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Characterization of a craft beer produced from plantain fruits (*Musa paradisiaca* L.)

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Abstract

The general objective of this study is to evaluate the microbiological quality, physicochemical and organoleptic characteristics of plantain beer enriched with sorghum. The methodological approach consisted of producing four types of plantain beer by incorporating sorghum. Conventional methods were used to determine the physicochemical characteristics and microbial loads. The results indicate yeast loads ranging from 7.2 to 7.6 log (CFU/mL) and 7.2 to 7.4 log (CFU/mL) respectively for the musts of plantain paste incorporated with sorghum and those without sorghum. The physicochemical characteristics presented pH values of 3.46 and 3.40 to 3.45, titratable acidity of 1.05% and 1.09 to 12.4%, dry extract refractometric of 0.52° Brix and 0.58 to 0.87° Brix, alcohol content of 2.80% and 3.31 to 6.23% respectively for the control beer and for the improved beers. Various organic acids, notably citric, acetic, lactic, fumaric, tanic and tartaric acids were determined with increasing contents depending on the level of sorghum incorporated. The loads of aerobic mesophilic germs (1.5±0.77 to 5.3±1.88 × 10 CFU/mL) and yeasts and molds (1.1±0.56 to 4.6±1.35 × 10 CFU/mL) obtained are below the threshold limit of microbiological criteria. No loads of total coliforms, *E. coli*, *S. aureus* and *Salmonella* were detected. Beers improved with 5%, 10% and 15% malted sorghum concentrations produced were of acceptable microbiological quality in relation to the limit threshold of microbiological criteria and their consumption could not present a risk for consumers.

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Introduction

Plantain (*Musa paradisiaca*) is a major food crop in humid tropical regions belonging to the genus Musa and the family Musaceae. It is the third tropical fruit crop in terms of tonnage with global production estimated at more than 44 million tonnes over an area of 6.7 million hectares (FAOSTAT, 2022). Considered the first food crop in Ivorian forest areas and the third food crop in Côte d'Ivoire, plantain has a experienced significant growth in recent years with production estimated at more than 1.8 million tonnes (FAOSTAT, 2022).

In Côte d'Ivoire, plantain is widely consumed and plays a very important nutritional and socioeconomic (Amoutchi *et al.,* 2020). Local consumption of plantain is estimated at 120 kg per inhabitant over a year in dried, boiled, roasted and fried form divided into several local dishes including foutou, aloco or frying of the cut ripe pulp, claclo or frying dough obtained at the advanced ripening stage, chips, cakes and in combination with other basic foods (Kouamé *et al.,* 2015). It is an energetic food that provides 120 kcal per 100 g and contributes approximately 70% of dietary energy availability in the world (Emaga *et al.,* 2007). Plantain is an important source of food and nutritional security for most farmers in sub-Saharan Africa (Ayeha *et al.,* 2023).

Despite food self-sufficiency in plantains in Ivory Coast, a real food security problem exists. It is a highly perishable fruit and vulnerable to postharvest spoilage due to their short shelf life under ambient conditions (Kikulwe *et al.,* 2018). Advanced ripening significantly loses its economic value (Ayeha *et al.,* 2023). During periods of abundance, post-harvest losses can reach 80% (Kitinoja *et al.,* 2018) in certain regions. These losses are mainly linked to poor conditions of harvest, transport, storage, post-harvest maturation, packaging and high temperature before reaching markets and then consumers (Swain *et al.,* 2017). If plantain are used in countries like India, the Philippines, China, Brazil and Togo for industrial processing and even some African countries including Uganda, Burundi, Cameroon, the Republic Democratic Republic of Congo and Madagascar to produce wine or beer (Malomo *et al.,* 2023) in order to promote them, it is not the case for Côte d'Ivoire where production is exclusively limited to traditional processing for food use. With the aim of contributing to the reduction of post-harvest losses of plantain in Côte d'Ivoire, the general objective of this study is to produce plantain beer by developing new products and characterizing its physico-chemical, functional and organoleptic potential.

Materials and methods

Study material

The study material consists of plantain fruits (*Musa paradisiaca* L.) of the four-month-old horn variety (Fig. 1A, 1B) and red sorghum grains (Fig. 1C).

