Nutritional content, phenolic composition and antioxidant activity of yoghurt enriched with baobab pulp (*Adansonia digitata*)

Batsirai Chipurura^{1*}, Michael Bhebhe², Maud Muchuweti²

Nutrafoods (2016) 15:135-143 DOI 10.17470/NF-016-1035-2

> Correspondence to: Batsirai Chipurura bchipurura@science.uz.ac.zw



Abstract

The objective of the study was to determine the nutritional content, phenolic content and antioxidant activity of yoghurt enriched with baobab pulp. Baobab yoghurt had significantly (p < 0.05) higher ash, crude fibre, vitamin C, iron, total phenolic content, total flavonoid content and DPPH scavenging activity than the control and commercial yoghurts. The ash, crude fibre, vitamin C and iron content was 3.7±0.01 g/100 g, 0.2±0.02 g/100 g, 18.25±0.02 mg/100 g and 0.33±0.01 mg/100 g, respectively. The total phenolic content, total flavonoid content and DPPH scavenging activity of the baobab yoghurt was 22.6±2.08 mg GAE/100 g, 4.23±0.25 mg CE/100 g and 19.03%, respectively. The following phenolic acids were detected in the baobab yoghurt: syringic, caffeic, *p*-dihydroxybenzoic, gallic, protocatechuic and chlorogenic acids. The present study showed that baobab yoghurt is a valuable source of nutrients and phenolic compounds compared to some commercial yoghurts.

Introduction

Adansonia digitata L. (baobab) is a deciduous tree that belongs to the Malvaceae family [1] and is native to arid Central Africa, semi-arid and subhumid sub-Saharan Africa and western Madagascar [2].

Rural communities in some African countries, such as Sudan, use every part of the baobab tree, namely, the leaves, seeds, pulp and bark [3] for food or medicine. The pulp from the fruit is eaten raw or made into various food products, such as seasoning, appetizers, frozen products, juices, jams and cereal-based gruels [4]. In addition to its food uses, the pulp is also traditionally used to treat fever, dysentery, diarrhoea, haemoptysis and microbial diseases [5]. The medicinal uses of baobab pulp are due to phytochemicals, such as vitamins, minerals, sterols, phenolics, triterpenes and saponins, in the pulp. Several studies have shown that these compounds have antioxidant, immunostimulant, antipyretic, analgesic, anti-inflammatory and hepatoprotective properties [6]. Vitamin C and other phytochemicals found in the pulp can reduce reactive oxygen species (ROS) such as superoxide, hydrogen peroxide, hydroxyl, hydroxyl ion and nitric oxide radicals, and are important in recycling vitamin E radicals [7]. The ROS radicals damage DNA, lipids, proteins and carbohydrates, resulting in a variety of human diseases such as

¹Institute of Food, Nutrition and Family Sciences, University of Zimbabwe, P.O. Box MP 167, Mount Pleasant, Harare, Zimbabwe ²Biochemistry Department, University of Zimbabwe, P.O. Box MP 167, Mount Pleasant, Harare, Zimbabwe *phone/fax: +263 775990494

neurodegenerative disorders (Alzheimer's disease, Parkinson's disease), cancer, age-related eye diseases, arthritis, diabetes and atherosclerosis [8].

In addition to the nutritional and health benefits associated with baobab fruit, processing of the pulp has many other advantages including the alleviation of poverty through creating employment, thus improving food security in rural communities [9]. As baobab fruit pulp has been acknowledged as a novel ingredient by European countries and the USA [4], African countries can earn foreign currency by exporting the pulp to Europe and North America. Globally, the importance of wild fruits as ingredients in commercial food products and as sources of phytochemicals has been overlooked by researchers and organizations involved in community development. This study, therefore, sought to determine the nutritional, phenolic content and antioxidant activity of baobab-flavoured yoghurt.

Materials and methods

Sources of ingredients and commercial yoghurts

Skim milk powder (SMP), sugar, modified starch, commercial (i.e. strawberry and forest fruit yoghurts) and baobab fruit pulp were bought from a Spar retail outlet in Harare. YoFlex culture (Chr. Hansen, Hoersholm, Denmark) containing strains of *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus* in a 1:1 ratio was bought from a Kefalos retail outlet in Harare. Tables 1 and 2 show the nutritional and phenolic content and antioxidant activity of the baobab pulp.

