



(RESEARCH ARTICLE)



Evaluation of sensory and microbiological quality of tigernut milk drink sweetened with date palm fruit

Obasi BC * and Mani VN

Department of Food Science and Technology, Faculty of Agriculture and Life Sciences, Federal University Wukari, Taraba State, Nigeria.

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Abstract

This study investigated sensory properties and microbiological quality of tigernut milk drink sweetened with date palm fruit. Six samples of the drink were prepared in various ratios, tigernut milk control (TMC 100%), date palm slurry control (DSC 100%) tigernut milk and date slurry (TMD1 90:10, TMD2 80:20, TMD3 70:30, and TMD4 60:40) respectively. The samples were pasteurized at 72°C for five minutes. The tiger nut milk were evaluated using 15-member semi-trained panelists on a 9-point hedonic scale and statistical analysis were carried out on their responses. The result for sensory evaluation revealed that there was significant difference ($P < 0.05$) among all the samples analyzed in terms of taste, aroma, colour, flavor and general acceptability. However, sample TMD4 was the most preferred in terms of all the parameters assessed which had a range from (6.1-7.5). Standard microbiological methods were used to isolate, characterize and identify microbial isolates. The result obtained for microbial load for total aerobic plate count, coliforms and fungi ranged from 1.90×10^3 – 1.26×10^6 , 3.20×10^3 – 1.6×10^6 , and 2.8×10^3 – 7.25×10^5 cfu/ml respectively. The result for microbial flora revealed the following species of microorganisms; *E. coli* (21%), *Staphylococcus aureus* (16%), *Proteus vulgaris* (19%), *Bacilli* (16%), *Salmonella typhi* (6%), and *Streptococcus* (8%). The study concluded that milk from tigernut and date palm fruit sources could serve as an alternative for diary milk, and that the date palm demonstrated the potential of being used as an alternative sweetener for tigernut milk drink production based on the general acceptability of the product.

Keywords: Date palm fruit; Microbial isolates; Quality; Sensory properties; Tiger nut milk drink

1. Introduction

Recently, milk from plant sources is gaining prominence. Some of this milk includes; Soy bean milk, rice milk, almond milk, oat milk, coconut milk and tigernut milk. Milk from plant sources are of particular interest owing to their abundance and potential to serve as substitutes for diary milk which is increasingly becoming scarce. The consumption of noncarbonated drinks has become increasingly important and demand largely based on their value, flavour, aroma and colour [1].

Tiger nut "*Cyperus esculentus lativum*" is an underutilized tuber of family Cyperaceae, which produces rhizomes from the base of the tuber that is somewhat spherical [2].It is a tuber that grow freely and is consumed widely in Nigeria, other parts of west Africa, east Africa, parts of Europe particularly Spain as well as in the Arabian Peninsula [3]. Tiger nut can be eaten raw, dried, roasted or grated and used as flour or vegetable milk. It is pressed for its juice to make beverage called "Chufa" [4]. It is known in Nigeria as "Aya" in Hausa, "Ofio" in Yoruba and "Akiusa" in Igbo where these varieties (black, brown and yellow) are cultivated [5]. Among these, the yellow variety is preferred over others because of its inherent properties such as large size, attractive color and fleshier nature. It also yield more milk upon extraction, contains lower fat and higher protein and less anti nutritional factors especially polyphenol [6].

* Corresponding author: Obasi BC

Date palm is a delicious fruit with a sweet taste and fleshy mouth feel texture that is nutritious, assimilative and energy producing. The main component of date palm is carbohydrate (mainly sugars, sucrose, glucose, and fructose), which may constitute about 70%. The sugars in date are easily digested and can immediately be moved to the blood after consumption and can quickly be metabolised to release energy for cell activities [7]. However, the fruits are under exploited and require more research attention. Being a good source of simple sugar such as glucose and fructose, it could be exploited as alternative sweeteners in foods and drinks thereby reducing added sugars in food products and averting health risk including obesity and diabetes. Healthy drinks have become an issue of interest as consumer awareness is moving towards sugar free, additive free and more natural drink, considering the recent trend in most soft drinks of containing high sugar, high pesticide residue and their attendant health risk. The lack of effective antimicrobial treatments at any step from planting to consumption of tigernuts suggested that pathogens introduced at any point may be present on the final food product [7].

