

## Processes for preserving and processing fish: Characterization and impact on microbiological quality in Lokossa, South-West Benin

Martinien Hospice Mahussi Assogba \*, Elie Linton and Issaka Youssao Abdou Karim

*Laboratory of Animal Biotechnology and Meat Technology, Department of Animal Production and Health, Polytechnic School of Abomey-Calavi, University of Abomey-Calavi, Abomey-Calavi, 01 BP: 2009 Cotonou, Benin.*

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### Abstract

The sanitary condition of consumers is comparable to the microbiological condition of fish. This study aimed at defining processing and preservation methods and determining their effect on the sanitary condition of fish in the Lokossa township. 27 processors were interviewed. 60 whole, pre-processed, smoked and fried samples of *S. melanotheron* and *C. guineensis* were taken. The identified processes were smoking (33%), frying (85.19%) and salting-drying (3.70%). All the processing operations began with trimming (scaling, gutting and cutting), drying and salting.

Both species had similar microbial counts ( $p > 0.05$ ) for total coliforms, faecal coliforms, *S. aureus* and *C. perfringens*, regardless of processing (whole, pre-processed, fried or smoked). FMAT counts and enterobacteria in *S. melanotheron* were significantly different between types of processing ( $p < 0.05$ ).

For *C. guineensis*, enterobacteria loads varied among pre-treated and whole fish, and smoked and fried fish. *E. coli* occurred in all samples, and 16.67% of the samples had Salmonella regardless of species. Minimizes the bacteriological load of the fish by smoking and frying under sanitary conditions.

**Keywords:** Food safety; Microbiology; Tilapia; *Coptodon guineensis*; *Sarotherodon melanotheron*; Health; Food technology

### 1. Introduction

Fish is an essential source of protein for humans and livestock. In 2022, fish production reached a record 223.2 million tonnes, with per capita consumption standing at 20.7 kg. This increase is linked to the growing global population and high protein requirements. Similarly, in Benin, the annual national production of fish is estimated at 88,677 tonnes, while the annual requirement is 202,000 tonnes. The deficit is made up by importing frozen fish (see references [2] and [3]). However, even if efforts are made to meet quantitative needs, a healthy diet and improved health are said to guarantee food security, according to [1].

In Benin, 1.1 million people are food insecure, and the phenomenon affects several departments. These include the Mono department, which has a food insecurity rate of 27% [4]. In order to overcome this issue, the target population in Mono needs to consume a varied, balanced and healthy diet. While this population has some access to cereals through agriculture, they have very limited access to protein-rich foods.

Fish is the most widely consumed animal protein in the diet of the population in this department because it is available and accessible to low-income households. This is due to the department's many lakes, including Ahémé and Toho, and

\* Corresponding author: Martinien Hospice Mahussi Assogba

its rivers, such as the Couffo and Mono, and their tributaries. Once caught, the fish are consumed either fresh or, more often, after processing, as they are highly perishable.