Macronutrients	Content (g/100 g)	Mineral/ vitamin	Content (mg/100 g)
Ash	3.70±0.01	Calcium	500.0±32.0
Crude fat	2.60±0.02	Magnesium	295.0±25.0
Crude protein	1.30±0.01	Potassium	2,500.0±200.0
Crude fibre	3.00±0.02	Zinc	3.00±0.05
		Iron	12.0±3.0
		Phosphorous	430.0±31.0
		Vitamin C	574.0±23.5
Table 1 - Macronutrient, mineral and vitamin C content of			

 Table 1 - Macronutrient, mineral and vitamin C content of baobab pulp

Preparation of experimental baobab yoghurt and control yoghurt

We previously optimized the ingredient levels for baobab-flavoured yoghurt using response surface methodology (RSM) [10], and 15 baobab-flavoured yoghurts with different amounts of baobab pulp, modified starch and sugar were formulated. A central composite rotatable design (CCRD) was used to evaluate the effects of the three ingredients on appearance, consistency on the spoon, consistency in the mouth, taste, overall acceptability, vitamin C content, total phenolic content and antioxidant activity. The baobab-flavoured yoghurt which obtained the highest score of 65.45% was formulated with 37.21 g baobab pulp, 1.85 g modified starch and 51.21 g sugar per litre of yoghurt [10], so these ingredient levels were used to produce the experimental yoghurts.

Experimental optimized yoghurt samples were produced in our laboratory according to a method modified from Tamime and Robinson [11]. First, SMP (125 g) was added to a mixture of sugar (51.21 g), modified starch (1.85 g) and potassium sorbate (50 mg), and the mixture was blended with an electric mixer (Phillips, South Africa) for 2 min. Warm (50°C) distilled water (1 litre) was added to the mixture, mixed for 1 min and allowed to stand for 2 h. The mixture was then heated and maintained at 85°C on a hot plate for 30 min. After heating, the mixture was cooled with ice to 43°C and inoculated with YoFlex culture (40 mg). The inoculated mixture was incubated at 43°C until pH was 4.6, and then cooled to 10°C. After 12-h storage, pasteurized baobab pulp (37.21 g) was

Total phenolic content (mg GAE/100 g)	559.30±19.01		
Total flavonoid content (mg CE/100 g)	200.00±10.00		
Proanthocyanidins (g LE/100 g)	8.3±0.1		
DPPH activity (%)	81.67±3.51		
Phenolic acids (mg/kg)			
Syringic acid	19.27±3.10		
Caffeic acid	7.9±1.80		
<i>p</i> -Dihydroxybenzoic acid	55±6.40		
Gallic acid	158.4±15.70		
Protocatechuic acid	117±8.45		
Table 2 - Composition of phenolic compounds and DPPH ac- tivity of baobab pulp			

added to the cooled yoghurt, and slowly mixed using an electric mixer (Phillips) for 1 min.

Baobab pulp was pasteurized using an ultrasonicator (model 90S, Westwood Ultrasonics, UK) for 10 min, and the sample temperatures then maintained between 32 and 35°C using ice to preserve heat-labile vitamins and phytochemicals.

The control yoghurt was made of 1 litre distilled water, 125 g SMP, 51.21 g sugar, 1.85 g modified starch and 50 mg potassium sorbate with no add-ed baobab pulp. All yoghurts were analysed after 7-day storage at 4°C.

Proximate composition

Moisture, crude fat, crude protein, crude fibre, ash and the energy content of yoghurts were analysed according to modified AOAC methods [12]. The carbohydrate content was the percentage of matter that remained after moisture, ash, fat and protein were determined. The energy levels of yoghurts were calculated as follows:

Energy (kJ)=(Crude protein×16.7)+ (Crude fat×37.7)+(Carbohydrate×16.7)

Vitamin C

Vitamin C was extracted following the AOAC method [12]. The yoghurt sample (5 g) was mixed with 100 ml of 1% acetic acid, and the mixture centrifuged at 3000 rpm for 10 min and left to stand for 20 min. The supernatant was collected for further analysis. For standardizing DCPIP, 20 ml of DCPIP solution (26 mg dissolved in 100 ml distilled water containing 21 mg sodium hydrogen carbonate) was titrated against ascorbic acid (50 mg in 50 ml metaphosphoric acid). After standardization, acetic acid (20 ml, 1%) was added to the filtered supernatant (5 ml) in a volumetric flask, and the samples titrated against DCPIP.

Mineral analyses

Ashed yoghurt samples (1 g) were digested by a mixture of HNO₃ (70%, 5 ml) and HCl (98%, 5 ml). The content of calcium, magnesium, potassium, zinc and iron was determined in the digested samples using a Perkin-Elmer 2280 emission and absorption spectrophotometer [12]. A photometer was used to determine phosphorous by the molyb-date-vanadium method [12].

