# QUALITY ATTRIBUTES OF A SALTED PORK PRODUCT, UNAM INUNG, IN CALABAR, NIGERIA

BY

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# CERTIFICATION

I certify that this work was carried out by Idongesit Philip AKPAN in the Department of Animal Science, University of Ibadan, under my supervision.

.....

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# DEDICATION

To my dearest father, Professor Philip Abraham Akpan, for showing me the path to academics!

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#### 'It wasn't easy but it was worth it'- CeCe Winans

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### ABSTRACT

*Unam inung* (Ui), a popular salted pork product produced and consumed locally in Calabar metropolis has dwindled in popularity due to the non-availability of standard recipe leading to high variation in its quality. A standard recipe for Ui could boost its production and also make it a popular commercial meat product. However, a standardised salt inclusion level and technique for Ui production is yet to be adequately documented. Hence, quality attributes of Ui prepared using varying table salt concentrations, pork cuts and processing methods were assessed.

Samples of Commercial Ui (CUi) from three processors (P1, P2, P3) were assessed for their proximate composition and Total Bacteria Count (TBC, log cfu/g) using standard procedures. Freshly harvested pork bellies were sectioned into five slabs 129.54 x 216.2 x 26.9mm dimension each weighing 500g. Each slab was dry-rubbed with 0, 5, 10, 15 and 20 % Table Salt (TS) w/w in a completely randomised design in triplicate. The slabs were stacked, sundried and reshuffled at two-day intervals for eight days. Weight Loss (WL, g), TBC, Lipid Oxidation (LO, mg/kg), and the proximate compositions were determined following standard procedures. Pork from belly, ham, shoulder, and loin of similar dimensions were sectioned into slabs and dry rubbed with 15% TS. The pork cuts were kept raw, smoked or sun-dried in a 3x4 factorial arrangement in a completely randomised design. Proximate composition and TBC were determined. Data were subjected to descriptive statistics, regression and ANOVA at  $\alpha_{0.05}$ .

The CUi had similar crude protein content while, the ash (%) in P2 (6.5±1.0) and P3  $(6.8\pm1.0)$  were significantly higher than in P1 (4.0±1.0). The ether extracts of 21.6±1.0% (P1) and 20.0±1.0% (P3) were similar but higher than 17.3±1.3% (P2). The TBC of 3.36 ±0.02 (P2) was higher than 3.17±0.20 (P3) and 3.07±0.11(P1). The WL reduced progressively with increased addition of TS from 32.5±6.0 (5%), 24.5±3.3 (10%), 21.3±1.0 (15%) to 17.1±0.6 (20%). Significantly lower TBC of 3.2±0.2 was in Ui rubbed with 15% TS than  $8.1\pm0.1$  (0%),  $5.5\pm0.1$  (5%),  $3.5\pm0.2$  (10%) and  $3.5\pm0.1$ (20%). The relationship between TS inclusion levels in Ui and TBC ( $R^2=0.99$ ) was significant. The LO of 0.616±0.01 (10%) and 0.634±0.012 (15%) were similar but higher than 0.174±0.012 (0%), 0.195±0.008 (5%) and 0.484±0.013 (20%). Ash (%) content of  $25.4\pm1.9$  and  $27.2\pm0.1$  in 15% and 20% TS slabs were higher than  $18.7\pm2.5$ and 9.2±1.1 in 10% and 5% TS slabs, respectively. Ether extracts of 25.3±3.1 (5%) and  $23.8\pm4.3$  (20%) were significantly higher than  $17.8\pm2.7$  (10%) while  $21.25\pm1.0$ (15%) was similar to 5, 10 and 20% treatments. The CP of 29.08±2.85 (5%) was significantly higher than 19.88±1.73 (10%), 23.32±2.50 (15%), and 23.46±4.06 (20%). Effects of interaction of processing methods and meat cuts on proximate composition of Ui were significant. The TBC of  $3.2 \pm 0.1$  in raw meat cut was significantly higher than  $3.06\pm0.01$  (sundried) and  $3.09\pm0.05$  (smoked) meat samples.

Dry-rubbing pork with 15% w/w table salt lowered total microbial load while smoking and sun-drying enhanced the shelf life.

Keywords: Salted pork, Meat cuts, Total bacteria count

Word count: 490

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# **CHAPTER ONE**

#### INTRODUCTION

### **1.1 Background of the study**

Pig production has been advocated as a medium for alleviating the low animal protein intake of Nigerians (Ajala and Osuhor, 2004; Adesehinwa *et al.*, 2007) which is below 65gm per caput daily recommendation stipulated by World Health Organization (WHO). This advocacy is due to the numerous attributes of the animal such as fast growth due to its short generation interval, high fecundity rate, ability to convert non-conventional feedstuff to meat, yields more meat compared to other animals and its ability to subsist in a new environment. etc. Osaro (1995) reported that the cost of pig production is low due to their fast growth rate.

Pig production is popular in Nigeria both in the Northern and Southern parts except among groups with religious edicts (Ajala *et al.*, 2007). In 2011, the pig population in Nigeria was reported to be 7.1 million in comparison to 145 million poultry, 72.5 million goat, 41.3 million sheep and 19.5 million cattle (National Agricultural Sample Survey, 2011). In 2020, the population of pigs was reported to be 7.99 million thousand heads of pigs in Nigeria rising from 865,000 thousand heads in 1971 to 7.99 million thousand in 2020 (World Data Atlas, 2020).

According to Ajala *et al.* (2007) insufficient dietary animal protein intake is a problem in Nigeria and as the population increases the weight of the problem will intensify. He further stated that the animal protein intake per day is less than the 65gm which is the minimum recommended level. About 8.4gm of 53.8gm of protein consumed by Nigerians comes from animal sources which means that animal products only contribute about 16% to protein consumption of Nigeria (Olayide *et al.*, 1978). Abiola *et al.* (2015) remarked that the consumption of products from pig is increasing such that the nation's production capacity cannot meet the demand hence supporting with importation.

In southern Nigeria, pigs are kept in commercial quantities and their meats are highly relished (Amaefule *et al.*, 2009). About 4.5% of meat consumption in Nigeria is obtained from pig (pork) (FAO, 2002; Istifanus *et al.*, 2010) and it is also regarded as the most widely consumed meat globally.

Pork is consumed as freshly cooked meat or processed into different products (Beynon, 1990). Processed meat refers to meat whose original form has been altered either by grinding, addition of additives or subjected to heat application to confer considerable shelf stability (Aduku and Olukosi, 2000). There are several processed products derived from pork such as sausages, cured and/or smoked meat such as bacon and ham. Bacon and ham are popular salted pork products but there are several local traditional salted products like *Boucane* from the reunion, *Lacòn gallègo* from the region of Galicia in Spain and *Unam inung in Calabar*.

*Unam inung* is a salted meat product made from pork and sold in Calabar metropolis of Cross River State of Nigeria. It was traditionally prepared by heavily dry- salting the pork belly, sun-drying for eight days until thoroughly dried and packed in clay pots. When required for the market, the product is retrieved, washed and boiled. It is usually consumed with a bland accompaniment such as boiled cassava chips, yam and plantain etc. This meat product was very popular in the seventies and eighties after the ban on a similar product from fish called "ekpomo". A section of the markets was devoted to the sale of this meat product. Presently this pork product is found in the hand of few aged traders who are also the processors.

There is variation in the quality of the product among processors. *Unam inung* which is similar to Salt pork was traditionally made from the belly of the pig but in recent times, the preparation of this meat product is not restricted to a particular meat cut as all portions of the pork carcass are utilised in its production. The use of traditional crock wares such as clay pots have been replaced to suit the present-day situation. The processing of this meat product is fast being lost in antiquity as processors are aging and the youngsters are more interested in white collar jobs thereby causing the popularity of this meat product to dwindle. Hence, the dearth of information on the processing of this product. Standardised production recipe is required to curb variation

in quality of the product. Sustaining this almost extinct meat product could create a variety in processed meat product available in the country and could go a long way to boosting the economy of the locality and the country at large. It would also increase the protein intake of the populace as Nigeria is still far beyond the stipulated protein intake recommended by World Health Organisation (WHO) which is 35g per caput per day while the daily protein intake of Nigerians is 3.5g per caput.

Until now, this meat product has not been extensively studied and it is still prepared by unscientific procedures. The product is characterised by a strong flavour due to lipolysis and free fatty acid oxidation during salting and sun drying. It is also affected by weather conditions of the locality as Calabar is situated in the Nigerian rain forest of the vegetation belt (F.G.N, 2002). It has a long period of rainfall compared to sunny weather which makes the production of *Unam inung* to be seasonal and hence a need for alternative processing method.

Few researches in this sphere have been documented. Solomon (1994) studied the outcome of salt levels and smoking on physicochemical parameters and consumer evaluation of *Unam inung*. Omelagu (2013) also assessed the efficiency of packaging materials in prolonging the shelf-stability of *Unam inung*.

# **1.2** Statement of the problem

The production technique of *Unam inung* is still traditional and lacks technological advancement to keep up with modern life style. Currently, this product is almost extinct from the market therefore, its production technique needs to be upgraded and documented

### 1.3 Justification

*Unam inung* which is the only traditional meat product from pork in Nigeria could create a variety in meat products as traditional meat products are predominantly made from beef such as *kilishi, kundi, danbunama, suya etc.* Utilization of pig in the production of *Unam inung* could create employment opportunity for the teeming population thereby boosting the economy of the nation. It can also ameliorate the protein insufficiency in the diet of Nigerians if *the Unam inung* venture is resuscitated.

By-products from pork processing industry could become a means of income generation. Proper documentation on the processing of *Unam inung* is necessary as information on it is scanty.

# 1.4 Objectives

# **General Objective**

The general objective of the study was to assess the quality attributes of *Unam inung* for consumption.

# **Specific objectives**

- 1. To assess the consumption pattern, nutritive value and qualitative attributes of commercially available of *Unam inung*
- 2. To determine the influence of different concentrations of salt on quality attributes of *Unam inung*
- 3. To evaluate the quality attributes of smoked and unsmoked *Unam inung* prepared from different meat cuts of pork carcasses

### **CHAPTER TWO**

#### **REVIEW OF LITERATURE**

### 2.1 Pork and its nutritional value

Pork refers to meat obtained from pigs. It is widely consumed globally which justifies its 38% contribution to meat production worldwide (McGlone, 2013). It is very common in western world especially in Asian clime where it is highly priced in their cookery. The usefulness of Pork cannot be overemphasized as it is also widespread. Its size makes it saleable as a whole or sectioned into wholesale cuts. All parts of the pork carcass are useful as its usually presented as fresh, cured and/or smoked. The intestines and fat from pork are used as ingredient and casing respectively in sausage production. The fat is also used for improvement of lean meat as barding and larding. Other parts such as the head, feet, skin are used for gelatine production while feet and hocks can be vended as sweet pickle. Despite all its usefulness, Pork suffers cultural discrimination and taboos.

Pork possesses excellent nutrients as it is rich in protein with all the nine essential amino acids required for development and maintenance of the body. The nine essential amino acid are histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine. It has protein content of 19-20% (Torres *et al.*, 2013). This adjudges lean pork as a healthy diet for healthy life style, weight control in diabetics.

Fat content of pork fall between 10 to 16 % in the absence of trimmings and other factors (Cormier, 1996). It comprises of saturated fatty acids, unsaturated fatty acids and essential fatty acids which is useful against heart diseases. The prominent fatty acids in pork are the oleic, palmitic and stearic acids. Myristic, palmitoleic, lauric and unsaturated fatty acids are less evident. Certain factors such as age, meat cut and diet affects the composition of fat. About 10% of oleic and linoleic acids are found in

streaky bacon and dorsal back than in intestine and fats in the abdomen. Linoleic acid increases with age.

In recent times, Pork fat and calories have been reduced as reported in many researches. A 77% decrease in the quantity of fat and 53% decrease in calories of cooked pork loin was observed between 1963-1983 according to USDA. Genetical and nutritional development have converted the old pork to light pork. The cholesterol, saturated fat and calories in pork is now comparable to beef and skinless chicken. This agrees with the 300 mg/day intake of cholesterol, < 9-10% of saturated fat and calories daily intake recommended by American Heart Association (Torres et al., 2013).

It is also full of vitamins especially the B vitamins and minerals including thiamine (vitamin B1), vitamin B12, phosphorus, niacin, riboflavin, vitamin B6, zinc, potasium, iron and magnesium. Pork has low sodium compared to beef and chicken. Its low sodium content makes it relevant in the diet of people with hypertension.

# 2.2 Pork carcass and cuts

Generally, carcasses are commonly cut up into primal and sub-primal cuts. These major cuts are cut up into retail cuts. Pork carcass soon after dressing is usually cut into halves. The carcass is laid on the slab, sawn through the middle of backbone into equal sides with the side to be cut facing upwards (FAO, 1991).

The Primal cuts in pork carcass comprise of Pork leg, Pork loin, Pork belly, and Pork shoulder while sub-primal cuts are further cut out from each primal cut. The Pork leg can be cut into Pork leg - butt portion, Pork leg - shank portion, Pork hock, Pork foot. The Pork loin can be cut into Pork loin rib end, Pork loin centre, Pork sirloin. The Pork belly remains whole. There is no further cut from belly. The Pork shoulder is cut up into Pork shoulder blade, Pork shoulder picnic, Pork jowl, Pork fore foot and Pork fore hock.

Figure 2. shows the Primal and Sub- primal cuts (BC cook Articulation Committee, 2015), the alphabets A-D represent the pork leg, E-G represents Pork loin, H-represents Belly and I-M represents the Pork shoulder.

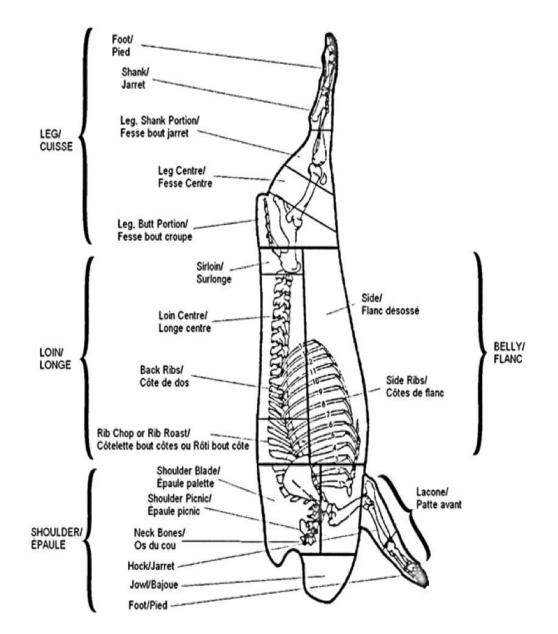


Figure 2.1: Pork carcass showing primal, sub-primal, and retail cuts

Source: BC Cook Articulation Committee. (2015)

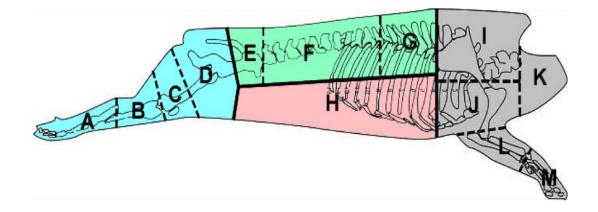


Figure 2.2: Pork primal and sub-primal cuts

(A) Pork foot (B) Pork hock (C) Pork leg shank portion (D) Pork leg butt portion (E)Pork sirloin (F) Pork loin centre (G) Pork loin rib end (H) Pork belly (I) Pork shoulderblade (J) Pork shoulder picnic (K) Pork jowl (L) Pork hock (M) Pork footSource: BC Cook Articulation Committee. (2015)

# 2.3 Pork products

Fresh pork can be cooked or preserved by curing for a period of time. Meat products derived from curing include bacon and ham. Pork is used in a variety of ways as fresh meat. Certain cuts and portion are popular in different region.

Meat products produced from pork fall under food from animal sources which provides essential proteins to human diets.

# 2.4 Processed pork

Pork is the most frequently used ingredient in sausage making. For instance, many oldstyle European sausages such as chorizo, fuet, Cumberland sausage and salami as well as numerous brands of breakfast sausages and the American hot dogs are also made from pork. In France, Charcuterie is a culinary practice of making sausages and other products from pork.

When raw pork is salt cured and sometimes smoked depending on the meat cut used, two products are derived, Ham and Bacon. Shoulders are made into Picnic shoulders and legs are made into hams when cured with salt and/or smoked. The side of the pork carcass particularly the loin produces round bacon and the belly produces streaky. With industrialization in the modern world, ham and bacon intake gets better and is prevalent.

Cured meat products are also known in non-western world. In China and Asia as a whole, their cooking style involves salted pork otherwise known as red roasted pork as preserved meat in their cuisine.

Any cut of meat from the belly, side or back that has been subjected to curing or smoking is known as Bacon. Bacon is used and consumed differently in different places. In central Europe, it is cooking ingredient in form of cubes (Lardons) appreciated for its flavour or a basis for fat. Besides bacons usage in cooking, the Italians, present it as thin slices of raw bacon (pancetta) in hors d'oeuvres.

Bacon is similarly used for barding roasts and other lean meat. It takes about 10 hours to smoke with different firewood. It could be consumed grilled, baked, or fried. Cuts of Bacon are known by different names for instance, in Australia, New Zealand and United Kingdom, an unsliced bacon from the side is called a 'Flitch' or slab bacon while 'Rasher' refers to a single slice. A Slice can also be referred to as Strip in northern part of America. Generally, slices of bacon are also called Collops. When a bacon has the skin on, as was traditionally, it is referred to as 'bacon rind' but there are rindless varieties too.

A variety of bacon cuts are available in Ireland and United Kingdom and are identified as "streaky bacon" and "streaky rashers". Back bacon is eaten as breakfast in Britain as well as Ireland. It is the meat obtained from the pigs back. In the United States, it is usually called "Canadian-style Bacon" or "Canadian Bacon".

Bacon according to USDA is the "cured belly of swine carcass". Other attributes of the bacon must be clearly marked e.g. "smoked pork loin bacon". Any Bacon labelled as "USDA Certified bacon" means that it had been treated of *Trichinella*.

# 2.5 Pork Processing

Pork processing starts from the slaughtering of the animal to the fabrication of the carcass into various products for subsistence and commercial use. Pork processing is usually carried out in two phases consisting of the Pig slaughter line and the carcass finishing. Some steps are involved in each of the phases.

# 2.5.1 Pig Slaughter Line

#### 1.Pre-slaughter handling

Pigs meant to be slaughtered are properly handled to obtain good quality pork. The way the animal is handled is a determinant of good quality (Miller, 2005). Preslaughter handling is paramount in the pork industry. The pigs are properly led to slaughter through stables and runways to minimize stress. If the animal to be slaughtered is stressed, it can lead to effects that are not desirable such as Pale Soft Exudative (PSE) and Dark Firm Dry (DFD) on the meat. Feed should not be given to the animal for about 12-24 hours to facilitate total bleeding and easy evisceration.

### 2. Stunning

The pigs are rendered unconscious before slaughtering. This procedure is in line with ethical and humane animal processing system. At this point either the chemical or electrical method of stunning can be applied on the animal. The meat derived from an animal that is stunned before slaughter is of superior quality because the animal is not stressed.

# 3. Bleeding and hollow knife systems

This system involves blades used for slaughtering and collecting blood from the slaughtered animal. It has either a horizonal or vertical bleeding system with appropriate non-automated workstation to innovative hollow knife for blood collection.

### 4. Scalding/ Dehairing/ Gambrelling

The removal of the hair takes place at this stage. The animal is drenched with boiling water or steam to aid the loosening of the hair follicle to release the hair. It can also be roasted over fire known as singeing. In the scalding method, the slaughtered animal is soaked in a scalding reservoir at a temperature range of 57°C to 63°C for 120 seconds to loosen the fur. If the temperature of the water is too low, the hair follicle will not be loosened and if it is too hot, the skin will be over heated and it will be difficult to remove the hair. Adequate temperature of the scald could be determined by rubbing the thumb on the skin of the animal to see if the fur comes off with ease (F.A.O., 1991). Thermometers and timers could also be used. Contamination should be minimized by changing the scalding water regularly. At slaughtering, the pig should be handled neatly and bleeding should be completed before soaking in water.

Dehairing is carried out with a bell scraper or knife. This manual method can be used to scrape off all the fur If the scald is effective. The carcass of the pig could also be skinned to remove the hair and dirt however, this is done when the skin is needed in the leather industry. The carcass is singed to remove the remaining hairs, shrink and set the skin.it also lessens the surface microorganisms. The black deposits from singeing are scrapped off and the carcass is perfectly cleaned before the commencement of evisceration. The carcass is hoisted up by the gambrel tendon to the rail of the dressing line conveyor.

#### 2.5.2 Carcass Finishing

This stage involves evisceration, chilling and cooling, cutting and dispatch. The internal organs of the pig are removed manually or automatically. It is then sent to chilling room or a chill tunnel in the case of a modern processing plant. this averts weight losses through reduction of temperature (Miller, 2006). The carcass can now be cut up to several cuts and moved to loading locations for dispatch to consumers.

#### 2.6 Pork consumption in Nigeria

In the last decade, pork consumption per capita was highest at 1.58kg in 2014, then dwindled to 1.47 kg in 2018 (FAO, 2005). Despite the decrease, Nigeria still ranked the highest in pork consumption per capita in 2018 comparing with other African countries like in Benin (0.690 kg), Cameroon (1.32 kg, Chad (0.040 kg) and Niger (0.070 kg) in 2018. The yearly consumption of meat per person in Nigeria is expected to rise three time its figure from 2012 to 2030 due to human population growth and per-capita food demand towards animal protein (FAO, 2018).

The fastest growing livestock subsector in the world including Nigeria is the pig and poultry industry (Nwachukwu and Udegbunam, 2020). Chauvin *et al.* (2012) pointed out that pig farming is an attractive venture for farmers in Nigeria and other underdeveloped nations as a result of the rising increment on the request for pork and its products. The consumption of pork is becoming popular and fast replacing other meat like beef, chevon, chicken and mutton with chicken being in close competition to pork (Pluhar, 2010).

## 2.7 Meat quality

The fulfilment derived by a consumer from eating meat is referred to as Meat quality (Jul and Zeuthen, 1981). It can be expressed in terms of hygienic quality, nutritional quality, organoleptic and serviceability. Organoleptic quality refers to quality as perceived by consumers in terms of colour, texture, juiciness and flavour which includes aromas. The nutritional quality which looks at how healthy a product is makes used of food constituents such as fats, carbohydrates and proteins. A typical meat is said to be nutritious if it displays a good amino acid, and poly unsaturated fatty acids profile. While hygienic quality considers the consumption of a product without harm.

This associated with contaminants such as microbial densities and chemical deposits in the product.

Meat quality concept as reported by Ingr (1989) and Jassim *et al.* (2011), puts together the attributes of meat while Ihenkoronye and Ngoddy (1985), relates it to some physical attributes such as the quantity of fat in between the muscle and the life span of the animal. These meat attributes are influenced by many internal (genotype and sex) and external factors such as production systems, nutrition and meat handling (ageing, frozen storage and cooking). Listrat *et al.* (2016) also added that several *in vivo* and after death factors have great impact on meat and fish quality.

