QUALITY, SENSORY AND STORAGE ATTRIBUTES OF CHEESE PRODUCED FROM COWMILK SUPPLEMENTED WITH TIGERNUT AND COCONUT MILK

BY

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CERTIFICATION

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DEDICATION

This work is dedicated to Almighty Allah, the source of knowledge and wisdom who guided and spared my life throughout my programme at the university and to my parents Mr. and Mrs. Abdulkareem for their tremendous contribution and support towards my educational pursuit.

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I express my sincere appreciation to the almighty Allah, the one who is free and far from all imperfections. He alone deserves to be praised, especially for keeping me alive and making it possible for me to complete this research work. I am forever indebted to Him.

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TABLE OF CONTENT

LIST OF FIGURES

LIST OF PLATES

LIST OF TABLES

ABSTRACT

Cheese is a concentrated dairy product produced by acid or rennet curdling of milk, stirring and heating the curd, draining off the whey, collecting and pressing the curd. In this study, Cheese was made from cow milk supplemented with tigernut milk and coconut milk in the percentage ratio of 25:25:50; 25:50:25; 50:25:25; 75:25:0; 75:0:25% cow-tigernut milk-coconut milk (v/v). The unsupplemented cow milk cheese was used as control. The quality, shelf life and sensory attributes of the cheese samples were examined. The effect of the supplementation was monitored as the cheese samples were stored at refrigeration temperature (4 ^{0}C) and examined for yield, total titratable acidity, proximate

analysis, microbial analysis and sensory quality. The percentage yield of the cheese samples showed significant differences ($p<0.05$) and ranged from 17.33 to 13.43% with increase in the percentage tigernut milk and coconut milk in the blends causing an increase and decrease in the percentage yield of the cheese samples respectively. The total titratable acidity decreased significantly ($p<0.05$) with decrease in percentage cow milk in the blend with the values ranging from 0.30 to 0.23%. The moisture content increased significantly ($p<0.05$) from 53.18% (C_{100}) to 59.66% ($C_{25}S_{50}N_{25}$) with increase in coconut milk resulting in an increase in moisture content. The protein content of the samples differed significantly ($p<0.05$) with values ranging from 18.77 to 14.59%. The fat content ranged from 21.49 to 12.90% with increase in coconut milk. The total ash content ranged between 3.03 and 1.47% while the carbohydrate content ranged from 10.72 to 2.81% with different level of supplementations being significantly different ($p<0.05$) from the control. Significant differences ($p<0.05$) was observed in the fungal and bacterial counts at different levels of storage period. Sample$C_{25}S_{50}N_{25}$ and$C_{75}S_{25}N_{0}$ showed the highest number of both bacterial and fungal counts respectively. There were significant differences ($p<0.05$) in color, texture, aroma and overall acceptability of the samples while the taste was judged to be similar. The study recommended that tigernut milk and coconut milk could be used either singularly or as a mix to supplement cow milk up to 50 % without adverse changes or effect on chemical properties, impairing the nutrients and acceptability of the final product. However 25% supplementation of cow milk with the milk substitutes proved to be the best blend based on consumer acceptability.

CHAPTER ONE

INTRODUCTION

1.1 Background of the study

Food is any substanceconsumed to provide nutritional support for an organism. It is usually of plant or animal origin, and contains essential nutrients, such as carbohydrates, fats, proteins, vitamins and minerals. The food substance is ingested by human or organism and assimilated by the cells to provide energy, maintain life and stimulate growth. Today majority of the food energy required by the ever increasingpopulation of the world is supplied by the food industry as opposed to the hunting and gathering and agriculture which are the two historical means by which human secure food (Balogun *et al.*, 2016). The right to food is a human right derived from the International Covenant on Economic, Social and Cultural Rights (ICESCR), recognizing the "right to an adequate standard of living, including adequate food", as well as the "fundamental right to be free from hunger". Food has its source from plant and animal which could be in form of cereals, legumes, root and tuber, fruit and vegetables, meat, fish and milk products.

As an agricultural product, milk is extracted from non-human mammals during or soon after pregnancy. Dairy farms produced about 730 million tonnes of milk in 2011,from 260 million dairy cows.The United States, India, China and Brazil are the world's largest exporters of milk and milk products. Throughout the world, more than six billion people consume milk and milk products. Over 750 million people live in dairy farming households (Hemme *et al.*, 2010). Milk is an essential part of daily diet for the growing children and expectant mothers. Milk is a major constituent of the diet; its quality assurance is considered essential to the welfare of a community (Marimuthu *et al.*, 2013). The principal constituents of milk include fat, protein, total solid, lactose and ash. In addition to this, milk contains several hundred minor constituents many of which includes milk fat, vitamins, metal ion and flavor compounds, which have a major impact on the nutritional, technological and sensory properties of milk and dairy products. It is a perishable commodity and also an excellent medium for microbial growth (Ogunlade *et al.*, 2017). In Nigeria, milk production is mainly done by the Fulani nomadic people who are pastoralists involved in the rearing of cattle moving from one location to another in search of green pasture. Due to lack of refrigeration facilities, the Fulani women process the surplus fresh milk into products such as non (fermented skimmed milk), mai shanu and soft, unripened cheese called “warankasi” or “wara” in short term (Adetunji and Babalobi, 2011).

Tigernut (*Cyperus esculentus*) are often used in various ways in traditional Nigerian cuisine, but tigernut milk is particularly well loved. Tigernuts are not nuts, they are tiny tubers (root vegetable) with a nut-like flavor and nut -like textureTigernut milk is naturally sweet, creamy and offers a luxurious, rich and nutty flavor. Tigernut milk is rich in monosaturated fat (oleic acid), as well as minerals (Like calcium, iron, magnesium, potassium and phosphorus) and vitamins C and E. Tigernuts are also a good source of probiotics (that is food for the good beneficial bacterial) like inulin and resistant starch. Tigernut milk can be made simply by soaking the tubers in water, blending and filtering. However, the addition of spice like cinnamon as well as sweetner is good. In Nigeria tigernut is not flavored from time to time with sweet spices, but also with alligator pepper.

Coconut (*Cocos nucifers*) grows extensively in Nigeria and is eaten as a snack usually for pleasure. Coconut is a fruit rich in fiber (aids digestibility), iron, and other minerals. Coconut also serves as an excellent source of raw materials for the development of dairy-like products.

Coconut milk is the liquid that comes from the grated meat of a brown coconut (it is different from coconut water). The colour and rich taste of coconut milk can be attributed to the high oil content. Most of the fat is saturated fat. Coconut milk has the following nutritional properties; protein (3%), fat (17% - 24%), and carbohydrate (2%). It has no cholesterol, contains many vitamins, minerals and electrolytes, including potassium, calcium and chloride (Balogun *et al*., 2016).Coconut milk is being used by confectionaries, bakeries, biscuits and ice cream Industries worldwide to enhance flavor and taste of various products. Coconut milk was found to be rich in calcium. The milk was reported to be high in minerals and vitamin content, while total saturated fat was 10% of the total energy (Adejuyiyan *et al*., 2014).

Cheese is a concentrated dairy commodity produced by acid or rennet coagulation or curdling milk, stirring and heating the curd, draining off the whey, collecting and pressing the curd. The cheese is ripened, cured, or aged to develop the flavour and texture. According to Balogun *et al* .(2016) cheese is a dairy product made by coagulating either whole milk, part-skim (low fat) milk, skim milk, or cream by removing much of the liquid portion while retaining the coagulum and the entrapped milk solids. Cheese is an excellent source of protein, fats and minerals such as calcium, iron and phosphorus, vitamins and essential amino acids, thus making it an important food in the diet of both old and young people (Oladipo and Jadesimi,2013).'Warankashi' is obtained by coagulating pasteurized milk at a specific temperature, pH and processing time with an appropriate coagulant. The preferred coagulant comes from Sodom apple leaf extract (*Calotropis procera*) because the cheese made with this coagulant has a sweeter flavour and a higher protein content compared to the cheese made with the other coagulants (Omotosho *et al*., 2011).The coagulated milk is poured into asmall basket or strainer in order to drain and to give the cheese the desired shape and size (Adetunji and Salawu, 2008). Hundreds of types of cheeses are produced based on their different characteristics and method of productions. Their styles, textures and flavours depend on the origin of the milk (including the animal's diet), whether they have been pasteurized or not, the butterfat content, the bacteria and the mold, the processing and aging. Herbs, spices or wood smoke may be used as flavouring agents (Fankhauser and David, 2007)

1.2 Justification

The manufacture of soft cheese (*wara*) is widespread in developing Africa countries and was thought to have originated in the Northern region of Nigeria due to the traditional cattle rearers (*Fulani*) access to fresh milk from cattle specie of *Zebu Indicus.* However many factors such as shortage of good quality milk, poor processing and preservation methods, poor hygiene practices, poor packaging and storage facilities have contributed to poor utilization and availability in areas where it might have been useful in alleviating protein and other nutritional deficiency. This often leads to importation of milk and milk products such as cheese which may adversely affect the nutrition and the socio-economic well-being of people especially low income earners and rural dwellers. Thus, there is a need to find alternatives making the use of plant milk supplementations a research field.Little research attention and inconsistency in findings have been major challenges on the production and blending ratio of cow milk to vegetable sourced milk such as tigernut milk, coconut milk, soy milk and Bambara nut milk. Therefore, the thrust of this study was to evaluate the chemical composition and sensory qualities of West African soft cheese 'warankashi' produced from blends of cow milk ,tigernut milk and coconut milk, documenting its keeping quality and keeping period and also increasing the utilization of blends of plant milk for the production of good quality cheese product. Tigernut milk is reported to have great potentials to supplement cow milk; it contains high quality protein and essential amino acids, essential minerals and vitamins. Unlike Cow milk it contains no cholesterol and can be consumed by lactose intolerant patient. It is a very rich source of highly valuable proteins, unsaturated fatty acids, soluble and insoluble dietary fibres, and isoflavones whose presence in everyday diet is very important (Hussein *et al*., 2016).

Coconut milk has the following nutritional properties; protein (3%), fat (17% - 24%), and carbohydrate (2%). It has no cholesterol, contains many vitamins, majorly vitamin E which helps in improving fertility, minerals and electrolytes, including potassium, calcium and chloride (Balogun *et al*., 2016). It has many uses, most of which build up the immune and the body's defence system (Balogun *et al*., 2016). Coconut milk is lactose free, unlike cow milk, and can be used as a milk substitute by those with lactose intolerance. Coconut utilization in Nigeria is very low as it is mainly eaten as a snack or shredded and fried to make a coconut candy. Coconut oil is still being used however the milk is under-utilized in the country (Balogun *et al*., 2016). Blend of cow milk, tigernut milk and coconut milk for the production of cheese will not only give a food product of improved quality but also a product which can be consumed by lactose intolerant patient, patient with low fertility and more so consumer who intend to improve his immune system and prevent cardio- vascular diseases, inflammatory, diabetes and cancer.

1.3 Objectives

1.3.1 General Objectives

The general objective of this study is to prepare cheese from blend of cow milk and differently sourced vegetable milk.

1.3.2 Specific Objectives

The specific objectives of this study are:

1) To produce cheese (warankashi) from cow milk supplemented with tigernut and coconut milk blend.
2) To evaluate the proximate composition of cheese produced from blends of cow milk and differently sourced vegetable milk.
3) To determine the yield and titratable acidity of cheese produced from blends cow and differently sourced vegetable milk.
4) To assess the microbial load of the cheese during storage.
5) To evaluate consumer acceptability of the cheese made from cow milk supplemented with different sourced vegetable milk.

CHAPTER TWO

LITERAURE REVIEW

2.1 Milk Composition

The Codex Alimentarius standards defined milk as the normal mammary secretion of milking animals obtained from one or more milking without either addition to it or extraction from it, intended for consumption as liquid milk or for further processing. This definition thereby precludes non-animal products which may resemble milk in color and texture (milk substitutes) such as soy milk, tigernut milk, rice milk, almond milk, and coconut milk (Codex, 1999).The earliest tribes of ancient Egypt and South-West Asia discovered sometime around 5000 BC that cow milk was a nourishing human food. While the ancient Egyptians recognized that cow milk was a wholesome and sustaining food they could have had little knowledge of its composition. The earliest evidence of knowledge of the composition of milk is dated at about 350 BC when Aristotle wrote "Casein, fat and water are all the known substances of milk (O'Connor, 1993).

From Southwest Asia domestic dairy animals spread to Europe (beginning around 7000 BC but did not reach Britain and Scandinavia until after 4000 BC and South Asia (7000–5500 BC). The first farmers in central Europe and Britain milked their animals. Pastoral and pastoral nomadic economies, which rely predominantly or exclusively on domestic animals and their products rather than crop farming, were developed as European farmers moved into the Pontic-Caspian steppe in the fourth millennium BC, and subsequently spread across much of the Eurasian steppe(Anthony,2007). Sheep and goats were introduced to Africa from Southwest Asia, but African cattle may have been independently domesticated around 7000–6000 BC. Camels, domesticated in central Arabia in the fourth millennium BC, have also been used as dairy animals in North Africa and the Arabian Peninsula. The earliest Egyptian records of burn treatments describe burn dressings using milk from mothers of male babies. In the rest of the world (i.e., East and Southeast Asia, the Americas and Australia) milk and dairy products were historically not a large part of the diet, either because they remained populated by hunter-gatherers who did not keep animals or the local agricultural economies did not include domesticated dairy species (Valenze, 2011).

Milk consumption became common in these regions comparatively recently, as a consequence of European colonialism and political domination over much of the world in the last 500 years. In the middle Ages, milk was called the "virtuous white liquor" because alcoholic beverages were safer to consume than water (Pecanac *et al*., 2013). Variations exist in the composition of milk for the various species. The composition of cow milk varies for a number of reasons which includes the individuality of the cow, the breed and age, stage of lactation, health of the cow, climatic conditions and herd management which includes feeding and general care. The total milk consumption (as fluid milk and processed products) per person varies widely from highs in Europe and North America to lows in Asia. However, as the various regions of the world become more integrated through travel and migration, these trends are changing, a factor which needs to be considered by product developers and marketers of milk and milk products in various countries of the world. Even within regions such as Europe, the custom of milk consumption has varied greatly. Consider for example the high consumption of fluid milk in countries like Finland, Norway and Sweden compared to France and Italy where cheeses have tended to dominate milk consumption.

The females of all mammal species can by definition produce milk, but cow's milk dominates commercial production. In 2011, FAO estimates that 85% of all milk worldwide was produced from cows. In the Western world, cow's milk is produced on an industrial scale and is by far the most commonly consumed form of milk. Dairy cattle such as the Holstein have been bred selectively for increased milk production. About 90% of the dairy cows in the United States and 85% in Great Britain are Holsteins. Other dairy cows in the United States include Ayrshire, Brown Swiss, Guernsey, Jersey and Milking Shorthorn (Dairy Shorthorn). Aside from cattle, many kinds of livestock provide milk used by humans for dairy products. These animals include buffalo, goat, sheep, camel, donkey, horse, reindeer and yak. The first four respectively produced about 11%, 2%, 1.4% and 0.2% of all milk worldwide in 2011 (Gerosa *et al*., 2012). Goff *et a*l. (2014) describes the approximate composition of milk of various species with the table below.

