

Development, quality assessment and antioxidant properties of cocoa based beverage produced from vegetable milk

Abstract

Developing countries are faced with the challenge of diet related diseases like diabetes mellitus, cancer, cardiovascular diseases and inadequate protein in their diet. The common beverage produced from animal milk was reported to have bad cholesterol which may lead to hypertension, hence the need to develop beverage from plant sources. Cocoa based beverage was developed from of Tigernut tubers (*Cyperus esculentus*), Soybean (*Glycine max*) and Alkalized Cocoa Powder sweetened with either natural or synthetic sweeteners (Date syrup, Sucrose and Aspartame). A commercially available sample was used as control. Physicochemical, amino, fatty, antioxidant properties and sensory evaluation were carried out using standard methods. The protein content of the formulated beverage ranged from 3.12g/100g in TCSW to 9.58g/100g in TCS and were significantly higher ($p < 0.05$) than TSBD (2.12g/100g) which was the control sample ;while Energy values (Kcal) ranged from 84.529kcal/100g in SCS to 468.51kcal/100g in TCS. The total phenolic content ranged from 136.11mg/g in SCS to 161.37mg/g in TCDS but was slightly lower than TSBD (165.85mg/g). In conclusion, the study established that the formulated beverage exhibited high amino acid profile and low fatty acid composition; hence the beverage drink may be suitable as functional foods for both old and young.

Keywords: anti-oxidant properties, cocoa – based beverage, sweeteners, tigernut milk, soy milk, sweeteners

Volume 9 Issue 1 - 2021

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Received: January 16, 2021 | **Published:** March 02, 2021

Introduction

Cardiovascular diseases are global metabolic illnesses that affect essential biochemical pathways in the body resulting into complications.¹ They are the leading causes of death worldwide. Developing countries are struggling to manage the deleterious effects of CVD along with growing complications of obesity, diabetes mellitus (Type I and especially Type 2), hypertension and sometimes renal kidney disease.² Elevation in the activity of nutritional antioxidants over the damaging effects of prooxidants has the potential to attenuate these diseases. There are also epidemiological evidences and interventional studies to correlate higher level of antioxidant-rich food uptake with lower incidence of CVD. Africans have the highest prevalence of CVD and have rather unusual risk factors characterized by high level of triglycerides and low density lipoprotein (LDL), low level of High Density Lipoproteins (HDL), glucose intolerance, insulin resistance and abdominal obesity. Hence there is an urgent need to explore various strategies to combat the increasing risk of CVD in the world as a whole. Underutilized tubers and medicinal plants with cardio protective and glucose lowering effects have played major roles in this aspect.³ Functional food is a current discovery in therapeutic food development. Its increasing interest reflects the fact that a specific diet has the ability to mitigate certain diseases through the presence of active phytochemicals absent in the major food but present in tubers. Plant-based functional foods are now getting more attention than ever before because of its myriad benefit to the society or to the whole mankind especially in the line of nutrition. The medicinal values of these plant-based foods lie in the phytochemicals and proteins that displayed physiological functions in the human body.⁴

Cocoa (*Theobroma cacao*) is one of Nigeria's major cash crop and its products have been reported to be rich sources of flavonoids known as procyanidins.⁵ Nigeria is among the top producers of cocoa in the world with increase in production from 367,000 – 421,300 tonnes.⁶ Tigernut (*Cyperus esculentus Lativum*) is an underutilized tuber that produces sweet nutlike taste widely grown in tropical and Mediterranean regions.⁷ Its Milk (having Spanish name “Horchata”) is a refreshing purely natural vegetable drink which is prepared with water, sugar and tigernuts has been reported to be high in dietary fibre and could be effective in the treatment and prevention of many diseases including colon cancer, diabetes and gastro intestinal diseases.⁸ Soybean, by virtue of its high protein content and generous amount of essential amino acids especially lysine, tryptophan and threonine, has been identified as a premier crop in finding solutions to problems of malnutrition in Nigeria.⁹ Soybean, which is cheap and easily available, contains isoflavones and provides useful quantities of phyto estrogens and those populations where there is significant use of these soy products are found to have a reduced risk of cancer and postmenopausal symptoms. Also, plant sterols similar in structure to cholesterol are found in soybeans oil and have been found to reduce low density Cholesterol. Sugar substitutes duplicate the effect of sugar with taste but usually with less energy.

Aspartame is an artificial, non-saccharide sweetener first sold under the brand name nutra sweet; it is approximately 200 times sweeter than sucrose or table sugar. Due to this property, even though aspartame produces 4 kcal/g when metabolized, the quantity needed to produce a sweet taste is so small that its caloric contribution is negligible.¹⁰ Date palm (*Phoenix dactylifera L*) is the major fruit crop in the kingdom of Saudi Arabia. It is one of the sweetest fruit

found in the world with good sources of macro elements like calcium, phosphorus, potassium and magnesium.¹¹

The cost of dairy milk is expensive leading to a dramatic decrease in its product consumption. In view of this, efforts have been made over the years to develop alternative milk-like products from vegetable sources.¹² Also, the common beverage produced from using these dairy milk is costly and they have been reported to be elevated in low density lipoprotein (LDL) that may lead to cardiovascular disease therefore extracts developed from plant sources have great potential as nutritional substitutes for cow milk. Previous work had been done on the production and evaluation of Soy- choco beverage with emphasis on the economic evaluation,¹³ fortified vegetable beverage was also produced by,¹⁴ and peanut soymilk was produced by.¹⁵ With the massive health benefits, the astringent taste of cocoa beverage without sweeteners has limited their usage as beverages drink. To promote the value-addition, the present study used tigernut milk, soymilk, alkalized cocoa powder and natural /synthetic sweeteners to formulate a beverage drink. The formulated beverages were analyzed for their physicochemical, antioxidant, amino/fatty acid composition and overall acceptability and its potential to act as nutraceutical beverage.

