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Assessment of microbial flora, characterization and public health implications of two species of smoked fish sold in 'Otuocha' Market in Anambra State

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Abstract

Microbiological assessment of fresh and smoked *Synodontis schall* and *Oreochromis niloticus* fish species was conducted. Three replicates of fresh and smoked fish samples were aseptically obtained from fishermen and open market in Otuocha area of Anambra State, Nigeria. The culture from organs of fresh fish and smoked samples were inoculated on Nutrient agar (NA.), *Salmonella Shigella* Agar (SSA), MacConkey Agar (MCA) and Potato Dextrose Agar respectively for heterotrophic bacteria and fungi counts. Colonial and microscopic characterizations of the isolates from the cultured media were examined. There no growth of coliform and *Salmonella sp* from tissue of the fresh fish and the smoked fish samples. Total plate counts of bacteria and fungi from gills, intestine and skin were found above 1 x 10^6 cfu/g safety limit recommended for foods. Bacteria count ranged from 1.27 x 10^7 cfu/g to 2.95 x 10^7 cfu/g and 1.0 x 10^6 cfu/g to 2.88 x 10^8 cfu/g for fungi. Six genera of bacteria namely *Salmonella sp*, *Escherichia coli*, *Staphylococcus sp, Enterococcus sp, Bacillus sp* and *Pseudomonas sp* were suspected from fish samples. Four fungi genera: *Saccharomyces sp, Penicillium sp, Rhizopus sp* and *Mucor sp* were indicated in this work. This assessment has revealed the level of gross contamination of fresh fish and safety of smoked fish samples from foodborne microorganisms.

Keywords: *Synodontis schall; Oreochromis niloticus*; Coliform; *Salmonella*; Smoked fish

1. Introduction

Fish is most important seafood in the world and especially in Nigeria and other Africa countries. This is because fish and fish products are important food component of the majority of people globally (FAO, 2016). Furthermore, Rahji and Bada (2010) reported that fish in human diets has risen to 2.66 million metric tons which constitute 41% of total animal protein consumed in Nigeria but represents about 14% of all animal proteins in a global basis (Abolagba and Melle, 2008). The popularity of fish is due to its availability and cheaper source of animal protein in human diet than meat (Akinwumi and Adegbehingbe, 2015; Olaleye and Abegunde, 2015). Adeyeye *et al*., (2015) and Ikutegbe and Sikoki, (2014) confirmed that fresh and smoked fish products are the most relatively and accessible animal protein source among the muscle food in developing countries in Africa.

Fish loses its freshness and its nutritional composition deteriorated due its perishable nature after death (Dehghani *et al*., 2018). The rate at which autolytic and microbial spoilage occurs in fresh fish varies after harvest; this depends on species, methods of harvesting, environmental factors and post-harvest handling. In developing countries preservation of fresh fish is a great challenge due to inadequate infrastructures especially in the rural areas; environmental and climate conditions (Anihouvi*et al.,* 2012). Many preservation methods including frying, drying, salting, fermentation

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(Adeyeye *et al*., 2015; Ikutegbe and Sikoki, 2014; Bako, 2004) has been adopted to prolong the shelf-life of fish from post-harvest. However, traditional smoking process gained more popularity among other methods due its uniqueness in flavour of smoked fish compared to other methods and convenience application of the process involved.

In reality foods harbor naturally microorganisms and unsafe owing to the presence of microorganisms which may invade human body (e.g *Salmonella, Escherichia coli, Listeria monocytogenes,* etc) (Olaleye and Abegunde, 2015; Ofred, 2009). These microorganisms multiply in number during processing. Growth of these organisms could be favored with nutrient content of the host food and contamination from atmosphere or utensils or workers. Some of these microorganisms produce toxins in food which have potential of poison agents cause symptoms to human consumption (Ofred, 2009).

