PHYSICAL, CHEMICAL AND QUALITY INDICES OF THREE IMPORTED FROZEN MARINE FISH SPECIES SOLD IN ZARIA, NIGERIA

BY

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DECLARATION

I, Abubakar Rabiat hereby declare that this dissertation was written by me and that it is a record of my own research work. This work has not been presented in any previous application for a higher degree. All cited works were adequately acknowledged through referencing.

_____	_____	_____
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CERTIFICATION

This dissertation titled "Physical and Chemical Quality Indices of Some Imported Frozen Marine Fishes Sold in Zaria, Nigeria" meets the regulation governing the award of the degree of Master of Science in the Department of Biological Sciences, Faculty of Science, Ahmadu Bello University, Zaria, and is approved for its contribution to knowledge and literary presentation.

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ABSTRACT

The physical and chemical quality indices of imported frozen marine fishes *Sardina pilchardus*, *Trachurus trachurus*, *Clupea harengus* sold at Sabongari Zaria were determined according to standard procedures. For the physical properties, the electrical properties, moisture content and pH of the samples evaluated. For *Sardina pilchardus* were 2.14 ± 0.58, 60.1 ± 6.30 and 7.04±0.10, respectively and for *Trachurus trachurus* the values were 2.92±0.53, 58.25±0.05 and 7.15 ± 0.04. *Clupea harengus*, they were 2.02 ± 0.323, 55.15 ± 0.45 and 6.98 ± 0.1, respectively. For chemical properties, the thiobarbituric Acid (TBA), Freefattyacid (FFA), Extractable protein nitrogen (EPN), formaldehyde, indole, and ash content values for *Sardina pilchadus* were 0.5 ± 0, 2.35 ± 0.25, 7.4 ± 5.3.80, 1.7 ± 0.35, 1.55 ± 0.05 and 1.35 ± 0.15, respectively, for *Trachurus trachurus*, the values were 0.45±0.05, 1.15±0.919, 3.1± 0.35, 1.4±0.10, 1.85±0.05 and 1.25±0.05, respectively and for *Clupea harengus*, the values were 0.45±0.071, 1.9±0.20, 5.2±2.10, 1.3±0.15, 1.9±0 and 1.13±0.43, respectively. Freefattyacid and Extractable protein nitrogen were highest in *Sardina pilchardus* (2.35) and (7.4) EPN respectively. In addition, quality index method was conducted by 24 panelists, *Sardina pilchardus* and *Clupea harengus* were graded as having good quality, *Trachurus trachurus* was graded to have satisfactory quality. All the physico-chemical indices indicated that the examined frozen samples were within permissible limit values set by National regulatory bodies.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 Fish Quality

Fish form the biggest group in the animal kingdom that is used for the production of animal-based food, of the more than 30,000 species. Only about 1000 of these species are commercially fished and used for food production (Zsuzanna *et al.*, 2011). According to FAO (2007), fish contributed more than 60% of the world supply of protein, especially in the developing countries. Consumer's demand for high quality, safe, and healthy food is increasing on a global basis (Sen, 2005). Consumer studies appear to indicate that quality is still the key buying cue for fish purchases (Alasalvar *et al*., 2011), and in order to secure the food safety, it is important to keep the quality of fishes at a high level in each link of the whole complex chain from catch to consumer.

FAO (2014) defined quality as the totality of features and characteristics of a product or service that bear on its ability to satisfy stated or implied needs. Fish quality is a complex concept involving a whole range of factors which for the consumer includes safety, nutritional quality, availability, convenience and integrity, fairness, eating quality and in obvious physical attributes of the species, size and product type (Abbas *et al*., 2008). Quality assessment of fishes has more to do with the determination of its shelf life or storage life, which is the amount of time that sea foods remain palatable. Different species of fishes have different shelf life which also varies depending on the oil levels, catch area, season, and duration of *rigor motis*, intrinsic conditions of the fish and how it was captured and handled (Huss, 1995). The quality of fish and fishery has become a major concern in the fish industry all over the world (Huss *et al.*, 2003).

Methods of processing and storage and its duration, generally causes depletion in essential nutrients and minerals in frozen and stored fin fishes (Arannilewa *et al*., 2005). Fish freshness is fundamental to fish quality. The state of freshness can be described by variety of definite properties of fish which is assessible by various indices (Bremner and Sakaquchi, 2000). Fish being one of the exceptionally perishable foods and as a result of globalization of food trade, fish products tends to be more perishable and more susceptible to rejection due to poor quality especially if the initial raw materials are of poor quality despite the technological developments in fish production (Huss *et al*., 2004).

Several methods are used to determine the quality of fish. These can be classified into sensory and instrumental methods (physical, chemical and microbiological methods). Physical methods involve the measurement of pH, texture or electrical properties (Chebet, 2010). Chemical methods rely on measurement of metabolite(s) produced during fish storage or distribution to obtain a quantitative fish index, chemical methods are rapid, quantitative and reproducible. The aim of microbiological examinations of fish products is to evaluate the presence of bacteria or organisms of public health significance and to give an impression of the hygienic quality of the fish including temperature abuse and hygiene during handling and processing (Huss 1995).

1.2 Statement of Research Problem

Imported frozen marine fishes flow through the marketing chain, the value addition can be influenced by varieties of factors like transportation and storage processes which can affect the quality of imported marine frozen fish (Bo, 2011). Duration of transportation from frozen state to Nigerian market may result in spoilage. There is dearth information on the quality of the frozen fish sold in Sabongari, Zaria.

1.3 Justification

Fish is a highly regarded food commodity by man. It is the cheapest source of animal protein, fish products are diverse and readily available and widely accepted across cultural or religion boundaries. According to Olokor *et al.* (2007), fish is one of the most nutritious foods. It contributes a considerable proportion of the protein diet in the developing nations particularly in Africa. As source of protein, the importance of fish in reducing protein deficiency in the country cannot be overemphasized (Ukoha, 1998). Adeyeye (2002) and Zalluoa *et al*. (2007) indicated that fish is rich in protein, contains low cholesterol level and high percentage of polyunsaturated fatty acids, liposoluble vitamins and essential minerals.

The health benefits related to seafood consumption especially fish have been well documented. Omega-3 fatty acid has proved to have protective effects in preventing coronary heart disease, reducing arrhythmias and thrombosis (Kinsella *et al*. 1990), lowering plasma triglycerides level (Ismail, 2005) and reducing blood clotting tendency. Brain and eye development have also been attributed to Docosa Hexaenoic Acid (DHA) in fish lipids.

Sardina pilchardus, *Trachurus trachurus*, and *Clupea harengus* are the major species of imported frozen fish sold in Zaria cutting across various economic classes. As a major fish product traded in Zaria, base line data establishing their qualities and safety is paramount.

1.4 Aim

The aim of this study is to evaluate the quality of some imported frozen marine fishes *Sardina pilchardus* (Sardine), *Trachurus trachurus (*Mackerel), *Clupea harengus* (Herring).

1.5 Objectives

The specific objectives are to determine the:

1. Physical indices: Electrical properties, pH and moisture content dictating the quality of some imported frozen marine fishes sold in Zaria

2. Chemical indices: Thiobarbituric, free fatty acid, extractable protein nitrogen, formaldehyde, indole, and ash content dictating the quality of some imported frozen marine fishes sold in Zaria

3. Quality of some imported frozen marine fishes sold in Zaria using quality index method, evaluating the parameters such as general appearance, eyes, gills, smell, shape and color of all samples

1.6 Hypotheses

1. Physical methods of assessing fish do not dictate significantly the quality of imported frozen marine fishes.

2. Chemical methods of assessing fish do not dictate significantly the quality of imported frozen marine fishes.

3. Quality index method of assessing fish does not dictate significantly the quality of imported frozen marine fishes.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Fish Demand and Supply

The Nigerian fishery industry is made up of three sub-sectors; the artisanal, the industrial and aquaculture. The major cultured species include tilapia, carp and catfish which have a high growth rate and are highly appreciated in Nigeria (Livestock and Aquaculture Watch, 2007). There is an increasing awareness for aquaculture in Nigeria; this has led to an increase in the turnout of the aquaculture industry. This boom in the industry has led to an increase in demand for *Clarias gariepinus*, which is the major farmed species. Yet the importation of frozen fish plays a key role in bridging the gap between the demand for and supply of fish from domestic sources. The major species imported are *Sardinnella eba* (Sardines), *Mackerel spp* (Mackerel) and Hake ("Panla"/stock fish) (Teslim, 2010).

Based on the economic tenets of demand and supply, it is known that several factors influence the consumers choice or demand for a particular product (e.g. color, size, price, trade restrictions e.t.c.). These factors help to describe inter product price relationship, showing the rate of increase or decrease in price or supply or demand of a product based on the fluctuations in the demand for another product. The demand for fish is estimated at 511,000 tones and the remaining is taken care of by importation and aquaculture. Nigeria is one of the largest importers of fish with official records indicating an average of five hundred and ninety eight thousand nine hundred and twenty nine tones (598,929) in 2014 estimated by Norwegian Seafood Council (Alicia 2015).

2.2 Fish Importation to Nigeria

Frozen seafood imports are widely is widely distributed in the country, through a network of privately owned cold stores located in major cities. The sea food is available frozen in the city markets, and also smoked for rural village markets. Sub- wholesalers always buy from whole-sellers to break bulk further except for promotional sales. The whole seller is the nucleus of distribution channels in Nigeria s retail sector and serves linearly every member in the distribution channel (Vaswani 2006). Nigeria has been a major importer of fishery product given its ever increasing consumption and lack of sufficient internal sources – market size – 1 million tones (Vaswnani 2006). Nigeria has to import about Seven hundred thousand Meter- Tone - second (700,000MTs) of fish valued about US $500 million annually, (Naoki *et al*., 2013). Nigeria is currently importing an estimated 1.9 million metric tones of fish valued at 125 billion per annum, the minister of Agriculture and rural development , Dr. Akinwumi has disclosed (Austin 2015).

Nigeria's fish supply came from four major sources in order of importance, namely; Importation, inland estuaries and coastal artisanal fishery, industrial trawl fishery and aquaculture (Naoki *et al*., 2013). The composition of imports is largely mackerels, cod, haddock, hakes, herrings and croakers caught off the coasts of Eastern Central Atlantic countries of Senegal and Mauritania and from the North Sea (Naoki *et al.,* 2013).