Extraction of phenolic compounds

Before extraction of the phenolic compounds, the yoghurt samples were defatted using hexane to minimize subsequent oxidation of the extracts. Yoghurt samples (50 g) were defatted with hexane (60 ml) by overnight stirring. The mixture was then filtered, and the residue extracted twice with hexane for 1 h. The defatted yoghurt samples (0.5 g) were then mixed with 5 ml of 50% methanol. The mixture was vortexed for 1 min and then ultrasonicated for 10 min. The ultrasonicated mixture was centrifuged at 3000 rpm for 10 min, and the resulting supernatant used in the following analysis.

Total content of phenolic compounds

The extracted yoghurt sample (50 μ l) was mixed with distilled water (950 μ l), followed by the Folin–Ciocalteau reagent (1 N, 500 μ l) and then sodium carbonate (2%, 500 μ l). The mixture was incubated at room temperature for 40 min, and then a Spectronic 20 Genesys spectrophotometer set at 725 nm was used to measure the absorbance of the mixture against a blank of 50% methanol. The total content of phenolic compounds was expressed as milligrams gallic acid equivalents per 100 g sample (mg GAE/100 g).

Total content of flavonoids

The extracted yoghurt sample (4 ml) was mixed with 2% methanolic $AlCl_3 \cdot 6H_2O$ (2 ml), and the mixture was incubated in a dark room for 15 min. A Spectronic 20 Genesys spectrophotometer at 430 nm was used to measure the absorbance of the extracts against a blank of 50% methanol. Quercetin was used as the standard, and the total content of flavonoids was expressed as milligrams of quercetin equivalents (QE) per 100 g of yoghurt extract (mg QE/100 g).

Total content of condensed tannins

The extracted yoghurt sample (0.5 ml) was added to butanol–HCl reagent (butanol:HCl 95:5 v/v). The mixture was added to 0.1 ml ferric reagent (2% ferric ammonium sulfate dissolved in 100 ml 2 N HCl). The mixture was vortexed for 2 min and then heated at 90°C in a water bath for 1 h. The mixture was cooled and a Spectronic 20 Genesys spectrophotometer set at 550 nm was used to measure the absorbance of the mixture against a blank of 50% methanol. The total content of condensed tannins was expressed as milligrams of leucocyanidin equivalents (LE) per 100 g of yoghurt extract (mg LE/100 g).

2.2-Diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging activity

Free radical scavenging activity was determined following the method of Kuda *et al.* [13]. The extracted yoghurt sample (20 μ l) was added to 2.980 ml methanolic solution of DPPH (0.0012 g/100 ml). A Spectronic 20 Genesys spectrophotometer set at 517 nm was used to measure the absorbance of the mixture against a blank of 50% methanol. Gallic acid (0.5 mg/ml) was used as a positive control. The scavenging activity was calculated as: % Scavenging activity=[1–(A517 nm sample/

A517 nm control)]

where A517 nm was the absorbance of the sample or control at 517 nm.

Phenolic acids

For extraction of phenolic acids, the defatted sample (0.5 g) was mixed with 5 ml of 50% methanol containing butylated hydroxytoluene (1 g/l) and 10 ml of 6 M HCl [14]. Nitrogen was then bubbled through each mixture for 1 min. The mixture was ultrasonicated for 10 min, and refluxed in a water bath at 85°C for 120 min. The refluxed mixture was filtered through a 0.22-µm filter unit (Millex-GV; Millipore, Molsheim, France) and made up to 100 ml with 50% methanol.

A Shimadzu HPLC system with a SCL-6B Shimadzu system controller, C-R AX Shimadzu Chromatopac, and a Shimadzu SPD-10 AV UV-Vis detector equipped with a Dynamax 60 Å C18 column was used for analysis of phenolic compounds. The eluent was made up of methanol, acetic acid and water (10:75:1). The phenolic standards (0.1 mg/ml) from Sigma Aldrich (St. Louis, USA), followed by 20 μ l of yoghurt samples were injected into the HPLC system. The system with a flow rate of 1 ml/min was run for 40 min, and detection of the phenolic acids was done at 280 nm. After each run, the system was reconditioned for 15 min before analysis of the next sample.

Statistical analysis

The results reported in this study are given as means±standard deviation. The parameters measured were compared using one-way analysis of variance (ANOVA) in Graphpad Prism, and means were separated by Tukey's post hoc test with p<0.05 considered significant.

Results and Discussion

Macronutrient and vitamin C content

Results for the macronutrients and vitamin C are shown in Table 3. Baobab yoghurt (82.5 ± 0.3 g/100 g) had the highest moisture content, while strawberry yoghurt (79.3 ± 0.6 g/100 g) had the lowest as shown in Table 3. Generally, yoghurts are high moisture products, and most of the reported values are above 80 g/100 g [14].

