

NUTRITIONAL AND MICROBIOLOGICAL QUALITY OF KUNUN- ZAKI BEVERAGE PRODUCED IN OWERRI MUNICIPAL

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ABSTRACT

Nutritional and Microbiological qualities of kunu-zaki hawked at different locations in Owerri municipal were assessed and evaluated. Three major locations of production and sales namely: Shell-camp, Ama-Hausa and Obinze all in Owerri municipal were selected for sampling of kunu-zaki. Laboratory prepared sample of kunu-zaki was used as control. Samples of kunu were evaluated for microbiological quality such as total bacteria and fungi counts also, biochemical characterization of bacteria and morphological characterization of fungi. Chemical properties which include proximate composition, titrable acidity, pH, total solid and minerals were also examined. Results showed that there was a significant difference ($P < 0.05$) on all the chemical properties of samples examined. The moisture content ranged from 85.14% to 89.57% while crude protein was between 2.20% and 4.09%. Ash content varied significantly ($P > 0.05$) with range 1.67% to 2.33%. Crude fiber content of 0.71% of control was higher than 0.68%, 0.53% and 0.44% found on the hawked kunu-zaki sample. Total carbohydrate was found within ranged values of 5.54% to 7.74%. Total solid content among kunu-zaki samples ranged from 12.54mg/100g to 14.32mg/100g, pH ranged 5.37 to 5.76 and acidity of the kunu-zaki samples varied but ranged from 0.23% of sample collected from Obinze location to 0.28% of Ama-Hausa kunu-zaki sample. Mineral contents of kunu-zaki samples differed significantly ($P < 0.05$), calcium content of 32.09mg/100g was higher than 31.21mg/100g, 27.31mg/100g and 12.53mg/100g. Potassium content ranged from 311.63mg/100g to 449.03mg/100g. Control laboratory sample was found with highest content of 11.74mg/100g magnesium, 4.81mg/100g iron and 30.26mg/100g vitamin C. Total bacteria counts of the kunu-zaki samples were not the same; control laboratory sample was least, 3.0×10^6 cfu/mL of heterotrophic bacteria, 3.0×10^6 cfu/mL of total Staphylococcus count and 9.0×10^6 cfu/mL of fungi counts. Bacteria identified are Bacillus spp, Klebsiella spp, Staphylococcus aureus and Salmonella while fungi suspected include Mucor spp, Saccharomyces spp, Aspergillus spp and Rhizopus. However, laboratory processed samples made kunu-zaki beverage were more acceptable with least microbial load. Good manufacturing and good hygiene practices should be given utmost importance during production to avoid microbial contamination that may cause food borne illness.

Keywords: Nutritional, microbiological, Kunun- zaki beverage

*Journal of the Faculty of Agriculture and Veterinary Medicine, Imo State University Owerri
 website: www.ajol.info*

<https://dx.doi.org/10.4314/jafs.v16i1.5>

INTRODUCTION

Food is any substance, usually composed of carbohydrates, fats, proteins and water, which can be eaten or drunk by humans for nutrition or pleasure (A.O.A.C, 2000). 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 originate from plant include alcoholic beverages made mostly from cereal grains (Adekunle, 2012). Kunu is an important non-alcoholic fermented beverages widely consumed in Northern parts of Nigeria especially during the dry season (Adelekan *et al.*, 2013). Kunu is cheap and the cereals used in its preparation are widely grown throughout the Savanna region of Nigeria such as Bauchi, Kano, Sokoto, and Katsina States (Adeyemi and Umar, 2011). It was consumed primarily (Adepoju *et al.*, 2012) used to be consumed mainly in the Northern parts but it is now widely acceptable in almost all parts of Nigeria, owing to its refreshing qualities. It is acceptable to people of all works of life and is being served at home and public places as food appetizer, refreshing drink and complementary food for infants. The various types of kunu processed and consumed in Nigeria include Kunu zaki, Kunu gyada, Kunu akamu, Kunu tsamiya, Kunu baule, Kunu jiko, Amshau and Kunu gayamba. However, kunu -zaki is the most commonly consumed. Kunu processing is mostly done by women with simple house hold equipment utensil. Depending on cereal availability, sorghum, maize, millet, guinea corn or rice are commonly used for kunu preparation (Alexander and Strette, 2001). Kunu zaki is one of the types of kunu (a non-alcoholic beverage) that is very popular in the northern part of Nigeria but it is being consumed now in all parts of the country (Adekunle, 2012).