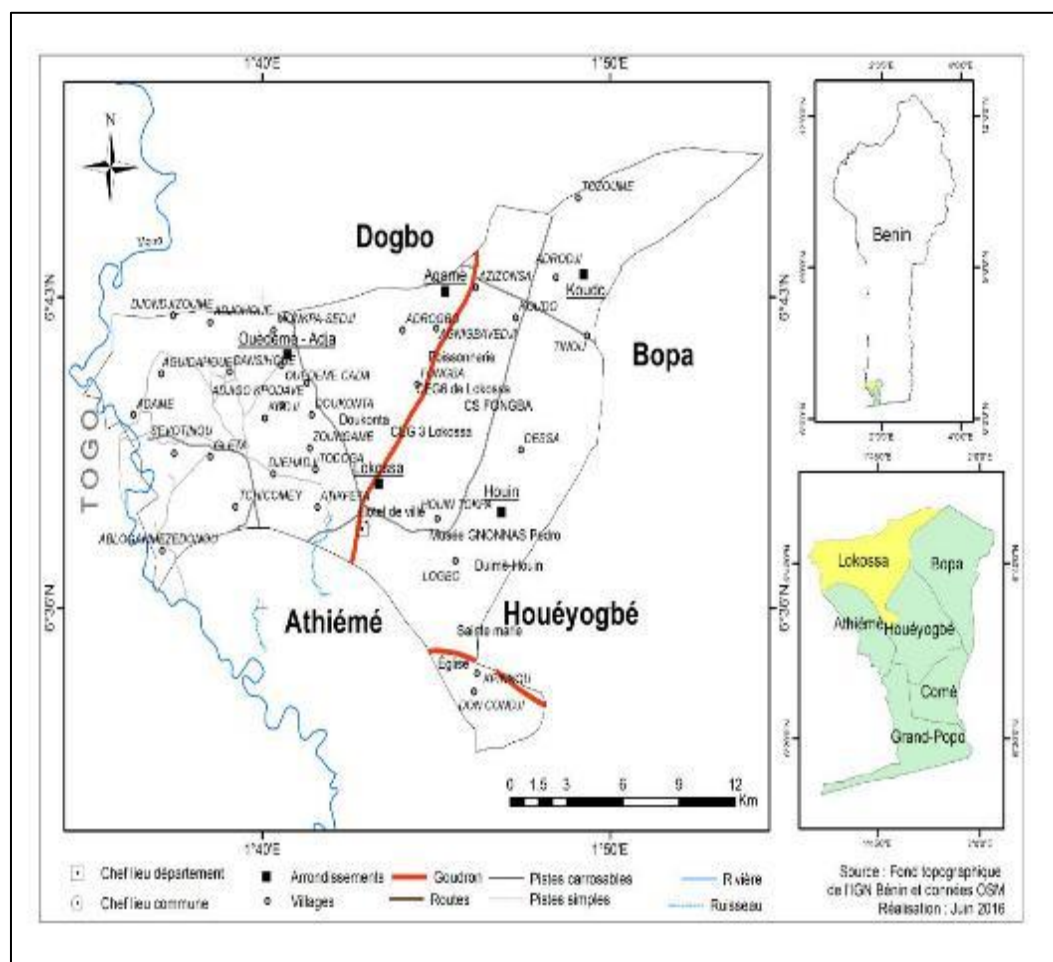
Immediately after capture, the muscles are practically sterile, containing only bacteria on the skin, gills and in the viscera [5]. Most of this bacterial flora (with the exception of *Clostridium botulinum*, *Vibrio para haemolyticus* and *Listeria monocytogenes*) is banal in nature, and therefore harmless or only responsible for altering the marketability of products [6].

Fish can be contaminated by pathogens during handling, processing and marketing [7]. There are different processes to enhance the preservation of fish in the Mono department. In this perspective, there is a necessity to review these processes with the aim of enhancing the quality of the final product. Sensory and sanitary quality are the most required qualities by the consumers. There has been some work to assess the microbiological quality of fish in the coastal departments (cf. references [8, 9] in the Atlantic at Abomey Calavi [10, 11, 12, 13, 14, 15, 9], and in the Mono and Ouémé departments [9].

The studies by [9] focused only on fresh fish. To ensure a healthy food supply in Mono, we have decided to examine the impact of processing and preservation methods on the hygiene of fish in Lokossa, a town in south-west Benin. This study will characterise these processes and assess the bacteria present in fresh fish before and after processing.

## 2. Materials and methods

### 2.1. Study area



**Figure 1** Geographical location of the township of Lokossa

Located in the north-western part of the Mono department, at an altitude of 31 meters, the township of Lokossa has a latitude of 6° 37' 60" North and a longitude of 1° 43' 0" east. It extends over 1,605 km and is one of 6 districts in the

department. It covers an area of 260 km<sup>2</sup>, which represents 16% of the Mono's surface area and 0.23% of Benin's total surface area (112,622 km<sup>2</sup>) [16].

Bordered to the North by the township of Dogbo in the Couffo department, to the South by the the township of Athiémé and Houéyogbé, to the east by that of Bopa and to the west by Togolese territory, this township is divided into five districts: Lokossa, Agamè, Koudo, Houin and Ouèdèmè-Adja.

These districts are subdivided into 8 town districts and thirty-seven (37) villages, making a total of forty-five (45) localities. The main town of the township (Lokossa) is located around 106 km from Cotonou (Benin's economic capital). It is also the capital of the Mono township [16]. Figure 1 shows the administrative map of Lokossa.

### 3. Methodology

Data were collected in two stages: a survey and microbiological analyses. The survey focused on fish processing in the Lokossa township, while the analyses focused on germ research.

#### 3.1. Survey of transformation processes/conservations

It carried out a survey of fish processing methods in two two districts of the township, namely Houin and Lokossa Centre, which run through Lakes Toho and Doukon respectively.

The questionnaire format solicited the following information: identification of respondent, activity carried out, dependents, kind of fish processed, processing units, cleanliness of processing unit, process and brief description of the same, and storage time.

