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Microbiological Quality and Food Safety Evaluation of Selected Processed and Raw Fish Sold in Open Markets in Awka, Anambra State, Nigeria

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Abstract

This study evaluated the microbiological quality and food safety status of both processed and raw fish sold in open markets in Awka, Anambra State, Nigeria. Four fish forms hot-smoked, cold-smoked, frozen, and barbecued were analyzed using standard microbiological methods. Total bacterial count (TBC), total fungal count (TFC), coliforms, and foodborne pathogens were determined using Nutrient Agar, MacConkey Agar, Eosin Methylene Blue Agar, Mannitol Salt Agar, Salmonella–Shigella Agar, and Sabouraud Dextrose Agar. Barbecued fish recorded **too** numerous to count (TNTC) bacterial growth, while frozen fish had the highest measurable mean TBC (6.35×10^5 CFU/g**).** Cold-smoked fish showed the lowest mean TBC **(**2.50×10^4 CFU/g**)** and no fungal growth. Identified bacteria included *Escherichia coli*, *Salmonella enterica*, *Salmonella typhimurium*, *Shigella flexneri*, *Enterobacter aerogenes*, *Proteus mirabilis*, *Enterococcus faecalis*, and *Staphylococcus aureus*. *Candida* spp. was the only fungal isolate detected. The presence of indicator organisms and pathogenic bacteria in both RTE and raw fish indicates poor hygiene, post-processing contamination, and potential public health risks. Improved sanitary practices and regulatory surveillance are recommended.

Keywords: Microbiological quality, food safety, Raw Fish, Processed Fish, foodborne pathogens, open markets.

1. Introduction

Fish has become an increasingly important source of protein and other elements necessary for the maintenance of a healthy body. It constitutes an important food component for a large section of the world population. The quality of fish is of major concern to the food processors, consumers, and public health authorities. Provision of safe, wholesome, and acceptable fish and fish products, through control of contamination, is essential from a food safety point of view. Fish is a major source of animal protein in Nigeria and is widely consumed in both edible and raw forms. Fish contributes about 60% of the world's supply of protein, and 60% of the developing world derives more than 30%

of their animal protein from fish (Emikpe *et al.,* 2011). Fish has a wide range of acceptability in most parts of the country due to its unique taste, flavor, and good texture (Olayemi *et al.,* 2012). However, fish is highly perishable and readily supports microbial growth due to its high moisture content and nutrient composition. In open markets, inadequate hygiene, lack of temperature control, and poor handling practices increase the risk of microbial contamination. The greatest risk to human health occurs due to the consumption of raw, inadequately cooked, or insufficiently processed fish and fish products. The symptoms of fish food poisoning include nausea, vomiting, diarrhea, abdominal cramps, headache, as well as muscle and joint pain.

It is well accepted that microorganisms are commonly present on fish surfaces, such as skin and gills, as well as the digestive tract and internal organs, for example, the kidney, liver, and spleen. However, fish and fish products, especially raw or undercooked products, have been involved in outbreaks associated with bacterial pathogens, biotoxins, histamine, viruses, and/or parasites (Galaviz-Silva *et al.,* 2019).

Processed fish products, such as smoked and barbecued fish, are consumed without further heat treatment, making post-processing contamination a critical food safety concern. The presence of pathogenic microorganisms such as *Salmonella* spp., *Escherichia coli*, and *Staphylococcus aureus* in fish has been linked to foodborne diseases globally. Despite high fish consumption in Awka, data on the microbiological safety of market-vended fish remain limited. Lack of proper awareness and Knowledge about microbial studies in fish is not known to the consumers, and this has led to diseases, especially in immune-compromised individuals, which could have been prevented with proper management. Some of these diseases, caused by microorganisms, have also been found to contain a resistance determinant that enhances their ability to infect consumers. For instance, *Escherichia coli* is known to survive well in aquatic environments, is highly adept at horizontal gene transfer and is a notorious vehicle for antibiotic resistance dissemination.