2.3 Effect of Storage Process on Frozen fish

Preservation by chilling and freezing are widely accepted by consumers even though other methods for preservation such as salting, drying, canning and smoking are applied (Mehta *et al*., 2011). The colder the fish, the better storing process, because it will help to minimize activities

of microbial or enzymatic spoilage (Tawari and Abowei, 2011). It is necessary that a sufficiently cold surrounding must be supplied to effect this change (Davies, 2006). Biochemical changes are identified to take place, as for example changes in the lipid and protein fractions during chilled and frozen storage. By enzymatic action, sugar is formed with the amino acids and forms the brownish or yellowish color (Laila *et al*., 2013). It has great impact on the nutritious value of fish and the health of the consumers for deterioration of fish during refrigerator storage (Gandotra *et al*., 2012). Oxidative deterioration of lipid can directly affect quality characteristics such as color, flavor, texture, nutritive value and safety (Wang, 2009). During frozen storage, enzymatic and non enzymatic rancidity highly influence the shelf life of marine product due to unsaturated lipid composition and the presence of pro oxidation molecules in their muscle (Aubourg *et al*., 2004).

2.4 Effect of Transportation on the Quality of Frozen Fish

The quality of frozen fish depends fundamentally on the quality of raw materials used and product manufacture, but can be jeopardized by failure to maintain product temperature at a suitable low level in any part of the cold chain including storage, transport, distribution and display in retail stores (David *et al.*, 2008). The rate of spoilage depends on handling during processing, acidity level, species of fish weather, mode of storage and temperature during transportation. Chemical breakdown of protein, fat and water contents contributes to quick spoilage of fish (Daramola *et al.*, 2007).

The fact that there are so many types of food with so many containers, temperature and handling requirements and so many modes of transportation available to the modern food company, it isn't difficult to see that independent of the mode of transportation, food ingredients are susceptible to

abuse and or contamination during transportation and storage (Larry, 2003). Transportation of products is often times done on open and non-refrigerated trucks which expose products to unfavorable storage condition (Vaswani, 2014). The impact of transportation mode of frozen fish has a tremendous effect on their resulting quality. Deviations in optimal storage temperature inevitably lead to decreasing product quality and shortening of shelf life, resulting in a depreciated product value.

2.5 Qualitative Changes that Occurs in Frozen Fish

Fish tissues possess high nutritional value and are therefore a recommended dietary component (Ausbourg *et al*. 2004). Lowering fish quality during frozen storage has been attributed to undesirable changes associated with lipid and proteins (Huidoboro and Tejada, 2004). Changes in lipid occur through hydrolysis and oxidation mechanism. Fish muscle proteins can undergo denaturation during frozen storage due to formation and accretion of ice crystals resulting in dehydration (Simeoniduo *et al*. 1997). Protein aggregation in frozen fish depends on storage temperature, temperature fluctuation, moisture change, storage time and enzymatic degradation (Bauchart *et al*., 2007).

Although freezing is an effective method of preserving foods, some deterioration in frozen food quality occurs during storage. The extent of quality loss depends on many factors, including the rate of freezing and thawing, storage temperature, temperature fluctuation, transportation, retail display and consumption (Boonsumrej *et al.,* 2007). In fatty species of fish such as herring (Clupeidae), mackerel (Scombridae) and Salmon (Salmonidae) the most serious cause of deterioration is fatty oxidation. Fish lipids contain high levels of polyunsaturated fatty acids, which are susceptible to attack by molecular oxygen, when lipid oxidation typically results in formation of aldehydes, alkyl radicals and semialdehydes (Chen *et al*., 2008). The chemical tests

for detecting oxidation and quantifying the extent of oxidation are based upon these changes. Thus, malonidialdehyde (MDA), a major degradation product of lipids oxidation and the most frequently method used for (MDA) determination is estimation of thiobarbituric acid reactive substances (TBARS) (Cordis *et al*., 1995). However, TBARS values go up during the initial phase of propagation stage and down in the termination stage of oxidation, the lipid oxidation leads also to objectionable changes of flavors, therefore sensory tests provide useful information related to quality loss (Mendes *et al.*, 2009).

2.6 Physical Methods of Assessing Fish Quality

Sensory changes occur in appearance, odor, taste and texture during storage of fish (Odoli, 2006). The electrical properties of skin and muscle change systematically after death and can be used as the basis of quality assessment of fish with the aid of an instrument (FAO, 2001). According to Odoli (2006), the pH of fish can give some valuable information about its condition. Measurements are carried out using a pH meter by placing the electrode either directly into the flesh or into a suspension of fish flesh in distilled water. The pH is considered as one of the important quality tests for evaluating the meat quality (Flecher, 1999). As a result, the pH of fish muscles remains high after death (>6.0) favoring microbial growth and enzymatic activities (Church, 1998). Electrical properties of muscle changes considerable by post mortem. The tissue typically goes through a rigor phase a few hours after having been sacrificed, and the disintegration of the tissue then develops gradually. Knowledge about the electrical changes during these stages may prove useful in different areas such as fish and meat. The properties are also useful in the detection of processing conditions or the quality of foods (Organ *et al.,* 1996). The variation in moisture content was due to the time taken in freezing the fish and the temperature of fish from which heat is steadily removed from the freezing point of water. The

temperature then remains almost stationary until most of the water turn to ice, when it again begins to fall rapidly as the frozen flesh is further cooled, (Magawata and Abdulmumin, 2015).

2.7 Chemical Methods of Assessing Fish Quality

The evaluation of food using chemical methods are considered to be more objective than sensory methods especially when it is done accurately using appropriate methods (Huss, 1995). Chemical methods of food evaluation are normally used to indirectly predict the level of a sensory attribute, which allows for immediate determination of freshness and to use chemical methods to serve this purpose, well set, quantified and standardized tolerance levels of chemical spoilage indicators need to be established (Huss, 1995).

Chemical methods rely on the measurement of metabolite(s) produced during fish storage or distribution to obtain a quantitative fish index. Chemical methods are rapid, quantitative and reproducible (FAO, 2001). Oily fish like herring and mackerel can become rancid; this is due to the reaction of the oil with oxygen in the air to create unpleasant odors and flavors. Oxidation occurs quite solely in iced fish, and the products of bacterial growth can render fish inedible before oxidation contributes much to the odors and flavors (FAO, 2001).

In frozen fish, bacterial action is reduced to negligible levels but oxidation of the oil, especially in oily fish, will continue during storage and will lead to a loss of eating quality. Some of the proteins in fish undergo changes, not fully understood during long periods of frozing, which lead to undesirable toughening of the flesh (FAO, 2001). In later stages of oxidation, secondary oxidation products will usually be present and therefore indicative of a history of autoxidation. These products comprise aldehydes, ketones, short chain fatty acids and others, many of which have unpleasant odors and flavors and which in combination, yield the fishy and rancid character associated with oxidized fish lipid. Some of the aldehydic secondary oxidation products react

with thiobarbituric acid, forming a reddish colored product that can be determined spectrophotometrically.

The chemical spoilage associated with fish during storage is mainly due to fish lipid degradation (auto- oxidation). In general, fish have high degree of poly unsaturated lipids than other food commodities (Huss, 1995). Other methods such as peroxide value and Thiobarbituric Acid also contribute to the chemical methods that are used to measure rancidity in fish and fish products generally. Free fatty acid content has been used to establish the grade of deterioration (DeKoning and Mol, 1991). Lipolysis of fats, lead to production of free fatty acids during storage. FFA are known to undergo further oxidation to produce low molecular weight compounds that are responsible for off-flavor and undesirable taste of fish products (Refsgaard *et al.* 2000). Also FFA has great influence on protein denaturation and texture deterioration by interaction with protein (Ladosa *et al*. 2004). Indole is an aromatic heterocyclic organic compound; it is widely distributed in natural environment and can be produced by varieties of bacteria as a degradation product of the amino acid tryptophan. It occurs naturally in human faeces and as an intense faecal odour.

2.8 Quality Index Method

European fisheries research institutes have developed a tool, by which sensory assessment is performed in a systematic way with an objective quality assessment method (Hyldig *et al*., 2007). The quality index method (Q1M), as it has come to be known, is a systematic, objective guideline that simplifies the quality-assessment process. It originated from Tasmania, during the late 1970s and early 1980s, by Tasmanian Food Research Unit (TFRU) of the Commonwealth Scientific and Industrial Research Organization (CSIRO), but most QIM schemes have been

developed in Europe, where the method is widely applied (Nielsen, 2005). QIM uses a practical rating system in which the fish is inspected and demerit points are recorded (Sen, 2005).

This approach was derived from understanding that during the storage of fish, changes occur that are readily detectable and often measurable. This is also in keeping with the fact that the vast majority of chemical, biochemical and microbiological tests on fish products start from zero, or a low value and increase with both temperature and period of storage (Hyldig *et al*., 2010). The use of a scaling method establishes robust data, reflecting the different quality levels of the fish in a simple and well -documented way.

The QIM is based on significant sensory quality parameters using well-defined characteristic changes of outer appearance attributed for raw fish and a score system from 0 to 3 demerit points (Hyldig and Green-Petersen, 2004; Martinsdottir, 2002). The attributes to be evaluated when applying, or developing the QIM are the skin, eyes, gills, and texture. The color of the gills, and of some species, the odor and the mucus of the skin are also evaluated (Martinsdottir, 2002). Each attribute is scored from 0 to 3 by novices, or experienced assessors, with low scores indicating the best quality.

The score allotted to each criterion is such that no single criterion could dominate and that the score values are easily to judged (Hyldig *et al*., 2010). The sum of all the attributes is called demerit points or QIM index points. This value increases linearly with the storage time on ice of a given fish. The score can be used to judge the handling and storage procedures, efficiencies of personnel, and duration of transport, and it is particularly useful in troubleshooting (Hyldig *et al.*, 2007). The main advantages in using QIM are that it is non-destructive and it takes into account the differences between species (Green-Petersen, 2010).

According to Nielsen (2005), QIMs are easier to use than some sensory methods and do not require any equipment other than the senses. All the quality descriptors are well defined, and most of the schemes include thorough instructions with illustrations. As a result, limited training is needed to achieve accurate results. Martinsdottir (2002) reported that the Quality Index increased linearly with the storage time on ice. The information was well suited to use in the production management. Quality index method is well suited to teach the inexperienced people to evaluate the fish, train the panelists and monitor the performance of the panelist since it is an objective method and includes instructions and easily understood illustrational materials.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Source of Fish Samples

The samples of imported frozen marine fishes were collected from the frozen fish depot in Sabongari, Zaria. Samples consist of three species namely; *Sardinna pilchardus, Trachurus trachurus* and *Clupea harengus* and for each species a total of thirty samples were purchased. Collection was carried out fortnightly-over a period of six weeks, to create replication. The samples were purchased and transported in ice-boxes to the Fisheries laboratory, Department of Biological Sciences, for morphometric measurements before analysis at the Analytical laboratory of Department of Chemistry, Ahmadu Bello University, Zaria.

