PUBLIC HEALTH RISKS AND MICROBIOLOGICAL ASSESSMENT OF FRESH AND SMOKED FISH IN PORT HARCOURT MARKETS

¹Emeka Samuel Obiora and ²Ngozi Patricia Amaechi

¹Department of Fisheries and Aquatic Environment, Rivers State University, Port Harcourt

²Department of Home Science and Management, Rivers State University, Port Harcourt

DOI: https://doi.org/10.5281/zenodo.13938888

Abstract: The microbiological quality of fresh and smoked fishes sold around selected markets (Mile 3, Mile 1 and Creek Road) in Port Harcourt City was carried out. The microbial count of the fresh fish was higher than that of the smoked ones. The bacterial count of the fresh fish ranged from 0.35×101cfu/g (Salmonella) to 1.6918×103 cfu/g(Total Bacterial Count) and the smoked one ranged 0.5×101cfu/g(Salmonella) to 1.042×103cfu/g (Total Bacterial Count). The fungal load of the fishes ranged between 1.867×102cfu/g for the smoked and 4.860×102,cfu/g) for the fresh ones. Statistically, there was no significant difference between the fresh and smoked fish with respect to microbial load at p<0.05. The moisture content of the fresh fish was higher (74.48±4.52) than that of the smoked fish (53.48±2.06) just like the pH of the fresh (6.57 ± 0.10) and smoked fish (6.17 ± 0.14) . The bacteria and fungi isolated from fish in the Creek road market (1550±265cfu/g, 381.50±1.75cfu/g) were significantly higher/different from those of the mile 3 market (1262.50±310cfu/g, 307.50±1.69cfulg) and the mile 1 market (1287.75±6.15cfu/g, 315±197cfu/g) at p<0.05. Bacillus substilis Krebsiella spp, Staphylococcus aureus and Streptococcus spp were found only on smoked fish while Acinetobacter spp, Corynecbacteruim spp, Flavobacterium spp, Enterobacter spp and Salmonella species were found on fresh. Bacteria like Escherichia coli, Micrococcus luteus, Pseudomonas aeruginosa, Proteus spp and Serratia spp were found on both fresh and smoked fish. Fungal species such as Penicillium expansum, Aspergillu spp, Fusarium spp, Rhizopus stolonifer and Mucor piriformis were found on both fresh and smoked fish. The microbial metries or count showed that they all exceeded the permissible limits of the World Health Organization (WHO), Standard Organization of Nigeria (SON) and the National Environmental Standard Regulation Enforcement Agency (NESREA). The microbial count showed some level of contamination under the influenced of pH and moisture. It is therefore crucial to implement strict hygiene and safety standards throughout the fish supply chain to mitigate these risks of consuming contaminated seafood.

Keywords: Comparative Evaluation, Microbiological Quality, Public Health Risks, Fresh and Smoke Fish, Port Harcourt

INTRODUCTION

Fish and fishery products are essential components of the human diet, providing a valuable source of high-quality protein, vitamins, minerals, and beneficial omega-3 fatty acids (FAO,2020). The consumption of fish and fish

products has been associated with numerous health benefits, including reduced risk of cardiovascular diseases, improved cognitive function, and enhanced immune system (Domingo,2016, Sioen et al.,2007). Consequently, there has been a growing global demand for fish and fish-based products in recent years.

In Nigeria, the consumption of smoked fish is a widespread and popular practice, particularly in the southern region, including the city of Port Harcourt (Adeyemi and Osilalu,2019). Smoked fish is a preferred choice among consumers due to its distinct flavor, extended shelf-life, and perceived health benefits (Eyo,2001). However, the microbiological quality of smoked fish is a major concern, as the smoking process and subsequent handling and storage conditions can significantly impact the growth and proliferation of pathogenic microorganisms (Obemeata et al.,2011, Gram and Huss,1997).

Microbiological contamination of fish and fish products can pose significant public health risks to consumers, leading to the transmission of foodborne illnesses, such as salmonellosis, listeriosis, and staphylococcal food poisoning (Huss, 1997, Omeiza et al., 2013). These pathogenic bacteria can be introduced during various stages of the production and distribution chain, including harvesting, processing, handling, and storage (ICMSF, 1998). Moreover, the consumption of contaminated fish and fish products can result in severe gastrointestinal symptoms, hospitalization, and even fatalities in vulnerable populations, such as the elderly, young children, and immuno compromised individuals (Scallan et al., 2011, Nyarko et al., 2011).

