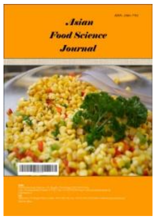

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Production and Physiochemical Evaluation of Fresh Cow Milk-based Beverage

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Authors' contributions

This work was carried out in collaboration among all authors. Author CNL designed and conducted the experiment, did the statistical analysis. Authors QMA and SPM contributed in the supervision of the experiment, paper formatting, grammar correction and approved the final manuscript. All authors read and approved the final manuscript.

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ABSTRACT

The aim of this work was to produce and evaluate the physiochemical properties of cow milk based beverage made from a blend of cow milk, honey, cocoa powder, ginger and garlic. The composition of the samples were as follows: sample A (1 L cow milk), sample B (1 L cow milk, 0.12 L honey, 25 g cocoa powder), sample B served as base ingredients for samples C and D and prepared 1% ginger, 0.5% garlic were added to samples C and D, respectively. Sample A and sample E (*vitamilk*), respectively served as control I and control II. The chemical properties and proximate composition of the milk beverage samples were investigated. The result of chemical properties showed that the pH values ranged from 6.11 to 6.52 and were all slightly acidic while the percentage titratable acidity had the highest value of 0.168 for sample D and the least value of 0.086 for sample A. In terms of proximate composition, sample A had the highest percentage moisture content of 90.75±0.21 while sample C had the least moisture content of 87.7±0.28. The %

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protein, carbohydrate, fat, and ash contents increased as 1% ginger and 0.5% garlic were added to samples C and D and were significantly difference (P<0.05) for all the samples. Sample C had the highest value of 5.15% protein content while sample A had the least value of 2.85%. The percentage ash content ranged from 0.96±0.007 to 0.58±0.007. The fiber content of all the samples were below 2.25±0.007. A similar trend was also observed for the values of minerals and vitamins which showed a significant difference (P<0.05). This work has shown that there is improved nutritional content of the samples B, C and D which will go a long way to alleviate malnutrition. The pH of all the samples were near neutral, this could negatively affect the shelf stability of the milk beverage as a result of possible microbial infestation and thus there is every need to keep the product always refrigerated before use or should be consumed fresh.

Keywords: Production; fresh milk; beverage; physiochemical; samples.

1. INTRODUCTION

Milk is a complex nutritious product of mammary gland secretion containing water, protein, fat, carbohydrate mainly lactose, mineral and vitamins [1]. Amongst all food, milk has the most complete and balanced nutritional contents. Milk is an essential component of the diet of about 6 billion people and the world production of milk stands at 730 million tons/year [2]. Milk and milk beverages, if sufficiently available, could efficiently cure many diseases caused by deficiencies in most Africans' diets. Apart from improving nutrition and health of members of a household, dairy also increases farmers' incomes [3]. Milk products are important commodities in the world market, providing many possibilities for new value added products to meet consumers' demand for convenience, nutrition, and health. Milk products are main constituents of the daily diet, especially for vulnerable groups such as infants, school children and the aged [4]. In rural Africa, approximately 70% of milk is utilized in the liquid form, while various indigenous dairy products such as cheese and yoghurt are produced under unhygienic conditions and practices [5]. Microbial contamination of milk and dairy products is a universal problem which affects dairy farmers leading to spoilage of milk and subsequent financial loss in profit.

Processing of dairy products gives small–scale dairy producers higher income than selling fresh milk and offers better opportunities to reach regional and urban markets and helps to deal with seasonal fluctuations in milk supply. The transformation of fresh milk into processed milk and milk products can benefit entire communities by generating off–farm jobs in milk collection, transportation, processing and marketing [6]. Natural ingredients may also be added to milk to produce different dairy products, add value to milk and above all extend the shelf life of the milk product. The quantities of these ingredients added to milk may be varied to obtain a product with good sensory properties, reduced microbial load, acceptable pH levels, and longer storage period.