3.2 Identification of Fish Samples

The samples of the imported frozen marine fishes were identified following the keys of Bally (2014). The fish samples were identified as: sardine, *Sardina pilchadus,* Walbaum 1972 (Plate I), mackerel, *Trachurus trachurus*, Linnaeus 1758 (Plate II) and herring, *Clupea harengus,* Linnaueus 1758 (Plate III).

Samples were rinsed under running tap water in preparation for analysis. Morphometrics of the identified samples, *Sardina pilchardus, Trachurus trachurus* and *Clupea harengus*, were determined. The weight in grams with the aid of measuring scale and the total length in centimeter with the aid of measuring board.



Plate 1: Lateral view of *Sardina pilchardus*



Plate II: Lateral view of *Trachurus trachurus*



Plate III: Lateral view of *Clupea harengus*

3.3 Physical Parameters for Assessing Fish Quality

3.3.1 Determination of Electrical Properties

A 5 gram portion of each fish sample was homogenized with 50ml of distilled water blended for 30seconds and the electrical property was determined using electrical conductivity meter, HACH Session 5 (Chebet, 2010).

3.3.2 Determination of Moisture Content

Moisture content was determined by oven drying method as described by FAO (2001). A suitable container of stainless steel with a loose fitting lid was weighed accurately after drying at 100℃ and cooled. Five g of the prepared sample was spread evenly over the bottom of the container, after putting the lid, the weight was determined again. The container was then placed in an oven at 101℃, the lid was removed and left for 24 hours, and the container was transferred to a dessicator after replacing the lid and allowed to cool at ambient temperature, the weight of the closed container was taken.

$$\text{Moisture content (\%)} = \frac{weight\ of\ wet\ materials - weight\ of\ dried\ materials}{weight\ of\ wet\ materials}$$

(FAO, 2001)

3.3.3. Determination of pH

The pH value of the fish was measured according to the method described by Nuno *et al.* (2013). A 5g portion of each sample was homogenized with 50ml of distilled water blended for 30 seconds and the pH value of the mixture was determined using pH meter, Hach Sension 1.

3.4 Chemical Parameters for Assessing Fish Quality

3.4.1 Determination of Thiobarbituric acid reactive substances (TBARS)

The levels of thiobarbituric acid reactive substances were determined according to procedure described by Natseba *et al*. (2005) and expressed as mg malonaldehyde / kg fish flesh. Two hundred mg of fish sample will be homogenized in a 25ml buthanol, 5ml of the homogenate was mixed well with 5ml of TBA agent in a capped test tube. The mixture was heated in water bath (95℃) for 2 hours and then cooled at room temperature. The color development was measured at 530nm as follows: where As is the absorbance of the sample against a blank sample of distilled water (Ab) (mg Malonaldehyde / kg fish flesh).

$$TBA = 50\,\frac{As - Ab}{200}$$ (Natseba *et al*., 2005)

3.4.2 Determination of Free Fatty Acid (FFA)

Free fatty acid was measured as described by Egan *et al.* (1997). One hundred and fifty g of comminuted fish sticks was mixed thoroughly with 250ml of chloroform for 10 minutes and filtered with Wattman filter paper. Ten ml of the filtered liquid will be used for oil sample weight after drying at 105℃. Free fatty acid content will be expressed as oleic acid percent by the titration of filtered liquid (2ml) containing 3 droplets of phenolphetalein with 0. 1N NaOH as follows:

(% oleic acid) FFA = $\frac{0.1\ NaOH\ used\ in\ titration\ x\ 28.2\ \ x\ 100}{oil\ sample\ x\ 1000}$ **(**Egan *et al*., 1997)

3.4.3 Determination of Extractable Protein Nitrogen

The extractable protein was evaluated according to the procedure described by Dyer *et al.* (1964). The samples were prepared by using a knife to dice representative samples of frozen fish muscle into small cubes. Twenty two g portion of the sample was weighed into sample cups and stored frozen. Four hundred and thirty ml of extracted buffer stored in the freezer until ice crystals are formed. Preweighed frozen sample and a 430ml aliquot chilled buffer were added into a blender jar, baffle was inserted into the blended jar and was blended for 1 minute and glass rod was used to reposition baffle and blend for 1 more minute, baffle was inserted again and blended for another 0.5 minute, extract was then poured into 600ml beaker, some portion was poured into centrifuge tube and centrifuged for 30 minutes at 13,000rpm. A portion of the supernant was decanted into 50ml beakers, and the sample was analyzed for protein.

The protein analysis was done using the procedure described by Dyer *et al.* (1964). Half, 1,2,3,4 and 5ml portion of standard BSA solution added into test tubes to make up 5ml distilled water. Two blanks were prepared with 5ml of distilled water, extract of aliquot (supernatants) containing 0.1 to 1mg protein nitrogen (0.6 to 6.0mg protein) with distilled water. Into each tube and one blank, 5ml of burette reagent was pipetted and stirred thoroughly on the vortex mixer, it was allowed to stand for 20 minutes at room temperature for development of full violet color. Spectrophotometer was set at 540 nm, to zero absorbance with turbidity blank in both reference and sample cells.

Absorbance of prepared samples measured, spectrophotometer was used again with burette blanks and absorbance of reacted standards and extracts against burette blank was measured. UV-VIS spectrophotometer model of UV 2550, made of Shimadzu was used.

3.4.4 Determination of Formaldehyde

The formaldehyde was determined using the procedure described by Bureau of Indian Standards (1997), which involved the extraction of the fish muscle tissues with trichloroacetic acid for the removal of proteins and treatment with acetyl acetone-ammonium acetate to form a colored formaldehyde derivative. The absorbance was measured with spectrophotometer. Five ml trichloroacetic acid extract of the fish muscle tissue was placed in a 50ml beaker, 10ml of distilled water was added, the solution was made alkaline with a few drops of 30 percent NaOH, and pH was adjusted to 6.0 with 5 % acetic acid. The solution was made up to 25ml with water and 5ml of it was mixed with 5ml of acetyl acetone-ammonium acetate reagent, the mixture was kept still for 50 minutes at 37℃ and color was read at a wave length of 410 nm. For blank, distilled water in duplicate was taken in place of sample, and for standard, series of tubes using 100µg formaldehyde standard solution were prepared with concentration ranging from 0-80µg. 6405/UV/VIS spectrophotometer, Jenway model England was use.

$$\text{Formaldehyde (mg,prcent)}=\frac{absorbance\ of\ sample \times concentration\ in\ standard \times 25 \times 100 \times 100}{absorbance\ of\ standard \times 5 \times 10 \times 10}$$

(BIS, 1997)

3.4.5 Determination of Indole

Indole was determined according to procedure described by BIS (1997). Forty g of fish was homogenized with 80ml ice cold trichloroacetic acid solution (TCA) in a waring blender for 1minute. Eighty ml ice cold light petroleum was added and blended for 1minute. The homogenated sample was transferred into 250ml centrifuge for 10 minutes at 10,000rev/min, supernatant was filtered through Whatman no. 1 paper under suction and filtrate was then

transfered into 250ml separatory funnel. When the 2 layers had separated, acid layer (lower) was transferred to second 250ml separatory funnel. Protein precipitate was denatured (separated by centrifugation) with 40ml light petroleum and filtered as described above, filtrate was transferred into second 250ml separatory funnel already containing TCA layer from the first extraction, shaken for 1minute and the layers were allowed to separate, acid layer was transferred to third separatory funnel and extract for third time with 40ml light petroleum.

All light petroleum extract was combined into 1 separatory funnel, indole was extracted with exactly 5ml freshly prepared Ehrlich's reagent by vigorously shaking for 1 minute and the rose indole complex formed was quantitatively transferred to Ehrlich's reagent layer. When the layers had separated, lower layer was transferred to 1cm path cell and read at 570nm against reagent blank. Standard curve prepared by measuring volume from 0.5 to 4ml indole solution (working solution) 80ml TCA was added into a separatory funnel, indole was extracted using the procedure described above to construct standard curve. Indole content was expressed as the amount of indole of microgram per 100g fish muscle. 6405 UV /VIS spectrophotometer, Jenway model, England was used.

3.4.6 Ash Content Determination

The term ash refers to the residue left after the combustion of the oven dried sample and is a measure of the total mineral content. Determination of ash content was done according to AOAC (1990).

Four crucibles were preheated in a muffle furnace at about 550°C. Each crucible was cooled in a dessicator and weighed Approximately 1g of each sample was weighed into the different

crucibles, the crucibles and their contents were transferred into the muffle furnace at 550°C and allowed to stay for 5 hours. The weights of the crucible contents were taken and recorded,

Percentage ash was calculated using the expression below:

$$\% \text{ ash} = \frac{weight\ of\ ash}{weight\ of\ dry\ sample} \times 100 \quad \text{(AOAC, 1990)}$$

3.5 Sensory Evaluation

Sensory evaluation of all samples was done by 24 trained panelists using Quality index method (QIM). Evaluation of the quality parameters such as; general appearance, eyes and gills' smell, shape, color of all samples were given demerit scores (0-4) and recorded in the Quality index form (QI form) according to (Larsan *et al*., 1992) (Appendix I). The scores for all the characteristics were summed up to give an overall sensory score, the quality index. Quality index method gives scores of zero to very fresh fish while increasingly larger totals result indicated higher level of deterioration in fish. The total mean of the quality parameter and characteristics of each fish sample was calculated and used to grade the fish samples into five grades A, B, C, D and E as described by Suleiman (2008) with modifications in terms of class limit and definite description of each grade, to suit this research's design, as shown below:

Grade	A	0-4.0	Excellent
	B	4.1-8.0	Good
	C	8.1-12.0	Satisfactory
	D	12.1-16.0	Tolerable
	E	16.1- 20.0	Poor

3.6 Statistical Analysis

Data obtained was subjected to analysis of Variance (Anova) and differences between the samples where present were ranked using Duncan's multiple range test at 5% level of significance. Pearson correlation was used to establish the relationship (where any) between physico-chemical parameters measured (SPSS, 1995). Correlation values at $P \leq 0.05$ levels were considered as statistically significant.

