Original article

Murici, fruit from the Cerrado of Minas Gerais, Brazil: physical and physicochemical characteristics, and occurrence and concentration of carotenoids and vitamins

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Murici, fruit from the Cerrado of Minas Gerais, Brazil: physical and physicochemical characteristics, and occurrence and concentration of carotenoids and vitamins.

Abstract – Introduction. The Cerrado is the largest biome in the state of Minas Gerais, Brazil; it presents an enormous biodiversity represented by fruit species, highlighting murici (*Byrsonima verbascifolia*). The objective of our study was to investigate the physical and physicochemical characteristics, occurrence and concentration of carotenoids, vitamin C, vitamin E and folates in murici fruits from the Cerrado of Minas Gerais, Brazil. **Materials and methods**. Titratable acidity was determined by volumetric neutralization, pH by potentiometry, soluble solids by refractometry, moisture using an oven, ash using a muffle furnace, protein by the micro-Kjeldahl method, total dietary fiber by the non-enzymatic gravimetric method and lipids by Soxhlet extraction; carotenoids and vitamin C were analyzed by HPLC-DAD, and vitamin E and folates by HPLC with fluorescence detection. **Results and discussion**. The murici pulp contained 7.47 g·100 g^{-1} carbohydrates, 5.13 g·100 g^{-1} lipids, 13.58 g·100 g^{-1} fibers and 71.58 g·100 g^{-1} moisture. The pH, titratable acidity and soluble solids were 3.93, 0.77 g citric acid·100 g^{-1} and 10.73 °Brix, respectively. The results indicated that the fruit is an excellent source of fiber and vitamin C (27.24 mg·100 g^{-1}), a good source of vitamin E (1819.72 µg·100 g^{-1}) for children and a source for adults and pregnant women. Folates were not found in murici. **Conclusion**. Murici stands out with regard to its nutritional value and can contribute significantly to the supply of nutrients, especially fiber and vitamin C. Thus, consumption of these fruits should be encouraged among families residing in the Cerrado and in other regions of Brazil.

Brazil / *Byrsonima verbascifolia* / fruits / physicochemical properties / proximate composition / vitamin C / vitamin E / B vitamins

Le murici, fruit du Cerrado du Minas Gerais au Brésil : caractéristiques, physiques et physico-chimiques, occurrence et concentrations des carotenoïdes et des vitamines.

Résumé – Introduction. Le Cerrado est le plus vaste biome de l'état de Minas Gerais au Brésil ; il dispose d'une importante biodiversité d'espèces fruitières, parmi lesquelles la présence du murici (*Byrsonima verbascifolia*) mérite d'être soulignée. L'objectif de notre étude a été d'étudier les caractéristiques physiques et physico-chimiques, l'occurrence et la concentration des carotenoïdes, de la vitamine C, de la vitamine E et des folates dans le fruit du murici collecté dans le Cerrado du Minas Gerais au Brésil. **Matériel et méthodes**. L'acidité titrable a été déterminée par neutralisation volumétrique, le pH par potentiométrie, les solides solubles par réfractométrie, l'humidité à l'aide d'un four, les cendres en utilisant un four à moufle, les protéines par la méthode de micro-Kjeldahl, les fibres alimentaires totales par la méthode de gravimétrie non-enzymatique et les lipides par l'extraction de Soxhlet ; des carotenoïdes et la vitamine C ont été analysés par HPLC-DAD, et la vitamine E et les folates l'ont été par HPLC avec détection de fluorescence. **Résultats et discussion**. La pulpe de murici contient 7,47 g·100 g^{-1} d'hydrates de carbone, 5,13 g·100 g^{-1} de lipides, 13,58 g·100 g^{-1} de fibres et 71.58 g·100 g^{-1} d'humidité. Le pH, l'acidité titrable et les solides solubles ont été, respectivement, de 3,93, 0,77 g·100 g^{-1} d'acide citrique et de 10,73 °brix. Les résultats ont indiqué que le fruit est une excellente source de fibres et de vitamine C (27,24 mg·100 g)$^{-1}$, une bonne source de vitamine E (1819,72 µg·100 g^{-1}) pour les enfants et une source pour les adultes et les femmes enceintes. Nous n'avons pas trouvé de folates dans le murici. **Conclusion**. Le murici se démarque grâce à sa valeur nutritive. Il pourrait contribuer de manière significative à l'apport en éléments nutritifs, notamment en fibres et vitamine C. La consommation de ce fruit devrait donc être stimulée auprès des familles résidant dans le Cerrado et dans d'autres régions du Brésil.

Brésil / *Byrsonima verbascifolia* / fruits / propriété physicochimique / composition globale / vitamine C / vitamine E / complexe vitaminique B

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1. Introduction

The Cerrado is considered the largest biome in the state of Minas Gerais, covering much of the northern and western portions of the state; it is the second largest in Brazil, only smaller than the Amazon rainforest. It covers an area of over two million square kilometers, representing approximately 22% of the national territory, distributed predominantly among the states of Minas Gerais, Goiás, Tocantins, Bahia, Ceará, Maranhão, Mato Grosso, Mato Grosso do Sul, Piauí, Rondônia and São Paulo [1, 2].

However, in recent decades, the Brazilian Cerrado has suffered severe degradation, where agricultural expansion, with extensive pastures and large monocultures, disregards the native vegetation and contributes to reduction of the original cover, thereby contributing to reducing its biodiversity [3]. According to the NGO Conservation International, the Cerrado is considered a "hotspot"; in other words, one of the richest and most threatened biomes on the planet [4].

