UTILIZATION OF SOYMILK AS MILK REPLACER IN FEEDING FRIESIAN X BUNAJI CALVES

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DEPARTMENT OF ANIMAL SCIENCE,
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DECLARATION

I declare that the work in this Dissertation entitled "**Utilization of Soymilk as Milk Replacer in Feeding Friesian x Bunaji Calves"** was carried out by me in the Department of Animal Science, Ahmadu Bello University, Zaria, under the supervision of Dr. S.M. Yashim and Dr. S.B. Abdu. The information derived from the literature has been duly acknowledged in the text and a list of references provided. No part of this Dissertation was previously presented for another degree or diploma at this or any other Institution.

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CERTIFICATION

This Dissertation entitled **UTILIZATION OF SOYMILK AS MILK REPLACER IN FEEDING FRIESIAN X BUNAJI CALVES** by **Ishaya Usman GADZAMA** meets the regulations governing the award of the degree of Masters of Science (Animal Science) of the Ahmadu Bello University, and is approved for its contribution to knowledge and literary presentation.

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DEDICATION

This Dissertation is dedicated first to the ALMIGHTY GOD, Who has made everything possible, then to my late father, Mallam Usman Ali Tampul and my late sister, Mrs Panam Usman Gadzama, who contributed tremendously to my success in life. I cannot forget their labour of love. I missed you dear father and sister. Rest on till we meet to part no more.

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ABBREVIATIONS USED

ABU = Ahmadu Bello University

ADF = Acid Detergent Fibre

ADWG = Average Daily Weight Gain

ANFs = Anti-Nutritional Factors

ANOVA = Analysis of Variance

AOAC = Association of American Analytical Chemists

APP = Agricultural Promotion Policy

ATA = Agricultural Transformation Agenda

BC = Before Christ

BL = Body Length

BUN = Blood Urea Nitrogen

CBN = Central Bank of Nigeria

CF = Crude Fibre

cm = Centimetre

CP = Crude Protein

CRD = Completely Randomized Design

CSC = Cotton Seed Cake

DM = Dry Matter

DMI = Dry Matter Intake

DRP = Dairy Research Programme

EDTA = Ethylene-Di-aminne-Tetra-acetic Acid

EE = Ether Extract

EU = European Union

FAO = Food and Agricultural Organization

FCR = Feed Conversion Ratio

FCT = Federal Capital Territory

FNRP = Feed and Nutrition Research Programme

GLM = General Linear Model

Hemi = Hemicellulose

HG = Heart Girth

Hgb = Haemoglobin

hrs = Hour (s)

HW = Height at Withers

IAR = Institute for Agriculture Research

Ig = Immunoglobulins

IITA = International Institute for Tropical Agriculture

JP = Jerusalem Pilgrimage

Kcal = Kilo Calories

Kg = Kilogram

L = Litre (s)

MCH = Mean Corpuscular Haemoglobin

MCHC = Mean Corpuscular Haemoglobin Concentration

MCV = Mean Corpuscular Volume

ME = Metabolizable Energy

mg = Milligram

mls = millilitres

mmol/L = Millimoles per Litre

MR = Milk Replacer

₦ = Nigerian Naira

N = Normal/Nitrogen

NaCl = Sodium Chloride (Common Salt)

NAPRI = National Animal Production Research Institute

NDF = Neutral Detergent Fibre

NFE = Nitrogen Free Extract

NH_3-N = Ammonia-Nitrogen

NRC = National Research Council

^oC = Degree Centigrade

OM = Organic Matter

PCV = Packed Cell Volume

% = Percent/Percentage

pH = potention of Hydrogen

RAN = Rumen Ammonia Nitrogen

RBC = Red Blood Cells

SAS = Statistical Analysis System

SEM = Standard Error of the Mean

SNF = Solid-Not-Fat

SPC = Soy Protein Concentrate

TP = Total Protein

TS = Total Solid

TVFA = Total Volatile Fatty Acid

UK = United Kingdom

WBC = White Blood Cells

WM = Whole Milk

ABSTRACT

An experiment was conducted in two stages. The first stage was to investigate the effect of soaking duration on the proximate composition and anti-nutritional factors (ANFs) of soybean (*Glycine max* (L.) Merril), while the second stage was to evaluate the performance of Friesian x Bunaji calves fed different ratios of soy:cow milk. In stage one of the experiment, soybean was sourced, cleaned and divided into five groups (intended for the experimental treatments). The first group was the control (unsoaked soybean), while the other groups were soaked in clean water for 12, 24, 48 and 72 hours, respectively. The water was changed twice after every 24 hours of soaking. Thereafter, the soybean was rinsed, sun-dried for 8 days, milled, sieved and then taken to the laboratory for analyses. Results of the proximate composition showed that unsoaked soybean

(control) had 40.28% crude protein, 14.11% fat and metabolizable energy of 3459.50 kcal/kg DM. While soybean soaked for 72 hours had 44.37% CP, 29.55% fat and ME value 5514.57 kcal/kg DM. Soaking improved the chemical composition as well as reduced the levels of anti-nutritional factors in the soybean especially at 72 hours duration. The crude protein content of the different ratios of soy:cow milk diets increased with increase levels of soymilk and varies from 3.29% in 0:100 to 5.23% in the diet containing 75:25 ratio of soy:cow milk. The total solid, fat, solid-not-fat and ash contents also followed similar pattern. In the second experiment, a growth trial was carried out to evaluate the feeding value of the best soaking duration (72hrs) in combination with cow milk at a ratio of 0:100 (control), 25:75, 50:50 and 75:25 soy:cow milk, respectively in the diets of Friesian x Bunaji calves. Sixteen calves with average body weight of 34.8±0.7kg were randomly assigned to four dietary treatments with 4 calves per treatment in a Completely Randomized Design. Results from the growth trial showed that there was no significant difference ($P>0.05$) in average total feed intake among calves fed the different ratios of soy:cow milk. Calves fed diet containing 25:75 ratio of soy:cow milk had the lowest total feed intake (409.64 kg), average daily feed intake (4.18 kg/day) and better total weight gain (74.25 kg), average daily weight gain (0.76 kg/day) and feed conversion ratio (5.52) as against (416.29 kg), (4.25 kg/day), (66.00 kg), (0.67 kg/day) and (6.31), respectively in calves fed cow milk alone (control).

Inclusion of soymilk at different ratios had positive influence on some linear body measurements (body length, height at withers and heart girth) of calves. The blood parameters measured were within the normal range for healthy calves, indicating that soaking of soybean in water for 72 hours had significantly reduced the levels of ANFs in the soymilk fed to calves. White blood cells and glucose were within the normal range ($4 – 12x10^9$/l) and (40 – 100mg/dl), respectively. Feeding different ratios of soy:cow milk had positive influence on rumen metabolites (rumen fluid temperature, pH, NH_3-N and TVFA) with 75:25 ratio of soy:cow milk raton having the highest concentration of RAN (13.84 mg/100ml). Sampling time significantly ($P<0.05$) influenced rumen temperature, pH, NH_3-N and TVFA concentrations. Digestibility of DM, OM, CP, NDF and ADF

were significantly (P<0.05) influenced by the inclusion of soymilk at different ratios. However, calves fed 25:75 and 75:25 ratios of soy:cow milk had better nutrient digestibility and nitrogen balance, respectively. The cost of cow milk per litre (₦98.00) was about 3 times higher than soymilk (₦33). Calves fed 25 and 50% soymilk had better feed cost per kg weight gain ₦496.10 and ₦469.67, respectively. Calves fed 75:25 ratio of soy:cow milk had the least total cost of feed consumed (₦ 30489.00). Indicating that feeding cow milk alone to calves was very expensive compared with feeding soy:cow milk at 75:25 ratio. It was concluded that soaking of soybean in water for 72 hours reduced the levels of ANFs and improved the nutritive value. Feeding 25:75 soy:cow milk improved live weight gain by 0.76 kg/day as against 0.67kg/day in calves fed cow milk alone. From an economic standpoint, feeding soy:cow milk at a ratio of 75:25 reduced the cost of feeding by (24.16%) as compared with feeding cow milk alone. Therefore, soaking of soybean in water for 72 hours was recommended for good result. And feeding calves with 25:75 ratio of soy:cow milk is recommended for better performance and feed conversion than feeding cow milk alone.

CHAPTER ONE

1.0 INTRODUCTION

Calves are the future producers of cow milk for human consumption, but depend solely on milk for their nutritional needs at the early stage of life. Milk is a unique food for calves and supplies a lot of nutrients essential for growth and organ development (Ghorbani *et al*., 2007). Post-natal feeding of dairy calves is very important for better health and growth especially in commercial as well as smallholder dairy farms (Khan *et al*., 2012). However, the indigenous breeds of cattle in Nigeria are naturally poor milk producers, as they do not produce enough milk for optimum sustainability of their calves talk less of having surplus for human consumption. This results in underfeeding or starvation of calves with a consequence of stunted growth and mortality. Shortage of nutrients especially during the early stages of life in calves reflects later in their whole life in terms of productive and reproductive performances (Roy *et al*., 2016). If suitable substitutes for milk are made available, the nutrition of infant pre-ruminants can be improved and survivability can be increased (Khan *et al*., 2012). The benefits of improved nutritional status of calves in the first 2 – 3 months of life may include reaching maturity age sooner, improved ability to withstand infectious challenges and increased subsequent milk production (Drackley, 1999).

Milk replacers (MR) are very good sources of liquid feed for calves (Roy *et al*., 2016). Khan *et al*. (2012) defined milk replacers as those feed ingredients or a mixture of such ingredients that can be used as substitute for whole milk in the diets of calves. They are made from by-products of milk with the addition of some other ingredients in such a way that the final product is comparable to whole milk. A typical MR comprises of skim milk powder, whey powder and vegetable oils. Crude protein content is usually 20%, and the fat content typically ranges from 15 to 22% (DM basis) (Mete *et al*., 2000).

In developed countries, alternatives to whole milk feeding to pre-ruminants are formulated using by-products of milk processing industry (Oliveira *et al*., 2015). Such practice is not feasible in a developing country like Nigeria where the demand for milk and milk products in human nutrition is constantly increasing as a result of the rapid increase in human population (ATA, 2013). The aim of using milk replacer is to cut down the cost of whole milk in calf rearing programme and to improve their performance (Mete *et al*., 2000). There are many types of low cost, high quality plant protein viz, soyflour, soymilk, soy protein concentrate and wheat protein which could be used in the formulation of milk replacers (Ghorbani *et al*., 2007). However, Roy *et al*. (2016) showed that soy protein is widely used in milk replacer formulations because of its high-quality protein content. Soybean contains up to 40% crude protein compared with 1.0 to 5.6% protein content of most animal milk (Ghorbani *et al*., 2007) and has an acceptable amino acid profile (Gernah *et al*., 2013). Soymilk not only provides protein but is also a good source of carbohydrate, fat, vitamins and minerals (Nitsan *et al*., 2005).

Soymilk, a novel milk replacer has been used for artificial rearing of young animals (Ghorbani *et al*., 2007). Few studies indicated that feeding soymilk as milk replacer to calves resulted to increased growth performance when compared to whole milk feeding (Bartlett *et al*., 2006; Ghorbani *et al*., 2007; Masum *et al*., 2009; Roy *et al*., 2016). Despite soybean's pivotal role in animal nutrition, it cannot be fed unprocessed because there are some anti-nutritional factors (ANFs) which limits its utilization. These ANFs include: protease inhibitors, tannins, phytates, oxalates, saponins, etc. Fortunately, the levels of these ANFs can be eliminated or reduced through soaking, cooking, sprouting, fermentation, toasting, etc (Soetan and Oyewole, 2009). In additon, soaking could reduce ANFs such as protease enzyme inhibitor, phytates, etc due to their partial or total solubilization and removal with the discarded solution (Prodanov *et al*., 2004). However, in calves with completely functioning rumens, ANFs and complex proteins are not as detrimental, and thus processing is not as crucial (Liener, 1994).

1.1 Justification of the Study

Milk replacer is an ideal liquid feed for pre-ruminants. Commercial milk replacers are scarce and expensive because they are mostly imported. Soybean is readily available, less expensive and can be cultivated locally. Soybean has potential for use as milk replacer formulations because of its high nutritive value. Soybean is a good source of high quality, relatively inexpensive protein. Soybean protein is often used in calf milk replacers and starters instead of milk proteins because of its low cost and its essential amino acid content which is similar to cow's milk. Soymilk is cheaper than cow milk therefore, successful replacement of cow milk with soymilk may reduce feed cost. However, to ensure proper growth and health of the calf, processing and subsequent solubility of the protein source are crucially important for determining the suitability of alternative proteins in milk replacers. Soaking as a processing method could be employed to improve the feeding value of soymilk. Successful replacement of cow milk with processed soymilk could also improve the nutrition and survivability of calves. The technology can easily be adopted by farmers to enhance the survival of the calves and have excess milk for sale. The use of soymilk as an alternative protein source in milk replacers is therefore a very vital area to explore in calf nutritional research.

1.2 Objectives of the Study

The broad objective of the study was to investigate the potentials of using soymilk as a replacement for cow milk in the diets of dairy calves. While the specific objectives were to evaluate:

1. The effect of soaking duration on the nutritional and antinutritional composition of soybean (milk quality of soybean).
2. The performance of Friesian x Bunaji calves fed different ratios of soymilk and cow milk.
3. The economic advantage of feeding different ratios of soy:cow milk in Friesian x Bunaji calves.

1.3 Research Questions

This study was carried out on the following research questions:

Research Question 1: Does soaking durations have any effect on nutritional and anti-nutritional contents of soybean?

Research Question 2: Does feeding different ratios of soy:cow milk have any effect on performance of Friesian x Bunaji calves?

Research Question 3: Does feeding different ratios of soy:cow milk have any economic advantage?

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Livestock Production in Nigeria

Nigeria is endowed with substantial animal resources, these include: 19.5 million cattle, 72.5 million goats, 41.3 million sheep, 157.8 million poultry, 7.1 million pigs, 974,499 donkeys, and 28,000 camels (APP, 2016). Livestock is an integral component of most of the agricultural activities and production systems prevalent in the country. Livestock plays a central role in sustaining millions of resource-poor farmers in different agroecological environments, ranging from pastoral and agro-pastoral to mixed farming systems that entirely depend on draft animals for the cultivation of cereal crops (Eromosele, 2014). Livestock products supply animal protein that improves the nutritional status of human beings. Livestock are used for various purposes including milk and meat production, draught power and manure utilization. They have economic security and social status functions as well. Agriculture contributes about 28.65% to Nigeria's Gross Domestic Product (GDP) (NBS, 2017), while the livestock sub-sector contributes only about 5.1% making Nigeria a net importer of protein food of animal origin (Annette *et al*., 2012). Cattle are the most predominant and highly valued livestock in Nigeria. They are kept for milk, beef, hide, traction and for many as a status symbol (APP, 2016).

2.2 Dairy Development in Nigeria

Milk is one of the most frequently used cattle product, however, many families cannot get enough milk for daily nourishment. Although traditional dairying in Nigeria starts centuries ago, industrial dairying is recent (Eromosele, 2014). Reports indicate that Nigeria has the potential of being a major milk producer in Africa (Annette *et al*., 2012). According to FAO (1988) report, the dairy industry provides a means of livelihood for a significant proportion of rural pastoral families in the sub-humid and semi-arid ecological zones of Nigeria.

For instance, about 183,000 rural households were said to have derived some income from the dairy industry in 1986 (Foraminifera, 2012). The ability of the dairy enterprise to generate regular income and to contribute to the household diet on a regular basis throughout the year is an advantage over other agricultural enterprises (Dairy Chain, 2014).

The various activities of the Nigerian dairy industry (viz: milk production, importation, processing, marketing and consumption) have been going on in the country for over 100 years. These activities are however, largely unorganized except for the relatively few processing firms that produce and market reconstituted milk products from imported powdered milk (Annette *et al*., 2012). Presently, the local production of milk is less than 1% of the total annual demand that has been estimated at 1.45 billion litres, making the total milk consumption in Nigeria less than 10 litres per head per day whereas the global average is about 40 litres per head per day (Yatai, 2013). In other parts of Africa, it is 28 litres per head per day. Nigeria spends about $1.3 billion on importation of milk annually. More so, with human population placed at over 187,916,768 and with an annual growth rate of 2.5% (NBS, 2017), there is a significant threat to the agricultural potentials of the country in terms of affording sustainable animal protein supply, especially as these animals are poorly managed (in terms of feed availability and disease), have poor genetic capacities and are rapidly being depleted (APP, 2016).

2.3 Dairy Production in Nigeria

Nigeria is a potential market for 1.3 million tons of milk valued at about ₦ 450 billion annually (Dairy Chain, 2014). The estimated total domestic fluid milk production in 2006 for example, is only about 600,000 litres (worth about ₦232.5 million) which entered formal marketing channels through corporate, public and other private milk collection co-operatives schemes from migrant herdsmen and the output of the few commercial dairy farms (Levin and Nzeka, 2009). The rest was either consumed by the producing families or traded informally within the producing communities. Imports of milk powder and other processed dairy products were estimated at $275 million in 2006 (Dairy Chain, 2014).

Average yield from this traditional system is 0.7 – 1.5 litres of fluid milk per day. During the dry season, this figure drops to about 0.5 litres. With supplementation using cotton seed cake, milk yield could be increased to between 1.5 and 2.0 litres (Annette *et al*., 2012). The "White Fulani" or "Bunaji" breeds are the dominant dairy breed in Nigeria (Malau Aduli, 1992), and the pastoralists own and maintain majority of them. These animals are mostly grazed on natural grass and legumes under the traditional system with little supplementary feeding regimes in some places. Migrant pastoralist move flocks over months and many miles to find pasture during the dry season, which often results in weight loss, low yields and sickness (Annette *et al*., 2012). A few commercial dairy farms maintain crossbreeds of Holstein Friesians, Brown Swiss and Montpellier for fluid milk production and the average yield is 8 to 18 litres of raw fluid milk per day (compared with the national average of only 4 litres). In temperate environment, the average yield of pure breeds under intensive management conditions is about 30 litres per head per day (Oni *et al*., 2001). Currently, most of Nigeria's dairy processors import milk powder and re-constitute it into liquid milk and other dairy products such as yoghurt, ice cream and confectioneries. Others repackage imported powdered milk into small affordable sachets (Foraminifera, 2012).

The imported dairy products (mostly milk powder) come from New Zealand, Australia, South America, the EU, India, Ukraine, Poland, and other smaller suppliers (Dairy Chain, 2014). Multi-national firms including Frieslandfoods (Netherlands), Glanbia (Ireland), Cussons-PZ (UK), Promasidor, etc; have either partnered or acquired some Nigerian dairy firms for re-constituting and/or packaging imported milk powder (Foraminifera, 2012). According to the Central Bank of Nigeria report of 2010, the increasing consumption trends have cost the government a substantial amount of foreign exchange to import dairy products into Nigeria (CBN, 1999).

2.4 Historical Background of Soybean

Historians believe that soybean (*Glycine max* (L.) Merrill) is one of the oldest crops grown by human beings (Liu, 1997). The botanical name of soybean is *Glycine hispida* or more commonly called *Glycine max* which was the genetic name given to it by Carl von Linne (Mattingly and Bird, 1945). Soybean is widely believed to have originated 4000 – 5000 years ago from Manchuria, a province (Northern and Central region) of China, with the first documented use of the plant by a Chinese emperor in 2838 B.C. (Liu, 1997). Soybean cultivation reached Africa in the late 1800s; although little is known of the countries to which it was first introduced (Macrae *et al*., 1993. The major soybean producing countries in the world are the United States, Brazil, China, Nigeria, India, Argentina, South Africa and Uganda (IITA, 2009).

2.4.1 Description of Soybean and its Origin in Nigeria

Soybean (*Glycine max* L.) is an annual herbaceous legume plant of the pea family Leguminosae and subfamily Papilionnidea (IITA, 2009). It is an important global legume that grows in the tropical, subtropical and temperate climates; like peas, beans, lentils and peanuts (Iwe, 2003). Soybean was first introduced to Nigeria in 1908. Later, introduction of the crop to the savanna ecology (IITA, 2009) in 1928 proved successful. It then spread into other parts of northern Nigeria, and soon became a cash crop in the Tiv division of Benue Province (now Benue State), which thereafter became the leading production centre (Iwe, 2003).

2.4.2 Soybean Production in Nigeria

According to a survey reported by IITA (2003), Benue State is the major producer of soybean in Nigeria. The current expansion in the production of soybean in Nigeria has been attributed to many years of research from the mid-1960s through the 1980s when Scientists adopted a nationally-coordinated approach to Soybean research (IITA, 2003).

Nigeria is the largest producer of the crop for human and livestock feeds in West and Central Africa (IITA, 2009). At present, the major soybean producing states in the country are Benue, Kaduna, Taraba, Plateau and Niger. Other growing areas include, Nasarawa, Kebbi, Kwara, Oyo, Jigawa, Borno, Bauchi, Lagos, Sokoto, Zamfara and FCT (IITA, 2009).

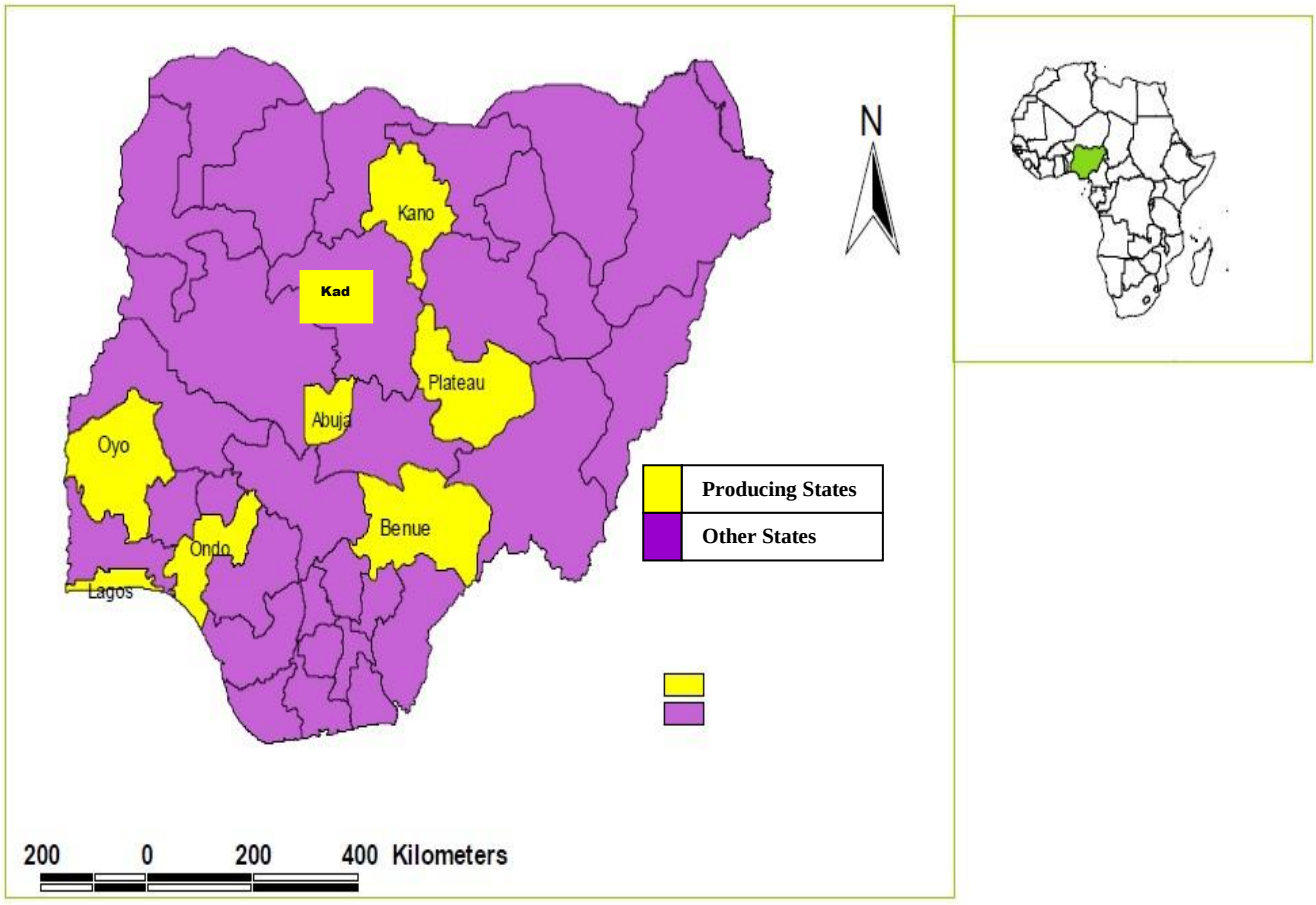


Figure 2.1: Map of Nigeria showing eight leading soybean producing states.
Source: IITA (2009).

