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Stabilization of the Traditional Sorghum Beer, "Tchoukoutou" using Rustic Wine-Making Method

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Abstract: Tchoukoutou is an opaque beer produced from red or brown sorghum. This beverage is consumed right after the manufacturing process otherwise the continual and uncontrolled fermentation turns it into an acidic drink unsuitable for consumption after few hours. The present study has combined the traditional brewing method, the rustic "champenoise" method of wine-making and heat treatment to produce a bottled and stabilized drink with a long shelf-life. The chemical analysis of the resulting product displayed pH 3.45±0.15 and an alcohol content of 3.28±0.40%. The sensory evaluation showed aspect and taste similar to those of the traditional brew. The microbiological analyses revealed that no germs survived the heat treatment. The product was kept for 6 months without any noticeable change of the organoleptic attributes.

Key words: Beverage, brasserie, cereal, champenoise method

INTRODUCTION

Sorghum bicolor (L.) Moench is the main cereal cultivated in African tropical countries. It constitutes 30% of the grains utilized and one of the staple grains in Africa. It is an ingredient in various dishes such as porridge, pudding, bread dough and semolina.

A part of Africa's yearly sorghum crop is allocated to opaque beer processing (Kayode *et al*., 2007). The sorghum beer is a refreshing drink well appreciated by a large number of consumers throughout Africa. This beer takes different designations according to the country of origin. In Western Africa: Burkina Faso, Mali and Ivory Coast it is known as Dolo. Ghanaians and Nigerians call it Pito and Burukutu or Otika respectively (Odunfa, 1985) In Togo, approximately 60% of the national production of sorghum is used to produce two kinds of sorghum brew: Tchakpalo and Tchoukoutou (OASID, 2004). The production and marketing of Sorghum beer remain women's activities from which they derive a substantial income.

The beer Tchoukoutou is sold and consumed in special cabarets that are popular bars and places of huge conviviality. Sorghum beer plays a fundamental socioeconomic role in northern Togo where it is abundantly served to maintain an atmosphere of merriment in all celebrations. This beer is also used in religious ceremonies, libations and traditional rituals.

The materials used for the beer processing remain rudimentary and are composed of large jars for mashing, cooking pots, and a basket of woven straws or a piece of cloth for filtering. The setting and processing conditions do not guarantee the sanitary safety of the product. The parameters of concentration, acidity, temperature, time, alcohol, and CO_2 content are not monitored during or after the manufacturing process, they are estimated by guesswork. This affects the consistency and safety of the product. The resulting product is alive and unstable, the organoleptic characteristics continue to change, and within 24 h it becomes too acidic to be consumed. The short shelf-life of this traditional brewing method limits its consumption to within a day of its production and hence its availability is limited to the seasonal availability of the sorghum grain. The present study was designed to combine the traditional brewing method and a rustic wine making technique for manufacturing stabilized Tchoukoutou beer with a long shelf-life without modifying the original organoleptic characteristics.

MATERIALS AND METHODS

Tchoukoutou brewing process: The *Sorghum bicolor* grains were selected and purchased in Lomé marketplace in October 2009 by five experienced women processors in Lomé market (Togo). In order to preserve the organoleptic properties required by the habitual

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consumers, the grains were processed into Tchoukoutou by the five women in their homes in accordance with the traditional practices. Briefly, the grains were soaked overnight at ambient temperature; afterwards the grains were germinated in open for 48 h before the grinding. The ground grains were mashed in water and boiled for approximately 2 h. The product was then filtered and soured overnight prior the 9 h boiling to obtain the wort. The wort was then fermented overnight by direct seeding with previous Tchoukoutou leaven.

Packaging and stabilization: For the packaging and the stabilization of tchoukoutou, the wort was prepared by the women as mention above. This wort was filtered and the brix was adjusted to 18-20 by boiling or diluting with water. Xanthan gum (E415) was added in variable proportions between 0.01-1% (m/v) in order to modify its viscosity and keep fine particles in suspension in the liquid. The wort was fermented using the champenoise method (Amerine and Singleton, 1977). Thus, the wort was seeded with 10% (v/v) leaven. The seeded wort underwent 2 steps fermentation. The first fermentation was assayed in entire batch for 20 h at ambient temperature. Afterwards, the fermenting wort was transferred into 33 centiliter bottles which were tightly sealed. The second fermentation was achieved in closed bottles at ambient temperature over 3 h. The bottles were then pasteurized in a water bath at 75-80ºC for 15 min in order to stop the fermentation. The time-temperature combination was determined after serial trials. The bottles were cooled after the heat treatment and were then ready for the consumption.

