

Mycoflora of Some Smoked Fish Varieties in Benin City Nigeria

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Abstract

A study of the mycoflora of six locally available and commonly consumed dried fish species namely; *Ethmalosa fimbriata* (bonga fish), *Tilapia sp.* (Banda mangala) *Gadus morhua* (stock fish), *Pseudotolithus typhus* (croaker), *Arius hendeloti* (cat fish) and *Drepane africana* (spade fish) was carried out in three sampling regimes. A total of thirty-six samples were randomly sourced from local markets in Benin City and cultured in two replicates per sample per batch in Saboraud dextrose agar (SDA). Six fungal isolates encountered in the study were *Aspergillus niger*, *A. flavus*, *Penicillium sp.*, *Fusarium sp.*, *Rhizopus sp.* and *Trichoderma sp.* in their order of decreasing frequency in all the fish samples. The highest mean mycoflora count (17.833×10^3 cfu) was recorded in *Tilapia sp.*, while the lowest mean value (11.16×10^3 cfu) was recorded in *Drepane africana*. *Aspergillus* species are known to produce aflatoxins which are carcinogenic (causing heptoma – cancer of the liver), acute hepatitis, reduced red blood cell and decreased immune system in man. *Fusarium sp.* is reported to produce fumonisin toxin and *Penicillium penicillic acid*. Prolonged intake of smoked fish with these metabolites may constitute potential public health hazard. Adequate cooking could help in reducing mycoflora of smoked fish.

Introduction

Preserving food and other perishable products like fish and meat generally involves processes that impede growth of micro organisms either by addition of growth inhibiting ingredients or adjusting storage conditions by freezing or drying. In preserving fish by smoking water activity in the fish is lowered to the point where the activity of spoilage micro organisms is inhibited (Okonta *et al.*, 2005). Smoking is the preferred method of fish preservation in most rural areas and riverine fishing communities. Akinola *et al.*, (2006) described several methods of fish preservation and reported the preponderance of fish preservation by smoking in most fishing communities in Nigeria owing to non-availability of electricity.

Salting, brining, addition of vinegar and the type of wood used for the smoking fire all contribute to the quality of smoked fish and salting combined with smoking reduced incipient spoilage resulting in extension of shelf life from two to six months (Tabor, 1984; Eyabi-Eyabi, 2000 and Omojowo, 2008). Traditional fish preservation by using hard wood in preference over soft wood for smoking yield more bacteriologically stable products (Davies, 2006) and the process also enhances texture and flavor of fish (Akande and Tobor, 1992).

Smoked fish is relished food item in many dishes in Nigeria and particularly in Benin City. It is therefore important to monitor the microbiological quality of smoked fish so as to prevent any health hazard that may arise from consumption of unwholesome smoked fish.

This study was undertaken to investigate possible contaminants present in smoked fish and by so doing, identify fungal species prevalent in smoked fish, their distribution, effects and possible public health implication of the presence of such mycoflora.

Materials and Methods

Six commonly available dried fish samples (*Ethmalosa fimbriata*, *Tilapia sp.*, *Gadus morhua*, *Pseudotolithus typhus*, *Arius hendeloti* and *Drepane africana*) were randomly sourced from local markets in Benin City. The samples were labeled A, B, C, D, E and F respectively and were carefully packaged in cellophane bags and taken to the laboratory for analysis. 10g of each fish sample were macerated and dissolved in 90ml of distilled water to obtain a stock solution. Serial dilutions were carried out for each of the samples and five replicates of each sample were prepared. Sterilized Saboraud dextrose agar (SDA) was poured into petri dishes containing antibiotic mixture and 1 ml of serial diluents of the samples under aseptic conditions. The plates were incubated for five days. The microbial isolates were observed for their cultural and morphological characteristics.

Results

The result of the total microbial colony counts expressed in cfu $\times 10^3$ obtained from two replicate cultures of three separated batches of sampling of six smoked fish species obtained from local markets in Benin City are shown in table 1.

