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Nutritional Composition and Antioxidant Properties of Instant Beverage Infusion from Blends of Moringa Leaf (*Moringa oleifera*), Zobo Calyx (*Hibiscus sabdariffa*) and Lemon Grass (*Cymbopogon citrates*)



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ABSTRACT

This study investigated the nutritional constituents and antioxidant properties of instant beverage infusions obtained from leaves of *Moringaoleifera*, *Hibiscus sabdariffa* and *Cymbopogon citrates* mixed in different proportions and packaged in tea bags as MZL1 (85% Moringa leaf + 10% zobo calyx + 5% lemongrass) and MZL2 (50% Moringa leaf + 45% zobo calyx + 5% lemongrass). The packaged beverage blends were stored at room temperature for four months and analyses were carried out at the two-month interval. The beverage with 85% Moringa leaf (MZL1) possessed the highest protein content (35.90%-48.17%). The vitamin C content of the beverages ranged from 51.30 mg/g – 67.50 mg/g, vitamin E (1.87 mg/g-3.47 mg/g) and vitamin A (788.52 unit/g-1009.20 unit/g). Beverage blends were found to contain iron (3.48 ppm-3.97 ppm), calcium (1.80 ppm-2.87 ppm), zinc (1.61 ppm-2.47 ppm), sodium (1.98 ppm-2.95 ppm) and potassium (0.75 ppm-0.78 ppm). The beverage blends demonstrated antibacterial activity. Beverage blends demonstrated free radicals scavenging properties with ABTS(1.05 millimole/g-1.24 millimole/g) and DPPH (85.32% -86.37%). It was observed that the beverage blends were scored above average for all attributes in the sensory analysis. The blends investigated could be a promising novel medicinal beverage infusion.



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1. INTRODUCTION

Moringa oleifera and *Hibiscus Sabdariffa* (Roselle) are used as medicinal and food ingredients in many parts of the World, including Nigeria and the rest of Africa. In northern Nigeria, both are highly sourced as food vegetables, particularly because of their health-promoting and disease-preventing properties strongly believed to be due to the presence of many phytochemicals in them. Moringa was reported to boost immune systems, a good source of vitamins and amino acids in addition to other essential nutrients and minerals[1] at levels higher than that recommended by the Food and Agriculture Organisation with patterns similar to that of soybean [2].

Roselle has been employed in the preparation of local, non-alcoholic beverage (zobo), tea, jam, industrial wine, and marmalade [3]. The anticholesterol action of *H. sabdariffa* (0.5% or 1%) was confirmed in rabbits fed with cholesterol for 10 weeks [4]. Furthermore, the antihypertensive action of *H. Sabdariffa* has been confirmed in rats with experimental hypertension [5].

The freshly cut and dried leaves of lemongrass have been used traditionally for making fragrances, flavoring food, and treating a wide variety of ailments due to its numerous phytochemical properties. The medicinal value of lemongrass has been attributed to citral, a volatile oil from the plants [6]. Also, polyphenols from lemongrass extract have been shown to relax and dilate the walls of the blood vessels [7].

In quest toward zero import dependence and food security, the food drink industry has come under scrutiny, as many synthetic drinks possess almost no food value, contain harmful or even carcinogenic chemicals which have been shown to aggravate diseases such as diabetes and high blood pressure [8]. This has precipitated research into local drinks or beverages which are cheap, readily available and possess nutritional, medicinal or pharmaceutical properties. Previous researches have reported medicinal properties of moringa leaf, zobo and lemon grass [1,3,6]. Therefore, this study is aimed at evaluating the phytochemical, nutritional and antioxidant properties of instant beverage from blends of *Moringaoleifera* leaves and *Hibiscus sabdariffa* calyxes using lemongrass as flavor.

2. MATERIALS AND METHODS

2.1 Sample collection

A considerable quantity of roselle calyx was purchased from the Local market in Owena, Ondo State, Nigeria. Fresh Moringa leaves, lime leaves and lemongrass were sourced from farms in Akure Metropolis, Ondo State, Nigeria. They were taken to the Department of Crop Soil and Pest Management of the Federal University of Technology Akure Nigeria for identification. All the chemicals and reagent used in this study were of analytical grade.

