Food Chemistry 121 (2010) 996–1002

Contents lists available at ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

Bioactive compounds and antioxidant capacities of 18 non-traditional tropical fruits from Brazil

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ARTICLE INFO

Article history:
Received 11 April 2009
Received in revised form 16 November 2009
Accepted 21 January 2010

Keywords:
Tropical fruits
Phenolics
Antioxidants
ABTS
DPPH
FRAP
β-Carotene bleaching

ABSTRACT

The bioactive compounds and antioxidant capacities of polyphenolic extracts of 18 fresh and dry native non-traditional fruits from Brazil were determined using ABTS, DDPH, FRAP and β-carotene bleaching methods. The study provides an adaptation of these methods, along with an evaluation of the compounds related to antioxidant potential. The results show promising perspectives for the exploitation of non-traditional tropical fruit species with considerable levels of nutrients and antioxidant capacity. Although evaluation methods and results reported have not yet been sufficiently standardised, making comparisons difficult, our data add valuable information to current knowledge of the nutritional properties of tropical fruits, such as the considerable antioxidant capacity found for acerola – *Malpighia emarginata* and camu-camu – *Myrciaria dubia* (ABTS, DPPH and FRAP) and for puçá-preto – *Mouriri pusa* (all methods).

1. Introduction

Tropical America is home to a great variety of fruit species and some of them have long been domesticated by native Amerindians. Species richness is associated with the geographical characteristics of the region, especially the heterogeneity of North and South America flora and partial overlapping between the Amazon region and lower Central America. A list of fruits from the tropics, including America, Asia, Australia and Africa, mentions over to 2000 species. In America alone, about one thousand species, belonging to 80 families, have been identified; of these, at least 400 occur in or stem from Brazil (Alves, Brito, Rufino, & Sampaio, 2008; Donadio, 1993; Martin, Campbell, & Ruberté, 1987).

Tropical fruit consumption is increasing on the domestic and international markets due to growing recognition of its nutritional and therapeutic value. Brazil boasts a large number of underexploited native and exotic fruit species of potential interest to the agroindustry and a possible future source of income for the local population. These fruits represent an opportunity for local growers to gain access to special markets where consumers lay emphasis on exotic character and the presence of nutrients capable of preventing degenerative diseases (Alves, Brito et al., 2008). Fruit consumption is no longer merely a result of taste and personal preference, but has become a concern of health due to the vital fruit nutrients content. In addition to essential nutrients, most fruits feature considerable amounts of micronutrients, such as minerals, fibres, vitamins and secondary phenolic compounds. Increasing evidence shows the importance of these micronutrients for human health. (Vasco, Ruales, & Kamal-Eldin, 2008; Veer, Jansen, Klerk, & Kok, 2000).

Over time, the different methodologies employed to evaluate antioxidant capacity *in vitro* have yielded conflicting and non-comparable results. Variations in sample preparation may also have affected results greatly and this is a problem deserving attention from researchers. Antioxidant capacity may be expressed using several different parameters, including peroxyl radical-scavenging capacity (ORAC – oxygen radical absorbance capacity, TRAP – total reactive antioxidant potential), metal reduction capacity (FRAP – ferric reducing antioxidant power, CUPRAC – cupric ion reducing antioxidant capacity), hydroxyl radical-scavenging capacity (the deoxyribose method), organic radical-scavenging capacity (ABTS

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doi:10.1016/j.foodchem.2010.01.037

– 2,2′-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid), DPPH – 2,2-diphenyl-1-picrylhydrazil) and amounts of lipid peroxidation products (TBARS, LDL oxidation, β-carotene co-oxidation) (Aruoma, 2003; Sánchez-Moreno, 2002).

FRAP, ABTS, DPPH and ORAC are the most widely used methods for determining antioxidant capacity *in vitro*. It is recommended that at least two (or even all) of these assays be combined to provide a reliable picture of the total antioxidant capacity of a foodstuff, provided the strengths, weaknesses and applicability of each type of assay are taken into account (Pérez-Jiménez et al., 2008). The β-carotene bleaching method is also popular. It evaluates the level of inhibition of free radicals generated during linoleic acid peroxidation (Duarte-Almeida, Santos, Genovese, & Lajolo, 2008). The aim of this work was to characterise the antioxidant capacity, along with a quantification of the major bioactive compounds, found in some underutilised tropical fruits from Brazil.

2. Materials and methods

2.1. Chemical reagents

The reagents used were 2,6-dichloroindophenol (DFI), 2,2-diphenyl-1-picrylhydrazil (DPPH·), 2,2′-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid) (ABTS·), 6-hydroxy-2,5,7,8-tetramethyl chroman-2-carboxylic acid (trolox), and 2,4,6-tris(2-pyridyl)-s-triazine (TPTZ) from Sigma Chemical Co., in addition to potassium persulfate from Acrós Organics, ferrous sulphate from Vetec and β-carotene from Merck.