Milk is the most complete food rich in nutrients complimenting those obtained from other animals and plant sources [7]. Natural ingredients such as honey, cocoa powder, garlic and ginger also contain good quantities of these nutrients thus, adding them to fresh milk is enhancing the nutritional preference of consumers in terms of improving human health and nutritional balance. Furthermore, these ingredients contains active components which play responsive roles in human health and nutrition: honey (peroxides and sugars), cocoa powder (antioxidants), ginger (gingerol, zingerone, shogaol) and garlic (allicin).

Keeping in mind the threats posed by spoilage and pathogenic microorganisms to both the milk industry and the smallholder dairy farmers, several procedures have to be applied in order to extend the shelf life of milk while adding value to it. This will drastically reduce the incidence of losses due to spoilage. This could be done by the production of milk beverage produced from cow milk using natural ingredients like garlic, ginger, honey and cocoa powder. These ingredients contain carbohydrates, proteins, fats, fiber, minerals and vitamins in varying proportions, that combine with milk to boost the nutrients intake of consumers [8].

In dietary guidelines for Americans, 2010, milk and its products are recommended because of their positive effect to dietary intake. In addition to the nutrients mentioned earlier, cow milk contains vitamins like A, B and D, minerals like calcium, phosphorus, potassium and iodine, which contribute to multiple and different vital functions in the human body. Calcium from these natural ingredients has good effect on strengthening the bones and potassium is known to control blood pressure [8]. To obtain the valuable nutritional benefits of milk, natural ingredients may also be processed with milk.

The main aims of this study were to develop milk beverage from a blend of cow milk and some natural ingredients (honey, cocoa powder, ginger, and garlic); to evaluate the physiochemical properties of the milk beverage samples developed, that are of particular interest for human health, thus giving improved nutritional composition of the milk for human consumption.

2. MATERIALS AND METHODS

2.1 Procurement of Materials

Garlic, ginger, honey cocoa powder and *Vitamilk* were purchased from Wurukum market, Makurdi. Cow milk was bought from vendors at Wadata market, Makurdi, Benue State, Nigeria, in September prior to production.

2.2 Preparations of Materials

2.2.1 Milk

Fresh cow milk was bought from vendors at Wadata market, Makurdi, Benue State, very early in the morning. It was strained and put into sterile containers and then pasteurization followed.

2.2.1.1 Pasteurization of the fresh milk

During the pasteurization process, 3 L of milk at a time was put in a double boiler over medium-high heat. A thermometer was probed into the milk and the milk was frequently stirred to help equalize temperature and prevent scorching. As the temperature reached 63°C, the heat was reduced and this temperature was maintained for 30 minutes [9]. It was then transferred into 1 L containers and cooled rapidly in an ice bath prior to processing.

2.2.1.2 Detection of adulteration in the milk

The fresh cow milk was tested for adulterants common the area namely: table sugar and water. The lactometer test for sugar showed negative while the water content of the milk was slightly higher than normal. It showed a value of 90.75% (Table 3) as compared to about 87.6% for unadulterated milk.

2.2.2 Ginger

Fresh mature rhizomes of ginger were separated, thoroughly washed, peeled and sliced (about 2 mm thickness) with stainless steel knife and then oven dried at 60°C for 48 h. It was then ground into powder using electric blender. The powder was sieved with a wire mesh of 4mm aperture to remove very rough particles sizes. The fine ginger flour was then kept in sterile bottles for further use.

2.2.3 Garlic

After removing the outer cover, garlic cloves were peeled off; washed with clean water; sliced with a stainless steel knife; and then oven dried at 60°C for 48h. It was then ground into powder using electric blender. The powder was further sieved with a wire mesh of 4mm aperture to reduce particle sizes. The fine flour was packed in sterile glass bottle and stored in dry and clean area according to Douglas et al. [10].

2.2.4 Honey

Pure, natural and unadulterated honey was bought and used in its natural form.

2.3 Recipes Formulation for Milk Beverage Samples

Composition of sample E was as seen on the packaging label. Modified sources: Regu , Abiola et al. [11,12].

