



## Comparative microbiological and physicochemical evaluation of different blends of tiger nut (*Cyperus esculentus*) and date (*Phoenix dactylifera* L.) beverages

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### ABSTRACT

**Background:** The rising cost of animal milk has led to studies being carried out on the formulation of plant-based milk products which can compare favourably in terms of nutritional content and general acceptability to consumers. **Objectives:** Hence, the objective of this study was to evaluate and compare the microbiological and physicochemical properties of different blends of tiger nuts and date juice. **Methods:** Fresh tiger nuts and dates were processed to formulate five beverage blends in these ratios 100:00 (A), 30:70 (B), 50:50 (C), 70:30(D), and 0:100 (E). The physicochemical parameters (pH, moisture content, total soluble solids (TSS), total titratable acidity (TTA), and carbohydrates) of the tiger nut-date blends were determined. **Results:** The values for the physicochemical parameters obtained are as follows; pH (6.20-6.51), Moisture content (87.11-89.21), TTA (0.021-0.034), TSS (10.79-12.89). Carbohydrates (12.11-13.61). Total plate counts of the beverage ranged from 0.1 to  $1.0 \times 10^3$  CFU/mL which is below the microbial limit of acceptance of  $2.0 \times 10^5$  CFU mL<sup>-1</sup>. The microorganisms isolated from the beverage are *Escherichia coli*, *Bacillus subtilis*, and *Staphylococcus aureus*. In addition, the 50:50 tiger nut-date beverage has the highest overall acceptance by the panelists compared to other blends. **Conclusions:** The tigernut-date beverage (50:50 tigernut-date blend) produced in this study could serve as an alternative to milk and milk products.

**Keywords:** Tiger nut; Date; Beverage; Moisture content; Sensory properties

### 1. INTRODUCTION

Beverages are generally drinks other than crops to develop beverages with good sensory water. The deficiency of necessary nutrients acceptance and nutritional value (Singh *et al.*, like fibre, protein, minerals, and vitamins in 2022).

carbonated drinks led to an improvement in the Tiger nut (*Cyperus esculentus*) is a root tuber development of alcoholic and non-alcoholic belonging to the family *Cyperaceae*. This tuber beverages. The increase in the consumption of has a unique name in different locality such as these beverages is based on their nutritive “aya” in Hausa; “aki awusa” in Igbo; “ofio” in value, flavour, and aroma (Eke-Ejiofor and Yoruba and “isipaccara” in Effik (Adedokun Beleya, 2018). However, many of these *et al.*, 2022). The slender rhizomes of tiger nut beverages are not self-sufficient in their form weak runners above the ground level that nutritional contents hence efforts are geared develop small-sized tubers at the tip of the towards identifying and harnessing the stem. Tiger nut tubers can reach about six nutritional potentials of some underutilized inches depth into the soil (Alam *et al.*, 2014).

Tiger nuts are consumed for their sweetness and nutritive value with significant proportion of protein, carbohydrate, sugars, lipid, dietary fibre and some specific minerals (Zilic *et al.*, 2022). Furthermore, some health benefits have been attributed to the consumption of tiger nuts and these include; treatment and prevention of colon cancer, heart disease, obesity, diabetes, and gastrointestinal disorders (Adejuyitan *et al.*, 2009). Due to increasing knowledge about the nutritive and health benefits of tiger nuts, several researchers have reported their potential applications in a number of consumer products made for the gastronomic, pharmaceutical/ medicinal, confectioneries, and the bio-fuel industries (Obinna-Echem and Torporo, 2018). Date (*Phoenix dactylifera* L.) is a flowering plant species in the palm family, *Arecaceae*, typically cultivated for its edible sweet fruit. The significance of the date in human nutrition predicated on its rich composition of carbohydrates, minerals, dietary fiber, vitamins, fatty acids, amino acids and protein (Hassan *et al.* 2016). Date is also an excellent material for producing refined sugar, concentrated juice, confectionery pastes and fermentation products (Ghazal *et al.*, 2016). Also, dates have strong antioxidant, anticancer and antiviral activities, as well as reduced risk of several chronic diseases such as coronary heart disease, cardiovascular disease, aging, atherosclerosis and neurodegenerative diseases (Al Harthi *et al.* 2015). Tiger nut and date fruits are highly nutritious crops that are gaining increasing attention due to their numerous health benefits. They are rich sources of minerals, dietary fibre, vitamins and