CHAPTER FOUR

4.0 RESULTS

The weight of the examined frozen fish for *S. pilchardus* were within the range of 0.08-1.2kg and length ranged from 32-34 cm, weight for *T. trachurus* were within the range of 1.3-1.6kg and the length were within the range of 37-42cm, weight for *C. harengus* were within the range of 1.8-1.9kg and the length were within range of 30-34cm.

4.1 Physical Indices of Imported Frozen Fish Samples

The values obtained for physical indices of frozen fish samples are represented in Table 4.1. The highest result obtained for electrical properties was seen in *T. trachurus* as having 2.92, and *C. harengus* with the lowest having 2.02. For moisture content, the highest value obtained was found in *S. pilchardus* with the value 60.1, and the lowest value in *C. harengus* was 55.15. The highest value for pH was seen in *T. trachurus* having 7.15, and lowest 6.98 was obtained in *C. harengus.* The values are represented as bars in Figure 4.1.

4.2 Chemical Indices of Imported Frozen Fish Samples

The values of evaluated chemical indices of frozen marine fishes are represented in Table 4.2. For TBA same result was obtained for both *T. trachurus* and *C. harengus* having 0.4mg and *S. pilchardus* as having 0.5mg. For FFA *S. pilchardus* was having the highest value of 2.35 and the lowest value of 1.15 in *T. trachurus*. The highest value of 7.4 for EPN was obtained in *S. pilchardus*, and the lowest value of 3.1 in *T. trachurus*. Formaldehyde test indicated *S. pilchardus* as having the highest value of 1.7, and the lowest value of 1.3 in *C. harengus*. Indole test indicated *C. harengus* with the highest value of 1.9, and the lowest value of 1.55 in *S.*

Table 4.1: Physical Indices of Frozen Fish Samples (Mean±SE)

Physical indices	***S. pilchardus***	***T. trachurus***	***C. harengus***	**Per.li**
Electrical properties (s/m)	2.14 ± 0.58^{a}	2.92 ± 0.53^{a}	2.02 ± 0.23^{a}	-
Moisture (%)	60.1 ± 6.30^{a}	58.25 ± 0.05^{a}	55.15 ± 0.45^{a}	60-70 *
PH	7.04 ± 0.10^{a}	7.15 ± 0.04^{a}	6.98 ± 0.14^{a}	6.5 °

Note: Means having different superscripts in the same row are not significant ($p \leq 0.05$)

Sources:

* FAO (2001)

* EOS (2005)

Per.li- Permissible limit

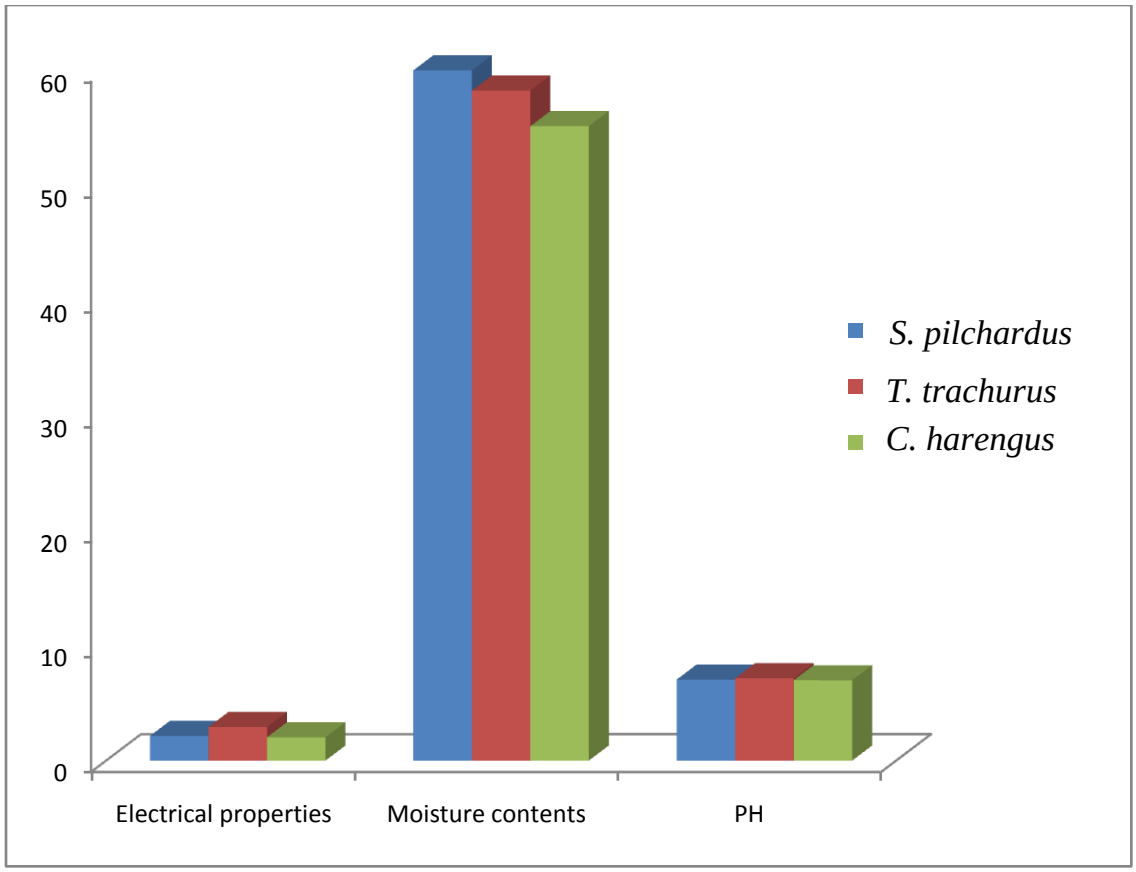


Fig 4.1: Physical indices of frozen fish samples

Table 4.2 Chemical Indices of Imported Frozen Fish Samples (Mean ± SE)

Chemical indices	***S. pilchardus***	***T. trachurus***	***C. harengus***	**Per.li**
Thiobarbituric acid (mg)	0.5± 0[a]	0.45 ± 0.05[a]	0.45±0.071[a]	° 4.5
Free fatty acid (%)	2.35±0.25[a]	1.25 ± 0.75[a]	1.9±0.20[a]	* 2.5
Extractable protein nitrogen (g)	7.4± 3.80[a]	3.1± 0.25[a]	5.2±2.10[a]	-
Formaldehyde (mg)	1.7±0.35[a]	1.4± 0.10[a]	1.3±0.15[a]	Φ10
Indole (mg)	1.55±0.05[b]	1.85±0.005[a]	1.9±0[a]	Φ2.5
Ash content (%)	1.35± 0.15[a]	1.25± 0.05[a]	1.13±0.43[a]	ϕ 1.5

Note: Means having different superscripts in the same row are significantly different (P≤ 0.05).

Sources:

° EOS (2005) Φ BIS (1997)

* FAO (2013) ϕ USDA(2010)

Per.li - Permissible limit

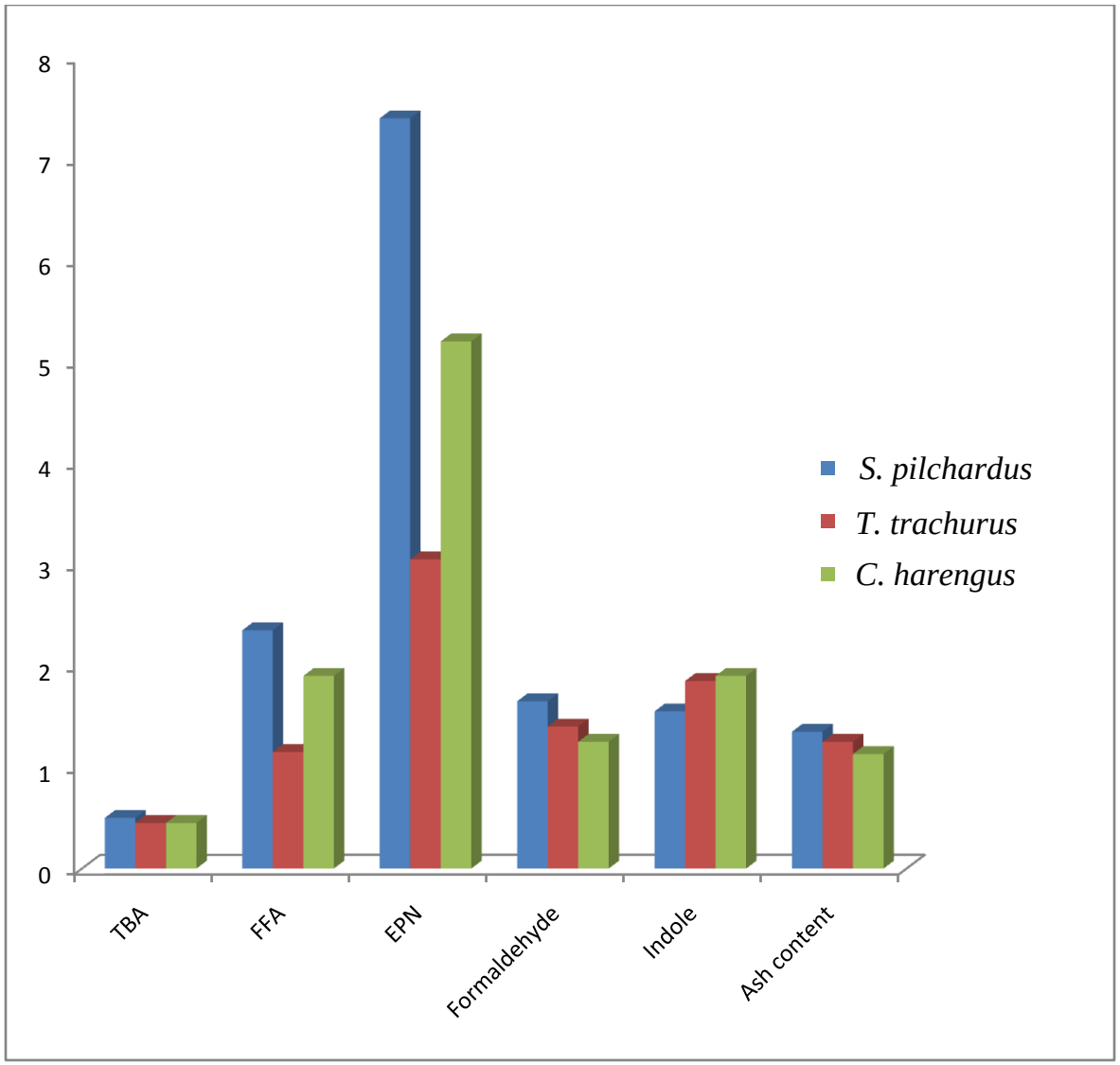


Fig 4.2: Chemical indices of frozen fish samples

Note:

TBA - Thiobarbituric acid

FFA - Free fatty acid

EPN - Extractable protein nitrogen

pilchardus. Highest value of 1.35 was found in *S. pilchadus* for ash content test and the lowest 1.13 in *C. harengus*. The values are represented as bars in Figure 4.2.