2.2 Methods

2.2.1. Sample preparation

The calyxes of Roselle were inspected and sorted to remove sticks and stones and were then sun-dried for two days. The fresh moringalime and lemongrass leaves were washed and air dried for 7days. After drying, they were ground in a laboratory grinder until fine particles were obtained and sieved. The blended samples were separately weighed and sealed separately in filter paper (used as tea bag) to prepare the beverage using the formulation; 85% moringa leaf+10% zobo calyx+5% lemongrass (MZL1) and 50% moringa leaf+45% zobo calyx+5% lemongrass (MZL2). The beverages were labeled and kept in an airtight container until analyzed.

2.2.2 .Determination of pH, bricks and total titratable acidity (TTA) of beverage blends

The pH and total titratable acidity of the samples were determined. Two grams of each of the samples were homogenized with 20ml of distilled water. The pH value of the homogenate was read using a pH meter and recorded. Total soluble solid was determined using Abbe refractometer. The amount of lactic acid in the sample was determined by titrating 20ml filtrate obtained from 2g of sample dissolved in 20ml of distilled water against 0.1M NaOH, using phenolphthalein as an indicator[9]. The titer value was then used to calculate the titratable acidity as percentage lactic acid.

$$\% \text{ Lactic acid} = \text{Titre value} \times 0.07 \quad (1)$$

2.2.3. Proximate analysis of the beverages

This was carried out using the method of Association of Official Analytical Chemists [9].

2.2.4. Determination of Vitamin content of beverage blends

2.2.4.1. Determination of vitamin C

The vitamin C content was determined using the ascorbic acid as the reference compound. Two hundred milliliter of the extract was pipette and mixed with 300 ml of 13.3% of TCA and 75 μ l of DNPH. The mixture was incubated at 37°C for 3 h and 500 ml of H₂SO₄ was added and the absorbance was read at 520nm [10].

2.2.4.2. Determination of vitamin A

About 2 g of the sample was mixed with 30 ml of absolute alcohol and 3 ml of 5% potassium hydroxide. The mixture was boiled gently under reflux for 30min in a stream of oxygen-free nitrogen. It was cooled rapidly by adding 30 ml of water and transferred into a separator, where it was washed with ether and the vitamin A was extracted by shaking for 1min. It was then washed evaporated down to about 5ml and the remaining ether was removed in a stream of nitrogen at room temperature. The residue was then dissolved in sufficient isopropyl alcohol, the extinction was measured and the wavelength of maximum absorption was used [11].

2.2.4.3. Determination of Vitamin E

About 1.0g of the sample was measured into 10ml of absolute alcohol with 20ml of 1M alcoholic sulphuric acid. The unsaponifiable matter was then extracted with diethyl ether. The residue was dissolved in 10ml absolute alcohol, the standard and the sample was transferred and 5ml of absolute alcohol was added followed by 1ml conc. nitric acid. The absorbance was measured at 470 nm against a blank containing absolute alcohol [12].

2.2.5. Antibacterial activity of beverage blends

The beverage extracts were tested for activity against bacteria using modified agar-well diffusion method [13]. Nine bacteria were collected from the Microbiology Department of The Federal University of Technology, Akure. Each was subcultured in McCartney bottles using nutrient agar and kept in the refrigerator. The microorganism used include; *Bacillus subtilis*, *Proteus spp.*, *Staphylococcus aureus*, *Serratiamar cescens*, *Bacillus cereus*, *Shigellaspp.*, *Pseudomonas spp.*, *Enterobacter sp.* and *Escherichia coli*. Five-hour broth cultures of the test bacteria adjusted to 10⁸cfu/ml and were applied on the surface of Nutrient

agar (HiMedia Laboratories Limited, Mumbai, India). A sterile flamed cork borer of 8 mm diameter size was used to punch four wells into each of the seeded plates and 0.5 ml of each extract was dispensed in each well. Controls were set up by filling wells with 1% of various solvents used. The plates were then incubated at 35°C for 24 h. The experiments were performed in duplicate and the means of the diameters of the inhibition zones were calculated.

