

**EFFECT OF SOME AROMATIC PEPPERS ON ORGANOLEPTIC  
AND MICROBIAL QUALITIES OF OVEN-DRIED TILAPIA  
FILLETS STORED AT AMBIENT TEMPERATURE**

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**NOVEMBER, 2025**

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**A PROJECT SUBMITTED TO THE DEPARTMENT OF AQUACULTURE AND  
FISHERIES MANAGEMENT, FACULTY OF AGRICULTURE, UNIVERSITY  
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AWARD OF BACHELOR OF AGRICULTURE (B. AGRIC) DEGREE WITH  
OPTIONS IN FISHERIES**

**NOVEMBER, 2025**

## **CERTIFICATION**

This is to certify that this project was carried out by **Queen Emediong IMO**, under my supervision in the Department of Aquaculture and Fisheries Management, Faculty of Agriculture, University of Benin, Benin City, Edo State, Nigeria.

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**Date**

## **DEDICATION**

The project work is specially dedicated to God Almighty for His hand of grace and favour upon my life all through my academic journey. This work is also dedicated to my darling Mother, Madam Blessing Bassey for her unwavering love, support and prayers.

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## ABSTRACT

This study was carried out to evaluate the effects of selected aromatic pepper spices on the organoleptic and microbial qualities of oven-dried *Oreochromis niloticus* (Nile tilapia) fillets stored at ambient temperature. Fresh fillets were seasoned with 2% (w/w) of three different pepper spices Scotch Bonnet (*Capsicum chinense*), Cayenne (*Capsicum annum*), and Bell Pepper (*Capsicum annum*) and compared with not spiced (control) samples. The spiced and not-spiced fillets were oven-dried at 100°C for 5 hours, packaged in sterile ziploc bags, and stored at room temperature ( $28 \pm 2$  °C) for 14 days. Microbiological, and sensory analyses were conducted at 7-day intervals to assess changes during storage.

Results revealed that the spice treatments and storage durations significantly ( $P < 0.05$ ) affected the sensory and microbial qualities of the fish samples. On Day 1, all samples exhibited high sensory ratings across appearance, aroma, taste, and overall acceptability (3.8 – 4.6), but these values declined progressively by Day 14. Cayenne and Bell Pepper-treated samples retained relatively higher flavour and acceptability scores than Scotch Bonnet and the control, suggesting mild preservative or antioxidant effects. Microbial counts increased with storage time, with bacterial loads ranging from  $7.0 \times 10^{-3}$  cfu/g in Bell Pepper-treated samples to  $240.0 \times 10^{-3}$  cfu/g in Cayenne-treated fillets samples after 14 days. *Proteus* sp., *Staphylococcus aureus*, and *Bacillus* sp. were among the predominant bacterial isolates, while *Aspergillus flavus*, *Mucor* sp., and *Penicillium* species were the most common detected fungi. Ultimately, bell Pepper proved to be the most effective in inhibiting microbial proliferation and maintaining product acceptability.

## CHAPTER ONE

### 1.0 INTRODUCTION

Fish is an essential source of food for people both rich and poor as reported by the Food and Agricultural Organization of the United Nation in the year 2000, that an estimate of about one billion people world-wide rely on fish as their source of protein (Saba and Rana, 2023). Fish is of more value compared to other animal sources because of its high-quality protein content (Iheanacho *et al.*, 2017). The quality of fish protein is based on its content of the most essential amino acids and important minerals such as phosphorus, calcium and magnesium (Chibueze *et al.*, 2022).

Fish however are highly susceptible to the process of deterioration and decay immediately they are captured or harvested due to the activities of microorganisms and enzymes contained within the fish and the environment of capture/harvesting and processing. Hence, proper fish handling, processing and preservation needs to be done to prevent the deterioration and damage of fish quality (Oli *et al.*, 2022). To increase storage life of fish, several preservative methods are employed to compensate for its perishability (Fitri 2022) and these preservation techniques prevent fish spoilage, lengthen shelf life by inhibiting activities of spoilage bacteria and metabolic changes that result in the loss of fish quality (Orire *et al.*, 2020). Some traditional fish processing methods such as smoking, drying, salting, frying, fermentation and or combination of these have been used to preserve process and improve shelf stability of fish (Chibueze *et al.*, 2022).

#### 1.1 Justification of the Study

Fish spoils rapidly due to high moisture, protein content, and microbial activity but according to Afolabi & Ogunbanwo in Antimicrobial efficacy of *Piper guineense* and *Allium sativum* on spoilage organisms of dried meat (2019), the use of aromatic peppers

and spices has become a promising natural alternative to synthetic preservatives because of their antioxidant and antimicrobial properties. The active components in these spices such as capsaicin and piperine have been shown to effectively inhibit microbial growth and delay lipid oxidation, which are the two major causes of spoilage in dried fish.

While smoking is widely used for fish preservation, according to Akinwumi *et al.*, (2016) on the Effect of different drying methods on quality characteristics of tilapia fish (*Oreochromis niloticus*) smoking often results in inconsistent drying, potential contamination with polycyclic aromatic hydrocarbons (PAHs) that are carcinogenic, and sometimes undesirable strong smoky flavors. Oven drying, in contrast, allows for precise control of drying conditions, uniform moisture removal, improved hygienic processing and reduced contamination risks.

Studies have been carried out to assess the preservative effects of aromatic peppers Okoro *et al.* (2020) Demonstrated that *Capsicum* spp extract reduced microbial load in smoked catfish during ambient storage and improved the taste of dried seafood products.

This study aims to evaluate these preservative effect of aromatic peppers on *O. niloticus*

Despite the numerous preservative studies, there is scarcity of data on treatments with indigenous and commonly accessible aromatic peppers at **ambient storage** conditions and shelf-life extension using natural preservatives and adequate integration of microbial and sensory analysis in preservation study.

This study aims to fill these gaps by evaluating the effects of aromatic peppers on the organoleptic and microbial of oven dried Nile tilapia (*O. niloticus*) fillets stored at ambient temperature and the findings will contribute to the development of easier, healthier, and preservation techniques that enhance consumer acceptability for fish in developing regions.

## **1.2 Aim of the Study**

The study was aimed at evaluating the effects of aromatic peppers on organoleptic and microbial qualities of oven-dried Nile tilapia (*O. niloticus*) fillets stored at ambient temperature. The objectives of the study were to determine the effects of aromatic pepper species (Scotch Bonnet, Cayenne pepper and bell pepper) on the:

- i. sensory attributes (taste, aroma, colour, and overall acceptability) of oven-dried Nile tilapia fillets stored at ambient temperature.
- ii. microbial loads of oven-dried Nile tilapia fillets stored at ambient temperature.

## CHAPTER TWO

### 2.0 LITERATURE REVIEW

#### 2.1 Fish and Nutrition

Fish is a vital component of human meals and a great source of important nutrients including vitamins, minerals, omega-3 fatty acids, and high-quality protein and depending on the species, fish have different nutritional profiles. Oily fish, such as salmon, mackerel, and sardines, are especially high in omega-3 fatty acids and these fats are essential for brain growth, cardiovascular health, and cognitive performance (Lanjenwa *et al.*, 2024). Eating fish has been linked to a number of health advantages, such as lowering the risk of heart disease, enhancing mental function, and promoting eye health (Lanjenwa *et al.*, 2024). Fish ranks as one of the most consumed foods worldwide, owing to its high nutritional value. It is recommended to consume between three to four servings of fish per week which is 85 grams per serving which amounts to 340 grams per week. Fish as a protein source, provides all essential amino acids such as tryptophan, lysine, methionine, and threonine which enhances digestibility. Additionally, fish is notable for its lipid composition, being rich in polyunsaturated fatty acids (PUFAs) like eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA), and docosapentaenoic acid (DPA), which offers cardiovascular and other health benefits. Fish and fishery products, are excellent sources of vitamins A, B-complex, D, and E, as well as minerals such as calcium, phosphorus, iodine, zinc, iron, and selenium (Horando *et al.*, 2024). Fish contains the approximate chemical composition of 75% water, 20% protein, 1–10% lipid, 1–2% minerals, and 0.1–1% carbohydrate (Listrat *et al.* 2016). This nutritional composition varies according to species, size, age, feeding habits, water temperature, etc. (Ahmed *et al.*, 2022). Omega-3 [(n-3)] fatty acids contained in fish have been linked to healthy aging throughout life. Recently, fish-derived omega-3 fatty acids EPA and DHA

have been associated with fetal development, cardiovascular function, and prevention of Alzheimer's disease. However, because our bodies do not efficiently produce some omega-3 fatty acids from marine sources, it is necessary to obtain adequate amounts through fish and fish products. Studies have shown that EPA and DHA are important for proper fetal development, including neuronal, retinal, and immune function. EPA and DHA may affect many aspects of cardiovascular function including inflammation, peripheral artery disease, major coronary events, and anticoagulation. EPA and DHA have been linked to promising results in prevention, weight management, and cognitive function in those with very mild Alzheimer's disease. (Swanson *et al.*, 2012). Fish consumption is also known to have health benefits for adults as consumption of fish lowers the risk of coronary heart disease (CHD) mortality. It is estimated that fish consumption reduces the risk of dying of coronary heart diseases by up to 36% resulting from the long-chain omega-3 fatty acids found in them. CHDs are a global health problem affecting all populations. A daily intake of 250 mg EPA+DHA per adult gives optimal protection against CHD<sup>3</sup> and for optimal brain development in children, the daily requirement is only 150 mg per day (Toppe, 2014). Aquatic foods can also improve human health by reducing micronutrient deficiencies that can lead to subsequent disease. Micronutrient deficiencies account for an estimated 1 million premature deaths annually. Nutrient-rich aquatic foods could provide food-based approaches to reducing nutrient deficiencies, with increasing access and consumption offering many advantages over nutrient supplementation for calcium, iron, zinc and vitamin. A from fish could provide a considerable proportion of the recommended dietary allowance (RDA) for adults and children under 5-years old (Hicks *et al.*, 2019)

### **2.3 Nile Tilapia (*Oreochromis niloticus*)**

Nile Tilapia (*O. niloticus*) is a popular and widely consumed aquatic product due to their distinct flavour and nutritional value (Elyana *et al.*, 2018). It is a rich source of essential amino acids, proteins approximately 16 g per 100 g, minerals like calcium, phosphorus, magnesium, sodium, and iron, and vitamins A, C, D, E, K, B6, and B12 (Penarubia *et al.*, 2022). Its by-products can also be processed into protein hydrolysates with potential uses in food, health, and pharmaceutical industries (Roslan *et al.*, 2014).



**Plate 1: Nile Tilapia (*Oreochromis niloticus*)**

## **2.4 Fish Spoilage**

Food spoilage means alteration of the original nutritional value texture and flavour making it harmful and unsuitable for consumption (Adenike, 2014). Fish spoilage is due to chemical, enzymatic or microbial activities (Ghaly *et al.*, 2010). One-fourth of the world's food supply and 30% of landed fish are lost through microbial activity alone. Fish spoilage results from three basic mechanisms: Enzymatic autolysis, oxidation, microbial growth. Low temperature storage and chemical techniques for controlling water activity, enzymatic, oxidative and microbial spoilage are the most common in the industry today (Ghaly *et al.*, 2010) and the resulting effects of these chemical and microbial activities include degradation of protein, development of oxidative rancidity, vitamin degradation, enzymatic reaction, and the action of microorganisms (Adenike,

2014). Spoilage microorganisms like *Pseudomonas*, *Bacillus*, *Aspergillus*, and *Penicillium* contribute to the degradation of food quality in texture, taste, and odour that makes up the organoleptic properties. Spoilage in fish is usually accompanied by change in physical characteristics, change in colour, odour, texture, colour of eyes, colour of gills and softness of the muscle (Getu and Misganaw, 2015).