2.2. Sample preparation

Table 1 shows the 18 fruits included in the study, along with their respective botanical identifications and geographical origins.

Table 1
List of the 18 tropical non-traditional Brazilian fruits included in the study.

Common name	Species	Family	Origin (City, State)
Açaí, assai	*Euterpe oleracea*	Arecaceae	Paraipaba, Ceará
Acerola	*Malpighia emarginata*	Malpighiaceae	Limoeiro do Norte, Ceará
Bacuri	*Platonia insignis*	Clusiaceae	Coelho Neto, Maranhão
Cajá, yellow mombim	*Spondias mombin*	Anacardiaceae	Limoeiro do Norte, Ceará
Caju, cashew apple	*Anacardium occidentale*	Anacardiaceae	Pacajus, Ceará
Camu-camu	*Myrciaria dubia*	Myrtaceae	Belém, Pará
Carnaúba	*Copernicia prunifera*	Arecaceae	Maracanaú, Ceará
Gurguri	*Mouriri guianensis*	Melastomataceae	Beberibe, Ceará
Jaboticaba	*Myrciaria cauliflora*	Myrtaceae	Serra de Ibiapaba, Ceará
Jambolão, java plum	*Syzygium cumini*	Myrtaceae	Trairi, Ceará
Juçara, jussara	*Euterpe edulis*	Arecaceae	São Paulo, São Paulo
Mangaba	*Hancornia speciosa*	Apocynaceae	Ipiranga, Piauí
Murici, nance	*Byrsonima dealbata*	Malpighiaceae	Fortaleza, Ceará
Murta	*Blepharocalyx salicifolius*	Myrtaceae	Crato, Ceará
Puçá-coroa-de-frade	*Mouriri elliptica*	Melastomataceae	Beberibe, Ceará
Puçá-preto	*Mouriri pusa*	Melastomataceae	Ipiranga, Piauí
Umbu	*Spondias tuberosa*	Anacardiaceae	Picos, Piauí
Uvaia	*Eugenia pyriformis*	Myrtaceae	Paraipaba, Ceará

The fruits were harvested and sent to the laboratory for pulp extraction. Two fruits (assai and jussara) required a special processing due to their highly fibrous epicarp and endocarp. Pulp and fibre were mechanically separated and weighed, and distilled water was added (1:2). The mass was homogenised and the inedible parts (fibre and pit) were discarded. Bacuri pulp was extracted manually with a knife and scissors, and the husk and seed were discarded. For the other 15 fruits, the pulp and peel were processed and only the seeds were discarded.

The bioactive compounds were determined by the following methodologies: vitamin C by the 2,6-dichlorophenol indophenol method (Strohecker & Henning, 1967), total anthocyanins and yellow flavonoids as described by Francis (1982), total carotenoids as by Higby (1962) and chlorophyll, following the procedure of Bruinsma (1963).

In preparation for the antioxidant assay, part of the pulp was kept fresh while the remainder was freeze-dried and stored at −80 °C prior to extraction and analysis. The moisture content was determined for all fruits (Table 2).

Due to different bioactive concentrations among the fruits, we performed pretesting for extractable polyphenols and total antioxidant capacity in order to determine the ideal sample size (g) for each method. The final fresh and dry weights for extraction were, respectively, assai: 5 g and 2 g; acerola: 2 g and 0.2 g; bacuri: 40 g and 10 g; camu-camu: 1 g and 0.2 g; carnauba: 20 g and 10 g; cashew apple: 20 g and 2 g; gurguri: 5 g and 4 g; jaboticaba: 3 g and 0.5 g; java plum: 5 g and 2 g; jussara: 2 g and 2 g; mangaba: 10 g and 2 g; murta: 5 g and 4 g; puçá-coroa-de-frade: 5 g and 2 g; puçá-preto: 5 g and 2 g; umbu: 20 g and 2 g; and uvaia: 6 g and 0.6 g; yellow mombin: 30 g and 4 g. The antioxidant capacity could not be determined for fresh samples of nance due to interference from oil contents. Thus, testing was limited to a dry sample weighing 0.8 g.

2.3. Extraction of antioxidants

The procedure developed by Larrauri, Rupérez, and Saura-Calixto (1997) was employed and is briefly described as follows: fresh and lyophilised samples from the pretesting stage were weighed (g) in centrifuge tubes and extracted sequentially with 40 ml of methanol/water (50:50, v/v) at room temperature for 1 h. The tubes were centrifuged at 25,400*g* for 15 min and the supernatant was recovered. Then 40 ml of acetone/water (70:30, v/v) was added to the residue at room temperature, extracted for 60 min and centrifuged. Methanol and acetone extracts were combined, made up to 100 ml with distilled water and used to determine antioxidant capacity and extractable polyphenol contents (Fig. 1).