The ash content of the yoghurts ranged from 3.0 ± 0.03 g/100 g for control yoghurt to 3.7 ± 0.01 g/100 g for baobab yoghurt. Igbabul *et al.* [15] reported a lower ash content in yoghurts (0.14–1.02 g/100 g) than found in this study. Therefore, baobab yoghurt is a good source of minerals.

The fat content of the yoghurts ranged from 2.9 ± 0.01 g/100 g for strawberry yoghurt to 3.6 ± 0.02 g/100 g for baobab yoghurt. The baobab yoghurt analysed in this study had a higher fat content than fruit yoghurts reported in the literature [15]. However, compared to meat and legumes, yoghurts are poor sources of fat.

The protein content of the yoghurt samples ranged from 2.6 ± 0.02 g/100 g for control yoghurt to 4.8 ± 0.02 g/100 g for strawberry yoghurt. The yoghurts analysed in this study are poor sources of protein compared to meat and meat products.

Baobab yoghurt (0.2±0.02 g/100 g) had the highest fibre content as fibre was not detected in the other yoghurt samples. Furthermore, baobab yoghurt had a higher fibre content than fruit yoghurts reported in the literature [15]. Baobab pulp is a rich source of fibre (Table 1) and contributed significant fibre to baobab yoghurt in this study. The red fibre from baobab pulp was reported to have higher antioxidant activity compared to other natural sources of antioxidants such as orange, kiwi, apple and strawberry pulp [16].

The carbohydrate content of the yoghurts ranged

Analysis	Baobab yoghurt	Control yoghurt	Forest fruit yoghurt	Strawberry yoghurt
Moisture (g/100 g)	82.5±0.3ª	81.4±0.3 ^{ab}	82.4±0.4ª	79.3±0.6 ^b
Ash (g/100 g)	3.7±0.01ª	3.0±0.03 ^b	3.2±0.03 ^c	3.5±0.02 ^d
Crude fat (g/100 g)	3.6±0.02ª	3.5±0.03 ^a	3.4±0.01 ^{ac}	2.9±0.01 ^b
Crude protein (g/100 g)	2.7±0.01ª	2.6±0.02 ^a	4.1±0.03 ^b	4.8±0.02 ^c
Crude fibre (g/100 g)	0.2±0.02ª	ND* ^b	ND ^b	ND ^b
Carbohydrate (g/100 g)	7.5±0.8ª	9.5±0.3ª	6.9±0.4ª	9.9±0.6ª
Energy (kJ/100 g)	306.06±18.2ª	334.02±8.5ª	311.88±14.1ª	354.82±10.5ª
Vitamin C (mg/100 g)	18.25±0.02ª	3.17±0.02 ^b	2.7±0.03 ^c	2.8±0.04 ^c

Results are presented as the mean±standard deviation of three different experiments

Mean±standard deviation values in the same row with the same letter are insignificantly different (p>0.05)

 Table 3 - Macronutrient and vitamin C content of baobab yoghurt, control yoghurt and commercial yoghurts

from 6.9 ± 0.4 g/100 g for forest fruit yoghurt to 9.9 ± 0.6 g/100 g for strawberry yoghurt, which is higher than the content reported by Ilyasoğlu et al. [17] for hazelnut-flavoured yoghurt.

Strawberry yoghurt (354.82±10.5 kJ/100 g) had the highest energy level, while baobab yoghurt (306.06±10.2 kJ/100 g) had the lowest. The energy levels of the baobab yoghurts in this study are within the 295.8–409.2 kJ/100 g range reported for hazelnut slurry fortified yoghurts [17]. Energy levels of food products are influenced by their protein, fat and carbohydrate contents; the low contents of these nutrients in this study are reflected in the low energy levels.

The vitamin C content of the yoghurts ranged from 2.7 ± 0.03 mg/100 g for forest fruit yoghurt to 18.25 ± 0.02 g/100 g for baobab yoghurt. The

Nutrient	Baobab yoghurt (mg/100 g)	Control yoghurt (mg/100 g)	Forest fruit yoghurt (mg/100 g)	Strawberry yoghurt (mg/100 g)
Calcium	97.6±10.0ª	86.6±12.0ª	105.5±12.0 ^a	114.43±8.0 ^a
Magnesium	28.0±3.0ª	18.0±2.0ª	20.5±4.0 ^a	25.8±3.0ª
Potassium	247.4±30.0 ^a	195.4±27.0 ^a	203.8±25.0 ^a	242.7±22.0 ^a
Zinc	0.25±0.01ª	0.22±0.01 ^{ac}	0.27±0.01 ^a	0.31±0.02 ^{ab}
Iron	0.33±0.01 ^a	0.20±0.02 ^b	0.003±0.001 ^c	0.076±0.010 ^d
Phosphorous	48.8±9.5ª	33.04±5.5ª	40.8±7.5ª	32.6±5.5ª

Results are presented as the mean \pm standard deviation of three different experiments

Mean \pm standard deviation values in the same row with the same letter are insignificantly different (p>0.05)

Table 4 - Mineral content of baobab, control and commercial yoghurts

consumption of 100 g of baobab yoghurt provides 30% of the recommended daily intake of vitamin C in pregnant women. The health benefits of vitamin C include the prevention and treatment of scurvy, the common cold, wounds, atherosclerosis and cancer [18].

