

Microbial and sensory quality of powdered beverage produced from blends of baobab pulp and moringa leave during storage

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International Journal of Science and Research Archive, 2023, 09(02), 786–794

Publication history: Received on 01 July 2023; revised on 16 August 2023; accepted on 18 August 2023

Article DOI: <https://doi.org/10.30574/ijrsra.2023.9.2.0646>

Abstract

The study investigated the microbial and sensory quality of powdered beverages produced from blends of baobab pulp and moringa leave flour. Samples were prepared and stored in sterile plastic containers and bottles, were stored at room temperature 27 ± 2 °C and in the refrigerator for 21 days. Sensory evaluation was done using the following parameters, appearance, mouth fill, taste, flavor and general acceptability. The samples most preferred in relation to all the parameters tested is samples A and B. Total aerobic plate count of the bacterial cells for control samples A-E ranged from 5.2×10^6 - 8.2×10^6 cfu/ml. Samples A-E ranged from 8.5×10^5 - 3.04×10^7 for plastic container and 1.15×10^5 - 1.6×10^7 for samples stored in bottles at room temperature, while samples stored in the refrigerator ranged from 5.6×10^5 - 2.3×10^7 for plastic containers and bottles 4.6×10^6 - 9.5×10^6 for week one. For week two samples A-E for plastic and bottles ranged from 3.7×10^5 - 8.5×10^5 and 3.2×10^5 - 9.6×10^6 for samples stored at room temperature and refrigerator respectively. For week three samples A-E stored in plastics and bottles at room temperature and refrigeration ranged from 3.5×10^5 - 1.7×10^6 and 1.0×10^5 - 3.0×10^5 . The bacterial isolates were characterized using morphological, biochemical tests and the identity of the isolates were confirmed with a microgen test kit. The species of bacteria identified includes: *Bacillus* Spp, *Escherichia coli*, *Klebsiella pneumonia*, *Staphylococcus aureus*, *Salmonella typhi* and *Shigella*. From this study it shows that the baobab pulp and moringa leave flour blend beverage should be stored in the refrigerator or consumed immediately after processing in order to maintain its organoleptic properties and avoid microbial contamination.

Keywords: Baobab pulp; Beverage; Moringa leave; Microbial; Sensory quality

1. Introduction

Baobab or *Adansonia digitata* L. belongs to the Malvaceae family [1] and is a deciduous tree native to arid Central Africa. Baobab is a very long-lived tree with multipurpose uses. The different plant parts are widely used as foods, shelter, clothing and medicines as well as material for hunting and fishing [2]. The baobab fruit is also used daily in the diet of rural communities in West Africa [3,4,5]. For example, the Hausa ethnic group uses the dried leaves as the main ingredient in a soup called miyankuka.

Moringa oleifera is one of the vegetables of the Brassica order and belongs to the family Moringaceae. Traditionally, besides being a daily used vegetable among people of different regions, the *Moringa* is also widely known and used for its health benefits. Among commoners, it has earned its name as 'the miracle tree' due to its amazing healing abilities for various ailments and even some chronic diseases.

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Moringa is capable of preserving food quality and reducing bacterial contamination due to mixed antioxidant, antibacterial and protease inhibiting properties [6,7].

It is well known that beverages are normally produced from different kinds of fruit especially orange, mango, and pineapple. These are seasonal fruits which makes it very expensive and also make it difficult for less privileged or the poor to readily afford it in some part of the country. [8] The enhancement of the production of cost-effective probiotic products made from locally available resources has the potential to lift rural communities which depend on it out of poverty by integrating them into sustainable markets. [9]. Therefore, this study will be of great impact and beneficial not only to rural communities but also because baobab pulp is an ideal candidate in the functional food market as it is very high in vitamin C, pectin and fiber content [9]. In addition to that, through the development of beverage using baobab pulp enriched with moringa leaf powder, the scientifically proven beverage can be adopted for commercialization by young entrepreneurs to improve the livelihoods of people living in areas where there is abundance.

2. Materials and methods

2.1. Sample Collection

Baobab fruit pulp capsules (*Adasonia digitata L.*) were purchased from Kaduna central market (Fig1), while moringa (*Moringa oleifera*) leaves were obtained from Federal University Wukari school farm (Fig 2), Taraba State, Nigeria.