Tenderness is the primary factor affecting meat quality. The lean surface texture is necessary in consumers ' assessment of meat quality (FAO, 1991). As important as the chemical constituents of meat is in the assessment of its quality, these constituents differ with the fattening rate of the animal, the specie and breed. (Gill, 1983). The peculiar muscular composition and structure makes up the distinctive properties of meat (Macrae *et al.*, 1997).

Relating to this, Brunso *et al.* (2005) stated that the quality of meat has triggered much interest and relevance lately. Dainty (1996) stated that meat quality evaluation can be carried out through microbiological, chemical and sensory analyses.

# 2.8 Meat preservation techniques

Meat after being slaughtered must be processed to prevent spoilage. Serval techniques come into play for the elongation of meat's storage stability. These techniques have been classified into different groups: preservation by moisture control, preservation by temperature control, preservation by direct microbial inhibition.

#### 2.8.1 Preservation by Moisture Control

This class of preservation method involves dehydration/drying, freeze drying or lyophilization, salting/curing and smoking.

#### 2.8.1.1 Dehydration or Drying:

When moisture or water is removed from meat, there is an increased concentration of water-soluble nutrients thereby becoming unavailable to microorganisms. The quantity of water present for microbial activity utilization is termed "water activity'(aW). Therefore, dehydration or drying decreases the water activity and inhibit the growth of microorganisms that causes spoilage. Ikeme (1990) observed that without water, most of the micro-organism and enzymes which cause food spoilage cannot operate. Examples of dehydration are sun drying, freeze drying and smoke drying etc.

The main physical factors involved in meat drying are rate of heat exchange, the variation in humidity amid the surrounding air and the meat surface, and rate at which water moves from the inner to the outer layers of the meat (Alonge, 1984). To hinder the proliferation of bacteria found in meat, there must be reduction of moisture and aW to 30% and 0.90 respectively (Alonge, 1984). The preservative effects of dehydration occur by lowering the aW which retards proliferation of microorganisms and stabilizes the dried products in the absence of cold storage.

#### **2.8.1.1.1** Sun drying:

Sun drying is an ancient technique of preservation which is still very important globally particularly in the tropics. (FAO, 1993). Meat drying occurs on exposure to sun rays, natural temperatures, proper circulation of air, humidity. The meat pieces are cut into uniform pieces and dried regularly. This allows the whole batch of meat to be dried with ease. Optimal meat drying is obtained by drying under low humidity and warm with minimal temperature change between day and night. Nevertheless, when good hygiene conditions and technological rules are adhered to, good results of meat dried under unfavorable conditions can be achieved. The concentration and extent to which the meat is dried hinges on temperature, air circulation and humidity. The meat will dry faster at high temperature, thorough air circulation and less humidity. Reduction of moisture in the meat occurs in a continuous cycle when moisture on the meat exterior layer evaporates into the atmosphere and moisture from the innermost part moves towards the exterior surface (F.A.O,1991). On the first day of drying, there is high moisture loss which decreases subsequently. About 60-70% weight losses are obvious after three four days of drying, the muscle and connective tissue shrink

causing alteration in the shape of meat in the process of drying. There is size and shape distortion on the meat as well as the texture becoming firmer and hard.

#### 2.8.1.1.2 Freeze-drying

This form of drying yields product with superior nutritive quality especially in restructured meat. The meat is kept in a vacuum with application of low heat, the moisture sublimes from the frozen meat into vapour. Drying in this method occurs in three stages: (i) freezing (solidification), (ii) primary drying (ice sublimation), and (iii) secondary drying (desorption of unfrozen water) (Ahn *et al.*, 2017)

The liquid suspension cools and creates pure water ice crystals at the freezing stage, increasing the concentration of bound water.

The material to be freeze dried is heated and subjected to a pressure lower than the triple point (at 0oC, pressure: 610 Pa) during the second stage, which is main drying (Rahman and Rerera, 2007). Permeable plugs are left on the frozen product due to ice sublimation. Lastly during the secondary drying stage, the bound water which is the remaining unfrozen water in the material is eliminated by a rise in temperature of chambers compartment (Abdelwahed *et al.*, 2006). Rehydration of freeze-dried meat is very poor and very tough even after cooking in liquid, (Aduku and Olukosi 1991). The advantages are that such product is light in weight, retains original shape and size and can easily be rehydrated.

### 2.8.1.1.3 Smoking

Smoking is among the earliest known methods of meat preservation (Pöhlmann *et al.*, 2012). This involves the use of a specifically smoking chamber where wood fuel is burned to a temperature of 300°C to generate smoke (Pal and Devrani, 2018). It permits smoke produced from firewood to be deposited on the exterior of the meat while the heat enters inside the products. This happens in partial burning of wood (Goulas and Kontominas, 2005) whereby higher molecular organic compounds are thermally broken down into compounds of lower molecular weight (pyrolysis). During decomposition of wood, reactions such as condensation, polymerization and oxidation also occur producing many other compounds from smoke.

Smoking extends the shelf life of foods in three ways: by generating heat, chemical, and also removing moisture from the surface of the material. Eliminating microorganisms from meat by heat during smoking is dependent on temperature and time. Wood smoke contains some synthetic compounds that act as antimicrobials which aids in preservation but these compounds alone are inadequate in preservation. Its preservative properties are achievable when it combines its antimicrobial and antioxidant properties (Kim *et al.*, 2014).

Smoking has other beneficial properties apart from preservation. It provides drying effect, required sensory properties, improved colour of cured meat, addition of antioxidants to the meat fat, antimicrobial and antifungal functions (Lingbeck *et al.*, 2014). The aldehydes and phenols precipitate as resins from the smokes and conveys luster on the meat.

Smoke contains some constituents such as phenols, aldehydes, organic acids, carbonyls, hydrocarbons, and alcohols. These constituents play different functions such as antioxidants in delaying rancidity of fat (Dineen *et al.*, 1999), aldehydes, phenols and carbonyls enhance colour and aroma or flavour of meat products. Smoke also contain Polycyclic aromatic hydrocarbons which has been associated with health risk such as cancer.

# 2.8.1.1.3.1 Smoking Techniques

Cold smoking, hot smoking, liquid smoking, and electrostatic smoking are the most common methods of smoking (Goncalves and Prentice-Hernandez, 1999).

### 1. Cold smoking

Cold smoking is usually performed at optimal temperature of 15-18°C which does not go beyond 30°C (Sigurgisladottir *et al.*, 2000) and generally takes a long time of about 10 hours to process though it leaves the inside of the meat uncooked. Meat at this temperature is expose to microorganisms as such cold smoking is usually combined with salt curing and/or fermentation. Some cold smoked meats are cooked to 160°F before consumption while some Salmon and mackerel that are cold smoked are eaten without re-cooking even though they still remain raw when eaten (Nummer and Andress, 2002). Although there is a description for commercial cold smoking published by the US FDA, Food scientists do not recommend cold smoking method due to the risk consumers are exposed to by consuming such product.

# 2. Hot smoking

Hot smoking is the method of smoking practiced in the tropics. It is usually carried out at temperatures ranging 55- 80°C and 120 - 150 °C which gives the product a quick flavour and colour (Alonge, 1984). It takes shorter time of about five hours to cook meat of fish to doness. Products from hot smoking such are hams are hard dried and keeps for a period of time.

# 3. Liquid smoking

Liquid smoke is derived from hard wood by condensation and fractional distillation of wood smoke with the removal of polycyclic hydrocarbons through filtration. It was developed to eliminate the process involved in smoking. When liquid smoke is applied externally to the meat as an aqueous spray prior to smoking, it conveys smoke-like flavours to it that is highly appreciated by consumers (Pal and Devrani, 2018). Liquid smoke is being manufactured and sold in technologically advanced countries. There is no trace of cancer implicating compounds like the 3 and 4 rings Polycyclic aromatic hydrocarbons that are usually found in wood fuel smoking (Hedrick *et al.*, 1994). In the preparation of liquid smoke, only the beneficial hydrocarbons are used while the harmful ones are discarded (Ikeme, 1990).

Liquid smoke is advantageous over the traditional smoking method in that it is easy to apply, it reduces the time of production of the smoked product, can penetrate the product through different routes such as curing brines by injection or dipping or by electrostatic spray (Knowles *et al.*, 1975). The drawback with this method is that the keeping quality of the smoked meat is poor together with the aroma as it gives off tarry or acidic flavour (Potthast *et al.*, 1984).

# 4. Electrostatic smoking method

The need for a speedy deposition of smoke birthed electrostatic smoking technique nonetheless this technique is not gaining popularly industrially as it should (Hedrick *et al.*, 1994). Smokes are passed through tunnels over electrical live wire at 20 - 60 kilowatts. The electrically charged smoke particles are passed into the smoking chamber over contrary charged surfaces of meat products on which they precipitate

quickly. One feature of this method is that it is mainly the particle or solid phase of the smoke that deposits on the meat products. Such products must be subsequently dried by infra-red radiation.

### 2.8.1.1.4 Salting

This involves the use of Salt as the lone ingredient in meat processing. It can also be referred to as corning or salt curing. When nitrate or nitrites are added to salt, it is regarded curing. Salt, otherwise referred to as table salt or sodium chloride, is chemically denoted as NaCl possessing sodium and chloride ion in a ratio of 1:1. It is a two-component chemical that forms an ionic bond when the positively charged electrons of the soft metal Sodium combine with the negatively charged electrons of chlorine. The ionic bond of Sodium chloride is known to be very strong such that dissolution in water does not influence the magnetic force of the electrons.

In the olden days, salt was regarded as an important article of trade which ranked along with gold (Pedrosa, 2017). The significance of salt in history was enormous such that words like salary, soldier and sausage had its origin from it (Kennedy, 2007). In human history, the commercial value of salt could be equated to oil. During the world wars, countries stockpiled salt for the preservation of food meant for their armies. It was also used as a form of tax. The "salt tax" of 1930 in India, caused a ban to be placed on the sale or production of salt in opposition to the British monopoly that was prominent. Presently, Sodium Chloride (NaCl) is mainly used in food for Preservation, technological and sensory purposes.

# 2.9 Salting methods

Salt is basically added to meat in two ways:

- 1. Dry application
- 2. wet application (brining)

#### 2.9.1 Dry salting

In this method of salt application, salted is applied in the dried form on the meat. This is also called 'Corning'. It was initiated in the Anglo-Saxon cultures. Corns or pellets of salt was used in dry curing meat. A typical example is the Irish Corned beef usually made from the brisket, of which any meat cut can be used.

# 2.9.2 Brining

In this method of salting, a solution of salt is made sometimes other ingredients are added such as erythorbate, nitrites and or sugar. Traditionally, salt was added in the brine solution until an egg could float on it. Nowadays, hydrometer is used to measure the specific gravity of the salt solution or the ingredients are adopted as specified in the recipe. Meat can be soaked in the brine after being mixed or the brine could be injected into the meat. Brine injection is known to speed up the process of curing. Brine cured meat is less salty than dry cured meat.

#### 2.10 Salt functionality in food

The functionality of salt in food production are highlighted as follows:

#### 2.10.1 Sodium chloride as a food preservative

Salts preserves food mainly by lessening the water available to microbes thus preventing the proliferation of putrefying bacteria (Inguglia *et al.*, 2019) thereby developing a dry membrane-like exterior (Leroi and Joffraud, 2000; Rorvik, 2000). Pegg and Honikel (2015) observed the shelf life of cured bacon and ham was extended with a reduction on the microbial growth in the presence of salt. The growth of microorganisms is prevented through the process of water removal from the cells of microbes by the action of salt. The loss of water from the cells of microbes continues until it becomes unconducive for growth and survivability.

The genus and species of microorganism determines the concentration of salt outside the cell required to inhibit microbial growth by plasmolysis. The growth of *C*. *perfringens* and *C. botulinum* are greatly suppressed by salt, *Salmonella* can be inhibited at about 3% salt concentration while *Listeria monocytogenes* and *Staphylococcus sp.* which are salt loving can thrive in concentrations as high as 12% and 20% respectively (Shelef and Seiter, 2005). The growth of detrimental organisms seen in salt treated products are subdued when salt levels are low (USDA FSIS 1997a). Additionally, salt denatures enzymes through reduction of their activities, by hindering catalysis and alteration of their cofactors (Ravishankar and Juneja, 2014). Products preserved by salt include cured meat products, such as dry-cured ham, bacon and ham canned fish, namely, sardines, tuna, mackerel, anchovies etc. (Pegg and Honikel, 2015; Featherstone, 2015).

#### 2.10.2 Texture enhancer

Salt changes the make-up of proteins and its relationship with water and fat, which influences the consistency of foods. In yeast bread production, salt influences the final texture. If salt is added in the right amount, the bulk in cheeses increase, meats become juicier, and breads can be dense. Salt influences WHC of meat and its derivatives and makes the it to be moist in texture.

#### 2.10.3 Flavour enhancer

Salt enhances savoury flavour although a salty flavour is frequently craved. It stabilizes sweetness and helps puts down other flavours, such as bitterness.

# 2.10.4 Source of Nutrient

Salt is regarded as a source of nutrient because of the presence of Sodium which is an essential nutrient required in small amounts in the body.

# 2.10.5 Binder

In processed meat, addition of salt reshuffles proteins which in turn makes it perform as a binder and emulsifier. This restructured protein prevents moisture and fat loss by holding the product together.

#### 2.11 Effect of salting on physicochemical properties of meat and fish

In fish processing, the chemical composition is essential as it affects the technological properties and the attributes of fish at storage. It is associated with muscle constituents such as moisture, ash content, fat and protein (Huss, 1988).

For microorganisms to proliferate in food, water must be accessible to them. When moisture content in food is restrained, it keeps for a longer period. The water needed by microorganisms is explained as water activity (aw) of the food or environment. Jay (2000) defined "Water activity as the ratio of water vapour pressure of the food substrate to the vapour pressure of pure water at the same temperature".

Clucas and Ward, (1996) reported that adding 'coarse salt' reduced the moisture content of fish by pulling out water from the fish and making it dried.

Methods of salting also affects the protein content as observed by Binici and Kaya, (2017). The authors compared Dry salting and Brine salting on Chub fish and observed a higher protein content in the latter. They attributed the higher protein content in dry salted chub fish to loss of moisture and concentration of other nutrients. The aforementioned authors established that high salt concentration causes loss of protein in dry salting method, but a contrary report from another study states that protein loss only occurs in brine salting method (Binici and Kaya, 2017). Several factors such as the fish type, the salt quantity, salting method and storage period may be responsible for the variance in the report.

Binici and Kaya (2017) observed an increment in ash content of both Dry salting method and brine salting of Chub stored for 120 days. The period of preservation and the amount of salt increases in salted fish. This was obvious in salt cured anchovy stored for 29 weeks

Salting simply decreases water activity (aw) which impedes spoilage microbial growth and also makes autolytic enzymes inactive (Ashie *et al.*, 1996, Horner, 1997)

# 2.12 Health risks of smoked meat

Polycyclic Aromatic Hydrocarbons (PAHs) and nitrosamines, both proven carcinogens, contaminate food during the smoking process. Theoretically, high risk of

gastrointestinal cancer is encountered by eating smoked foods. According to Fritz and Soos, (1980) there are indications that smoked foods may contain carcinogens. These authors also noted that frequent intake of smoked foods correlates significantly with high incidence of intestinal cancer as presented by epidemiological studies. Alonge (1988) stated that Polycyclic Aromatic Hydrocarbons are prevalent and may pose health hazards in Nigeria.

#### 2.13 Sodium Intake

Meat and various foods from it, are said to contribute about 15-25% of the overall consumption of sodium chloride in a day (WHO, 2003). The World Health Organization endorses about 5g of salt to be consumed daily and this amount is equal to 2g of sodium daily (WHO, 2003). Nevertheless, these nutritional recommendations are surpassed in many technologically advanced countries.

In Spain for instance, the Spanish Food Safety and Nutrition Agency (AESAN) began a strategy to lessen salt intake in 2008. The goal of the salt reduction plan was to decrease the current value of NaCl intake from 9.7 g to 8.0 g daily by 2014.

The difficulty in cutting down salt in dry-cured meat is based on the double role it plays in preserving and disinfecting the meat. This is possible because Sodium chloride lessens the amount of water available to microbes and stabilize microbial growth thereby prolonging storage quality of the product. (Hutton, 2002, Taomina, 2010). In addition, Salt affects proteins found within muscles by making it soluble and forming gel to develop and optimum texture (Desmond, 2006).

#### 2.14 Implication of moisture in meat

The quantity of water present food and other materials is usually referred to as "water content" or "moisture content" (Park, 1996). It is an important measurable element in meat making up about 75% of the weight of meat. It gives information on yield and quantity of solids in food as well as commercial worth, storage attribute and value of food products (Pomeranz and Meloan, 1994). Moisture is a vital component of food such that foods are classified according to their moisture. Food having moisture content above 70% are classed as Perishable, about 50-60% moisture is non-perishable while less than 15% are stable food (Ahmad *et al.*, 2018). High moisture content in

foods reduces the storage value of food because it aids the growth of microorganisms which causes spoilage. Moisture also affects colour, firmness and aroma of meat.

Moisture in meat tissue exist mainly as free water in muscle fibers and connective tissues. Throughout the application of heat, curing or other processing procedures and even storage, the remaining water though, small in the myofibril is known as bound water. The ability of the water to be withheld in the muscle fiber on application of pressure during processing is known as Water holding capacity. If the muscle fiber is disrupted, WHC of the muscle will be altered and it will improve the storage quality of meat. Processing and preservation techniques are responsible for the final moisture content of meat (Kamruzzaman, 2016).

#### 2.15 Microbiology of meat

The microbiology of muscle tissues hinges on the functional state of the animal, the extent of carcass pollution the process of slaughtering, temperature, storage and distribution (Nychas, 2008). Some microorganisms are found in the intestines of the animal along with the surroundings from where the animal thrived (Jay *et al.*, 2005). Psychrotropic bacteria and other microbes were isolated from skins, carcasses and working exteriors inside an abattoir (Gill, 2005).

Different categories of microorganisms thrive on meat depending on the type, the storage environment and the processing that the meat is subjected to (Gill and Molin,1991). Some pathogenic microorganisms in food (meat) are described below:

# 2.15.1 Bacillus

Bacillus spore forming, positive gram staining, rod shaped, bacteria that are commonly found in soil, air and water. This genus possesses strains that are detrimental to humans and cause food borne illness, and also beneficial stains that serve as probiotics for animals (Ray, 2004). *B. cereus* is a facultative toxin-producing anaerobe. It is often implicated in sickness originating from ingestion of food which manifest in stomach upset and other discomforts (Frazier and Westhoff, 2006).

#### 2.15.2 Shigella

This is a rod-shape, gram positive, non- spore forming and motile enterobacteria. There are transmitted mostly in overcrowded population with poor sanitation. Shigella outbreak can be caused by polluted food and water (Ray, 2004). The disease of shigella is called shigellosis otherwise known as dysentery; a disease accompanied by running watery stool without blood stain.

# 2.15.3 Yersinia

Yersinia species are gram negative, rod-shape bacteria usually found both in the environment and intestines of mammals and mollusc. It can be seen in snail, in view of its mode of locomotion and feelings. There are two major species of pathogenic Yersinia, viz; *Yersinia enterocolitica* and *Yersinia pestis* (Frazier and Westhoff, 2006). In pathogenicity, it resembles *Vibrio* although their colonies are smaller than *Vibrio* (Ray, 2004).

#### 2.15.4 Vibrio cholera.

*Vibrio cholera* is gram negative bacteria which is shaped like a comma. When first isolated, they look curvy in shape but can also straighten out as rods during culture. The bacterium has a flagellum at one cell pole as well as pili. *V. cholera* is a can survive in the absence or presence of oxygen but it relies on fermentation. It can also tolerate strong alkaline condition with high salt concentration. *Vibrio cholera* adheres to and multiplies on the cells lining the small intestine of snail. The bacteria produce a potent enterotoxin (Frazier and Westhoff,1986). The infection caused by *Vibrio cholera* is called rice-water; a disease accompanied by fast running watery stool that resemble rice-wash water (Ray, 2004)

#### 2.15.5 Salmonella

This microorganism has the shape of a rod, does not form spores, does not response to crystal violet staining. They belong to the enterobacteriaceae with cell diameters between approximately 0.7 and  $1.5\mu m$ . they utilize organic sources to obtain energy from redox reactions. and are facultative anaerobes. The detection of

salmonella is usually carried out on ferrous sulphide media because when grown on this media, the produce hydrogen sulphide (Ray, 2004).

Salmonella can subsist in non-living medium for a long period of time as they were seen in desiccated excreta of two and a half years old (Ray, 2004). It could be eliminated by Ultraviolaet radiation. Prevention against salmonella requires heating food to a temperature not less than  $75^{\circ}$  C in 10mins so the heat penetrates the interior of the food (Frazier and Westhoff, 1986). A salmonella infection causes vomiting, fever and headache.

#### 2.15.6 Escherichia coli ([E. coli)

This is a gram negative, facultative anaerobic, rod-shaped bacterium that is commonly found in the colon of higher mammals. It measures between  $1.1\mu m$  to  $6.0\mu m$ . Majority of the E. *coli* strains are not pathogenic but some serotypes are involved in severe food poisoning in the hosts, and sometimes cause products to be recalled due to food contamination. They do not move, response negatively to oxidase, citrate and vapour pressure, while their response to catalase and methyl red are positive (Frazier and Westhoff, 1986). Escherichia coli is regarded as an indicator organism because its presence in food depicts feacal contamination (Clarence *et al.*, 2009; Edema *et al.*, 2008; Okonko *et al.*, 2008). If ingested causes stomach cramps and diarrhea (Ray, 2004).

# 2.15.7 Staphylococcus

These bacteria respond positively to crystal violet staining and can thrive in the presence and absence of oxygen. They can be spherical, oval or round in shape, hence cocci. They are also pathogenic. Talaro *et al.* (1996) reported that staphylococcal food poisoning is the rampant in most countries. Some authors have reported the isolation of Enterotoxigenic Staphylococcus strains and E. coli strains from foods that have been isolated from food that caused infection (Adesiyun, 1995). Although *E. coli and S. aureus* are common in human and animal flora, their presence in food (suya) indicates extreme human processing (Adamolekun and Adamolekun,1992). The pathogenic member of staphylococcus is *S. aureus* which is association with boils, carbuncle and

endocarditis. *S. epidermis* and *S. Saprohyticus* catalase positive, oxidase negative, and they exist in high concrentration of salt (NaCl) (Frazier and Westhoff, 1986).

#### 2.16 Meat spoilage

Meat is a highly nutritious food but it is not shelf stable because it forms a medium for microbial growth (Jay *et al.*, 2005). Due to its chemical composition, deterioration sets in soon after slaughter till the period it is consumed.

