

Jaboticaba (*Myrciaria jaboticaba* (Vell.) O. Berg) peel powder produced by convective drying process: a rich anthocyanin product

M.C. Pessanha de Araujo Santiago^{1,a}, R. Galhardo Borguini¹, L. Da Silva de Mattos do Nascimento¹, E.C. de Oliveira Braga², V. De Carvalho Martins³, A.C.M. Senna Gouvêa³, F. Marques Peixoto^{3,4}, S. Pacheco¹, R.I. Nogueira⁵ and R.L. de Oliveira Godoy¹

¹ Laboratório de Cromatografia Líquida, Embrapa Agroindústria de Alimentos, Av. Das Américas, 29501, Guaratiba, Rio de Janeiro, RJ, 23020470, Brazil

² Instituto de Química, Universidade Federal do Rio de Janeiro, Av. Athos da Silveira Ramos, 149, Bloco A, Ilha do Fundão, Rio de Janeiro, RJ, 21941909, Brazil

³ Departamento de Ciência e Tecnologia dos Alimentos, Universidade Federal Rural do Rio de Janeiro, BR 465, km 7, Seropédica 8, RJ, 238908000, Brazil

⁴ Departamento de Farmácia, Universidade Estadual da Zona Oeste, Av. Manuela Caldeira de Alvarenga, 1203, Campo Grande, Rio de Janeiro, RJ, 23070-200, Brazil

⁵ Planta Piloto de Secagem, Embrapa Agroindústria de Alimentos, Av. Das Américas, 29501, Guaratiba, Rio de Janeiro, RJ, 23020470, Brazil

Summary

Introduction – Jaboticaba is a Brazilian fruit with a high concentration of bioactive compounds, mainly anthocyanins, in the peel. Although this fruit is an underutilized crop, it has a high productivity and requires practically no specific cultivation treatments. Due to the high perishability of the fruit and the difficulty of digesting its peel, facilitating the processing and consumption of them is a challenge. Although the chemical composition of jaboticaba fruits has been described previously, it is very important both for consumers and for the food industry to know the behavior of some compounds in the processed fruit, especially the behavior of the bioactive substances. The objective of this work is to obtain a product, in powder, based on jaboticaba peel through a simple and low-cost technology, and to chemically characterize and evaluate its application as a nutraceutical product. **Materials and methods** – Jaboticaba peel powder was obtained by convective drying and its anthocyanin content and stability were evaluated. Other flavonoids, phenolic acids, carotenoids and sugars content and antioxidant activity were also assessed. **Results and discussion** – The powder showed a high content of anthocyanins and ellagic acid, and other bioactive substances. The product also preserved its anthocyanin content over a 4-month-storage period. **Conclusion** – This powder product can be considered a potential functional ingredient and also a potential natural colorant, which could substitute synthetic dyes and at the same time be a source of bioactive compounds.

Keywords

Brazil, jaboticaba, *Myrciaria jaboticaba*, natural ingredient, bioactive compounds, underutilized species

Significance of this study

What is already known on this subject?

- Jaboticaba peel is known as a source of bioactive compounds, mainly anthocyanins.

What are the new findings?

- The product was obtained from the whole peel and by using a simple and low-cost technology (convective drying), which differs from products already reported (freeze-drying and spray-drying).
- The powder showed high anthocyanin stability, an important trait for possible use as functional ingredient.

What is the expected impact on horticulture?

- The present study can contribute to add value to this Brazilian underutilized fruit and to promote its productive chain.
- The simple drying technology can be easily transferred and used by small-scale farmers, to diversify and secure their income.

Résumé

La poudre d'écorce du fruit du jaboticaba (*Myrciaria jaboticaba* (Vell.) O. Berg) produite par séchage par convection: un produit riche en anthocyanes.

Introduction – Le jaboticaba est un fruit brésilien dont l'écorce (ou la peau) contient une forte concentration de composés bioactifs, principalement des anthocyanes. Bien que cet arbre fruitier soit une culture sous-utilisée, il a une productivité élevée et ne nécessite pratiquement aucun entretien de culture spécifique. En raison de la haute périssabilité du fruit et de la difficulté à digérer sa peau, la transformation

^a Corresponding author: manuela.santiago@embrapa.br.

post-récolte et la consommation humaine restent à optimiser. Il est très important à la fois pour les consommateurs et pour l'industrie alimentaire de connaître le comportement de certains composés dans le fruit transformé, en particulier le comportement des substances bioactives. L'objectif de ce travail est d'obtenir un produit, en poudre, à base d'écorce de fruit du jabuticaba à travers une technologie simple et peu coûteuse, de caractériser chimiquement et d'évaluer son application comme produit nutraceutique. Matériel et méthodes - La poudre d'écorce du fruit du jabuticaba a été obtenue par séchage par convection et sa teneur en anthocyanes et sa stabilité ont été évaluées. Les teneurs en autres flavonoïdes, acides phénoliques, caroténoïdes et sucres et l'activité anti-oxydante ont également été évaluées. Résultats et discussion - La poudre présentait une teneur élevée en anthocyanes et en acide ellagique, ainsi que d'autres substances bioactives. Le produit a conservé sa teneur en anthocyanes sur une période de stockage de 4 mois. Conclusion - Le produit en poudre peut être considéré comme un ingrédient fonctionnel potentiel et également un colorant naturel potentiel, qui pourrait remplacer les colorants synthétiques et en même temps être une source de composés bioactifs.

Mots-clés

Brésil, jabuticaba, *Myrciaria jaboticaba*, ingrédient naturel, composés bioactifs, espèce sous-utilisée

Introduction

Among the jabuticaba species *Myrciaria jaboticaba*, popularly known as jabuticaba Sabará, is the species most widely cultivated in Brazil, mainly in the southeastern part of the country. Their fruits are usually thin-skinned, small, always sweet and mature in October and November. The fruits are consumed in their natural state and as sweets and jellies (Lorenzi *et al.*, 2006). Many fruits of Brazilian biodiversity, such as jabuticaba, are perishable and poorly exploited sensory and nutritionally, as well as for use in processed products, being only produced and consumed locally (Gurak *et al.*, 2014). Although this fruit is an underutilized crop, it has a high productivity and requires practically no specific cultivation treatments. It is very common in domestic orchards in the South and Southeast Brazilian regions.