2.4.3 Importance of Soybean

Among the legumes, soybean is an important crop due to its nutritional value and its wide utilization at household as well as at industrial levels (Perkins, 1995). The crop has a unique chemical composition on an average dry matter basis; it contains about 40% protein and 20% oil. With this, it ranks highest in terms of protein content (both in quality and quantity), among all food crops and second highest oil content among all food legumes.

The oil is highest in terms of quality. It contains a high proportion of unsaturated fatty acids such as linolenic and lineic acids hence; it is a healthful oil (Iwe, 2003). Soy protein contains all the essential amino acids most of which are present in amounts that closely match those required from humans or animals. For example, 1kg of soybean contains as much protein as, 2kg of boneless meat or 45 cups of cow milk or 5 dozens of eggs (Perkins, 1995). It has a protein-digestibility corrected amino acid score, very close to 1, the highest rating possible and the same rating for animal proteins such as egg white and casein (Liu, 2000). Soybean is widely known to be cheap, easily available and a good source of rich and cheap protein compared to animal protein and other nutrients (Perkins, 1995).

2.4.4 Utilization of Soybean by Animals

Due to its high protein content and good nutritional value, soybean was primarily used as animal feed. Soybean have been a significant source of plant proteins for livestock feed industries for many years (Dari, 2009). Its universal acceptability in animal feed is as a result of important attributes such as relatively high protein content and suitable amino acid profile except methionine, minimal variation in nutrient content, ready availability year-round, and relative freedom from intractable antinutritive factors if properly processed and its relatively low cost (FAO, 2004). Therefore, its production and consumption will continue to grow as a preferred source of alternate high quality protein in animal diets (IITA, 2003).

2.4.5 Anti-nutritional Factors in Soybean

Full fat soybean contain anti-nutritional factors that reduce the digestibility, bio availability of nutrients and utilization of amino acids in monogastric and immature ruminants (Maidala *et al.,* 2013). This is the reason why animals fed on unprocessed soybean do not grow properly. Some of the ANFs in soybean include: protease inhibitors, tannins, phytates, oxalates, saponins, etc (Soetan and Oyewole, 2009).

2.4.5.1 Protease Inhibitors

Protease inhibitors represent 6% of the protein present in soybean seed. They prevent the proper functioning of the enzymes responsible for digesting protein in the diet (Grieshop *et al*., 2001). Protease inhibitors can block either trypsin or chymotrypsin, reduce the hydrolysis of dietary protein, decrease amino acid absorption and thereby reduce digestibility (Roy *et al*., 2010). Large amounts prevent proper digestion of soy; however, processed soy foods contain only 5% or less of the naturally occurring protease inhibitors (Joshi *et al.*, 1989). Protease inhibitors, which are ANFs for monogastric animals, do not exert adverse effects in ruminants because they are degraded in the rumen (Grieshop *et al*., 2001).

2.4.5.2 Tannins

Tannins are mostly water soluble phenolic compounds with a molecular weight greater than 500 and with the ability to precipitate proteins from aqueous solution. They occur almost in all vascular plants. They prevent bloating (Lalles, 1993) and more importantly reduce degradation of protein in the rumen, thus making available to the hind gut intact plant protein (Grieshop *et al*., 2001). Consequently, such feeds with moderate content of tannins may be good sources for enhancing high nitrogen retention (Lalles, 1993), growth rate, as well as growth hormone (Liener, 1994). Feeds with more than 5% condensed tannins cause low intake, reduced digestibility and cause poor growth in ruminants (Roy *et al*., 2010). Tannins or their oxidized products can form complexes with amino acid, protein, enzymes which adversely affect their digestibility (Khandelwal *et al*., 2010). In ruminants, dietary condensed tannins (2 – 3%) have been shown to impart beneficial effects because they reduce the wasteful protein degradation in the rumen by the formation of a protein-tannin complex (Ramakrishna *et al*., 2006).

2.4.5.3 Phytates

They are also considered as antinutrients due to the fact that they bind iron and calcium preventing their availability thus, reducing their proper absorption (Ramakrishna *et al*., 2006). A lot of phytates in diets decrease availability of calcium, phosphorus and zinc. The phytates decrease also activity of enzymes (pepsin, trypsin and amylase) as well as availability of protein, amino acids, starch and energy (Liener, 1994).

2.4.5.4 Oxalates

Oxalate is considered an anti-nutrient because it inhibits calcium absorption. Oxalate like phytates bind minerals like calcium and magnesium and interfere with their metabolism (Soetan and Oyewole, 2009). Dietary oxalate can be degraded by rumen microbes into CO_2 and formic acid. Ruminants adapted to diets with high oxalate content can tolerate oxalate levels that are lethal to non-adapted animals (Bamualin *et al.,* 1980). Ruminants, unlike monogastric animals, can consume considerable amounts of high oxalate feeds without adverse effects, due principally to microbial decomposition in the rumen (Joshi *et al.*, 1989).

2.4.5.5 Saponins

Saponins are characterised by a bitter taste and foaming properties. Saponins retard growth rate in non-ruminants, due primarily to reduction in feed intake (Bamualin *et al.*, 1980). In ruminants, saponins may undergo bacterial degradation in the rumen, and may not retard the growth. Nevertheless, recent studies indicate that they inhibit microbial fermentation and synthesis in the rumen (Liener, 1994). The adverse effects of saponins can be overcome by repeated washing (soaking) in water which makes the feed more palatable by reducing the bitterness associated with saponins (Joshi *et al.*, 1989).

2.4.6 Methods of Alleviating the ANFs in Soybean

Increasing the nutritional quality of soybean and other legumes can be accomplished by several processing methods such as soaking, toasting, boiling, salt treatment, fermentation, germination, pressure cooking, cooking, urea treatment, etc (Akande and Fabiyi, 2010). The methods of processing the seeds to eliminate antinutritional factors have been a major challenge to most farmers (Okagbare and Akpodiete, 2006). In calves with completely functioning rumen, antinutritional factors and complex proteins are not as detrimental, and thus processing is not as crucial (Liener, 1994). Several studies have shown that, soaking and sprouting improves the nutritive value of legumes (Soetan and Oyewole, 2009; Pele *et al*., 2016). Preparation of many indigenous foods from grains involves soaking in water, which is ultimately a household art (Liener, 1994). In additon, soaking could reduce ANFs such as protease enzyme inhibitor, phytates, certain minerals and α-galactosides due to their partial or total solubilization and removal with the discarded solution (Prodanov *et al*., 2004). It is against this background that this research work attempt to evaluate the effect of soaking duration as a local processing method on proximate composition and anti-nutritional factors of soybean.

2.4.6.1 Soaking as a Processing Method

Soaking is a unit operation that involves the immersion of materials (seeds, leaves and nuts) in water for the purpose of sprouting, fermentation or softening of seeds. Soaking is a common local processing method in Africa that has been reported to influence the properties of legume seeds (Roy *et al*., 2010). It is one of the processing methods used to improve the nutritional value of soybean. During soaking, seeds absorb water thereby making the feed material to become soft, extensible, flowable, loss of crispness, hardness or toughness and fermentation of starch components occurs simultaneously (Liener, 1985). It also leads to the breakdown of several components into simpler compounds with alteration in texture, flavour, aroma and taste (Liener, 1994).

Soaking could be one of the processes to remove soluble anti-nutritional factors, which can be eliminated with the discarded solution. However, some metabolic reactions can take place during soaking which will affect some of the constituent compounds (Grieshop *et al*., 2001). Pele *et al*. (2016) investigated the effect of traditional processing methods on the nutritional and anti-nutritional properties of soybean. Soybean was subjected to different processing methods which include: sun drying and milling, soaking for 24 hours, sun drying and milling, soaking for 12 hours, de-hulling, sun drying and milling and sprouting for 120 hours, sun drying and milling (Pele *et al*., 2016). The research revealed that these processing methods significantly improved the nutritional quality and reduced the anti-nutritional properties of soybean. Also, the study showed that protein content of soybean was significantly increased while the fat content significantly reduced due to soaking, de-hulling and sprouting. Similarly, the phytic acid which hinders minerals absorption was also significantly reduced due to sprouting. And the fat content of soybean which could have a negative effect on rancidity was significantly reduced due to soaking, de-hulling and sprouting (Pele *et al*., 2016).

2.4.6.2 Alleviation of ANFs by Rumen Microbial Activity

Ruminant animals have a symbiotic relationship with rumen microorganisms. The rumen environment (slightly acidic) provides many reductive and hydrolytic reactions which, in the majority of cases, decrease the biological activity of allelochemicals before absorption from the tract (Bridges, 2005). Anaerobic degradation of flavonoid and hydrolysable tannins by mixed rumen microbes has also been demonstrated. Such rumen microbes are present in small numbers and their growth rate is slow.

Anaerobic microbial degradation of condensed tannins has also been demonstrated. Dietary oxalate can be degraded by rumen microbes into CO_2 and formic acid. Ruminants adapted to diets with high oxalate content can tolerate oxalate levels that are lethal to non-adapted animals (Bridges, 2005).

2.4.7 How to Mix and Administer Calf Milk Replacer

Costello (2012) reported that, the water used to mix milk replacer should always be below 45°C otherwise the proteins which are essential to calf performance will be destroyed. He added that cow milk should be fed between a temperature of 37 – 39 ^{o}C to stimulate a strong oesophageal groove reflex, which will help to prevent milk entering the rumen. The spillage of milk into the rumen will increase the risk of scours and result in poor growth of calf. The author also suggested that to mix MR correctly, digital weighing scale should be used to weigh the MR powder accurately. To mix MR with a total solid concentration of 12.5%, 125g of MR powder should be dissolved in 875ml water to make up 1 litre mixture. Using a full litre of water will lead to a weaker (11.1% total solids) concentration in the milk replacer. Half of the water should used at a tempereture below 45°C. All the MR powder can be added, whisked until smooth, and the the remaining water can be added and then whisk again (Costello, 2012). The temperature of the mixture should be kept between 37 – 39 °C and then feed according to live body weight of the animal (usually 10% of body weight is used for better performance of calf).

2.4.8 How to Mix and Administer Milk Replacer Containing Soy Protein

In an experiment to investigate the utilization of soymilk as milk replacer for raising calves, Masum *et al*. (2009) reported that, soymilk was prepared from large size, thin hull and not more than 1 year old whole soybean. One kg whole soybean was ground in a soy flour mill and 125g powder was dissolved in 1000ml of water and strained with fine muslin cloth. Thereafter, the soymilk was boiled at 100°C for 15 minutes with constant stirring. After proper cooling, the authors reported that the soymilk was supplied to calves. On the other hand, another batch of soymilk was prepared in the same way and offered to another group of calves without straining. Masum *et al*. (2009) also reported that whole milk and soymilk were administered to calves two times daily, one litre in the morning and one litre in the afternoon.

In another experiment, Sarker *et al*. (2015) determine the effect of soymilk as milk replacer on growth, carcass characteristics, rumen development, ovarian morphology and testicular development in kids. The soymilk was prepared from yellow soybean, which were ground to produce soy flour. An amount of 125g of soyflour was dissolved in 1 litre of water by stirring. The mixture was then boiled at 100 ^{o}C for 10 – 15 minutes with constant stirring. After proper cooling, the authors reported that the soymilk was mixed with whole milk and was supplied to the kids. The first treatment which comprised of 8 kids, were reared on 100% whole milk, the second on 75% whole milk + 25% soymilk, and the third on 50% whole milk + 50% soymilk. Pandiyan *et al*. (2012) conducted an experiment to evaluate the effects of soymilk and ewe milk blend on the growth performance of suckling Mecheri lambs. A total of 250g of soybean was roasted until the beans get brown in colour. The roasted soybean was cooled under room temperature and soaked overnight in water containing 0.3% sodium bicarbonate solution. The soybean was washed with water 2 – 3 times and blanched in boiling water for 45 – 60 minutes by gently stirring. There after, the beans were manually dehulled and 2kg of luke warm water was added and ground well. The mixture was boiled for 2 minutes and filtered through a muslin cloth to remove the solid particles. Finally, the authors reported that the soymilk was pooled together with ewe milk and thoroughly mixed. The lambs on treatment one were fed exclusively on ewe's milk (100%), while those on treatments 2 and 3 received 50:50% and 75:25% soy:ewe milk.

2.5 Feeds for Calves

In the pre-weaning period, the digestive tract of the calf is not fully developed and most of the feed is fed as liquid. Liquid feeds are the primary nutrient source for calves in the first weeks of life, and they by-pass the reticulum and rumen via closure of the oesophageal groove (Heinrichs and Lesmeister, 2005).

The formation of the esophageal groove sends liquid feeds directly into the stomach compartment that will digest them best – the omasum followed quickly by the abomasums (Heinrichs, 2005). Apart from drinking water, the three types of liquids used are colostrums, whole milk and milk replacers.

2.5.1 Colostrum

The milk produced in the first few days after parturition is called colostrum. The composition of colostrum differs from whole milk, especially with the higher content of protein. The high protein content of colostrum is mainly due to immunoglobulins (Ig) (Nocek *et al*., 1984) that are crucial for the passive immune system of the newborn calf. Because of limited (or no) transfer of Ig over the placenta, the calf is born with a very low immune status and fully dependent on a passive transfer of Ig from the colostrum (Stott *et al*., 1981). The first hours after birth are critical because the intestine of the calf gradually becomes less permeable for the large Ig molecules (Quigley *et al*., 2006). In practice, the content of Ig in milk decreases gradually after the first milking (Stott *et al*., 1981). There are three types of Ig's; Ig-G (80-90%), Ig-A (5%) and Ig-E (7%) (Larson *et al*., 1980). Although the Ig-G is the quantitatively most important in ruminants, all the types need to be present to obtain an optimal immunity (Logan *et al*., 1974).

2.5.2 Whole Milk

Whole milk (cow milk) is the natural liquid feed for calves, but it is also the most expensive liquid feeding. Heinrichs and Lesmeister (2005) reported that whole milk was the main dairy calves liquid feed before 1956. Whole milk (WM) is highly digestible and considered an ideal feed for the newborn calf as it matches the required levels and types of nutrients in early growth and development in life (NRC, 2001). Whole milk consists on average of 26% protein, 30% fat and 38% lactose on dry matter (DM) basis. Excess WM is considered a cheap feed compared to milk replacers.

High performance farms in Norway use excess WM and unsaleable milk, like milk with elevated cell count and transition milk, in their calf rearing when available (Bekkevoll and Helberg, 2009). In general, the use of such milk have not caused any elevated scouring or long term negative effects in first lactation heifers (Kesler, 1981). However, use of discarded milk from cows on antibiotic treatment is not recommended due to the possible resistance to certain bacteria (Kesler, 1981).

2.5.3 Effect of Feeding Whole Milk on Calf Performance

The appropriate and ample supply of milk to calf is important for the animal's performance. During the last several decades, several scientists have studied the common practice of providing limited amount of milk to get better performance of calves (Khan *et al*., 2007). Their studies showed that restricted milk decreased weight gain, increased the risk of disease and abnormal behaviour in calves whereas the study by Jasper and Weary (2002) reported that greater quantity of milk improves growth rate and feed efficiency. Drackley (2008) reviewed that milk whole milk feeding accelerated growth, enhanced nutrition and growth in young calves. The research reported by (Hill *et al*., 2008) claimed that the calves fed about 8 kg of milk daily gained 0.9kg of body weight per day. The utilization of milk-based diets improves digestion and assimilation of nutrients compared to dry diets in calves due to higher digestibility and nutrient profile in ingredients of milk origin in the milk replacer (Davis and Drackley, 1998; Heinrichs, 2005). Poor growth was due to insufficient supply of nutrients from milk and sudden change can cause scour and reduced growth rate, which ultimately increased the weaning time (Hill *et al*., 2008).

2.5.4 Milk Replacers

Milk replacers (MR) were first manufactured in the 1950's (Davis and Drackley, 1998) and became available commercially because the price of milk was too high and was expensive to feed whole milk to calves raised on the farm (Bridges, 2005). Milk replacer is a form of liquid feed that is incorporated into the diet of young, growing animals.

If used properly, MR is an excellent substitute for whole milk (Davis and Drackley, 1998). Bridges (2005) defined MR as a liquid feed fed to young animals prior to weaning. Milk replacers are usually made up of ingredients such as skim milk powder (60 – 75%), vegetable or animal fat (15 – 25%), butter milk powder, whey protein (5 – 10%), soy lecithin (1 – 2%) and vitamin-mineral premix. A small proportion of other ingredients like glucose, non-milk protein and cereal flour can also be used (Ghorban *et al*., 2007). Many dairy calves are fed milk replacers prior to weaning to reduce costs for the dairy producer and make whole milk available for sale (Davis and Drackley, 1998).

Calves may be started on a milk replacer when they are about 4 – 6 days old, but the switch from whole milk to milk replacer should be gradual. Abrupt changes will increase the likelihood of nutritional scours and stress (Oliveira *et al*., 2015). Initially, milk replacers were manufactured with gruel ingredients (linseed meal, wheat middlings), but the use of spray-dried milk, skim milk and dried whey, together with improvements on fat homogenization at manufacturing resulted in high quality products (NRC, 2001). There are two main protein sources in milk replacer: milk (dried skim milk, whey protein concentrate, dried whey, sodium caseinate) or vegetable (soy protein, wheat protein, potato protein). Vegetable sources are normally used to reduce MR costs. Milk replacers can be manufactured with a variety of ingredients and levels of nutrients to match management requirements of a variety of farms. Additives, including vitamins, antibiotics, coccidiostats, or ionophores, can be supplied in the milk replacer. Typically, these cannot easily be used in whole milk or waste milk feeding systems (NRC, 2001). Calf performance and digestible amino acid absorption can be improved by the inclusion of Thrionine, Methionine and Lysine in MR containing soy protein (NRC, 2001). More recently, high quality animals' proteins include red blood cell protein and plasma proteins have been used in some MR formulations (Jaster *et al*., 1990).

Commercial milk replacer composed of whole milk powder, whey powder, lactose, soy protein isolate, vitamin-mineral premix, coccidiostat, flavorant, growth promoter and antioxidant additives. The chemical composition, expressed in dry matter of commercial milk replacer is as follows: 88% DM, 20% CP, 12% EE, 45% Lactose, 15% Ash, 0.8% Calcium and 0.4% Phosphorus (Oliveira *et al*., 2015). Milk replacers vary widely in protein content. Crude protein usually varies between 18 – 24% whereas milk contains approximately 25 – 26% crude protein on a dry matter basis (Davis and Drackley, 1998).

According to NRC (2001), milk replacers should contain at least 22% crude protein on a dry matter basis. The best milk replacers contain at least 20% crude protein on an as fed basis from primary milk sources. The subsequent crude protein level in milk replacers formulated from non-milk protein is generally higher than the protein in the entire milk-protein milk replacers, in an attempt to compensate for decreased protein digestibility and amino acid utilization (Davis and Drackley, 1998). However, the content of protein necessary for calf growth depends on the amount fed, the amount of starter feed consumed, the energy density of the milk replacer and starter as well as the source of protein (Drackley, 1999). Convenience, economics and biosecurity are some of the factors that make using milk replacer appealing to dairy producers (Oliveira *et al*., 2015). Diseases such as Johne's, salmonella, *Escherichia coli*, Bovine Viral Diarrhea, pasteurella and mycoplasma can be transmitted through unpasteurized milk (NRC, 2001). The comparison of quality of MR with whole milk have been reported to give similar performance for the parameters observed (Heinrichs and Lesmeister, 2005).

2.5.5 Alternative Proteins in Milk Replacers for Dairy Calves

During the pre-weaning period, calves need a high quality liquid feed, normally provided as milk or milk replacer. Milk replacer is a very acceptable source of nutrients due to the consistency and nutrient content of the product.

The milk replacer industry is based on the use of whey protein concentrate as main source of protein, however increase in the demand for dairy proteins are increasing the price of whey protein, and consequently milk replacers (Karszes, 2005). Alternative proteins for use in calf milk replacers and starters are typically selected based on how close their amino acid profile matches that of milk proteins. Among all the alternative sources currently used, soybean protein demonstrates some desirable characteristics such as availability, price and chemical composition (Toullec *et al*., 1994).

However, substituting 40% of the CP from whey protein with soy protein generally decreases growth rates in pre-ruminant calves. The causes for this decrease are attributed to the presence of anti-nutritional compounds, lowered digestibility, and increased endogenous protein production (Karszes, 2005). Therefore to ensure proper growth and health of the calf, processing and subsequent solubility of the protein source are crucially important for determining the suitability of alternative proteins in milk replacers. Calves require a high quality protein to grow properly (Quigley, 1999).

Soybean protein is often used in calf milk replacers and starters instead of milk proteins because of its low cost and its essential amino acid content is similar to cow's milk (Karszes, 2005). Although soybean protein is most widely used and thoroughly researched as the main alternative protein used for calf diets, there are many other alternative proteins available. Alternative protein ingredients such as wheat, fish, corn, pea, rapeseed, field beans, peas, blood, meat, bone, egg, sunflower, barley, oats, beet pulp and potatoes, have also been used and researched as protein sources in milk replacers (Quigley, 1999). The use of alternative sources of vegetable and animal protein in milk replacers and starters for calves, instead of using milk or milk protein based products, has increased as a result of increased milk prices and costs of raising calves, along with the increasing importance of milk protein for humans (Quigley, 1999).

