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Characterization of Oil Extracted from Buriti Fruit (*Mauritia flexuosa*) Grown in the Brazilian Amazon Region

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Abstract Five samples of buriti oil from industrial and artisanal suppliers were characterized in terms of nutritional quality parameters (nutraceutical levels) and acidity. As a first screening, each sample was analyzed by titration, spectrophotometry and an HPLC method, and the results were compared. As expected, artisanal samples showed lower acidity and higher levels of carotenes and tocols (tocopherols and tocotrienols). A blend of industrial and artisanal samples in suitable proportions was completely characterized in terms of analytical and physico-chemical properties, i.e., fatty acid composition, iodine value, partial and total acylglycerol contents, refractive index (40 °C), saponification value, unsaponifiable matter, acidity (expressed as % of oleic acid), peroxide value, phosphorus content, oil stability index, tocol and carotene concentrations. The results of the present study showed that buriti oil is a valuable source of monounsaturated fatty acids, and vitamins A and E. No previous work in the literature has analyzed buriti oil to this extent. The chromatographic method using HPLC was effective in qualifying and quantifying tocopherols, tocotrienols and carotenes.

Keywords Buriti oil · Carotenes · Tocopherol · Tocotrienol · HPLC

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Introduction

Buriti (*Mauritia flexuosa*) is a palm tree native to Brazil that normally grows in permanently or periodically flooded areas along rivers, forests and savannas (north-central Brazil and Venezuela) [1]. Its fruit has a hard, red and squamous shell that covers a soft and oily pulp, with color variations ranging from dark yellow to reddish (after complete ripening) [2]. According to Ferreira [1], it is possible to extract around 45 kg of buriti oil from 1,000 kg of ripened fruits.

Previous work [2, 3] showed that buriti oil has a high concentration of monounsaturated fatty acids (MUFA). The MUFAs in buriti oil are higher than those in olive and Brazil nut oils, which are know to have high-quality nutritional oils with blood cholesterol-lowering properties. In addition, the low concentration of polyunsaturated fatty acids (PUFA) gives buriti oil a high oxidative stability.

The nutraceutical fraction of buriti oil consists of tocopherols [2, 3] and carotenes [2–7], which have nutritional importance as antioxidants [8, 9] and pro-vitamin A [5, 10], respectively, is well-recognized. In fact, Mariath et al. [5] demonstrated the effectiveness of homemade buriti candy (from the pulp) in the treatment and prevention of xerophthalmia in children of Northeastern Brazil. There are also indications that oral supplementation of vitamin A to newborn babies of underdeveloped regions can reduce

mortality by at least 15% [10]. Thus, buriti oil seems to be a viable alternative for increasing vitamin A ingestion, for example, blended with other oils for culinary use. Previous studies reported concentrations of 800 mg/kg of total tocopherols [2] in pressed buriti oil and 19,300 mg/kg of total tocopherols in buriti oil extracted by the supercritical means [3]. It should be emphasized that no previous work has analyzed the fractional contents (α-, β-, δ- and γ-) of tocopherols and tocotrienols in buriti oil.

Composition and, consequently, the nutritional value of crude buriti oil can vary with season and the extraction process. Also, inappropriate post-harvest handling of buriti fruits can lead to high oil acidity levels (hydrolysis of triacylglycerols), which reduces the oxidative stability, decreases the smoke point, and induces vitamin oxidation.

We screened five buriti oil samples from industrial and artisanal suppliers on the basis of acidity and total carotene and tocopherol contents. The resulting data allowed us to formulate a suitable blend of industrial and artisanal samples that was completely characterized.

Materials and Methods

Three samples of buriti oil (E1, E2 and E3) were acquired from industrial suppliers and two artisanal samples (A1 and A2) were purchased at a popular market Araguaia River region (Tocantins, Brazil). Tocantins is an inland state of north-central Brazil, forming the boundary between the Amazon Rainforest and the coastal savanna. Quality parameters, including acidity, carotenes and tocopherols of the oil were determined.

Acidity

Acidity was determined according to method 2201 of IUPAC [11] by using an automatic titrator (808 Titrando, METROHM).

Carotenes

Determined by using UV/VIS-spectrophotometer (Lambda 40, Perkin Elmer), as suggested by PORIM [12]. The calibration curve was done using a pure standard of β-carotene (Fluka).