While several studies have investigated the microbiological quality of smoked fish in different regions of Nigeria, there is limited research on the comparative assessment of the microbiological quality between smoked and fresh fish sold in the Port Harcourt metropolitan area (Adeyemi, & Osilalu, 2019, Odu, & Ukpoma, 2013). This information is crucial for understanding the potential public health risks associated with the consumption of these fish products and developing targeted interventions to improve food safety.

Therefore, the present study aims to conduct a comparative assessment of the microbiological quality of smoked and fresh fish sold in selected markets in Port Harcourt City, Nigeria, and to evaluate the potential public health impact on consumers. The findings from this research will contribute to the existing knowledge base and provide valuable insights for policymakers, regulatory authorities, and the fish processing industry to enhance the microbial safety and quality of fish products in the study area.

Materials and Methods.

Study Area and Sample Collection:

The study was conducted in Port Harcourt City, the capital of Rivers State, Nigeria. Port Harcourt is located in the southeastern region of Nigeria and serves as the capital of Rivers State. The city is situated on the eastern bank of the Bonny River, approximately 40 kilometers inland from the Atlantic Ocean. Its geographic coordinates are approximately 4.8156° N latitude and 7.0495° E longitude (Figure 1). It is a major commercial and industrial hub in the southern region of the country, with a large population and thriving fish markets (Adeyemi and Osilalu, 2019). Port Harcourt is surrounded by tropical rainforest vegetation, featuring a diverse range of plant species. The area is known for its dense foliage, including hardwood trees, shrubs, and various grasses. The rich biodiversity supports both terrestrial and aquatic ecosystems, particularly in the adjacent mangrove swamps.

A total of 120 fish samples of Silver catfish (Chrysichtys nigrodigitatus) were purchased from the three markets (Mile 3, Mile1and Creek Road) in Port Harcourt. These samples included 60 smoked and 60 fresh fish

representing one of the most common fish type consumed in the study area (Eyo, 2001). The fish samples were collected in sterile bags and transported to the laboratory for microbiological analysis within 2 hours of collection (Gram and Huss, 1996).

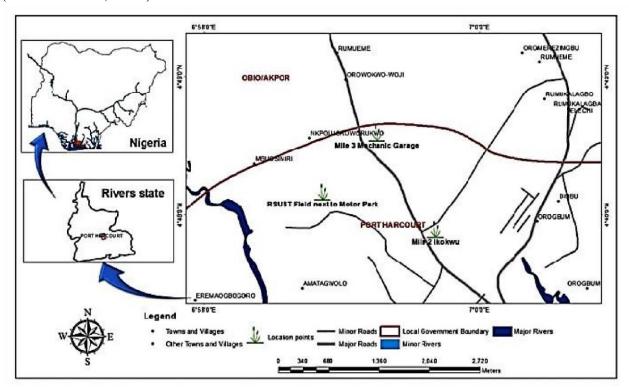


Figure 1: Map Showing the Sampling Locations in Port Harcourt Microbiological Analysis:

Upon arrival at the laboratory, the fish samples were processed for microbiological examination. The skin, gills, and intestinal contents of the fish were aseptically homogenized, and serial dilutions were prepared using sterile saline solution (0.85% NaCl) (ICMSF, 1998).

Enumeration of Total Viable Counts (TVC):

To determine the total viable counts (TVC) of bacteria, aliquots of the appropriate dilutions were pour-plated onto Plate Count Agar (PCA) and incubated at 37°C for 24-48 hours. The number of colony-forming units (CFU) per gram of fish sample was then calculated (ICMSF, 1998).

Isolation and Identification of Pathogenic Bacteria:

For the isolation and identification of pathogenic bacteria, the fish samples were inoculated onto selective media, including Salmonella-Shigella (SS) Agar for the isolation of Salmonella and Shigella species (Obemeata et al.,2011), Mannitol Salt Agar (MSA) for the isolation of Staphylococcus species (Huss,1997) and Eosin Methylene Blue (EMB) Agar for the isolation of Escherichia coli (Scallan et al.,2011).