A B C

Fig. 1. Photographs of mature green plantain fruits (A) and 21 day senescing (B) and red sorghum grains (C)

Biological material

The biological material was *Saccharomyces cerevisiae* yeast purchased in a supermarket in the commune of Cocody and used for the fermentation of plantain must.

Collection of plantain samples and sorghum grains

Plantain samples mature green of approximately 7 kg each of the four-month-old horn variety were collected in the markets of three communes of Abidjan, notably Abobo, Adjamé and Yopougon. At each of the sites, three samples were collected, giving a total of nine samples for all three sites. All collected samples were labeled, placed in hermetically sealed plastic packaging and then transported to the laboratory.

At the same time, four samples of red sorghum grains of 1 kg each were purchased at the Cocody market in

the commune of Abidjan and sent to the laboratory in order to follow the malting process (Fig. 2).

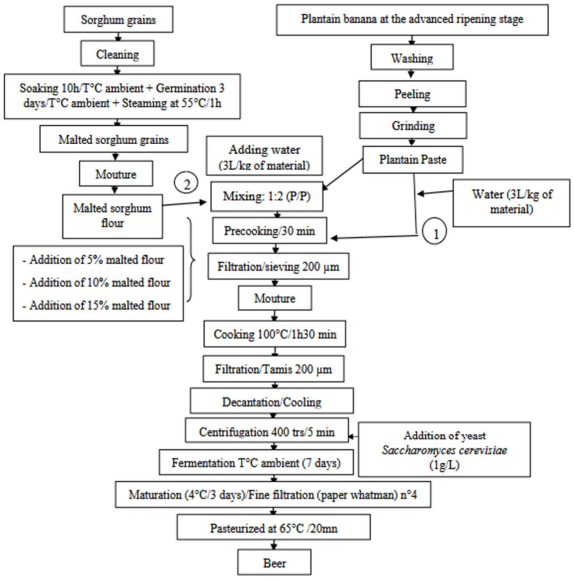

Fig. 2. Plantain beer production diagram
1- Process for producing control beer or pure plantain beer; 2- Process for producing improved beer or associated plantain-sorghum beer.

Production of plantain beer

The production of plantain musts was carried out according to the method of Ourega *et al.* (2016) with some modifications, namely the pure plantain method for the production of control beer and the plantain-sorghum method with different concentrations of malted sorghum for the production of improved beer (Fig. 2).

In the pure plantain method, plantain fruits collected at the mature green stage were stored in sealed plastic packages for 21 days where they reached a very advanced ripening stage (senescence phase). Then, the fruits were washed, peeled, weighed and crushed using an electric blender (Laboratory blender, Model 38BL 40, USA) to obtain a paste. A quantity of 1 kg of plantain paste obtained was taken and then 3 L of distilled water were added. After homogenization, a fraction of approximately 2 L of the mixture was precooked for 30 min at a temperature of 55°C then filtered to obtain pure plantain must. The must obtained is cooked at 100°C for 1 hour 30 minutes and is filtered again. After cooling and decanting, the must was centrifuged at 4000 rpm for 5 min. After centrifugation, the must was inoculated with the yeast *Saccharomyces cerevisiae* in the proportion of 1 g per liter of must and the mixture was subjected to fermentation for 7 days at room temperature in the laboratory. After the alcoholic fermentation phase, the fermented must was matured at 4°C for 3 days, then filtered on Whatman N°4 paper to obtain the pure plantain beer or control beer.

In the plantain-sorghum method at different concentrations of sorghum (5%, 10% and 15%), the must obtained from the plantain-sorghum mixture was used for the production of improved beers under the same conditions as the control beer.

In total, four types of beers were produced from plantain fruits, water, sorghum flour and *Saccharomyces cerevisiae* yeast (Table 1). The beers produced were used for physicochemical, phytochemical and microbiological analyses. Subsequently, the different beer samples were pasteurized at 65°C/20 minutes for sensory analyses.

Study of yeast growth during the fermentation of plantain musts

Yeast growth was determined according to the method of Biesta-Peters *et al.* (2010) carried out in the different plantain must solutions prepared from the production diagram in Fig. 2.