The chemical composition of jabuticaba fruit has been described in some papers (Batista *et al.*, 2014; Wu *et al.*, 2012) that has shown its value as a source of bioactive compounds. According to Wu *et al.* (2012), jabuticaba and its products can be considered functional foods due to the high antioxidant capacity of the fruit. These authors also emphasize the importance of choosing appropriate processing conditions in order to degrade as little as possible the bioactive compounds originally present in the fruit.

Leite-Legatti *et al.* (2012) found expressive contents of total phenolic compounds and anthocyanins in jabuticaba peel powder, besides its high antioxidant activity. Two major anthocyanins, delphinidin and cyanidin-3-*O*-glucoside, were identified. The authors also reported that jabuticaba peel extracts showed antiproliferative effects against leukemia and were active against prostate cancer cells.

Due to the high perishability of the fruit and the difficulty to digest its peel, which has a rigid texture and therefore requires hard chewing, improved methods to facilitate the processing and preservation of jabuticaba fruit could be very useful. The use as a supplement could be an option to facilitate the jabuticaba peel consumption and improve the anthocyanins ingestion. According to Zeisel (1999), food supplements that have a concentrated form of a bioactive component from a particular food in a non-food matrix are called nutraceuticals. Such products have the purpose of promoting beneficial health effects, depending on if the substances in which they are rich are biologically available. In the case of anthocyanins, some studies which have already concerned the anthocyanins bioavailability, related that their bioaccessibility and absorption are pointed to be higher in gastric mucosa level than in intestinal site (Fernandes *et al.*, 2012; Peixoto *et al.*, 2016).

This work differs from others already presented in the literature, because in most of them the jabuticaba peel product is obtained from freeze-drying process (Gurak *et al.*, 2014; Alezandro *et al.*, 2013; Leite *et al.*, 2011) or by spray-drying the extract of jabuticaba peel (Silva *et al.*, 2014). Silva *et al.* (2013) studied the influence of different carrier agents and temperatures for the production of jabuticaba extracts by spray-drying microencapsulation evaluating some parameters as the anthocyanins retention. Silva *et al.* (2014) also used spray-drying process to produce a natural pigment powder with functional properties. The use of a low-cost process facilitates the technology transfer to small farmers. Moreover, when the whole peel is used, and not only its extract, it is possible to obtain a richer product considering its bioactive compounds content, besides a lower waste generation. The objective of this work is to obtain a product, in powder, based on jabuticaba peel through a simple and low-cost technology (convective drying), to chemically characterize and to evaluate its application for a possible use as a nutraceutical product. Furthermore, this work evaluated the stability of the anthocyanins in the powder product, which may be added to foods as a natural dye and/or be consumed alone as a nutraceutical.

Materials and methods

Samples

Jabuticaba fruits (10 kg) of the *Myrciaria jaboticaba* species (Sabará) were collected in October (spring season) 2016 in the Joaquim Egídio district of Campinas, São Paulo, Brazil (46°53'10.99"S, 22°51'0.572"W). The ripe fruits were selected and separated in three samples of 500 fruits each. The fruits were washed and the peels were manually removed. The peels were submitted to a convective drying process on the same day.

Chemicals and standards

Acetonitrile, methanol, acetone, petroleum ether, and methyl *tert*-butyl, all of HPLC grade, and ethanol of ACS grade, were purchased from Tedia™ (Ohio, USA). Formic acid (98%), sulfuric acid (65%), hydrochloric acid (37%) and phosphoric acid (85%) were also purchased from Tedia™ (Ohio, USA). The ultrapure water was obtained from a Milli-Q™ Gradient 10A System (Merck Millipore Corporation, Massachusetts, USA). ABTS (2,2-azino-bis(3-ethylbenzo-thiazoline-6-sulfonic acid)), fluorescein and trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) were

purchased from Sigma-Aldrich™ (Missouri, USA). Standards of sugars: glucose and fructose (95%) and phenolic acids (95%) were purchased from Sigma Aldrich™ (USA). Standards of carotenoids and anthocyanins were isolated from natural sources by HPLC at the Embrapa Food Technology Laboratory (Rio de Janeiro, Brazil) with a purity >95% (Gouvêa *et al.*, 2012).

Fruit parts

Six jabuticaba fruits were individually weighed in order to establish the mean weight per fruit. Peel, pulp and seeds of the same six fruits were separated, and each part was individually weighed to define the content percentages (%).

Convective drying process

The jabuticaba peels were placed on trays in a single layer and subjected to dehydration at a temperature of 60 °C and air velocity of 1 m s⁻¹ for 22 h on a convective layer dryer developed by Embrapa Food Technology (Rio de Janeiro, Brazil) (Santiago *et al.*, 2016). The dried fruit peels were ground using a blender and stored in aluminum and polyethylene packs at room temperature (25 °C) until analysis.

Nutraceutical preparation

The capsules were prepared with the jabuticaba peel powder, which was previously sieved (150 mesh or 106 µm) and encapsulated following GMP (Good Manufacturing Practices). About 30 hard gelatin capsules were produced in a manual encapsulator with size 03 and 0.2 g powder was conditioned in each of them.

Jabuticaba peel powder centesimal composition

The analyses of moisture, ash, lipid, fiber and proteins followed the standard methods of the Association of Official Analytical Chemists (2010). The total carbohydrate content was determined by the difference of the other analyzed components. Centesimal composition analyses were done in duplicate.

