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Original article

Microbiological and nutritional quality of banku a fermented maize and cassava-based food consumed in West Africa

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Abstract

Banku is a food made of fermented maize flour and fermented cassava dough. It is produced and consumed by the Togolese, Nigerian and Ghanaian peoples living in Côte d'Ivoire. Not only is this food less known by the original inhabitants of the Côte d'Ivoire but the nutritional and sanitary quality of this food is not defined. However, its preparation remains empirical and is done in unhygienic conditions. Thus, objective of this study is to promote banku through its nutritional and sanitary characteristics in order to ensure consumer safety. Two localities (Grand-Bassam and Gonzagueville) of Côte d'Ivoire where there is a strong foreign community were chosen to take the various samples. The analyses were carried out by biochemical and microbial properties during the production of banku. The results showed that the fermented maize flour had the highest titratable acidity with a value of $0.31 \pm 0.01\%$. The phytate content was lower in the mixture of fermented maize flour and cassava dough with a value of 8.81 ± 0.57 mg/100 g DM. After cooking, the mesophilic aerobic germs load in the banku indicated (5.6 ± 0.4) × 103 CFU/g which is below the standard prescribed by CODINORM. The promotion of banku in Côte d'Ivoire would contribute to food self-sufficiency and growth of the country's economy. The nutritional and microbiological properties are in accordance with the CODINORM (2001) standard. Banku can be safely consumed in Côte d'Ivoire and worldwide.

Keywords Maize flour, Cassava dough, Fermentation, Nutritional and microbiological quality, Banku

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Introduction

Fermentation is also used by indigenous populations in developing countries, particularly in the many traditional technologies for processing cassava and maize [1]. Fermented products derived from cassava (Manihot utilissima) and maize (Zea mays L.) include banku. Banku is a Ghana dish that is prepared with a mixture of fermented maize and cassava contained in a smooth whitish paste, cooked in boiling water [2]. Banku is served as a side dish with soup, stew, or in a pepper sauce with fish [3]. In Côte d'Ivoire, this dish is produced and consumed by the Togolese, Nigerian and Ghanaian people. It is an unknown food in Côte d'Ivoire and its production relies on empirical knowledge, based on the traditional experience of the foreign producer. Its production involves a combination of steps in which the mixture of maize flour and cassava dough is fermented for three days before being kneaded by hand to obtain a suitable consistency for subsequent cooking. This method of production does not seem to offer a guarantee to curb the proliferation of microorganisms. For example, hand-kneading or numerous hand manipulations during production are favorable to the contamination and growth of many microorganisms [4]. According to [5], food safety in Africa is crucial. This concerns fermented foods in particular. They are produced in the absence of hygiene practices, production and marketing. In addition, the production sites are not very healthy and the premises are not very adequate. Other problems are often encountered by producers, such as the lack of control over manufacturing parameters such as temperature, fermentation processes, etc. [5]. Fermentation is an ancestral process of food processing that has three major advantages overall, namely improving the nutritional quality of food; improving the sanitary quality of food, improving the organoleptic quality of food. These benefits are produced by certain microorganisms of interest to the food industry. These include lactic acid bacteria and yeasts. In fact, lactic acid bacteria, thanks to the organic acids they produce, guarantee food safety, as well as providing it with specific flavor and texture characteristics. Work carried out by [6] showed that lactic acid bacteria help to reduce cyanide levels in cassava and are responsible for the sour taste in fermented products due to the production of lactic and acetic acids. Yeasts had previously been identified as the second most predominant germs involved in cassava-based foods, after lactic acid bacteria. Cassava-based foods after lactic acid bacteria capable of contributing to flavor development in fermented products [7]. A poorly done fermentation, often leads to a product of inconsistent quality, poor hygiene, low nutritional value and short shelf life [8]. Similarly, the presence of antinutritonal substances can reduce the nutritional value of foods by interfering with the absorption of certain minerals (iron, calcium, zinc), and the digestibility of proteins and carbohydrates. In some cases, they can potentially cause toxicity and health problems when present in the body in excessive quantities. Thus, to ensure the health of consumers and to promote a new traditional food on the Ivorian territory, it is necessary to know the microbiological and nutritional characteristics of banku. On the one hand, this study will allow us to put in place strategies for rapid and early detection of microbiological and anti-nutritional hazards and prevention of food-borne diseases for the production of banku of consistent and healthy quality. On the other hand to solve the problem of food self-sufficiency in Côte d'Ivoire. Until today, there are no studies on the biochemical composition and the sanitary quality of this food which could be integrated in the food habits of the people of Côte d'Ivoire and even in the whole world. Therefore, the general objective of this study is to valorize the *banku* in order to promote its consumption on the African continent and in the whole world.

