



Sapucaia nut (*Lecythis pisonis* Cambess) and its by-products: A promising and underutilized source of bioactive compounds. Part I: Nutritional composition and lipid profile



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ABSTRACT

The nutritional composition of the sapucaia nut, cake and shell, the nut and cake minerals content and the lipid profile of the nut oil (fatty acids, tocopherols, phytosterols and triacylglycerols) were determined. The nuts and cake exhibited a high content of lipid (47.9 to 60.8 mg 100 g⁻¹), protein (15.8 to 19.5 mg 100 g⁻¹), dietary fiber (16.5 to 22.6 mg 100 g⁻¹) and provided an excellent source of selenium (26.4 to 46.94 μg g⁻¹). The oil contained a high amount of unsaturated fatty acids (39.7 to 45.4% of oleic and 32.2 to 46.6% of linoleic acids) and presented a high Oxidative Stability Index (8.57–12.95 h) indicating the presence of antioxidant compounds in the oil. The major triacylglycerols in the sapucaia oil were LLO, PLO, LOO, POO, OOO, PLL and LLL. The main bioactive lipids identified in the oil were γ-tocopherol (19.2 to 28.5 mg 100 g⁻¹) and β-sitosterol (92.8 to 194 mg 100 g⁻¹). The results showed that the sapucaia nut and its by-products are a promising natural source of bioactive and nutritional compounds and when present in the diet can contribute to the maintenance of human health. In addition, the nut and by-product represents a promising raw material for the food industry.

1. Introduction

Nuts such as almonds, cashew nuts, walnuts, pistachios, hazelnuts, pecans, macadamia and Brazil nuts have received special attention from researchers due to their unique combination of nutrients. Studies have confirmed that a daily intake of nuts associated to a healthy diet contributes to the prevention of cardiovascular disease, type II diabetes and cancer (Aune et al., 2016; Nishi et al., 2014; Yang, 2009). The beneficial effects on health probably are due to the synergistic interaction of the bioactive constituents present in the nuts. Nuts provide large amounts of healthy monounsaturated and polyunsaturated fats which help to regulate blood cholesterol and prevent cardiovascular diseases (Nishi et al., 2014). They are also rich in fiber and phytosterols that contribute to the reduction of the cholesterol re-absorption in the

intestine (Robbins, Shin, Shewfelt, Eitenmiller, & Pegg, 2011; Salas-Salvadó, Bulló, Pérez-Heras, & Ros, 2006; Shahzad et al., 2017). In addition, nuts contain tocopherols (vitamin E), minerals (calcium, iron, zinc, potassium and magnesium), antioxidant minerals (selenium, manganese and copper) and phenolic compounds that may reduce oxidative stress and inflammation (Cardoso, Duarte, Reis, & Cozzolino, 2017; Colpo et al., 2014; Naozuka, Vieira, Nascimento, & Oliveira, 2011; Robbins et al., 2011). The low content of sodium and high content of potassium contribute to maintain a normal blood pressure (Ndanuko et al., 2017).

Brazil, due to its size and the existence of different biomes, has one of the largest world reserves of native plant species that have not been fully investigated. Such plants may present significant nutritional and economic potential. Sapucaia nut (*Lecythis pisonis* Cambess), from the

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botanical family of *Lecythidaceae*, is a species present in the Amazon region and Atlantic forest, mainly in the states of Amazonas, Pará, Rondonia, Piauí, Pernambuco, Maranhão, Bahia, Espírito Santo, Minas Gerais, and Rio de Janeiro (USDA, 2017; Vallilo, Tavares, Aued-Pimentel, Campos, & Moita Neto, 1999). The fruits present a weight between 1 and 2.5 Kg and the average yield of sapucaia nuts per tree is 75 kg. Each fruit contain between 30 and 50 nuts and each nut weighs between 4 and 14 g (Wickens, 1995). The nuts are rich in proteins, essential fatty acids and minerals (Denadai et al., 2007; Naozuka et al., 2011; Teixeira, Ávila, Ribani, Silveira, & Ribani, 2017; Teixeira, Ghazani, Corazza, Marangoni, & Ribani, 2018; Vallilo et al., 1999). Although sapucaia nuts are consumed by the local population, their nutritional and phytochemical compositions are unclear and the economic and socio-cultural potential of the nuts have not been explored. Only a few studies have investigated the chemical composition of sapucaia nuts and its by-products. In addition, since there is a demand for this raw material but it is not produced on a large scale, there is a high extractive potential for sapucaia nut, being possible increase their exploitation.

In this study, the nutritional and phytochemical composition of sapucaia nuts, cake, oil and shell were determined. This knowledge is fundamental for encouraging its sustainable production and consumption by the population. In addition, the exploitation of this oleaginous fruit may help the preservation of the ecosystem and the socioeconomic development of the local population and it is also a promising raw material for the food industry.

2. Materials and methods

2.1. Samples

Sapucaia nuts (*Lecythis pisonis* Cambess) were collected from native trees in Teresina - Piauí, Brazil (05° 05' 21" S; 42° 48' 07" W; 72 m of altitude, and average annual temperature and relative humidity of 26.2 °C and 71%, respectively) and provided by the Brazilian Agricultural Research Corporation (EMBRAPA Meio-Norte) (samples A1, A2 e A3). The sample B1 was collected in Viçosa - Minas Gerais, Brazil (20° 45' 14" S; 42° 52' 53" W; 648 m of altitude, and average annual temperature and relative humidity of 18.5 °C and 66%, respectively) and provided by the Federal University of Viçosa (UFV). All samples (about 600 g each) were harvested in October 2016.

2.2. Chemical reagents

The standards and Rh, Ca, Mg, Cu, Zn, Mn, Se, Na, Sn, Pb, Cd, Cr, Ni, Al and Mo stock solutions were purchased from Sigma-Aldrich Co. (St. Louis, USA). The fatty acid standards (a mix with 37 components, linolenic acid methyl ester isomer mix, all-cis-7,10,13,16,19-docosapentaenoic acid, linoleic acid and conjugated methyl esters) were purchased from Merck (Darmstadt, Germany). The tocopherols standards, α -, β -, γ - and δ -tocopherol, and the phytosterols standards, β -sitosterol, estigmasterol, campesterol and brassicasterol, were purchased from Merck (Darmstadt, Germany). Nitric acid (65% m/m) was purchased from Merck (Darmstadt, Germany) and purified by quartz double sub-boiling distillation (Kürner Analystechnik, Rosenheim, Germany). Solvents of analytical grade were used in the chemical analysis and they were purchased from Vetec Fine Chemicals (Xerem, Brazil) and Sigma-Aldrich Co. (St. Louis, USA).