A mixture of microorganisms that have been isolated from exposed tiger nuts included *Bacillus subtilis*, *Staphylococcus aureus*, *Aspergillus flavus*, *A. niger*, *Fusarium solani*, *Saccharomyces cerevisiae*, *S. fubiligera* and *Candida pseudotropicalis*, with varied percentage frequencies of occurrence, which rendered the tiger nuts unwholesome, except for the unexposed samples, which recorded relatively lower microbial loads [8]. The presence of pathogenic *E. coli*, *S. faecalis*, and *S. aureus* usually constitutes a direct proof of faecal contamination of irrigation water [9]. [10] conducted an assessment of microbiological safety of tigernut milk in the cape coast metropolis of Ghana and found out that the most predominantly encountered species were *E.coli* and *Bacillus* spp which had 18.9% each. Others included *Enterococcus* spp (16.2%), *S. aureus* and *P.aeruginosa* (13.5% each) and *Streptococcus* spp (10.8%).

Tigernut milk is suitable as alternative beverage for celiac patients who are intolerant to gluten and lactose. It has high nutritional qualities and potential health beneficial bioactive compounds. However, most of the available information reveals that tiger nut and date palm is underutilized in Nigeria and still confined to local use only, hence the need to determine the effect of date palm inclusion on the tigernut milk drink production. Therefore, the aim of this study is to evaluate the sensory and microbiological quality of tigernut-milk drink sweetened with date palm fruits in order to ascertain its acceptability and safety.

2. Material and methods

2.1. Sample collection

Fresh yellow tigernut (*Cyperus esculentus*), and date palm fruit (*Phoenix dactylifera L.*) were obtained from yelwa market in Bauchi, Bauchi State, Nigeria. The samples were then transported to the Food Science and Technology Department, Federal University Wukari for analyses in clean polythene bags.

2.2. Experimental design

Completely randomized design were used in the study and the principal factors were tiger nut and date palm fruit slurry to produce six samples.

2.3. Sample preparation

Fresh tigernuts (untreated tigernuts) were used for the preparation of tigernut milky juice extract (plain tigernut milk) and was sweetened using date palm fruit juicy extract.

2.4. Tigernut milk drink Production

The method as described by [11] with little modification were used for the production of tigernut-milk drink from fresh tiger nuts. Fresh tiger nuts were manually sorted and cleaned to remove foreign particles and unwanted materials. The fresh tiger nuts were milled into slurry and water was added to extract the milk at a ratio of 1:3.

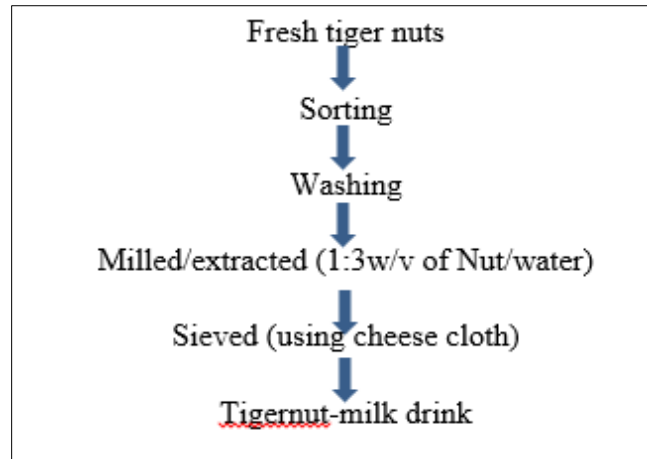
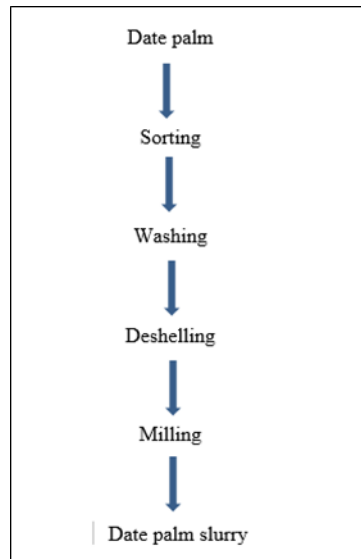


Figure 1 Flow chart for tigernut milk production. Source: Udeozor [11]

2.5. Production of date palm fruit slurry

The date palm fruits was sorted, washed and milled using a dry milling machine. Date palm fruit juice was then extracted at a ratio of 1:3 w/v of date slurry to water



Source: [11]