2.2.6. Mineral Determination

Two grams of sample was placed in a crucible, ashed in a muffle furnace at 550°C for 5 h and transferred into the desiccator to cool. The washed sample was dissolved with 1ml nitric acid and 1ml HCl and made up to 100 ml. This was used to analyze for Mg, Na, Ca, K, Mn, Cu, Zn and Fe. The atomic absorption spectrophotometer was used to determine these elements [9].

2.2.7. Phytochemical Analyses

2.2.7.1. Determination of oxalates

Oxalate was determined by soaking 1g of the beverage sample in 75ml of 1.5M H₂SO₄ for 1h and then filtered through a No 1 Whatman filter paper. About 25 ml was taken out of the filtrate, poured inside a conical flask and titrated hot (80-90°C) against 0.1M KMnO₄ until a pink colour that persist for 15 s was obtained [14].

2.2.7.2. Determination of saponins

About 2g of the finely ground sample was weighed into a 250ml beaker and 100ml of n-butanol acid was added. The mixture was shaken for 5 h to ensure uniform mixing. The mixture was then filtered with No 1 Whatman filter paper into 100ml beaker containing 20ml of 40% saturated solution of magnesium carbonate. The mixture obtained was again filtered through No 1 Whatman filter paper to obtain a clean clear solution. About 1ml of the solution was taken into 50ml volumetric flask using pipette, 2ml of 5% iron (iii) chloride (FeCl₃) solution was added and made up to the mark with distilled water. It was allowed to stand for 30min for the colour to develop. The absorbance is read against the blank at 380nm [15].

2.2.7.3. Determination of tannin

About 0.2 g of beverage sample was weighed into 10 ml of 70% aqueous acetone and was properly covered. The bottles were put in an ice bath shaker for 2 h at 30⁰C. Each solution was then centrifuged and the supernatant stored in ice. About 0.2ml of each solution was pipetted into the test tube and 0.8 ml of distilled water was added. Standard tannic acid solutions were prepared and 0.5 ml of Folin Ciocateau reagent was added to both samples and standard followed by 2.5ml of 20% Na₂CO₃. The solutions were vortex and allowed to incubate for 40 min at room temperature, its absorbance was read at 725 nm against a reagent blank [16].

2.2.7.4. Determination of phytate

About 4 g of sample was soaked with 100 ml of 2% HCl for 3 h and then filtered through a No 1 Whatman filter paper. Twenty five milliliter of the filtrate was taken and 5ml of 0.3% of ammonium thiocyanate solution was added as indicator, after which 53.5ml of distilled water was added to give it the proper acidity. This was titrated against 0.00566 g per milliliter of standard iron (III) chloride solution that contained about 0.00195 g of iron per milliliter until a brownish yellow colouration persist for 5min [17].

2.2.7.5. Determination of alkaloids

Five grams of the sample and 200 ml of 10% acetic acid in ethanol was allowed to stand for 4 min in a beaker. This was filtered and extract was concentrated on a water bath to one quarter of the original volume. Concentrated ammonium hydroxide was added dropwise to the extract until the precipitation was completed. The whole solution was allowed to settle and the precipitate was collected and washed with dilute ammonium hydroxide and then filtered. The residue was then alkaloid which was dried and weighed [18].

$$\% \text{ Alkaloid} = \frac{W_3 - W_2}{W_1} \times 100 \quad (2)$$

2.2.8. Evaluation of Antioxidant Properties

2.2.8.1. Determination of total phenol

About 0.2ml of the extract was mixed with 2.5 ml of 10% FolinCiocalteau's reagent and 2ml of 7.5% Sodium carbonate. The reaction mixture was incubated at 45⁰C for 40 min, and the

absorbance was measured at 700nm in the spectrophotometer. Gallic acid was used as standard phenol [19].

2.2.8.2. Determination of total flavonoid

About 0.2ml of the extract was added to 0.3ml of 5% NaNO₃. After 5min, 0.6ml of 10% AlCl₃ was added and after 6min, 2ml of 1M NaOH was added to the mixture followed by the addition of 2.1ml of distilled water. Absorbance was read at 510nm against the reagent blank and flavonoid content was expressed as mg rutin equivalent [20].

2.2.8.3. Determination of ferric reducing property

About 0.25 ml of the sample was mixed with 0.25ml of 200mM of Sodium phosphate buffer at pH 6.6 and 0.25 ml of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20min, thereafter 0.25ml of 10% TCA was also added and centrifuged at 2000rpm for 10min. One millilitre of the supernatant was mixed with 1ml of distilled water and 0.1% of FeCl₃ and the absorbance was measured at 700nm [21].