antioxidants, which make them suitable in food and beverage production. The combination of tiger nut milk and date milk could result in a healthier beverage rich in antioxidants, vitamins, and minerals with several health benefits (Chinedu, *et al.*, 2022). Hence there is need to carry out a comparative analysis of the microbiological, physiochemical and sensory properties of different blends of tiger nut milk and date juice.

## **2. Materials and Methods**

### **2.1. Collection of sample**

Samples of fresh tiger nut (*Cyperus esculentus*) and date (*Phoenix dactylifera* L.) were purchased from Agbara market, Ogun State, Nigeria

### **2.2. Preparation of Sample**

The fresh tiger nuts and dates were sorted separately to remove unwanted materials that may affect the quality of beverage produced. The sorted tiger nuts and dates were washed thoroughly with distilled water to further remove dirt and debris.

### **2.3. Production of tiger-nut milk**

The extraction of tiger nut milk was carried out using the method described by Udeozor (2012) with little modification. The sorted tiger nuts were washed thoroughly with distilled water to further remove dirt and debris. The nuts were soaked in distilled water at ratio 1:3 of the nuts to the distilled water for 12 hours to soften them. The soaked nuts was milled using a warming blender and the slurry sieved to extract the milk. The extracted milk was transferred into sterile bottle and kept in the freezer for further analysis.

## 2.4. Production of Date Juice

The sorted date fruit was further washed to remove dirt and other extraneous materials. The flesh and the seed of the fruit were further separated by manual peeling. The date flesh was blended to a fine paste using a warring blender and later centrifuged (Sanchez-Zapata *et al.*, 2011)

## 2.5. Production of Tigernut- Date Beverage

Five different blends of Tigernut-Date beverage in the ratio 100:0 (A), 70:30 (B), 50:50 (C), 30:70 (D), and 0:100 (E) were prepared. The appropriate ratio of the tiger-nut and date was prepared and milled using a warring blender to obtain a homogenized beverage. Each of the blend is prepared with precision to ensure accurate ratios and consistency. The resulting tiger nut and date juice blends were pasteurized at 65°C for 30 minutes. (Figure 1). The beverage was subjected to microbiological, sensory and physicochemical analysis.

## 2.6 Isolation and Identification of Bacteria Associated with The Tiger-nut and Date Beverage

Microorganisms associated with tiger nut-date beverage were isolated and cultured on Nutrient agar and MacConkey agar (BDH Chemicals, UK) for bacteria isolation and enumeration. Identification of the bacteria isolates were identified according to the methods of Cowan and Steel (1993) for bacteria identification.

## 2.7. Determination of Physicochemical Properties

Physicochemical properties of the tiger nut-date beverage such as moisture content, pH, Carbohydrates, Total soluble solids (TSS) and Total titratable acidity (TTA) were carried out according to the methods of Association of Official Analytical Chemists (AOAC, 2012).

