Anti–Nutrients And Physicochemical Properties Of A Defatted Moringa Seed Kunun–Zaki Sweetened With A Natural Sweetener Serendipity Berry (*Dioscoreophyllum Cumminsii*)

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ABSTRACT

This study aimed at investigating the anti-nutrients and physicochemical properties of a defatted moringa seed kunun-zaki sweetened with serendipity berry (*Dioscoreophyllum cumminsii*). The natural sweetener berry was obtained from a farm sorted and washed. The fleshy parts were removed from the seed in the fruits, dried and ground into powder. Millet kunun-zaki was prepared with defatted moringa seed powder. kunun-zaki of 250 ml was sweetened with serendipity berry and sucrose (control) at varied ratios of 1.0g, 2.0g, 3.0g, 4.0g and 5.0g respectively wherein defatted moringa powder of 1.5g was used to enrich the sweetened kunun-zaki. Standard assay methods were used to analyse the anti nutrients and physicochemical properties of the samples namely: kunun-zaki sweetened with serendipity berry 0-5g (STK0, STK1, STK2, STK3, STK4 and STK5) and sucrose (MKG1, MKG2, MKG3, MKG4 and MKG5).
The results revealed that the anti-nutrients composition of kunun-zaki sweetened with serendipity berry had oxalate, tannin, alkaloid, phytate and cyanide ranged between (0.03-0.10mg/100g), (6.00-8.70 mg/100g), (0.03-0.11 mg/100g), (0.38-0.40 mg/100g) and (1.30-1.34 mg/100g) respectively. Also, kunun-zaki sweetened with sucrose had (0.07-.0.09 mg/100g), (0.06-0.07 mg/100g), (1.81-1.89 mg/100g), (7.66-7.74 mg/100g) and (0.08-0.09 mg/100g) for oxalate, tannin, alkaloid, phytate and cyanide respectively. The results of physicochemical contents showed that total solid, titratable acidity, pH, brix value and specific gravity of kunun-zaki sweetened with serendipity berry and sucrose had the following values (5.67-7.74%), ( 0.07-0.09), (6.20-6.63), (4.00-10.10) and ( 1.00-1.04mg/m³) and sucrose ( 3.01-4.40%), ( 0.01-0.02), (6.09-6.23), (2.61-4.01) and (1.01-1.07mg/m³) respectively. The brix value present in this sweetener is protein base. The protein is the sweetest known naturally occurring substance up to 3,000 times sweeter than sucrose and approximately 100,000 times as potent sugar

Introduction

For many years the Nigerian soft drink industry has been heavily dependent on imported raw materials in order to conserve foreign exchange and emphasis is now on the development of indigenous beverages and the country’s attention has begun to shift toward the local sourcing of raw materials for economic development (Ahmed, et al., 2003). kunun-zaki is an indigenous fermented beverage made from un sprouted cereal grains (Adeyemi and Umar, 1994). The drink has its origin in the northern parts of Nigeria but is now popular in almost all the states in Nigeria (Gaffa, et al., 2002a). Cereal grains form a major source of dietary nutrients for all people, particularly those in the developing countries (Akpapunam, et al. 1997). However, the nutritional quality of cereal grains and sensory properties of the products are inferior due to
lower protein contents, deficiency of certain essential amino acids, lower protein and starch availabilities, presence of certain antinutrients and coarse nature of the grains (Adeyemi and Umar 1994). Kunun-zaki processing is mostly done by women using simple household equipment and utensils depending on cereal availability, the unsprouted cereal grains used for kunun-zaki processing are sorghum, maize, millet and rice is mostly non composite proportions. Though, kunun-zaki is popular and fast becoming a household technology in the country, the cereal grains used are selected randomly and diversely. For instance, some women use millet only or sorghum only, while others use only maize. Some also use sorghum with rice, while others use millet with sorghum (Gaffa et al., 2002a). The most popular cereals used are sorghum and millet. When kunun-zaki is made from sorghum the final product is a light-brown liquid. When made from millet the resulting liquid product is milky white (Gaffa, et al., 2002b).