The biodiversity of the Brazilian Cerrado is represented by the variety of fruit species found in this biome, including *Byrsonima verbascifolia* belonging to the family Malpighiaceae, which is known in Brazil as douradinha-falsa, mirici, muriciz-inho, orelha-de-burro and orelha-de-veado. The murici, fruit of *B. verbascifolia,* can be consumed *in natura* by the population. When ripe, it presents a yellow color with a strong odor similar to that of stale cheese [5, 6]. The pulp is fleshy and soft, and can also be consumed in the form of juices, jellies, ice creams and liquors [6].

However, despite the use of murici pulp for various purposes, there is no data on the nutritional content of the fruit in the specialized literature and Brazilian food composition tables; especially, there is no data in relation to concentrations of carotenoids and vitamins studied with reliable analysis methods such as high-performance liquid chromatography (HPLC). Thus, the objective of our study was to physically and physicochemically characterize and investigate the occurrence and concentration of carotenoids and vitamins, as well as categorize murici from the Cerrado of Minas Gerais, Brazil, with regard to its nutritional value.

2. Materials and methods

2.1. Raw material

Murici fruits (*B. verbascifolia*) were collected directly from random trees during its harvest season (November 2010 to March 2011) in an area of native vegetation typical of the Cerrado, located in the northern region of the state of Minas Gerais, Brazil, in the city of Januária (lat. 15°29' S and long. 44°21' W).

2.2. Collection and preparation of the samples

To obtain five replicates the collection area was divided into five sub-areas, where approximately 200 g of murici (60 units) were collected in each area. In the laboratory, fruits were selected according to the degree of maturation and absence of injuries, based on the characteristic parameters of color and texture of the fruit. Ripe fruits were considered those with predominantly yellow skin and soft texture. The morphologically perfect and completely ripe fruits were washed with tap water to eliminate surface dirt from the site collection and dried on paper towels.

The pulp obtained was homogenized using a domestic food processor (Faet Multipratic, MC5, Brazil), packaged in polyethylene bags, labeled, wrapped in aluminum foil and stored at (–18 ± 1) °C.

2.3. Standards of carotenoids and vitamins

The standards of α-carotene and β-carotene were isolated from concentrated extract of carrot, while β-cryptoxanthin and lycopene were isolated from extracts of papaya and tomato, respectively, by open column chromatography, according to Rodrigues-Amaya [7]. L-ascorbic acid was purchased

from Sigma-Aldrich® (Germany). The vitamin E standards (α-, β-, γ- and δ-tocopherol and tocotrienol) were purchased from Calbiochem®, EMD Biosciences, Inc. (USA). The folate standards used [(6S)-5,6,7,8-sodium tetrahydrofolate (THF), (6S)-5-methyl-5,6,7, 8-tetrahydrofolate (5-MTHF) and (6S) 5-formyl-5,6,7,8-tetrahydrofolate (5-FTHF)] were provided by Merck-Eprova®(Switzerland).

2.4. Physical characterization

Individual measurements of diameter and length were carried out on 30 murici fruits using a digital caliper rule (Mitutoyo, Brazil). The mass of fruit (MFr), mass of the pulp (MPu) and seed mass were obtained by individual direct weighing on a semi-analytical balance (Gehaka, BG 2000, Brazil).The pulp yield was calculated using the equation [(MPu/MFr) × 100].

2.5. Physicochemical analyses

The physicochemical analyses were performed in three repetitions. Titratable acidity, soluble solids and pH were determined according to the methodologies proposed by the Instituto Adolfo Lutz [8]; moisture, ash, protein, lipids and total dietary fiber were determined according to the methods of the Association of Official Analytical Chemistry [9]. Carbohydrate concentrations were estimated by the equation: [100 – (% moisture + % fat + % protein + % total dietary fiber + % ash)]. The total energy was estimated considering the conversion factors of 4 kcal·g^{-1} for protein and carboydrate, and 9 kcal·g^{-1} for lipids [9].

2.6. Extraction and analyses of carotenoids and vitamins

The extraction and analysis were performed in five repetitions. During the steps of extraction and analysis, the samples and extracts were protected from both sunlight and artificial light with the use of amber glass bottles, aluminum foil and blackout curtains, and they were also protected from oxygen by using lids and environments with nitrogen gas in glass bottles.

2.7. Carotenoids

The occurrence and concentration of α-carotene, β-carotene, β-cryptoxanthin and lycopene were investigated in murici pulp. Extraction was performed using the method proposed by Rodriguez-Amaya *et al.* [10].

About 5 g of pulp were weighed, supplemented with 60 mL of cooled acetone (divided into three volumes of 20 mL), homogenized in a micro-crusher (Marconi, MA 102, Brazil) for approximately 3 min, and vacuum-filtered in a Büchner funnel using filter paper. Then, the filtrate was transferred in three fractions to a separatory funnel containing 50 mL of cooled petroleum ether. Each fraction was washed with distilled water for complete removal of acetone. Anhydrous sodium sulfate was added to the ether extract to remove any residual water. Subsequently, the ether extract was concentrated using a rotary evaporator (Tecnal, TE-211, Brazil) at (35 ± 1) °C and transferred to a 25-mL volumetric flask, where the volume was completed with petroleum ether. This extract was then transferred to a hermetically sealed amber glass bottle and stored at (–18 ± 1) °C.

For chromatographic analysis, an aliquot of 10 mL of the extract was evaporated under a flow of nitrogen gas, and the dry residue was dissolved in 2 mL HPLC-grade acetone (Tedia, Brazil). The extracts were filtered through HV Millex filter units made of polyethylene, with 0.45 µm porosity (Millipore, Brazil).