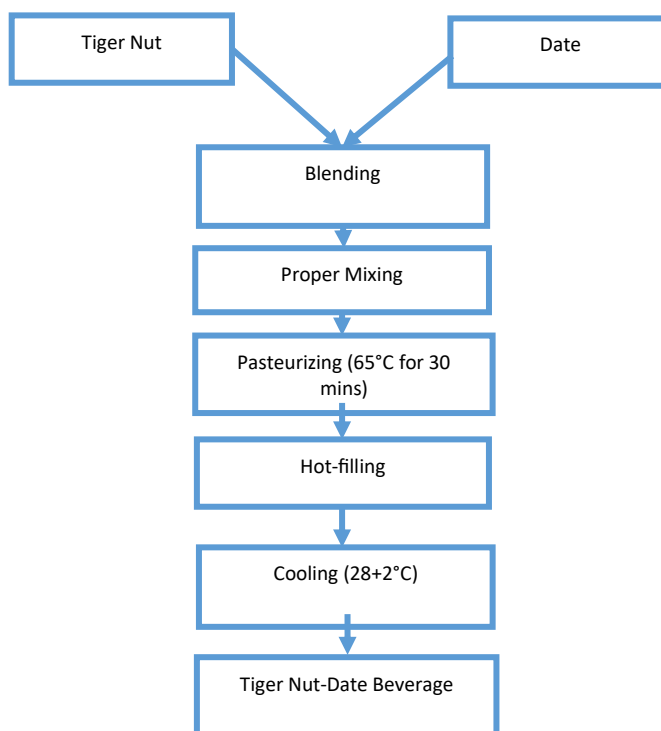


Figure 1: Flowchart of tigernut-date beverage production

## 2.8. Sensory Evaluation

Five samples of tiger nut-date beverage were subjected to sensory evaluation by 10 untrained panellists consisting of 400 level students of the department of Biological sciences, Crawford University, Igbesa who were familiar with beverages. Samples of the tiger nut beverage were coded and presented to the panelists using white transparent disposable cups. Water was provided for mouth wash in between evaluations. Panelists were asked to evaluate the samples for colour, flavour, taste, mouth feel and overall acceptability using a 5-point hedonic scale (5 = strongly like to 1 = strongly dislike) (Sanful, 2009)

## 2.9. Statistical Analysis

Mean and standard deviation of the duplicated data were analysed while the significance were determined using ANOVA at 95% confidence interval (p value <0.05).

## 3. Results and Discussions

### 3.1 Bacterial counts of freshly prepared tiger nut with date juice

The bacterial counts of the freshly prepared tiger nut-date beverage ranged between 0.1 to 1.0 x 10<sup>3</sup> CFU/mL and is presented in Table 1. The microbial counts of the beverage in this study are below the microbial limit of acceptance which is 2.0×10<sup>5</sup> CFU mL<sup>-1</sup> for dairy milk by Codex Alimentarius Commission (2002). The report from this study is similar to that of exposed and unexposed tiger nut beverage with microbial loads of 1.2×10<sup>3</sup> and 0.2×10<sup>3</sup> CFU mL<sup>-1</sup> respectively (Onovo and Ogaraku, 2007).

### 3.2. Identification of Bacteria Associated with the Tiger Nut and Date Beverage

The identification of bacteria present in the tiger nut-date beverage samples investigated was based on their colonial characteristics, Gram staining reaction and Biochemical tests (Voges Proskauer, Catalase, Citrate, Indole, Maltose, Lactose, Glucose, Fructose). The microorganisms isolated from the tiger nut-date beverage were identified as *Staphylococcus aureus*, *Escherichia coli* and *Bacillus subtilis* (Table 2). In a similar study conducted by Nyarko *et al.* (2011) on the microbiological assessment of tiger nut, it was observed that the predominant microorganisms present were *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli* and *Bacillus* spp. *Bacillus* are spore-forming bacteria that are commonly found in soil and water. Therefore, their presence in tigernut beverage could indicate the use of contaminated water or inadequate washing of soil from the tigernut tubers. These microorganisms which could be from the nose, hand, skin and clothing of handlers, coughing, talking and sneezing droplets which could settle on the food during production and storage (Omonigho and Osubor, 2002)

The presence of *Staphylococcus aureus* could have been introduced after processing through cross contamination from the environment and the body of the tigernut beverage handlers. This organism has been reported to be of serious concerns in food safety as it is known to produce enterotoxins which is responsible in certain foodborne illness (Udeozor and Awonorin, 2014).

