Occurrence of Fungi Species in Some Fisheries Products from Retail Markets in Lagos, Nigeria

Olaitan Olajuyigbe¹, Esther Amusan¹ and Kafayat Yusuf²

Abstract

Fish is a major source of protein that is easily prone to contamination by micro-organisms because of its composition. In this study, the fungal floral of fresh and smoke-dried fish samples from five major markets in Lagos, Nigeria were investigated. The fungi were isolated by plating out serially-diluted samples on Potato Dextrose Agar at room temperature for three to five days. A total of sixty-four fungal isolates were obtained and identified to belong to the genera, *Aspergillus*, *Penicillium*, *Trichoderma*, *Fusarium*, *Rhizopus*, *Mucor* and *Neurospora*. However, yeasts and *Aspergillus* sp. were the most predominant organisms. This research showed that most of the fish samples from the selected markets are contaminated with fungi.

Key words: Fish, Fungal Floral, Mycosis, Mycotoxins, Market.

1. Introduction

Fish is an important source of human dietary protein worldwide, especially in Africa (Kumolu and Ndimele, 2011), fish and fisheries products constitute about 49% of the total animal protein intake in Nigeria (Ajani, 2008). Adebayo-Tayo *et al.* (2008), mentioned that fish is better digested in human digestive system than beef. However, fresh fish samples are generally accompanied by microbes which are generally part of the normal floral of the fish as a result of the environment from which the fish is collected as well as the handling procedures. Invariably, pathogenic microorganisms could be present as a result of the pollution of water bodies or sometimes as a result of the treatments given to the fish before processing. Eventually, most or all of these pathogens are destroyed during processing. Most fish species possess very good preservation qualities after being salted, sun-dried or smoke-dried (Madu *et al.*, 1984). Unfortunately, these processed products are sometimes still susceptible to fungi infestation. Fungi are diverse in nature and cause of an infection known as "mycosis" (Abbey, 1995). Their presence in fish could result in food-borne infection, food poisoning or mycotoxicosis (resulting from the ingestion of the mycotoxins produced by toxigenic fungi).

¹ Department of Fish Technology and Product Development, Nigerian Institute for Oceanography and Marine Research, P.M.B. 12729, V.I., Lagos, Nigeria.

² Department of Microbiology, University of Lagos, Akoka, Lagos, Nigeria

^{*} Corresponding authors, e-mail: olaitan_afolabi@yahoo.com

Post-processing contamination of fish by fungi could be due to inadequate processing or improper handling and storage of the fish. Painfully, the presence of fungi in fish may constitute a serious public health hazard and may also lead to economic losses. This risk is exacerbated by the fact that smoke-dried fish is sometimes eaten without any further processing.

This research evaluated the different types of fungi species associated with fishes sold in the five major fish markets located in Lagos, Nigeria. The findings were used to create awareness on potential public health hazards associated with fish and the need for adequate regulations of microorganisms in fishery products in Nigeria.

2. Materials and Methods

2.1 Collection of Samples

Three commonly available fin and shell fish (*Pseudotolithus typhus* (croaker), *Micromesistius poutasou* (blue whiting) and *Peanus notialis* (pink shrimp) were randomly sourced from five major fish markets (Makoko, Liverpool, Mushin, Badagry and Iyanapaja) in Lagos. A total of eighty-one samples were purchased, they included thirty-five samples of fresh and forty-six smoke-dried samples. The fresh samples were kept in ice boxes (the ratio of ice to fish was 1: 1) while the smoke-dried samples were collected in sterile polythene bags and taken to the laboratory immediately for analysis within four hours.

2.2 Isolation of Fungi

The fungi species associated with the samples were isolated using standard method (APHA, 1992). Ten grams of samples were homogenized in 90ml sterile distilled water and serially diluted. One milliliter of selected dilution (10⁻¹, 10⁻³, and 10⁻⁵) was plated out in duplicates. Sterile Potato Dextrose Agar (PDA), into which 0.01g/l of ampicillin had been added (to suppress bacteria growth) was poured into the plate and thoroughly mixed with the diluted samples. The plates were allowed to solidify and incubated at 30±2°C for 3–5 days.