2.2. Preparation of baobab pulp powder

Whole baobab fruits were weighed, and their hard woody shells carefully crushed to expose the white flesh powder surrounding the seeds. The pulp was separated from the seeds by grinding using pestle and mortar. The sample was sieved using a 40 mesh sieve to obtain a fine powder. The powder was weighed and immediately packed in polyethylene bags sealed and stored in a dark cool place until when needed for analysis and preparation of reconstitution drinks. The procedure is presented in the figure 3 below.

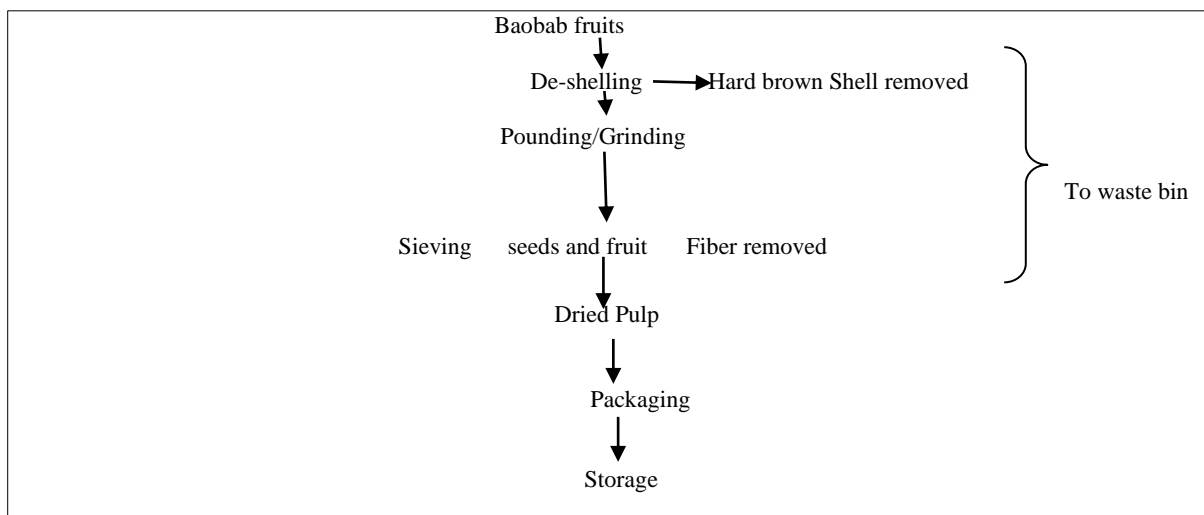


Figure 1 Flow chart for the Processing of Baobab Fruit Pulp Powder

Source: [9]

2.3. Preparation of moringa leaves powder

The method of [11] was used as described. Moringa leaves were collected after removal of the inedible portion (dirt, stones, and seeds). Collected leaves were washed and rinsed with distilled water. Drying of the leaves was done at room temperature $27 \pm 2^\circ\text{C}$. Dried leaves were ground using mortar and pestles. The sample was sieved using a standard sieve mesh of 0.5 mm – 1.0 mm pore size screen to obtain a fine ground leaf powder. The ground powder was oven dried at 50°C for 30 minutes to reduce the moisture content. The moringa powder was packaged in air-tight polythene bags for analysis.