Materials and methods

Sample source and collection

The material consisted of four (04) samples taken during the production of banku: the maize flour obtained after fermentation, the fermented cassava dough, the mixture of the maize flour and the fermented cassava dough and finally the product obtained after cooking « the banku ». Fermentations were carried out in ROUND plastic bowls D. 36 8L POLA of 28 cm in diameter and 12.5 in height covered tightly with clean and sterile linen or in GILAC HACCP food plastic bins - volume 12 to 55 L, of height 18 cm and diameter 32,5 cm. These samples were taken in two (02) localities (Grand-Bassam and Gonzagueville). These localities were chosen because they are home to Togolese, Nigerian and Ghanaian banku consumers. Gonzagueville is a district of the commune of Port Bouet (Abidjan). While Grand-Bassam is a city located 14 km from Gonzagueville. In each locality, five producers were selected. For each visit, four samples were taken from a producer: 500g of fermented maize flour, 500g of fermented cassava dough, 500g of the mixture of fermented maize flour and fermented cassava dough and finally 500g of the product obtained after cooking (the banku). Three visits were made to each producer. A total of 120 samples were taken from the 10 women farmers, 12 samples per farmer. These samples were packaged in sterile "Stomacher" bags, placed in a cooler containing carbohydrate ice at 4°C and transported to the central laboratory of the NANGUI AB-ROGOUA University in Abidjan, Côte d'Ivoire where they were refrigerated at 4°C before any analysis.

Production follow-up

For production monitoring, all production units participated. Grand Bassam and Gonzagueville were chosen as the production zones. The banku processors were observed during production in order to learn about the different stages of banku production. This monitoring helped to explain certain results in this study.

Biochemical analysis

Sample acidity

pH and titratable acidity were determined by the method of [9]. Forty grams of samples were ground in 300 ml of distilled water in a porcelain mortar and then centrifuged at 4000 tours/min for 30 min. The pH was determined on 50 ml of the supernatant using a pH-meter (P107 Consort). Total titratable acidity (TTA) was determined by titrating 10 ml of supernatant used for pH determination against 0.1 M NaOH using phenolphthalein as indicator. TTA was calculated as percentage.

Macronutrient contents

The dry matter content were determined gravimetrically in an oven at 105 °C and every 24 hours the weight of the sample was determined until a stable weight was obtained [10]. Watersoluble carbohydrates were determined by the phenol sulphuric acid method according to [11] and the values were expressed in mg/100 g of dry matter, while the reducing sugars were quantified as described by [12] and expressed in mg 100 g⁻¹ of dry matter.

Phenolic compounds content

Total polyphenol contents were determined using the colorimetric method of Folin-Ciocalteu [13]. The absorbance was read at 725 nm against a blank without extract taken as reference. Quantification of total polyphenols is done according to a linear calibration line (y=ax + b) performed by a standard extract gallic acid at different concentrations (0 to 1000 µg ml⁻¹). Polyphenol content is expressed as gallic acid equivalents in milligrams per 100 grams of dry matter.

$$Polyphénols\left(mg/100g\right)=\frac{OD_{725}\times5\times10^{3}}{5,04\times m_{e}}$$

Calibration line: $OD_725 = 5.04$, mass (mg) Gallic acid, R2 = 0.992, me : mass (g) of the sample.

Flavonoid content was measured by colorimetric assay using the method described by [14] using aluminum chloride. To a volume of 0.5 mL of each methanolic extract was successively added 0.5 mL distilled water, 0.5 mL aluminum chloride (10%, w/v), 0.5 mL sodium acetate (1M), and 2 mL distilled water. Then, the tubes were allowed to stand for 30 min at room temperature. The optical density reading was taken by spectrophotometer at 415 nm (BK-UV1000

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spectrophotometer, Biobase, Qingdao, China), compared to a control containing distilled water instead of methanolic extract. Finally a concentration range of quercetin from 0 to 0.1 mg/mL was performed for the calibration curve. The total flavonoid content was expressed as mg quercetin equivalent per 100 g dry matter

Flavonoids
$$(mg/100g) = \frac{0D_{415} \times 2 \times 10^3}{18, 12 \times m_e}$$

Calibration line: $OD_415 = 18.12$, mass (mg) Quercetin, **R2** = 0.99. **me :** mass (g) of the sample.