Table 1 Sample formulations / blend ratio of the study

Sample	control (mL)	Tigernut milk (mL)	Soymilk (mL)	CMC (mL)	Cocoa powder (mL)	Aspartame	Date syrup	Sucrose
TCSW		494	-	0.5	5	0.5	-	-
TCDS		489.5	-	0.5	5	-	5	-
TCS		489.5	-	0.5	5	-	-	5
TCO		494.5		0.5	5			
SCSW		-	494	0.5	5	0.5	-	-
SCDS		-	489.5	0.5	5	-	5	-
SCS			489.5	0.5	5	-	-	5
SCO			494.5	0.5	5	-	-	-
TSBD	500							

Determination of proximate composition of the cocoa - based beverage

Proximate composition was determined as described by.¹⁹ Moisture content was determined by oven drying methods, Fat content was done using soxhlet extractor, Total ash content using muffle furnace, protein content was determined using standard kjeldahl procedure and carbohydrate was determined by difference. The pH content was determined using the Eco-testr pH1 (Japan). Titratable acidity, free fatty acid, viscosity and sugar content were determined according to standard methods.

Antioxidant capacity determination of the cocoa-based beverage

Determination of total phenolic content: The total phenolic content of the sample was determined as described by.²⁰ 0.2ml of the sample

Materials and methods

Food materials sources

The Tigernut tubers (*Cyperus esculentus*) variety and soybean (*Glycine max*) were purchased from the Main Market in Akure. The food materials were authenticated at the Department of Crop, Soil and Pest Management, Federal University of Technology Akure, Nigeria. All other reagents and kits used were of analytical grade.

Processing of food material

Tigernut milk was extracted as described by;¹⁶ Soymilk was also extracted as described by.¹⁷ Alkalized cocoa powder (5ml) and 0.5ml of carboxyl methyl cellulose (CMC) were mixed with Tigernut milk and soymilk with the addition of 0.5ml of Aspartame, 5ml of Date syrup and 5ml of sucrose respectively. The mixture was blended in a Q- link blender and then homogenized and pasteurized at 72°C for 5 mins¹⁸ with slight modifications. It was then rapidly cooled and packaged in Polyethylene terephthalate bottles for further analysis. A commercially available sample was used as control. The sample formulation was illustrated in Table 1.

was mixed with 2.5ml of 10% folin ciocalteaus reagent and 2ml of 7.5% sodium carbonate. The reaction mixture was subsequently incubated at 45°C for 40 mins, and the absorbance was measured at 765nm in the JENWAY UV- Visible sourced from Germany. A standard curve was prepared and used to translate the measured absorbance valued to gallic acid equivalents (GAE).

Ferric reducing antioxidant property (FRAP) determination: The reducing property of the extract was determined by the method of.²¹ A 2.5ml of the extract was mixed with 2.5ml of 200 mM of Sodium phosphate buffer pH 6.6 and 2.5ml of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 min, thereafter 2.5 ml of 10% trichloroacetic acid was added and the mixture was centrifuged at 2000rpm for 10min, 1ml of the supernatant was mixed with 1ml of distilled water and 0.1% of Ferric chloride and the absorbance was measured at 700nm while the ferric reducing power was then calculated and the values expressed in mg/g.

$$FRAP = \frac{(Absorbance\ sample * Concentration\ of\ Standard * 1000)}{(Absorbance\ of\ Standards * Concentration\ of\ sample)}$$

Determination of free radical scavenging ability (DPPH): The free radical scavenging ability of the extract against DPPH (1, 1-diphenyl-2-picrylhydrazyl) using the method of.²² The extract (1ml) was mixed with 1ml of 0.4mM methanolic solution containing DPPH radicals, the mixture was left in the dark for 30mins and the

absorbance was measured at 516nm. The percentage antioxidant activity was calculated as follows: The scavenging activity of samples corresponded to the intensity of quenching DPPH. The antioxidant activity were expressed as percentage inhibition and calculated as follows:

$$\%inhibition = \left[\frac{(Absorbance\ of\ control - Absorbance\ of\ sample)}{Absorbance\ of\ control} \right] \times 100$$

ABTS scavenging ability determination: The ABTS (2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid) (ABTS) scavenging ability of the beverage was determined according to the method described by.²³ The ABTS was generated by reacting an (7mM).ABTS aqueous solution with $K_2S_2O_8$ (2.45 mM, final concentration) in the dark for 16

hours and adjusting the absorbance at 734nm to 0.700 with ethanol. 0.2ml of the appropriate dilution of the extract was then added to 2.0ml of ABTS solution and the absorbance was read at 734nm after 15mins. The Trolox equivalent antioxidant capacity (TEAC) was subsequently calculated using trolox as the standard.