**Table 1: Bacterial count (CFU mL<sup>-1</sup>) of freshly prepared tiger nut with date juice**

Samples	Bacterial count (10 <sup>-5</sup> ) (Cfu/ml)
T-D1	0.1 x10 <sup>3</sup>
T-D2	1.0 x10 <sup>3</sup>
T-D3	0.4 x10 <sup>3</sup>

VP: Voges Proskauer

GNB: Gram Negative Bacillus

GPC: Gram Positive Cocci

T-D: Tiger nut-Date beverage

+: Positive

- : Negative

T-D 1-3: Tiger nut date beverage samples

**Table 2: Identification of bacteria isolated from the tiger nut-date beverage.**

Sample	Gram/Shape	VP	Motility	Catalase	Citrate	Indole	Maltose	Lactose	Glucose	Fructose	Probable Isolates
T-D 1	GNB	-	+	+	-	+	-	+	+	-	<i>Escherichia coli</i>
T-D 2	GNB	+	+	+	+	-	+	-	+	+	<i>Bacillus subtilis</i>
T-D 2	GNC	+	-	+	+	-	+	+	+	+	<i>Staphylococcus aureus</i>

### 3.3 Determination of physicochemical properties

The physicochemical properties of the tiger nut-date beverage was investigated and reported in table 3.

#### 3.3.1 Moisture content

There is a proportional relationship between the moisture content and the quantity of the tiger nut in the beverage. The moisture content of the beverage increased from 87.11 g/100 g in blend A to 89.21 g/100 g in blend E as the quantity of the tiger nut decreases from 100 % to 0 % in the tiger nut-date beverage. The report on the moisture content from the tiger nut beverage from this study is similar to that of Okyere and Odamtten (2014) where the values are in the same range. The high moisture will encourage microbial growth on the beverage which could lead to reduced shelf life and instability of the beverage (Akande and Okunola, 2011).

#### 3.3.2 Total Soluble Solids (TSS)

The total soluble solids in the beverage decreased from 12.89 % in sample A to 10.79 % in sample E. The TSS reduced with reduction in the quantity of tiger nut in the tiger nut-date beverage. These values are below the minimum standard for sweetened dairy milk (28%) (Codex Alimentarius, 2011). The TSS of the tiger nut –date beverage from this study is in the same range from the TSS reported in the study on different cultivars of tiger nut milk by Okyere and Odamtten (2014). Furthermore, these results are similar to the results of Nonye *et al.* (2023) who reported total solids of 13.9–14.8% for tiger nut beverage.

### 3.3.3 Carbohydrate

The carbohydrate content of the different blends decreased with increasing dates composition from 13.61 g in 100% tiger nut blend (A) to 12.11 g in 0 % tiger nut blend (E). Carbohydrate content in the present study is similar to that reported by Ariyo *et al.* (2021). However, the carbohydrate content of the tiger nut is lower compared to the report of Ukwuru *et al.* (2011). The variations in the carbohydrate composition could be attributed to the difference in the cultivar of tiger nut and other growth parameters/enhancers which also affect the nutritional composition of the crop (Ariyo *et al.*, 2021).

#### 3.3.4 Total Titratable Acidity (TTA)

The Total Titratable Acidity content of the tiger nut-date beverage ranged from 0.021% to 0.034 %. Sample E has the lowest TTA content while sample A has the highest. This is due to the fact that sample A has the highest content of tiger nut which is high in fibre content. The hydrolysis of these compound during fermentation could increase glucose and maltose levels, which are converted into organic acids or alcohols by microbial activity and which directly influences the acidity of the environment (Granito and Alvarez, 2006). Furthermore, the acidity of the beverage could have been influenced by several factors such as the initial microbial load of milk, fermentation period and the hygienic conditions of the production process (Imbachí-Narváez *et al.*, 2017).

#### 3.3.5 pH

The pH values of the tiger nut-date beverage



under this study is in the range of 6.12-6.51. In a similar study, Udeozor (2012) reported a pH of 6.7 for tiger nut milk showing that tiger nut is less acidic. This pH makes tiger nut beverage a tolerable drink for patients with ulcer and other related diseases that are sensitive to acidic beverage. The pH values obtained from the present study is close to the pH range of 6.5 - 6.8 reported by Wakil *et al.* (2014) for

tiger nut milk samples from three varieties of tiger nuts. On the contrary, the pH values were higher than the pH of 4.7 reported by Babatuyi *et al.* (2019) for tiger nut milk. The differences in the pH values of tiger nut milk reported by different authors could be attributed to variations in soil composition on which the nuts were cultivated.