The majority of the processed fish (smoked and barbecued fish) and raw fish marketed in Awka, Anambra state, are sold in open markets and so are exposed to dust, contamination, and high temperature, which supports spoilage, encouraging the growth of microorganisms, which might facilitate increased transmission and prevalence of pathogens. Moreover, poor personal hygiene, such as neglecting to wash hands after handling contaminated materials encourage contamination of fish with pathogenic organisms. This study, therefore, assessed the microbiological quality and food safety status of selected processed and raw fish sold in open markets in Awka, Anambra State. However, there are other objectives of this study which includes isolating and identifying bacteria associated with the frozen fish, hot and cold smoked fish and barbecued fish sold in open markets in Anambra State, Nigeria, reinforcing the need for proper sanitary measures to avoid bacterial contamination of fish and raising awareness on the potential risks associated with poor handling of fish both at processed and raw stages.

1.1 Preservation Processes and Microbiological Implications in Fish Products

Fish is among the most perishable food commodities due to its high moisture content, neutral pH, and rich nutrient profile, which make it highly susceptible to microbial spoilage and quality deterioration. To extend shelf life and ensure food safety, various preservation techniques such as smoking,

freezing, drying, and barbecuing are employed globally, especially in regions without advanced cold chain infrastructure.

Smoking remains a widely applied traditional method. Cold-smoking involves exposing fish to smoke at low temperatures (20–30 °C), which imparts flavor and reduces moisture but keeps the product in the temperature danger zone, potentially favoring pathogenic growth if not properly controlled. Hot-smoking, conducted at higher temperatures (>145 °F internally), not only imparts desirable sensory qualities but also kills vegetative microbes, making the product safer; however, post-processing contamination risks persist if hygiene practices are inadequate (Oshoke, J. O., *et al*., 2025). Modern hygienic controls, such as the application of acidified sodium chlorite on cold-smoked fish surfaces, have been shown to reduce aerobic plate counts and improve safety, highlighting the importance of sanitation and handler knowledge in processing environments (Fernandes et. al., 2018).

Freezing is one of the most effective long-term preservation methods, significantly slowing microbial growth and biochemical degradation. Frozen fish stored under controlled, low-temperature conditions can maintain quality attributes close to fresh fish for extended periods (). Proper packaging, such as water-barrier films and glazing, is critical to prevent freezer burn and oxidative rancidity, which compromise texture and flavor.

Recent research underscores the evolving landscape of fish preservation. Advanced industry studies on combinations of freezing and hot-smoking demonstrate that processing order influences texture and product stability, with freezing before smoking yielding firmer, higher-quality fillets in Atlantic mackerel (Brunton et al., 2019). Additionally, multi-omics analyses of cold-smoked salmon have revealed distinct spoilage dynamics tied to microbial community structures and metabolic profiles, suggesting that predictive spoilage models may improve quality control.

Emerging strategies beyond traditional methods include bio-based coatings and active packaging to retard spoilage microbes, and integrated cold chain management systems incorporating IoT-based monitoring to optimize preservation from harvest through distribution (Abdel-Reheem, *et al*., 2023). These innovations aim to reduce postharvest losses while maintaining nutritional value and consumer safety.

Despite these advancements, basic challenges remain; artisan fish smoking and handling in open markets often lack standardized control measures, increasing the risk of contamination by spoilage organisms and foodborne pathogens. Therefore, combining traditional methods with improved sanitation, cold chain optimization, and modern antimicrobial interventions is essential to enhance fish safety and quality in both local and industrial contexts.

2. MATERIALS AND METHODS

2.1 Sample Collection

Four types of fish, smoked, cold-smoked, barbecued, and frozen (raw), were collected aseptically from Eke Awka, Second Market, Ifite, and Abakiliki Street markets. Samples were transported in sterile containers to the microbiology laboratory for analysis.