Twenty-seven women were interviewed during the survey: twenty-three at Houin district and four at central Lokossa. They were randomly chosen from among the most diligent and regular fish processors.

#### 3.2. Fish sampling

The Houin district, and more specifically the Duimè riverbank, has been chosen to collect fish samples for microbiological analysis. The site is packed with fishermen, fishmongers and processors. Two species of fish (*Coptodon guineensis* and *Sarotherodon melanotheron*), the most heavily fished in the township, were selected for data collection to assess microbiological quality.

Samples of fresh, pre-treated, fried and smoked fish were acquired from fishmongers and processors in the arrondissements of Lokossa center and Houin (Township of Lokossa). Microbiological analyses were carried out at the Laboratory of Animal Biotechnology and Meat Technology of the Department of Animal Production and Health of the Polytechnic School of Abomey-Calavi of the University of Abomey-Calavi.

Each sampled fish was aseptically placed in a Stomacher bag and then in a cooler fitted with carboglass. In total, the sanitary quality study was carried out on 30 fish of each species (Table 1).

**Table 1** Sampling plan

Types of fish	Fish species	Number of fish sampled
Whole fish	<i>C. guineensis</i>	10
	<i>S.melanotheron</i>	10
Pre-treated fresh fish	<i>C. guineensis</i>	10
	<i>S.melanotheron</i>	10
Fried fish	<i>C. guineensis</i>	5
	<i>S.melanotheron</i>	5
Smoked fish	<i>C. guineensis</i>	5
	<i>S.melanotheron</i>	5
Total		60

### 3.3. Microbiological analyses

#### 3.3.1. Microorganisms tested in samples

The flora tested were : Total Aerobic Mesophilic Flora (TAMF), total and fecal coliforms (with a search for *Escherichia coli*), which provide information on the freshness of the flesh and the slaughtering conditions, respectively; *Pseudomonas*, which are psychotrophic germs that indicate fish spoilage and can develop in fish kept at ambient temperatures (25 to 40°C), as well as pathogenic germs such as *Salmonella*, *Staphylococci* and Sulfite-Reducing Anaerobes (SRA).

Samples were analysed in accordance with ISO standards specific to each germ:

- TAMF : [17];
- *Salmonella* : [18];
- Suspected pathogens staphylococci: [19];
- Sulfite-Reducing Anaerobes (SRA) : [19];
- Total coliforms :[20];
- Fecal coliforms : [20];
- *E.coli* : [21];
- *Pseudomonas* : [22].

### 3.4. Test sample, preparation of dilutions and inoculum

Dilutions are prepared for microbiological examination in accordance with standard NF V08-010. Twenty-five grams of flesh from the head, body, tail and gills of the fish were aseptically removed into a sterile Stomacher bag, then 225 ml of buffered peptone water was added and the whole homogenised in the Stomacher for 30 seconds to one minute. The crushed material was then left to stand for a maximum of 45 minutes at room temperature to revive the germs. This mixture thus constituted the mother suspension, which was used to prepare successive dilutions in geometric progression of reason 10. The various dilutions were carried out using this stock solution. One millilitre of the said solution ( $10^{-1}$ ), taken after homogenisation, was inoculated into 9 ml of sterile Tryptone salt or diluent (0.1%peptone + 0.85% NaCl) to obtain dilution  $10^{-2}$ .

After homogenisation, 1 ml of this  $10^{-2}$  dilution was also inoculated into 9 ml of sterile Tryptone Salt to obtain dilution  $10^{-3}$ . This was followed by dilutions  $10^{-4}$  and  $10^{-5}$ .

### 3.5. Criteria for assessing the good bacteriological quality of fish

For good fish quality, the microbiological criteria below were used to assess the analytical results obtained in our work. These are:

- Absence of Sulfite-Reducing Anaerobes (SRA) in 1 g ;
- Absence of pathogenic germs, in particular *Salmonella*, in 25 g ;
- Absence of coliforms in 1 g ;
- Total" aerobic flora < 25 germs/g for freshly obtained meat and <500 germs/g for cold-preserved meat.

### 3.6. Statistical analysis

Survey data on the processing of fresh fish and fish frying and smoking processes in the township were analysed using the *Proc Freq* procedure in [23], and frequency significances were determined using the chi-squared test.