Table 1: Approximate composition of milk from various species of mammals.

Constituents	Unit	Cow	Goat	Sheep	Water buffalo
Water	G	87.8	88.9	83.0	81.1
Protein	G	3.2	3.1	5.4	4.5
Fat	G	3.9	3.5	6.0	8.0
Saturated fatty acids	G	2.4	2.3	3.8	4.2
Monounsaturated fatty acids	G	1.1	0.8	1.5	1.7
Polyunsaturated fatty acids	G	0.1	0.1	0.3	0.2
Carbohydrate (lactose)	G	4.8	4.4	5.1	4.9
Cholesterol	Mg	14	10	11	8
Calcium	Mg	120	100	170	195
Energy	Kcal	66	60	95	110
	Kj	275	253	396	463

Source: USDA (2007).

The composition of milk differs widely among species. Factors such as the type of protein; the proportion of protein, fat, and sugar; the levels of various vitamins and minerals; and the size of the butterfat globules, and the strength of the curd are among those that may vary. On the average, it contains 3.4% protein, 3.6% fat, and 4.6% lactose, 0.7% minerals and supplies 66 kcal of energy per 100 grams. These compositions vary by breed, animal, and point in the lactation period. The protein ranges for the four breeds of Jersey, Zebu, Brown swiss and Holstein-Friesian is 3.3% to 3.9%, while the lactose range is 4.7% to 4.9%. (Goff *et al*., 2014). Research has also proven that seasonal factors have great impact on the composition of the milk. Casein to fat ratio are higher in milk produced during May and August, while fat in milk from February is higher and total protein is lower than in months of May and August. Age is not an important factor affecting the composition of milk although there appears to be a tendency for the fat content to decrease with increasing age. However, the health of the cow may affect milk composition considerably. Cows suffering from mastitis or inflammation of the udder give milk low in fat, casein and lactose and high in chlorides. The composition of milk varies appreciably over the period of lactation. The milk given immediately after calving (colostrum) contains a very high percentage of total solids (up to 19%) due mainly to the very high content of protein and fat. During the first week after calving there is a progressive change towards normal composition. The quality of food also has an effect on the composition of the milk. Poor quality feed depresses the protein content and continuous underfeeding results inmilk of lower fat content (Balogun *et al.,* 2016). The composition of milk is very important for the manufacture of dairy products. The yield of butter and cheese obtained from milk depends on the quantities of the major constituents present in milk. Butter yield depends on the fat content of the milk while cheese yield depends on the fat and protein contents. In addition, depending on the type of cheese being made, the ratio of fat to protein (casein) in the milk will affect the quality of the cheese.

2.1.1 Milk Production

Milk is the source of nutrients and immunological protection for the young cow. The gestation period for the female cow is 9 months. Shortly before calving, milk is secreted into the udder in preparation for the new born. At parturition, fluid from the mammary gland known as colostrums is secreted. This yellowish colored, salty liquid has very high serum protein content and provides antibodies to help protect the newborn until its own immune system is established. Within 72 hours, the composition of colostrums returns to that of fresh milk, allowing to be used in the food supply.The period of lactation, or milk production, then continues for an average of 305 days, producing as much as 9000 or more kg of milk. This is quite a large amount considering the calf only needs about 1000 kg for growth.Within the lactation, the highest yield is 2-3 months post-parturition, yielding 40-50 L/day. Within the milking lifetime, a cow reaches a peak in production about her third lactation, but can be kept in production for 5-6 lactations if the yield is still good.

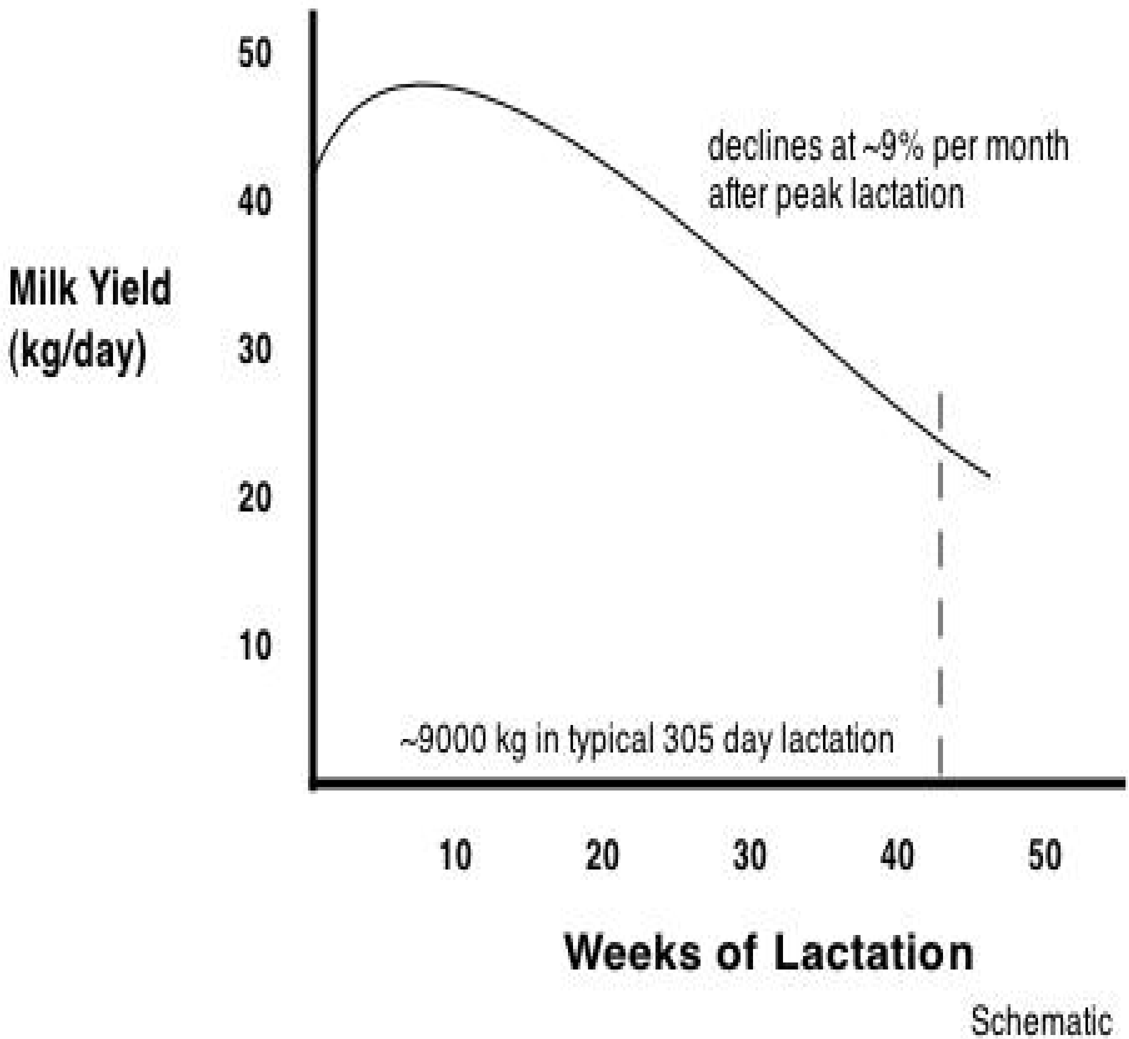

Figure 1: Graphical representation of milk yield of a cow

Source: (FAOSTAT, 2010)

About 1-2 months after calving, the cow begins to come into heat again. She is usually inseminated about 3 months after calving so as to come into a yearly calving cycle.

FAO reported Israel dairy farms as the most productive in the world, with a yield of 12,546 kilograms (27,659 lb.) milk per cow per year. This survey over 2001 and 2007 was conducted by ICAR (International Committee for Animal Recording) across17 developed countries. The survey found that the average herd size in these developed countries increased from 74 to 99 cows per herd between 2001 and 2007. A dairy farm had an average of 19 cows per herd in Norway, and 337 in New Zealand. Annual milk production in the same period increased from 7,726 to 8,550 kg (17,033 to 18,850 lb) per cow in these developed countries. The lowest average production was in New Zealand at 3,974 kg (8,761 lb) per cow. The milk yield per cow depended on production systems, nutrition of the cows, and only to a minor extent different genetic potential of the animals. What the cow ate made the most impact on the production obtained. New Zealand cows with the lowest yield per year grazed all year, in contrast to Israel with the highest yield where the cows ate in barns with an energy-rich mixed diet. The milk yield per cow in the United States, the world's largest cow milk producer, was 9,954 kg (21,945 lb) per year in 2010. In contrast, the milk yields per cow in India and China – the second and third largest producers were respectively 1,154 kg (2,544 lb) and 2,282 kg (5,031 lb) per year. (FAOSTAT, 2012).

2.1.2 Milk Utilization

Milk produced under hygienic conditions from healthy cows should contain no more than 50 000 bacteria per milliliter. It is important that milk, whether it is for liquid consumption or for the manufacture of dairy products, is of good hygienic quality. Milk that is of poor hygienic quality may cause disease to humans and will result in poor quality products with low consumer acceptability. Milk produced under unhygienic conditions or from unhealthy cows may cause illnesses to humans including tuberculosis, brucellosis, sore throats, diarrhea and abdominal pains. Preventing the entrance and subsequent growth of bacteria in milk presents a constant challenge to those concerned with milk production and processing. The nutritive value of milk makes it essential that it is handled and treated with care. The bacteria that are normally in milk as it comes from the cow as well as bacteria from exterior sources, e.g. the surroundings and utensils, must be prevented from growing. Milk is an ideal medium for the growth of bacteria and if it is kept at above 16^{o}C the bacteria present will multiply rapidly thereby causing deterioration in quality and a reduction of its shelf-life. At temperatures below 10^{o}C the growth of bacteria is considerably reduced (O'Connor, 1993).

A 2008 review found evidence suggesting that consumption of milk is effective at promoting muscle growth (Roy, 2008). Some studies have suggested that conjugated linoleic acid, which can be found in dairy products, is an effective supplement for reducing body fat (Whigham *et a*l., 2007). With regards to the claim of milk promoting stronger bones, there has been no association between milk consumption or excess calcium intake and a reduced risk of bone fractures. In 2011, *The Journal of Bone and Mineral Research* published a meta-analysis examining whether milk consumption might protect against hip fracture in middle-aged and older adults. Studies could find no association between drinking milk and lower rates of fractures (Bischof-ferrari *et al*., 2011). In order to prevent spoilage, milk can be kept refrigerated and stored between 1 and 4 °C (34 and 39 °F) in bulk tanks. Most milk is pasteurized by heating briefly and then refrigerated to allow transport from factory farms to local markets. The spoilage of milk can be forestalled by using ultra-high temperature (UHT) treatment. Milk so treated can be stored unrefrigerated for several months until opened but has a characteristic "cooked" taste. Condensed milk, made by removing most of the water, can be stored in cans for many years, unrefrigerated, as can evaporated milk. The most durable form of milk is powdered milk, which is produced from milk by removing almost all water. The moisture content is usually less than 5% in both drum- and spray-dried powdered milk.

When raw milk is left standing for a while, it turns "sour". This is the result of fermentation, where lactic acid bacteria ferment the lactose in the milk into lactic acid. Prolonged fermentation may render the milk unpleasant to consume. This fermentation process is exploited by the introduction of bacterial cultures (e.g. *Lactobacilli sp.*, *Streptococcus sp.*, *Leuconostoc sp.*, etc.) to produce a variety of fermented milk products. The reduced pH from lactic acid accumulation denatures proteins and causes the milk to undergo a variety of different transformations in appearance and texture, ranging from an aggregate to smooth consistency. Some of these products include sour cream, yogurt, cheese, buttermilk, kefir, and kumis, ice milk, pudding, hot chocolate and French toast. Milk is often added to dry breakfast cereal, porridge and granola. Milk is often served in coffee and tea. Steamed milk is used to prepare espresso-based drinks such as cafe latte (Yiu *et al*., 2006).

In Nigeria, the Fulani pastoralists process surplus fresh milk into various stable products like West African soft cheese (Warankashi), Nono (fermented skimmed milk) and Mai- shanu. The West African soft cheese which is the typical type of cheese found in Nigeria has a shelf life of 2-3 days when immersed in the whey. Milk is an extremely nutritious food. It is an aqueous colloidal suspension of proteins, fats and carbohydrates that contains numerous vitamins and minerals. Many of the pathogenic bacteria encountered do not grow well in milk but remain viable for undesirable lengths of time. Processing and utilization of dairy products in African countries is not well developed (Oladipo *et al.*, 2013).

2.2 Tigernut Production

Tigernuts are not actually nuts but tubers found on the root of a sedge plant. It was first discovered 4000 years ago and comes in several varieties. The tubers were originally cultivated by ancient Egypt's populations at the Nile valley. Their cultivation was subsequently extended throughout other areas with temperate climate and fertile soil. Reports have shown that tigernuts came to Spain from Africa (IHS, 2005; HBR, 2005; CVNews, 2006; Deatra, 1999). Tigernuts are edible tubers with a sweet nutty flavour. Other common names for these tubers are "earth almond" and "yellow nut sedge". They are quite hard and are generally soaked in water before consumption. In Egypt and the Mediterranean nut sedges were used as sources of food, medicine and perfumes.

Tigernut tubers were routinely roasted and consumed by nursing mothers. . The dried ground tubers were used in coffee and chocolate drinks. Oil extracted from the tubers was an ingredient in soap maikng as well as a lubricant for fine machinery. The leafy plant parts of the nut sedge were fed to livestock. Egyptians made very efficient use of the nut sedge. They used them in cultivation as early as 2400 BC. One such example of tigernuts is depicted in a wall painting of an Egyptain tomb in 15th century BC (Deatra, 1999). In the painting, workers are shown to be weighing the nuts while a scribe records their work. In another part of the same tomb, instructions were written for eating the tubers as sweets after grinding and adding honey. Tigernut tubers have been found in the tombs and are considered to be locally domesticated in Egypt. This gives the impression that the tubers were greatly valued by the Egyptian people as a food source (Deatra, 1999).

2.2.1 Nutritional composition of tigernuts and its products FAO (1988) and TTSL (2005) showed that tigernut tubers are rich in starch (20-30% of DW) and fat (20-28% DW) with small quantities of protein which is about twice of that of cassava. Table 2.1 showed the energy and nutrient content of tiegrnuts as reported by other researchers (Umerie *et al*.,1997); TTSL, 2005; Addy and Eteshola, 1984; Temple *et al*., 1990). Tigernuts have relatively higher fat content and gross energy, and in this regard compared better with nuts than that of cereals which also belong to the same other Cyperales. Research has been done on the oil extracted from the seeds of yellow nut sedge (*Cyperus esculentus* var. *esculentus*) as a non-conventional oilseed. This study was used to determine oil substitutes for more conventionally used oil types such as tigernut, palm and olive oils. Non-conventional oils would be less expensive and therefore more available to poorer (developing) countries.