Plate 1: BARBEQUED FISH



Plate 2: HOT SMOKED FISH



Plate 3: FROZEN FISH



Plate 4: COLD SMOKED FISH

MATERIALS USED

The materials used include Autoclave, Incubator, Weighing balance, Bunsen burner, Wire loop, Hot air oven, Cotton wool, Spatula, Aluminum foil, Microscope, Beakers, Conical flask, Slides, Micropipette, Petri dishes, Reagent bottles, Test tubes, Calibrated measuring cylinder, Nutrient agar, Methyl red, Kovac's reagent, Potassium hydroxide (KOH), Hydrogen peroxide (H2O2), Oxidases reagent and Peptone water.

SAMPLE ANALYSIS

One gram (1g) of the fish samples was weighed out aseptically and introduced into 10ml of sterile peptone water for bacteria, it was properly shaken to homogenize the sample. A 10-fold serial dilution of each of the sample was carried out using peptone water as the diluents. 0.1ml of appropriate dilutions (10-2) of the sample were inoculated by plate method into sterile plates of Nutrient agar (NA), Eosin-Methylene Blue Agar (EMB), MacConkey Agar, Mannitol Salt Agar (MSA) and Salmonella-Shigella agar (SSA) plates for the culture of bacteria. The culture plates were incubated at 37°C aerobically for 24-48hours for bacteria; while Saboraud Dextrose Agar (SDA) was used for fungi. Developing colonies on Nutrient agar for bacteria and SDA for fungi were counted to obtain total viable. Discrete colonies for the bacteria were obtained by sub culturing into Nutrient agar plates and were subsequently identified using standard methods.

Total Bacterial Count (TBC) and Total Fungal Count (TFC) were calculated thus:

TBC/TFC = (N) x 10 VD

Where TBC: Total Bacterial Count

TFC: Total Fungal Count

TCC: Total Coliform count

V: Volume plated

D: Dilution Factor



Plate 5: The petri dishes containing the cultured species

Characterization and identification of bacteria

Identification of the bacterial isolates was accomplished by the observation of colonial characteristics, Gram reaction and biochemical tests (Chessbrough, 1984). The characterization of the isolates were performed, by employing Gram staining reaction, Catalase test, Citrate test, Sugar fermentation test, Coagulase test, Motility test, Oxidase test, Urease test, Indole test, Methyl Red and Voges proskauer test as described by Bergey's Manuel of Determinative Bacteriology, 9th edition (1994).

Gram reaction

Thin smear of the isolate was made on clean, non-greasy, dust-free slides, air dried and heat fixed. The smear was flooded with crystal violet and allowed to remain on the slide for 60 seconds. Thereafter, the crystal violet was washed off with gentle running water. Again, the slide was flooded with slide with Gram's iodine, allowed to remain for 60 seconds and washed off. The slide was decolorized with acetone-alcohol mixture. The slide was counter-stained with safranin for 60 seconds and rinsed with tap water and allow to air dry. The slide was then viewed under oil immersion lens microscope (× 100). Purple colour indicated Gram–positive organisms while red or pink colour indicated Gram-negative organisms.

Catalase test

Exactly 3ml of 3% solution of hydrogen peroxide (H2O2) was transferred into a sterile test tube. Then, 3 loopful of a 24-hour pure culture of the test bacteria were inoculated into the test tube. The

tube was observed for immediate bubbling indicative of a Positive, while no bubbling indicated a negative reaction.

Motility test (hanging drop method)

A loopful of 18–24-hour broth culture of the test bacteria was placed at the centre of a clean grease-free cover-slip. Carefully, the cover slip was inverted and placed over the concave portion of a hanging drop slide. The cover-slip/slide arrangement was observed for motility at X100 magnification on a compound microscope. Care was taken to not interprete "drift" or "Brownian motion" as motility. Results were recorded as motile or non-motile.

Oxidase test

All bacteria that are oxidase positive are aerobic, and can use oxygen as a terminal electron acceptor in respiration. This does NOT mean that they are strict aerobes. Bacteria that are oxidase-negative may be anaerobic, aerobic, or facultative; the oxidase negative result just means that these organisms do not have the cytochrome c oxidase that oxidizes the test reagent. They may respire using other oxidases in electron transport.

