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> > **Original Research Article**

Nutritional, Functional and Sensory Attributes of Ready-to-use Cocoa Beverage

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Abstract

Increased consumer awareness of the health advantages of plant-based foods and beverages has resulted in a preference for functional beverages over traditional sugar-laden drinks and beverages that just satisfy thirst but give little or no nutritional or health benefits. This study carried out the formulation and sensory evaluation of cocoa beverages enriched with calcium and zinc. Nutritional composition, functional properties and anti-nutritional factors were investigated using standard analytical methods of the Association of Official Analytical chemist. Data were analyzed by ANOVA and results were expressed as mean and standard deviation. The result of proximate indicates that moisture, ash, and fibre content were within the normal range. The result also shows that the blend formulated contained an appreciable amount of vitamin A and minerals elements such as K, Na, Ca, and Zn which are public health importance. The result of functional properties indicate that bulk density range from $(0.763 - 0.579 \text{ g/cm}^3)$, swelling index (0.7 - 0.32 ml/g), reconstitution index (3.0 - 1.1 g/ml), water absorption capacity (0.6 - 0.58 ml/g) and pH (6.78-6.63). Therefore, on the basis of this study, it can be concluded that the formulated blends serve can serve as a good sources of macro and micro nutrients to combat hidden hunger.

Keywords: Beverage, Cocoa, Micronutrients, Functional properties and hidden hunger.

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INTRODUCTION

Cocoa beans are an important agricultural commodity and also a key raw material in cocoa beverage powder and chocolate production. It is derived from beans of the cacao tree Theobroma cacao L. after the maturity period from the fruits of the parent's trees (Bussy, Ottaviani, et al., 2021). Raw cocoa beans are one of the world's most nutritious fruits, helping to treat a wide range of diseases and clinical situations including oxidative stress, stress and depression, cardiovascular disease, and cancer. However, studies have revealed that polyphenol molecules, which have the capacity to act as antioxidants, are responsible for the aforementioned claims and health advantages of cocoa (Ayoubi et al., 2021; Tee et al., 2021). The phenolic content of cocoa beans accounts for 12-18% of its dry mass, and includes phenolic acids, flavonoids, simple phenols, and benzoquinones, among other compounds. Cocoa beans are a good source of lipids, carbs, proteins, vitamins, and minerals constituents in addition to their phenolic content (Stachyshyn et al., 2021).

Globally, rising demand for foods that give more than just nourishment but also promote health and well-being has led in stratospheric rise in the global market for functional foods and preventative or protective foods with health claims (Faiqoh et al., 2021; Oladeji and Badmus, 2014; Tee et al., 2021). Plants provide the raw materials for such foods. Cocoa powder is one plant product with such claims that it is widely accepted by consumers not only in Nigeria but around the world. However, research have revealed that consumer acceptance of cocoa powder has begun to drop around the world due to its aftertaste and astringency, which has resulted in a slew of disadvantages for the cocoa beverage industry (Al Aribah et al., 2020; Tang et al., 2021). Green and black tea, despite having a higher antioxidant capacity than cocoa powder and other flavonoids-rich foods like red wine, garlic, blueberry, and strawberry, have replaced cocoa powder in the global market due to these characteristics. Cocoa powder is two times more antioxidants source than red wine and also its antioxidants are triple than those in green tea and

blueberries which are good sources of antioxidants (Pérez-Ramírez *et al.*, 2021; Polanowska *et al.*, 2021).

Nutritionally, cocoa powder is a rich source of protein, contain a significant amount of vitamin A, nicotinic acid and riboflavin, minerals such as iron, calcium, copper, zinc, magnesium, sodium, and phosphorous (Stachyshyn et al., 2021). Thus, it may serve the dual purpose of food enriched with vital health-promoting nutrients and bioactive compounds.Currently, ready-to-drink cocoa powder has been developing to become an alternative beverage produced as consumer-friendly to harness the aforementioned health and nutritional benefits (Al Aribah et al., 2020). However, in the course of producing ready-to-use cocoa, some nutritional content of the cocoa significantly decreases.