Fish and shellfish are common vehicles of transmitting food borne diseases (Ofred, 2009). The diseases caused by contaminated foods constitute one of the most widespread health problems (Olaleye and Abegunde, 2015). Pilet and Leroi (2011) reported that fish and fish products contribute 10%–20% of food-borne diseases; and thus microbiological safety of foods becomes an important public health concern in order to achieve food security. Bacteria such as *Staphylococcus aureus*, *Salmonella* spp., *Escherichia coli*, and *Listeria monocytogenes* have been fingered in food-borne causing organisms in fish and fish products(Likongwe*et al*., 2018; Udochukwu *et al*. 2016; Adeyeye *et al*., 2015; Ayeloja*et al*., 2018; Nunoo and Kombat, 2013). Another concern is the contamination by fungi. However, at present it has also been found that in certain areas the aquatic environment is a natural reservoir for *Vibrio cholerae*. Microbiologists are of the opinion that the presence of *Escherichia coli* in food indicates faecal contamination in food. However, there is scarcity of scientific information on prevalence of pathogens on fish and other street vended or traded food in Nigeria markets thus the need for this study with the objective of determining microbial flora that are prevalent on smoked fish sold in Otuocha markets in Anambra State Nigeria.

2. Materials and methods

2.1. Sample Collection, Identification and Preparation

2.1.1. Fish Sampling

Three (3) replicates of two common species of the fresh fish samples were bought from local fishermen at the fish landing site in Otuocha community area of Anambra State. The fish species were identified using standard taxonomic and description indices described by Sikoki and Francis (2007). Three replicates of the smoked fish samples of the identified species studied in this work were obtained randomly from the retailers at different sales-point in Otuocha market of Anambra State. Fresh and smoked samples were aseptically packaged in laboratory sterile plastic bags and transported to the laboratory for analysis.

3. Microbiological analysis

3.1. Preparation of Media

The media used are Nutrient agar (NA.), *Salmonella Shigella* Agar (SSA), MacConkey Agar (MCA) and Potato Dextrose Agar. All media used were prepared according to the manufacturers' instructions. The mean counts of bacteria and mould in colony forming units per gram (cfu/g) of samples were determined.

3.2. Preparation of Cultures

Fresh fish samples were dissected into flesh/tissue, gills, intestine and skin and the smoked fish samples were macerated under aseptic conditions. The samples were serially diluted and appropriate dilutions (0.1 mL) of each samples was inoculated on different agar media. All cultures were incubated in duplicate at 37ºC for 24 – 48 hours. The bacteria were inoculated on Nutrient Agar for 24-48 hours, *Salmonella Shigella* Agar (SSA) for 24 hours and Potato dextrose agar (PDA) for 24 hours. Colonies on plates containing 30 - 306 colonies were counted and multiplied by the dilution factor (Cheesbrough, 2002; ICMSF, 2005).

Microbiological profile (coliform and moulds) of the samples was determined based on cultural microscopic and biochemical characteristics of representative colonies from plates of coliform and moulds purified by repeated streaking and sub-culturing, respectively (ICMSF, 2005). Moulds were identified using the wet mount method described by Cheesbrough (2002).

Table 1 shows mean total plate count of culture fish samples. The mean values are varied among the samples; where $4.1x10^9$cfu/g and $7.8x10^8$cfu/g were total heterotrophic bacteria found on the skin of fresh *Synodontis schall* and *Oreochromis niloticus* fish samples respectively. These values were the highest total bacteria counts when compared to 1.87 $x10^7$cfu/g, and $1.27x10^7$cfu/g of tissue, gills and intestine of *Synodontis schall* specie; as well as $1.11x10^5$cfu/g; $2.69x10^9$cfu/g and $2.95x10^9$cfu/g of *Oreochromis niloticus* specie. The colony counts obtained in fish samples organs with exception of tissue organ are more than the recommended count of 1x10 cfu/g (ICMSF, 2005) fish for good bacteriological quality in both species. This variation on mean total plate counts could be attributed to rate of exposure of the organs (more importantly the fish flesh and gill) to polluted water environment. The least TPC counts recorded on flesh/tissue samples could be as a result of gross contamination during post-harvest fish handling which depend on hygienic practices of the handlers. However the pattern of total heterotrophic plate counts found on the cultured fish samples organs in this work were in line with the findings of Olayemi *et al*. (2012), Ibrahim *et al.* (2014); Olaleye and Abegunde (2015) who reported higher bacteria counts were above recommended safety limit for fish products.

