



## Microbiological Quality of "Soumbala", an African Locust Bean (*Parkia biglobosa*) Condiment Sold in the Markets of Abidjan, Côte d'Ivoire

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### Authors' contributions

This work was carried out in collaboration among all authors. Authors KO and BZBIA designed the study, performed the statistical analysis. Author ADMP managed the analyses of the study and wrote the first draft of the manuscript. Authors YKM and KNR managed the literature searches. All authors read and approved the final manuscript.

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### ABSTRACT

**Aims:** Produced in an artisanal way from the fermentation of the seeds of *Parkia biglobosa*, "Soumbala", is a condiment very appreciated in several African countries including Côte d'Ivoire. This study was conducted to assess the microbiological quality and fungal profile of this condiment sold in the markets of nine communes in Abidjan.

**Study Design:** Food safety.

**Place and Duration of Study:** Soumbala's samples were collected in the markets during the month of October 2019 and analyzed at the Laboratory of Microbiology and Food Biotechnology of the Nangui Abrogoua University, Côte d'Ivoire.

**Methodology:** For this purpose, 27 samples of "Soumbala" were taken from the different markets and analysed. The loads of the different microorganisms (mesophilic aerobic germs, total coliforms,

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moulds) were determined by counting after culture in agar medium. The microbiological quality has been assessed according to Directive 2005/2073/EC. The physico-chemical composition (pH, titratable acidity, moisture content) of "Soumbala" has been determined according to standard methods. The identification of the mould strains isolated from the different samples was carried out using the identification keys.

**Results:** The results of the various physico-chemical parameters obtained ranged from 13.81 to 20.31%, 5.58 to 6.50 and 3.73 to 9.06 % for moisture content, pH and titratable acidity, respectively. The mesophilic aerobic germ loads of 7.21 to 7.70 log<sub>10</sub> cfu/g determined in the analyzed samples are above the acceptability limit (6 log<sub>10</sub> cfu/g) applied in this study. The maximum loading of total coliforms was 3.92 log<sub>10</sub> cfu/g. All mould loads are below the acceptability limit. The mould strains isolated and identified from the identification keys belong to the genus *Aspergillus*.

**Conclusion :** A public health risk related to the consumption of "Soumbala" could exist if these moulds produced mycotoxin.

**Keywords:** "Soumbala"; microbiological quality; fungal profile.

## 1. INTRODUCTION

Condiments are aromatic products used to season culinary preparations and can be of animal (meat broth), mineral (salt) or vegetable (mustard) origin [1]. In Africa, traditional condiments obtained by fermentation of fish or legume products are an important part of the diet of the population, and among the condiments of vegetable origin is African mustard. This mustard is derived from the spontaneous fermentation of oilseeds such as those of the African locust bean *Parkia biglobosa*, which leads to the production of a condiment that is well known in many households in West Africa. Several names are attributed to this condiment namely nétérou in Senegal [2,3], dawadawa in Niger, irù in Nigeria, affitin in Benin [4], "Soumbala" in Mali, Burkina Faso and Côte d'Ivoire. This condiment has several virtues and is consumed for its high nutritional and organoleptic values. These qualities have earned him scientific interest among some researchers in Burkina Faso, Senegal, Benin and Nigeria [5,6,7,8]. This work has shown that in the fermentation of *Parkia biglobosa* seeds, the dominant microflora is made up of spore-forming germs of the *Bacillus* genus which are responsible for the development of very particular aromas.

In Côte d'Ivoire, particularly in Abidjan, the galloping urbanization and the high concentration of populations constitute an important market for the consumption of food products from inland cities and bordering countries. Among these products are perennial and food crops, but also seasoning condiments such as "Soumbala" marketed under unhygienic conditions. According to Azokpota et al. [4], "Soumbala", due to its high

water and nutrient content, is a suitable medium for the proliferation of microorganisms. In addition, the natural fermentation process, heavy handling, improper transportation, and poor packaging increase the risks of exposure to varieties of microorganisms, some of which could be pathogenic.