Tigernut oil is 80% unsaturated fatty acid, mainly oleic (64.2 – 68.8 %) and this shows that tigernut oil has a good potential as a substitute for imported olive oil (Deatra, 1999; Mc

Namara, 2004; TTSL, 2005). Fat in diets provide twice much energy as carbohydrate or protein, thus low fat diets are recommended to aid weight control. Different types of fat (fatty acids) have different effects on health and the risk of diseases states such as coronary heart disease (CHD). Saturated fatty acids (SFA) increase levels of blood cholesterol and should be avoided whenever possible. There is evidence that the replacement of SFA with 20 monounsaturated fatty acid (MUFA) may have a favorable effect on the risk of CHD. Venho *et al.* (2000) investigated types of fat intake in relation to CHD risk in women and reported that for every increase of 5% in energy from MUFA there is a decrease in CHD relative risk of 0.81%.

Tigernut is a good source of phosphorous, potassium and iron. It also contains magnesium, calcium, zinc, copper, sodium and manganese (TTSL, 2005). Phosphorus found in plant is usually bound to a compound called phytate meaning that it is poorly absorbed from the gut into the body. Phosphorous (P)**,** together with calcium, constitutes the bulk of the mineral substance of the bones and teeth. It plays a part in the formation of ATP (an energy compound indispensable for "activating" glucose, fatty acids, etc) and in improvement of intellectual performance. Phosphate is important in the body. It helps regulate acidity/ alkalinity by acting as a buffer (Moore, 2004). Potassium (K) is important in maintaining electrolyte and chemical balance between the tissue cells and the blood. K is the most important neural element in intracellular behaviour. It plays a part in numerous enzymatic reactions and in important physiological processes, such as cardiac rhythm, nervous conduction, and muscular contraction. Iron (Fe) in food is often in a complex form. Vitamin C aids in the absorption of iron. Vitamin C is a reducing agent and changes Fe into a more easily absorbed form. An acid medium also helps Fe absorption. Consequently, Fe helps prevent anaemia. Zinc has a wide variety of functions in the body and is found in all body tissues. It is involved in many enzyme reactions including those involved in energy generation from carbohydrate, fat and protein. It also has a role in cell division, the transport of carbon dioxide and oxygen in the blood and also in immunity. Since it has a wide range of role in the body, symptoms of zinc deficiency are also wide-ranging and include a delay in wound healing, poor appetite, a suppressed immune system and poor growth (Moore, 2004; Wardlaw and Kessel, 2002). Magnesium is also involved in many enzyme systems and in particular those involving the currency of energy in the body, ATP. Magnesium is also required for the synthesis of proteins, the production of energy and muscle contraction (Moore, 2004). Research studies have suggested that a low intake of magnesium may increase the risk of coronary heart disease (Al-Delaimy *et al*., 2004) and type 2 diabetes (Lopez-Ridaura *et al*., 2004).

2.2.2 Importance of tigernuts

Nutritional and health importance of tiger nuts Tigernuts and its products are rich in carbohydrates, mono-, di-, and polysaccharides (TTSL, 205; Moore, 2004). They contain relatively high levels of protein, oleic acid (monounsaturated fatty acid which has a bigger resistance to chemical decomposition) and fat (TTSL, 2005). Tigernuts have excellent nutritional quality with a fat composition similar to olive oil and rich mineral content, especially phosphorus and potassium (FAO, 1988; Moore, 2004). Tigernut oil has a mild, pleasant flavour and is considered as food oil similar but superior in quality to olive oil. The polyunsaturated fatty acid content (linoleic acid & linolenic acid) is enough to cover daily minimum needs of about 10 g (TTSL, 2005), Moore (2004) oil has high content of Vitamin E (alpha-tocopherol), and thus higher oxidative stability than other oils**,** due to its content of polyunsaturated fatty acids and gamma-tocopherol.

Tigernuts may need to rely significantly on its health benefits, promoting a rich monounsaturated fatty acid content, high vitamin E levels and prebiotic qualities (NUTRA, 2005; Moore, 2004). Vitamin E, an antioxidant which protects the body form free radical attack, is vital for the maintenance of cell membranes. It may also play an important role in delaying cells from aging thereby improving the elasticity of skin. Vitamin E is good for treatment of acne and other skin „alterations". It is particularly important in areas of the body exposed to oxidative stress such as the lungs and the red blood cells. Vitamin E may reduce the risk of cancer and CHD due to its role as anti oxidant, however research in this area is 22 currently inconclusive (Moore, 2004; Wardlaw and Kessel, 2002). In supra-nutritional doses, Vitamin E has been
Claim to benefit diseases associated with oxidative stress including cardiovascular disease, cancer , Alzheimer's and Parkinson's diseases (Brigelius-Flohe, 2002). Tigernut oil has therapeutic properties as it reduces "bad" cholesterol (LDL-cholesterol) and increases the "good" one (HDL-cholesterol). It can also reduce levels of triglycerides in blood, reduce risk of formation of bloody clots, produce dilatation in veins and prevent arteriosclerosis**.** Tigernuts may play an important role in the prevention and nutritional therapy for cardiac pathologies, due to its high content of monounsaturated fatty acids (Oleic acid) to improve metabolism and health (TTSL, 2005; Moore, 2004). Tigernut oil exhibits positive effects on digestive secretions (gastric, pancreatic and bile), due to high content of oleic acid, the most powerful stimulator of production of Cholecistokinine (TTSL, 2005). Tigernuts may prevent heart attacks, thrombosis and activate blood

circulation. The high contents of soluble glucose in tigernuts prevent cancer. Recently, some investigators discovered that they reduce the risk of suffering colon cancer. Tigernuts have relative antioxidant capacity, because they contain considerable amount of water-soluble flavonoid glycoside (a phytochemical). Consumption of antioxidant could protect the immune system of malnourished populations. The intake of antioxidant containing foods may delay the progression of HIV infection to AIDS (ONRG, 2005) The high fibre content of tigernuts combined with its delicious taste makes them ideal for healthy eating. The high content of fiber content of tigernut has a good effect on digestion (TTSL, 2005). This is because fibre stimulates digestive juices, contributes to a longer feeling of fullness and speeds up transit in the intestinal tract and so prevents constipation. Tigernut may have prebiotic qualities, a result of the short chain carbohydrates called oligosaccharides, which feed probiotic bacteria helping to promote intestinal health (NUTRA, 2005). Moore (2004) reported that levels of oligosaccharides have not been measured in tigernut, however they were found in the milky drink "*horchata*". The oligosaccharides, which are short chain carbohydrates and have shown the most promise as potential prebiotics. Recent research has also suggested that oligosaccharides may increase the absorption of the minerals calcium and magnesium. These effects were observed with doses in the range of 5-10 g per day (Delzenne, 2003). The amino acid profile of tigernuts is dominated by arginine . Although arginine is not an essential amino acid, it has been termed „conditionally essential‟. It is essential in the fetus and the neonate. In adults it may have a role in disease states especially where tissue is being broken down such as in sepsis or trauma (Wu *et al.*, 2000). The area of 23 arginine remains an exciting area of nutrition research, however it must be noted that some of the effects may require pharmacological doses, at a much higher level than that supplied by our regular diet (Moore, 2004). Many of the postulated beneficial roles of arginine are related to the fact that it is a precursor for nitric oxide (NO). NO is a vasodilator produced by the endothelial cells of the vascular system and has an important role in the regulation of the cardiovascular system. This „endothelium-derived relaxation" is impaired in conditions such as diabetes, high blood pressure and high plasma cholesterol (Pieper *et al*.,1996) demonstrated in animal studies that oral administration of L-arginine could normalize endothelial relaxation in diabetic rats. Guigliano *et al.* (1997) however, showed that intravenous infusion of L-arginine (3-5g) to humans could reduce blood pressure in diabetic men. In men with high blood cholesterol levels, 21g per day of intravenously administered arginine improved endothelium derived relaxation. This intravenous dose is much higher than the level of arginine consumed in a usual diet (Moore, 2004).

Tigernuts are free from gluten cholesterol. They have very low sodium content (TTSL, 2005). Scientific analysis on the "nutritional and dietetic aspects of tigernuts" (Farré, 2003), "digestive aspects of tigernuts" (Bixquert, 2003) and "effects of tigernuts on heart diseases and related aspects" (Valls, 2003) concluded that tigernuts have high content of oleic acid, have positive effects on cholesterol levels due to high content of vitamin E. Tigernuts are suitable for diabetic persons, ideal for children, older persons and sportsmen and are very healthy. For many years, tigernuts have been considered to have adequate properties to fight respiratory infections, and some stomach illnesses (CVNews, 2006). The scientific analysis by Zimmermann (1987) concluded that tigernuts reduce the risk of colon cancer and are suitable for diabetic and obese persons. To this date, *horchata* is considered an effective remedy for diarrhea, according to popular tradition in Valencia, Spain CVNews, 2006). "*Horchata"*, a natural sweet tasting vegetable milk can be extracted directly from tigernuts and used as a refreshing drink which can also serve as substitute for cow milk. The following characteristics make "horchata" a perfect substitute of vegetable milk:

- It is ideal milk for persons that don"t tolerate gluten (celiacs) or that are allergic to cow milk and its derivates**.**
- It helps in reduction of LDL ("bad") cholesterol and increases HDL ("good") cholesterol because of its high contents of oleic acid and Vitamin E, which has an antioxidant effect on fats.
- The high content of oleic acid and the amino acid arginine prevents arteriosclerosis.
- It is suitable for diabetic persons.
- It is recommended for persons with digestion disorders, flatulence and diarrheas, because of the content of digestive enzymes (lipase, catalase and amylase).
- It is high phosphorus, potassium, calcium, magnesium and iron
- .It has considerable amounts of vitamins C and E (TTSL, 2005 and Moore, 2004).

2.3Coconut

Coconut (*Cocos nucifera L*.)is a monocotyledonous plant of the familyArecaceae and the monospecific genus Cocos.Recent theory indicates that it originated in Polynesia (Wikipedia, 2016). Almost every part of the coconut tree can be used in either making commercial products or meeting the food requirements of rural communities (Teulat et. al., 2000). This palm can be found growing over most of the islands and coasts of the subtropics and tropics (Dowe, 2010).Coconut plays a significant role in the economic, cultural and social life of over 80tropical countries(Oyoo*etal*., 2015). Over the years, the coconutpalm has been referred to as “the tree of the Heavens” and “tree of a hundred uses” (Rillo, 1999) which indicates its remarkable usefulness and qualities. It is a major source of income for rural families and plays an important role in wealth generation and improving the quality of life in many tropical countries. Sustainable yields can be increased by providing high quality planting materials along with improved management of the coconut plant.At the small scale farming level, coconut is an important contributor to food security. At the industrial level, value-added products of coconut areimportant sourcesof employment and income in rural communities (Singh et al., 2008).

Botanically, the coconut fruit is a drupe, not a true nut. Like other fruits, it has three layers: the exocarp, mesocarp, and endocarp. The exocarp, mesocarp and endocarp. The exocarp and mesocarp make up the husk of the coconut. The coconuts sold in the shops of non-tropical countries often have had the exocarp removed. The mesocarp is composed of a fiber calledcoir, which has many traditional and commercial uses. The shell has tree germination pores (stoma) or ‘eyes’ that are clearly visible on its outside surface once the husk is removed. A full sized coconut weighs about 1.44kg(3.21b). It takes around 6000 full grown coconuts to produce ton copra (Bourke *et al.,* 2009). Each and every part of the plant is useful in one way or another and not even an inch of the tree goes waste. The coconut palm is intertwined with life itself from the food they eat to the beverage they drink and derive almost everything necessary to sustain the life. All the daily needs like household utensils, baskets, cooking oil, furniture and cosmetics are made from coconut palm.

2.3.1 Historical development of coconut

The origin of coconut palm is the subject of controversy. Cook (1901) was one of the earliest modern researchers to draw conclusion about the location of origin of *Cocos nucifera* based on its current day worldwide distribution. He hypothesized that coconut originated in the Americans based on the fact that America coconut populations predated European contact and because he considered pan tropical distribution by ocean current improbable. Theo Heyerdahl later used his hypothesis of the American origin to support his theory that the pacific islanders originated in South America. However, more evidence exists for indo-pacific origin ether around Melanesia or the Indian ocean (Jackson, 2011). The oldest fossils known of modern coconut dating from Eocene period from around 37 to 55 million years ago were found in Australia and Indian. However, older palm fossils such as some of nipa fruit have been found in America (Jackson, 2011).

Botanists place the origin of coconut palm in Papua New Guinea area, in some very distant past, on basis of occurrence of the nearest botanical relatives (Child, 1974). Harries (1990) argues its origin in Malaysia and stated that the distribution of Cocos spp.

is a relic of Gondwanaland. The current theory suggests it to be native to Malaysia, a bio-geographical region roughly defined as an area that includes Southeast Asia, Indonesia, Australia, New Guinea, and several Pacific Island groups. The Indian subcontinent and South Asia designated as Hindustani center, an important region of diversity of crop plants among the eight centers, has been identified as a secondary center of origin of *Cocos nucifera*(Randhawa, 1980). In the Hawaiian Islands, the coconut is regarded as a Polynesian introduction, first brought to the islands by early Polynesian voyagers from their homelands in the South Pacific. Human beings seem to have no role in the spread of coconut to various places as these can float for very long periods, and then sprout when they lodge on the shore. This was dramatically demonstrated when coconuts were found growing on an island created by volcanic activity in Krakatoa in 1929–30 (Child, 1974).

2.3.2 Production and cultivation

Coconut palms are grown in more than 90 countries of the world, with a total production of 61 million tonnes per year. Most of the world production is in tropical Asia, with Indonesia, the Philippines, and India accounting collectively for 73% of the world total (FAO 2013). Coconut palms grow throughout thetropics in a band around the world from 25°North and 25° South of the equator. The palm can be found in Southeast Asia, Indonesia, India, Australia, the Pacific Islands, South America, Africa, the Caribbean, and the southern extremes of North America. The ideal growing conditions for coconut palms include free-draining aerated soil often found on sandy beaches, a supply of fresh groundwater, humid atmosphere, and temperatures between 27°C and 30°C. It thrives on sandy soils and is highly tolerant of salinity and prefers areas with abundant sunlight and regular rainfall (1,500–2,500 mm [59–98 in] annually), which makes colonizing shorelines of the tropics relatively straightforward. Coconuts also need high humidity (at least 70–80%) for optimum growth, which is why they are rarely seen in areas with low humidity. However, they can be found in humid areas with low annual precipitation such as in Karachi, Pakistan, which receives only about 250 mm (9.8 in) of rainfall per year, but is consistently warm and humid. Coconut palms require warm conditions for successful growth, and are intolerant of cold weather. Some seasonal variation is tolerated, with good growth where mean summer temperatures are between 28 and 37 °C (82 and 99 °F), and survival as long as winter temperatures are above 4–12 °C (39–54 °F); they will survive brief drops to 0 °C (32 °F). Severe frost is usually fatal, although they have been known to recover from temperatures of −4 °C (25 °F). They may grow but not fruit properly in areas with insufficient warmth (Chan *et al*., 2006).

2.3.3 Nutritional value of coconut

According to United State Drug Administration (USDA), per 100g serving with 354 calories, raw coconut meat supplies a high amount of total fat 33g and carbohydrate 24g. Micronutrients contained in significant content include Pantothenic acid (Vitamin B5), having 20% of the Daily value and the dietary minerals such as iron, phosphorus and zinc.