$$\%Scavenging\ ability\ of\ sample = \left[\frac{Absref - Abssample}{Absref} \right] \times 100$$

Amino acid profile and fatty acid composition of cocoa based beverage

Large proportions of the formulated beverage and control samples were poured into empty petri dish and oven dried overnight at a temperature of 60°C. The samples were allowed to cool, scraped and stored for further analysis. The amino acid profiles of the experimental samples were determined according to the method described by.¹⁹ The experimental samples were digested using 6N HCl for 24 h. Amino acids were determined using the Beckman Amino Acid Analyzer (model 6300; Beckman Coulter Inc., Fullerton, Calif., USA) employing sodium citrate buffers as step gradients with the cation exchange post-column ninhydrin derivatization method. The data were calculated as grams of amino acid per 100 g crude protein of the sample.

Fatty acid composition was determined as described by.¹⁹ each beverage sample was weighed into the extraction thimble. 200 mL of petroleum ether 40-60°C boiling range was measured and then added to the dried 250 mL capacity flask. The covered porous thimble with the sample was placed into the condenser of the soxhlet extractor arrangement that has been assembled. The lipid was extracted for 5 hours. The extracted flask with the oil was oven dried at 105°C for 1 hour. The flask containing the dried oil was cooled in the desiccators and the weight of the cooled flask with the dried oil was measured. Methyl esters and analyser were prepared and the fatty acid parameters calculated using standard methods.

Consumers preference of cocoa -based beverage from vegetable milk

The cocoa based beverage prepared was evaluated for their sensory qualities and general acceptability. A scoring test technique was used to determine which of the products was most preferred. A 15- member semi trained taste panel carried out the rating of soychoco and tigerchoco samples. Each of the panellists rated the samples on the basis of colour, taste, aroma, mouth feel and overall acceptability using a nine-point Hedonic scale (9 =like extremely; 5=neither like or dislike;1=dislike extremely). The scores from the rating were subsequently analyzed.²⁴

Statistical analysis

Except otherwise stated, all analyses were performed in triplicate and the data generated were analyzed using one way analysis of variance(ANOVA) using SPSS version 20.0. The means were separated using the Duncan New Multiple Range Test and significance was accepted at $p \leq 0.05$.

Result and discussion

Proximate composition and physical properties of cocoa -based beverage from vegetable milk using different sweetening agents

The proximate composition of Tiger-soy beverage drink and control samples are presented in Table 2. The moisture contents ranged from 80.88g/100g in sample TCS to 95.08g/100g in sample SCS. The moisture content of the formulated beverage was higher than the control sample. Comparatively, the moisture content of the produced beverage was within the acceptable limit recommended for beverage samples.²⁵ Previous researches had stated that high moisture content is a characteristic of liquid based beverage.²⁶ In comparism with other findings, it was observed that the moisture content of the beverage drink in this present study were similar to the values reported by²⁷ for the production and evaluation of Tigernut milk flavoured with moringa leaf extract and In contrary to other study, the values were higher than values (61.1–64.21g/100g) reported by²⁸ for chemical composition and sensory properties of soytigernut cheese. The difference between the moisture content observed in this study and other reports could be attributed to the blending of food materials, processing and the addition of other sweetening agents.

The protein content of the beverage sample ranged from 2.05g/100g in TSBD – 6.42g/100g in SCDS. Majority of the formulated beverages were significantly ($p < 0.05$) higher than the control sample. The high protein content may be suitable for growth, development and replacement of worn out tissues in infant and adults, thus the developed beverage may be more satisfactory in providing balanced nutrients as compared to the control sample.

Table 2 Proximate composition (g/100g) and Energy value (Kcal/100g) of cocoa - based beverage from vegetable milk using different sweetening agents

Sample	Moisture	Crude protein	Crude fat	Total ash	Crude fibre	CHO	Energy value	Dry matter
TCSW	92.25±0.08 ^a	3.12±0.01 ^e	0.52±0.01 ^c	0.84±0.16 ^b	ND	3.27±0.33 ^f	126.317±0.84 ^e	7.75±0.06 ^c
TCDS	88.30±0.07 ^b	5.45±0.02 ^d	0.57±0.01 ^c	0.86±0.11 ^b	ND	4.82±0.16 ^e	192.998±2.16 ^c	11.70±1.33 ^b
TCS	80.88±0.00 ^c	9.58±0.10 ^a	0.60±0.02 ^c	0.85±0.10 ^b	ND	17.12±0.54 ^a	468.51±2.07 ^a	19.12±2.21 ^a
TCO	90.22±0.04 ^a	0.39±1.10 ^g	0.62±0.00 ^c	0.21±0.04 ^d	ND	8.45±0.01 ^c	172.17±1.01 ^d	9.78±1.33 ^b
SCSW	93.20±0.13 ^a	4.16±0.01 ^c	0.58±0.03 ^c	0.52±0.06 ^b	ND	1.54±0.29 ^e	117.056±1.57 ^f	6.80 ± 0.22 ^d
SCDS	92.32±0.21 ^a	5.42±0.01 ^b	0.51±0.04 ^c	0.86±0.07 ^b	ND	0.98±0.01 ^g	126.107±1.60 ^d	7.68 ±0.33 ^c
SCS	95.08±0.02 ^a	3.22±0.02 ^e	0.55±0.03 ^c	0.58±0.12 ^c	ND	0.60 ±0.11 ^h	84.529±1.55 ^g	4.92 ±0.11 ^e
SCO	89.34±0.22 ^b	2.11±0.14 ^f	1.18±0.33 ^b	0.46±0.01 ^c	ND	6.91±0.34 ^d	195.12±1.11 ^c	10.66±0.28 ^b
TSBD	81.99±0.14 ^c	2.05±0.03 ^g	1.85±0.11 ^a	1.40±0.10 ^a	ND	12.71±1.12 ^b	316.237±2.77 ^b	18.01±1.57 ^a