**Table 3: Physicochemical properties of tiger nut-date beverage**

Parameter	A (100:0)	B (70:30)	C(50:50)	D (30:70)	E (0:100)
Moisture content	87.11	87.41	88.77	89.09	89.21
Total Soluble Solids (TSS)	12.89	12.59	11.23	10.91	10.79
Carbohydrate	13.61	13.22	12.79	12.47	12.11
Total Titratable Acidity (TTA)	0.034	0.032	0.027	0.024	0.021
pH	6.12	6.20	6.34	6.48	6.51

### 3.4 Sensory Evaluation of the tiger nut-date beverage

The sensory properties of the different blends of the tiger nut-date beverage are indicated in Table 4. The given results are from a sensory evaluation of different blends of tiger nut and date juice. The blends are represented by the ratio of tiger nut to date juice, ranging from 100:00 (pure tiger nut) to 0:100 (pure date juice). The sensory evaluation provides insights into the taste, aroma, appearance, sweetness, and general acceptability of the different tiger nut-date blends. The taste scores are highest for sample C (50:50 Tiger nut-date beverage blend) with  $3.3 \pm 1.42$ , indicating a balanced and desirable taste profile. The aroma scores are highest for sample A with  $3.4 \pm 1.67$ , while the appearance scores are highest for sample D with

$3.4 \pm 1.43$  blend. The beverage became sweeter with  $4.0 \pm 0.94$  as more date juice was added in sample C, suggesting a preference for increased sweetness. Finally, the optimum value of  $4.0 \pm 1.29$  was obtained with sample C, indicating a favorable overall impression of the tiger nut-date beverage by the panelists. The general acceptability of the blends is promising for commercialization of the blends and conforms to reports on orange-tiger nut beverage (Ukwuru *et al.*, 2011). Increasing taste could be attributed to the sweetening properties of date palm fruits (El Hadrami and Al-Khayri, 2012) and general acceptability could be a result of satiety and flavour associated with the beverage (Mansour *et al.*, 2012).

**Table 4: Sensory Evaluation analysis of tiger nut-date beverage**

Parameter	100:00(A)	70:30(B)	50:50(C)	30:70(D)	0:100(E)
<b>Taste</b>	2.1 ± 1.14	2.5 ± 1.34	3.3 ± 1.42	1.5 ± 1.04	2.0 ± 1.26
<b>Aroma</b>	3.4 ± 1.67	2.5 ± 1.03	2.6 ± 0.97	3.1 ± 1.51	2.9 ± 1.51
<b>Appearance</b>	3.3 ± 1.56	2.7 ± 1.05	3.0 ± 1.05	3.4 ± 1.43	3.2 ± 1.40
<b>Sweetness</b>	2.6 ± 1.58	2.8 ± 1.29	4.0 ± 0.94	2.3 ± 1.42	2.5 ± 1.57
<b>General Acceptability</b>	2.6 ± 1.26	2.3 ± 0.85	4.0 ± 1.29	3.4 ± 1.36	3.0 ± 1.34

**Conclusion**

The need for indigenous and cheaper alternative to the imported milk and milk products with high nutritive values in this part of the world is influencing the choice of consumers on the consideration of readily affordable plant-based milk products. This study showed that the blended mixture of tiger nut and date juice significantly impact the physicochemical and sensory properties of the resulting beverage. The carbohydrate content and total titratable acidity of the blends vary with different proportions of tiger nut and date. The sensory evaluation indicates that the 50-50% Tiger nut-date beverage is generally acceptable and preferred to the other blends by the panelists. Therefore, the results from the study showed that the inclusion of date to the beverage formulation improved the nutrient composition and sensory attributes of the tiger nut milk thus making it a cheaper alternative to the expensive milk and milk products.

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