Antioxidant capacity

The jabuticaba peel powder antioxidant capacity was measured by two methods: ABTS•⁺ radical-scavenging capacity and oxygen radical antioxidant capacity (ORAC). For both assays, the antioxidant compounds were extracted using 1 g of the sample and solutions of methanol/water (50:50 v/v) and acetone/water (70:30 v/v) (Rufino *et al.*, 2010). Analyses were performed in triplicate.

The ABTS radical specie was prepared by mixing 38.4 mg of ABTS and 6.6 mg of potassium persulfate and dissolved in ultrapure water and left in the dark at room temperature for 16 h before use. The ABTS solution was diluted with ethanol PA to an absorbance of 0.70 ± 0.02 at 734 nm. After the addition of 30 µL of the sample extract or trolox standard to 3 mL of diluted ABTS solution, the absorbance was recorded 6 min after mixing at 734 nm. The blank assay used ethanol (Rufino *et al.*, 2010).

For the ORAC assay fluorescein was used as the reference reagent (Zulueta *et al.*, 2009). In a black microplate, with a fluorescein 78 nM solution as reference, the fluorescence of the reaction medium was read, showing the oxidation of fluorescein. The analysis was performed at 37 °C in a microplate reader with excitation wavelength of 485 nm and emission wavelength of 535 nm.

For both assays the antiradical activity was expressed as trolox equivalents, using a trolox calibration curve.

Sugar analysis

The sugars were extracted according to Macrae (1998) using 1 g of sample and ultrapure water in an ultrasonic bath for 20 min, followed by the addition of 5 mL of acetonitrile and then the extract was filtered. This was analyzed by a high performance liquid chromatograph (Waters™ Alliance 2690/5), a Waters™ refractive index detector model 2410, and Empower™ software. A Zorbax™ Carbohydrate Agilent column 30 cm × 4.6 mm was used with the following parameters: a flow of 1.0 mL min⁻¹, injection volume of 20 µL, isocratic elution mode with a mobile phase of acetonitrile: water (75:25 v/v) and run time of 20 min. Analyses were performed in triplicate.

The sugars were quantified by external standards, based on calibration curves made with commercial analytical standards of fructose, glucose and sucrose.

Total carotenoids

The carotenoids were extracted according to Rodriguez-Amaya (2001). Approximately 1 g of jabuticaba peel powder was weighed and then manually macerated in a porcelain mortar with 3 g of celite (silver carbonate) and 20 mL acetone. The mixture was vacuum filtered in a glass funnel with a sintered plate. The extraction procedure was repeated until the sample no longer exhibited the characteristic color of carotenoids. The acetone extract was transferred quantitatively to a separator funnel containing 30 mL of petroleum ether and washed, at least five times, with 200 mL ultrapure water. The ether extract was filtered through anhydrous sodium sulfate, collected in 100 mL volumetric flasks and completed with petroleum ether. A 5 mL aliquot was separated for a further saponification reaction with 5 mL of potassium hydroxide (KOH) solution 10% in methanol (10:90, v/v) at room temperature and a reaction time of 16 h. After 16 h, the level of total carotenoids in the saponified sample extract was determined by spectrophotometry at 450 nm. Analyses were performed in triplicate.

Analysis of flavonoids and phenolic acid profiles

The analysis of free phenolic acids was performed according to the method described by Pérez-Jiménez (2008). The extraction was carried out by first mixing 1 g of the jabuticaba peel powder in 4 mL of methanol:water (50:50 v/v, pH 2) followed by mechanical stirring for 1 h and centrifugation (5,000 × g) for 10 min. The supernatant was collected (Extract 1). Then, 4 mL of acetone:water (70:30; v/v) was added to the residue and the mechanical stirring and centrifugation steps were repeated. The supernatant was collected (Extract 2). Then 3 mL of the supernatants (Extracts 1 and 2) were mixed and transferred to a 1.5 mL vial for chromatographic injection.

The sample residues were submitted to the extraction of hydrolyzed phenolics according to the method described by Frighetto and Bacca (2012). Alkaline hydrolyses were carried out with 5 mL of a solution (NaOH 2 M containing 1% of ascorbic acid and EDTA 10 mM) added to the samples followed by heating at 61–63 °C for 60 min., after which 1.5 mL of HCl 6 M was added immediately for the acid hydrolysis. The mixture was vortexed for 10 sec, left until it cooled to room temperature, followed by centrifugation (2,700 rpm) for 10 min. The supernatant was collected and 6.5 mL of ethyl acetate was added. The organic phase was separated and extraction with ethyl acetate was repeated. The organic fraction was dried under a nitrogen gas (N₂) flow and then diluted in methanol for chromatographic analysis.

The analyses of the two fractions (free and hydrolyzed phenolics), were performed in a high performance liquid chromatograph (HPLC Alliance Waters™ model 2690/5), with a Waters™ photodiode array detector model 2996 (270, 310 and 370 nm), a Thermo Hypersil BDS C₁₈ column (100 × 4.6 mm × 2.4 μm), a 1.0 mL min⁻¹ flow rate, an injection volume of 10 μL, a run time of 28 min, and an elution mode gradient using an aqueous solution of 0.0015% phosphoric acid (95%) and acetonitrile (5%). At 12:00 min the acetonitrile concentration was increased to 12%, at 18:00 min to 20%, and at 20:00 min to 50% acetonitrile. The acetonitrile concentration was maintained at 5% until 25:00 min and then returned to the initial condition (5%). The quantification of the flavonoids and phenolic acids was done using external standards.

Anthocyanin analysis

The anthocyanins were extracted using 0.01 g of the powder obtained in this work and 10 mL of methanol/formic acid solution (90:10 v/v) in an ultrasonic bath with subsequent centrifugation until discoloration of the solution (Brito *et al.*, 2007). Then, an aliquot of the extract was dried and diluted with 200 μL of 5% formic acid solution in water:methanol (90:10 v/v). Analyses were carried out in a Waters™ Alliance 2695 system, using a Waters™ 2996 photodiode array detector, with a Thermo™ Scientific C₁₈ BDS (100 mm × 4.6 mm; 2.4 μm) column, a flow rate of 1.0 mL min⁻¹, a column temperature of 40 °C, an injection volume of 20 μL and using a gradient elution method with acetonitrile and formic acid (Gouvêa *et al.*, 2015). Analyses were performed in triplicate.