4.3 Quality Indices of Imported Frozen Fish Samples

The scores of characters measured of *S. pilchardus* are presented in Figure 4.3, with gill shape having the best character and stiffness of the skin having the highest score.

The scores of characters measured for *T. trachurus* are shown in Figure 4.4, with gill color having the best character and blood spot on gill cover having the highest score obtained.

The scores of the characters measured for *C. harengus* are as depicted in Figure 4.5, with gill color, eye color and skin brightness having the best quality and stiffness of the skin having the highest score.

4.4 Relationship between Physicochemical Indices of *S. pilchardus*

The correlation of physicochemical analysis of *S. pilchardus* is shown in Table 4.3. There was significant relationship between physicochemical indices of *S. pilchard*. There was strong positive correlation (r = 0.731) between electrical properties (EP) and moisture content (mc). The correlation of pH with electrical properties (r = 0.108) was positively weak, while with moisture content (r = -0.599) was negatively correlated. TBA correlation negatively with EP (r = -0.655) and pH (r = -0.822) and weak and positively correlated with mc (r = 0.037). Free fatty acid correlate significantly ($P<0.05$) with pH (r = 0.999), while EP (r = 0.133) was positively weak, mc (r = -0.579) and TBA (r = -0.836) were negatively strong. There was strong and positive correlation of EPN with EP (r = 0.832) and mc (r = 0.987), while pH (r = -0.462), TBA (-0.125) and FFA (r = -0.440) were weak and negatively correlated. All the parameters correlated

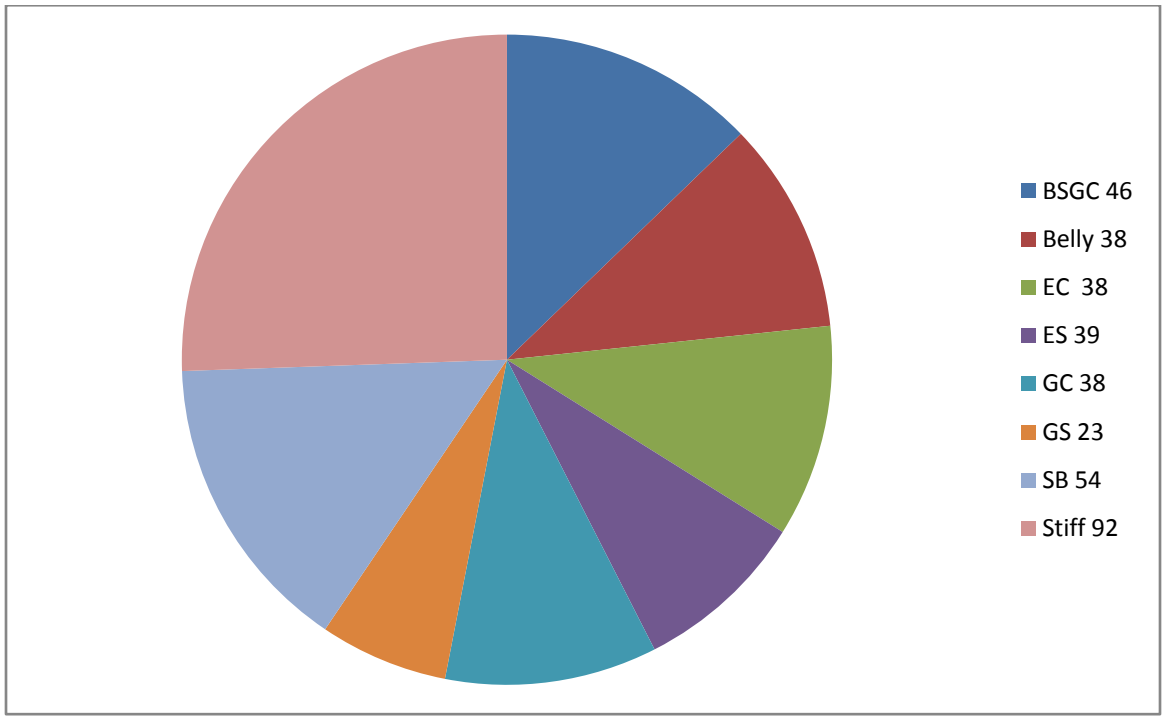


Figure 4.3: Scores of characters measured for *S. pilchardus*

Legend:

Belly	- Belly firmness	GC	- Gill color
BSGC	- Blood spot on gill cover	GS	- Gill shape
EC	- Eye clarity	SB	- Skin brightness
ES	- Eye shape	Stiff	- Stiffness of the skin

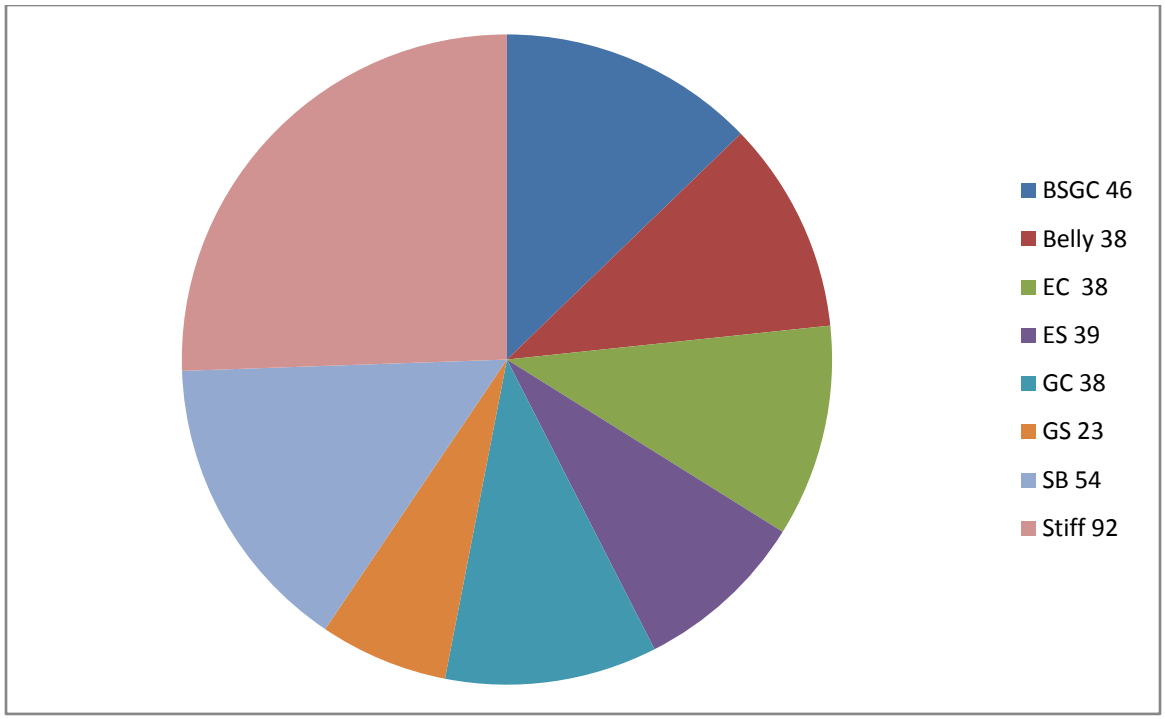


Figure 4.4: Scores of characters measured for *T. trachurus*

Legend:

Belly	-	Belly firmness	GC	-	Gill color
BSGC	-	Blood spot on gill cover	GS	-	Gill shape
EC	-	Eye clarity	SB	-	Skin brightness
QI	-	Quality index	ES	-	Eye shape

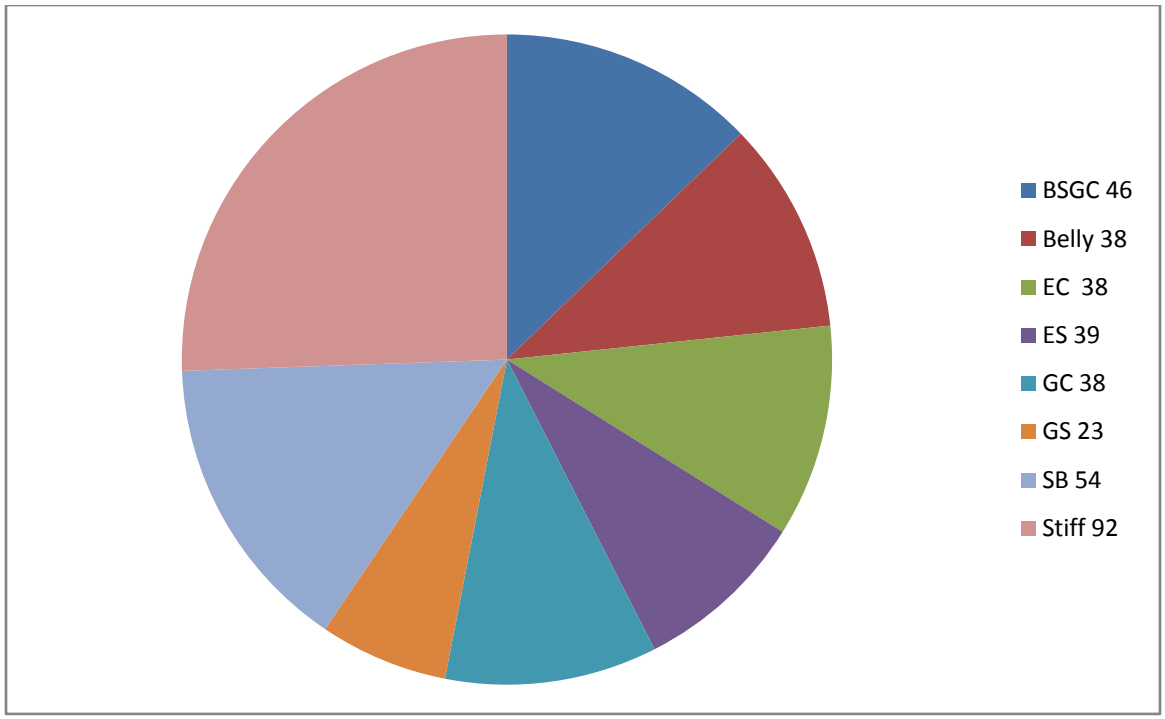


Figure 4.5: Scores of characters measured for *C. harengus*

Legend:

Belly	-	Belly firmness	GC	-	Gill color
BSGC	-	Blood spot on gill cover	GS	-	Gill shape
EC	-	Eye clarity	SB	-	Skin brightness
QI	-	Quality index	ES	-	Eye shape

positively with formaldehyde except TBA (r = -0.904) that was negatively correlated. There was weak and negative correlation of indole with mc (-0.214), EPN (r = -0.053) and strongly negatively with TBA (r = -0.984), EP (r = 0.510), pH (0.910), FFA (r = 0.920) and formaldehyde (r = 0.814) were strong and positively correlated. Ash contents correlate positively weak with EP (r = 0.189) and formaldehyde (r = 0.567) but significantly ($p<0.05$) positive with FFA (r = 0.998) and indole (r = 0.941), while at mc (r = -0.531), TBA (r = -0.866) EPN (-0.388) were negatively correlated.