2.3.4 Utilization of coconut

The multiplicity and versatility of uses of coconut tree can be best judged by an Indonesian saying: “There are as many uses for the coconut as there are days in the year”.

It is considered as one of the ten most useful trees in the world, and one among the five Devavrikshas (God’s trees) known in India, providing food for millions, especially in the tropics. Around 83 functional uses of various parts of palm listed range from food to stuffing of coir in pillows, preparation of beds, ropes, mats, utensils of daily use such as spoons, drainers, brooms, chains, toddy drawers, door mats, floor mats, musical instruments, furniture, cots, rosary boxes, brush, fuel, scoops, containers, oil bottles, toothbrushes, hooka-bases, neck belts and blinds for bulls used for plowing and oil crushing, cricket bats, and various types of children’s toys (Watt, 1889). Coconut provides five types of food products: coconut water, coconut milk, sugar, oil, and meat. The cavity of the immature nut is filled with “water” containing sugars; the water has been used as a refreshing drink since ancient times as mentioned by Kalidasa (380–413 AD) (Aiyer, 1956). Coconut water is sterile until the coconut is opened (unless the coconut is spoiled).

The various parts of the coconut have a number of culinary uses. The seed provides oil for frying, cooking, and making margarine. The white, fleshy part of the seed, the coconut meat, is used fresh or dried in cooking, especially in confections and desserts such as macaroons. Desiccated coconut or coconut milk made from it is frequently added to curries and other savory dishes. Coconut flour has also been developed for use in baking, to combat malnutrition (Grimwood, 1975). Coconut chips have been sold in the tourist regions of Hawaii and the Caribbean. Coconut butter is often used to describe solidified coconut oil, but has also been adopted as a name by certain specialty products made of coconut milk solids or puréed coconut meat and oil. Dried coconut is also used as the filling for many chocolate bars. Some dried coconut is purely coconut, but others are manufactured with other ingredients, such as sugar, propylene glycol, salt, and sodium metabisulfite. Shredded or flaked coconut is used as a garnish on some foods (Roehl, 1996). Some countries in Southeast Asia use special coconut mutant called Kopyor coconut (Kopyor in Indonesia) or macapuno (in the Philippines) as dessert drinks. Discussed below are some of the important uses of coconut components

- **Coconut water:** Coconut water serves as a suspension for the endosperm of the coconut during its nuclear phase of development. Later, the endosperm matures and deposits onto the coconut rind during the cellular phase. It is consumed throughout the humid tropics, and has been introduced into the retail market as a processed sports drink. Mature fruits have significantly less liquid than young, immature coconuts, barring spoilage. Coconut water can be fermented to produce coconut vinegar. Per 100-gram serving, coconut water contains 19 calories and no significant content of essential nutrients (Paniappan, 2012).The water and flesh from young coconuts contains the full range of B vitamins, with the exception of B6and B12. Young coconut water is also high in minerals, particularly calcium (for bones), magnesium (for the heart), and potassium (for muscles). An average young coconut provides 3 grams of dietary fiber which helps proper digestion and elimination. Fresh coconut water is very high in electrolytes much higher than most sports drinks. This makes it a great choice for athletes and children who exercise a lot. Finally, the water of young coconuts is completely sterile and so close to the structure of human blood that it can be transfused directly. In fact, it was regularly used during World War II for wounded soldiers when blood plasma was low. Coconut water with sandalwood paste is used for bathing (Mittre, 1991).

- **Coconut sugar:** The cloudy liquid is easily boiled down to syrup, called jaggery, coconut molasses, or dhiyaa hakuru and then crystallized into a rich dark sugar, almost exactly like maple sugar. Sometimes it is mixed with grated coconut for candy as kamamai in Kiribati and ‘addu bondi’, in the southernmost atoll, Addu Atoll. Boiled toddy, known as jaggery, with lime makes a good cement (Mittre, 1991). Coconut sugar is classified as a low glycemic index food and is considered to be healthier than traditional white and brown sugar. It can be used as a 1:1 sugar substitute for coffee, tea, baking, and cooking.Coconut sugar has high mineral content and is a rich source of potassium, magnesium, zinc, and iron. In addition to this it contains vitamins B1, B2, B3, and B6. Compared to brown sugar, coconut sugar has twice the iron, four times the magnesium, and over 10 times theamount of zinc.
- **Coconut oil:** Another product of the coconut is coconut oil. It is commonly used in cooking, especially for frying. It can be used in liquid form as would other vegetable oils, or in solid form as would butter or lard. Dried meat of coconut becomes copra and is processed for oil. In India, people make a vegetarian butter called ‘ghee’ from coconut oil; it is also used in infant formulations. Coconut oil has been used as a cooking medium in South India and Southeast Asia since ancient times. It is of course primary culinary fat in Kerala. Coconut oil is extensively used as a hair oil and body oil, for burning wicked oil lamps, and as lubricant. It has medicinal properties and is used in cosmetics. Much of the fat in coconut oil is saturated which is in form of lauric acid. Coconut oil may be a better alternative to partially hydrogenated vegetable oil when solid fats are

required (Tarrago-Trani *et al.*, 2006). In addition virgin coconut oil is composed mainly medium chain triglycerides which may not carry the same risks as other saturated fats. Early studies on the health effects of coconut oil is on partially hydrogenated coconut oil which creates trans fats and not virgin coconut oil which has a different health risk profile (kintanar, 1998).

2.3.5 Coconut milk

Coconut milk, not to be confused with coconut water, is obtained primarily by extracting juice by pressing the grated coconut white kernel or by passing hot water or milk through grated coconut, which extracts the oil and aromatic compounds. It has a total fat content of 24%, most of which (89%) is saturated fat, with lauric acid as a major fatty acid. When refrigerated and left to set, coconut cream will rise to the top and separate from the milk. The milk can be used to produce virgin coconut oil by controlled heating and removal of the oil fraction. A protein-rich powder can be processed from coconut milk following centrifugation, separation, and spray drying (Naik *et al.*, 2012).Coconut milk contains a large proportion of lauric acid, a saturated fat that raises blood cholesterol levels by increasing the amount of high-density lipoprotein cholesterol which is also found in significant amounts in breast milk and sebaceous gland secretions (Amarasiri et al., 2006). This may create a favourable blood cholesterol profile, though it is unclear if coconut oil may promote atherosclerosis through other pathways (Mensink et al., 2003). According to a nutritional journal on obesity (2003), coconut milk is rich in medium-chain fatty acids (MCFAs), which the body processes differently from other saturated fats. MCFAs may help promote weight maintenance without raising cholesterol levels.

2.3.6 Health benefit of coconut milk

According to (seed guides, 2004) coconut milk nutrition contains a very high level of saturated fats, but the saturated fats found in coconut milk are mainly short and medium chain fatty acids, which are usually not stored by the body as fats. Instead, such short and medium chain fatty acids have found to provide instant energy to the body. Apart from these fatty acids, coconut milk contains many other essential nutrients, which can be attributed for several health benefits.

Some of the most important benefits of coconut milk are explained below:

- A major part of the fats found in coconut milk is lauric acid, which has been found to exhibit antibacterial, antifungal and antiviral properties. This fatty acid can boost the immune system and its diseases fighting ability.
- Lauric acid can also be helpful in maintaining the elasticity of the blood vessels and in keeping them clean, which can lower the risk for conditions like, atherosclerosis and heart disease.
- Coconut milk also contains several antioxidant compounds, which can provide protection against the harmful free radicals and their damaging effects on the body cells and tissues.
- Coconut milk can improve the health of the digestive system and promote digestion. It can relieve the symptoms of stomach ulcers and acid reflux disease as well.

- Coconut milk can give about 22% of the recommended daily allowance of iron. With such a high level of iron, it can help to treat anaemia caused by iron deficiency.
- Apart from these, coconut milk may help to relax the nerves and the muscles, control blood sugar and reduce joint inflammation.
- Coconut contains vitamin E which helps in improving fertility.

2.4History and Origin of Cheese

The origin of cheese making is lost in unrecorded history. There is evidence to suggest that cheese was made as far back as 700 BC. There are numerous references to cheese making in the Bible while the writings of Homer and Aristotle indicate that cheese was made from the milk of cows, goals, sheep, mares and asses. Around 3 AD trade in cheese between countries especially on sea routes became so great that the Roman emperor Diocletian had to fix maximum prices for the cheese (O' Connor, 1993). Cheese is an ancient food whose origins predate recorded history. There is no conclusive evidence indicating where cheesemaking originated, either in Europe, Central Asia or the Middle East, but the practice had spread within Europe prior to Roman times and, according to Pliny the Elder, had become a sophisticated enterprise by the time the Roman Empire came into being (Subbaraman, 2012). The earliest evidence of cheese-making in the archaeological record dates back to 5,500 BCE, in what is now Kujawy, Poland, where strainers with milk fats molecules have been found.Earliest proposed dates for the origin of cheesemaking range from around 8000 BCE, when sheep were first domesticated. Since animal skins and inflated internal organs have, since ancient times, provided storage vessels for a range of foodstuffs, it is probable that the process of cheese making was discovered accidentally by storing milk in a container made from the stomach of an animal, resulting in the milk being turned to curd and whey by the rennet from the stomach.

There is a legend – with variations – about the discovery of cheese by an Arab trader who used this method of storing milk. The earliest cheeses were likely to have been quite sour and salty, similar in texture to rustic cottage cheese or feta, a crumbly, flavorful Greek cheese.

Cheese produced in Europe, where climates are cooler than the Middle East, required less salt for preservation. With less salt and acidity, the cheese became a suitable environment for useful microbes and molds, giving aged cheeses their respective flavors. The earliest ever discovered preserved cheese was found in the Taklimakan Desert in Xinjiang, China, and it dates back as early as 1615 BCE (Watson *et al.,* 2014).

2.4.1 Production of cheese

Cheese is made in almost every country of the world and there exist more than 2000 "varieties. Despite the large number of varieties cheese may be classified into different groups, i.e. ripened and unripened cheese, cheese with low or high fat content and cheese with soft or hard consistency. Unprocessed milk held at high ambient temperate tires has a shelf-life from 2-3 hours up to 24 hours. Cheese, however, has a shelf-life from 4-5 days up to five years depending on the variety. Cheese therefore provides an ideal vehicle for preserving the valuable nutrients in milk and making them available throughout the year. Although cheese making survived as an art form for more than 7000 years the advance of scientific knowledge has led to a better understanding of the raw material, milk, and the cheese making and ripening process. A number of developments have taken place which aids the cheese maker to produce a better and more consistent quality cheese.

These developments include the findings of Pasteur in 1857 that bacteria harmful to the cheese process as well as ,pathogenic microorganisms could be destroyed by heat; the introduction of pure cultures of microorganisms (starters) to produce acid at a reliable and consistent rate; the refinement of the extraction of rennet from calves resulting in better quality cheese curd and the development of objective methods, e.g. the acidometer, for the assessment of curd and cheese quality (O'Connor 1993).

In 2014, world production of cheese from whole cow milk was 18.7 million tonnes, with the United States accounting for 29% (5.4 million tonnes) of the world total followed by Germany, France and Italy as major producers (table). Other 2014 world totals for processed cheese include (Workman, 2016).

- from skimmed cow milk, 2.4 million tonnes (leading country, Germany, 845,500 tonnes)
- from goat milk, 523,040 tonnes (leading country, South Sudan, 110,750 tonnes)
- from sheep milk, 680,302 tonnes (leading country, Greece, 125,000 tonnes)
- from buffalo milk, 282,127 tonnes (leading country, Egypt, 254,000 tonnes)

During 2015, Germany, France, Netherlands and Italy exported 10-14% of their produced cheese. The United States was a marginal exporter (5.3% of total cow milk production), as most of its output was for the domestic market. The manufacture of wara cheese is widespread in Nigeria and a similar cheese called Wogachi is made in the northern provinces of Benin republic, a French speaking country to the west of Nigeria. The Fulani's of Northern Nigeria are traditionally cattle rearers and they have access to fresh milk from Zebu Bos indicuscattles. Wara cheese making is thought to have started in this region and as a result of the normadic lifestyle of the Fulani was spread to other parts of Northern Nigeria, Kwara, Oyo, Ogun, Ondo and the Benin republic (Bamidele, 2006).

There are many types of cheese, with around 500 different varieties recognized by the International Dairy Federation,more than 400 identified by Walter and Hargrove, more than 500 by Burkhalter, and more than 1,000 by Sandine and Elliker.The varieties may be grouped or classified into types according to criteria such as length of ageing, texture, methods of making, fat content, animal milk, country or region of origin, etc.—with these criteria either being used singly or in combination,but with no single method being universally used.The method most commonly and traditionally used is based on moisture content, which is then further discriminated by fat content and curing or ripening methods.Some attempts have been made to rationalize the classification of cheese—a scheme was proposed by Pieter Walstra which uses the primary and secondary starter combined with moisture content, and Walter and Hargrove suggested classifying by production methods which produces 18 types, which are then further grouped by moisture content (Patrick, 2000).

2.4.2 Processing and nutrient composition

The principle of cheese processing is based on the coagulation of the protein in milk, during which about 90% of the milk fat is encapsulated. The coagulated mass is called curd, the remaining liquid is called whey. Curd consists mainly of milk proteins (casein) and milk fat; while whey mainly contains water, milk sugar (lactose), protein (serum proteins) and B vitamins. Due to the fact that wara is made of milk it contains various microflora and lactobacillus species which makes it susceptible to spoilage within a short period of time. Traditionally, it is preserved in its whey which last barely a day or two, it's being boiled in water to make it tough so as to increase its keeping quality which can last 3-4 days in a chilled condition.

Cheese is usually stored in its whey and consumed fresh, but this can only last for 3-5 days after which spoilage occurs. It is sometimes fried and used as a meat-substitute in stews and soups, or smoke-dried to enhance its keeping qualities. However, all these increase its shelf life by only a few extra days or few weeks at best. The capacity to preserve cheese like any other food is directly related to the level of technological development. The slow progress in upgrading traditional food processing and preservation techniques in West Africa contributes to food and nutrition insecurity in the sub-region. Simple, low-cost, traditional food processing techniques are thus essential to not only eradicatestarvation and Protein-Energy Malnutrition in the sub region, but also alleviate poverty by minimizing food wastage and generating income (Oladipo *et al.*, 2012).

Cheese is obtained by coagulating pasteurized milk at a specific temperature, pH and processing time with an appropriate coagulant. The preferred coagulant comes from

Sodom apple leaf extract (Calotropis procera) because the cheese made with this coagulant has a sweeter flavor and a higher protein content compared to the cheese made with the other coagulants (Omotosho *et al.*, 2011). The coagulated milk is poured into a small basket or strainer in order to drain and to give the cheese the desired shape and size. Hundreds of types of cheeses are produced based on their different characteristics and method of productions. Their styles, textures and flavors depend on the origin of the milk (including the animal's diet), whether they have been pasteurized or not, the butterfat content, the bacteria and the mold, the processing and aging. Herbs, spices or wood smoke may be used as flavoring agents (Omotosho *et al.*, 2011).

'Warankashi' is consumed in its fresh unripened state, fried or used as a meat-substitute in stews and soups. Cheese is an excellent source of protein, fat and minerals such as calcium, iron and phosphorus, vitamins and essential amino acids and therefore is an important food in the diet of both young and old people (Oladipo *et al.*, 2012).