The quantification of the main anthocyanins was carried out using external standards, based on calibration curves made with analytical standards isolated in the laboratory according to Gouvêa *et al.* (2012).

The anthocyanin stability of the peel powder was evaluated every 30 days, over a 4-month period. Time zero was the analysis immediately after the drying process. The samples were kept at room temperature, in individual aluminum and polyethylene packs and without exposure to light until analysis by HPLC.

The capsules were kept in the plastic packaging of high-density polyethylene, at room temperature and under light protection, for a period of 3 months. The capsules were analyzed in 4 times (T₀: analysis after encapsulation, T₁: anal-

ysis after 30 days, T₂: analysis after 60 days, T₃: analysis after 90 days). For each time, powders contained in 3 capsules were mixed and homogenized and analyzed in triplicate.

Degradation kinetic study

The anthocyanin stability study was evaluated by establishing a mathematic model for the deterioration kinetic. It was obtained the kinetic order and the degradation rate constant, which enabled the jabuticaba peel powder anthocyanins half-life (t_{1/2}) determination. The kinetic analysis was performed using the software R version 3.2.4 (R Foundation for Statistical Computing, Vienna, Austria).

Results and discussion

Fruit parts

The mean weight per fruit was 7.6 ± 0.6 g. The percentage of the jabuticaba fruit parts was 27.4 ± 5.1% peel, 56.8 ± 6.5% pulp and 15.8 ± 2.2% seed.

Convective drying process

The drying process of the jabuticaba peel showed a yield of 18% powder from the peels. The powder had an intense purple color (Figure 1) and was obtained using only the peel of the fruit, which contributed to the intense color of the same, which would have been lighter in hue if other parts of the fruit (pulp and seeds) had been used in the processing. This is a positive aspect that enhances the possible use of the powder obtained as a natural colorant. Furthermore, it was possible to obtain a powder with a high concentration of anthocyanins without prior extraction of these pigments.

Jabuticaba peel powder centesimal composition

The centesimal composition of the powder was determined in order to characterize and to establish references to allow future comparisons with powder obtained from other fruits, as well as different drying processes. The results were expressed as g 100 g⁻¹: moisture (11.21 ± 0.15), lipids (0.39 ± 0.04), ash (2.90 ± 0.01), protein (5.69 ± 0.02), dietary fiber (22.64 ± 0.00) and carbohydrate (57.17 ± 0.22).

As described by Santiago *et al.* (2016) for the jamelão peel powder, which is also a fruit from the *Myrtaceae* family, the jabuticaba peel powder contains high amounts of dietary fibers and also low lipid contents.

Antioxidant capacity

The jabuticaba peel powder antioxidant capacity by ABTS^{•+} assay gave a value of 659 μmol trolox g⁻¹. Rufino *et al.* (2010) reported an antioxidant activity of 317 μmol trolox g⁻¹ for freeze dried aqueous-organic extracts of jabuticaba; they also used the ABTS^{•+} assay.

The jabuticaba peel powder antioxidant capacity measured by ORAC assay was 992 μmol trolox g⁻¹, which is in accordance with the ORAC values (985.9 to 1792.3 μmol trolox g⁻¹) reported by Kang *et al.* (2012) for açaí, another Brazilian berry with high content of anthocyanins.

Wang *et al.* (1997) studied the relationship of the ORAC assay with different anthocyanins and observed that cyanidin-3-*O*-glycoside presented the highest antioxidant activity, most probably due to its hydroxylation pattern. This fact highlights even more the antioxidant capacity of the product obtained from the jabuticaba peel, which presents the cyanidin-3-*O*-glycoside anthocyanin representing more than 90% of its total monomeric anthocyanins as described below.

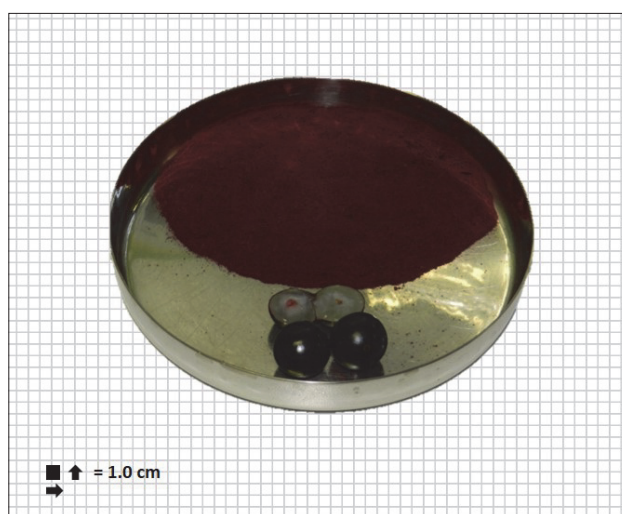


FIGURE 1. Jabuticaba fresh fruit and the powder obtained from its peel.

TABLE 1. Contents of the phenolic compounds (mg 100 g⁻¹) in jabuticaba peel powder. Results reported are mean values ± standard deviations (analysis in triplicate).

| Phenolic fractions | Rutin | Quercetin | Protocatechuic acid | Gallic acid | Ellagic acid | <i>p</i> -coumaric acid |
|--------------------|--------------|-------------|---------------------|----------------|----------------|-------------------------|
| Free | 11.13 ± 0.29 | 4.70 ± 0.17 | 5.53 ± 0.21 | 31.70 ± 2.36 | 120.87 ± 5.64 | 10.03 ± 0.21 |
| Hydrolyzed | 4.90 ± 0.44 | 3.07 ± 0.38 | 9.10 ± 1.11 | 385.67 ± 74.65 | 164.50 ± 12.80 | 3.67 ± 0.76 |
| Total | 16.03 | 7.77 | 14.63 | 417.37 | 285.37 | 13.70 |

Sugar analysis

These macronutrients are essential to the organism because they provide energy and essential components for the growth and maintenance of the body. Moreover, it is important to have information about the sugar content since their concentration in food can also influence the sensory evaluation for the development of new products.