Also, meat spoilage is linked with the type, composition and population of microbes as well as the nature of energy substrate in meat. Temperature, oxygen, enzymes, accessible moisture, light, and microorganisms are all interconnected elements that affect the preservation value of meat. These factors have a negative impact on the sensory qualities of meat, either individually or collectively. Fresh beef has a maximum storage stability of 24 hours at room temperature (Nychas et al., 2008). Spoilage is said to occur when food becomes obnoxious to the olfactory of the consumers due to metabolites released by the microorganisms contaminating it. (Paulsen and Smulders, 2003). On the whole, spoilage is dependent on the discretion of the consumer, which is determined by the financial prowess and perception of the person involved and the degree of variation (Nychas et al., 2008).

### 2.16.1 Types of meat spoilage

Meat spoilage is usually categorised on availability or non-availability of oxygen and well as the causal microorganism.

#### 2.16.1.1 Spoilage under aerobic conditions

Bacteria may cause the following under aerobic conditions:

# 1. Surface slime

This may be produced by species of *Pseudomonas, Cinenterobacter, Moraxella, Alcaligens, Bacillus, Streptococcus, Leuconostoc* and *Micrococcus* (Frazier and Westhoff, 2006). Temperature and availability of moisture influences microorganisms causing surface slime.

# 2. Off-odour and off taste

Undesirable odour and taste that arise in meat are caused by microbes growing on the exterior of the meat. Any defect that gives a sour odour maybe due to volatile acids e.g., formic, lactic, butyric, and propionic acids or even the growth of yeast (Frazier and Westhoff,2006). When oxygen is available, yeast may thrive on the periphery of the meat triggering slime production, breaking down fat, giving off offensive odour /taste and changing colour due to pigments in the yeast. Moulds growing in the presence of oxygen can produce black spot, white spots, stickiness, decomposition of fats and odour/taste.

# 3. Variation in meat pigment

Microorganisms produce oxidizing compounds during meat spoilage which causes variation in meat pigment from red to gray, brown and green. Hydrogen sulphide or peroxides produced by bacteria species such as of *lactobacillus* and *leuconostoc* are reported to cause the greening of sausage (Frazier and Westhoff, 2006).

#### 2.16.1.2 Spoilage under anaerobic conditions

Microorganisms growing both in the presence and absence of oxygen are capable of causing spoilage in the absence of oxygen (Frazier and Westhoff, 1986). Bacteria may cause the following under anaerobic conditions:

# 1. Souring

This refers taste and odour occurring due to spoilage in the absence of oxygen. This could be caused by formic, acetic, butyric, proponic and higher fatty acids or other organic acids such as lactic and succinic acid (Gadner,1981). These acids are formed by the action of microorganism on meat.

#### 2. **Putrefaction**

This is the breakdown of protein in the absence of oxygen thereby relasing offensive compounds like hydrogen sulphide, ammonia and amines. It is produced by species of bacteria that responds positively to cystal violet staining.

Temperature is similarly paramount in this type of spoilage. At temperature of 0°C, microbial attack is restricted to mould and yeast. These include many of the type that produce sliminess, discolouration and spots of growth on the surface and many that can cause souring.

# 2.17 Identification of meat spoilage

Meat spoilage is identified on the basis of negative changes caused by microorganisms, inherent meat enzymes and insects (Jay *et al.*, 2005). The microorganisms in meat in the cause of spoilage, use up many substrates and produce new compounds which can be determined in meat. Detection of meat spoilage is done through physical and chemical methods. Physically, changes in colour, formation of slime, whiskers stickiness are observations of meat spoilage

1) Amino acids are sources of many off flavour compounds. Off flavours are associated with meat spoilage at low temperature, The production of sulfide and mercaptin can be measured to ascertain meat spoilage (Paulsen and Smulders, 2003). Chemical determination for the presence of ammonia, indole, skatol and trimethyamine etc., is performed to determine microorganisms of spoilage in meats.

2) From the onset of spoilage in meat displays a simultaneous increment in Ph, bacterial counts and water holding capacity of meat proteins.

3) High peroxide value indicates chemical spoilage of meat and products.

# 2.18 Factors affecting the growth of microorganisms in meat

Food microbiology deals with the detection and control of disease causing and putrefying microorganisms. Due to the nutrient dense nature of meat, it becomes a medium for proliferation of microorganisms. Microbial growth therefore is affected by several factors which are categorized as Extrinsic and Intrinsic factors. These factors include environment where the meat is stored and processed as well as the chemicals that may be present in the meat or added (Willie *et al.*, 2011).

### 2.18.1 Intrinsic factors

The composition of meat is a vital intrinsic factor that influences microbial proliferation. When food is mainly made up of carbohydrate, mould and yeast predominate; this may lead to bacteria colonization. When food contains large quantity of proteins (meat and dairy products), bacterial growth can produce a variety of changes – spoilage bacteria will predominate and degrade the accumulated waste product of bacteria and the PH will gradually increase. Cooking produces a lower oxidation- reduction potential that brings about a reducing environment for microbial growth.

With the presence of accessible proteins and other growth factors, a perfect media for the development of aerobic and anaerobic organisms is prepared.

#### 2.18.2 Extrinsic factors

Food preservation is based on extrinsic factors because they are easily controlled than intrinsic factors. Temperature is a necessary extrinsic factor in determination of the degree of food spoilage. For instance, spoilage problem can occur with minimally processed, concentrated frozen citrus products (Willie *et al.*, 2011). Its preparation does not require heat treatment. Humidity is another essential factor in controlling food spoilage. In a high relative humidity, microbial growth occurs very fast regardless of low temperatures (especially if the refrigerator is poorly managed in a unfrozen state). Dried foods left in a wet environment traps moisture on the surface and this aids the proliferation of microbes.

There are other extrinsic factors influencing the growth of microorganisms in meat. Some methods are employed to bring about these factors. The purpose of each method is to eradicate the population of putrefying and pathogenic microorganisms while upholding the quality of meat. Such factors include;

1. Refrigeration at 4°C or temperature below 10°C

- 2. High temperature drying in oven or fire flame.
- 3. Pasteurization.
- 4. Salting-chemical preservatives.
- 5. Radiation and irradiation.
- 6. Packaging.

Some microorganisms associated with meat spoilage are Shigella, Salmonella, *Clostridium prefrengenes, Bacillus spp., E. coli, Vibrio-cholera, Yersinia, Staphylococcus aureus, proteus mirabilis* (produce proteolytic enzymes that degrade the meat into toxic compound e.g ammonia).

# 2.19 Microorganisms of meat spoilage

Meat deteriorates when microbial growth on meat, produces changes that can be seen, touched and mouth felt as metabolites are released. The visible growth may appear as slime due to formation of colonies, degradation of nutrients such as carbohydrates, proteins lipids etc, which affects texture and production of off-odor and off-flavour (Jackson *et al.*, 2001; Gram *et al.*, 2002).

*Pseudomonas spp., Enterobacteriaceae, Brochothrix thermosphacta,* and lactic acid bacteria (LAB) are frequently linked to meat deterioration. Their ability to cause spoilage hinges on storage conditions (Borch *et al.,* 1996).

LAB are highly significant meat putrefying bacteria having many species which include *Lactobacillus, Leuconostoc, Pediococcus and Streptococcus.* They are anatomically related to a group of fastidious and abundant gram-positive organisms.

Microorganisms contaminate meat through sources like slaughtering of sick animals, exposure to flies by processing near the sewage or dumpsite, washing with dirty water by the butcher, transportation on ramshackle automobile, use of unsterilized knife and other utensils in addition to contaminated spices. During slaughtering, the sterile animal tissue is exposed to contamination with members of the *Enterobacteriaceae* found in the gut of the animal such as *Salmonella species* and *Escherichia coli*, and Lactic acid found on humans and animals in the surrounding (Lawrie and Ledward 2006). Lawrie and Ledward (2006), and Alexander *et al.* (1998) reported that *Enterococci* and *Clostridia* were picked out from lymph node of animals used as meat.

Another group of bacteria commonly identified in meat spoilage are the gramnegative proteolytic bacteria which breaks down proteins and gives off bad odour (Hamman, 1997).

*Micrococcaceae* and *Staphylococcaceae* families are another common bacterial group found in meat aside the enteric organisms. The most common forms are coagulasenegative *Staphylococci* that can survive aerobically or anaerobially and are salt tolerant. *Staphylococcus* carnosus, *Staphylococcus xylosus*, and *Staphylococcus kocuria* are the most prevalent strains. These organisms, on the other hand, are completely harmless and pose no threat.

*Enterobacteriaceae* are used as post processing measures for contamination. This group of bacteria decreases when processed because of its response to low water activity and low pH (Fernandez-Lopez *et al.*, 2008).

A lot of food scientists have researched the detection of the enteric bacteria as a pointer to the hygienic conditions of food, environment and water. The incidence of Enterobacteriaceae in meat suggested the presence of microbiological and toxigenic bacteria, posing a public health risk (Mira, 1989). The meat handling work surface was found to be the source of Enterobacteriaceae on the meat. Stiles and Lai-King (1981) stated that the presence of Enterobacteriaceae in ground beef is an indicator of direct or indirect enteric contamination of meat. The prevalence of Enterobacteriaceae in meat products can be linked to fecal contamination of meat handlers' hands, tools, and handling surfaces at all stages of processing.

Staphylococci are halophilic microorganisms that can survive at high salt concentrations, they are the microorganisms most closely linked to product safety (Casaburi *et al.*, 2007). A species of staphylococci known as Staphylococcus aureus (*S. aureus*) causes staphylococcal food intoxication, a rapid symptom manifesting stomach flu. *S. aureus* is prevalent in environment as well as sensory organs of man. Staphylococcal food-borne disease (SFD) is a global disease emanating from the infection of food by *S. aureus* enterotoxins that was previously formed.

Muscle tissue softening, slime and offensive odour production is an attribute of bacterial spoilage (Ames *et al.*, 1991). Therefore, microbial and biochemical quality evaluations are essential to certify any processed product's food safety (Azam *et al.*, 2003).

Consumers are concerned about the microbial safety of seafood as the outbreaks of infection linked to eating fish are instigated by *Salmonella, Staphylococcus, Vibrio sp* and *Bacillus cereus* (Flemming *et al.*, 2000).

According to research, the growth of Pseudomonas spp. causes meat spoilage at low temperature. Other spoilage bacteria are capable of growing too but their growth will be insignificant compared to the entire population of microbes (García-López *et al.*, 1998).

Meat's maximum bacteria level is  $10^7$ - $10^9$  cfu/cm<sup>2</sup> during refrigerated storage, and meat products have a maximum bacteria level of  $10^7$ - $10^8$  cfu/g (Borch *et al.*, 1996).

Fungi can be found in a number of locations in the environment and they can utilize substrates from various nutrients and organic acids which explains its prevalent incidence. Nonetheless, high pH, water activity and temperature does not favour their existence. They are also relatively unaffected by reduced pH, water activity, and temperature.

In the early stage of storage of nuggets samples, coliforms were not present probably because of a log phase delay caused by a lower metabolic rate triggered by an abrupt change in the physical environment or exhaustive heating of the goods during processing. The absence of the Enterobacteriaceae and fungi could be attributable to the application of heat, sanitary practices maintained during processing, and antibacterial properties of preservatives utilized, according to Choi and Chin (2003).

The acceptable limits for total aerobic bacterial and fungal counts regarded as satisfactory for readymade foods are in the order of  $\leq 10^3$  and  $10^4$  to  $10^5$  (ICMSF, 1996).

Total bacterial count of perishable food is used to evaluate its quality and shelf-life. However, high count may be attributed to unsanitary methods of production or exposure to conditions favouring bacterial proliferation as asserted by Sharma *et al.* (1996). The total aerobic bacterial count is tended to indicate the level of microorganisms in products (FDA, 2001). Devatkal and Mendiratta (2001) found that microbial counts increased as storage time increased in restructured pork rolls.

According to previous research, the maximum acceptable Total Viable Count in meat consumed by man is 5 log10 CFU/g (Lawrie and Leward, 2006).

Plate counts with representative sample units fewer than 5 X  $10^5$  CFU/g are regarded good quality, between 5 X  $10^5$  and  $10^7$  slightly acceptable quality, and plate counts of more than  $10^7$  are considered undesirable (ICMSF, 1986-44 p).

Mould can proliferate over a wide range of temperature. Therefore, one can find mould particularly in all foods at almost any temperature under which food is held. Besides mould can assists in the decomposition and may produce poisonous substances namely mycotoxins which are detrimental to living beings (Frazier and Wasthoff, 1983). The number of moulds present in a product is indicative of the proper sanitation and

quality of the product. Moulds can assist in putrefactive processes and in other cases, they may impart a mouldy odour and taste of food stuffs.

# 2.20 Proximate composition of meat

The percentage composition of carbohydrate, protein, lipid and ash is usually referred to as proximate composition. It is altered by various factors including stage of age, nutrition, sex and season (Pushparajan *et al.*, 2012). The proximate composition of meat is useful in assessment of the nutritional quality since the chemical constituents are evident in biochemical contents (Nagabhushanam and mane, 1978). In fish processing, it influences yield, odor, flavor, texture, and fat oxidation stability. Many authors have reported that the proximate composition is influenced by management practices and body characteristics of the animal (*Ćirković et al.*, 2012; Ljubojević *et al.*, 2015).

## 2.21 pH of meat

In meat products, the pH is very significant as it influences physicochemical and organoleptic attributes of the meat product (Goli, *et al.*, 2007). pH in meat varies due to breakdown of muscle tissue soon after death and when substances are added to meat in technological processes (Gault, 1985).

A fish's freshness or decay is determined by its pH value. The pH of fresh fish is almost neutral, it declines to due to lactic acid as a result of death, afterwards, it rises due to spoilage. The disordering of the oxidation-reduction balance, as well as the effects of enzymes and bacteria are all contributing to this increase. (Varlık *et al.*, 1993).

Fresh fish has a pH of virtually neutral, which decreases owing to lactic acid after death and then rises as rotting sets in. This rise is due to a disruption of the oxidation-reduction equilibrium, as well as the effects of enzymes and microbes. (Varlk *et al.*, 1993).

Deterioration of a product is depicted by rise in pH. In normal circumstances, meat pH value decreases during postmortem as lactic acid is formed from glycogen. The low pH-value prolongs shelf life and flavour of meat (FAO, 2004). According to Heinz and Hautzinger (2007), the pH value for pork and its products is within 5.50- 6.20. The pH of meat products is necessary for their storage. Low pH values constitute unconducive conditions for microbial activity thereby enhancing the storage value of the product.

The level of salt and salting method affect pH as demonstrated in the production of pastirma. High salt level with dry curing method produced lower pH compared to low salt brine cured method (Aksu and Kaya 2002). A rise in pH was seen during the processing of pastrma, according to Kaban (2009). Proteolytic alterations between muscle and çemen paste, according to Uguz *et al.*, (2010), could cause insignificant increase in the end products. Increment in pH of Italian dry cured ham in comparison to raw ham were linked to the let out of low molecular weight nitrogen compounds in relation to the external and internal activities of protein breaking enzymes (Virgili *et al.*, 2007). Other researchers have also observed high pH due to salt level in dry cured hams (Arnau *et al.* 1997).

Kumar *et al.*, (2007) described the reduced pH of meatballs during storage to be linked to bacterial activity involving the catabolism of carbohydrate into organic acids, mostly lactic acid thereby reducing pH. During the salting stage in Kaddid processing, Azaier, *et al.* (2011) noticed significantly (p <0.05) declining pH values. He explained that the decrease in the pH of the meat proteins resulted from addition of salt in the salting phase.

The level of microbial spoilage may be detected by pH (Eyo, 2011). This happens when alkaline compounds like ammonia accrue from microbial activity thereby increasing pH (Erkan and Ozden, 2008). The measurement of pH is necessary for fish quality evaluation and it also serves as an index (Pacheco-Aguilar *et al.*, 2000). pH in the flesh and gills of fish plays an important role on its freshness with regards to bacterial growth. When the pH of fish flesh and gills are low, the growth of bacteria

will be slow and the other way round (Okeoyo *et al.*, 2009). Post mortem pH affects WHC and has also been known as a vital physical attribute of meat (Soeparno, 2005).

#### 2.22 Water holding capacity (WHC) of meat

The WHC in meats relates to the quantity of free water let out by the meat on application of external force. The value of WHC in meat production cannot be over emphasized because it impacts the commercial and eating qualities of meat (Oeckel *et al.*, 1999). The arrangement of the myofibrillar proteins makes it easy for water to diffuse in the meat, thus influencing the WHC of meat (Huff-Lonergan and Lonergan, 2005; Kristensen and Purslow, 2001, Melody *et al.*, 2004).

It influences the physical characteristics of meat implying that these physical characteristics are dependent on the WHC (John *et al.*, 2006) such as colour, texture, firmness, juiciness and tenderness of cooked meat. It also affects the shrinkage of meat as confirmed by Barbera and Tassone (2006). High WHC makes meat tender after cooking (Young *et al.*, 2004). The electric charges of myofibrillar protein groups are altered when NaCl is applied to meat products thereby changing the WHC of the meat. Chloride anions have a strong affinity for positively charged protein groups, whereas sodium cations have a poor affinity for negatively charged protein groups (Cobos and Diaz., 2014). Once the pH value rises higher than the isoelectric point, the negative charges of the chloride ions rises as it binds to proteins. This shifts the isoelectric points to a reduced pH, makes the proteins of the myofibrils to repel, thereby opening the structure to improve water binding in meat (Cobos and Diaz, 2014)

Ions also boost the water holding capacity of connective tissue proteins. On the contrary, as soon as the pH falls beneath the isoelectric point, the ions of chloride counteract the positive charges of the protein group thereby lowering the water holding capacity and the positive charges. As pH values increases, the muscle proteins swell up indicating an increase in WHC (Westphalen *et al.*, 2005). This is probably as a result of increase in the hydrogen bond amid the protein and water when pH is high (Westphalen *et al.*, 2005). On application of NaCl to meat products, the negative charges of protein rise as a result of the affinity of chloride ions and proteins. These negative charges on protein repels the myofilament causing the myofibrils to swell and increase binding capacity (Ruusunen and Puolanne, 2005)

Several factors including ionic strength, pH, osmotic pressure, development of rigor mortis and sarcomere length affects Water holding capacity whose action alters the components of the cell. (Offer and Knight 1988). He further stated that water in the muscle is present between the myofills while a higher proportion is bound within spaces amid the actin and myosin filaments in the cells. High water content in the muscles enhances economic value and meat quality by boosting the sensory characteristics of the meat.

Salt increases the WHC of meat derivatives by pulling out the proteins in the myofibrill which gels upon heating (Chantrapornchai and McClements, 2002)

# 2.23 Thiobarbituric Acid Reactive substances (TBARS)

The usage of TBARS has frequently become pointer towards the level of oxidation of lipids (Ojagh, *et al.*, 2010). It assesses the malonaldehydes produced during the oxidation process. The Oxidation of Lipids manifests in formation of a wide range of compounds. These compounds possess disagreeable sensory (Hernandez *et al.*, 2009).

Torres *et al.* (1988) proved that NaCl could promote lipid meat oxidation, with a stronger effect in Charque (Torres *et al.*, 1989). Increased thiobarbituric acid values were obvious due to high lipid oxidation and volatile metabolite formed in aerobic packaging. Tarladgis *et al.* (1960), Devatkal and Mendiratta (2001) both observed that during storage, the TBA values of several meat and meat products increased. Lorenzo and Purrinos, (2013) disclosed the occurrence of significant (P< 0.001) increases in TBARS of muscular portion throughout the process of salting and afterwards. Similar trends of high malonaldehyde contents were observed in Lacon and Ham respectively (Garrido *et al.*, 2009; Melgar *et al.*, 1990)

# 2.24 Histology

Structure of muscle tissues are studied in Histology. Tissue refers to a collection of cells which may be identical, similar or dissimilar together with the products outside their cells that performs the same function (Banks, 1993). Histologically, meat constituents include skeletal muscle fibers, connective tissues adipose tissue and others etc. the study of histology makes it easy to detect tissues directly in products derived

from meat. The methods of histological studies enable the assessment of processed meat quality (Sadeghinezhad *et al.*, 2016).

Histological study makes the direct detection of tissues in meat products possible (Georgier and Vitanov, 1995) and the determination of chemical alterations of mineral content and pH of meat products by examining the structural characteristics in muscle fibres. The cross section of fiber area and extracellular spaces in meat samples at different pH was studied by Sharedeh *et al.* (2015)

Muscle fibers are one of several muscle structure elements linked to the quality of meat (Aguilera, 2005). Thickness, quantity, and type of the muscle fibre, all have an impact on meat texture. Heating produces major changes in muscle structure. The connective tissue protein and collagen have an impact on the machine-driven characteristics of meat. The amount of collagen in the meat, as well as its ability to be soluble when heated, affects the texture. The variations that occur in meat when heat is applied is dependent on temperature and time while cooking methods affects how soft or tender the meat will be (Vasanthi, *et al.*, 2007).

# **CHAPTER THREE**

# MATERIALS AND METHODS

#### **3.1** STUDY ONE (Phase 1)

# Assessment of Consumption Pattern and Nutritive value of Unam inung

#### 3.1.1 Location and study area

This study took place at Watt market in Calabar, Cross River State of Nigeria where the product is usually sold. Calabar municipality lies on latitude 4.97°N and longitude 88.35°E. It is 6.14m above sea level with rainfall ranges of 42.0 to 1401.0 mm/month. It is a metropolitan city.

# 3.1.2 Sampling procedure

Consumers were painstakingly sought for in Calabar municipality and at the point of sale of the product while processors were sought for in all the markets in Calabar municipality but were found only in Watt market.

# **3.1.3** Instrument for data collection:

A structured questionnaire was designed and administered to 150 consumers. However, a total of 146 were retrieved from the Consumers.

The questionnaire assessed Consumer's Awareness, Preference and Hygiene consideration, Sensory characteristics, Purchasing and consumption pattern of *Unam inung*. The background characteristics of the consumers was also assessed.

# 3.1.4 Statistical analysis

Data obtained were subjected to simple descriptive statistics and Chi square using SPSS version 20 (IBM SPSS, 2011).

# Phase 2: Qualitative evaluation of commercially available *Unam inung*

# **3.1.2.1** Experimental site:

The experiment was carried out at the Department of Animal Science laboratory, University of Uyo.

# 3.1.2.2 Sample Collection and Experimental Design

The ready-to-eat *Unam inung* was purchased from three different processors at Watt market and immediately transported to Animal Science laboratory at University of Uyo for the various analyses. The design of the experiment used was the completely randomized design (CRD) with the products from each processor representing a treatment and was replicated three times.

# 3.1.2.3 Parameters measured

The following Analyses were carried out:

# **3.1.2.3.1 Proximate analysis**

Proximate analysis of the pork product was carried out following the procedures of AOAC (1999). Parameters analysed were Moisture content, Crude Protein, Ash, and ether extract.

# 3.1.2.3.2 Microbiological analysis

Microbial analysis was carried out immediately after obtaining *Unam inung* from the processors to determine the Total Bacterial Count (TBC) and Total Fungi counts (TFC) and also to identify the microorganisms present in the samples.