With the exception of the work carried out by Camara et al. [9], little scientific work relating to Soumbala exists in Côte d'Ivoire, yet this condiment is used in the preparation of many traditional culinary dishes and could be a source of microbial contamination if it is of poor sanitary quality. Food safety is therefore still threatened by many pathogens responsible for a variety of diseases [10]. Thus, the prevention of these risks requires frequent analysis of food products intended for human consumption. The objective of this study is to determine the microbiological quality and fungal profile of "Soumbala" sold in the markets of the communes of the city of Abidjan.

## 2. MATERIALS AND METHODS

### 2.1 Sample Collection

Soumbala samples were taken in the markets of nine communes in Abidjan. These are the communes of Treichville, Abobo, Port bouët, Cocody, Attecoubé, Yopougon Koumassi, Adjame and Marcory. In each market, three samples of Soumbala of about 500 g each were taken from the vendors, for a total of 27 samples analyzed. The various samples were taken in stomacher bags and transported to the laboratory in a cooler for microbiological and physico-chemical analyses.

## 2.2 Physico-Chemical Analyses

### 2.2.1 Moisture content

The moisture content was determined according to the method of AOAC (1990) which is based on the loss of mass of the sample in the oven at 105°C until a constant mass is obtained. Thus, 5 g of peanut paste was introduced into a glass capsule of known mass ( $m_0$ ). The capsule containing the sample of total mass ( $m_1$ ) has been placed in an oven (Memmert, Germany) which is set at 105±2°C for a period of 24 hours. After removing from the oven, the capsule was cooled in a desiccator. After cooling, the whole content (sample + capsule) is weighed, and the mass is recorded. The operation is then repeated every 2 hours until a constant mass ( $m_2$ ) is obtained, and the moisture content is determined by the following formula:

$$\text{Moisture content (\%)} = \frac{(m_1 - m_2)}{(m_1 - m_0)} \times 100 \quad (1)$$

### 2.2.2 pH determination

For pH determination, 10 g of each Soumbala sample are dissolved in 100 mL sterile distilled water. After homogenization, the pH is measured with a digital pH meter (WTW multi line P4) previously calibrated with pH 7 phosphate and pH 4 acetate buffer solutions.

### 2.2.3 Titratable acidity determination

Titrate acidity was determined using the method described by Amoah-Awua et al. [11]. Ten grams of "Soumbala" were homogenized in 100 mL of distilled water. After filtration, 2 to 3 drops of 1% phenolphthalein are added to 10 mL of filtrate taken for the test sample and the mixture is determined with 0.1N sodium hydroxide solution. The end of the determination is marked by a pale pink colouring. The acidity rate expressed as a percentage is obtained according to the following formula:

$$\% \text{ Acidity} = \frac{V(\text{NaOH}) \times N(\text{NaOH}) \times 0.09 \times 100}{V(\text{test sample})} \quad (2)$$

## 2.3 Microbiological Analysis

Enumeration of microorganisms was carried out according to the agar dilution method. Ten grams from each Soumbala sample were homogenized with 90 mL of buffer peptone water (Bio-rad, France), and serial decimal dilutions were performed. The enumeration of mesophilic aerobic germs was carried out according to the

standard (NF ISO 4833 -2003) on Plate count Agar (Bio-rad, France). The inoculated Petri dishes were incubated at 37°C/24 hours. Total coliform counts were performed in the mass on Violet Red Bile Lactose Agar (Bio-rad, France) according to NF ISO 4832-2006. The inoculated Petri dishes were incubated at 37°C/24 hours. Mould enumeration was performed on Sabouraud chloramphenicol agar (Bio-rad, France) according to ISO 6611 :2004. The inoculated Petri dishes were incubated at 30°C/72 hours. The microbial loads expressed in colony forming units (cfu/g) per gram.