Whitman No.1 filter paper was soaked with the substrate tetramethyl-p-phenylenediamine dihydrochloride. The filter paper was moistened with sterile distilled water. Then the test colony was picked with wooden or platinum loop and smeared in the filter paper. The inoculum was observe the area around the inoculated paper for a color change to deep blue or purple within 10-30 seconds. Positive and negative quality controls were also set up (Positive control: *Pseudomonas aeruginosa*; B. Negative control: *Escherichia coli*). Positive was indicated by development of dark purple color (indophenols) within 10 seconds. Negative: Absence of color

Urease test using Christensen's urea agar

The urease test identifies those organisms that are capable of hydrolyzing urea to produce ammonia and carbon dioxide. It is primarily used to distinguish urease-positive Protease from other Enterobacteriaceae.

Heavy inoculum from an 18- to 24-hour pure culture was used to streak the entire Christensen's Urea Agar slant surface. Adequate care was taken not to stab the butt, as it will serve as a color control. The tubes were incubated with loosened caps at 35 C. The slants were observed for a color change at 6 hours, 24 hours, and every day for up to 6 days. Urease production would be indicated by a bright pink (fuchsia) color on the slant that may extend into the butt. Note that any degree of pink is considered a positive reaction. Prolonged incubation may result in a false-positive test due to the hydrolysis of proteins in the medium. To eliminate protein hydrolysis as the cause of a positive test, a control medium lacking urea was also set up. Rapidly urease-positive Proteae (*Proteus spp., Morganella morganii, and some Providencia stuartii strains*) will produce a strong positive reaction within 1 to 6 hours of incubation. Delayed-positive organisms (e.g., *Klebsiella or Enterobacter*) will typically produce a weak positive reaction on the slant after 6 hours, but the reaction will intensify and spread to the butt on prolonged incubation (up to 6 days). The culture medium will remain a yellowish color if the organism is urease-negative

Indole test

A loopful of an 18-24-hour culture was used to inoculate the test tube containing 3 ml of sterile tryptone water. Incubation was done at 35–37℃ first for 24 hours and further for up to 48 hours. Test for indole was done by adding 0.5 ml of Kovac's reagent, shaken gently and then examined for a ring of red colour in the surface layer within 10 minutes, indicative of a positive reaction. Absence of red colour indicated a negative reaction.

Methyl red test

Exactly 5 drops of methyl red indicator were added to an equal volume of a 48-hour culture of the isolate in Methyl red–Voges Proskauer (MR-VP) broth. The production of a bright red colour indicates a positive test, while a yellow colour indicates a negative test after vigorous shaking.

Voges-prausker test

Exactly 2ml of the 18-24 hours culture of the test organism growing on MR-VP broth was aseptically transferred into a sterile test tube. Then 0.6ml of 5% α-naphthol was added, followed by 0.2ml of 40% KOH (NB: These reagents needed to be added in this order). The tube was shaken gently to expose the medium to atmospheric oxygen and then allowed to stand undisturbed for 15-30 minutes. A positive test was indicated by the presence of a red colour after 15-30 minutes, indicative of the presence of diacetyl, the oxidation product of acetoin (Test was always considered invalid after one hour because VP-negative cultures may produce a copper-like colour, false positive). Lack of pink-red colour denoted a negative reaction.

Citrate test

A 24h old culture was inoculated into test tubes containing sterile Simmons Citrate agar slant and then incubated for 24hours. A positive test was indicated by a change from green to blue colour on the surface of the Simmons Citrate agar slant. No colour change indicated a negative reaction.

Sugar fermentation test

Each of the isolate was tested for its ability to ferment a specific sugar. 1g of the sugar and 1g of peptone water were dissolved in 100ml of water. 5ml of the solution were transferred into clean test-tubes using sterile pipettes. The test-tubes containing peptone water and sugar were added Durham's tube which were placed inversely and bromothymol blue as an indicator. These were sterilized for 10minutes and allowed to cool before inoculating the inocula. The test-tubes were incubated for 3days. The production of acid and gas or acid only indicated utilization of sugars. Acid production was indicated by change in colour of the medium from green to yellow while gas production was observed by presence of gas in the Durham's tubes.