Tocopherol Content

Chromatographic analyses were carried out on a Shimadzu equipment series LC-20AT instrument (Kyoto, Japan) equipped with a quaternary pump, a manual sampler, a degasser, and a SPD-M20A spectrophotometric detector model, which was set at 292 nm. Chromatographic separations of the compounds were achieved at 30 °C, using a normal-phase column CLC-SIL (Shimadzu, 250 × 4.6 mm i.d.; 5 μm particle size) with a guard column (10 × 4.6 mm) purchased from Shimadzu (Kyoto, Japan). The column was equilibrated and eluted under gradient conditions using a flow rate of 1.0 mL min^{-1}. The gradient condition was: 0–7 min 99.5% hexane and 0.5% isopropanol; 7–12 min linear gradient 0.5–1% isopropanol; 12–20 99.0% hexane and 1.0% isopropanol; 20–23 min reconditioning step of the column was 0.5% isopropanol isocratic for 7 min. The chromatographic run time for each analysis was 30 min. Samples were dissolved in hexane and aliquots of 20 μl were injected into HPLC system. A validation chromatographic run included a set of calibration samples assayed in triplicate and quality control samples at three levels in triplicate. The standard calibration curves for known amounts of tocopherols, ranging from 2.50 to 500.00 μg mL^{-1}, were linear ($R > 0.999$).

Oil Blends

After the initial screening, the industrial sample E2, whose quality parameters most resembled those of the artisanal samples, i.e., low acidity and high nutraceutical value (Table 1) was chosen for the blended oil. E2 had the lowest acidity value and the highest contents of nutraceutical (total tocopherols and carotenes) of the industrial oil samples. The blended oil consisted of 15 kg of E2 and 3 kg from each of the artisanal oils for a total of 21 kg. Because of the limited production of artisanal oils, a more economical product can be made from a blended oil and still retain its high quality parameters. The blended oil was then characterized for quality.

Table 1 Acidity (expressed as % oleic acid), total carotenes (mg kg^{-1}), α- and β- tocopherols (mg kg^{-1}) for artisanal and industrial samples of buriti oil

	E1[a]	E2[a]	E3[a]	A1[b]	A2[b]
Acidity	6.22 ± 0.01	2.01 ± 0.06	4.14 ± 0.03	1.77 ± 0.02	1.14 ± 0.02
Total carotenes	252 ± 9	564 ± 16	664 ± 19	1890 ± 112	1661 ± 65
α-Tocopherol	367 ± 22	523 ± 9	329 ± 7	1343 ± 6	1099 ± 9
β-Tocopherol	213 ± 4	269 ± 4	683 ± 23	647 ± 2	918 ± 10

[a] Samples from industrial suppliers

[b] Samples from artisanal suppliers

Blend Characterization

Separation of Acylglycerols

Triacylglycerols (TAG), diacylglycerols (DAG), monoacylglycerol (MAG) and free fatty acids (FFA) were separated by size exclusion chromatography according to the AOCS official method Cd 11c-93 [13]. Samples were dissolved to 1% in tetrahydrofuran (THF), and eluted using THF at a flow rate of 1 mL min^{-1} in a size-exclusion high performance liquid chromatography system (Perkin Elmer 250, USA) equipped with a refractive index detector (Sicon Analytic, Germany) and a size exclusion column (Jordi Gel DVB 300 × 7.8 mm, 500 and 100 Å, USA).

Fatty Acid Profile

The blended oil was analyzed by gas chromatography for the fatty acid methyl esters to determine the fatty acid composition, according to the official method (1–62) of the AOCS [13]. Samples were prepared in the form of fatty acid methyl esters according to the official method (2–66) of the AOCS [13]. Analytical conditions: chromatograph, CGC Agilent 6850 GC System (USA); column, DB-23, Agilent (50% Cyanopropyl-methylpolysiloxane), 60 m × 0.25 mm × 0.25 μm; helium as the carrier gas at a rate of 1.0 mL min^{-1}; injection temperature of 250 °C; column temperature of 110 °C for 5 min, 110–215 °C (rate of increase 5 °C min^{-1}), 215 °C for 24 min; detection temperature of 280 °C. The fatty acid methyl esters were identified by comparison with external standards purchased from Nu Check Inc. (Elysian, IL). The quantification was accomplished by internal normalization.