## 2.4 Fungal Profile

After enumeration, the isolated mould strains were transplanted onto Sabouraud agar with chloramphenicol and identified on the basis of macroscopic and microscopic characteristics using identification keys [12]. Macroscopic examination takes into account the surface and underside of the Petri dishes containing the purified mould strains. On the surface, the characteristics observed are the appearance of the colonies (powdery, cottony...), the shape, colour, size. On the reverse side, the observation is made on the ability of the mycelium to penetrate the agar, the colour and the presence or absence of pigment. Microscopic examination consisted of observing the appearance of the mycelium (whether or not partitioned), the shape of the conidia or spores, the conidial heads and that of the conidiophores, the presence or absence of metules and the arrangement and shape of the phialides. To carry out this examination, part of the colony to be identified is stained with methylene blue on a slide and observed with an optical microscope at objective x 40.

## 2.5 Statistical Analysis

Means, standard deviations and the analysis of variance (ANOVA) were performed using Statistica software (Stat., Soft, Inc., 7.1). The study of correlations between microbiological and physico-chemical parameters was carried out using the ADE4TkGUI software.

## 3. RESULTS

### 3.1 Physico-Chemical Parameters of the Various Samples

Table 1 presents the results of the physico-chemical analysis of the different samples taken in the nine communes. The moisture content of the different samples ranges from 13.21 to

20.13% with the highest rate in the Yopougon samples and the lowest rate in the Attecoubé samples. All "Soumbala" samples have an acidic pH. Abobo samples recorded the lowest pH (5.58) and Attecoubé samples recorded the highest pH (6.5). The titratable acidity values obtained ranged from 3.73 to 9.06%. These values were obtained in the Yopougon and Adjame samples, respectively. No significant differences were recorded for the moisture content, pH and titratable acidity of the different samples from one commune to another.

### 3.2 Microbial Load from the "Soumbala" Samples

The average loads in total coliforms, mesophilic aerobic germs and moulds of the different samples of Soumbala taken in the nine communes of the city of Abidjan are presented in Table 2. The average loads of total coliform in the samples range from 0 to 3.92 log<sub>10</sub> cfu/g. The samples from the communes of Adjame, Treichville and Attécoubé do not show total coliforms. The highest load of total coliforms was obtained in the samples from Yopougon. The average loads of the mesophilic aerobic germs are of the same order in all the samples of the nine communes. The highest load was obtained in the Port bouët samples (7.7 log<sub>10</sub> cfu/g) and the lowest load in the Koumassi samples (7.21 log<sub>10</sub> cfu/g). Mould loads range from 1.56 to 2.88 log<sub>10</sub> cfu/g. The maximum and minimum loads are obtained in the samples from Attécoubé and Abobo, respectively. No significant differences are recorded for total coliforms, mesophilic aerobic germs and moulds in the different samples from one commune to another. All mesophilic aerobic germs loads are above the threshold value M, in contrary to mould loads, which are below the corresponding threshold value M. With the exception of the samples from

the communes of Yopougon and Port bouët, the total coliform loads of the other communes are below the standard.

### 3.3 Correlations between Microbial Loads and Physico-Chemical Parameters

A positive correlation is observed between moisture content and mesophilic aerobic germs loads. pH and titratable acidity are positively correlated with mould loads. Total coliform loads are negatively correlated with moisture content, pH and titratable acidity. mesophilic aerobic germs loads are negatively correlated to pH and titratable acidity (Fig. 1).

### 3.4 Fungal Profile

On the basis of identification keys taking into account macroscopic and microscopic characters, only one fungal genus was identified. This is the *Aspergillus* genus (Table 3).