#### **2.4.1 Enzymatic Spoilage**

Autolytic activities by endogenous enzymes of seafoods result in products that initially cause loss in the characteristic fresh odour and taste of fish and then softens the flesh. These changes start short after the death of seafood animals and progresses to produce a number of volatile compounds which give the products their spoilage characteristics (Mahmud *et al.*, 2018).

#### **2.4.2 Oxidative Spoilage**

Lipid oxidation is a major cause of deterioration and spoilage for fish and typically involves the reaction of oxygen with the double bonds of fatty acids (Ikape *et al.*, 2017). Therefore, fish lipids which consist of poly unsaturated fatty acids are highly susceptible to oxidation and during this process, lipase split the glycerides forming free fatty acids which are responsible for the off flavour, frequently referred to as rancidity, reducing the oil quality in the fish. (Ikape *et al.*, 2017).

#### **2.4.3 Microbial Spoilage**

Microbial growth and metabolism is a major cause of fish spoilage which produce amines, biogenic amines such as putrescine, histamine and cadaverine, organic acids, sulphides, alcohols, aldehydes and ketones with unpleasant off-flavors (Ikape *et al.*, 2017). According to Gram and Huss, the high composition of non-protein nitrogen compounds and low acidity (pH>6) of the flesh of seafoods are the major cause of their spoilage, as these condition favor the growth of spoilage microorganisms. These

microbes in turn produce metabolites that affect the organoleptic properties of the products and render them undesirable attributes (Mahmud *et al.*, 2018).

## **2.5 Shelf Life**

Various food products have different spoilage times that are referred to as shelf-life (Humaid and Jamal, 2014). Components of fish body decompose and new compounds formed during spoilage, which lead to protein degradation and lipid oxidation, as well as changes in fish odour, flavour, and texture (Mei *et al.*, 2019). Therefore, it is of importance to develop effective treatment methods to preserve the quality and extend the shelf life of fish (Sarker *et al.*, 2021).

## **2.6 Fish Processing and Preservation**

Fish is highly perishable and the deterioration process begins immediately after catch, since the bacterial growth and invasion on the fish are prevented by the body's natural defense system during life but after death the defense system breaks down and the bacteria multiply and invade the flesh (Abolagba and Uwagbai, 2011). To this effect, several processing methods and preservatives commonly artificial preservatives have been employed in extending the shelf life for longer periods. However, due to the adverse reactions to these preservatives, consumers have expressed disagreement with the utilization of additives and so natural preservatives have been analyzed as suitable alternatives for sustainable food preservation (Sulieman *et al.*, 2016).

### **2.6.1 Fish Processing**

The term "fish processing" refers to the series of procedures that take place with fish and fish products in the period between when the fish are caught or harvested and when the final products are sold to consumers. The purpose of fish processing and preservation is to deliver fish that is consumable to the final consumer (Santiago *et al.*, 2024). Fish processing is necessary because, in spite of the nutritional importance of fish, it is highly

perishable especially in the hot climatic regions where average temperature is warm enough to enhance proliferation of mycodeteriogens (Abiala *et al.*, 2020). Where there is no access to refrigeration or ice, these hot climatic regions emphasizes on the physical, chemical and biological processes that lead to spoilage and deterioration of freshly caught fish. However, reducing moisture content through drying, smoking or curing will result in stable source of protein that can be transported consumers (Sulieman *et al.*, 2012).

Fish and seafood preservation is a crucial concern for the food industry, as they are excellent culture media for microorganisms due to intrinsic and extrinsic factors, such as high nutritional composition, water content, and pH, which favor microbial growth (Pierozaan *et al.*, 2023). To prolong the shelf life of fish, it is preserved by many processes including oven drying, sun drying, solar drying, canning and smoking among others (Babarinde *et al.*, 2016). Dried fish is a major component of harvested fisheries in many countries including Nigeria (Food and Agricultural Organization, 2006). About 25 to 30% of the world fish catch is consumed in the dried, salted, smoked form or combination of these processes (Aliya *et al.*, 2012).

### **2.6.2 Fish Handling**

Post-harvest, handling, processing and transportation of fish require particular care in order to ensure proper quality and safety. Retaining the nutritional values of the fish, preserving the benefits of its rich composition and avoiding costly and debilitating effects of fish-borne diseases are vital. Many different techniques have been used to preserve fish quality and to increase shelf life. Techniques are designed to inhibit or reduce the metabolic changes that lead to fish spoilage by controlling specific parameters of the fish and/ or its environment (FAO, 2009). Fish handling and processing includes

bleeding, gutting, beheading, filleting, skinning and trimming before fillets are bought by consumers (Penarubia *et al.*, 2022).

### **2.6.2.1 Gutting**

Fish quality and shelf life can be increased much more if they are bled and the viscera removed, limiting access to most spoilage bacteria. The gutting process, therefore, destroys the reservoir of these digestive enzymes and bacteria there by reducing the process of autolysis in the fish (Santiago *et al.*, 2024).

### **2.6.2.2 Filleting**

Filleting is the removal of bone from fish by separating the flesh (fillets) from the head, backbone, and internal organs, usually resulting in two boneless or semi-boneless pieces. This method enhances the presentation, ease of cooking, and consumer preference, especially in value-added fish products Akinneye *et al.* (2007). Changes in consumer consumption expectations as well as the development of reliable cold-chains, has made fresh pre-processed fishery products such as fillets more popular among consumers because of their convenience for processing and cooking. Value addition through production of fish fillets has been an effective strategy to increase consumer acceptability and commercial values to ensure better utilization of tilapia (Sarker *et al.*, 2021). However, the filleting process results in a significant amount of by-products that can be converted to nutritious low-cost fish products. By-products containing heads, frames and belly flaps, and parts of the viscera like liver and roe are good sources of high quality proteins, lipids with long-chain omega-3 fatty acids, micronutrients like vitamins A, D, riboflavin (B2) and niacin (B3) as well as minerals such as iron, zinc, selenium and iodine (Olsen *et al.*, 2014). The fillet yields in industrial processing is species-dependent and may leads to a significant removal of parts of the fish, such as heads, bones, guts or by-products (Penarubia *et al.*, 2022).

### **2.6.2.3 Drying**

Drying as a preservative method, expels water from biological materials and lessens their initial volumes, which consequently inactivates enzymes and restricts bacteria growth by reducing the water activity in the food products while preserving the product stability and nutrients, which in turn minimizes spoilage and prolongs shelf life (Cao *et al.*, 2021). Natural drying methods like sun drying is a very simple process where fish are placed in sunlight and allowed to dry until the moisture in their bodies evaporates because of the sun's heat (Kalita *et al.*, 2020). Nonetheless, sun drying is limited to seasonal weather variation and faced with the problem of infestations by insects like blowflies and mites which can cause extensive damage resulting in heavy financial loss. To combat this, fish processors sometimes use insecticides directly on dry fish for the prevention of these insect infestation. When these fish are consumed, these insecticides applied are harmful to human health. In order to minimize the sun drying related problems some improved drying technology such as oven drying was developed to process fish. The oven being an enclosed drying system can prevent insect infestation on dried fish product and improve the hygienic and nutritional status of the dried fishery products (Jahan *et al.*, 2018) as opposed to more primitive alternatives like sun drying.

#### **2.6.2.3.1 Sun Drying**

Traditionally, preserving fish involves the action of the sun and wind to effect drying by evaporation. Sun drying is the simplest and cheapest method of air drying process used for foods. It can be done as open sun drying or solar tent drying. Effective open sun drying depends mainly on the environmental temperature, relative humidity and wind speed. However, products dried using this method, are subjected to insects' and birds infestations (Mahmud *et al.*, 2018)

#### **2.6.2.3.2 Smoke Drying**

Two methods of smoking are practiced for fishery products, cold smoking and hot smoking, depending on the temperature used. Products treated with cold smoking is carried out at around 30°C following salting and drying treatment of fish muscle. Hot smoking on the other hand, involves treatment at a temperature range of 70–80°C and results in cooked product that do not require subsequent processing (Mahmud *et al.*, 2018). Most consumers of fish in Nigeria consume smoked fish. It is relished for its taste and aroma as well as longer shelf life as a result of the combined effects of dehydration, antimicrobial and antioxidant activities of several smoke constituents mainly: formaldehyde, carboxylic acids and phenols.

#### **2.6.2.3.3 Oven Drying**

Oven-drying has been shown to significantly enhance the nutritional, chemical, and microbiological quality of dried fish compared to traditional methods such as sun drying and open rack drying. Studies indicate that while advanced techniques like mechanical drying, solar tent drying, solar tunnel drying, low-temperature vacuum drying, and freeze-drying yield high-quality products, electric oven-drying stands out for its ability to improve both product quality and shelf life. Specifically, electric oven-drying helps preserve protein quality and reduces lipid oxidation, thereby maintaining the overall nutritional integrity of the fish (Santiago *et al.*, 2024). In recent times, smoking kilns and artificial dryers are used to obtain products of high quality (Omodara *et al.*, 2012). According to Jahan *et al.*, 2018, solar drying produced better quality dried fish compared to that of sun drying due to reduction in insect infestation and other contaminants.

#### **2.6.3 Fish Preservation**

Artificial preservatives have played a crucial role in extending the shelf life and preserving the quality of fish food for prolonged periods (Sulieman *et al.*, 2016). To maintain the quality, several synthetic preservatives like butylated hydroxytoluene

(BHT), tert-butylhydroquinone (TBHQ), and butylated hydroxy anisole (BHA) (Santiago *et al.*, 2014) are added to food products to prevent changes in texture and colour, development of unpleasant flavour and rancid odour, and loss of nutrients of seafood during storage. However, the use of these preservatives have been linked to potential health hazards which include, biological toxicity and carcinogenic consequences. Hence, natural preservatives with excellent antioxidant and antimicrobial properties have been extensively searched and implemented as safe alternatives in seafood processing, with the sole purpose of extending shelf-life (Olatunde and Benjakul, 2018). The polyphenols or phenolic compounds present in the plant are important natural antioxidants. Also, phenolic extracts from vegetables, cereals grains, fruits, and spices have positive effects in preventing lipid oxidation (Santiago *et al.*, 2024).

#### **2.6.3.1 Aromatic Pepper Spices**

Spices are known for their antimicrobial values and potentials as food preservatives. Among the spices, **aromatic peppers** stand out due to their rich content of bioactive compounds such as capsaicinoids and flavonoids, which contribute to their strong antimicrobial and antioxidant properties. (Srinivasan, 2014).

#### **2.7 Properties and Composition of Pepper Spices**

Through extensive research, aromatic spices have been proven to have good preservative qualities due to the antioxidant and antimicrobial compounds in them. Pepper belongs to the Solanaceae family, it is the world's most produced and consumed vegetable and considered one of the oldest food additives in human nutrition. Peppers contain aromatic compounds like alcohol, aldehyde, and ester. Phenolic compounds contained in aromatic peppers positively affect human health and significantly influence food quality factors such as colour, taste, and aroma (Polat *et al.*, 2024). The antimicrobial and antioxidant properties of spices and herbs play an important role in food preservation (Srinivasan,

2014). Flavonoids, phenolic compounds, sulfur-containing compounds, tannins, alkaloids, phenolic diterpenes and vitamins are some of the bioactive components that make up the antioxidant properties of spices. The ability of spices to prevent the formation of free radicals, destroy them, repair oxidative damage and get rid of damaged molecules is what gives them their antioxidant properties (Carlsen *et al.*, 2010). Some species of pepper spices commonly used in fish food preservation and processing include:

### **2.7.1 Scotch Bonnet (*Capsicum chinese*)**

The mostly consumed pepper, appreciated for its colour, flavour, and aroma. It is used as food ingredients both industrially and in traditional cuisine (Mariano *et al.*, 2022). Scotch bonnet pepper presents significant health-promoting potentials such as, supporting digestion, improving blood flow etc. This is mainly attributed to bioactive compounds such as flavonoids, capsaicinoids, carotenoids, contained in them. (de Aguiar *et al.*, 2019).