Kunu zaki is produced from millet and sorghum grains. The process of manufacture involves wet milling of millet grains with spices (ginger, clove, black pepper, ehuru) and sweet potato (*Ipomea botatas*) but sweet potato is optional (Adeyemi and Umar, 2011). The short shelf-life of this beverage is however a major problem faced by their producers and consumers (Adekunle, 2012). Also 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. A large number of lactic acid bacteria, coliforms, *E. coli*, molds and yeast have been reportedly implicated in food spoilage as they use the carbohydrate content of the foods for undesirable fermentation processes (Alexander and Strette, 2001). Research carried out showed that *E.coli*, *Staphylococcus* and *Salmonella* are the major cause of food borne diseases (AOAC, 2000.). The problem of food borne disease or the assessment of the microbiological quality of food cannot be overemphasized, as a result of this; this study will be of an added knowledge to the existing ones. The general objective of this study is to determine the microbiological and nutritional quality of Kunu beverage produced from different Hausa locations in Owerri, Imo State, Nigeria.

MATERIALS AND METHODS

Sample collection

The kunu samples used in this study were procured from different location in Owerri Municipal Council in Imo State. The locations include Shell camp, Ama-Hausa and Obinze.

Laboratory preparation of Kunu-zaki

The millet, ginger (dried) and sugar were all purchased from Eke Ukwu market, Owerri, Imo State. The materials were sorted and only those in good condition were used.

Production of Kunu-zaki

Laboratory sample of Kunu-zaki were produced from 10 grams of millet as shown in Table 3.1. The sample were washed, soaked overnight and wet milled with equal volume of water. Ginger was added to each of the blends prior to milling as indicated in Table 3. 1. Each of the wet milled samples was divided into two equal parts. Two parts were prepared into a thick porridge using boiled water. The remaining one part of the wet milled samples were added back to their respective thick porridges and stirred. They were allowed to stand overnight for about 15 hours to allow for fermentation and flavour development. The fermented samples were sieved using muslin cloth to give the different kunu-zaki samples which were used for subsequent analysis.

Proximate Composition Analysis

This was carried out according to the method of(AOAC, 2000)

Moisture Content Determination

Two grams of each of the sample was weighed into dried weighed crucible. The samples was put into a moisture extraction oven at 105⁰C and heated for 3h. The dried samples was put into desiccators, allowed to cool and reweighed. The process was reported until constant weight was obtained. The difference in weight was calculated as a percentage of the original sample

Percentage moisture

$$= \frac{W2 - W1}{W2 - w3} \times 100$$

Where

W1 = Initial weight of empty dish, W2 = Weight of dish + undried sample, W3 = Weight of dish + dried sample

Ash Content Determination

Two grams of each of the samples was weight into crucible, heated in a moisture extraction oven for 3h at 100⁰C before being transferred into a muffle furnace at 550⁰C until it turned white and free of carbon. The sample was then removed from the furnace, cooled in a desiccator to a room temperature and reweighed immediately. The weight of the residual ash was then calculated as Ash Content

$$\text{Percentage Ash} = \frac{\text{Weight of Ash}}{\text{Weight of original of smaple}} \times \frac{100}{1}$$

Crude Protein Determination

The micro kjeldahl method described by (AOAC, 2000) was used. Two grams of each of the samples was mixed with 10ml of concentrated H₂SO₄ in a heating tube. One table of selenium catalyst was added to the tube and mixture heated inside a fume cupboard. The digest was transferred into distilled water. Ten millimeter portion of the digest mixed with equal volume of 45% NaOH solution and poured into kjeldahl distillation apparatus. The mixture was distilled and the distillate collected into 4% boric acid solution containing 3 drops of methyl red indicator. A total of 50ml distillate was collected and titrated as well. The sample was duplicated and the average value taken. The Nitrogen content was calculated and multiplied with 6.25 to obtain the crude protein content. This is given as percentage Nitrogen

$$= \frac{(100 \times N \times 14 \times VF)I}{100 \times Va}$$

Where,

N= Normality of the titrate (0.1N)