## 4. DISCUSSION

The different samples of "Soumbala" taken in the markets of the nine communes of Abidjan have acid pH values between 5.58 and 6.50. The different acid pH values obtained in the "Soumbala" sold in Abidjan could be linked to the non-respect of the fermentation time (40 to 72 hours) of the seeds of *Parkia biglobosa* during the production of this condiment. Indeed, during the production of "Soumbala", the fermentation process of the seeds varies the pH. Thus the acid pH on the first day reaches an alkaline pH at the end of fermentation [13]. This result is different from that of Hongbété et al. [7]. These authors obtained in samples of "Afitin" in Benin, a condiment similar to "Soumbala", alkaline pH. According to Parkouda et al. [14], the fermentation of nere seeds is alkaline.

**Table 1. Moisture content, pH and titratable acidity values of the different samples of "Soumbala"**

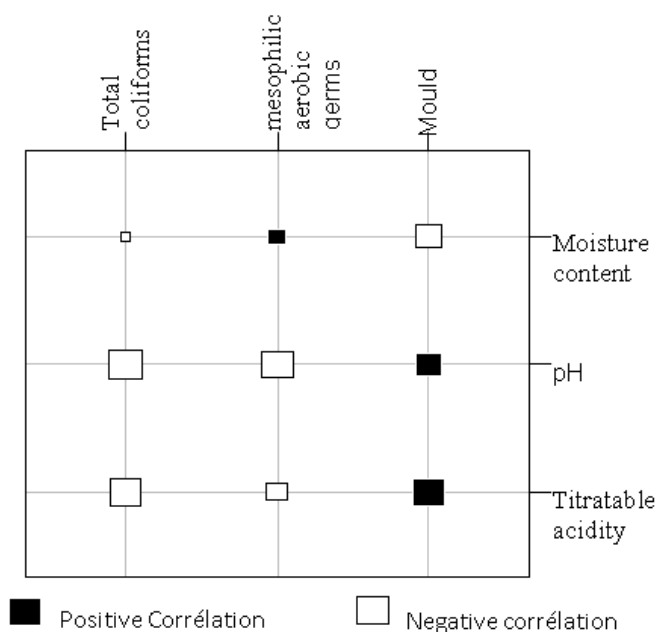
Communes	Moisture content (%)	pH	Titratable acidity (%)
Yopougon	20,13±1,55 <sup>a</sup>	5,71±0,48 <sup>a</sup>	3,73±1,27 <sup>a</sup>
Cocody	20,10±0,34 <sup>a</sup>	5,93±1,02 <sup>a</sup>	4,80±0,60 <sup>a</sup>
Port-bouet	13,21±0,96 <sup>a</sup>	5,65±0,08 <sup>a</sup>	6,76±2,92 <sup>a</sup>
Abobo	16,20±4,71 <sup>a</sup>	5,58±0,12 <sup>a</sup>	4,60±2,22 <sup>a</sup>
Marcory	15,20±2,21 <sup>a</sup>	6,09±0,25 <sup>a</sup>	3,80±2,35 <sup>a</sup>
Adjame	13,81±3,49 <sup>a</sup>	6,47±0,04 <sup>a</sup>	9,06±3,17 <sup>a</sup>
Koumassi	14,67±1,74 <sup>a</sup>	6,38±0,15 <sup>a</sup>	5,66±0,56 <sup>a</sup>
Treichville	16,47±1,22 <sup>a</sup>	6,39±0,17 <sup>a</sup>	5,60±0,43 <sup>a</sup>
Attecoubé	13,81±3,49 <sup>a</sup>	6,50±0,04 <sup>a</sup>	7,70±1,83 <sup>a</sup>

Means assigned the same letter in the same column are not significantly different at the 5% threshold.