The total ash content ranged between 0.58g/100g in sample SCS - 1.40g/100g in TSBD. The values are within the range of the total ash content of most liquids based beverages as reported by.²⁹ The crude fat content of the beverage is similar to the values reported by³⁰ for beverages. The carbohydrate content of the cocoa- based beverages ranged between 0.60g/100g - 12.71g/100g with the control sample having the highest percentage of carbohydrate content. Carbohydrates are inexpensive source of food energy. The developed beverage may not contribute much sugar to the blood of the consumer, hence may be preferred by diabetic patients.

The result of the physical properties of the formulated beverage and the control sample is shown in Table 3. From the table, the pH ranged from 5.06-6.84. The values gotten here is similar to pH values

(6.12–6.71) for Tigernut milk reported by¹⁶ and.³¹ The beverage sample generally have high pH values and this could be attributed to the raw materials used for this study. The TTA of the formulated beverage ranged from 0.153–0.324%. Tigerchoco sweetened with date syrup showed significantly higher titratable acidity value with an inverse correlation with the pH value. Also, SCDS has the lowest titratable acidity value with an inverse relationship with the pH value which means that there is an inverse relationship with the pH and TTA of this study which is similar to the report of.³² The brix ranged between 1.8-10° brix with the control sample having the highest value. The high value in TSBD may be attributed to the sugar content of the beverage. Samples produced from Tigernut milk have higher brix content as compared to those produced from soymilk and this may be attributed to the natural sugar present in tigernut tubers.

Table 3 Physical properties of cocoa - based beverage from vegetable milk using different sweetening agents

Sample	pH	Density	Titratable acidity (%)	Total dissolve solid (mg/L)	Brix
TCSW	5.57±0.21 ^b	1.0042±0.01 ^b	0.153±0.02 ^c	7.00±2.42 ^d	6.33±1.44 ^d
TCDS	5.28±0.81 ^b	1.0213±0.01 ^b	0.324±0.05 ^a	11.5±1.11 ^c	10.00±0.72 ^c
TCS	5.47±0.76 ^b	1.0237±0.03 ^b	0.315±0.02 ^a	15.00±2.11 ^b	12.22±1.44 ^b
TCO	6.50±0.26 ^a	0.9833±0.10 ^c	0.028±0.04 ^e	11.22±0.26 ^b	4.66±0.33 ^e
SCSW	6.73±0.34 ^a	1.0071±0.01 ^b	0.126±0.03 ^c	3.50±0.21 ^g	3.22±1.38 ^f
SCDS	6.84±0.33 ^a	1.0095±0.02 ^a	0.135±0.11 ^c	4.50±1.37 ^f	4.12±0.13 ^e
SCS	6.42±0.03 ^a	1.0118±0.11 ^b	0.155±0.22 ^c	17.0±1.69 ^a	5.00±0.21 ^d
SCO	6.93±0.39 ^a	0.9763±0.02 ^c	0.090±0.19 ^d	10.23±0.24	2.17±0.01 ^g
TSBD	5.06±0.29 ^c	1.0933±0.04 ^a	0.233±0.33 ^b	5.50±3.33 ^e	14.00±0.01 ^a

Antioxidant properties of cocoa -based beverage from vegetable milk using different sweetening agents

Figures 1–4 illustrates the Antioxidant properties of cocoa beverage. Plant food contains classes and types of antioxidant and knowledge of their antioxidant potential. The consumption of beverages has been inversely associated with morbidity and mortality from degenerative diseases. 1,1 Di-phenyl-2-picrylhydrazyl (DPPH) radical is a commonly used procedure which has been confirmed against several other assays for antioxidant activities. DPPH radical are capable of accepting electron or hydrogen to become diamagnetic

molecule. The beverage formulations neutralized the DPPH radical from deep violet colour to colourless or pale yellow with this property allowing visual monitoring of the reaction recorded as % DPPH inhibition.³³ The actual values obtained for the DPPH scavenging ability of the beverages ranged from 23.11% to 40.02% with the control sample having the highest percentage of the scavenging ability and sample SCSW has the lowest value. The values reported in this is in agreement with the scavenging ability of beverages as described by³⁴ for total antioxidant potential of fruit juices, beverages and hot drinks consumed in Egypt and it is also similar to that reported by.¹⁶ The addition of sweeteners in this study might have contributed to the