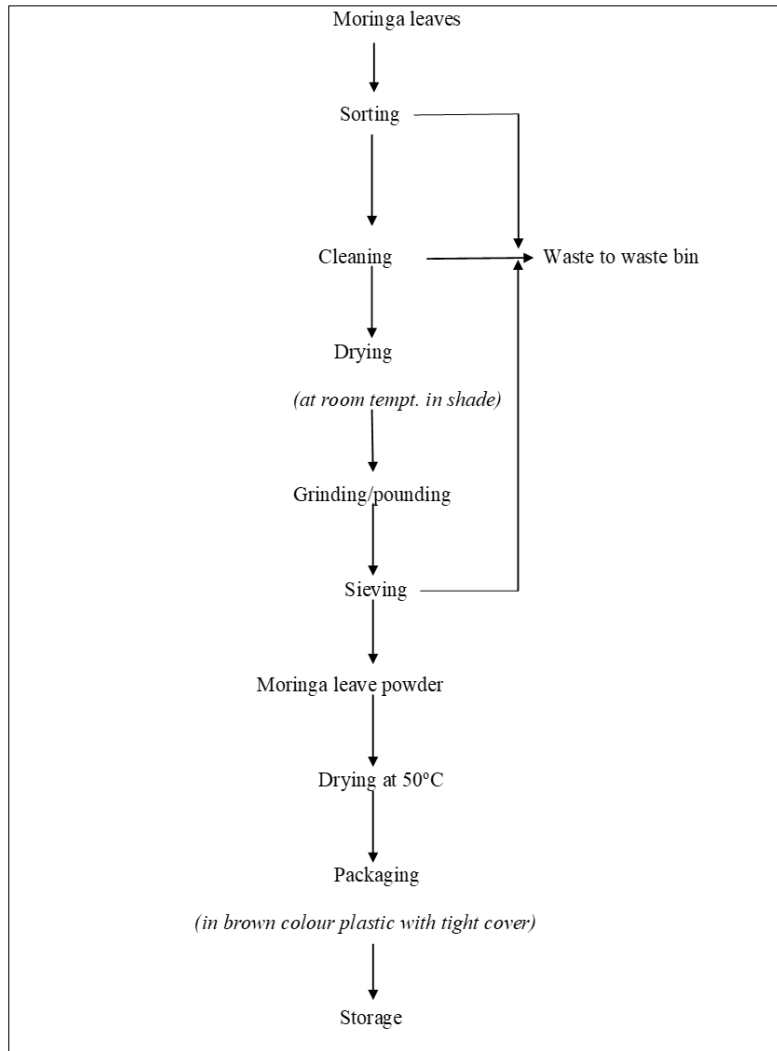


Figure 2 Flow Chart for Processing of Moringa Leaves Powder. Source: [10]

2.4. Preparation of experimental samples

Five grams (5g) of powdered baobab pulp was weighed, prepared and blended with moringa leaf powder at different proportion of 5%, 10%, 15%, 20%, and 25% respectively. The Samples were package in a plastic containers and bottles and stored at ambient ($27 \pm 2^{\circ}\text{C}$) and refrigeration temperatures for a period of 21days for analysis.

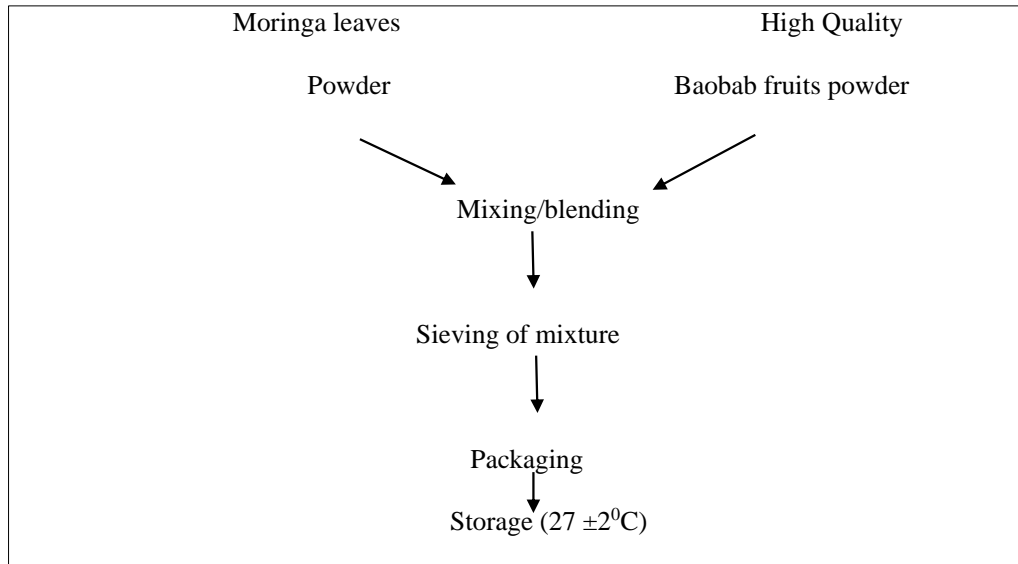


Figure 3 Flow Chart for Processing of blends of Boabab Pulp and Moringa Leaves powder

Source: [11, 12] with Some Modification

2.5. Isolation and Enumeration of Bacterial Cells

The method as describe by [13] were used for the enumeration of the bacterial cells using culture techniques. The beverage samples were subjected to ten – fold dilution with sterile saline as diluent. 0.1ml of the ten -fold dilutions of the desired dilution were inoculated into the various sterile culture plates of MacConkey and Nutrient agar media in duplicates using pour plate techniques. The inoculated samples were incubated at 37°C for 24h. At the end of the incubation period, all enumerations were expressed as colony forming unit per milliter (cfu/ ml).The MacConkey agar was used for total coliform count (TCC) and Nutrient agar for total heterotropic bacterial count (THBC).