VF= Total volume of the digest= 100ml

T= Titre Value

Va= Aliquot Volume distilled

Fat content determination

Two grams of the sample was loosely wrapped with a filter paper and put into the thimble which was fitted to a clean round bottom flask, which has been cleaned, dried and weighed. The flask contained 120ml of petroleum ether. The sample was heated with a heating mantle and allowed to reflux for 5h. The heating was then stopped and the thimbles with the spent samples kept and later weighed. The difference in weight was received as mass of fat and is expressed in percentage of the sample.

$$\text{The percentage oil is percentage fat} = \frac{W2-W1}{W3} \times \frac{100}{1}$$

Where,

W1 = weight of the empty extraction flask

W2 = weight of the flask and oil extracted

W3 = weight of the sample

Crude fibre determination

Two grams (2g) sample and 1g asbestos were put into 200ml of 1.25% of H₂S₀₄ and boiled for 30 minutes. The solution and content then poured into Buchner funnel equipped with muslin cloth and secured with elastic band. This was allowed to filter and residue was then put into 200ml boiled NaOH and boiling continued for 30 minutes, then transferred to the buchner funnel and filtered. It was then washed twice with alcohol, the material obtained washed thrice with petroleum ether. The residue obtained was put in a clean dry crucible and dried in the moisture extraction oven to a constant weight. The dried crucible was removed, cooled and weighed. Then, difference of weight (i.e. loss in ignition) is recorded as crucible fibre and expressed in percentage crude fibre

$$= \frac{W1 - W2}{W3} \times \frac{100}{1}$$

Where,

W1 = weight of sample before incineration

W2 = weight of sample after incineration

Wt = weight of original sample

Carbohydrate content determination

The nitrogen free method described by (AOAC, 2000) was used. The carbohydrate is calculated as weight by difference between 100 and the summation of other proximate parameters as Nitrogen free Extract (NFE) percentage carbohydrate

$$(NFE) = 100 - (m + p + F_1 + A + F_2)$$

Where

m = moisture

p = protein

F₁ = Fat

A = ash

F₂ = Crude fibre

Determination of mineral composition of samples

The methods of (Chowdhurys and Punia, 2007) were used for the determination of minerals (Ca, Mg, K, Na, P and Fe) by atomic absorption spectrophotometer. One gram samples, in triplicate were dry, ashed in a muffle furnace at 550⁰C for 8h until a white residue of constant weight were obtained. The minerals were extracted from ash by adding 20.0 mL of 2.5% HCl, and this was transferred quantitatively to a 50 mL volumetric flask. It was diluted to volume (50 mL) with

deionised water, stored in clean polyethylene bottles and mineral contents determined using an atomic absorption spectrophotometer (Perkin- Elmer, Model 2380, USA)

Determination of microbial population

Microbial count was done according to ICMSF (Jango-Cohen, 2005). Ten grams (10gmL) of each sample was serially diluted into 10- folds dilution using sterile peptone water, and then homogenized by shaking vigorously. An aliquot portion (0.1ml) of 3rd up to 5th dilution was inoculated in duplicate onto the potato dextrose agar (PDA), and nutrient agar (NA) (MA) for the isolation of heterotrophic fungi and bacteria respectively. Potato dextrose agar plates were incubated at ambient temperature (28^o+02^oC) for fungal growth. The nutrient agar plates was spread evenly with a sterile spreader and incubated for 24-48h at different temperatures 37^oC for total viable count (Alexander and Strette, 2001).

Biochemical reaction of microbes

Species that cannot be distinguished by their cultural microscopic characteristics may exhibit distinct difference in their biochemical reactions is provided by (Alexander and Strette, 2001).

Carbohydrates Fermentation Test

The ability of organisms to ferment sugars could be used as a distinguish factor. An aliquot portion of the basal medium (peptone sugar water) was dispersed into a test-tube and filled by gently inversion with the medium. The cultures (twenty-four hours old) was inoculated into the sterilized medium and incubated for forty eighth hours at 37^oC. A color change from red to yellow (with phenol red indicator) and air space in the Durham tubes was considered positive, indicating acid and gas production respectively.

Catalase Test

Aerobic and aero-tolerant organisms have the ability to produce an enzyme-catalase that splits H₂O₂, a toxic compound produced in the respiratory process to oxygen and water. To test for the presence of catalase, a loop full of test culture (twenty four hours old) was placed on a clean, grease free slide. Add a loopful of the H₂O₂ and mix thoroughly with the edge of a slide. Effervescence due to the liberation of oxygen indicates a positive catalase reaction.