# **3.1.2.3.3** Estimation of microbial densities

Microbial loads were assessed by the pour plate method reported by Harigan and McCane (1990). Using standard microbial techniques, logarithmic dilution of the samples was conducted down the dilution gradient to the third factor (10<sup>-3</sup>) in sterile water using a 1ml pipette. A sample of 1 gram was homogenized in 9ml of sterile water to form the aliquot. 1ml of the aliquot (sample supernatant) was pipetted and mixed in 9ml of sterile water in another test-tube and shaken vigorously thereafter 1ml of the desired dilution was plated out in duplicate set on nutrient agar amended with cycloheximide (Nystatin) at 100uml to prevent fungal growth (Essien *et al.*, 2006). The plates were nurtured at 37°C for 24-48 hours. Discrete microbial populations after incubation were enumerated, studied and recorded as colony forming unit per gram (cfu/g) of meat. The desired diluent was also plated out in duplicate sets on sabouraud dextrose agar sets to which streptomycin at 30uml to prevent bacterial growth. The plates were nurtured at 28-30°C for 5-7 days. Microbial Colonies after incubation were counted and enumerated.

#### **3.1.2.3.4** Characterization and Identification of bacterial isolates

Bacterial isolates were characterized and identified based on their cultural and morphological as well as microscopic examination and biochemical characteristics following the methods described by Holt *et al.*, (1994) and Cowan, (1985). The biochemical tests conducted to assist in the identification include- Gram stain, catalase test, urease production coagulase test, oxidase test, indole production test, citrate test, methyl red test, vogesproskauer, starch hydrolysis, nitrate reduction, hydrogen sulphide production and sugars fermentation.

# 3.1.2.3.4.1 Identification of Bacteria

The bacteria isolated were subjected to the under listed biochemical analyses:

#### 3.1.2.3.4.2 Gram Stain Reaction

On a dry clean glass slide devoid of grease was smeared the test organism with a germ-free wire loop and a drop of distilled water. This slide was air dried and heat fixed (Brooks *et al.*, 1991).

Crystal violet stain was poured on the smear for sixty (60) seconds then shielded with Gram's iodine (Lugol iodine) and left to react for half a minute. Plenty tap water was used to wash the slide and the smear was counterstained with a solution of safranin for 60 seconds. It was cleaned with water and air dried. Using oil immersion objective (x100), the smear was viewed microscopically. Gram positive bacteria were detected by black, purple or blue colouration. The red coloured bacteria depicted the gram-negative bacteria

# **3.1.2.3.4.3** Motility test

Using the "hanging drop method", the test organisms were injected into peptone water and nurtured for 24 hours. The test organism was smeared on a cover slip which was covered with a clean slide. The cover slip was quickly inverted creating a culture drop in a hanging position. Microscopic observation was carried out at x100 lens. Organisms that actively moved from one place to another were observed as Motile while those that remained in one spot vibrating were seen as non-motile.

### 3.1.2.3.4.4 Coagulase test

A smear of the isolate was set down on a dirt-free slide and mixed gently with a loopful of blood serum using a sterile loop. Within ten seconds of the mixture, if the organisms clumped together, it indicated a positive outcome but the absence of clumping indicated a negative outcome. Staphylococus aureus is usually identified in this test.

# 3.1.2.3.4.5 Starch hydrolysis test

Identification of organisms that produce amylase, an enzyme which hydrolysis starch is usually carried out by this test. The test organism was injected with a prepared starch agar and incubated for 48 hours. Thereafter, 2-3 drops of gram's iodine were added to the incubated plates, a blue-black colour change of the medium indicated the presence of starch thus a negative result while a brown colour change specified a positive result (Priest, 1977).

# 3.1.2.3.4.6 Methyl red test

In a test tube containing a 2-day old broth culture of the isolates, about two -three drops of methyl red indicator were added. Colour change occurred with red showing that acid can be produced in glucose solution by the organism while yellow meant negative result.

#### 3.1.2.3.4.7 Urease test

A sterile Urea base medium (Urea Agar) was used to inoculate the test organism and it was incubated at 37°C for one day. A purple colouration showed presence of urease while no change in colour showed absence of urea.

# 3.1.2.3.4.8 Citrate test

A 100ml of water and 1 g of Simmons citrate Agar were mixed in a conical flask and the mixture was decontaminated in an autoclave. The test organism was inoculated at 37°C for 24 hours. A positive citrate test was specified by a green to blue colour change while green colour specified negative citrate test.

# 3.1.2.3.4.9 Catalase test

The making of the Catalase enzyme by the isolates was determined by this test. Using a sterile wooden loop, a small amount of the organism from a 3-day old colony was applied on a sterile slide together with 3 drops of 3% hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>)

solution freshly prepared and a cover slip placed over it. An immediate production of active bubbles specified a positive result while absence of bubble depicted negative outcome (Cheesebrough, 1984).

#### **3.1.2.3.4.10** Salt tolerant test

Six (6g) of NaCl in 100ml of Nutrient agar (6%) was sterilized by autoclaving for 15min at 121°C and allowed to set. The medium was injected with the test organism and nurtured for 48 hours. The organism that grows on the medium at that salt concentration indicated positive result while those that did not grow indicated negative result.

# 3.1.2.3.4.11 Indole test

The ability of the organisms to convert tryptophan to indole is determined in this test. Indole is produced by reductive removal of the amine group from tryptophan. Tryptophanase speeds up the deamination reaction whereby the amine group of the tryptophan is removed and indole, pyruvic acid and ammonia are produced. The isolated microbial population of the test organism was made into emulsion in tryptophan broth and nurtured at 37°C for 24 hours. In addition to the broth culture was 0.5ml of Kovac's reagent and shaken gently. The production of indole was ascertained when the reaction turned red at the point of contact with the reactants while production of yellow colour indicates absence of indole.

# **3.1.2.3.4.12** Voges Proskauer test (VP test)

Identification of glucose fermenting bacteria that produces acetylmethylcarbinol.

First, 5% of Alpha Naphtol was added followed by 40% KOH solution in absolute ethanol. Barret's reagent was carefully added to a test tube already containing the test broth solution. Within a duration of 15 minutes of adding the Barret's reagent, a change in colour occurred. A pink-red colour signified positive result while lack of pink-red colour signified negative result

#### 3.1.2.3.4.13 Oxidase test

This test detects if the organism possesses the cytochrome oxidase enzyme. Strips of whatman's No.1 filter paper were soaked with in 1% tetramethyl-p-phenylene diamine dihydrochloride solution. With a wooden applicator, a smear of the test organism was applied on the moistened filter paper. The filter paper changes colour from light pink to dark purple indicating positive oxidase tests, while absence of colouration indicated a negative result.

#### 3.1.2.3.4.14 Sugar fermentation test

One gram of each of the sugar used, 90ml of distilled water, 1ml of peptone water were added and mixed in a conical flask. Then 10ml of phenol red solution was also dropped into the mixture in the conical flask. About 10ml of the mixture was dispensed in test tubes plugged to Durham's tube and cotton wool. The test tubes were purified at 121°C for 15 minutes and left to cool. The test organisms were then injected and nurtured at 37°C for 24 hours. Colour variation from red to yellow designated acid production, a change from red to yellow with air bubbles indicates acid and gas production while absence of change indicated no acid production.

### 3.1.2.3.5 Characterisation and identification of fungi isolates

Fungal isolates were characterized and identified as describe by Fawole. and Oso (2004). The identification was based on cultural, morphological and vegetative as well as reproductive features.

#### **3.1.2.5** Statistical analysis

Data gathered fro the experiment was subjected to Analysis of Variance using SAS (1999) package.

# STUDY TWO: Quality attributes of *Unam inung* as influenced by different salt levels

# 3.2.1 Sample preparation

The unskinned pork bellies were trimmed of connective tissues and sectioned into anterior and posterior sections of 500g per slab. The pork slabs were dry rubbed with salt (NaCl) at 0, 5, 10, 15, 20 % of the green slab weight. Each slab was randomly allotted to one of the five treatments, with two slabs from each anatomical position being assigned to the same treatment to eliminate positional bias.

The dry rubbed slabs were stacked in a woven basket to aid aeration in alternate layers for a period of eight days. The slabs were over-turned at two days interval and the weights and other parameters were measured.

# 3.2.2 Parameters measured

# 3.2.2.1 Proximate composition

Proximate analysis of the salted pork was determined in accordance with the procedures of AOAC (1999). Parameters analysed were Moisture content, Crude Protein, Ash, and ether extract.

# 3.2.2.2 рН

The pH measurement was performed with a digital pH instrument (HANNA 4824) using the methods of AOAC (1999). Five grams of the representative salted pork subjected to different concentration of salt, were uniformly weighed, combined with 50ml of distilled water and blended for 1min until a smooth homogenous solution was obtained. The microcomputer pH meter was dipped in the mixture and allowed to equilibrate for one minute before the reading was taken. Three readings were taken and averaged as the pH of each sample.

# **3.2.2.3** Water Holding Capacity (WHC)

This was determined using the Filter Paper Press Method (FPPM) with adaptation from Suzuki *et al* (1991). One gram of the salted pork was weighed and positioned in between two 12.5 cm diameter Whatman No1 filter paper (Schleicher and schuell. cat N01001 125) and pressed in between two 13cm x14cm plexi-glass with the aid of a vice for 60 seconds.

The sample after pressing was removed, wrapped in a foil and dried in the oven at 100°C for one day with the aim of determining its moisture content. On a tracing paper, the area of the pressed sample and the area made wet by water as seen on the filter paper were traced to determine the amount of water let out from the meat sample. These two areas on the tracing paper were transferred to a graph sheet where the figures were derived.

Thus WHC:

 $WHC = 100 - [(Ar-Am) \times 9.47]$ 

(Wm X Wo)

Where

Ar = Area of water released from meat cm<sup>3</sup>

 $Am = Area of meat sample cm^3$ 

Wm = Weight of meat sample in mg

Wo = Moisture content of meat

9.47 = constant factor

# **3.2.2.4 Drip loss**

Drip loss was determined by the process described by Ibraheem and Abdullahi (2000) and was calculated using the formula

Drip loss =  $\underline{Purge weight}$  X 100 Sample weight

# 3.2.2.5 Microbial analysis

The pork samples were collected for microbial analysis in a sterile Aluminium foil. The samples were immediately analyzed on getting into the laboratory for the Total Bacterial count (TBC), Total Fungal Count (TFC), Total Pathogenic Bacterial Count (TPBC), *E. coli, Samonella/Shigella* count

# **3.2.2.6** Thiobarbituric Acid Reactive substances (TBARS)

The Thiobarbituric acid method as described by by Pikul *et al.*, (1989), at intervals of 0, 7, 14, and 21 days of storage was carried out. A sample of 5g was added into a test tube having a capacity of 50ml, blended with 15ml of sterile water using a laboratory mortar and pestle. One ml of the normalized sample was transferred into a disposable test tube 13 x 100mm butylated hydroxyanisole ( $50\mu$ l, 10%) where 2ml Thiobarbituric acid (TBA) was added. The mixture was vortexed and then incubated in a boiling water bath for 15min to develop colour. The sample was then cooled in cold water for 10min, vortexed again and centrifuged for 15 minutes. The absorbance of the resulting supernatant solution was determined at 535nm against a blank containing 1ml of deionized distilled water and 2ml of TBARS was expressed as milligrams of malondialdehyde per kilogram of sample.

### 3.2.3 Statistical analysis

Data obtained was subjected to Analysis of Variance in a Completely Randomized Design where significance differences occurred the means were separated using Duncan Multiple Range Test (DMRT) on SPSS version 20 (IBM SPSS, 2011).

# **3.3 STUDY THREE: Evaluation of the quality of smoked and unsmoked** *Unam inung*

#### **3.3.1** Collection of samples

Meat cuts from belly, ham, shoulder and loin were purchased from a butcher at Nung Udoe, Ibesikpo, Akwa Ibom State. Table salt, firewood and polyethene were purchased from Akpan Andem market in Uyo, Akwa Ibom State. The different cuts were subjected to the optimal salt level from Study 2 which was 15% salt (NaCl) per 500g of pork.

#### **3.3.2** Sample preparation

The meat cuts were trimmed of connective tissues and sectioned into 500g per slab. The sectioned pork was dry rubbed with salt (NaCl) at 15 % of the green slab weight. At the end of the salting phase, each pork slab was sectioned into three, one third was set aside for smoking, another third was subjected to sun-drying and the last third was treated as raw representing the control.

# 3.3.3 Smoking

One salted half of the pork was subjected to smoking in a in a drum kiln. The fire was made using wood fuel from Hardwood until smoke was produced. The temperature of the kiln was sustained between 50 - 60°C. The salted pork was smoked to doneness with smoked flavour, golden brown colour, and a lustrous surface. The smoking was carried out for 9 hours.

#### **3.3.4** Experimental Design

This experiment was on a 3x4 factorial arrangement in a Completely Randomized Design. The processing methods and Meat cuts served as the independent factors. Raw, Smoked and Sun-dried were levels of the Processing methods and Belly, Ham, Loin, and Shoulder were levels of the Meat cut.

#### 3.3.5 Parameters measured

The following were measured on the Raw meat, the product after sun drying and the product after smoking

#### 3.3.5.1 Proximate analysis

Proximate analysis of the raw, smoked and sundried meat cuts were determined in accordance with the procedures of AOAC (1999). Parameters analysed were Moisture content, Crude Protein, Ash, and ether extract.

# 3.3.5.2 Microstructure of the meat samples

The Microstructure of the meat was assessed on the raw, sundried and smoked *Unam inung* through Histological Technique (Heamatoxylin and Eosin Method (Drury, 1990) and Histomorphological/Stereological analysis method (Gundersen *et al.*, 1999).

# **3.3.5.3** Procedures for histological Technique (Heamatoxylin and Eosin Method (Drury, 1990)

The Muscle tissue was carefully dissected out, trimmed of all fat and blotted dry to remove any blood. They were weighed and volume determined by water displacement and then fixed in 10% formal saline (fixation). The fixed tissues were dehydrated in graded series of ethanol (70% for 7 hours and 90% for 12 hours) and subsequently passed through absolute alcohol for one hour and cleared in xylene.

As soon as it cleared, the tissues were infiltrated in molten paraffin wax (Paraplast) in the oven at 58°C. Three changes of molten paraffin wax (impregnation) at one-hour interval were made, the mounted sections to be cut by the rotary microtome were oriented perpendicularly to the long axis of the tissues. Thereafter, the tissues were embedded in wax and blocked out

The sections were designated "vertical sections". Serial sections of  $5\mu$ m thick were obtained from a solid block of tissue (microtomy) fixed on clean albuminized slides to prevent sections from pulling off the slides and later stained with haematoxylin and eosin staining techniques, after which they were passed through ascending grade of

alcohol, cleared in xylene and mounted in DPX mountant and observed under a digital light microscope.

# 3.3.5.4 Histomorphological/Stereological analysis method (Gundersen *et al.*, 1999)

Muscle fibers were identified histologically and isolated under 100X magnification and the 400x magnification respectively. The measurement and counting procedure were performed on the muscle spindle and fibers of the cell nuclei of the entire Muscle. The number of the Muscle spindle was estimated using Image-Pro analysis software, incorporated into Tissuegnostic digital microscope with a motorized stage (Lang MS 316) (for the step lengths on the X, Y-axis), under 100X and 400X magnification. The thickness of the tissue measured and the movements in the Z-axis were controlled using a microcator (Heidenhain, Germany). For counting 100–200 myocites nuclei per spindle/fibres, according to the dissector principle, the area of the counting frame was 20,449 µm and the step length for the X and Y axis was 1250 µm. Image acquired was digitalized and segmented to expose region of interest (ROI). ROI such as fibre/spindle length, width, Area, Diameter, and volume, be measured using a whole slide based on the calibration of the microscope following segmentation, filtered, refined and condensed and the measurements were taken.

# **3.3.6** Microbial analysis

Microbial analysis of the raw, smoked and sundried meat cuts was carried out on the 8<sup>th</sup> day of storage to determine the Total Bacterial Count (TBC) and Total Fungi counts (TFC).

#### **3.3.7** Statistical Analysis

The GLM procedures of SPSS version 20 was used to for analysis of Variance of all data in this experiment. Significant interaction means were further analysed by simple main effects of both independent variables, and means were separated by Turkey's HSD test using SPSS version 20 (IBM SPSS, 2011).

# **CHAPTER FOUR**

#### RESULTS

# 4.1 STUDY 1 Phase 1: Assessment of Production, Consumption Pattern and Nutritive Value of *Unam inung*

#### 4.1.1 Background characteristics of *Unam inung* consumers

The Demographic and Socio-economic characteristics of Unam inung consumers as presented in Table 4.1 shows that consumers interviewed were from Edo state (3.42%), Akwa Ibom state (21.23%), Cross river state (73.97%) and Ebonyi state (0.69%). These consumers according to tribe were Efiks (64.44%), the Ibibios (21.23%), ikom (7.54%), Benin (3.42%) and the Igbos (1.37%). 60.27% of them were married while 39.73% were single. Out of which 15.75% were males and 84.25% were females. The age categories of the consumers were 4.79% for 11-20 years, 23.29% for 21-30 years, 46.57% for 31-40years, 24.66% for 41-50 years and 0.69% for 51 years and above. All (100%) of the consumers were Christians. Majority (48.63%) of the consumers were civil servants, 32.19% were into business, 0.69% was a trader and 15.75% were students. 36.85% of the consumers had tertiary education and 22.60% had secondary education and 20.55% had primary education. Most (41.08%) of the consumers had a monthly income of N30,000.00 and above, followed by 30.82% of consumers who earned  $\mathbb{N}15,000.00 - \mathbb{N}30,000.00, 19.80\%$  did not disclose their monthly income and 7.53% earned  $\ge 7,500.00 - \ge 10,000.00$ . The household composition of the consumers assessed by number of Children and number of wives showed that 39.73% had no children, while 27.40% had one child each, and 18.49% had two children and 14.38% had three children and above. In terms of number of wives, 21.74% had no wives and 78.26% had one wife. There was no consumer with two or three wives.

Characters	Frequencies	Percentage (%)
State of Origin		
Edo	5	3.42
Akwa Ibom	31	21.23
Anambra	1	0.69
Cross River	108	73.97
Ebonyi	1	0.69
Total	146	100.00
Tribe		
Benin	5	3.42
Ibibio	31	21.23
Efik	97	66.44
Ikom	11	7.54
Igbo	2	1.37
Total	146	100.00
Marital Status		
Married	88	60.27
Single	58	39.73
Total	146	100.00
Gender		
Male	23	15.75
Female	123	84.25
Total	146	100.00
Age		
11-20	7	4.79
21-30	34	23.29
31-40	68	46.57
41-50	36	24.66
≥51	1	0.69
Total	146	100.00
Religion		
Christianity	146	100.00
Occupation		
Civil servant	71	48.63
Business	47	32.19
Trader	1	0.69
Student	23	15.75
Other	4	2.74
Total	146	100.00
Level of Education		
Primary	30	20.55
Secondary	33	22.60
Tertiary	83	56.85
Total	146	100.00
Income level		

 Table 4.1. Background characteristics of Unam inung consumers

7,500- 10,000	11	7.53
15,000- 30,000	61	41.78
30,000 and above	45	30.82
Others	29	19.86
Total	146	100.00
No. of Children		
None	58	39.73
One	40	27.40
Two	27	18.49
Three and above	21	14.38
Total	146	100.00
No. of wives		
None	5	21.74
One	18	78.26
Two	0	0
Three and above	0	0
Total	23	15.75

# **4.1.2** Consumers response to awareness, preference and hygiene considerations of the *Unam inung*

Consumers response to awareness, preference and hygiene considerations of the *Unam inung* are presented in Table 4.2. All (100%) consumers interviewed had been consuming the product. There was no consumer who had not eaten *Unam inung* before. 91.10% of the consumers liked the product while 8.90% were indifferent i.e neither liked or disliked the product. None of the consumer disliked the product outrightly. All (100%) the consumers said they preferred pork to be used in making *Unam inung*. There was no consumer that preferred mutton or beef for the production of *Unam inung*. 93.84% of the consumers said the *Unam inung* was prepared under hygienic conditions, 0.68% said otherwise while 5.48% were indifferent i.e neither hygienic nor unhygienic.

Variable	Frequencies	Percentage (%)
Ever consumed the p	product	
Yes	146	100
No	0	0
Total	146	100
Likeness for the prod	luct	
Yes	133	91.10
No	0	0.0
Others	13	8.90
Total	146	100
Meat type for Unam	inung	
Pork	146	100
Mutton	0	0
Beef	0	0
Total	146	100
Hygienically prepare	d	
Yes	137	93.84
No	1	0.68
Others	8	5.48
Total	146	100.00

 Table 4.2. Consumer's Awareness, Preference and Hygiene consideration

 Variable
 Frequencies

 Percentage (%)

### 4.1.3 Sensory characteristics of *Unam inung*

The Sensory characteristics of *Unam inung* is shown in Table 4.3. The taste of *Unam inung* was investigated in terms of saltiness. All (100%) the consumers said that *Unam inung* is salty. No consumer said otherwise. A total of 86.30% of the consumers attested to peculiar aroma of the product, 16% said otherwise while 4% of the consumers were indifferent. Majority (99.32%) of the consumers confirmed that the product develops odour quickly if not properly handled as well as getting bad rapidly.

Variable	Frequencies	Percentage (%)
Taste (Saltiness)		
Yes	146	100
No	0	0.00
Total	146	100.00
Smell (Aroma)		
Yes	126	86.30
No	16	10.96
Others	4	2.74
Total	146	100.00
Odour		
Yes	145	99.32
No	1	0.68
Total	146	100.00
Spoilage		
Yes	146	100.00
No	0	0
Total	146	100.00

 Table 4.3. Sensory characteristics of Unam inung

### 4.1.4 Consumers responses on the purchasing pattern of *Unam inung*

Consumers responses on the pattern of purchase of *Unam inung* are presented on Table 4.4, a total of 54% of the consumers purchased *Unam inung* weekly, 36% of the consumers purchased fortnightly, 7% purchased monthly and 2% purchased daily. The quantity of *Unam inung* purchased by the consumers was to the scale of 1kg as represented by 98% of the consumers while 1% buys up to 2kg.

Variable	Frequencies	Percentage (%)
Frequency of purchase		
Daily	3	2.05
Weekly	79	54.11
Fortnightly	53	36.30
Monthly	11	7.53
Total	146	100.00
Quantity of purchase		
1kg	144	98.63
2kg	2	1.37
3kg	0	0.00
>4kg	0	0.00
Total	146	100.00

## Table 4.4. Purchasing pattern of Unam inung

### 4.1.5 Consumers responses on the Consumption pattern of *Unam inung*

Consumers responses on the Consumption pattern of *Unam inung* are presented on Table 4.5, The product was mostly consumed on the same day of purchase as indicated by 79.45% while 19.86% of the consumers consumed it immediately after purchase. It is normally eaten with boiled cassava chips (*Edita Iwa*) as revealed by 97.26% of the consumers from the study, 2.06% of the consumers consumed it with other kinds of food, only one person prefers to consume it alone. The consumers' preference to consume it with accomplishment may be due to the saltyness of the product.