Table 1. Experimental device for the production of plantain beer

Quantities of ingredients	Plantain beer			
	BT	B5%	B10%	B15%
Plantain paste (g)	1000	950	900	850
Distilled water (L)	3	3	3	3
Sorghum flour (g)	0	50	100	150
Yeast (g)	1 g/l	1 g/l	1 g/l	1 g/l

BT: Control beer without sorghum flour; B5%: Beer improved with 5% malted sorghum flour; B10%: Beer improved with 10% malted sorghum flour; B15%: Beer improved with 15% malted sorghum flour.

The plantain must solutions were then inoculated with the baker's yeast *Saccharomyces cerevisiae* at a rate of 1 g/L and fermented at room temperature in the laboratory for 7 days, or 168 hours. The optical density (OD) of each fermented must solution was determined with a spectrophotometer (BK-UV1000, China) at 600 nm at 24-hour intervals against a blank consisting of the uninoculated must. Subsequently, 1 mL of each culture was taken to carry out successive dilutions of which aliquots of 0.1 mL of each dilution were spread on YPDA agar, and then the Petri dishes were incubated at 30°C for 72 hours in an incubator (BJPX-B80II, China). After incubation, all characteristic yeast colonies (white, creamy, ovoid, smooth and shiny) were counted by eye, then the microbial loads were calculated and the results were expressed in UFC/mL (Unit Forming Colony per milliliter) using the following formula determined.

Physico-chemical analyzes of plantain beers

Determination of pH and titratable acidity

The pH was determined following the method of Le Coq (1965). To do this, the electrode of a pH meter (P604, Consort, France) was introduced directly into 10 mL of sample and the pH reading was read from the value displayed on the pHmeter screen.

As for titratable acidity, 10 mL of the sample was titrated with a sodium hydroxide solution of normality 0.1 N until it turned pale pink after the addition of 2 to 3 drops of 1% phenolphthalein. The results obtained constitute the average of three tests and the titratable acidity rate was expressed as a percentage of lactic acid calculated using the following formula below (Amoa-Awua *et al.*, 2007).

$$\% \text{ Titratable acidity} = \frac{\text{Vol}_{\text{NaOH}} \times \text{N}_{\text{NaOH}} \times 0{,}09 \times 100}{\text{PE}}$$

Vol $_{NaOH}$: volume of sodium hydroxide solution,

N_{NaOH} : Normality of sodium hydroxide solution

PE: Test sample (10 mL)

0,09: milliequivalent gram of lactic acid

Determination of refractometric dry extract

The refractometric dry extract was determined by the AOAC (1995) method. A drop of the plantain beer samples was placed on the glass of the pocket refractometer (Model Atago pocket refractometer) to evaluate the quantity of suspended matter. The value of the refractometric dry extract was read under light, at the level of the eyepiece of the device. Three tests are carried out for each sample.

Determination of alcohol level

The alcohol content of the beer was determined according to the method described by OBI *et al.* (2015) where the difference in measurement between the density before and after fermentation was used. The percentage of alcohol per volume of beer was determined from the following formula:

$$\% \text{ alcohol} = \frac{[1{,}05 \times (D_i - D_f)/D_f \times 100]}{0{,}79}$$

D_i : Initial density

D_f : Final density

1,05 : Mass of alcohol per 1 g of CO_2 released

0,79 : Density of ethanol

Determination of organic acids

The method used for the determination of organic acids was that described by Coulibaly *et al.* (2021). The samples (10 mL) were first centrifuged (13,000 rpm for 3 min) and the supernatants were filtered using a 0.45 µm millipore filter (Sartorius AG, Goëttingen, Germany). Organic acids (citric, tartaric, lactic, fumaric, acetic and tanic acids) were separated and quantified by high-performance liquid chromatography (HPLC). The device is equipped with a diode array UV detector (Agilent Technologies, 1200 series) and an Aminex HPX 87H column (300 mm x 7.8 mm, Bio-Rad, France) coupled to a refractometer (Agilent Technologies , 1200 series). The oven temperature was set at 50°C. The eluent was 5 mM sulfuric acid at an elution rate of 0.5 mL/min and the UV detector was set at the wavelength of 210 nm. A volume of 20 µL of the samples was injected and the analysis lasted 35 min. Calibration is carried out using solutions containing the different compounds to be analyzed in the linearity ranges of each product. Data acquisition and processing are

carried out using Chromoleon software ©Dionex 1996-2001 version 6.70 SP2a Build 1871.