2.4 Production of Milk Beverage Samples

Honey and cocoa powder were added to pasteurized cow milk and mixed using an electric blender to obtain a homogenized mixture (sample B). Replicates of this mixture served as the based ingredients for samples C and D. 1% of ginger powder was added to the base ingredients to obtain sample C while 0.5% of garlic powder was added to the base ingredients to produce sample D. Each sample produced was blended and filtered with a 0.2 mm aperture filter cloth. They were then stored in 60cL labelled sterile containers prior to physiochemical evaluation.

2.5 Physiochemical Analyses

All the physiochemical analyses of the samples were carried out in the Food Science and Biochemistry Laboratories of Benue State University, Makurdi, Nigeria.

Table 1. Milk beverage composition per sample

Component	Milk beverage composition per sample				
	A	B	C	D	E
Cow milk (L)	1	1	1	1	0.33
Honey (L)	0	0.12	0.12	0.12	0.05
cocoa powder (g)	0	25	25	25	8
Ginger (g)	0	0	10	0	3.5
Garlic (g)	0	0	0	5	0

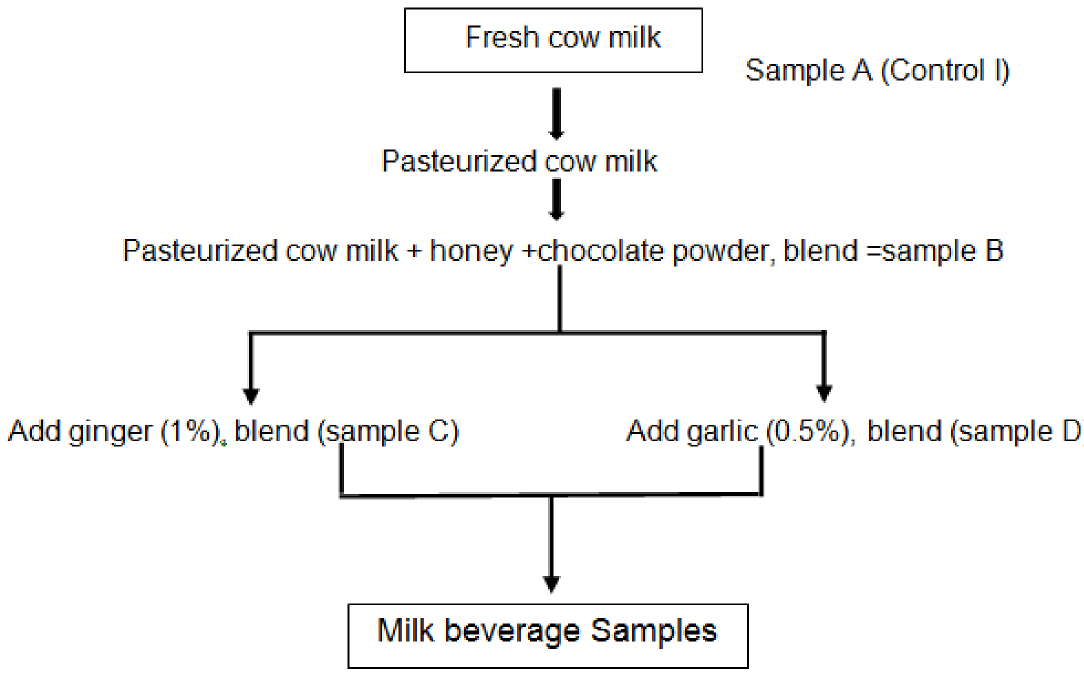

Fig. 1. Flow chart for the production of the milk beverage samples
**Sample E is Vitamilk obtained from grocery shop (control II)*

2.5.1 Determination of pH

The pH was determined at room temperature using a digital pH meter. The pH meter was calibrated with buffer standards of pH 4 and pH 10 prior to use. 50 mL of each sample was placed in a beaker and the probe of the pH meter inserted into each sample and pH value recorded [13].