Majority of the consumers (59.59%) are willing and ready to buy the product if found in shops while 30.14% are not willing to buy from shop and 10.27% were indifferent. There is no constraint (cultural taboo) to eating the product as revealed by the response of the consumers (100%).

Variable	Frequencies	Percentage (%)
Time of consumption		
Same day	116	79.45
Immediately	29	19.86
Days after purchase	1	0.69
Total	146	100.00
Mode of consumption		
Alone	1	0.68
Local snack (edita iwa)	142	97.26
Others	3	2.06
Total	146	100
Readiness to buy from shops		
Yes	87	59.59
No	44	30.14
Others	15	10.27
Total	146	100.00
Cultural belief		
Yes	0	0
No	146	100.00
Total	146	100.00

## Table 4.5. Consumption pattern of Unam inung

### 4.1.6 Consumers level of Education as related to their responses

The relationship between the level of Education of consumers and their responses were analysed by Chi square test of independence and presented in Table 4.6. The level of Education showed significant relationship with Frequency of purchase, time of consumption, likeness, smell, readiness to buy from shops and hygiene status of *Unam inung*. Quantity of purchase, mode of consumption, taste and odour were independent of level of Education of the consumers of *Unam inung*.

Variable	X <sup>2</sup> value	df	p-value	remark
Frequency of purchase	26.25	6	0.00	Sig
Quantity of purchase	2.68	2	0.26	NS
Time of consumption	11.68	4	0.02	Sig
Mode of consumption	1.61	4	0.81	NS
Readiness to buy product				
from shop	12.62	4	0.01	Sig
Likeness	7.53	2	0.02	Sig
Taste	4.62	2	0.99	NS
Smell	10.35	4	0.04	Sig
Odour	3.89	2	0.14	NS
Hygiene	16.20	4	0.00	Sig

## Table 4.6. Test of relationship between the level of education of Unam inung

consumers and their responses

Sig: significant; NS: Not significant

### Phase 2: Qualitative evaluation of commercially available *Unam inung*

# **4.1.2.1** Microbial load (log cfu/g) of the commercially available *Unam inung* samples

The microbial load of the commercially available *Unam inung* samples are shown in Table 4.7. The Total Bacterial Counts from the three sources varied significantly from each other. Products from Processor 2 had the highest counts  $(3.36 \pm 0.02)$  followed by products from Processor 3  $(3.17 \pm 0.20)$  and products from Processor 1  $(3.07 \pm 0.11)$ . Total fungal counts (TFC) also varied across the sources significantly. Products from Processor 3 had the highest counts  $(2.87 \pm 0.02)$  followed by products from Processor 2  $(2.72 \pm 0.02)$  and products from Processor 1  $(2.60 \pm 0.04)$ .

 Table 4.7. Microbial load (log cfu/g) of the commercially available Unam inung samples

Parameters	Processor 1	Processor 2	Processor 3	SEM
TBC*	$3.07 \pm 0.11^{\circ}$	$3.36\pm0.02^a$	$3.17 \pm 0.20$ <sup>b</sup>	0.44
TFC	$2.60{\pm}0.04^{c}$	$2.72\pm0.02^{\text{b}}$	$2.87\pm0.02^{a}$	0.39

<sup>abc</sup> Means with different superscripts are significantly different (p<0.05) TBC: Total bacterial count; TFC: Total fungal count \* Acceptable limit: < 7 log cfu/g

# 4.1.2.2 Proximate composition (% DM) of the commercially available *Unam inung* samples

The Proximate composition (%DM) of the commercially available *Unam inung* samples are presented on Table 4.8. The moisture content of the samples from the three sources were not significantly different but ranged from  $46.67\pm1.00 - 47.02\pm2.00$ . The crude Protein content ranged from  $21.87\pm1.00 - 23.05\pm1.00$  but did not vary significantly. The Fat content was highest in Processor 1 ( $21.64\pm1.00$ ) followed by Processor 3 ( $20.01\pm1.01$ ) and least in Processor 2 ( $17.30\pm1.30$ ) and were significantly different. Ash content in products obtained from Processors 2 ( $6.50\pm1.00$ ) and 3 ( $6.84\pm1.00$ ) were significantly different from those obtained from Processor 1( $4.00\pm1.00$ ).

Parameters (%)	Processor 1	Processor 2	Processor 3	SEM
Moisture	47.02±2.00	46.95±1.00	46.67±1.00	0.41
Crude Protein	22.14±1.00	23.05±1.00	21.87±1.00	0.34
Fat	21.64±1.00 <sup>a</sup>	17.30±1.30 <sup>b</sup>	20.01±1.01 <sup>a</sup>	0.71
Ash	$4.00 \pm 1.00^{b}$	$6.50 \pm 1.00^{a}$	6.84±1.00 <sup>a</sup>	0.53

 Table 4.8. Proximate composition (%DM) of the commercially available Unam

 inung samples

<sup>a,b</sup> Means with different superscript on the same row are significantly (P<0.05) different

#### 4.1.2.3 Incidence of bacterial species in commercially available *Unam inung*

Table 4.9 shows the occurrence of microbiological organisms in the commercially available *Unam inung* samples. A total of twelve (12) bacteria were isolated from the *Unam inung* samples from the three processors. *Staphylococcus epidermidis* and *Aerococcus viridans* were the most commonly isolated bacteria occurring three times each at 25% followed by *Bacillus spharius, Staphylococcus aureus, Streptococcus avium* which occurred twice each at 16.67%. *Staphylococcus aureus* and *streptococcus avium* were absent in products from Processor 1 but present in Processor 2 and 3 respectively. A reverse trend is seen in *Bacillus spharius* which occurred in Processor 1 and 2 but absent in Processor 3.

Samples	Bacillus spharius	Staphylococcu s epidermidis	Staphylococcu s aureus	Streptococcu s avium	Aerococcus viridans	
Processor 1	+	+	-	-	+	
Processor 2	+	+	+	+	+	
Processor 3	-	+	+	+	+	
Total	2	3	2	2	3	
% Occurrence	16.67	25	16.67	16.67	25	

 Table 4.9. Incidence of bacterial species in commercially available Unam inung

+ = Positive present; - = Negative absent

### 4.1.2.4 Incidence of fungal species in commercially available *Unam inung*.

Table 4.10 show the occurrence of fungal species in commercially available *Unam inung*. The products from processor 1 had *Rhyzopus stolonifera* and *Pennicilium citrinum* with a 20% occurrence while products obtained from Processor 2 had Candida *pseudotropicalis, Candida tropicalis* and *Sacccaharomyces estuary* at 30% occurrence. The Unam inung from Processor 3 had *Verticillium. albo-atrum, Rhyzopus stolonifera, Candida pseudotropicalis, Candida tropicalis, Sacccaharomyces estuary* at 50% occurrence.

Fungal species	Processor 1	Processor 2	Processor 3
Verticillium. albo-atrum	-	-	+
Rhyzopus stolonifer	+	-	+
Pennicilium citrinum	+	-	-
Candida pseudotropicalis	-	+	+
Candida tropicalis	-	+	+
Sacccaharomyces estuary	-	+	+
% occurence	20	30	50

Table 4.10. Incidence of fungal species in commercially available Unam inung

+ = Positive present, - = Negative absent

### 4.1.2.5 Morphological and biochemical characteristics of bacterial isolates

The morphological characteristics biochemical characteristics and names of bacterial species in the sample are presented in Table 4.11. The bacteria identified were *Bacillus spharicus*, *Staphylococcus epidermidis*, *Staphylococcus aureus*, *Streptococus avium* and *Aerococcus viridans*.

	Cell shape	Gram stain	Catalase	Coagulase	Motility	Methyl Red	Citrate	Urase	Spore	VP	Indole	Oxidase	Starch	Glucose	Lactose	Maltose	Manitol	Xylose	probable Organism
1	Rod	+	+	-	+	+	+	+	+	-	-	+	-	00	00	00	00	Ao	Bacillus spharicus
2	Cocci	+	+	-	-	+	-	-	-	+	-	-	-	Ag	Ag	Ag	Ag	00	Staphylococcus epidermidis
3	Cocci	+	+	+	-	+	-	+	-	+	-	-	-	Ag	Ag	Ag	Ag	00	Staphylococcus aureus
4	Cocci	+	-	-	-	-	-	-	-	-	-	-	-	Ao	00	00	00	00	Streptococcus avium
5	Cocci	+	-	-	-	+	+	-	-	-	-	-	-	Ao	Ao	Ao	Ao	00	Aerococcus viridans

Table 4.11. Morphological and biochemical characteristics of bacterial isolates
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+ = positive reaction, - = negative reaction, Ag = acid with gas production, Ao = acid, no gas production, 00 = no acid, no gas,
 VP = Voges Proskaur

# STUDY 2: Quality attributes of *Unam inung* as influenced by different concentrations of salt (NaCl)

#### **4.2.1 Proximate composition**

The proximate composition of *Unam inung* salted at different levels is shown in Table 4. 12. Moisture, Crude protein, ether extract, and ash could not be determined for the 0% (Control) samples due to spoilage. The moisture content of *Unam inung* with 10%, 15%, and 20% salt concentrations were higher (P< 0.05) than that of 5% salt concentration. There was no statistical (P>0.05) difference between *Unam inung* with 10%, 15%, and 20% salt concentrations.

The Crude protein content of *Unam inung* salted at the five different levels showed an inverse pattern to moisture. However, *Unam inung* with 5% salt concentration recorded the highest (29.08%) Crude protein content while the least was observed at 10% (19.88%). *Unam inung* with 10%, 15%, and 20% salt concentrations were all similar statistically (P>0.05) but different (P<0.05) from that of 5% salt concentration.

The ether extract showed an increase with increasing level of salt from 10% to 20% (17.75 - 23.75) salt concentration. However, it reduced from 5% to 10% (25.33 to 17.75%) salt concentration. Pork treated with 10% salt was similar to that treated with 15% but was significantly lower than that of 20% (23.75%) and 5% (25.33%) salt concentration.

The ash content increased as the salt concentrations increased. *Unam inung* treated with 5% salt had the lowest (9.17%) ash content and was different (P<0.05) from that of 10% (18.66%), T4 (25.37%) and T5 (27.18%). *Unam inung* with 15% and 20% were significantly similar (P> 0.05 but different (P<0.05) from 5% and 10% salt concentrations.

			Salt concentration (%)						
Parameters	0	5	10	15	20	SEM			
Moisture	ND	$30.05{\pm}2.19^{\mathrm{b}}$	$44.14 \pm 1.68^{a}$	$45.32{\pm}3.67^a$	$44.82 \pm 3.39^{a}$	1.77			
Crude Protein	ND	$29.08{\pm}2.85^a$	$19.88 \pm 1.73^{b}$	$23.32{\pm}2.50^{\text{b}}$	$23.46{\pm}4.06^{b}$	4.29			
Ether Extract	ND	$25.33 \pm 3.06^{a}$	$17.75{\pm}2.65^{\mathrm{b}}$	21.25±1.02 <sup>ab</sup>	$23.75{\pm}4.28^a$	0.99			
Ash	ND	9.17±1.14 <sup>c</sup>	$18.66 \pm 2.49^{b}$	$25.37{\pm}1.92^{a}$	$27.18 \pm 0.14^{a}$	1.86			

 Table 4.12: Proximate composition (%) of Unam inung at different concentration of salt

<sup>abc</sup> Means with different superscript on the same row are significantly (P < 0.05) different.

**ND**: Not determined due to spoilage

#### 4.2.2 Weight loss of *Unam inung* at two days interval of *Unam inung*

Weight loss of *Unam inung* at two days interval is presented on Table 4.13. *Unam inung*, with 20% salt recorded the least loss  $(9.00 \pm 1.43g)$  at the first interval of weighing which was similar to that with 10% salt (12.89 ± 4.67g). *Unam inung* with 5% salt recorded the highest (15.63±2.47g) weight loss and was similar to 15% salting (14.12±2.13g) but both were different (P<0.05) from that of 20% salt concentration. The treatment with 0% salt concentration did not keep beyond 24 hours so no determinations were done on it.

At the second interval, *Unam inung* with 20% salt recorded the least (P< 0.05)  $(14.74\pm1.29g)$  and was similar statistically (P>0.05) to 15% salt concentration  $(19.15\pm0.94g)$ . *Unam inung* with 5% still recorded the highest  $(29.23\pm5.79g)$  weight loss just as in the first interval which was different (P<0.05) from other treatments.

By the third interval of weight loss assessment, the *Unam inung* with 5% salt concentration still exhibited the highest  $(32.54\pm6.00g)$  weight loss which was different (P<0.05) from other treatments. *Unam inung* with 10% and 15% salt concentrations were similar  $(24.45\pm3.34g; 21.33\pm1.01g)$  while 15% was also similar to 20%. The least  $(17.11\pm 0.64g)$  weight loss was recorded by *Unam inung* with 20% salt concentration.

Parameters	s Salt concentration (%)					
	0	5	10	15	20	_
Day3	ND	$15.63 \pm 2.47^{a}$	12.89±4.67 <sup>ab</sup>	14.12±2.13 <sup>a</sup>	9.00±1.43 <sup>b</sup>	0.91
Day5	ND	29.23±5.79 <sup>a</sup>	21.62±3.60b	$19.15 \pm 0.94^{bc}$	14.74±1.29 <sup>c</sup>	1.57
Day7	ND	$32.54{\pm}6.00^{a}$	24.45±3.34b	$21.33 \pm 1.01^{bc}$	17.11±0.64 <sup>c</sup>	1.65

 Table 4.13. Weight loss (g) of Unam inung at two days interval

<sup>abc</sup> means with different superscript are significantly(p>0.05) different. ND: not

determined due to spoilage

# **4.2.3** Drip loss (%) and water holding capacity (%) of *Unam inung* treated with different levels of salt

The Drip loss and Water holding capacity of *Unam inung* treated with different concentrations of salt was assessed at two days intervals as shown in Table 4.14. At day 3, *Unam inung* with 20% salt concentration recorded the least drip loss (9.00%) which was similar statistically (P>0.05) to those treated with 10% salt (11.35%). The highest drip loss (15.63%) was observed in *Unam inung* with 5% salt which was similar to those on 15% salt concentration (14.12%).

Drip loss on day 5 showed that *Unam inung* with 5% salt had the highest (P<0.05) drip loss of 19.63% which differed from the other treatments (10%, 15% and 20% -11.11%, 6.21% and 6.73% respectively). Drip loss of meat samples on 10%, 15% and 20% were all similar statistically (p>0.05) and ranged from 6.21% to 11.11%.

As at day 7, *Unam inung* on 5% salt concentration recorded the highest (4.98%) while those with 15% concentration had the least (2.78%). *Unam inung* with 5%, 10%, and 20% were similar statistically (P>0.05), while those on 10%, 15% and 20% were also similar.

The water holding capacity (%) for all the treatment were statistically (P>0.05) similar and ranged from 24.60% in 5% concentration to 28.00% in 20% concentration.

Parameters		Sa	SEM			
	0	5	10	15	20	
Day 3	ND	15.63 <sup>a</sup>	11.35 <sup>bc</sup>	14.12 <sup>ab</sup>	9.00 <sup>c</sup>	1.06
Day 5	ND	19.63 <sup>a</sup>	11.11 <sup>b</sup>	6.21 <sup>b</sup>	6.73 <sup>b</sup>	1.91
Day 7	ND	4.98 <sup>a</sup>	4.04 <sup>ab</sup>	2.78 <sup>b</sup>	3.16 <sup>ab</sup>	0.62
Water holding capacity	ND	24.66	25.65	26.00	28.00	1.81

 Table 4.14. Drip loss (%) and Water holding capacity (%) of Unam inung treated

 with different levels of Salt

 $^{abc}$  means with different superscript are significantly (p< 0.05) different. ND: not determined due to spoilage

# 4.2.4 pH values of *Unam inung* treated with different salt concentration at two days interval

Table 4.15 shows the pH of *Unam inung* treated with different concentrations of salt at two days interval. The pH of *Unam inung* on the first day of processing at 0% concentration recorded the least value of 6.80 which was significantly different from those of other concentrations (5, 10, 15, 20 %). The salted meat samples with salt concentrations (5, 10, 15, 20 %) were similar (P>0.05) and ranged from 7.36 (5%) to 7.91 (20%).

At the  $3^{rd}$  day of storage, the sample with 0% concentration had decomposed and the pH could not be measured while the pH of all the salt treated samples decreased with the highest concentration (20%) having the highest (P< 0.05) pH (7.40) than (6.78) of the least concentration (5%) of salt. The samples with 10% and 15% concentrations were statistically similar (P>0.05) to both 5% concentration and 20% concentration.

By the 5<sup>th</sup> day, the pH (7.32) of the sample with the least salt concentration (5%) increased and was higher (P < 0.05) than the pH (6.73, 6.76 and 6.86) of samples with 10%, 15%, and 20% concentration respectively. The pH of the samples with 10%, 15%, 20% concentrations decreased and were similar (P>0.05).

Parameters	arameters Salt concentration (%)					
	0	5	10	15	20	_
Day 1	6.82 <sup>b</sup>	7.36 <sup>a</sup>	7.48 <sup>a</sup>	7.72 <sup>a</sup>	7.91 <sup>a</sup>	0.18
Day 3	ND	6.78 <sup>b</sup>	7.09 <sup>ab</sup>	7.26 <sup>ab</sup>	7.40 <sup>a</sup>	0.18
Day 5	ND	7.82 <sup>a</sup>	6.73 <sup>b</sup>	6.76 <sup>b</sup>	6.86 <sup>b</sup>	0.21

 Table 4.15. pH values of Unam inung at days interval

<sup>abc</sup> Means with different superscript on the same row are significantly (P < 0.05) different

ND: Not determined due to spoilage

# **4.2.5** Microbiological load (log Cfu/g) and Regression analysis Total Bacteria Count and Total Fungi Count of *Unam inung* treated with different salt concentration

The microbiological load of *Unam inung* treated with different salt concentration is shown on Table 4.16. The Total Bacteria Count (TBC) was highest  $(8.05\pm0.09 \text{ Cfu/g})$  in the Control sample with 0% salt and lowest observed with 15% salting  $(3.16\pm0.23 \log \text{Cfu/g})$ .

The Total Fungal Count (TFC) was highest in *Unam inung* with 0% salt concentration  $(7.81\pm0.05 \log Cfu/g)$  and least in that with 15% salt  $(2.39\pm0.13 Cfu/g)$ .

The Total Pathogenic Bacteria Count recorded highest  $(6.97 \pm 0.06 \log \text{Cfu/g})$  count in samples with 0% salt concentration while those with 15% salting was lowest  $(2.30 \pm 0.79 \log \text{Cfu/g})$ . In addition, the count reduced with increased salt level from 0% to 15% salt concentration but increased at 20% salt concentration for TVC, TFC and TPBC.

*E. coli* was only observed in the control sample (0%) at a count of  $7.40 \pm 0.03 \log \text{Cfu/g}$ . *E. coli* count in the salted meat samples were not detected.

Salmonella/Shigella count was high in Unam inung with 0% salt concentration  $(7.39\pm 0.02 \log \text{Cfu/g})$  and least in  $(0.3\pm 0.65 \text{ Cfu/g})$  in 10% salt concentration. The count reduced with increasing salt levels from 0% to 10% salt concentration. However, the count was not observed in Unam inung with 15% salt concentration but a value of 0.75 log Cfu/g was obtained in samples with 20% salt concentration.

The result of regression analysis for TBC (fig 2) showed that salt inclusion accounted for 99.5% of the effect on Total Bacteria Count. The linear and quadratic regression were significant (P< 0.011) while the cubic regression was not significant (P> 0.915).

On Figure 3, the regression result for Total Fungal count indicates that salt inclusion accounted for 97% of the effect on TFC. The linear regression was significantly (P < 0.001) different.

Parameters	Salt concentration (%)						
	0	5	10	15	20	-	
TBC	8.05±0.09 <sup>a</sup>	$5.51 \pm 0.14^{b}$	3.52±0.18 <sup>c</sup>	3.16±0.23 <sup>d</sup>	3.47±0.14 <sup>c</sup>	0.43	
TFC	$7.81 \pm 0.05^{a}$	3.68±0.20 <sup>b</sup>	$2.47{\pm}0.63^{cd}$	$2.39{\pm}0.13^{d}$	3.04±0.47 <sup>c</sup>	0.44	
TPBC	$6.97 \pm 0.06^{a}$	$3.41{\pm}0.19^{b}$	$2.35{\pm}0.14^{c}$	2.30±0.79 <sup>c</sup>	$2.45 \pm 0.35^{\circ}$	0.42	
E. Coli	7.40±0.03 <sup>a</sup>	$0.00{\pm}0.00^{\rm b}$	$0.00\pm0.00^{b}$	$0.00\pm0.00^{b}$	$0.00\pm0.00^{b}$	0.67	
SSC	7.39±0.02 <sup>a</sup>	1.39±1.00 <sup>b</sup>	0.3±0.65°	$0.00 \pm 0.00^{c}$	$0.75\pm0.5^{bc}$	0.64	

 Table 4.16. Microbial load (log Cfu/g) of Unam inung treated with different concentrations of salt

<sup>abc</sup> Means with different superscript on the same row are significantly (P< 0.05) different.</li> **TVC**: Total Bacteria count; **TFC**: Total fungal count; **TPBC**: Total pathogenic bacterial count; SSC: Salmonella Shigella count.

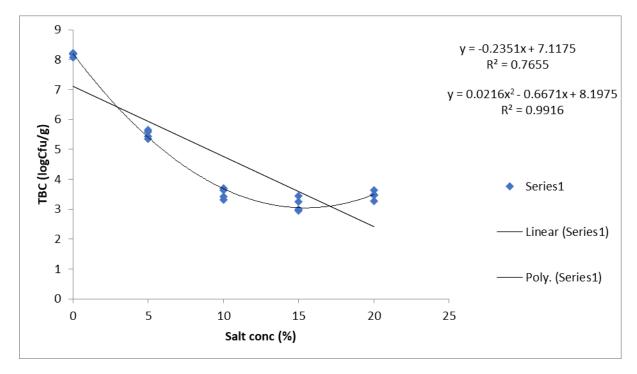


Figure 4.1. Total Bacteria Count (log cfu/g) of *Unam inung* treated with different concentration of salt

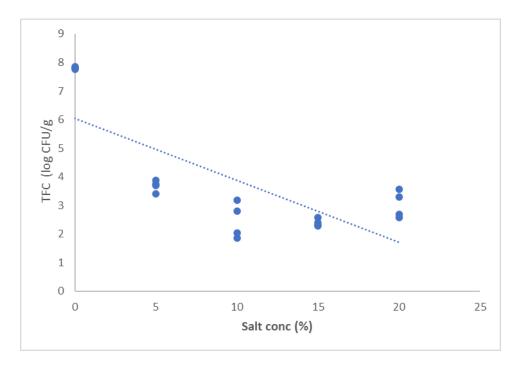


Figure 4.2. Total Fungal Count (log cfu/g) of *Unam inung* treated with different concentration of salt

#### 4.2.6 Thiobarbituric acid test (mg/kg) of *Unam inung* at days interval

The Thiobarbituric acid test (mg/kg) of *Unam inung* treated at different salt concentrations were assessed at days intervals. The results are presented on Table 4.17.