2.3. Sample preparation and lipid extraction

The sapucaia nuts were cracked and the sapucaia oil was obtained using a hydraulic press TE-098 Tecnal® (Sao Paulo, Brazil). The samples were pressed three times and the obtained oil was centrifuged for 10 min at 4.000 rpm. The oil samples were stored in amber vials (flushed with nitrogen) at -24 °C for further use in the analyses. The

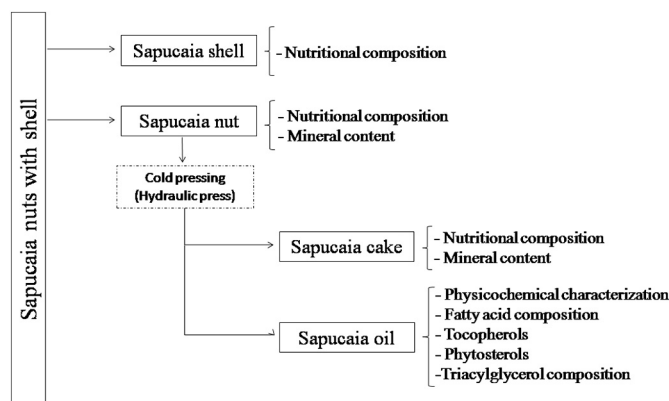


Fig. 1. Analyses carried out on sapucaia nuts, cake, oil and shell.

extraction yield (EY) was calculated according to the equation:

$$EY (\%) = \left[\frac{\text{weight of extracted oil}}{\text{weight of crushed almonds}} \right] \times 100 \quad (1)$$

The physicochemical characterization, fatty acid composition, tocopherol, phytosterol and triacylglycerol content of the extracted oil were determined. The nutritional composition and mineral content of the sapucaia nuts and of the resulting cake generated after pressing of the nuts were determined. Fig. 1 show a schematic diagram of the analyses carried out on sapucaia nuts, cake, oil and shell.

2.4. Nutritional composition

The nutritional composition of the sapucaia nut, cake, and shell was performed following the methods recommended by the Association of Official Analytical Chemicals (AOAC, 2005). The moisture content (925.09) was determined by oven-drying the samples at 105 °C until a constant weight was reached. The ash content (923.03) was determined by incineration at 550 ± 15 °C. The crude protein content (N × 6.25) was estimated by the macro-Kjeldahl method (920.87). The total lipids (920.85) were determined by the Soxhlet extraction method. The total dietary fiber, was analyzed by the enzymatic-gravimetric method (991.43). Total carbohydrates were calculated by difference according to the Eq. (2):

$$\text{Total carbohydrates (g 100 g}^{-1}\text{)} = 100 - (\text{g fat} + \text{g protein} + \text{g ash} + \text{g fiber}) \quad (2)$$

Total energy was calculated according to the Eq. (3):

$$\text{Energy (kcal 100 g}^{-1}\text{)} = 4 \times (\text{g proteins} + \text{g carbohydrates}) + 9 \times (\text{g fat}) \quad (3)$$

2.5. Mineral content

The samples (0.1 g) were digested using a MLS-1200 Milestone microwave oven (Soriso, Italy), with 6 mL of HNO₃ and 1 mL of H₂O₂, with the applied power varying from 250 to 600 W for 25 min in closed PFA vessels. The digested samples were diluted appropriately with deionized water. Rhodium (10 µg L⁻¹) was used as the internal standard for all determinations. External calibration was carried out using aqueous solutions prepared from a multi-element stock standard solution containing all analytes. The elements selenium, calcium, magnesium, manganese, zinc, copper, chrome, nickel, molybdenum, sodium, aluminum, cadmium, tin and lead were determined by inductively coupled plasma mass spectrometry (ICP-MS) using a Perkin Elmer SCIEX, model NexIon 300D (Shelton, USA). The operating parameters of the ICP-MS equipment were: sampling/skimmer cones: platinum; RFpower: 1100 W; signal measurement: continuous; auto lens: on;

detector voltage: pulse (1250 v) and analog (–2290 v); gas flow rate: main (15.0 L min⁻¹) and nebulizes (1.05 L min⁻¹). The data on limit of detection (LOD), limit of quantitation (LOQ) and determination coefficients (R²) of each mineral are available in Supplementary material (Table S1).

2.6. Physicochemical characterization of the oil extracted from sapucaia nut

The physicochemical characteristics of the sapucaia nut oil were determined according to methods of the American Oil Chemists' Society (AOCS, 2004): refractive index (Cc 7-25); relative density (Cc 10a-25); color measurement (Cc 13b-45); smoke point (Cc 9a-48); acid value (Cd 3d-63); peroxide value (Cd 8-53); specific extinction (232 and 270 nm) (Ch 5 – 91); *p*-anisidine value (Cd 18-90) and oil stability index (Cd 12b-92).

2.7. Fatty acid composition

The fatty acids were determined by the Ce 1a-13 method (AOCS, 2004). The fatty acids methyl esters were prepared following the procedure described by Hartman and Lago (1973). The analyses were performed using a gas chromatograph 3900 Varian® (Palo Alto, USA). The chromatographic conditions were as follows: capillary column (CP-Sil 88, Chrompack) with 100 m length, 0.25 mm inner diameter, and 0.2 µm film; and a flame ionization detector (FID). Operating conditions: split of 1:50; detector temperature of 310 °C; injector temperature of 270 °C; oven temperature of 120 °C 2 min⁻¹, 120–220 °C (2.2 °C min⁻¹) and 220–235 °C (1.5 °C min⁻¹), remaining 235 °C 15 min⁻¹; auxiliary gas (make up gas) nitrogen (30 mL min⁻¹); carrier gas: hydrogen (1 mL min⁻¹); volume injected of 1 µL. Each supernatant was injected once into the chromatograph. The qualitative composition was determined by the comparison of peak retention times with the respective standards for fatty acids. The quantitative composition was determined by area normalization and expressed as mass percentage.