enhanced DPPH inhibition. Sample SCDS and SCS have better free radical scavenging potential in all the beverage samples. Researchers have reported that the antioxidant effect relates to the development of reductones and reductones was also reported to be terminators of free radical chain reaction.³⁵ The antioxidant activity of beverage samples can be related to their reducing ability. The ferric reducing property (FRAP) range between 3.22 and 18.34mg/g, the antioxidant activity measures the electron donating capacity.³⁶ The presence of reducers causes the conversion of Fe^{3+} /ferricyanide complex to the ferrous

form which serves as a significant indicator of its antioxidant capacity. The values for SCSW, SCDS, and SCS is similar to the values reported by.³⁷ The ability to chelate transition metals is considered to be due to an antioxidant mechanism.³⁸ The result revealed that all the samples chelate Fe (II) but sample TCDS has improved FRAP (18.34mg/g) as it is significantly higher than the other formulated beverages and it also shows better affinity towards the ferric cyanide content to have enhanced ion hydrogen transfer from ferric to ferrous.

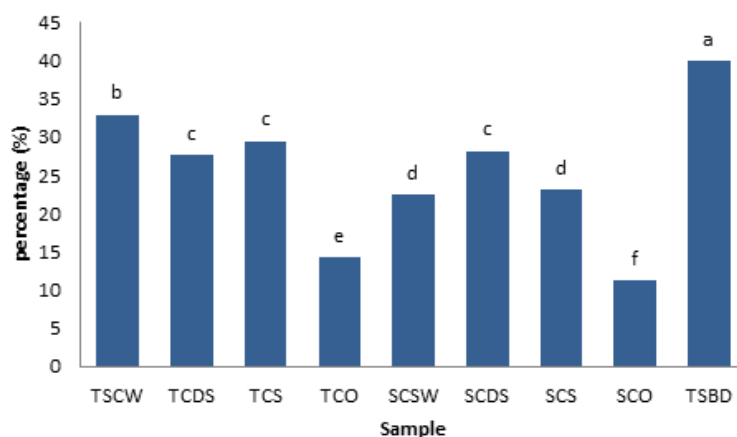


Figure 1 DPPH Content of cocoa - based beverage from vegetable milk using different sweetening agents Values are mean±SD (n=3). Bars with different alphabets are significantly different (p<0.05).

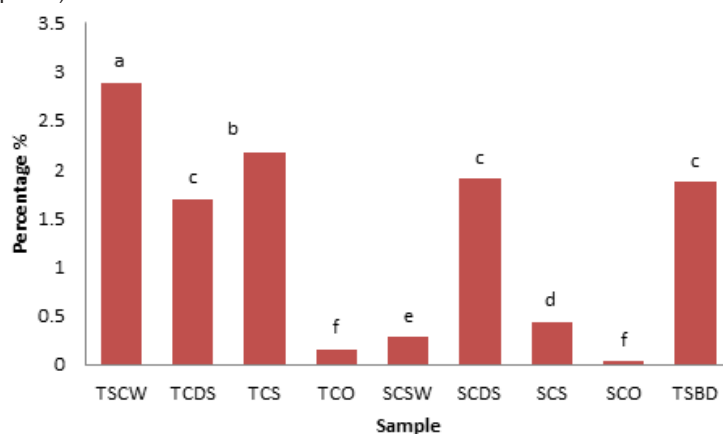


Figure 2 ABTS Content of cocoa - based beverage from vegetable milk using different sweetening agents Values are mean±SD (n=3). Bars with different alphabets are significantly different (p<0.05).

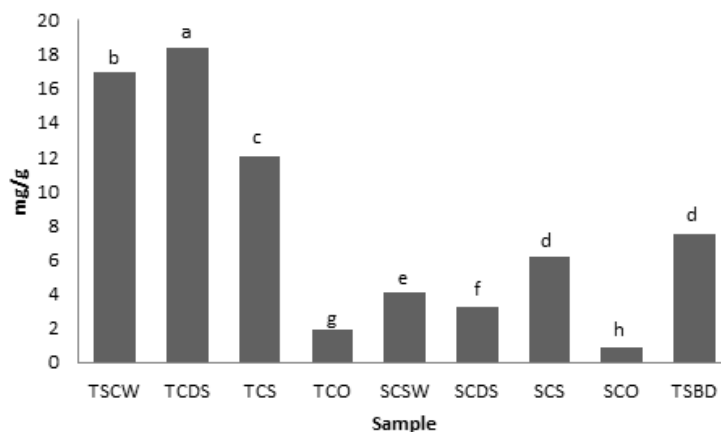


Figure 3 Ferric Reducing Antioxidant Power of cocoa - based beverage from vegetable milk using different sweetening agents Values are mean±SD (n=3). Bars with different alphabets are significantly different (p<0.05).

