



Fungi Associated with the Spoilage of Smoke Dried Fish in Open Market in Osogbo, Osun-State, Nigeria

**Francis Olusegun Oladeji ^{a*}, Olanrewaju John Adebayo ^b,
Abolayo Tawakalitu Tope ^c, Makanjuola Bosede Christiana ^d
and Olawuni Timothy Ifeoluwa ^e**

^a Department of Environmental Health Science, Fountain University, Osogbo, Osun-State, Nigeria.

^b Department of Environmental Health Science, Faculty of Basic Medical and Health Science, Lead City University, Ibadan, Nigeria.

^c Department of Environmental Health, Nigeria Navy School of Health Sciences, Offa, Kwara State, Nigeria.

^d Ekiti State College of Health Science and Technology, Ijero-Ekiti, Nigeria.

^e Interlink Polytechnic, Ijebu-Jesa, Osun-State, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Man primarily consumes dried fish as a source of nutrition. It has been proven that eating fish can act as a vector for the spread of some "mycological pathogens," especially in people with weakened immune systems. This investigation was conducted to provide specific information on the existence of various fungal species linked to the contamination of smoke-dried fish sold in Osogbo, Osun State's open markets. This investigation was conducted to provide specific information on the existence of various fungal species linked to the contamination of smoke-dried fish sold in Osogbo,

*Corresponding author: E-mail: franciscohealth3@gmail.com;

Osun State's open markets. Using potato dextrose agar (PDA) and sabouraud dextrose agar (SDA) and microscopy, the study for microbial contamination was conducted. In pure culture, *Mucor spp.*, *Aspergillus spp.*, *Rhizopus spp.*, and *Fusarium spp.* were isolated and identified as the organisms. Following the incubation time, the total fungal counts of samples of smoked fish throughout a 7-day storage period are displayed in Tables 1 and 2. On the first two days of the investigation for the used samples, there was no fungus count. Some samples had a low fungus count on the third day of the research. However, from the fourth day to the seventh day, all of the samples demonstrated positive fungi growth. As the length of storage grew, the fungal burden (count) rose as well. Thus, for all samples that were analyzed, the seventh day revealed the greatest fungal count. *Mucor spp.*, *Fusarium oxysporum*, *Aspergillus spp.*, *Rhizopus spp.*, and *Penicillium spp.* are the isolated organisms. This study demonstrates that the fungal infestation in examined fish is extremely high. This might be the main reason for spoiling and astringency, and it might also increase the risk to the public's health and cause financial losses. This suggests the need for veterinary and public health collaboration through a fish regulatory program. Health education should be given to fish processors on a safe method of preservation in order to prevent or minimize fungal contamination.

Keywords: Contamination; preservation; smoke-drying; fish; *Aspergillus*; spoilage; infestation.

1. INTRODUCTION

"In Nigeria, eating smoked and smoke-dried fish without additional or sufficient cooking is rather prevalent. Smoke-dried fish are frequently infected with microorganisms like bacteria, yeasts and molds both during processing and when displayed openly in market places" [1]. It was claimed that the majority of sellers of smoked and smoke-dried fishes disregarded safe preservation procedures. Among them include inadequate ventilation, sloppy preparation hygiene, exposure to a fly infestation, and pests' simple access to the storage environment [2].

Fish is an excellent source of protein-rich nutrients. However, if contaminated and improperly processed, processed fish that has been smoked or smoke-dried may be susceptible to the proliferation of microorganisms [3]. The main cause of concern for the deterioration of fish products is the growth of pathogenic organisms like molds and yeasts as well as other non-microbial activities such lipid oxidation (Martin, 2010). Food preservation procedures often involve adding substances that block microbial development or modifying storage conditions by freezing or drying, as well as preserving fish and meat that are biodegradable or putrescible [4]. Fish smoking consists of two steps. Wet hot smoking and dry hot smoking are two of them. Temperatures that are high enough to cook the fish for consumption are needed for these methods.

Wet hot smoking fish takes roughly one to two hours and produces a versatile, moist product with a moisture percentage of 40–50%. Fish with

a moisture content of 10-15% can be produced via dry hot smoking, which typically follows the earlier process and lasts for longer periods of time (up to days).