Coagulase Test

This test differentiates *Staphylococcus aureus* which produce the enzyme coagulase from the non-coagulase producing *staphylococcus* and *micrococcus* species. The enzyme coagulates causes plasma to clot by converting fibrinogen to fibrin. Place a drop of physiological saline on each end of a slide. Emulsify a colony of the test organism on each of the drop to make two thick suspensions. Add a drop of plasma to one of the suspension and mix gently. Look for clumping of the organism within ten seconds.

Oxidase Test

Some organisms have the tendency of forming a complex mixture with the oxidase reagent (para-amino dimethyl phenylenedimine monochloride and water) and appearing purple on filter paper soaked with the reagent.

Procedure

Few drops of freshly prepared oxidase reagent will be placed on sterile filter paper. Test cultures (Twenty four hours old) were streaked on the filter paper soaked with the reagent. The appearance of a purple color after ten seconds indicates positive reaction.

Imvic Test

This test consists of four different tests; they are Indole production, Methyl-red test, Voges Proskauer tests and the physiological properties of microorganisms. They are especially useful in the differentiation of Gram negative intestinal bacilli, particularly *Escherichia coli* and the *Enterobacter-klebsiella* group.

Indole Production: Indole is produced in triptone broth by the enzyme of certain organisms. Triptone broth is rich in amino acid tryptophan which can be used by some bacteria as source of carbon, energy as well as nitrogen. Tryptophan is degraded to indole pyruvic acid and ammonia by some microorganisms. A loopful of test culture (Twenty four hour old) was inoculated into the triptone broth and incubated for two days. Into six milliliters of culture broth a three milliliters of Kovac's reagent was added from aqueous layer, colour change to red is a positive test.

Methylred and Voges-Proskauer tests must be considered together from the MR and VP test, i.e. MR +, VP- or MR-, VP+. Methyl red test was performed to demonstrate the capacity of different organisms to produce acid from the fermentation of sugar (dextrose). Methyl red positive organisms produce a red coloration when five drops of methyl red-indicator is added into forty-eight hour old MR-VP culture. The Voges-Proskauer test demonstrates the ability of organisms to produce acetone from glucose metabolism. Some organisms metabolize glucose to produce pyruvic acid which is further broken down to yield butane-diol and acetyl-methyl carbonyl as an intermediate product. Into one milliliter of the culture add one milliliter of six percent alcoholic solution of alpha-naphthol (α -naphthiol) and one milliliter of 16% Kott and stand for 15-20 minutes. Development of red to pink colour is a positive test.

Citrate Test

The citrate test was performed by inoculating into organic synthetic medium in which sodium citrate is the only sources of carbon and energy. In sodium citrate broth (Koser's citrate medium), the presence of growth (turbidity) is a positive test result.

Motility Test

The ability of an organism to move is determined by the presence of one or more flagella. In the laboratory, motility test of micro organisms, especially bacteria is demonstrated by the hanging drop method. A loopful of broth culture of the test organism was dropped at the centre of Vaseline made into a cycle on clean grass free microscopic slide. A cover slip is placed on top of the Vaseline and inverted. Motility was observed with the low magnifications (10^{\times} and 40^{\times}). The appearance of darting or rigging in the broth signifies motility. (Jango-Cohen, 2005), (Alexander, and Strette, 2001), Buchanan consulted in the identification of the organisms taking into consideration the test performed.

Statistical analysis

Data were subjected to Analysis of Variance (ANOVA version 20.0) and Turkey's test was used for comparison of means.

RESULTS

The proximate composition of kunu zaki sample is shown in Table 4.1. The proximate composition showed significant difference ($p < 0.05$) among the samples. The moisture content ranged from 85.14-89.57%. The kunu sample from shell camp has the highest moisture content of 89.57%, followed by kunu sample from Obinze and Ama-hausa with 88.35% and 87.69% respectively while control sample has the least value of 85.14%. The protein content varies significantly from 2.20-4.09%. The control sample has the highest protein value of 4.09% followed by kunu sample from Ama-Hausa and Obinze with 3.77% and 2.84% while kunu sample from Shell camp has the least value of 2.20%. The Ash content ranged between 1.67% and 2.33%. The sample control has the highest value of 2.33% while kunu sample from Ama-Hausa has the least value of 1.67%. The crude fiber content varies from 0.44-0.71%. The control sample has the highest crude fiber value of 0.71% followed by kunu sample from Shell camp and Obinze with 0.68% and 0.53% while kunu sample from Ama-Hausa has the least value of 0.44%. The carbohydrate content varies between 5.54-7.74%. The control sample has the highest value of 7.74% while kunu sample from shell camp has the least value of 5.54%.

