

Evaluation of the Nutritional and Microbiological Quality of Kunun (A Cereal Based Non-Alcoholic Beverage) in Rivers State, Nigeria

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Citation

E Essien, C Monago, E Etor. *Evaluation of the Nutritional and Microbiological Quality of Kunun (A Cereal Based Non-Alcoholic Beverage) in Rivers State, Nigeria*. The Internet Journal of Nutrition and Wellness. 2009 Volume 10 Number 2.

Abstract

Evaluation of the nutritional and microbiological quality of laboratory processed and hawked kunun drinks (A Cereal Based Non-Alcoholic Beverage) in different locations of Port Harcourt Metropolis; south-south Nigeria was investigated. Chemical analysis showed that the crude protein content of the hawked kunun drinks was higher (0.67 ± 0.01 - $1.98 \pm 0.01\%$) than that of the laboratory processed kunun samples (0.48 ± 0.02 - $0.80 \pm 0.00\%$), while the total solids were highest in the laboratory processed samples than the hawked Kunun drinks. However, there were no significant differences between the P^H and carbohydrate contents of the hawked kunun drinks and that of the laboratory processed kunun samples. Microbial analysis showed high mean total viable and coliform counts in the hawked drinks while the laboratory processed kunun drinks harbored no coliform bacteria. The microbes found associated with the hawked drinks are *Bacillus subtilis*, *B. cereus*, *Streptococcus faecium*, *Staphylococcus epidermis*, *Micrococcus acidiphilis*, *Escherichia coli*, while *Streptococcus faecium* *Bacillus subtilis* and *Micrococcus acidiphilis* were found in the laboratory prepared kunun samples

INTRODUCTION

Food is any substance, usually composed of carbohydrates, fats, proteins and water, which can be eaten or drunk by animals including humans for nutrition or pleasure. Items considered food may be sought from plants, animals or other categories such as fungus and fermented products (Jango-cohen, 2005). In many developing Countries like Nigeria, people depend mostly on indigenous technology for food preparations especially food of plant origin. Some of these foods that originates from plant includes on alcoholic beverages made mostly from cereal grains. In addition to filling a basic human need, beverages form part of the culture of human society (Larry).

Kunun is a traditional non-alcoholic fermented beverage widely consumed in the Northern parts of Nigeria especially during the dry season (Adeyemi and Umar, 1994). Kunun is cheap and the cereals used in its preparation are widely grown throughout the savannah region of Nigeria such as Bauchi, Kano, Sokoto and Katsina States (Agoa, 1987). Kunun is consumed anytime of the day by both adult and children as breakfast drink or food complement, It is usually used as appetizer to entertain visitors in rural and urban centers and is commonly served at social gatherings

Onuorah et al. (1987; Amusa and. Odunbaku, 2009).

Although there are various types of Kunun processed and consumed in Nigeria including Kunun zaki, Kunun gyada, Kunun akamu, Kunun tsamiya, Kunun baule, Kunun jiko, Amshau and Kunun gayamba. However, kunun zaki is the most commonly consumed.

Kunun processing is mostly done by women using simple household equipment and utensil. Depending on cereal availability, Sorghum, Maize, millet, Guinea corn or rice are commonly used for kunun preparation. According to Odunfa and Adeyeye (1985) the traditional processing of Kunu involves the steeping of grains, wet milling with spices (ginger, cloves pepper), wet sieving and partial gelatinization of the slurry, followed

by the addition of sugar and bottling. The processed Kunun is usually packed for sale either in plastic bottles or in large containers and distributed under ambient temperature or cooled in a refrigerator where available. Kunun-zaki processed from sorghum grains contains 11.6% protein, 3.3% fat, 1.9% ash and 76.8% carbohydrate and arrays of amino acid (Lichtenwalner et al., 1979).

However, this nonalcoholic beverage is however becoming

more widely accepted in several other parts of Nigeria, including Port Harcourt metropolis, owing to its refreshing qualities.

The preparation of this beverage has become technology in many homes in the rural communities and more recently in the urban areas where commercial production due to support from the government through the poverty alleviation scheme, has helped to alleviate poverty among the people.

In developing nations like Nigeria, it has not been possible to have control over the processing of hawked foods, because most of the vendors lack the adequate knowledge of food processing and handling practices. As such, there is likely to be a high risk of chemical and microbial contamination. A large number of lactic acid bacteria, coliforms, molds and yeast have been reportedly implicated in food spoilage as they use the carbohydrate content of the foods for undesirable fermentation processes (Odunfa, 1988; Ojokoh et al., 2002; Amusa et al., 2005). Brief fermentation usually occurs during kunun processing (steeping of the grains in water over an 8-48hrs period), and is known to involve mainly lactic acid bacteria and yeast (Odunfa and Adeyeye, 1985).