2.5.5.1 Soy Products

Soybean is the leading protein source used in livestock diets around the world as it contains high quality protein that is essentially equivalent to milk protein (Liener *et al*., 1985). It also contains many of the essential amino acids that are deficient in most cereal grain-based diets commonly fed as energy sources to livestock (Stobo and Roy, 1979). Soy products have been tried extensively as substitutes for milk protein with varying results due to the physical and chemical treatments they receive. The success of soy protein is based on availability and low cost compared with other protein sources such as skim milk and whey protein (Davis and Drackley, 1998).

Early reports showed that soy flour as a sole source of protein for calves resulted in poor performance and high mortality (Stobo and Roy, 1979). The decrease in digestibility appears to be related to the fact that soybean proteins contain different groups of anti-nutritional compounds such as trypsin inhibitors, antigenic proteins, indigestible carbohydrates, lectins, tannins, phytate and saponins which can be detrimental to the performance of calves (Xu *et al*., 2002). Since then a lot of work has been done to determine the detrimental factors which resulted in such poor performance. Acid or alkali pre-digestion of soy flour was found to result in marked improvement in growth of calves fed a milk replacer containing only soybean protein (Touchette *et al*., 2003).

2.5.6 Other Ingredients Used as Milk Relacer

Other sources of plant protein have been tried as a substitute for milk protein in milk replacers for calves. Other alternative plant protein sources include: potato, wheat, pea and corn (Toullec *et al*., 1994). Alternative sources originating from animals are animal plasma protein (Quigley and Bernard, 1996), egg protein (Touchette *et al*., 2003), fish protein (Petchey, 1982) and meat solubles (Davis and Drackley, 1998; Tanan, 2005).

2.5.6.1 Fish Protein

Fish protein is also used in calf diets. Fish protein concentrate incorporated into the milk replacer at 35% resulted in acceptable growth and performance of calves (Huber and Slade, 1967; Campos and Huber, 1982). Previously, Sleiman and Huber (1971) found that an incorporation rate of 40% fish protein concentrate did not adversely affect calf performance, and Opstevedt *et al*. (1978) concluded even higher levels could be fed if grain and hay were also available to the calf. Huber (1975) found that calves performed satisfactorily when fed milk replacers containing 70% fish protein concentrate if grain and hay were available as well. Gorrill and Nicholson (1969) found that calf performance was similar for calves fed whole milk, milk replacer containing all milk protein, or milk replacer with 50% of the protein coming from isopropanol-extracted fish protein concentrate and 50% coming from milk proteins. Huber and Slade (1967) also found that feed conversion rates were lower for calves on fish flour than calves on all milk replacers and Makdani *et al*. (1974) found similar results with calves fed fish protein concentrate prepared by various processing methods. However, poor performance was recorded when calves were fed milk replacer containing only fish flour protein (Huber and Slade, 1967) or only from fish protein concentrate (Makdani *et al*., 1974). Several other sources of alternative proteins have been studied less extensively as well.

2.5.6.2 Pea Protein

Bhatty and Christison (1980) found that pea protein isolate could replace up to 50% of milk proteins in a milk replacer without any major adverse effects in calf growth or performance. They additionally concluded that pea protein concentrate contained higher levels of indigestible starch and oligosaccharides that prevented it from being utilized at the same rates of incorporation as pea protein isolate.

2.5.6.3 Whey Powder

Whey powder, which is made from the liquid material created as a by-product of cheese production, can be substituted for whole milk proteins in all-milk replacers successfully in calf diets at a rate of up to 60% of the total protein (Cruywagen and Horn, 1984). Acid whey powder can also be added to milk replacers at a level of 23% to help adjust the pH (Gorrill and Nicholson, 1972). Adjusting the pH improves calf performance by slowing the rate of abomasal emptying and lowering chymptrypsin and trypsin activities in the intestinal digesta (Gorrill and Nicholson, 1972).

2.5.6.4 Rapeseed Flour and Oil

Gorrill *et al*. (1976) found that rapeseed flour and oil could be incorporated at 30% level in young calf diets. They also found that dehulled commercial high-glucosinolate rapeseed meal or rapeseed flour didn't appear to be a suitable source of nutrients for calf milk replacers due to poor digestibility and high contents of antinutritional and allergenic factors.

2.5.6.5 Potatoes

Potatoes (Hinks *et al*., 1975) and evaporated milk (Noller *et al*., 1956) can also be used in place of cow's milk proteins in calf rations. These alternative proteins have been used successfully in place of whole milk proteins in calf milk replacers.

2.5.6.6 Blood Protein

Quigley *et al*. (1999) has done several studies with blood protein and found that calves on diets including hydrolyzed spray-dried red blood cells performed equally to calves on all milk replacers, even with higher levels of incorporation.

2.5.6.7 Liquid Egg

Liquid egg is another alternative protein that can be used in milk replacers and can effectively replace up to 10% of the milk proteins in the calf diet (Touchette *et al*., 2003). Quigley (2001) reported that spray-dried whole eggs did not provide enough nutrients to support adequate calf growth when incorporated into a milk replacer as the sole source of protein.

2.5.6.8 Barley, Oats or Molassed Beet Pulp

Finally, Cafrey and McAlesse (1964) found that MR can be based on barley, oats or molassed beet pulp without affecting calf performance. They also found that of the protein supplements they examined, soybean and linseed were the most palatable with skim milk powder, while fish protein was the least palatable, which is valuable information since palatability is the main factor in getting calves to consume liquid feeds at an early age. In addition, sunflower-based MR are also suitable for feeding young calves in place of milk proteins (Mandibaya *et al*., 1999).

2.5.7 Factors that Influence Calf Performance on Alternative Protein

Since two of the main factors that influence calf performance on alternative protein based milk replacers and starters are the proportion of the total protein replaced with the alternative protein and the age of the calf, incorporation rates of alternative proteins in calf diets profoundly impacts how the calves perform on the alternative protein (Wolfswinkel, 2009). Many alternative proteins are suitable for young calves as long as they do not exceed a certain incorporation percentage. The maximum incorporation rate is different for each alternative protein source. Also, the manner in which the alternative protein source is processed has effect on overall calf performance (Wolfswinkel, 2009).

2.5.8 Digestive Disturbances in Calves fed Alternative Proteins

Digestive disturbances in calves fed alternative proteins are often due to the inability of the calf to secrete the appropriate enzymes for the digestion of non-milk nutrients (Caugant *et al*., 1992). Disturbances that occur as a result include inhibition or acceleration of abomasal emptying, impaired curd formation in the abomasum, altered rate of digesta flow through the small intestine, altered gastrointestinal tract morphology, abnormal salt and water exchange, decreased nitrogen absorption, and the creation of antinutritional factors (Williams *et al*., 1976). Other factors that influence calf performance on alternative protein based milk replacers and starters are the proportion of the milk protein replaced with the alternative protein and the age of the calf (Akinyele and Harshbarger, 1983; Caugant *et al*., 1992); younger calves and those fed a higher proportion (above 80%) of alternative protein sources are more likely to respond poorly.

2.6 Effect of Milk Replacer on Calf Performance

The research conducted by Hill *et al*. (2008) demonstrated that milk replacer can support equivalent or greater calf performance if milk replacer is supplemented with amino acid and fatty acid. Similarly the research conducted by Bar-Peled *et al*. (1997) support the idea of increased rate of milk replacer feeding to increase the rate of gain. Bartlett *et al*. (2006) reported that conventional milk replacer containing 20 – 22% CP increased lean tissue growth.

2.6.1 Effect of Soybean Based MR on Growth Performance of Calves

Calves at birth possess all the physical characteristics of an adult ruminant (omasum, abomasum, reticulum, and rumen); however, the presence of a fold of tissue that leads from the base of the esophagus to the reticulo-omasal orifice (oesophageal groove) causes young calves to behave as simple stomached animals. For this reason, newborn calves depend more on dietary ingredients that are easier to digest than their adult counterparts (Blum, 2006).

In this early period of pre-ruminant digestion, the abomasum is the primary gastric compartment, comprising 50 to 70% of the total volume and because of this the activity and digestive capacity of the other three compartments is minor (Davis and Drackley, 1998). Besides the physical modifications in the gastrointestinal tract, the predominance of the abomasum and intestines in digestion of young animals leads to the production of certain enzymes, which forces the calf to behave as a monogastric animal. These enzymes are more adapted to the digestion of milk components because of evolutionary adaptation (Davis and Drackley, 1998).

Additionally, Akinyele and Harshbarger (1983) found that gains for calves fed soy protein concentrate milk replacers were better when only 30% of the milk protein was replaced by soy protein concentrate compared to calves fed milk replacers containing over 84% soy protein. Ghorbani *et al*. (2007) studied the effect of partial replacement of whole milk with soymilk on pre-weaning calf performatnce and weaning costs. The results suggested that feeding soymilk up to 50% of the milk diet maintained health during the first 2 to 4 weeks of age when the neonate calf is highly susceptible to non-milk proteins and plant anti-nutrients. Campos and Huber (1982), however, found that replacement of 50% of the milk protein with soy protein concentrate did not result in significantly lower average daily gains in comparison to calves fed the all milk replacer. Colvin and Ramsey (1968) also found that satisfactory growth could be achieved in calves fed with a milk replacer containing 86% soy protein concentrate and Gorrill and Nicholson (1969) found that 70% could be replaced with soy protein concentrate. If grain and hay is provided in addition to the milk replacer, up to 90% of the milk protein can be replaced with soy protein concentrate. In summary, upwards of about 85% incorporation of soy protein concentrate is acceptable when just milk replacer is supplied to the calf and up to 90% of the protein can be replaced if concentrates and hay are offered.

The performance of calves fed milk replacer containing soy protein concentrate (SPC) differs according to the animal age. In animals younger than 3 weeks of age and with milk replacer containing 50% protein replacement with SPC, a decrease in average daily gain (32.5%) and feed conversion (33%) was observed during the first two weeks of age (Peisker, 2001). The use of high levels of soy flour in milk replacer decreased growth rates in calves (Colvin and Ramsey, 1969). However, in older animals the decrease was only 7.1% and 5.9% compared to all milk ingredient replacers (Lalles, 2000). Masum *et al*. (2009) conducted an experiment to study the utilization of soymilk as milk replacer for raising calves. The calves of group A were reared by cow milk, group B reared by strained (sieved) soymilk and group C reared by unstrained (unsieved) soymilk. The authors found that there was no significant difference among the three groups of calves fed cow milk or soymilk. Non-significant differences were also reported for increment in wither height and heart girth of the calves.

2.6.2 Effect of Soybean Based MR on Nutrient Digestibility and Nitrogen Retention

Results of nutrient digestibilities and nitrogen retention showed considerable improvement when calves were fed milk replacers containing soy protein (Masum *et al*., 2009). Calves were able to use soybean based milk replacer more effectively. However, relatively poor growth, digestibility and nitrogen retention resulted when cooked soybean, soybean meal, or lightly cooked soybean flour were used (Porter and Hill, 1963) and several studies have shown that diets containing more than 30 – 40% of their protein treated by heating or fat-extraction or untreated soy flour nearly always proved unsuccessful when fed to calves and have led to diarrhea, weight loss, poor growth rate and sometimes even death (Colvin and Ramsey, 1968). This poor performance could be due to the high levels of starch in soybean meal and soy flour, which require further processing other than cooking to become digestible to the calf (Gorrill and Nicholson, 1969).

Caugant *et al*. (1992) found the differences in amino acid digestibilities of soy flour and soy protein concentrate diets were minimal, suggesting that the processing used to prepare soy flour can vastly improve its nutritional value by breaking down starches that are indigestible to the young calf. Colvin and Ramsey (1969) presented data from calves fed milk replacers containing soy flour that would support this observation as well. When they treated soy flour by soaking it in acid, they doubled the growth of the calves fed the milk replacer containing acid-treated soy flour in comparison with the calves on the untreated soy flour.

Another way to overcome the poor digestibility of soy flour is to feed it in combination with another higher quality protein such as whey protein concentrate. Rindsig and Bodoh (1977) used soy flour in combination with whey powder and colostrum to create a milk replacer that was suitable for young calves. Ground, raw soybean is not an acceptable source of protein for young calves in milk replacers when incorporated at a level of 40% (Williams and Knodt, 1950). Kwiatkowska (1972) found that up to 30% of protein from the milk replacer could be supplied in the form of solvent-extracted soybean meal with no significant differences in nitrogen retention. When soybean meal supplied 73% of the total protein in a milk replacer, calves grew poorly due to low protein digestibility and decreased fat and ash absorption (Nitsan *et al*., 2005).

2.6.3 Economic Benefits of Feeding Soybean Based Milk Replacers

Economics of animal production are becoming a vital component in farm management. Finding opportunities to reduce cost without compromising animal performance is a high priority in dairy farming (El-jack and Ahmed, 2012). Nutrition, health, reproduction, and calving management are areas where improvement can be made (Karszes, 2005). The first month of the calves life are the most demanding, because newborn calves depend completely on the feed they receive to survive; therefore they need high quality feed and intensive care (Karszes, 2005).

Among the costs to raise a heifer, feed accounts for 49% of the total costs invested, followed by labour that accounts only for 17%. The most inefficient stage is from birth to 90 kg (pre-weaning), when animal's feed and management cost is high, but weight gain accumulated in that period is less than 8% of the total weight accumulated in animal's life (Karszes, 2005). El-jack and Ahmed (2012) carried out an experiment to examine the cost effective feeding system of buffalo male calves for quality meat production. Results obtained from that study showed that feed cost per kg weight gain for group of calves fed milk replacer at 4% of body weight + early weaning diet for the first 45 days and later on early weaning diet only was 52 and 25% less than calves fed milk replacer at 8% of body weight and those fed milk replacer at 4% of body weight + early weaning diet on ad libitum basis.

Economic analysis also showed that use of soymilk was cheaper than cow milk. And was concluded that soymilk as milk replacer could be used successfully for raising dairy calves (Masum *et al*., 2009). Results showed that milk replacer feeding is 11.39% less expensive than whole milk (Khan *et al*., 2012). Results obtained by (Ghorbani *et al*., 2007) introduced soymilk as an economic substitute for whole milk in calf-rearing. Therefore, calves can be successfully reared on milk replacer at a lower cost than with whole milk, thus offering a potential increase in economic returns for dairy farmers (Khan *et al*., 2012).

2.6.4 Causes of Growth Depression in Calves fed Soybean Based Milk Replacers

The chemical composition of soybean indicates adequate protein levels and reasonable amino acid profiles for growth. In fact, soybean appears to possess fairly high levels of essential amino acids similar to that of cow's milk (FAO, 1988). Despite the general increase in CP content for the soy ingredients used in milk replacer, digestibility varies depending on the overall composition of the milk replacer, origin of the ingredients and the animal's age (Davis and Drackley, 1998).

Higher CP digestibility is normally observed in skim milk, whey protein and skim milk protein concentrate; however, digestibility increases with increasing age, no matter the quality of the ingredients. It was reported that the cause of decreased performance in calves fed soy protein could be associated with decrease apparent digestibility (Shakya *et al*., 2016). Decreases in digestibility are normally attributed to the presence of antinutritional factors and protein fractions associated with low digestibility (Roy *et al*., 2016). The anti-nutritional factors like ß-galactomannans and lectins can disturb the physiological processes of digestion and absorption of nutrients, and cause immunological reactions in the gut that decrease energy supply to the tissues (Tomkins and Jaster, 1991).

Several studies have found that the anti-nutritional factors (trypsin inhibitor, lectins, antigenic proteins, polyphenols, oligosaccharides, phytates, saponins) found in soy proteins causes decreased growth, low feed conversion efficiency and poor health of the animals (Tomkins and Jaster, 1991). The factors associated with reduced performance in soy protein sources containing anti-nutritional factors (trypsin inhibitors, lectins, antigens) which can influence protein digestion and absorption, but also the anti-nutritional factors can influence carbohydrate digestion, mineral utilization and vitamin availability (Tomkins and Jaster, 1991).

Other factors constraining animal performance from soybased MR are the improper or inadequate processing of the soybean and reconstitution at high temperatures (Lalles, 1993). Digestive disturbances in calves fed alternative proteins are often due to the inability of the calf to secrete the appropriate enzymes for the digestion of non-milk nutrients (Lalles, 1993). Disturbances that occur as a result include inhibition or acceleration of abomasal emptying, impaired curd formation in the abomasum, altered rate of digesta flow through the small intestine, altered gastrointestinal tract morphology, abnormal salt and water exchange, decreased nitrogen absorption, and the creation of antinutritional factors (Lalles, 1993).

Other factors that influence calf performance on alternative protein based milk replacers and starters are the proportion of the milk protein replaced with the alternative protein and the age of the calf (Heinrichs, 2005). Results from that experiment reinforced the idea that the replacement of a large part of protein from dairy origin with soy protein depresses animal performance and diet digestibility. Younger calves and those fed a higher proportion of alternative protein sources are more likely to respond poorly (Heinrichs and Jones, 2003).

2.7 Solid Feeds

2.7.1 Calf Starter

Calf starter is a crucial link to proper ruminal development and successful weaning. Since intake of dry feed initiates rumen development and allows early weaning, the availability and intake of calf starters is important to calves prior to weaning (Quigley, 1999). Calf starter quality is important to ensuring good rumen development and to allow early weaning (Heinrichs, 2005). Calf starters fed to calves may be meals (unpelleted), pelleted or textured type feeds. Ingredients used in calf starters generally reflect the type of starter. Textured feeds, for example, generally contain a pellet, which contains protein, carbohydrate sources plus vitamins and minerals, along with other grains, such as rolled oats and corn (Davis and Drackley, 1998). All starters must meet the nutrient requirements of the calf. It is also important to note that the 1989 NRC requirements for calf starters assume that hay would be offered to calves in addition to starter (Heinrichs, 2005).

2.7.1.1 Variables that Contribute to Differences in Calf Starter Intake

There are a number of variables that contribute to differences in calf starter intake as all starters are not the same (Davis and Drackley, 1998). The milk feeding program affects calf starter intake based on the amount being fed and the percentage of protein and fat in the milk replacer. Water is an essential nutrient for the calf and its availability affects starter intake (Heinrichs, 2005).

Other factors include the formulations and physical form of the starter, the birth weight of the calf, the genetic makeup of the calf and even the gender has an effect on intake. And finally, the housing, management and environment the calf is exposed to has a large impact on how well the calf consumes the starter offered (Quigley, 1999).

2.7.2 Forages

Hay is often preferred to straw or silage as a mean of bulk in the diet (Hill, 2005). High intakes of hay could be detrimental to development as it fills up the stomach and reduces the intake of grain/concentrates (Shain *et al*., 1993). However, Hill *et al*. (2005) found that young calves wasted 3 times more than they consumed when offered straw or hay. Other alternatives to forages are coarsely ground grain or other fibrous feed like coarsely ground starters/grains and soybean hulls (Shain *et al*., 1993).

2.7.3 Effect of Milk Replacer on Starter Intake

Khan *et al*. (2007) described that calves consumed starter ration at 14 day of age, the intake of starter ration increases as feeding milk is reduced. Similar results were reported by Terre *et al*. (2009). Ghorbani *et al*. (2007) reported that calves fed milk or milk replacer consume as much starter feeds as those fed higher amount of milk before weaning. However, when unlimited amount of milk are offered to young calves they begin to chew solid feed at 2nd week of life. Baldwin *et al*. (2004) reported that if calves are not given solid feed at early age, they will begin to consume bedding materials, indicating a growing hunger for solids as the calf age.

2.7.4 Effect of Liquid Feeding on Rumen Development

Ad libitum milk feeding to calves is known to delay the initiation of ruminal fermentation and development because of depressed intake of solid feed (Baldwin *et al*., 2004). The newborn calf is born with a sterile tract and the inoculation of microbes is achieved by the cow licking the calf, microbes on feed particles and even microbes in water droplets in the environment (Mann, 1963).

Depending on the feeding, especially the amounts of milk, the digestive tract might be fully developed at around 8 weeks of age (Tamate *et al*., 1962). In free grazing ruminants, Wardrop and Coombe (1961) suggested dividing the development of the digestive tract into three periods. The non-ruminant phase, where the young ruminant is mainly dependent on milk feeding from the mother, a transition phase at the age of 3 – 8 weeks, at which the young ruminant has started to consume vegetation, and the last stage from 8 weeks and onwards, where it is considered to be an adult ruminant. Butyric acid, and to some extent propionic acid, from the digestion of easily digested carbohydrates, are the main energy source for rumen epithelial growth and development (Britton and Krehbiel, 1993). Therefore, calves fed only milk have little papillae growth (Warner *et al*., 1956) as the milk by-passes the fore-stomachs through the reticular groove and into the abomasum.

After weaning feeding calves are governed by digestive capacity, physio-chemical attribute of solid feed and change in absorption metabolites (Lesmeister and Heinrichs, 2004). It has been known that calves can grow a lot faster when they are supplied with more nutrients (Britton and Krehbiel, 1993). Large amount of milk before weaning may delayed rumen development and physiochemical properties of rumen and cause lower solid feed consumption. Thus, restricted milk feeding should be introduced to encourage calves to eat concentrate and minimize cost for relatively expensive liquid milk (Lesmeister and Heinrichs, 2004). The mortality of calves increases from 5% to as much as 20% if colostrum is offered later than 24 hours post-parturition compared to within the first 6 hours limit. In practice, the immune status of the calf can be determined from the serum level of immunoglobulins (Margerison and Downey, 2005).

2.8 Calf Rearing Systems

The calf rearing period, which is the period from birth until 6 months after weaning, is a period with many risks. The highest mortality rates occur at this period. The newly born calf is very weak and susceptible to diseases. During this period it needs extra care, with particular attention to good feeding, routine health check up, hygiene and housing (Davis and Drackley, 1998).

2.8.1 Conventional Rearing

Until recently, little attention has been on the nutrition in the pre-weaning period. The live weight gain in the pre-weaning period was thought to be of less importance as the calves have been thought to be able to compensate for this growth through compensatory growth (Owens *et al*., 1993) post-weaning. Therefore, the calves have been given restricted amounts of milk/MR at rates of 8 – 10% of body weight, aiming at growth rates far less than the genetic potential. In contrast, intakes of 16 – 24% of body weight were measured in calves fed WM/MR *ad libitum* (Davis and Drackley, 1998). Restricted amounts of liquid feed normally leads to increased intake of dry feed like grain/concentrates at earlier age (Hopkins, 1997), which again leads to early weaning. Other arguments for early weaning is improved calf health and less labour required for the calf rearing and reduced feed costs in the heifer rearing, as liquid feed normally is more expensive than dry feed (Davis and Drackley, 1998).

2.8.2 Intensive Rearing

The view on calf and heifer rearing for replacement is changing. More focus is now on feed quality and amounts of nutrient intake in the pre-weaning period, as several studies have concluded that an elevated nutrient intake in the pre-weaning period is a good investment in the replacer herd as positive effects both in early and later life was revealed (Shamay *et al*., 2005; Drackley *et al*., 2007; Terré *et al*., 2009).