Studies have shown a decrease in minerals such as magnesium, zinc, calcium, and sodium of cocoa after production (Anagbogu et al., 2021). Calcium and Zinc are elements of public health importance as their deficiency in one'sdiet could lead to a clinical condition (Radomska et al., 2021). Hence, incorporating Zinc and Calcium-rich ingredients into the final product toincrease their content in the final cocoa derived products is recommended (Rajput, 2021; Rojo-Poveda et al., 2021). Vegetables like pumpkin and eggshell were reported to be a good source of some mineral elements of public health importance like zinc and calcium. Incorporating them into the final cocoa powder product would significantly increase the level of zinc and calcium of the formulation. Moreover, vegetables can also be useful to enrich the flavour of cocoa-derived products. Therefore, this study aims at the formulation of ready-to-use cocoa beverage powder enriched with zinc and calcium so that it could an alternative in combating hidden hunger.

MATERIALS AND METHODS

Procurement and processing of material

The food materials such as cocoa powder, pumpkin, and eggshells were purchased from Birnin Kebbi New Market,Kebbi State. The pumpkin and eggshells werewashed to remove the dirt. Then soak in 1% normal saline (NaCl) for 5minutes to getrid of microbes and the pumpkin was peeled and cutinto thin slices. The samples of pumpkin and eggshells were shade-driedto avoid loss of nutrients. Then the eggshells and sliced pumpkin were grounded separately, sieved using a 1mm pore sieve and stored in an airtight container.

Nutrient Analysis

Proximate composition was determined in triplicateusing standard procedures of the Association of OfficialAnalytical Chemists (AOAC, 1999). The moisturecontent was determined by the oven drying method. Crudeprotein was determined by Micro-Kjeldahl Method. Fatwas determined by soxhlet extraction utilizing hexane assolvent. The crude fibre was determined by the neutralizationmethod(Method 962.09). Ash content was determined by the dry ashing method of AOAC (Method 923.03) (AOAC, 1999).

Carbohydrate Estimation

Carbohydrate content was determined by difference (%Carbohydrate) = [100-(%Protein + %Moisture+ %Ash+% Fibre% + %Crude Lipid)] (Mathew *et al.*, 2015).

Determination of some selected minerals

Selected mineral content (Iron, Zinc, Magnesium, Phosphorus, Potassium and Calcium) were determined using atomic absorption spectrophotometer (AAS) (Shimadzu AA-6200 Tokyo, Japan) according to the AOAC method (AOAC, 1996).

Determination of amino acid profile of the formulated blend

The amino acid profile was determined with Technicon Amino Acid Analyzer (TSM-1) using Norleucine as internal standard (Adeyeye and Afolabi, 2004).

Determination of Functional Properties pH

The pH was measured by making a 10% (w/v) powdered suspension of each sample in distilled water. Each sample was mixed thoroughly in a beaker, and the pH was recorded with an electronic pH meter (Model PHN-850, Villeur-Banne, France) (Mathew *et al.*, 2015).

Bulk Density (BD)

Bulk density was determined according to Onwuka (2005) using the formula below. 20g sample was poured into a 100 ml graduated cylinder. The cylinder was tapped 40 to 50 times and the bulk density was calculated as weight per unit volume of sample.

was calculated as weight per unit volume of sample. Bulk density $\frac{g}{cm^3} = \frac{\text{weight of the sample}}{\text{volume of the sample}}$

Water AbsorptionCapacity (WAC)

From the ground sample, 1g was weighed into conical graduated centrifuge tubes of known weights and mixed with 10cm³ of distilled water for one minute with a glass rod. The tubes were centrifuged at 5000 rpm for 30 min. The volume of the supernatant was discarded and each tube together with its content was reweighed as water absorbed per gram of sample. The gain in mass was the water absorption capacity of the flour sample. The volume difference gave the volume of water absorbed per gram sample. Absorption capacity is expressed in grams of water absorbed per gram of sample (Onwuka, 2005).