Fungal identification

Isolation and Characterization of the Fungi.

This was done based on the description of the gross morphological appearance of fungal colonies on the SDA culture medium and the slide culture and lacto phenol cotton blue wet mount staining techniques for microscopic evaluation under X100 and X40 magnification of the microscope; with reference to the Manual of Fungal Atlases (Frey et al., 1979; Barnett and Hunter, 2000; Watanabe, 2002; Ellis et al., 2007).

RESULTS:

The results of the microbial analysis of different fish samples are as shown in the tables below

Table 1: Total Microbial Count and Diversity of Bacteria Isolated from the fish

SAMPLE	NUTRIENT AGAR			SDA			Identity of Fungi on SDA
	X	Y	Mean TBC	x	y	Mean TFC	
HS	32	34	$3.30x10^5$			$5.50 x10^4$	*Candida spp*
CS	2	3	$2.50 x10^4$	Nil	Nil	Nil	Nil
FF	62	65	$6.35 x10^5$			$5.60 x10^5$	*Candida spp*
BQ	TNTC	TNTC	TNTC	4	4	$5.50 x10^4$	*Candida spp*

Key:

TNTC- Too Numerous to Count

HS- Hot-smoked (Ready to eat)

CS- Cold-smoked (Ready to eat)

FF- Frozen Fish (Non-Ready to eat)

BQ- Barbeque (Ready to eat)

TBC- Total Bacteria Count

TFC- Total Fungal Count

SDA- Sabouraud Dextrose Agar

X – First Count of the bacteria/fungi

Y – Second Count of the bacteria/ fungi.

Table 2: Counts and Identities of Various Organisms found on the Fish Samples

SAMPLE	Mannitol Salt Agar		EOSINE METHYLENE AGAR		MacConkey Agar		Samonella Shigela Agar	
	N	Identity	N	Identity	N	Identity	N	Identity
HS	Nil	Nil	*3*	*Escherichia coli*	2 1	*Enterococcus faecalis* *Salmonella typhimurium*	1 1	*Salmonella enterica* *Shigella flexneri*
CS	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
FF	1	*Staphylococcus aureus*	*37* *3*	*Escherichia coli* *Proteus mirabilis*	24 1	*Enterobacter aerogenes* *Enterococcus faecalis*	3 25	*Salmonella enterica* *Shigella flexneri*
BQ	Nil	Nil	*17*	*Escherichia coli*	13 35	*Enterobacter aerogenes* *Salmonella typhimurium*	2 14	*Salmonella enterica* *Shigella flexneri*

Table 3: Morphological and Biochemical Identifications of the Various Bacterial Isolates.

Form	Surface	Colour	Margin	Elevation	Opacity	Gram	Cat	Mot	Ind	MR	VP	Cit	Lac	Glu	Suc	Fru	Mal	Oxi	Ure	Identity
Circular	Glistening	Cream	Entire	Raised	Transparent	- Rod	+	+	-	+	+	+	+	+	+	-	+	-	+	*Proteus mirabilis*
Circular	Smooth	Yellowish	Entire	Raised	Opaque	+ cocci	+	+	-	+	-	-	AG	AG	A	A	AG	-	+	*Staphylococcus aureus*
Circular	Smooth	Greyish/ Colourless	Entire	Convex	Translucent	- Rod	+	-	Var	+	-	-	-	+	-	+	var	-	-	*Shigella flexneri*
Circular	Smooth	Greyish/ white	Lobate	Low convex	Translucent	- Rod	+	+	-	+	-	+	-	+	-	+	+	-	-	*Salmonella enterica*
Circular	Smooth	Cream	Entire	Convex	Opaque	+cocci	-	-	-	-	+	-	+	+	+	-	+	-	-	*Enterococcus faecalis*
Circular	Smooth	Whitish	Entire	Convex	Translucent	- Rod	+	+	+	+	-	-	+	+	var	-	-	-	-	*Escherichia coli*

