological stress and improving feed assimilation. The inclusion of 4% fossil shell flour in the diets of WAD rams also improved the digestibility of dry matter, crude protein, crude fibre and ash.

The mean values of all blood parameters observed were within the normal range recommended for healthy sheep except for white blood cell for sheep on 0% inclusion of fossil shell flour. The higher values observed for white blood cell for sheep on 0% inclusion level of fossil shell flour compared to other treatments could be an indication that fossil shell flour is effective against elimination of parasites and their toxins. The inclusion of fossil shell flour in the diets of WAD sheep up to 6% has no negative effect on the haemathological and serum parameters observed in this study.

The high potential gas production observed in all the treatments is due to the highly fermentable nature of the diets. The diets with 2% and 4% inclusion levels of fossil shell flour depicted better degradability because of their higher gas production compared to other treatments this could be attributed to their high OMD (organic matter digestibility). The diets with 4% inclusion of fossil shell flour produced the highest methane gas this reflects its high gas production.

The performance of pregnant ewes was assessed in terms of weight changes during pregnancy, gestation length and weight loss during lactation, type of birth, lambs birth and weaning weights and mortality. All ewes gained weight during pregnancy with those on 0% inclusion of fossil shell flour having the highest weight. Dry matter intake was highest for pregnant ewes on 2%

inclusion of fossil shell flour and least for those on 4% inclusion of fossil shell flour. The inclusion of 2% fossil shell flour in the diets of pregnant WAD ewes improved dry matter intake. During lactation all the ewes lost weight across the treatments, however, inclusion of 2% of fossil shell flour in the diets of lactating ewes can help to reduce or mitigate the weight loss.

6.2 Conclusions

From the results obtained from this study it can be concluded that:

- The positive response in terms of improved weight gain and nutritive performance by the West African dwarf sheep indicated that the inclusion of 4% fossil shell flour will improve the growth performance of West African Dwarf sheep.
- The inclusion of fossil shell flour up to 6.0% in the diet of West African dwarf sheep has no negative effect on the blood parameters observed and therefore no detrimental effect on the health of the sheep.
- Inclusion of 2.0% fossil shell flour in the diets of West African dwarf sheep improved dry matter intake and reduced weight loss during lactation.
- The improvements observed in weight gain, feed conversion ratio, dry matter intake during pregnancy and weight loss reduction during lactation are paramount in improving the performance of our indigenous sheep (West African dwarf Sheep).

6.3 Recommendations

- For good accomplishment in the organic livestock farming system, the inclusion of 2-4% fossil shell flour in the diets of West African Dwarf sheep is recommended.
- Future study should be carried out on reproductive performance of West African Dwarf sheep fed diets supplemented with fossil shell flour to investigate and establish its effect on sperm production and quality and the repeatability of twinning rate.
- For improved performance of pregnant West African Dwarf ewe, the inclusion of 2% Fossil shell flour in their diets is recommended.
- Future study should be carried out to study the interactions and effect of fossil shell flour on rumen microbes

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APPENDIX

APPENDIX I: ANOVA TABLE FOR PACKED CELL VOLUME (%)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	208.1691687	69.3897229	2.46	0.1124
Error	12	337.8366750	28.1530562		
Corrected Total	15	546.0058437			

R-Square	Coeff Var	Root MSE	Mean
0.381258	15.13741	5.305945	35.05188

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APPENDIX II: ANOVA TABLE FOR RED BLOOD CELL (mm^3 $x10^6$)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	3	56.9213250	18.9737750	0.94	0.4517
Error	12	242.1778500	20.1814875		
Corrected Total	15	299.0991750			

R-Square	Coeff Var	Root MSE	Mean
0.190309	29.73856	4.492381	15.10625

APPENDIX III: ANOVA TABLE FOR WHITE BLOOD CELL (mm^3 $x10^3$)

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	85.0240750	28.3413583	2.28	0.1317
Error	12	149.3023000	12.4418583		
Corrected Total	15	234.3263750			

R-Square	Coeff Var	Root MSE	Mean
0.362845	32.10651	3.527302	10.98625

APPENDIX IV: ANOVA TABLE FOR HAEMOGLOBIN (g/dl)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	13.80860000	4.60286667	1.04	0.4102
Error	12	53.12770000	4.42730833		
Corrected Total	15	66.93630000			

R-Square	Coeff Var	Root MSE	Mean
0.206295	17.94939	2.104117	11.72250

APPENDIX V: ANOVA TABLE FOR MEAN CORPUSCULAR VALUE (μ^3)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	18.6909688	6.2303229	0.29	0.8328
Error	12	259.1079750	21.5923312		
Corrected Total	15	277.7989438			

R-Square	Coeff Var	Root MSE	Mean
0.067282	18.49593	4.646755	25.12313

APPENDIX VI: ANOVA TABLE FOR MEAN CORPUSCULAR HAEMOGLOBIN CONCENTRATION (%)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	2.50432500	0.83477500	0.72	0.5576
Error	12	13.86065000	1.15505417		
Corrected Total	15	16.36497500			

R-Square	Coeff Var	Root MSE	Mean
0.153030	3.259858	1.074734	32.96875

APPENDIX VII: ANOVA TABLE FOR TOTAL PROTEIN (g/dl)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	2.67231875	0.89077292	5.70	0.0116
Error	12	1.87627500	0.15635625		
Corrected Total	15	4.54859375			