2.6. Characterization and Identification of the isolates

The purified bacterial isolates were used for characterization and identification using morphological (cultural morphology of colonies), microscopic (Gram staining reactions) and biochemical tests (reactions with appropriate sugars and biochemical reagents) as described by [14]. The identity of the isolates were confirmed to species level using microgen kit (MicrogenTMGnAa+B-ID system) Surrey, United Kingdom.

2.7. Sensory evaluation

The sensory quality of the powdered baobab beverage were evaluated using twenty untrained panelists, comprising of student and staff of the Department of Food Science and Technology from Federal University Wukari. A nine-point Hedonic scale with one (1) representing “extremely dislike” and nine (9) “extremely like” was used. The qualities assessed were appearance, mouth fill, taste, flavor, and general acceptability [15].

2.8. Statistical analysis

All the experiments were conducted in duplicates in completely randomized design. The data were subjected to analysis of variance (ANOVA). Means with significant different were separated by the least significant difference (LSD) test. Significance was accepted at $p < 0.05$.

3. Results and discussion

3.1. Sensory quality

Figure 7 shows the result for sensory evaluation. Significant difference ($p < 0.05$) between mean scores were found for appearance, mouth fill, taste, flavor, and general acceptability. The sample with 1.3% powdered baobab pulp and moringa leave blend had the highest score for taste, flavor, appearance, and general acceptability. The sample with 0.8% was the least preferred by the panelist for almost all the attributes tested. The sample with the concentration of 1.3% had significantly higher rating in all attributes tested except for sample 0.5% ($6.75 \pm 1.37ab$) which had significantly

higher rating for appearance. Also 1% ($6.35 \pm 1.38a$) blend beverage sample showed significantly higher rating for general acceptability no significant difference ($p < 0.05$) between samples 1.3% and 0.8% indicating that 0.8% sample compared favorably with 0.5% ($5.90 \pm 1.71a$) and 0.3% ($6.15 \pm 2.03a$). Powdered baobab pulp and moringa leave beverage blend showed significantly higher rating for general acceptability for sample with the concentration of (0.5%), however no significant difference ($p < 0.05$) between 1.3% and 1% samples. No significant difference ($p < 0.05$) existed between all the samples incorporated with moringa leave powder for appearance, mouth fill, taste and flavor.

3.2. Microbial Quality of Powdered baobab pulp and moringa blended beverage

The result in Table 1 shows the total aerobic count of the bacterial cells obtained from the various blends of powdered baobab and moringa leave beverage stored for twenty one days at ambient ($27 \pm 10C$) and refrigeration ($5 \pm 10C$) temperatures. The microbial population of powdered beverage for control sample A-E ranged from 5.2×10^6 - 1.04×10^7 cfu/ml. Sample stored in plastic container for week one for samples A-E ranged from 8.5×10^5 - 3.04×10^7 while samples stored in bottles ranged from 1.15×10^5 - 1.6×10^7 , for samples stored in a plastic container for week two ranged from 3.7×10^5 - 8.5×10^6 while samples stored in bottles ranged from 1.02×10^6 - 8.2×10^6 and samples stored in a plastic container for week 3 ranged from 3.0×10^5 - 2.6×10^6 while samples stored in bottle ranged from 2.1×10^5 - 7.3×10^5 . The total viable count levels for all the samples A-E are not within the safety limit of $< 10^5$ colony forming unit for powdered baobab pulp and moringa leave beverage.

From the result obtained it shows that the count kept increasing with days in the samples stored at room temperature for both plastic and bottle. This could be due to an increase in acidity or reduction in potential oxygen. This result agrees with the work of [16]. But for the refrigerated sample the count kept reducing from first week to the third week for the stored samples.