In most Nigeria cities, the sales and consumption of this locally made beverage is high due to the high cost other non-alcoholic drinks. This drink is usually hawked in the motor parks, military barracks, school premises and market places.

This research was therefore conducted to investigate the nutritional and microbiological qualities of this hawked non-alcoholic drink called Kunun-Zaki in selected locations of Port Harcourt Metropolis, south- south Nigeria.

MATERIALS AND METHODS

Hawked Kunun drinks samples were purchased from four locations in in PortHrcourt Metropolis- Rumuokoro market, Mile 1 and Mile 3 motor parks and the Army barracks in Bori camp. Five samples were purchased in each location, properly labeled and placed in plastic containers. These samples were respectively brought to the Biochemistry and Microbiology laboratory of the Biochemistry and Microbiology Departments of the University of Port Harcourt for biochemical and microbiological analysis respectively. A Kunun drink sample was also prepared in the laboratory using Millet and Guinea corn respectively. The samples were purchased from the Bori camp Mammy market.

The drink samples were also subjected to the same analysis.

BIOCHEMICAL ANALYSIS

Biochemical analysis carried out on the Kunun drink samples include the pH, the % ash, % moisture content, % total solid, protein, total titrable acidity, carbohydrate content and ether extract (AOAC, 1990).

MICROBIAL ANALYSIS

Isolation of microbes associated with Kunun zaki drinks: Ten fold dilutions of each kunun samples were made using 0.1% peptone water. Appropriate dilutions were made and 0.1 mL of the diluted samples were pour plated in triplicate plates on Plate Count Agar (PCA) for viable count, Eosin Methylene Blue (EMB) for *Escherichia coli* count, Manitol Slant Agar (MSA) for *Staphylococcus* count, Salmonella/Shigella agar for *Salmonella/Shigella* counts, and Brilliant bile broth (BGBB) for coliform test. All plates were incubated for 48hrs at 30C.

Identification of the microbial isolates: Colonies were selected randomly, bacteria cultures were characterized and identified using various morphological and biological test such as gram stain, spore stain, motility, catalase, coagulase, indole, MR –VP, urease, citrate, Oxidase and sugar fermentation. Pure cultures of each isolate were obtained by streaking the specific colonies on suitable media and incubated appropriately; these were maintained in an agar slant in McCartney bottles. The identification of the microbial isolates was based on classification Scheme proposed by Harrigan and McCance (1976), Buchanan and Gibbson (1974) and Collin and Lyne (1984). The identification was based essentially on morphological and biochemical reactions.

Result obtained was subjected to analysis of variance using one – way ANOVA. Differences between means were separated using Duncan's Multiple Range Test (Steel and Torrie, 1980, Duncan, 1955).

RESULTS AND DISCUSSION

Results of the biochemical analyses of Hawked and laboratory processed kunun are shown in Table 1.

Results presented in Table 1 shows that the highest crude protein % was found in the hawked Kunun zaki samples obtained from Mile 1 market ($1.98 \pm 0.01\%$), followed by Rumuokoro market ($0.98 \pm 0.02\%$) while samples from Mile 3 had the lowest protein content ($0.67 \pm 0.05\%$). The kunun

drinks prepared with guinea corn had the highest protein content of $0.80 \pm 0.01\%$ while that prepared with millet had a protein content of $0.48 \pm 0.02\%$. Ash content ranged from $0.42 \pm 0.10\%$ to $1.38 \pm 1.08\%$ in the hawked kunun samples while the laboratory prepared samples had ash contents of 1.14 ± 0.56 and 1.09 ± 0.05 respectively.

Figure 1

Table 1: The Nutritional properties of Hawked and laboratory processed Kunun-zaki

Location	Moisture (%)	Ash (%)	Protein (%)	CHO (%)	pH	Total Solids	Titrate acidity (g/100g)
Mile 1 motor park/Market	88.35 ± 0.999	1.38 ± 1.08	1.98 ± 0.01	78.90 ± 0.19	4.13 ± 0.01	13.66 ± 0.29	0.45 ± 0.00
Mile 3 motor park/Market	94.05 ± 0.20	0.72 ± 0.03	0.67 ± 0.01	72.20 ± 0.20	4.35 ± 0.01	6.23 ± 1.01	0.12 ± 0.01
Rumuokoro Market	94.01 ± 0.02	0.42 ± 0.10	0.98 ± 0.02	82.65 ± 0.10	4.33 ± 0.01	6.41 ± 1.24	0.13 ± 0.01
Bori camp Army Barracks	88.46 ± 0.23	0.30 ± 0.10	0.78 ± 0.05	80.16 ± 0.11	4.14 ± 0.01	13.94 ± 1.27	0.35 ± 0.01
Laboratory prepared Kunun with Guinea corn	85.98 ± 0.05	1.14 ± 0.56	0.80 ± 0.10	89.40 ± 0.26	4.23 ± 0.00	16.51 ± 1.17	0.27 ± 0.01
Laboratory Prepared Kunun with millet	86.38 ± 0.04	1.09 ± 0.05	0.48 ± 0.02	82.97 ± 0.16	4.22 ± 0.01	14.18 ± 0.92	0.25 ± 0.01