The term “intensive rearing” is here defined as an elevated nutrient intake during the pre-weaning period of the calves through WM or MR with protein levels like in WM, fed at feeding levels close to *ad libitum*, aiming at levels of average daily gain of 800 g in the pre-weaning period. This level of growth rate in early life is only possible through liquid feed like milk or milk replacers (Davis and Drackley, 1998).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Location of the Study

The study was conducted at the National Animal Production Research Institute (NAPRI), Ahmadu Bello University, Shika-Zaria, Nigeria. Shika is located within the Northern Guinea Savanna ecological zone of Nigeria between latitude 10°11'N and longitude 7°8'E, at an altitude of 650m above sea level (Ovimaps, 2016). The area receives a mean annual rainfall of 1150mm which commences from May and last till October. Following the wet season is a period of dry cool weather known as 'harmattan' which marks the onset of the dry season. This extends from mid-October to March when the hot weather sets in. At this period, the mean minimum and maximum temperatures range from 12 – 28°C during the cold 'harmattan' season and 20 – 36°C in the hot season. The mean relative humidity is 21% and 72% during the 'harmattan' and the rainy seasons respectively (IAR, 2016).

3.2 Source of Feed Materials

The soybean variety used for this study was Samsoy II and was purchased from an open market in Giwa Local Government Area of Kaduna State. theWhile the other feed materials *Digitaria smutsii* hay, concentrate diets and cow milk (Friesian x Bunaji milk) were obtained from Dairy Research Programme of NAPRI, Shika-Zaria.

3.3 Stage One of the Experiment: Effect of Soaking Duration on Nutritional and Anti-Nutritional Composition of Soybean.

3.3.1 Preparation of Soyflour

The collected soybean was cleaned by winnowing and hand picking of stones and debris. The cleaned soybean were divided into five groups labelled as (A, B, C, D and E) each consisting of about 0.5 kg of soybean. The soybean in the first group (A) which served as the control were not soaked in water rather they were sun-dried for a day on a stainless tray, milled into flour and sieved with the use of 0.04mm sieve.

The resultant soyflour was stored in polythene bags and then taken to the laboratory for chemical analysis. While the soybean in the remaining groups (B, C, D and E) were soaked in excess water in plastic containers for 12, 24, 48 and 72 hours, respectively. The water was changed twice after every 24 hours. This allows for greater leaching of anti-nutritional factors, minerals, soluble sugars and non-protein nitrogen. After each soaking period, the soybean were rinsed with clean tap water and sun-dried for 8 days on a stainless tray. The dried soybean were milled into flour and sieved with the use of 0.04mm sieve. The resultant soyflour was stored in polythene bags and were taken to the laboratory for chemical analysis. The flowchart of the processing method is shown in figure 3.1.

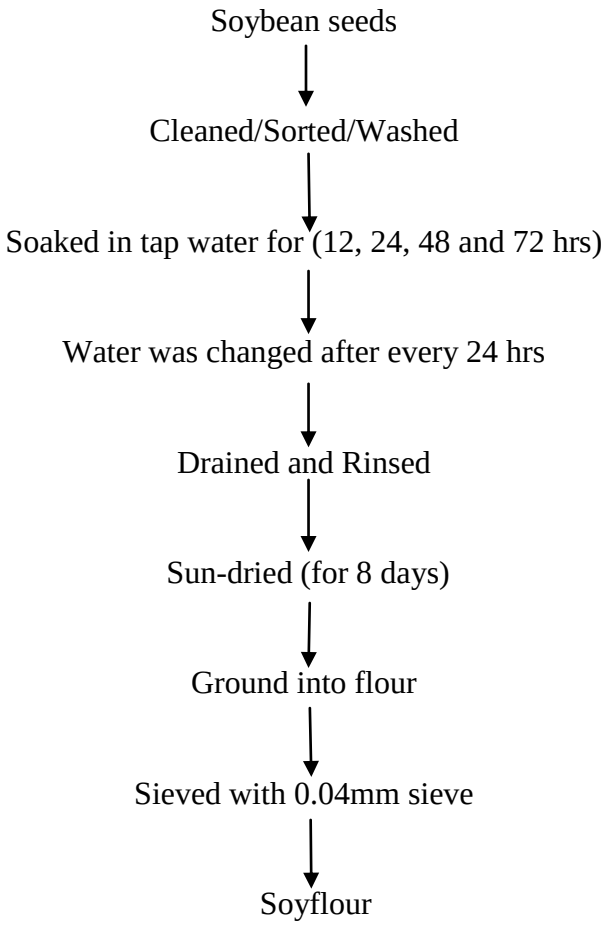


Figure 3.1: Flowchart for Soybean Processing into Soyflour.
Source: Revised from Pele *et al*. (2016).

3.4 Stage Two of the Experiment: Growth Performance of Friesian x Bunaji Calves.

3.4.1 Experimental Animals, Design and Management

Sixteen (16) Friesian x Bunaji dairy calves of mixed sexes (8 males and 8 females) aged between 2 – 3 weeks, with live weight of 34.8±0.7kg from the Dairy Research Programme of the National Animal Production Research Institute (NAPRI), Ahmadu Bello University (ABU), Shika-Zaria were used for the growth trial. The calves were identified with ear tags, weighed and randomly distributed into four (4) dietary treatments consisting of 4 calves per treatment in a Completely Randomized Design (CRD). The calves were housed in an open-sided, well-ventilated building (calf-pen) with a concrete floor equipped with both feeding and watering troughs. In the morning, each of the experimental animal received one litre of the different ratios of soy:cow milk between 08:00 – 09:00am, and another one litre was administered in the evening between 04:00 – 05:00pm. In addition, the experimental animals were given weighed amount of concentrate diet and *Digitaria smutsii* hay to stimulate rumen development. The concentrate diet was made up of maize, maize offal, cotton seed cake (CSC), bone meal, common salt (NaCl) and vitamin-mineral premix (Table 3.1). Fresh and clean drinking water was also provided daily *ad libitum*. Proper sanitary measures were obeserved to protect the calves against parasitic infestations and other contagious diseases. The floor of the calf house was washed thoroughly using a hand brush, broom, detergent and Izal. The watering and feeding troughs were also washed daily before the morning feeding. The growth trial was carried out for a period of 98 days (between October, 2016 to February, 2017).

3.4.2 Experimental Diets

Ten (10) litres of Friesian x Bunaji milk from the Dairy Research programme of NAPRI was collected at 08:00am and used for the morning feeding, while another 10 litres was collected at 04:00pm and used for the evening feeding, respectively.

The experimental diets were prepared immediately after fresh milk collection. Soymilk was prepared in batches according to standard procedure. To formulate one litre of soymilk, 125g of the 72 hrs soaked soyflour was dissolved in 1000ml of clean tap water (Sarker *et al*., 2015). Thereafter, 6 litres of soymilk was prepared by dissolving 750g of the 72 hrs soaked soyflour in 6000 ml of clean tap water. The mixture was homogenized by constant stirring to prevent coagulation. The soymilk was added to fresh cow milk in a stainless container at the ratio of 0:1000mls (which served as the control); 250:750mls; 500:500mls and 750:250mls soymilk:cowmilk respectively before feeding to the calves. The different ratios of the soy:cow milk used are as follows:

- **Experimental Diet 1 =** 0% soymilk + 100% cow milk (control)
- **Experimental Diet 2 =** 25% soymilk + 75% cow milk
- **Experimental Diet 3 =** 50% soymilk + 50% cow milk
- **Experimental Diet 4 =** 75% soymilk + 25% cow milk

Table 3.1: Ingredient Composition of Concentrate Diet fed to Friesian x Bunaji Calves.

Ingredients	**Percentage (%)**
Maize	22.00
Maize Offal	22.75
Cotton Seed Cake	52.00
Bone Meal	2.00
Common Salt (NaCl)	1.00
Vitamin-Mineral Premix	0.25
Total	**100.00**

3.4.3 Milk Sample Collection

Milk sample was collected from Friesian x Bunaji dairy cows that were kept on the Dairy Research Programme farm of NAPRI, Shika, Zaria. To avoid contaminations, the plastic containers used for milk sample collection were cleaned by soaking them for 24 hours in 10% HNO_3, then in distilled water for another 24 hours after which the containers were rinsed with more distilled water, dried in an oven before being used for milk sample collection.

The milk samples in the cleaned plastics containers were quickly transported and stored in ice packed cooler at 0 ± 6^{0}C and stored in a deep freezer at -4^{o}C pending laboratory analyses.

3.4.4 Feed Intake and Live Weight Changes

Feed offered and leftovers (orts) were recorded daily. Concentrate diet and hay were offered in the morning in separate feeding troughs; feed leftovers were weighed the following morning and total feed intake was determined by difference. Body weight changes of calves were recorded fortnightly before the morning feeding using an analog weighing scale graduated in kg. Total weight gain was calculated from the difference between initial body weight and final body weight. Average daily weight gain (ADWG) was determined by dividing the total weight gain by the number of days of the feeding trial (i.e. 98 days). Feed conversion ratio (FCR) was calculated from the ratio of average daily feed intake to average daily weight gain and feed cost per kg gain were calculated as secondary data.

3.4.5 Linear Body Measurements

Anatomical reference points for linear body measurements described according to the methods of Yakubu *et al*. (2009) were used. All the measurements were taken manually using a flexible measuring tape calibrated in centimetres (cm). Caution was taken to avoid animal movement during measurement by manually restraining the animal while standing on a flat surface with the head held up in an unforced position. The following linear body measurements were taken:

***3.4.5.1 Body Length (BL)*:** This was measured as the distance from point of the shoulder blades to the point of the pin bone (or ischium) in centimetre.

***3.4.5.2 Height at Withers (HW)*:** This was measured as the highest point over the scapulae vertically to the ground or measured from the highest point on the dorsum of the animal to the ground surface at the level of front legs.

***3.4.5.3 Heart Girth (HG)*:** This was measured as the circumference of the body at a point immediately behind the fore legs, perpendicular to the body axis.

3.4.6 Blood Collection

Blood samples were collected from all the experimental animals (16) at end of the growth trial. Five (5) mls of blood sample was drawn through the jugular vein using hypodermic needles with syringe and was emptied into two separate sets of vacutainer tubes. Two (2) mls of the collected blood sample was poured into a plastic bottle containing anti-coagulant [ethylene diaminetetracetic acid (EDTA)]. The vials were capped immediately and the contents were tilted twice gently to mix the blood with the EDTA as reported by Aye (2013). The remaining 3mls was emptied into another plastic bottle that donot contain EDTA.

3.4.7 Rumen Fluid Sampling

At the end of the growth trial, all the experimental animals were used for rumen fluid collection to evaluate the effect of feeding different ratios of soymilk and cow milk on some rumen metabolites. Rumen fluid samples were collected using stomach tube at 0hr before morning feeding and subsequently at 3, 6 and 9 hours post feeding, respectively. The stomach tube used had a probe attached at the posterior end which was carefully inserted into the rumen of the calf through the oesophagus, after restraining the animal. The pump was pressured to collect the rumen liquor into a stainless container. A thermometer and digital pH meter were used immediately to determine the temperature and pH respectively. The samples were strained through a double layer cheese cloth to remove extraneous materials. Twenty (20) mls of the rumen fluid was measured into well-labelled plastic bottles containing equal volume (20mls) of 0.1 normal sulphuric acid (H_2SO_4) solution to trap the ammonia Nitrogen (NH_3-N). After each collection, the rumen fluid samples were immediately stored in a freezer at a temperature of -4^oC pending laboratory analysis.

3.4.8 Metabolism Trial

The aim of this study was to determine the nutrient digestibility and nitrogen balance of the experimental diets. At the end of the growth study, all the experimental animals were carefully transferred to metabolism crates fitted with facilities for separate collection of faeces from urine as reported by (Osuji *et al*., 1993). During the digestibility trial however, the animals were maintained on the same treatment diets as in the growth trial. Clean drinking water was provided *ad libitum*. An adjustment period of 10 days was observed before a 7 day collection period, during which feed offered, left over (orts), urine and faeces collected were weighed and recorded at 08.00hours every day.

Nitrogen loss from urine was prevented by collecting urine for each animal into a plastic container acidified with 100mls of 0.1N tetraoxosulphate (VI) acid (H_2SO_4) placed under the metabolism crate. Ten percent (10%) of the daily urine output was taken from each animal and bulked in well-labelled plastic bottles stored in a refrigerator. At the end of the 7 day's collection period, 10% of the total urine bulked from each of the experimental animal was sub-sampled and kept in the refrigerator pending analysis (Osuji *et al*., 1993). Daily total faecal output were also collected, weighed and sample was taken for dry matter determination. The representative samples were bulked and sub-sampled, milled, sieved using a 0.04mm sieve and stored in polyethene bags pending analyses (Osuji *et al*., 1993).

3.5 Economic Analyses

The market price of the various feed ingredients at the time the experiment was conducted was used to compute and compare the total cost of feed consumed during the growth trial and feed cost per kilogram weight gain. This was used to determine whether feeding different ratios of soymilk and cow milk was economical or not.

3.6 Laboratory Analyses

The chemical composition of feed samples (milk from Friesain x Bunaji dairy cows, soybean at different duration of soaking, soy:cow milk mixtures, *Digitaria smutsii* hay and concentrate diets), rumen fluid, blood, urine and faecal samples were determined. Dry matter content of soybean at different duration of soaking, *Digitaria smutsii* hay, concentrate diets and faecal samples were determined by oven drying at 60^{o}C for 48 hours. Crude protein content (Nitrogen x 6.25) of the feed samples and urine were determined using the Micro Kjeldahl procedure of the Association of Official Analytical Chemists (AOAC, 2005). Total ash was determined by charring the samples in a muffle furnace at 500^{o}C for about 3 hours. Ether extract (EE) and crude fibre (CF) were determined according to standard procedures of (AOAC, 2005). While nitrogen free extract (NFE) was determined by subtraction (on DM basis): NFE = DM – % CP + % CF + % EE + % Ash.

Neutral detergent fibre (NDF), acid detergent fibre (ADF) and hemicelluloses were determined by the method of Van Soest *et al*. (1991). Milk and soymilk samples were analyzed for gross composition (moisture, total solids, solid-not-fat, fat, protein and ash). The total solid (TS) was determined according to the method described by Bradley Jr (2003) by drying 5g of milk samples to a constant weight at 105^{o}C for 3 hours (AOAC, 2005). Fat content was estimated by the Gerber method (AOAC, 2005).

Solid-not-fat (SNF) was calculated as the difference between TS and fat content (SNF = TS – fat). While milk protein (Nitrogen x 6.38) was determined by semi-micro distillation method using Kjeldahl and Makhamps apparatus and ash content was determined by igniting the dried samples at 500^{o}C (AOAC, 2005). For blood samples collected, the heparinised blood was used for haematological studies which include: packed cell volume, haemoglobin, red blood cell, white blood cell, lymphocytes, neutrophils and basophils.

While the blood samples in the set of bottles containing no anti-coagulant were kept in the refrigerator at a temperature of about –4°C for about 3 hours to aid sedimentation. The samples were later spun in a centrifuge at 3000 rpm for 10 minutes and the serum was separated and stored at –10°C for analysis. The sera for each of the animals were then used to assay for the serum metabolites (Total Protein, Globulin, Albumin, Glucose, Blood Urea Nitrogen and Creatinine) according to methods descibed by Etana *et al*. (2011).

Rumen ammonia concentration and total volatile fatty acids (TVFA) were determined according to standard procedures of AOAC (2005). Tannin contents were determined according to methods described by (Makkar, 2000). Oxalate and phytate were determined according to methods described by (Oke, 1969) and (Reddy *et al*., 1988), respectively. Other ANFs (saponins and protease inhibitors) were determined according to procedures described by (AOAC, 2005). All chemical analyses were carried out at the Central Laboratory of NAPRI, ABU, Shika-Zaria according to standard procedures.

3.7 Statistical Analyses

Data generated from this study were analyzed using the General Linear Model (GLM) procedure of Statistical Analysis System (SAS, 2002) to see the response of the experimental animals to measured parameters. Significant differences among treatment means were compared using Dunnette's Test in the SAS package. The models used for the feeding trial and rumen fluid sampling time were as follows:

3.7.1 Model for the Feeding Trial

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where:

Y_{ij} = j^{th} observation of the i^{th} ratios of soy:cow milk;

μ = Overall mean;

T_i = effect of the i^{th} ratio of soy: cow milk on performance (i = 0:100, 25:75, 50:50 and 75:25%);

e_{ij} = random error.

3.7.2 Model for Rumen Fluid Sampling Time

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where:

Y_{ij} = j^{th} observation of the i^{th} sampling time of rumen fluid;

μ = Overall mean;

T_i = effect of the i^{th} sampling time on rumen metabolites (i = 0, 3, 6 and 9 hours);

e_{ij} = random error.

CHAPTER FOUR

4.0 RESULTS

4.1 Stage One of the Experiment: Effect of Soaking Duration on Nutritional and Anti-Nutritional Compositon of Soybean.

4.1.1 Chemical Composition of Soybean at Different Soaking Duration (DM basis).

The result of the nutrient composition of soybean subjected to different duration of soaking (on dry matter basis) is presented in Tables 4.1. The dry matter content ranged from 95.53 in the soybean soaked for 48 hours to 96.91% in the unsoaked soybean. The crude protein content ranged from 40.28 in the unsoaked soybean to 46.60% in soybean soaked for 24 hours. The crude fibre, ether extract and ash content ranged from 5.16 to 9.50%, 14.11 to 29.55% and 2.95 to 8.47%, respectively. While nitrogen free extract (NFE) ranged from 11.01 in soybean soaked for 48 hrs to 24.55% in the unsoaked, and the metabolizable energy (ME) ranged from 3459.50 in the unsoaked soybean to 5631.61 kcal/kg DM in soybean soaked for 48 hours.

4.1.2 Chemical Composition of Friesian x Bunaji Milk and SoyMilk (wet basis).

The result of the chemical composition of milk from Friesian x Bunaji cows and soymilk soaked for 72 hours is presented in Table 4.2. The cow milk had a total solid, fat, solid-non fat, protein and ash content of 12.44, 2.90, 9.54, 3.29 and 0.75%, respectively. While soymilk had a total solid, fat, solid-non fat, protein and ash content of 87.11, 12.89, 3.30, 9.59, 5.55 and 0.82%, respectively.

Table 4.1: Proximate Composition of Soybean at Different Duration of Soaking (DM basis)

	Duration of Soaking (hours)				
Parameters (%)	**0**	**12**	**24**	**48**	**72**
Dry Matter	96.91	96.63	96.03	95.53	96.18
Crude Protein	40.28	45.44	46.60	45.53	44.37
Crude Fibre	9.50	6.17	5.16	8.46	7.60
Ether Extract	14.11	18.48	24.78	29.26	29.55
Ash	8.47	3.71	3.78	3.29	2.95
Nitrogen Free Extract	24.55	22.83	15.71	11.01	11.71
ME[1]	3459.50	5333.04	5459.87	5631.61	5514.57

[1] = Kcal/kg DM; DM = Dry Matter; ME= Metabolizable Energy

Table 4.2: Chemical Composition of Friesian x Bunaji Milk and Soymilk (Wet basis).

Parameters (%)	Friesian x Bunaji Milk	Soymilk (soaked for 72 hrs)
Moisture	88.30	87.11
Total Solid	11.70	12.89
Fat	2.69	3.30
Solid-Not-Fat	9.01	9.59
Protein	3.29	5.55
Ash	0.67	0.82

4.1.3 Anti-Nutritional Factors in Soybean at Different Duration of Soaking

The results of the anti-nutritional factors (ANFs) of the unsoaked and soaked soybean are presented in Table 4.3. Soaking of soybean in water has significant ($P<0.05$) effect on the levels of some ANFs studied. Unsoaked soybean had 4.39, 7.09, 0.42, 1.04 and 2.46 mg/100ml levels of phytateas, protease inhibitors, tannins, oxalates and saponins, respectively. Soaking for 12 hours reduced the levels of phytateas from (4.39 to 2.21mg/100ml), protease inhibitors (7.09 to 6.03mg/100ml), tannins (0.42 to 0.36 mg/100ml), oxalates (1.04 to 1.00 mg/100ml) and saponins (2.46 to 1.06 mg/100ml), respectively. Increasing the soaking duration resulted in an increased reduction in the levels of all the ANFs studied. Soaking for 72 hours had the highest level of percent reduction in ANFs (90.89, 46.54, 57.14. 19.23 and 60.16 for phytates, protease inhibitors, tannins, oxalates and saponins, respectively).

4.2 Chemical Composition of Concentrate Diet and *Digitaria smutsii* Hay

The results of the chemical composition of concentrate diet and *Digitaria smutsii* hay used for the growth trial are presented in Table 4.4. The dry matter and crude protein contents of the concentrate diet and *Digitaria smutsii* hay were 91.65 and 13.85%, 95.54 and 7.76%, respectively. While the crude fibre, ether extract, ash, NFE, NDF, ADF and hemicellulose contents of the concentrate diet were 14.70, 18.88, 11.94, 32.28, 23.43, 68.50 and 42.80%, respectively. However, *Digitaria smutsii* hay had 10.50% CF, 13.65% EE, 8.53% ash, 55.10%, NFE, 49.29% NDF, 63.73% ADF and 9.88% hemicellulose as shown in Table 4.4.

4.3 Chemical Composition of Different Ratios of Soy:Cow Milk fed to Calves

Table 4.5 shows the results of the chemical composition of the different ratios of soy:cow milk fed to Friesian x Bunaji calves. The moisture content differed significantly ($P<0.05$) and ranged from 81.98 in the diet containing 75:25 ratio to 88.30% in 0:100 ratio (the control diet). There was a significant ($P<0.05$) increase in total solids as the level of soymilk inceased in the diet.

Table 4.3: Anti-Nutritional Factors in Unsoaked and Soaked Soybean (DM basis).

	Duration of Soaking (hours)				
Parameters (mg/100ml)	**0**	**12**	**24**	**48**	**72**
Phytates	4.39	2.21	1.29	1.75	0.40
Protease inhibitors	7.09	6.03	5.72	4.91	3.79
Tannins	0.42	0.36	0.28	0.24	0.18
Oxalates	1.04	1.00	0.96	0.94	0.84
Saponins	2.46	1.06	1.04	1.02	0.98
% Reduction in ANFs					
Phytates	-	49.66	70.62	60.14	90.89
Protease inhibitors	-	14.95	19.32	30.75	46.54
Tannins	-	14.29	33.33	42.86	57.14
Oxalates	-	3.85	7.69	9.62	19.23
Saponins	-	56.91	57.72	58.54	60.16

DM = Dry Matter; ANFs = Anti-Nutritional Factors

The fat, solid-non fat, protein and ash content also follow similar trend. Total solids, fat and solid-not-fat were higher in diet containing 75:25 ratio (18.02, 4.75 and 13.27%) but not significantly (P>0.05) different from 50:50 ratio (16.52, 3.75 and 12.77%), respectively. However, protein and ash contents in the diet containing 75:25 ratio of soy:cow milk were higher (5.23 and 1.45%) compared with (3.29 and 0.67%) of protein and ash in the control diet.