2.5.2 Measurement of titratable acidity

The titratable acidity was measured by titrating 15 mL of each milk beverage sample with 0.1 M sodium hydroxide until the substance reached a pH value 8.2, corresponding to the end point of phenolphthalein. Readings were done using pH meter [14,15]. When this value was reached, the spent NaOH volume was recorded and the acid percentage of the substance was calculated using the formula:

$$\text{Titratable acidity} = \frac{\text{Titre value x M x 90 x 100}}{\text{Volume of sample x 1000}}$$

Where,

M = Molar concentration of NaOH

2.5.3 Proximate analysis

2.5.3.1 Moisture content determination

Moisture content was determined using the oven-drying method as outlined by AOAC, [16]. Two (2) grams of samples was weighed into preweighed dry crucibles and heated in hot air oven at 105°C for 3 h. Cooling and weighing of dried samples were done at intervals of 30 minutes until there was no weight change after three successive weight measurements. The electronic balance was used in taking weight measurements. The final weight of dried samples was noted and the moisture content calculated as:

$$\%Moisture= \frac{(\text{Wt of crucible+Fresh}) - \text{Wt of crucible+dry sample}}{\text{Weight of fresh sample}} X100$$

2.5.3.2 Determination of protein

The amount of protein was determined by total-Kjeldahl Nitrogen method according to AOAC, [16]. Two (2 g) of sample was digested in concentrated sulphuric acid with one Kjeldahl tablet followed by neutralization using 40% sodium hydroxide and distillation. The resulting solution was titrated with 0.1N hydrochloric acid using a mixed indicator (methyl red and bromocresol green). The Percent nitrogen (N) concentration was calculated using the following equation

$$\%\ Nitrogen= (S-B) \times N \times 0.014 \times D \times \frac{100}{W} \times V$$

Where,

D = dilution factor, T = titre value = (S-B), W = weight of sample,

Crude protein was obtained by multiplying the corresponding total nitrogen (N) content by a conventional factor of 6.25

Thus: Crude protein (%) =% N ×6.25

2.5.3.3 Determination of fat

Crude fat was determined by the Soxhlet Extraction method as outlined by [16]. Two (2) grams of dried samples was weighed into preconditioned and weighed (W0) extraction thimble and placed in the Soxhlet Extraction apparatus. Fat content of the samples was extracted using organic solvent (petroleum ether) and boiled under reflux for 6 hours. The extraction thimbles was then removed and dried in an oven at 105^{0}C for 30 minutes then cooled and weighed (W1). Percentage of fat content was calculated using the following formula:

$$\%\ Crude\ fat = \frac{\text{Weight of fat in sample}}{\text{Weight of dry sample}} X100$$

$$= \frac{W0-W1}{\text{Weight of dry sample}} X\ 100$$

2.5.3.4 Determination of total ash

Two (2) grams of samples was weighed in dry crucibles, carbonized on a hot plate and heated on a muffle furnace at 600°C for 8 hours after which it was cooled in a desiccator and weighed. Ash content was determined by the difference in weight after cooling the samples in desiccators to ambient temperatures AOAC, [16].

$$\%\ ash\ content = \frac{\text{Weight of incinerated sample}}{\text{Weight of fresh sample}} X100$$

2.5.3.5 Determination of carbohydrate content

The lactose content was estimated by the method, as described by AOAC, [16] according to the following equation:

% Carbohydrate =100 − (moisture + fat + protein + ash) %

2.5.3.6 Determination of mineral content

Analysis of potassium(K) and phosphorus (P) contents of the samples were carried out using flame photometry method AOAC, [17]. The other elemental contents (Ca, Mg and Zn) were determined, after wet digestion of sample ash with an Atomic Absorption Spectrophotometer [17]. All determinations were carried out in duplicates.

2.5.3.7 Determination of vitamin content

Vitamins A and D were determined according to the methods of AOAC, [17].

3. RESULTS AND DISCUSSION

The result obtained from the physiochemical analysis of milk beverage samples are presented in the Tables 2, 3, 4 and 5.