The fructose (14.02 ± 0.11 g 100 g⁻¹) and glucose (12.39 ± 0.12 g 100 g⁻¹) sugars were detected in jabuticaba peel powder and they give the product a sweet taste, favorably influencing the acceptance by consumers.

Total carotenoids

The total content of carotenoids was 1,192.00 ± 33.40 µg 100 g⁻¹ in the jabuticaba peel powder. This powder showed a total content of carotenoids approximately seven times higher than the one found by Batista *et al.* (2016) for jambó (*Syzygium malaccense*) freeze-dried peel (158 µg 100 g⁻¹), which is a fruit belonging to the same family as jabuticaba. Romualdo *et al.* (2015) evaluated the chemical composition of açaí pulp powder (freeze dried pulp) and obtained a similar content for total carotenoids (1,112.67 ± 34.96 µg 100 g⁻¹).

Although the jabuticaba peel powder cannot be considered a rich source of carotenoids, the consumption of this product would help increasing the intake of these natural antioxidants.

Analysis of flavonoids and phenolic acids

Gallic and ellagic acids were the main phenolic acids

found in jabuticaba peel powder (Table 1; Figure 2). Their concentrations increased after the hydrolysis step, which suggests the presence of gallotannins and ellagitannins in the fruit. The presence of ellagitannins was corroborated by Alezandro *et al.* (2013), who also studied jabuticaba fruit.

According to Abe *et al.* (2012), the main sources of ellagic acid among fruits consumed by the Brazilian population are those belonging to the *Myrtaceae* family, such as jabuticaba. The quantities of ellagitannins decrease as the jabuticaba ripens, while the anthocyanins increase significantly. Jabuticaba, due to its very pleasant flavor, is a promising source of ellagic acid derivatives in the diet.

Other compounds, such as rutin, quercetin, protocatechuic acid and *p*-coumaric acid, although in lower concentrations, were also detected (Table 1; Figure 2). Such compounds, especially quercetin and their derivatives, are reported to be potentially beneficial to health (Coskun *et al.*, 2015).

The presence of these phenolic compounds in jabuticaba peel powder, as other bioactive compounds, such as anthocyanins and carotenoids, could bring potential health benefits since they contribute to the total antioxidant capacity of the product.

Anthocyanin analysis

The anthocyanins delphinidin-3-*O*-glucoside and cyanidin-3-*O*-glucoside were identified in the jabuticaba peel powder (Figure 3). Leite-Legatti *et al.* (2012) previously identified the same anthocyanin profile in the peel of jabuticaba fruit also from the Sabará species.

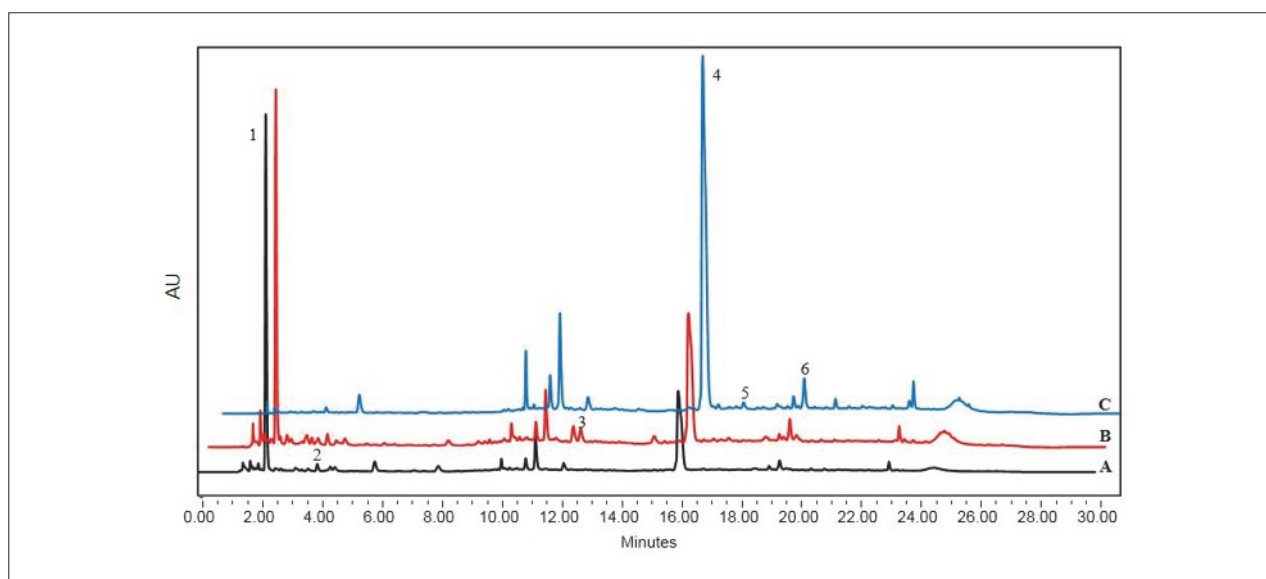


FIGURE 2. Chromatograms of the hydrolyzed phenolics present in jabuticaba peel powder: Peak identifications – peak 1: gallic acid; peak 2: protocatechuic acid; peak 3: *p*-coumaric acid; peak 4: ellagic acid; peak 5: rutin; peak 6: quercetin. In black (A): chromatogram extracted at 270 nm. In red (B): chromatogram extracted at 310 nm. In blue (C): chromatogram extracted at 370 nm. AU: Absorbance unit.