Mineral content of yoghurts

Results for the mineral content of the yoghurts are shown in Table 4. Strawberry yoghurt (114.43±8.00 mg/100 g) had the highest calcium content, while

the control yoghurt (86.60±12.00 mg/100 g) had the lowest. In this study, baobab pulp significantly contributed to the calcium content of the baobabflavoured yoghurt, consumption of 100 g of which provides 36% of the adequate daily intake of calcium in infants. Baobab pulp is an excellent source of calcium (Table 1) and has a higher calcium content than milk and some dairy products [11].

The magnesium values of yoghurt ranged from 18.0±2.0 mg/100 g for control yoghurt to 28.0±3.0 mg/100 g for baobab yoghurt.

Consumption of 100 g of baobab yoghurt provides 37% of the recommended daily allowance of magnesium for infants aged 7–12 months. Magnesium has structural roles in bones, cell membranes and chromosomes [19].

The potassium content varied from 195.4±27.0

mg/100 g for control yoghurt to 247.4±30.0 mg/100 g for baobab yoghurt. Baobab yoghurt had higher potassium content compared to pineapple, strawberry and wild berry fruit yoghurts with concentrations of 128.5, 119.1 and 120.9 mg/100 g, respectively [20]. Consumption of 100 g of baobab yoghurt provides 35% of the recommended daily allowance of potassium for infants aged 7–12 months. This essential dietary mineral and electrolyte is important for muscle contraction, nerve impulse transmission and heart function [21]. Strawberry yoghurt had the highest zinc content at 0.31±0.02 mg/100 g, while the control yoghurt had the lowest at 0.22±0.01 mg/100 g. The reported zinc content of strawberry (0.32 mg/100 g) and peach (0.28 mg/100 g) yoghurt is comparable to results obtained in this study [20]. Consumption of 100 g of baobab yoghurt provides 8% of the recommended daily allowance of zinc for infants aged 7–12 months. Although the zinc levels found in this study are low, additional zinc from the baobab-flavoured yoghurt may provide therapeutic benefits for children with diarrhoea and other diseases [22].

The iron values of the studied yoghurts ranged from 0.003 ± 0.020 mg/100 g for forest fruit yoghurt to 0.33 ± 0.01 mg/100 g for the baobab yoghurt. Sanchez-Segarra *et al.* [20] reported a lower iron content for strawberry (0.12 mg/100 g) and peach (0.05 mg/100 g) yoghurt compared to the baobab yoghurt analysed in this study. The consumption of 100 g of baobab yoghurt provides only 3% of the recommended daily allowance of iron for infants aged 7–12 months and therefore is a poor source of iron. In developing countries, iron deficiency is associated with considerable health costs, poor pregnancy outcomes, decreased productivity and impaired school performance [23].

Baobab yoghurt ($48.8\pm9.5 \text{ mg}/100 \text{ g}$) had the highest phosphorous content, while strawberry yoghurt ($32.6\pm5.5 \text{ mg}/100 \text{ g}$) had the lowest. Consumption of 100 g of baobab yoghurt provides 18% of the recommended daily allowance of phosphorous for aged infants 7–12 months. Phosphorous is important for preventing osteoporosis, which is a global health problem [24].

The variations in the nutritional composition

among the yoghurts may be due to several factors that influence the nutritional quality of fruits, including soil, weather and climatic factors, fertilizer application, postharvest handling and storage [25]. Also, the variations may be attributed to the different amounts of fruit and milk in the product, and the different methods of extraction and analysis [26]. Furthermore, variation in the nutritional composition may be attributed to genetic factors because the fruits used to make the yoghurts belonged to different genera.

Total content of phenolic compounds

Total phenolic content varied from 22.67 ± 2.08 mg GAE/100 g for baobab yoghurt to 0.87 ± 0.3 mg GAE/100 g for control yoghurt, as shown in Table 5. In this study, the total content of phenolic compounds in baobab yoghurt was higher than that in fruit yoghurts reported by other researchers, indicating that baobab yoghurt is an important source of antioxidant components.

Karaaslan *et al.* [27] reported the concentration of phenolic compounds in grape and callus yoghurts as 7.90±10.05 mg GAE/100 g and 7.5±0.32 mg GAE/100 g, respectively.