Microbiological analyzes in beers produced

Enumeration of aerobic mesophilic germs (NF ISO 4833-1: 2013)

The aerobic mesophilic germs were cultured aseptically in the mass of Plate Count Agar (PCA) at 30°C. To do this, 1 mL of each decimal dilution was dispersed in two Petri dishes. The PCA agar, sterilized (121°C/15 mins) and cooled to 45°C in a water bath, was transferred to each of these boxes. After homogenization and solidification of the agar, a second layer of PCA agar was poured on top of the first. The Petri dishes were then incubated at 30°C for 72 hours. The valid count of colonies enumerated is 30 to 300.

Enumeration of E. coli and coliforms (ISO 4832: 2006)

E. coli chromogenic agar was used for the detection and enumeration of *E. coli* and coliforms. Inoculation was carried out by incorporating 1 mL of the stock suspension or successive decimal dilutions into the mass. Then, 12 to 15 mL of the VRBL agar supercooled at 45°C was poured into Petri dishes. The mixture was gently homogenized by rotating movements. After solidification, a second layer of approximately 5 mL of agar was made. Finally, the Petri dishes were incubated for 24 hours at 37°C for total coliforms and 44°C for *E. coli*. Likely characteristic colonies will be dark blue to purple in color for *E. coli* and pink to red for other coliforms. The valid count of colonies enumerated is 15 to 150.

Enumeration of yeasts and molds (NF V08-059: 2002)

The enumeration of the fungal flora was carried out after the aseptic distribution of 0.1 mL of each of the respective dilutions into Petri dishes containing the previously prepared Sabouraud Chloramphenicol agar. Seeding was done by spreading 0.1 mL of the stock suspension or dilutions on the surface of the solidified agar in sterile Petri dishes. Subsequently, the Petri dishes were incubated at 30°C for 24 to 72 hours. Yeast colonies appear whitish, smooth, rounded with a bakery odor and a diameter of 0.5 to 2 mm after 24 to 48 hours. The molds were filamentous, fluffy, cottony, powdery and granular, sometimes invasive or not after 72 hours. The valid count of colonies enumerated is 15 to 150.

Enumeration of Staphylococcus aureus (ISO 6888-1: 2021)

Baird Parker agar was the medium used for the detection and enumeration of *Staphylococcus aureus*. Seeding was done by spreading 0.1 mL of the stock suspension or dilutions on the surface of the solidified agar in sterile Petri dishes. The Petri dishes were incubated at 30°C for 24 hours. *Staphylococcus aureus* forms black colonies with a light halo around the colonies. The valid count of colonies enumerated is 15 to 150.

Salmonella enumeration

The enumeration and detection of *Salmonella* was carried out according to the method of Hendriksen (2003) in several stages. First, pre-enrichment in non-selective medium, followed by enrichment in selective medium and culture on selective agar. To carry out pre-enrichment in non-selective medium, 25 mL of beer were homogenized in 225 mL of previously sterilized buffered peptone water then incubated at 37°C for 24 hours. Then, 1 mL of the pre-enriched culture was transferred using a sterile pipette into 10 mL of previously prepared sterile Vassiliadis Rappaport broth. The whole was incubated at 37°C for 24 hours. The characteristic colonies with regular outlines, green with bluish shades and/or not with black centers on Hektoen which will be observed, will be taken to be re-inoculated on nutrient agar in order to obtain pure cultures. The enrichment culture will be streaked onto Salmonella-Shigella (SS) agar and incubated at 37°C for 24 hours. Presumptive *Salmonella* colonies that are opaque and translucent with a black center (H_2S positive) will be identified for confirmation.

Determination of the microbiological quality of beer

The plan for interpreting the microbiological quality of the beer obtained is the three-class plan for assessing the microbiological quality of food (Regulation CE, 2022).