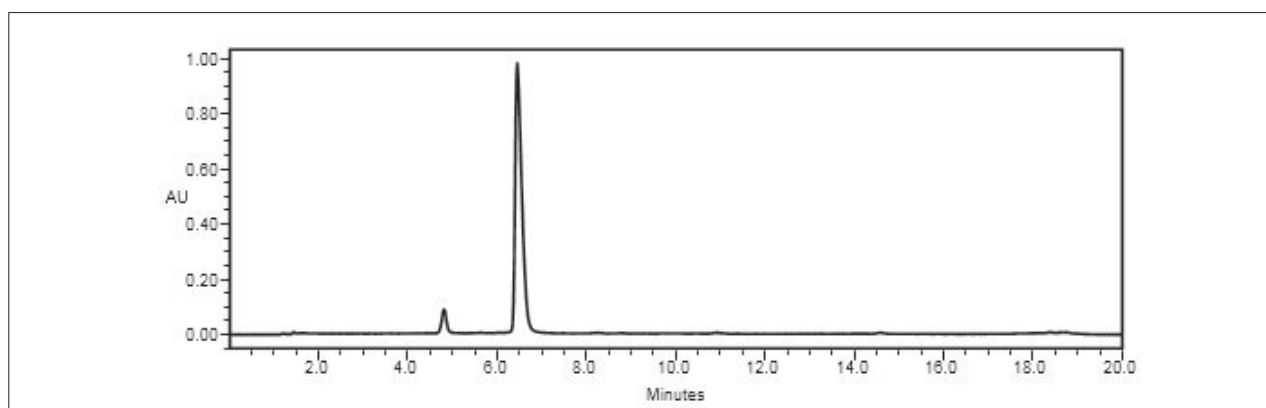


FIGURE 3. Chromatograms of the anthocyanins from jabuticaba peel powder. Peak identifications – peak 1: delphinidin-3-*O*-glucoside; peak 2: cyanidin-3-*O*-glucoside. AU: absorbance unit.

Silva *et al.* (2014) aimed to obtain a pigment powder from the aqueous extract of the jabuticaba depulping residue by spray-drying using maltodextrin as the carrier agent and to assess the potential of this pigment to act as an antioxidant and an antimicrobial agent. They suggest that the jabuticaba depulping residue could be used to produce a natural pigment with functional properties. However, the anthocyanins content found by these authors was much lower (7.00 to 21.60 mg 100 g⁻¹) than the one found in the present study (504.39 mg 100 g⁻¹), which was attributed to the dilution caused by the addition of encapsulating agents required for the drying process used. This fact further enhances the process of convective drying used in this study to obtain natural dye since it is a simple and low-cost technology that preserves the bioactive compounds.

The mathematical model used to evaluate the anthocyanins stability showed that these compounds degradation followed a zero order reaction kinetics, which characterized

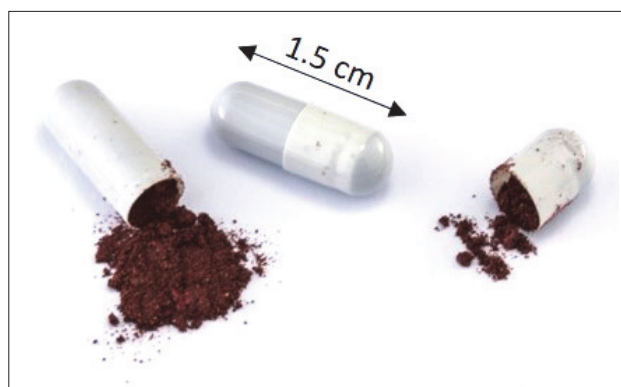


FIGURE 4. Capsules prepared with jabuticaba peel powder.

a linear regression behavior (linear equation $y = 0.3825 \times -1.9827$; correlation coefficient $r^2 = 0.970888$), where the anthocyanins content decreases with higher storage times. The obtained results showed a degradation constant (k) of 0.3825 (mg 100 g⁻¹ day⁻¹) and a half-life ($t_{1/2}$) of 665.7 days for the jabuticaba peel powder anthocyanins, which indicates that the powder can maintain its quality as a source of anthocyanins for a long period of time (Table 2).

The bioaccessibility of the anthocyanins of the jabuticaba peel powder obtained in the present study was already evaluated by Peixoto *et al.* (2016) under gastro-intestinal conditions. The gastric and intestinal absorption of anthocyanins were also examined for the same authors in order to estimate anthocyanin transport efficiency. *In vitro* digestion coupled to epithelium absorptive models (gastric MKN-28 and intestinal Caco-2) was applied independently for each phase. The bioaccessibility of anthocyanins after gastric digestion was 13%, whereas intestinal bioaccessibility was 10%. The assays showed that, applying the MKN-28 cell model, the anthocyanin transport efficiency was 19.7%, whereas using the Caco-2 cell model this result was 0.8%. In conclusion gastric mucosa plays an important role in anthocyanin's absorption.

The capsules prepared with jabuticaba peel powder (Figure 4) showed a high content of anthocyanins during the 3 months of storage with an average retention of 80% of these compounds, showing a good potential for use as a nutraceutical. Complementary evaluations, such as different capsules materials and longer stability studies, are still necessary.