Carotenoid analysis was carried out by HPLC using a HPLC system (Shimadzu, SCL 10at VP, Japan) coupled to a diode array detector (DAD) (Shimadzu, SPD-M10A, Japan), and the chromatographic conditions developed by Pinheiro-Sant'Ana *et al.* [11]: a chromatographic column (Phenomenex Gemini RP-18, 250 mm × 4.6 mm, 5 mm), equipped with a guard column (Phenomenex ODS, 4 mm × 3 mm), a mobile phase of methanol:ethyl acetate:acetonitrile (70:20:10, v/v/v) at a flow rate of 2 mL·min^{-1}, injection of sample: 50 µL; running time 15 min. Chromatograms were obtained at 450 nm.

Vitamin A concentrations were calculated according to the recommendations of the Institute of Medicine [12], in which 1 Retinol Activity Equivalent (RAE) corresponds to 1 µg of retinol, 12 µg of β-carotene, or 24 µg of other provitamin A carotenoids.

2.8. Vitamin C

The extraction and analysis of ascorbic acid and conversion of dehydroascorbic acid into ascorbic acid were performed according to the conditions proposed by Campos *et al.* [13].

For the extraction, about 5 g of pulp were ground for about 3 min in 15 mL of extraction solution composed of ultrapure water supplemented with metaphosphoric acid 3%, acetic acid 8%, sulfuric acid 0.8% and EDTA 0.0294%. The extract was centrifuged (Fanem, Excelsa Baby II-206R, Brazil) at 4,000 rpm (1,789 *g*) for 15 min, vacuum-filtered on filter paper in a Buchner funnel, and diluted to 25 mL in a volumetric flask with ultrapure water. Subsequently, the extract was again centrifuged at 14,000 rpm (21,913 *g*) for 5 min and the supernatant stored under refrigeration (5 ± 1) °C until analysis.

For the conversion of dehydroascorbic acid into ascorbic acid, a 1.0-mL aliquot of the extract obtained in the extraction of ascorbic acid was pipetted into an amber glass bottle and added to 1.0 mL of 1.2 M Trizma buffer solution (pH 9.0) containing 40 mM dithiothreitol (DTT) (Sigma-Aldrich, Germany) to increase the pH to near neutrality (pH 6.0). The extract was kept at rest for 10 min at room temperature and protected from light. Subsequently, the pH of the extract was reduced to 2 by the addition of 0.5 mL of 0.4 mM H_2SO_4.

The ascorbic acid analyses were performed by injection of 30 µL of the extract previously filtered in filter units with porosity of 0.45 µm. The analyses were performed using the same HPLC system employed in the analysis of carotenoids and the following chromatographic conditions were used: HPLC-DAD system, RP-18 chromatography column (Lichrospher 100, 250 mm × 4 mm, 5 mm), mobile phase consisting of ultrapure water with 1 mM NaH_2PO_4, 1 mM EDTA and pH adjusted to 3.0 with H_3PO_4, and mobile phase flow of 1.0 mL·min^{-1}. Chromatograms were obtained at 250 nm [13].

The total concentration of vitamin C was calculated by the sum of ascorbic acid and dehydroascorbic acid found in the samples.

2.9. Vitamin E

The extraction and analysis of the eight components of vitamin E (α-, β-, γ- and δ-tocopherols and α-, β-, γ- and δ-tocotrienols) were performed according to Pinheiro-Sant'Ana *et al.* [14].

Approximately 10 g of pulp were supplemented with 4 mL of heated ultrapure water [(about (80 ± 1) °C], ten mL of isopropyl alcohol, one mL of hexane containing BHT 0.05% and 5 g of anhydrous sodium sulfate. Gradually, twenty-five mL of the extraction solvent mixture (hexane:ethyl acetate, 85:15, v/v) were added to the suspension. After these procedures, the suspension was homogenized in a microcrusher at average speed for 1 min. Once ground, the sample was vacuum-filtered through filter paper in a Büchner funnel, and the residue was maintained in an extraction tube.

The extraction was repeated with the addition of 5 mL of isopropyl alcohol and 3 mL of the solvent mixture, with subsequent homogenization and vacuum filtration. Then the extract was concentrated in a rotary evaporator at (70 ± 1) °C for about 2 min, transferred to a volumetric flask and the volume completed to 25 mL with the solvent mixture.

After extraction, five mL of the extracts were dried in nitrogen gas, redissolved in 2.0 mL of HPLC-grade hexane and filtered through filter units with porosity of 0.45 µm. The analysis of the different components of vitamin E was performed by HPLC (Shimadzu SCL 10AD VP, Japan) comprising a high-pressure pump (Shimadzu LC-10AD VP, Japan), an autosampler with a 50-µL loop (Shimadzu SIL-10AF, Japan) and a fluorescence detector (Shimadzu, RF10AXL, Japan).

The chromatographic conditions used for the analysis included: fluorescence detection (excitation at 290 nm and emission at 330 nm); a Luna chromatographic column (Phenomenex, Si60, 250 mm × 4 mm, 5 mm), equipped with a guard column (Phenomenex, Si60, 4 mm × 3 mm), a mobile phase composed of: hexane:isopropanol:acetic acid in the proportions of 98.9:0.6:0.5, a mobile phase flow rate of 1.0 mL·min^{-1} and injection of the sample: 10 µL. The total concentration of vitamin E was calculated by the sum of the components of vitamin E identified in the samples [14].