Statistical analysis

Statistical analysis of the data was carried out using R software (R i386 3.1.2). It made it possible to calculate the means and standard deviations of the data obtained. The different parameters analyzed were then subjected to an analysis of variance (ANOVA). For this purpose, repeated measures ANOVA and Duncan's multi-range tests were used. In the event of a significant difference between the parameters studied, the classification of the means was done according to the Duncan test. The significance level is $\alpha = 0.05$. Statistical differences with a probability value less than 0.05 ($P<0.05$) are considered significant. When the probability is greater than 0.05 ($P>0.05$), the statistical differences are not significant.

Results and discussion

Growth kinetics during the fermentation of plantain musts

At the level of the pure plantain must for obtaining the control beer (BT), the curve presents an exponential phase from 0 to 48 hours of fermentation with average loads of 7.2 to 7.4 log (UFC/ mL). As for the other musts incorporated with 5% (B5%), 10% (B10%) and 15% (B15%) of malted sorghum flour, the curves each present an exponential phase of growth from 0 to 72 hours of fermentation with average loads ranging from 7.2 to 7.5 log (UFC/mL) similarly for the B5% and B10% and from 7.2 to 7.6 log (UFC/mL) for B15% (Fig. 3). The fermentation kinetics of different plantain musts is linked to the yeast growth cycle which allows the transformation of sugars contained in the must. The high yeast loads recorded in sorghum musts incorporated for 72 hours corroborate those of the work of Boli *et al.* (2023) who reported exponential phases of yeast growth during the first 72 hours of fermentation. These results could be due to sorghum which could hydrolyze the starch contained in the musts. Indeed, according to Ciocan *et al.* (2023), sorghum grains contain amylase fractions involved in starch hydrolysis during brewing. These results could also be due to hydrolytic enzymes synthesized by certain microorganisms in the must and which are capable of hydrolyzing fermentable sugars, notably sucrose, into glucose and fructose used by yeasts during energy metabolism to produce alcohol and carbon dioxide as reported by Rasbold *et al.* (2021) and Osiebe *et al.* (2023).

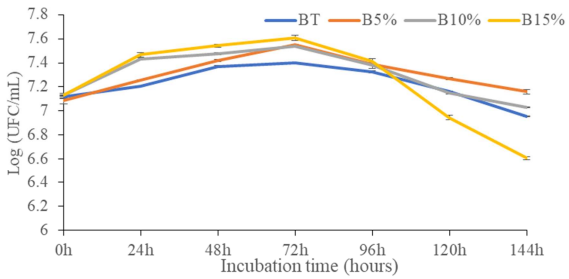

Fig. 3. Kinetics of yeast growth during the alcoholic fermentation of plantain musts

Each of the different growth phases is followed by a stationary phase at 72 hours and a decay phase until the end of fermentation with an average load of 6.9 log (CFU/mL) for the control Each of the different growth phases is followed by a stationary phase at 72 hours and a decay phase until the end of fermentation with an average load of 6.9 log (CFU/mL) for the control and of 7.2 log (UFC/mL) for B5%, 7. 0 log (UFC/mL) for B10% and 6.6 log (UFC/mL) for B15% for the beers ameliored. The decay phase observed after 72 hours of fermentation could be due to chemical and physical modifications of the must. Indeed, chemical and physical modifications of the environment create conditions not favorable to complete fermentation, in particular the alcohol itself which, at a certain concentration in the fermenting must, could systematically attack and inhibit the yeasts which origin of its production (De Cliff and Ndayiragije, 2013). Furthermore, this result could also be explained by the production of inhibitors by the yeast itself in the must during fermentation. Indeed, Cisilotto *et al.* (2023) reported that it

sometimes happens that yeasts release into the fermenting must compounds that are toxic to it, notably ethanol, carbon dioxide and medium-chain saturated fatty acids which can exert a negative influence and cause a significant loss of vitality and viability on yeasts in the must. Verdenal *et al.* (2023) also reported that factors such as insufficient assimilable nitrogen in the must could explain a significant risk of reduction or cessation of alcoholic fermentation.