**Plate 2: Scotch Bonnet (*Capsicum chinese*)**

### **2.7.2 Cayenne pepper (*Capsicum anuum*)**

A fruit-vegetable commonly found in human daily food, as they are extremely popular for the huge content of vitamin C and total soluble phenolics compounds, higher than other vegetables commonly recognized as a source of this substance. Scientific research has proven that, *capsicum annum*, is the only crop that produce alkaloid compound called capsaicinoid responsible for its hot taste. Fresh Cayenne pepper is a very good

source of vitamin C and E as well as provitamin A and carotenoids. Also, it is known for antioxidant properties. Vitamin C, including ascorbic acid and its oxidation product (dehydroascorbic acid), has many biological activities in the human body due to its antioxidant property



**Plate 3: Cayenne Pepper (*Capsicum annum*)**

### **2.7.3 Bell pepper (*Capsicum annuum*)**

In human consumption, bell peppers are an important source of nutrient such as in providing carotenoids, phenols, vitamin C, foliates. In peppers, there are phytochemical property that have many biochemical and pharmacological properties which includes antioxidants, anti-inflammatory, antiallergenic and anti-carcinogenic activities (Lee *et al.*, 2005). It has been proven that, ripe red peppers can reduce the risk of cancer (Nishino *et al.*, 2009) and considering the antimicrobial and antioxidant attributes found in bell pepper, investigations have explored their potential utility as antimicrobial agents for food preservation, with the aim of managing foodborne pathogens and mitigating product spoilage (Sulieman *et al.*, 2023).



## **Plate 4: Bell pepper (*Capsicum annum*)**

### **2.8 Application of Aromatic Pepper Spices in Fish Preservation**

Aromatic pepper contain alkaloid (e.g. piperine), terpenes, flavones and volatile oils (e.g. piperlyne) that exhibit detoxification, hypotensive, and anticancer activities (Butt *et al.*, 2012; Meghwal and Goswami, 2013; Yoon *et al.*, 2015). Pepper is also used as a preservative and flavour enhancer in meat and meat-based products (Thiel *et al.* 2014).

Polyphenols are natural antioxidants abundant in herbs that demonstrates antioxidant and antimicrobial effects in various fish and fish products. Phenolic compounds serve as processing aids for texture modification in fish fillets (Maqsood *et al.*, 2013). Oxidation affects pigments, flavours, lipids, proteins, and vitamins in foods, leading to spoilage, particularly on polyunsaturated fatty acids (PUFA), which results in unpleasant rancid odours and harmful secondary oxidation products.

Protein and lipid oxidation contribute to the deterioration of nutritional values, sensory qualities, and water-holding capacity in food products (Suleman *et al.*, 2020). In pepper spices, the antioxidant properties of polyphenolic compounds in herbs and spices have substantial effects against bacteria, yeast, mould, and fungi (Kurutas, 2016).

### **2.9 Organoleptic Properties of Fish**

The term organoleptic is based on the smell, taste organs, tongue and olfactory system. Sensory evaluation takes into account all the senses and physiological systems that are associated with each one. Basic tastes are perceived on the tongue and aroma is perceived by the olfactory system. Sensory evaluation also considers texture which is perceived through multiple systems of the body. Organoleptic properties are the aspects of food, water or other substances creates a unique experience via the senses including taste, sight, smell and touch (Zhou *et al.*, 2016; Chuahan and Sharma 2003)

## **CHAPTER THREE**

### **3.0 MATERIALS AND METHODS**

#### **3.1 Collection of Fish Samples**

Fresh Nile tilapia (*Oreochromis niloticus*) was obtained from a reputable fish vendor in Yanga Market Benin City. A total of 5 samples of uniform size weighing 200g each was purchased and these were transported in sterile polythene containing ice chips to the laboratory.

#### **3.2 Collection of Pepper Samples**

The pepper spices used for this study include the Scotch bonnet (*Capsicum chinese*), Cayenne pepper (*Capisicum anuum*), and Bell pepper (*Capsicum anuum*). These were purchased in their fresh form from Oba market in Oredo Local government Area, Benin City.

#### **3.3 Identification of Fish Sample**

The Nile tilapia (*O. niloticus*) was identified in the market based on its distinct morphological characteristics. The Nile tilapia (*O. niloticus*) used in this study, possesses a laterally compressed body covered with large and shiny scales. Its body coloration is generally grayish or silvery with 6 to 9 dark vertical bands visible along the sides, and the belly is whitish or light gray. The dorsal fin is long and continuous, extending nearly to the tail, which is rounded and marked with faint vertical stripes.

#### **3.4 Electric Oven Type**

The electric oven consists of a metallic, thermally insulated chamber that minimizes heat loss and maintains a stable internal temperature. A compact countertop electric oven of standard Century model was employed for the oven-drying process. The oven features adjustable temperature and timing controls, providing a stable enclosed heating

environment suitable for uniform dehydration and prevention of insect contamination during drying.

### **3.5 Processing Procedures**

#### **3.5.1 Preparation of Pepper Samples**

Each of the three pepper species (Scotch bonnet, Cayenne and bell pepper) were sorted separately, to ensure that only fresh and healthy ones were used. These were washed thoroughly in clean water to remove dust, dirt, and other possible contaminants. After washing, each of the pepper samples were blended separately with 2ml of water into a semi-fine paste, then sieved to reduce moisture content and the paste placed in sterile beakers.

#### **3.5.2 Preparation of Fish Samples**

The fish samples were washed thoroughly with clean tap water to remove all adhering debris. Each fish was then descaled manually using the blunt knife end and filleted by cutting along the dorsal line from just below the gill cover, following the backbone toward the tail using a sterile stainless-steel knife, which yielded two symmetrical fillets from each fish. The fillets were further trimmed to remove any remaining bones, fins, or skin to obtain uniform, boneless portions and rinsed thoroughly to ensure complete removal of viscera and blood residues making it suitable for subsequent treatment and drying.

The cleaned and drained fillets were then weighed using a sensitive scale to give an average weight of 60.3g per fillet and divided into 4 portions of 150g each before being subjected to the pepper spice treatments. The fish Samples were treated with spices equivalent to 2% of their fresh weight in this study as this concentration has been reported to effectively inhibit microbial growth and delay lipid oxidation without adversely affecting sensory quality (Shamima *et al.*, 2007). The portions were marinated

in four different bowls, the first was control and so it was marinated in distilled water only, the second portion was marinated in Scotch Bonnet pepper solution of 1.206g pepper which is 2% of the fillet weight and 100ml of water, the third portion was marinated in Cayenne pepper solution of 1.206g pepper and 100ml of water while the fourth portion was marinated in Bell pepper solution of 1.206g pepper and 100ml of water.

All portions were marinated for 1 hour and flipped on either side every 15 minutes to ensure even absorption.



**Plate 5: Tilapia Fillets**



**Plate 6: Blended and Sieved Pepper Species**

### **3.5.3 Drying Process**

The fish samples that were treated with the pepper spices were carefully arranged on the racks of the oven in a way that prevented contamination by touch or dripping from other treatments. The drying process was carried out for about 5 hours at 100°C internal temperature till a constant weight was achieved. Periodically, during smoking, the fish samples were checked and flipped so as to allow even distribution of heat and after smoking, the fish samples were removed from the oven and left to cool.

### **3.5.4 Storage**

After drying, the fillets were left to cool at room temperature, packed in Ziploc bags and sealed to prevent absorption of moisture and microbial impact from the atmosphere

### **3.5.5 Processed fish sample analysis**

Fresh and treated oven-dried *O. niloticus* were taken to the laboratory and analyzed to check for microbial loads. Bacterial and fungal analyses were carried out on the samples. The stored fish was subjected to a weekly analysis of bacterial and fungal load for a period of three weeks.

## **3.6 Microbial Analysis**

### **3.6.1 Materials**

Gloves, nose mask, petri dishes, absolute ethanol (sterilizing agents), autoclave, masking tapes, marker pens, McCartney bottles, paper towels, pipette, wash bottle, test tubes, measuring scale, microscope, disinfectant were the materials used for microbial analysis.

### **3.6.2 Sterilization of materials**

All containment materials such as pipettes, test tubes and wash bottles used in this study were thoroughly washed in detergent and rinsed in clean water and allowed to drip-dry to ensure that they were grease free. The glass wares were properly sterilized in an

autoclave at 121°C for 15 minutes. Commercial Petri dishes which had been sterilized was used.

### **3.6.3 Method**

For the microbial analysis of the samples, the method adopted were that as described by Roberts (1978). Bacteria and fungi were isolated for confirmation, thus Nutrient agar and Potato dextrose agar were prepared using the manufacturer's instructions.

### **3.6.4 Nutrient Agar**

Fourteen grams (14g) of Nutrient agar was mixed into 500ml of distilled water and stirred to ensure proper mixing. The mixture was autoclaved at 121°C for 15 minutes and then allowed to cool to 45°C. The media was poured into an already inoculated Petri dish; the plates were inoculated with pasture pipette and incubated at 37°C for 24 hours, allowed to solidify and then inverted. Nutrient agar (NA) plated were inverted during the incubation to avoid contamination by water of condensation which will merge colony forming unit. Nutrient Agar was used for the isolation of bacteria.

### **3.6.5 Potato Dextrose Agar**

Potato dextrose agar was used for the isolation of fungi according to Robert (1978). The medium was prepared by dissolving 20g of potato dextrose agar (PDA) into distilled water into a 500ml conical flask, after which it was stirred to ensure proper mixing, the mixture was then autoclaved 121°C for 15 minutes and allowed to cool for 45°C. PDA was poured into already inoculated sterile disposable Petri dishes and allowed to solidify.

### **3.6.6 Isolation and Enumeration of Bacteria and Fungi**

The serial dilution method of Harrigan and McCance (1976) was aseptically carried out in tubes. Sterile test tubes were used for the ten-fold dilution. Test tubes labelled:  $10^{-1}$ ,  $10^{-2}$ ,  $10^{-3}$ ,  $10^{-4}$ , were used for each of the samples and one gram (1.0 gm.) of each of the samples was weighed out, mixed with nine milliliters of sterile distilled water into a test

tube. One milliliter (1.0 ml) of the aliquot was obtained from each of the samples and transferred into the test tube labelled  $10^{-1}$  and mixed properly. One milliliter (1.0 ml) aliquot was then transferred serially from the tube ( $10^{-1}$ ) to tubes labeled  $10^{-2}$ ,  $10^{-3}$ , and  $10^{-4}$ , in that order. This was done for each of the samples A B, C and D at the end of each serial dilution, 1.0 ml left in the pipette tip was discarded. Aliquots from the appropriate tubes were then used to inoculate appropriate media for isolation of fungi using the pour plate method (Bourbonnais, *et al.*, 1995)

The pour plate technique was used for the isolation of fungi during the study. The agar media used were prepared according to manufacturers' instruction. The plates were labelled appropriately, and with the aid of a 0.1 ml pipette aliquots of the appropriate dilutions were inoculated into the agar plates. The agar plates were then incubated at room temperature ( $28\pm 2^{\circ}\text{C}$ ) for 3- 5 days.