TABLE 2. Anthocyanin stability evaluation of jabuticaba peel powder. Reported contents (in mg 100 g⁻¹) are mean values \pm standard deviations (analysis in triplicate).

| Storage time (days) | Delphinidin-3- <i>O</i> -glucoside | Cyanidin-3- <i>O</i> -glucoside | Total monomeric anthocyanin |
|---------------------|------------------------------------|---------------------------------|-----------------------------|
| 0 | 34.39 \pm 0.14 | 470.00 \pm 20.68 | 504.39 \pm 20.54 |
| 30 | 32.03 \pm 0.20 | 469.40 \pm 1.41 | 501.43 \pm 1.21 |
| 60 | 31.88 \pm 0.75 | 467.10 \pm 28.12 | 498.98 \pm 28.87 |
| 90 | 31.44 \pm 0.32 | 448.14 \pm 20.41 | 479.60 \pm 20.70 |
| 120 | 30.24 \pm 0.90 | 444.59 \pm 37.88 | 474.83 \pm 36.94 |

Conclusion

Jaboticaba peel powder studied in this work presented a great variety of bioactive compounds in its composition, such as anthocyanins, other flavonoids, phenolic acids and carotenoids. The high stability of its anthocyanin content adds value to this product, since this is an important characteristic for the possible use as a nutraceutical and/or a natural colorant.

In terms of the increasing worldwide demand for healthier products, this powder can be considered a natural functional ingredient, which could substitute synthetic dyes and at the same time be a source of bioactive compounds, especially anthocyanins and ellagic acid. These information can contribute to add value to this Brazilian underutilized fruit and to promote its productive chain. Jaboticaba could be considered as a super fruit, like açai, once its peel is rich in anthocyanins and other bioactive compounds and is normally discarded during the *in natura* fruit consumption.

References

- Abe, L.T., Lajolo, F.M., and Genovese, M.I. (2012). Potential dietary sources of ellagic acid and other antioxidants among fruits consumed in Brazil: Jaboticaba (*Myrciaria jaboticaba* (Vell.) Berg). *J. Sci. Food and Agric.* *92*, 1679–1687. <https://doi.org/10.1002/jsfa.5531>.
- Alezandro, M.R., Dubé, P., Desjardins, Y., Lajolo, F.M., and Genovese, M.I. (2013). Comparative study of chemical and phenolic compositions of two species of jaboticaba: *Myrciaria jaboticaba* (Vell.) Berg and *Myrciaria cauliflora* (Mart.) O. Berg. *Food Res. Int.* *54*, 468–477. <https://doi.org/10.1016/j.foodres.2013.07.018>.
- Association of Official Analytical Chemists (AOAC) (2010). *Official Methods of Analysis of AOAC International*. 18th edn, 3rd rev. (Gaithersburg, MD, USA: AOAC).
- Batista, A.G., Silva, J.K., Cazarin, C.B.B., Biasoto, A.C.T., Sawaya, A.C.H.F., Prado, M.A., and Maróstica Júnior, M.R. (2016). Red-jambo (*Syzygium malaccense*): bioactive compounds in fruits and leaves. *LWT Food Sci. and Technol.* *76*, 284–291. <https://doi.org/10.1016/j.lwt.2016.05.013>.
- Batista, A.G., Lenquiste, S.A., Cazarin, C.B.B., Silva, J.K., Luiz-Ferreira, A., Bogusz Jr, S., Hantao, L.W., Souza, R.N., Augusto, F., Prado, M.A., and Maróstica Jr, M.R. (2014). Intake of jaboticaba peel attenuates oxidative stress in tissues and reduces circulating saturated lipids of rats with high-fat diet-induced obesity. *J. Funct. Foods* *6*, 450–461. <https://doi.org/10.1016/j.jff.2013.11.011>.
- Brito, E.S., Araujo, M.C.P., Alves, R.E., Carkeet, C., Clevidence, B.A., and Novoty, J.A. (2007). Anthocyanins present in selected tropical fruits: acerola, jambolão, jussara and guajiru. *J. Agric. and Food Chem.* *55*, 9389–9394. <https://doi.org/10.1021/jf0715020>.
- Coskun, O., Kanter, M., Korkmaz, A., and Oter, S. (2005). Quercetin, a flavonoid antioxidant, prevents and protects streptozotocin-induced oxidative stress and β -cell damage in rat pancreas. *Pharm. Res.* *51*, 117–123. <https://doi.org/10.1016/j.phrs.2004.06.002>.
- Fernandes, I., de Freitas, V., Reis, C., and Mateus, N. (2012). A new approach on the gastric absorption of anthocyanins. *Food Funct.* *3*, 508–516. <https://doi.org/10.1039/c2fo10295a>.
- Frighetto, R.T.S., and Bacchan, M. (2012). Quantification of constitutive phenolic acids of soybean [*Glycine max* (L.) Merrill] by high performance liquid chromatography (HPLC). *Embrapa Meio Ambiente: Jaguariúna* (in Portuguese).
- Gouvêa, A.C.M.S., Santiago, M.C.P.A., Pacheco, S., Godoy, R.L.O., and Cabral, L.M.C. (2012). Anthocyanins standards (cyanidin-3-O-glucoside and cyanidin-3-O-rutinoside) isolation from freeze-dried açai (*Euterpe oleracea* Mart.) by HPLC. *Food Sci. and Technol.* *32*, 43–46. <https://doi.org/10.1590/S0101-20612012005000001>.
- Gouvêa, A.C.M.S., Melo, A., Santiago, M.C.P.A., Peixoto, F.M., Freitas, V., Godoy, R.L.O., and Ferreira, I.M.P.L.V.O. (2015). Identification and quantification of anthocyanins in fruits from *Neomitranthes obscura* (DC.) N. Silveira, an endemic specie from Brazil by comparison of chromatographic methodologies. *Food Chem.* *185*, 277–283. <https://doi.org/10.1016/j.foodchem.2015.02.086>.
- Gurak, P.D., Bona, G.S., Tessaro, I.C., and Marczak, L.D.F. (2014). Jaboticaba pomace powder obtained as a co-product of juice extraction: a comparative study of powder obtained from peel and whole fruit. *Food Res. Int.* *62*, 786–792. <https://doi.org/10.1016/j.foodres.2014.04.042>.
- Kang, J., Thakali, K.M., Xie, C., Kondo, M., Tong, Y., Ou, B., Jensen, G., Medina, M.B., Schauss, A.G., and Wu, X. (2012). Bioactivities of açai (*Euterpe precatoria* Mart.) fruit pulp, superior antioxidant and anti-inflammatory properties to *Euterpe oleracea* Mart. *Food Chem.* *133*, 671–677. <https://doi.org/10.1016/j.foodchem.2012.01.048>.
- Leite, A.V., Malta, L.G., Riccio, M.F., Eberlin, M.N., Pastore, G.M., and Maróstica Júnior, M.R. (2011). Antioxidant potential of rat plasma by administration of freeze-dried jaboticaba peel (*Myrciaria jaboticaba* Vell. Berg). *J. Agric. and Food Chem.* *59*, 2277–2283. <https://doi.org/10.1021/jf103181x>.
- Leite-Legatti, A.V., Batista, A.G., Dragano, N.R.V., Marques, A.C., Malta, L.G., Riccio, M.F., Eberlin, M.N., Machado, A.R.T., Carvalho-Silva, L.B., Ruiz, A.L.T.G., Carvalho, J.E., Pastore, G.M., and Maróstica Júnior, M.R. (2012). Jaboticaba peel: antioxidant compounds, antiproliferative and antimutagenic activities. *Food Res. Int.* *49*, 596–603. <https://doi.org/10.1016/j.foodres.2012.07.044>.
- Lorenzi, H., Bacher, L., Lacerda, M., and Sartori, S. (2006). *Brazilian Fruits & Cultivated Exotics (for consuming in natura)* (São Paulo: Instituto Plantarum de Estudos da Flora).
- Macrae, R. (1998). *HPLC in Food Analysis. Food Science and Technology – A Series of Monographs*: (New York: Academic Press).
- Peixoto, F.M., Fernandes, I., Gouvêa, A.C.M.S., Santiago, M.C.P.A., Borguini, R.G., Mateus, N., Freitas, V., Godoy, R.L.O., and Ferreira, I.M.P.L.V.O. (2016). Simulation of *in vitro* digestion coupled to gastric and intestinal transport models to estimate absorption of anthocyanins from peel powder of jaboticaba, jamelão and jambo fruits. *J. Funct. Foods* *24*, 373–381. <https://doi.org/10.1016/j.jff.2016.04.021>.
- Pérez-Jiménez, J., Arranz, S., Taberner, M., Díaz-Rubio, M.E., Serrano, J., Goñi, I., and Saura-Calixto, F. (2008). Updated methodology to determine antioxidant capacity in plant foods, oils and beverages: Extraction, measurement and expression of results. *Food Res. Int.* *41*, 274–285. <https://doi.org/10.1016/j.foodres.2007.12.004>.
- Rodríguez-Amaya, D.B. (2001). *A Guide to Carotenoid Analysis in Foods* (Washington: International Life Sciences Institute).
- Romualdo, G.R., Fragoso, M.F., Borguini, R.G., Santiago, M.C.P.A., Fernandes, A.A.H., and Barbisan, L.F. (2015). Protective effects of spray-dried açai (*Euterpe oleracea* Mart) fruit pulp against initiation step of colon carcinogenesis. *Food Res. Int.* *77*, 432–440. <https://doi.org/10.1016/j.foodres.2015.08.037>.
- Rufino, M.S.M., Alves, R.E., Brito, E.S., Pérez-Jiménez, J., Saura-Calixto, F., and Mancini-Filho, J. (2010). Bioactive compounds and antioxidant capacities of 18 non-traditional tropical fruits from Brazil. *Food Chem.* *121*, 996–1002. <https://doi.org/10.1016/j.foodchem.2010.01.037>.
- Santiago, M.C.P.A., Gouvêa, A.C.M.S., Peixoto, F.M., Borguini, R.G., Godoy, R.L.O., Pacheco, S., Nascimento, L.S.M., and Nogueira, R.I. (2016). Characterization of jamelão (*Syzygium cumini* (L.) Skeels) fruit peel powder for use as natural colorant. *Fruits* *71*, 3–8. <https://doi.org/10.1051/fruits/2015041>.