The analyses revealed presence of microbial contaminations of varying degrees in both stored sample under room and refrigerated temperatures for week 3 as shown in Table 2. Stored samples at (ambient temperature) in plastic containers were the most microbiologically contaminated with bacterial cells. However, the refrigerated sample for week three for sample (D) has the lowest number of bacterial contaminants (1.2×10^7 Cfu/ml). The detected microbial contamination from the stored sample under ambient temperature exceeded the acceptable limits while those stored in the refrigerator did not. The result obtained from the study showed little /no antimicrobial effect with the moringa leave powder which was meant to serve as a control for the growth of bacterial cells especially for the samples stored under ambient condition.

3.3. Characterization and Identification of bacterial isolates from powdered baobab pulp and moringa leave beverage blend

The results obtain from bacterial characterization and identification revealed the presence of *Bacillus* Spp , *Staphylococcus* spp, *Shigella* and *Pseudomonas* species. The confirmed identity of the isolates are as shown in Table 2. The organism identified includes the following *E.coli*, *Klebsiella pneumonia*, *Salmonella typhi* and *Staphylococcus aureus*. The prevalence of organisms are as follows; sample A (1.3%) *E.coli*, sample B (1%) *Klebsiella pneumonia*, sample C (0.8%) *Salmonella typhi*, sample D (0.5%) *Staphylococcus aureus* and sample E (0.3%) *Salmonella typhi*.

The presence of these microflora could be due to the fact that these organisms are spore formers and are known as common environmental contaminants; nevertheless, they have been implicated as food borne pathogens [17]. The presence of *Salmonella* spp, *E. coli*, and *Klebsiella* calls for concern as these organisms are frequently associated with poor sanitary practices and could be a pointer to danger of possible food borne infection. *E. coli* and *Salmonella* spp are especially of fecal origin and have been implicated in numerous food borne diseases.



Figure 4 Baobab fruit pulp capsule



Figure 5 *Moringa oleifera* leaf



Figure 6 *Moringa oleifera* leaf powder

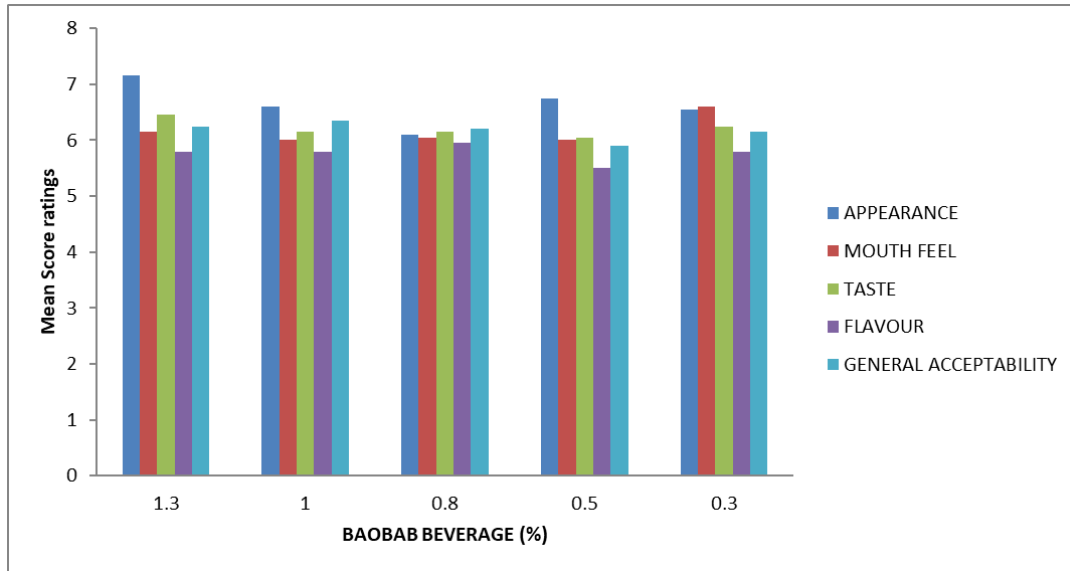


Figure 7 Mean organoleptic scores for the different blends of baobab and moringa beverage

Table 1 Total aerobic plate count of powdered baobab pulp and moringa leave blended beverage of bacterial cells (cfu/ml) during Storage