2.2.8.4. Determination of free radical scavenging ability

About 1ml of the beverage extract was mixed with 1ml of 0.4mM methanolic solution of 1, 1-diphenyl-2-picrylhydrazyl (DPPH), the mixture was left in the dark for 30min before measuring the absorbance at 516nm [22].

2.2.8.5. Test for ABTS scavenging ability

The chemical 2, 2'-azino-bis (3-ethylbenthiazoline-6-sulphonic acid) (ABTS) was used to determine the scavenging ability of the beverage extract. About 0.2ml of the appropriate dilution of the extract was added to 2.0ml of ABTS solution and the absorbance was read at 732nm after 15mins. The Trolox equivalent antioxidant capacity was subsequently calculated [23].

2.2.9. Sensory Evaluation

Sensory evaluation was carried out according to Solomakos *et al.* [24]. The sensory test was carried out at the end of four months after all the analyses. Each panelist was provided with the different beverage blends packaged in a tea bag and Lipton was used as a control.

2.2.10. Statistical Analysis

Data from three replicates were subjected to Analysis of Variance (ANOVA) using statistical package for the social science 17.0 for Windows (SPSS 17.0) and the means separated using Duncan Multiple Range Test (DMRT).

3. RESULTS AND DISCUSSION

3.1. Chemical properties of beverage blend

The pH profile, brix and TTA of the beverages during storage is shown in Table 1a. At the start of the experiment the pH was from 4.40 in 50%, Moringa leaves + 45% Zobo + 5% Lemongrass sample (MZL2) to 5.88 (Lipton), at the second month of storage it ranged from 3.50 to 5.10 and at the fourth month 3.37-4.80. Contrast, the total titratable acidity was found to increase as storage progressed with values of 2.10g/l-2.58g/l at zero-day and 3.40g/l-8.83g/l on the 4th month of storage. The low pH value may probably be as result of zobo added to the blends. This is in line with the observations of Awhin [25], which stated that zobo is slightly acidic with a pH of 3.5 which reduces with time and does not pose any health threat. As the pH value decreased, TTA was found to increase and similar observation was reported [26].

3.2. Proximate composition of beverage blend

The result of proximate composition of beverages on Table 1b revealed that beverage sample with 85% Moringa leaf (MZL1) possessed highest protein content (35.90% -48.17%) followed by Lipton, the control sample (18.16%-40.25%) and the least value was from beverage with 50% Moringa leaf (15.00%-36.22%). The crude fiber content of beverage MZL2 (23.10%-21.42%) and Lipton(34.23% -20.30%) were observed to decrease during storage while sample MZL1 recorded increase in fiber content (21.83% -25.64%).The results of the proximate composition revealed that the beverage became a concentrated source of all the nutrients required for maintenance of good human health. Lakshmi and Vimla [27] showed that moringa leaf contains good amounts of protein and fiber in various dry samples. In addition, protein content of the beverage samples were higher than many of the commonly consumed green leafy vegetables, spinach (2%), mint (4.8%) and 15% - 30% for vegetables [28]. The beverage blends can also be rich sources of fiber, which agrees with the fact that vegetables are natural broom for the body.

3.3. Vitamin content of beverage blend

The vitamin C content (Table 2a) of the beverage samples at 0 day revealed that the values ranged from 51.30 mg/g – 67.50 mg/g, vitamin E (1.87 mg/g-3.47 mg/g) and vitamin A (788.52 unit/g-1009.20 unit/g). The calyces of zobo have been found to be rich in vitamins, natural carbohydrate, protein [29] and minerals [30]. Drastic reduction in vitamin content of the beverage blends and Lipton were observed during the 4 month storage yielding 8.14 mg/g-18.71 mg/g for vitamin C, 1.35 mg/g-2.38 mg/g for vitamin E and 693.19 unit/g-997.24 unit/g for vitamin A. Results further showed that Lipton contained lowest vitamin content while the beverage blend with 85% Moringa leaf (MZL1) possessed the highest values. This result may be an indication that the beverage blends could be rich sources of vitamins to the consumer.