The Generalised Linear Model procedure (*Proc GLM*) was used for analysis of variance. The factors used to explain variation in bacterial loads were treatment mode, fish species, and the interaction between treatment mode and species. The F-test was used to determine the significance of each effect, and the means were calculated and compared using the Student's t-test. Frequencies of *Pseudomonas*, *E. coli* and *Salmonella* were calculated using the *Proc Freq* procedure in SAS and compared using the chi-squared test and two-tailed Z-test.

For each relative frequency, a 95% confidence interval (CI) was calculated. This was done using the following formula:

$$CI = 1,96 \sqrt{\frac{[P(1 - P)]}{N}}$$

P is the relative frequency. N is the sample size.

## 4. Results

### 4.1. Activities carried out by processors

Several activities were carried out by processors in the township of Lokossa (table 2). All the respondents process and sell fish, and 11.11% of them also farm. Those who also fish and rear livestock represent only 3.70% of the women surveyed. In addition to farming, the women were involved in other activities such as selling porridge (15.38%) and other products (30.77%).

**Table 2** Activities carried out by processors in Lokossa

Variables		Numbers	Percentage (%)	CI
Agricultural activities	Agriculture	27	11.11b	11.85
	Breeding	27	3.70b	7.12
	Fishing	27	3.70b	7.12
	Fish farm	27	0.00b	0.00
	Fish processing and sales	27	100.00a	0.00
Other activities	Sale of porridge	27	15.38ab	13.61
	Retailer of various products	27	30.77a	17.41
	Production of palm oil	27	7.69b	10.05
	Production of palm oil, food sales	27	7.69b	10.05
	Production and sale of "akassa"	27	7.69b	10.05
	Processing and sale of "come"	27	7.69b	10.05
	Clothes launderer	27	7.69b	10.05
	Catering	27	7.69b	10.05
Main activities	Agriculture	27	0.00b	0.00
	Breeding	27	0.00b	0.00
	Fishing	27	0.00b	0.00
	Fish farm	27	0.00b	0.00
	Fish processing and sales	27	77.78a	15.68
	Other activities	27	11.11b	11.85

Percentages between the classes of the same column followed by different letters differ significantly at the threshold of 5% ; CI: Confiance Interval; % : percentage.

### 4.2. Processed fish species and processing methods

Table 3 shows the different species of fish processed in the township of Lokossa and the techniques used. The fish species most commonly used for processing is tilapia (92.59%), whose proportion is higher ( $p < 0.001$ ) than that of the other species: dentex, sea bream and horse mackerel. The proportions of dentex, sea bream and processed horse mackerel were 11.11%, 7.41% and 7.41% respectively of all fish and did not differ significantly between them ( $p > 0.05$ ). The processing methods used in the township are: smoking, frying and salting-drying.

Of these processes, frying was the most used (85.19%), followed by smoking (33%) and salting-drying (3.70%).

Finally, cold preservation, drying, smoking and frying were the preservation techniques used in the township. Preservation by drying was the most used, with 77.79% of processors surveyed using a preservation technique.