Physicochemical Analysis

The following physicochemical properties of the blended oil were determined: refractive index (RI) at 40 °C, saponification value (SV), iodine value (IV), unsaponifiable matter (UM), peroxide value (PV) and phosphorus content according to AOCS official methods [13]: Cc 7-25, Cd 3a-94, Cd 1c-85, Ca 6a-40, Cd 8-53 and Ca 12-55, respectively. Oil color was determined by a Lovibond Tintometer according to AOCS official method Cc 13e-92 [13], using a 1-in. cell. All analyses were done in triplicate.

Density and Viscosity

Densities and viscosities of buriti oil are already reported in the literature, and the temperature-dependence of these two properties can be found in Ceriani et al. [6].

Oil Stability Index

The oxidative stability was measured according to the AOCS method Cd 12b-92 [13], in OSI at 110 °C and a flow rate of 9.0 air L h^{-1}, using a sample of 5 g.

Carotene and Tocopherol Contents

The contents of α-, β-, γ- and δ-tocopherols and α-, γ- and δ-tocotrienols were determined by HPLC as described, using α-, β-, γ- and δ-tocopherol standards (Calbiochem, USA). Carotene was characterized by the same methodology, using β-carotene (Fluka, Germany) as pure standard. The β-carotene nominal standard was analyzed using a 6-min run, with an isocratic mobile phase of hexane and isopropanol (99.5:0.5, v/v). The standard calibration curve for known amounts of β-carotene, ranging from 0.50 to 500.00 μg mL^{-1}, was linear ($R > 0.999$).

Tocotrienols were quantified based on the area of tocopherol homologues. The β-carotene content was also determined by UV/VIS-spectrophotometry (described above) for comparison purposes.

Results and Discussion

A large variability prevailed in the buriti oil samples (Table 1). Artisanal samples had better quality parameters. Acidity values, total carotene and total tocopherol contents of the industrial samples ranged from 2.01 to 6.22%, 252 to 664 mg kg^{-1}, and 580 to 1012 mg kg^{-1}, respectively. Sample E2 had an acidity value significantly lower than the other industrial samples, intermediate concentrations of total carotene and β-tocopherol, and the highest content of α-tocopherol. Based on these observations, E2 was selected as a blend component.

The artisanal A1 and A2 samples had 1,890 and 1,661 mg kg^{-1} total carotenes, and 1,990 and 2,017 mg kg^{-1} tocopherols (α and β), respectively. As can be seen, A1 and A2 carotene contents are higher than those found in fruits traditionally recognized as good sources of vitamin A, such as the guava, pitanga, papaya and passion fruit [4]. A1 and A2 have α- and β-tocopherol contents within the same range as wheat germ oil [14].

Blended Buriti Oil

The class of acylglycerols and fatty acid profile of blended buriti oil is shown in Table 2. Blended buriti oil is composed essentially of TAG (91.4%), but relevant levels of partial acylglycerols (MAG and DAG) were also detected (0.9 and 4.1%, respectively). The FFA concentration of blended buriti oil determined by size exclusion

Table 2 Total and partial acylglycerols and fatty acid profile of buriti oil (blend)

Acylglycerols		(% mass)
TAG		91.9
DAG		4.1
MAG		0.9
FFA		3.1
Fatty acids	Trivial name (abbreviation)	(% mass)
C12:0	Lauric (L)	0.03
C14:0	Myristic (M)	0.08
C16:0	Palmitic (P)	16.78
C16:1	Palmitoleic (Po)	0.32
C17:0	Margaric (Mg)	0.08
C17:1	Margaroleic (Mo)	0.07
C18:0	Stearic (S)	1.77
C18:1	Oleic (O)	74.06
C18:2	Linoleic (Li)	4.94
C18:3	Linolenic (Ln)	1.04
C20:0	Arachidic (A)	0.12
C20:1	Gadoleic (Ga)	0.53
C22:0	Behenic (Be)	0.09
C24:0	Lignoceric (Lg)	0.09