#### **3.6.6.1 Identification and Characterization of Isolates**

The identification of fungi isolates was based on their morphological and cultural characteristics of Barnet and Hunter, 1974. Agar cultures of each isolate were used in determining their cultural characteristics. 3 to 5 days old culture were examined to study their cultural and morphological characteristics and the observations made were on features such as growth pattern, colony margin, reverse plate appearance, spore shape and attachment, pigmentation of spores and texture (Boussard. *et al.*, 2012).

#### **3.6.6.2 Morphological Test**

Morphological characteristics were examined under a microscope.

### **3.7 Organoleptic (Sensory) Evaluation**

Sensory evaluation was out on days 0 and 7, by 20 semi-trained panelists made up of students and staff of the Aquaculture and fisheries management department who were selected to evaluate aroma, taste, and texture for their familiarity with dried fish products.

The attributes assessed included Colour, aroma, taste, texture, and overall acceptability and were evaluated using the 5-Point Hedonic scale. The dried fillet samples were coded and presented randomly to avoid bias. Water and crackers was provided for rinsing their palates between samples.

### **3.8 Experimental Design**

The experimental design was made up of two factors;

- i. Pepper treatments (Scotch bonnet, Cayenne and Bell pepper)
- ii. Storage duration (1 week, 2 weeks and 3 weeks)
- iii. The experimental design was therefore to be three pepper specie (Scotch bonnet, Cayenne and Bell pepper) x 3 storage times (1 week, 2 weeks and 3 weeks) factorial, in a completely randomized design (CRD). Experimental trials were replicated three times.

### **3.9 Statistical Analysis**

Analysis of Variance table (ANOVA) was performed to ascertain differences between the oven-dried fillet samples with different pepper treatments (Scotch bonnet, Cayenne and Bell pepper) and the control. Data Analysis was done using GenStat software version 12.1. All analysis was duplicated three times using Ducan Multiple Range Test (DMRT).

## CHAPTER FOUR`

### 4.0

### RESULTS

#### **4.1 Effects of Pepper spices on the Mean Organoleptic qualities of dried Tilapia fillets**

Table 1 shows the effects of different pepper spices on the sensory attributes (appearance, aroma, flavour, taste, and overall acceptability) of oven-dried *O. niloticus* fillets. The mean sensory scores ranged between 2.6 and 3.3 on a 5-point hedonic scale, suggesting moderate consumer acceptability.

In terms of appearance, fillets treated with cayenne and scotch bonnet peppers recorded the highest mean scores (3.3), followed by the control (3.2) and bell pepper (3.0). The aroma was also highest in samples treated with cayenne pepper (3.1), followed by the control (3.0), while bell pepper and scotch bonnet pepper treatments both had identical mean values (2.9). These variations, however, were not statistically significant ( $p > 0.05$ ). Furthermore, flavour and taste scores were relatively low (2.6-2.9) across all treatments, and the differences observed were likewise not statistically significant ( $p > 0.05$ ). Although spiced samples appeared slightly more appealing (3.0-3.3) than the control (3.2), the mean differences were within the LSD benchmark, confirming that the variations among treatments were statistically insignificant ( $p > 0.05$ ).

#### **4.2 Effects of Storage Periods and Pepper Spices Treatments on the Mean Organoleptic Qualities of Treated Dried Tilapia Fillets**

The effects of different pepper spices and storage periods on the organoleptic quality of treated dried tilapia fillet as shown in table 2 indicates that on Day 1, both treated and untreated samples recorded high sensory scores, ranging from a mean score of 3.7 to 4.6 on a 5-point hedonic scale, indicating very good acceptability. The control and Cayenne pepper-treated fillets samples had the highest aroma scores (4.6 and 4.5, respectively), while Bell pepper and Scotch bonnet pepper treatments also showed

**Table 1: Effects of Pepper spices on the Mean Organoleptic qualities of oven-dried Tilapia fillets**

Seasoning	Parameters				
	Appearance	Aroma	Flavour	Overall Acceptability	Taste
Bell Pepper	3.0 <sup>a</sup>	2.9 <sup>a</sup>	2.9 <sup>a</sup>	2.9 <sup>a</sup>	2.8 <sup>a</sup>
Cayenne Pepper	3.3 <sup>a</sup>	3.1 <sup>a</sup>	2.7 <sup>a</sup>	3.1 <sup>a</sup>	2.8 <sup>a</sup>
Control	3.2 <sup>a</sup>	3.0 <sup>a</sup>	2.7 <sup>a</sup>	2.9 <sup>a</sup>	2.7 <sup>a</sup>
Scotch Bonnet Pepper	3.3 <sup>a</sup>	2.9 <sup>a</sup>	2.6 <sup>a</sup>	2.8 <sup>a</sup>	2.6 <sup>a</sup>
LSD	0.4425ns	0.3936ns	0.4475ns	0.2988ns	0.3105ns

good sensory appeal, particularly in appearance (4.2 and 4.3 respectively). All samples however, exhibited high overall acceptability scores, with mean values ranging from 4.0 to 4.3 but the differences among treatments were not statistically significant ( $p > 0.05$ ). After one week (7 days of storage), the sensory parameters were observed to decline significantly ( $P < 0.05$ ) across all treatments. The appearance scores dropped in range to 1.8–2.4, while aroma decreased to 1.5–1.8. In addition, flavour (1.6 across all samples), taste (1.6–1.7), and overall acceptability (1.6–2.0) values followed the same downward trend. The cayenne pepper-treated fillets retained relatively better sensory attributes in overall acceptability (2.0) than the control (1.7), scotch bonnet and bell pepper (1.6 each). Analysis of variance (ANOVA) revealed that both storage period and spice treatment significantly influenced ( $p < 0.05$ ) the organoleptic attributes of oven-dried *O. niloticus* fillets.

### **4.3 Effects of Storage Period per Week on the Overall Mean Organoleptic Qualities of Dried Tilapia Fillets**

In Table 3 reveals the mean sensory ratings of oven-dried *O. niloticus* fillets treated with different pepper spices and stored under ambient conditions for two weeks.

At Day 1, all samples recorded high sensory scores across all parameters, with mean scores ranging from 3.8 to 4.6. Specifically, appearance scores were highest in cayenne pepper, control, and scotch bonnet pepper treatments with identical mean scores (4.3), while bell pepper recorded a slightly lower score (4.2). Aroma scores ranged from 4.3 to 4.6, with the control sample showing the highest value (4.6). Flavour ratings varied between 3.7 and 4.2, and overall acceptability ranged from 4.0 to 4.3, with bell pepper achieving the highest panel rating (4.3). Taste values were similarly high across

**Table 2: Effects of storage periods and Pepper Spices Treatments on the Mean Organoleptic qualities of dried Tilapia fillets**

Day	Treatment	Appearance	Aroma	Flavour	Overall	
					Acceptability	Taste
1	Bell Pepper	4.2 <sup>a</sup>	4.4 <sup>a</sup>	4.2 <sup>a</sup>	4.3 <sup>a</sup>	4.1 <sup>a</sup>
	Cayenne Pepper	4.3 <sup>a</sup>	4.5 <sup>a</sup>	3.8 <sup>a</sup>	4.2 <sup>a</sup>	4.0 <sup>a</sup>
	Control	4.3 <sup>a</sup>	4.6 <sup>a</sup>	3.8 <sup>a</sup>	4.1 <sup>a</sup>	3.9 <sup>a</sup>
	Scotch Bonnet Pepper	4.3 <sup>a</sup>	4.3 <sup>a</sup>	3.7 <sup>a</sup>	4.0 <sup>a</sup>	3.7 <sup>a</sup>
7	Bell Pepper	1.8 <sup>b</sup>	1.5 <sup>b</sup>	1.6 <sup>b</sup>	1.6 <sup>b</sup>	1.6 <sup>b</sup>
	Cayenne Pepper	2.3 <sup>b</sup>	1.8 <sup>b</sup>	1.6 <sup>b</sup>	2.0 <sup>b</sup>	1.6 <sup>b</sup>
	Control	2.2 <sup>b</sup>	1.6 <sup>b</sup>	1.6 <sup>b</sup>	1.7 <sup>b</sup>	1.7 <sup>b</sup>
	Scotch Bonnet Pepper	2.4 <sup>b</sup>	1.6 <sup>b</sup>	1.6 <sup>b</sup>	1.6 <sup>b</sup>	1.6 <sup>b</sup>
		0.4425	0.3936	0.4475	0.4226	0.4391

treatments, ranging from 3.7 to 4.1. The differences observed among treatments on Day 1 were not statistically significant ( $p > 0.05$ ).

However, by Day 7, there was a statistically significant decline ( $p < 0.05$ ) in all sensory attributes across the treatments. Appearance scores dropped to a range of 1.8–2.4, aroma to 1.5–1.8, and flavour to 1.6 in all samples. Likewise, taste decreased to 1.6–1.7, and overall acceptability reduced to between 1.6 and 2.0.

Analysis of variance (ANOVA) indicated that storage period had a significant effect ( $p < 0.05$ ) on all sensory parameters measure thereby confirming the statistical differences observed between Day 1 and 7.

## **4.2 Result of the Determination of Microbial Load of oven-dried Tilapia fillet**

### **4.2.1 Effects of pepper spices on the Microbial loads of dried Tilapia fillets in $10^{-3}$ Cfu/gm**

In Table 4, the effects of different pepper spices on the microbial load (bacterial and fungal counts) of oven-dried *O. niloticus* fillets is expressed in  $10^{-3}$  cfu/g. The microbial assessment revealed variations in both bacterial and fungal counts among the different spice-treated samples and the control.

The Cayenne pepper-treated samples exhibited the highest bacterial count ( $86.8 \times 10^{-3}$  Cfu/g), significantly ( $P < 0.05$ ) higher than those of other treatments. On the contrary, samples treated with bell pepper ( $7.3 \times 10^{-3}$  Cfu/g) and Scotch bonnet pepper ( $11.1 \times 10^{-3}$  Cfu/g) recorded lower bacterial counts, suggesting that these spices possessed better antimicrobial effects compared to cayenne. The fresh sample ( $25.3 \times 10^{-3}$  Cfu/g) and the control ( $44.4 \times 10^{-3}$  Cfu/g) showed moderate bacterial loads, indicating that spice addition reduced bacterial growth compared to untreated samples.

Similarly, the fungal counts ranged from  $20.7 \times 10^{-3}$  to  $41.3 \times 10^{-3}$  Cfu/g. The lowest fungal count was recorded in the fresh sample ( $20.7 \times 10^{-3}$  Cfu/g), while the control had the highest fungal growth ( $41.3 \times 10^{-3}$  Cfu/g). Among the spice treatments,

**Table 3: Effects of storage periods per week on the overall Mean Organoleptic qualities of dried spiced Tilapia fillets**

Storage Period	Parameters				
	Appearance	Aroma	Flavour	Overall Acceptability	Taste
Day1	4.3 <sup>a</sup>	4.4 <sup>a</sup>	3.8 <sup>a</sup>	4.1 <sup>a</sup>	3.9 <sup>a</sup>
Day 7	2.2 <sup>b</sup>	1.6 <sup>b</sup>	1.6 <sup>b</sup>	1.7 <sup>b</sup>	1.6 <sup>b</sup>
LSD	0.3129	0.2783	0.3164	0.2113	0.2196

Cayenne, Bell Pepper, and Scotch Bonnet recorded moderate fungal loads ( $24.7\text{--}28.0 \times 10^{-3}$  Cfug), suggesting that the spices were effective in suppressing fungal proliferation during storage.