**Table 3** Processed fish species, processing and storage techniques

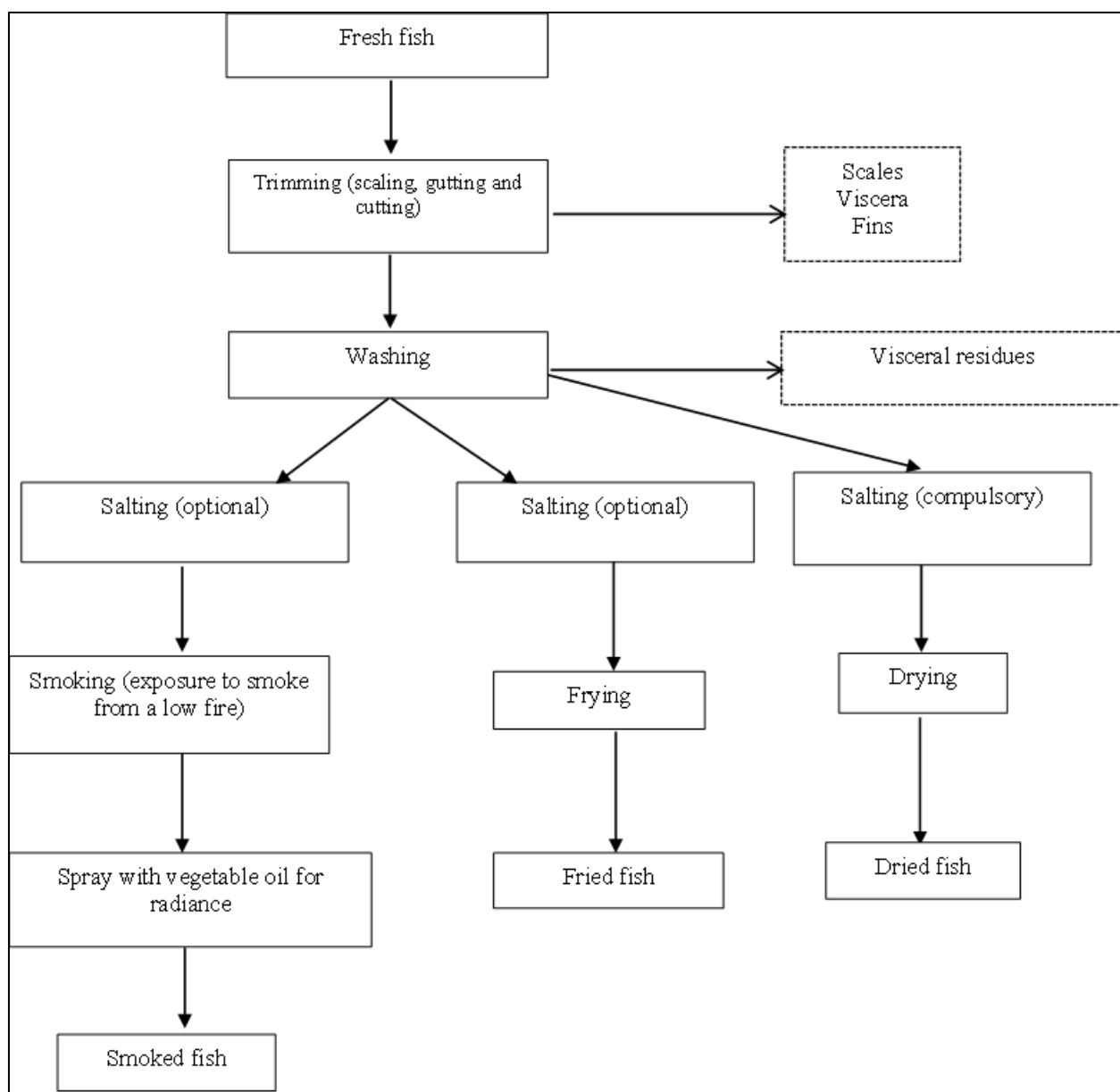
Variables		Township of Lokossa		
		Numbers	Percentage (%)	CI
Species of fish purchased for processing and preservation	Tilapia	27	92.59a	9.88
	Dentex	27	11.11b	11.85
	Dorade	27	7.41b	9.88
	Horse mackerel	27	7.41b	9.88
Processing methods used	Smoking	27	33.33b	17.78
	Frying	27	85.19a	13.40
	Salting and drying	27	3.70c	7.12
	Production of lanhouin	27	0.00c	0.00
Types of conservation technique	Ice preservation	27	25.93b	16.53
	Regular drying	27	77.79a	15.68
	Regular smoking	27	3.70c	7.12
	Other: regular heating in oil	27	3.70c	7.12

Percentages between the classes of the same column followed by different letters differ significantly at the threshold of 5% ; CI: Confidence Interval; % : percentage.

#### 4.3. Description of fish processing and preservation processes

Figure 2 describes the various fish processing and preservation processes in the township of Lokossa.

All processing begins with trimming (scaling, gutting and cutting), and these trimming stages vary depending on the species. The fish were then washed or cleaned to remove any residues. The rest of the process depended on each processing method. For smoking, the cleaned fish is drained, salted (optional operation) soaked in oil and exposed to the smoke over a low fire. During smoking, the fish is sprayed with peanut oil. This process took around 4 hours for 50 kg of fish. As for frying, after trimming, the fish were cleaned and rinsed with water, drained, then salted (optional) before being fried in red oil or peanut oil. This process took around 3 and a half hours for 50 kg of fish. Salting-drying was compulsory after trimming and draining. The fish were then exposed to the sun. This process lasted between 48 and 72 hours, sometimes longer depending on the humidity in the air. Finally, to preserve the processed fish, processors used drying and smoking techniques. Each processing procedure corresponds to a processing diagram (Figure 2).