There was no significant difference between the Carbohydrate contents and the P^H of the hawked and the laboratory prepared samples while the Total titratable acidity of samples obtained from Mile 1 Motor park/market and Bori camp army barracks were significantly higher (0.45 ± 0.10 and $0.35 \pm 0.10\%$ respectively) than samples obtained from Mile 3 Motor park and Rumuokoro markets (0.12 ± 0.01 and $0.13 \pm 0.03\%$ respectively). Total solids of the laboratory prepared samples were significantly higher than in the hawked samples (Table 1).

Six different microbes were found associated with the Hawked (marketed) Kunun in Port Harcourt metropolis while the laboratory processed samples harboured five (Table 2). These microbes include *Lactobacillus plantarum*, *Bacillus cereus*, *Streptococcus faecium*, *S. lactis*, *Staphylococcus aureus*, *Micrococcus acidiphilus* and *Escherichia coli*. In both samples *L. plantarum*, *S. lactis* and *Bacillus subtilis* had the highest rate of occurrence while the least was *E. coli*.

Figure 2

Table 2: The incidence of Bacteria found associated with hawked kunun zaki in selected locations in PortHarcourt metropolis

Location	<i>B. cereus</i>	<i>B. subtilis</i>	<i>L. plantarum</i>	<i>S. epidermis</i>	<i>M. acidophilus</i>	<i>S. faecium</i>	<i>E. Coli</i>
Mile 1 motor park/Market	+	-	+	+	+	-	-
Mile 3 motor park/Market	+	-	+	+	-	-	-
Rumuokoro Market	+	-	+	-	+	-	+
Bori camp Army Barracks	+	-	+	-	-	-	-
Laboratory prepared Kunun with Guinea corn	-	-	+	-	-	-	-
Laboratory Prepared Kunun with millet	-	+	+	-	-	-	-

+ = present
- = absent

Results in Table 3 indicate that the highest total viable count of 8.00×10^6 cfu/ml was recorded for samples obtained from Rumuokoro market followed by samples obtained from Mile 3 Motor park/market (7.40×10^6 cfu/ml), while the least mean total viable of was recorded for samples obtained from Bori camp and Mile 1 markets.

Figure 3

Table 3: The total microbial load of hawked Kunun-zaki in selected locations in PortHarcourt metropolis

Location	Mean Total Viable Count (cfu/ml $\times 10^6$)	Salmonella Shigella count (cfu/ml $\times 10^1$)	Staphylococci Count (cfu/ml $\times 10^2$)	Mean total coliform count (cfu/ml $\times 10^4$)
Mile 1 motor Park/Market	3.21 ± 0.09	Nil	3.5 ± 0.03	5.80 ± 0.22
Mile 3 motor Park/Market	7.40 ± 0.19	Nil	3.1 ± 0.01	3.50 ± 0.15
Rumuokoro Market	8.00 ± 0.21	Nil	Nil	2.9 ± 0.10
Bori camp Army Barracks	2.60 ± 0.10	Nil	Nil	4.30 ± 0.20
Laboratory prepared Kunun with Guinea corn	1.60 ± 0.04	Nil	Nil	Nil
Laboratory Prepared Kunun with millet	1.02 ± 0.02	Nil	Nil	Nil

These values were however significantly lower than values recorded for the laboratory prepared kunun samples (1.60×10^6 cfu/ml and 1.02×10^6 cfu/ml respectively). The coliform count ranged from 2.9×10^6 cfu/ml to 5.8×10^6 cfu/ml in the hawked samples while no coliform was found associated with the laboratory processed samples. There was no observable growth of salmonella/shigella in both the hawked and laboratory prepared samples, while the hawked samples from Mile 1 and Mile 3 Motor Park/ markets had

staphylococcus counts of 3.5 ± 0.03 and $3.1 \pm 0.01 \times 10^2$ cfu/ml respectively. No staphylococcus was observed in the Bori camp, Rumuokoro and the laboratory prepared samples.

