Microbial and Sensory Quality of Freshly Processed and Reconstituted "Kununzaki" - A Nigerian Millet Based Beverage

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Abstract

The microbial and sensory qualities of freshly processed and reconstituted Kununzaki beverages prepared from steeped millet grains were carried out. The samples were analysed for Aerobic plate, Fungi, Coliform, Staphylococcal, Salmonella and Shigella counts. The samples were also evaluated for difference and preference. The study showed that the reconstituted beverage had better microbiological quality with detectable difference between the two samples with the fresh sample being preferred.

Key words: Millet grains, Kununzaki, microbial quality, sensory quality.

Introduction
"Kunuzaki" is a non-alcoholic beverage made from millet grain. It is widely consumed in many parts of Northern Nigeria, especially during the dry season (Adeyemi and Umar, 1994). It can also be prepared from fermented sorghum, maize or a mixture of the three. It is sometimes referred to as "arrere" in most areas where it is consumed and usually retailed in empty table water bottles or in larger containers in comparison with carbonated drinks (Tahir and Oyawole, 1993). Kununzaki is very 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 (Agboola, 1987). The traditional process of manufacture involves wet-milling of millet or sorghum grains with water and spices (ginger, cloves, red and black pepper), followed by wet sieving, partial gelatinization of the slurry, sugar addition and bottling. (Onuorah et al., 1987). The traditional manufacture is crude as the basic processes are not standardized and levels of ingredients such as spices and mixtures are not quantified. Furthermore, a wide variation exists in the method of preparation depending on taste and cultural habit, which explains the lack of consistency in product quality. For instance, some people prefer "Kununzaki" with much pepper or sweet taste while some prefer it with little or no pepper or sugar (Adeyemi and Umar, 1994). "Kununzaki" is highly susceptible to spoilage because of its high moisture content. This gives it a short shelf life due to microbial activities. Adeyemi and Umar, (1994) and Bankole et al., (1999) reported a decrease in pH and an increase in titrable acidity of commercial sample of Kununzaki and "Kunutsamiya" (a related product manufactured also from millet) during storage. These changes were significantly higher for samples stored at ambient conditions than under refrigeration. (Onuorah et al., 1987).

In view of the short shelf life of freshly processed Kununzaki, the objective of this research is to produce Kununzaki powder that can be reconstituted as instant beverage (in order to reduce bulkiness thus making it easier for marketing ) and to prolong its shelf life. The microbiological and sensory qualities of the two samples are also herein investigated.

**Materials and Methods**

**Preparation of Fresh Kununzaki Beverage**

According to Adeyemi and Umar, (1994), 1 kg of millet grains were cleaned and steeped in twice its volume of water (2L) for 24 h. Thereafter the steeped grains were washed and spices added. The amount of spices (w/w) added were 0.65% ginger, 0.25% red pepper, 0.05% cloves and 0.05% black pepper. The steeped millet grains and spices were then wet milled and wet sieved to remove the shafts after which the supernatant was decanted from the slurry. The slurry was divided into two equal halves with one half added to boiling water while stirring for 2mins., cooled to a temperature of 35 ± 2°C and subsequently added to the remaining half slurry. The mixture was
sweetened with 8% (w/v) granulated sugar and mixed properly to obtain the freshly processed Kununzaki beverage.

**Preparation of Reconstituted Kununzaki Beverage**

"Kununzaki" was prepared according to Adeyemi and Umar, (1994) without sweetening. The resultant mixture was centrifuged at 583 revs⁻¹ followed by decanting to remove excess water. The paste obtained was dried in a cabinet dryer at 60° ±2°C for 18 h. The resultant flakes were milled and sieved (0.25 mm mesh) to obtain dry Kununzaki powder (fig. 1). Kununzaki beverage was then prepared from the dry Kununzaki powder by reconstituting with water (1:10) and sweetening at 8% (w/v) with sugar.

**Moisture Content and pH determination**

Moisture content of fresh and reconstituted `Kununzaki` beverages were determined using the method of AOAC, (1984). The pH of the samples was determined using the method described by Pearson, (1991) after standardizing with buffer solutions at pH 4 and 7.

**Microbiological Quality**

The aerobic plate counts were determined by diluting the fresh and reconstituted Kununzaki beverages decimally and plating 1ml of the aliquots separately on triplicate plates of Nutrient agar (Oxoid) and incubating at 28° ± 2°C for 48h.

The Fungi counts were determined by plating 1 ml aliquots of the samples on acidified Potato Dextrose Agar (Oxoid) and incubating at room temperature (28 ± 2°C) for 48h. Observed colonies were subcultured to obtain pure cultures which were incubated for 5-7 days, subsequently isolated and identified using standard methods (Barnett and Hunter, 1972). The presumptive Coliform counts were determined by plating 1ml of the serially diluted samples on MacConkey agar No 3 (Oxoid) and incubating at 35 ±2°C for 24h. Confirmation was by fermentation of Lactose and Indole at 42 ±2°C after 24h incubation.