The inoculated plates were incubated at 37°C for 24-48 hours. Presumptive colonies were further identified using standard biochemical and morphological tests, such as Gram staining, catalase, oxidase, and API identification systems (Omeize et al., 2011).

Fungal Enumeration

Fungal counts were determined by inoculating 1 g of the homogenized sample onto Potato Dextrose Agar (PDA) plates, followed by incubation at 25°C for 5-7 days. The colonies were counted and expressed as CFU/g.

pH and Moisture Content Determination

pH Measurement: The pH of each fish sample was measured using a calibrated pH meter. A 10 g sample was mixed with 90 mL of distilled water, stirred for 30 minutes, and the pH was recorded.

Moisture Content: Moisture content was determined by the oven-drying method. A 5 g sample was weighed and dried in an oven at 105°C until a constant weight was achieved. The moisture content was calculated as a percentage of the initial weight.

Statistical Analysis:

The data collected from the microbiological analysis were subjected to appropriate statistical tests, including one-way analysis of variance (ANOVA) and post-hoc comparisons, to determine the significant differences in the microbiological quality between smoked and fresh fish samples. The level of statistical significance was set at p < 0.05 (Nyarku et, al.2011).

Ethical Considerations:

The study protocol was reviewed and approved by the Ethics Committee of the Rivers State University, Nkpolu-Oroworukwo, Port Harcourt, Nigeria. Informed consent was obtained from the fish vendors before sample collection and the confidentiality of the data was maintained throughout the study (WMA, 2013).

RESULTS

The results of the study showed that the microbial count of the fresh fish was higher than that of the smoked fish with the fresh ranging from $0.35\times10^{\circ}$ cfu/g (Salmonella) to 1.6918×10^{3} cfu/g(Total Bacterial Count) and the smoked one ranging from 0.5×10^{1} ,cfu/g (Salmonella) to 1.0416×10^{3} ,cfu/g(Total Bacterial Count) (Table1) respectively. The fresh fish also had higher fungal load $(4.860\times10, \text{ cfu/g})$ than the smoked ones $(1.8667\times10^{2}\text{cfu/g})$ (Table1). Statistically, Table 2 showed that there was no significant difference between the fresh and smoked fish with respect to microbial load at p<0.05.

The moisture content of the fresh fish was higher (74.48 ± 4.52) than that of the smoked fish (53.48 ± 2.06) just like the pH of the fresh (6.57 ± 0.10) and smoked fish (6.17 ± 0.14) Table 2) **Table 1: Mean Value**

Fish Type	Total Bacte	Fungal Count		
	Count (CFU/g	(CFU/g)	(CFU/g)	(CFU/g)
Fresh	1691.83±191	325.8±55	1.63	486.67±66.8
	$(1.691x10^3)$	$3.258x10^2$	0.35×10^{1}	4.86×10^{2}
Smoked	1041.6 ± 276	54.58 ± 23	0.50 ± 0.55	186.67±59.99
	$(1.044x10^3)$	5.458×10^{1}	0.5×10^{1}	1.867×10^{2}

of Microbial Load of Fresh and Smoked Fish ((Chrysichtys nigrodigitatus) in the Study Area

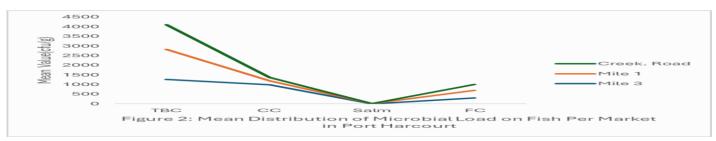
Table 2: Test for the Microbial Loa d of the Fresh and Smoked Fish in the St udy Area

Fish Type	Mean SD		t	df	Sig(2-tailed
Fresh 43.34	17 646.8	0	1.634	5	0.163
Smoked	48.6350	70.51	1.690	5	0.152

Table 3: pH Value and Moisture Content of Fresh and Smoked Fish in the Study Area

Parameters	Fresh Fish	Smoked Fish
pH Value	6.57±0.10	6.16±0.14
Moisture Content	74.48±4.52	53.48±2.66

The bacteria and fungi isolated from fish in the Creek road market $(1550\pm265\text{cfu/g}, 381.50\pm1.75\text{cfu/g})$ were significantly higher/different from those of the mile 3 market $(1262.50\pm310,\text{cfu/g}, 307.50\pm1.69\text{cfu/g})$ and the mile 1 market $(1287.75\pm6.15\text{cfu/g}, 315\pm197\text{cfu/g})$ at p<0.05(Figure 2).