Anti-nutritional compounds

The determination of phytates in the sample was performed according to the method described by [15] Latta and Eskin (1980) using Wade's reagent. One (1) g of sample was homogenized in 20 ml of HCl (0.65 N) under stirring for 12 h at room temperature. The mixture was centrifuged at 3000 rpm for 40 min using a centrifuge (SIGMA 3-16P, Germany). A 0.5-ml sample of each supernatant was taken followed by addition of 3 ml of Wade's reagent. Then the tubes were allowed to stand for 20 min in the dark and the absorbance reading was done with a spectrophotometer (BK-UV1000 spectrophotometer, Biobase, Qingdao, China) at 490 nm against the control containing no extract. Finally, a calibration curve was performed using a phytic acid range of concentration from 0 to 10 mg/mL. Results were expressed as mg phytic acid equivalent (PAE)/100 g dry matter (DM).

$$Phytates (mg/100g) = \frac{OD_{490} \times 4}{0.033 \times m_e}$$

Calibration line: OD_490 = 0.033 mass (μ g) Phytate sodium, **R2** = 0.99., **me :** mass (g) of the sample.

The method used for the determination of oxalates is that described by [16]. Two (2) grams of sample were dried and ground and homogenized in 25 mL of H2SO4 (3M) under magnetic stirring for 1 h at room temperature. The mixture was then filtered through Whatman filter paper or paper towels. Then, a 25 mL volume of this filtrate was titrated under heat with 0.05 M potassium permanganate (KMnO₄) solution until a persistent pink turn for 30 seconds. The oxalate content expressed in mg 100 g⁻¹ dry matter was obtained by the following equation:

$$Oxalates (mg/100g) = \frac{2, 2 \times V_{eq} \times 100}{m_e}$$

Veq: volume (mL) of KMnO₄ poured at the equivalence, **me:** mass (g) of the sample.

The determination of tannins was performed as described by [17] A volume of 1 mL of methanolic extract was taken and to this volume was added 5 mL of vanillin reagent (0.1 mg vanillin in 70% (v/v) hydrochloric acid). Then, the tubes were allowed to stand for 20 min in the dark and the

absorbance reading on a spectrophotometer (BK-UV1000 spectrophotometer, Biobase, Qingdao, China) was taken at 500 nm against the blank containing distilled water in place of the methanolic extract. Finally, a calibration curve was performed using a tannic acid range of concentration from 0 to 0.1 mg mL-1. The results were expressed as mg tannic acid equivalent (TEA)/100 g dry matter (DM).

$$Tannins (mg/100g) = \frac{OD_{500} \times 10^3}{3, 11 \times m_e}$$

Calibration line: $OD_500 = 3.11$, mass (mg) Tannins acid, **R2** = 0.99, **me :** mass (g) of the sample.

Microbial analysis

The samples of our food to be analyzed were prepared in the proportions (g/mL), according to the technique described by French Standard (NF) ISO 6887-V08-010-6 (2013). Ten (10) grams of our food were introduced into a sterile glass bottle containing 90 mL of buffered peptone water (Conda, Spain) previously autoclaved (121 °C, 45 min, 1bar). The mixture obtained after manual homogenization by shaking for 2 min corresponds to the stock solution. Successive decimal dilutions were then prepared from this stock solution ranging from 10⁻¹ to 10⁻⁸.

In accordance with standard NF ISO 4833-2003, 1 mL of the stock suspension and 1 mL of the decimal dilutions produced were duplicated on different empty petri dishes, then added to PCA (Plate Count Agar) agar medium supercooled at 45°C. After homogenization and solidification, a second layer of PCA medium was poured into each dish. Petri dishes were then incubated at 30 °C in an oven for 24 to 72 h after solidification. To count mesophilic aerobic germs on PCA agar, petri dishes containing 30 to 300 were selected for counting.