**Figure 2** Diagram of fish processing in the township of Lokossa

#### 4.4. Microbiological quality of fish

##### 4.4.1. Effect of treatment method and species on the microbial load in fish

Table 4 shows the effect of treatment and species on microbial load. For each of the fish species (*S. melanotheron* and *C. guineensis*), the microbial loads were similar for total Coliforms, fecal Coliforms, *Staphylococcus aureus*, *Clostridium perfringens* ( $p > 0.05$ ) whatever the treatment method (whole, pre-treated, fried and smoked).

However, the highest Total Aerobic Mesophilic Flora (TAMF), load ( $p < 0.05$ ) was obtained on whole *S. melanotheron* samples. TAMF loads were identical on the other treated *S. melanotheron* samples.

For Enterobacteria, the highest loads were obtained in whole and pretreated *S. melanotheron* and in whole and pretreated *C. guineensis*. No cases of Enterobacteria were obtained in the fried and smoked samples of either species.

**Table 4** Effect of treatment method and species on the microbial load in fish

Types of treatments	Load of <i>S. melanotheron</i>												
	Total Mesophilic (CFU/g)		Aerobic Flora		Total coliforms (CFU/g)		Fecal coliforms (CFU/g)		<i>Staphylococcus aureus</i> (CFU/g)		<i>Clostridium perfringens</i> (CFU/g)		Enterobacteri a (CFU/g)
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	DS	
Whole fish	1330000.00a	89566.85	925.00a	671.75	722.5a	958.12	37.75a	31.46	26.75a	28.63	236.5a	25.59	
Pre-processed fish	642166.67b	113844.19	0.00a	00.00	1400a	0.00	11.25a	3.88	33.00a	2.82	226.60a	0.28	
Fried fish	620000.00b	0.00	0.00a	0.00	0.00a	0.00	0.00a	0.00	0.00a	0.00	0.00b	0.00	
Smoked fish	320000.00b	0.00	0.00a	0.00	0.00a	0.00	17.50a	0.00	0.00a	0.00	0.00b	0.00	
Significance test	*		NS		NS		NS		NS		*		
	Load of <i>C. guineensis</i>												
Whole fish	737833.33a	714884.95	925.00a	671.75	300.00a	212.132	8.75a	9.54	45.00a	7.77	184.00a	23.20	
Pre-processed fish	1037500.00a	83 674 307	1250.00a	212.13	800.00a	848.52	18.75a	15.9	70.25a	45.6	269.00a	20.6	
Fried fish	141333.33a	0.00	0.00a	0.00	0.00a	0.00	30.00a	0.00	0.00a	0.00	0.00b	0.00	
Smoked fish	189333.33a	0.00	0.00a	0.00	0.00a	0.00	0.50a	0.00	0.00a	0.00	0.00b	0.00	
Significance test	NS		NS		NS		NS		NS		*		

NS :  $p > 0.05$  ; \* :  $p < 0.05$  ; DS : Standard Deviation ; Means between the classes of the same column followed by different letters differ significantly at the threshold of 5%; CFU/g: Colony Forming Unit per gram



#### 4.5. Frequencies of Salmonella, Pseudomonas and Escherichia coli

The frequency of *Pseudomonas* in *S. melanotheron* was higher (50%) than in *C. guineensis* (16.67%). *Pseudomonas sp* was found in whole fish and pretreated fish (50% in each case), while no cases were observed in fried and smoked fish (Table 5). The frequency of salmonella was 50% in the case of pre-treated fish, while no salmonella cases were found in the case of whole, fried and smoked fish. The frequency of the presence of salmonella in the samples analysed was 16.67% for each of the species (*S. melanotheron* and *C. guineensis*) (Table 6).

Finally, *Escherichia coli* was present in all the fish samples analysed (Table 7).

**Table 5** *Pseudomonas* frequencies

Source of variation		Number	Percentage of <i>Pseudomonas sp</i> (%)	
			Presence	CI
Species	<i>S. melanotheron</i>	12	50.00a	40.01
	<i>C. guineensis</i>	12	16.67a	29.82
Treatments	Whole fish	8	50.00a	49.00
	Fried fish	5	0.00b	0.00
	Smoked fish	5	0.00b	0.00
	Pre-treated fish	8	50.00a	49.00

Percentages between the classes of the same column followed by different letters differ significantly at the threshold of 5% ; CI: Confiance Interval; % : percentage.

**Table 6** *Salmonella* frequencies

Source of variation		Percentage of <i>salmonella sp</i> (%)		
		Number	Presence	CI
Species	<i>S. melanotheron</i>	12	16.67a	29.82
	<i>C. guineensis</i>	12	16.67a	29.82
Treatments	Whole fish	8	0.00b	0.00
	Fried fish	5	0.00b	0.00
	Smoked fish	5	0.00b	0.00
	Pre-treated fish	8	50.00a	49.00

Percentages between the classes of the same column followed by different letters differ significantly at the threshold of 5% ; CI: Confiance Interval; % : percentage.