Baobab pulp is an excellent source of phenolic compounds (Table 2), and in this study contributed extra phenolic compounds to baobab yoghurt. These phenolic compounds have antimicrobial and antioxidant activities [28].

Total content of flavonoids

Baobab yoghurt (4.23±1.00 mg CE/100 g) had the highest total content of flavonoids, while control yoghurt (0.38±0.08 mg CE/100 g) had the lowest, as shown in Table 5. The total content of flavonoids was higher in baobab yoghurt than in the control, indicating that baobab pulp supplemented the flavonoids in the product. Table 2 indicates that baobab pulp is a good source of flavonoids, which contribute to the anticarcinogenic, antiallergic, antiviral, antiinflammatory and antiproliferative activities of foods [29].

Sample	Total phenolic content (mg GAE/100 g)	Flavonoids (mg CE/100 g)	Proanthocyanidins (g LE/100 g)	Antioxidant activity (%)
Baobab yoghurt	22.67±2.08ª	4.23±0.25 ^a	5.00±0.50 ^a	19.03±0.25ª
Control yoghurt	0.87±0.15 ^b	0.38±0.08 ^b	0.00 ± 0.00^{b}	11.40±0.2 ^b
Forest fruit yoghurt	5.17±0.76 ^c	1.80±0.26 ^c	7.97±0.25 ^c	10.57±0.40 ^c
Strawberry yoghurt	8.33±1.53 ^c	2.80±0.20 ^d	3.00±0.20 ^d	15.47±0.25 ^d

Results are presented as the mean \pm standard deviation of three different experiments Mean \pm standard deviation values in the same column with the same letter are insignificantly different (p>0.05)

 Table 5 - Total phenolic content, flavonoids and DPPH free radical scavenging activity of yoghurts and baobab pulp

Content of proanthocyanidins

Proanthocyanidins were not found in the control yoghurt (Table 5) but were present in forest fruit yoghurt (7.97±0.25 g LE/100 g) at levels lower than those found in various fruits. Baobab yoghurt had a significantly higher proanthocyanidin content (5.00 ± 0.25 g LE/100 g) than that reported by Nhukarume *et al.* [30] in a baobab beverage and other beverages prepared from fruits indigenous to Zimbabwe. Proanthocyanidins are bioavailable and have better protective effects against diseases than nutritional antioxidants such as vitamin C and β -carotene [31].

Content of phenolic acids

The contents of phenolic acids in the yoghurts are shown in Table 6. Syringic acid (0.72±0.01 mg/ kg) was found in baobab yoghurt but not in the other yoghurts. Although caffeic acid is common in fruits and vegetables, in this study the acid was only detected in the baobab and forest fruit yoghurts. Baobab yoghurt had the highest content of p-dihydroxybenzoic, which, however, was not detected in the control and strawberry yoghurts. Baobab yoghurt also had the highest content of gallic acid (6.60±2.20 mg/kg), which also was not detected in the forest fruit and strawberry yoghurts. Protocatechuic acid was detected in all yoghurts, with content ranging from 1.20±0.12 mg/ kg for the control yoghurt to 8.90±1.34 mg/kg for the forest fruit yoghurt. Chlorogenic acid content was 2.35±0.25 mg/kg for strawberry yoghurt, but was not detected in the other yoghurts. Most phenolic acids were identified in baobab yoghurt, which had higher phenolic acid contents than commercial yoghurts. The phenolic compounds identified in the baobab pulp, baobab yoghurt and commercial yoghurts are common plant metabolites with antioxidant activity [32]. Therefore, the phenolic acids identified in the yoghurts probably contributed to the total phenolic content and DPPH free radical scavenging activity of the products. Although fruits are good sources of phenolic acids, in this study low levels of and sometimes no phenolic acids were detected in the yoghurts, most likely because the low pH in the product could have induced degradation of these compounds [33]. Also, the yoghurt starter culture can metabolize phenolic acids to form biologically inactive compounds [34]. Phenolic acids have antioxidant, antifibrosis, antithrombosis, antitumour, antihypertension, antifibrosis and antivirus properties [35].

DPPH radical scavenging activity

The DPPH free radical scavenging activities of the yoghurts varied from $10.57\pm0.40\%$ for baobab yoghurt to $19.57\pm0.25\%$ for forest fruit yoghurt, as shown in Table 5. Baobab pulp has been reported to be a good source of dietary antioxidants including vistamin C, fibre and various phenolic compounds [4]. This study also showed that baobab pulp had a high content of vitamin C, fibre and phenolic compounds, and these compounds may have contributed to the antioxidant activity of baobab yoghurt. The phenolic compounds found in baobab pulp (Table 2) could help prevent diseases, such as cancer, caused by free radicals.