In accordance with NF ISO 7954-1988, 0.1 mL of inoculum corresponding to dilutions 10⁻³, 10⁻⁴ was surface-seeded in a Petri dish containing 15 mL of Sabouraud chloramphenicol medium. Incubation took place at 37 °C for 3 to 5 days. Petri dishes of Sabouraud chloramphenicol agar containing 15 to 150 characteristic yeast colonies were considered for enumeration. Yeast colonies appeared whitish, smooth and bulging, with a diameter of 0.5 to 2 mm.

The medium used for lactic acid bacteria enumeration is MRS (Man Rogosa Sharp) agar in accordance with ISO 15 214 (1998). Seeding was done by spreading 0.1 mL stock suspension or decimal dilutions retained on the surface of the agar previously poured and cooled in a petri dish. Petri dishes were incubated anaerobically in jars for 48 h at 30 °C. Plates with colony counts between 15 and 150 were counted.

Neutral red crystal violet bile lactose agar (VRBL agar) was used for coliform enumeration. Inoculation was car-

ried out in the mass with 1 mL of inoculum in sterile Petri dishes. Then 12 to 15 mL of supercooled medium at 45 °C was poured into the Petri dishes containing the inoculum. The resulting mixture was homogenized by gentle manual agitation. After solidification, a second layer of 4 mL of the same medium was poured. Incubation was carried out for 24 h at 30°C for total coliforms (NF ISO 4832 (V08-015), 2006) and 44 °C for fecal coliforms (NF ISO 4832 (V08-060), 2009). Colonies appeared red, purplish and round. All characteristic colonies present in plates containing 15 to 150 colonies were counted.

Bacillus was counted on plates Mossel agar (AES Laboratoire, COMBOURG France). The stock solution or decimal dilutions were placed in an 80 °C water bath for 10 min, then cooled immediately. This treatment destroys the vegetative forms of the microorganisms. The medium used for *Bacillus* research and enumeration was Mossel agar. Inoculation was carried out by spreading 0.1 mL of the mother suspension or decimal dilutions on the surface of agar previously poured and cooled in petri dishes. Incubation took place at 30 °C for 24 to 48 h. *Bacillus* colonies on Mossel agar are pink with the presence of a clear, opaque halo around the culture, or yellow with the absence of an opaque halo. Presumptive *Bacillus* colonies present in plates containing 15 to 150 colonies were counted.

According to NF ISO 16140, 2003, *E. coli* inoculation was carried out by spreading 0.1 mL of the mother suspension or decimal dilutions on the surface of RAPID'*E. coli* agar, previously poured and cooled in petri dishes. Incubation took place at 37 °C for 24 h.. On RAPID'*E. coli* agar, *E. coli* colonies appear purple to pink. *E. coli* colonies present in plates containing 15 to 150 colonies were counted.

Salmonella was detected using [18] multi-stage method: pre-enrichment on non-selective medium, followed by enrichment on selective medium and isolation on selective agar. For pre-enrichment on non-selective medium, 25 g of our sample is added to 90 mL of peptone water in a sterile flask. The well-homogenized mixture is incubated at 37 °C for 24 hours. Then, for enrichment in selective medium, 1 mL of the pre-enriched culture was pipetted into 10 mL of sterile Rappaport Vassiliadis broth. Incubation took place for 24 h at 37 °C. Each enrichment culture was streaked onto *Shigella-Salmonella* agar (SS, Oxoid). Incubation took place at 37°C for 24 h. On SS agar, presumptive colonies were colorless, transparent with or without a black center.

According to NF ISO 6888-1/1999, inoculation was carried out by spreading 0.1 mL of the mother suspension or decimal dilutions on the surface of Baird Parker agar, which had been poured and cooled in petri dishes. Incubation took place at 37°C for 48 h. On Baird Parker agar, presumptive

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S. aureus colonies were either shiny black, entire, convex, surrounded by clear zones extending into the opaque medium, or shiny black, entire, convex, without well-defined clear zones, or dark grey. Presumptive *Staphylococcus aureus* colonies on plates containing 15 to 150 colonies were counted.