4.5 Relationship between Physicochemical Indices of *T. trachurus*

Table 4.4 is showing the correlation of physicochemical properties of *T. trachurus*. There was significant ($p<0.05$) correlation between the *T. trachurus* physicochemical indices. Moisture contents and EP (0.0100) was weakly and positively correlated. There was also weak correlation between pH and EP (r = 0.388) but strong at mc (r = 0.925). The correlation between TBA and EP (r = 0.987) was strongly positive, with mc (r = 0.170) and pH (r = 0.532) were weak. There was strong and positive correlation of FFA with EP (r = 0.987), pH (r = 0.843) and TBA (r = 0.094) except with mc (r = 0.575). The correlation of EPN with EP (r = 0.964), TBA (r = 0.908) and FFA (r = 0.642) were strong; it was weak and negatively correlated with mc (r -0.258). There was positive correlation of formaldehyde with all the corresponding physicochemical indices. It correlated significantly ($p<0.05$) TBA (0.998); while with EP (r = 0.994), FFA (r = 0.879) and EPN (r = 0.930) were strong but weak with mc (r = 0.115) and pH (r = 0.484). Indole correlated positively strong with mc (r = 0.938) and pH (r = 0.737) but weakly with FFA (r = 0.257). It correlated negatively weak with EP (r = -0.337), TBA (r = -0.181), EPN (r = -0.576) and formaldehyde (r = -.0235). Ash contents correlated strongly and positively with the

Table 4.3: Correlation of Physiochemical indices of *S. pilchardus*

	EP	**Mc**	**pH**	**TBA**	**FFA**	**EPN**	**Forma**	**Indole**	**Ash cont.**
EP	1								
Mc	0.731	1							
PH	0.108	-0.599	1						
TBA	-0.655	0.037	-0.822	1					
FFA	0.133	-0.579	0.999*	-0.836	1				
EPN	0.832	0.987	-0.462	-0.125	-0.440	1			
Formal.	0.915	0.394	0.500	-0.904	0.522	0.537	1		
Indole	0.510	-0.214	0.910	-0.984	0.920	-0.053	0.814	1	
Ash cont.	0.189	-0.531	0.997	-0.866	0.998*	-0.388	0.567	0.941	1

* = Significant at 0.05

Note:

EP - Electrical properties

Mc - Moisture content

TBA - Thiobarbituric acid

FFA - Freefattyacid

EPN - Extractable protein nitrogen

Formal - Formaldehyde

Ash Content - Ash contents

Table 4.4: Correlation of Physicochemical indices of *T. trachurus*

	EP	**Mc**	**pH**	**TBA**	**FFA**	**EPN**	**Formal.**	**Indole**	**Ash cont.**
EP	1								
Mc	0.010	1							
PH	0.388	0.925	1						
TBA	0.987	0.170	0.532	1					
FFA	0.823	0.575	0.843	0.904	1				
EPN	0.964	-0.258	0.128	0.908	0.642	1			
Formal.	0.994	0.115	0.484	0.998*	0.879	0.930	1		
Indole	-0.337	0.938	0.737	-0.181	0.257	-0.576	-0.235	1	
Ash cont.	-0.442	0.893	0.655	-0.292	0.145	-0.666	-0.345	0.993	1

* = Significant at 0.05

Note:

EP - Electrical properties

Mc - Moisture content

TBA - Thiobarbituric acid

FFA - Freefattyacid

EPN - Extractable protein nitrogen

Formal - Formaldehyde

Ash Content - Ash contents

following: mc (r = 0.893), pH (r = 0.665) and indole (r = 0.993) but it was negatively correlated with EP (r = -0.442), TBA (r = -0.292), formaldehyde (r = -0.345) all were weak and strongly negative with EPN (r = -0.666)

4.6 Relationship between Physicochemical indices of *C. harengus*

The correlation of physicochemical indices of *C. harengus* is shown in Table 4.5. Moisture contents and EP (r = -0.359) correlated negatively. There was also negative correlation between pH and EP (r = 0.439) but strongly positive with mc (r = 0.996). There correlation between TBA and EP (1.00) was positive and perfect and it was weakly negative with mc (r = -0.369) and pH (r = 0.449). The FFA correlated strongly positive with EP (r = 0.995) and TBA (0.994), while mc (r = -0.263) and pH (r = -0.346). The correlation of EPN with EP and TBA were perfectly (r = 1.000) and strongly positive with FFA (r = 0.994), while mc (r = -0.357) and pH (r = -0.436). There was positive correlation between formaldehyde and the corresponding physicochemical indices. It correlated strongly with FFA (r = 0.643) but weakly with EP (r = 0.562), mc (r = 0.570), pH (r = 0.496), TBA (r = 0.553) and EPN (r = 0.564). Similarly, indole correlated positively with EP (r = 0.126), TBA (r = 0.225) and EPN (r = 0.128) were weakly, while mc (r = 0.881), pH (r = 0.836) and formaldehyde (r = 0.891) were strongly positive. Ash contents correlated strongly with mc (r = 0.604) and pH (r = 0.671), and indole (r = 0.154) was weakly correlated. There was strong negative correlation of ash contents with EP (r = -0.961), TBA (r = -0.964) and EPN (r = -0.964) but weakly negative with formaldehyde (r = -0.311).

Table 4.5: The Correlation of Physicochemical indices of *C. harengus*

	EP	**Mc**	**pH**	**TBA**	**FFA**	**EPN**	**Formal**	**Indole**	**Ash cont.**
EP	1								
Mc	-0.359	1							
pH	-0.439	0.996	1						
TBA	1.000**	-0.369	-0.449	1					
FFA	0.995	-0.263	-0.346	0.994	1				
EPN	1.000**	-0.357	-0.436	1.000**	0.995	1			
Formal.	0.562	0.570	0.496	0.553	0.643	0.564	1		
Indole	0.126	0.881	0.836	0.115	0.225	0.128	0.891	1	
Ash cont.	-0.961	0.604	0.671	-0.964	-0.928	-0.960	-0.311	0.154	1

** = Significant at 0.01

Note:

EP - Electrical properties

Mc - Moisture content

TBA - Thiobarbituric acid

FFA - Freefattyacid

EPN - Extractable protein nitrogen

Formal - Formaldehyde

Ash Content - Ash contents

CHAPTER FIVE

5.0 DISCUSSION

5.1 Physical Indices of Frozen Fish Samples

In this study, *C. harengus* had low hydrogen ion concentration; Sugumar *et al*. reported low pH inhibits most of bacterial activity. The highest pH was found in *T. trachurus* but it did not exceed the acceptable limit. The mean pH value of 6.9 to 7.1 in this study is similar to the postmortem pH of 6.0 to7.1 stated by Pacheco-Anguilar *et al*. (2000).

Similar result of the moisture content obtained was reported by Magawata *et al. (*2015) for *Clupea harengus.* The natural moisture content of sardines was 74.27%/100g, which decrease during preservation period reaching 52%/100g (Marilia *et al.* 2003). Range of moisture content for mackerel 74 – 56%, similar result was obtained for Atlantic mackerel 58.80% (Oladunni *et al*. 2013). Low moisture content reduces spoilage and it indicates the good quality of sea food.

5.2 Chemical Indices of Frozen Fish Samples

Thiobarbituric acid value is an important indicator of rancidity especially in seafood product. The TBA value of the examined samples indicated that the frozen fish samples were acceptable according to the Egyptian Organization for Standardization and Quality Control (EOS, 2005) which recommended that the permissible limit for TBA be not more than 4.5mg malonaldehyde / Kg. Similar result was reported by Hassan (2011).

The marine fish species contains low freefattyacid, the studied samples of the frozen fish were within the acceptable level of 2.5% by FAO (2013). *T. trachurus* has been reported as having significantly higher protein content than *S. pilchardus* as reported by Paul and Vivian (2011), in this study it has been observed that *T.trachurus* has higher protein content than *S. pilchardus*

(7.4) and *C. harengus* (5.2) due to low extractable protein nitrogen content recorded for *T. trachurus* (3.1). The result of Formaldehyde found in mackerel was similar to that reported by Noordiana *et al.* (2011). Ash is a measure of the mineral content of a food item (Omotosho and Olu, 1995). The ash content of the species of this study matches with that reported by USDA (2010) for mackerel. Ash content of the frozen fish products examined remained is to those reported by Magawata *et al*. (2015) for *Clupea harengus*. Range for the ash content gave an indication that the fish samples may be good sources of minerals such as calcium, potassium and magnesium.

The result of indole range obtained (1.6 to 1.9) indicates that the frozen fish samples examined are within the acceptable limit according to the Bereau of Indian Standard (1997), which recommended that permissible limit of Indole be not more than 2.5mg/kg.

5.3 Quality Index Method of Frozen Fish Samples

Quality index method is based on the significant sensory parameters for fish using many parameters and a scoring system from 0 to 4 demerit points (Jonsdottir, 1992). The quality index score grading in this study graded sample A of a good quality with a mean hedonic points of 5.6. Sample B of a satisfactory quality with mean hedonic points of 8.3 and sample C of a good quality with mean hedonic points of 5.3.