Phenolic acid	Baobab yoghurt (mg/kg)	Control yoghurt (mg/kg)	Forest fruit yo- ghurt (mg/kg)	Strawberry yoghurt (mg/kg)
Syringic	0.72±0.01ª	ND*	ND	ND
Caffeic	0.40±0.02 ^a	ND	0.25±0.01 ^b	ND
<i>p</i> -Dihydroxybenzoic	2.50±0.20ª	ND	1.90±0.12 ^b	ND
Gallic	6.60±2.20ª	0.40±0.01 ^b	ND	ND
Protocatechuic	5.50±1.10ª	1.20±0.12 ^b	8.90±1.34 ^c	1.50±0.15 ^b
Chlorogenic	ND	ND	ND	2.35±0.25ª

*The detection limits ranged from 0.08 mg/kg for caffeic acid to 0.5 mg/kg for gallic acid Results are presented as the mean \pm standard deviation of three different experiments Mean \pm standard deviation values in the same row with the same letter are insignificantly different (p>0.05)

Table 6 - Phenolic acid content of baobab, control and commercial yoghurts

The wide variations in the composition of phenolic compounds and antioxidant activin ity among the yoghurts may be due to different amounts of fruit in the product, different groups of phenolic compounds in the fruits, and different methods of extraction and analysis [26]. In this study, the observed total phenolic

content, total content of flavonoids and DPPH activity of control yoghurts were probably due to the presence of phenolic compounds in milk, and other reducing substances, such as dairy proteins. In any case, the clear increase observed in baobab yoghurts compared to control yoghurts indicates baobab pulp phenolic compounds were present in the product.

Conclusion

Baobab yoghurt was found to be a valuable source of minerals, crude fibre, vitamin C, iron and phenolic compounds compared to some commercial yoghurts. The antioxidant activity of the baobab yoghurt may be attributed to its vitamin C, fibre, flavonoid, proanthocyanidin and phenolic acid content. Therefore, baobab yoghurt can be used as a functional food to prevent diseases such as cancer and diabetes, which are caused by free radicals.

Acknowledgements

The authors would like to thank DAAD and the University of Zimbabwe Research Board for financial support.

Conflict of Interest

The authors declare that they have no conflict of interest.

REFERENCES

- Bremer B, Bremer K, Chase MW, Reveal JL, Soltis DE, Soltis PS (2013) An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG II. Bot J Linn Soc 141:399–436
- Yazzie D, VanderJagt DJ, Pastuszyn A, Okolo A, Glew RH (1994) The amino acid and mineral content of baobab (*Adansonia digitata* L.) leaves. J Food Comp Anal 7:189–193
- Gebauer J, El-Siddig K, Ebert G (2002) Baobab (*Adansonia digitata* L.): a review on a multipurpose tree with promising future in the Sudan. Gartenbauwissenschaft 1:55–60
- Chadare FJ, Linnemann AR, Hounhouigan JD, Nout MJR, Van Boekel M (2008) Baobab food products: a review on their composition and nutritional value. Crit Rev Food Sci Nutr 49:254–274
- Sidibe M, Williams JT, Hughes A, Haq N, Smith RW (2000) Crops for the future, Vol 4. Baobab, *Adansonia digitata* L. International Centre for Underutilised Crops, Southampton, UK