Tryptone Sulfite Cycloserine agar is recommended for the detection and enumeration of *Clostridium perfringens*. For petri dish culture, heat the product to be tested to destroy vegetative forms and activate spores. Place 1 mL of the product to be tested, or its decimal dilutions, in sterile Petri dishes and add 15 ml of liquefied TSC agar to each dish within 15 min, mix thoroughly and leave to solidify. Incubation takes place anaerobically at 37 °C for 24 h. Finally, count the colonies surrounded by a black halo, due to the reduction of sulfite to iron sulfide precipitate. Take the reading very soon after removal from the jar.

Statistical analysis

The assays were performed in triplicate and the data presented are the means and standard deviations of these three determinations calculated with Excel 2016. Means obtained after different treatments were compared by analysis of variance (ANOVA) using Duncan's multiple comparison test at 5% level of significance.

Results and Discussion

Production of banku

Banku is a little-known food in Côte d'Ivoire, unlike in other countries such as Togo, Nigeria and Ghana. It is a Ghanaian dish prepared with a mixture of fermented maize and cassava contained in a smooth, whitish paste, cooked in boiling water [2]. Its preparation involves soaking the raw material (a mixture of maize and manioc) in water for 24 hours. After grinding, the maize flour and cassava dough are placed in a large container (the maize flour and cassava paste) with just enough hot water to moisten it completely. There are no strict rules regarding the ratio of maize flour to cassava dough. In most cases, 3 to 4 volumes of corn paste are mixed with 1 volume of cassava paste. The mixture is mixed well by hand and the container is covered with a clean cloth for a three-day fermentation in a warm place at 37°C or in the upper part of the refrigerator. When properly fermented, it should have a slightly sour, but not unpleasant aroma, like a rising bread dough. An over-fermented mixture does not taste good. The fermented dough should be stirred and kneaded continuously with the hands to obtain a suitable consistency for the subsequent baking at 100 °C. To cook, the fermented dough is put in a large pot and left steamcooked for twenty minutes or more, stirring constantly and vigorously. The *banku* should become thick and stiff. If it becomes too dry, water can be added if necessary. Form the *banku* into balls about the size of tennis balls. *Banku* can be served hot or cold at room temperature (Figure. 1).

Biochemical parameters during banku production

To ensure consumer health and to promote a traditional food that is new to Côte d'Ivoire, the nutritional and health parameters of the flour, dough, flour-dough mixture and the finished product called *banku* were studied. Thus, the fermented maize flour shows a pH of 4.18 ± 0.52 with an acidity level of 0.31 ± 0.01 %. While the fermented cassava dough records a pH of 3.58 ± 0.04 with an acidity level of 0.27 ± 0.04 . Under the same storage conditions, the pH of the mixture of fermented cassava flour and fermented cassava dough 3.53 ± 0.04 with an acidity level of 0.18 ± 0.01 . However, the pH of the final product (*bankou*) is 4.07 ± 0.08 with an acidity level of 0.05 ± 0.01 (Table 1).

Table 1. Samples acidification

Samples	pН	Titratable Acidity (%)
FMF	4.18 ± 0.52^{a}	0.31 ± 0.01^{a}
FCD	3.58 ± 0.04^{b}	0.27 ± 0.04^{a}
MMCF	3.53 ± 0.04^{b}	$0.18 \pm 0.01^{ m b}$
FP	$4.07\pm0.08^{\rm a}$	$0.05\pm0.01^\circ$

Notes: Values are means of triplicate samples, each analyzed in triplicate (n =9) \pm standard deviation Within the same column; mean values followed by a different alphabetical letter are statistically different (p \leq 0.05) (Duncan multiple t-test); **FMF** : Fermented maize flour ; **FCD** : Fermented cassava dough ; **MMCF** : Mixture of fermented maize flour and fermented cassava dough ; **FP** : Final product .

These pH values observed in the fermented samples and in the *banku* could be explained by the production of organic acids, notably lactic acid and acetic acid by lactic bacteria. In fact, lactic acid bacteria constitute the majority flora of fermented cassava and corn products. Similar remarks were obtained in the study conducted by [19] and [20] on the physicochemical characteristics of fermented cereal-based foods. It should be noted that the acidity obtained in the final product (banku) complies with the requirements of the Ivorian standard NI 03-08-003 on attiéké, the results of [21] on attiéké and those of other work on fermented cassava-based foods [22, 23].