Serendipity berry fruit (*Dioscoreophyllum cumminsii*) is a tropical rain forest vine that belongs to the family menispermaccae. It grows in a dense humid tropical forest region of west and central Africa (Inglett and May, 1969: Oselebe and Nwankiti, 2005). In Nigeria the plant grow in the relatively undisturbed rainfall areas of southern Nigeria. The intense sweetener of the fruit of *Dioscoreophyllum cumminsii* called serendipity berry. The serendipity berry was revealed to be a protein (Inglett, 1976). Monellin is a sweet protein found in the fruit of serendipity berry (*Dioscoreophyllum cumminsii*) (Abiodun and Akinoso, 2014). The protein is the sweetest known naturally occurring substance up to 3,000 times sweeter than sucrose and approximately 100,000 times as potent sugar on a molar basis (Inglett, 1976 and Faruya, et al., 1983, Penarrubia, et al., 1992). Monellin could thus replace sugar in foods for diabetics and dieter (Oselebe and Nwankiti, 2005).
A strong and rapidly growing *Moringa oleifera* Lamarck (fam. Moringaceae) tree is widely cultivated due to its high adaptability to environmental conditions. It’s considered as one of the most useful trees in the world because almost all parts of this plant can be used as in food, in medicines and for industrial purposes (Anwar *et al.*, 2007). In many countries, there are huge efforts to spread the use and cultivation of *M. oleifera*, since it is a significant source of fats, proteins, beta-carotene, vitamin C, iron, potassium, and other nutrients with low toxicity of seeds and leaves. For these reasons, some parts of this plant have drawn much attention and have been studied for its various biological activities, including anti-atherosclerotic, immune-boosting, anti-cardiovascular diseases, antiviral, antioxidant and antimicrobial, anti-inflammatory (Kumar, *et al.*, 2013). Properties and tumor suppressive effects in skin papillomagenesis, hepatocarcinoma cancer, colon cancer, and myeloma (Khalafalla *et al.*, 2010).

This study aimed at evaluating the anti-nutrients and physicochemical properties of kunun-zaki enriched with defatted moringa seed flour and sweetened with a natural sweetener serendipity berry (*Dioscoreophyllum cumminsii*).

**Materials and methods:**

**Materials:** Cereal grains millet (*Pennisetum typhoideum*) and *Moringa oleifera* seeds were obtained from Institute of Agricultural Research and Training (IAR&T) Moor Plantation, Ibadan, Oyo State. Sweetener serendipity berry (*Dioscoreophyllum cumminsii*) was obtained from a farm at Esa-odo, Osun state.

**Methods:** Processing of defatted moringa seeds and serendipity berry: Moringa seeds were sorted to remove the defected ones and the sorted seeds were dehulled to remove the seed coat.
The seeds were dried, grinded into powder and wrapped with filter paper. Thereafter, it was placed inside ethanol for 3 days, dried and packaged for further use. **Processing of serendipity berry:** The sorted fruits were washed. The seeds were separated from the fleshy parts. The fleshy parts were dried and milled into powder and the product obtained was stored for further use. The method of Gaffa and Ayo, (2002) was adopted for processing of kunun-zaki. The quantities of kunun-zaki and defatted moringa seed flour was constant (250ml and 1.5g) but that of the sweetener was varied (1.0g, 2.0g, 3.0g, 4.0g and 5.0g). Also sucrose was used to sweeten kunun-zaki which serves as control. (1.0g, 2.0g, 3.0g, 4.0g and 5.0g).