The result showed bacteria such as Bacillus substilis Krebsiella spp, Staphylococcus aureus and Streptococcus spp were found only on smoked fish while Acinetobacter spp Corynecbacteruim spp, Flavobacterium spp, Enterobacter spp and Salmonella were found on fresh fish (Table 4). Bacteria like Escherichia coli, Micrococcus luteus, Pseudomonas aeruginosa, Proteus spp and Serratia spp were found on both fresh and smoked fish. Similarly, fungi such as Penicillium expansum, Aspergillu spp, Fusarium spp, Rhizopus stolonifer and Mucor piriformis were found on both fresh and smoked fish.

Table 5 showed the main values and the pernissibe.limits of the national and international agencies of the microbial metrics. The mean values of the microbial metries or count showed that they all exceeded the permissible limits of the World Health Organization (WHO), Standard Organization of Nigeria (SON) and the National Environmental Standard Regulation Enforcement Agency (NESREA). Total bacteria count in the fish was 1366.75 ± 408.293 cfu/g against the permissible limit of <1000 cfu/g for fresh and smoked fish while fungal count was 336.67 ± 167.947 cfu/g against<100 cfu/g.

Table 5: Bacterial and Fungal Isolates on Fresh and Smoked Fish in the Study Area

S/N	Bacteria Species	Fresh	Smoked	Fungal species	Fresh	Smoked
		Fish	Fish		Fish	Fish
1	Acinectobacta spp	+		Aspergillus expansium	+	+
2	Bacillus substilis		+	Fusarium spp	+	+
3	Corynebacterium spp	+		Mucor piriformis	+	+
4	Escherichia coli	+	+	Rhizopus stolonifera	+	+
5	Flavobacterium spp	+		Saccharomycus spp	+	+
6	Klebsiella spp		+			
7	Enterobacter spp	+				
8	Micrococcus luteus	+	+			
9	Pseudomonas aeruginosa	+	+			
10	Staphylococcus epidermidis	+				

11	Staphylococcus aureus		+		
12	Streptococcus spp		+		
13	Proteus spp	+	+		

DISCUSSION

The observed difference microbial load in fresh than dried fish in the study could be attributed to the hygienic condition under which they were handled (Tiamigu et al; 2011). This is in line with the assertions by the researchers (leroi et al; 1998 Huss 1995, Mez-Guillen et al, 2002, AdebayoTayo et al 2009, FAO 2009, Sattar et al 2000) that maintaining high hygienic standard at every stage from harvesting to consumer handling is essential for controlling microbial load of both fresh and dried fish. The observed influence of moisture content and pit on the microbial load of the fresh and dried fish in their study is In agreement with the finding of Udochukwu et al (2016) in Benin City. Huss (1995) opined that high moisture content in fresh fish facilitate the growth of bacteria while Adebayo Tayo et al (2009) disclosed that low moisture content in fish reduce microbial loads in dried fish. According to Omojola (2005) moisture content and pH interact to influence the microbial ecology of fish.

The presence of the bacteria such as Bacillus substilis, Klebsiella spp, Staphylococcus aureus and Streptococcus spp on smoked fish in this study is in consonance with the observation of Udochukwu et al (2016) in Benin City. According to Adesiyun et al (2006) the presence of Staphylococcus aureus is of safely concern because they produce enterotoxins that lead to food poisoning and can be introduced into fish through contaminated hands, surfaces or processing equipment. Claucus and ward (1996) reported Staphylococcus aureus to occur naturally as microflora of fish and shellfish. Kosygin et al (1990) also reported Bacillus substilis, Staphylococcus aureas Proteus mirabilis Klebsiella spp, Salmonella typical and Streptococcus spp to be associated with smoked fish through human handlers, air and soil. Adebayo-Tayo et al (2009) opined that the presence of Enterobacter spp in the fish sample is a strong indication of faecal contamination which poses risks for food safety and public health. Efstathiou et al; (2011) also regarded Enterobacter spp as indicator organisms in seafood products. Ipki and offem (2008) reported Staphylococcus aureus Escherichia coli and Pseudomonas fluoresecens, Salmonella spp from the gills, intestine and whole body of catfish Clarias gariepinus and seafood in Malaysia.