At the point of preparation (Day 0), *Unam inung* with 20% salt concentration recorded the highest (0.036  $\pm$ 0.005mg/kg) (P< 0.05) while the least was observed for those with 10% salt concentration (0.018  $\pm$ 0.004). *Unam inung* with 0% salt concentration (0.021 $\pm$ 0.04mg/kg) was similar (P>0.05) with those preserved with 5% salting (0.028 $\pm$ 0.004mg/kg), and 15% (0.028 $\pm$ 0.005mg/kg), while that with 15% was similar with those preserved with 0% salt concentration.

By day 3, *Unam inung* with 10%, 15% and 20% salt concentration recorded were significantly (P>0.05) higher  $(0.150\pm 0.038, 0.150\pm 0.038$  and  $0.139\pm 0.08$ mg/kg) than 0%  $(0.072\pm 0.012$ mg/kg) but 5% salt concentration  $(0.114\pm 0.012$ mg/kg)was similar to both all other treatments.

At day 5, *Unam inung* samples with different salt concentrations showed no particular trend for the lipid oxidation.

At day 7, TBARs in 10% ( $0.616\pm0.008$ mg/kg) and 15% ( $0.634\pm0.012$ mg/kg) were similarly higher (P< 0.05) than those with 20% salt concentration ( $0.484\pm0.013$ mg/kg), 5% salt concentration ( $0.195\pm0.008$ mg/kg) and 0% salt concentration ( $0.174\pm0.012$ mg/kg).

Parameter	0%	5%	10%	15%	20%	SEM
S						
Day 0	$0.021 \pm 0.004^{bc}$	$0.028 \pm 0.004^{b}$	0.018±0.004°	0.028±0.005 <sup>b</sup>	$0.036 \pm 0.005^{a}$	0.002
Day 3	0.072±0.012 <sup>b</sup>	$0.114 \pm 0.012^{ab}$	0.150±0.038ª	0.150±0.038 <sup>a</sup>	0.139±0.008ª	0.010
Day 5	0.276±0.012 <sup>b</sup>	0.206±0.005 <sup>e</sup>	$0.257 \pm 0.000^{\circ}$	$0.231 \pm 0.009^{d}$	0.320±0.008ª	0.011
Day 7	$0.174 \pm 0.012^{d}$	0.195±0.008°	$0.616 \pm 0.008^{a}$	0.634±0.012 <sup>a</sup>	0.484±0.013 <sup>b</sup>	0.053

Table 4.17. Thiobarbituric acid test (TBARS) determined at days intervals of 0, 3, 5 and 7 days of storage in mg/kg

<sup>abcd</sup> Means with different superscript on the same row are significantly (P < 0.05) different

### Study 3: Evaluation of the quality attributes of differently processed Unam inung

# **4.3.1** Effect of processing methods and meat cuts on the proximate composition (%) of *Unam inung*.

The effect of processing methods and meat cuts on the proximate composition (%) of *Unam inung* is presented on Table 4.18. For processing methods, the raw *Unam inung* recorded the highest moisture content (65.02%) which was varied (P< 0.05) from Sundried (43.56%) and Smoked (32.29%). The higher the heat, the lower the moisture content. For Ash, the reverse was the case as it rather increased with increased in heat. Smoked *Unam inung* was highest in Ash content (5.74%) which also varied (P<0.05) from Sundried (3.22%) and raw (1.51%). The Protein content was highest in Smoked *Unam inung* (27.32%) and this was higher (P<0.05) than protein content of Sundried (22.28%) and raw (19.90%). Increasing heat led to increased Protein content in methods of meat processing. The Fat content was highest in Sundried (22.50%) which was similar (P>0.05) to the smoked *Unam inung* (22.02%). Both Smoked and Sundried fat content in *Unam inung* were higher (P<0.05) than the fat content in Raw (10.23%).

The proximate composition of the Meat cuts (Belly, Ham, Shoulder and Loin) showed that moisture content was highest (48.40%) in Belly while the least (P<0.05) was observed in Ham (45.17%). All Meat cuts were varied (P<0.05) from each other. For Ash, Loin had the highest (P<0.05) content (3.82%) that was significantly different from the other meat cuts (Belly, Ham and Shoulder being 3.23%, 3.52% and 3.37% respectively). The Protein content was least (21.01%) in Loin and Highest (25.80%) in shoulder. Protein content was varied (P< 0.05) from each other. The Fat content was lowest (16.71%) in Shoulder and the highest value observed in Belly (20.38%). Ham and Loin were similar in Fat content (17.84% and 18.07%).

Parameter	Moisture	Ash	Protein	Fat
Processing method				
Raw	$65.02 \pm 0.53^{a}$	1.51±0.31°	19.90±1.11 <sup>c</sup>	10.23±0.65 <sup>b</sup>
Sundried	$43.58 \pm 2.24^{b}$	$3.22 \pm 0.57^{b}$	$22.28 \pm 2.23^{b}$	22.50±2.41ª
Smoked	$32.29 \pm 2.04^{c}$	5.74±0.11 <sup>a</sup>	27.32±3.83 <sup>a</sup>	$22.02 \pm 2.02^{a}$
SEM	0.07	0.01	0.22	0.19
Meat Cut				
Belly	$48.40{\pm}12.94^{a}$	$3.23{\pm}1.88^d$	21.93±2.05 <sup>c</sup>	20.38±7.19 <sup>a</sup>
Ham	45.17±15.96 <sup>d</sup>	$3.52{\pm}1.81^{b}$	23.94±5.29 <sup>b</sup>	17.84±5.53 <sup>b</sup>
Shoulder	46.56±14.85 <sup>c</sup>	3.37±2.07°	25.80±4.66ª	16.71±5.61 <sup>c</sup>
Loin	$47.71 \pm 13.88^{b}$	$3.82{\pm}1.68^{a}$	$21.01{\pm}1.54^{d}$	18.07±6.10 <sup>b</sup>
SEM	0.07	0.01	0.25	0.22
P-value				
Processing method	< 0.01	< 0.01	< 0.01	< 0.01
Meat Cut	< 0.01	< 0.01	< 0.01	< 0.01
PM x MC	< 0.01	< 0.01	< 0.01	< 0.01

 Table 4.18. Effects of processing methods and meat cuts on the proximate composition of *Unam inung*

<sup>abcd</sup> Means on the same column within each subset with different superscripts are significantly different (P< 0.05) PM: Processing method; MC: Meat cut

# **4.3.2** Interaction effects of processing methods and meat cuts on the proximate composition of *Unam inung*.

The interaction effect of processing method X meat cuts is presented on Table 19. Under moisture content of *Unam inung*, Raw X Ham recorded the highest content (65.45%) while that of Smoked X Ham (29.43%) was the least (P<0.05). The Raw X meat cut values were higher and significantly (P<0.5) different from those of Sundried X Meat cut. The smoked X meat cut were the least in values for Moisture.

Ash content was highest in Smoked X Meat cut, ranging from 5.62% in Loin to 5.85% in Shoulder. However, the Smoked Loin was similar to Smoked Belly also Ham and Shoulder. Sundried X Meat cuts was next to Smoked X meat cuts while Raw X Meat cut had the least values.

High (P<0.05) Protein content were recorded in Smoked X meat cut for Shoulder and Ham (31.33% and 30.43% respectively). The least values for Protein were observed in Raw X ham of 18.59%.

Sundried X meat cut (Belly) recorded the highest fat content value (25.84%) which was significantly different from others irrespectively of processing method or meat cut. The least value for fat was observed for Raw X meat cut (shoulder) of 9.41% and all values here were significantly lower than those of Sundried X meat cut and Smoke X meat cut.

Processing	Meat cut	Moisture	Ash	Protein	Fat
method					
Raw	Belly	64.29±0.05 <sup>c</sup>	$1.43\pm0.04^{h}$	20.42±0.33 <sup>e</sup>	$10.85{\pm}0.30^{\rm f}$
	Ham	$65.45{\pm}0.04^a$	1.78±0.03 <sup>g</sup>	18.59±1.38 <sup>d</sup>	$10.64{\pm}0.02^{\rm f}$
	Shoulder	$65.35{\pm}0.04^{ab}$	1.06±0.03 <sup>i</sup>	$20.64 \pm 1.00^{d}$	$9.41{\pm}0.03^{g}$
	Loin	64.99±0.57 <sup>b</sup>	1.76±0.04 <sup>g</sup>	19.96±0.27 <sup>d</sup>	$10.01{\pm}0.60^{fg}$
Sundried	Belly	$46.29{\pm}0.05^d$	$2.63\pm0.02^{f}$	$20.74 \pm 0.10^{d}$	$25.84{\pm}~1.02^{a}$
	Ham	40.64±0.01 <sup>g</sup>	2.95±0.02 <sup>e</sup>	22.79±1.35°	$20.03{\pm}0.05^{e}$
	Shoulder	$42.59{\pm}0.03^{\rm f}$	$3.21\pm0.01^d$	25.43±0.18 <sup>b</sup>	$21.50{\pm}~1.52^{d}$
	Loin	44.78±0.00 <sup>e</sup>	4.09±0.02 <sup>c</sup>	$20.17{\pm}0.20^d$	$22.64 \pm 1.02^{cd}$
Smoked	Belly	$34.63{\pm}0.02^h$	5.64±0.03 <sup>b</sup>	24.63±0.47 <sup>b</sup>	$24.46{\pm}0.03^{b}$
	Ham	$29.43{\pm}0.03^k$	5.83±0.02 <sup>a</sup>	$30.43 \pm 0.3^{a}$	$22.84{\pm}0.03^{c}$
	Shoulder	$31.74{\pm}0.11^{j}$	5.85±0.03 <sup>a</sup>	$31.33{\pm}0.18^a$	$19.22\pm0.02^{\text{e}}$
	Loin	$33.36 \pm 0.50^{i}$	5.62±0.03 <sup>b</sup>	$22.90{\pm}~1.15^{c}$	$21.57{\pm}0.60^d$
SEM		2.31	0.30	0.67	1.00

 Table 4.19. Interaction between the Processing methods and meat cuts on the proximate composition of Unam inung

<sup>abcdefghi</sup> Means on the same column with different superscripts are significantly different (P < 0.05)

#### 4.3.3 Effect of processing methods and meat cuts on the histology of *Unam inung*

The effect of processing methods and meat cuts on the histology of *Unam inung* is presented on Table 4.20.

Processing methods had no effect (P>0.05) on the histological parameters (area, width, length, volume and diameter) measured.

For meat cuts, there were significant differences (P<0.05) in area and diameter, while width, length and volume were not affected (P>0.05) by the cuts.

Belly was lowest (P<0.05) in area (2384.66  $\mu$ m<sup>2</sup>) and significantly different from Ham, Shoulder and Loin (5717.47  $\mu$ m<sup>2</sup>,46069.80  $\mu$ m<sup>2</sup>, and 4815.53  $\mu$ m<sup>2</sup>). The ham, shoulder, and loin were similar statistically (P>0.05).

Belly was highest (P<0.05) in diameter (681.33 $\mu$ m) and this was significantly different from Ham, Shoulder and Loin (402.73  $\mu$ m, 252.27  $\mu$ m and 253.60  $\mu$ m respectively). The Ham, Shoulder and Loin were all similar (P>0.05) in diameter.

Parameter	Area (µm <sup>2</sup> )	Width (µm)	Length (µm)	Volume (µm <sup>3</sup> )	Diameter (µm)
Processing method					
Raw	$4362.95 \pm 2681.86$	403.50±194.30	500.35±245.51	4358.70± 1989.17	$389.15 \pm 335.84$
Sundried	3726.90±3048.56	$353.25{\pm}237.58$	464.55±257.05	3993.75± 2498.11	412.85±301.20
Smoked	5055.95±2792.95	$299.30 \pm 140.50$	387.30±188.78	$4047.50 \pm 2534.87$	$390.45 \pm 268.57$
SEM	582.95	41.09	41.43	495.94	53.02
Meat cut					
Belly	2384.66± 1956.72 <sup>b</sup>	$317.67 \pm 116.02$	$406.20 \pm 207.39$	3946.13±1551.17	681.33±289.06ª
Ham	$5717.47 \pm 3097.64^{a}$	323.73±189.63	$432.61 \pm 150.14$	4701.60± 2673.44	402.73±267.31 <sup>b</sup>
Shoulder	4609.80±2682.22 <sup>a</sup>	403.13±259.92	$536.01 \pm 280.23$	$4170.47 \pm 2727.94$	$252.27 \pm 208.06^{b}$
Loin	4815.53±2676.27ª	$363.53{\pm}200.86$	428.00±272.88	$3715.07 \pm 2258.94$	253.60± 212.51 <sup>b</sup>
SEM	673.13	47.45	47.84	572.66	61.22
P-value					
Processing method	0.28	0.21	0.15	0.85	0.94
Meat cut	0.01	0.56	0.23	0.66	< 0.01
PM x MC	0.37	0.04	< 0.01	0.04	0.07

Table 4.20. Effects of Processing methods and meat cuts on the histomorphormetry of *Unam inung* 

<sup>abcd</sup> Means on the same column within similar subset with different superscripts are significantly different (P< 0.05) **PM**: Processing method; **MC**: Meat cut

# 4.3.4 Interaction effects of processing methods and meat cuts on the histomorphormetry of *Unam inung*

The Interaction effects of processing methods and meat cuts on the histomorphormetry of *Unam inung* is presented on Table 4.21. Sundried X shoulder recorded the highest (P<0.05) value (786.00 $\mu$ m) for length which was similar statistically (P>0.05) with Raw X Loin (675.40  $\mu$ m), Raw X Belly (593.40  $\mu$ m) and Sundried X Ham (530.60  $\mu$ m) but different from the rest. The least value for length was recorded for Sundried X Loin (207.60  $\mu$ m).

The highest value for width was recorded for Sundried X Shoulder (551.00  $\mu$ m) and thus significantly different (P<0.05) from Sundried X Belly (248.20  $\mu$ m), Smoked X ham (215.80  $\mu$ m) and (the least) Sundried X Loin (207.60  $\mu$ m). Sundried X Shoulder for width was statistically similar(P> 0.05) with the remaining interactions.

The least values for Volume was recorded for Sundried X Loin (2266.80  $\mu$ m<sup>3</sup>) which was similar (P>0.05) with Smoked X Ham (2515.60  $\mu$ m<sup>3</sup>) but significantly different (P<0.05) from Sundried X Ham (6273.80  $\mu$ m<sup>3</sup>). Sundried X Ham was similar with the other interactions not mentioned.

Processing	Meatcut	Length(µm)	Width(µm)	Volume(µm <sup>3</sup> )
method				
Raw	Belly	$593.40 \pm 158.63^{abc}$	$388.80 \pm 143.32^{ab}$	$4918.20 {\pm}~1614.70^{ab}$
	Ham	392.60± 140.65 <sup>cde</sup>	399.40± 193.35 <sup>ab</sup>	$5315.40 \pm 2247.49^{ab}$
	Shoulder	$340.00 \pm 291.02^{cde}$	$340.00 \pm 291.02^{ab}$	$3425.20 \pm 1318.46^{ab}$
	Loin	$675.40 \pm 246.76^{ab}$	$535.80 \pm 76.49^{a}$	$3776.00 \pm 2519.69^{ab}$
Sundried	Belly	334.00± 166.79 <sup>cde</sup>	$248.20{\pm}~64.33^{b}$	$3501.60 \pm 1020.59^{ab}$
	Ham	530.60± 146.27 <sup>abcd</sup>	$406.20 \pm 248.22^{ab}$	6273.80± 2489.90 <sup>a</sup>
	Shoulder	786.00± 150.95ª	$551.00 \pm 325.98^{a}$	$3932.80 \pm 3069.74^{ab}$
	Loin	$207.60 \pm 58.33^{e}$	$207.60 \pm 58.33^{b}$	$2266.80 \pm 1563.45^{b}$
Smoked	Belly	$291.20 \pm 175.20^{de}$	$316.00 \pm 100.37^{ab}$	$3418.60 \pm 1731.06^{ab}$
	Ham	374.80± 139.80 <sup>cde</sup>	$215.80{\pm}~54.04^{b}$	$2515.60 \pm 2056.87^{b}$
	Shoulder	482.20± 193.93 <sup>bcd</sup>	$318.40\pm 64.93^{ab}$	$5153.40 \pm 3593.26^{ab}$
	Loin	401.00± 241.23 <sup>cde</sup>	347.20± 253.23 <sup>ab</sup>	$5102.40 \pm 2008.72^{ab}$
SEM		30.12	25.34	299.43

 Table 4.21. Interaction between the Processing methods and Meat cuts on the

 Histomorphometry of Unam inung

 $^{abcd}$  Means on the same column with different superscripts are significantly different (P< 0.05)

#### 4.3.5 Histology of muscles

The histologic sections of the raw, sundried and smoked *Unam inung* are captured the photomicrographs on Plates 1-12. The photomicrographs AB – AS represents the raw meat cuts: AB- raw Belly, AH- raw Ham, AL- raw Loin, and AS- raw shoulder. BB- BS represents the sundried meat cuts: BB sundried Belly, BH- sundried Ham, BL- sundried loin, BS- sundried shoulder. CB-CS represents the smoked meat cuts: CB- smoked Belly, CH- smoked Ham, CL- smoked Loin, and CS- smoked shoulder

These photomicrographs show the Histologic section of the muscle at two magnifications, (x100) and (x400). The upper photomicrographs depicts (x100) while the lower depicts (x400) stained by H&E method.

Plates 1-4 bearing AB-AS revealed normal cellular profile of Muscle spindle, Myocytes, Perimysium, Endomysium Interstitial space, Blood vessel, Nucleus, Adipocytes, Connective Tissue and Muscle fibers within normal cellular architecture.

Plates 5-8 bearing BB-BS revealed distortion and splitting of muscle fiber and spindle as compared to control group.

Plates 9-12 bearing CB- CS revealed distortion and splitting of muscle fiber and spindle as compared to control group

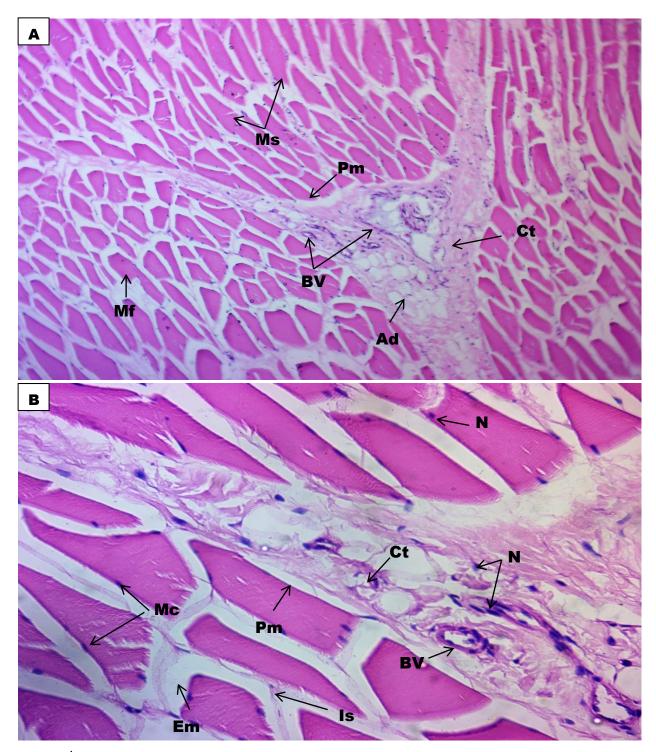


Plate 4.1 Photomicrographs of raw belly at magnification A(x100) and B(x400) stained with H&E method.

*Keys: Muscle spindle* (*Ms*) *Myocytes* (*Mc*), *Interstitial space* (*Is*), *Blood vessel* (*Bv*) *Nucleus* (*N*), *Adipocytes* (*Ad*), *Connective Tissue* (*Ct*), *Muscle fibers* (*Mf*), *Perimysium* (*Pm*), *Endomysium* (*Em*)

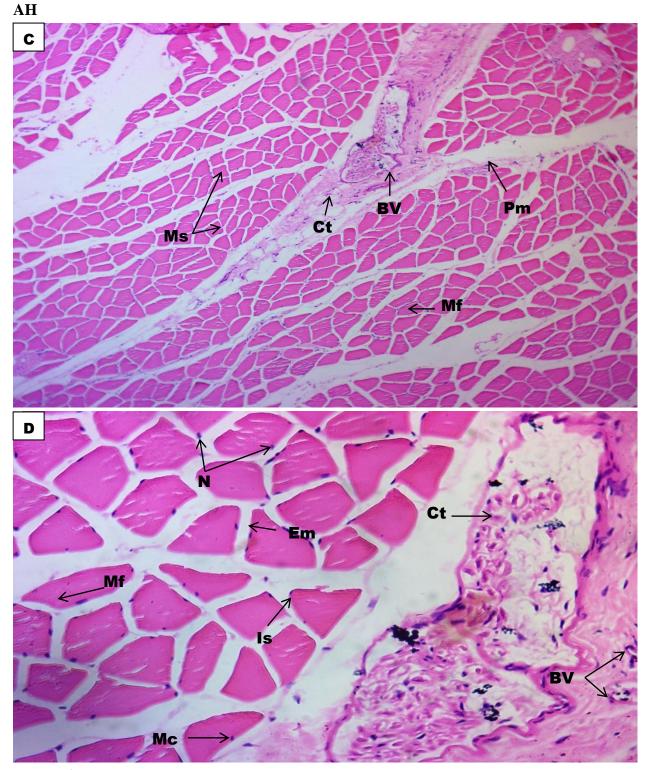


Plate 4.2 Photomicrographs of raw ham at magnification C(x100) and D(x400) stained with H&E method.

*Keys: Muscle spindle* (*Ms*) *Myocytes* (*Mc*), *Interstitial space* (*Is*), *Blood vessel* (*Bv*), *Nucleus* (*N*), *Connective Tissue* (*Ct*), *Muscle fibers* (*Mf*), *Endomysium* (*Em*) *Perimysium* (*Pm*),

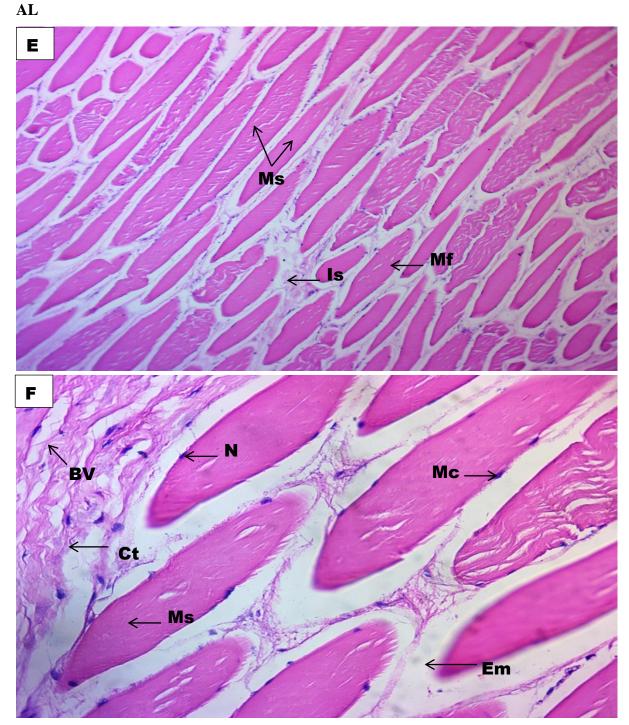


Plate 4.3. Photomicrographs of raw loin at magnification E(x100) and F(x400) stained with H&E method.