Physico-chemical characteristics of plantain beers

Table 2 presents the physico-chemical characteristics of plantain beers of the different beers produced. All beers produced have an acid pH varying from 3.4 to 3.46. The highest value (3.46) was obtained with the control beer while the lowest value (3.4) was obtained in the beer improved with 15% malted sorghum flour. However, no significant difference (P>0.05) was observed between the pH of the different beer samples. Regarding titratable acidity, the values vary from 1.05 to 1.24%. The highest value (1.24%) was found in the beer improved with 15% malted sorghum flour while the smallest value (1.05%) was obtained in the control beer without the addition of sorghum flour. However, no significant difference (P>0.05) was observed between the titratable acidity of the beer samples. The results of this study showed that the pH of plantain beer is acidic. Our results corroborate those of Ourega *et al.* (2016) who reported acidic pH values during the production of plantain beers. This acidification would be linked to the activity of microorganisms during fermentation. Indeed, according to Tran *et al.* (2022), yeasts acidify their environment through the fermentation of metabolized sugars, notably sucrose converted into glucose and fructose for the production of organic acids, which contributes to lowering the pH of the environment. Beers improved with sorghum have a relatively acidic pH than that of the control beer without sorghum. This result confirms the work of Ourega *et al.* (2016) who reported acidic pH values in plantain beer.

Table 2. Organic acid contents of different types of plantain beer

Plantain beers	Physico-chemical characteristics			
	pH	Titratable acidity (%)	ESR (°Brix)	Alcohol (%)
BT	3,46±0,1^{a}	1,05±0,02^{a}	0,52±0,10^{b}	2,80±0,1^{b}
B5%	3,45±0,2^{a}	1,09±0,08^{a}	0,58± 0,12^{b}	3,31±0,4^{b}
B10%	3,42±0,1^{a}	1,15±0,04^{a}	0,78±0,10^{a}	5,07±0,3^{a}
B15%	3,40±0,1^{a}	1,24±0,0^{a}	0,87±0,10^{a}	6,23±0,6^{a}

In the same line, values bearing the same letters are not significantly different at the 5% threshold. BT: Control beer without sorghum flour; B5%: Beer improved with 5% malted sorghum flour; B10%: Beer improved with 10% malted sorghum flour; B15%: Beer improved with 15% malted sorghum flour.

At the refractometric dry extract level; the soluble sugar content varies from 0.52±0.24 to 0.87±0.37° Brix. The highest soluble sugar content (0.87±0.37° Brix) was obtained in the control beer, whereas the lowest (0.52±0.24° Brix) was obtained in the control beer without addition of malted sorghum flour. However, a significant difference (P<0.05) was observed between the refractometric dry extract of the beer samples. Sugars constitute the main source of energy for the metabolism of yeasts introduced during fermentation. The high sugar content of improved beers could be due to the sorghum incorporated into their preparation process. Indeed, Ciocan *et al.* (2023) indicated that the starch of sorghum grains leads to a high production of sugars during the enzymatic activity responsible for the hydrolysis of fermentable sugars including sucrose into glucose and fructose during brewing.

The alcohol content of the different beers produced varies from 2.8% to 6.23%. The highest rate (6.23%) was obtained in beer improved with 15% malted sorghum flour, followed by 5.07% and 3.31% in beers improved respectively with 10% and 5% malted sorghum flour. The lowest alcohol level (2.8%) was obtained with the control beer without sorghum.

However, a significant difference (P<0.05) was observed between the alcohol levels of the different beers. Beers improved with sorghum have the highest alcohol levels compared to the control beer without sorghum. These results could be due to the high sugar content in the improved beers which would be used much more by the yeasts for the production of alcohol.

Our results confirm those of Yao *et al.* (2021) who indicated that they must containing more sugar contains the highest alcohol level. Furthermore, Canonico *et al.* (2019) and Coulibaly *et al.* (2021) reported the degradation of fermentable sugars (glucose and fructose) metabolized by *Saccharomyces* yeasts for high alcohol production. The alcohol levels of improved beers obtained (3.81 to 6.23%) in this study are within the range of alcohol levels (2 to 8%) of most commercialized industrial beers (De Cliff and Ndayiragije, 2013). The alcohol levels obtained in this study made it possible to classify the beers produced into three categories, namely light beer, normal beer and strong beer. This complies with article B.02.130-B.02.135 of the food and drug regulations concerning the names used for tasting beers, in particular extra-light beer at extra strong beer (RAD, 2023). Furthermore, our results are higher than those obtained (1.92 to 5.05%) in honey beer by Yao *et al.* (2021) in Côte d'Ivoire. This could be explained by the partial transformation of the sugars contained in the must. Indeed, according to De Cliff and Ndayiragije (2013), the difference in the alcohol content of beer is due to the partial transformation of sugars into alcohol and carbon dioxide (CO_2) inducing the presence of non-fermentable sugars in the starting wort which ends up in the beer.