Salmonella and Shigella counts were determined by inoculating the SS agar (Oxoid) plates heavily with the specimens and incubating at 35 ±2°C for 24 h. Non lactose fermenters form colourless colonies while lactose fermenters which produced pink or red colonies were counted.
Presumptive Staphylococcal counts were determined by inoculating Staphylococci medium 110 (Oxoid) with the specimens and incubating at 30 ±2°C for 72h. Pigmented colonies surrounded by bright yellow zones (halo) resulting from mannitol fermentation were counted. Confirmation was by positive coagulase test.

**Sensory Quality**

Triangle test was used to differentiate the samples. Coded Kununzaki beverages were served to 10 judges. Each judge received three coded samples, two being similar and one odd. The judges were asked to identify the odd samples. The difference analysis was then carried out using the MSTAT computer package. The preference test was carried out using a 9 point Hedonic scale (where 1 and 9 represent dislike extremely and like extremely respectively). The Kununzaki beverages were presented to a 10-member panel consisting of people who are familiar with the product. The mean scores received for each beverage was compared using t-test (Ihekoronye and Ngoddy, 1985).

**Results and Discussion**

**Moisture and pH**

The mean moisture content of the fresh Kununzaki beverage was 89.27±2% and 8.66 ± 0.8 % for the reconstituted powder. This indicates that the ratio of moisture in both samples is approximately 10: 1 respectively. The average pH of the fresh sample was 3.15 ± 0.1 while that of the reconstituted sample was 3.91± 0.2. There was less acidity in the reconstituted sample because of water removal (Adeyemi and Umar, 1994).

**Microbiological Quality**

The mean Aerobic plate, Fungi, Coliform Staphylococci, Salmonella and Shigella Counts (cfu/ml) for fresh and reconstituted beverages are shown in [table 1](#). The Aerobic plate counts for fresh Kununzaki beverage ranged from 3.53 x 10^4 to 5.98 x 10^4 with a mean of 4.42 x 10^4 (cfu/ml). The Aerobic plate counts for the reconstituted Instant beverage ranged from 0.46 x 10^4 to 2.8 x 10^4 (cfu/ml) with a mean value of 1.60 x 10^4 which was lower to that of fresh Kununzaki. This was due to the low moisture content of the reconstituted powder from which the beverage was made as a result of the drying process to which the Kununzaki paste was subjected to. The drying process would have destroyed some microorganisms while the low moisture content of the reconstituted powder would
have restricted further microbial growth since micro-organisms do not thrive well in an environment of low water activity (Ihekoronye and Ngoddy, 1985).

The Fungi counts ranged from $1.20 \times 10^4$ to $1.70 \times 10^4$ (cfu/ml) with a mean of $1.48 \times 10^4$ for the fresh beverages, while that of the reconstituted samples were $0.3 \times 10^4$ to $0.55 \times 10^4$ (cfu/ml) with a mean of $0.42 \times 10^4$. The low incidence of fungi in the reconstituted beverage was presumed to be due in part to destruction of some of the fungi during the drying process and inhibition of fungi growth in the dry powder as a result of low water activities. The different types of moulds isolated from fresh and reconstituted Kununzaki beverages are shown in Table 2. The presence of fungi in a product is undesirable and in particular, the species of *A. flavus* and *A. niger*. Toxigenic strains of *A. flavus* have been known to produce aflatoxin, a potent hepatotoxic and carcinogenic agent (Uraih and Ogbadu, 1980). *A. niger* is also known to produce protocathenic and oxalic acids which are toxic metabolites (Avdesh and Prakash, 1968).

Coliform counts ranged from $2.25 \times 10^3$ to $3.0 \times 10^3$ (cfu/ml) with a mean of $2.63 \times 10^3$ (cfu/ml) for the fresh beverage, while for the reconstituted beverage, the range was $0.70 \times 10^3$ to $0.83 \times 10^3$ with a mean of $0.76 \times 10^3$. The presence of Coliforms in the Kununzaki beverage is an indication of faecal contamination. It may also be due to the contamination from water and other ingredients used in processing. In particular, it had been shown from previous reports that the microbiological quality of water supplies to some communities in Nigeria is poor with Coliform counts exceeding recommended level (Tahir and Oyawole, 1993). The lower Coliform counts obtained in the reconstituted beverage may be due to the fact that Coliform bacteria is thermolabile and most of the Coliforms might have been destroyed by heat during the process of drying the Kununzaki paste. Also, the mean Staphylococci counts in the fresh beverage was $0.2 \times 10^1$ while none was isolated from the reconstituted beverages. This may be due to the fact that staphylococcus bacteria is also easily destroyed by heat. No Salmonella and Shigella species were isolated from both fresh and reconstituted beverages indicating that they are not health hazards in these beverages.