 $WAC = \frac{\text{density of water} \times \text{volume absorbed}}{\text{weight of the sample}}$

Reconstitution Index (RI)

From the ground sample, five grams of each sample was dissolved in 50 cm³ of boiling water. The mixture was agitated for 90 seconds and was transferred into a 50 cm³ graduated cylinder and the volume of the sediment was recorded after settling for 30 minutes (Onwuka, 2005).

RI
$$(^{Cm^3}/g) = \frac{\text{Volume of the sediment}}{\text{weight of the sediment}}$$

Swelling Index (SI)

The swelling index was determined according to Ukpabi and Ndimele (1990). From each sample, 3g were transferred into clean, dry, and graduated (50cm³) cylinders. The samples were gently levelled and the volumes noted. Distilled water (30 cm³) was added to each sample. The cylinder was swirled and allowed to stand for 60 min while the volume change (swelling) was recorded every 15 min. The ratio of the initial volume to the final volume gave the swelling index.

SI
$$(cm^3/g) = \frac{\text{change in volume of the sample}}{\text{change in weight of the sample}}$$

Determination of Wettability

Triplicate samples were weighed and, in each case, 1.00 g was introduced into a 25 cm³ measuringcylinder with a diameter of 1 cm and a finger wasplaced over the end of the cylinder. The mixture wasinverted and clamped at a height of 10 cm from the surface of a 250 cm³ beaker containing 100 cm³ of distilled water. The finger was removed to allow the test material to be dumped. Wettability was taken as the time required for the sample to become completelywet (AOAC, 2005).

Qualitative Phytochemicals Screening

The phytochemical analysis of the blend samples was conducted according to AOAC (2005).

Test for Alkaloids

The test solution was acidified with acetic acid and a drop of Mayer'sreagent was added. A white precipitate indicated the presence of an alkaloid.

Test for Flavonoids

Few drops of conc. HCl were added to the samples of cocoa beverage, a red colour appeared to indicate the presence of flavonoid.

Test for Glycoside

The extract was filtered and sugar was removed by fermentation with Baker's yeast. The acid was removed by precipitation with Ba(OH)₂. The remaining extract contained the glycosides. The hydrolysis of the solution was done with a concentration of sulfuric acid and after the hydrolysis indicate the presence of sugar through the help of Fehhling's solution.

Test for Steroids

To 2ml of the extract in test-tube, 6ml chloroform and 4ml of concentrated H_2SO_4 were carefully added by the side of the test tube. The upper layer turned red and sulfuric acid layer showed yellow with green fluorescence. This indicated the presence of steroids.

Test for Tannins

To the extract, 3 drops of 0.1% ferric chloride were added. A brownish-green precipitate indicated the presence of tannins.

Test for Phenols

To 2 ml of plant extract was added 2ml of 5% aqueous ferric chloride. The formation of blue colour indicated the presence of phenols.

Test for Saponins

To 2ml of the methanolic extract, 3ml of distilled water were added and shaken vigorously for about 5minutes. The formation 2cm layer of foam which in turn persist for 10minutes indicated the presence of saponins.

Test for Terpenoids

2mls of chloroform were dissolved in 5ml of plant extract and 3ml of concentrated H_2SO4 were carefully added. Formation of reddish colouration at the inter-phase indicated a positive result for the terpenoids.

Sensory Evaluation of the Composite Blends

Sensory evaluation was determined according to USAID (2015). The test formulations were judged by semi-trained panellists. Five points hedonic rating scale was used. Scores were defined as 1 for dislike extremely or bad, 2 for like slightly only or tolerable, 3 for like or good, 4 for like very much, 5 for like extremely or excellent.

Statistical Analysis

Data were reported as means \pm standard error of the mean of triplicate determination. One-way analysis of variance (ANOVA) was used to establish significant differences (*P*<0.05). Values were analyzed statistically using Graph Pad PRISM (Statcon, Witzenhausen, Germany).