2.10. Folates

The occurrence of three folate forms (THF, 5-MTHF and 5-FTHF) was investigated, with the extraction and analysis performed according to Della Lucia *et al.* [15]. Approximately 5 g of the pulp were ground in 20 mL of phosphate buffer solution 0.1 M, pH 6.0, containing ascorbic acid 1% and 2-mercaptoethanol 0.1%. The suspension was centrifuged at 4,000 rpm (1,789 *g*) for 15 min, vacum-filtered in a Buchner funnel using filter paper, and diluted to 25 mL in a volumetric flask with ultrapure water. Then, the extract was heated for about 12 min in a water bath at (100 ± 1) °C and cooled in an ice bath until the temperature fell below (37 ± 1) °C.

The cooled extract was centrifuged again at 4,000 rpm (1,789 *g*) for 15 min and submitted to deconjugation of the polyglutamates into monoglutamates. For deconjugation, rat plasma (100 µL) containing the enzyme conjugase (γ-glutamyl carboxypeptidase) was added to 3 mL of the previously obtained supernatant, and the extract was incubated in a water bath at (37 ± 1) °C for 3 h. Then, the extract was heated in boiling water for 5 min to inactivate the enzyme.

Extract purification was performed using an ion-exchange column with a stationary phase of Q-Sepharose Fast Flow (Pharmacia, USA). The column was pre-conditioned with methanol (Tedia, Brazil) and water (1:1), and the extract was applied to the column at a flow rate of 2 drops per second. Then, retained folates were eluted in 1.5 mL of sodium acetate (0.1 M) containing 10% NaCl, 1% ascorbic acid and 0.1% 2-mercaptoethanol.

Analyses were performed by injection of 50 µL of the extracts previously filtered through filter units with a porosity of 0.45 µm into the same system used for analysis of vitamin E, complemented with a mobile phase degassing system utilizing helium (Shimadzu DGU-2, Japan). The chromatographic conditions used for analysis included: HPLC system; a ShimPack 100 RP18 chromatography column (150 mm × 4.6 mm, 4.6 µm) (Merck, Germany), and a mobile phase composed of phosphate buffer (30 mM NaH_2PO_4, pH adjusted to 2.3 with H_3PO_4) as eluent A and acetonitrile as eluent B.

The binary gradient utilized was as follows: from (0 to 5) min, 94% of eluent A + 6% of eluent B; from (5 to 25) min, linear gradient to 75% of eluent A + 25% of eluent B, from (25 to 33) min, 75% of eluent A + 25% of eluent B, from (33 to 35) min, return to initial conditions followed by a stabilization period of up to 50 min. The mobile phase flow rate was 0.7 mL·min^{-1}. The mobile phase was degassed with helium gas for 15 min at 100 kPa before initiating the analyses and 50 kPa during runs [15].

2.11. Identification and quantification of carotenoids and vitamins

Qualitative identification of the compounds was performed by comparing the retention times obtained for standards and samples analyzed under the same conditions. In addition, carotenoids and ascorbic acid were identified by comparison of the absorption spectra of the standards and samples using the DAD, and the folates and vitamin E by co-chromatography.

Quantitation of the compounds was performed using external standardization curves. Appropriate dilutions were made from standard solutions in order to achieve concentrations comparable with those observed in fresh pulp of murici. For this,

solutions for each compound present in the pulp of the fruit (β-carotene, ascorbic acid, α-tocopherol and β-tocotrienol) were prepared at different concentrations.

Construction of standard curves was taken by injection, in duplicate, with six increasing concentrations of the standard solutions in the range between 0.003 µg and 0.112 µg for β-carotene, 0.155 µg and 7.750 µg for ascorbic acid, 0.0010 µg and 0.1042 µg for α-tocopherol, and 0.0020 µg and 0.5100 µg for β-tocotrienol. Thus, there was a linear correlation between the peak areas and concentrations of each compound injected.

Quantification of compounds in murici was performed on the analytical curves and regression equations achieved for β-carotene (y = 1421302.230x + 3563.819; R^2 = 0.999); ascorbic acid (y = 1394501.207x – 115382.946; R^2 = 0.993); α-tocopherol (y = 93284137.0896x + 47566.8009; R^2 = 0.997), and β-tocotrienol (y = 24797142.844x + 285288.289; R^2 = 0.998). The real concentration was obtained by calculations based on the dilution factors.

2.12. Quality control of the analytical methods

Tests of recovery, linearity, repeatability, limit of detection (LOD) and limit of quantification (LOQ) were performed for quality control of the analytical methods used in the analysis of carotenoids and vitamins.

The recovery tests were performed by addition of standards (β-carotene, ascorbic acid, α-tocopherol and β-tocotrienol) to the samples. The amount of added standard ranged from 50% to 100% of the initial concentration observed in murici pulp. The percentage of recovery was calculated using the equation: % recovery = [(final concentration of compound – concentration of added standard) / (initial concentration of compound) × 100]. All procedures were performed in triplicate.

The determination of the linearity range of the compounds was determined by injection, in duplicate, of six standard solutions with different concentrations, using the same chromatographic conditions used for the analysis of the extract. The data obtained from the peak areas were used for linear regression analysis. The coefficient of determination (R^2) obtained in each case was used to evaluate the linearity [16].

Repeatability tests were performed by extraction and analysis in five replications of the same repeat containing vitamins found. The repeatability was evaluated by calculating the relative standard deviation (RSD) of the peak areas and retention times of the analyzed components [16]. The limit of detection (LOD) was performed by successive dilutions of standards followed by determination of the smallest detectable amount. The LOD was established as three times the amplitude of the baseline noise and the limit of quantification as ten times the LOD [17].