Gram: Gram reaction

Cat: Catalase test

Mot: Motility test

Ind: Indole test

MR: Methyl-red test

VP: Voges-Proskauer test

Cit: Citrate Utilization test

Sugar Fermentation Tests:

Lac: Lactose Fermentation

Glu: Glucose Fermentation

Suc: Sucrose Fermentation

Fru: Fructose Fermentation

Mal: Maltose Fermentation

Oxi: Oxidase

Ure: Urease

Table 4: Colonial and Microscopic Identifications of the Fungal Isolate

	Colony morphology	Microscopy	Identity
	Colonies (SDA) white to cream-coloured smooth, glabrous yeast-like.	Spherical to ellipsoidal budding blastoconidia, 2-6 x 3-7 µm in size. Ascospores may be produced on 5% malt extract or cornmeal agar after 5-30 days at 25OC.	***Candida sp.***

RESULT

The result presented in Table 1 shows that the fish sample BQ (Barbequed fish) has the highest bacteria count which was too numerous to count, followed by FF (frozen fish) with a mean TBC of 6.35×10^5, HS of 3.30×10^5 and CS of 2.50×10^4. Using nutrient agar medium, Table 1 also shows the total fungal count using SDA, only one species of fungi was identified which *is Candida sp*. The fish sample FF has the highest number of *Candida sp* with a mean TFC of 5.60×10^5 while BQ and HS (Hot smoked) have the same mean TFC and CS (Cold smoked) fish did not show any TFC. All the fish samples analysed have Candida sp while CS did not have any specie of fungi present.

Table 2 shows the result obtained from MSA (Mannitol Salt Agar), EMB (Eosine methyl blue agar) and Mac Conkey Agar to identify and count various organisms discovered in the fish sample. Using MSA, no bacteria were found in Hot smoked, cold smoked fish and Barbequed fish, while FF was shown to contain a bacterium which was identified as *Staphylococcus aureus*. The types of bacterial and fungal isolates in both fresh and smoked fish samples are similar to those previously observed in other studies by Akinjogunla *et al*.and Wogu and Maduakor.

Using EMB AGAR *3 E. coli* was identified in HS, *37 E. coli* and 3 *Proteus mirabilis* in FF and *17 E. coli* in BQ. Using this medium nothing was found in CS fish sample.

Using MacConkey agar bacteria present in this medium was counted and identified; 2 *Enterococcus faecalis* and 1 *Salmonella typhimurium*, present in HS, in FF we identified *24 Enterobacter aerogenes* and *1 Enterococcus faecalis* and in BQ there was *13 Enterobacter aerogenes* and *35 Salmonella typhimurium*, while CS had no organism present.

In Table 3, various tests using reagents were carried out to isolate and identify various bacteria in the fish samples. In Table 3, their forms, margin, elevation, surface, opacity and colours were also identified. Sugar fermentation tests were also conducted in order to identify sugars that can be fermented by bacteria which also aided us in identifying various bacteria.

Table 4 shows the result using microscopy in which a cream-coloured yeast like organism was observed and the shape viewed was spherical with a size of 2-6×3-7µm.The specie identified was *Candida sp.*

5.0 Discussion

The present study is the evaluation of microbial profile of select ready and non- ready to eat fish vended in open market in awka, Anambra state. The results from this study revealed the presence of fungi species namely; *Candida* sp. in all the fish samples but cold smoked fish. The absence of fungi in the fish samples could be as a result of proper handling by vendors or good hygiene practiced by