2.4. Total phenolics determination

Total polyphenols were determined by the Folin–Ciocalteu method (Obanda & Owuor, 1997) in supernatant. Extracts (1.0 ml) were mixed with 1 ml of Folin–Ciocalteu reagent (1:3), 2 ml of 20% sodium carbonate solution and 2 ml of distilled water. After 1 h, absorbance at 700 nm was read in the spectrophotometer. Results were expressed as g gallic acid equivalents (GAE)/100 g.

2.5. $ABTS^{\cdot+}$ assay

The $ABTS^{\cdot+}$ assay was based on a method developed by Miller et al. (1993) with modifications. $ABTS^{\cdot+}$ radical cations were produced by reacting 7 mM ABTS stock solution with 145 mM potassium persulfate and allowing the mixture to stand in the dark at room temperature for 12–16 h before use. The $ABTS^{\cdot+}$ solution was diluted with ethanol to an absorbance of 0.70 ± 0.02 at

Table 2
Bioactive compounds (mg/100 g fresh matter[a]) and humidity (%) in 18 non-traditional Brazilian tropical fruits.

Fruits	Vitamin C	Total anthocyanins	Yellow flavonoids	Total carotenoids	Chlorophyll	Moisture
Açaí, Assai	84.0 ± 10	111 ± 30.4	91.3 ± 20.6	2.8 ± 0.4	20.8 ± 3.8	84.1 ± 2.8
Acerola	1357 ± 9.5	18.9 ± 0.9	9.6 ± 1.4	1.4 ± 0.1	n.d.	91.0 ± 0.2
Bacuri	2.4 ± 0.3	0.3 ± 0.2	16.9 ± 1.7	–	n.d.	73.7 ± 8.0
Cajá, yellow mombim	26.5 ± 0.5	–	7.1 ± 0.7	0.7 ± 0.0	n.d.	86.4 ± 0.9
Caju, cashew apple	190 ± 5.7	9.5 ± 4.6	63.8 ± 26.5	0.4 ± 0.1	n.d.	86.9 ± 0.6
Camu-camu	1882 ± 43.2	42.2 ± 17.0	20.1 ± 4.4	0.4 ± 0.0	n.d.	89.8 ± 0.5
Carnaúba	78.1 ± 2.6	4.1 ± 0.1	66.4 ± 2.3	0.6 ± 0.2	4.2 ± 0.2	70.7 ± 0.6
Gurguri	27.5 ± 0.2	3.3 ± 0.2	41 ± 1.5	4.7 ± 0	n.d.	74.7 ± 3.7
Jaboticaba	238 ± 2.2	58.1 ± 0.9	147 ± 42.5	0.32 ± 0.1	n.d.	85.9 ± 0.4
Jambolão, java plum	112 ± 5.8	93.3 ± 3.4	70.9 ± 1.2	0.51 ± 0.1	0.9 ± 0.2	84.9 ± 0.3
Juçara, Jussara	186 ± 43.3	192 ± 43.2	375 ± 87.6	1.9 ± 0.5	21.5 ± 4.1	90.2 ± 1.3
Mangaba	190 ± 1.91	0.4 ± 0.11	15 ± 1.1	0.3 ± 0.05	n.d.	90.8 ± 1.2
Murici, nance	148 ± 4.0	0.5 ± 0.1	13.8 ± 0.5	1.1 ± 0.1	n.d.	60.6 ± 0.7
Murta	181 ± 1.8	143 ± 0.5	207 ± 8.2	0.5 ± 0.1	5.0 ± 0.5	74.1 ± 2.2
Puçá-coroa-de-frade	41.1 ± 6.7	3.7 ± 0.8	17.7 ± 2.0	3.4 ± 0.1	n.d.	62.6 ± 0.2
Puçá-preto	28.9 ± 1.4	103 ± 21.6	143 ± 12.6	4.2 ± 0.4	5.6 ± 1.1	64.1 ± 0.8
Umbu	18.4 ± 1.8	0.3 ± 0.2	6.9 ± 1.7	1.0 ± 0.2	n.d.	87.9 ± 0.1
Uvaia	39.3 ± 5.2	1.13 ± 0.1	17.5 ± 1.6	1.7 ± 0.1	n.d.	89.3 ± 1.2

[a] Mean value ± standard deviation; $n = 3$; n.d. = not determined.