Table 2: Nutrients in cheese and other food products per 100gram

Food	Protein (mg)	Fat (mg)	Calcium (mg)	Iron (mg)	Thiamine (mg)
Cheddar cheese	26	33.5	800	0.5	0.04
Cottage cheese	13.6	4.0	60	0.1	0.07
Yoghurt	5.0	1.0	180	0.09	0.09
Whole meal bread	8.8	2.7	23	2.5	0.26
Egg	12.3	10.9	52	2.0	0.09
Potato	2.1	0.1	8	0.5	0.11
Butter	0.4	82.0	15	0.16	0.00

Source: (O'Connor, 1993).

According to USDA the nutritional value of cheese varies widely. Cottage cheese may consist of 4% fat and 11% protein while some whey cheeses are 15% fat and 11% protein, and triple-crème cheeses are 36% fat and 7% protein. In general, cheese is a rich source (20% or more of the Daily Value, DV) of calcium, protein, phosphorus, sodium and saturated fat. A 28-gram (one ounce) serving of cheddar cheese contains about 7 grams (0.25 oz.) of protein and 202 milligrams of calcium. Nutritionally, cheese is essentially concentrated milk: it takes about 200 grams (7.1 oz) of milk to provide that much protein, and 150 grams (5.3 oz) to equal the calcium.

2.4.3 Cheese in African diet

Compared with the quantities produced in Europe and North America the amount of cheese produced in Africa is quite small. Most of the cheese produced in Africa is made on a small scale and generally at farm l%.There is very little scientific information available on the cheese made at farm level; the recipe or the process is passed on from parents to children by observation and practical experience. The quality of the farm-made cheese can be variable because the ingredients and techniques used are so dependent on local conditions and available facilities. Different methods of coagulation of the milk may be used, e.g. the juice extract from local growing plants, acid precipitation combined with heating and rennet coagulation may be employed. In addition, defined starters are generally not used (they may not always be available and are expensive) and therefore acid production by the naturally occurring microorganisms or whey from previous batches are likely to be rather variable. Even though produced in small quantities cheese is a very valuable food and source of nutrients and cash for many people in Africa (O'Connor, 1993).

The processing and utilization of dairy products in many African countries is not well developed. The Masai tribesmen of Kenya and Tanzania who herd large numbers of zebu cattleand consume large quantities of milk have not developed a cheesemaking tradition. Nomads in developing arid countries have suffered the tragedy of hunger in recent years. It would be nutritionally advantageous if the idea of producing cheese, a stable and nutritious food which can be stored for periods of time, were introduced to tribal chiefs(Bamidele, 2006).

The production of cheese in African countries has been increasing this millennium from 430,000 metric tonnes in 1990 to 743,000 metric tonnes in 2002 (FAO, 2002). This increase is expected to continue as more diversification takes place and more food processing is encouraged. Egypt has the highest cheese production of African countries and accounts for about 67 % of African cheese production. Nigeria produced 7022 metric tonnes of cheese in 1994, and production increased slightly to 8000 metric tonnes in 2003. Most African countries increased production for total cheese (all kinds) between 1994 and 2003 except Zimbabwe that suffered a decrease in production during this period.There is very little scientific information available on the cheeses made in Africa. Recipes and processes are passed from generation to generation by observation and practical experience (Bamidele, 2006).

In countries with a dairy industry cheese provides an ideal vehicle for preserving the valuable nutrients of milk. Cheese is an excellent source of protein, fat and minerals such as calcium, iron and phosphorous, vitamins and essential amino acids, making it an important food in the diet of both young and old. In Nigeria, due to the lack of industrial production of traditional cheese varieties the nutritional benefits of cheese have not been utilized fully. The soft Wara cheese produced in Nigeria at farms makes use of local ingredients. The vegetable rennet used for production of Wara cheese is produced from a native sodom apple plant (*Calotropisprocera*) which can be cultivated all year round. Therefore, there is no need for imported rennet. A better understanding of the mechanism of action of this plant rennet is required if cheeseproduction is to be carried out on a larger scale using sodom apple extract as the coagulant. Cheesemaking in Africa is largely dictated by tradition. Due to shortage of milk, cheese production is expensive and powdered milk and cheese may be imported. The cheese produced is generally consumed very soon after manufacture, primarily because of the poor shelf life under ambient conditions. The problems are further compounded by the fact that during periods of surplus milk production the prices for milk, butter and cheese are considerably lower than in periods of lower milk production(Bamidele, 2006).

2.4.4 Ingredients used in cheese production

There are a number of ingredients which are essential for cheesemaking but some ingredients, e.g.colouring, added chemicals etc. are not required for all varieties of cheese (O'Connor, 1993).

- **Milk:** Good quality milk from the cow, sheep, goat etc is required. Knowledge of its chemicalcomposition and bacteriological quality is desirable if cheese of consistent quality is to be made.
- **Starter:** Certain cheese varieties require starters (pure cultures of lactic acid bacteria) containing organisms with specific functions, e.g. flavor development.

he recipe will indicate the type and quantity of starter to be used and temperature conditions.

- **Colour:**The recipe and the market will determine if coloring matter should be used. Occasionally it is required to bleach the original color of the milk and to whiten the curd.
- **Chemicals:** Chemicals such as calcium chloride and sodium nitrate are recommended in recipes for some varieties of cheese to improve curd quality and prevent the growth of organisms which may causeproblems during the ripening or maturing of the cheese.
- **Coagulant:** Rennet is the usual coagulant used but the juice extract ofsome fruits and plants, e.g. lemonsand *Calotropis procera* may be used for some cheese varieties.
- **Salt:** Salt (sodium chloride) may be added to some varieties of cheese, the quantity and method of addition depending on the recipe. Salt may be added directly to the mill or curd pieces; it may be rubbed into the finished cheese or the cheese may be immersed in a brine solution.

2.4.5 Steps in conventional cheese production

Below are listed the principal steps in cheese making. Not all these steps are used for all cheese varieties and such steps as may be used will be determined by the recipe (O'Connor, 1993).

- **Milk treatment:** Milk may be heat treated, e.g. 73^0C for 15 seconds, to destroy pathogens and reduce microbial numbers. The milk may be standardized, i.e. the fat content may be increased or reduced or the casein-to-fat ratio may be adjusted.

- **Starter:** Good quality starter is required. The type and quantity will be determined by the cheese recipe. For some cheese varieties commercial starter preparations are not used; natural fermentation or whey from the previous lot of cheese made may be used.
- **Coagulation:** Various coagulants are used, e.g. rennet and lemon juice. The coagulants bring about, under defined conditions of temperature, quantity and time, the coagulation of the milk into a firm jelly-like mass.
- **Cutting the coagulum:** The coagulum may be cut with knives into curd particles of a defined size, e.g. 1-2 cm, or it may be ladled into containers or cheese moulds. The cutting or ladling of the coagulum is a very important step in the manufacture of some cheese varieties as it determines the rate of acid development and the body (firmness) and texture of the cheese.
- **Heating or cooking the curd:** Heating (40-45^{0}C) the curds and whey affects the rate at which whey is expelled from the curd particles and the growth of the starter microorganisms. For some cheese hot water may be added to the curds and whey.During heating the curds and whey may be stirred to maintain the curd in the form of separate particles.
- **Whey removal:** After heating and stirring and when the curd particles have firmed and the correct acid development have taken place the whey is removed allowing the curd particles to match together.
- **Curd texturing:** It is a characteristic of some cheese varieties that the curd mass is allowed to develop a texture along with further whey drainage and acidity development.

- **Milling the curd:** When the curd has reached the desired texture it is broken up into small pieces to enable it to be salted evenly. Milling the curd can be done either by hand or mechanically.
- **Salt addition:** Salt may be added to the cheese curd as described above or it may be incorporated in the finished cheese by immersion in a brine solution. The addition of salt retards the growth of lactic organisms and slows down acid production. Salt also retards or prevents the growth of bacteria which may cause flavorand other defects in the cheese.
- **Moulding or hooping the curd**: After milling (and salting) the curd pieces are packed into moulds or containers the size and shape of which are determined by the variety of cheese made. The moulds may be made either of metal, plastic or wood.
- **Pressing**: Pressing the curd in the moulds assists in some whey removal and compacts the curd pieces into the shape of the mould. The temperature of the curd and the extent of pressing are critical to the quality and appearance of the final cheese. The pressing may be done by the application of weights or, as in large-scale cheese manufacture, by mechanical cheese presses.
- **Removal of cheese from the mould**: When the cheese has been pressed for the prescribed duration it is removed from the press and the mould. The cheese thus obtained may be further treated by bandaging in cheese cloth, by the application of colour or coating the cheese with wax or a thin layer of butter. Such operations help lie cheese to maintain its shape and enhance its appearance.

- **Storage:** The cheese variety will determine the period and the conditions (temperature and humidity) of storage. Market conditions including consumer preferences and prices may exert an influence on the duration and conditions of storage.

Not all of the above steps are necessary in the manufacture of all cheese varieties. Fresh unripened cheese which can be consumed almost immediately after manufacture requires only the initial steps of heating the milk, coagulation and breaking the coagulum and the separation of the curds and whey as seen in the local method of producing cheese.

2.4.6 Steps in local cheese production

- **Milk treatment:** Milk is pasteurized at 73^{o}C for 15 seconds in order to destroy pathogenic and spore forming microbes and also to reduce the microbial load. The fat content of the milk and the casein to fat ratio may also be adjusted
- **Coagulation:** Several coagulants such as alum, extracts of sodom apple or pawpaw leaf, lemon juice, steeped water, calcium chloride have been used. The coagulant aids the coagulation of the milk into a firmly jelly-like mass, under certain conditions of temperature, quantity and time.
- **Whey removal:** After heating and stirring and when the curd particles have firmed and the correct acid development have taken place the whey is removed allowing the curd particles to match together.
- **Curd texturing:** Curds are allowed to develop textures along with further drainage and acidity development which helps to enhance the characteristics of the cheese variety.

Fresh milk

↓

Sieving (to remove dirt particles)

↓

Pasteurization (72^{0}C, 20mins)

↓

Cooling (40^{0}C)

↓

Addition of Sodom apple extracts (35g/3litres of milk)

↓

Cooking (70^{0}C, 20mins)

↓

Scum removal

↓

Curd cutting to facilitate whey expulsion

↓

Whey drainage (2mins)

↓

Warankashi

Figure 2:Local Cheese Production

Source: Ukeyima *et al.,* 2010.

CHAPTER THREE
MATERIALS AND METHODS

3.1 Material Collection and Preparation

Sodom apple leaves (*Calotropis procera*) and fresh cow milk will be obtained from a nomadic settlement located within ikole-Ekiti (precisely Asin) Local Government Area of Ikole, Ekiti State, Nigeria. The fresh cow milk was collected from a healthy cow aseptically, packaged in a sterile white container, and placed in a cooler containing ice crystal. This helped to prevent postharvest contamination and the increase in temperature during the long transportation. The tiger nut and coconut was purchased from a local market in Ekiti. Other necessary processing

equipment and reagents were obtained in the Laboratory of Food Science Department Federal University OyeEkiti. The production process was carried out carefully using the method described by Balogun*et al*. (2016), and Hussein *et al*. (2016) which is a traditional processing method.

3.2 Production of Tigernut Milk

Tigernut milk was produced by modified method described by Belewu and Belewu(2007). The tigernut was selected, the tigernut was then washed and then hydrated, then the juice (thick and milky liquid) was extracted from the tigernut using a blender, the tigernut milk was then stored (It is important if it is not going to be consumed same day to keep it in constant movement using mixer at low temperature or frozen).

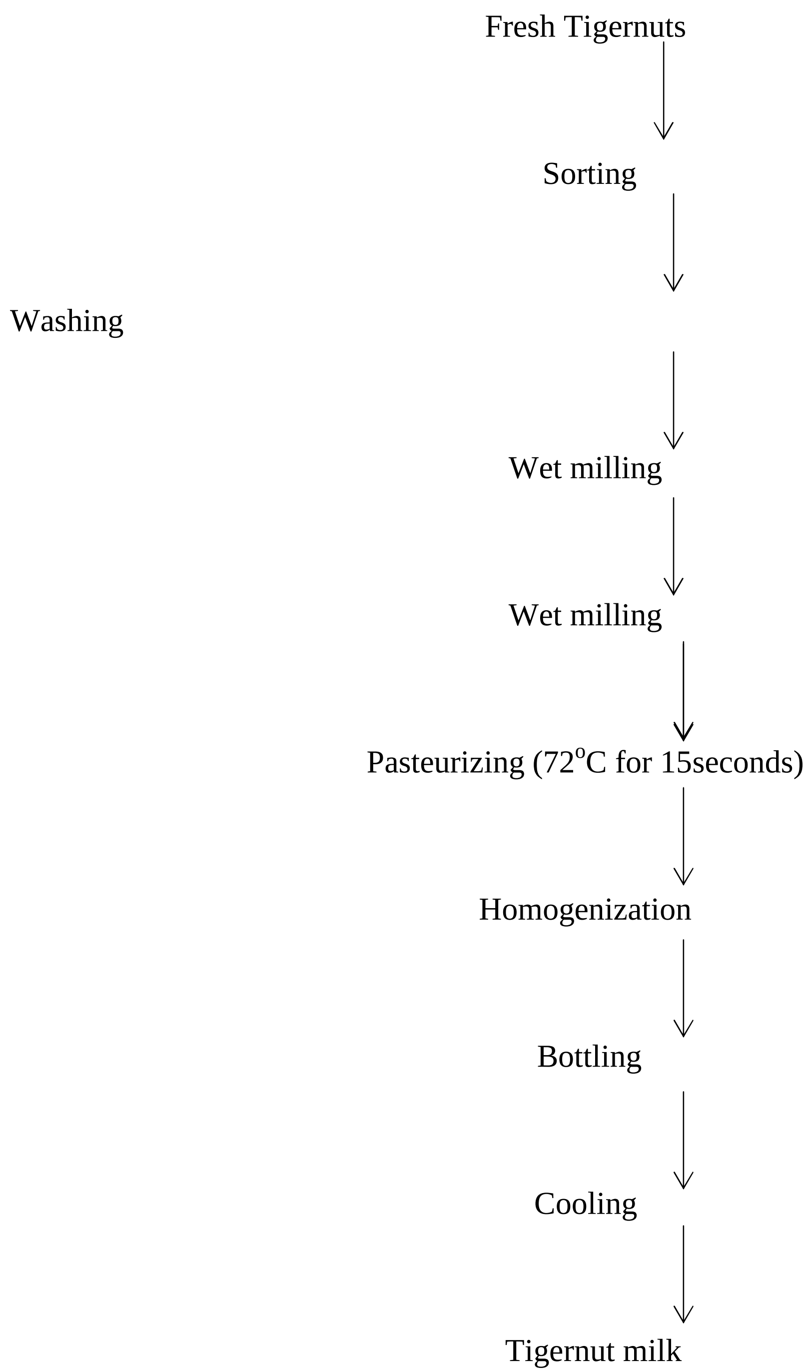

Figure 3:Production of Tigernut Milk

Source: (Balogun *et al*., 2016)**.**

3.3 Production of Coconut Milk

A whole coconut was purchased, shelled, and shredded using a traditional coconut grater. Coconut milk was produced by mixing the shredded pulp with an equal weight of warm distilled water (60°C) in a blender, filtered through a double-layered cheese cloth, and manually squeezed with a twisting motion to extract most of the milk. The extracted emulsion was pasteurized and stored at 30°C before the production of cheese and used within 24 hours of production (Balogun *et al*., 2016).