handlers. This is similar to the findings of Nwiyi and Onyeabor (2012) that rough handling results in a faster spoilage rate. The fungal contamination of this fish samples could be attributed to improper sanitation, handling, storage, transportation and marketing of fish this is similar with the findings of Hassan (2003). This study also revealed seven bacterial species namely; *E. coli*, *Enterococcus faecalis*, *Salmonella sp., Proteus mirabilis, Enterobacter aeorogenes, Shigella flexneri* and *Staphylococcus aureus. Escherichia coli* is one of the major pathogens associated with post-harvest fish spoilage. This finding is in agreement with previous findings by Gram and Huss (2001), who reported that this organism is one of the major causes of microbial spoilage of fresh fish after capture and the microbial count on the different media suggests contamination. However, in a related study, Emikpe *et al.* reported that contamination of fish from enteric bacteria of human and animal origins may be responsible for various food spoilages. *Escherichia coli* in all the samples may be due to its ubiquitous nature as it could be found in almost all environment including human skin, water and air during processing. This result corroborated that of Majeed and MacRae (1991) who observed that most of the bacteria flora associated with spoilage of fish were Gram negative rod bacilli such as *E. coli,* and *Salmonella* species. Also, the value of E. coli was higher in frozen fish than other 3 fish samples (table 2). This is probably due to the fact that frozen fish have a higher moisture content than the other samples. Thermo sensitive bacteria like *Enterobacteriaceae* are used as indicators of hygiene conditions and contamination of food after cooking (Health Protection Agency, 2009). The detection of *E. coli* in samples (HS, FF AND BQ) also suggested a contamination by fecal matter from animal or human origin due to poor adherence to hygiene practices which has been considered as one of the main factors of contamination in this study. The presence of coliform is an indication of poor sanitation of the hawkers and could also be a result of possible feacal contamination. The presence of *E. coli, Enterococcus spp., Salmonella* sp. and *Staphylococcus aureus* is a health concern since these microorganisms carry virulent genes. The presence of Staphylococcus *aureus*, signifies that the fish might have been contaminated from source as a result of improper handling practices by processors. *S. aureus* is common inhabitants of the skin and mucous membranes and 20% to 50% of normal healthy adults may be carriers of *S. aureus* **(**Talaro & Talaro 1999). Improper handling and improper hygiene might lead to the contamination of food and this might eventually affect the health of the consumers. The presence of *Staphylococcus aureus* a normal flora of skin and mucous membrane of humans can be attributed to human contact during handling and processing (Dalgaard *et al*., 2006). *Staphylococcus aureus* produces a variety of extra cellular enzymes and toxins that have been found to be responsible for food poisoning and can rapidly develop resistance to many antimicrobial agents and pose therapeutic problems (Thrower, 2000). *Staphylococcus aureus* has long been recognized as one of the most important pathogens that causes disease in humans. Consumption of fish or fish products containing this can lead to food poisoning. The procedures carried out to maintain and preserve the quality of these fish samples from the time they are transported to various stores or retailers till the time they get to the final consumer can lead to the introduction of *Staphylococcus aureus.* Okareh and Erhahon (2015) also confirmed that *Staphylococcus sp*. are pathogenic strains which could cause food poisoning due to the heat stable *Staphylococcus* enterotoxin which is resistant to gastrointestinal enzymes. Similarly, Morales et al. (2014) claimed that Staphylococcal food contamination is usually traced to workers who are carriers and/or in contact with inadequately cleaned equipmen*t S. aureus* been one of the pathogens isolated from the frozen fish is a normal flora in man but not in fish, its presence in the samples could be attributed to