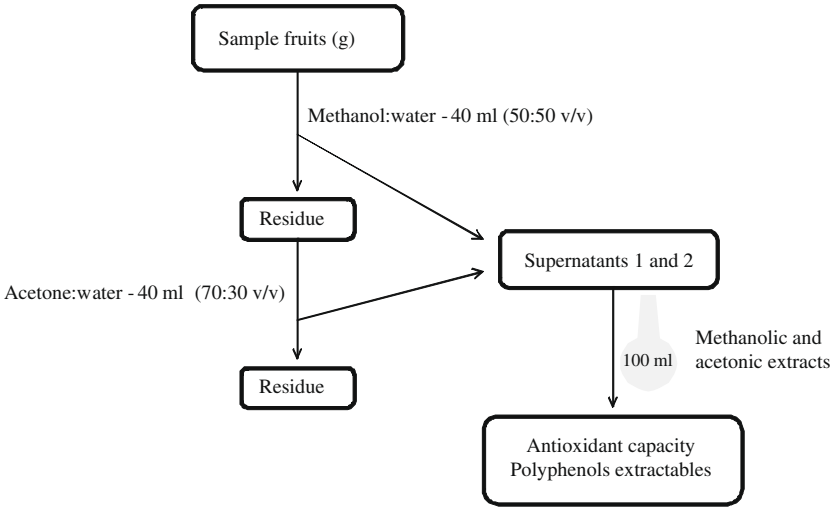

Fig. 1. Flow chart showing determination of antioxidant capacity of aqueous-organic extracts.

734 nm. After the addition of 30 μl of sample or trolox standard to 3 ml of diluted $ABTS^{\cdot+}$ solution, absorbances were recorded at 6 min after mixing. Ethanolic solutions of known trolox concentrations were used for calibration and the results were expressed as μM trolox/g fruit.

2.6. DPPH· (free radical-scavenging) assay

The antioxidant capacity was determined by the modified DPPH· method (Brand-Williams, Cuvelier, & Berset, 1995) which is based on the quantification of free radical-scavenging with modifications. A methanol solution containing 0.06 mM DPPH· was prepared. After adjusting the blank with methanol, an aliquot of 100 μl of fruit extract was added to 3.9 ml of this solution. The decrease in absorbance at 515 nm was measured at 1 min intervals for the first 10 min, and then at 5 min intervals until stabilisation. Based on preliminary study, the times required to obtain DPPH· readings of each fruit were as follows: assai, 120 min; acerola, 10 min; bacuri, 180 min; caju, 30 min; camu-camu, 5 min; carnauba, 120 min; gurguri, 60 min; jaboticaba, 60 min; java plum, 90 min; jussara, 60 min; mangaba, 30 min; murta, 30 min; nance, 240 min; puçá-coroa-de-frade, 120 min; puçá-preto, 150 min; umbu, 180 min; uvaia, 120 min; yellow mombin, 180 min. The antioxidant capacity was expressed as the concentration of antioxidant required to reduce the original amount of free radicals by 50% (EC_{50}) and values expressed as g fruit/g DPPH·.

2.7. Ferric reducing antioxidant power (FRAP) assay

The antioxidant capacity of each sample was estimated by FRAP assay, following the procedure described in the literature (Benzie & Strain, 1996) with modifications. Briefly, 2.7 ml of freshly prepared FRAP reagent (TPTZ, $FeCl_3$ and acetate buffer) at 37 °C was mixed with 90 μl of fruit extract and 270 μl of distilled water. Using a blank containing FRAP reagent as reference, absorbance at 595 nm was determined at 30 min. Aqueous solutions of known Fe (II) concentrations in the range of 100–1500 μM (Fe_2SO_4) were used for calibration.

2.8. β-Carotene bleaching method

The antioxidant capacity of each sample was estimated by the β-carotene bleaching method, following the procedure described in the literature (Marco, 1968) with modifications. The spectropho-

tometric assay is based on β-carotene oxidation (discolouring) induced by the products from linoleic acid oxidative degradation. Solutions were prepared by mixing 5 ml of β-carotene/linoleic acid system solution and 0.4 ml of fruit extract/trolox solutions at different concentrations. The mix was kept in a water bath at 40 °C. Spectrophotometric readings were made at 470 nm 2 min after the mixing and then at 15–120 min intervals. Results were expressed as oxidation inhibition percentages, as the absorbance of successive samples decreased in relation to trolox.

2.9. Statistical analysis

Assays were performed in triplicate for each sample. Results were expressed as mean values ± standard deviation (SD). To determine whether the bioactive compounds contributed to the antioxidant capacity, Pearson's correlation coefficients were calculated, at 1% and 5% probability, using the Student's *t* test for all variables.

3. Results and discussion

3.1. Quantification of bioactive compounds

The fruits included in this study play an important economic role, either in the international market or locally in certain countries of tropical America.

The results for vitamin C, total anthocyanins, yellow flavonoids, total carotenoids and chlorophyll obtained in this research are shown in Table 2 and reveal that most of the fruits contained considerable amounts of vitamin C, especially camu-camu (1882 mg/100 g) and acerola (1357 mg/100 g).