The presence of Aspergillus spp, Rhizopus sp Fusarium spp and Penicillium spp in smoked fish species could be attributed to reabsorption of moisture from the environment during storage which led to the growth of microorganism in addition to poor handling, processing and display (Christiana et al; 2010). The exceedance of bacterial and fungal counts in fish in this study could be primarily caused by poor hygiene, practices, inadequate processing, improper storage and consumer mishandling (Adebayo-Tayo et al 2009 Huss 1995, Efstathiou et al; 2011)). The health implication of consuming contaminated fish ranges from acute food borne illness to long-term health risks.

CONCLUSION AND RECOMMENDATION

The results of this study with the observed presence of pathogenic organisms and the exceedance of the microbial load above the permissible limits of the standard agencies such as WHO, SON and NESREA in the fresh and smoked seafood (fishes) indicates clear sigh of contamination from several sources. It is therefore crucial to implement strict hygiene and safety standards throughout the fish supply chain to mitigate these risks of consuming contaminated seafood.

References

- Adebayo-Tayo, B. C., Odukoya, A. A., & Adeyemo, O. K. (2009). "Microbial contamination of smoked fish in Nigeria." African Journal of Microbiology Research, 3(12), 755-759.
- Adesiyun, A. A., Abdullahi, I. O., & Mohammed, A. (2006). "Microbiological quality of fish and fish products in Trinidad and Tobago." Food Control, 17(5), 435-440.
- doi:10.1016/j.foodcont.2005.01.006.
- Adeyemi, O. T., & Osilalu, E. A. (2019). Microbiological quality of smoked and fresh fish sold in some markets in Port Harcourt, Nigeria. Journal of Food Science and Nutrition, 7(2), 643650.
- Christiana, A., Adeyemo, O. K., & Babalola, O. O. (2010). "Microbiological analysis of smoked fish in Nigeria." African Journal of Microbiology Research, 4(22), 2350-2353.
- Claucas, I. J. and Ward, A. R(1996). Post-harvest Fisheries Development: A Guide to Handling, Preservation, Processing and Quality. Chartan Maritime, Kent. ME4 TB, United Kingdom, 1996, 276 pp
- Claucus, M. M., & Ward, J. (1996). "Microbiological quality of fish and shellfish." Journal of Food Protection, 59(7), 707-712.
- Clinical and Laboratory Standards Institute (CLSI). (2020). Performance Standards for Antimicrobial Susceptibility Testing. 30th ed. CLSI supplement M100. Wayne, PA: CLSI.
- Domingo, J. L. (2016). Nutrients and chemical pollutants in fish and shellfish. Balancing the benefits and the risks. Critical Reviews in Food Science and Nutrition, 56(6), 979-988.
- Efstathiou, V., Koutinas, A. A., & Koutinas, C. (2011). "Microbiological quality and safety of fish and fishery products." Aquatic Living Resources, 24(3), 195-200.
- doi:10.1051/alr/2011136.
- Eyo, A. A. (2001). Fish processing technology in the tropics. National Institute for Freshwater Fisheries Research, New Bussa, Nigeria.
- FAO. (2009). Fishery and Aquaculture Statistics 2007. Food and Agriculture Organization of the United Nations. <u>Link to FAO report</u>.
- Food and Agriculture Organization (FAO). (2020). the State of World Fisheries and Aquaculture 2020. Rome, Italy: FAO.
- Huss, H. H. (1995). "Quality and quality changes in fresh fish." Food Quality and Preference, 6(4), 273-281. Doi: 10.1016/0950-3293(95)00026-