In other hand, the dry matter content or water content undergoes changes during the preparation of *banku*. The highest dry matter content observed during *banku* production was in the fermented maize flour 61.25 ± 3.69 g 100/g FM and the lowest in the final product 25.16 ± 3.49 g100/g FM. The observed differences are significant at the 5 % level. Also, the results show the presence of total sugars in the four samples studied and the highest value was found in the fermented maize flour $7.74 \pm 0.05 \text{ mg } 100/\text{g DM}$, and the lowest was in the final product $0.51 \pm 0.04 \text{ mg } 100 \text{ g}^{-1}$ DM. However, reducing sugars were found only in the fermented maize flour with a content of $9.17 \pm 0.50 \text{ mg } 100/\text{g}$ DM. The observed differences are significant at the 5% level (Table 2).

Table 2. Content of some macronutrients

Samples	Dry matter (g/100g FM)	Total sugars (mg/100g DM)	Reducing sugars (mg/100g DM)	
FMF	(g/100g FW) 61.25 ± 3.69 ^a	7.74 ± 0.05^{a}	9.17 ± 0.50^{a}	
FCD	$41.95\pm4.82^{\mathrm{b}}$	$0.59\pm0.13^{\text{b}}$	$0.00\pm0.00^{\rm b}$	
MMCF	41.54 ± 2.03^{b}	$1.17 \pm 0.15^{\circ}$	$0.00\pm0.00^{\mathrm{b}}$	
FP	$25.16 \pm 3.49^{\circ}$	0.51 ± 0.04^{b}	$0.00\pm0.00^{\mathrm{b}}$	

Notes: Values are means of triplicate samples, each analyzed in triplicate $(n = 9) \pm$ standard deviation. Within the same

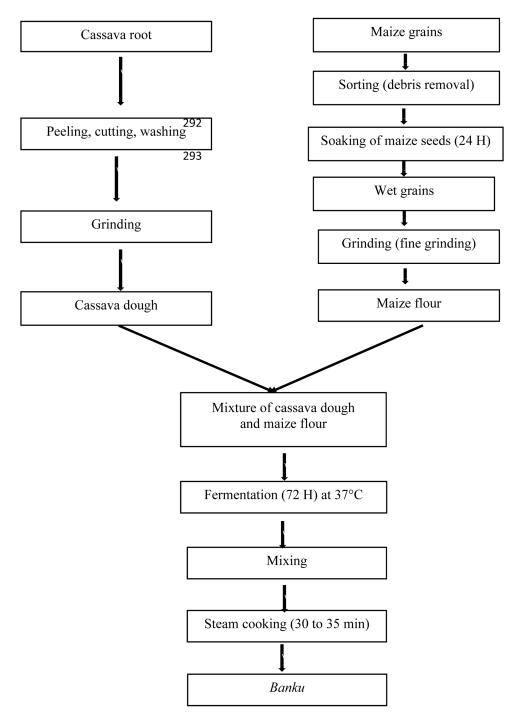


Fig. 1. Production diagram of *banku*

Remark : Cassava dough and maize flour can be fermented separately for 72 hours at 37°C and then mixed together after fermentation.

column; mean values followed by a different alphabetical letter are statistically different ($p \le 0.05$) (Duncan multiple t-test); **FMF :** Fermented maize flour ; **FCD :** Fermented cassava dough ; **MMCF :** Mixture of fermented maize flour and fermented cassava dough ; **FP :** Final product ; **DM:** Dry matter ; **FM:** Fresh matter.

Indeed, the different levels of dry matter and sugars encountered during the production of *banku* would not only be related to the raw material but also and especially to certain unit operations such as fermentation and cooking where a large amount of water is eliminated during the preparation of the fermented food [24]. During this production process, an elimination of these substances, especially starch, occurs during the fermentation stage, which allows a degradation of starch into fermentable sugars. This observation is also made by [25] and [8], during the fermentation of cassava dough.

Similarly, the reduction of phenolic compounds changes from 342.26 ± 4.55 to $47.95 \pm 2.50 \text{ mg}100/\text{g}$ DM for total polyphenols and 40.65 ± 0.46 to $7.77 \pm 0.39 \text{ mg} 100/\text{g}$ DM for total Flavonoids (Table 3).