Figure 2 Flow chart for date palm fruits slurry production

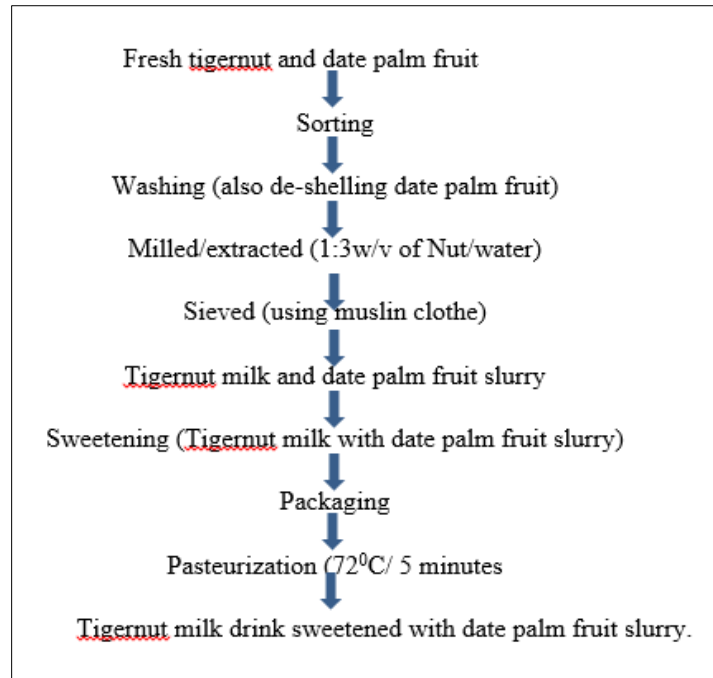
2.6. Formulation of sweetened tigernut milk drink

Tigernut milk drink was properly sweetened in different ratios. 100% tigernut milk drink and 100% date palm slurry was used as control and substituted up to 40% date palm fruit slurry respectively. This is as shown in Table 1 below.

Table 1 Sample formulation table

Sample Codes	TMC	DSC	TMD1	TMD2	TMD3	TMD4
Ratio	100	100	90:10	80:20	70:30	60:40

Keys: TMC = Tigernut milk drink control; DSC= Date palm fruit slurry control; TMD= Tigernut milk drink sweetened with date palm fruit slurry



Source: [11]

Figure 3 Flow chart for tigernut-milk drink sweetened with date palm fruits formulation

2.7. Sensory Evaluation

The method as described by [12] was used for the evaluation of the samples. Samples were assessed for colour, flavour, taste, aroma and overall acceptability using 15-member semi-trained panelists on 9-point hedonic scale with (9) = extremely like and (1) = extremely dislike. The samples were presented to the panelists. All the panelists were drafted from among students and staff members of Food Science and Technology Department.

2.8. Microbiological Analysis

2.8.1. Preparation of Media

The enumeration of bacteria cells and fungi count from the samples of tigernut milk sweetened with date palm fruit was done using Nutrient agar, Macconkey agar, and Potato dextrose agar. They were prepared according to the manufacturers (Titan Biotech Ltd) instruction and sterilized by autoclaving at 121°C for 15 minutes at 15 pounds per square inch (PSI).

2.8.2. Isolation and Enumeration

Total bacterial count was determined using the method as described by [13]. The stock solution was prepared by dissolving 1ml of the sample of tigernut milk drink sweetened with date palm fruit in 9ml of sterile peptone water, serial dilution (10 fold) was carried out (1:10, 1: 100, 1:1000...10,000). 0.1ml of appropriate dilutions (10⁻² and 10⁻⁴) was placed on various agar plates using pour plate method and incubated at 37°C for 18-24 hr for total bacteria and coliform count. For fungi 1ml amount of appropriate dilutions (10⁻² and 10⁻⁴) was also poured into the plates of potato dextrose agar and incubated at room temperature 28±1°C for 3 to 5 days. All enumeration was expressed as colony forming unit per milliliter (cfu/ml).

2.8.3. Purification and maintenance of microbial isolates

Bacteria isolates were transferred into fresh agar medium of isolation and incubated at 37°C for 24hr. Pure colonies of bacteria were maintained and stored at 4°C until needed.

2.8.4. Identification and characterisation of the isolates

Bacteria isolates were identified and characterized based on their cultural, morphological and biochemical tests as described by [13,14]. Biochemical tests included: indole, catalase, citrate utilization, oxidase, hydrogen sulfide production, Triple-Sugar Iron agar (TSI) etc.

2.9. Statistical Analysis

Data obtained was expressed as mean \pm standard deviation (S.D.), analysed by analysis of variance (ANOVA) and comparison between means was carried out using the Duncan test at 95% confidence level. The SPSS statistical software version 20 was used for the analyses of experimental results.