The chemical properties of Kunu zaki is presented in Table 4.2. The Total solid content of the samples was not the same. Total solid of 14.32mg/100g of the control kunu samples was higher than 13.46mg/100g of Ama-Hausa, 13.02mg/100g of Obinze and the least 12.54mg/100g was found on sample collected from Shell-camp. The pH varies from 5.37 -5.76. the kunu sample from Obinze has the highest pH value of 5.76 while the kunu sample from Ama-Hausa sample has the least value of 5.37. The Acidity ranged from 0.23- 0.28mg/100. The kunu sample from Ama-Hausa has the highest acidity value of 0.28mg/100g while kunu sample from Obinze has the least value 0.23mg/100g.

The mineral and Vitamin C composition of kunu zaki is presented in table 4.3. The calcium content varies from 12.53-32.09mg/100g. The control sample has the highest calcium value of 32.09mg/100g while the kunu sample from Obinze has the least value of 12.53mg/100g. The potassium content ranged between 311.63-449.03mg/100g. The control sample has the highest potassium value of 449.03mg/100g followed by kunu sample from Obinze and Shell camp with 412.03mg/100g and 399.08mg/100g while kunu sample from Ama-Hausa has the least value of 311.63mg/100g. The magnesium content varies significantly from 5.44-11.74mg/100g. The control sample has the highest magnesium value of 11.74mg/100g while kunu sample from Ama-Hausa has the least value of 5.44mg/100g. The control sample has the highest value of iron and vitamin C with 4.81mg/100g and 30.26mg/100g respectively while kunu sample from Ama-Hausa has the least value of iron and vitamin C with 1.19mg/100g and 22.14mg/100g respectively.

DISCUSSION

Proximate composition

The proximate composition of kunu zaki sample is shown in Table 4.1. The proximate composition showed significant difference ($p < 0.05$) except the crude fiber that show no significant difference ($p > 0.05$) among the samples. The moisture content ranged from 85.14-89.57%. The moisture content of the kunu zaki is high. High moisture content in food influence protection against micro-organism which affect the stability and safety of food, hence refrigeration or freeze would be required to extend the shelf life of the product. The high moisture content found on the samples suggests that they can be used as refreshing drink that quenches thirst. This is typical of refreshing drinks. The moisture content of the beverages was however in line with the one as reported for juice from *Spondias mombin* and extract of *Hibiscus sabdariffa* (Adepoju and Oladejo, 2012). Water is the most important constituent of every living cell. It takes part in various chemical and biochemical processes including hydrolytic breakdown of nutrients during digestion, is continually lost from the body through urine, stools, sweat, and is expelled through respiration. Water imbalance leads to serious conditions such as dehydration which may be fatal (Pelczar *et al.*, 1993). The protein content varies significantly from 2.20-4.09%. there is significant difference ($p < 0.05$) among the kunu samples examined. The protein content of the samples in this work was higher than the value reported by (Chowdhurys, and Punia, 2007), and this may be attributed to the raw material adopted during the production of beverages. The improved protein content of kunu-zaki recorded in this study may be beneficial to consumers of the products as majority of people in Nigeria could not afford protein sources of food such as meat and egg for economic reasons. The ash content ranged between 1.67% and 2.33%. (Iwe, 2002) reported that ash content of food is a measure of mineral element present in food stuff. (Adelekan *et al.*, 2013) reported increase in the

ash content of kunun-zaki enriched with extract of sesame seeds over the control sample. Values of the ash contents in the knun-zaki recorded by the research workers are similar to those obtained in the present study. However, (Iwe, 2002) and (Adelekan *et al.*, 2013) obtained ash content of higher values in their finding on kunu-zaki than those recorded in this study. The difference could be attributed to the types of cereals or raw materials used in the production of the beverage in the different studies. Different cereal types have abilities to contribute to the ash content of kunu- zaki as a result of the differences in their ash compositions. The crude fiber of the samples showed significant difference ($p < 0.05$) with statistical analysis. This could be as a result of higher content of fiber contained in the raw materials used. The carbohydrate content of the sample showed significant difference ($p < 0.05$). The high carbohydrate content of kunun-zaki indicates a good source of energy needed for human activity (Iwe, 2002).