R-Square	Coeff Var	Root MSE	Mean
0.587504	6.476308	0.395419	6.105625

APPENDIX VIII: ANOVA TABLE FOR INITIAL WEIGHT (Kg)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	0.15760000	0.05253333	0.15	0.9285
Error	12	4.24000000	0.35333333		
Corrected Total	15	4.39760000			

R-Square	Coeff Var	Root MSE	Mean
0.035838	4.575970	0.594418	12.99000

APPENDIX IX: ANOVA TABLE FOR FINAL WEIGHT (Kg)

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	0.87080000	0.29026667	1.44	0.2801
Error	12	2.42000000	0.20166667		
Corrected Total	15	3.29080000			

R-Square	Coeff Var	Root MSE	final Mean
0.264617	3.181531	0.449073	14.11500

APPENDIX X: ANOVA TABLE FOR DAILY WEIGHT GAIN (Kg)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	996.852300	332.284100	13.82	0.0003
Error	12	288.560000	24.046667		
Corrected Total	15	1285.412300			

R-Square	Coeff Var	Root MSE	Mean
0.775512	13.22387	4.903740	37.08250

APPENDIX XI: ANOVA TABLE FOR DAILY FEED INTAKE (Kg)

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	4067.431500	1355.810500	29053.1	<.0001
Error	12	0.560000	0.046667		
Corrected Total	15	4067.991500			

R-Square	Coeff Var	Root MSE	Mean
0.999862	0.052563	0.216025	410.9825

APPENDIX XII: ANOVA TABLE FOR FEED CONVERSION RATIO

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	46.1080000	15.3693333	3.29	0.0580
Error	12	56.0000000	4.6666667		
Corrected Total	15	102.1080000			

R-Square	Coeff Var	Root MSE	Mean
0.451561	19.62077	2.160247	11.01000

APPENDIX XIII: ANOVA TABLE FOR GESTATION LENGTH

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	48.5000000	16.1666667	1.40	0.2904
Error	12	138.5000000	11.5416667		
Corrected Total	15	187.0000000			

R-Square	Coeff Var	Root MSE	Mean
0.259358	2.261100	3.397303	150.2500

APPENDIX XIV: ANOVA TABLE FOR DAM WEIGHT AT MATING

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	17.2500000	5.7500000	0.07	0.9729
Error	12	934.5000000	77.8750000		
Corrected Total	15	951.7500000			

R-Square	Coeff Var	Root MSE	Mean
0.018125	28.12647	8.824681	31.37500

APPENDIX XV: ANOVA TABLE FOR WEIGHT GAIN DURING PREGNANCY

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	42.4218750	14.1406250	2.22	0.1380
Error	12	76.3125000	6.3593750		
Corrected Total	15	118.7343750			

R-Square	Coeff Var	Root MSE	Mean
0.357284	28.11741	2.521780	8.968750

APPENDIX XVI: ANOVA TABLE FOR WEIGHT BEFORE PATURITION

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	117.8750000	39.2916667	0.58	0.6371
Error	12	807.8750000	67.3229167		
Corrected Total	15	925.7500000			

R-Square	Coeff Var	Root MSE	Mean
0.127329	18.59502	8.205054	44.12500

APPENDIX XVII: ANOVA TABLE FOR WEIGHTAT PATURITION

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	76.0468750	25.3489583	0.41	0.7498
Error	12	744.8125000	62.0677083		
Corrected Total	15	820.8593750			

R-Square	Coeff Var	Root MSE	Mean
0.092643	19.52795	7.878306	40.34375

APPENDIX XVIII: ANOVA TABLE FOR DAM WEIGHT AT WEANING

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	111.9066687	37.3022229	0.61	0.6219
Error	12	735.1896750	61.2658063		
Corrected Total	15	847.0963437			

R-Square	Coeff Var	Root MSE	Mean
0.132106	20.16130	7.827248	38.82313

APPENDIX XIX: ANOVA TABLE FOR DAILY WEIGHT GAIN PREGNANT EWES

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	1888.819600	629.606533	134.92	<.0001
Error	12	56.000000	4.666667		
Corrected Total	15	1944.819600			

R-Square	Coeff Var	Root MSE	Mean
0.971206	3.611547	2.160247	59.81500

APPENDIX XX: ANOVA TABLE FOR FEED CONVERTION RATIO (FCR) OF PREGNANT EWES

The ANOVA Procedure

Dependent Variable:

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	120.7872750	40.2624250	8.63	0.0025
Error	12	56.0075000	4.6672917		
Corrected Total	15	176.7947750			

R-Square	Coeff Var	Root MSE	Mean
0.683206	14.28004	2.160392	15.12875

APPENDIX XXI: ANOVA TABLE FOR FEED INTAKE OF PREGNANT EWES

The ANOVA Procedure

Dependent Variable

Source	DF	Sum of Square	Mean Square	F Value	Pr > F
Model	3	25456.68750	8485.56250	2006.44	<.0001
Error	12	50.75000	4.22917		
Corrected Total	15	25507.43750			

R-Square	Coeff Var	Root MSE	Mean
0.998010	0.217690	2.056494	944.6875