Coconut fruit

↓

Deshelling

↓

Washing

↓

Size reduction

↓

Blending(with warm water)

↓

Sieving

↓

Pasteurizing (75^{o}C for 20 minutes)

↓

Cooling

↓

Coconut milk

Figure 4: Production of Coconut milk

Source: (Balogun *et al.*, 2016)

3.4 Production of Cheese (Warankashi)

The portion of the sieved milk for warankashi production from the cow, tigernut and coconut was measured. The variation in the measurement was done according to the method described by Balogun *et al*. (2016) with modification. Cheese samples were produced using 1000ml as standard with formulation by partial substitution of cow milk with coconut milk and tigernut milk at varying proportions, to create a range of 75:0:25; 75:25:0; 50:25:25; 25:5025; 25:25:50 (v/v). The hundred percent (100%) cow milk warankashi was produced and used as control.The sodom apple leaves were used as a coagulant. They were washed and pounded using a mortar and pestle. 15 g of the pounded sodom apple leaf was added to each of the milk blends.

The method of Balogun *et al*., (2016) with some modifications was used to produce the cheese. Fresh cow milk, tigernut milk and coconut milk blend was transferred into a metal pot. The metal pot was then placed over a slow burning fire and heated to a temperature of 50 °C. 15 g of pounded Calotropis procera leaves was added to the warmed milk. The milk was heated slowly. The metal pot was removed from the fire to cool to about 40 °C, and the leaves were sieved out. The metal pot was then placed on a slow burning stove to boil until it reached boiling point. The milk was kept at the boiling point until it coagulated and there was a visible separation of curds and whey. The curds were then scooped using a scooping spoon into a small set of small raffia baskets (akpere-wara) placed over a container for whey collection. The raffia basket helped to give the final cheese product with a desired shape. After a few minutes, the curd was formed and placed in a clean container containing cool water (Balogun *et al*., 2016)

Milk Blends

↓

Addition of coagulum (*Calotopis procera*)

↓

Boiling (50^{o}C, 10minutes)

↓

Cooling (20^{o}C)

↓

Remove plant parts

↓

Boiling (95^{o}C, 20minutes)

↓

Curd

↓

Draining

↓

Wara (Cheese)

Figure 5: Production of Cheese(warankashi)

Source: (Balogun *et al*., 2016)

Table 3: Research methods for the production of cow/cow-tigernut-coconut cheese blend

Sample	Cow milk (%)	Tigernut milk (%)	Coconut milk (%)	Sodom Apple leaves (g)
C_{100}	100	-	-	15
$C_{75}T_0$ CC_{25}	75	0	25	15
$C_{75}T_{25}CC_0$	75	25	0	15
$C_{50}T_{25}CC_{25}$	50	25	25	15
$C_{25}T_{50}CC_{25}$	25	50	25	15
$C_{25}T_{25}$ CC_{50}	25	25	50	15

Keys:

C_{100}: 100% cow milk**; $C_{75}T_0CC_{25}$: (7**5% cowmilk+25% coconut milk**) $C_{75}T_{25}CC_0$: (**75% cowmilk + 25% Tigernut milk**) $C_{50}T_{25}CC_{25}$: (**50% cow milk + 25% Tigernut milk + 25% coconut milk**) $C_{25}T_{50}CC_{25}$: (**25% cow milk +50% Tigernut milk + 25% coconut milk**) $C_{25}T_{25}CC_{50}$: (**25% cow milk + 25% Tigernut milk+ 50% coconut milk**).**

Plate 1: Cheese produced from Cow-tigernut-coconut milk blends

Plate 2: Cheese produced from Cow-tigernut-coconut milk blends

3.5 Proximate Analysis

The proximate composition which includes moisture content, fat, protein, crude fibre, ash and carbohydrate were all carried out on the cheese samples using the AOAC 2010 method. The percentage yield and total titratable acidity was carried out using method described byBalogun *et al*. (2016).

3.5.1 Determination of moisture content of cow-tigernut-coconut cheese

Apparatus: oven, Petri-dishes, desiccators, weighing balance

Methods: The Petri dishes were washed with distilled water and kept in oven at temperature of 105°C for some minute to condition them for use. The Petri dishes were transferred to the desiccators to cool.The empty Petri dishes were weighed using the digital analytical balance, 3g of the samples were weighed into the Petri dishes, and they were transferred into the oven for drying for about 3hours. The Petri dishes containing the sample were then transferred to the desiccators to cool for 10 minutes and were weighed. Drying and weighing was repeated until constant weight was achieved.

Calculation

Weight of empty petri-dish = W_1

Weight of petri-dish + sample before drying = W_2

Weight of petri-dish + oven dried sample = W_3

Total weight loss = W_2-W_3

Weight of sample = W_2-W_1

% Moisture = W_2-W_3x 100

W_2-W_3 1

3.5.2Determination of fat content of cow-tigernut-coconut cheese

Apparatus: Soxhlet apparatus and accessories, oven, desiccator and analytical balance

Reagents: Petroleum ether spirit

Methods: Two grams of each sample was placed in a weighed filter paper and was tied together. The filter paper plus sample was then transferred a fat free extraction, thimble and plug lightly with ab cotton wool. The thimble was placed in the extractor and fitted up with reflux condenser and 250ml flask is then filled to ¾ of its volume with petroleum ether (b.pt 40^{o}C- 60^{o}C). The soxhlet flask extractor plus condenser set were placed on the heater. The heater was put on for 6 hours with constant running water from the tap for condensation of ether vapour. The set was constantly watched for ether leaks and the heat source was adjusted appropriately for ether to boil gently the ether was let to siphon over several times (over at least 10-12 times) until it is short of siphoning. It is after this is noticed that any ether content of extractor is carefully drained into the ether stock bottle. After 6 hours, the heating mantle was put off, in a single containing sample is then removed and dried in an oven to a constant weight

Calculation:

Weight of sample = weight after drying – weight of filter paper

Weight of oil/loss = initial weight before extraction – weight of sample

$$\% \text{ crude fat} = \frac{\text{Weight loss}}{\text{Initial weight}} \times \frac{100}{1}$$

3.5.3 Determination of Fatty Acidsof cow-tigernut-coconut cheese

Standard methods of the AOAC (1996) were used to determine the fatty acids content of the cheese (Method 989.05). The esterswere analyzed using a gas chromatograph (GC) system Model HP 6890 series (USA). The capillary column: Modelnumber, Agilent 1909 1 N- 116; Hp-INNOWax.

3.5.4Determination of crude protein contentof cow-tigernut-coconut cheese

Apparatus: Analytical balance, digestion flasks, heating mantle, hot plate, 50ml burette, 10ml pippete, 10ml measuring cylinder, 100ml beaker.

Reagent: Copper sulphate, sodium sulphate, boric acid indicator, hydrochloric acid, distilled water, methyl red-bromocresol green mixed indicator, concentrated H_2SO_4.

Digestion

Two grams of sample was weighed carefully into kjedahl digestion flask to ensure that all sample material got to the bottom of the flask. 0.5grams of copper sulphate, 10g of sodium sulphate and 25ml of concentrated sulphuric acid was added into the digesting flask containing the weighed sample. Heat was then applied slowly at first to prevent undue frothing. Digestion continues for 3 hours until the digester becomes clear pale green. It was left to cool and 100ml of distilled water was rapidly added. The digestion flask was rinsed 2-3 times and the rinsing was added to the bulk.

Distillation

Markham distillation apparatus was used for distillation. The distillation apparatus was steamed up and about 10ml of the digest was added into the apparatus via a pipette and allowed to boil. 10ml of sodium hydroxide was added from the measuring cylinder so that no ammonium is lost. It was distilled into 50ml of 2% boric acid containing screened methyl red indicator.

Determination:

5ml of the digest was pipetted into the body of the apparatus via the small funnel aperture. 5ml of 10% (W/V) sodium hydroxide was added through the same opening with the 5ml pipette. The mixture was steam-distilled for 2 minutes into a 50ml conical flask containing 10ml of 2% boric-acid plus mixed indicator solution and it was placed at the receiving tip of the condenser. The boric-acid plus indicator solution changed colour from blue to green which showed that all the ammonia liberated has been trapped.

Titration

The green colour solution obtained was then titrated against 0.01N HCl contained in a 50ml burette. At the end point or equivalent point, the green colour turned to blur colour which indicate that all nitrogen trapped as Ammonium Borate [$(NH_4)_2BO_3$] had been removed as Aluminum chloride (NH_4Cl). The percentage nitrogen in this analysis was calculated using the formula:

%Nitrogen= Titre valuex Atom mass of Nitrogen x Normality of HCl used x 4

The crude protein content is determined by multiplying percentage Nitrogen by a constant factor of 6.25

Therefore % Crude Protein = % Nitrogen x 6.25

3.5.5 Determination of amino acids contentof cow-tigernut-coconut cheese

The amino acids content cheese was determinedaccording to the procedure of Amado*et al.,* (1983). Thesample analyzed by using an amino acid analyzer LC 3000Epoendrof, Germany. The instrument condition was: flowrate, 0.2 ml/min, pressure of buffer from 0.0 to 50.0 bars,pressure of reagent from 0.0 to 150.0, reaction temperature 123 °C.

3.5.6Determination of crude fibre contentof cow-tigernut-coconut cheese

Apparatus: heating mantle, crucibles, furnace, sieve cloth, fibre flask, funnel, analytical weighing balance and desiccator.

Reagents: 0.255N H_2SO_4, 0.313N NaOH

Determination: Two grams of the de-fatted sample was accurately measured into the fibre flask and 100ml of 0.255N of H_2SO_4 was added. The mixture was heated for 30 minutes with the heating mantle. The hot mixture was allowed to cool and filtered through a filter paper. The filtrate obtained was thrown off and the residue was returned to the fibre flask to which 100ml of (0.3131N NaOH) was added and heated for another 30 minutes. The mixture was cooled and filtered through a filter paper. The filter paper plus the residue were transferred into a crucible and oven-dried at 105°C for 3 hours to drive off moisture. The oven-dried crucible containing the filter paper was cooled in a desiccator and later weighed to obtain the weight W_1. The crucible with weight W_1 was transferred to the muffle furnace for ashing at 550°C for 4 hours. The crucible containing grey ash (free of carbonaceous material) was cooled in the desiccator and weighed to obtain W_2. The difference W_1-W_2 gave the weight of fibre. The percentage fibre was obtained by the formula:

$$\%\text{Crude Fibre} = \frac{d \quad w \ o \ r \qquad b \qquad A\ hi \quad - w \ o \ r \qquad A \qquad A\ hi}{w \quad ht\ o \quad the\ sa} \times 100$$

3.5.7Total ash determination of cow-tigernut-coconut cheese

Apparatus: Porcelain crucibles, desiccator, analytical balance and muffle furnace

Two grams of the sample was weighed and transferred into a weighed porcelain crucible. This was transferred and combusted in a muffle furnace set at 550°C and was left for about 4 hours, about this time the sample had turned white and was free of carbon. The crucibles containing ash was cooled at room temperature in a desiccator and weighed. This was done in duplicate; the percentage ash was then calculated from the formula below:

$$\%\text{ Total Ash} = \frac{w \quad ht\ o \quad c \qquad + a\ h - w \quad ht\ o \quad c}{W \quad ht\ o \quad sa} \; x \; 100$$

3.5.8Determination of carbohydrate contentof cow-tigernut-coconut cheese

The total carbohydrate content of each of the sample was derived by difference from the results obtained from other parameters as indicated below.

% Total Carbohydrate= 100- (% moisture content + % crude protein + % crude fat + % total ash + % crude fat)

3.5.9 Determination of percentage yield of cow-tigernut-coconut cheese

The yield was determined by a method described by Balogun *et al*. (2016). The yield of warankashi from the cow milk-coconut milk-tigernut milk blends/mix and the whole cow milk was determined by the calculation as follows:

$$\text{Yield of warankashi (\%)} = \frac{\text{weight of warankashi}}{\text{weight of milk}} \times \frac{100}{1}$$

Weight of milk = volume (ml) of cow milk-coconut milk-tigernut milk or whole cow milk used

Weight of warankashi = weight (g) of warankashi (either from blends or whole cow milk) produced.

(Assuming 1g = 1ml).

3.5.10 Total titratable acidity

Apparatus: beaker, burrete funnel and filter paper

Reagent: phenolpthalene, 0.01N of NaOH

Ten grams of the sample was dissolved in 30 ml of distilled water in a beaker and stirred. The mixture was then filtered into a 100ml standard volumetric flask. The filtrate was made up to 100 ml. 10 ml of the filtrate was pipetted into a beaker and 1 drop of phenolphthalein was added. The mixture was then titrated against the standard 0.01 N sodium hydroxide solution until a light pink colour was attained. The reading of the burette was recorded. This was done in duplicates.

Calculations:

$$\frac{N(NaOH) \times \text{Titre Value} \times \text{Lactic-Acid value} \times \text{dilution factor}}{10ml} \times \frac{100}{1}$$

N= normality of NaOH (0.01) Dilution factor = 10

Titratable acidity for lactic acid (constant) = 0.09 Unit = gram of acid/mol

3.5.11 Sensory evaluation

Sensory evaluation will be conducted using a 9-point hedonic scale and a semi-trained panel consisting of 20 member's familiar with the consumption of warankashi. The panelists will be instructed to evaluate the coded samples for colour, taste, texture, aroma, and overall acceptability. Each sensory attribute was rated on a 9point hedonic scale (1 = dislike extremely and 9= like extremely) (Iwe, 2003).

3.6 Microbiological Analysis of Cow-Tigernut-Coconut Cheese

The media used for enumeration of microorganisms in this study includes nutrient agar and potato dextrose agar (PDA) agar. The media were prepared according to the manufacturer's specification. These media were sterilized in an autoclave at 121^{O}C for 15minutes .

One gram of each sample was dissolved in sterile de-ionized water and serially diluted. One milliliter of appropriate dilutions was seeded on plate count agar using spread plate method, and the medium was then incubated at 37°C for 24 h. The plate count agar was examined and colonies present were counted and recorded after incubation at 37°C for 24 h, to get the total colony count in CFU/g. This was done to monitor the microbial load for 14 days at 7 days interval and the shelf-life of each sample was also monitored.