The Least Significant Difference (LSD) values for bacterial (46.4) and fungal counts (5.356) indicate statistically significant differences among treatments ( $P < 0.05$ ).

#### **4.2.2 Effects of Storage Period and Treatments on the Mean Microbial Loads of Dried Tilapia Fillets**

Table 5 shows the effects of different pepper spice treatments and storage durations on the bacterial and fungal loads of oven-dried *O. niloticus* fillets, expressed in  $10^{-3}$  Cfug. The microbial counts increased progressively with storage time across all treatments, indicating that storage duration significantly ( $P < 0.05$ ) influenced microbial proliferation in the dried fish samples.

At day 0, the bacterial counts were generally low across all treatments, ranging from  $5.3 \times 10^{-3}$  Cfug in bell pepper-treated samples,  $6.0 \times 10^{-3}$  Cfug in scotch bonnet pepper-treated samples to  $7.0 \times 10^{-3}$  Cfug in cayenne-treated samples. The control sample also recorded a low count ( $6.3 \times 10^{-3}$  Cfug), reflecting effective drying and lower initial contamination. Similarly, the fungal counts ranged from  $8.0 \times 10^{-3}$  to  $20.7 \times 10^{-3}$  Cfug. By day 7, both bacterial and fungal counts increased moderately across all treatments, with the control sample showing  $11.3 \times 10^{-3}$  Cfug for bacteria and  $47.3 \times 10^{-3}$  Cfug for fungi. Among spiced samples, bell pepper ( $7.0 \times 10^{-3}$  Cfug) and Scotch bonnet ( $10.0 \times 10^{-3}$  Cfug) showed relatively lower microbial loads than cayenne ( $13.3 \times 10^{-3}$  Cfug) and the control ( $11.3 \times 10^{-3}$  Cfug). These variations were not statistically significant ( $P > 0.05$ )

At day 14, there was a marked increase in microbial counts across all treatments. The Cayenne pepper-treated sample had the highest bacterial counts ( $240.0 \times 10^{-3}$  Cfug) and a high fungal counts ( $42.0 \times 10^{-3}$  Cfug), indicating significant spoilage and possible

post-processing contamination. In contrast, Bell pepper ( $9.7 \times 10^{-3}$  Cf/g) and Scotch bonnet ( $17.3 \times 10^{-3}$  Cf/g) treatments maintained comparatively low bacterial loads, the observed differences were statistically significant ( $P < 0.05$ ) suggesting stronger antimicrobial stability. The control sample had high bacterial ( $115.7 \times 10^{-3}$  Cf/g) and fungal ( $61.7 \times 10^{-3}$  Cf/g) counts, confirming that untreated dried fish deteriorates more rapidly under ambient storage conditions.

#### **4.3 Effects of Storage Periods on the Overall Mean Fungal Counts of Treated Tilapia Fillets**

The graph in Figure 7 illustrates changes in the mean fungal counts of oven-dried *O. niloticus* fillets over a 14-day storage period. The fungal population increased progressively with time, from  $12.9 \times 10^{-3}$  Cf/g on Day 0 to  $29.9 \times 10^{-3}$  Cf/g on Day 7, and further to  $41.7 \times 10^{-3}$  Cf/g by Day 14. This continuous rise in fungal counts indicates that storage duration had a significant ( $P < 0.05$ ) effects on fungal proliferation in the dried tilapia samples

#### **4.4 Effects of storage periods on the overall Mean bacterial counts of dried tilapia fillets**

The graph in Figure 8 shows the changes in the mean fungal counts of oven-dried *O. niloticus* fillets over a 14-day storage period. The bacterial population increased progressively with time, from  $10 \times 10^{-3}$  Cf/g on Day 0 to  $13.4 \times 10^{-3}$  Cf/g on Day 7, and further to  $81.6 \times 10^{-3}$  Cf/g by Day 14. This continuous rise in fungal count indicates that storage duration had a significant ( $P < 0.05$ ) effect on the bacterial growth in the dried tilapia samples.

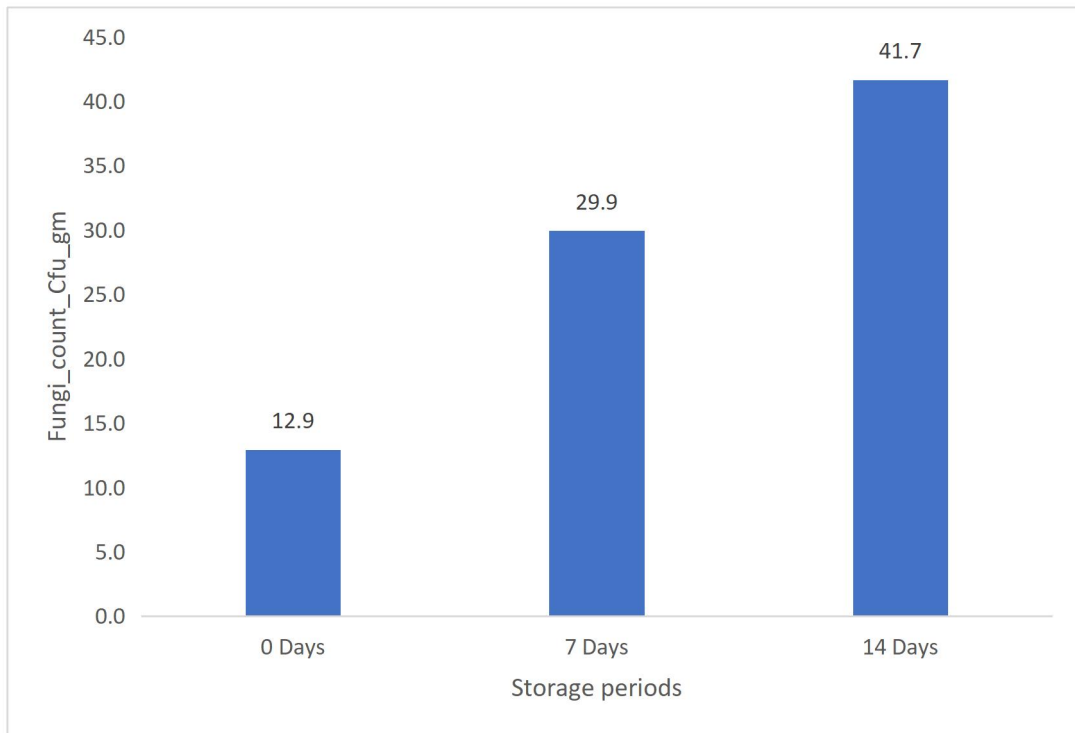
**Table 4 : Effects of pepper spices on the Microbial loads of dried Tilapia fillets in  $10^{-3}$  Cfu/gm**

<b>Treatments</b>	<b>Bacterial counts</b>	<b>Fungi counts</b>
Control	44.4 <sup>ab</sup>	41.3 <sup>a</sup>
Fresh	25.3 <sup>b</sup>	20.7 <sup>c</sup>
Bell Pepper	7.3 <sup>b</sup>	28.0 <sup>b</sup>
Cayenne	86.8 <sup>a</sup>	24.7 <sup>bc</sup>
Scotch Bonnet	11.1 <sup>b</sup>	26.2 <sup>bc</sup>
LSD	46.4	5.356

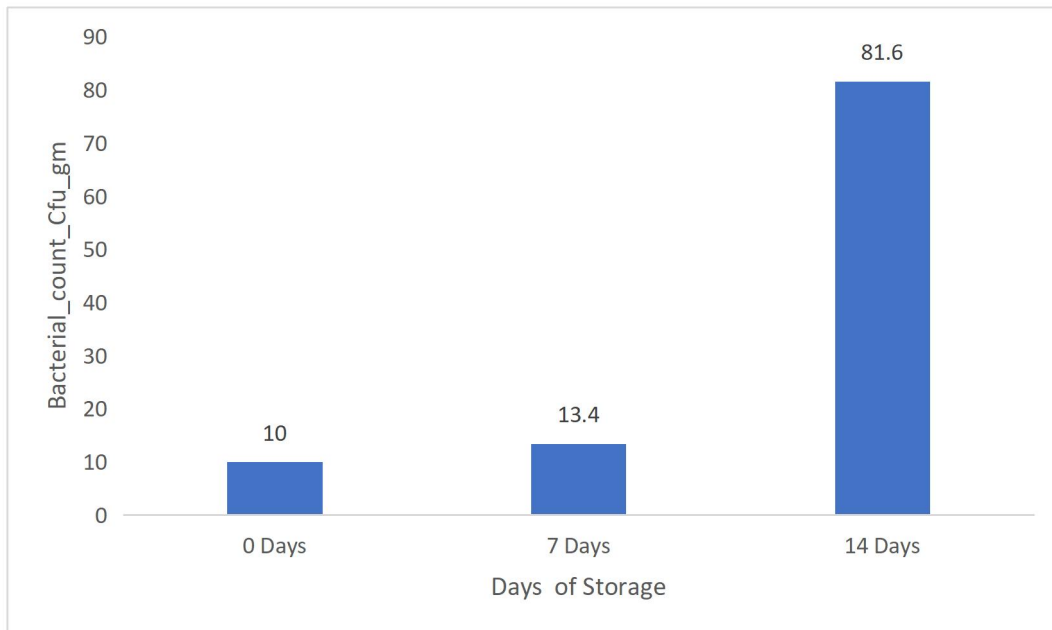
**Table 5: Effects of Storage period and Treatments on the Mean Microbial loads of Oven-dried Tilapia fillets in 10<sup>-3</sup> Cfug/m**

Storage periods	Treatments	Bacterial count	
		Cfu/gm	Fungi count Cfu/gm
0	Control	6.3 <sup>c</sup>	15.0 <sup>fghij</sup>
	Fresh	25.3 <sup>c</sup>	20.7 <sup>efgh</sup>
	Bell Pepper	5.3 <sup>c</sup>	8.0 <sup>j</sup>
	Cayenne	7.0 <sup>c</sup>	10.3 <sup>gi</sup>
	Scotch Bonnet	6.0 <sup>c</sup>	10.7 <sup>ghj</sup>
7	Control	11.3 <sup>c</sup>	47.3 <sup>b</sup>
	Fresh	25.3 <sup>c</sup>	20.7 <sup>efg</sup>
	Bell Pepper	7.0 <sup>c</sup>	31.7 <sup>cd</sup>
	Cayenne	13.3 <sup>c</sup>	21.7 <sup>ef</sup>
	Scotch Bonnet	10.0 <sup>c</sup>	28.3 <sup>de</sup>
14	Control	115.7 <sup>b</sup>	61.7 <sup>a</sup>
	Fresh	25.3 <sup>c</sup>	20.7 <sup>efghi</sup>
	Bell Pepper	9.7 <sup>c</sup>	44.3 <sup>b</sup>
	Cayenne	240.0 <sup>a</sup>	42.0 <sup>b</sup>
	Scotch Bonnet	17.3 <sup>c</sup>	39.7 <sup>bc</sup>
		80.36	9.276

**Fig. 1: Effects of storage periods on the overall mean Fungi counts of Treated Tilapia Fillets**



**Fig. 2: Effects of storage periods on the overall mean bacterial counts of treated tilapia fillets**



#### 4.5 Microbial Frequency and Diversity of Organisms Isolated from Oven-Dried *O. niloticus* Fillets during Storage

Table 6 presents the total percentage frequency and diversity of bacterial and fungal organisms isolated from the oven-dried *O. niloticus* fillets treated with different pepper species after drying. The samples were coded as: A (Scotch bonnet pepper), B (Bell pepper), C (Cayenne pepper), D (Control), and E (Fresh sample).