2.8. Tocopherols

The sample extraction and preparation of standards were carried out according to Panfili, Fratianni, and Irano (2003). Between 0.1 and 1.2 g of oil was weighed in a 10 mL flask and the volume was completed with *n*-hexane. The solution was homogenized on a tube shaker for one minute. The extract was filtered through a Gelman Acrodisc LC13 PVDV 0.45 µm pore size syringe filter (PALL Life Sciences, Ann Arbor, USA) before analysis. The tocopherol content of the sapucaia nut oils was determined by HPLC using a Shimadzu Prominence Chromatograph (Shimadzu®, Kyoto, Japan). The analysis conditions used were as follows: fluorescence detector at 294 nm excitation and 326 nm emission wavelengths; silica column Si-60 Merk (125 nm × 4 nm × 5 µm) with a column temperature of 24 °C and a flow rate of 1.5 mL min⁻¹; mobile phase: *n*-hexane/ethyl acetate/acetic acid (97.3: 1.8: 0.9 v/v/v). Peak identification was performed by comparing sample retention times with standards. For analytical quantification, analytical curves from standard compounds were used.

2.9. Phytosterols

The analysis of phytosterols was performed according to Bragagnolo and Rodriguez-Amaya (1993). Samples were subjected to direct saponification and the unsaponifiable matter was extracted with hexane and analyzed using a gas chromatograph 3900 Varian® (Palo Alto, USA). The chromatographic conditions for the analysis were: automatic injection (1 µL); capillary column (Chrompack CP-Sil 88, 100 m long, 0.25 mm internal diameter, 0.20 µm de film); split injection (1:50); detector temperatures at 300 °C; injector temperature at were 250 °C; column temperature was initially set at 150 °C and held for 1 min, then

the column was heated at 150–300 °C at a rate of 10 °C min⁻¹, and held at 300 °C for 10 min; auxiliary gas (make up gas) nitrogen (30 mL min⁻¹); carrier gas: hydrogen (30 mL min⁻¹). Peak identification was performed by comparing the retention times of the samples with the retention times of the standards.

2.10. Triacylglycerol composition

The triacylglycerols (TAGs) of the sapucaia nut oil were determined according to the method reported by Segura, Silva, Soares, Gioielli, and Jachmanián (2011), by High-Performance Liquid Chromatography (HPLC) using a chromatograph Shimadzu Prominence 20A (Shimadzu® Corporation, Kyoto, Japan). 5 mg of oil were dissolved in 1 mL of acetone and mixed by stirring. The chromatographic separation of the compounds was performed using the following HPLC conditions: the injection volume was 1 µL; detector Shimadzu ELSD-LTII equipped with an evaporative light scattering; two columns Supelcosil™ C18 (25 cm × 4.6 mm × 5 µm), connected in series and operated at approximately 20 °C; mobile phase was acetone/acetonitrile (1:1) with a flow of 1 mL min⁻¹ and an increasing linear gradient of chloroform (20% to 60 min). This solvent composition remained constant for 20 min, then it returned to the starting composition at 85 min. Peaks were identified using pure TAG standards and considering the order of elution, according to the corresponding equivalent carbon number (ECN). Two replicate analyses were performed and the average values were reported.

2.11. Statistical analysis

All analyses were carried out in duplicate or triplicate and the results were expressed as means ± standard deviation (SD). The analysis of variance (ANOVA) was carried out and the mean values were compared with Tukey's test considering a 5% level of significance (*p* < 0.05). The statistical analysis of the results was performed using the software Statistica® 7.0 (2008).

3. Results and discussion

3.1. Nutritional composition of sapucaia nut, cake and shell

Table 1 shows that sapucaia nuts have a high lipid (47.97 to 60.76 g 100 g⁻¹) and protein content (15.80 to 19.49 g 100 g⁻¹), which contributes to a high energy value (534.13 to 615.74 kcal 100 g⁻¹). The sample from Minas Gerais (B1) presented a significantly lower (*p* < 0.05) lipid content and a significantly higher (*p* < 0.05) protein content when compared to the samples from Piauí (A1, A2 and A3). The nuts are also rich in dietary fiber and inorganic compounds. Vallilo et al. (1999), Denadai et al. (2007) and Carvalho, Costa, Souza, and Maia (2008) reported similar results for lipid content, protein and ash for sapucaia nuts. The dietary fiber content in the samples of this study was higher than that reported by Denadai et al. (2007), and Carvalho et al. (2008) (5.67 and 7.0 g 100 g⁻¹, respectively). The high value of dietary fiber in the nuts may contribute to several health benefits, such as the reduction of the blood cholesterol and of the postprandial glucose response. In addition, the consumption of dietary fibers has been associated to a reduction in the risk of developing coronary heart diseases and diabetes (Salas-Salvadó et al., 2006).

The cake, obtained from the pressing process of sapucaia nuts, contained a high amount of lipids, which is related to the low oil yield (51%, 45% 54% and 30% for samples A1, A2, A3 and B1, respectively). Costa and Jorge (2012) using the same extraction process reported average oil yields of 59% for sapucaia nut. Rabadán, Álvarez-Ortí, Gómez, Alvarruiz, and Pardo (2017) reported low yield for pistachio oil obtained by hydraulic press 24.5%. The higher lipid content in the cake obtained from sample B1 may be due to the higher nut moisture content since the extraction efficiency is affected by the sample's temperature

Table 1
Nutritional composition (g 100 g⁻¹) of sapucaia nut, cake and shell.