3.7 Statistical Analysis

SPSS 16.0 was used to statistically analyze the data obtained from the study. Results obtained are triplicate determinations and were subjected to an analysis of variance to determine the significant differences among the samples, and the means were separated using the Duncan test. A completely randomized research design was used.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Proximate Composition of Cheese Produced from Cow-Tigernut-Coconut Milk Blends

The result of proximate composition of cheese produced from cow-tigernut-coconut milk blends is presented in Table 4. There were significant (p<0.05) differences in all the parameters evaluated. This difference could be attributed to the addition of coconut milk and tigernut milk at different levels and the resultant difference in the proportion of cow milk present in the samples. The moisture content of cheese (*warakanshi)* samples ranged from 53.92% to 56.66% with samples containing $C_{75}T_0CC_{25}$ having the lowest moisture content while sample $C_{25}T_{25}CC_{50}$ contains the highest moisture content and no significant difference was observed between samples $C_{25}T_{25}CC_{50}$ and $C_{50}T_{25}CC_{25}$as shown in Table 4. This result shows that the blending of cow-tigernut-coconut milk significantly (p<0.05) influenced the moisture content of cheese(*warankashi)* products. The moisture content increase with addition of vegetable sourced milk. This can be further proved from samples with lowest moisture content havingzero percent in either coconut milk, tigernut milk or both as seen in samples $C_{75}T_0CC_{25}$; $C_{75}T_{25}CC_0$; and C_{100}. The result agrees with the findings of Balogun *et al. (*2016) who reported increase in moisture content as the level of coconut increases

(66.69% and 71.86% for cheese produced from 100% cow milk and 70:30% cow-coconut milk respectively).

The protein content of cheese produced from cow-tigernut-coconut milk blends shows variations with additions of tigernut milk and coconut milk. The protein content ranged from 14.59% to 18.77% with sample $C_{25}T_{25}CC_{50}$ having the lowest protein content and this may be due to the high amount of coconut milk in the blend since coconut milk contains the least amount of protein of all the three milk used, while $C_{75}T_{25}CC_{0}$ had the highest protein content. Sample $C_{75}T_{0}CC_{25}$ has the second highest protein content and is significantly different to the control sample C_{100}. This result is similar to the findings of Balogun *et al.* (2016) who reported that sample with 75:25% of cow-coconut cheese has the highest protein content of her samples.

Hussein *et al.* (2016) also reported increase in the crude protein as the proportion of tigernut milk is increased in the blend for the cow- tigernut cheese production. (25:75% cow- tigernut milk 'warankashi' had the highest value (18.50%) followed by 50:50 % cow- tigernut milk '*warankashi'* (16.50%) and 75:25 % cow- tigernut milk '*warankashi'* (15.60%) had the lowest value). The differences in the findings may be due to many factors such as differences species of tigernut used, the maturity and nutrient composition of the tigernut and coconut as well as the amount of protein available in the milk after the extraction process.

There are significant difference in the fat content of cheese produced from cow-tigernut-coconut milk blends with samples $C_{75}T_{25}CC_{0}$ and $C_{25}T_{25}CC_{50}$of cow-tigernut-coconut cheese having a value of 12.90% and 21.49% respectively. The value of control sample (C_{100}) was 19.51% which is close to the highest fat content although significantly ($p<0.05$) different. The fat content of the samples can be seen to be dependent on the proportion of cow milk and coconut milk in the samples as Balogun *et al.* (2016) reported that the fat content increases with increase in the coconut milk and this is as a result of the high fat content of the milk and supporting this was that coconut milk is rich in saturated fatty acids, particularly lauric acid and fat is important as a source of energy in the human body. Hussein *et al.* (2016) reported that the fat content of the 'warankashi' decreases as the percentage composition of tigernut milk increases suggesting that the fat content is dependent largely on the cow milk. Similar results were reported by Okorie and Adedokun (2013) for the blending of cow-bambara nut milk 'warankashi', Adejuyitan *et al.* (2014) for tigernut -coconut 'warankashi' and Chikpah *et al.* (2016) for cow-tigernut milk. The value of fat found in this study was closely related to 13.4% fat recorded by Uaboi-Egbenni*et al*. (2010) and 20% reported by Mustafa *et al.* (2013). This result implies that cow- tigernut -coconut milk 'warankashi' blended with up to 25% tigernut-milk and about 50% of coconut milk will be a good source of energy in human diet.

The ash content in foodstuffs is a measure of mineral element in food. The ash content of 'warankashi' products varied significantly ($p<0.05$) as sample $C_{75}T_{25}CC_{0}$ had a value of 3.03% closely followed by sample $C_{50}T_{25}CC_{25}$. Thus it can be deduced that increase in the percentage cow milk (i.e. above 50%) leads to increase in the ash content. This is supported by the findings of Balogun *et al.* (2016) who reported that there is decrease in the ash content as there is increase in the 100% cow-milk (1.02%) while in the cheese with added coconut milk it decreased from 0.88%-0.32% with 5% and 30% proportions of added coconut milk respectively. The increased in ash content with the addition of 25% of tigernut milk and coconut milk in the produced cow- tigernut-coconut milk ‘warankashi’ implies that it is a good source of minerals.

The crude fibre serve functions like increasing digestibility, lowering of plasma cholesterol levels, decreasing the incidence of colon cancer and others (Adejuyitan *et al.*, 2014). No noticeable value of crude fiber was detected.

Total carbohydrate content of the cheese samples ranged from 2.81% to 10.72% as seen in samples $C_{25}T_{25}CC_{50}$ and $C_{75}T_{25}CC_{0}$ showing that there are significant ($p<0.05$) differences between the samples. Sample $C_{25}T_{50}CC_{25}$ is not significantly ($p<0.05$) different to the sample with the highest carbohydrate content ($C_{75}T_{25}CC_{0}$). The control sample (C_{100}) has a carbohydrate content of 6.06% and differed significantly to the other samples. This result indicates that the total carbohydrate content is dependent on the tigernut milk level and this view can be strengthened by taking into account the high carbohydrate content of tigernut reported by Oladele*et al.* (2007) as containing 3.50-3.78% of water, 32-46% crude protein, 15.5-24.7% fat and crude ash of 3.97-4.25% and carbohydrates content from 41.2 to 46.99% on a dry matter basis.

Table 4: Proximate composition of cow-tigernut-coconut cheese

Samples	Moisture	Protein	Fat	Ash	Carbohydrate
(%)	(%)	(%)	(%)	(%)	
C_{100}	$55.18^{c}\pm0.25$	$16.75^{b}\pm1.58$	$19.51^{b}\pm0.74$	$2.52^{c}\pm0.02$	$6.06^{bc}\pm1.06$
$C_{75}T_{0}CC_{25}$	$53.92^{c}\pm0.81$	$17.25^{ab}\pm0.00$	$20.46^{ab}\pm0.53$	$2.72^{bc}\pm0.16$	$5.67^{c}\pm0.12$
$C_{75}T_{25}CC_{0}$	$54.59^{c}\pm0.01$	$18.77^{a}\pm0.16$	$12.90^{d}\pm0.22$	$3.03^{a}\pm0.14$	$10.72^{a}\pm0.06$
$C_{50}T_{25}CC_{25}$	$59.32^{a}\pm1.10$	$16.53^{b}\pm0.74$	$13.84^{d}\pm1.42$	$2.84^{ab}\pm0.17$	$7.49^{b}\pm0.91$
$C_{25}T_{50}CC_{25}$	$57.49^{b}\pm0.00$	$15.48^{bc}\pm0.43$	$15.71^{c}\pm0.01$	$1.92^{d}\pm0.15$	$9.42^{a}\pm0.29$
$C_{25}T_{25}CC_{50}$	$59.66^{a}\pm0.13$	$14.59^{c}\pm0.14$	$21.49^{a}\pm0.57$	$1.47^{e}\pm0.64$	$2.81^{d}\pm0.53$

Values are mean ±S.D. means with same superscript across a columns are not significantly ($p<0.05$) different

Sample keys: C_{100}: (100% cow milk) $C_{75}T_{0}CC_{25}$: (75% cow milk + 25% coconut milk) $C_{75}T_{25}CC_{0}$: (75% cow milk + 25% tigernut milk) $C_{50}T_{25}CC_{25}$: (50% cow milk + 25% tigernut milk + 25% coconut milk) $C_{25}T_{50}CC_{25}$: (25% cow milk + 50% tigernut milk + 25% coconut milk) $C_{25}T_{25}CC_{50}$: (25% cow milk + 25% tigernut milk + 50% coconut milk).

4.2 Percentage yield and Total Titratable Acidity of Cow-Tigernut-Coconut Cheese

The percentage yield and titratable acidity of cheese produced from blends of cow-tigernut-coconut milk were presented in Table 5. It was observed from the result that there were significant ($p<0.05$) differences in the percentage yield of the cheese samples which ranged from 13.43% of sample ($C_{25}T_{25}CC_{50}$) to 17.33% of the cheese sample ($C_{25}T_{50}CC_{25}$).This differences may be generally due to the nutrient composition of the various materials used in the blends, the nature and specie of the food materials and the quantities of the materials used in the production. The sample $C_{25}T_{50}CC_{25}$ was observed to have the highest yield, this was in agreement with the findings of Adedokun *et al.,* (2013) who reported increase in percentage yield of cheese as there was increase in the bambara milk in the blend for the cheese production (28.05% and 41.1% for 100% cow-milk cheese and 50% bambara milk supplementation respectively). There was also no significant difference between samples C_{100} and $C_{75}T_{25}CC_{0}$. The relatively low value of the percentage yield in the cheese produced from the higher proportion of cow milk may be due to the specie of the animal, age, sex, feeding habits and type of feeds given to the animal and the time and season of milking. It can be deduced from the findings that the percentage yield may depend on the available protein for curdling by enzyme.

Thus, it can be said that increase in the amount of protein rich materials used in supplementations in the production of cheese will lead to increase in the percentage yield of the cheese as seen in this research and also the findings of Adedokun *et al.,* (2013).

Balogun *et al.,* 2016 also reported that percentage yield decreases with increase in the percentage coconut milk in the blend. (26.71% for 100% cow milk to 13.55% to 70:30 proportion of cow-coconut mix). However as reported by Balogun *et al.,* (2016) the principles of cheese making involves the removal of water from milk with a consequent six to ten-fold concentration of protein, fat, minerals and vitamins by the formation of protein coagulum which further shrinks to expel whey. Therefore, the decline in the percentage yield of the cheese sample $C_{25}T_{25}CC_{50}$ can be attributed to the high proportion of the coconut milk which is also in agreement with the findings of Balogun *et al., (*2016) and also shows that coconut milk has partial potential to be used in the production of cheese.

Titratable acidity (TA) and pH are interrelated in terms of acidity, but have different impacts on food quality (Sadler and Murphy, 2010). The total acid available to react with sodium hydroxide solution during titration is TA whiles the pH gives a measure of the strength of the acid in food. Sadler and Murphy (2010) reported that the impact of an acid on food flavour is much more determined by TA than pH. The total titratable acidity (TTA) was found to be between 0.23% and 0.30%. The significant ($p<0.05$) differences between the samples can be attributed to the differences in the ratio of the cow milk in the blend as the samples with 25 percent cow milk as seen in samples with $C_{25}T_{50}CC_{25}$ and $C_{25}T_{25}CC_{50}$ have lower titratable acidity compared to samples with cow milk of 50 percent and above. This is supported by the findings of Abdel Moneim *et al.*(2012) who reported that cheese samples produced from cow milk has higher titratable acidity than those produced from goat milk and mixtures of both milk (0.23 ± 0.01%, and 0.13 ± 0.01% for cow and goat milk mozzarella cheese respectively). It was reported by

Adedokun *et al.,* (2013) that changes in pH and total titratable acidity depend on the effects of cheese ripening and condition of storage after cheese production.

Table 5: Percentage yield and total titratable acidity of Cow-Tigernut-Coconut Cheese

Samples	Yield (%)	TTA (%)
C_{100}	14.32^{bc}±0.90	0.29^{a}±0.02
$C_{75}T_{0}CC_{25}$	15.33^{b}±1.53	0.27^{a}±0.02
$C_{75}T_{25}CC_{0}$	14.33^{bc}±0.61	0.30^{a}±0.02
$C_{50}T_{25}CC_{25}$	13.38^{c}±0.58	0.30^{a}±0.03
$C_{25}T_{50}CC_{25}$	17.33^{a}±0.83	0.23^{b}±0.01
$C_{25}T_{25}CC_{50}$	13.43^{c}±0.67	0.24^{b}±0.03

Values are mean ±S.D. means with same superscript across a columns are not significantly (p<0.05) different

Sample keys: C_{100}: (100% cow milk) $C_{75}T_{0}CC_{25}$: (75% cow milk + 25% coconut milk) $C_{75}T_{25}CC_{0}$: (75% cow milk + 25% tigernut milk) $C_{50}T_{25}CC_{25}$: (50% cow milk + 25% tigernut milk + 25% coconut milk) $C_{25}T_{50}CC_{25}$: (25% cow milk + 50% tigernut milk + 25% coconut milk) $C_{25}T_{25}CC_{50}$: (25% cow milk + 25% tigernut milk + 50% coconut milk).

4.3 Microbiological quality of Cow-Tigernut-Coconut Cheese

The microbial count result showed that the growth rate of all microorganism increased as the number of days increased. The increase in the number of microorganism in the cheese samples may partially result from the curdling and partially from the retention of these microorganisms in the curd as the whey is run off. The increase in the microbial load may also be due to factors such as the moisture content of the milk samples in the blend, the nutrient composition of the cheese samples, environmental factors, storage condition, the differences in the pH and titratable acidity of the cheese products and milking process, milk collection and cheese making process as stated by Oladipo and Jadesimi (2012).

Table 6 below shows the growth of bacteria as cheese samples were stored at refrigeration temperature for three (3) days. There was significant ($p<0.05$) difference between the samples at different days of analysis with sample $C_{25}T_{50}CC_{25}$ having the highest bacteria growth in all the days of storage. This may be due to the highest ratio of the tigernut milk used in the blend as tigernut milk contains high amount of nutrients which can support microbial growth as it was reported by Vaneys *et al.* (2004) that tigernut milkcontains 5.6-11.5% of water, 9-14% crude protein, 15.5-24.7% fat and crude ash of 4.5-6.5% and carbohydrates content from 31.7 to 31.85% on a dry matter basis. In addition, control sample (C_{100}) was observed to have a value of 83.33 with the lowest growth of bacteria at day 1 been the sample $C_{75}T_0CC_{25}$ with a mean value of 66.67.

There was no significant ($p<0.05$)difference between samples $C_{25}T_{50}CC_{25}$ and $C_{25}T_{25}CC_{50}$ at day 1 and also samples $C_{50}T_{25}CC_{25}$and $C_{25}T_{50}CC_{25}$ at day 3. It can be deduced from the result that the rate of bacteria growth increases with increase in the addition of tigernut milk in the blend. This is also visible in the result shown in Table 7 with the bacteria count increasing with increase in the tigernut milk and overall with increase in the storage time. Sample $C_{25}T_{25}CC_{50}$ has a total bacteria count of 1.9 x 10^6cfu/g and sample $C_{25}T_{50}CC_{25}$ having a count of 1.8 x 10^6 cfu/g and at 48 and 72hours of storage the bacteria count of the sample increased to 4.5 x 10^6 and 8.4 x 10^6 cfu/g respectively. This increase is higher than that of the control sample C_{100} which has bacteria count of 8.3 x 10^5cfu/g, 1.9 x 10^6cfu/g and 7.9 x 10^6cfu/g for day 1, 2 and 3 respectively. This finding shows that cheese produced from blends of cow milk particularly with tigernut milk or other milk from plant sources get spoilt faster and thus preservation methods such as drying, frying and use of chemicals can be employed together with refrigeration to further enhance the keeping period and quality of the cheese as reported in the findings of Oladipo and Jadesimi. (2012).