3.4. Mineral composition of beverage blend

The mineral content of beverages is shown in Table 2b. It was observed that the iron content of the beverage blends (3.84 ppm-3.97 ppm) was higher than that of Lipton (2.72 ppm). Iron is an essential component of hemoglobin, an erythrocyte protein that transfers oxygen tissue, which carry the oxygen in the blood and the muscle [31]. Calcium content of the beverage blends ranged from 1.80 ppm-2.87 ppm while Lipton has 1.33 ppm. Calcium is one of the most important minerals for the growth, formation of and maintenance of healthy teeth and reproduction of the human body. Beverage blends contain potassium (0.75 ppm-0.78 ppm) and Sodium (1.98 ppm-2.95 ppm). Sodium and potassium are important in the diet due to their role in blood pressure regulation [32].

3.5. Antibacterial activity of beverage blend

Ethanol extract of beverages were found to produce inhibition zones (11.00 mm -18.00 mm) against some pathogenic microorganisms tested with the exception of *Bacillus subtilis* and *Pseudomonas aeruginosa* and the activity appeared to be stable during storage (Table 3). Aqueous extracts of *M. oleifera* was found to be inhibitory against many pathogenic bacteria, including *S. aureus*, *B.subtilis*, *E. coli*, and *P.aeruginosa* in a dose-dependent manner [33].

3.6. Phytochemical content of beverage blend

Result of the phytochemical content of the beverage samples and control shows that values for saponin (66.45 mg/g -136.62 mg/g) tannin, (10.86 mg/g -23.66 mg/g) and phytate (4.28 mg/g -9.84 mg/g) increased slightly with storage while that of oxalate decreased (Table 4). Ingesting saponins has been linked with a decrease in overall blood cholesterol. Saponins enhance the effectiveness of oral vaccines by improving their absorption because of increasing gut mucosal permeability, which facilitates absorption of large molecules contained in vaccines [34]. When phytic acids binds minerals in the gut, it prevents the formation of free radicals, thus making it an antioxidant. Bioactive components have been found to be associated with the activity of biological systems. The bioactive compounds that have been isolated from the extract [35] explained the inhibitory activity demonstrated by the crude extract from *Eleutherineamericana*. We may infer that the antioxidant and antimicrobial properties demonstrated by the formulated beverage samples were because of the phytochemicals in the leaves.

3.7. Antioxidant activity of beverage blend

The antioxidant properties of beverage blends and Lipton is shown in Figure 1. Results revealed that the ferric reducing antioxidant power (FRAP) of the beverages ranged from 25.62 mg/g-46.24 mg/g, phenol (66.52 mg/g-231.03 mg/g) and flavonoid (17.67 mg/g-42.4 mg/g). The free radical scavenging (DPPH) properties of the beverages were 86.37%-85.32%, 85.58%-81.57% and 81.57%-80.34% for 0 day, 2nd month and 4th month of storage respectively. It was observed that as storage progressed there were increases in the values of FRAP, phenol and flavonoid content of both beverage blends and Lipton while ABTS and free radical scavenging activity (DPPH) was decreasing. Reducing the power of a compound is related to electron transferability of the compound that could lead to the neutralization of free radical [36]. The observed increase in reducing power of the blends during storage suggested that they are good electron donors. Beverage blends obtained from cocoa, zobo and ginger demonstrated excellent antioxidant properties [37].

3.8. Sensory report of beverage blend

The response of the panelist on the sensitivity analyses of the beverage blends is shown in Table 5. The attributes include appearance (6.27-7.13), aroma (6.13-7.47), colour (6.07-6.57), flavor (6.07-7.53) and overall acceptability (6.27-7.22). Lipton was scored significantly

higher than the beverage blends in appearance and overall acceptability, while Lipton and beverage blend MZL2 were significantly higher than MZL1 in flavor and aroma and no significant differences were observed in the scores for color and taste for Lipton and beverage blends. It was observed that the beverage blends were scored above the average for all attributes examined.

4. CONCLUSION

This study has shown that the beverage blends contain essential nutrient needed by the body for maintenance of good health. In addition, the beverage blends possess phytochemicals that exhibited antioxidant properties. It was observed that some of these properties were preserved during the four-month shelf life study. Therefore, packaging of beverage blends in this form could be a very good way to improve the accessibility of people to medicinal plants.