3.1 pH and Titratable Acidity

The results shown in Table 2 revealed that the mean pH values of the five milk beverage samples ranged from 6.3-6.52. The mean titratable acidity values of samples A, B, C, D and E were 0.116, 0.150, 0.136, 0.168, and 0.086 respectively. pH is a measure of the hydrogen ions (H+) activity in solution. The pH values of all the samples were, however, lower than those reported by [18] but higher than those reported by [19]. The mean pH values obtained for the five samples of milk beverage analyzed from this work indicated that they were near neutral pH and since lower pH (acidic) in food helps to reduce the activity of spoilage microorganism it implies that all the milk beverage samples may have low shelf stability and should be stored under refrigeration or should be consumed fresh.

Titratable acidity (TTA) measures the predominant acid present in the milk beverage samples. From the results presented in Table 2, the values of TTA were slightly less than those reported by Klaenhammer et al. [18]; they cited 0.19%. The TTA usually increases with storage due to increase production of lactic acid by lactic acid producing bacteria. The score of 0.17% or less of TTA is desirable for safe consumption of the product according to International Food Standard (IFS).

3.2 Proximate Composition

The mean values of the proximate composition such as moisture, carbohydrate, protein, fat, ash and fiber contents of the formulated milk beverage samples are presented in Table 3. Few reports were found in the literature concerning the determination of the chemical composition of unfermented milk drinks. Therefore, the results of the present work were compared with more of fermented dairy drinks. Proximate composition is important in determining the quality raw materials and it is often used as a basis of establishing the nutritional value and overall acceptance of the formulated food products [20].

From the results, the moisture of the milk beverage samples ranged from 87.7% (sample C) to 90.75% (sample A). Moisture provides a measure of the water content of a food material and it is also an index of storage stability of food products [21]. High moisture contents in food has been shown to encourage microbial growth [22]. In this work, sample C had the least percent of moisture content and this could be justified by the thickness of the sample due to the dry ingredients added like ginger and cocoa powder. A similar trend is observed in sample D which had dry garlic. From the results, there was significant difference ($P < 0.05$) in moisture content of milk beverage samples. Sample A had the highest moisture content and this could be due to adulteration of milk by unscrupulous farmers or milk dealers. Barham et al. [23], reported that water is the easiest way and cheap source for adulteration of milk. These values of moisture content are almost equal with those reported by Johanson et al. [19] and Dublin-Green et al. [19,24] and all these values were close to the earlier findings, from 80% to 90% [19]. Water serves as a medium for solution and colloidal suspension for the other components present in milk.

The carbohydrate content of the samples ranged from 3.95% (sample A) to 6.63% (sample C). The mean values of the carbohydrate content of the milk beverage samples were all significantly different at 5% level of significance. The difference was accounted for the fact that honey, cocoa powder, and ginger contributed a huge amount of carbohydrate in samples B, C and D coupled with the lactose in cow milk. Sample A (control I) had only lactose. Eckles et al. [25], reported that lactose, or milk sugar, is the main carbohydrate found in milk. It's broken down into its subunits - glucose and galactose in human digestive system.

The protein content of the milk beverage samples ranged from 2.85% (sample A) to 5.15% (sample C). The mean values of the protein content for the milk beverage samples were significantly different at $P<0.05$. The reason for sample C having the highest protein content could be that the ginger added to the sample contained substantial amount of protein that added up to the protein content of the milk beverage sample. Garlic also contains a small quantity of protein and with other ingredients gave sample D a protein value of 4.31%. Also honey, cocoa powder contain good quantities of protein and this accounted for the high protein content in samples B, C and D. These values are higher than those reported by Eckles et al., [25] because of the above mentioned reason. Proteins help to build and maintain lean muscle. The high protein and calorie content of the ingredients added to milk beverage samples could solve the problem of protein-calorie malnutrition in Africa especially for children and the elderly.