2.3 Frequency of Occurrence of Fungal Isolates

The frequency of occurrence (FO) of fungal isolates is the relative number of times a particular isolates is obtained. It was calculated using the equation below:

$$FO = \frac{Total\ number\ of\ Isolation\ of\ an\ isolate}{Total\ number\ of\ isolates}\ X\ 100$$

2.4 Identification of Isolates

Discrete colonies were sub-cultured onto fresh PDA plates and the isolates were identified based on their cultural/ macroscopic characteristics (colour, colony morphology, pigmentation, texture) and microscopic characteristics. The microscopic characteristics were observed by preparing wet mount of the fungi in cotton blue in lacto-phenol, covered with a cover slip and observed under x4, x10 and x40 objective lens. The structure of the sporangia, conidial and mycelium were observed following the method of Barnet and Barry (1972).

3. Results and Discussion

The total fungal count ranged from 0 to 4.0 x 10⁵. A total of sixty-four fungal isolates were obtained. Twenty nine isolates were obtained from fresh samples, while thirty-five isolates were obtained from smoke-dried fish samples. They were identified to belong to genera such as Aspergillus, Penicillium, Fusarium, Rhizopus, Mucor, Neurospora, and Trichoderma. Yeasts were also isolated. The fungi species associated from the selected fishery products from some markets in Lagos state are similar to those obtained from the western part of Nigeria as reported by Fafioye et al., (2008). Essien et al. (2005) and Iyayi (2011) also isolated Aspergillus, Fusarium and Penicillium, Rhizopus and Trichoderma from fish products sold in Nigerian markets. Adelaja et al., 2013, isolated Fusarium, Mucor and Aspergillus species from smoked fish from Oyan Lake and Ogun waterside of Ogun state, Nigeria. The occurrence of fungi isolates from the different markets is reported in Table 1. Aspergillus niger and yeasts were isolated from all the markets while Neurospora sp. and a yet to be identified fungus were only isolated at Makoko and Badagry markets respectively. Table 2 shows the frequency of occurrence of the fungi isolates. Yeast and Aspergillus sp had the highest frequency of occurrence (32.81% and 25.01% respectively), while the least was Neurospora sp (1.56%). The presence of these fungi on the fish samples can be attributed to poor sanitary conditions in the market environments coupled with unhygienic handling and processing techniques. All the markets were littered with refuse and some of them even have refuse dumps being burnt right inside the markets. Emmanuel and Hontoyon (2016), stated that the fungi contaminants could have been deposited on the samples during handling by consumers who tend to select attractive sizes of fish during sales. Adebayo-Tayo et al., (2008), noted that some market environments are usually unhygienic, and may also be a source of contamination. The markets environment encouraged the proliferation of flies and other pests which could be carriers of microorganisms. Proper and adequate storage conditions of different products should be encouraged. Stores should be well ventilated in order to prevent the build-up of moisture in products and measures should be taken to prevent the exposure of products to pests. It is pertinent to state that the fungi isolated are potential pathogens. The occurrence of fungi in most of the samples is also an indication of the likely presence of mycotoxins. Mycotoxins are secondary metabolites of some fungi. This may pose a serious public health hazard as mycotoxins have been reported to be carcinogenic, teratogenic, mutagenic and immunosuppressive (IARC, 1993). AFB1 (one of the aflatoxins produced by toxigenic Aspergillus flavus, has been reported to be a major cause of hepatocellular carcinoma (cancer of the liver). Aflatoxin (a group of mycotoxins) has also been reported to have been implicated in cases of acute hepatitis in man (Eaton and Groopman, 1994). There is also a need for regulatory bodies to set and enforce standards for microbiological regulations of fish and fisheryproducts.

Table 1 Occurrence of Fungal Isolates in Five Different Markets

Isolate	Makoko	Liverpool	Mushin	Badagry	lyanapaja
Aspergillus niger	+	+	+	+	+
Aspergillus flavus	-	-	+	+	+
Penicillium sp	+	+	+	+	-
Trichoderma sp	+	+	+	+	-
Fusarium sp	+	-	+	-	+
Rhizopus sp	-	+	+	-	-
<i>Mucor</i> sp	+	-	-	-	+
Neurospora sp	+	-	-	-	-
Unidentified fungus	+	-	-	+	-
Yeast	+	+	+	+	+