RESULTS AND DISCUSSION

Constituent	Composition			
	F1	F2	F 3	
Moisture content	3.0 ± 0.06^{a}	3.5±0.04 ^b	3.5±0.03 ^b	
Ash	8.0±0.01 ^a	6.0±0.02 ^b	$6.5 \pm 0.04^{\circ}$	
Protein	4.55±0.1 ^a	4.38±0.33 ^b	4.46±0.21 ^c	
Fiber	0.5 ± 0.07^{a}	0.5±0.01 ^a	0.5±0.01 ^a	
Lipid	2.5±0.21 ^a	3.0±0.14 ^b	2.0±0.83 ^a	
Carbohydrate	81.45±2.33 ^a	82.62 ± 2.22^{a}	83.04±2.06 ^a	

Table 1: Percentage Proximate composition of the formulated blend

Values are mean \pm standard error of the mean (SEM) of triplicate determinations, Values with the same superscript in the same row differ significantly at (P < 0.05).

Table 2: Selected Micronutrient content of the formulated blends

Micronutrient	Composition		
	F1	F2	F3
Sodium (Na)	167.5±1.03 ^a	200.0±1.03 ^b	200.0±1.28 ^b
Calcium (Ca)	300 ± 2.06^{a}	305 ± 2.33^{b}	335±2.18 ^b
Potassium (K)	1.50±0.03 ^a	0.90 ± 0.01^{b}	$0.75 \pm 0.03^{\circ}$
Magnesium (Mg)	0.60 ± 0.01^{b}	0.60 ± 0.03^{b}	0.60 ± 0.01^{b}
Zinc (Zn)	3.29±0.30 ^a	3.2 ± 0.33^{b}	$3.18 \pm 0.08^{\circ}$
Vitamin A(mg/100g)	451.88 ± 3.82^{a}	414.96±3.02 ^b	402.71±3.18 ^b

Values are mean \pm standard error of the mean (SEM) of triplicate determinations, Values with the same superscript in the same row differ significantly at (P < 0.05).

Table 3: Essential amino acid profile of the composite blends

Parameter (g/100g)	F1	F2	F3
Histidine	1.53±0.03 ^a	1.69 ± 0.21^{b}	$1.79 \pm 0.02^{\circ}$
Isoleucine	5.63±0.83 ^a	5.70 ± 0.13^{b}	6.02±0.23 ^c
Lysine	10.69±1.03 ^a	10.87 ± 0.91^{b}	$11.08 \pm 0.30^{\circ}$
Methionine	0.40 ± 0.01^{a}	0.48 ± 0.05^{a}	0.59±0.01 ^a
Threonine	1.50±0.1 ^a	1.61 ± 0.81^{b}	2.00±0.01 ^c
Tryptophan	0.34±0.01 ^a	0.39±0.03 ^a	0.42 ± 0.01^{a}
Valine	3.83 ± 0.8^{a}	3.98 ± 0.3^{b}	4.21±0.9 ^c

Values are mean \pm standard error of the mean (SEM) of triplicate determinations, Values with the same superscript in the same row differ significantly at (P < 0.05).

Table 4: Functional Properties of the Formulated cocoa product

Parameters	Composition		
	F1	F2	F3
BD	0.76 ± 0.03^{a}	0.74 ± 0.03^{b}	$0.58 \pm 0.03^{\circ}$
SI	0.44 ± 0.01^{a}	0.7 ± 0.08^{b}	$0.32 \pm 0.03^{\circ}$
WAC	0.64 ± 0.02^{a}	0.63 ± 0.03^{b}	$0.58 \pm 0.01^{\circ}$
RI	3.0±0.1 ^a	1.2 ± 0.8^{b}	$1.1 \pm 0.01^{\circ}$
pН	6.3 ± 0.98^{a}	6.76 ± 0.23^{b}	$6.78 \pm 0.28^{\circ}$

Values are mean \pm standard error of the mean (SEM) of triplicate determinations, Values with the same superscript in the same row differ significantly at (P < 0.05).