Chemical analyses: Chemical analyses were performed on the five samples of traditional beer and five samples of bottled beer. The samples were collected from sellers before the fermentation and after fermentation in the morning between 9 and 11 O'clock. At this period consumers claimed that the Tchoukoutou is supposed to preserve its optimal organoleptic characteristics. Samples of bottled beer were collected from five laboratory preparations. For each samples, the total dry matter of the wort was determined using a refractometer (Euromex, HC type 0-32, Holland). The attenuation limit accounting for the amount of sugar that has been transformed into alcohol was calculated by dividing the difference between the Brix values before and after fermentation on the brix of the wort before the fermentation.

The pH was measured with an electronic pH-meter (WTW type pH 330i) and the acidity was estimated by titration with a sodium carbonate 0.1 N using a phenolphthalein as indicator. The density and the alcohol contents were determined with by pycnometry (AOAC, 1984).

Shelf-life assessment: The bottles were divided into three lots that underwent the following treatments for a total period of 6 months. The first lot was exposed to sunlight; the second was stored in darkness and the third in a refrigerator. This allows estimating the delay time for which the product was expected to retain its optimum organoleptic properties. Samples were taken after 1, 2 and 6 months for evaluation of the sensory properties and the determination of the date of minimum freshness.

Microbiological analysis: All media used in this part were purchased from Biorad (France). Microbial quality control was assayed for traditionally brewed Tchoukoutou and six months stored bottled Tchoukoutou. The traditionally brewed Tchoukoutou samples were collected from the same women at the selling sites. For the microbiological assay, 10 mL samples were diluted in 90 mL sterile peptone. Serial decimal dilutions were performed from these solutions. Each dilution was used for the enumeration of the following microorganisms:

- Total aerobic bacteria with Plate Count Agar (PCA) after 24 h incubation at 30ºC (NF V08-051. 1999).
- Total coliforms and thermotolerant coliforms on Violet Red Bile Lactose (VRBL) after 24 h incubation at 30 and 44ºC, respectively (NF V08-050, 1999).
- Sulphite Reducing Bacteria (SRB) by Most Probably Number (MPN) with tryptone-sulfite neomycin broth after 20 h incubation at 44ºC (XP V08-061, 2005)
- Yeast and moulds with Sabouraud-Cloremphenicol after 3 to 5 days incubation at 30ºC (NF V08-059, 2002).
- The lactic and acetic acid bacteria on Man Rogosa Sharpe (MRS) at 30ºC for 24 to 72 h (ISO 15214, 1998)
- *Salmonella* spp.: Preenrichment with buffered peptone water at 37ºC for 24 h, enrichment with Rappaport Vassiliadis soya broth at 37ºC for 24 h and isolation on, Hektoen and *Salmonella shigella* agar at 37ºC for 24 h (NF V08-052, 1997)

Microbial quality of samples was assayed using the critical limits of French Association of Normalisation (AFNOR). Catalase test was performed by adding hydrogen peroxide on a single colony. The presence of catalase was revealed by the formation of gas bubbles. Oxidase tests were performed using available commercial oxidase disks purchased from Bio Rad (France).

Sensory assessment: A panel of 80 habitual consumers completed sensory evaluation of bottled beer freshly produced and beer preserved over 1, 3 and 6 months. The test method used was the attribute rate/scaling (Stone and Sidel, 2004) and the characteristics involved were color,

acidic taste, flavor, foam stability, viscosity, appearance and overall acceptability.

Statistical analysis: Experiments were conducted on five samples and results of chemical characteristics values were expressed as mean with standard deviation. Means were compared using epi-info software version 6. Statistical significance was set at $p<0.05$.

RESULTS AND DISCUSSION

Packaging and conservation are the main problems encountered by processors of local beers in Africa. The present study aimed to bottle Tchoukoutou the traditional sorghum-based beer after double fermentation process. The analysis of the flowchart showed that the manufacturing process of Tchoukoutou is an empirical processing method; however it is fairly close to the current industrial processing of the modern barley-based beer in its main phases such as steeping, malting and brewing. However removing rootlets of the germinated grains after malting, and clarification of the brew by the removing the yeasts and solid particles at the end of the controlled fermentation, are not required in Tchoukoutou processing. The latter is also similar to the brewing process of cloudy wort practiced in the North of France and comprising an infusion mashing and a decoction mashing (Perisse *et al*., 1959).