Camu-camu is considered a highly nutritious food. Other authors (Alves, Filgueiras, Moura, Araújo, & Almeida, 2002) have reported vitamin C contents (2600 mg/100 g pulp) for this fruit even higher than the levels observed in the present study. Acerola is almost as rich in vitamin C as is camu-camu, because the content of vitamin C can vary from 0.8% to 3.5% (Alves, Chitarra, & Chitarra, 1995; Alves, Filgueiras, Mosca, & Menezes, 1999; Alves, Filgueiras, Mosca, Silva, & Menezes, 2008).

Anthocyanins are brightly-coloured compounds responsible for much of the red, blue, and purple colouring of fruits. They are especially abundant in berries such as blueberries and blackcurrants (Kähkönen, Hopia, & Heinonen, 2001). Assai and jussara, sometimes called palm berry, display a characteristic purplish-black colouring due to their large contents of anthocyanins (111 and 192 mg/100 g), flavonoids (91.3 and 375 mg/100 g) and chlorophyll (20.8 and 21.5 mg/100 g), respectively. Pozo-Insfran, Brenes, and Talcott (2004) concluded that anthocyanins were the predominant contributing factor to the antioxidant capacity of assai, which was found to be higher than that of muscadine grape juice and that of several berries, such as high-bush blueberries, strawberries, raspberries, blackberries and cranberries.

Puçá-preto was shown to be an excellent source of total anthocyanins (103 mg/100 g) as were the myrtaceans, murta (143 mg/100 g), java plum (93.3 mg/100 g), jaboticaba (58.1 mg/100 g) and camu-camu (42.2 mg/100 g), with levels comparable to those of other well-known fruit sources of anthocyanins. These values are in the same range as those reported for tropical fruits. In comparison, strawberries contain 21 mg/100 g, red grapes 27 mg/100 g, red raspberries 92 mg/100 g, cherries 122 mg/100 g, blackberries 245 mg/100 g and cultivated blueberries 387 mg/100 g (Wu et al., 2006).

Carotenoids are not only important vitamin A precursors, but display a considerable level of antioxidant activity. The fruits included in this study contained carotenoids in the range 0.3 mg/100 g (mangaba) to 4.7 mg/100 g (gurguri). The latter is, by any standards, a rich source of carotenoids. The most obvious exception is perhaps the wine palm (*Mauritia vinifera*; 48.9 mg/100 g), one of the most important vitamin A precursors in the Brazilian flora (Godoy & Rodriguez-Amaya, 1998).

3.2. Polyphenols and antioxidant capacity

3.2.1. Extractable polyphenols

The amount of extractable polyphenols varied greatly among the fruit species (Tables 3 and 4). Following the example of Vasco et al. (2008), who tested 17 fruits from Ecuador for polyphenol contents, we classified our fruits into three categories: low (<100 mg GAE/100 g), medium (100–500 mg GAE/100 g) and high (>500 mg GAE/100 g) for samples based on fresh matter, and low (<1000 mg GAE/100 g), medium (1000–5000 mg GAE/100 g) and high (>5000 mg GAE/100 g) on dry matter.

Table 3
Polyphenols and antioxidant capacity in aqueous-organic extracts of 18 non-traditional Brazilian tropical fruits based on fresh matter.[a]

Fruits	Extractable polyphenols mg GAE/100 g	DPPH· EC_{50} (g/g DPPH·) [b]	$ABTS^{\cdot+}$ μmol trolox/g	FRAP μmol Fe_2SO_4/g	β-Carotene bleaching % O.I. [c]
Açaí, assai	454 ± 44.6	4264 ± 1381	15.1 ± 4.1	32.1 ± 6.5	31.9 ± 3.2
Acerola	1063 ± 53.1	670 ± 64.5	96.6 ± 6.1	148 ± 16	n.d.
Bacuri	23.8 ± 0.7	n.d.	n.d.	n.d.	n.d.
Cajá, yellow mombim	72.0 ± 4.4	9397 ± 64.8	7.8 ± 0.2	11.8 ± 0.2	92.7 ± 1.1
Caju, cashew apple	118 ± 3.7	7142 ± 205	11.2 ± 0.04	22.9 ± 0.7	25 ± 8.9
Camu-camu	1176 ± 14.8	478 ± 1.2	153 ± 2.6	279 ± 1.5	n.d.
Carnaúba	338 ± 36.4	3549 ± 184	10.7 ± 0.2	15.5 ± 0.4	87.7 ± 2.7
Gurguri	549 ± 22.2	1385 ± 102	35.5 ± 1.6	70.4 ± 7.8	69.7 ± 8.2
Jaboticaba	440 ± 9.9	1472 ± 16.9	37.5 ± 1.4	87.9 ± 1.9	90.7 ± 0.1
Jambolão, java plum	185 ± 3.8	3025 ± 65.4	29.7 ± 0.3	35.5 ± 1.4	67.6 ± 3.1
Juçara, jussara	755 ± 8.3	1711 ± 46	78.3 ± 13.3	84.9 ± 16.1	70.8 ± 7.9
Mangaba	169 ± 21.5	3385 ± 349	14.6 ± 1.8	18.3 ± 1.6	n.d.
Murici, nance	n.d.	n.d.	n.d.	n.d.	n.d.
Murta	610 ± 17.7	936 ± 33.3	49.1 ± 0.2	108 ± 2.3	74 ± 9.2
Puçá-coroa-de-frade	268 ± 4.8	1272 ± 51.4	38.5 ± 1.2	84.9 ± 1.3	77.3 ± 1.4
Puçá-preto	868 ± 51.0	414 ± 14.4	125 ± 9.7	208 ± 3.9	85.9 ± 7.4
Umbu	90.4 ± 2.2	7074 ± 218	6.3 ± 0.2	17.2 ± 0.3	63.4 ± 8.4
Uvaia	127 ± 3.3	3247 ± 392	18 ± 0.8	38.4 ± 4.1	79.8 ± 5.9