3. Results and discussion

3.1. Sensory evaluation of tigernut milk drink sweetened with date palm fruit

The results for sensory evaluation of tigernut milk drink sweetened with date palm fruit is presented in Table 2. The results for taste, aroma, color, flavor and general acceptability, among the six samples of tigernut milk drink sweetened with date palm fruit ranged from 6.33 – 8.1, 6.5 – 7.2, 6.93 – 7.13, 6.3 – 7.6, and 6.1–7.5 respectively with significant differences ($P < 0.05$). The result revealed that sample TMD4 was the most preferred in terms of all the parameters evaluated in relation to taste, aroma, colour, flavor and general acceptability. Therefore the overall choice for TMD4 could be attributed to the aroma and its general acceptability as compared to TMC, DSC, TMD1, TMD2 and TMD3. [15] suggested that plant-based dairy alternatives that have similar sensory characteristics to dairy milk were more accepted, and found that the more similar the sensory profile of a dairy milk alternative in terms of aroma, colour, taste and flavor is to dairy milk, the more it is accepted [16]. This could be implied as the reason for acceptability and appeal of TMD4 by the judges.

Table 2 Means sensory scores of tigernut milk drink sweetened with date palm fruit

Sample Code	Ratio	Taste	Aroma	Colour	Flavor	General Acceptability
TMC	100	6.67 ^a \pm 1.78	6.67 ^a \pm 1.36	6.93 ^a \pm 1.03	6.29 ^a \pm 1.67	6.60 ^a \pm 1.60
DSC	100	8.13 ^{ab} \pm 0.99	7.13 ^a \pm 1.55	6.87 ^a \pm 1.13	7.60 ^{ab} \pm 1.02	7.13 ^a \pm 1.25
TMD1	90:10	6.33 ^a \pm 1.45	6.53 ^a \pm 1.64	6.27 ^a \pm 1.71	6.60 ^{ab} \pm 1.24	6.07 ^a \pm 1.39
TMD2	80:20	7.40 ^{ab} \pm 1.06	6.77 ^a \pm 1.35	7.13 ^a \pm 1.25	6.93 ^{ab} \pm 1.39	7.13 ^a \pm 1.13
TMD3	70:30	7.40 ^{ab} \pm 2.25	6.73 ^a \pm 1.22	6.60 ^a \pm 1.22	6.87 ^{ab} \pm 1.73	7.20 ^a \pm 1.26
TMD4	60:40	7.13 ^{ab} \pm 2.25	7.20 ^a \pm 1.32	6.47 ^a \pm 1.22	6.53 ^a \pm 1.73	7.53 ^a \pm 1.46

Values are mean \pm standard deviation of 15 panelists. Means within each column not followed by the same superscript are significantly different ($P < 0.05$) from each other using Duncan multiple range test.

3.2. Microbial load of tigernut milk drink sweetened with date palm fruit

Table .3 shows the total aerobic plate count for bacteria cells which ranged from 1.90×10^3 – 1.26×10^6 cfu/ml for TMC, DSC, TMD1, TMD2, TMD3 and TMD4 respectively. The total coliforms count from tigernut milk drink sweetened with date palm fruits ranged from 3.20×10^3 – 1.6×10^6 cfu/ml for TMC, DSC, TMD1, TMD2, TMD3 and TMD4 respectively. The fungi count from tigernut milk drink sweetened with date palm fruits ranged from 2.8×10^3 – 7.25×10^5 cfu/ml for TMC, DSC, TMD1, TMD2, TMD3 and TMD4 respectively.