Akande et al. [2] noted that "smoke drying of fish is as old as a man and is one of the long-use local forms of preservation methods that is mostly used by most fishing communities and wholesale fish dealers in a significant locality in several underdeveloped nations like Nigeria. Many foodborne mould and possibly yeasts may also be hazardous to human health because of their ability to produce toxic metabolites known as mycotoxins, which are stable compounds that are not destroyed during food processing. Smoked and Smoke-drying, have been known to give fish products desirable taste and odors, preserve and prolong the shelf-life of the products conveniently at ambient conditions".

"Even though the organism may not survive during food preparation, the preformed toxin may still be present. Certain moulds and yeasts may also cause allergic reactions or may cause disease in Humans. Research has shown that Smoke-drying practices pose serious health implications especially when these microorganisms and /or toxins find their way into the digestive system. It was also recorded that consumption of smoked-dried fish has been reported to cause outbreak of several cases of human gastroenteritis, severe diarrhea and food poisoning" [5]. The creature might not live while the food is being prepared, but the toxin might still be there. Some yeasts and molds can also make people allergic to them or make them sick. According to research, smoke-drying procedures

have substantial negative health effects, especially when germs and/or pollutants enter the digestive system. Additionally, it was noted that eating smoked-dried fish has been linked to an increase in occurrences of food poisoning, severe diarrhea, and gastroenteritis in humans [5]. According to Swaminarayan and Sparling's 2007 paper, a number of bacteria isolated from various fish species have toxins, and their toxin continued to accumulate in fish flesh even after salting and storage periods, leading to major systemic dysfunctions and risks to the general public's health.

2. MATERIALS AND METHODS

2.1 Study Area

The research was conducted in Osogbo, the Nigerian state of Osun's capital. The state was split from Oyo state in 1991, and Osogbo is the capital city, housing the offices of both the Osogbo and Olorunda local governments (located in the city's Oke Baale and Igbonna, respectively). The capital city of the state is located in latitude 4.5418° E and longitude 7.7827° N. Civil servants, paramilitaries, soldiers, traders, farmers, and artisans make up the majority of the population in Osogbo. The community has a wealth of natural resources as well.

2.2 Sample Collection

From September to October 2021, samples of smoke dried fish were randomly chosen and purchased from two main fish markets in Osogbo, Osun State, Nigeria (Oja Oba and Igbonna Markets). The samples collected for the study were transported to the lab for processing and analysis to check for the presence of fungi in well-prepared sterile polythene bags.

2.3 Isolation and Identification of Fungi Agents

500ml of distilled water was used to dissolve 32.5 grams of Sabouraud dextrose agar. To ensure a homogenous mixture, it was heated and stirred. Aluminum foil was placed over the conical flask's mouth after it had been corked with non-absorbent cotton wool. It was correctly labeled and autoclaved at 121 °C for 15 minutes to sterilize it.

The collected samples were serially diluted, and 0.5ml of the diluents was added to the sterile

Petri plates. PDA and SDA that had already been prepared and sanitized were added to the plates. Mycoflora was isolated using Warcup's serial dilution and culture plate exposure method. For purification, the isolated organisms were subcultured, and they were then recognized using conventional microbiological and biochemical analytical techniques (Warcup, 2000).

2.4 Data Analysis

Utilizing percentage and rates, the frequencies and market distribution of fungal isolates were identified. Tables and figures were used to present the results.

3. RESULTS AND DISCUSSION

Following the incubation time, Tables 1 and 2 display the total fungal counts of smoked fish samples over a 7-day storage period. On the first two days of the investigation, there was no fungal count for the used samples. Some samples had a low fungus count on the third day of the experiment. However, from the fourth day to the seventh day, all of the samples demonstrated positive fungi growth. As the length of storage increased, the fungal burden (count) grew. Thus, for all samples that were analyzed, the seventh day revealed the greatest fungal count. *Mucor spp.*, *Fusarium oxysporum*, *Aspergillus spp.*, *Rhizopus spp.*, and *Penicillium spp.* are the isolated organisms.

Table 1 displays the identification of the isolated people using colony morphology and cultural traits. The outcome of this experiment suggests that the fish's habitat is an independent variable. Room temperature, humidity, storage conditions, and fish handling are the constants (control variables).