Chemical properties of Kunu zaki

The chemical properties of kunu zaki are presented in Table 4.2. All the samples were slightly acidic with pH range of 5.37 to 5.76. This could be attributed to lactic acid fermentation of the beverage which led to the production of lactic acids which caused slight acidity of the samples (Adebayo *et al.*, 2010). The low value of TTA for samples indicates poor fermentation which could have caused poor production of lactic acid. This invariably led to higher pH value of this sample when compared with the other samples.

Mineral and Vitamin C Composition of Kunu zaki

Table 4.3 showed the mineral and vitamin C composition of Kunun zaki samples. The calcium content varies from 12.53mg/100g to 32.09mg/100g. Calcium helps in the regulation of muscle contractions and transmission of nerve impulses as well as bone and teeth development (Adelekan *et al.*, 2013). The potassium content ranged between 311.63mg/100g and 449.03mg/100g. Potassium is essential for its important role is the synthesis of amino acids and protein. The increases in the content of the minerals recorded in the Kunu-Zaki sample could therefore be of nutritional advantage to consumers of the products. There is significant difference ($p < 0.05$) on the magnesium, iron and zinc content among the sample examined..The increase in the mineral content in the samples could therefore justify the need to enrich the beverage with source that are rich in other nutrients lacking in cereals normally adopted in its production (Chowdhurys, and Punia, 2007). Minerals are of great importance in diet as they play important roles in body metabolism.

Microbiological quality of Kunu zaki

The microbes found associated with both the hawked and the laboratory prepared kunun sample comprises of bacteria which includes, *Bacillus subtilis*, *Staphylococcus aureus*, *Klebsiella sp*, and *Salmonella sp* the mould which are, *Apergillus niger*, *Rhizopus spp* and *Mucor spp* and the yeast

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which includes and *Saccharomyces cerevisiae*. 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. The presence and the activities of these fermenters might be responsible for the souring of taste usually observed if not consumed within six hours of processing (Odunfa. and Adeyeye, 2005.) found that *Bacillus* and *Lactobacillus* species were readily found in foods of low acid content like juices and beverages where they produce organic acids. *Bacillus* species are spore formers whose spores could survive high temperatures of processing. The thermotolerant 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. Some of these associated microbes have been implicated in food poisoning out breath of some food materials (Amusa and. Odunbaku, 2009). However, (Adebayo, *et al.*, 2010) reported that staphylococcus spp level of 10^8 ml⁻¹ is considered potential hazardous to consumers. These fungi also have high survival rate of their spores hence their presence in these food drinks could have emanated from the air as air since most of the hawked hkunu drinks are not usually well covered. The presence of coliform bacterial in the hawked kunu drinks in Nigeria is a source of concern because the teaming populace relies on these drinks as alternative to the bottled soft drinks whose price is becoming unaffordable.

CONCLUSION

The results obtained from the different analyses of the Kunu-zaki beverages showed that the laboratory processed sampled did not alter the proximate, chemical and mineral properties of the beverage rather it helped to improve these properties thereby making them more acceptable. However, microbial content of these hawked marketed Kunu-zaki drinks was higher and showed high microbial load which may be potential pathogens to consumers.

RECOMMENDATION

Good manufacturing and good hygiene practices should be given utmost importance during production to avoid microbial contamination that may cause food borne illness.