**Analyses:** Anti-nutrients and physicochemical evaluation: Anti-nutritional content such as oxalate, phytate, alkaloid, tannin and cyanide were carried using Dairo (2001) and Sahore and Amani (2012). Physicochemical properties (pH, titratable acidity, total solid, brix values and specific gravity,) were determined using AOAC (2006)

**Statistical Analysis:** All determinations were carried out in triplicate. Data were subjected to statistical analysis using SPSS 16.0 and means were separated using Duncan multiple Ranges Test (DMRT)

**Results and discussion:**

**Table 1: Anti–nutritional contents of defatted moringa seed kunun-zaki sweetened with serendipity berry**

<table>
<thead>
<tr>
<th>Sample (g)</th>
<th>Oxalate (g/100g)</th>
<th>Tannin (g/100g)</th>
<th>Alkaloid (g/100g)</th>
<th>Phytate (g/100g)</th>
<th>Cyanide (g/100g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>KS</td>
<td>0.03±3.38&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>6.00±4.81&lt;sup&gt;c1&lt;/sup&gt;</td>
<td>0.03±3.26&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>0.45±3.32&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.30±5.01&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>
Table 2: Anti-nutritional contents of defatted moringa seed kunun-zaki sweetened with sucrose

<table>
<thead>
<tr>
<th>Sample</th>
<th>Oxalate (mg/100g)</th>
<th>Tannin (mg/100g)</th>
<th>Alkaloid (mg/100g)</th>
<th>Phytate (mg/100g)</th>
<th>Cyanide (mg/100g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MKG_1</td>
<td>0.09±2.45&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>0.06±3.70&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.87±5.10&lt;sup&gt;e&lt;/sup&gt;</td>
<td>7.66±5.57&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>0.09±2.26&lt;sup&gt;cd&lt;/sup&gt;</td>
</tr>
<tr>
<td>MKG_2</td>
<td>0.08±1.23&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.07±3.67&lt;sup&gt;d&lt;/sup&gt;</td>
<td>1.89±5.45&lt;sup&gt;d&lt;/sup&gt;</td>
<td>7.78±3.99&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.09±2.63&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td>MKG_3</td>
<td>0.08±2.56&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.06±2.78&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>1.89±4.57&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>7.74±2.18&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.09±3.44&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>MKG_4</td>
<td>0.07±2.58&lt;sup&gt;de&lt;/sup&gt;</td>
<td>0.06±4.14&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>1.85±4.20&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.70±3.81&lt;sup&gt;de&lt;/sup&gt;</td>
<td>0.09±2.87&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>MKG_5</td>
<td>0.07±2.66&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.06±3.56&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>1.81±3.44&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.70±2.34&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.08±3.85&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