**Table 7** Frequencies of *Escherichia Coli*

Source of variation		Percentage of <i>E. coli</i> (%)		
		Number	Presence	CI
Species	<i>S. melanotheron</i>	12	100.00	0.00
	<i>C. guineensis</i>	12	100.00	0.00
Treatments	Whole fish	8	100.00	0.00
	Fried fish	5	100.00	0.00
	Smoked fish	5	100.00	0.00
	Pre-treated fish	8	100.00	0.00

I CI: Confiance Interval ; ; % : percentage.

## 5. Discussion

### 5.1. Microbiological quality of fish

#### 5.1.1. Total Aerobic Mesophilic Flora (TAMF)

For *Coptodon guineensis*, there was no significant difference between the mean TAMF loads for whole fish, pre-treated fish, smoked fish and fried fish. On the other hand, a difference was noted between the TAMF load of fresh fish and those of smoked, fried and pre-treated fish for *Sarotherodon melanotheron*. The species therefore has an effect on the TAMF load. These results differ from those of Assogba (2018a and b) who noted that there was no difference between the TAMF loads of four species (*S. melanotheron* and *C. guineensis*; *Trachurus Trachurus* and *Scomber Scombrus*) and those of [24] on horse mackerel and mackerel.

Several authors have noted this similarity of charge between species. These include: [10] in Benin, [25] in Togo and [26] in Nigeria on artisanally smoked fish. The difference in loads of *S. melanotheron* observed between fresh fish and smoked, fried and pre-treated fish was reported by [25] in Togo following an assessment of the microbiological quality of artisanally smoked fish, and by [5] and [7] for artisanally smoked fish in Ivory Coast intended for export.

Low loads of TAMF were observed in fried and smoked fish compared with whole and pretreated fish, whatever the species. The TAMF load of pretreated *S. melanotheron* fish was higher than that of whole fish.

This difference could be explained by the pollution of intestinal germs during fish evisceration.

[27] have confirmed this, stating that the risks of contamination of the flesh by fecal flora are significant. The reduction in the bacterial flora of fried fish is due to the action of heat. The reduction in the bacterial flora of smoked fish could be due to the effect of smoking. [28] reports that smoke contains many volatile compounds such as phenols and polyphenols with strong antiseptic or bacteriostatic properties.

In addition, despite the bactericidal effect of the processes, a high load of TAMF on the finished product is due to contamination of the products after processing, as a result of various handling operations. Indeed, most fish processors do not apply basic hygiene rules. The design of smoking and frying workshops does not take into account the application of good manufacturing practices (separation of clean and soiled sectors, forward motion or non-interlacing of production lines).

### 5.2. Total coliforms

The total coliform load did not vary between whole fish, smoked fish, pre-treated fish and fried fish for either *S. melanotheron* or *C. guineensis*. These results are similar to those observed in Benin by [15] on the same species and by [24] between horse mackerel and mackerel. These authors observed that total coliform loads did not change according to species. [7] and [25] observed similar results in Ivory Coast and Togo respectively.

Contrary results were obtained by [15] where the loads of fresh fish (*S. melanotheron*; *C. guineensis*) decreased when fried and smoked. The same trends were observed by [29] when fresh horse mackerel ( $0.64 \times 10^3$  CFU/g vs  $0.0042 \times 10^3$  CFU/g) and mackerel ( $0.77 \times 10^3$  CFU/g vs  $0.0012 \times 10^3$  CFU/g) were smoked. The differences observed are due to the fact that frying and smoking had a bactericidal effect on total coliforms.

### 5.3. Fecal coliforms

There was no significant difference between the fecal coliform loads of whole, pre-treated, smoked and fried fish for either *S. melanotheron* or *C. guineensis*. Contrary to our results, [24] observed on horse mackerel and mackerel that the fecal coliform load was higher in fresh fish than in smoked fish.

The same results were obtained by [29] for horse mackerel ( $0.37 \times 10^3$  CFU/g vs  $0.0039 \times 10^3$  CFU/g) and mackerel ( $0.36 \times 10^3$  CFU/g vs  $0.0012 \times 10^3$  CFU/g). According to [29], processing has an effect on germ reduction in fish.