2.13. Categorization of murici as a source of nutrients and fiber

The categorization of a portion of murici pulp as a source of fiber and vitamins for children (4–8 years), pregnant women and adult men (19–30 years) was performed according to Philippi, who classifies food as a "source" supplying nutrients when it provides 5–10% of the Dietary Reference Intake (DRI), "good source" when it supplies 10–20% of the DRI and "excellent source" of nutrients when supplying more than 20% of the DRI [18].

2.14. Experimental design and statistical analysis

A completely randomized design with five replications, represented by the five batches of each fruit, was used for analyses of the concentration of carotenoids and vitamins, with three replicates for physicochemical analyses. Data was stored in spreadsheets using the Microsoft Office Excel software system, version 2007. Calculation of the mean, standard deviation and range of parameters was performed using SAS software (Statistical Analysis System), version 9.2 (2008), licensed to the Federal University of Viçosa (UFV), Brazil.

3. Results and discussion

3.1. Physical characterization

The murici fruits presented a rounded shape, slightly flattened, with a thin bright yellow skin. Their pulp is juicy, fleshy, oily and yellowish, with a strong odor, containing a small seed in its interior (*figure 1*).

The average pulp mass of each fruit was 2.78 g, ranging from 1.43 g to 4.41 g with yield of 79.04% (*table I*), which may result in good technological use of the fruit for the development of products. The average pulp mass was higher than the 1.99 g found by Gusmão *et al.* [19] in fruit of the same species collected in the municipality of Montes Claros, Minas Gerais, Brazil. The pulp yield was also higher than the 63% reported by Araújo *et al.* [20] in fruits collected in the Cerrado region of the state of Alagoas, Brazil. These different results may be due to soil and climatic differences between the places of fruit collection.

3.2. Physicochemical characterization

The soluble solids concentration found in the murici pulp of fruit from Minas Gerais, Brazil (10.73 °Brix) (*table II*) was similar to that reported by Guimarães and Silva [21] (10.67 °Brix) in fruits collected in the state of Goiás, and higher than that found by Canuto *et al.* [22] in fruits from the Amazon (1.5 °Brix). The higher soluble solids content found in our study compared with that reported in previous studies may be explained by the difference in the states (which have different climates) in which the

Figure 1.
Whole fruit and fruit pulp of murici (*Byrsonima verbascifolia*).

Table I.
Mean ± standard deviation of physical characteristics of 30 fruits of murici (*Byrsonima verbascifolia*) from the Cerrado (Januária, Minas Gerais, Brazil).

Parameters	Diameter (cm)	Height (cm)	Mass (g)			Pulp yield (%)
			Fruit	Seeds	Pulp	
Mean	1.82 ± 0.17	1.36 ± 0.11	3.51 ± 0.87	0.73 ± 0.19	2.78 ± 0.71	79.04 ± 3.09
Minimum	1.40	1.10	1.74	1.23	1.43	71.43
Maximum	2.10	1.60	5.51	0.31	4.41	84.69

fruits were collected, and also by their ripeness. The contents of soluble solids correlate with the sugar and organic acid concentrations, which are parameters of interest for the marketing of fresh fruit due to the preference of consumers for sweet fruits [22].

Another criterion for the classification of fruit flavor, odor, stability and quality is the determination of the titratable acidity [23]. The titratable acid concentration we found in Minas Gerais (0.77 g citric acid·100 g^{-1}) (*table II*) was similar to that found by Canuto *et al.* [24] in fruit pulp of the same species collected in the Amazon (1.0 g citric acid·100 g^{-1}). The [soluble solids/titratable acid] ratio obtained in our study was high (3.93). This parameter is related to fruit quality in terms of maturity and flavor, showing the balance between sugars and acidity which makes it more palatable, highlighting its sweet and pleasant taste [23].

The murici pulp in the fruits we collected in Minas Gerais presented a pH greater than the 3.42 reported by Guimarães and Silva [21] in fruits collected in Goiás, and pH 3.7 observed by Canuto *et al.* [24] in fruits of the Amazon.

The concentration of dietary fiber in murici fruits from the Cerrado of Minas Gerais, Brazil, was 13.58 g·100 g^{-1} (*table II*); there, this fruit may be considered an excellent source of this component for children, adults and pregnant women, since the consumption of a portion of fruit pulp (86 g) may contribute over 40% of daily needs in accordance with the recommendation of the Institute of Medicine [25]. Due to the high content of dietary fiber, murici can help control intestinal transit and reduce both serum glucose and cholesterol [26].

We observed a lower moisture content than that reported by Guimarães and Silva (75.87 g·100 g^{-1}) [21] and by Silva *et al.* (80.64 g.100 g^{-1}) [27] in fruits of the same species collected in the state of Goiás. The high moisture concentration in murici pulp, along with the fragility of its skin, makes this fruit highly susceptible to enzymatic and microbial spoilage, which complicates its conservation.

The murici pulp from the Cerrado in Minas Gerais showed carbohydrate concentrations greater than the 5.96 g·100 g^{-1} observed by Silva *et al.* in fruits from the Cerrado collected in the state of Goiás, Brazil, [27]; however, this value is lower than the 19.62 g·100 g^{-1} observed by Guimarães and Silva in fruits also collected in the state of Goiás [21]. The lipid concentration was greater than the 2.19 g·100 g^{-1} [27] and 3.02 g·100 g^{-1} [21] reported for fruits collected in the state of Goias, Brazil.