Table 3. Phenolic compounds content

Sample	Total polyphenols (mg/100g DM)	Total flavonoids (mg/100g DM)	
FMF	342.26 ± 4.55^{a}	40.65 ± 0.46^{a}	
FCD	108.13 ± 2.62^{b}	11.26 ± 0.66^{b}	
MMCF	$108.80 \pm 3.48^{\rm b}$	$14.13 \pm 0.40^{\rm b}$	
FP	$47.95 \pm 2.50^{\circ}$	$7.77 \pm 0.39^{\circ}$	

Notes: Values are means of triplicate samples, each analyzed in triplicate (n =9) \pm standard deviation. Within the same column; mean values followed by a different alphabetical letter are statistically different (p \leq 0.05) (Duncan multiple t-test); **FMF :** Fermented maize flour ; **FCD :** Fermented cassava dough ; **MMCF :** Mixture of fermented maize flour and fermented cassava dough ; **FP :** Final product ; **DM:** Dry matter Anti-nutritional compounds notably Phytates, àxalates, tannin are also reduced during fermentation. The highest phytate content was found in the final product (86.42 ± 1.63) mg 100/g DM. While the highest oxalate content was in the fermented maize meal (132.00 ± 5.50) mg 100/g DM and the highest tannin content was found in the fermented corn meal (185.32 ± 2.41) mg100/g DM (Table 4).

Table 4. Anti-nutritional compounds content

Samula	Phytates	Oxalates	Tanins
Sample	(mg/100 g DM)	(mg/100 g DM)	(mg/100 g DM)
FMF	84.93 ± 0.62^{a}	$132.00 \pm 5.50^{\mathrm{a}}$	185.32 ± 2.41^{a}
FCD	$85.29 \pm 1.86^{\mathrm{a}}$	$40.33 \pm 3.18^{\mathrm{b}}$	$22.94 \pm 1.59^{\text{b}}$
MMCF	84.81 ± 0.57^{a}	$31.17 \pm 3.18^{\text{b}}$	54.77 ± 1.52°
FP	86.42 ± 1.63^{a}	$25.67 \pm 3.18^{\circ}$	22.51 ± 1.47^{b}

Notes: Values are means of triplicate samples, each analyzed in triplicate (n =9) \pm standard deviation. Within the same column; mean values followed by a different alphabetical letter are statistically different (p \leq 0.05) (Duncan multiple t-test); **FMF** : Fermented maize flour ; **FCD** : Fermented cassava dough ; **MMCF** : Mixture of fermented maize flour and fermented cassava dough ; **FP** : Final product ; **DM**: Dry matter

The reduction of phenolic compounds and Anti-nutritional compounds observed during the preparation of the *banku* would be due to the fermentation step of the different foods used content varied during the production of *banku*. Indeed, according to [26], it is known that spontaneous fermentation reduces the concentration of anti-nutritional substances and phenolic compounds. However, the analysis of physicochemical parameters indicates that the analyzed *banku* complies with the [27] CODINORM (2001) standard.

Microbial analysis during banku production

Concerning microbiological analysis, the table 5 shows the evolution of the microorganism load of fermented maize flour, fermented cassava dough, the mixture of maize flour

Microbial load of samples (CFU/g)				
Germs	FMF	FCD	MMCF	FP (Banku)
AMG	$(1.2 \pm 1.1) \times 10^{7a}$	$(2.6 \pm 2.8) \times 10^{8a}$	$(1.5 \pm 1.3) \times 10^{8a}$	$(5.6 \pm 0.4) \times 10^{3a}$
TC	ND	ND	$(2.7 \pm 1.9) \times 10^{0a}$	ND
FC	ND	ND	$(9 \pm 0.1) \times 10^{1a}$	ND
Yeasts	$(4.3 \pm 0.4) \times 10^{4b}$	$(5.4 \pm 0.4) \times 10^{4b}$	$(6.4 \pm 0.6) \times 10^{5b}$	ND
LAB	$(2.2 \pm 4.4) \times 10^{7a}$	$(1.9 \pm 0.5) \times 10^{6a}$	$(6.4 \pm 0.6) \times 10^{6a}$	ND
E. coli	ND	ND	ND	ND
S. aureus	$(2.8 \pm 0.9) \times 10^{3b}$	ND	$(9.5 \pm 0.9) \times 10^{0b}$	ND
Bacillus	$(5.9 \pm 1.5) \times 10^{2b}$	$(2.2 \pm 0.3) \times 10^{2c}$	$(5.2 \pm 0.2) \times 10^{3a}$	ND
Clostridium	ND	ND	ND	ND
Salmonella	Absent	Absent	Absent	Absent