Table 5: Qualitative Phytochemical Screening of cocoa beverage

Parameters	F1	F2	F3
Tannins	+	+	+
Saponins	+	+	+
Flavonoids	+	+	+
Terpenoids	+	+	+
Cardiac glycosides	+	+	+
Alkaloids	+	+	+
Anthraquinones	+	+	+
Phenols	+	+	+

Key: + = presence; - = absence.

Sample	Colour	Aroma	Taste	Mouth-Feel	Overall acceptability
F1	5	5	5	4	4.75
F2	4	5	4	3	4.0
F3	5	4	3	3	3.75

Table 6: Sensory Attributes of Formulated Blends

DISCUSSION

The nutrient compositions of the formulated blends as shown in Table 1 indicated that moisture content of the formulated samples ranging from 3.0 and 3.5 which is slightly high than the values recorded by (Moreira et al., 2017). However, this study recorded lesser moisture contents than (Oladeji and Badmus, 2014). It's possible that the high moisture level stems from the high moisture content of included substances such as pumpkin. Ash content was the second-highest of the proximate composition after carbohydrate withvalues of 8.0, 6.5, and 6.0 percent respectively. There was a significant difference (P<0.05) for ash contents among the formulated samples. The amount of ash in food materials indicates how much minerals it contains (Bussy, Hewitt, et al., 2021; Yusuf et al., 2020). The ash values found in the study suggest that the integrated materials are good mineral suppliers.

The low fibre content was recorded in both samples. This is not an anomaly and is in line with previous studies (Bursa *et al.*, 2021). Furthermore, the greater material cocoa of the formulated blend has low fibre contents (Stachyshyn *et al.*, 2021). The protein content of the formulations is higher than the values reported by (Tee *et al.*, 2021). However, the study reported lower values of protein than the earlier study (Sperkowska *et al.*, 2021). Similarly, both samples have a high carbohydrate content, which is necessary for energy production. As a result, the formula can also be used as an energy drink.

Table 2 show the values of some selected minerals of public health significance such as Ca, Na, K, P, and Mg. The level of Kand Na reported in this study coincide with the values recorded by Mathew *et al.*, (2015). However, Yusuf *et al.*, (2020), reported higher values of Mg and K than the current study. Except for Na, K, and Mg, there was a consistent decrease in the values of minerals as cocoa powder proportion decreased. Similarly, as the amount of combined nutrients drops, so does the amount of calcium. Cocoa products with a high K content are useful sources of K.

The level of beta-carotene ranges from 451.88, 414.96, and 402.71 between the formulations. There was a significant difference (P<0.05) in beta-carotene levels among the blend formulations. This study recorded high values of beta-carotene than the previous studies by (Kharat and Deshpande, 2017). The inclusion of pumpkin as a formulation ingredient, which is a

strong source of vitamin A, may account for the high vitamin A levels observed.

The test for functional properties that bulk density, water absorption capacity, and reconstitution index was higher in F1; pH and swelling index were high in F3 than every other sample. Both samples recorded a low swelling index. When the swelling index is high, more water is needed to make the ready-to-drink cocoa beverage infusion. (Djikeng *et al.*, 2018). Similarly, lower values of bulk density were recorded in both samples, indicating the less heaviness of the drink infusion. From a nutritional point of view, lower bulk density is preferred for food samples (Ayo-omogie *et al.*, 2019; Yusuf *et al.*, 2020; Yusuf *et al.*, 2021).

The sensorial evaluations through organoleptic property indicated a significant difference (P<0.05) in the overall acceptability, consistency, aroma, and mouthfeel of the formulations. However, F1 has the highest overall acceptance than other formulations which translate to the good keeping quality of the formulation.

CONCLUSION

According to the findings of the current study, fortification of cocoa powder as ready-to-drink beverage with pumpkin seed and eggshell would improve not only the mineral but also the nutrient content. As a result, the formulation can be utilized not only as an energy supplement, but also to battle hidden hunger among young and adult groups in Nigeria and other poor countries throughout the world.

Conflict of Interest: The authors declare no conflict of interest.

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