In the murici pulp of the fruit we studied, the protein and ash concentrations were also higher than those reported by Silva *et al.* (0.72 g·100 g^{-1} and 0.78 g·100 g^{-1}, respectively) [27] and Guimarães and Silva (0.86 g·100 $g^{-1}$and 0.63 g·100 g^{-1}, respectively) [21] in fruit pulps found in the Cerrado of the state of Goiás, Brazil. The energy value of murici evaluated in our study was higher than the 46.43 kcal·100 g^{-1} observed by Silva *et al.* [27]; however, it was lower than the 109.10 kcal·100 g^{-1} which was found by Guimarães and Silva in a study with fruits from the Cerrado of Goiás, Brazil [21].

3.3. Carotenoids and vitamins

3.3.1. Quality of analytical methods

Tests for quality control of the analyses performed in our study showed that the assay conditions were reliable, with low probability of carotenoid and vitamin loss during extraction: they allowed the detection of low concentrations of the analyzed compounds (*table III*).

The recovery percentages of carotenoid and vitamin standards added to the murici extracts were excellent, ranging from 91.7% to 98.0% (*table III*). The repeatability test showed low values of relative standard deviation for peak areas (RSD-PA) and retention times (RSD-RT), which adds to the reliability of the analyses. A wide linearity range was observed for each compound examined and the coefficients of determination (R^2) were greater than 0.993. The limit of detection (LOD) for carotenoids and vitamins ranged from 0.025 to 12.321 µg·mL^{-1}. The limit of quantification

(LOQ), considered as 10 times the value of the LOD, ranged from (0.251 to 123.21) μg·mL^{-1}. Therefore, the LOD and LOQ obtained show that very small amounts of the vitamins can be detected and quantified by the method employed.

3.3.2. Qualitative composition

Murici pulp showed the presence of β-carotene (retention time-RT: 8.30 min), ascorbic acid (RT: 3.51 min), α-tocopherol (RT: 6.45 min) and β-tocotrienol (RT: 11.05 min). In the murici samples we studied, we did not identify α-carotene, β-cryptoxanthin, lycopene, β-, γ- and δ-tocopherol, α-, γ- and δ-tocotrienol or the three forms of folate (*figure 2*).

3.3.3. Vitamin content

Studies on the concentration of vitamin C, vitamin A, vitamin E and folates in murici fruits from the Cerrado of Minas Gerais, Brazil, were not found in the literature (*table IV*). In pulp of murici, the presence of vitamin C was observed both in the reduced (ascorbic acid) and in the oxidized forms (dehydroascorbicdehydroascorbic acid), where ascorbic acid corresponds to 82.92% of the total vitamin C concentration. The concentration of vitamin C found in murici pulp was higher than that of fruits conventionally consumed by the Brazilian population: pitanga (26.3 mg·100 g^{-1}), tangerine (21.0 mg·100 g^{-1}), melon (18.0 mg·100 g^{-1}) and grapes (10.8 mg·100 g^{-1}); it was similar to that found in mango (27.7 mg·100 g^{-1}) [28]. The concentration of vitamin C in murici pulp was also higher than that of other fruits found in the Cerrado of Minas Gerais, Brazil, such as boiled pequi (*Caryocar brasiliense* Camb.) (14.33 mg·100 g^{-1}) [29], jatobá (*Hymenaea stigonocarpa*) (8.91 mg·100 g^{-1}) [30], araticum (*Annona crassiflora* Mart.) (5.23 mg·100 g^{-1}) [31] and tamarind (*Tamarindus indica* L.) (4.79 mg·100 g^{-1}) [32].

The concentration of vitamin A observed in murici was similar to nectarine (0.15 mg·100 g^{-1}) and higher than that found in strawberries (0.02 mg·100 g^{-1}) [33] and mangaba (*Hancornia speciosa*) (7.5 mg·100 g^{-1}) in the Cerrado of Minas