Sample	Parameter (dry basis)						
	Moisture (g 100 g ⁻¹)	Ashes (g 100 g ⁻¹)	Lipids (g 100 g ⁻¹)	Protein ^a (g 100 g ⁻¹)	Total dietary fiber ^{**} (g 100 g ⁻¹)	Carbohydrate (g 100 g ⁻¹)	Energy value (kcal 100 g ⁻¹)
Nuts							
A1	4.15 ± 0.07 ^b	3.53 ± 0.20 ^a	60.76 ± 0.55 ^a	15.80 ± 0.31 ^b	16.51 ± 0.87 ^a	4.90 ± 0.21 ^b	616.57 ± 6.64 ^a
A2	4.31 ± 0.03 ^b	3.34 ± 0.20 ^a	57.49 ± 0.67 ^b	15.83 ± 0.22 ^b			
A3	4.29 ± 0.09 ^b	3.36 ± 0.01 ^a	59.48 ± 0.62 ^{ab}	16.21 ± 0.47 ^b			
B1	9.15 ± 0.11 ^a	3.34 ± 0.36 ^a	47.99 ± 0.15 ^c	19.49 ± 0.43 ^a	22.63 ± 0.28 ^b	6.50 ± 0.04 ^a	534.13 ± 3.27 ^b
Cake							
A1	7.61 ± 0.14 ^b	5.17 ± 0.34 ^a	32.41 ± 2.48 ^{ab}	26.78 ± 1.00 ^b	31.23 ± 0.11 ^a	5.11 ± 0.13 ^a	414.31 ± 1.74 ^b
A2	6.39 ± 0.10 ^c	5.02 ± 0.12 ^a	31.98 ± 0.96 ^{ab}	26.54 ± 0.03 ^b			
A3	6.68 ± 0.07 ^c	5.07 ± 0.09 ^a	31.36 ± 0.25 ^b	26.70 ± 0.26 ^b			
B1	10.89 ± 0.10 ^a	3.35 ± 0.27 ^b	37.01 ± 0.47 ^a	29.67 ± 0.20 ^a	27.99 ± 0.36 ^b	1.98 ± 0.04 ^b	460.43 ± 3.55 ^a
Shell							
A1	10.98 ± 0.07 ^b	3.36 ± 0.01 ^a	0.46 ± 0.01 ^b	3.62 ± 0.17 ^a	75.22 ± 1.86 ^a	17.25 ± 1.90 ^a	88.49 ± 7.49 ^a
A2	10.31 ± 0.02 ^b	3.29 ± 0.01 ^a	0.41 ± 0.05 ^b	3.82 ± 0.01 ^a			
A3	10.86 ± 0.03 ^b	3.17 ± 0.07 ^a	0.61 ± 0.05 ^a	3.88 ± 0.08 ^a			
B1	13.44 ± 0.06 ^a	2.73 ± 0.10 ^b	0.63 ± 0.01 ^a	3.99 ± 0.07 ^a	80.89 ± 1.00 ^a	11.75 ± 0.83 ^b	68.64 ± 3.61 ^a

A1, A2, A3: sapucaia nuts from Piauí; B1: sapucaia nut from Minas Gerais. Mean ± S.D. (n = 3). Different letters in the same column presented difference at a significant level (Tukey test, p < 0.05).

^a Nitrogen conversion factor = 6.25.

^{**} The fiber analysis for the samples from Piauí were made for a pool of the samples.

and moisture (Rabadán et al., 2017). Moisture, ash, protein and dietary fiber contents of sapucaia cake were higher than those obtained from the nut because the oil separation concentrates the other components in the residual cake. The protein content was significantly higher (p < 0.05) for sample B1 when compared to samples A1, A2 and A3. On the other hand, the ash, dietary fiber and carbohydrate content for sample B1 was significantly lower (p < 0.05).

Dietary fiber represented the largest fraction of the composition of the sapucaia shell (between 75.22 and 80.89 g 100 g⁻¹), followed by the carbohydrates (11.75–17.11 g 100 g⁻¹) and proteins (3.62–3.99 g 100 g⁻¹). The ashes were significantly higher (p < 0.05) for the samples A1, A2, and A3 (3.17–3.36 g 100 g⁻¹) when compared with sample B1 (2.73 g 100 g⁻¹). The lipid content represented the smaller fraction of the sapucaia shell and was significantly higher (p < 0.05) for the samples A3 and B1.

The significant differences observed on the nutritional composition of the sapucaia nuts, cakes and shells obtained from different regions may be related to environmental factors, soil composition, geographical location, harvest period and genetic factors (Yada, Huang, & Lapsley, 2013).

3.2. Mineral content of sapucaia nuts and cake

The sapucaia nuts and cake contained 13 mineral elements (Table 2). The selenium (Se) concentration in the nut ranged between 26.4 and 46.9 µg g⁻¹ and between 28.7 and 48.5 µg g⁻¹ in the cake. The results indicate that the intake of a single sapucaia nut (2 g) may be sufficient to reach the recommended adult daily intake of selenium (55 µg/day), not exceeding the Tolerable Upper Intake Level (UL) (Supplementary material Table S2) (FAO/WHO, 2001; Food & Nutrition Board, 2004). Selenium, as a constituent of selenoproteins, is an essential micronutrient playing an important role in numerous physiological functions such as the conversion of the prohormone thyroxine (T4) to the active thyroid hormone triiodothyronine (T3), and also the conversion of inactive reverse T3 to diiodothyronine. The antioxidant activity of Se is also related to the prevention of age-related illnesses such as cancer and cardiovascular diseases (Kumar & Priyadarsini, 2014). Selenium has the ability to increase mercury (Hg) excretion and eliminate reactive oxygen species induced by Hg, indicating a protective action against the Hg (Sakamoto et al., 2013). A

selenium daily intake < 40 µg has been considered deficient. The selenium deficiency may lead to Keshan disease (an endemic cardiomyopathy with myocardial insufficiency), cancer and cardiovascular diseases (Roman, Jitaru, & Barbante, 2014). On the other hand, an intake of > 400 µg/day of Se has been associated with toxic effects, including selenose, the symptoms of which are loss of hair and nails, skin lesions and disorders of the nervous system (Roman et al., 2014). Considering the level of Se detected in a single sapucaia nut a daily intake exceeding 8–10 nuts may therefore exceed the toxicity limit for Se.

In other studies, Lemire, Fillion, Guimarães, and Mergler (2010) reported a Se content in the sapucaia nut between 5.01 and 14.36 µg g⁻¹. The Brazil nut, which has a content of Se between 0.2 and 512 mg kg⁻¹, is considered the food with the highest content of Se (Silva Júnior et al., 2017).

Sapucaia nuts are also rich in calcium (Ca) and magnesium (Mg). The Supplementary material (Table S2) shows the Recommended Dietary Allowance (RDA) for minerals for adults (19–50 years). Ca and Mg are important minerals in the diet. The first is related to bone health and protection against bone loss. On the other hand, Mg is related with activation of enzymatic systems (Fardellone, 2015). Denadai et al. (2007), Carvalho et al. (2012) and Naozuka et al. (2011) reported greater amounts of Ca and Mg in sapucaia nut samples from different regions of Brazil (1720 and 2790 µg g⁻¹; 1821 and 3434 µg g⁻¹; and 1798 and 3151 µg g⁻¹, respectively) when compared to the results of this study. However, smaller amounts for Ca (880 µg g⁻¹) and similar quantities for Mg (1465 µg g⁻¹) were reported by Vallilo et al. (1999) in sapucaia nut.