Table 3 Blend of buriti oil physical–chemical characteristics

Color (red units)	60.0 ± 0.00
Color (yellow units)	9.00 ± 1.7
Acidity (% as oleic acid)	3.12 ± 0.01
Iodine value (g of I·100 g of oil^{-1})	74.64 ± 0.9
Saponification value (mg of KOH·g of oil^{-1})	192.88 ± 19.53
Refractive index (40 °C)	1.4610 ± 0.0
Unsaponifiable matter (%)	0.5 ± 0.0
Phosphorus (mg kg^{-1})	1.0 ± 0.0
Peroxide value (mequiv kg^{-1})	14.2 ± 0.1
Oxidative stability by Rancimat (h)	18.3 ± 0.11
Density (mg mL^{-1}) 25 °C	0.909[a]
Viscosity (mPa s) 25 °C	62.461[a]
Tocopherols (mg kg^{-1})	
α-Tocopherol	614 ± 5
β-Tocopherol	687 ± 8
γ-Tocopherol	50 ± 2
γ-Tocotrienol	12 ± 1
δ-Tocopherol	136 ± 0
δ-Tocotrienol	18 ± 1
Total tocopherol	1517 ± 13
Carotenes (mg kg^{-1})	
Spectrophotometry	1003 ± 20
HPLC	900 ± 10

[a] Referring to Ceriani et al. [6]

chromatography was 3.1%. The acidity value obtained by titration [11] and expressed as % of oleic acid was coincident (Table 3). This is not surprising, if one considers the relevance of oleic acid in the fatty acid profile of buriti oil (Table 2). Although 3.1% acidity can be considered high, similar values are commonly found in other pulp oils, such as crude palm oil [15].

As shown in Table 2, palmitic, oleic and linoleic acids are the most abundant fatty acids present in buriti oil. Concerning fatty acid composition, buriti oil can be considered similar to olive oil [16]. Results shown in Table 2 are in accordance with previous works that present the fatty acid compositions of buriti oil [2, 6].

Table 3 shows a variety of physical and chemical characteristics of buriti oil. The color in terms of red and yellow units (60.0R + 9.00Y) is due to the presence of β-carotene. In comparison with palm oil [17], a rich source of carotenes, the blended buriti oil tested in this work has lower yellow and higher red unit values. The values reported by Tan et al. [17] for two palm oil samples are 3.2R + 19.5Y and 2.6R + 16.7Y. The iodine value (74.64 g of I·100 g^{-1} of oil), the refractive index (1.4610 at 40 °C) and the saponification value (192.88 mg of KOH·g^{-1} of oil) are within the range as that of olive oil [16].

The low phosphorus content of buriti oil (1 mg kg^{-1}) enables the purification by physical refining. As reported by O'Brien [16], one of the requirements for physical refining is that the phosphorus content of the oil should be lower than 5 mg kg^{-1} before the steam stripping step.

The buriti oil tested in this work was very stable to oxidation (18.3 h OSI), and can be considered as an oil with high oxidative stability [18], equivalent to olive oil. This result confirms that the high percentage of oleic acid contributes to the high oxidative stability; the remaining antioxidant activity is attributed to the minor components of the oil, the tocopherols and pigments [19].

Despite the high oxidative stability given by OSI (18.3 h), the blended buriti oil had a peroxide value of 14.2 mequiv kg^{-1}. Peroxide values of fresh oils are usually lower than 10 mequiv kg^{-1} [20]. However, Codex Alimentarius [21] establishes a peroxide value up to 15.0 mequiv/kg for cold pressed fats and virgin oils. Peroxides are the first and least stable oxidation products and easily decompose to more stable secondary products [22]. Therefore, peroxides are present in oils in the initial oxidation stage. The purchased oils were probably not fresh, and were at the initial stage of oxidation.

The unsaponifiable matter (0.5%) was found to be lower than the corresponding values for soybean (0.6–1.6) and corn oils (1.3–2.3), and within the same range as those of safflower (0.3–0.6) and palm oils (0.15–0.99) [16].