This is due to the effect of heat, which significantly reduces the initial germs, which explains the absence of fecal coliforms in fried and smoked fish. These results show that the fish were not contaminated after processing by the operator. In fact, heat-tolerant coliforms are indicative of poor hygiene conditions, in this case staff hygiene [25].

They are found in the digestive tracts of humans and animals (fish). Their presence in whole and pretreated fish is due to contamination of the viscera from pretreated fish. [30] found results different from those of our study on the same species with the presence of fecal coliforms in fried and pretreated fish, which in his study indicates cross-contamination (after processing) from humans to the finished product.

#### 5.3.1. *Staphylococcus aureus* and *Clostridium perfringens*

For both species, there was no significant difference between the staphylococci and clostridium loads for whole fish, pretreated fish, smoked fish and fried fish. These results are similar to those of [24] on horse mackerel and mackerel. In fact, *Staphylococcus aureus* is a germ that is not part of the fish flora, but its natural habitat is the skin and mucous membranes of humans and animals.

Its presence in the fried and smoked fish in this study, with the exception of the zero-load obtained in the smoked *S. melanotheron*, indicates human contamination after processing and therefore a failure to comply with hygiene rules.

As for *Clostridium perfringens*, it is a highly ubiquitous bacterium that is widely distributed throughout the environment (soil, sediment, sewage, slurry, cadavers, dust, plant surfaces).

Its presence in whole and pre-treated fish may be due to polluted water in water bodies and soil where the fish are treated. Its absence from fried and smoked fish in this study is due to the effect of heat. According to [28], a temperature of 63 °C and 75 °C destroys germs and heat-labile enterotoxins.

#### 5.4. Enterobacteria

For both species, the enterobacteria loads of whole and pretreated fish are higher than those of fried and smoked fish.

Contrary results were obtained in studies carried out by [31] where no load was observed in horse mackerel and mackerel stored at -18°C. The microbial loads of whole and pre-treated fish were similar to each other, as were those of fried and smoked fish, which were both zero. Enterobacteria are fecal contamination germs. Their absence in the processed products reflects the effect of heat during processing, which contributed to their destruction.

In addition, the application of and compliance with hygiene rules guarantees a healthy finished product free from fecal contamination germs.

#### 5.5. Salmonella, *Escherichia coli* and *Pseudomonas*

The frequency of salmonella was 50% in the pre-treated fish compared with 0% in the whole fish. *S. melanotheron* had the highest salmonella load, 50% compared with 16.67% for *C. guineensis*. [30] identified 26.67% salmonella in *S. melanotheron* and 30% in *C. guineensis* at the riverbank and market in Abomey-Calavi. The presence of *E. coli* was 100% in all species despite the treatments carried out. The presence of these two pathogenic germs is therefore proof of fecal contamination during the handling of fish before and after processing.

This contamination could occur during the various processing/preservation processes (daily smoking, regular smoking and drying) where processed fish are handled and exposed to the open air by processors who do not pay much attention to the rules of hygiene and good practice. The maximum permitted load for *E. coli* must be less than 100 CFU/g, while for salmonella, no presence is tolerable in 25g of sampled flesh. In addition, the frequency of *Pseudomonas sp* was high in both whole fish and pre-treated fish. In addition, 50% of *S. melanotheron* compared with 16.67% of *C. guineensis* presented this load. According to [32] *Pseudomonas sp* is a germ that spoils fish stored on ice at 0°C. These germs are subject to rapid spoilage.

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## 6. Conclusion

This study shows that fish processing methods in the Lokossa township include frying, smoking and salting. Fish processing is essentially a female activity. Of these processes, frying is the most common. Microbiological analysis shows that fresh, whole and pre-processed fish have a high bacterial load. However, the frying and smoking processes reduce the total aerobic mesophilic bacteria (TAMF) load in *S. melanotheron*.

The presence of total coliforms, fecal coliforms, *Staphylococcus aureus* and *Clostridium sp* in the two species indicates that there is poor hygiene during processing and preservation, from reception to processing. The presence of Salmonella and *E. coli* in all the fish species, whether processed or not, indicates that the fish are not safe for consumption.

To promote processed fish quality and working and residential conditions for processors, the following is suggested: organize the fish processing and preservation business; and schedule regular training for processors in proper production and hygiene practices before, during, and after processing and preservation.

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## Compliance with ethical standards

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### *Disclosure of conflict of interest*

No conflict of interest in our manuscript

### *Statement of ethical approval*

Ethical standards have been respected.

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