Table 2 shows the total colonies of fungal species observed in the media used, potato and sabouraud dextrose agar from smoked fishes. The trend in terms of positivity rate also similar to what was observed in this Table 2 which shows that *Rhizopus spp.* having the highest occurrence of '32' which is (26%) followed by *fusarium spp.* 26 (21.14%). *Penicillium spp.* were the least occurring of '16' (13.01%). The fungi isolated in this study are comparable to the observation of Abolude et al. [6], in his study of fresh water, fungi and smoked fish in eggs and brood stock of *C. gariepinus* zaria.

Table 1. Cultural and morphological characteristics of identified fungi

Fungi Isolates	Cultural characterization	Morphological characterization	Sample location
<i>Aspergillus spp.</i>	Yellow or yellowish-green colonies distinct margin	Conidiophores arise from a foot cell. Club shaped vesicles	A and B
<i>Penicillium spp.</i>	Fast growing colonies in green colour with dense conidia	Branched Conidiophores with chains of conidia looks like a brush	D and B
<i>Rhizopus spp.</i>	white cottony mycelia, with black dots and covers the entire plate		D
<i>Mucor spp.</i>	Large white colonies which turns into black later	Erect sporangiophores are formed. Sporangiophore swells at the tip to form <i>sporangia</i> which are globular shaped. <i>Columella</i> is present	
<i>Fusarium oxysporum</i>	White to cream, flat woolly to cottony and spreading	Presence of large <i>conidia</i> and erect <i>sporangiospores</i>	D and B

Table 2. Total colonies of fungal species observed in Potato dextrose agar media and Sabouraud dextrose agar media from smoked fishes

S/N	Isolated Species	No of Colonies In DPA	No of Colonies in SDA	Total	% Contribution
1	<i>Aspergillus spp.</i>	10	14	24	19.51
2	<i>Penicillium spp.</i>	07	09	16	13.01
3	<i>Rhizopus spp.</i>	09	23	32	26.02
4	<i>Mucor spp.</i>	08	17	25	20.32
5	<i>Fusarium spp.</i>	08	18	26	21.14
Total colonies of all the fungal species				123	

This finding is similar with the study of Alawodi and Bichi[7] in which they reported that dam's environment and the equipment used in fish smoking were more contaminated than fish farms. This might have been as a result of smoked fish handler activities.

It is important to note that the majority of the fungal agents isolated were of medical significance. The presence of *Aspergillus spp.* and *Penicillium spp.*, could lead to mycotoxin elaboration and when consumed, they could cause mycotoxin poisoning.

However, Refia et al. (2004) revealed that *Penicillium spp.*, *Aspergillus spp.*, and *Rhizopus spp.* are normal mycoflora present in most fish. The fungi recovered from this investigation were similar to findings by other authors.

According to Akande and Tobor, who were cited by Sani et al., the environment where fish are displayed in the market is typically unhygienic and could constitute another avenue for microbial contamination. As a result, the contamination of smoke-dried fish by numerous different species of fungi as observed in this study and many other earlier studies is not surprising. Akande, Tobor, Sani, and others correctly observed that almost often, retailers show smoke-dried fish samples in open trays close to garbage dumps, which encourage fungal growth by air droplets.

The fungi that were found over the course of this study endeavor are opportunistic infections that

are significant in both human and veterinary medicine. When foods like smoke-dried fish that have been exposed to contamination are consumed, the presence of toxigenic fungi, such as some species of *Aspergillus*, *Penicillium*, and *Candida*, increases the risk of mycotoxins being produced, which may cause gastrointestinal and metabolic disturbances.

Only strains of *Aspergillus flavus* exhibited aflatoxigenic generating features among the molds found by Job et al. While Bukola et al. discovered aflatoxins B1 and G1 in all the *Aspergillus* species isolated in their investigation in Uyo, Osibona et al. discovered aflatoxin and ochratoxin A in stored fish samples as a result of the *Aspergillus* and *Penicillium* species. Sani et al. did confirm that many fungal taxa have virulence factors that lead to the production of toxins in favorable predisposing environments.

4. CONCLUSION

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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