[a] Mean value ± standard deviation; $n = 3$; n.d. = not detected.
[b] Concentration of antioxidant required to reduce the original amount of free radicals by 50%.
[c] Oxidation inhibition.

Table 4
Polyphenols and antioxidant capacity in aqueous-organic extracts of 18 non-traditional Brazilian tropical fruits (dry matter).[a]

Fruits	Extractable polyphenols mg GAE/100 g	DPPH· EC_{50} (g/g DPPH·)	$ABTS^{\cdot+}$ μmol Trolox/g	FRAP μmol Fe_2SO_4/g	β-Carotene bleaching % O.I. [b]
Açaí, assai	3268 ± 527	598 ± 164	64.5 ± 19.2	220 ± 32.9	76.1 ± 6
Acerola	10,280 ± 77.7	49.2 ± 2.5	953 ± 34.1	1996 ± 47	n.d.
Bacuri	1365 ± 43.3	6980 ± 854	18.1 ± 3.7	16.1 ± 1.4	74.9 ± 0.7
Cajá, yellow mombim	579 ± 12.9	1064 ± 162	40.7 ± 2.2	97.6 ± 0.6	84.9 ± 3.4
Caju, cashew apple	830 ± 26.5	906 ± 78.2	79.4 ± 15.7	154 ± 7.8	44.6 ± 11.7
Camu-camu	11,615 ± 384	42.6 ± 1.4	1237 ± 33.8	2502 ± 74.5	n.d.
Carnaúba	830 ± 28.3	4877 ± 24.3	16.4 ± 0.2	18.8 ± 0.1	94.2 ± 3.2
Gurguri	1364 ± 24.8	360 ± 32.7	136 ± 20.1	274 ± 15.7	97.5 ± 0.7
Jaboticaba	3584 ± 90.9	138 ± 3.1	317 ± 2.7	635 ± 11.9	90.6 ± 0.6
Jambolão, java plum	1117 ± 67.1	938 ± 46.9	125 ± 10.8	173 ± 10.8	88.4 ± 6.8
Juçara, jussara	5672 ± 55.9	70.1 ± 4.8	606 ± 142	834 ± 142	96.1 ± 2.5
Mangaba	935 ± 37	890 ± 69.1	65.6 ± 7.4	163 ± 11.7	34.7 ± 12.3
Murici, nance	2380 ± 104	238 ± 17.7	412 ± 13	334 ± 3.9	61.5 ± 1.6
Murta	2055 ± 75.7	363 ± 27.4	166 ± 4	299 ± 22.4	92.5 ± 0.6
Puçá-coroa-de-frade	1047 ± 77	316 ± 2	161 ± 3	380 ± 0.4	95.9 ± 1.2
Puçá-preto	2638 ± 48.9	65.6 ± 2.4	346 ± 21.7	909 ± 28.4	99.1 ± 0.5
Umbu	742 ± 19	933 ± 109	77 ± 15.4	143 ± 1.3	79.3 ± 14.6
Uvaia	1930 ± 129	276 ± 22.2	182 ± 14.2	408 ± 34.9	63.7 ± 5.3

[a] Mean value ± standard deviation; $n = 3$; n.d. = not detected.
[b] Oxidation inhibition.

The fresh and dried fruits richest in polyphenols were, respectively, camu-camu (1176 mg GAE/100 g and 11,615 mg GAE/100 g), acerola (1063 mg GAE/100 g and 10,280 mg GAE/100 g) and puçá-preto (868 mg GAE/100 g and 2638 mg GAE/100 g), indicating that these fruits are excellent sources of polyphenols.

The fresh and dried fruits classified as having intermediate polyphenol contents were assai (454 mg GAE/100 g and 3268 mg GAE/100 g) and jaboticaba (440 mg GAE/100 g and 3584 mg GAE/100 g), respectively. Finally, fresh fruits classified as low in polyphenols included umbu, yellow mombin and bacuri; dried fruits included mangaba, carnauba, cashew apple, umbu and yellow mombin.