4.4 Stage Two of the Experiment: Performance of Friesian x Bunaji Calves fed Different Ratios of Soy:Cow Milk.

4.4.1 Effect of Feeding Different Ratios of Soy:Cow Milk on Intake and Live Weight Changes of Friesian x Bunaji Calves.

The results of feed intake (dry and liquid feeds) and weight changes of calves fed different ratios of soy:cow milk are presented in Table 4.6. The average total dry matter intake of calves on 0:100 ratio of soy:cow milk (i.e. the control) was significantly (P<0.05) higher (2.25kg/day) than the 2.18kg/day in calves fed 25:75 ratio of soy:cow milk and 2.22 kg/day in 50:50 ratio of soy:cow milk. Average total DM intake of calves on 50:50 SCM diet (2.22kg/day) was not different from calves on 75:25 ratio of soy:cow milk. Total feed intake (kg) and average daily feed intake (kg/day) also followed similar pattern. The results of the weight changes of the experimental animals in Table 4.6 showed that there was a significant difference (P<0.05) in the final body weight (kg) of calves across the treatments. Calves fed 25:75 ratio of soy:cow milk had higher final weight gain (109.75kg) though not significantly (P>0.05) different from calves fed 50:50 ratio of soy:cow milk (106.6kg).

Similar pattern was also observed in total weight gain (kg), average daily weight gain (kg/day) and feed conversion ratio (FCR). The results of this study revealed that calves fed 25:75 ratio of soy:cow milk had significantly (P<0.05) better live weight gain (0.76 kg/day) and feed conversion ratio of (5.52) indicating better conversion of feed to body tissues compared with 0.67kg/day with FCR of 6.31 in calves fed the control diet and 0.59kg/day with FCR of 7.11 in calves fed 75:25 ratios of soy:cow milk, respectively.

Table 4.4: Chemical Composition of Concentrate Diet and *Digitaria smutsii* Hay.

	Parameters (%)								
Feed Materials	**DM**	**CP**	**CF**	**EE**	**Ash**	**NFE**	**NDF**	**ADF**	**Hemi**
Concentrate Diet	91.65	13.85	14.70	18.88	11.94	32.28	23.43	68.50	42.80
Digitaria smutsii	95.54	7.76	10.50	13.65	8.53	55.10	49.29	63.73	9.88

DM = Dry matter; CP = Crude Protein; CF = Crude Fibre; EE = Ether Extract; NFE = Nitrogen Free Extract; NDF = Neutral Detergent Fibre; ADF = Acid Detergent Fibre; Hemi = Hemicellulose

4.4.2 Effect of Feeding Different Ratios of Soy:Cow Milk on Linear Body Measurements of Friesian x Bunaji Calves

Table 4.7 shows the effect of feeding different ratios of soy:cow milk on linear body measurements of Friesian x Bunaji calves. On an average, total increment in body length were 18.25, 20.25, 17.00 and 21.25 cm for calves fed 0:100, 25:75, 50:50 and 75:25 ratios of soy:cow milk, respectively. While weekly changes in body length of calves fed 0:100, 25:75, 50:50 and 75:25 ratios of soy:cow milk were 1.59, 1.45, 1.14 and 1.66 cm, respectively. Daily changes in body length in the same order for the four treatment groups were 0.23, 0.21, 0.16 and 0.24 cm, respectively.

Total height changes from the beginning up to week 14 were 20.75, 18.50, 16.75 and 20.50 cm for calves fed 0:100, 25:75, 50:50 and 75:25 ratios of soy:cow milk, respectively. Weekly height changes during that time were 1.77, 1.18, 1.30 and 1.68 cm, respectively. On the other hand, daily height changes were 0.25, 0.17, 0.19 and 0.24 cm for calves fed 0:100, 25:75, 50:50 and 75:25 ratios of soy:cow milk, respectively. Total heart girth changes from initial to week 14 of the experiment were 24.25, 23.25, 23.75 and 25.25 cm for calves fed 0:100, 25:75, 50:50 and 75:25 ratios of soy:cow milk, respectively. Weekly heart girth changes during that time were 1.73, 1.66, 1.70 and 1.80cm, respectively. On the other hand, daily heart girth changes were 0.25, 0.24, 0.24 and 0.26 cm for calves fed 0:100, 25:75, 50:50 and 75:25 ratios of soy:cow milk, respectively.

4.4.3 Effect of Feeding Different Ratios of Soy:Cow Milk on Some Haematological Parameters of Friesian x Bunaji Calves.

The result of the effect of feeding different ratios of soy:cow milk on haematological parameters of Friesian x Bunaji calves is presented in Table 4.8. There was significant ($P<0.05$) difference in the packed cell volume (PCV) across the treatments. Calves fed 75:25 ratio of soy:cow milk had higher PCV percenatage (19.53), while calves fed the control diet had the lowest PCV value (15.01%).

Table 4.5: Chemical Composition of Different Ratios of Soy:Cow Milk fed to Calves.

	Ratios of Soy to Cow Milk (%)				
Parameters (%)	**0:100**	**25:75**	**50:50**	**75:25**	**SEM**
Moisture	88.30^{a}	84.80^{b}	83.48^{c}	81.98^{d}	0.21
Total Solids	11.70^{b}	15.20^{ab}	16.52^{a}	18.02^{a}	0.83
Fat	2.69^{b}	3.02^{ab}	3.75^{a}	4.75^{a}	0.53
Solid-Not-Fat	9.01^{b}	12.18^{ab}	12.77^{a}	13.27^{a}	0.39
Protein	3.29^{b}	4.35^{ab}	4.40^{ab}	5.23^{a}	0.67
Ash	0.67^{c}	0.99^{b}	1.08^{b}	1.45^{a}	0.09

^{abcd} means with different superscripts within the same row differed significantly ($P < 0.05$).
SEM = Standard Error of Mean.

Table 4.6: Performance of Friesian x Bunaji Calves fed Different Ratios of Soy:Cow Milk

	Ratios of Soy:Cow Milk (%)				
Parameters	**0:100**	**25:75**	**50:50**	**75:25**	**SEM**
Dry Matter Intake (kg/day)					
Concentrate Intake	1.96[a]	1.92[b]	1.95[a]	1.96[a]	0.01
Digitaria smutsii Hay Intake	0.28	0.26	0.27	0.27	0.01
Total Dry Matter Intake	2.25[a]	2.18[c]	2.22[b]	2.22[b]	0.01
Liquid Feed Intake					
Liquid Feed Intake (L/h/day)	2.00	2.00	2.00	2.00	0.00
Total Feed Intake (kg)	416.29[a]	409.64[c]	413.14[b]	413.91[b]	0.93
ADFI (kg/day)	4.25[a]	4.18[c]	4.22[b]	4.22[b]	0.01
Live Weight Changes					
Initial Weight (kg)	34.50	35.50	34.00	35.25	2.43
Final Weight (kg)	100.50[ab]	109.75[a]	106.00[a]	93.50[b]	3.83
Total Weight Gain (kg)	66.00[b]	74.25[a]	72.00[a]	58.25[c]	2.64
ADWG (kg/day)	0.67[b]	0.76[a]	0.73[ab]	0.59[c]	0.03
Feed Conversion Ratio	6.31[b]	5.52[a]	5.74[ab]	7.11[c]	0.29

[abc] means with different superscripts within the same row differed significantly (P <0.05).
L = Litre; h = Head; ADFI = Average Daily Feed Intake; ADWG = Average daily weight gain;
SEM = Standard Error of Mean.

Table 4.7: Effect of Feeding Different Ratios of Soy:Cow Milk on Linear Body Measurement of Friesian x Bunaji Calves

Parameters Studied (cm)		Ratios of Soymilk to Cow Milk (%)				
		0:100	25:75	50:50	75:25	SEM
Body Length (BL)	Initial	68.00	69.50	70.00	69.50	1.60
	Final	86.25^{b}	89.75^{ab}	87.00^{b}	90.75^{a}	1.47
	Total Change	18.25^{b}	20.25^{c}	17.00^{d}	21.25^{a}	0.13
	Weekly Change	1.59^{b}	1.45^{c}	1.14^{d}	1.66^{a}	0.009
	Daily Change	0.23^{b}	0.21^{c}	0.16^{d}	0.24^{a}	0.001
Height at Withers (HW)	Initial	77.50	76.50	78.00	76.25	1.95
	Final	98.25^{a}	95.00^{ab}	94.75^{b}	96.75^{a}	1.65
	Total Change	20.75^{a}	18.50^{d}	16.75^{c}	20.50^{b}	0.10
	Weekly Change	1.77^{a}	1.18^{d}	1.30^{c}	1.68^{b}	0.007
	Daily Change	0.25 ^{a}	0.17^{d}	0.19^{c}	0.24^{b}	0.001
Heart Girth (HG)	Initial	81.50	84.75	81.75	81.25	2.05
	Final	105.75	108.00	105.50	106.50	1.34
	Total Change	24.25^{ab}	23.25^{b}	23.75^{b}	25.25^{a}	0.71
	Weekly Change	1.73^{a}	1.66^{b}	1.70^{a}	1.80^{a}	0.05
	Daily Change	0.25^{ab}	0.24^{b}	0.24^{b}	0.26^{a}	0.007

^{abcd} means with different superscripts within the same row differed significantly (P<0.05).
SEM = Standard Error of Mean.

The result of white blood cell (WBC) showed significant (P<0.05) difference across treatments. Calves fed 75:25 and 50:50 ratios of soy:cow milk were significantly (P<0.05) higher and those fed 0:100 and 25:75 ratios of soy:cow milk had significantly (P<0.05) lower and similar WBC. Haemoglobin concentration also followed similar pattern. While for MCHC, calves fed 0:100 and 25:75 ratios of soy:cow milk had significantly (P<0.05) higher and similar MCHC of 29.91 and 28.58g/dl, respectively followed by (25.08g/dl) in calves fed 50:50 ratio of soy:cow milk, the least MCHC value (21.66g/dl) was recorded in calves fed 75:25 ratio of soy:cow milk.

4.4.4 Effect of Feeding Different Ratios of Soy:Cow Milk on Some Serum Biochemical Indices of Friesian x Bunaji Calves

The results of feeding different ratios of soy:cow milk on some serum biochemical indices of Friesian x Bunaji calves are presented in Table 4.9. Serum total protein differed significantly (P<0.05) across the treatments and was highest (6.32g/dl) in calves fed 75:25 ratio of soy:cow milk though not statistically different from calves fed 50:50 (6.28g/dl) and 0:100 (6.12) ratios of soy:cow milk. The lowest serum protein (5.51g/dl) was observed in calves fed 25:75 ratio of soy:cow milk. Albumin, glucose and creatinine also followed similar pattern. But the amount of globulin was highest (2.44g/dl) in calves fed 0:100 ratio of soy:cow milk (the control) and lowest in calves fed 75:25 ratio of soy:cow milk though not signifcantly (P>0.05) different from calves fed 25:75 and 50:50 ratio of soy:cow milk. There was a significant (P<0.05) difference in blood urea nitrogen (BUN) across the treatments. Calves fed 50:50 ratio of soy:cow milk had the highest BUN of (9.79mg/dl) while calves fed 25:75 ratio of soy:cow milk had the lowest BUN concentration of (7.13mg/dl) and was statistically similar with calves fed the control diet (7.60mg/dl).

Table 4.8: Effect of Feeding Different Ratios of Soy:Cow Milk on Some Haematological Parameters of Friesian x Bunaji Calves.

	Ratios of Soymilk to Cow Milk (%)					
Parameters	**0:100**	**25:75**	**50:50**	**75:25**	**SEM**	**Range[1]**
PCV (%)	15.01^{c}	18.67^{ab}	17.67^{b}	19.53^{a}	0.85	24 – 46
WBC (x10^{9}/l)	10.28^{b}	10.66^{b}	11.65^{a}	12.46^{a}	0.49	4 – 12
RBC (x10^{12}/l)	6.52	7.22	6.44	7.40	0.51	0 – 20
Hgb (g/dl)	9.81^{b}	9.61^{b}	10.43^{a}	10.78^{a}	0.25	8 – 15
MCV (fl)	49.93^{b}	49.15^{b}	54.55^{a}	50.27^{b}	0.71	40 – 60
MCH (pg)	15.47^{a}	15.36^{ab}	15.18^{b}	15.91^{a}	0.25	11 – 17
MCHC (g/dl)	29.91^{a}	28.58^{a}	25.08^{b}	21.66^{c}	1.02	30 – 36
Differential WBC (%)						
Lympocytes	68.88^{a}	66.44^{ab}	67.08^{a}	64.78^{b}	1.48	45 – 75
Neutrophils	26.82^{a}	18.11^{c}	20.97^{b}	24.40^{a}	1.42	15 – 33
Basophils	0.68^{b}	0.94^{a}	0.81^{ab}	1.10^{a}	0.08	0 – 2

^{abc} means with different superscripts within the same row differed significantly (P <0.05).

PCV = Packed Cell Volume; WBC = White Blood Cells; RBC = Red Blood Cells; Hgb = Haemoglobin;

MCV = Mean corpuscular volume; MCH = Mean corpuscular haemoglobin;

MCHC = Mean corpuscular haemoglobin concentration; SEM = Standard Error of Mean.

[1]**Source:** Merck Veterinary Manual (2016).

Table 4.9: Effect of Feeding Different Ratios of Soy:Cow Milk on Serum Biochemical Indices of Friesian x Bunaji Calves

	Ratios of Soy to Cow Milk (%)					
Serum Indices	**0:100**	**25:75**	**50:50**	**75:25**	**SEM**	**Range[1]**
Total Protein (g/dl)	6.12^{a}	5.51^{b}	6.28^{a}	6.32^{a}	0.17	6.7 – 7.5
Albumin (g/dl)	3.68^{b}	3.34^{c}	4.11^{a}	4.32^{a}	0.15	2.5 – 3.8
Globulin (g/dl)	2.44^{a}	2.17^{b}	2.17^{b}	2.00^{b}	0.09	3.0 – 3.5
Glucose (mg/dl)	81.72^{b}	73.87^{c}	76.38^{bc}	96.20^{a}	2.87	40 – 100
Blood Urea Nitrogen (mg/dl)	7.60^{c}	7.13^{c}	9.79^{a}	8.99^{b}	0.30	10 – 25
Creatinine (mg/dl)	0.47^{b}	0.46^{b}	0.60^{a}	0.58^{a}	0.03	0.5 – 2.2

^{abc} means with different superscripts within the same row differed significantly ($P < 0.05$).

g/dl = gram per decilitre; mg/dl = milligram per decilitre; SEM = Standard Error of Mean.

[1]**Source:** Merck Veterinary Manual (2016).

4.4.5 Effect of Feeding Different Ratios of Soy:Cow Milk on Mean Rumen Metabolites of Friesian x Bunaji Calves.

The result of the effect of feeding different ratios of soy:cow milk on mean rumen metabolites in Friesian x Bunaji calves is presented in Table 4.10. There was no significant difference ($P>0.05$) in the rumen fluid temperature and pH across the treatments. The rumen ammonia nitrogen (RAN) significantly ($P<0.05$) differ among treatment diets with 25:75 ratio of soy:cow milk having the lowest (11.34 mg/100ml) though not statistically different from animals fed 50:50 ratio of soy:cow milk but animals on 75:25 ratio of soy:cow milk the highest RAN value (13.84 mg/100ml) (Figure 4.1). The RAN value (12.70 mg/100ml) obtained in calves fed 0:100 ratio of soy:cow milk was significantly ($P<0.05$) higher than animals fed 25:75 and 50:50 ratios of soy:cow milk.

The mean values for TVFA across treatments showed that calves fed 0:100 ratio of soy:cow milk had the least (26.63 mmol/l) TVFA concentration, though not significantly ($P>0.05$) different from calves on 25:75 ratio of soy:cow milk. Statistically, animals fed 50:50 and 75:25 ratios of soy:cow milk had similar TVFA concentration (31.31 and 31.69 mmol/l), respectively though not significantly ($P>0.05$) higher than animals fed 25:75 ratio of soy:cow milk (29.25 mmol/l). The result of this study showed increase TVFA concentration as the ratio of soymilk in the diets increased from 0 to 75% (Figure 4.1).

4.4.6 Effect of Sampling Time (Hours) on Rumen Fluid Temperature, pH, Ammonia and TVFA of Friesian x Bunaji Calves fed Different Ratios of Soy:Cow Milk.

The result of the effect of sampling time on mean rumen metabolites in Friesian x Bunaji calves fed different ratios of soy:cow milk is presented in Table 4.11. Sampling time significantly ($P<0.05$) influenced rumen temperature, pH, NH_3-N and TVFA concentrations. The rumen temperature 6 hours post feeding (29.22 ^oC) was significantly ($P<0.05$) higher than the values obtained at 0, 3 and 9 hours post feeding (18.91, 25.66 and 24.31 ^oC), respectively (Figure 4.5).

Table 4.10: Effect of Feeding Different Ratios of Soy:Cow Milk on Rumen Fluid Temperature, pH, Ammonia and TVFA of Friesian x Bunaji Calves.

	Ratios of Soy to Cow Milk (%)				
Rumen Metabolites	**0:100**	**25:75**	**50:50**	**75:25**	**SEM**
Temperature (^{o}C)	24.13	24.31	25.13	24.53	0.50
pH	6.93	6.81	6.92	6.88	0.11
NH_3-N (mg/100ml)	12.70^{b}	11.34^{c}	11.78^{c}	13.84^{a}	0.37
TVFA (mmol/l)	26.63^{b}	29.25^{ab}	31.31^{a}	31.69^{a}	1.15

^{abc} means with different superscripts within the same row differed significantly ($P < 0.05$).

NH_3-N = Rumen Ammonia Nitrogen; TVFA = Total Volatile Fatty Acids; ^{o}C = Degree Centigrade; SEM = Standard Error of Mean

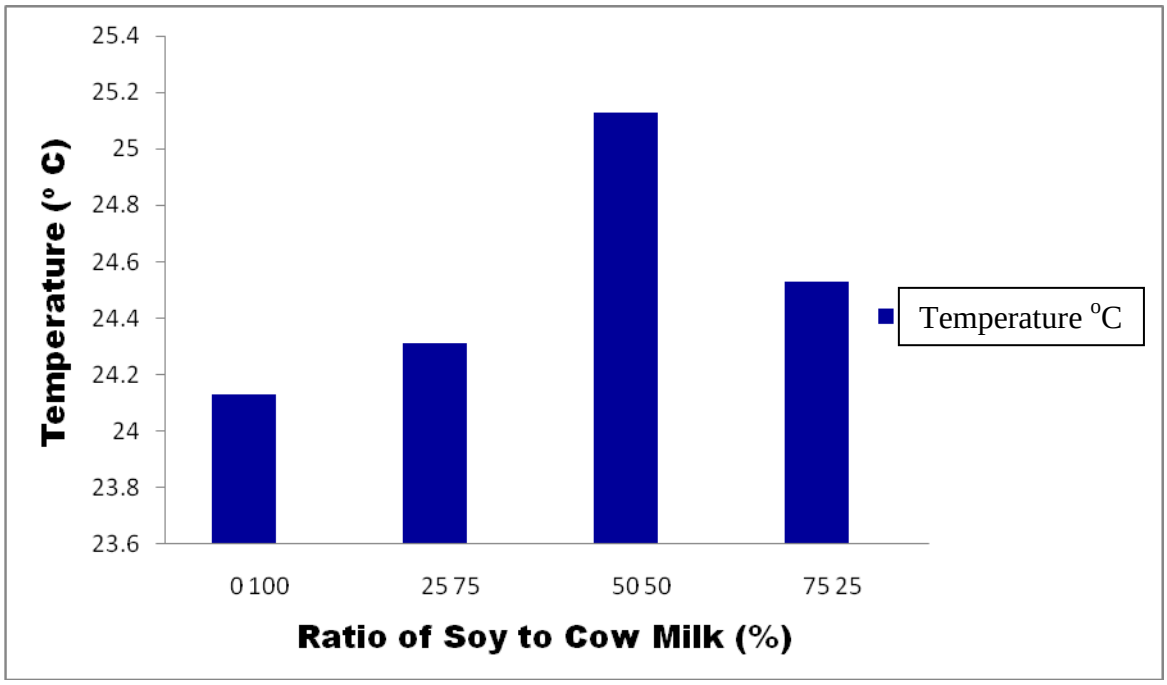


Figure 4.1: Effect of Feeding Different Ratios of Soy:Cow Milk on Rumen Fluid Temperaturevof Friesian x Bunaji Calves.

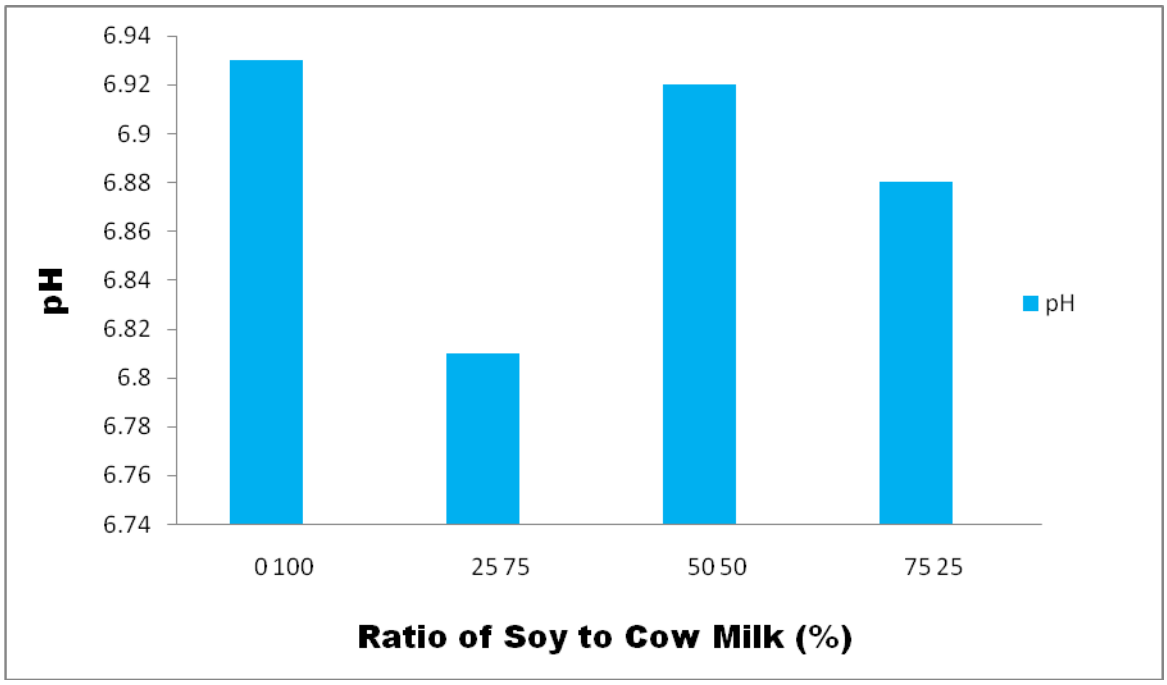


Figure 4.2: Effect of Feeding Different Ratios of Soy:Cow Milk on Rumen Fluid pH of Friesian x Bunaji Calves.