Table 3. Organic acid contents of different types of plantain beer

Organic acids (mg/L)	Plantain beers			
	BT	B5%	B10%	B15%
Citric acid	0,80±0,12^{b}	2,13±0,04^{a}	2,16±0,05^{a}	2,26±0,02^{a}
Acetic acid	0,13±0,02^{b}	1,97±0,01^{a}	1,97± 0,01^{a}	1,97±0,01^{a}
Lactic acid	0,30±0,04^{a}	Nd	Nd	Nd
Fumaric acid	1,23±0,12^{a}	1,15±0,00^{a}	0,33±0,07^{b}	0,32±0,09^{b}
Tanic acid	10,44±0,1^{d}	17,78±0,46^{c}	37,84±0,15^{b}	63,43±0,71^{a}
Tartaric acid	Nd	82,17±0,3^{c}	138,42±0,4^{b}	425,27±0,2^{a}

In the same line, values bearing the same letters are not significantly different at the 5% threshold. BT: Control beer without sorghum flour; B5%: Beer improved with 5% malted sorghum flour; B10%: Beer improved with 10% malted sorghum flour; B15%: Beer improved with 15% malted sorghum flour. nd: not detected.

Organic acid contents of different of beer

Table 3 indicates the content of the different organic acids present in the different types of beer. The organic acids detected during this study were citric, acetic, lactic, fumaric, tanic and tartaric acids. Tartaric acid was the major organic acid in the beers produced followed by tanic acid, citric acid, acetic acid, fumaric acid and lactic acid. The highest tartaric acid content (425.27±0.2 mg/L) was obtained in B15% beer, followed by 138.42±0.4 mg/L in B10% beer and 82.17 ±0.3 mg/L in B10% beer. No traces were detected in BT beer.

However, a significant difference (P<0.05) was observed between the different tartaric acid contents of the different beers. The high rate of tartic acid in this study could be due to the fact that this acid constitutes the base of the organic acids of plantain beer and would be the major acid (Ourega *et al.,* 2016). The highest tanic acid content (63.43±0.71 mg/L) was obtained in B15% beer, followed by 37.84±0.15 mg/L in B10% beer, 17.78 ±0.46 mg/L in B10% beer and 10.44±0.01 mg/L in BT beer. The highest citric acid content (2.26±0.02 mg/L) was obtained with B15% beer followed by 2.16±0.05 mg/L in B10% beer and 2.13± 0.04 mg/L in B5% beer. The lowest citric acid content (0.80±0.12 mg/L) was obtained with the control beer BT. In terms of acetic acid, the highest content (1.97± 0.00 mg/L) was practically similar in all samples of improved beers,

notably B15%, B10% and B5%, unlike the control beer. The lowest content (0.13± 0.02 mg/L) was observed in Control beer without sorghum flour (BT). A significant difference ($P<0.05$) was observed between the citric and acetic acid contents of the control BT beer and the improved beers (B5%, B10% and B15%).

As for fumaric acid, the highest content (1.23±0.12 mg/L) was obtained with BT beer, followed by B5% beer (1.15±0.00 mg/L), B10% beer (0.33±0.07 mg/L) and B15% beer (0.32±0.09 mg/L). Beer acidity is caused by excessively low pH due to high levels of organic acids in beer. The contents of the different organic acids studied, notably citric acid, acetic acid, fumaric acid and tanic acid, were higher in the improved beers than in the control beers. This could be due to the sorghum added during the production of the improved beers. Indeed, according to Kim *et al.* (2021), those carbohydrates are essential nutrition for the survival of microorganisms for the production of organic acids. These results could also be explained by the high soluble sugar content in improved beers. Indeed, Li *et al.* (2017) and Su *et al.* (2019) reported that sugars are degraded for the production of organic acids as final metabolic products. Lactic acid was detected only in BT beer (0.3± 0.04 mg/L) while it was not detected in the different improved beers. This result could be due to the fact that the lactic acid bacteria responsible for the production of lactic acids would be inhibited during the production processes of plantain must in the presence of sorghum. A significant difference ($P<0.05$) was observed between the fumaric and lactic acid contents of the different beers.