Note: + Positive - Negative

Table 2 Frequency of Occurrence of Fungal Isolates

Isolates	No. from Fresh	No. from Smoked	Total No.	Frequency of
	Samples	Samples		Occurrence (%)
Aspergillus niger	3	7	10	15.62
Aspergillus flavus	1	5	6	9.38
Penicillium sp	3	3	6	9.38
Trichoderma sp	2	4	6	9.38
Fusarium sp	6	1	7	10.94
Rhizopus sp	1	2	3	4.69
Mucor sp	1	1	2	3.13
Neurospora sp	1	0	1	1.56
Unidentified fungus	1	1	2	3.13
Yeast	10	11	21	32.81
Total	29	35	64	100

4. Conclusion

This study revealed that fresh and smoke-dried fish samples from Lagos markets are contaminated with fungi. These smoke-dried fishery products are generally regarded as "ready to eat". However, it is advisable that more efforts should be made in enlightening the fish sellers and consumers on the need for adequate cooking of smoked-dried fish before consumption to reduce the risk of food-borne infections. Also, proper orientation should be given to the fish sellers on ways of ensuring precautionary microbiological measures and taking proper hygienic steps during processing, sale and storage of fishery products. They should be educated on the proper temperature and time of storage of their products to reduce the ease of proliferation of microorganisms. Standard packaging materials should also be used for packaging of fishery products.

References

Abbey, S.D. 1995. Foundation in Medical Mycology. Bidsol and Company. Lagos. Pp. 35

Adebayo-Tayo, B.C., Onilude, A.A. and Patrick, U.C. 2008. Mycofloral of Smoke-Dried Fishes

Sold in Uyo, Eastern Nigeria. World Journal of Agricultural Sciences 4(3): 346–350.

Adelaja, O.A., Olaoye, O.J., Ikenweiwe, N.B. and Ashley-Dejo, S.S. 2013. Comparison of microbial load associated with smoked fish (*Chrysichthys nigrodigitatus*) from Oyan

- Lake and Ogun waterside in Ogun State, Nigeria. Global Journal of Science Frontier Research Agriculture and Veterinary 13(8): 200–213.
- APHA, 1992. Compendium of methods for the microbiological examination of foods, (Vanderzant, C. and Splittstoesser, D. Eds), APHA Washington DC. Pp. 3
- Barnet, H.L. and Barry, B.H. 1972. Illustrated genera of imperfect fungi. 3rd Edition. Burgess Publication Co., Minneapolis. Pp. 63–70.
- Eaton, D. L. and Groopman, J. D. 1994. The toxicology of Aflatoxins. Academic Press New York. Pp. 117.
- Emmanuel, B.E. and Hontoyon, E.W. 2016. Microbiological Quality of Cold Smoked Sardine, Sardinella maderensis (Lowe, 1839) in four markets in Lagos Metropolis, Lagos State, Nigeria. Journal of Aquatic Science 31(1): 167–184.
- Essien, J.P., Ekpo, M., Brooks, A.A. 2005. Mycotoxigenic and Proteolytic Potential of Mould, Associated with smoked shark fish (*Chlamydoselachus anguincus*). Journal of Applied Science and Environmental Management. 9(3): 55–57.
- Fafioye, O.O., Efuntoye, M.O. and Osho, A. 2002. Studies on the fungal infestation of five traditionally smoke-dried fresh water fish in Ago-Iwoye, Nigeria. Mycopathologia, 154: 177–179.
- Fafioye, O.O., Fagbohun, and Olubanjo O.O. 2008. Fungal Infestation and Nutrient Quality of Traditionally Smoke-Dried Freshwater Fish. Turkish Journal of Fisheries and Aquatic Sciences. 8: 7–13.
- IARC, 1993. Monographs on the evaluation of carcinogenic risks to humans: Some naturally occurring substances, food items and constituents, heterocyclic aromatic amines andmycotoxins. International Agency Res. Cancer. Geneva. 56: 489–521.
- Madu, C.T., Okoye, F.E., Sado, E.K., Omorinkoba, W.S., Bankole, W.O. and Ita, E.O. 1984. A preliminary report of induced breeding trials with the mud fish (*Clarias anguillaris*) KLRI Animal Report. Pp. 144–159.
- Wogu, M.D. and Iyayi, A. D. 2011. Mycoflora of Some Smoked Fish Varieties in Benin City Nigeria. Ethiopian Journal of Environmental Studies and Management. 4(1): 36–38.