contamination from personnel and the environment (Wagner, 2013). Similarly, cross-contamination via utensil (Roche *et al*., 2003. Harrington, 2010, and Berrang *et al*., 2013). The presence of *Salmonella* sp. in the fish samples is an indication that the water used for processing was faecally contaminated. *Salmonella* sp. usually is not a fish pathogen, rather the consumption of *Salmonella* contaminated feed and water causes this infection. The impact of this pathogen in human, particularly in the very young or older generation includes gastroenteritis, abdominal cramps, enteric fever and bacteremia resulting from ingestion of uncooked or poorly cooked fishes. *Shigella flexneri* was the third common bacteria observed in this study, it is gram negative and does not ferment lactose but ferments glucose. They can be located in water and transmitted by contaminated water.it causes diarrhea, fever, headache and nausea. The presence of *Enterobacter aeorogenes* is a clear indication of environment and faecal pollution either from human, animals or poor handling practices. *Proteus mirabilis* which was identified in fish sample FF is an opportunistic pathogen whose presence might be due to faecal contamination. It primarily affects individuals whose immune system is compromised, even though the number of *Enterococcus faecalis* present was little it still indicates that the water used for processing was faecally contaminated. The result of this analysis supports the work of (Ayeloja *et al*., 2018) who carried out microbiological analysis of smoked fish processed in Benin. . Total fungal counts from the fish ranged between 5.50×10^4 - 5.6×10^5 CFU/g denoting that all the evaluated fish were microbiologically certified as been of good quality hence the counts were within the permissible level of international standard. ICMSE (1986) accepted limit of the total coliform count for frozen fish is <100 MPN/g. The total bacteria count from the fish samples as recorded in this study was between $2.50\times10^4 – 6.35\times10^4$ CFU/g. It is generally accepted that fish with microbial load of $>10^6$cfu/ml is likely to be at the stage being unacceptable from the microbiological point of view and unfit for human consumption (Cheesbroughl, 2000). The total bacterial count on fish rarely indicates the quality of the fish but it gives an indication of the risk of spoilage induced since each of these organisms had different ways of affecting health conditions of consumers of such contaminated fish (Gram *et al.*, 2000). Also, there was a significantly higher bacterial and fungal load in the hot smoked, frozen and barbequed fish samples Thatcher and Clark (1973) earlier report that the kind and number of hygiene practice during handling and smoking processes. The reason could as well be an indication of partial dehydration during smoking and unhygienic display at market stalls, as earlier observed by *Dike-Ndudim et al*. (2014). The reason for this contamination is mostly because the vendors neither underwent any form of formal training in food preparation nor did they attempt to seek it. According to FAO (1997), fish handlers should have the necessary knowledge and skills to enable them to handle food hygienically. Systems should be put in place to ensure that food handlers remain aware of all procedures necessary to maintain the safety and suitability of food. FAO (1999) recommends that every vendor/helper of food should undergo a basic training in food hygiene before licensing.

Conclusion

The findings of this study revealed that the ready to eat (Cold smoked, hot smoked fish and barbequed) fish and non-ready to eat (frozen) fish vended in open markets in awka, Anambra state are not always of satisfactory microbiological status and represent potential sources of foodborne diseases. The quality of raw material, poor hygiene practices, and inappropriate handling practices during processing and selling are factors that contribute to the unsatisfactory microbiological quality

of these products. Processors and sellers should be trained on good hygiene and handling practices in order to produce a safe and sound product for consumption.

The presence of these microorganisms in the fish samples cause spoilage and food poisoning. Microbes were isolated from the fish samples and it signals that microorganisms in fish can cause a health hazard to consumers if not hygienically handled and properly processed before consumption and labour loss to sellers if adequate preservation of fish is not relatively considered .as important factor. Though the microbial load recorded from the fishes is minimal and can be accepted, the pathogenic microorganisms isolated emphasized that fish can serve as a possible vehicle for human infection. Every home in Nigeria consumes fish on daily basis in their diet. The consumed fish are either in inform of dried, smoked, boiled or fried, barbequed and to date, there is no literature or data on disease outbreak as a result of consuming ready or non- ready to eat fishes

Fish should not just be nutritionally balanced but microbiologically safe as well. Foodborne illness can be prevented by good hygiene practices such as the use of Good Manufacturing Practices (GMP) and Hazard Analysis Critical Control Point (HACCP) application in the chain of fish production and processing Therefore government should enforce strong food safety regulations according to WHO and FAO whose permissible limit for microbial load in fish is $<10^6$cfu/ml

In addition, vendors need to be educated on food safety and hygienic practices.

Recommendation

- The need for microbial assessment of fish should be emphasized to reduce possible contamination.
- Fish can support the growth of pathogenic (food poisoning) bacteria and must be kept at certain temperatures to minimize the growth of any pathogens that may be present in the ready or non-ready to eat fish, to prevent the formation of toxins in the fish**.**
- Care should be taken during handling and processing of fishes
- Minimize microbial contamination by raising proper awareness of the health hazards it can cause.
- Education of fish mongers on the importance of maintaining good hygiene levels, and the use of treated water for fish processing are recommended.

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