Table 1 Mean Total plate counts (TPC) on fresh and smoked fish samples

Fish organs	Total Heterotrophic Bacteria Counts (cfu/g)	Total Heterotrophic Fungal Counts (cfu/g)	Total Coliform counts (cfu/g)	Total counts on SSA (cfu/g)
Fresh *Synodontis schall*				
Flesh/tissue	3.7×10^4	2.8×10^3	No growth	2.0×10^3
Gills	1.87×10^7	1.8×10^7	2.0×10^4	3.6×10^7
Intestine	1.27×10^7	3.1×10^7	1.0×10^4	3.2×10^7
Fresh skin	4.1×10^9	1.0×10^6	1.0×10^6	6.5×10^6
Fresh *Oreochromis niloticus*				
Flesh/tissue	1.11×10^5	1.9×10^4	No growth	1.1×10^4
Gills	2.69×10^9	2.88×10^8	$2.34\ 10^8$	2.75×10^8
Intestine	2.95×10^9	1.72×10^8	1.5×10^7	1.05×10^7
Fresh skin	7.8×10^8	3.6×10^7	5.0×10^6	3.0×10^6
Smoked fish				
S-SS	2.0×10^6	2.2×10^6	No growth	No growth
S-ON	7.2×10^6	4.2×10^6	No growth	No growth

There was no coliform bacteria growth in tissue organs of fresh fish species. Coliform counts between 2.0×10^4cfu/g and 2.34×10^8cfu/g of gills, intestine and skin organs were recorded in both fish species; these counts were above 1x10cfu/g recommended for food safety (ICMSF, 2005). The presence of coliform bacteria is used as indicator for fecal contamination in food especially in water environment where the fish lives. The growth counts of bacteria on *Salmonella Shigella* Agar (SSA) were not the same across the fresh fish organs. The counts ranged from 2.0×10^3cfu/g to 3.6×10^7cfu/g of *Synodontis schall* specie organs and 1.1×10^4cfu/g to 2.75×10^8 cfu/g of *Oreochromis niloticus* fish organs.

The total fungi counts of the fish samples were not the same across the organs of the fish. Tissue organ of the two species of the fish samples had the least counts of 2.8×10^3cfu/g and 1.9×10^4cfu/g for *Synodontis schall* and *Oreochromis niloticus* fish species respectively. The gills and intestine organs of the fish species had highest heterotrophic fungi counts of 1.8×10^7cfu/g and 3.1×10^7cfu/g for *Synodontis schall* and 2.88×10^8cfu/g and 1.72×10^8cfu/g *Oreochromis niloticus* respectively. More so, the fungi population found in this fish samples are higher than $<10^6$cfu/g a safe fungal load for food (Cheesbrough, 2002), hence, a look at the high fungi counts found in work suggest health risk exposed to consumers' without adequate post-harvest preservation.

The mean total heterotrophic plate counts and fungi counts on smoked samples of the two species of fish used in this work were varied. Bacteria counts of 2.0×10^6cfu/g and 7.2×10^6cfu/g were isolated while fungi counts were of 2.2×10^6cfu/g and 4.2×10^6 cfu/g from *Synodontis schall* and *Oreochromis niloticus* species respectively. In both bacteria and fungi counts were found slightly above1x10cfu/g recommended for food safety (ICMSF, 2005). This variation and total

heterotrophic counts in this recorded could explained by the fact that the samples were collected from various sellers where the quality of the raw material varied, as well as handling and hygiene practices (Anihouvi *et al.* 2019). There was no growth of coliform bacteria and *Salmonella Shigella* agar from smoked fish samples. This result suggested absent of coliform bacteria and salmonella bacteria; this implies that the smoked fish were safe from foodborne disease organisms.

Table 2 and Table 3 shows the colonial and microscopic characteristics of bacteria isolates cultured from fresh fish organs. *Salmonella sp* and *Shigella sp* were suspected on *Salmonella Shigella* agar cultured and *Escherichia sp* were revealed from MacConkey agar respectively. These suspected organisms from the fresh fish organs may constitute a hazard to consumers' health (Dierick *et al.*, 2005; EFSA and ECDC, 2016) Culture from smoked fish samples showed any colonial and microscopic in both media. These results further corroborate the findings in Table 1. However, the absence of *Salmonella sp, Shigella sp* and *Escherichia coli* from smoked fish samples from Otuocha market Anambra indicated that there was no contamination with enteric organisms by handlers during retail sales. This could be attributed to the effectiveness of smoke kiln used (Olayemi *et al.,* 2012), level of hygiene and environmental sanitation practice in the market.