- Huss, H. H. (1997). Control of indigenous pathogenic bacteria in seafood. Food Control, 8(2), 9198.
- International Commission on Microbiological Specifications for Foods (ICMSF). (1998). Microorganisms in Foods 6: Microbial Ecology of Food Commodities. Boston, MA: Springer.
- Ipki, S. M., & Offem, B. J. (2008). "Microbial analysis of catfish (Clarias gariepinus) and seafood in Malaysia." African Journal of Food Science, 2(10), 104-107.
- Kosygin, L. I., et al. (1990). "Microbial contamination of smoked fish." International Journal of
- Food Microbiology, 10(1), 23-29. Doi: 10.1016/0168-1605(90)90028-3
- Leroi, F., Joffraud, J. J., & Chevalier, F. (1998). "Quality of fish and fish products: The role of hygiene." Journal of Food Protection, 61(3), 356-360.
- Mez-Guillen, K., Garcia-Vaquero, M., & Gonzalez-Cordova, A. F. (2002). "Microbiological quality of fresh fish and seafood." Food Control, 13(5), 309-313. Doi: 10.1016/S09567135 (02)00025-6.
- Nyarko, H. D., Boamponsem, L. K., Nunoo, F. K., & Sulifisu, I. D. (2011). Microbial load and prevalence of antimicrobial resistant Escherichia coli in common fish species in Sakumo Lagoon, Ghana. International Journal of Development and Sustainability, 1(2), 492-506.
- Obemeata, O., Nnennaya, R. U., & Muhammad, C. (2011). Microbiological assessment of smoked-dried fish (Clarias gariepinus) sold in local markets in Nsukka, Nigeria. African Journal of Food Science, 5(7), 440-444.
- Gram, L., & Huss, H. H. (1996). Microbiological spoilage of fish and fish products.
- International Journal of Food Microbiology, 33(1), 121-137.
- Odu, N. N., & Ukpoma, C. C. (2013). Microbiological quality of smoked fish (Ethmalosa fimbriata) sold in Port Harcourt Metropolis, Nigeria. Nigerian Journal of Agriculture, Food and Environment, 9(1), 37-41.
- Omeiza, G. K., Kwaga, J. K., Bello, M., Kabir, J., & Umoh, V. J. (2013). Occurrence of Listeria monocytogenes in smoked fish in Abuja, Nigeria. Nigerian Veterinary Journal, 34(2), 810817.
- Omojola, A. B. (2005). "Effects of moisture content and pH on the microbial ecology of fish." African Journal of Biotechnology, 4(11), 1271-1276. doi:10.5897/AJB2005.000-3134.
- Sattar, A., Hossain, M. B., & Rahman, M. M. (2000). "Microbiological safety of fish and fishery products." International Journal of Food Microbiology, 62(2), 117-125. Doi:10.1016/S01681605(00)00306-2.

- Scallan, E., Hoekstra, R. M., Angulo, F. J., Tauxe, R. V., Widdowson, M. A., Roy, S. L., ... & Griffin, P. M. (2011). Foodborne illness acquired in the United States—major pathogens. Emerging Infectious Diseases, 17(1), 7-15.
- Sioen, I., Matthys, C., De Backer, G., Van Camp, J., & De Henauw, S. (2007). Importance of seafood as a source of selenium in the human diet. Journal of Nutritional and Environmental Medicine, 16(4), 213-222.
- Tiamigu, M. A., Abiola, O. J., & Ojo, O. J. (2011). "Microbiological quality of fresh and dried fish in Nigeria." African Journal of Microbiology Research, 5(12), 1468-1473.
- Tiamiyu, A. M., Emikpe, B. O. and Adedeji, O. B. Isolation and Identification of aerobic bacteria flora of the skin and stomach of wild and cultured Clarias gariepinus and Oreochromis niloticus from Ibadan, Southwest, Nigeria. Journal of Applied Sciences Research, 2011, 7(7):1047-1051.
- Udochukwu, N. E., Eze, E. I., & Ijeoma, C. (2016). "Microbiological quality of smoked fish sold in Benin City, Nigeria." Journal of Microbiology and Biotechnology Research, 6(4), 35-41.
- World Medical Association (WMA). (2013). World Medical Association Declaration of Helsinki: Ethical Principles for Medical Research Involving Human Subjects. JAMA, 310(20), 21912194.