Notes: Values are means of triplicate samples, each analyzed in triplicate $(n = 9) \pm$ standard deviation. On the same line; mean values followed by a different alphabetical letter are statistically different ($p \le 0.05$) (Duncan multiple t-test). AMG: Aerobic mesophilic germs; *E. coli: Escherichia coli; S. aureus: Staphylococcus aureus*; TC: Total coliforms, FC: Fecal coliforms, LAB: Lactic acid bacteria ND: Not detected, FMF : Fermented maize flour ; FCD : Fermented cassava dough ; MMCF : Mixture of fermented maize flour and fermented cassava dough ; FP : Final product.

and fermented cassava dough and the final product during the production process. The loads of aerobic mesophilic germs (AMG) varied with values ranging from (1.2 ± 1.1) ×10⁷ CFU/g in the fermented maize flour and (2.6 ± 2.8) ×10⁸ CFU/g in the fermented cassava dough. After mixing the maize flour and the fermented cassava dough, the loads of AMGs were $(1.5 \pm 1.3) \times 10^8$ CFU/g. In the final product, the aerobic mesophilic germs (AMG) load decreased to $(5.6 \pm 0.4) \times 10^3$ CFU/g (Table 5).

This microbial load was composed of coliforms, lactic acid bacteria, Bacillus, Salmonella, yeast and Staphylococcus aureus. According to [28] the total aerobic mesophilic flora is the primary contamination flora of food and the source of contamination of foodstuffs comes from the environment, dust, cross contamination and contamination due to handling. This flora of contamination is often made up of enterobacteria, Bacillus, Staphylococci, lactic bacteria or other potentially pathogenic agents. Their presence beyond the standards is due to a lack of hygiene in processing and poor storage conditions [28]. The presence of certain microorganisms in the mixture could be due to contamination of the ingredients by the hands of female producers during production, but also by domestic animals. These germs provide information on the lack of hygienic handling, inefficiency of processes, lack of ownership of premises and equipment used for production [29].

After cooking, the banku (final product) recorded a microbial load in mesophilic aerobic germs indicating the absence of coliforms, lactic acid bacteria, Bacillus, Salmonella, yeast and Staphylococcus aureus. The absence of all these germs searched shows that these microorganisms were eliminated during cooking. According to [28], the cooking temperature of 88 \pm 3.33 °C and a cooking time of 35 \pm 1.37 min are sufficient to eliminate all vegetative forms except spore forming forms. The observed mesophilic aerobic germs value indicates the presence of others in the banku that were not sought in our study. However, this mesophilic aerobic germs load in the *banku* is largely lower than the requirements of the Ivorian standard on fermented foods. The microbiological quality of the banku would be satisfactory and could therefore be consumed without danger to the health of the consumer. It should be noted that in all samples analyzed, clostridia and E. coli were not detected. This indicates that the lactic acid bacteria would have produced enough lactic acid during the fermentation to inhibit the development of Clostridium and E. coli. Indeed, lactic acid bacteria, thanks to the organic acids they produce, prevent the growth of most pathogenic germs [30, 31] and guarantee food safety, as well as providing them with very specific characteristics of flavour and texture.

Conclusion

The present study evaluated the biochemical and microbiological characteristics of banku, a fermented food consumed by living foreigners in Côte d'Ivoire. From a nutritional point of view, banku, a product of fermented maize flour and cassava ferment, would contribute significantly to improving the calorific intake of West African populations. In poor families, it could find its place as a substitute for some food, since its nutritional properties are in accordance with the CODINORM 2001 standard. In terms of sanitary quality, not only does banku contain no pathogens but also its level of contamination in mesophilic aerobic germs is lower than the microbiological criteria prescribed by CO-DINORM 2001. In sum, the microbiological quality of the banku is satisfactory. Other studies could follow on from this, such as setting up an HACCP system for production or using selected starters to help control the flavor and aroma of these products. Above all, producers need to be made aware of and trained in the basic rules of hygiene, the microbial hazards involved in poor hygiene practices, and the means of preventive and corrective action. We also need to train producers in Good Manufacturing Practices (GMP) and Good Hygiene Practices (GHP).

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