Table 2 Colonial and microscopic characteristics of bacteria isolates on *Salmonella Shigella* Agar

Fish organs	Colonial characteristics	Microscopic characteristics	Motility	Spore formation	Most probable identity
Fresh *Synodontis schall*					
Flesh/tissue (SFM)	Black shiny fish eye colonies	Gram negative rods in singles	+	-	*Salmonella* sp
Gills (SFG)	Black shiny fish eye colonies	Gram negative rods in singles	+	-	*Salmonella* sp
Intestine (SFI)	Black shiny fish eye colonies	Gram negative rods in singles	+	-	*Salmonella* sp
Fresh skin (SFS)	Black shiny fish eye colonies	Gram negative rods in singles	+	-	*Salmonella* sp
Fresh *Oreochromis niloticus*					
Flesh/tissue (NFM)	Moist and shiny light pink colonies	Gram negative slender rods	-	-	*Shigella* sp
Gills (NFG)	Black shiny fish eye colonies	Gram negative rods in singles	+	-	*Salmonella* sp
Intestine(NFI)	Black shiny fish eye colonies	Gram negative rods in singles	+	-	*Salmonella* sp
Fresh skin (NFS)	Moist and shiny light pink colonies	Gram negative slender rods	-	-	*Shigella* sp
Smoked fish					
S-SS	nil	nil	nil	nil	nil
S-ON	nil	nil	nil	nil	nil

Key: SFM – *Synodontis* fresh muscle, SFG – *Synodontis* fresh gill, SFI – *Synodontis* fresh intestine, SFS – *Synodontis* fresh skin; NFM - *Niloticus* fresh muscle, NFG - *Niloticus* fresh gill, NFI - *Niloticus* fresh intestine, NFS - *Niloticus* fresh skin; S-SS - Smoked *Synodontis schall;* S-ON -Smoked *Oreochromis niloticus*

Table 3 Colonial and microscopic characteristics of bacteria isolates on MacConkey Agar

Sample code	Colonial characteristics	Microscopic characteristics	Motility	Spore formation	Most probable identity
Fresh *Synodontis schall*					
Flesh/tissue	No growth	No growth			
Gills (SFG)	Small circular moist and shiny pink colonies	Gram negative rods predominantly in singles	+	-	*Escherichia coli*
Intestine (SFI)	Small circular moist and shiny pink colonies	Gram negative rods predominantly in singles	+	-	*Escherichia coli*
Fresh skin (SFS)	No growth	No growth	nil	nil	nil
Fresh *Oreochromis niloticus*					
Flesh/tissue (NFM)	Small circular moist and shiny pink colonies	Gram negative rods predominantly in singles	+	-	*Escherichia coli*
Gills (NFG)	Small circular moist and shiny pink colonies	Gram negative rods predominantly in singles	+	-	*Escherichia coli*
Intestine (NFI)	Small circular moist and shiny pink colonies	Gram negative rods predominantly in singles	+	-	*Escherichia coli*
Fresh skin (NFS)	Small circular moist and shiny pink colonies	Gram negative rods predominantly in singles	+	-	*Escherichia coli*
Smoked fish					
S-SS	No growth	No growth	nil		nil
S-ON	No growth	No growth	nil		nil

Key: SFM - *Synodontis* fresh muscle, SFI -*Synodontis* fresh intestine, SFG -*Synodontis* fresh gill, SFS -Synodontis fresh skin**; B**NFM -*Niloticus* fresh muscle, NFG -*Niloticus* fresh gill, NFI -*Niloticus* fresh intestine, NFS -*Niloticus* fresh skin**;** S-SS - Smoked *Synodontis schall,* S-ON -Smoked *Oreochromis niloticus*

Furthermore *Staphylococcus* sp, *Enterococcus* sp, *Bacillus sp*and *Pseudomonas* sp (Table 4 and Table 5) were the foodborne pathogens suspected from cultures from fresh fish organs isolated on Nutrient agar. Three pathogenic organisms namely *Bacillus* sp*, Staphylococcus* sp and *Enterococcus* sp (Table 6) were revealed from smoked fish samples. These suspected organisms may have contaminated fresh and smoked fish samples through environment, fishing equipment and handlers (Okonko *et al*., 2008). Most of the suspected food pathogens identified in this work cause diseases in human (Herman *et al.* 2011; Adelaja *et al.,* 2013).