Table 6: Bacteria count of Cow- Tigernut-Coconut cheese during storage

Samples	Storage period (days) 1	2	3
C_{100}	$83.33^{cd}\pm5.77$	$193.33^{c}\pm11.55$	$793.33^{ab}\pm170.10$
$C_{75}T_{0}CC_{25}$	$66.67^{d}\pm11.55$	$293.33^{bc}\pm83.27$	$733.35^{ab}\pm2.13$
$C_{75}T_{25}CC_{0}$	$125.33^{bc}\pm18.90$	$266.67^{bc}\pm130.13$	$786.67^{ab}\pm61.10$
$C_{50}T_{25}CC_{25}$	$133.33^{b}\pm23.09$	$353.33^{ab}\pm50.33$	$893.33^{a}\pm57.74$
$C_{25}T_{50}CC_{25}$	$180.00^{a}\pm20.00$	$450.00^{a}\pm86.60$	$840.00^{a}\pm69.28$
$C_{25}T_{25}CC_{50}$	$199.33^{a}\pm51.00$	$400.00^{ab}\pm50.00$	$650.00^{b}\pm50.00$

Values are mean ±S.D. means with same superscript across a columns are not significantly ($p<0.05$) different

Sample keys: C_{100}: (100% cow milk) $C_{75}T_{0}CC_{25}$: (75% cow milk + 25% coconut milk) $C_{75}T_{25}CC_{0}$: (75% cow milk + 25% tigernut milk) $C_{50}T_{25}CC_{25}$: (50% cow milk + 25% tigernut milk + 25% coconut milk) $C_{25}T_{50}CC_{25}$: (25% cow milk + 50% tigernut milk + 25% coconut milk) $C_{25}T_{25}CC_{50}$: (25% cow milk + 25% tigernut milk + 50% coconut milk).

Table 7: Bacterial count of Cow-Tigernut-Coconut cheese during storage

	Storage period (Days)		
Samples	1	2	3
C_{100}	8.3×10^5	1.9×10^6	7.9×10^6
$C_{75}T_0 CC_{25}$	6.7×10^5	2.6×10^6	7.3×10^6
$C_{75}T_{25}CC_0$	1.3×10^6	2.7×10^6	7.9×10^6
$C_{50}T_{25}CC_{25}$	1.3×10^6	3.5×10^6	8.9×10^6
$C_{25}T_{50}CC_{25}$	1.8×10^6	4.5×10^6	8.4×10^6
$C_{25}T_{25}CC_{50}$	1.9×10^6	3.8×10^6	6.5×10^6

Sample keys: C_{100}: (100% cow milk) $C_{75}T_0CC_{25}$: (75% cow milk + 25% coconut milk) $C_{75}T_{25}CC_0$: (75% cow milk + 25% tigernut milk) $C_{50}T_{25}CC_{25}$: (50% cow milk + 25% tigernut milk + 25% coconut milk) $C_{25}T_{50}CC_{25}$: (25% cow milk + 50% tigernut milk + 25% coconut milk) $C_{25}T_{25}CC_{50}$: (25% cow milk + 25% tigernut milk + 50% coconut milk).

There was significant ($p<0.05$) difference in the total fungal count of the cheese samples with increase in storage time. It was observed from Table 8 that there was increase in the total number of fungal with addition of tigernut milk and coconut milk in the sample. The control sample C_{100} has a value of 1.67 which is low compared to sample $C_{75}T_{25}CC_{0}$ which has a greater value of 100. Sample $C_{75}T_{25}CC_{0}$ shows increase in spoilage over the storage days while also having the highest number of fungal growth on a daily basis.

Decrease in fungal growth with increase in coconut milk was observed as sample $C_{50}T_{25}CC_{25}$ showed a steady value of having the least number of growths during days of storage. Table 9 shows the colony forming unit per gram of the cheese sample with the control samples 100% cow milk cheese having a fungal count of 1.7×10^{4}, 7.3×10^{5} and 9.3×10^{5} cfu/g for the three days of storage these findings was in contrast with the findings of Dauda. (2017) who reported no fungal growth for storage at day one although this may be due to the difference in samples as the report was on fried cheese. The increase in the fungal count as the storage days increases is also in line with the findings. Dauda. (2017) also reported that increase in the fungal growth could be due to favorable environmental condition and the acidity of the cheese samples at storage. Sample $C_{75}T_{25}CC_{0}$ has the highest fungal growth with values ranging from (1.0×10^{6} cfu/g), (1.7×10^{6} cfu/g) and (1.7×10^{6} cfu/g) for the three days of storage although there are no changes in growth for day 2 and day 3. The study showed that tigernut supplemented cheese is very prone to both bacterial and fungal spoilage and refrigeration temperature alone is not enough to maintain its quality and enhance its keeping period. Thus there is need for combined preservation methods to increase the shelf life, maintain the quality and enhance the safety and wholesomeness of the cheese product.

Table 8: Fungal count of cow-tigernut-coconut cheese during storage

	Storage period (Days)		
Samples	1	2	3
C_{100}	$1.67^{c}\pm1.53$	$73.33^{b}\pm20.82$	$93.33^{c}\pm5.77$
$C_{75}T_{0}CC_{25}$	$12.00^{c}\pm2.00$	$76.67^{b}\pm15.82$	$106.67^{c}\pm11.55$
$C_{75}T_{25}CC_{0}$	$100.00^{a}\pm20.00$	$166.67^{a}\pm30.55$	$173.33^{a}\pm23.09$
$C_{50}T_{25}CC_{25}$	$10.00^{c}\pm2.00$	$16.00^{c}\pm5.29$	$20.00^{d}\pm2.00$
$C_{25}T_{50}CC_{25}$	$5.00^{c}\pm1.00$	$53.33^{b}\pm6.11$	184.00 ± 14.42
$C_{25}T_{25}CC_{50}$	$43.33^{b}\pm5.77$	$80.00^{b}\pm20.00$	133.33 ± 11.54

Values are mean ±S.D. means with same superscript across a columns are not significantly ($p<0.05$) different

Sample keys: C_{100}: (100% cow milk) $C_{75}T_{0}CC_{25}$: (75% cow milk + 25% coconut milk) $C_{75}T_{25}CC_{0}$: (75% cow milk + 25% tigernut milk) $C_{50}T_{25}CC_{25}$: (50% cow milk + 25% tigernut milk + 25% coconut milk) $C_{25}T_{50}CC_{25}$: (25% cow milk + 50% tigernut milk + 25% coconut milk) $C_{25}T_{25}CC_{50}$: (25% cow milk + 25% tigernut milk + 50% coconut milk).

Table 9: Fungal count of cow-tigernut-coconut cheese during storage

	Storage period (Days)		
Samples	1	2	3
C_{100}	1.7×10^4	7.3×10^5	9.3×10^5
$C_{75}T_0CC_{25}$	1.2×10^5	7.7×10^5	1.1×10^6
$C_{75}T_{25}CC_0$	1.0×10^6	1.7×10^6	1.7×10^6
$C_{50}T_{25}CC_{25}$	1.0×10^5	1.6×10^5	2.0×10^5
$C_{25}T_{50}CC_{25}$	5.0×10^4	5.3×10^5	8.4×10^5
$C_{25}T_{25}CC_{50}$	4.3×10^5	8.0×10^5	1.3×10^6

Sample keys: C_{100}: (100% cow milk) $C_{75}T_0CC_{25}$: (75% cow milk + 25% coconut milk) $C_{75}T_{25}CC_0$: (75% cow milk + 25% tigernut milk) $C_{50}T_{25}CC_{25}$: (50% cow milk + 25% tigernut milk + 25% coconut milk) $C_{25}T_{50}CC_{25}$: (25% cow milk + 50% tigernut milk + 25% coconut milk) $C_{25}T_{25}CC_{50}$: (25% cow milk + 25% tigernut milk + 50% coconut milk).

4.4 Sensory Analysis of Cheese produced from blends of Cow-Tigernut-Coconut Milk

The mean sensory scores for cheese samples produced with varying proportions of added tigernut milk and coconut milk are shown in Table 10. There was no significant difference in the taste of all the six samples of cheese provided thus the taste was very palatable and acceptable by the consumers. There was significant ($p<0.05$) difference in colour, texture, aroma and overall acceptability where the colour of sample $C_{75}T_{25}CC_0$ is not significantly different to C_{100} having similar value of 7.00 respectively.

The sample with highest percentage of tigernut milk in the cow-tigernut-coconut cheese blend was rated the lowest in colour with a value of 5.80. This is similar to the findings of Igor *et al.* (2006) and Hussein. (2016) who reported that in term of appearance, 100 % cow milk 'warankashi' was rated the best 6.43 followed by 75:25% cow-tigernut milk '*warankashi*' 5.57 and also that the cow- tigernut milk 'warankashi' with higher percentage of tigernut milk was rated the least 5.00. This was reported to be due to discolouration of the *'warankashi'* as the substitution of tigernut milk increases. He further explained that the addition of tigernut milk in cheese analogue resulted into a slight brownish discoloration leading to reduced acceptability. Sample $C_{75}T_0CC_{25}$with a value of 6.80 was in contrast with the findings of Balogun *et al.* (2016) who reported no significance difference in the colour between 100% cow milk and 75-25% cow-coconut cheese blend.

The texture of the control sample C_{100} with a value of 6.65 was the most acceptable and was significantly ($p<0.05$) different to the other samples. This was in contrast with the findings of Hussein. (2016) who reported that sample with 100% cow milk has the lowest texture 4.56 while the result supports the findings of Balogun *et al.* (2016) who reported that 100%cow milk has the best texture.

In terms of aroma the sample $C_{75}T_{25}C_{0}$ of cow-tigernut-coconut cheese has the highest score of 6.45 followed by sample $C_{50}T_{25}C_{25}$ with 6.00 although not significantly different from the control sample C_{100}. These findingswere in contrast with the findings of Hussein. (2016) who reported that the aroma of 25:75 % cow-tigernut- milk 'warankashi' were rated lowest. However, Ekanem. (2017) reported sample containing 75:30% cow-coconut milk cheese blend to have the highest value for aroma (6.85) which may be due to the aroma and flavor of the essential oils in the coconut milk.

The sample $C_{75}T_{25}C_{0}$ and $C_{75}T_{0}C_{25}$ has the highest overall acceptability which was followed by the control sample C_{100} and sample $C_{50}T_{25}C_{25}$. This denotes that samples with coconut and tigernut milk supplements are generally accepted by the consumer and can be used in the production of cheese commercially.

Table 10: Sensory properties of cow-tigernut-coconut cheese

Samples	Appearance	Texture	Taste	Aroma	Overall acceptability
C_{100}	$7.00^{a}\pm1.41$	$6.65^{a}\pm1.09$	$5.15^{a}\pm1.90$	$5.40^{ab}\pm1.85$	$6.10^{ab}\pm1.86$
$C_{75}T_{0}$ CC_{25}	$6.80^{ab}\pm1.28$	$6.50^{ab}\pm1.70$	$5.35^{a}\pm2.13$	$5.20^{ab}\pm1.96$	$6.30^{a}\pm1.75$
$C_{75}T_{25}CC_{0}$	$7.00^{a}\pm1.56$	$6.50^{ab}\pm1.61$	$6.05^{a}\pm1.67$	$6.45^{a}\pm1.99$	$7.00^{a}\pm1.45$
$C_{50}T_{25}CC_{25}$	$6.25^{ab}\pm1.48$	$6.45^{ab}\pm1.57$	$5.85^{a}\pm1.95$	$6.00^{ab}\pm1.89$	$6.10^{ab}\pm1.77$
$C_{25}T_{50}CC_{25}$	$5.80^{b}\pm1.61$	$6.00^{ab}\pm1.86$	$5.35^{a}\pm2.16$	$5.50^{ab}\pm1.85$	$5.75^{ab}\pm1.83$
$C_{25}T_{25}CC_{50}$	$5.95^{b}\pm1.64$	$5.30^{b}\pm2.72$	$4.80^{a}\pm2.19$	$5.05^{b}\pm2.16$	$5.40^{ab}\pm1.85$

Values are mean ±S.D. means with same superscript across a columns are not significantly ($p<0.05$) different

Sample keys: C_{100}: (100% cow milk) $C_{75}T_{0}CC_{25}$: (75% cow milk + 25% coconut milk) $C_{75}T_{25}CC_{0}$: (75% cow milk + 25% tigernut milk) $C_{50}T_{25}CC_{25}$: (50% cow milk + 25% tigernut milk + 25% coconut milk) $C_{25}T_{50}CC_{25}$: (25% cow milk + 50% tigernut milk + 25% coconut milk) $C_{25}T_{25}CC_{50}$: (25% cow milk + 25% tigernut milk + 50% coconut milk).

CHAPTER FIVE

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

The result from this study shows increase in the yield, protein, moisture content, fat and ash content with increasing proportion of the coconut and tigernut milk at different levels in the blend which shows high possibility of using a milk analogue from a vegetable source such as coconut milk and tigernut milk in cheese production. The increase in yield of the supplemented cheese makes it economical and cost effective. The results of the microbiological evaluation showed that samples with tigernut milk and coconut milk supplements have high bacteria and fungal count thus very prone to spoilage. The result of consumer oriented test from this study shows that 75:25:0% and 75:0:25% cow-tigernut -coconut milk 'warankashi' were very accepted by the consumer with no significant difference in the other samples. Therefore tigernut milk and coconut milk could be used both individually and together to supplement cow milk up to 50 % without much negative effect on chemical properties, impairing the nutrients and acceptability of the final product. However 25 % proved to be the best blend based on consumer acceptability thus, it is recommended.

5.2 Recommendations

Cheese making in developing countries most especially in Nigeria is largely dictated by traditional means and due to the shortage of milk, milk and milk products such as cheese may be imported into the country which can adversely affect the socio-economic well-being of people especially low income earners and rural dwellers. Thus, milk from plant sources should be encouraged in the production of cheese as this would not only reduce the overdependence on cow milk but also reduce cost and improve the health of the consumer as research has proven that vegetable sourced milk such as tigernut milk, coconut milk, tigernut milk and bambara milk used extensively in supplementation of cow milk has health benefits such as prevention of cardio-vascular diseases, cancer, dental caries, and helps in building up the immune system and body defense. Further research can be carried out on mineral composition of the cow- tigernut - coconut cheese blend as well as ways to further improves the keeping quality and keeping period of the blended cheese without adverse effects on the cheese quality.

The major findings in this research therefore include the following:

Milk from plant source can be effectively used in the production of high quality cheese without any negative effects on the nutrient and chemical composition of the cheese.

Cow milk is essential in the production of cheese due to the casein present that ensures coagulation and also to ensure effective consumer acceptability and improved quality of the cheese.

The Inconsistency in findings in supplementation of cow milk with vegetable sourced milk for cheese production was discovered to be due to the factors such as differences in coagulant, differences in the quality of cow milk used, differences in the species and quality of the vegetable sourced milk and variation in production process.

Cheese produced from cow milk supplemented with plant milk has more nutrient, better quality and higher yield thus very economical and can be used in alleviating protein and other nutrient deficiency in developing countries.

Cheese produced from blends of cow-coconut and tigernut cheese is prone to spoilage thus there is need for effective preservation methods, and good manufacturing and hygiene practices.

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