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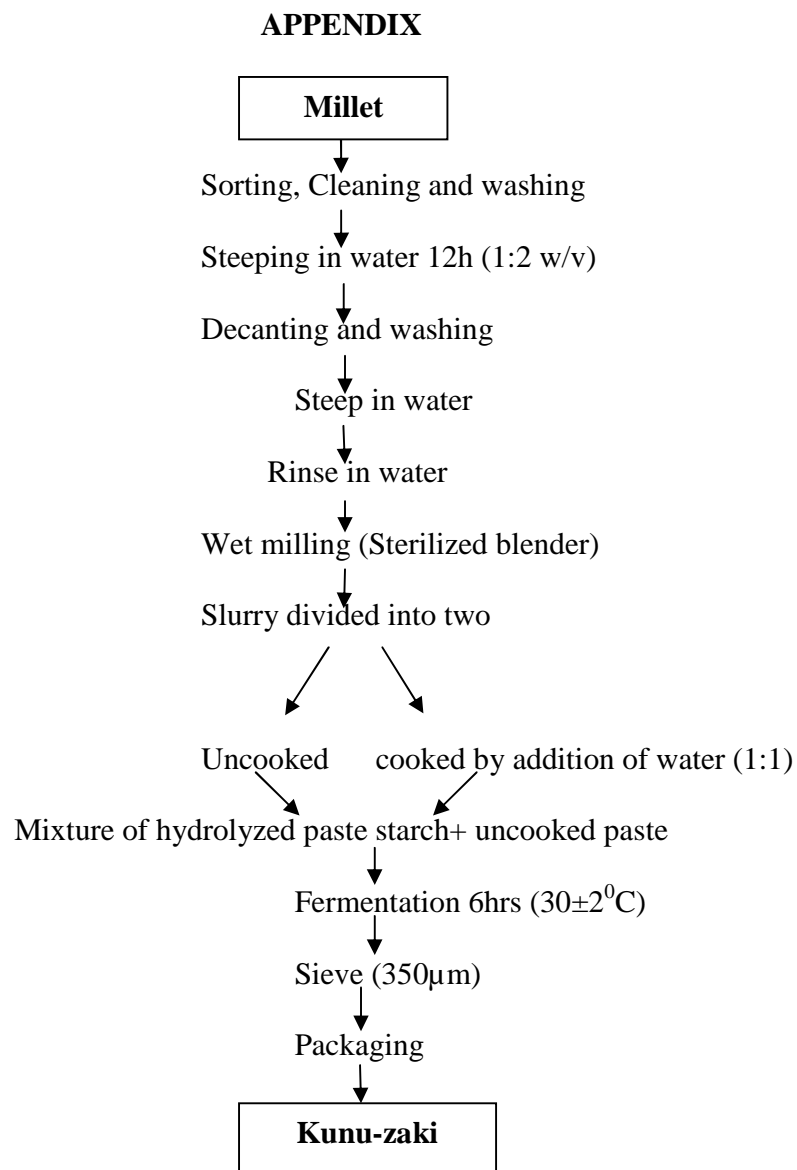


Figure 1: Chart for Kunu-zaki production using millet

Proximate composition of Kunun-zaki from Owerri Municipal

Table 1: Proximate composition of kunu zaki

| Sample | Moisture % | Protein % | Ash % | Crude fiber % | Carbohydrate % |
|---------------------------------|--------------------------|-------------------------|-------------------------|--------------------------|---------------------------|
| Shellcamp kunun sample | 89.57 ^a ±0.02 | 2.20 ^d ±0.03 | 2.02 ^b ±0.03 | 0.68 ^a ±0.02 | 5.54 ^d ±0.09 |
| Ama-hausa kunun sample | 87.69 ^c ±0.03 | 3.77 ^b ±0.02 | 1.67 ^c ±0.03 | 0.44 ^c ±0.02 | 6.44 ^b ±0.06 |
| Obinze kunun sample | 88.35 ^b ±0.03 | 2.84 ^c ±0.03 | 2.09 ^b ±0.01 | 0.53 ^b ±0.0 | 6.19 ^c ±0.01 |
| Laboratory control kunun sample | 85.14 ^d ±0.03 | 4.09 ^a ±0.04 | 2.33 ^a ±0.03 | 0.71 ^a ±0.01 | 7.74 ^a ±0.03 |
| LSD | 0.027 | 0.029 | 0.025 | 0.015 | 0.059 |

Mean values with the different letters are significant ($p < 0.05$) in the same column, LSD=Least significant difference.

Total solid, pH and acidity content of Kunun-zaki from Owerri Municipal

Table 2: Chemical properties of Kunu zaki

| Sample | Total solid mg/100g | pH | Acidity % |
|---------------------------------|--------------------------------|-------------------------|-------------------------|
| Shellcamp kunun sample | 12.54 ^d ±0.01 | 5.49 ^c ±0.01 | 0.26 ^b ±0.01 |
| Ama-Hausa kunun sample | 13.46 ^b ±0.01 | 5.37 ^d ±0.01 | 0.28 ^a ±0.01 |
| Obinze kunun sample | 13.02 ^c ±0.02 | 5.76 ^a ±0.01 | 0.23 ^c ±0.02 |
| Laboratory control kunun sample | 14.32 ^a ±0.01 | 5.61 ^b ±0.01 | 0.26 ^b ±0.01 |
| LSD | 0.016 | 0.009 | 0.002 |

Mean values with the different letters are significant ($p < 0.05$) in the same column, LSD=Least significant difference.