The double fermentation used in the present study allowed producing alcohol and saturating the brew with CO_2. The first fermentation conducted in the conditions similar to those of the traditional ones resulted in the traditional Tchoukoutou, a red or brown cloudy liquid containing yeasts and other tiny solids in suspension. Since the fermentation took place in an open container at ambient temperature, the carbon dioxide escaped resulting in a beverage not saturated with CO_2 and flat or not fizzy. The second fermentation achieved in sealed bottles trapped the carbon dioxide in the product, producing the desired bubbly carbonated drink. This step must be closely monitored to prevent an overproduction of CO_2 and a resulting breakage of bottles in the pasteurization step. Indeed serial tests allowed us to set the fermentation at ambient temperature for 3 h.

The effect of the xanthan gum used to maintain solid particles in suspension and limit the buildup of sediment was partially satisfactory at a rate of 0.03% (m/v). However it was noted that this additive contributed to the improvement of the foam stability as pointed out by Runkel (1976). As a result an effervescent and bubbling beer with an appearance similar to the traditional Tchoukoutou was obtained.

The results of the chemical characteristics of the traditional and bottled Tchoukoutou are presented in the Table 1. The recorded pH were 3.50±0.65 and 3.45±0.15 for the traditional beer and the bottled beer, respectively. These two pH were not statistically different ($p = 0.902$), similarly there was no statistical significance between the two beers in term of acidity and density (Table 1). However, according to the acidity and the pH, the two types of beer were sufficiently acidic and this characteristic would contribute to guarantee their safety. Contrary to the pH of traditional *Tchoukoutou* which is often variable due to fermentation by contaminating acidic bacteria, that of the bottled drink remained steady during storage. The ethanol content in the samples of *Tchoukoutou* were statistically different ($p = 0.047$), indeed the traditional *Tchoukoutou* (4.13%) was more alcoholic than the bottled (3.28%). Similarly the attenuation limit of the traditional beer was grater than the bottled one ($p = 0.01$). According to the value of alcohol content *Tchoukoutou* remains in the range of 2-5%, characteristic of drinks assigned to the class of beers. However, the recorded amount is slightly higher than those recorded for sorghum beer (2-3%) in other studies (Agu and Palmer, 1998; Briggs *et al*., 2004).

Table 1: Chemical characteristics of the two types of T*choukoutou*

Chemical characteristics	Traditional *Tchoukoutou*	Bottled *Tchoukoutou*
pH	3.50 ± 0.65	3.45 ± 0.15
Acidity (g/L)	1.94 ± 0.06	1.94 ± 0.06
Density	1.07 ± 0.00	1.07 ± 0.00
Alcohol contents (% vol.)	4.13 ± 0.34	3.28 ± 0.40
Attenuation limit	28.65 ± 2.10	26.50 ± 1.80

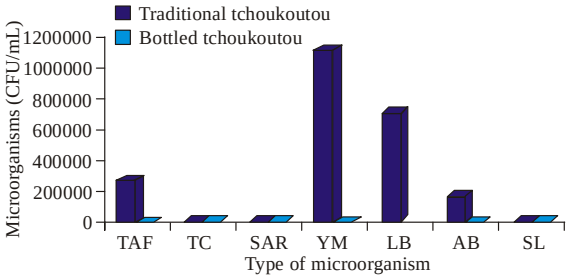

Fig.1: Identified microorganisms in the two types of *Tchoukoutou* TAF: Total aerobic flora; TC: Thermophilic coliforms; SRB: sulfite-reducing anaerobes; YM: Yeasts and molds; LB: lactic bacteria; AB: Acetic bacteria; SL: *Salmonella*

Microbiological analyses results (Fig. 1) showed that the bottled Tchoukoutou did not contain any microbes after pasteurization proving that the thermal treatment was sufficient to destroy the fermenting microorganisms. On the other hand, the traditional beverage in which the fermenting microbes were still alive was loaded with yeasts, molds, lactic and acetic bacteria. Fortunately it was observed that pathogenic and poisoning microbes (*Salmonella* and sulfite reducing bacteria) and food sanitary indicator microorganisms (coliform) were absent.