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TABLE 1A- Chemical properties of fresh and stored beverage blends at four months

Storage time	Beverage blends	pH	Brix (⁰ Bx)	Total titratable acidity (g/l)
0 day	Lipton	5.88±0.02 ^a	1.00±0.00 ^a	2.10±0.020 ^c
	MZL1	5.76±0.02 ^a	0.80±0.10 ^a	2.58±0.02 ^a
	MZL2	4.40±0.02 ^b	0.90±0.10 ^a	2.29±0.01 ^b
2months	Lipton	5.10±0.11 ^a	1.00±0.0 ^b	5.02±0.01 ^a
	MZL1	4.90±0.02 ^b	1.17±0.06 ^a	3.27±0.01 ^b
	MZL2	3.50 ±0.02 ^c	0.83±0.06 ^c	2.29±0.01 ^b
4months	Lipton	4.80±0.10 ^b	1.10±0.01 ^a	8.35±0.30 ^a
	MZL1	4.50±0.02 ^b	1.00±0.01 ^b	3.40±0.30 ^b
	MZL2	3.37±0.46 ^c	0.90±0.01 ^c	8.83±0.34 ^a

Values are means ± standard deviation of three determinations. Values with different superscripts along columns are statistically significant p≤0.05

Lipton (control);MZL1 = 85% Moringa leaves + 10% Zobo + 5% Lemongrass;

MZL2 = 50% Moringa leaves + 45% Zobo + 5% Lemongrass

TABLE 1B-Proximate composition (%) of fresh and stored beverage blends after four months

Storage time	Sample	Moisture	Fat	Crude fiber	Protein	Ash	CHO
0 day	Lipton	14.3±0.58 ^a	5.67±2.52 ^c	34.23±0.25 ^a	18.16±0.29 ^b	6.45±0.4 ^c	21.17±2.02 ^a
	MZL1	16.67±2.89 ^a	11.67±2.52 ^a	21.83±0.76 ^c	35.90±0.36 ^a	10.12±0.34 ^a	5.05±0.78 ^b
	MZL2	17.33±1.53 ^a	9.33±2.89 ^b	23.10±0.36 ^b	15.00±0.20 ^c	10.50±0.70 ^a	24.73±0.61 ^a
2months	Lipton	11.45±0.04 ^c	4.16±0.2 ^c	31.50±1.32 ^a	22.0±1.0 ^b	5.48±0.2 ^c	26.74±1.87 ^b
	MZL1	14.38±0.48 ^{ab}	6.72±0.86 ^a	22.99±2.06 ^c	40.2±0.2 ^a	9.85±0.01 ^a	5.86±1.48 ^c
	MZL2	15.23±0.32 ^a	6.63±0.86 ^a	23.00±0.21 ^c	16.62±0.35 ^c	7.86±2.75 ^b	30.67±3.80 ^a
4months	Lipton	10.99±0.96 ^a	5.82±0.83 ^{ab}	20.30±0.01 ^e	40.25±0.05 ^b	5.07±0.01 ^a	17.56±1.80 ^b
	MZL1	7.48±0.43 ^b	4.68±0.43 ^c	25.64±0.01 ^a	48.17±0.29 ^a	4.8±0.05 ^b	9.23±0.98 ^c
	MZL2	6.80±1.01 ^b	6.27±0.15 ^a	21.42±0.03 ^d	36.22±0.23 ^c	3.98±0.01 ^c	25.31±1.20 ^a

Values are means ± standard deviation of three determinations. Values with different superscripts along columns are statistically significant $p \leq 0.05$.