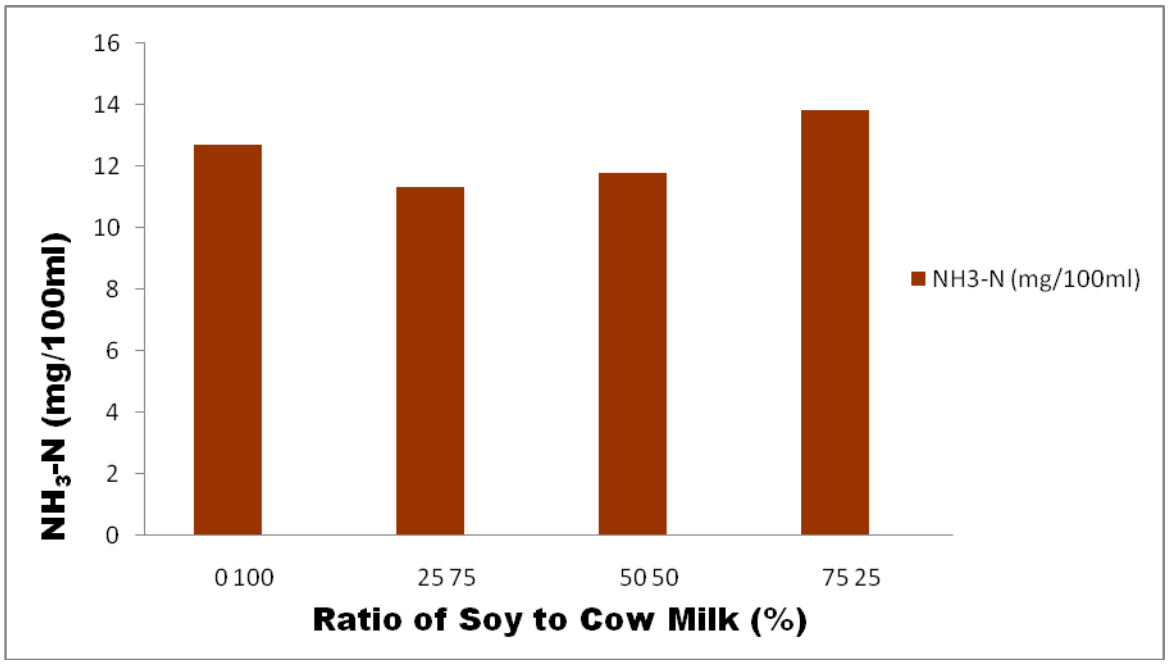


Figure 4.3: Effect of Feeding Different Ratios of Soy:Cow Milk on NH_3-N Concentration of Friesian x Bunaji Calves.

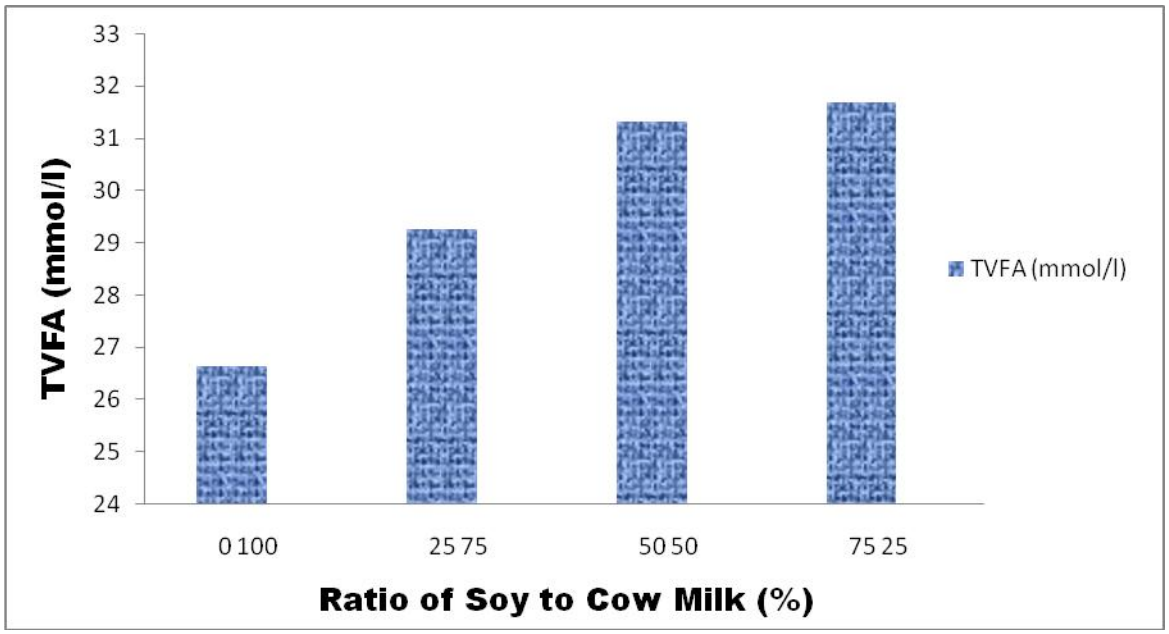


Figure 4.4: Effect of Feeding Different Ratios of Soy:Cow Milk on TVFA Concentration of Friesian x Bunaji Calves.

Rumen pH at 3, 6 and 9 hours (7.04, 6.28 and 6.14, respectively) post feeding were significantly ($P<0.05$) lower compared to the result obtained at 0 hour (8.08) post feeding (Figure 4.6). The effect of sampling time on rumen NH_3-N concentration at 3 and 9 hours (13.44 and 13.57 mg/100ml, respectively) post feeding were statistically similar but significantly ($P<0.05$) higher compared to the results obtained at 0 and 6 hours (11.08 and 11.56 mg/100ml, respectively) post feeding (Figure 4.7). The effect of sampling time on total volatile fatty acids concentration revealed an increase in TVFA concentration as the sampling time increased from 0 hour (22.00 mmol/l), 3 hours (24.69 mmol/l) up to 6 hours (37.00 mmol/l) post feeding, respectively. There was a decline in TVFA concentration at 9 hours (35.19 mmol/l) post feeding (Figure 4.8).

4.4.7 Nutrient Digestibility and Nitrogen Balance in Friesian x Bunaji Calves fed Different Ratios of Soy:Cow Milk.

The result of the nutrient digestibility and nitrogen balance in Friesian x Bunaji calves fed different ratios of soy:cow milk is presented in Table 4.12. Dry matter (DM) digestibility was higher in calves fed soy:cow milk at 25:75 (49.74%) and 50:50 (48.07%). The lowest DM digestibility was observed in calves fed 75:25 (45.34%) ratio of soy:cow milk. Organic matter (OM) digestibility was statistically higher in calves fed 75:25, 50:50 and 25:75 ratios of soy:cow milk, while calves on 0:100 ratio of soy:cow milk had significantly lowest OM digestibility (58.13%) compared to calves on the other dietary treatments. Crude protein (CP) digestibility was significantly higher (69.70%) in calves fed 25:75 ratio of soy:cow milk. Statistically, calves on the control diet had the lowest CP digestibility (62.94%). The digestibility of neutral detergent fibre (NDF) and acid detergent fibre (ADF) followed similar trend. The results of nitrogen intake showed significant ($P<0.05$) difference across treatments. Calves fed 25:75 and 50:50 ratios of soy:cow milk had significantly ($P<0.05$) higher and similar nitrogen intake. While those fed 75:25 and 0:100 (control) were statistically similar and lower.

Table 4.11: Effect of Sampling Time on Rumen Fluid Temperature, pH, Ammonia and TVFA of Friesian x Bunaji Calves.

	Sampling Time (Hours)				
Rumen Metabolites	**0**	**3**	**6**	**9**	**SEM**
Temperature (^{o}C)	18.91^{d}	25.66^{b}	29.22^{a}	24.31^{c}	0.39
pH	8.08^{a}	7.04^{b}	6.28^{c}	6.14^{c}	0.09
NH_3-N (mg/100ml)	11.08^{c}	13.44^{a}	11.56^{b}	13.57^{a}	0.22
TVFA (mmol/l)	22.00^{b}	24.69^{b}	37.00^{a}	35.19^{a}	1.63

^{abc} means with different superscripts within the same row differed significantly ($P < 0.05$).

NH_3-N = Rumen Ammonia Nitrogen; TVFA = Total Volatile Fatty Acids; ^{o} C = Degree Centigrade; SEM = Standard Error of Mean

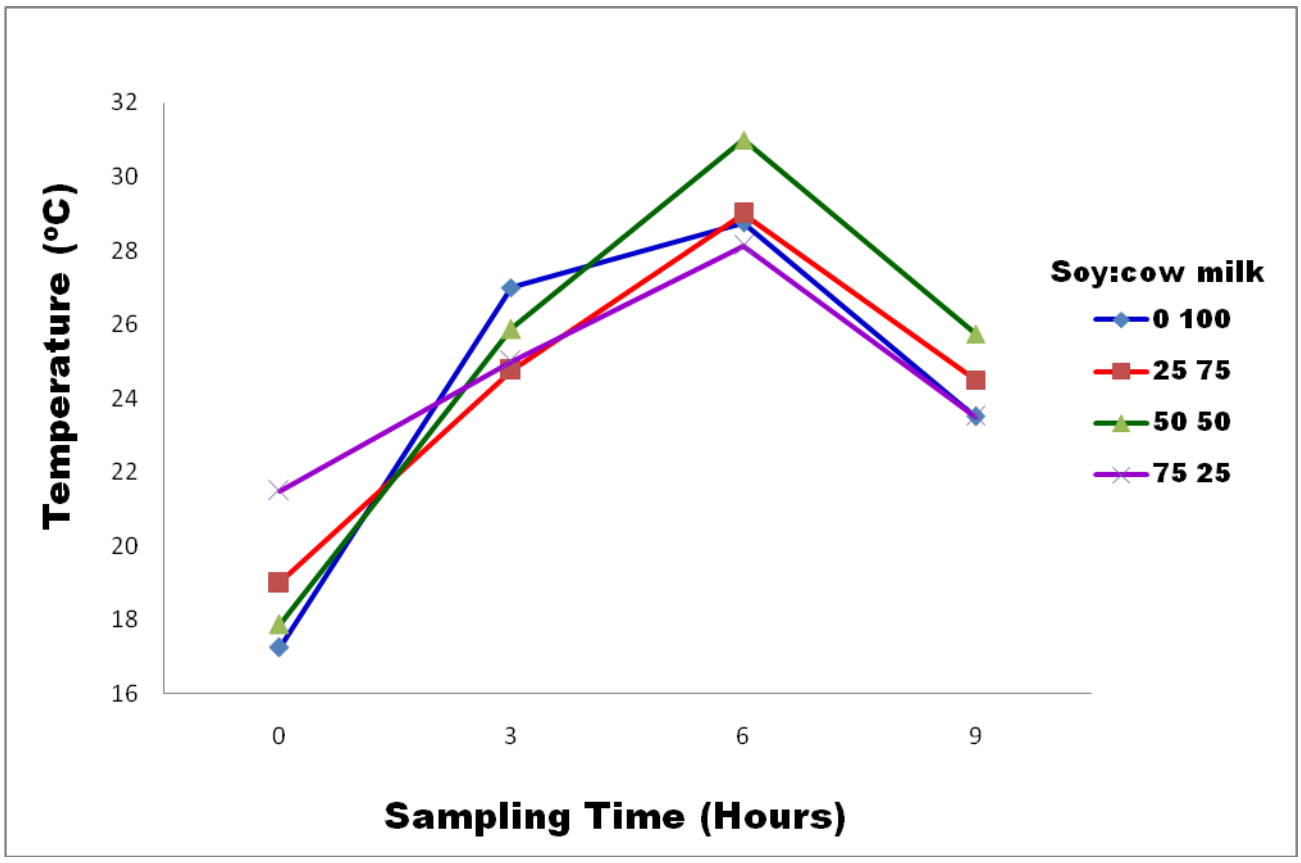


Figure 4.5: Trend of Temperature (^{o}C) of Rumen Fluid of Calves fed Different Ratios of Soy:Cow Milk.

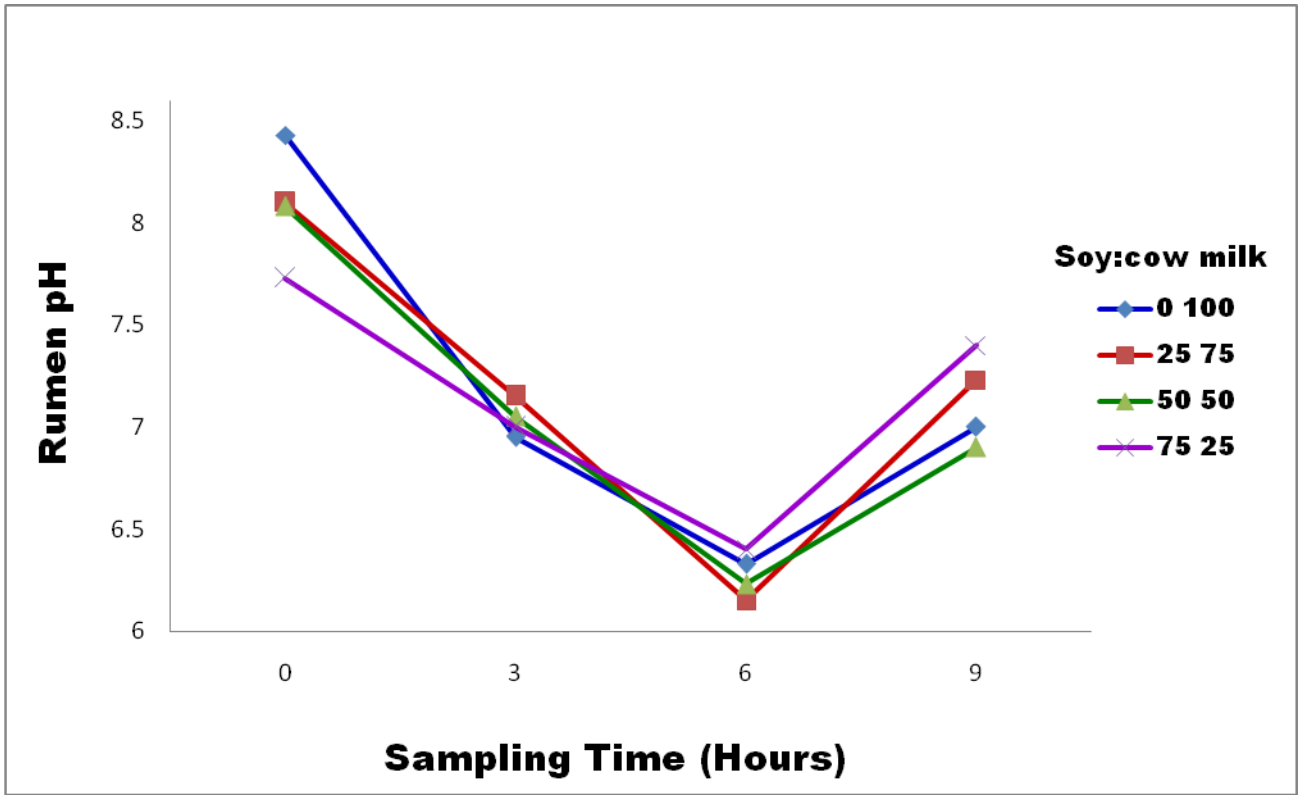


Figure 4.6: Trend of pH in the Rumen Fluid of Calves fed Different Ratios of Soy:Cow Milk.

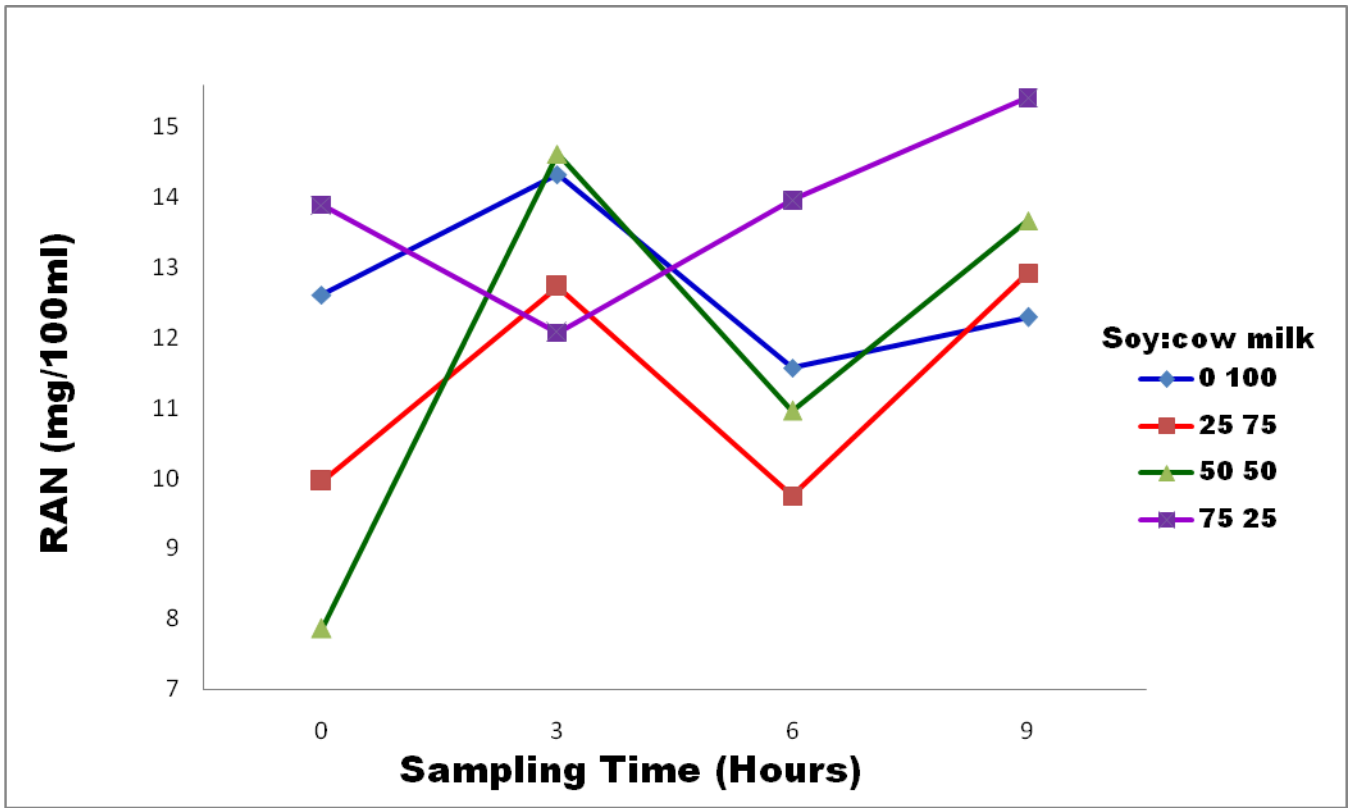


Figure 4.7: Trend of NH_3-N Concentration (mg/100ml) of Calves fed Different Ratios of Soy:Cow Milk.

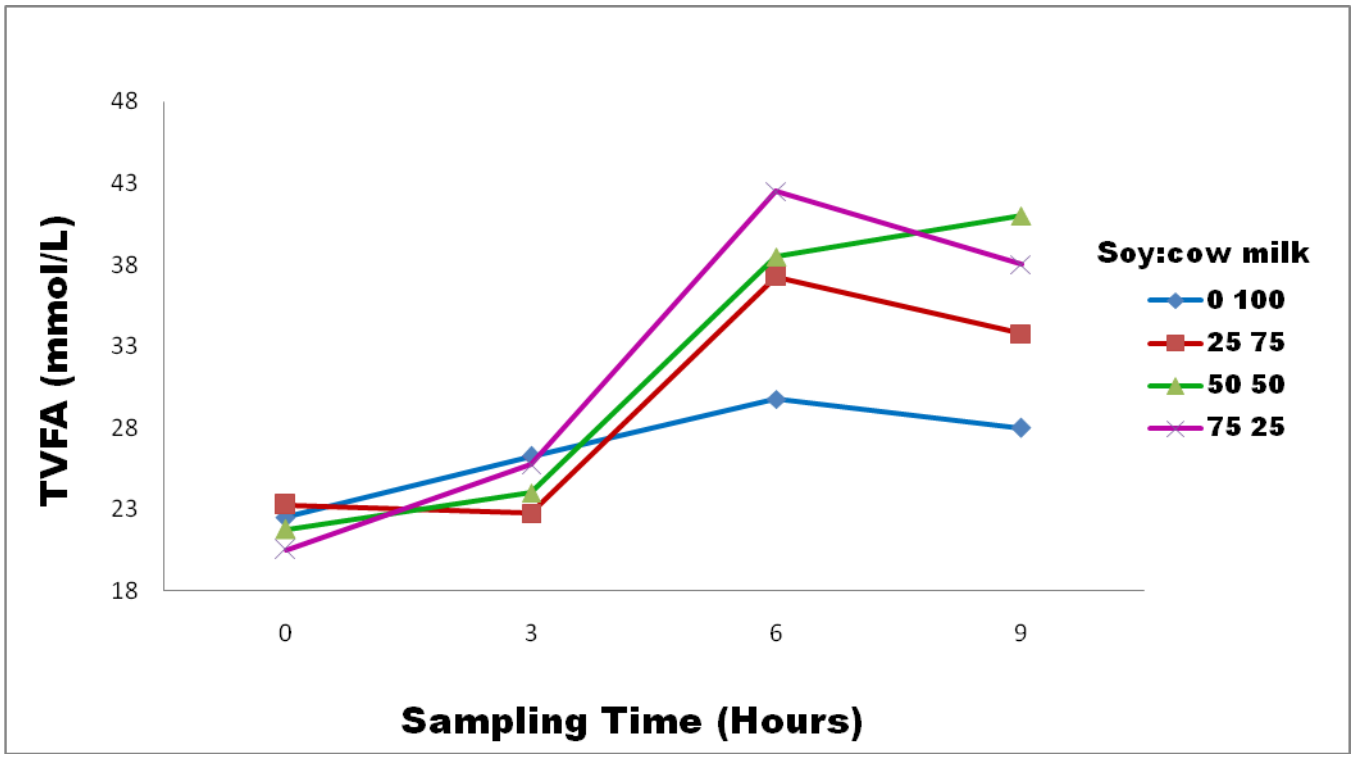


Figure 4.8: Trend of TVFA Concentration (mmol/l) of Calves fed Different Ratios of Soy:Cow Milk.

Urinary nitrogen output did not show any significant (P>0.05) difference across treatments. But faecal nitrogen and total nitrogen output decreased significantly (P<0.05) with increased levels of soymilk in the diets. Calves fed 75:25, 50:50 and 25:75 ratios of soy:cow milk had statistically similar and high (47.79, 42.53 and 40.84 g/day), respectively with those fed 75:25 ratio of soy:cow milk having the highest (47.79 g/day) nitrogen retention. While those fed 0:100 ratio of soy:cow milk (control) had the lowest (33.67 g/day) nitrogen retention.