Four (4) fungi general namely *Saccharomyces* sp*, Penicillium sp, Rhizopus* sp and *Mucor* sp are suspected from fresh fish organs (Table 7) and smoked fish (Table 8) samples respectively. The few mycoflora suspected from fish samples in this work was in line with reports of Akani and Nwankwo (2019); Sani *et al*., (2016) and Samuel *et al*., (2015). Despite few fungi general found in this work, it is important to recall that the total heterotrophic fungi counts (Table 1) from fresh *Synodontis schall* organ samples falls within $\log_{10}$ x 10^6cfu/g safety limit (ICMSF, 2005; Cheesbrough, 2002) recommended for food but slightly higher in fresh *Oreochromis niloticus* organ samples. The absence of fungi growth from smoked fish samples this work implies the samples may not pose any public health risk(s) to consumers' with respect to mycoflora threat.

Table 4 Colonial and microscopic characteristics of bacteria isolate from fresh *Synodontis schall* on Nutrient Agar

Sample code	Colonial characteristics	Microscopic characteristics	Motility	Spore formation	Most probable identity
Flesh/tissue SFM	Smooth and shiny golden yellow colonies Small circular moist and shiny cream colonies Dull and dry serrated flat cream colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad Gram positive cocci in chains Gram positive beaded rods in short chains	- - +	- - +	*Staphylococcus* sp *Enterococcus* sp *Bacillus* sp
Gills SFG	Smooth and shiny golden yellow colonies Small circular moist and shiny cream colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad Gram positive cocci in chains			*Staphylococcus* sp *Enterococcus* sp
Intestine SFI	Smooth and shiny golden yellow colonies Small circular moist and shiny cream colonies Bluish-green moist colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad Gram positive cocci in chains Gram negative rods in singles	- - -	- -	*Staphylococcus* sp *Enterococcus* sp *Pseudomonas* sp
Fresh skin (SFS)	Smooth and shiny golden yellow colonies Small circular moist and shiny cream colonies Dull and dry serrated flat cream colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad Gram positive cocci in chains Gram positive beaded rods in short chains	+ + +	_ _ _	*Staphylococcus* sp *Enterococcus* sp *Bacillus* sp

Key: SFM – Synodontis fresh muscle**;** SFG – Synodontis fresh gill **;**SFI – Synodontis fresh intestine **;** SFS - Synodontis fresh skin

Table 5 Colonial and microscopic characteristics of bacteria isolate from fresh *Oreochromis niloticus* on Nutrient Agar

Sample code	Colonial characteristics	Microscopic characteristics	Motility	Spore formation	Most probable identity
Flesh/tissue (NFM)	Smooth and shiny golden yellow colonies Small circular moist and shiny cream colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad Gram positive cocci in chains	+ +	_ _	*Staphylococcus* sp *Enterococcus* sp
Gills (NFG)	Smooth and shiny golden yellow colonies Small circular moist and shiny cream colonies Dull and dry serrated flat cream Bluish-green moist colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad Gram positive cocci in chains Gram positive beaded rods in short chains Gram negative rods in singles			*Staphylococcus* sp *Enterococcus* sp *Bacillus* sp *Pseudomonas* sp
Intestine (NFI)	Smooth and shiny golden yellow colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad			*Staphylococcus* sp

	Small circular moist and shiny cream colonies Bluish-green moist colonies	Gram positive cocci in chains Gram negative rods in singles			*Enterococcus* sp
Fresh skin (NFS)	Smooth and shiny golden yellow colonies Small circular moist and shiny cream colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad Gram positive cocci in chains			*Staphylococcus* sp *Enterococcus* sp

Key: NFM -*Niloticus* fresh muscle **;**NFG -*Niloticus* fresh gill**;** NFI - *Niloticus* fresh intestine **;** NFS - *Niloticus* fresh skin

Table 6 Colonial and Microscopic Characteristics of Bacteria Isolate from Smoked Fish on Nutrient Agar

Sample code	**Colonial characteristics**	**Microscopic characteristics**	**Motility**	**Spore formation**	**Most probable identity**
S-SS	Mucoid slimy rough cream colonies Dull and dry serrated flat cream colonies	Gram positive rods in chains Gram positive beaded rods in short chains			*Bacillus* sp *Bacillus* sp
S-ON	Smooth and shiny golden yellow colonies Small circular moist and shiny cream colonies Dull and dry serrated flat cream colonies	Gram positive cocci predominantly in clusters, few in pairs and tetrad Gram positive cocci in chains Gram positive beaded rods in short chains			*Staphylococcus sp* *Enterococcus* sp *Bacillus sp*

Key: S-SS - Smoked *Synodontis schall***;** S-ON - Smoked Oreochromis niloticus

Table 7 Colonial and Microscopic Characteristics of Fungal Isolates from fresh on Potato Dextrose Agar