**Sensory Quality**

Difference test conducted on the fresh and reconstituted beverages using the triangle test, showed that there was a detectable difference between the two samples at both 1% and 5% level of probabilities. Table 3 shows the mean sensory scores for fresh and reconstituted Kununzaki beverages using the 9-point Hedonic scale. The preference test showed that there was significant difference in the level of preference of the two beverages and that the fresh
beverage was preferred by the judges at 5% level of probability. There was no significant difference between the fresh and reconstituted beverages for colour and sweetness at (P< 0.05). The differences in aroma and spiciness may be due to loss of volatile components while drying (Hui, 1992, Adeyemi and Umar, 1994) while the difference in consistency may be due to poor reconstitution of Kununzaki powder. Thus the reconstituted Kununzaki beverage had better microbiological quality than the fresh Kununzaki beverage while the sensory quality of the reconstituted sample was poorer.

Acknowledgement

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References


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Fig 1: Flow chart for production of dry Kununzaki powder

Millet grains

- Cleaning
- Steeping
- Wet milling
- Wet-sieving
- Settling
- Decanting

Slurry

- Spices
- Overtails
- Supernant

Half slurry added to boiled water with stirring for 2mins.

- Cooled to (32 ± 2°C)

Mixing

- Centrifuging

Decanting

- Water

Paste
Dry Kununzaki powder

- Sieving (0.25mm) mesh
- Milling
- Flakes
- Drying
Table 1: Mean Microbial counts (cfu/ml) for Fresh and Reconstituted Kununzaki Beverages +.

<table>
<thead>
<tr>
<th>Kununzaki Samples +</th>
<th>Aerobic Plate Counts</th>
<th>Fungi Counts</th>
<th>Coliform Counts</th>
<th>Staphylococci Counts</th>
<th>Salmonella and Shigella Counts</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh</td>
<td>4.42 x 10^4</td>
<td>1.48 x 10^4</td>
<td>2.6 x 10^3</td>
<td>0.2 x 10^1</td>
<td>&lt;10*</td>
</tr>
<tr>
<td>Reconstituted</td>
<td>1.60 x 10^4</td>
<td>0.42 x 10^4</td>
<td>0.76 x 10^3</td>
<td>&lt;10</td>
<td>&lt;10</td>
</tr>
</tbody>
</table>

+ = Kununzaki Samples were analysed in triplicates
* = less than the minimum number that can be counted using the methodology applied.
**Table 2. Moulds isolated from Fresh and Reconstituted Kununzaki Beverages.**

<table>
<thead>
<tr>
<th>Fresh Kununzaki Beverage</th>
<th>Reconstituted Kununzaki Beverage</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Aspergillus flavus</em>. Link</td>
<td><em>A. niger</em>. van Tieghem</td>
</tr>
<tr>
<td><em>A. niger</em>. van Tieghem</td>
<td><em>A. terrus</em>. Thom</td>
</tr>
<tr>
<td><em>A. terrus</em>. Thom</td>
<td><em>A. flavus</em>. Link</td>
</tr>
<tr>
<td><em>Mucor racemosum</em>. Thom</td>
<td></td>
</tr>
</tbody>
</table>
| *Neurospora sitophilla*. Sheer and Dodge | }
**Table 3: Mean sensory scores for Fresh and Reconstituted Kununzaki Beverages**

<table>
<thead>
<tr>
<th>Kununzaki</th>
<th>Colour</th>
<th>Aroma</th>
<th>Sweetness</th>
<th>Spiciness</th>
<th>Consistency</th>
<th>Overall Acceptability</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh</td>
<td>7.8&lt;sub&gt;a&lt;/sub&gt;</td>
<td>7.6&lt;sub&gt;a&lt;/sub&gt;</td>
<td>7.7&lt;sub&gt;a&lt;/sub&gt;</td>
<td>7.1&lt;sub&gt;a&lt;/sub&gt;</td>
<td>7.7&lt;sub&gt;a&lt;/sub&gt;</td>
<td>7.9&lt;sub&gt;a&lt;/sub&gt;</td>
</tr>
<tr>
<td>Reconstituted</td>
<td>7.5&lt;sub&gt;a&lt;/sub&gt;</td>
<td>6.3&lt;sub&gt;b&lt;/sub&gt;</td>
<td>7.1&lt;sub&gt;a&lt;/sub&gt;</td>
<td>6.4&lt;sub&gt;b&lt;/sub&gt;</td>
<td>6.0&lt;sub&gt;b&lt;/sub&gt;</td>
<td>6.9&lt;sub&gt;b&lt;/sub&gt;</td>
</tr>
</tbody>
</table>

Means with the same subscript letters are not significantly different at P ≤ 0.05