The oxalate content in the kunun-zaki with serendipity berry and sucrose are respectively in (Tables 1 and 2). Oxalate is an anti-nutritional factor which interferes with metabolic processes, so that growth and bioavailability of nutrients are negatively influenced. These factors stand as indices for judging the nutritional value of any given food substance (Binita and Khtarpaul, 1997). The oxalate content of the kunun-zaki with serendipity berry for samples KS_0 and KS_1
(0.03 and 0.07mg/100g) are significantly not different while samples KS$_3$ and KS$_4$ (0.09 and 0.09mg/100g) are also not significantly different. Sample KS$_5$ (0.10mg/100g) is significantly different from every other samples but the values significantly reduced as the quantity of sucrose added to kunun-zaki increased. Samples MKG$_1$ had the highest value (0.09mg/100g), MKG$_2$ and MKG$_3$ (0.08mg/100g) had the same values while samples MKG$_4$ and MKG$_5$ had the same values (0.07mg/100g). However the reported value for the oxalate obtained in this present study was lower than the established toxic level of 2-5g (Birgitta and Caroline, 2000). The lower values of oxalate contents obtained in both kunun-zaki sweetened with serendipity berry and sucrose could be connected with the fact that oxalates are water soluble and heat labile since kunun-zaki undergoes soaking (fermentation) and cooking. According to Ladeji, et al., (2004) oxalate can bind to calcium in food thereby rendering calcium unavailable for normal physiological and biochemical roles such as the maintenance of strong teeth, nerve impulse transmission as clotting factor in blood and cofactor in enzymatic reactions. Tannin content in kunun-zaki with serendipity berry ranged between (6.00-8.70mg/100g). There were no significant difference ($P > 0.05$) between the samples and the Concentration on of tannin increased in kunun-zaki with serendipity berry. Tannin concentration in kunun-zaki with sucrose ranged between (0.06-0.07mg/100g) and the concentration was lower compared to the kunun-zaki with serendipity berry. This is in agreement with report of Olosunde, et al (2014) who reported a decrease in tannin content of kunun-zaki enriched with moringa seed flour. Tannins inhibits the activities of some enzymes like trypsin, amylase and lipase by forming insoluble complexes with protein and divalent ions such as Fe$^{2+}$ and Zn$^{2+}$ thereby reducing their absorption in the body (Elegbede, 1998). Alkaloid content of samples of kunun-zaki with serendipity berry and sucrose had the values between (0.03-0.85mg/100g) and (1.81-1.89g/100g). The contents of
alkaloid did not differ significantly in both set of samples of kunun-zaki (serendipity berry and sucrose). The sample with sucrose had higher values of alkaloid compared with samples with serendipity berry. However, the two set of samples had higher values of alkaloid contents than alkaloid in mango juice 0.01mg/100g (Fawomola, 2010). High alkaloid contents cause toxicity when ingested by human being (Abiodun, et al., 2014). Phytate contents of kunun-zaki with serendipity berry and sucrose were (0.45, 0.38, 0.38, 0.40, 0.40, 0.40 and 0.40 g/100g) and (7.66, 7.78, 7.74, 7.70 and 7.70 g/100g). No significant differences (P< 0.05) were observed between the two set of samples of kunun-zaki (serendipity berry and sucrose). The values recorded for both the set of samples of kunun-zaki (serendipity berry and sucrose) were lower relative to 8.25and 4.25mg/100g reported by white sesame seed and black millet respectively (Odunmolu, 1992). This could be attributed to the types of processing (fermentation) and the moisture content of the samples (Oladele, et al., 2009). Khan, et al., (1990) stated that the higher the moisture content, the higher the phytate loss. Fermentation has been reported to be the most effective in reducing phytates (Fagbemi, et al., 2005). The cyanide contents of kunun –zaki with serendipity berry and sucrose had values which ranged from (1.28-1.34mg/100g) and (0.08-0.09mg/100g). The results obtained in this present study indicated that the cyanide levels found was low. The lethal dose range for humans of cyanide ingested is estimated to be only 0.5-3.5mg/kg body weight (Bradbury, 1991). Therefore, this beverage could be free from cyanide toxicity since it has values lower than the established toxic level.
Table 3: Physicochemical composition of defatted moringa seed kunun-zaki sweetened with serendipity berry

<table>
<thead>
<tr>
<th>Sample (g)</th>
<th>Total solid (%)</th>
<th>Titratable acidity</th>
<th>pH</th>
<th>Brix value</th>
<th>Specific gravity (mg/m³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>KT₀</td>
<td>5.67±2.03</td>
<td>0.08±2.10</td>
<td>6.20±2.67</td>
<td>4.00±3.66</td>
<td>1.00±3.88</td>
</tr>
<tr>
<td>KT₁</td>
<td>6.28±3.14</td>
<td>0.09±3.14</td>
<td>6.36±3.07</td>
<td>6.30±3.20</td>
<td>1.04±3.34</td>
</tr>
<tr>
<td>KT₂</td>
<td>6.93±2.19</td>
<td>0.08±2.81</td>
<td>6.40±3.14</td>
<td>8.75±4.15</td>
<td>1.04±4.05</td>
</tr>
<tr>
<td>KT₃</td>
<td>7.04±1.33</td>
<td>0.09±2.02</td>
<td>6.45±4.25</td>
<td>9.25±2.57</td>
<td>1.04±2.47</td>
</tr>
<tr>
<td>KT₄</td>
<td>7.25±3.32</td>
<td>0.09±2.43</td>
<td>6.35±2.58</td>
<td>9.30±3.60</td>
<td>1.04±6.57</td>
</tr>
<tr>
<td>KT₅</td>
<td>7.74±1.37</td>
<td>0.07±2.13</td>
<td>6.63±1.07</td>
<td>10.10±5.45</td>
<td>1.04±4.20</td>
</tr>
</tbody>
</table>