Sample A - *Ethmalosa fimbriata* (locally known as bonga fish) had microbial count ranging from 10×10^3 - 18×10^3 cfu; sample B -

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Tilapia sp. (locally known as banda mangala) had appreciably higher microbial load 14×10^3 – 20×10^3 cfu and for sample C - *Gadus morhua* (locally known as stock fish), the microbial count ranged from 10×10^3 - 19×10^3 . The total colony forming units recorded in the replicate samples of samples D (*Pseudotolithus typhus*, locally known as croaker fish) and sample E (*Arius hendeloti*, locally called cat fish) were 8×10^3 – 19×10^3 cfu and 12×10^3 – 15×10^3 cfu respectively; while sample F - *Drepane africana* (locally called spade fish) microbial count ranged from 11×10^3 – 14×10^3 cfu. One-way analysis of variance of the samples showed significant difference in samples A and D ($P < 0.05$). Sample F recorded the lowest mean microbial count value of 11.167×10^3 cfu and the highest mean microbial count of 17.833×10^3 cfu was recorded for sample B.

Table 2 shows the frequency distribution of the six fungal isolates from three sampling regimes of the six smoked fish samples. The results are presented in order of descending frequencies of the fungal isolates. *Aspergillus niger* had the highest overall frequency while *Trichoderma* sp. was the least. Frequencies of the fungal isolates were generally highest during the first sampling and lowest during the second sampling.

Discussion

The six fungal isolates encountered in this study were *Aspergillus niger*, *A. flavus*, *Penicillium* sp, *Fusarium* sp. *Rhizopus* sp. and *Trichoderma* sp. and they are the most common microbes associated with smoked fish. The most frequently recorded isolate in the fish samples was *Aspergillus niger* and this is an indication of its ubiquitous nature. The Several species of yeasts and *Aspergillus* have been isolated from salted and dried meat and fish

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products (Graikoski 1973) and these species are known to produce toxic substances. Bukola *et al.*, (2008) detected aflatoxins B1 and G1 concentrations ranging from 1.50 – 8.10 μ g/kg and 1.81 – 4.5 μ g/kg respectively. This finding is instructive as consumption of contaminated smoked fish could pose serious health problems. Aflatoxins have been implicated in cases of acute hepatitis in man and they are also known to be carcinogenic causing hepatoma (Eaton *et al.*, 1994).

The highest mean mycoflora count value 17.833×10^3 cfu was recorded in sample B - *Tilapia* sp., while the lowest mean value 11.16×10^3 cfu was recorded in sample F - *Drepane africana* with no significant difference in their mycoflora variability ($P > 0.05$). This is an indication of the differences in both processing and handling of both smoked fish samples. The preponderance of these molds with their weak proteolytic potential and ability to elaborate proteases in smoked fish is an indication of their ability to cause spoilage. This result and effect was corroborated by Eyo (1992) and Hussain *et al.*, (1993) with additional species of the genera *Penicillium* like *P. italicum*, *P. viridatus*; *Candida tropicalis* and *Absidia* sp.

Improper smoking and drying of fishes may lead to insect infestation, fungal attack, fragmentation and degradation of the product (Banwart, 1989 and Eyo, 1992). Since most of the molds isolated were possible contaminants rather than originating from the fish samples, better preservation and handling (drying and storage) would reduce mycoflora proliferations. It is therefore important that both artisanal fishermen and marketers should adopt better preservation methods and smoking kilns should not be over crowded during fish drying.

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Table 1 Microbial colony count of smoked fish samples

sample	colony count x10 ³ cfu						P-value	
	1 st Sampling		2 nd Sampling		3 rd Sampling			Mean
A	18	11	13	10	18	17	14.5	P< 0.05
B	20	18	19	14	16	20	17.833	P> 0.05
C	11	10	18	19	19	15	15.333	P> 0.05
D	19	16	10	8	16	17	14.333	P< 0.05
E	12	14	12	15	15	13	13.5	P> 0.05
F	10	11	11	10	11	14	11.167	P> 0.05

Table 2 Frequency of occurrence of fungal isolates per sample

Fungal isolates		% frequency		
		1 st Sampling	2 nd Sampling	3 rd Sampling
1	<i>Aspergillus niger</i>	97	88	92
2	<i>A. flavus</i>	89	86	89
3	<i>Penicillium</i> sp.	81	83	83
4	<i>Fusarium</i> sp.	75	75	77
5	<i>Rhizopus</i> sp.	69	67	69
6	<i>Tricoderma</i> sp.	58	55	61