**Table 2. Microbial load from the "Soumbala" samples**

Communes	Microorganisms ( $\log_{10}$ cfu/g)		
	Total coliforms	mesophilic aerobic germs	Mould
Yopougon	3,92 $\pm$ 0,03 <sup>a</sup>	7,51 $\pm$ 0,15 <sup>a</sup>	2,35 $\pm$ 0,32 <sup>a</sup>
Cocody	1,30 $\pm$ 2,25 <sup>a</sup>	7,41 $\pm$ 0,09 <sup>a</sup>	2,78 $\pm$ 0,15 <sup>a</sup>
Port-bouet	3,63 $\pm$ 0,57 <sup>a</sup>	7,70 $\pm$ 0,09 <sup>a</sup>	2,71 $\pm$ 0,37 <sup>a</sup>
Abobo	0,92 $\pm$ 1,60 <sup>a</sup>	7,36 $\pm$ 0,13 <sup>a</sup>	1,56 $\pm$ 1,40 <sup>a</sup>
Marcory	1,87 $\pm$ 1,81 <sup>a</sup>	7,31 $\pm$ 0,28 <sup>a</sup>	1,59 $\pm$ 1,38 <sup>a</sup>
Adjame	<1	7,42 $\pm$ 0,12 <sup>a</sup>	2,79 $\pm$ 0,13 <sup>a</sup>
Koumassi	2,18 $\pm$ 1,99 <sup>a</sup>	7,21 $\pm$ 0,31 <sup>a</sup>	2,43 $\pm$ 0,33 <sup>a</sup>
Treichville	<1	7,47 $\pm$ 0,02 <sup>a</sup>	2,72 $\pm$ 0,17 <sup>a</sup>
Attecoube	<1	7,31 $\pm$ 0,17 <sup>a</sup>	2,88 $\pm$ 0,04 <sup>a</sup>
Threshold (M)	3	6	4

Means assigned the same letter in the same column are not significantly different at the 5% threshold.

**Fig. 1. Graphical presentation of showing correlation between different parameters**

The moisture levels (13.21 to 20.13%) recorded in the different samples are similar to those reported by Camara et al. [9]. The moisture content of food products plays a decisive role during their conservation. It is a parameter that significantly affects the storage and growth rate of microbial contaminants [15].


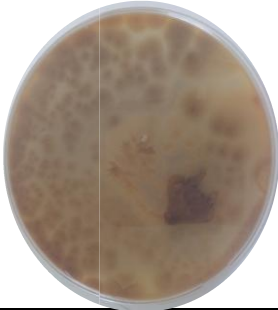
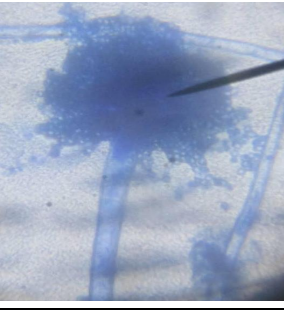

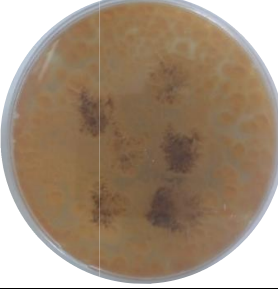
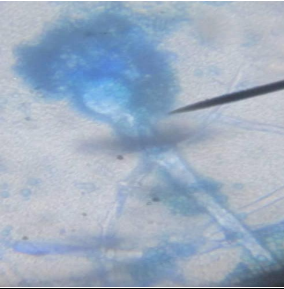

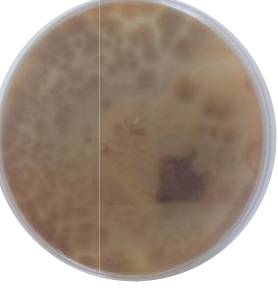
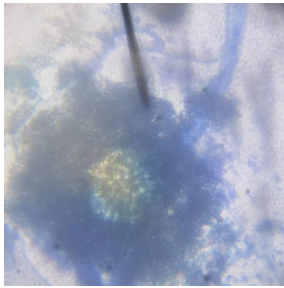