Table 4. Microbial loads of different plantain beers

Plantain beers	Microbial loads (CFU/mL)					
	GAM	C.T.	*E. coli*	L.M.	*S. aureus*	*Salmonella*
BT	$(5,3±1,88)10^{a}$	00	00	$(4,6±1,35)10^{a}$	00	ND
B5%	$(2,6±1,63)10^{b}$	00	00	$(1,6±0,71)10^{b}$	00	ND
B10%	$(2,1±1,09)10^{bc}$	00	00	$(1,4±0,14)10^{b}$	00	ND
B15%	$(1,5±0,77)10^{c}$	00	00	$(1,1±0,56)10^{b}$	00	ND
Standard*	$< 10^{3}$	< 10	< 10	$< 10^{2}$	< 10	ND/25mL

In the same column, the averages of loads marked with the same letter are not statistically different at the 5% threshold. BT: Control beer without sorghum flour; B5%: Beer improved with 5% malted sorghum flour; B10%: Beer improved with 10% malted sorghum flour; B15%: Beer improved with 15% malted sorghum flour; GAM: Mesophilic aerobic germs; C.T.: Total coliforms; L.M.: Yeasts and molds; ND: Not detected; CFU: Colony Forming Unit. *: EC Regulation, (2022).

Microbiological quality of plantain beers

Table 4 presents the microbiological quality of different plantain beers enriched with sorghum. Aerobic mesophilic germs and yeasts and molds were the only microorganisms isolated. The loads of aerobic mesophilic organisms vary from 1.5±0.77 × 10 CFU/mL to 5.3±1.88 × 10 CFU/mL and are significantly different ($P<0.05$) from one beer to another.

Regarding yeasts and molds, the loads vary from 1.1±0.56 to 4.6±1.35 × 10 CFU/mL Control beer has a higher yeast and mold content ($P<0.05$) than other beers. The microbiological quality of the different beers made it possible to detect the loads of aerobic mesophilic germs and yeasts and molds. These results are consistent with those of Suiker and Wösten (2022) who reported the presence of fungi, notably yeasts and molds, in beer and beer-based products. The presence of these microorganisms in the different beers produced would be normal given that their high loads have been reported during the fermentation of plantain must (Kambiré *et al.,* 2023). Furthermore, all the microbial loads detected are below the threshold limit of the microbiological criteria concerning aerobic mesophilic germs and yeasts and molds respectively 10^{3} CFU/mL and 10 CFU/mL for

vegetable drinks generally pasteurized at 70°C (Regulation EC, 2022). These low charges could be explained by pasteurization which is the last important step in beer production because we allows a significant reduction in microbial growth during beer production.

No loads of total coliforms, *E. coli, S. aureus* and *Salmonella* were detected. Loads of total coliforms, *E. coli*, *S. aureus* and *Salmonella* were not detected in any of the beers produced.

These results could be due to pasteurization or even heating the must to 100°C during the production stage. Indeed, according to Suiker and Wösten (2022), the heating step to 100°C during must production effectively inactivates the harmful microbes that have accumulated up to this step. The non-detection of certain germs studied in the beers produced could also be due to the beer itself. Indeed, Roselli *et al.* (2024) reported that alcohol increases heat by significantly inhibiting the microorganisms within it. These same authors indicated that beer resists the growth and survival of microorganisms. This is believed to be due to the combination of a series of intrinsic antimicrobial factors in beer including the presence of ethanol, bitter compounds, low pH, high CO_2 concentrations, low O_2 levels and a general lack of nutrients such as fermentable carbohydrates, vitamins and amino acids (Menz *et al.*, 2009).

Conclusion

This study, which is a contribution to the reduction of post-harvest losses of senescent plantain through the production of beer, gave a good yield in production must. Beers improved with 5%, 10% and 15% malted sorghum concentrations presented the highest contents of soluble sugars and alcohol. Tartaric acid was the major organic acid in the beers produced. The different beers produced are of acceptable microbiological quality in relation to the limit threshold of microbiological criteria and their consumption could not present a risk for consumers.

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