4.5 Economic Benefits of Feeding Different Ratios of Soy:Cow Milk to Friesian x Bunaji Calves.

The economic analysis of feeding different ratios of soy:cow milk to Friesian x Bunaji calves is presented in Table 4.13. The cost of soy:cow milk ration decreased with increase in the soymilk component. Cost of 0:100 soy:cow milk was ₦ 19, 208.00. While the cost of 25:75, 50:50 and 75:25 soy:cow milk rations were ₦ 16, 023.00, ₦ 12, 838.00 and ₦ 9, 653.00, respectively. Cost of concentrate and *Digitaria smutsii* hay per kg ranged from ₦ 14.71 to ₦ 14.72 and ₦ 5.99 to ₦ 6.01, respectively. Statistical analysis showed that calves fed 0:100 soy:cow (control) had significantly (P<0.05) highest total cost of feed consumed (₦40, 201.11) and those fed 75:25 had the significantly (P<0.05) lowest total cost of feed consumed (₦30, 489.00). The result of feed cost per kg weight gain showed significantly (P<0.05) difference across treatments. Calves fed 0:100 ratio of soy:cow milk had significantly (P<0.05) higher (₦627.62/kg) and those fed 25:75 and 50:50 had significantly (P<0.05) lower (₦ 496.10/kg and ₦ 469.67/kg) and similar feed cost per kg weight gain. Percent feed cost reduction was significantly (P<0.05) higher (24.16%) in calves fed 75:25 followed by (16.37%) in calves fed 50:50 ratio of soy:cow milk. The best percent feed cost reduction was recorded in calves fed 25:75 ratio of soy:cow milk (9.32%). The results of

the ecomomic analysis in this study showed that it was economical to use soymilk at different ratios in the diets of dairy calves compared to feeding cow milk alone.

Table 4.12: Nutrient Digestibility and Nitrogen Balance in Friesian x Bunaji Calves Fed Different Ratios of Soy:Cow Milk.

	Ratios of Soy to Cow Milk (%)				
Digestibility Coefficient (%)	**0:100**	**25:75**	**50:50**	**75:25**	**SEM**
Dry Matter	46.35^{ab}	49.74^{a}	48.07^{a}	45.34^{b}	1.82
Organic Matter	58.13^{b}	64.27^{a}	66.93^{a}	68.01^{a}	1.90
Crude Protein	62.94^{c}	69.70^{a}	65.65^{b}	67.05^{b}	1.22
NDF	57.81^{b}	59.61^{a}	54.43^{c}	55.78^{c}	0.74
ADF	60.62^{c}	63.15^{a}	61.55^{b}	59.27^{d}	0.42
Nitrogen Balance (g/day)					
Nitrogen Intake	74.36^{b}	78.90^{a}	76.32^{ab}	75.89^{b}	1.35
Urinary Nitrogen	5.49	5.24	4.86	3.42	1.98
Faecal Nitrogen	35.20^{d}	32.82^{c}	28.93^{b}	24.68^{a}	0.59
Total Nitrogen Output	40.69^{d}	38.06^{c}	33.79^{b}	28.10^{a}	1.29
Nitrogen Retained	33.67^{b}	40.84^{a}	42.53^{a}	47.79^{a}	3.49
Nitrogen Retained (% N-Intake)	45.28^{b}	51.76^{b}	55.73^{ab}	62.97^{a}	4.44

^{abcd} means with different superscripts within the same row differed significantly (P <0.05).

NDF = Neutral Detergent Fibre; ADF = Acid Detergent Fibre; g =gram; N = Nitrogen; % = Percentage; SEM = Standard Error of Mean.

Table 4.13: Economic Analysis of Feeding Different Ratios of Soy:Cow Milk to Calves.

	Ratios of Soy to Cow Milk (%)				
Parameters	**0:100**	**25:75**	**50:50**	**75:25**	**SEM**
Cost of Concentrate Consumed (₦)	2831.00[a]	2764.45[c]	2812.27[b]	2818.89[ab]	8.76
Cost of *Digitaria* Consumed (₦)	167.25[a]	154.35[b]	156.45[b]	157.80[b]	3.79
Cost of Liquid Feed Consumed (₦)	19208.00[a]	16023.00[b]	12838.00[c]	9653.00[d]	919.43
Total Cost of Feed Consumed (₦)	40201.11[a]	36454.58[b]	33619.03[c]	30489.00[d]	925.44
Feed Cost/Kg Weight Gain (₦/Kg)	627.62[a]	496.10[bc]	469.67[c]	534.46[b]	25.13
% Feed Cost Reduction	-	9.32[c]	16.37[b]	24.16[a]	3.10

[abcd] means with different superscripts within the same row differed significantly (P <0.05).
Cost of Concentrate per kg = ₦ 14.72; Cost of *Digitaria smutsii* per kg = ₦ 6.01; Cost of Cow Milk per litre = ₦ 98; Cost of Soymilk per litre = ₦ 33; Kg = Kilogram; L = Litre; ₦ = Naira; % = Percent;
SEM = Standard Error of Mean.

CHAPTER FIVE

5.0 DISCUSSION

5.1 Stage One of the Experiment: Effect of Soaking Duration on Nutritional and Anti-Nutritional Compositon of Soybeans.

5.1.1 Chemical Composition of Soybeans at Different Soaking Duration (on DM basis).

The chemical composition of soybeans at different duration of soaking was comparable to values earlier reported by Pele *et al.* (2016). The high DM content observed in all the samples could be attributed to adequate drying of the soybean seeds after soaking which reduced the moisture content. Moisture content is very important in food storage and processing. Moisture affects the shelf life and nutrition of food products. Dari (2009) reported that soyflour should have a low moisture content to avoid contamination by mycotoxins and fungus. Aruah *et al*. (2012) stated that the moisture content of any food is an index of its water activity and is used as a measure of stability and susceptibility to microbial contamination, indicating that soybeans can be processed into flour after soaking and drying and the soyflour could be kept in a polythene bag for sometime without microbial spoilage and deterioration in quality.

There was an increase in crude protein content with increase duration of soaking which could be attributed to complex biochemical changes that occur during hydration which led to breaking down of protein constituent by enzymes into simple compounds that are used to make new compounds (Maidala *et al*., 2013). Similar results were reported by Pele *et al.* (2016) for increase in crude protein content which was attributed to sprouting which increased the bioavailability of protein content in the soybeans. The reduction in protein content in soybeans soaked for 24 hours compared with soybeans soaked for 72 hours

could be attributed to leaching of amino acids, which are the building blocks of proteins as reported by (Maidala *et al*., 2013). However, the results of this study showed that soaking did not reduce the crude protein content significantly.

The increased fat content observed in this study was attributed to the synthesis of fatty acid as a result of soaking the soybeans in water. This supports the report of Hanssen (2003) who observed increase in fat content after 12 hours of soaking soybeans. But is different from the report of Sharma *et al*. (2011) who observed decrease in fat content of sprouted soybeans which was attributed to the utilization of fat as energy source during sprouting period or the hydrolysis of fat to glycerol and fatty acids in the presence of lipolytic enzymes. However, soaking decreased the crude fibre, ash and carbohydrate contents of the soybeans. The decreased in carbohydrate might be due to increase in alpha-amylase activity.

The alpha-amylase breaks down complex carbohydrates to simple and more absorbable sugars, which are utilized by the seeds during the early stages of soaking, thereby resulting to loss in dry matter content (Sarker *et al*., 2015). Legumes have been reported to be good sources of ash (Aruah *et al*., 2012). Ash content refers to the inorganic substance which translates into high mineral contents (Sharma *et al*., 2011). Soaking reduced the ash content of the soybeans which could be as a result of biochemical reaction. As the content of CF and NFE reduced from the soybean due to leaching of soluble carbohydrates (oligosaccharides), the other components such as crude protein and fat increased, thus improving the nutritional value of the soymilk. Soybean processing treatments like soaking was reported to substantially reduce the content of fermentable, soluble carbohydrates (Maidala *et al*., 2013). The higher fat contents in soybeans soaked for 48 and 72 hours might be responsible for the higher values of metabolizable energy (5631.61 and 5514.57 kcal/kg DM, respectively), obtained in this study.

5.1.2 Chemical Composition of Friesian x Bunaji Milk and SoyMilk (Wet basis).

Milk is a complex mixture of fat, proteins, carbohydrate, minerals, vitamins and other miscellaneous constituents dispersed in water (Harding, 1999). Milk composition including moisture, protein, fat, total solid and solid-non fat are an important indicators of milk quality (Alabi, 2005). The percentage of total solid obtained for Friesian x Bunaji milk was lower than the value reported by (Ahmed *et al*., 2002) in Holstein Friesian cow milk in Pakistan. A similar result was obtained by (Ibeawuchi and Dalyop, 1995) in White Fulani cow milk. The low value recorded for total solid content in this study when compared with other researches may be due to low nutrition and other management practices adopted. The milk protein content of Friesian x Bunaji milk obtained in this study was comparable with values reported by (Ndubueze *et al.*, 2006) for grazing White Fulani cows. And even for Friesian x Bunaji cattle in Nigeria (Ibeawuchi and Umoh, 1990). The total solids of cow milk obtained in this study was slightly below the value reported by (Sarker *et al*., 2015) which might be due to the difference in breed and feed.

The milk fat content obtained in this study was slighlty lower than the value reported by Roy *et al.* (2016). Higher milk fat was also reported by Sarker *et al*. (2015). The low milk fat content observed in this study might be attributed to age of cows from which the milk was collected, breed of the cows, stage of lactation and nutrition. The breed of cow whose milk were used were a cross between tropical (Bunaji or White Fulani) and temperate (Holstein Friesian), so the milk fat will not be as high as those from pure breeds of Friesian or other dairy cattle like Jersey. Fayeye *et al*. (2013) reported that Jersey breed of dairy cattle had higher milk fat than Holstein Friesian breed. Egbowon (2004) reported that milk fat content decreases at mid-lactation and continue to decrease until the end of lactation.

Also, underfeeding can cause lower proportion of milk components. Similarly, Nickerson (1999) reported that genetic factor, nutrition, environment and milking management practices have important effect on milk composition and quality.

The protein content of the 72 hours soaked soymilk observed in this study on wet basis (5.55%) was slightly above the value (4.0%) reported by Sarker *et al*. (2015) for soymilk obtained by boiling soybeans at 100 ^{o}C for 15 minutes. But similar to the values of (4.5%) reported by Odumodu (2010) for soybeans soaked for 18 hours. The total solids (12.89%), fat (3.30%) and ash (0.82%) obtained in this study for soymilk were slightly higher than the (10.70%), (2.70%) and (0.60%) reported by Sarker *et al*. (2015) for total solids, fat and ash, respectively. This study found that soaking increased the protein content of the soybeans. This is consistent with the report of Pele *et al*. (2016) who found that soaking increased the CP content of soybeans soaked for 12 and 24 hours.

5.1.3 Anti-Nutritional Factors in Soybean at Different Duration of Soaking (DM basis).

Anti-nutritional factors are compounds which reduce the nutrient utilization and/or feed intake of plants or plant products used as animal feeds. The major anti- nutrients found in soybeans are phytates, protease inhibitors, tannins, oxlates and saponins (Tadele, 2015). The range of phytates (0.40 to 4.39 mg/100ml) observed in this study is lower than the levels (5.45 to 8.05%) reported by Pele *et al*. (2016) for soybeans soaked for 12, 24 and 72 hours. The reduction of phytates observed in this study could be due to leaching and as the seeds were preparing for sprouting. Osman (2007) has reported reduction in phytic acid content in soybeans subjected to different processing methods (soaking, cooking and germination) which was attributed to leaching in water. Processing of soybeans by soaking in water was effective in enhancing the proximate composition and reducing anti-nutritional factors of soybean. Mikic *et al*. (2009) reported that phytic acid content in soybean seeds and products was about 1 – 1.5% of dry matter. The range of levels for

protease inhibitors (3.79 to 7.09 mg/100ml) observed in this study was within the range of (4 – 8mg/100g) and (4.91 to 7.00%) reported by Peisker (2001) and Pele *et al*. (2016), respectively for soybeans soaked in water.

However, the range of tannins (0.18 to 0.42 mg/100ml) reported in this study is much lower than the (19.23 to 25.23 mg/100g) reported by Pele *et al*. (2016) for soybeans soaked for 12, 28 and 72 hours. The difference could be due to the variety of soybean used and the soil on which it was cultivated which could have influenced its chemical composition. Protease inhibitors, tannins, oxlates and saponins also followed similar pattern. This showed soaking had decreased the ANFs of the soybeans which implies improvement in the nutritional quality of the soybeans for effective utilization by the animals. The soybeans with the least level of ANFs are the ones soaked for 72 hours. This implies that as the duration of soaking increases, the anti-nutritional content in the soybeans decreases. This is in line with the report of Mikic *et al*. (2009) who observed that, amongst the various processing methods (boiling, toasting and soaking*)* employed on soybeans, soaking method was found to reduce the levels of various anti-nutritional substances. This also agress with the report of Liener (1994) who found that phytates can be reduced through soaking or other forms of processing.

Duration of soaking had profound effect on anti-nutritional levels of the soybeans. The highest percent reduction in ANFs was recorded in soybeans soaked for 72 hours which could be attributed to leaching and as the seeds were preparing to germinate. Germination had earlier been reported to lower the levels of ANFs in legume seeds depending upon the type of bean and germinating conditions (Akpanum and Achinewhu, 1985). The extent of the reduction of ANFs in soybeans also depends on the species, pH and length and conditions of soaking. Liu and Markakis (1987) also observed that soaking of soybeans in water at 22^{o}C for 24 hours had no effect on the ANFs. Therefore, the longer the period of soaking the lesser the level of ANFs.

5.2 Chemical Composition of Concentrate Diet and *Digitaria smutsii* Hay.

The dry matter (95.54%) content of *Digitaria smutsii* hay observed in this study was higher than (90.40%) reported by Yashim (2014) but slightly higher than (94.29%) reported by Yusuf (2016). The high DM content observed in this study could be attributed to the season of harvest and the curing process and storage. The crude protein content (7.76%) observed in this study for *Digitaria smutsii* hay was higher than (5.36%) reported by Yashim (2014) but slightly lower than (9.63%) reported by Yusuf (2016). Crude protein content of hay is dependent on the stage at which the hay was harvested, duration of curing and also the leaf to stem ratio. The neutral detergent fibre (49.29%) is lower than (69.23%) reported by Yashim (2014).

However, the acid detergent fibre (63.73%) observed for digitaria hay in this study was higher than the (33.88%) reported by Yashim (2014). The differences in the chemical composition of the *Digitaria smutsii* hay could be attributed to variation in the nutrient content of the soil, weather, stage of maturity, method of harvest, curing and also storage. The crude protein content (13.85%) of the concentrate diet in this study was similar to 13.88% reported by Iriso *et al*. (2016) for Friesian x Bunaji crossbred heifers. Whilk the NDF (23.43%) was lower than (35.13%) reported by Iriso *et al*. (2016) but ADF (68.50%) reported in this study was higher than 23.68% reported by Iriso *et al*. (2016). The differences in NDF and ADF observed could be attributed to the type of nature of the ingredient used in the ration formulation. And could also be as a result of human error during the laboratory analysis.

5.3 Chemical Composition of Different Ratios of Soy:Cow Milk fed to Calves.

The mixture of soy:cow at different ratios showed significant ($P<0.05$) increase in nutritional content as compared with cow milk or soymilk alone. The similar fat content in the diets containing different ratios of soymilk might be attributed to the low fat content in cow milk which had not affect the total fat content in the mixture much. However, the protein content of the mixture of soymilk and cow milk were higher than those of cow milk alone. The non-significant ($P>0.05$) difference observed in the protein contents of the diet containing soymilk in addition to cow milk might be due to the low protein content of cow milk used. Other authors reported higher fat and protein contents in cow milk but the low levels obtained in this study may be attributed to breed of cow, feeding and stage of lactation.

5.4 Stage Two of the Experiment: Growth Performance of Friesian x Bunaji Calves fed Different Ratios of Soy:Cow Milk.

5.4.1 Effect of Feeding 72 hrs Soaked Soymilk and Cow milk on Intake and Live Weight Changes of Friesian x Bunaji Calves fed *Digitaria smutsii* Hay as Basal Diet.

The total dry matter intake observed in this study which ranged between 2.18 to 2.25 kg/day was higher than the range (0.47 to 0.56 kg/day) reported by Roy *et al*. (2016) for calves fed soybeans based milk replacer. The higher DMI observed in this study could be attributed to the inclusion levels of soymilk in the diets of the calves which stimulate intake. This finding showed that feeding different ratios of soy:cow milk stimulated solid feed intake in the calves. Reducing milk feeding had been reported to hasten intake of starter feeds and promotes rumen development in calves (Hussain *et al*., 2009). The similarities observed in the total dry matter intake in this study could be attributed to group

housing of the calves. Group housing was shown to influence calf feed intake in comparison with individual housing (Montoro *et al*., 2013).

The total weight gain of 66.00kg and 58.25kg obtained in this study for calves fed cow milk alone and 75:25 ratio of soy:cow milk were comparable with 69.50kg and 59.00kg reported by Jasper and Weary (2002) for Holstein calves fed cow milk *ad libitum* and conventional milk replacer, respectively. The higher total weight gain of 74.25kg and 72.00kg obtained in this study for calves fed 25:75 and 50:50 ratios of soymilk and cow milk could be attributed to better feed utilization as result of low consumption of ANFs.

The average daily weight gain obtained in this study which range from 0.59 to 0.76 kg/day was higher than the range of 0.18 to 0.31 kg/day observed by Roy *et al*. (2016). However, the ADWG (0.76 kg/day and 0.59 kg/day) obtained in this study for calves fed 25:75 and 75:25 ratios of soymilk and cow milk, respectively were similar to 0.78 kg/day and 0.48 kg/day reported by Jasper and Weary (2002) for Holstein calves fed cow milk *ad libitum* and conventional milk replacer respectively. This study showed that calves fed 25 and 50% soymilk gained weight much more rapidly as compared with calves on cow milk alone. The significant drop in total weight gain (58.25 kg) and ADWG (0.59 kg/day) observed in calves fed 75:25 soy:cow milk might be attributed to the high levels of soymilk in the diet. Khorasani *et al*. (1989) also observed similar pattern with the use of high levels of soyflour in milk replacer which decreased growth rates in calves.

In addition, Nitsan *et al*. (2005) reported that inclusion of soybean meal upto 73% of the total protein in milk replacer resulted in poor calf performance due to low protein digestibility and decreased fat and ash absorption. However, it was earlier shown that the two main factors that influence calf performance on soybean based milk replacers are the proportion of the total protein replaced with the soybeans and the age of the calf (Roy *et al*. (2016).

Feed conversion (FCR), which explains the transformation capacity of dietary nutrients in body tissue, was lower for calves fed 25:75 ratio of soymilk and cow milk though not statistically different from calves on 50:50 soy:cow milk. The FCR observed in this study was relatively better in calves fed 25:75 ratio of soymilk and cow milk than those fed cow milk alone. This implies that calves fed 25:75 ratio of soymilk and cow milk had better feed utilization and transformation capacity to body tissue compared to calves on the other treatments.

5.4.2 Effect of Feeding Different Ratios of Soy:Cow Milk on Linear Body Measurement of Friesian x Bunaji Calves.

Linear body measurements provide good information on performance, productivity and carcass characteristics of animals (Ige *et al.*, 2015). Linear measurements have been used to predict performance characteristics in cattle (Olutogun *et al.*, 2003). Linear body measurements are also important because they allow the farmer to track the animal's skeletal development and deposition of body mass (Ige *et al.*, 2015). In addition to providing body size, it can also be used to estimate live weight of animals (Gizaw, 1995). Statistical analysis showed that there was significant differences ($P<0.05$) between total, weekly and daily increment in body length for all the four groups of calves. The findings of this study were not in agreement with the reports of (Masum *et al*., 2011) when calves were fed whole milk and soymilk replacers. The higher body length obtained in this study could be attributed to higher dry matter intake and the levels of soymilk used in the diet which promotes better skeletal deveploment.

Statistically, there was significant difference in total, weekly and daily height gain of the four groups of calves. The result of this study is not in agreement with the results of Masum

et al. (2011) who found that withers height gain was not significantly different (P>0.05) among calves fed whole milk with vitamin fortified soymilk.

And the reports of Rahman *et al.* (1988) and Mete *et al.* (2000) who found that feeding soymilk based milk replacer to calves did not have any significant influence on wither height. However, the height at withers for calves in this study were within the range reported by Azevedo *et al*. (2016) for Holstein x Gyr crossbred calves fed increasing amounts of milk replacer powder in whole milk. But are higher than those reported by Masum *et al*. (2009) for crossbred calves fed soymilk replacer. This implies that soymilk had a positive effect on wither heights of calves in this study. This finding is consistent with the report of Lee *et al.* (2009) who used milk and milk replacer in feeding dairy calves and obtained a significant increment in wither height.

Statistically, there were significant differences in total, weekly and daily changes in heart girth for calves fed 0:100, 25:75, 50:50 and 75:25 ratios of soy:cow milk, respectively. This is different from the report of (Masum *et al*., 2011) who observed that gain in heart girth among the groups of calves fed whole milk and soymilk replacer were not significant. This study showed that increasing the amounts of soymilk in the diets of Friesian x Bunaji calves had positive effect on linear body measurements. The differences in linear body mesurements observed in this study with that of other authors might be due to dietary factors (i.e. the concentration and levels of soymilk and cow milk in the diets) and also the dry matter intake.

This study found that soymilk positively affects the deposition of structural tissues, resulting in a further development of the animals. It was further inferred that the negative effect of some of the ANFs in soymilk on linear body measurements were minimized by feeding it together with cow milk. The cow milk tend to mask the effects of ANFs that may be present in the diets. Blome *et al*. (2003) and Bartlett *et al*. (2006) corroborated that calf

growth rate increases when protein is provided in synchrony with energy in the liquid feed. However, variations in linear body measurements observed within treatments might be due to individual difference when taking the measurements.

5.4.3 Effect of Feeding Different Ratios of Soy:Cow Milk on Haematological Parameters of Friesian x Bunaji Calves.

Haematology is the study of morphology and physiology of blood (Etim *et al*., 2014a). Packed cell volume (PCV) which is also known as haematocrit (HT or Hct) or Erythrocyte Volume Fraction (EVF) is the percentage (%) of red blood cells in blood (Isaac *et al*., 2013). The PCV obtained in this study (15.01%), (18.67%) and (17.67%) for calves fed 0:100, 25:75 and 50:50 ratios of soymilk and cow milk, were lower than 27.30%, 26.90% and 27.50% reported by Sarker *et al*. (2015) for goat kids fed the aforementioned ratios of soymilk and cow milk. The PCV values obtained in this study fell within the normal range of 24 – 46% for healthy calves as reported by (Merck Veterinary Manual, 2016). This implies that the inclusion of soymilk in the diets of calves was ideal and adequate and the animals were not anaemic. Increased PCV shows a better transportation and thus results in an increased primary and secondary polycythemia (Isaac *et al*., 2013).

Furthermore, Chineke *et al*. (2006) and Etim *et al*. (2014a) reported that high PCV values indicated either an increase in number of red blood cells or reduction in circulating plasma volume. Bello and Tsado (2013) stated that PCV within the normal range suggests that there was no toxic factor (such as haemaglutinin) in the feed of the animal which may adversely affect blood formation. White blood cells (WBC) followed similar pattern but were still within the normal range (4 – 12%) reported by (Merck Veterinary Manual, 2016). Although calves fed 75:25 ratio of soymilk to cow milk had slightly higher WBC which might be attributed to the high levels of soymilk used in the diet which means higher consumption of possible residuals of ANFs by the calves on that treatment. Higher concentration of WBC in the blood of an animal implies that the animal is trying to fight the effect of some undesirable elements in the diet consumed. Etim (2010) reported that

normal range of WBC indicate that the animals are healthy because increase in WBC counts above the normal range is an indication of allergic conditions, anaphylactic shock and certain parasitism.