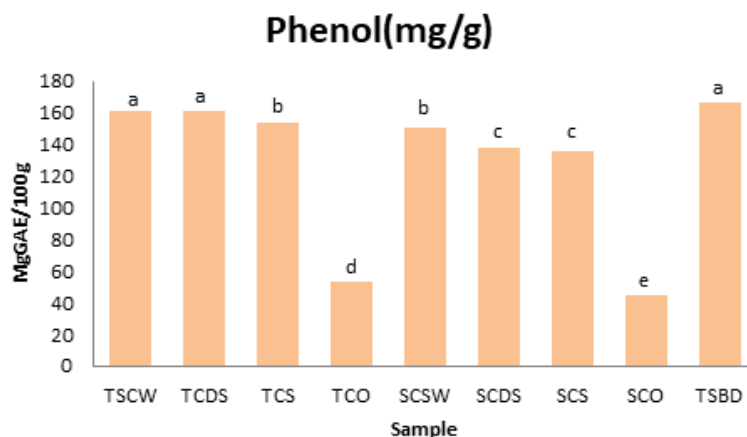


Figure 4 Total Phenolic content of cocoa - based beverage from vegetable milk using different sweetening agents Values are mean±SD (n=3). Bars with different alphabets are significantly different (p<0.05).

Cocoa beverage generally contain polyphenols that have diverse beneficial biochemical and antioxidant effects.³⁹ The beverages prepared from tigernut milk had the highest phenolic content while those prepared from soymilk had the lowest phenolic content. The values reported for Tiger choco beverages (160.82, 161.37, 153.22mgGAE/g) is similar to the values reported by⁴⁰ for the antioxidant activities of beverage blends from Tigernut, hibiscus sabdaniffa and moringa oleifera extracts. Total phenolic content has direct relationship with antioxidant property of the beverage particularly they can modulate rennin–angiotensin converting enzymes and hence reducing hypertension. Recent researchers have identified certain food polyphenols as health promoting agents because of their ability to act as antioxidants and free radical scavengers. As anti-cancer agents, polyphenols act by removing carcinogenic agents through chelation, then modulating cancer cells signally and cell cycle progression, inducing various enzymes activities and promoting apoptosis.⁴¹ In the last decades, several studies have demonstrated significant decrease of plasma antioxidants in the causes of diabetes and its associated complications such as endothelial dysfunction and atherosclerosis.⁴² For instance, evidence has shown that low levels of

plasma antioxidants are more pronounced in elderly diabetic subjects. Thus, the rationale for the therapeutic use of antioxidants in the treatment and prevention of cardiovascular disease and other health related complications is strong.

Amino acid profile and fatty acid composition of cocoa based beverage produced from vegetable milk using different sweetening agents

Amino acid profile and fatty acid composition of the formulated beverage and control sample are presented in Tables 4&5. The total essential amino acid ranged between 24.05 in TSBD to 36.15 in SCDS while the total non-essential amino acid ranged between 32.18 in TSBD to 58.31 in SCDS. It was observed that sample SCDS has the highest proportion of both essential and non-essential amino acid composition; this could be attributed to the high protein content in soybean and Date palm respectively. The most abundant amino acid is the glutamic acid and the limiting amino acid is Cysteine. Glutamic acid performs a number of functions in the body such as strengthening of the immune system and regulations of acid –base balance and protection against cell toxins.⁴³ Its abundance in this study is beneficial.

Table 4 Amino acid (mg/100g) composition of cocoa - based beverage from vegetable milk using different sweetening agents

Sample	TCSW	TCDS	TCS	TCO	SCSW	SCDS	SCS	SCO	TSBD
Non-essential									
Glycine	4.26 ^d	4.92 ^b	4.35 ^c	3.65 ^e	6.22 ^a	6.29 ^a	6.24 ^a	5.05 ^b	1.38 ^f
Alanine	3.09 ^d	3.33 ^c	3.26 ^c	3.11 ^c	4.06 ^b	4.33 ^a	4.10 ^b	3.90 ^b	2.99 ^e
Serine	2.97 ^e	3.39 ^c	3.06 ^d	2.63 ^f	3.82 ^b	4.92 ^a	3.96 ^b	4.80 ^a	1.88 ^g
Proline	3.00 ^d	3.50 ^c	3.41 ^c	2.23 ^f	3.47 ^c	4.11 ^a	3.26 ^d	3.70 ^b	1.41 ^e
Aspartic	10.95 ^d	11.05 ^c	10.15 ^d	10.21 ^d	14.79 ^a	14.83 ^a	13.97 ^b	10.95 ^d	10.31 ^d
Glutamic	17.22 ^d	17.62 ^c	17.05 ^e	11.33 ^f	19.03 ^b	19.22 ^a	19.05 ^b	16.52 ^e	11.66 ^f
Tyrosine	2.01 ^d	2.16 ^c	2.13 ^c	1.83 ^e	2.41 ^b	2.73 ^a	2.43 ^b	2.67 ^a	1.66 ^e
Cysteine	0.69 ^c	0.79 ^f	0.99 ^c	0.44 ^c	1.77 ^a	1.88 ^a	1.69 ^a	1.21 ^b	0.89 ^c
ΣNEAAS	44.19 ^d	47.75 ^c	44.40 ^d	35.43 ^f	55.57 ^b	58.31 ^a	54.70 ^b	48.82 ^c	32.18 ^e

Table Continued...