Mn, Zn and Cu are metals required in trace amounts for a large number of physiological functions. Mn is a cofactor for many enzymes, including hydrolases, lyases, superoxide dismutase (SOD) and glutamine synthetase (Parmalee & Aschner, 2016). Zn plays an important role in the normal functioning of immune system and acts as ionic signalling in large number of cells (Wani et al., 2017). Cu is an essential cofactor and a structural component of a number of important enzymes which are involved in redox reactions (Scheiber, Mercer, & Dringen, 2014). Denadai et al. (2007) and Carvalho et al. (2012) reported higher values for Mn (80.69 µg g⁻¹, 48 µg g⁻¹, respectively), Zn (40.37 µg g⁻¹, 44 µg g⁻¹, respectively) and Cu (32.76 µg g⁻¹, 23 µg g⁻¹, respectively) when compared with the results of this study. Naozuka et al. (2011)

Table 2
Mineral content ($\mu\text{g g}^{-1}$) of sapucaia nuts and cake.

Samples	Elements ($\mu\text{g g}^{-1}$)													
	Se	Ca	Mg	Mn	Zn	Cu	Cr	Ni	Na	Mo	Al	Cd	Pb	Sn
Nuts														
A1	34.8 ± 6.19 ^b	1249 ± 2.39 ^a	1682 ± 1.76 ^b	38.3 ± 2.31 ^a	11.8 ± 1.03 ^b	11.6 ± 2.45 ^{bc}	0.41 ± 0.061 ^{ab}	0.85 ± 0.265 ^b	5.34 ± 0.54 ^a	Nd	4.56 ± 0.665 ^b	0.35 ± 0.024 ^a	0.51 ± 0.049 ^a	7.48 ± 1.21 ^c
A2	46.9 ± 3.87 ^a	1168 ± 4.20 ^b	1572 ± 4.38 ^c	41.4 ± 1.87 ^a	20.9 ± 2.26 ^a	16.9 ± 0.632 ^a	0.45 ± 0.013 ^a	0.48 ± 0.043 ^b	2.22 ± 0.307 ^b	Nd	1.65 ± 0.254 ^b	0.33 ± 0.005 ^a	0.45 ± 0.010 ^a	11.09 ± 0.03-1 ^{bc}
A3	44.9 ± 2.24 ^a	1057 ± 2.77 ^c	1740 ± 4.81 ^a	22.9 ± 5.75 ^b	17.3 ± 3.19 ^{ab}	15.4 ± 0.461 ^{ab}	0.30 ± 0.004 ^b	0.74 ± 0.006 ^b	3.85 ± 0.412 ^{ab}	Nd	2.87 ± 0.457 ^{ab}	0.36 ± 0.014 ^a	0.46 ± 0.018 ^a	14.6 ± 0.84-6 ^{ab}
B1	26.4 ± 0.777 ^b	1063 ± 5.18 ^c	1138 ± 0.073 ^d	30.6 ± 0.507 ^{ab}	10.2 ± 0.221 ^b	7.00 ± 0.169 ^c	0.30 ± 0.007 ^b	3.85 ± 0.018 ^a	2.39 ± 0.738 ^b	Nd	2.63 ± 0.189 ^b	0.31 ± 0.008 ^a	0.45 ± 0.013 ^a	17.5 ± 1.26 ^a
Cake														
A1	42.9 ± 1.04 ^a	2473 ± 4.02 ^a	2872 ± 2.19 ^b	56.5 ± 1.21 ^a	30.2 ± 3.78 ^a	26.9 ± 5.66 ^a	0.47 ± 0.004 ^a	2.44 ± 0.291 ^b	9.67 ± 0.345 ^a	Nd	5.22 ± 0.218 ^a	0.35 ± 0.028 ^a	0.52 ± 0.076 ^a	14.5 ± 2.25 ^a
A2	48.5 ± 1.79 ^a	1852 ± 7.20 ^c	2673 ± 4.47 ^c	57.2 ± 2.65 ^a	35.7 ± 1.11 ^a	30.5 ± 3.94 ^a	0.47 ± 0.028 ^a	1.16 ± 0.097 ^b	3.93 ± 1.01 ^b	Nd	4.79 ± 0.008 ^a	0.34 ± 0.010 ^a	0.57 ± 0.095 ^a	13.5 ± 1.85 ^a
A3	46.1 ± 3.90 ^a	1742 ± 5.91 ^c	3555 ± 5.65 ^a	59.3 ± 5.46 ^a	32.6 ± 1.25 ^a	23.9 ± 3.17 ^{ab}	0.44 ± 0.085 ^a	1.66 ± 0.029 ^b	8.21 ± 0.095 ^{ab}	Nd	6.12 ± 0.916 ^a	0.36 ± 0.002 ^a	0.60 ± 0.071 ^a	19.1 ± 0.898 ^a
B1	28.7 ± 3.21 ^b	1583 ± 5.46 ^d	1872 ± 2.49 ^d	42.8 ± 7.61 ^a	14.9 ± 1.54 ^b	10.3 ± 1.51 ^b	0.40 ± 0.037 ^a	5.58 ± 0.971 ^a	8.87 ± 2.29 ^{ab}	Nd	5.09 ± 0.349 ^a	0.36 ± 0.001 ^a	0.60 ± 0.021 ^a	18.1 ± 1.29 ^a

A1, A2, A3: sapucaia nuts from Piauí; B1: sapucaia nut from Minas Gerais. Mean ± S.D. (n = 3). Different letters in the same column presented difference at a significant level (Tukey test, $p < 0.05$). Nd: not detected.

also found higher values for Mn ($64.3 \mu\text{g g}^{-1}$) and Zn ($39.3 \mu\text{g g}^{-1}$) but reported lower values for Cu ($9.2 \mu\text{g g}^{-1}$).

It has been reported that chromium (Cr) and nickel (Ni) are related to risk reduction for chronic diseases such as osteoporosis and diabetes. They are also beneficial to central nervous system function and bone development (Nielsen, 2014). The RDA established for Cr is $35 \mu\text{g/d}$ and $25 \mu\text{g/d}$, for male and female, respectively; and for Ni is 1 mg/day (FAO/WHO, 2001; Food & Nutrition Board, 2004). Carvalho et al. (2012) reported higher values for Ni ($5 \mu\text{g g}^{-1}$) in sapucaia nuts when compared to the present study, and the Cr levels were not detected. The concentration of sodium (Na) in samples of sapucaia nut and cake were low (mean of 3.35 and $7.67 \mu\text{g g}^{-1}$, respectively) considering that the international bodies of nutrition recommend an intake for Na of 1.5 g/day . Denadai et al. (2007) also detected low levels of Na ($5.8 \mu\text{g g}^{-1}$) in samples of sapucaia nut.