Silva, M.C., Souza, V.B., Thomazini, M., Silva, E.R., Smaniotto, T., Carvalho, R.A., Genovese, M.I., and Favaro-Trindade, C.S. (2014). Use of the jaboticaba (*Myrciaria cauliflora*) depulping residue to produce a natural pigment powder with functional properties. *LWT – Food Sci. and Technol.* *55*, 203–209. <https://doi.org/10.1016/j.lwt.2013.08.026>.

Silva, P.I., Stringheta, P.C., Teófilo, R.F., and Oliveira, I.R.N. (2013). Parameter optimization for spray-drying microencapsulation of jaboticaba (*Myrciaria jaboticaba*) peel extracts using simultaneous analysis of responses. *J. Food Engin.* *117*, 538–544. <https://doi.org/10.1016/j.jfoodeng.2012.08.039>.

Wang, H., Cao, G., and Prior, R.L. (1997). Oxygen radical absorbing capacity of anthocyanins. *J. Agric. Food Chem.* *45*, 304–309. <https://doi.org/10.1021/jf960421t>.

Wu, S., Dastmalchi, K., Long, C., and Kennelly, E.J. (2012). Metabolite profiling of Jaboticaba (*Myrciaria cauliflora*) and other dark-colored fruit juices. *J. Agric. and Food Chem.* *60*, 7513–7525. <https://doi.org/10.1021/jf301888y>.

Zeisel, S.H. (1999). Regulation of 'Nutraceuticals'. *Science* *285*, 1853–1855. <https://doi.org/10.1126/science.285.5435.1853>.

Zulueta, A., Esteve, M.J., and Frígola, A. (2009). ORAC and TEAC assays comparison to measure the antioxidant capacity of food products. *Food Chem.* *114*, 310–316. <https://doi.org/10.1016/j.foodchem.2008.09.033>.

Received: Dec. 20, 2017

Accepted: May 15, 2018