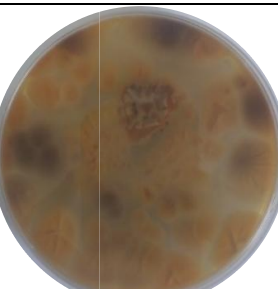
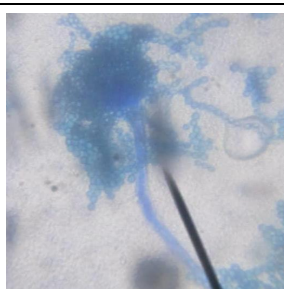
From one commune to another, the physico-chemical parameters of the different samples of "Soumbala" analysed do not vary significantly. The "Soumbala" in the markets of the nine communes of Abidjan could come from the same production area or from an identical process (fermentation time).

Samples of " Soumbala " showed high loads of mesophilic aerobic germs. This result could be explained by the high water content of this product, which would favour the growth of mesophilic aerobic germs after stopping the fermentation process. These values are high compared to those reported in the work of Camara et al. [9] i.e.  $2.8 \cdot 10^4 \pm 1.7 \cdot 10^3$  to  $1.8 \cdot 10^5$  cfu/g on the microbiological study of the same biological material. These different loads are positively correlated with moisture content. This parameter could have an influence on the growth of these microorganisms in "Soumbala" sold in the markets of the communes of Abidjan. The high load of mesophilic aerobic germs would

favour a strong spoilage of the product and would constitute a risk for the presence of pathogenic germs [16]. According to Kasse et al. [16], the level of variability in mesophilic aerobic germs from one seller to another could come from the production process, or from contamination during the sale and/or conservation of leftovers, but could also depend on the density of street traffic, which influences

environmental hygiene and therefore the contamination of the product. The mesophilic aerobic germs loads of all the samples of "Soumbala" from the nine communes are higher than the acceptability limit ( $6 \log_{10}$  cfu/g). These samples are therefore of unsatisfactory microbiological quality with regard to these germs.

**Table 3. Macroscopic and microscopic characteristics of fungal contaminants isolated from "Soumbala" samples**

Macroscopic aspects		microscopic aspect	Fungal Genus
Obvers	Backhand		
			<i>Aspergillus</i> sp1
			<i>Aspergillus</i> sp2
			<i>Aspergillus</i> sp3
			<i>Aspergillus</i> sp4

The presence of total coliforms in samples of "Soumbala" could be due either to a lack of hygiene in the manufacturing process or to faulty storage and/or marketing conditions. The loads obtained in this study are lower than those reported by Azokpota et al. [4]. The different physico-chemical parameters studied have no influence on total coliform loads. These loads are mainly due to an intake linked to poor hygiene practices.

Mould loads are below the microbiological criteria set for these microorganisms. The positive correlation between pH, acidity and mould is due to the cosmopolitan nature of mould. Indeed, molds are able to grow in acidic foods.

*Aspergillus* is the genus of mould identified in the various samples. This result is consistent with that of Roukaya et al. [17] in a study of the same biological material in Niger. According to Meyer et al. [18], moulds of the genus *Aspergillus*, *Penicillium* and *Fusarium* are known to be contaminants of agricultural products and/or for their ability to produce toxic secondary metabolites that can have immediate harmful effects on the body such as acute poisoning, immune deficiency or cancer.

## 5. CONCLUSION

The objective of this study was to assess the microbiological quality and the mycoflora of "Soumbala" sold in the markets of nine communes of Abidjan in order to contribute to the fight against food insecurity. This study shows that "Soumbala" sold in markets of the nine communes of Abidjan is of unsatisfactory microbiological quality. The study of correlations has shown that physico-chemical parameters such as moisture content, pH and titratable acidity have an influence on the loadings of certain microorganisms (mesophilic aerobic germs, moulds). The different strains of mould isolated from "Soumbala" samples belong to the genus *Aspergillus*. On the basis of the results obtained, it would first of all be very useful to recommend to the consumer to add "Soumbala" during culinary preparations and to encourage producers and sellers to adopt adequate hygienic conditions.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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