Minerals content of Kunun-zaki from Owerri Municipal

Table 3: Mean values of mineral composition of kunu zaki

| Sample | Calcium mg/100g | Potassium mg/100g | Magnesium mg/100g | Iron mg/100g | Vitamin C mg/100g |
|---------------------------------|---------------------------|---------------------------|--------------------------|-------------------------|--------------------------|
| Shellcamp kunun sample | 12.53 ^a ±17.72 | 399.08 ^c ±0.06 | 6.89 ^c ±0.01 | 2.22 ^c ±0.01 | 26.59 ^c ±0.01 |
| Ama-hausa kunun sample | 31.21 ^a ±0.01 | 311.63 ^d ±0.04 | 5.44 ^d ±0.02 | 1.19 ^d ±0.02 | 22.14 ^d ±0.02 |
| Obinze kunun sample | 27.31 ^a ±0.04 | 412.03 ^b ±0.04 | 8.09 ^b ±0.04 | 2.76 ^b ±0.01 | 27.32 ^b ±0.02 |
| Laboratory control kunun sample | 32.09 ^a ±0.04 | 449.03 ^a ±0.04 | 11.74 ^a ±0.05 | 4.81 ^a ±0.01 | 30.26 ^a ±0.03 |
| LSD | 8.860 | 0.045 | 0.032 | 0.015 | 0.022 |

Mean values with the different letters are significant ($p < 0.05$) in the same column, LSD= Least significant difference.

Microbiological quality of Kunun-zaki from Owerri Municipal

Table 4: Microbial Counts *cfu/mL* of the samples Analyzed

| Samples/count | THBC | TCC | TSSC | TSC | TFC |
|---------------------------------|-------------------|-------------------|-------------------|-------------------|-------------------|
| Shellcamp kunun sample | 5.5×10^7 | 5.2×10^5 | 0 | 6.0×10^5 | 3.0×10^5 |
| Ama-hausa kunun sample | 5.2×10^6 | 8.0×10^4 | 0 | 4.7×10^5 | 2.1×10^5 |
| Obinze kunun sample | 8.0×10^6 | 3.2×10^6 | 3.0×10^4 | 3.2×10^5 | 4.0×10^5 |
| Laboratory control kunun sample | 3.0×10^6 | 0 | 0 | 3.0×10^6 | 9.0×10^4 |

Key

THBC= Total heterotrophic bacteria count

TCC = Total *coliform* count

TSSC = Total *salmonella/shigella* count

TSC = Total *staphylococcus* count

TFC = Total *fungi* count

Table 5: Biochemical identification of pure culture of bacteria isolate

| Isolate Code | Gram Reaction | Catalase | Coagulase | Notifility | Indole | Citrate | Methyl red | Vogues Proskauer | Probable organism |
|--------------|---------------|----------|-----------|------------|--------|---------|------------|------------------|------------------------------|
| 1 | +ve Rod | + | - | + | - | + | - | + | <i>Bacillus sp</i> |
| A2 | -ve Rod | + | - | - | - | + | - | + | <i>Klebsiella sp</i> |
| A3 | +ve Cocci | + | + | - | - | + | - | + | <i>Staphylococcus aureus</i> |
| B1 | +ve Rod | + | - | + | - | + | - | + | <i>Bacillus sp</i> |
| B2 | -ve Rod | + | - | - | - | + | - | + | <i>Klebsiella sp</i> |
| B3 | +ve Cocci | + | + | - | - | + | - | + | <i>Staphylococcus aureus</i> |
| C1 | +ve Rod | + | - | + | - | + | - | + | <i>Bacillus sp</i> |
| C2 | -ve Rod | + | - | - | - | + | - | + | <i>Klebsiella sp</i> |
| C3 | +ve Cocci | + | + | - | - | + | - | + | <i>Staphylococcus aureus</i> |
| C4 | -ve Rod | + | - | + | - | + | + | - | <i>Salmonella sp</i> |
| D1 | +ve Rod | + | - | + | - | + | - | + | <i>Bacillus sp</i> |
| D2 | +ve Cocci | + | + | - | - | + | - | + | <i>Staphylococcus aureus</i> |