Toxic and non-essential elements such as aluminum (Al), cadmium (Cd), lead (Pb) and tin (Sn) were also identified in sapucaia nut and cake. According to the Agency for Toxic Substances and Disease Registry, the limit of Al is 1 mg kg^{-1} of weight/day (ATSDR, 2017). The maximum level recommended for Cd is $0.05 \text{ mg kg}^{-1}/\text{day}$ and for Pb in vegetables is 0.2 mg kg^{-1} (Codex Alimentarius, 1996). There is no recommended limit for the ingestion of Sn. In this study none of the samples exceeded the values considered as toxic by the Agency for Toxic Substances and Disease Registry and Codex Alimentarius Commission. Heavy metals in food derive from several sources such as soil polluted by heavy metals (where they may stay for longer periods because of their higher affinity with organic matter), pesticides and fertilizers (Zang, Wang, Ashraf, Qiu, & Ali, 2017).

3.3. Physicochemical analysis of the sapucaia nut oil

Table 3 shows some characteristics of identity and quality of the sapucaia nut oil. The refraction index (between 1.469 and 1.470 at 20°C) indicated that the oils are rich in unsaturated fatty acids. Density at 20°C ranged between 0.88 and 0.92 g/cm^3 and the Lovibond color (30.0–40.0 for yellow units and 0.8–1.0 for red units) indicated a yellow color for the oil. The oil showed a peroxide value in accordance with the Brazilian National Health Surveillance Agency (ANVISA) for unrefined cold-pressed oils (maximum of 15 mEq kg^{-1} for virgin oils) (Brazil, 2005). The peroxide value and specific extinction (232 and 270 nm) indicated adequate oxidative quality for the oil. The Oxidative Stability Index (OSI) of the sapucaia nut oil was between 8.57 and 12.95 h, which reflects its fatty acid composition and the presence of antioxidant compounds in the oil. The OSI was significantly ($p < 0.05$) higher for samples A1, A2 and A3 when compared to sample B1, due to high oleic acid content. Costa and Jorge (2012) found a higher oxidative stability (24.89 h) of the sapucaia oil extracted by cold pressing at 100°C . Teixeira et al. (2017) reported an OSI of 7.18 h of the sapucaia nut oil extracted by Soxhlet, and 13 h for sapucaia nut oil extracted by Blich & Dyer. Teixeira et al. (2018) reported an OSI of 9.29 h (110°C) of the sapucaia nut oil extracted using supercritical CO_2 . The results obtained in this work were similar for the induction period reported for Brazil nut (8.24 h), canola (8.63 h), hazelnut (8.88 h), pecan (9.87), corn (9.96 h), and soybean (12.0 h) oils (Castelo-Branco, Santana, Di-Sarli, Freitas, & Torres, 2016). Although there is no established minimum value for the induction period for vegetable oils, the longer the induction period, the greater the oxidative stability of the oil.

3.4. Fatty acid composition, content of tocopherols and phytosterols in samples of sapucaia oil

Ten different fatty acid were identified in the sapucaia nut oil. Oleic acid was predominant in the A1, A2 and A3 samples (39.7 to 45.4%), and linoleic acid in the sample B1 (46.6%). Among the saturated fatty acids, palmitic and stearic acids were the major fatty acids (11.1–15.2% and 7.50–8.44%, respectively). The sapucaia nut oil is a source of

Table 3
Physicochemical characteristics of sapucaia nut oil.

Parameter	Samples			
	A1	A2	A3	B1
Refraction index (20 °C)	1.469 ± 0.001 ^a	1.469 ± 0.001 ^a	1.469 ± 0.001 ^a	1.470 ± 0.001 ^a
Density (g/cm ³)	0.91 ± 0.003 ^a	0.91 ± 0.001 ^a	0.92 ± 0.006 ^a	0.88 ± 0.002 ^b
Smoke point (°C)	209.0 ± 2.83 ^b	210.5 ± 0.70 ^b	210.0 ± 2.83 ^b	217.5 ± 1.77 ^a
Acid value (g oleic acid 100 g ⁻¹)	0.19 ± 0.02 ^b	0.19 ± 0.01 ^b	0.20 ± 0.01 ^b	0.34 ± 0.02 ^a
Peroxide value (mEq Kg ⁻¹)	1.47 ± 0.11 ^b	1.53 ± 0.44 ^b	1.60 ± 0.02 ^b	2.52 ± 0.12 ^a
Specific extinction – 232 nm (%)	0.74 ± 0.02 ^b	0.78 ± 0.02 ^b	0.73 ± 0.02 ^b	1.46 ± 0.02 ^a
Specific extinction – 270 nm (%)	0.03 ± 0.01 ^b	0.07 ± 0.02 ^b	0.02 ± 0.01 ^b	0.39 ± 0.01 ^a
p-Anisidine value	Nd	Nd	Nd	Nd
Oxidative stability index – OSI (h)	12.95 ± 0.06 ^a	12.88 ± 0.07 ^a	12.75 ± 0.22 ^a	8.57 ± 0.23 ^b
Color Lovibond (5¼ in) [*]	30.0 Y/1.0 R	40.0 Y/1.0 R	30.0 Y/1.0 R	40.0 Y/0.8 R

A1, A2, A3: sapucaia nuts from Piauí; B1: sapucaia nut from Minas Gerais. Mean ± S.D. (n = 3). Different letters in the same line presented difference at a significant level (Tukey test, *p* < 0.05). Nd = not detected.

* Y = yellow, R = red.

unsaturated fatty acids, since it contained an unsaturated ratio between 0.27 and 0.30%. The fatty acid composition of the sapucaia nut oil obtained for samples from Piauí were similar to the results reported by Vallilo et al. (1999) and Costa and Jorge (2012). Similar results obtained for samples from Minas Gerais were reported for Teixeira et al. (2017) and Teixeira et al. (2018) who found oleic acids as major component in sapucaia nut oil, followed by linoleic and palmitic acids. It has been reported that the substitution of dietary saturated fat by oleic acid and/or polyunsaturated fatty acids may reduce the cardiovascular risk, mainly by reducing blood cholesterol levels (Nishi et al., 2014).

The γ-tocopherol was the main tocopherol identified in sapucaia nut oil, representing from 88 to 95% of the total content of these compounds. Low concentrations of α- and δ-tocopherol were also identified.

Table 4
Fatty acid composition, tocopherols and phytosterols of sapucaia oil.