*Keys: Muscle spindle* (*Ms*) *Myocytes* (*Mc*), *Interstitial space* (*Is*), *Blood vessel* (*Bv*) *Nucleus* (*N*), *Connective Tissue* (*Ct*), *Muscle fibers* (*Mf*), *Endomysium* (*Em*)

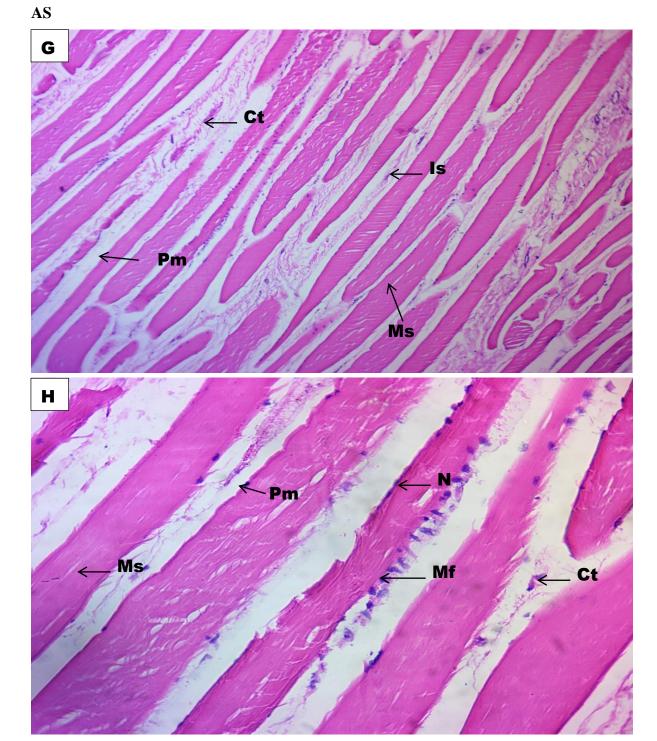


Plate 4.4 Photomicrographs of raw shoulder at magnification G(x100) and H(x400) stained with H&E method.
Keys: Muscle spindle (Ms), Interstitial space (Is), Blood vessel (Bv) Nucleus (N), Connective Tissue (Ct), Muscle fibers (Mf), Perimysium (Pm)



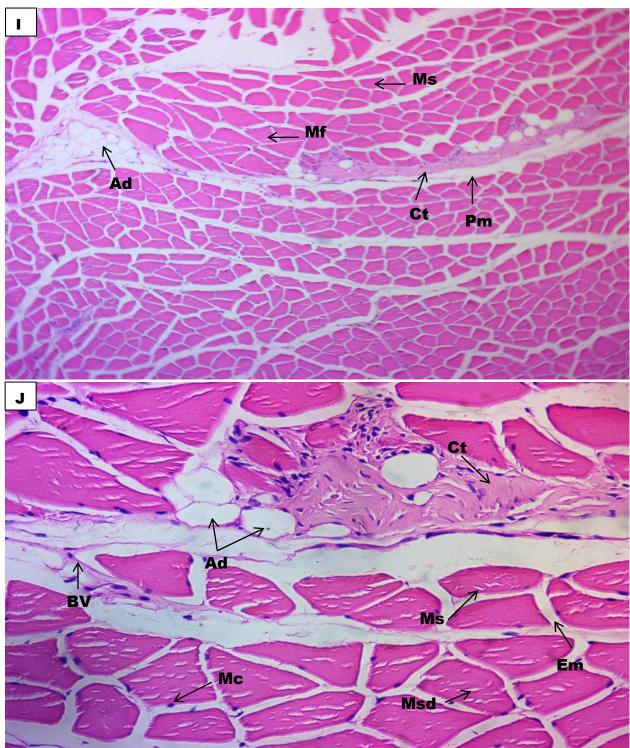
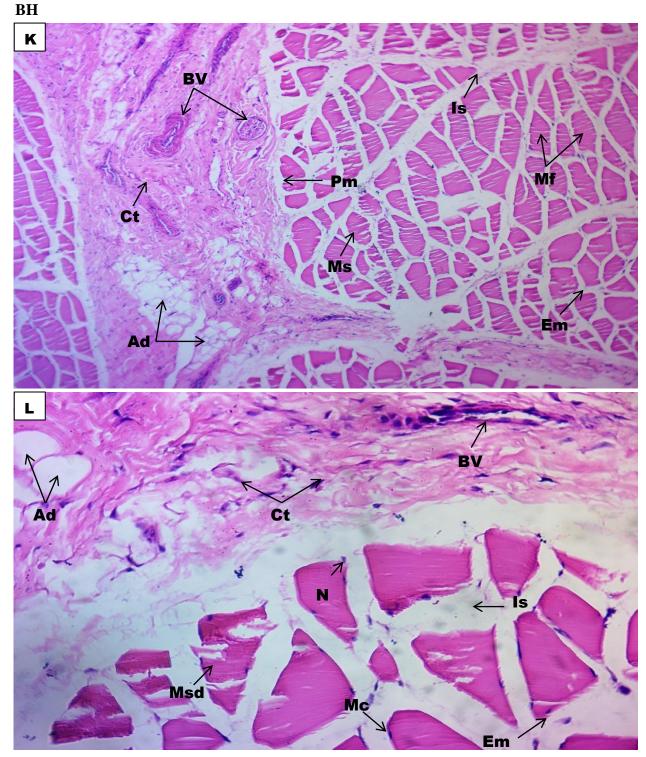


Plate 4.5 Photomicrographs of sundried belly at magnification I(x100) and J(x400) stained with H&E method.

**Keys:** Muscle spindle (**Ms**) Myocytes (**Mc**), Interstitial space (**Is**), Blood vessel (**Bv**), Nucleus (**N**), Adipocytes (**Ad**), Connective Tissue (**Ct**), Muscle fibers (**Mf**), Muscle spindle distortion (**Msd**), Perimysium (**Pm**), Endomysium (**Em**)



*Plate 4.6 Photomicrographs of sundried ham at magnification* K(x100) *and* L(x400) *stained with* H&E *method.* 

**Keys:** Muscle spindle (**Ms**) Myocytes (**Mc**), Interstitial space (**Is**), Blood vessel (**Bv**), Nucleus (**N**), Adipocytes (**Ad**), Connective Tissue (**Ct**), Muscle fibers (**Mf**), Muscle spindle distortion (**Msd**), Perimysium (**Pm**) and Endomysium (**Em**).

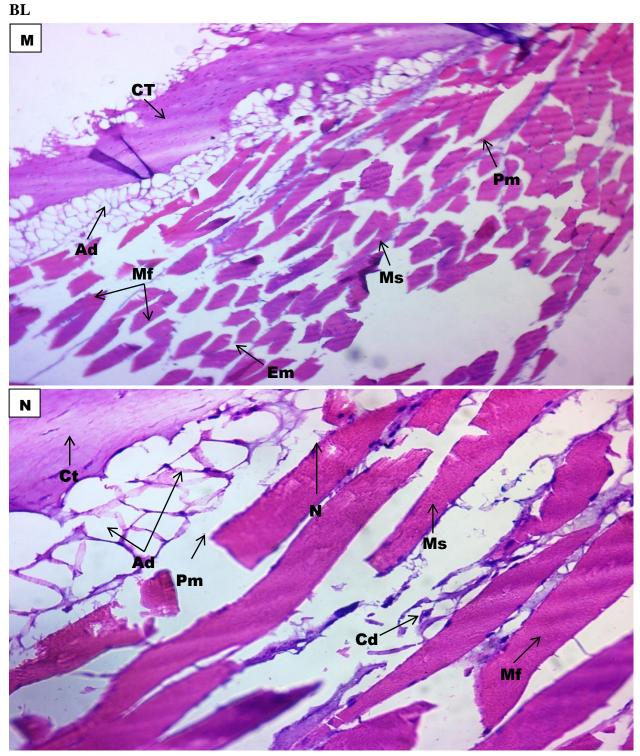
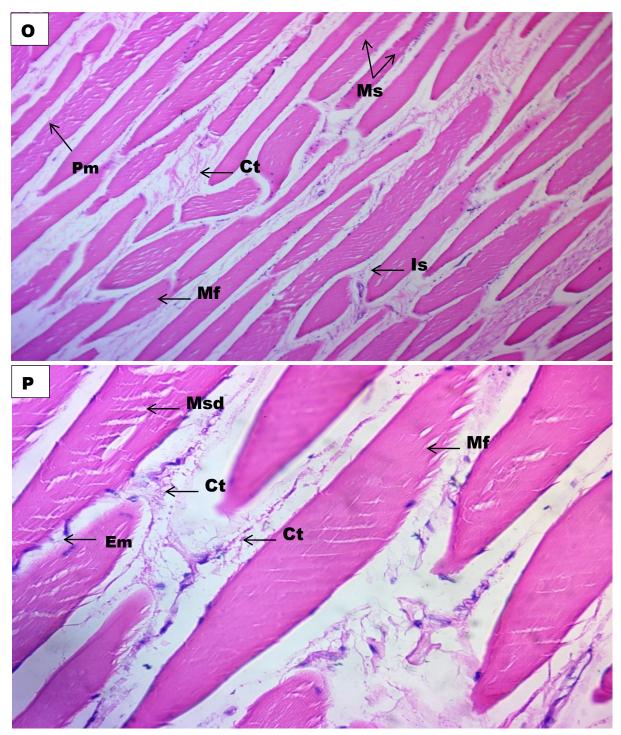


Plate 4.7 Photomicrographs of sundried loin at magnification M(x100) and N(x400) stained with H&E method.

*Keys: Muscle spindle* (*Ms*), *Interstitial space* (*Is*), *Blood vessel* (*Bv*), *Nucleus* (*N*), *Adipocytes* (*Ad*), *Connective Tissue* (*Ct*), *Muscle fibers* (*Mf*), *Muscle spindle distortion* (*Msd*) and Perimysium (*Pm*) and Connective tissue distortion (*Cd*)





*Plate 4.8 Photomicrographs of sundried shoulder at magnification* O(x100) *and* P(x400) *stained with* H&E *method.* 

**Keys:** Muscle spindle (**Ms**) Myocytes (**Mc**), Interstitial space (**Is**), Blood vessel (**Bv**), Nucleus (**N**), Adipocytes (**Ad**), Connective Tissue (**Ct**), Muscle fibers (**Mf**), Muscle spindle distortion (**Msd**), Perimysium (**Pm**), Endomysium (**Em**)

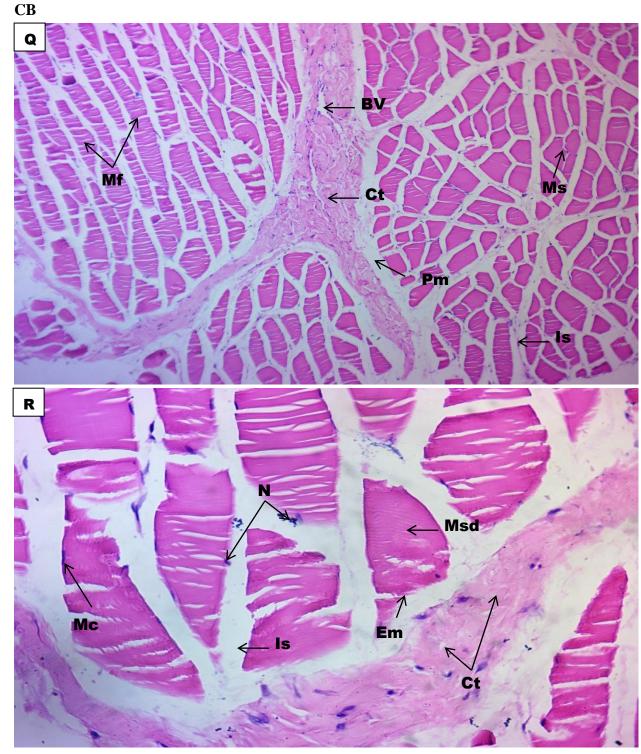


Plate 4.9 Photomicrographs of Smoked belly at magnification Q(x100) and R(x400) stained with H&E method.

**Keys:** Muscle spindle (**Ms**) Myocytes (**Mc**), Interstitial space (**Is**), Blood vessel (**Bv**), Nucleus (**N**), Adipocytes (**Ad**), Connective Tissue (**Ct**), Muscle fibers (**Mf**), Muscle spindle distortion (**Msd**), Perimysium (**Pm**) and Endomysium (**Em**).

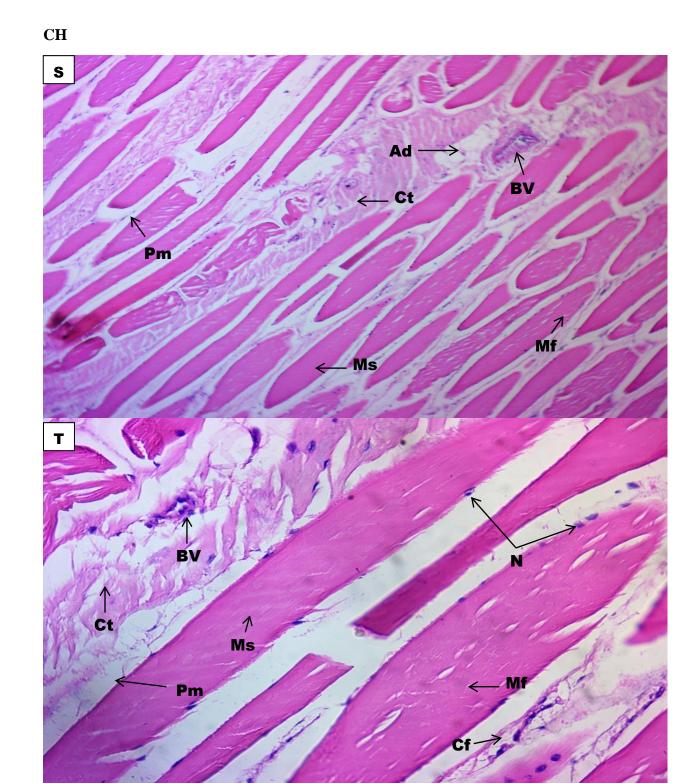


Plate 4.10 Photomicrographs of smoked ham at magnification S(x100) and T(x400) stained with H&E method.

Keys: Muscle spindle (Ms), Interstitial space (Is), Blood vessel (Bv), Nucleus (N), Adipocytes (Ad), Connective Tissue (Ct), Muscle fib ers (Mf), Muscle spindle distortion (Msd) and Perimysium (Pm)

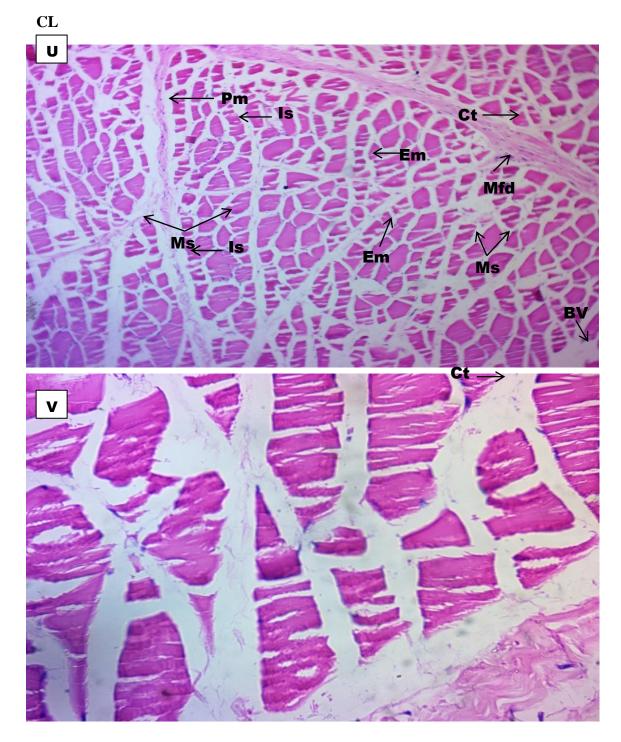


Plate 4.11 Photomicrographs of smoked loin at magnification U(x100) and V(x400) stained with H&E method.

*Keys: Muscle spindle* (*Ms*), *Interstitial space* (*Is*), *Blood vessel* (*Bv*), *Nucleus* (*N*), *Adipocytes* (*Ad*), *Connective Tissue* (*Ct*), *Muscle fibers* (*Mf*), *Muscle spindle distortion* (*Msd*), *Perimysium* 

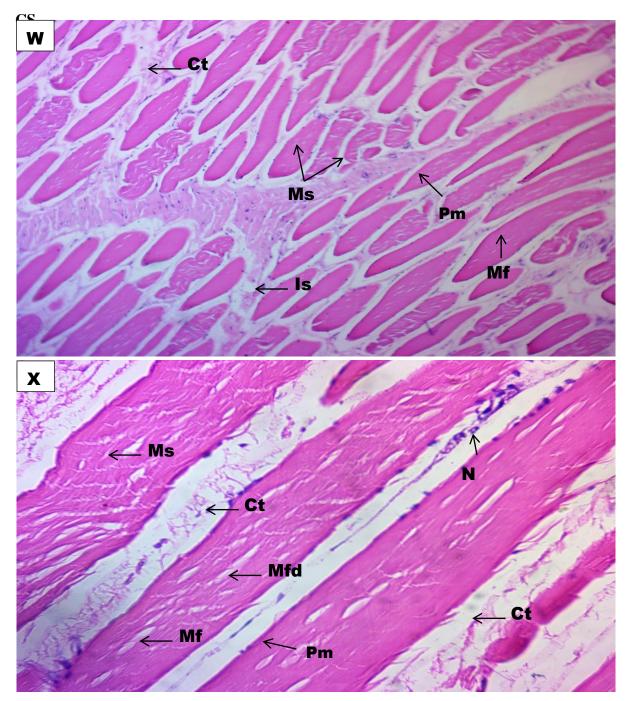


Plate 4.12 Photomicrographs of smoked shoulder at magnification W(x100) and X(x400) stained with H&E method.

**Keys:** Muscle spindle (**Ms**) Myocytes (**Mc**), Interstitial space (**Is**), Blood vessel (**Bv**), Nucleus (**N**), Adipocytes (**Ad**), Connective Tissue (**Ct**), Muscle fibers (**Mf**), Muscle spindle distortion (**Msd**) and perimysium (**Pm**)

#### 4.3.6 Main effect of total bacterial and total fungal count (log cfu/g) in *Unam inung*

The main effect of total bacterial and total fungal count (log cfu/g) in *Unam inung* is presented on Table 4.22. Total Bacterial Count (TBC) was highest (P<0.05) in Raw (processing method) with 3.22 log cfu/g and 3.06 log cfu/g respectively). Sundried and Smoke (3.09 log cfu/g and 3.06 log cfu/g). Sundried and Smoked were similar in terms of TBC.

The TFC was highest (P<0.05) in Smoked (4.65log cfu/g) and the least (P<0.05) was recorded in Raw (2.34 log cfu/g). Sundried which had 3.58 log cfu/g varied (P<0.05) from both Raw and Smoked.

The meat cuts showed no variation (P>0.05) in both TBC and TFC however, the interaction of the processing method and meat cut for TFC were significant (P<0.05).

Parameter	TBC	TFC
Processing method		
Raw	$3.22 \pm 0.12^{a}$	$2.34 \pm 0.27^{\circ}$
Sundried	$3.09 \pm 0.05^{b}$	$3.58 \pm 0.04^{b}$
Smoked	$3.06 \pm 0.01^{b}$	$4.65 \pm 0.03^{a}$
SEM	0.02	0.01
Meat cut		
Belly	$3.16 \pm 0.12$	$3.67{\pm}0.87$
Ham	$3.11 \pm 0.09$	$3.45 \pm 1.06$
Shoulder	$3.12 \pm 0.12$	$3.42 \pm 1.13$
Loin	3.10±0.10	$3.55 \pm 0.94$
SEM	0.03	0.02
P-value		
Processing method	<0.01	<0.01
Meat cut	0.41	<0.01
PM X MC	0.72	<0.01

 Table 4.22. Effect of Total Bacterial Count and Total Fungal Count in Unam inung

 $^{abc}$  Means on the same column with different superscripts are significantly different (P< 0.05) PM: Processing method; MC: Meat cut

#### 4.3.7 Interaction of effect of total fungal count in Unam inung

The Interaction of effect of total fungal count in *Unam inung* is presented on Table 4.23. The highest value for TFC was seen in smoked X meat cut. The belly, ham, shoulder, and loin were all similar statistically (P > 0.05) (4.70, 4.61, 4.65 log Cfu/g respectively) but different (P < 0.05) from Sundried X meat cut and Raw X meat cut.

Sundried X meat cut ranged from 3.53- 3.62log Cfu/g. Belly, ham, and Shoulder (3.62, 3.56, 3.6 log Cfu/g. Sundried X meat cut was significantly different from Raw X meat cut.

Raw X meat cut recorded the least value of 2.03 log Cfu/g for Shoulder and its highest was observed in Belly which varied (P < 0.05) from shoulder and Belly (2.67log Cfu/g). Ham (2.17 log Cfu/g) also varied (P < 0.05) from Shoulder and Belly, more so, different (P < 0.05) from Loin (2.48 log Cfu/g).

<b>Processing Method</b>	Meat cut	TFC
Raw	Belly	$2.67 \pm 0.07^{d}$
	Ham	$2.17 \pm 0.07^{f}$
	Shoulder	$2.03 \pm 0.12^{\text{g}}$
	Loin	$2.48 \pm 0.03^{e}$
Sundried	Belly	$3.62 \pm 0.01^{b}$
	Ham	$3.56 \pm 0.03^{b}$
	Shoulder	$3.60 \pm 0.02^{b}$
	Loin	$3.53 \pm 0.02^{\circ}$
Smoked	Belly	$4.70 \pm 0.01^{a}$
	Ham	$4.62 \pm 0.06^{a}$
	Shoulder	$4.61 \pm 0.06^{a}$
	Loin	$4.65 \pm 0.06^{a}$
SEM		0.16

 Table 4.23. Interaction effect of Total Fungal Count in Unam inung

<sup>abcd</sup> Means on the same column with different superscripts are significantly different (P < 0.05)

# **CHAPTER FIVE**

# DISCUSSION

# 5.1.1 Assessment of Production, Consumption Pattern and Nutritive Value of *Unam inung*

The Demographic and socio-economic characteristics of consumers (Table 4.1) revealed that although *Unam inung* is typical of the Efiks in Cross River State, it is also relished by non – Cross Riverians resident in Calabar; a factor favoured by non-constraint to pork consumption in southern Nigeria (Amaefule *et al.*, 2009; Ajala *et al.*, 2007).