White blood cells are known to act as defensive mechanism against toxic factors in the body. The WBC counts obtained in this study agrees with the findings of Sarker *et al*. (2015) who observed elevated serum antibody in calves fed soy products. Reports had also shown that animals with high concentration of WBCs are capable of generating antibodies in the process of phagocytosis and have high degree of resistance to disease (Etim *et al*., 2014b). However, the WBC counts obtained in this study fall within the normal range of (4 – 12 $x10^9$/l) reported by (Merck Veterinary Manual, 2016).

Red blood cells (RBC) serve as a carrier of haemoglobin in the body of the animal. And the haemoglobin reacts with oxygen carried in the blood to form oxyhaemoglobin during respiration (Isaac *et al*., 2013). The values of RBCs in this study were within the normal range of (0 – 20 $x10^{12}$/l) reported for healthy calves (Merck Veterinary Manual, 2016). This could be due to the fact that soybean is a rich source of minerals and vitamins (such as iron, zinc, copper, thiamine, riboflavin, niacin and patholenic acid) which are well-known hematinics (McArthur *et al*., 1988) and are essential in the formation of RBCs (Ganong, 2005).

Haemoglobin (Hgb), Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH) and Mean corpuscular haemoglobin concentration (MCHC) were all within the normal ranges of 8 – 15g/dl, 40 – 60 fl, 11 – 17pg and 30 – 36g/dl, respectively reported for healthy calves (Merck Veterinary Manual, 2016). This means that the addition of soymilk in the diets of calves increased the protein content and its utilization. This finding is consistent with the report of Bello and Tsado (2013) who found that haemoglobin values within normal range imply that the dietary proteins were of high quality. The differential WBC (lymphocytes, neutrophils and basophils) observed in this study fall within the

normal range for healthy calves. This means that the levels of ANFs in the processed soybeans did not affect the immune system of the animals.

This agrees with Chineke *et al.* (2006) who found that lymphocytes, neutrophils and basophils offer protection against toxins and infectious organisms in the body of the animal. Thus, significantly high differential WBC may imply the presence of ANFs in the blood. Etim *et al*. (2014b) corroborated that the immune status of an animal is a function of lymphocytes and neutrophils. And that when these cells fall within the normal range, it's an indication that the feeding pattern did not affect the immune system.

5.4.4 Effect of Feeding Different Ratios of Soy:Cow Milk on Some Serum Biochemical Indices of Friesian x Bunaji Calves.

Blood biochemical indices are considered important in evaluating the health status of animals as a result of feeding, disease or low production (Hagawane *et al*., 2009). Total protein significantly ($P<0.05$) increased with increased levels of soymilk in the diets of the calves. The serum total protein (5.51 to 6.12 g/dl) obtained in this study were within the range (6.12 to 6.49 g/dl) reported by Geiger *et al*. (2014) in Holstein calves fed varying milk replacers with or without direct-fed microbial supplementation. The improvement of serum total protein observed in this study might have been associated with improved dietary intake as a result of the inclusion of soymilk. However, the serum TP concentration obtained in this study fall within the normal range (6.7 – 7.5 g/dl) reported by (Merck Veterinary Manual, 2016) for healthy animals. This is in line with the reports of Hagawane *et al*. (2009) who indicated that TP concentrations are usually used as an appraisal of nutritive status of animals reflecting improved feed intake and metabolism.

The albumin concentration is also an important indicator of nutritional status of animals. Albumin performs many functions in the body of an animal including maintaining the osmotic pressure that causes fluid to remain within the blood stream instead of leaking out

into the tissues (Hagawane *et al*., 2009). The serum albumin (3.34 to 4.32 g/dl) obtained in this study were similar to the range (4.1o to 4.80 g/dl) reported by (Lee *et al*., 2009) in Holstein calves fed milk replacer containing different amount of energy and protein.

But the values of albumin obtained in this study were slighlty above the normal range (2.5 – 3.8 g/dl) for healthy animals which suggest that the calves were not dehydrated. This is because, albumin levels are dependent on the state of hydration of the body. An animal that is dehydrated will have an artificially high albumin level, which would return to normal when the dehydration is corrected (Hagawane *et al*., 2009). This study showed that the inclusion of soymilk at different ratios in the diet of calves did not lead to dehydration. Globulins as a component of serum TP are known to play important role in blood clotting, osmotic function, blood viscosity, defensive function, buffering action, capillary permeability, conservative and transport function as well as serve as reserve proteins (Hagawane *et al*., 2009). The globulin concentrations obtained in this study were within the normal range (3.0 – 3.5 g/dl) for healthy calves (Merck Veterinary Manual, 2016).

Blood glucose concentration is regarded as one of the indicators of energy status in ruminants (Ganong, 2005). The liver is the pre-dominant site of glucose synthesis in the ruminant. Glucose supply is crucial for maintenance and productive functions in growing ruminants (Hagawane *et al*., 2009). The blood glucose concentration observed in this study which ranged between 73.87 to 96.20mg/dl was higher than the range of (40.00 to 49.75mg/dl) reported by Roy *et al*. (2016). But is consistent with the values (70.75 to 77.75 mg/dl) reported by Geiger *et al*. (2014) in Holstein calves fed varying milk replacers with or without direct-fed microbial supplementation. The high blood glucose observed in this study could be attributed to the treatment effect of soymilk used in the diet of the calves. This increment could also be attributed to the physiological shift in the primary energy source from glucose to volatile fatty acids (VFA) when the rumen in young calves becomes functional (Hammon *et al*., 2002). However, the blood glucose levels in this study were

within the normal range (40 – 100 mg/dl) reported by (Merck Veterinary Manual, 2016) for healthy calves.

The reported range of blood urea nitrogen (BUN) in this study (7.13 to 9.79 mg/dl) was lower than 17.37 to 23.00mg/dl reported by Roy *et al*. (2016) for calves fed soybeans based milk replacer. The BUN in this study increased significantly ($P<0.05$) with increased levels of soymilk in the diet. The significant ($P<0.05$) increase in blood urea nitrogen levels (9.79 mg/dl) obtained in calves fed 50:50 ratio of soymilk and cow milk could be attributed to the high utility value of the diet. Similarly, other workers (Khan *et al*., 2007) also demonstrated increased BUN in calves with increase levels of soymilk in the diet. The concentrations of BUN obtained in this study fall within the normal range of 10 – 25mg/dl reported by (Merck Veterinary Manual, 2016) for healthy calves. Higher concentration of BUN is an index of renal dysfunction (Khan *et al*., 2007). However, Harris (2009) reported that BUN may be a better indicator of hydration than renal function, and an elevated BUN would imply dehydration. This study found that calves fed the different ratios of soymilk were not dehydrated because an elevated level of BUN would imply dehydration. Significantly lower ratios denote acute tubular necrosis, low protein intake, starvation or severe liver disease as reported by Hagawane *et al*. (2009).

Serum creatinine is an important indicator of renal health because it is an easily measured by-product of muscle metabolism that is excreted unchanged by the kidneys (Harris, 2009). The serum creatinine (0.47 to 0.60 mg/dl) obtained in this study were lower than the values (1.50 to 2.2 mg/dl) reported by Lee *et al*. (2009) in Holstein calves fed milk replacer containing different amount of energy and protein. It was found that the values of creatinine obtained in this study were within the normal range (0.5 – 2.2 mg/dl) reported for health calves (Merck Veterinary Manual, 2016). This supports the findings of Hammon *et al*.

(2002) who reported that serum creatinine concentration in calves fed high protein milk replacer were within the safe range, suggesting normal liver and renal functions in calves.

5.4.5 Effect of Feeding Different Ratios of Soy:Cow Milk on Mean Rumen Metabolites of Friesian x Bunaji Calves.

The temperature and pH of the rumen fluid of calves in this study were not significantly affected ($P>0.05$) by the different ratios of soy:cow milk in the diets. The similar rumen fluid temperature observed across the treatments in this study showed no differences in heat evolution from microbial fermentation activity in the diets (Figure 4.1). The temperature of a healthy bovine tends to range from 37.8 to 40.0 °C. Similarly, the rumen fluid temperature obtained were within the relative constant range values (38.00 – 40.00^{o}C) for continuous microbial fermentation as reported by (Demba and Liang, 2011). Moreover, rumen contents tend to ferment at 40.00 °C so this works as a central self-heating system. Rumen pH is one of the variables which can profoundly influence microbial population. Cellulolytic bacteria and methanogenic bacteria are severely affected once the rumen pH falls below 6.0 (Church, 1998). The rumen pH values obtained in this study were within (5.50 to 7.00) for optimum rumen microbial fermentation as reported by (Church, 1998). Rumen pH is an important factor that measures the alkalinity and acidity of rumen contents (Church, 1998). The rumen pH values obtained in this study were within the range of values (6.00 and 7.20) for maximum microbial growth reported by (Demba and Liang, 2011).

Rumen ammonia nitrogen (NH3–N) concentration was significantly affected ($P<0.05$) by the different ratios of soy:cow milk. The variation obtained in NH_3-N concentration might be due to ammonia production during microbial metabolism which is the major source of nitrogen used for biosynthesis of microbial cells. Isah *et al*. (2014) reported that the concentration of the NH_3-N in the rumen is a consequence of the balance between its

production, absorption and utilization by microbes. Fermentation of proteinous feed in the rumen produces NH_3-N, many ruminal bacteria can grow with ammonia as the main source of nitrogen, but some require amino acid.

Eryavuz *et al*. (2003) reported that NH3–N concentration has a good profile with values between 12 and 15mg/100ml as a minimum rumen fluid to maximize rumen microbial protein synthesis. The values observed in this study were similar to the finding of (Kisseda *et al*., 2010) who reported that must suitable rumen NH_3-N concentrate levels for microbial activities range between 5 and 20mg/100ml. The significant ($P<0.05$) increase in the TVFA concentration observed in this study may probably be due to increased in the concentration of ammonia in the rumen (Figure 4.1). Calves fed 75:25 ratio of soy:cow milk exhibited the highest total volatile fatty acids though not statistically different from those on 50:50 ratio of soy:cow milk. This indicates that there was efficient utilization of the ingested feed material leading to a favourable rumen environment and subsequent yield of rumen metabolites. Micro-organisms yield in the rumen is an index or a function of the amount of microbial proteins made available to the ruminants daily (Minson, 1983).

5.4.6 Effect of Sampling Time (Hours) on Mean Rumen Metabolites in Friesian x Bunaji Calves fed Different Ratios of Soy:Cow Milk.

The higher (29.22^{o}C) rumen fluid temperature obtained at 6 hours after feeding (Figure 4.2) could be attributed to the different ratios of the soy:cow milk fed. As temperature is usually cooler in the morning after resting and warmer in the afternoon or night after long day of muscular activity. Conversely, the rumen fluid pH (8.08) at 0hr was above neutral (slightly alkaline). But the pH slightly dropped to (6.14) at 9 hours post feeding (Figure 4.3). This could be attributed to the buffering effect as a result of reflex action of salivation which occured in the morning in expectation for feeding. This could also be attributed to feeding which stimulated the microbes in the rumen and thus started degrading the diets thereby releasing TVFAs which slightly drops the rumen pH. However, the rumen fluid pH fall

within the normal range (6.00 to 7.00) and (6.55 to 6.77) reported by (Reddy and Reddy, 1999) and Geiger *et al*. (2014) respectively, in Holstein calves fed varying milk replacers with or without direct-fed microbial supplementation.

This means that the rumen pH of animals in this study was adequate for normal rumen microbial fermentation. The pH values obtained in this study were above the minimum of 6.0 recommended by Mould and Orskov (1984) below which cellulolytic digestion in the rumen could be inhibited. Rumen ammonia nitrogen (RAN or NH_3-N) is a measure of the extent of rumen microbial protein degradation. Rumen ammonia nitrogen is an important factor in determining the utilization of nitrogen in the rumen (Jokthan, 2006). The highest concentration of RAN observed at 9 hours post feeding indicated that there was high degradation of nitrogen in the rumen (Figure 4.4). The rumen NH_3-N concentration obtained in this study is higher than the values reported by (Satter and Slyter, 1974) for optimum microbial function and the values (2.00 to 8.00mg/100ml) reported by (Drewnoski and Poore, 2012) for high producing ruminant livestock. The differences observed in the present study and that of other workers could be attributed to the nitrogen content of soymilk fed and the level of inclusion in the liquid feed. Powel *et al.* (1981) reported that RAN between 3.60 – 17mg/100ml promoted microbial protein production.

Total Volatile Fatty Acids (TVFA) are the main energy sources for ruminants. Thus, their concentration in the rumen fluid of the experimental calves gave an indication of the energy values of the diets. It appears that the TVFA concentrations in this study were positively influenced by the inclusion of soymilk in the diets of the calves (Figure 4.5). The range of TVFA (22.00 to 37.00 mmol/l) obtained in this study were higher than the range of (10.5 to 12.8mmol/l) reported by Kandil *et al.* (1996) for calves fed milk replacer. The increase in TVFA (37.00 mmol/l) at 6 hours post feeding could be associated with adequate digestibility of the feed material in the rumen. The ability of calves to digest solid feed material in the rumen is a function of the age of the animal, development of the rumen and quality of feed that is provided from liquid and solid feed (Baldwin *et al*., 2004). To

maintain physiological functions of the rumen, Iqbal *et al*. (2014) reported that fibre should be an important part of the ration.

Abdelsamei *et al*. (2005) reported that the consumption of calf starter and water led to the production of volatile fatty acids in the rumen which in turn stimulates rumen development.

5.4.7 Nutrient Digestibility and Nitrogen Balance in Friesian x Bunaji Calves fed Different Ratios of Soy:Cow Milk.

Nutrient digestibility in animals is the classical and direct method for estimating feed digestion by ruminants; hence studies on digestibility of ruminant feeds are very important as they allow for the estimation of nutrients actually available for ruminant nutrition (Yashim *et al*., 2016). The results obtained in this study on dry matter, organic matter and crude protein digestibility were lower than the values reported by Azevedo *et al*. (2016) for Holstein x Gyr crossbred calves fed increasing amounts of milk replacer powder in whole milk. Also, the dry matter and crude protein digestibility obtained in this study for calves fed 25:75 ratio of soymilk and cow milk was lower than those reported by (Rahman *et al*., 2016) in calves fed soybeans, shotti and skim milk based milk replacers. This might be due to the differences in the type, quantity and chemical composition of the liquid feeds used.

Davis and Drackley (1998) reported that digestibility varies depending on the overall composition of the milk replacer, origin of the ingredients and the animal's age. Improvement in the digestibility of dry matter and crude protein by the calves fed 25:75 ratio of soymilk and cow milk could be attributed to the additive effects of soymilk and cow milk feeding as compared to calves fed only cow milk. It could also be as a result of soaking which reduced the levels of ANFs in the soybean diets thereby improving protein degradation in the rumen. This agrees with earlier report of Njoku and Evbuomwan (2014) that the presence of ANFs lowers the solubility of proteins entering the abomasum and

small intestine for digestion. However, decreased crude protein digestibility was reported with increased soybean protein in milk replacers for young calves (Erickson *et al*., 1989).

The significant difference in organic matter digestibility observed in this study is similar with the report of Rotger *et al*. (2006) who stated that a change in feed ingredients alters the amount of OM and its fermentation rate in the rumen. The digestibility of feed was reported to be influenced by the composition, available energy and retention time in the gastrointestinal tract (Abdu *et al*., 2012). The digestibility of NDF and ADF were better in calves fed soymilk and cow milk in the ratio of 25:75 compared with calves fed 100% cow milk. The difference observed in NDF and ADF digestibility could be attributed to the disparity in the ratios of soymilk used and the composition of the diets. The improvement in calves performance and overall increase in digestibility could be due to the elimination of ANFs found in soybeans by soaking in water for 72 hours. Although nitrogen intake was significantly different across treatments, positive nitrogen balance was recorded in calves fed soymilk. Calves that were fed soymilk milk had highest nitrogen retention as compared with calves fed only cow milk. This finding differs from the report of Shakya *et al*. (2016) who found that retention of digested nitrogen is often lower for calves receiving non-milk proteins not properly processed when compared with calves fed milk protein which suggests poor availability of essential amino acids in the non-milk proteins. This indicates that the nitrogen obtained from soymilk contributed to the total nitrogen balance in the calves.

The results of nitrogen retention obtained in this study were lower than the values reported by (Rahman *et al*., 2016) in calves fed soybeans, shotti and skim milk based milk replacers. Nitrogen retention is the major indicator used to assess the protein nutrition status of ruminant livestock (Abdu *et al*., 2012). The low nitrogen retention observed in this study

could be attributed to the CP content of the soymilk and cow milk diets supplied to the calves. This supports the findings of Sarwar *et al*. (2003) who observed that nitrogen retention depends on good digestibility and utilization of nutrients. However, the values fall within the normal range of (52.80 – 64.6%) reported by Azevedo *et al*. (2016).

The positive nitrogen balance obtained in this study signified that anti-nutrient contents of soybeans did not depress their utilization by the calves. This shows the significance of soaking as a processing method. Conversely, relatively poor growth, digestibility and nitrogen retention resulted when cooked soybeans, soybean meal or lightly cooked soybean flour were used in feeding calves (Gorrill and Thomas, 1967).

5.5 Economic Benefits of Feeding Different Ratios of Soy:Cow Milk to Friesian x Bunaji Calves.

Among the costs to raise a heifer, feed accounts for 49% of the total costs invested (Karszes, 2005). Inclusion of soymilk in the diets of calves reduced the cost of liquid feed consumed compared to feeding cow milk alone. The cost of feed per kg weight gain was influenced by feeding soymilk in this study. Animals fed cow milk alone had the highest feed cost per kg weight gain as compared with calves on treatments containing soymilk. The reduction in feed cost per kg weight gain in calves fed 75:25, 25:75 and 50:50 showed that inclusion of soymilk reduced the cost of production. This finding agreed with the report of Khan *et al*. (2012) who found that feeding soybean based milk replacer to calves was less expensive to feeding cow milk. This is also in agreement with (Ghorbani *et al*., 2007) who reported soymilk as an economic substitute for whole milk in calf rearing. The higher feed cost per kg weight gain observed on animals fed cow milk alone could be attributed to the cost of cow milk (₦ 98 per litre) which was about 3 times the cost of one litre of soymilk (₦33). Calves fed 75:25 ratio of soy:cowmilk had better percent feed cost reduction compared with those on 25:75 ratio of soy:cowmilk which could be attibuted to level of soymilk used which was cheaper compared with cow milk which was more expensive. The study found that the inclusion of 25 and 50% soymilk resulted in a better

and more cost effective feed as compared with cow milk alone. This study also found that the use of soymilk was cheaper than cow milk. Therefore, calves can be successfully reared on soy:cow milk combinations at lower cost than with cow milk alone, thus offering a potential increase in economic returns for farmers.

CHAPTER SIX

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

From the results of this study, the following conclusions were made:

- Soaking of soybeans in water for 12, 24, 48 and 72 hrs generally improved the chemical composition and reduced the levels of ANFs. However, soybeans soaked for 72 hrs had the best percent reduction in ANFs.

- Calves fed diet containing 25:75 ratio of soy:cow milk had the lowest total feed intake (409.64 kg) and average daily feed intake (4.18 kg/day) and better total weight gain (74.25 kg), average daily weight gain (0.76 kg/day) and feed conversion ratio (5.52).

- Inclusion of soymilk at different ratios had positive influence on linear body measurements (body length, height at withers and heart girth) of calves.

- All the blood parameters measured were within the normal range for healthy calves. Indicating that soaking for 72 hours significantly lowered the levels of ANFs in soymilk and did have any deleterious effect on the health of calves.

- Feeding different ratios of soy:cow milk had positive influence on rumen fluid temperature, pH, NH_3-N and TVFA with 75:25 soy:cow milk raton having the highest concentration of RAN (13.84 mg/100ml). However, pH levels remained within acceptable range for optimum rumen fermentation (6.36 – 7.31).

- Sampling time significantly ($P<0.05$) influenced rumen temperature, pH, NH_3-N and TVFA concentrations.

- Digestibility of DM, OM, CP, NDF and ADF were significantly ($P<0.05$) influenced by the inclusion of soymilk at different ratios. However, calves fed 25:75

and 75:25 ratios of soy:cow milk had better nutrient digestibility and nitrogen balance compared to other treatments.

- The cost of cow milk per litre (₦98.00) was about 3 times more expensive than soymilk (₦33). Calves fed 25 and 50% soymilk had better feed cost per kg weight gain (₦496.10 and ₦469.67), respectively.

- Calves fed 75:25 ratio of soy:cow milk had the least total cost of feed consumed (₦ 30489.00). Indicating that feeding cow milk to Friesian x Bunaji calves was very expensive as compared with feeding soy:cow milk at 75:25 ratio. Feeding soy:cow at a ratio of 75:25 reduced cost by (24.16%) as compared with feeding cow milk alone.

6.2 Recommendations

- Soaking of soybeans in water is recommended for performance and best at 72 hours.

- Feeding of 25:75 ratio of soy:cow milk is recommended for better performance and feed conversion efficiency.

- Further study is recommended on the use of other ingredients as milk substitute and to compare the results with this study.

6.3 Contribution To Knowledge

- Soaking of soybeans for 72 hours improved the nutritive value of soybeans and reduced the levels of anti-nutritonal factors (phytates, protease inhibitors, tannins, oxalates and saponins by (90.89, 46.54, 57.14, 19.23 and 60.16%), respectively.

- Feeding of calves with soymilk at a ratio of 25:75 improved live weight gain by 0.76 kg/day as against 0.67kg/day in calves fed cow milk alone.

- Feeding soy:cow milk at a ratio of 75:25 reduced the cost of feeding by (24.16%) as compared with feeding cow milk alone.

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APPENDICES



Appendix I: Soybeans soaked in excess water for 72 hours after which they were rinsed.



Appendix II: Rinsed soybeans ready for drying (drying lasted for 8 days).



Appendix III: A Friesian x Bunaji Cow being Milked and Mixture of Soy:Cow Milk in Stainless Containers Ready for Feeding to Calves.



Appendix IV: Bucket-feeding of the Different Ratios of Soy:Cow Milk to Friesian x Bunaji Calves.



Appendix V: Feeding Weighed Amount of Concentrate Diet and *Digitaria smutsii* Hay; and Clean Drinking Water was Provided *ad libitum* to the Calves During the Growth Trial.



Appendix VI: Weighing Friesian x Bunaji Calves During the Study.



Appendix VII: Rumen Fluid Collection using a Stomach Tube and Determination of Temperature and pH using a Thermometer and Digital pH Meter, respectively.



Appendix VIII: Friesian x Bunaji Calves Feeding in their Individual Crates During Metabolism Trial.



Appendix IX: Faecal and Urine Samples Collected from Calves During Metabolism Trial.