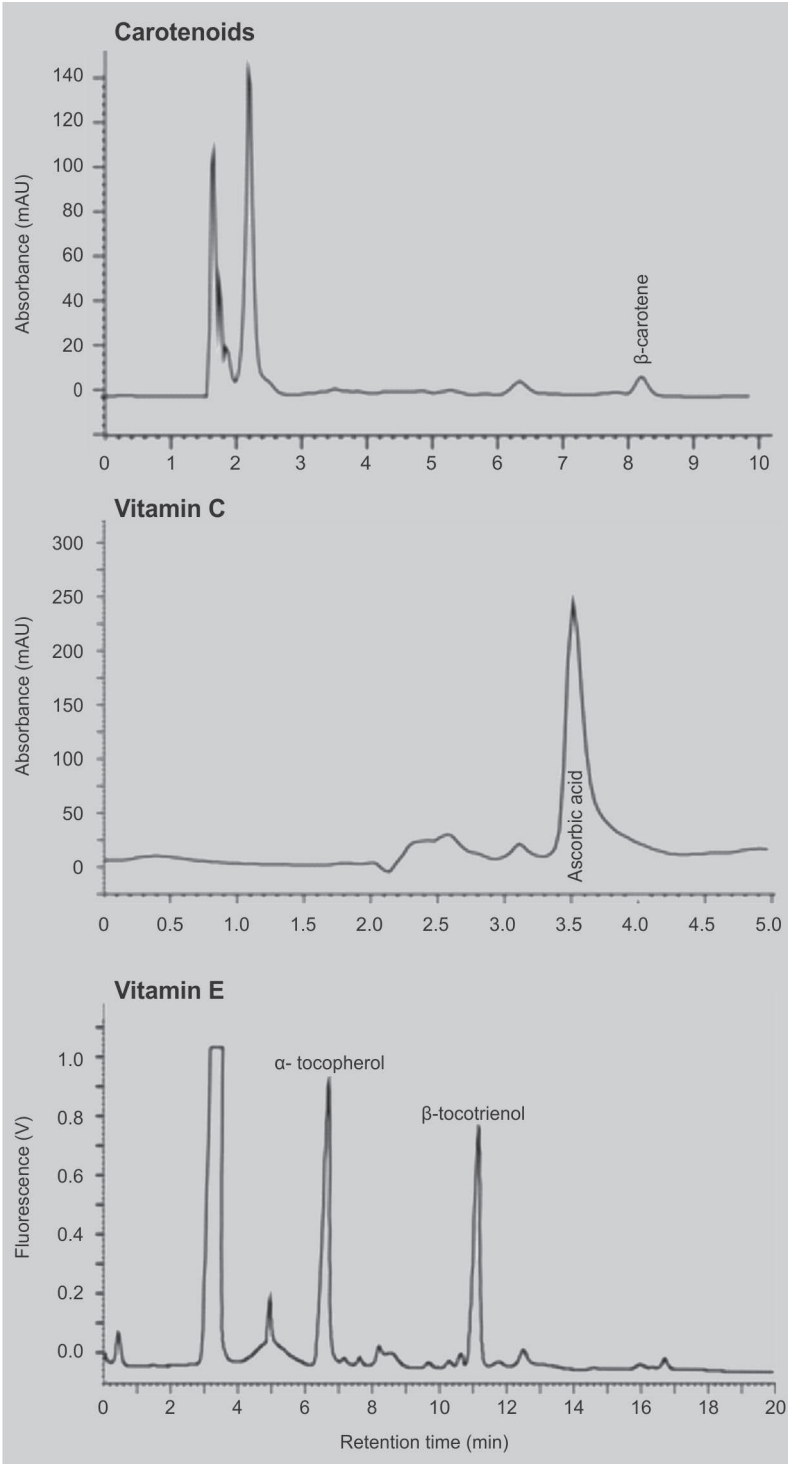

Figure 2.
HPLC analysis of carotenoids, vitamin C and vitamin E in murici pulp (*Byrsonima verbascifolia*) from the Cerrado of Minas Gerais (Januária, Minas Gerais, Brazil).

Table II.
Mean ± standard deviation for physicochemical characteristics and total energy of the pulp of murici (*Byrsonima verbascifolia*) from the Cerrado (Januária, Minas Gerais, Brazil). Values are expressed in fresh matter, mean of three repetitions.

Soluble solids (SS) (°Brix)	Titratable acidity (TA) (g citric acid·100 g^{-1})	[SS/TA] ratio	pH	Moisture	Ash	Proteins	Total dietary fibers	Lipids	Carbohydrates	Total energy value (kcal·100 g^{-1})
				(g·100 g^{-1})						
10.73 ± 1.22	0.77 ± 0.00	13.94 ± 1.63	3.93 ± 0.02	71.58 ± 2.27	0.93 ± 0.08	1.26 ± 0.06	13.58 ± 0.14	5.13 ± 0.45	7.47 ± 2.00	81.23 ± 10.70

Table III.
Repeatability, limits of detection and quantification, linearity range, and recovery of carotenoids and vitamins in the pulp of murici (*Byrsonima verbascifolia*) from the Cerrado (Januária, Minas Gerais, Brazil).

Compounds	Repeatability		Limit of detection (µg.mL^{-1})	Limit of quantification (µg.mL^{-1})	Linearity range (µg)	Recovery (%)
	RSD-PA	RSD-RT				
β-carotene	1.05	0.60	6.422	64.221	0.2060 – 6.0321	95.5
Ascorbic acid	1.58	0.59	12.321	123.214	0.1550 – 7.7500	91.7
α-tocopherol	0.72	0.92	0.025	0.251	0.0104 – 0.1040	93.4
β-tocotrienol	0.86	0.90	0.034	0.342	0.0170 – 0.1700	98.0

RSD-PA: relative standard deviation of the peak areas.

RSD-RT: relative standard deviation of retention times.

Table IV.
Concentration of carotenoids and vitamins in 100 g of pulp of murici (*Byrsonima verbascifolia*) from the Cerrado (Januária, Minas Gerais, Brazil). Values are expressed in fresh matter (mean of five repetitions).

Parameters	Carotenoids	β-carotene	Vitamin C	Ascorbic acid	Dehydroascorbic acid	Vitamin A (RAE·100 g^{-1})	Vitamin E	α-tocopherol	β-tocotrienol
	(mg·100 g^{-1})						(µg·100 g^{-1})		
Mean ± standard deviation	0.16 ± 0.04	0.16 ± 0.04	27.23 ± 4.91	22.58 ± 5.76	4.64 ± 2.14	13.37 ± 3.22	1816.71 ± 417.97	949.68 ± 413.54	867.03 ± 87.29
%	100.0	100.0	100.0	82.92	17.08	–	100.0	52.3	47.7

RAE: Retinol Activity Equivalent.