*Unam inung* consumption cuts across both married and single people. The high consumption frequency by the married may be attributed to the caring culture typical of the efik women who buy *Unam inung* as '*mkpo uyong udua*' (appetizer) for their husbands to keep them while the main meal is being prepared. This high frequency is also evident in the gender of consumers as majority of them were females.

Consumption of *Unam inung* cuts across all ages but majority of the consumers were in the category of 31- 40years which reveals that these age group are the active home-makers who would want to treat their household to the *Unam inung* delicacy while the main meal is being prepared. Moreover, Unam inung is a snack that is relished by the working-class citizenry who also fall within this age bracket.

Calabar is a large city where majority of the populace are Christians and there is no religious taboo on the consumption of pork. Most of the consumers were civil servants probably because civil service is the predominant business of the Cross riverians followed by those into business while few of them were students, hence, the high frequency of civil servants in Calabar. It could be concluded that all the consumers had basic education.

Education is important in our day-to-day life style because it enables consumers to understand the nutritional and safety implications of what they consume. According to Akinnusi (2004), illiteracy and a lack of knowledge about the nutritional composition and health advantages of snails may make acceptance and consumption difficult. This same assertion probably applies to the consumption of *Unam inung*.

The highest percentage (41.78%) of the consumers earned between \$15,000 - \$30,000, 30.82% earned \$30,000 and above, 7.53% earned \$7,500 - \$10,000. 19.86% of the consumers earned no income. They were either students or Children that were sent to buy the product.

The household size in this study revealed that those that had no children (39.73%) consumed the product more than those that have one (27.40%), two (18.49%), three and more (14.38%). This may be because a combination of the product with local snack (*edita iwa*) can satisfy as a meal and it requires little or no further preparation. Out of the 23 males that purchased the product, 21.74\% of them were not married and 78.26\% had one wife each. This reveals that polygamy is low in this study area.

Table 2 reveals that all the consumers had eaten the product before the time of the interview and had had knowledge of it. 91% of them expressed likeness for it while 9% of them were Undecided. They neither liked nor disliked the product. They consumed it because it had been a delicacy in the locality.

The sensory characteristics in Table 3 revealed that 97% of the consumers do not like the salty taste of the product. 86% of the consumers said the product develops odour quickly if not properly handled. Majority said the product was hygienically prepared as they believe that salt will inhibit the growth of micro-organisms.

All the consumers (100%) confirmed that the *Unam inung* goes bad easily if not properly handled due to the fatty nature of the meat which will aid rancidity. Generally, pork (100%) was preferred by all for *Unam inung* since it has been the only meat type processed in this manner.

On the pattern of purchase (Table 4), 54% of the consumers purchased *Unam inung* weekly, 36% of the consumers purchased fortnightly,7% purchased monthly and 2%

purchased daily. The quantity of *Unam inung* purchased by the consumers was to the scale of 1kg as represented by 98% of the consumers while a 1% buys up to 2kg. The product was mostly consumed on the same day of purchased as indicated by 79% while 19% of the consumers consumed it immediately it is bought. It is normally eaten with boiled cassava chips (*Edita Iwa*) as revealed by 97% of the consumers from the study, 2% of the consumers consume it with other kinds of food, only one person prefers to consume it alone. The consumers' preference to consume it with accomplishment may be due to the saltyness of the product.

Majority of the consumers (60%) are willing and ready to buy the product if found in shops probably due to the advancement of technology and the consciousness of consumers for wholesome and well packaged food while 30% are not willing to buy from shop and 10% were indifferent. There is no constraint (religious taboo) to the consumption of the product as revealed by the consumers. This is could be due to the fact that Calabar metropolis is dominated by Christians.

#### 5.1.2 Qualitative evaluation of commercially available *Unam inung*

Microbial load in the samples were considerably low. The Total bacterial count and the Total fungal counts were within the permissible level of microbial standards (6 log cfu/g of sample) in cooked meat products as reported by Jay (1996). The reason for the low count could be attributed to the salt and heat application through the salting and cooking processes. Salt is known to be a preservative because it reduces the water activity of foods. It could be noted that water activity is the amount of unbound water that is available for microbial growth and chemical reaction in a food medium. Salt is able to decrease water activity through the association of sodium and chloride ion with water (Fennema, 1996). Addition of salt to meat also cause microbial cells to lose water through osmosis thereby inhibiting growth (Davidson, 2001). Salt also limits the solubility of oxygen to some microorganisms, deter enzymes activities in the cells and force cells to spend energy to eliminate sodium ions from the cell which also inhibits growth (Shelef and Seiter, 2005) The variations in Total Bacterial and Fungal counts obtained in this research could be attributed to the utensils, equipment, handling and processing methods used by the

processors of this meat. This view corresponds with the findings of Igene *et al.* (1988) which stated that quality variation in suya exist from one processor to another due to unstandardized methods of preparation leading to inconsistent product quality

The presence of *Staphylococcus aureus* may be caused by application of salt to the meat during processing as affirmed by Gilbert and Harrison (2001) who reported that meat preserved with salt encourages the growth of *Staphylococcus aureus*. Boles *et al.* (2000) also stated that *Staphylococcus aureus* requires about 6.5% Sodium Chloride for growth and is usually found in salty meat products. However, contamination of food with *Staphylococcus spp.* are mainly through physical contact as it is a normal flora of the human skin (Gilbert and Harrison 2001).

*Staphylococcus spp.* are part of the normal human flora frequently found in the nose, respiratory tract, and on the skin. The prevalence of *Staphyloccocus epidermidis* may be due to fact that they are transmitted by the carrier (processors) (Cheesbrough, 2000; Mankee, 2003). They are responsible for a number of common infections. *Bacillus spp* are soil microflora, vegetation and food. They are usually implicated in food borne diseases. When ingested, it causes gastrointestinal illness with nausea, vomiting and diarrhea. It is also linked to serious infection in host whose immune system has been compromised to cause septicemia and endophthalmitis which can lead to vision loss (McDowell *et al.*, 2021). *Aerococcus viridans* are airborne present in the environment.

The moisture content and protein of *Unam inung* were similar across the sources. Moisture is a vital component of food such that foods are classified according to their moisture. Food having moisture content above 70% are classed as Perishable, about 50-60% moisture is non-perishable while less than 15% are stable food (Ahmad *et al.*, 2018). High moisture content in foods reduces the storage value of food because it aids the growth of microorganisms which causes spoilage. Moisture also affects colour firmness and aroma of meat.

The variation in Fat content may be attributed to the meat cut used. The fat content may have increased due the concentration of nutrients as a result of loss of moisture. (Fakolade *et al.*, 2008) while the high content of the ash may be as a result of the amount of salt

applied on the samples during salting. Although Torres *et al.* (1994) and Oladejo *et al.* (2011), observed that ash content increases on application of heat.

# 5.2 Quality attributes of *Unam inung* as influenced by different salt levels

Salt is used as a preservative in meat processing. This function is achieved through the dehydration of moisture from the meat tissues through osmosis by lowering the water activity of the meat and making it unfavourable for the growth of microorganism thereby prolonging the storage duration of the meat.

The application of salt on the meat samples reduced the moisture content of the salted pork. The moisture content of pork is about 70%. The treatment with the least concentration of salt had the highest moisture loss as evident in the values obtained for weight loss. From the inclusion of 10% concentration of salt upwards, the moisture content increased and was similar across treatment. This showed that water was pulled out of the meat cells and there was an uptake of salt through osmosis. The meat became saturated with salt. This happened until there was an equilibrium between the meat cells and the environment thereby, causing moisture to be bound to the meat cells.

Various authors (Xargayo *et al.*, 1998, Medynski *et al.*, 2000 and Puolanna *et al.*, 2001) noted that sodium chloride raises the rate at which muscle protein become soluble, boosts the ability of tissues to bind water and makes better the pH of the meat.

The result of this study does not agree with the reduced but similar moisture content at certain salt levels (Ferreira *et al.*, 2013). Solomon *et al.* (1994), also reported a linear decrease in moisture content as the concentration of salt increased.

The linear increase in ash content could be attributed to the amount of salt added to the meat samples because the total amount of minerals present in a food are summed up as ash. This agrees with the works of Ferreira *et al.*, 2013, Solomon *et al.*, 1994.

The increase in pH upon salting on the first day could be due to the application of NaCl. Ogunsola and Omojola (2003) reported an increase in pH with increasing salt levels for freshly salted meat. Sodium chloride raises the ionic concentration of the meat matrix and enhances water binding among other functional characteristics. The result reported herein tallies with that of Alonge, (1984) for smoked meat.

The decrease in pH after three days of storage may be attributed to the loss of phosphate during salting due to osmotic dehydration by NaCl. This was also observed by Uguz *et al.* (2010) in his Pastirma comparison studies.

On the 5<sup>th</sup> day, the increase in the pH of the least concentration (5%) of salt may be triggered by microbial and enzymatic activities on meat. (Ahmad *et al.*, 2005; Mahmoud *et al.*, 2006; Virgili *et al.*, 2007). However, high pH is indicative of poor quality of a product.

Application of microbiological methods is the only way to obtain information about the hygiene status of places, equipment and food. A suitable means of achieving this is by assessing the product after processing against contamination and its storage quality (Zhou *et al.*, 2011).

The Total viable count signifies the hygienic quality of a product. The microbial load in the Control sample indicates cross contamination during slaughtering of the animal. This was carried out on the floor. The reduction of the TVC of the salted pork could linked to the antimicrobial activity of salt as it inhibited the growth of microbes. Salt is known to have a bacteriostatic effect in meat products, and some greater concentrations will slow or even halt bacterial growth. The influence of the different concentrations on the microbiological quality showed that 15% concentration of Salt inhibited microbes best compared to 20% sample. This may be as a result of the proliferation of halophilic microbes in the 20% samples. Pathogenic microbes were not found in Salt treated samples mainly due to the bacteriostatic effect of Salt. Salt pulls out water from bacterial cell through the process of osmosis until there is an equilibrium with salt concentration of the cell. Water now becomes unavailable to the microbes and their growth is inhibited (Hajmeer *et al.*, 1999), thereby leading to extension of shelf life of the product (Marsdeen, 1980).

Oxidative degradation of lipids in food from muscles are popularly determined using TBAR analysis as asserted by Ockerman (1981). The degree of oxidation of *Unam inung* was measured by TBARs methods. TBARs values increased significantly (p<0.05) as the

salt concentration increased. Two authors also recorded increased malonaldehyde content in Lacón and Hams samples respectively after salting (Garrido *et al.*, 2009 and Melgar *et al.*, 1990). The higher values of the TBARs in salt treated samples on the last day of the experiment exhibited the pro-oxidant nature of salt as it is known to quickens lipid oxidation.

# 5.3 Evaluation of the quality of Smoked and Sundried Unam inung

The method of processing plays a vital role on the nutrient contents of fish (meat) and substantial variation in nutrients have earlier been reported by Akinneye *et al.* (2010).

These processing methods have different applications, techniques and significant influence on the chemical, physical and nutritional composition of processed product. This is because heating, freezing and exposure to high concentration of salt lead to chemical and physical changes.

The moisture content in smoked and sundried *Unam inung* obtained in this study was within the range of 29.43- 46.29%. This slightly varied with the moisture range of 30 - 40 % reported by Fakolade and Omojola (2008) for beef and Camel Kundi. It was higher than the moisture content recorded for kilishi processed by sun-drying and oven drying (Egbunike and Okubanjo, 1999) and also higher than the range of  $17.40 \pm 1.15\%$  in Istrian sausage to  $42.80\pm 0.56\%$  in Kraški pršut (Pleadin *et al.*, 2016). However, the Smoked meat cuts were drier than the sundried meat cuts apparently as a result of dehydration leading to moisture loss during smoking. This could have been caused by heat and temperature of the smoking process as Gomez *et* al. (2020) stated that smoking affected the meat according to the duration of smoking

The result of the Ash content agrees with the works of Ferreira *et al.* (2013) and Solomon (1994) who reported increases in ash content as the salt level increased. The afore mentioned authors reported that ash increased with heat application. The high ash content of smoked *Unam inung* may be attributed to wood ash particulates that deposited on the meat during smoking and the dry salt that was applied on the meat during sun drying and also the uptake of NaCl by the meat samples all through salting. Curing generally increases the ash content of meat.

The fat content obtained from the present study was similar to the fat content analysed by Ferreira *et al.* (2006) for Alheria as it was within the same range of 9.41 - 25.84%. The high fat contents may be attributable to reduction of moisture during salting and smoking which led to nutrient concentration. This tallies with the results of Żmijewski *et al.* (2006) who stated that fat inversely correlated with water as is usually seen in numerous fish species.

Protein content of sundried and smoked *Unam inung* ranged between  $20.17 \pm 0.20 - 31.33 \pm 0.18\%$  and were higher than the content of Raw *Unam inung*. This showed that the processing methods improved the protein content of *Unam inung*. Protein is regarded as the most important constituent in nutrition and processing because it describes the quality of raw meat and how appropriate it is in processing (Heinz and Hautzinger, 2007). The finished processed meat is judged by its protein content. Pleadin *et al.* (2016), stated that protein content of traditional meat products of Croatia and Slovenia to be within the range of  $20.93 \pm 0.21\% - 36.38 \pm 0.46\%$ . He also noted that meat products with total proteins greater than 20% are of high quality (Pleadin *et al.*, 2016).

On the contrary, the protein content of *Unam inung* in this study were lower than the protein content observed by Soniran and Okubanjo (2002) in pork loin roast cooked to three internal temperatures at 65°C, 75°C and 85°C respectively.

The result of this study on proximate composition is in consonance with the results that stated that moisture content reduced while other nutrients such as ash, crude protein and lipid content were improved during smoking of Matrinxa fillet (Franco *et al.*, 2010). Processing methods therefore causes variation in nutrient content of meat as asserted by Türkkan *et al.*, (2008).

The processing method did not influence the histology of *Unam inung* rather the different meat cuts exhibited some variation. The interaction effect of the processing method and the meat cuts was evident in length, width and volume of the meat cells. Sundried loin was the shortest in length and the narrowest in width and also had the least volume. The Sundried Shoulder cut had the longest fibre and the widest width but the Sundried Ham had the highest volume. All other cuts were similar in their attributes.

The histologic section of the raw muscle revealed that it was not affected while that of the Sundried and smoked samples were slightly affected.

On the whole, processing methods improves the storage stability of meat and its derivatives as retorted by Mirsha *et al.* (2017). The processing methods vividly influenced the total bacterial count of *Unam inung* by reducing the microbial load. The lower Total Bacterial count observed in the Sundried and the Smoked *Unam inung* compared to the raw samples may be attributed to the application of Salt and heat through Sun drying and Smoking. Lawrie (1991), stated that there is a reduction in the growth of microorganisms and improvement of keeping quality whenever meat is subjected to curing and smoking. Evaporation of moisture occurring from both methods led to reduction in the quantity of water available for microbes to grow and multiply. Ikeme (1990) also, stated that most of the compounds in wood smoke exhibit either bacteriostatic or bactericidal properties; it is believed that formaldehyde (a carbonyl compound) accounts for most of the preservative action of smoke. Smoke compounds are efficient at retarding microbial spoilage due to the production of chemical and physical barrier against microorganisms through the deposition of resins occurring from condensation of formaldehyde and phenol, surface drying and coagulation of protein.

The high TBC in the raw samples may have been through contamination of the meat during slaughtering with equipment used and contact with the environment. Carcass dressing is usually carried out on the floor which yields poorly handled meat. Transportation and the way the meat is handled during sale also contribute to contamination of the carcass.

The meat cuts on the other hand did not vary in their Total Bacterial count. The interaction effect of the processing methods and meat cuts was also not significant. The levels of the processing methods were not dependent on the meat cuts.

The processing method and the meat cuts affected the total fungal counts of *Unam inung*. The fungal effects on meat cut depended on the levels of the processing method. The smoked meat cuts had the highest fungal counts followed by the sundried meat cuts and the least was seen in the raw meat cuts. The reason for this may be because fungi are known to thrive on drier substances.

#### **CHAPTER SIX**

#### SUMMARY, CONCLUSION AND RECOMMENDATIONS

## 6.1 Summary of findings

- 1. Out of the total respondents, about 73.97% of the consumers were from Cross river state, 84.25% were females and 60% were married
- 2. Majority of the consumers earned N15,000 –N30,000 and 91.1% expressed their likeness for the product and 93.84% said the product was hygienically prepared
- 3. *Unam inung* was frequently purchased at weekly basis by 54.11% of the consumers and 98.63% of the consumers bought 1lb of the product
- 4. About 79.45% respondents consumed the product on the same day of purchase and the main accompaniment of consumption of *Unam inung* is *edita iwa* (97.26%)
- 5. The total bacterial and fungal counts of the commercially available *Unam inung* were within the acceptable limit of < 7log cfu/g
- 6. The moisture and crude protein of the commercially available *Unam inung* ranged from 46.67 47.02% and 21.87- 23.05%, respectively
- 7. The moisture content of *Unam inung* was high in 10%, 15%, and 20% salt treated samples compared to that 5%
- 8. The crude protein was higher at 5% treatment compared to 10%, 15%, and 20%
- 9. On the eighth day of the experiment, the samples with 5% salt concentration had the highest weight loss

- 10. The pH of the salt treated samples increased compared to control
- 11. Raw ham had the highest moisture content compared to others
- 12. Smoked ham and shoulder had the highest CP compared to others
- 13. Sundried belly had the highest fat content compared to others but raw shoulder and loin were similar and had the least fat content
- 14. Total fungi count was higher among the smoked meat cuts compared to the sundried and raw meat cuts

# 6.2 CONCLUSION

This research assessed the quality attributes of *Unam inung*, a salted pork product in Calabar, Nigeria. Three studies were carried out for the assessment of this product.

Study one was done in two phases, the first phase involved the assessment of Consumption Pattern and Nutritive value of *Unam inung*. It was deduced from the study that majority of the consumers were married women from Cross river state who are aware of the product and had been consuming it. Even though they liked the product, they complained it is usually salty and preferred it to be made from pork. These consumers were high income earners who expressed likeness for the product and attested that it was hygienically prepared. Most of the consumers bought *Unam inung* to the tune of 1kg every week but usually consume the product on the same day of purchase with edita iwa as an accompaniment. They are willing to buy it from shops if made available. There exists an important relationship between a level of education of the consumers to the frequency frequency of purchase, time of consumption, readiness to buy from shops, likeness, odour and hygienic quality of the product.

In the second phase of the study, Qualitative evaluation of commercially available *Unam inung* was carried out. The commercially available *Unam inung* was high in protein and ash content and the microbial load was within the permissible recommended limit of  $< 7\log$  cfu/g of cooked meat.

In Study two, the quality attributes of *Unam inung* as influenced by different salt levels were assessed. There was an outright spoilage of the control samples without salt treatment. Moisture content was least in the sample with least concentration of salt and increased similarly in samples with higher concentrations of salt. There was a linear increase in ash content as the salt concentration increased. Fat and Crude protein content also increased in the salt treated samples. Lipid oxidation increased with increased salt concentration while 15% salt concentration produced *Unam inung* the least microbial load.

Lastly, in Study three, the quality attributes of differently processed *Unam inung* was evaluated. The processing methods (Sun drying and Smoking) used, reduced moisture and increased the other nutrient components of *Unam inung*. The microstructure of the meat was slightly affected by the processing methods while the microbial load was also reduced

# 6.3 **Recommendations**

- The application of table salt by dry rubbing on the pork preserved the meat and also reduced microbial growth on the meat, therefore researches into other preservative methods should be carried out.
- The production of *Unam inung* is still in the traditional phase, researches leading to the application of modern technology in its production should be encourage to upgrade it.
- 3) The processing methods enhanced the shelf life of the pork and also reduced microbial load therefore other forms of drying like oven drying should be utilized to enhance safety of the product.
- 4) The consumers of *Unam inung* are willing to purchase it from shops if made available, hence, appropriate packaging for convenience should be researched.
- 5) There is demand for *Unam inung*, and this adds up to the variety of meat products in Nigeria, therefore, production of *Unam inung* should be encouraged by provision of loans, raw material and processing facilities to the processors.

# 6.4 Contributions to knowledge

- 1. Incorporation of 15% dry salt into pork inhibited the growth of micro-organisms in *Unam inung* thereby promoting the keeping quality of the product.
- 2. The advancement of technology and the consciousness of consumers for wholesome and well package product could increase the willingness and readiness of *Unam inung* in shops and supermarket.
- 3. Processing methods such as sun-drying and smoking reduced moisture and increased the nutrient composition in *Unam inung*
- 4. Smoking as an alternative method of processing enhanced the qualities of *Unam inung* and can be used during the rainy seasons for its production

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# APPENDIX

#### UNIVERSITY OF IBADAN DEPARTMENT OF ANIMAL SCIENCE

# **CONSUMER'S QUESTIONAIRE**

This questionnaire seeks to select your responses on the acceptability of *Unam inung* (a cured meat product) for consumption. Kindly assist in providing accurate information on the question raised by ticking the appropriate answers.

# **SECTION A**

PERSONAL DATA

- 1) STATE:
- 2) TRIBE: (a)Yoruba (b) Igbo (c) Hausa (d) others
- 3) GENDER: (a) Male (b) Female
- 4) RELIGION: (a) Christianity (b) Islam (c) Traditional
- 5) OCCUPATION: (a) Civil Servant (b) Business person (c)Trader
- 6) LEVEL OF EDUCATION: (a) Primary (b) Secondary (c) Tertiary (d) No formal
- 7) INCOME PER MONTH (a) N7,500 N10,000 (b) N15,000 30,000 (c) N30,000 and above
- 8) HOUSE HOLD COMPOSITION:
- i. Number of wives: (i) None (ii) One (iii) Two (iv) Three and above
- ii. Number of children: (i) None (ii) One (iii) Two (iv)Three and above

#### **SECTION B**

- 9) Have you ever eaten *Unam inung*? (a) Yes (b) No
- 10) Do you like it? (a) Yes (b) No
- If No, please answer No. 11, if Yes please skip No. 11.
- 11) Please rank, using numbers (1-5) in order of importance, each of the following reasons for disliking *Unam inung*.

I do not like the taste of <i>Unam inung</i> (salty)	
I do not like the smell of <i>Unam inung</i>	

It develops odour quickly	
It gets spoint easily	

- 12) Which meat type do you prefer for *Unam inung*? (a) Pork (b) Beef (c) mutton
- 13) How frequently do you buy *Unam inung*? (a) daily (b) weekly (c) fortnightly (d) monthly
- 14) How much of *Unam inung* do you purchase in a month? (a)1 kg (b) 2kg (c) 4kg (d) above 4kg
- 15) When do you consume the product? (a) immediately (b) same day (c)days after purchase
- 16) Do you consume *Unam inung* alone or with other snacks? (a) Alone (b) with other snacks
- 17) Would you readily buy Unam inung if available in the shops? (a) Yes (b) No
- 18) Do you have any cultural belief behind purchasing Unam inung? (a) Yes (b) No