The fat content of the milk beverage samples ranged from 1.84% (sample E) to 4.86% (sample C). The fat content of all the samples were significantly different at $P<0.05$. The milk beverage sample with the highest value was sample C (4.86%) and the lowest was sample E (1.84%). Cocoa powder and ginger contain fat that may have added up to the existing fat in cow milk in sample B, C and D. Sample E (control II) may have been partially defatted.

The ash content of the formulated milk beverage samples ranged between 0.58% (sample E) and

0.96% (sample C). The ash contents of the milk beverage samples were significantly different at P<0.05 for all the samples. Sample C had the highest ash content of 0.95% while the least was sample A. The value of ash content increased in sample D when ginger was added to the sample. Sample C that had ginger gave the highest ash content and the samples with cocoa powder had similar increase in ash content. Olusegun et al. [26], reported increase in the ash contents of *kunun zaki* enriched with extract of sesame seeds over the control sample without the extract. Ash content is an indication of mineral content of the milk beverage samples.

The fiber content of the milk beverage samples ranged from 0.00% (sample A) to 2.25% (sample C). From the result presented in Table 3, the crude fiber contents were significantly different at P<0.05 for all the samples. Sample A (control I) had the lowest while sample C had the highest. Empty cow milk (sample A) has no fiber but ingredients like cocoa powder, ginger (sample C), garlic (sample D) had a substantial quantity of fiber and that accounted for high fiber in those samples. Thus, the fiber present in these milk beverage samples may contribute to positive effects on the consumer's health considering the induced changes in intestinal microbiota and systemic effects on the host's intestinal lumen [27].

3.3 Minerals and Vitamins

3.3.1 Minerals

The results of mineral content of the milk beverage samples presented in Table 4 showed that the concentration of Ca, Zn, K, P and Mg in the milk beverage samples ranged from 93.76 mg/100mL (sample A) to 193.37 mg/100 mL (sample C), 1.765 mg/100 mL (sample E) to 3.115 mg/100 mL (sample C), 271.02 mg/100 mL (sample A) to 585.63 mg/100 mL (sample C), 51.76 mg/100 mL (sample A) to 88.125 mg/100 mL (sample C) and 33.62 mg/100 mL (sample E) to 88.125 mg/100 mL (sample C). Generally, the mineral potassium (K) from the sample A had the highest value of 413.835mg/100 mL, while zinc (Zn) has the least value of 1.655 mg/100 mL from the milk beverage sample E. The increase in the quantity of these mineral is due to the addition of ginger (sample C), garlic (sample D), honey and cocoa powder (samples B, C and D). Minerals are nutritionally important components in food and they could be classified as essential or non-essential elements. Minerals are essential for health and as such are part of all aspect of cellular function and they are involved in structural components of human beings [28]. Some mineral elements form an integral part of enzyme or protein structure. They are vital for normal growth, maintenance, effective immune system and prevention of cell damage [28]. Calcium (Ca) together with phosphorus (P) help build and maintain strong bones and teeth and reduce the risk of stress fractures and osteoporosis later in life. They also play a role in promoting normal blood pressure [29].

3.3.2 Vitamins

The lowest value of vitamin A was recorded in sample E (27.1 µg/100mL) and the highest value was in sample C (33.8 µg/100mL). For vitamin D, the highest mean score was recorded by sample C (0.551 µg/100mL) while the least was recorded by sample E (0.335 µg/100mL).The vitamins examined in this work were vitamins A and D (fat soluble). The means values for Vitamins A and D of the milk beverage samples are shown in Table 5. The high values of vitamins A and D in sample C could be as a result of addition of honey, cocoa powder and ginger which content a certain quantity of these vitamins. Fulgoni et al. [29], reported that cocoa powder contains vitamins A, D, and other water soluble vitamins.. Vitamin A is very important for good vision, healthy skin, and a healthy immune system while vitamin D helps absorb calcium for healthy bones [29].