A total of 41 microbial isolates were obtained throughout the storage period, comprising 17 bacterial and 24 fungal isolates. This result indicates that both bacterial and fungal species were present on the fish fillets after, although fungal isolates occurred slightly more frequently (58.4%) than bacterial ones (41.5%).

##### 4.5.1 Bacterial Isolates

Five bacterial species were identified across the treatments which included *Proteus sp.*, *Streptococcus epidermidis*, *Micrococcus sp.*, *Staphylococcus aureus*, and *Bacillus sp.* Among these, *Proteus sp.* had the highest frequency of occurrence (5 isolates, 12.2%), followed by *Streptococcus epidermidis* and *Staphylococcus aureus* (4 isolates, 9.8% each) then *Micrococcus sp.* (3 isolates, 7.3%) while *Bacillus sp.* had the lowest (1 isolate, 2.4%). In terms of treatment distribution, bacterial occurrence was highest in the fresh (E) and Scotch bonnet pepper-treated (A) samples, each with 4 isolates (23.5%), followed by the Bell pepper (B), Cayenne (C), and Control (D) samples which each recorded 3 isolates (17.6%).

Bacterial diversity, representing the number of different species present per sample, ranged from 3 to 4 species. The highest diversity (4 species) was observed in the Scotch bonnet (A) and fresh (E) samples, while the others had a slightly lower diversity of 3 species.

#### 4.5.2 Fungal Isolates

A total of seven fungal species were identified which were; *Aspergillus flavus*, *Mucor sp.*, *Mucor mucedo*, *Penicillium oxalicum*, *Penicillium italicum*, *Botrytis sp.*, *Cladosporium sp.*, *Saccharomyces sp.*, and *Cryptomonas sp.*

Among these, *Aspergillus flavus* recorded the highest frequency of isolation (5 occurrences, 12.2%), followed by *Penicillium oxalicum* (4 occurrence, 9.8%), then *Penicillium italicum* and *Mucor sp.* (3 occurrence, 7.3%), *Mucor mucedo* (2 occurrence, 4.8%) while *Botrytis sp.*, *Cladosporium sp.*, and *Cryptomonas sp.* each had the lowest (1 isolate, 2.4%).

Fungal occurrence was slightly higher in the fresh sample (E), which recorded the greatest number of isolates (6, 25%), followed by the Scotch bonnet (A) and Cayenne pepper (C) samples with 5 isolates each (20.8%). The Bell pepper (B) and Control (D) treatments had comparatively fewer fungal isolates (4 each, 16.7%).

Fungal diversity values were similar across most treatments, ranging from 3 to 4 species, with the Scotch bonnet (A) and fresh sample (E) showing the highest diversity (57.1%).

#### 4.5.3 Total Microbial Distribution

Overall, the combined bacteria and fungi counts revealed that microbial occurrence was highest in the fresh sample (E) with 10 total isolates (24.4%), followed by Scotch bonnet (A) with 9 isolates (22%), and Cayenne pepper (C) with 8 isolates (19.5%). The Bell pepper (B) and control (D) samples each recorded the lowest microbial frequencies (7 isolates, 17%).

**Table 6: Total percentage frequency and diversity of organisms isolated from the fish at day 1**

Organisms	Freq.	% Freq.	Samples				
			A	B	C	D	E
<i>Proteus sp</i>	5	12.2	✓	✓	✓	✓	✓
<i>Streptococcus epidermis</i>	4	9.8	✓	✓	✓	x	✓
<i>Micrococcus sp</i>	3	7.3	✓	x	x	✓	✓
<i>Staphylococcus aureus</i>	4	9.8	✓	x	✓	✓	✓
<i>Bacillus sp</i>	1	2.4	x	✓	x	x	x
Bacteria frequency	17		4	3	3	3	4
Bacteria % frequency		41.5	23.5	17.6	17.6	17.6	23.5
Bacteria diversity	5		4	3	3	3	4
Bacteria % diversity		29.4	80	60	60	60	80
<i>Aspergillus flavus</i>	5	12.2	✓	✓	✓	✓	✓
<i>Mucor sp</i>	3	7.3	✓	✓	x	x	✓
<i>Mucor muccido</i>	2	4.8	✓	x	x	x	✓
<i>Penicillium oxalicum</i>	4	9.8	x	✓	✓	✓	✓
<i>Penicillium italicum</i>	3	7.3	x	✓	✓	x	✓
<i>Botrytis sp</i>	1	2.4	✓	x	x	x	x
<i>Cladosporium sp</i>	1	2.4	x	x	x	✓	x
<i>Saccharomyces sp</i>	4	9.8	✓	x	✓	✓	✓
<i>Cryptomonas sp</i>	1	2.4	x	x	✓	x	x
Fungi frequency	24		5	4	5	4	6
Fungi %frequency		58.4	20.8	16.7	20.8	16.7	25
Fungi diversity	7		4	3	3	4	4
Fungi %diversity		29.2	57.1	42.9	42.9	57.1	57.1
Bacteria/Fungi frequency	41		9	7	8	7	10
Bacteria/Fungi %frequency		99.9	22	17	19.5	17	24.4
Bacteria/Fungi diversity	12		8	6	6	7	8
Bacteria/Fungi %diversity		29.3	66.7	50	50	58.3	66.7

#### 4.6 Frequency and Diversity of Microorganisms Isolated from oven-dried *O. niloticus* Fillets

Table 7 presents the total frequency and percentage distribution of microorganisms isolated from oven-dried *O. niloticus* fillets treated with different pepper species coded as samples A (Scotch bonnet pepper), B (Bell pepper), C (Cayenne pepper) and D (Control), all stored under ambient conditions for 7 days. A total of 31 microbial isolates were identified, comprising 16 bacterial and 15 fungal species, indicating that both bacteria and fungi were actively involved in the microbial colonization of the stored fillets.

##### 4.6.1 Bacterial Frequency and Diversity

The bacterial isolates identified included *Proteus sp.*, *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus sp.*, *Micrococcus sp.*, and *Bacillus sp.* The most frequently occurring bacterial species were *Proteus sp.* and *Micrococcus sp.* (12.9% each), followed by *Staphylococcus aureus* (9.7%), *Escherichia coli* (6.5%), and *Streptococcus sp.* (6.5%). *Bacillus sp.* recorded the lowest frequency (3.2%).

The bacterial frequency was highest in sample A (31.3%), followed by samples C and D (25% each), while sample B had the lowest value (18.8%). The bacterial diversity also followed a similar pattern, with sample A (Scotch Bonnet pepper) recording the highest diversity (83.3%), while Samples C (Cayenne) and D (Control) both recorded moderate diversity (66.7%), whereas sample B (Bell pepper) showed the least bacterial diversity (50%).

##### 4.6.2 Fungal Frequency and Diversity

Four fungal genera were identified which were *Aspergillus flavus*, *Botrytis sp.*, *Aspergillus tamaritii*, and *Saccharomyces sp.* Among these, *Aspergillus flavus*, *Botrytis sp.*, and *Saccharomyces sp.* were the most frequently occurring (12.9% each), while *Aspergillus tamaritii* occurred at a lower frequency (9.7%).

**Table 7: Total percentage frequency and diversity of organisms isolated from the fish by day 7**

Organisms	Freq.	Samples				
		% Freq.	A	B	C	D
<i>Proteus sp</i>	4	12.9	✓	✓	✓	✓
<i>Staphylococcus aureus</i>	3	9.7	✓	x	✓	✓
<i>Escherichia coli</i>	2	6.5	✓	x	✓	X
<i>Streptococcus sp</i>	2	6.5	✓	x	x	✓
<i>Micococcus sp</i>	4	12.9	✓	✓	✓	✓
<i>Bacillus sp</i>	1	3.2	x	✓	x	X
Bacteria frequency	16		5	3	4	4
Bacteria % frequency		51.7	31.3	18.8	25	25
Bacteria diversity	6		5	3	4	4
Bacteria % diversity		37.9	83.3	50	66.7	66.7
<i>Aspergillus flavus</i>	4	12.9	✓	✓	✓	✓
<i>Botrytis sp</i>	4	12.9	✓	✓	✓	✓
<i>Aspergillus tamari</i>	3	9.7	✓	✓	✓	X
<i>Saccharomyces sp</i>	4	9.8	✓	✓	✓	✓
Fungi frequency	15		4	4	4	3
Fungi %frequency		48.4	26.7	26.7	26.7	20
Fungi diversity	3		3	3	3	3
Fungi %diversity		20	100	100	100	100
Bacteria/Fungi frequency	31		9	7	8	7
Bacteria/Fungi %frequency		100.1	29.0	22.5	25.8	22.5
Bacteria/Fungi diversity	9		8	6	7	7
Bacteria/Fungi %diversity		29.0	88.9	66.7	77.8	77.8

The fungi frequency across treatments showed that samples A, B, and C recorded equal frequencies (26.7% each), while sample D (Control) had a slightly lower value (20%). Fungal diversity was consistent across all samples, with three fungal species isolated per treatment (100% relative diversity).

#### 4.6.3 Overall Microbial Distribution

The combined bacterial and fungal frequencies summed to 31 isolates, with bacteria accounting for 51.7% and fungi 48.4% of total isolates. The overall microbial occurrence was highest in sample A (29.0%), followed by sample C (25.8%), while samples B and D each recorded 22.5%. The microbial diversity pattern also indicated that sample A harbored the highest number of different microorganisms (88.9%), followed by samples C and D (77.8% each) while sample B had the least (66.7%).

### 4.7 Frequency and Diversity of Microorganisms Isolated from oven-dried *O. niloticus* Fillets

Table 8, presents the total frequency and diversity of microorganisms isolated from oven-dried *O. niloticus* fillets treated with different pepper spices and stored under ambient conditions for 14 days. A total of 35 microbial isolates were obtained, comprising 15 bacterial and 20 fungal species across all treatments.

#### 4.7.1 Bacterial Isolates

Six bacterial species were identified from the samples: *Escherichia coli*, *Bacillus subtilis*, *Staphylococcus aureus*, *Micrococcus sp.*, *Staphylococcus epidermidis*, and *Proteus sp.* Among these, *Proteus sp.* exhibited the highest frequency of occurrence (11.4%), followed by *Staphylococcus aureus* and *Staphylococcus epidermidis* (8.6% each).

The bacterial frequency was highest in Scotch Bonnet pepper-treated fillets (Sample A) with 5 isolates, followed by the Control (Sample D) with 4 isolates. Bell pepper (B) and Cayenne pepper (C) treatments each recorded 3 bacterial isolates. The corresponding percentage frequencies were 33.3%, 20%, 20%, and 26.7% respectively.

In terms of bacterial diversity, Scotch Bonnet (A) and Control (D) samples showed the highest bacterial diversity (80%), followed by Bell pepper (B) (60%) and Cayenne pepper (C) (40%).

#### 4.7.2 Fungal Isolates

Ten fungal species were isolated across the treatments: *Aspergillus tamarii*, *Aspergillus flavus*, *Penicillium sp.*, *Penicillium italicum*, *Penicillium oxalicum*, *Mucor sp.*, *Mucor mucido*, *Saccharomyces sp.*, *Cryptomonas sp.*, *Fusarium sp.*, and *Trichoderma viride*. Among these, *Aspergillus flavus*, *Mucor sp.*, *Mucor mucido*, and *Saccharomyces sp.* recorded the highest frequencies (8.6% each).