Table 4: Physicochemical composition of defatted moringa seed kunun-zaki sweetened with sucrose

<table>
<thead>
<tr>
<th>Sample (g)</th>
<th>Total solid (%)</th>
<th>Titratable</th>
<th>pH</th>
<th>Brix value</th>
<th>Specific gravity ±(mg/m³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MKG₁</td>
<td>3.80±1.23</td>
<td>0.01±3.12</td>
<td>6.23±3.65</td>
<td>2.61±3.65</td>
<td>1.06±3.63</td>
</tr>
<tr>
<td>MKG₂</td>
<td>3.82±3.38</td>
<td>0.01±3.44</td>
<td>6.09±3.63</td>
<td>2.82±2.75</td>
<td>1.07±3.63</td>
</tr>
<tr>
<td>MKG₃</td>
<td>3.01±2.25</td>
<td>0.01±2.57</td>
<td>6.09±3.78</td>
<td>3.01±3.29</td>
<td>1.01±5.50</td>
</tr>
<tr>
<td>MKG₄</td>
<td>4.40±3.76</td>
<td>0.02±2.40</td>
<td>6.09±4.44</td>
<td>4.01±4.66</td>
<td>1.07±3.46</td>
</tr>
<tr>
<td>MKG₅</td>
<td>4.40±3.00</td>
<td>0.02±4.52</td>
<td>6.09±5.63</td>
<td>4.01±2.42</td>
<td>1.07±2.62</td>
</tr>
</tbody>
</table>
Tables 3 and 4. Shows the values of the physicochemical composition of kunun-zaki with serendipity berry and sucrose. In Table 3, the total solid increased steadily as the quantity of serendipity berry increased (5.67-7.74%) such a trend was also reported for kunun-zaki enriched with moringa seed flour (Olosunde, et al., 2014). Table 4(3.01-4.40%), the total solid for kunun-zaki with sucrose had lower values compared to kunun-zaki with serendipity berry this could be as a result of the presence of serendipity berry since total solid is an index of the cloudiness. Total solid in juice are sugars, organics and pectins. Also the contents of the dry matter and solids are important quality attributes and characteristics that determines the taste of a drink (Wakowiak-Tomezak, et al., 2008). The total titratable acidity of kunun-zaki (serendipity berry and sucrose) was also low ranging from (0.07-0.09) and (0.01-0.02). The pH of kunun-zaki with serendipity berry and sucrose are (6.20-6.62) and (6.03-6.09). However, the pH levels of the samples were alkaline, which is still within the standard level of pH for kunun-zaki. Brix value (sugar content) is a good indicator of consumer appraisal in relation to the beverage quality (Abiodun and Akinoso, 2014). The brix values of kunun-zaki with serendipity berry and sucrose are as follows, (4.00, 6.30, 8.75, 9.25, 9.30 and 10.10) and (2.61, 2.82, 3.01, 4.01, and 4.01). There were higher values of brix value in kunun-zaki with serendipity berry than the values in kunun-zaki with sucrose. The protein in serendipity berry is the sweetest known naturally occurring substance up to 3,000 times sweeter than sucrose and approximately 100,000 times as potent sugar on a molar basis (Inglett, 1976 and Faruya, et al., 1983, Penarrubia et al.). Although, they both followed the same trend as the serendipity berry and sucrose increases the values of the brix value increased for the two set samples. Specific gravity of all the samples
were significantly different (P < 0.05). The specific gravity ranged between (1.00-1.04mg/m^3) for kunun-zaki with serendipity berry and (1-01-1.07 mg/m^3) for kunun-zaki with sucrose.

**Conclusion**: The results obtained in this study showed that the anti-nutritional factors in kunun-zaki sweetened with serendipity berry and sucrose were significantly low. Kunun-zaki with serendipity berry had appreciable amount of brix value (sugar content) and the sugar present in this sweetener is protein base. It has low anti-nutritional factors and therefore, could not pose health hazard to consumer. Thus, it can replace sugar in foods for diabetics and dieter. Also, the sweetener compared relatively well with sucrose (sugar) used in the beverage industries.

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