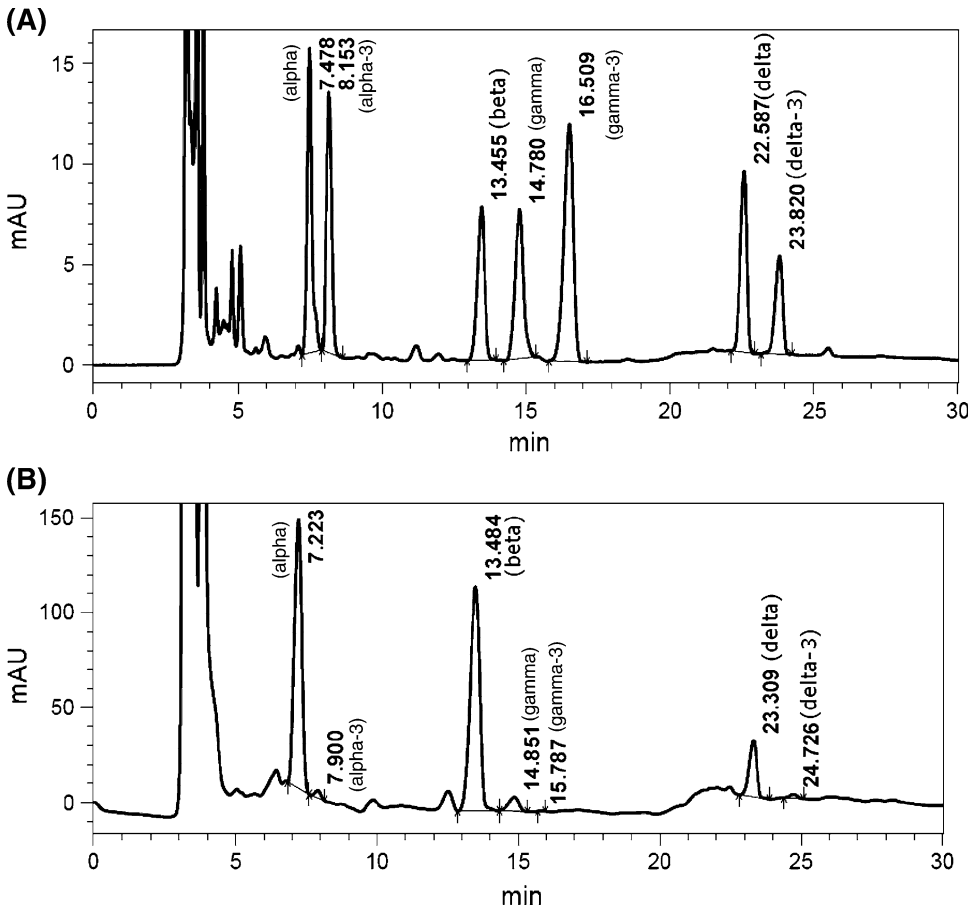

Fig. 1 HPLC chromatograms: qualitative standard (**a**) and blend (**b**). CLC-SIL column; CLC SIL(4) guard column; gradient mobile phase hexane–isopropanol; 1 mL min^{-1} flow; injected 20 μL; photo-diode array detection: 292 nm. The labels alpha, beta, gamma and delta correspond to α-tocopherol, β-tocopherol, γ-tocopherol, and δ-tocopherol, respectively

The blended buriti oil has all fractions of tocopherols: α-, β-, γ- and δ-. The most important tocopherols were α- and β- fractions. The β-tocopherol (687 mg kg^{-1}) appeared in the highest concentration, followed by α-tocopherol (614 mg kg^{-1}), δ-tocopherol (136 mg kg^{-1}) and γ-tocopherol (50 mg kg^{-1}). The blend also had high concentrations of γ- and δ-tocotrienol (12 and 18 mg kg^{-1}, respectively). The α-tocotrienol concentration was below the limit of quantification, so it could not be considered (Fig. 1).

Figure 1 shows representative chromatograms obtained from a qualitative standard sample (A) and a buriti oil sample (B), both were prepared in hexane. The chromatograms depicted in Fig. 1 were obtained according to the described chromatographic conditions. The selectivity and specificity for α-, β-, γ- and δ-tocopherols and α-, γ- and δ-tocotrienols is indicated by the sharp and symmetrical peaks, as well as the lack of significant interfering peaks from other oil components. Concomitantly with this analysis, it was also possible to quantify carotenes using HPLC combined with photodiode array detector. As one can see in Table 3, carotene concentration given by UV/VIS-spectrophotometry (1,003 mg kg^{-1}) was 10% higher than the value found by HPLC (900 mg kg^{-1}). This fact is in accordance with the ones reported in the literature [4], confirming that β-carotene is approximately 90% of the total carotenes of buriti oil. It is of note that UV/VIS-spectrophotometry is an easier and cheaper method than HPLC, and it seems to give reliable results. As pointed out by Wong et al. [23], the presence of tocopherols does not interfere in the UV/VIS-spectrophotometry analysis for carotenes. This statement is confirmed in this work, and also holds for HPLC analysis, in which tocopherols and carotenes have very different retention times.

Our analyses showed that the nutritional value of buriti oil is due to the high levels of oleic acid, carotenes and tocopherols. The first screening for quality properties revealed that important variability was found in industrial and artisanal samples. We believe that buriti oil has potential for culinary use in blends with other oils, thus, increasing the levels of vitamins A and E in the blends.

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