Gerais [34]. The concentration of vitamin E observed in the murici pulp was represented by the compounds α-tocopherol and β-tocotrienol, corresponding to 52.3% and 47.7% of the total content, respectively (*table IV*). Murici showed a considerable vitamin E concentration, higher than that found in kiwi (1450 µg·100 g^{-1}) and peach (1230 µg·100 g^{-1}) [28]. The concentration was also higher than that of other fruits found in the Cerrado of Minas Gerais, Brazil, such as jatobá (495.54 µg·100 g^{-1}) [30], araçá (*Psidium firmum* O. Berg) (336.33 µg·100 g^{-1}) [35], baked pequi (170.81 µg·100 g^{-1}) [29] and tamarind (108.78 µg·100 g^{-1}) [32].

3.3.4. Nutritional value of murici pulp as a source of vitamins and dietary fiber

To study the potential contribution of murici pulp to supplying the daily recommendation of vitamins for children, adults and pregnant women, pulp portions were calculated according to the National Health Surveillance Agency/ANVISA, Resolution 39, which recommends the presence of 70 kcal in a single fruit portion [36].

Consumption of 30 units of murici fruit, equivalent to 86 g of pulp (70 kcal), can contribute to supplying more than 40% of the recommended daily fiber intake for children, adults and pregnant women, and thus can be considered an excellent source of this nutrient for the three groups (*table V*).

The murici fruit is also highlighted due to its potential for daily supply of vitamin C. Consuming only one portion of the fruit is sufficient to supply 93.7% of the daily vitamin C requirement for children, 26.0% for adults and 29.3% for pregnant women; it is therefore considered an excellent source of this vitamin [18].

Murici may also be considered a source of vitamin E for the three groups studied. The potential contribution of vitamin E was calculated based on the concentration of α-tocopherol, as recommended by the Institute of Medicine [37]. It is noteworthy, however, that the presence of β-tocotrienol found in murici is also very important because of its potent antioxidant action and positive effect on the prevention of chronic diseases such as diabetes mellitus type 2 [38].

4. Conclusion

Murici stands out with regard to its nutritional value, and may contribute to supplying daily nutritional requirements,

Table V.
Potential contribution of the pulp of murici (*Byrsonima verbascifolia*) from the Cerrado of Minas Gerais to the supply of the daily recommendation of vitamins according to the recommended portion (86 g the pulp = 30 units the fruit = 70 kcal).

Concentration per serving (calculation based on Resolution no. 39) [35].

Dietary fiber (g)	Vitamin A (µg)	Vitamin C (mg)	Vitamin E (µg)
11.67	11.49	23.43	1562.38

Calculation based on the RDA (Recommended Dietary Allowance) [36, 37, 39].

People	Dietary fiber		Vitamin A		Vitamin C		Vitamin E	
	RDA (g)	% of contribution	RDA (µg)	% of contribution	RDA (mg)	% of contribution	RDA (µg)	% of contribution
Children	25	46.68	400	2.87	25	93.70	7000	11.40
Adults	25	46.68	800	1.43	90	26.02	15000	5.44
Pregnant	28	41.67	770	1.48	85	29.27	15000	5.44

particularly dietary fiber and vitamin C, but this fruit is also a source of vitamin E and contains provitamin A. This fact justifies encouraging its consumption by families living in the Cerrado and also in other regions of Brazil.

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El murici, fruto del Cerrado de Minas Gerais en Brasil: características físicas y físico-químicas, casos y concentraciones de los carotenos y de las vitaminas.

Resumen – Introducción. El Cerrado es el bioma más vasto del estado de Minas Gerais en Brasil. Dispone de una importante biodiversidad de especies fruteras, entre las cuales merece distinguirse la presencia del murici (*Byrsonima verbascifolia*). El objetivo de nuestro trabajo fue el estudio de las características físicas y físico-químicas, los casos y la concentración de los carotenos, de la vitamina C, de la vitamina E y de los folatos en el fruto de murici, cosechado en el Cerrado de Minas Gerais, en Brasil. **Material y métodos**. Se determinaron la acidez valorable mediante neutralización volumétrica, el pH mediante potenciometría, los sólidos solubles mediante refractometría, la humedad con la ayuda de un horno, las cenizas mediante el empleo un horno de mufla, las proteínas mediante el método de micro-Kjeldahl, las fibras alimentarias totales mediante el método gravimétrico no enzimático y los lípidos mediante extracción Soxhlet. Se analizaron los carotenos y la vitamina C mediante HPLC-DAD, y la vitamina E y los folatos mediante HPLC con detección de fluorescencia. **Resultados y discusión**. La pulpa de murici contiene 7,47 g·100 g^{-1} de hidratos de carbono, 5,13 g·100 g^{-1} de lípidos, 13,58 g·100 g^{-1} de fibras y 71.58 g·100 g^{-1} de humedad. El pH, la acidez valorable y los sólidos solubles fueron, respectivamente, de 3,93, 0,77 g·100 g^{-1} de ácido cítrico y de 10,73 °Brix. Los resultados indicaron que el fruto es una fuente excelente de fibras y de vitamina C (27,24 mg·100 $g)^{-1}$, una buena fuente de vitamina E (1819,72 µg·100 g^{-1}) para los niños y una fuente para adultos y mujeres embarazadas. No encontramos folatos en el murici. **Conclusión**. El murici se destaca gracias a su valor nutritivo. Podría contribuir de manera significativa al aporte de elementos nutritivos, sobre todo de fibras y vitamina C. Por lo tanto, el consumo de este fruto debería impulsarse entre las familias residentes en el Cerrado y en otras regiones de Brasil.

Brasil / *Byrsonima verbascifolia* / frutas / propriedades fisicoquímicas / composición aproxinada / vitamina C / vitamina E / vitaminas B