- De Caluwé E, Halamová K, Van Damme P (2009) Baobab (*Adansonia digitata* L.): a review of traditional uses, phytochemistry and pharmacology. In: Rodolfo H, Simon JE, Ho CT (eds) African natural plant products: new discoveries and challenges in chemistry and quality. Oxford University Press, USA, pp 51–84
- Hamid AA, Aiyelaagbe OO, Usman LA, Ameen OM, Lawal A (2010) Antioxidants: its medicinal and pharmacological applications. Afr J Pure Appl Chem 4:142–151
- Wang X, Wang W, Li L, Perry G, Lee H, Zhu X (2014) Oxidative stress and mitochondrial dysfunction in Alzheimer's disease. Biochem Biophys Acta 1842:1240–1247
- Nyanga LK (2012) Ziziphus mauritiana (masau) fruits fermentation in Zimbabwe: from black-box to starter culture development. Doctoral dissertation, Wageningen University, the Netherlands
- Chipurura B, Muchuweti M (2013) Postharvest treatment of *Adansonia digitata* (baobab) fruits in Zimbabwe and the potential of making yoghurt from the pulp. Int J Postharvest Technol Innov 3:392–402
- Tamime AY, Robinson RK (2007) Tamime and Robinson's yoghurt: science and technology. Woodhead Publishing, Cambridge
- 12. AOAC (1990) Official methods of analysis. 15th edn. Arlington, VA, USA
- Kuda T, Tsunekawa M, Goto H, Araki Y (2005) Antioxidant properties of four edible algae harvested in the Noto Peninsula, Japan J Food Comp Anal 18:625–633
- Sikorska M, Matlawska I, Glowniak K, Zgorka G (2000) Qualitative and quantitative analysis of phenolic acids in *Asclepias syriaca* L. Acta Pol Pharm 57:69–72
- Igbabul B, Shember J, Amove J (2014) Physicochemical, microbiological and sensory evaluation of yoghurt sold in Makurdi metropolis. Afr J Food Sci Technol 5:129–135
- Besco E, Braccioli E, Vertuani S, Ziosi P, Brazzo F, Bruni R (2007) The use of photochemiluminescence for the measurement of the integral antioxidant capacity of baobab products. Food Chem 102:1352–1356
- Ilyasoğlu H, Yılmaz F, Burnaz NA, Baltacı C (2015) Preliminary assessment of a yoghurt-like product manufactured from hazelnut slurry: study using response surface methodology. LWT-Food Sci Technol 62:497–505
- Naidu KA (2003) Vitamin C in human health and disease is still a mystery? An overview. Nutr J 2:7
- Rude RK, Shils ME. Magnesium (2006) In: Shils ME, Shike M, Ross AC, Caballero B, Cousins RJ (eds) Modern nutrition in health and disease. 10th edn. Lippincott Williams and Wilkins, Baltimore, pp 223–247



- Sanchez-Segarra PJ, Garca-Martinez M, Gordillo-Otero MJ, Diaz-Valverde A, Amaro-Lopez MA, Moreno-Rojas R (2000) Influence of the addition of fruit on the mineral content of yoghurts: nutritional assessment. Food Chem 71:85–89
- Peterson LN (1997) Potassium in nutrition. In: O'Dell BL, Sunde RA (eds) Handbook of nutritionally essential minerals. Marcel Dekker, New York, pp 153–183
- 22. Black RE (2003) Zinc deficiency, infectious disease and mortality in the developing world. J Nutr 133:1485S–1489S
- 23. Zimmermann MB, Hurrell RF (2007) Nutritional iron deficiency. Lancet 370:511–520
- 24. Carr AC, Frei B (1999) Toward a new recommended dietary allowance for vitamin C based on antioxidant and health effects in humans. Am J Clin Nutr 69:1086–1107
- 25. Hornick SB (1992) Factors affecting the nutritional quality of crops. Am J Altern Agric 7:63–68
- 26. Bravo L (1998) Polyphenols: chemistry, dietary sources, metabolism, and nutritional significance. Nutr Rev 56:317-333
- Karaaslan M, Ozden M, Vardin H, Turkoglu H (2011) Phenolic fortification of yogurt using grape and callus extracts. LWT-Food Sci Technol 44:1065–1072
- 28. Shetty K, Wahlqvist ML (2003) A model for the role of the proline-linked pentose-phosphate pathway in phenolic phytochemical bio-synthesis and mechanism of action for human health and environmental applications. Nutr Asia Pac J Clin 13:1–24

- 29. Yao LH, Jiang YM, Shi J, Tomas-Barberan FA, Datta N, Singanusong R (2004) Flavonoids in food and their health benefits. Plant Food Hum Nutr 59:113–122
- Nhukarume L, Chikwambi Z, Muchuweti M, Chipurura B (2010) Phenolic content and antioxidant capacities of Parinari curatelifolia, *Strychnos spinosa* and *Adansonia digitata*. J Food Biochem 34:207–221
- Bagchi D, Bagchi M, Stohs SJ, Das DK, Ray SD, Kuszynski CA (2000) Free radicals and grape seed proanthocyanidin extract: importance in human health and disease prevention. Toxicology 148:187–197
- Shahidi F, Wanasundara UN, Amarowicz R (1994) Natural antioxidants from low-pungency mustard flour. Food Res Int 27:489–493
- Kapasakalidis PG, Rastall RA, Gordon MH. Extraction of polyphenols from processed black currant (*Ribes nigrum* L.) residues. J Agric Food Chem 54:4016–4021
- 34. Redeuil K, Bertholet R, Kussmann M, Steiling H, Rezzi S, Nagy K (2009) Quantification of flavan-3-ols and phenolic acids in milk-based food products by reversed-phase liquid chromatography-tandem mass spectrometry. J Chromatogr A 1216:8362–8370
- Jiang RW, Lau KM, Hon PM, Mak TC, Woo KS, Fung KP (2005) Chemistry and biological activities of caffeic acid derivatives from *Salvia miltiorrhiza*. Curr Med Chem 12:237– 246