Sample	TCSW	TCDS	TCS	TCO	SCSW	SCDS	SCS	SCO	TSBD
Essential									
Histidine	1.29 _e	1.99 ^c	1.43 ^d	1.04 ^f	2.03 ^c	2.55 ^a	2.13 ^b	2.21 ^b	1.06 ^f
Phenylalanine	2.29 _e	2.66 ^d	2.39 ^e	2.00 ^f	4.37 ^c	4.77 ^b	4.22 ^c	4.44 ^c	6.00 ^a
Methionine	1.26 _c	1.37 ^b	1.21 ^d	1.15 ^d	1.36 ^b	1.48 ^a	1.40 ^a	1.48 ^a	0.37 ^e
Valine	3.72 _b	3.94 ^b	3.44 ^b	3.21 ^b	4.23 ^a	4.54 ^a	4.43 ^a	4.35 ^a	3.22 ^b
Tryptopan	0.92 _b	1.02 ^a	0.89 ^b	0.69 ^c	1.11 ^a	1.64 ^a	1.33 ^a	1.55 ^a	0.93 ^b
Threonine	2.59 _b	2.66 ^b	2.63 ^b	2.18 ^b	3.00 ^a	3.19 ^a	3.06 ^a	3.00 ^a	1.88 ^c
Isoleucine	3.02 ^b	3.22 ^b	3.14 ^b	2.89 ^c	4.00 ^a	4.77 ^a	4.22 ^a	4.11 ^a	4.19 ^a
Leucine	6.11 ^b	6.29 ^b	6.11 ^b	2.60 ^d	7.00 ^a	7.89 ^a	7.22 ^a	7.55 ^a	4.22 ^c
Lysine	4.88 ^b	5.05 ^a	4.95 ^b	4.01 ^b	5.05 ^a	5.22 ^a	5.11 ^a	5.22 ^a	2.18 ^c
ΣEAAAS	26.08 ^d	28.20 ^c	26.19 ^d	19.77 ^e	32.15 ^b	36.05 ^a	33.12 ^b	33.91 ^b	24.05 ^d

Table 5 Fatty acid composition of cocoa based beverage from vegetable milk using different sweetening agents

Sample	TCSW	TCDS	TCS	TCO	SCSW	SCDS	SCS	SCO	TSBD
Saturated									
Caprylic acid	0.01 ^d	0.09 ^c	0.02 ^d	0.14 ^e	0.20 ^b	0.22 ^b	0.37 ^a	0.30 ^a	0.00 ^f
Capric acid	0.00 ^e	0.03 ^d	0.09 ^c	0.18 ^b	0.20 ^b	0.21 ^a	0.21 ^a	0.21 ^a	0.00 ^e
Lauric acid	0.02 ^d	0.04 ^d	0.10 ^c	0.25 ^a	0.17 ^b	0.16 ^b	0.11 ^c	0.10 ^c	0.00 ^e
Myristic acid	0.03 ^d	0.07 ^c	0.13 ^c	0.11 ^c	0.27 ^a	0.21 ^b	0.22 ^b	0.09 ^c	0.00 ^e
Palmitic acid	13.62 ^d	17.31 ^c	17.61 ^c	14.23 ^d	21.33 ^b	23.50 ^a	23.00 ^a	18.11 ^c	20.31 ^b
Margaric acid	0.02 ^c	0.04 ^c	0.06 ^c	0.02 ^c	0.16 ^b	0.13 ^b	0.20 ^a	0.07 ^c	0.08 ^c
Stearic acid	6.22 ^b	6.33 ^b	3.60 ^d	1.22 ^e	4.21 ^c	4.11 ^c	4.42 ^c	3.63 ^d	7.11 ^a
Behenic acid	0.01 ^c	0.22 ^a	0.09 ^b	0.02 ^c	0.11 ^b	0.10 ^b	0.21 ^a	0.11 ^b	0.21 ^a
Lignoceric acid	0.22 ^c	3.22 ^a	0.89 ^c	0.10 ^c	1.12 ^b	1.10 ^b	1.16 ^b	1.05 ^b	0.16 ^c
Arachidic acid	0.05 ^b	0.10 ^b	0.62 ^a	0.05 ^b	0.08 ^b	0.07 ^b	0.10 ^b	0.09 ^b	0.89 ^a
ΣSFA	20.20 ^d	27.45 ^b	23.21 ^c	16.32 ^e	27.85 ^b	29.81 ^a	30.00 ^a	23.76 ^c	28.76 ^b
Monounsaturated									
Palmitoleic acid	0.30 ^c	1.23 ^b	1.06 ^b	1.30 ^b	1.33 ^b	1.46 ^b	1.55 ^b	1.60 ^b	5.11 ^a
Oleic acid	54.26 ^b	53.31 ^b	54.40 ^b	64.10 ^a	52.01 ^b	53.10 ^b	52.22 ^a	54.90 ^a	32.97 ^c
Erucic acid	0.39 ^b	0.26 ^b	0.31 ^b	0.28 ^b	1.69 ^a	1.75 ^a	1.88 ^a	0.48 ^b	0.06 ^c
ΣMUFA	54.95 ^b	54.80 ^b	55.77 ^b	65.68 ^a	55.03 ^b	56.31 ^b	55.65 ^b	56.98 ^b	38.14 ^c
polyunsaturated									
Linoleic acid	23.96 ^b	17.05 ^d	20.16 ^c	10.37 ^f	14.49 ^e	11.37 ^f	11.84 ^f	12.22 ^f	26.95 ^a
Linolenic acid	0.71 ^c	0.56 ^c	0.68 ^c	1.00 ^d	1.22 ^b	1.22 ^b	1.22 ^b	1.13 ^d	6.12 ^a
Arachidonic acid	0.18 ^b	0.14 ^c	0.18 ^b	1.33 ^a	1.31 ^a	1.29 ^a	1.29 ^a	1.44 ^a	0.03 ^d
ΣPUFA	24.85 ^b	17.75 ^d	21.02 ^c	12.70 ^e	17.02 ^d	13.88 ^e	14.35 ^e	14.79 ^e	33.10 ^a