A recent study yielded similar findings for total phenol contents in fresh acerola from Ceará, Brazil (1056 mg GAE/100 g) (Alves, Brito et al., 2008). In another study, involving 14 myrtaceans (Reynertson, Yang, Jiang, Basile, & Kennelly, 2008), results were also close to our own: 10,100 mg GAE/100 g, 3160 mg GAE/100 g and 995 mg GAE/100 g for dried camu-camu, jaboticaba and java plum, respectively.

3.2.2. Measurement of antioxidant capacity

Antioxidant capacity, as determined by DPPH·, $ABTS^{\cdot+}$, FRAP and β-carotene methods, is shown in Tables 3 and 4.

When testing fresh and dry matter, respectively, by the DPPH· method, the most antioxidant fruits were puçá-preto (EC_{50} = 414 and 65.6 g/g DPPH·), camu-camu (EC_{50} = 478 and 42.6 g/g DPPH·) and acerola (EC_{50} = 670 and 49.2 g/g DPPH·), indicating an association between antioxidant capacity and phenol contents and, in the case of acerola and camu-camu, between antioxidant capacity and vitamin C contents as well (Table 5). In another study of the antioxidant capacity of fresh samples of acerola from Ceará, Brazil, results were slightly higher than our own (839 g/g DPPH·) (Alves, Brito et al., 2008). The banana passion fruit (*Passiflora mollissima*) produced in tropical America also boasts a large antioxidant capacity (407 g fresh fruit/g DPPH·), not unlike the species evaluated in the present study (Vasco et al., 2008).

Organised in decreasing order of antioxidant capacity, measured on fresh matter by the DPPH· method, our fruits ranked as follows: yellow mombin > cashew apple > umbu > assai > carnauba > mangaba > uvaia > java plum > jussara > jaboticaba > gurguri > puçá-coroa-de-frade > murta > acerola > camu-camu > puçá-preto. For dry matter the order observed was: bacuri > carnauba > yellow mombin > java plum > umbu > cashew apple > mangaba > assai > murta > gurguri > puçá-coroa-de-frade > uvaia > nance > jaboticaba > jussara > puçá-preto > acerola > camu-camu.

When evaluated by the ABTS· method, our fruits ranged from 6.3 to 153 μmol trolox/g (fresh matter) and from 16.4 to 1237 μM trolox/g (dry matter). The corresponding figures for the FRAP method were 11.8–279 and 16.1–2502 μmol Fe_2SO_4/g, respectively.

When organised in order of increasing antioxidant capacity, measured by the ABTS· method, the ranking was: umbu < yellow mombin < carnauba < cashew apple < mangaba < assaí < uvaia < java plum < gurguri < jaboticaba < puçá-coroa-de-frade < murta < jussara < acerola < puçá-preto < camu-camu for fresh matter. For dry matter, it was: carnauba < bacuri < yellow mombin <

Table 5
Pearson's correlation coefficients (r) between bioactive compounds and antioxidant capacity (fresh matter) of 18 non-traditional Brazilian tropical fruits.

R	Vitamin C	Anthocyanins	Flavonoids	Carotenoids	Chlorophyll	Polyphenols	DPPH·	$ABTS^{\cdot+}$	FRAP
Anthocyanins	−0.00								
Flavonoids	−0.10	0.67**							
Carotenoids	−0.23	−0.06	−0.14						
Chlorophyll	0.17	0.57	0.55	0.93*					
Polyphenols	0.70**	0.32	0.20	0.25	0.66				
DPPH	−0.38	−0.21	−0.26	−0.32	0.12	−0.72**			
ABTS	0.70**	0.13	0.03	0.14	0.36	0.92**	−0.68**		
FRAP	0.70**	0.04	0.01	0.14	0.15	0.89**	−0.69**	0.97**	
β-Carotene	−0.45	−0.10	0.20	0.07	−0.60	−0.16	−0.13	−0.11	−0.12

* Significant at $p < 0.01$.
** Significant at $p < 0.05$.

assai < mangaba < umbu < cashew apple < java plum < gurguri < puçá-coroa-de-frade < murta < uvaia < jaboticaba < puçá-preto < nance < juçara < acerola < camu-camu.

The corresponding sequence based on tests with the FRAP method was: yellow mombin < carnauba < umbu < mangaba < cashew apple < assai < java plum < uvaia < gurguri < puçá-coroa-de-frade < jussara < jaboticaba < murta < acerola < puçá-preto < camu-camu for fresh matter, and bacuri < carnauba < yellow mombin < umbu < cashew apple < mangaba < java plum < assai < gurguri < murta < nance < puçá-coroa-de-frade < uvaia < jaboticaba < jussara < puçá-preto < acerola < camu-camu for dry matter.