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CHAPTER SIX

6.0 SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

6.1 Summary

The research was done to evaluate the quality of some imported frozen marine fishes sold at Sabongari Zaria. Methods used for determination of the quality of the fishes were physical, chemical and quality index methods. The physico-chemical indices indicate that all the examined frozen fishes were within permissible limits set by certified regulatory bodies and possessed satisfactory quality indices. Among all the examined samples, *Trachurus trachurus* performed best with less protein denatured, due to the less EPN result obtained. Quality index methods was done by 24 panelist, *S. pilchadus* and *C. harengus* were graded as having good quality, *T. trachurus* was graded to have satisfactory.

6.2 Conclusions

Based on the objectives of this research, the following conclusions were arrived at:

- Prominent physical indices dictating the quality of the imported frozen marine fish are, Electrical properties (2.92), pH (7.04) for *T. trachurus,* Electrical properties (2.14), pH (7.04) for *S. pilchardus* and Electrical properties (2.02), pH (6.98) for *C. harengus*
- Promient chemical indices dictating quality of imported frozen marine fishes are TBA for both *T. trachurus* and *C. harengus* was (0.4) and (0.5) for *S. pilchardus,* FFA and EPN were highest in *S. pilchardus* (2.45) and (7.4) respectively
- Prominent features dictating quality of imported frozen marine fishes using Quality Index Method are Stiffness of the skin(1.2) and Skin brightness(0.7) for *S. pilchardus*, Blood spot on gill cover (1.9,0.9) and Stiffness of the skin (1.7,1) for *T. trachurus* and *C. harengus* respectively

6.3 Recommendations

It is therefore recommended that:

- *Trachurus trachurus* is recommended over *Sardina pilchardus* and *Clupea harengus*.

-Proper transportation and storage system should be maintained to retain the quality of the imported frozen fishes.

REFERENCES

Abbas, K. A., Mohammed, A., Jamilah, B., and Ebrahimian, M. (2008). A review on correlations between fish freshness and pH during cold storage. *American Journal of Biochemical Biotechnology*, **4** (4): 46-421.

Adeyeye, E. I. (2002). Determination of the chemical composition of the nutritional valuable parts of male and female common West African freshwater crabs *Sudananaute africanus. International Journal of Food Science Nutrition,* 53: 189-196.

Alasalvar, C., Grigor, J. M. and Ali, Z. (2011). Practical evaluation of fish quality by objective, subjective, and statistical testing. In: Alasalvar, C., Shahidi, F., Miyashita, K., Wanasundra, U., (Eds). *Handbook of Seafood Quality, Safety and Healthy Applicators*. New Delhi: Blackwell Publishing Ltd;p.13-29.

Alicia Villegas (2015).Pelagic round up, Russian ban hits Iceland , Nigeria import quotas. Retrieved on 11 01 2016, from http:// www. under currentnews. Com/.2015….

AOAC. (1990). *Official methods of analyses of the association of analytical Chemists,* 15th Ed. Washington DC: 66-68

Arannilewa, S. T., Salawu, S. O., Sorungbe, A. A. and Olasalawu, B.B. (2005). Effect of frozen period on the chemical, microbioloqical and sensory quality of frozen tilapia fish *(Sarotherodum galiaenus). African Journal of Biotecthnology*, **4** (8): 85-855.

Ausbourg, S.P., Perez-Alonso, F. and Gallardo, J.M. (2004). Studies on rancidity inhibition in frozen horse mackerel, *European Journal of Lipid Science and Technology*, 106:232-240.

Austin Oboh. (2015). Nigeria import 125 billion worth of fish annually. Independent News paper Limited. April 29 2015 P. 8

Bally, N. 2014. *Sardina pilcharus* (Walbum, 1792). In: Froese, R. and Pauly D. (Eds). (2014). Fish Base. Accessed through: World Register of Marine Species from http:\\www.marine. species .org \alpha. Php? P = taxdetails &id = 126421 on 24-11-2014.

Bally, N. 2014. *Trachurus trachurus* (Linneaus1758). In: Froese R. and Pauly D. (Eds.) Fish Base. Accessed through: World Register of Marine Species. On 8-11-2014. From http:\\www. Marine species org\alpha.php? p = taxdetails&id=126822.

Bally, N. 2014. *Clupea harengus* (Linneaus 1758). In: Froese R. and Pauly D. (Eds.) Fish Base. Accessed through: World Register of Marine Species. On 8-11-2014. From http:\\www. Marine species org\alpha.php? p = taxdetails&id=126822.

Bauchart, C., Champbon C., Mirand, P. P., Savary I., Auzeloux, D., Resbond and Morzel M. (2007). Peptides in rainbow trout (*Oncorhynchus mykus)* muscle subjected to ice storage and cooking. *Journal of Food Chemistry*, **100**: 1566-172.

Boonsumrej, S., Chaiwannichsiri, S., Tantratian S., Suzuki T. and Takai R. (2007). Effect of freezing and thawing on the quality changes of tiger shrimps *(Penaeus monodon)* frozen by air blast and cytogenic freezing . *Journal of Food Engineering.* 80: 292-299.

Bo Wei (2011). The Cold chain Management in Supermarket Case Study on the Fresh Food Logistics in a Supermarket. Retrieved on 10/10/2014 from www.diva/portal.org/smash/.../fulltext02.

Bremner. H. A., and Sakaquchi, M. (2000). A critical look at whether 'freshness' can be determined. *Journal Aquatic Food Production Technology*, 9: 5-25.

Bureau of Indian Standards (1997). *Indian standard on prawns (shrimps) frozen specification.* 3rd Revision, New Delhi 110002.

Chebet, L. (2010). Methods for Evaluation of fish freshness and quality. Retrieved on 10/10/2014 from www.skemman.is/.../Masters_thesis_2010_Lilli...

Chen Y., Nguyen, C., Semmens, K., Beamer, S. and Jaczynski J. (2008). Chemical changes in omega -3-enhanced farmed rainbow trout (*Oncorhynchus mykiss)* fillets during abusive temperature storage. *Food Control,* 19: 599- 608.

Church, N. (I998). Feature MAP and crustaceans - sensory enhancement. *Food Science Technology Today,* 12 (2): 73-82.

Cordis, A. D., Maulik, N. and Das D.K., (1995). Detection of oxidation stress in heart by estimating the dinitrophynylhyrazine derative of malonaldehyde. *Journal of Molecular Cell Cardinal,* 21:1645-1653.

David, A., David, R., Kathyn, B. (2008). Frozen foods handling and storage, world food logistics organization. New York, pp1-12.

Daramola, J.A., Fasakin, E. A., and Adeparusi, E. O. (2007). Changes in Physicochemical and Sensory Characteristics of smoked-dried fish species stored at ambient temperature. African Journal of Food Agriculture Nutrition and Development, **7** (6): 98-108

Davies, R. M. (2006). Mechanization of fish industry in Nigeria state and potential for self sufficiency in fish production. Proceedings of the one –Day workshop on intensive fish farming, USA. August 19 2006. pp.10

De Koning, A. J and Mol, T. H. (1991). Quantitative quality test for frozen fish soluble protein and free fatty acid content as quality criteria for hake *(Merluccius capensis)* stored at -18°C. *Journalof Science Food Agriculture*, **54** (3): 449-458.

Dyer, W. J., Brockerhoff, H., Hoyle, R. J., and Fraser D. I. (1964). Polyphosphate treatment of frozen cod. In protein extractability and lipid hydrolysis. *Journal of Fish Research of Board of Canada,* 21:101.

Egan, H., Krik, R .S. and Sawyer, R. (1997). *Pearson's chemical Analysis of food* (9thed.). Churchil Livingstone Edinburgh,UK, pp.609-634.

Egyptian Organization for Standardization and Quality Control (EOS) (2005). *Chilled and frozen fish*. ES; 3494, pp 889-892.

Erkan, N. and Ogden, O. (2008). Quality Assessment of Whole and Gutted Sardines (*Sardina pilchadus)* stored in ice. *International Journal Food Science,* 43:1549-1559.

Flecher D L. (1999). Broiler beast meat color, variation pH, and texture. Poultry Science; 78: 1323-1327 FAO/Codex. Guidelines for the Sensory Evaluation of Fish and Shellfish in laboratories.CAC/GL, 31:3-33.

Food and Agriculture Organization. (2001). *Measuring Fish Composition.* Retrieved on 28/8/14 from www.fao.org/wards /fan.../x5957eo|....

Food and Agriculture Organization. (2007). *Protein and Amino Acid Requirement in Human Nutrition.* WHO Press.150- 179 p.

Food and Agriculture Organization. (2014). *Quality of fish and fish products*. Fisheries and Agriculture for a World Without Hunger. Retrieved on 6/8/14 from www.fao.org \fishery\ topic\1521\en

Gandotra, R. S., Shaman, M. K. and Gupta, S. (2012). Effect of chilling and freezing on fish muscle. *Journal of Pharmacology Biological Science*, 2: 5-9

Green-Peterson D.M.B. (2010). Sensory Quality of Seafood- in the Chain from Catch to Consumption (PhD) Thesis. Copenhagen, Denmark: University of Denmark.

Hassan, F. (2011). Quality evaluation of some fresh and imported frozen seafood. *Advance Journal of Food Science and Technology*, **3**(1):83 -88.

Huidoboro A. and Tejada M. (2004). Gilthead sea bream *(Sparus aurata),* suitability for freezing and commercial alternatives. *Journal Science Food and Agriculture,* 84: 1405-1413.

Huss H. H. (1995). *Quality and Quality changes in fresh fish.* Food and Agriculture Organization Fisheries Technical Paper. Food and Agriculture Organization, United Nations, Rome. pp.348

Huss, H. H., Ababouch, L., and Gram, L. (2003). *Assessment and Management of Seafood Safety and Quality* Rome. Food and Agriculture Organization of the United Nations. FAO Fisheries Technical Paper 444. p239

Huss, H. H., Ababouch, L., and Gram, L. (2004). *Assessment and Management of Seafood Safety and Quality.* Rome Italy. Food and Agriculture Organization of the United Nations. FAO Fisheries Technical Paper 444

Hyldig, G., Bremme, R. A. Martinsdottir, E. and Schelvis R. (2007). *Quality Index Method*. In: Nollet L.M.L., Boylston, T., Chen, F., (Eds). *Handbook of Poultry and Seafood Quality.* Oxford. Blackwell, p.499-510.

Hyldig, G.,and Green -Peterson, D.M.B. (2004). Quality Index Method- an objective tool for determination of sensory quality. *Journal of Aquaculture Food Production Technology,* **13** (4): 71-80.