The total fungal frequency (57.5%) was slightly higher than that of bacteria (42.9%), showing that fungal proliferation was more pronounced during storage.

Cayenne pepper (C) and Control (D) samples had the highest fungal frequencies (30% each), followed by Scotch Bonnet (A) (25%) and Bell pepper (B) (15%). Fungal diversity was highest in the cayenne pepper-treated fillets (83.3%) scotch bonnet and control samples each exhibited moderate fungal diversity (50%), while Bell pepper recorded the lowest (40%).

#### 4.7.3 Overall Microbial Distribution

The total microbial (bacteria and fungi) frequency was highest in scotch bonnet (A) samples (28.6%), followed by cayenne (C) and control (D) (25.7% and 28.6%, respectively). The least frequency (22.9%) was observed in bell pepper (B) treated fillets. Similarly, microbial diversity was greatest in Scotch Bonnet (A) and Control (D) (63.6% each).

**Table 8: Total percentage frequency and diversity of organisms isolated from the fish by day 14**

Organisms	Freq.	Samples				
		% Freq.	A	B	C	D
<i>Escherichia coli</i>	1	2.9	x	x	x	✓
<i>Bacillus subtilis</i>	2	5.7	✓	✓	x	✓
<i>Staphylococcus aureus</i>	3	8.6	✓	x	✓	✓
<i>Micococcus sp</i>	2	5.7	✓	x	x	✓
<i>Staphylococcus epidermidis</i>	3	8.6	✓	✓	✓	X
<i>Proteus sp</i>	4	11.4	✓	✓	✓	✓
Bacteria frequency	15		5	3	3	4
Bacteria % frequency		42.9	33.3	20	20	26.7
Bacteria diversity	5		4	3	2	4
Bacteria % diversity		33.3	80	60	40	80
<i>Aspergillus tamari</i>	1	2.9	✓	x	x	x
<i>Aspergillus flavus</i>	3	8.6	✓	✓	✓	x
<i>Penicillium sp</i>	2	5.7	x	x	✓	✓
<i>Penicillium italicum</i>	1	2.9	x	x	x	✓
<i>Penicillium oxalicum</i>	1	2.9	x	x	x	✓
<i>Mucor sp</i>	3	8.6	✓	x	✓	✓
<i>Mucor mucido</i>	3	8.6	✓	x	✓	✓
<i>Saccharomyces sp</i>	3	8.6	✓	✓	x	✓
<i>Cryptomonas sp</i>	1	2.9	x	✓	x	x
<i>Fusarium sp</i>	1	2.9	x	x	✓	x
<i>Trichoderma viridi</i>	1	2.9	x	x	✓	x
Fungi frequency	20		5	3	6	6
Fungi %frequency		57.5	25	15	30	30
Fungi diversity	6		3	2	5	3
Fungi %diversity		30	50	40	83.3	50
Bacteria/Fungi frequency	35		9	7	8	7
Bacteria/Fungi %frequency		100.4	28.6	22.9	25.7	28.6
Bacteria/Fungi diversity	11		7	5	7	7
Bacteria/Fungi %diversity		31.4	63.6	45.5	63.6	63.6

## CHAPTER FIVE

### 5.0

### DISCUSSION

#### 5.1 Effects of Pepper Spices on the Organoleptic Qualities of Dried Tilapia Fillets

The use of different pepper spices such as Scotch Bonnet (*Capsicum chinense*), Cayenne (*Capsicum annum*), and Bell Pepper (*Capsicum annum*) had a measurable but statistically non-significant ( $P > 0.05$ ) effect on the organoleptic qualities of oven-dried *O. niloticus* fillets. The parameters that were evaluated include; appearance, aroma, flavour, taste, and overall acceptability, to collectively determine consumer preference.

At day 1, the sensory scores across all pepper treatments were relatively high, ranging from 2.6 to 3.3 on a five-point hedonic scale, which showed moderate to good acceptance. The Cayenne Pepper and Scotch Bonnet Pepper treatments recorded slightly higher mean scores in appearance (3.3) and aroma (3.1) as compared to the control and Bell Pepper treatments. This can be attributed to the distinct volatile compounds such as capsaicin present in these peppers, which enhanced the aromatic profile of the fillets (Ade-Omowaye *et al.*, 2015; Srinivasan, 2016).

As storage continued from Day 1 to Day 7, a decline in sensory quality was observed across all parameters. Mean scores for appearance, aroma, flavor, and taste fell below 2.0, showing a reduction in consumer acceptability. The decrease in sensory quality could be linked to oxidative rancidity and microbial activity, which contribute to the development of off-flavors and discoloration during storage overtime (Majumda *et al.*, 2018). The results showed that samples treated with Bell Pepper and Cayenne Pepper retained slightly higher flavor and overall acceptability scores than the control and Scotch Bonnet treatments, implying that these spices may have mild preservative or antioxidant properties that help delay sensory deterioration (Olatunde *et al.*, 2022)

Bell Pepper-treated samples, in particular, maintained a relatively appealing appearance and aroma during early storage. This could be due to its high content of carotenoids and flavonoids, which not only enhance color stability but also slows down lipid oxidation (Olatunde *et al.*, 2022). The overall sensory decline during storage was consistent with the trend observed in the weekly mean organoleptic scores, where Week 1 samples (average 4.1 - 4.3) were rated significantly higher than those in Week 2 (average 1.6 - 1.7). This pattern indicates that, although spices may impart beneficial initial effects on sensory quality, ambient storage conditions remain a critical factor in preservation of product quality (Sampels, 2015).

## **5.2 Effects of Pepper Spice and Storage Duration on the Microbial Loads of oven-dried Tilapia Fillets**

In this study, the bacterial and fungal counts of oven-dried Nile tilapia (*O. niloticus*) fillets treated with Scotch Bonnet, Cayenne, and Bell Pepper were evaluated immediately after drying, after one week, and after two weeks of ambient storage. The results showed that both spice treatment and storage duration had measurable effects on microbial load, although the influence of time was more pronounced which corresponds to the deductions of Siddique and Aktar (2011) that storage duration significantly affects microbial and nutritional stability in dried fish, reinforcing your result that time exerts a stronger effect on microbial proliferation than spice treatment alone.

### **5.2.1 Effects of Pepper Spices on the Microbial Loads of Dried Tilapia Fillets After Drying**

Immediately after drying (Day 0), the treatments, Bell Pepper treated samples exhibited the lowest bacterial count ( $5.3 \times 10^{-3}$  Cfu/g), followed by Scotch Bonnet ( $6.0 \times 10^{-3}$  Cfu/g), suggesting that these spices possessed notable antimicrobial properties in accordance with the findings of de Aguiar *et al.*, (2019) who reported that certain

bioactive compounds such as flavonoids, capsaicinoids, present in pepper inhibit bacterial proliferation.

### **5.2.2 Effects of Pepper Spices on the Microbial Loads of Dried Tilapia Fillets After 7 days**

After 7 days of storage, there was a moderate increase in both bacterial and fungal counts across all samples. The bacterial count ranged from  $7.0 \times 10^{-3}$  to  $25.3 \times 10^{-3}$  CfU/g, while the fungal count increased from  $10.3 \times 10^{-3}$  to  $47.3 \times 10^{-3}$  CfU/g. The rise in microbial numbers may be attributed to moisture reabsorption from the atmosphere, which creates a favorable environment for microbial growth (Majumdar *et al.*, 2018). However, samples treated with Cayenne Pepper and Bell Pepper still recorded relatively lower counts compared to the control, suggesting in accordance to Rubió *et al.*, (2013) that the bioactive compounds isolated from different spices shows positive antimicrobial activity against some of the most common microorganisms that affect the food quality and decrease the shelf life of food.

### **5.2.3 Effects of Pepper Spices on the Microbial Loads of Dried Tilapia Fillets After 14 days**

After 14 days of storage, microbial count increased significantly ( $P < 0.05$ ). The fillets that were treated with Cayenne Pepper recorded the highest bacterial load ( $240.0 \times 10^{-3}$  CfU/g), followed by the Control ( $115.7 \times 10^{-3}$  CFU/g), while Bell Pepper and Scotch Bonnet samples had much lower values ( $9.7 \times 10^{-3}$  and  $17.3 \times 10^{-3}$  CfU/g, respectively). The fungal counts also followed the same pattern, with the highest count observed in the control ( $61.7 \times 10^{-3}$  CfU/g). The continuous rise in microbial load during storage may be attributed to temperature fluctuations and the breakdown of lipid and nitrogenous compounds, which accelerate oxidative deterioration and consequently promote microbial spoilage Rasul *et al.* (2022).

The relatively high microbial count in samples treated with Cayenne pepper after two weeks may be due to its higher initial moisture content or possible spice contamination introduced during processing. While the lower microbial in fillets treated with Bell pepper suggests superior preservative efficacy and this effect may be attributed to its rich composition of carotenoids, phenolic acids, and antioxidant compounds, whose antimicrobial and antioxidant properties have been recognized for their role in inhibiting bacterial and fungal proliferation and reducing spoilage (Sulieman *et al.*, 2023).

Ultimately, it was observed that while drying and spicing significantly reduced initial contamination, ambient storage for more than 7 days led to rapid microbial multiplication, especially in samples that already had higher moisture content and these supports the conclusion made by the Agriculture Institute in 2023, which s advices that combination of natural spices with effective packaging and proper moisture regulation will significantly improve the microbial stability and extends the shelf life of dried fish products.

### 5.3 Discussion of Microbial Frequency and Diversity of Organisms isolated after Day 1

The frequency and diversity results in Table 6 revealed that a total of 41 microbial isolates, comprising 17 bacterial and 24 fungal species, were obtained from oven-dried *O. niloticus* fillets treated with different pepper spices and stored under ambient conditions. The predominance of fungi (58.4%) over bacteria (41.5%) suggests that fungal growth was more favored under the storage environment, possibly due to the reabsorption of moisture content under storage conditions that typically support mold proliferation on dried fish products (Adu-Gyamfi, 2008).

### 5.3.1 Bacterial Occurrence and Diversity

The bacterial species isolated *Proteus* sp., *Streptococcus epidermidis*, *Micrococcus* sp., *Staphylococcus aureus*, and *Bacillus* sp. are commonly associated with dried and smoked fish products, which indicates post-processing contamination or environmental exposure during handling and storage (Adebayo-Tayo *et al.*, 2012). Among these, *Proteus* sp. showed the highest frequency (12.2%), while *Bacillus* sp. was the least frequent (2.4%). The presence of *Proteus* and *Staphylococcus aureus* in particular may indicate contamination from human contact or unsanitary handling surfaces, since both organisms are known to be part of normal human skin flora (Jega *et al.*, 2019).

Bacterial frequency was highest in the fresh and Scotch bonnet pepper-treated samples (23.5% each), followed by Bell pepper, Cayenne pepper, and the control (17.6% each). The reduced bacterial counts in most spiced samples compared to the untreated control suggests that the pepper spices exerted varying degrees of antibacterial activity. This is consistent with the findings of Sulieman *et al.* (2023), who reported that *Capsicum* species possess notable antimicrobial compounds such as capsaicin, carotenoids, and phenolics, which can inhibit bacterial growth in food systems.