Table 2. Chemical properties of milk beverage samples

Samples	pH	% Titratable Acidity (TTA)
A	6.320 ± 0.014^{bc}	0.116 ± 0.015^{b}
B	6.495 ± 0.007^{d}	0.150 ± 0.010^{d}
C	6.520 ± 0.014^{de}	0.136 ± 0.005^{c}
D	6.300 ± 0.014^{b}	0.168 ± 0.005^{e}
E	6.110 ± 0.014^{a}	0.086 ± 0.005^{a}
ANOVA (sig.)	0.001	0.001

Values are mean ± standard deviation of mean. Values with different superscripts are significantly different (p<0.05) for Duncan multiple range test

Table 3. Proximate composition of milk beverage samples

Samples	Moisture (%)	Carbohydrate (%)	Protein (%)	Fat (%)	Ash (%)	Fiber (%)
A	90.75±0.21[e]	3.95±0.07[a]	2.85±0.07[a]	2.55±0.07[bc]	0.68±0.007[ab]	0.00±0.00[a]
B	88.45±0.07[bc]	4.85±0.07[c]	4.26±0.07[c]	2.34±0.07[b]	0.83±0.007[c]	1.35±0.07[b]
C	87.70±0.28[a]	6.63±0.07[e]	5.15±0.07[e]	4.86±0.07[e]	0.96±0.007[e]	2.05±0.07[e]
D	88.15±0.07[c]	5.15±0.07[d]	4.31±0.07[d]	2.96±0.07[d]	0.87±0.007[d]	1.63±0.04[d]
E	88.65±0.07[d]	4.46±0.07[b]	3.93±0.07[b]	1.84±0.07[a]	0.58±0.007[a]	1.48±0.04[c]
ANOVA (Sig.)	0.001	0.001	0.001	0.001	0.001	0001

Values are mean ± standard deviation of mean. Values with different superscripts are significantly different (p<0.05) for Duncan multiple range test

Table 4. Minerals (mg/100 mL)

Samples	Ca	Zn	K	P	Mg
A	93.760±0.014[a]	1.815±0.007[b]	271.020±0.014[a]	51.765±0.007[a]	34.515±0.007[c]
B	146.035±0.007[c]	2.850±0.000[c]	415.735±0.007[c]	80.435±0.007[c]	33.955±0.007[b]
C	193.370±0.014[e]	3.155±0.007[e]	585.635±0.007[e]	88.125±0.007[e]	58.660±0.014[e]
D	173.155±0.007[d]	3.045±0.007[d]	511.025±0.007[d]	84.355±0.007[d]	45.160±0.014[d]
E	143.950±0.000[b]	1.765±0.021[a]	345.260±0.014[b]	73.375±0.007[b]	33.625±0.007[a]
ANOVA (Sig.)	0.001	0.001	0.001	0.001	0.001

Values are mean ± standard deviation of mean. Values with different superscripts are significantly different (p<0.05) for Duncan multiple range test

Table 5. Vitamins (µg/100 mL)

Samples	Vit A	Vit D
A	28.6±0.007[b]	0.352±0.000[b]
B	31.3±0.000[d]	0.515±0.007[c]
C	33.8±0.014[e]	0.551±0.014[e]
D	32.3±0.014[c]	0.457±0.014[d]
E	27.1±0.007[a]	0.335±0.021[a]
ANOVA (Sig.)	0001	0.001

Values are mean ± standard deviation of mean. Values with different superscripts are significantly different (p<0.05) for Duncan multiple range test

4. CONCLUSION

This study showed that the five milk beverage samples were conveniently produced from a blend of cow milk and natural ingredients: honey, cocoa powder, ginger and garlic. These ingredients provided an opportunity for value to be added to cow milk which in its natural form is not palatable and do not yield high profit. The milk beverage produced is an excellence source of vitamins, calcium, phosphorus, potassium, proteins and other nutrients so should be consumed by both children and adults. The pH of all the samples were near neutral pH, this could negatively affect the shelf stability of the milk beverage samples as a result of possible microbial infestation and thus there is every need to keep the products always under refrigeration before use or should be consumed fresh.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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