Table 6 Consumer preference for cocoa based beverage from vegetable milk using different sweetening agents

Sample	Appearance	Taste	Mouth feel	Aroma	Overall acceptability
TCSW	8.11±1.13 ^c	7.03±1.33 ^d	6.33±0.10 ^d	7.18±0.02 ^b	7.05±1.56 ^d
TCDS	8.48±1.21 ^a	7.38±0.03 ^b	6.90±1.21 ^b	7.30±0.13 ^a	7.90±1.79 ^b
TCS	8.31±1.05 ^b	7.18±0.09 ^c	6.72±1.51 ^c	7.18±0.02 ^b	7.28±1.64 ^h
TCO	8.10±1.00 ^c	2.07±0.01 ^h	4.22±0.01 ^h	7.16±0.00 ^b	4.44±1.32 ^b
SCSW	6.22±0.01 ^f	4.22±0.10 ^g	5.00±0.01 ^g	6.40±0.26 ^c	6.02±0.31 ^g
SCDS	6.88±1.06 ^d	5.12±0.06 ^e	5.82±1.31 ^e	6.50±0.11 ^d	6.97±0.08 ^e
SCS	6.48±0.08 ^e	4.88±0.20 ^f	5.21±1.06 ^f	6.40±0.22 ^c	6.28±0.01 ^f
SCO	6.18±0.02 ^f	1.88±0.22 ^g	4.23±0.23 ^g	6.22±0.50 ^d	3.55±0.00 ^g
TSBD	8.52±1.20 ^a	8.02±1.21 ^a	7.08±1.63 ^a	7.33±0.16 ^a	8.86±1.23 ^a

The control sample had the least amino acid composition in this study; this observation indicates that the formulated beverage is suitable to provide essential amino acid for physiological needs in both children and adults. The free fatty acid composition of the formulated beverage and control sample is illustrated in Table 5.

The result showed that the total saturated fatty acid of the beverage ranged from 20.20mg/100g in TCSW to 30.00mg/100g in SCS. For monounsaturated fatty acid, the value ranged from 38.14mg/100g in TSBD to 56.31mg/100g in SCDS also, for polyunsaturated fatty acid, the value ranged from 13.88mg/100g in SCDS to 33.10mg/100g in TSBD. The mono and poly unsaturated fatty acid of the formulated beverage samples was much higher than the control samples. This observation could be attributed to the inclusion of Tigernut and soybean in the production of the beverage. This observation agreed with⁴⁴ recommendations that advocated for the addition of vegetable oil in the formulation of food in order to increase the energy density and to facilitate fat-soluble vitamin absorption. The MUFA and PUFA proportions in this study were similar to those previously reported by.⁴⁵ It was also discovered that among the fatty acid compositions of the beverages oleic acid, Palmitic acid and Linoleic acid were the most abundant.

Evidence has shown that linoleic acid and α -linoleic acid are precursors to decosahexanoic acid and arachidonic acid. The decosahexanoic acid and arachidonic acid are critical for brain growth and development, particularly of the neutral development in the first 6 months of life and that low intake of these fatty acids may lead to impaired cognitive, visual and motor skill development^{46, 47} so therefore, the formulated beverage may essential for both old and young because of its fatty composition.

Consumer preference of cocoa based beverage produced from vegetable milk using different sweetening agents

The consumer acceptability of cocoa based beverage produced from Tigernut milk and soymilk with different sweetening agents is illustrated in Table 6. The appearance, Taste, mouth feel, Aroma and overall acceptability ranged as follows; 6.22 in SCSW – 8.52 in TSBD, 4.22 in SCSW -8.02 in TSBD, 5.00 in SCSW -7.08 in TSBD, 6.40 in SCSW-7.30 in TSBD and 6.02 in in SCSW-8.86 in TSBD respectively. It was observed that all the samples scored high in appearance and Aroma. However, samples TCO and SCO had the least score in taste and overall acceptability probably due to absence of sweeteners. Based on overall acceptability, the control sample TSBD scored highest (8.86) followed by TCDS (7.90). The disparity in these two samples could be due to the difference in food composition,

processing and familiarity in the taste of the control samples by the panelists.

Conclusion

Process technology for the production of cocoa- based beverage using vegetable milk and different sweetening agents was established. Base- line information on the Physicochemical, antioxidant, amino acid, fatty acid compositions and sensory properties were provided. The addition of sweeteners enhanced the antioxidant potential of the beverages and reduced the astringent taste of the cocoa powder if consumed alone. All the formulated beverages were found to exhibit optimum characteristics. This study was an eye opener in the utilization of cocoa powder, tigernut/soymilk and sweeteners in the production of beverages. Further studies on the shelf life of the formulated beverage are required so as to make certain how long it will hold its quality.

Acknowledgments

None.

Conflicts of interest

Author declares there is no conflicts of interest.

Funding

None.

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