Lipton (control);

MZL1 = 85% Moringa leaves + 10% Zobo + 5% Lemon grass;

MZL2 = 50% Moringa leaves + 45% Zobo + 5% Lemon grass

TABLE 2A- Vitamin content of beverage blends after four months of storage

Storage time	Sample	Vitamin C (mg/g)	Vitamin E (mg/g)	Vitamin A (unit/g)
0 day	Lipton	51.30±1.77 ^b	1.87±0.01 ^c	788.52±0.00 ^a
	MZL1	53.49±0.45 ^b	2.10±0.01 ^c	845.76±0.0 ^a
	MZL2	67.50±28.93 ^b	3.47±0.01 ^c	1009.20±0.0 ^a
2 month	Lipton	20.52±5.04 ^b	1.58±0.01 ^c	711.05±0.04 ^a
	MZL1	12.79±0.04 ^b	1.84±0.01 ^c	791.43±0.04 ^a
	MZL2	50.99±8.41 ^b	2.79±0.01 ^c	998.12±0.01 ^a
4 month	Lipton	18.71±2.57 ^b	1.35±0.01 ^c	693.19±0.01 ^a
	MZL1	8.14±0.64 ^b	1.54±0.02 ^c	721.19±0.01 ^a
	MZL2	15.33±0.26 ^b	2.38±0.01 ^c	997.24±0.01 ^a

Values are means ± standard deviation of three determinations. Values with different superscripts along rows are statistically significant $p \leq 0.05$

Lipton (control);

MZL1 = 85% Moringa leaves + 10% Zobo + 5% Lemon grass;

MZL2 = 50% Moringa leaves + 45% Zobo + 5% Lemon grass

TABLE 2B-Mineral composition (ppm) of beverage blends at day 0

Minerals	Lipton	MZL1	MZL2
Potassium	0.85±0.0 ^a	0.75±0.0 ^b	0.78±0.0 ^b
Sodium	2.30±0.58 ^b	1.98±2.52 ^c	2.95±0.155 ^a
Magnesium	0.53±0.03 ^c	1.10±0.10 ^a	0.85±0.05 ^b
Calcium	1.33±0.06 ^c	2.87±0.06 ^a	1.80±0.0 ^b
Iron	2.72±0.03 ^b	3.97±0.01 ^a	3.84±0.01 ^a
Zinc	2.51±0.01 ^a	1.61±0.01 ^b	2.49±0.01 ^a
Copper	1.61±0.01 ^a	0.26±0.01 ^b	1.63±0.01 ^a
Manganese	0.37±0.01 ^a	0.23±0.01 ^b	0.32±0.03 ^a
Phosphorus	0.21±0.02 ^a	0.16±0.01 ^b	0.26±0.01 ^b

Values are means ± standard deviation of three determinations. Values with different superscripts along rows are significantly different $p \leq 0.05$.

MZL1 = 85% Moringa leaves + 10% Zobo + 5% Lemon grass;

MZL2 = 50% Moringa leaves + 45% Zobo + 5% Lemon grass;

TABLE 3. Antibacterial activities (mm) of beverages from day 0 to 4th month of storage

Bacteria	0 day			2 months			4 months		
	Lipton	MZL1	MZL2	Lipton	MZL1	MZL2	Lipton	MZL1	MZL2
<i>Bacillus subtilis</i>	0	0	0	0	6	3	0	0	0
<i>Enterobacter Aerogenes</i>	18	11	16	11	16	13	16	12	6
<i>Pseudomonas aeruginosa</i>	3.2	0	0	0	0	0	13	4	5
<i>Proteus mirabilis</i>	17	18	11	8	8	9	16	8	13
<i>Serratia marcescens</i>	13	16	18	17	18	14	21	6	12
<i>Shigella flexneri</i>	11	0	0	11	0	8	23	8	8
<i>Escherichia coli</i>	13	14	16	10	16	14	18	12	10
<i>Bacillus cereus</i>	18	13	18	11	10	10	16	12	12
<i>Staphylococcus aureus</i>	15	0	0	11	0	0	0	0	0