Isolate Code	WEEK ONE plastic bottle) storage conditions (Control)		WEEK TWO plastic container) storage conditions		WEEK TWO (for bottles) ON STORAGE		WEEK THREE (for plastic container) ON STORAGE		WEEK THREE (for bottles) ON STORAGE		WEEK FOUR (for plastic container) ON STORAGE		WEEK FOUR (for bottle) ON STORAGE	
	A.T °C	R.T °C	A.T °C	R.T °C	A.T °C	R.T °C	A.T °C	R.T °C	A.T °C	R.T °C	A.T °C	R.T °C	A.T °C	R.T °C
A	1.04×10 ⁷	8.2×10 ⁶	3.04×10 ⁷	2.3×10 ⁷	1.5×10 ⁷	9.8×10 ⁶	8.5×10 ⁶	1.2×10 ⁶	8.2×10 ⁶	6.3×10 ⁶	7.0×10 ⁵	3.0×10 ⁵	5.2×10 ⁵	3.1×10 ⁶
B	7.2×10 ⁶	9.4×10 ⁶	1.24×10 ⁷	7.4×10 ⁵	1.15×10 ⁵	4.0×10 ⁶	5.7×10 ⁶	9.6×10 ⁶	7.0×10 ⁶	9.2×10 ⁶	1.7×10 ⁶	8.0×10 ⁵	4.8×10 ⁵	2.4×10 ⁵
C	7.8×10 ⁶	1.12×10 ⁷	1.62×10 ⁷	5.6×10 ⁵	1.45×10 ⁶	7.6×10 ⁶	4.0×10 ⁶	3.6×10 ⁶	8.6×10 ⁶	6.3×10 ⁶	9.0×10 ⁵	2.0×10 ⁵	6.6×10 ⁵	3.8×10 ⁵
D	8.4×10 ⁶	1.04×10 ⁷	8.5×10 ⁵	9.0×10 ⁵	1.6×10 ⁷	8.3×10 ⁶	3.7×10 ⁵	7.0×10 ⁵	1.02×10 ⁶	9.1×10 ⁶	3.0×10 ⁵	1.0×10 ⁵	7.3×10 ⁵	4.1×10 ⁵
E	5.2×10 ⁶	6.7×10 ⁶	1.4×10 ⁷	8.4×10 ⁵	2.04×10 ⁶	9.5×10 ⁶	6.5×10 ⁵	3.2×10 ⁵	5.4×10 ⁶	7.4×10 ⁶	2.6×10 ⁶	9.0×10 ⁵	2.1×10 ⁵	6.7×10 ⁵

Samples A-B: Shows percentage of powdered baobab pulp and moringa leave blended beverage 1.3,1,0.8,0.5,0.3 respectively; Where A.T = ambient temperature; R.T = refrigeration temperature.

Table 2 Bacterial Species Isolated From Powdered Beverage Produced From Blends Of Baobab Pulp And Moringa Leave During Storage

	LYS	ORN	H ₂ S	GLU	MAN	XYL	ONP	IND	URE	VP	CIT	TDA	IDENTITY
A	+	-	+	-	-	-	+	+	-	+	+	-	<i>E.coli</i>
B	+	-	-	-	-	-	+	+	-	+	+	-	<i>Klebsiella pneumonia</i>
C	-	-	+	-	-	-	+	-	-	+	+	-	<i>Salmonella typhi</i>
D	-	-	-	+	-	+	+	+	-	+	+	-	<i>Staphylococcus aureus</i>
E	+	-	+	-	-	-	+	-	-	+	-	-	<i>Salmonella typhi</i>

KEY: Samples : A-E : Shows percentage of powdered baobab pulp and moringa blended beverage 1.3, 1, 0.8, 0.5, 0.3 respectively. LYS= Lysine, ORN= Ornithine, H₂S= Hydrogen sulphide, GLU= Glucose, MAN= Manitol, XYL= Xylose, ONP= Ortho-Nitrophenol, IND= Nitrate, URE= Urease, VP= Vogues proskauer, CIT= Citrate, TDA= Deaminase Reagent.

4. Conclusion

The addition of powdered moringa leave to powdered baobab pulp improved the quality of the product processed in relation to appearance, taste, flavor and general acceptability of the beverage produced. The sample with 0.5% blend was the most generally preferred. The microbial quality of the beverage stored in the refrigerator was within the safety limit of <105 cfu/ml per sample unit. Therefore, this study will be of great impact to the enhancement of the production of cost effective probiotic products made from locally available resources which has the potential to lift rural communities which depend on it out of poverty by integrating them into sustainable market.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to disclosed.

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