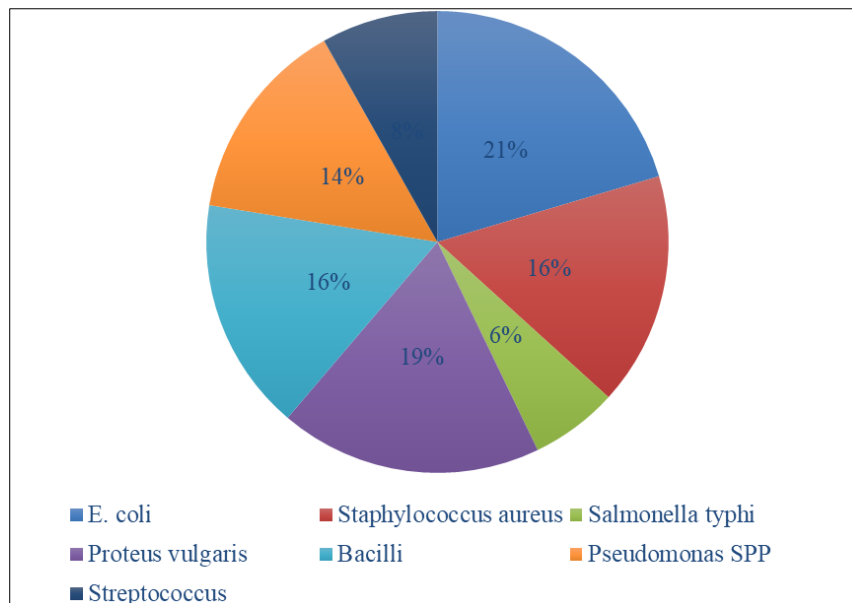
A high microbial count was observed in the analysed tiger nut milk drink this was not surprising as the tiger nut milk production systems are sometimes done under unhygienic conditions [17], with no authorized agency to monitor their microbial quality and safety. The presence of high microbial load could also be as a result of contamination from the milling equipment used in the processing. The result of this study is in agreement with the report by other researchers [18,19] who obtained a similar result.

Table 3 Total aerobic plate count (TAPC) of bacterial cells (Cfu/ml) obtained from tigernut milk drink sweetened with date palm fruit

Isolate Code	Ratio (%)	Total aerobic plate count (cfu/ml)	coliform count (cfu/ml)	fungi count (cfu/ml)
TMC	100	1.46x10 ⁴	2.8 x10 ³	2.8 x10 ³
DSC	100	1.90x10 ⁴	1.27 x10 ³	3.0 x10 ⁵
TMD1	90:10	1.26x10 ⁶	1.77x10 ⁴	No growth
TMD2	80:20	1.0x10 ⁶	1.68 x10 ⁵	7.25 x10 ⁵
TMD3	70:30	6.3x10 ³	3.20 x10 ³	No growth
TMD4	60:40	2.0x10 ⁴	2.0x10 ⁴	1.80x10 ⁴

3.3. Morphological and biochemical characteristics of bacterial strains isolated from tigernut milk drink sweetened with date palm fruit

The results in Fig 4. showed the identity of the microorganisms isolated from tiger nut milk drink sweetened with date palm fruit. The microbial species included *E. coli* (21%), *Staphylococcus aureus* (16%), *Proteus vulgaris* (19%), *Bacilli* (16%), *Salmonella typhi* (6%), and *Streptococcus* (8%). The result of this study is in line with the report of [20], who also isolated some diverse microbial species associated with tigernuts as obtained from the study. The presence of pathogenic *Escherichia coli*, *Streptococcus spp faecalis* and *Staphylococcus aureus* usually constitute a direct proof of faecal contamination of irrigation water[14]. The occurrence of *Staphylococcus aureus* and *Escherichia coli* in the analysed sample was not surprising as it corroborates with the study of [20] ; who reported the isolation of *S. aureus*, *S. pyogenes*, *P. aeruginosa*, *A. flavus*, *Rhizopus spp*, *Bacillus subtilis*, *Proteus spp*, and *S. cerevisiae* in hawked kunu and zobo drinks sold within Lagos State University of Technology campus, Ogbomoso, Nigeria. The identified non-lactic acid bacteria (*E. coli*, *Bacillus species* and *Proteus species*) reported in this study has also been reported by other authors [10,21,22]. The thermotolerant nature of *Bacillus* spores ensures survival at pasteurization temperatures and hence, their presence in the tigernut milk drink sweetened with date palm fruit samples that have been subjected to heat treatment during processing.

**Figure 4** Prevalence of microorganisms isolated from tigernut milk drink sweetened with date palm fruits

4. Conclusion

The overall acceptance of the panelists in terms of taste, color, flavor, aroma and general acceptability indicated that tigernut milk sweetened with date palm fruit products are appealing and could gain substantial acceptance by the public. The microbial species identified in the milk shows some level of microbial contamination which may not pose a serious threat as to the safety of consuming milk from tigernut and date palm fruit sources. This is because the products analysed satisfied the FAO/WHO microbial acceptable limit (5.30 log₁₀ cfu/ml) for dairy milk. The findings in this study considering sensory and microbiological evaluation of tigernuts milk drink sweetened with date palm fruits demonstrated the potential of using date as alternative sweetener for tigernut milk drink production.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

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