TABLE 4-Phytochemicals of fresh and stored beverage blends after four months

Storage time	Sample	Saponins (mg/g)	Tannins (mg/g)	Oxalate (mg/g)	Phytate (mg/g)	Alkaloids (%)
0 day	L	64.30±0.01 ^c	12.55±0.13 ^a	1.45±0.01 ^c	9.84±0.0 ^a	3.55±0.07 ^c
	MZL1	66.45±0.64 ^b	10.86±0.3 ^b	1.84±0.05 ^b	9.84±0.06 ^a	10.3±0.28 ^a
	MZL2	67.40±0.13 ^a	11.14±0.07 ^b	2.85±0.05 ^a	4.28±0.06 ^b	7.2±0.28 ^b
2 Months	L	53.38±0.16 ^c	13.50±1.56 ^a	1.40±0.19 ^b	4.13±0.01 ^b	4.56±0.06 ^c
	MZL1	68.45±0.04 ^b	11.15±0.28 ^{ab}	0.58±0.07 ^c	4.13±0.01 ^b	12.2±0.23 ^a
	MZL2	69.40±0.10 ^a	11.15±1.21 ^{ab}	2.71±0.25 ^a	5.69±0.12 ^a	8.3±0.28 ^b
4 Months	L	15.0±14.14 ^c	23.66±1.34 ^a	0.62±0.01 ^c	3.35±0.07 ^c	9.5±0.14 ^b
	MZL1	136.62±0.16 ^a	20.35±0.48 ^{bc}	0.73±0.01 ^b	4.06±0.08 ^b	17.15±0.92 ^a
	MZL2	101.41±0.58 ^b	22.98±0.38 ^{ab}	0.82±0.01 ^a	8.17±0.10 ^a	15.95±0.35 ^a

Values are means ± standard deviation of three determinations. Values with different superscripts along columns are significantly different.

L = Lipton (control);

MZL1 = 85% Moringa leaves + 10% Zobo + 5% Lemon grass;

MZL2 = 50% Moringa leaves + 45% Zobo + 5% Lemon grass

TABLE 5-Sensory evaluation of the beverage blends at day 0

Sample	Appearance	Aroma	Colour	Flavor	Taste	Overall acceptability
L	7.13±0.90 ^a	7.47±0.70 ^a	6.57±2.33 ^a	7.53±0.42 ^a	7.40±0.60 ^a	7.22±0.40 ^a
MZL1	6.27±1.14 ^b	6.13±0.90 ^b	6.07±1.70 ^a	6.07±1.72 ^b	6.80±0.90 ^a	6.27±0.20 ^b
MZL2	6.60±1.11 ^{ab}	6.80±1.60 ^a	6.07±1.30 ^a	6.93±0.20 ^a	6.80±0.60 ^a	6.64±0.30 ^{ab}

Values are means ± standard deviation of three determinations. Values with different superscripts along columns are significantly different.

L = Lipton (control);

MZL1 = 85% Moringa leaves + 10% Zobo + 5% Lemon grass;

MZL2 = 50% Moringa leaves + 45% Zobo + 5% Lemon grass

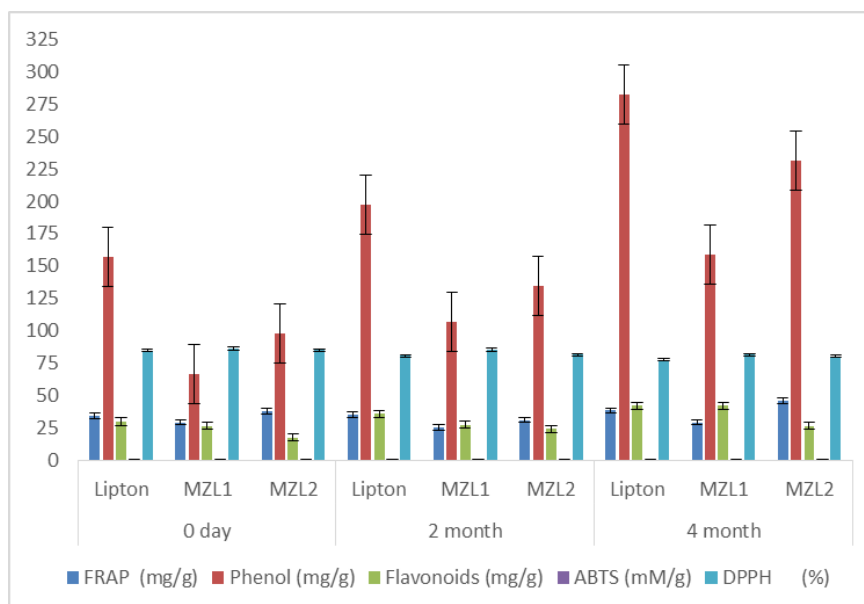


FIGURE 1-Antioxidant properties of beverage blends from day 0 to 4months of storage