Parameter	Samples			
	A1	A2	A3	B1
Fatty acids (%)				
Myristic acid (C 14:0)	0.10	0.09	0.09	0.07
Palmitic acid (C 16:0)	15.2	12.9	14.4	11.1
Palmitoleic acid (C 16:1)	0.35	0.21	0.21	0.24
Margaric acid (C 17:0)	0.08	0.08	0.07	0.07
Stearic acid (C 18:0)	7.93	8.44	7.91	7.50
Oleic acid (C18:1 n9c)	39.7	45.4	44.4	40.0
Linoleic acid (C 18:2 n6c)	40.0	32.3	32.2	46.6
γ-linolenic acid (C 18:3)	0.27	0.26	0.27	0.22
α-Linolenic acid (C 18:3 n3c)	0.35	0.26	0.32	0.22
Cis-11-eicosenoic acid (C 20:1)	0.07	0.07	0.08	0.06
Total saturated fatty acids	23.3	21.5	22.5	18.8
Total monounsaturated fatty acids	40.1	45.7	44.7	34.2
Total polyunsaturated fatty acids	36.6	32.8	32.7	47.0
S/U [*]	0.30	0.27	0.29	0.23
Tocopherols (mg 100 g⁻¹)				
α-Tocopherol	1.12 ± 0.03 ^b	0.75 ± 0.09 ^b	1.10 ± 0.01 ^b	2.24 ± 0.16 ^a
β-Tocopherol	Nd < 0.02 [*]	Nd < 0.02 [*]	Nd < 0.02 [*]	Nd < 0.02 [*]
γ-Tocopherol	28.5 ± 0.49 ^a	21.2 ± 0.13 ^b	26.8 ± 0.83 ^a	19.2 ± 0.17 ^b
δ-Tocopherol	0.28 ± 0.02 ^b	0.26 ± 0.02 ^b	0.41 ± 0.01 ^a	0.43 ± 0.02 ^a
Total tocopherols	29.9 ± 0.54 ^a	22.2 ± 0.24 ^b	28.3 ± 0.85 ^a	21.8 ± 0.32 ^b
Vitamin E (UI 100 g ⁻¹)	6	4	5	5
Phytosterols (mg 100 g⁻¹)				
β-sitosterol	93.7 ± 0.78 ^c	101.3 ± 1.34 ^b	92.8 ± 1.49 ^c	193.9 ± 1.84 ^a
Stigmasterol	11.2 ± 0.19 ^b	11.8 ± 0.36 ^b	9.92 ± 0.42 ^c	13.2 ± 0.09 ^a
Campesterol	8.60 ± 0.49 ^{bc}	9.11 ± 0.32 ^{ab}	8.42 ± 0.28 ^{bc}	9.63 ± 0.26 ^a
Brassicasterol	Nd < 1.50	Nd < 1.50	Nd < 1.50	Nd < 1.50

A1, A2, A3: sapucaia nuts from Piauí; B1: sapucaia nut from Minas Gerais. Fatty acid (n = 1). Mean ± S.D. (n = 3). Different letters in the same line presented difference at a significant level (Tukey test, *p* < 0.05). Nd = not detected.

* S/U: Saturated to unsaturated ratio.

Table 5
Triacylglyceride (TAG) composition of sapucaia oil.

TAGs	ECN	Samples (%)			
		A1	A2	A3	B1
LLL	42	6.40 ± 0.35 ^b	5.91 ± 0.01 ^b	6.00 ± 0.15 ^b	12.6 ± 0.20 ^a
LLO	44	12.9 ± 0.43 ^b	12.9 ± 0.68 ^b	11.6 ± 0.03 ^b	20.7 ± 0.14 ^a
PLL	44	6.92 ± 0.32 ^b	6.41 ± 0.04 ^b	6.42 ± 0.18 ^b	10.0 ± 0.04 ^a
LOO	46	14.1 ± 0.11 ^a	14.9 ± 0.98 ^a	14.7 ± 0.44 ^a	14.1 ± 0.05 ^a
PLO	46	16.5 ± 0.19 ^a	16.4 ± 0.50 ^a	16.6 ± 0.41 ^a	16.8 ± 0.05 ^a
PPL	46	2.51 ± 0.34 ^a	1.33 ± 0.26 ^b	1.92 ± 0.21 ^{ab}	1.73 ± 0.03 ^{ab}
OOO	48	11.93 ± 0.59 ^a	12.5 ± 0.38 ^a	13.3 ± 0.48 ^a	6.85 ± 0.13 ^b
POO	48	15.6 ± 0.48 ^b	16.7 ± 0.47 ^{ab}	17.4 ± 0.39 ^a	10.9 ± 0.21 ^c
PPO	48	2.43 ± 0.10 ^{ab}	1.52 ± 0.18 ^{ab}	1.52 ± 0.01 ^b	2.52 ± 0.46 ^a
PPP	48	1.91 ± 0.08 ^a	1.81 ± 0.41 ^a	1.91 ± 0.24 ^a	0.60 ± 0.01 ^b
SOO	50	4.95 ± 0.21 ^a	5.69 ± 0.41 ^a	5.92 ± 0.36 ^a	2.51 ± 0.03 ^b
SSL	50	2.74 ± 0.05 ^a	2.91 ± 0.44 ^a	2.50 ± 0.41 ^a	0.92 ± 0.01 ^b
SOS	52	0.64 ± 0.02 ^{ab}	1.00 ± 0.22 ^a	0.70 ± 0.16 ^{ab}	0.31 ± 0.01 ^b

A1, A2, A3: sapucaia nuts from Piauí; B1: sapucaia nut from Minas Gerais. Mean ± S.D. (n = 3). Different letters in the same line presented difference at a significant level (Tukey test, p < 0.05). O: oleic acid; L: linoleic acid, P: palmitic acid; S: stearic acid. ECN = equivalent carbon number.

The chemical compounds identified in the sapucaia nut oil such as tocopherols, phytosterols, and oleic and linoleic acids when present in the diet can contribute to the maintenance of a healthy life. Studies have associated a nut rich diet to a reduced incidence of coronary heart diseases, and to other beneficial effects on hypertension, cancer, inflammation and cholesterol-lowering effect. The reported effects may be associated with a synergistic interaction of the many bioactive constituents of nuts (tocopherols, phytosterols, oleic and linoleic acids) which may all favorably influence human physiology (Aune et al., 2016; Cardoso et al., 2017; Nishi et al., 2014; Yang, 2009).