Hyldig, G., Martinsdottir, E., Sveinsdottir, K., Schelvis, R. and Bremmer, A. (2010). Quality Index Method. In: Nollet L.M.T., Toldra, F., (Eds). *Handbook of Seafood Products analysis.* New Jersey, CRCPress, p. 463-481.

Ismail, H. M., (2005). The role of omega-3 fatty acids in cardiac protection: An overview, *Front. Biological Science,* 10: 1079-1088.

Jonstdottir, S. M., Hyldig, G., Nielsen, J., Bleechmoie,T. and Silberg, S. (1999). Rapid PC based sensory methods. *Information Fish International*, 2: 54-56.

Kinsella, J. E., Lokesh, B., and Stone, R. A. (1990). Dietary n-3 polyunsaturated fatty acid amelioration of cardiovascular diseases: Possible mechanisms. *America Journal of Clinical Nutrition*, 52: 1- 28.

Kostuch, S. and Olley, J. (2009). *Protein changes in frozen fish.* Retrieved on 20/10/2015 from www.tandfonline.com/doi/abs/10.1080/10408397609527218# ViZp-DpDk.

Ladosa, V., Barros-Velazquenz, J., Gallardo, J. M and Auboug. A. (2004). Effect of advanced chilling methods on lipid damage during sardine (*Sardina pilchardus*) storage. *European Journal of Lipid Science and Technology*, 106: 844-850.

Laila, D. L., Wahidzu. Z. and Tajul. A.Y. (2013). Effect of chilled frozen storage on the physico-chemical, microbial and sensory quality of farmed, Bighead carp (*Hypophythalmichythys nobilis*). *Journal of Fisheries and Aquatic Science*, 8:686-696.

Larry K. (2003). Transportation: The Squeaky Wheel of the Food Safety System. *Food Safety Magazine* .November 17. Retrieved on 4/14/2014 from www.food safety magazine .com/magazine.

Larsen, E. P., Heldbo, J., Jerspersen, C .M. and Nielsen, J. (1992). *Development of a Standard for Quality Assessment on Fish for Human Consumption*. In; *Quality Assurance in the Fish Industry*. Hiss. In H.H., Jacobson, M., Liston, J. (Eds.) Elsevier, Amsterdam .pp.351-358

Livestock and Aquaculture Watch. (2007). Fishing boom in Nigeria. *The Monthly Journal for livestock and Aquaculture*, **2** (11): 6.

Magawata, I., and Abdulmumin, I. (2015). Quality of frozen fish sold in some selected selling points within Katsina metropolis. *Food Science and Quality Management.* 37: 32-38

Marilia, O., Sergio, D. P., Claudio, R. G. and Liafenaz, A. (2003). Monitoring the sardine (*Sardinella brasiliensis*) fermentation process to obtain Anchovies. *Scientia Agriiola* , **60** (3): 511-517.

Martinsdottir E. (2002). *Quality Management of Stored Fish*. In : Bremmer A,(Ed). *Safety and Quality Issues in Fish Processing.* Hirtshals, Woodhead Publishing Limited, United Kingdom. p.360-378.

Mehta,N. K., Elavarasan, K., Reddy, A., and Shamasundar A.M. (2011). Effect of ice storage on the functional properties of proteins from a few species of fresh water fish (Indian major

carps) with special emphasis on gel farming. *Journal of Food Science Technology*, 51 (4) : 655-663

Mendes, R., Cardoso, C. and Pestana, C. (2009). Measurement of malondialdehyde in fish. A comparison study between HPCL methods and the traditional spectrophotometric test. *Food Chemistry*, 112: 1038-1045.

Naoki, N., Masayuki, I. and Bill, C. (2013). Fisheries in Nigeria Report to Minstry of Agriculture, Nigeria Government. Retrieved on 7/11/2014, from http:// systemseng. Jp/.../Nigeria-Fisheries - Report.

Natseba, A., Lwalinda, L., Kakura, E., Muyanja, C.K and Muyonga, H.H., (2005). Effect of pre-freezing icing duration on quality changes in frozen Nile perch *(Lutes niloticus). Food Research International*, 38:469-474.

Nielsen, D. (2005). Quality Index Method provides objectives seafood assessment. *Global Aquaculture Advocates*, 36-38.

Nordiana, N., Fatimah, A. B. and Farhana, Y.C. B. (2011). Formaldehyde content and quality characteristics of selected fish and seafood from wet markets. *International Food Research Journal*, 18: 125-136.

Nuno, M., Soares, S., Mendes, T., Antonia, A. and Vicente, A. (2013). Effect of Chisosan based solution applied as edible coating. *Journal of Food Engineering*. Retrieved on 23-8-2014, from www.elsevier.com\locate\j Food eng. 119: 316-323

Odoli, C. O. (2006).The effect of temperature abuse at the beginning of storage on the quality and shelf life of fresh water Arctic charr (*Salvelinus alpines*). *Fisheries Training Programme*, pp. 1-46.

Oladunni, O., Oladele, and O., Adelaja, O. (2013).Trace Metal, Proximate Composition and Anatomical properties of Four Fish Commonly Consumed in South –western Nigeria. *International Conference on Food and Agriculture Sciences.* AICSIT, Singapore, 2010. Vol. 55. Retrieved on 23-4-2015 from www.icmeeb. Org/program-2013 pdf.

Olokor, J. O., Ihuahi, F. S. and Omojowo, B. A. (2007). *Hand book practical fisheries technology*. National Institute of Fresh Water Fisheries (NIFFR), New Bussa, Niger State Rem-Thomass press pp l.

Omotosho, J. S., and Olu, O.O., (1995). The effect of food and frozen storage on the nutrient composition of some African fishes. *Revenue Biology Tropical*, 43:289-295.

Organ, G. M., Suerre, G. and Peyman, M. (1996). Electrical properties of fish muscle post mortem. *Medical and Biological Engineering and Computing*, the 1st International conference on Bioelectromagnetism, June 9- 13, 1996. Tampere, Finland.

Pacheco-Anguilar R., Lugo-Sanchez M.E. and Robels-Burgueno, M.R. (2000). Postmortem Biochemical characteristics of Monterey sardine muscle stored at O°C, *Journal Food Science*, 65:40-47

Pomeranz,Y.and Meloan, C. E.(1971) . In Food Analysis: Theory and Practices AVI, Yatchting Publishing Corporation West port, pp. 481-510.

Paul, J. U., and Vivian, A. N. (2011). The Biochemical Composition of Three Exotic Fish Delicacies: *Scomber scombrus,Trachurus trachurus* and *Sardina pilchard* frozen and imported into Nigeria, *Pakistan Journal of Nutrtion*, **10** (12): 1158-1162.

Sen, D. P. (2005). *Advances in fish Processing Technology*. Allied Publishers Pvt Limited New Delhi.

Simeonidou, A., Govaris A., and Vareltzis, B., (1997). Quality assessment of seven Mediterranean fish species during storage. *Food Research International*, 30:479-483.

Sikorski, K. and Zdzislaw, E. (2009).Protein Changes in Frozen Fish. Retrieved on 20/10/2015 from www.tand fonline.com/10408397609527218 3#.ViZP-DpDke8.s

Sotelo, C .G., Pineiro, C. and Perez -martin, R. I. (1995). Denaturation of fish proteins during frozen storage; role of formaldehyde. Zeitschrift for lebensmittel. Untersuchung und-forschung, 200: 14-23

Statistical Package for the Social Scientist (SPSS), (1999). SPSS for Windows. Release 100.01. Standard Version. 256 pp.

Suleiman, B. and Abdullahi, S. A. (2008). Consumer Acceptance of *Heterotis niloticus* processed into kilishi stored over time in Zaria, Nigeria. *Biological and Environmental science Journal for the Tropics*, **5** (3):122-125.

Tawari ,C. C. and Abowei, J. F. N. (2011). Traditional fish handling and preservation in Nigeria. *Asian Journal of Agricultural Science*, 3:427-436.

Teslim, Bada. (2010). Market delineation study of the fish market in Nigeria; An application of co integration Analysis. *Journal of Agricultural Sciences*. **2** (3): 101-112

Ukoha, O. O. (1998). Fishery sector of Nigeria: Analysis of the output performance and the way forward. In: *Agricultural Development in the 21st Century*, Tweeten, L., and S.C. Thompson (Eds). Black well Publishing Limited, NewYork, pp. 22-28.

USDA, 2010. U.S. Department of Agriculture, Agricultural Research Service National Nutrient Database for Standard Reference 23. Nutrient laboratory. http:// www.ars.usda.gov/ba/bhnre//nd/.

Vaswani (2006). Nigeria fish Vaswani Nigeria. Retrieved on 9/8/2014, from www. Nigeria fish.org/…

Vaswani (2014). Nigeria fish Vaswani Nigeria. Retrieved on 5/11/2014, from www.nigeria fish.org/...

Wang, T. (2009). *Oxidation deterioration of lipids can directly affect quality characteristics such as color, flavor, texture, nutritive value and safety.* PhD thesis, Faculty of food science and nutrition school of health sciences, University of Iceland.

Ward, A. G. (1963). The Nature of the Forces Between Water and the Macromolecular Constituents of Food. *In: Recen*t *Advances in Food Science*, Vol 3. Leitch, J. M. and Rhodes D.N. (Eds.) Butterworths Publishing Limited, London.

Zalluoa, P. A., Hsua,H.Y., Terwedowa, H. and Zang, T. (2007). Impact of seafood and fruit consumption on bone minerals density. *Mauritas,* 56:1-11.

Zsuzanna, G. J., Zsuzanna, S. and Istan, C. (2011). Fish meal quality and Safety. *Technologija Mesa*, **52** (1): 97-105.

APPENDIX I

Quality Assessment Scheme used to identify the quality index demerit score

Quality parameters	Character	Score
General appearance	Skin	0 bright, shining 1 bright 2 dull
	Bloodspot on gill cover	0 none 1 small, 10-30% 2 Big, 30-50% 3 very big 50-100%
	Stiffness	0 stiff, in rigor mortis 1 elastic 2 firm 3 soft
	Belly	0 firm 1 soft 2 belly burst
	Smell	0 fresh, seaweed/metallic 1 neutral 2 musty/sour 3 stale meat/rancid
Eyes	Clarity	0 clear 1 cloudy
	Shape	0 normal 1 plain 2 sunken
Gills	Color	0 characteristics red 1 faded, discolored
	Smell	0 fresh, seaweed/metallic 1 neutral 2 sweaty/slightly rancid 3 sour stink/stale, rancid
Sum of scores		(Min. O and max. 20

Larsen *et al.,* 1992