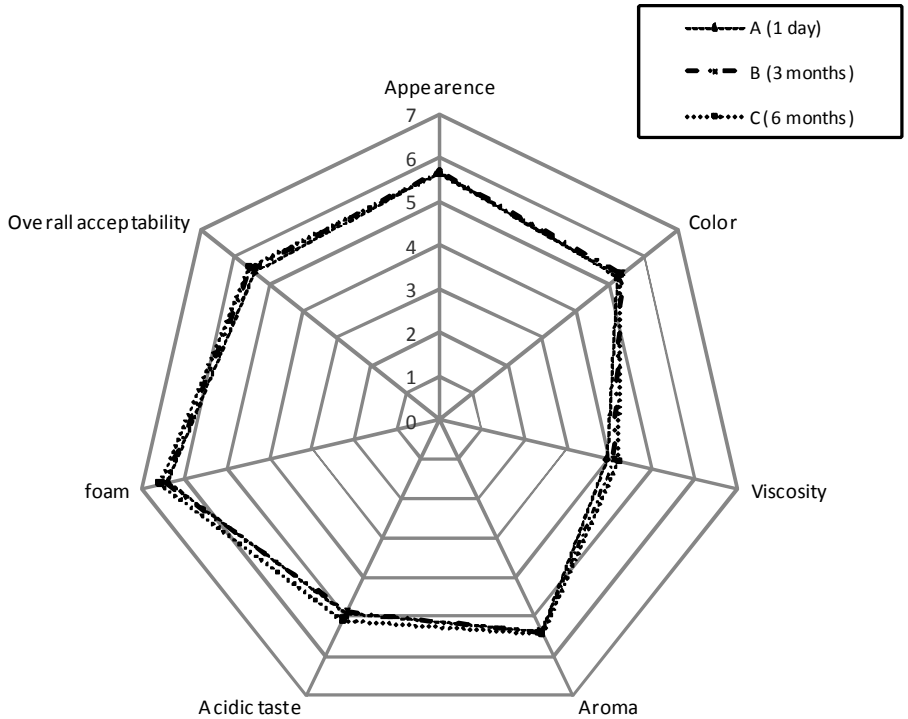

Fig. 2: Diagram of average ranking scores (on 9 points basis) for organoleptic attributes of the bottled drink: Freshly made and stored for 3 or 6 months

It can be inferred from these results that the spontaneous and non controlled fermentation of the traditional beer is achieved by a mixture of yeasts associated with lactic and acetic bacteria. The most representative microorganisms were yeasts, lactic, and acetic bacteria. These results are in accordance with those of previous works indicating that the distribution of the microorganisms isolated from the African troubled beers depended on the conditions of brewing and the source of the brewing ingredients (Ekundayo, 1969; Faparusi *et al*., 1973; Nout, 1980; Odunfa, 1985; Sanni and Lönner, 1993; Sefa-Dedeh *et al*., 1999). According to Konlani *et al*. (1996), *Saccharomyces cerevisiae* represented about 55-90% of the total yeasts in the beer of sorghum in Togo and in Burkina Faso.

Samples exposed to sunlight at ambient temperatures presented slight color fading with a buildup of a sedimentary deposit. On the other hand, the refrigerated samples and those stored in the shade did not show any changes apart from the sediment. Of the last two types of samples aforementioned, the preference was for the bottles kept in the refrigerator. The designed tchoukoutou products that underwent a sensory assessment (Fig. 2) at different stages of storage: freshly made, stored for 3 or 6 months had slight differences with regard to all the organoleptic attributes considered, but these differences were not significant ($p>0.05$). It can be inferred that the bottled and stabilized tchoukoutou could be kept up to 6 months without profound changes of the organoleptic attributes. By using the double fermentation technique of the rustic champenoise method of wine-making and pasteurization, we produced a consumer accepted bottled tchoukoutou with a long shelf-life. On the whole, this work defines the conditions for a standardized production of bottled tchoukoutou exhibiting an improved image of modernity by its presentation in bottles, its convenience of consumption and its long shelf-life. This improvement of the traditional processing method which resulted in a modernized product is likely to contribute to the development and expansion of this local drink.

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