3.5. Triacylglyceride (TAGs) profile of sapucaia oil

The TAGs dilinoleo olein (LLO), palmito linoleo olein (PLO), linoleo diolein (LOO), palmito diolein (POO), triolein (OOO), palmito dilinoleo (PLL) and trilinoleo (LLL) were the major triacylglycerols in sapucaia

Table 6
Nutritional composition of sapucaia, Brazil and cashew nuts.

Parameter	Sapucaia nuts ^a	Brazil nuts	Cashew nuts
Energy value (kcal 100 g ⁻¹)	534–617	643–710.6 ^{AB}	570–600.23 ^{AC}
Protein (g 100 g ⁻¹)	15.8–19.5	14.5–18.58 ^{AB}	18.5–22.67 ^{AC}
Carbohydrate (g 100 g ⁻¹)	4.90–6.50	7.60–15.1 ^{AB}	19.86–29.1 ^{AC}
Total dietary fiber (g 100 g ⁻¹)	16.5–22.6	7.9 ^A	3.7–3.92 ^{AC}
Total lipids (g 100 g ⁻¹)	47.9–60.8	63.5–67.20 ^{AB}	46.3–47.79 ^{AC}
Ca (μg g ⁻¹)	1057–1249	1460–1800 ^{AB}	330.0–640.5 ^{AC}
Mg (μg g ⁻¹)	1138–1740	3250–3650 ^{AB}	2370–2770.0 ^{AC}
Mn (μg g ⁻¹)	22.9–41.4	15.9–16.5 ^{AB}	11.0 ^A
Zn (μg g ⁻¹)	10.2–20.9	35.1–42.0 ^{AB}	47.0–49.8 ^{AC}
Cu (μg g ⁻¹)	7.00–16.9	14.0–17.9 ^{AB}	19.2 ^A
Se (μg g ⁻¹)	26.4–46.9	11.48–36.1 ^{BE}	0.0102 ^C
Total saturated fatty acids (mg 100 g ⁻¹)	18.8–23.3	24.5–26.7 ^{DE}	20.20–20.22 ^{DE}
Total monounsaturated fatty acids (mg 100 g ⁻¹)	34.2–45.7	29.97–38.6 ^{DE}	56.87–58.13 ^{DE}
Total polyunsaturated fatty acids (mg 100 g ⁻¹)	32.7–47.0	36.8–43.0 ^{DE}	21.03–22.22 ^{DE}
α-Tocopherol (mg 100 g ⁻¹)	0.75–2.24	5.44–8.29 ^{DE}	0.36–1.48 ^{DE}
γ-Tocopherol (mg 100 g ⁻¹)	19.2–28.5	11.62–14.63 ^{DE}	5.56–5.72 ^{DE}
β-sitosterol (mg 100 g ⁻¹)	92.8–194	62.7–132.54 ^{DE}	111.0–176.8 ^{DE}
Stigmasterol (mg 100 g ⁻¹)	9.92–13.2	5.5–5.7 ^{DE}	0.7–11.67 ^{DE}
Campesterol (mg 100 g ⁻¹)	8.42–9.63	1.2–2.69 ^{DE}	8.6–10.53 ^{DE}

^a Samples of sapucaia nuts from Piauí and Minas Gerais (A1, A2, A3 and B1). ^ATACO (2011), ^BSantos et al. (2013), ^CSousa, Fernandes, Alves, Freitas, and Naves (2011), ^DYang (2009), ^ERobbins et al. (2011).

nut oil (Table 5). The triacylglycerols found in sapucaia nut oil predominantly contained oleic and linoleic acids and less palmitic and stearic acids, which is compatible with the fatty acid profile found in the samples. The TAG composition of the sapucaia nut oil obtained in this study is similar to results reported by Teixeira et al. (2018).

3.6. Nutritional composition and lipid profile of the sapucaia nut compared with Brazil nut and cashew

The Brazil nut and cashew are nuts native from Brazil and known worldwide as foods rich in bioactive compounds. The sapucaia nut, despite its significant nutritional potential, is still an underutilized food material. Table 6 shows that the sapucaia nut presents a lower content of oil than Brazil nut and higher than cashew nut. On the other hand, the protein content is similar to Brazil nuts and cashew nuts. The dietary fiber content of the sapucaia nut is higher than that reported in the literature for Brazil nuts and cashew nuts.

Sapucaia and Brazil nuts are rich in Se. In sapucaia nuts, the content of Ca is higher than cashew nuts and lower than Brazil nuts. The content of Mg and Zn are higher in Brazil and cashew nuts when compared with sapucaia nuts. On the other hand, the Mn content is higher in sapucaia nut when compared to Brazil and cashew nuts and the Cu content is similar for all nuts.

The sapucaia nut presents a lower concentration of saturated fatty acids than Brazil nuts and cashew nuts. The content of mono-unsaturated and polyunsaturated fatty acids of the sapucaia nut are similar to Brazil nuts. On the other hand cashew nuts have a higher content of monounsaturated fatty acids and lower for polyunsaturated fatty acids when compared to the sapucaia nut. The sapucaia nut presents higher content of γ-tocopherol when compared to Brazil nuts and cashews nuts.

These results showed that sapucaia nuts presents potential health benefits and may be used to improve the diet of the Brazilian population. Its nutritional and phytochemical composition is comparable and, in some cases, better than other representative Brazilian native nuts such as Brazil nuts and cashews.

4. Conclusions

The evaluation of the nutritional composition and lipid profile of sapucaia nut and its by-products showed a significant nutritional potential for these raw materials. Significant differences were observed for the nutritional composition of the sapucaia nuts, cakes and shells between samples obtained from different regions. This underutilized nut present desirable nutritional and phytochemical composition, comparable to other representative native Brazil nuts. The high protein, dietary fiber and minerals in sapucaia nut and cake indicated that this raw material may be used as a nutrient-rich ingredient in different applications. The selenium content may limit the direct consumption of the nuts but, on the other hand, the ingestion of Se in regions of high Hg exposure may be important because of the protective effect of Se against Hg. As the sapucaia nut oil is an excellent source of bioactive compounds, such as oleic and linoleic acids, γ-tocopherol and β-sitosterol, it may contribute to the maintenance of human health. The results of this work represent a step forward for knowledge of this underutilized raw material. The data obtained may be used to stimulate the local production and consumption of this type of nut, leading to its economic development. In addition, the nuts may be used as a promising raw material for the food industry.

Conflict of interest

The authors confirm that there are no conflicts of interest associated with this publication.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.foodres.2018.03.028>.

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