### 5.3.2 Fungal Occurrence and Diversity

Fungal isolates were dominated by species of *Aspergillus*, *Penicillium*, *Mucor*, and *Cladosporium*, which are well known spoilage fungi associated with stored and dried fish (Ndife *et al.*, 2018). The most frequently isolated fungus was *Aspergillus flavus* (12.2%), followed by *Penicillium oxalicum* and *Saccharomyces* sp. (9.8% each). The high frequency of *A. flavus* is particularly concerning due to its potential to produce aflatoxins, which are toxic secondary metabolites that can compromise food safety (Feruke-Bello *et al.*, 2024).

The occurrence of *Mucor* sp. and *Penicillium italicum* in several samples further indicates possible airborne contamination during storage, as these species are commonly found in air and dust environments (Sissoko *et al.*, 2025). However, their relatively lower occurrence in the pepper-treated samples suggests that the antimicrobial and antioxidant properties of the spices likely limited their growth. Previous studies have demonstrated that essential oils and phenolic extracts from *Capsicum* species can suppress mycelial growth and spore germination of common foodborne fungi (Sepúlveda *et al.*, 2024).

Fungal frequency and diversity were highest in the fresh (25%) and Scotch bonnet-treated (20.8%) samples, while Bell pepper and control samples had lower values (16.7%). This aligns with the report of Amuneke *et al.* (2020) whose study demonstrated the effectiveness of natural spice in controlling microbial population and pathogens.

### 5.3.3 Overall Microbial Distribution

When bacterial and fungal frequencies were combined, the total microbial load was highest in the fresh sample (24.4%) and Scotch bonnet pepper treatment (22%), while the lowest values were recorded in the Bell pepper and control samples (17% each). This pattern indicates that although spice treatments suppressed microbial proliferation to some degree, variations existed among the spice types. The superior performance of Bell pepper may be linked to its relatively high levels of carotenoids and phenolic acids, which have both antioxidant and antimicrobial activity (Sulieman *et al.*, 2023).

Overall, the results support the conclusion that pepper spices can contribute to microbial control in dried fish, but their efficacy is species-dependent. The microbial frequency and diversity data suggest that Bell pepper and Cayenne pepper provided better preservative protection than Scotch bonnet pepper, possibly due to differences in chemical composition and capsaicin concentration (Puvaca *et al.*, 2014; Agriculture Institute, 2023). These findings highlight the potential of natural spices as alternative

food preservatives, offering both safety and shelf-life benefits under ambient storage conditions.

#### 5.4 Discussion of Microbial Frequency and Diversity of Organisms isolated after 7 Days

The microbial profile of oven-dried *O. niloticus* fillets treated with different pepper spices, after 7 days revealed the presence of both bacterial and fungal contaminants, indicating that microbial growth occurred despite the drying process. A total of 31 isolates were obtained, comprising 16 bacterial and 15 fungal species. This finding aligns with the reports of Olatunde *et al.*, (2022) and Rasul *et al.*, (2022), who similarly observed the persistence of microbial populations in dried fish products stored under ambient conditions due to rehydration, oxidative changes, and environmental contamination.

##### 5.4.1 Bacterial Occurrence and Diversity

Among bacterial isolates, *Proteus* sp. and *Micrococcus* sp. were the most frequently occurring species, accounting for 12.9% each as well as *Staphylococcus aureus* and *Escherichia coli* accounting for 9.7 and 6.5% respectively. These bacteria are common spoilage organisms associated with post-processing handling and environmental exposure of dried fish (Omoruyi *et al.*, 2019). The presence of *Bacillus* sp., although occurring less frequently (3.2%), confirms the study of Rasul *et al.*, 2022 who reported that it is a known spore-former capable of surviving high temperatures and low moisture conditions.

The bacterial frequency was highest in Scotch Bonnet-treated fillets (sample A) 31.3 %, indicating that despite the known antimicrobial properties of capsaicinoids, environmental and moisture factors may have favored microbial survival. Similar trends were noticed by Akinbisoye *et al.*, (2020), who then deduced that the moisture content of smoked catfish is of great importance during storage, the lower it is the better the storability. The relatively lower bacterial diversity in Bell Pepper (sample B) treatment

suggests stronger inhibition of bacterial growth, possibly due to variations in spice phytochemical concentration or drying uniformity (Sogo *et al.*, 2022).

#### 5.4.2 Fungal Occurrence and Diversity

Fungal isolates included *Aspergillus flavus*, *Botrytis* sp., *Aspergillus tamarii*, and *Saccharomyces* sp., with *A. flavus* and *Botrytis* sp. being the most prevalent (12.9% each). These genera are commonly associated with the spoilage of dried fish and other protein foods during ambient storage in alignment with the findings from Osibona *et al.* (2018). Fungal diversity was consistent (100%) across all treatments, indicating that the ambient storage environment was the primary determinant of fungal persistence, rather than spice type. This observation corresponds with Rasul *et al.* (2022), who reported that fungal growth on dried fishery products is predominantly influenced by environmental conditions and packaging methods, rather than compositional factors alone.

#### 5.4.3 Overall Microbial Trends

The combined bacterial and fungal frequencies revealed that Scotch Bonnet treated fillets (sample A) had the highest microbial load and diversity, while Bell Pepper (sample B) and Control (sample D) recorded the lowest. This pattern may be attributed to differences in spice composition bell pepper, rich in carotenoids and phenolic acids, has demonstrated higher antioxidant and antimicrobial efficacy corresponding with the notion of Wahba *et al.*, (2010) that Phenolic compounds, polyphenols, and flavonoids are important antibacterial substances in plants. The persistence of microbial isolates across all treatments reinforces that while spices can slow microbial proliferation, they cannot fully prevent contamination without complementary preservation measures such as vacuum packaging or refrigeration (Agriculture Institute, 2023).

### 5.5 Discussion of Microbial Frequency and Diversity in Oven-Dried *O. niloticus* Fillets after 14 days

### 5.5.1 Bacterial Occurrence and Diversity

Six bacterial species were identified, including *Escherichia coli*, *Bacillus subtilis*, *Staphylococcus aureus*, *Micrococcus sp.*, *Staphylococcus epidermidis*, and *Proteus sp.* The most frequently isolated bacterium was *Proteus sp.* (11.4%), followed by *Staphylococcus aureus* and *Staphylococcus epidermidis* (8.6% each). These organisms are commonly associated with post-handling contamination and spoilage of fish product and presence of *E. coli* indicates possible faecal contamination, which as observed by Adu-Gyamfi (2006) results from water pollution and contamination of equipment.

*Bacillus subtilis* and *Micrococcus sp.* were among the more resilient species detected across treatments. These organisms are known spore-formers and can survive heat treatment such as oven-drying and their persistence despite spice treatment as suggests in accordance to Jega *et al.*, (2019) that drying alone may not entirely eliminate heat-resistant microorganisms but can reduce their activity to sub-lethal levels.

The bacterial frequency and diversity were highest in the Scotch Bonnet-treated samples (A) and control (D), each recording up to four species (80% diversity). Conversely, Bell pepper (B) and Cayenne pepper (C) treatments exhibited reduced bacterial diversity (60% and 40%, respectively), indicating some inhibitory effect attributable to the antimicrobial compounds in these spices, notably phenolic acids, carotenoids, and capsaicinoids (Sulieman *et al.*, 2023).

### 5.5.3 Fungal Occurrence and Diversity

Ten fungal genera were isolated, with *Aspergillus flavus*, *Mucor* sp., *Mucor mucido*, and *Saccharomyces* sp. recording the highest frequencies (8.6% each). The high occurrence of *Aspergillus* and *Penicillium* species in stored samples is consistent with reports by Omoruyi and Abolagba (2016), who identified these fungi which as saprophytes that proliferate even in the absence of moisture.

Among treatments, Cayenne pepper (C) and Control (D) samples exhibited the highest fungal frequencies (30%), while Scotch Bonnet (A) and Bell pepper (B) had slightly lower frequencies (25% and 15%, respectively). This suggests in alignment to Frantianni *et al.*, (2020) that the bioactive compounds in these peppers particularly the capsaicin and phenolic compounds exert antifungal activity.

### 5.4 Overall Microbial Distribution

The combined bacteria and fungi frequency showed that Scotch Bonnet (A) and Control (D) samples had the highest total microbial frequency (28.6%), while Bell pepper (B) recorded the lowest (22.9%). Microbial diversity followed a similar pattern, with Scotch Bonnet, Cayenne (C) and Control samples exhibiting the broadest range of isolates (63.6%), while Bell pepper (B) recorded lower diversity (45.5%).

This pattern suggests that while spice treatments had measurable antimicrobial effects, none of the treatments completely suppressed microbial diversity. Similar observations have been made by Okonji *et al.* (2020) who emphasized that dried fish products are prone to gradual microbial recontamination, when packaging and handling are not strictly controlled.

## CHAPTER SIX

### 6.0 CONCLUSION AND RECOMMENDATIONS

#### 6.1 Conclusion

The study was carried out to evaluate how different pepper spices specifically bell pepper (*Capsicum annuum*), cayenne pepper (*Capsicum annum*), and scotch bonnet pepper (*Capsicum chinense*) influence the organoleptic and microbial quality of oven-dried tilapia fillet under ambient storage.

The results showed that spice treatment had a significant effect on both the organoleptic and microbial qualities of the oven-dried tilapia fillets. However, samples treated with Bell pepper (*Capsicum annuum*) exhibited superior sensory qualities such as better appearance, aroma, taste, flavour, and overall acceptability when assessed by panelists using the 5-point Hedonic scale as compared to those treated with Cayenne and Scotch bonnet peppers, as well as the control which was not treated with any spice. This improved performance could be attributed to the high levels of carotenoids, phenolic compounds, and antioxidants present in bell pepper, which are known to inhibit microbial growth and delay oxidative degradation.

In terms of microbial quality, the pepper treated samples, particularly those treated with bell pepper, also recorded lower bacterial and fungal counts as compared to the control sample all through the storage period. This goes to prove that the bioactive compounds in peppers possess antimicrobial properties that can suppress the activities of spoilage microorganisms. Regardless of this, the overall microbial load increased progressively as the storage durations increased, showing that oven-dried fish products are still susceptible to contamination under ambient temperature conditions.

Despite the initial preservative benefits imparted by pepper treatments, the sensory scores for all samples declined with time, suggesting that ambient storage is a limiting factor in maintaining the quality of processed fish and fish products.

The study therefore concludes that the use of aromatic peppers, especially bell pepper, is a natural and effective method for improving the shelf life and organoleptic qualities of oven-dried Nile tilapia fillets. Be that as it may, proper storage practices and controlled environmental conditions are key contributors in sustaining quality over extended storage periods.

## **6.2 Recommendations**

- It is recommended that fish processors incorporate the use of natural aromatic spices such as bell pepper during in drying and preservative processes as these spices offer natural antioxidant and antimicrobial effects, which improves product quality while minimizing the use of synthetic preservatives.
- Since ambient temperature which is prone to fluctuation increases lipid oxidation and microbial spoilage, maintaining a controlled temperature of storage conditions is vital; for example cold rooms or re Fridgerators especially in commercial settings.
- Future studies should be carried out to investigate the use of other indigenous spices and their effects on dried fish preservation.
- Small and medium scale fish processors or even end consumers can benefit from adopting spice treatment as preservative techniques, since these methods are affordable, easily accessible, culturally acceptable, and environmentally friendly.

This study therefore establishes that the application of aromatic peppers, particularly bell pepper, holds substantial promise as a sustainable, low cost, and natural alternative approach to improving the quality and shelf life of oven-dried Nile tilapia fillets under ambient storage conditions.

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