Sample code	**Colonial characteristics**	**Microscopic characteristics**	**Identity of isolates**
Fresh *Synodontis schall*			
Flesh/tissue (SFM)	Small circular cream colonies Small smooth moist and shiny golden yellow colonies, White thread-like flat colonies	Large gram positive oval budding cells Gram positive spherical budding cells	*Saccharomyces* sp *Saccharomyces* sp
Gills (SFG)	Moist and shiny butyrous cream colonies Dirty green dry spores enclosed in white periphery	Gram positive ellipsoidal budding cells Hyphae septate. Conidia mop head like	*Saccharomyces* sp *Penicillium sp*
Intestine (SFI)	Small circular cream colonies Small smooth moist and shiny golden yellow colonies White thread-like flat colonies	Large gram positive oval budding cells Gram positive spherical budding cells	*Saccharomyces* sp *Saccharomyces* sp
Fresh skin (SFS)	Tall white filamentous hyphae with orange spores at the tip	Non septate hyphae. Spores enclosed in a sporangium	*Rhizopus sp*
Fresh *Oreochromis niloticus*			
Flesh/tissue (NFM)	Small circular cream colonies	Large gram positive oval budding cells	*Saccharomyces* sp

	Small smooth moist and shiny golden yellow colonies	Gram positive spherical budding cells	*Saccharomyces* sp
Gills (NFG)	Small circular cream colonies Small smooth moist and shiny golden yellow colonies Dirty green dry spores enclosed in white periphery	Large gram positive oval budding cells Gram positive spherical budding cells Hyphae septate. Conidia mop head like	*Saccharomyces* sp *Saccharomyces* sp *Penicillium sp*
Intestine (NFI)	Small circular cream colonies Small smooth moist and shiny golden yellow colonies	Large gram positive oval budding cells Gram positive spherical budding cells	*Saccharomyces* sp *Saccharomyces* sp
Fresh skin NFS)	Small circular cream colonies Small smooth moist and shiny golden yellow colonies	Large gram positive oval budding cells Gram positive spherical budding cells	*Saccharomyces* sp *Saccharomyces* sp

Key: SFM – *Synodontis* fresh muscle, SFG – *Synodontis* fresh gill, SFI – *Synodontis* fresh intestine, SFS - *Synodontis* fresh skin; NFM -*Niloticus* fresh muscle, NFG -*Niloticus* fresh gill, NFI -*Niloticus* fresh intestine, NFS -*Niloticus* fresh skin

Table 8 Colonial and Microscopic Characteristics of Fungal Isolates from Smoked Fish on Potato Dextrose Agar

Sample code	**Colonial characteristics**	**Microscopic characteristics**	**Identity of isolates**
S-SS	Small circular cream colonies Small smooth moist and shiny golden yellow colonies Short white cotton wool like mycelia	Large gram positive oval budding cells Gram positive spherical budding cells Hyphae non septate. Spores enclosed in a sporangium. Sporangiophore septate	*Saccharomyces* sp *Saccharomyces* sp *Mucor* sp
S-ON	Small circular cream colonies Small smooth moist and shiny golden yellow colonies White thread-like flat colonies	Large gram positive oval budding cells Gram positive spherical budding cells	*Saccharomyces* sp *Saccharomyces* sp *Saccharomyces* sp

Key: S-SS - Smoked *Synodontis schall***;** S-ON -Smoked *Oreochromis niloticus*

4. Conclusion

The findings from this work revealed that the organs such as skin, gills and intestine of species of fresh fish studied showed presence of coliform, *Salmonella* sp and pathogenic organisms at growth counts above safety limit. This suggested heavy polluted environment of the source of fresh fish naturally; unhygienic post-harvest handling practice of the fishermen. These bacteria found in this work on fresh organs should be treated as foodborne indicators, since they are capable of causing serious infections and food poisoning.

The smoked fish samples sold in Otuocha market area of Anambra satisfactory safe from coliform and *Salmonella* contaminations. Foodborne pathogens and four general of fungi organisms were found in smoked fish samples at counts that falls on upper (10^6cfu/g) safety limit level recommended for foods. The smoking process and hygiene practices in the market may have helps in preservation of the smoke fish samples.

Compliance with ethical standards

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Disclosure of conflict of interest

No conflict of interest to be disclosed.

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