DISCUSSION

Results of the experiment indicated that the highest crude protein content was found in the hawked Kunun samples compared to the laboratory prepared samples. Reasons for this might have been as a result of some of the additives added to the processed kunun samples. However, the protein contents of these Kunun drinks were very low probably because most of it might have been lost during processing. According to Hamad and Fields (1979), much of the protein in cereals is usually located in the testa and germ which are usually sifted off during processing. The protein contents obtained in this study were lower than that observed in sorghum-based kunun drinks by Amusa and Odunbaku, (2009). This may have been due to the use of Guinea corn and millet to prepare the drinks. However, Lichtenwalner et al. (1979) had also reported that the protein content of sorghum based kunun gruel was 11.6%. There was no significant difference between the Carbohydrate contents and the P^H of the hawked and the laboratory prepared samples. Results obtained for carbohydrate contents of all the samples in Port Harcourt metropolis were similar to those obtained for hawked sorghum based kunun drinks in south-western Nigeria by Amusa and Odunbaku, (2009). This confirms that most cereal grains contain similar carbohydrate contents. It is a known fact that fermentation which usually occurs during kunun processing usually involves mainly lactic acid bacteria and yeast (Odunfa and Adeyeye, 1985).

Results obtained for total titrable acidity of the hawked samples from two locations were higher than values obtained for the laboratory prepared samples. Values obtained for the laboratory prepared samples (0.27 ± 0.03 and 0.25 ± 0.01 g/100g respectively) compared well with values obtained by Amusa and Ashaye (2009) in kunun drink which ranged between 0.21 and 0.28 g/100g. High total solids content of the laboratory prepared samples compared to the hawked samples may be due to loss of slurry during the wet milling of the cereal grains.

The microbes associated with the kunun samples include *Lactobacillus plantarum*, *Bacillus cereus*, *Streptococcus faecium*, *S. lactis*, *Staphylococcus aureus*, *Micrococcus acidiphilis* and *Escherichia coli*.

The presence of some of these organisms are not surprising as most of them are known to thrive in medium rich in fermentable substrates such as sugars which often led to the production of acids after fermentation. Odunfa and Adeyeye (1985) reported that *L. plantarum* was the predominant organism in the fermentation responsible for lactic acid production, while, *S. lactis* and *Micrococcus acidiphilis* are known to be involved in fermentation of agricultural produce. The presence and the activities of these fermenters might be responsible for the souring of taste usually observed if the drinks are not consumed within six hours of processing. *Bacillus* species are spore formers whose spores could survive high temperatures of processing. The thermophilic nature of the spores of these microbes ensures survival at pasteurization temperatures and hence their presence in the Kunun samples that have been subjected to heat treatment during processing. The ropiness associated with the fermented drink has been associated with the presence of both *Pseudomonas* spp. and *Bacillus subtilis* (Adegoke et al., 1993).

Some of these associated microbes have been implicated in food poisoning outbreak of some food materials (Sartory and Howard, 1992). The presence of *Escherichia coli* in food is an indication of faecal contamination of product. According to Sartory and Howard (1992), the presence of *E. coli* in water indicates faecal contamination and most of the coliforms found associated with the hawked Kunun are known to be causative agents of food borne gastroenteritis and bacterial diarrhoea diseases (Jiwa et al., 1981; Onuorah et al., 1987).

The presence of coliform bacterial in the hawked kunun drinks is not unexpected since the source of water used in many parts of Port Harcourt metropolis is tap or borehole water. Coliforms have been reportedly associated with tap water popularly consumed in some towns in Nigeria (Adegoke et al., 1993). Amusa and Ashaye (2009) also reported that the presence of coliforms in hawked kunun drink samples in south-western Nigeria were as a result of the use of contaminated water, containers, as well as dirty environment where the kunun samples were being processed and even hawked. No coliform was found associated with the laboratory prepared kunun drink samples where sterile water was used.

The presence of *Staphylococcus* species in the Mile 1 and Mile 3 samples (Table 3) were possible contaminants from handlers. *Staphylococcus aureus*, a mesophile have been

implicated in food poisoning outbreak of some food material. Odunfa (1988) reported that *Staphylococcus aureus* levels of 10^8 /ml are considered potential hazardous to consumers.

This is a source of concern in Nigeria because the teeming populace relies on these drinks as alternatives to the bottled canned drinks whose price is becoming unaffordable. It is therefore suggested that kunun drinks should be properly processed to avoid microbial contamination. While treated municipal water or clean water should be used during for processing and dilution of the processed drinks to avoid contamination with enteropathogenic bacteria.

Since spices have been reported to inhibit microbial growth (Zaika et al., 1983; Adegoke and Skura, 1994; Adebisin et al. 2001), the addition of spices to the processed kunun drinks is highly advocated. Health education training should be organized regularly for the processors by the health workers on the importance of cleanness of their environment and the use sterilized packaging materials. Since kunun zaki drink is highly relished by both young and adult in many Parts of Nigeria, with proper processing and packaging it will not only alleviate the longing for fluid intake in warm tropical climates but would also provide a cheaper and more nutritive drink due to its therapeutic properties (Bestshart, 1982), than the more costly, sugar laden soft drinks and processed fruit juices in the market.

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