It may be concluded from the ABTS$^{\cdot+}$, DPPH$^{\cdot}$ and FRAP assays that acerola, camu-camu and puçá-preto have high levels of antioxidants. Fruits in general display great variations in antioxidant capacity when evaluated as fresh samples by the ABTS$^{\cdot}$ method. However, compared to findings published elsewhere (Vasco et al., 2008), the non-traditional species included in our study ranked relatively high.

The β-carotene bleaching method is widely used in laboratories around the world. Since no high temperatures are required, the antioxidant capacity of thermo-sensitive vegetable extracts may be determined and qualitatively evaluated. In the present study, antioxidant capacity was determined from the ability of samples to inhibit β-carotene bleaching caused by free radicals generated during linoleic acid peroxidation. Antioxidant capacity was classified as high (>70%), intermediate (40–70%) or low (<40%) levels of oxidation inhibition (O.I.) (Hassimotto, Genovese, & Lajolo, 2005). Fresh samples of yellow mombin, carnauba, gurguri, jaboticaba, jussara, murta, puçá-coroa-de-frade, puçá-preto and uvaia displayed high levels of O.I. The intermediate group included java plum and umbu. Only two fruits (assai and cashew apple) displayed less than 40% O.I.

In the evaluation of lyophilised samples, 12 fruits showed high levels of O.I., while only three (cashew apple, nance and uvaia) presented intermediate levels. No fruits scored below 40% O.I. Acerola and camu-camu could not be tested by this method at any concentration, possibly because the high content of vitamin C in these fruits interfered in the system as a pro-oxidant factor. A similar study (Alves, Brito et al., 2008), using the β-carotene bleaching method, likewise did not detect antioxidant capacity in acerola.

Vitamin C is the most abundant hydrosoluble antioxidant in plants. However, ascorbic acid displayed pro-oxidant activity in the β-carotene system and so may have influenced our findings for acerola. Pro-oxidant activity has previously been reported for ascorbic acid when using the β-carotene bleaching method or the liposome method (Hassimotto et al., 2005). The pro-oxidant behaviour of ascorbic acid has been described elsewhere (Kalt, Forney, Martin, & Prior, 1999) and appears to be due to the formation of ascorbyl radicals during oxidation.

3.3. Correlation between study variables

Correlation coefficients for vitamin C, total anthocyanins, yellow flavonoids, total carotenoids, chlorophyll and extractable polyphenols, measured by the ABTS$^{\cdot}$ assay, DPPH$^{\cdot}$ assay, FRAP assay and β-carotene bleaching method are shown in Table 5.

A positive and significant correlation was found in this study between vitamin C-extractable polyphenols ($r = 0.70$), ABTS$^{\cdot}$ ($r = 0.70$) and FRAP ($r = 0.70$). Polyphenols and DPPH$^{\cdot}$ were negatively and significantly correlated ($r = -0.72$; $p < 0.05$); this is due to the fact that the DPPH$^{\cdot}$ method yields inversely proportional results. There was also a positive and significant correlation of polyphenols ($p < 0.05$) and ABTS$^{\cdot}$ ($r = 0.92$) and FRAP ($r = 0.89$) methods.

No correlation was observed between the β-carotene bleaching and any of the study variables. In another study (Hassimotto et al., 2005), the correlation between antioxidant capacity and vitamin C could not be established with the β-carotene method and liposome method because vitamin C was a pro-oxidant in both systems. Other authors (Kalt et al., 1999) reported a negative influence of vitamin C, showing ascorbate content and antioxidant capacity to be negatively correlated ($r = -0.80$) for strawberries, raspberries and high- and low-bush blueberries.

In conclusion, it is not always a simple task to choose the most appropriate method to determine antioxidant capacity. The FRAP and ABTS$^{\cdot+}$ methods are generally indicated for hydrophilic compounds, while the β-carotene bleaching method is suitable for lipophilic compounds. The DPPH$^{\cdot}$ method may be employed routinely with aqueous-organic extracts containing hydrophilic and lipophilic compounds. Regarding the considerable amounts of vitamin C (camu-camu and acerola), anthocyanins (Myrtaceaes – murta, java plum, jaboticaba and camu-camu), carotenoids (Melastomaceae – gurguri, puçá-preto e puçá-coroa-de-frade) and phenolic compounds, our results indicate promising perspectives for the exploitation of non-traditional tropical fruit species with considerable levels of nutrients and antioxidant capacity. The considerable amounts of anthocyanins for several of the non-traditional fruits included in this study are likely to draw attention to these species as potential commodities. Although evaluation methods and results reported have not yet been sufficiently standardised, making comparisons difficult, the data add valuable information to current knowledge on the nutritional properties of tropical fruits, such as the considerable antioxidant capacity found for acerola and camu-camu (ABTS$^{\cdot+}$, DPPH$^{\cdot}$ and FRAP) and for puçá-preto (all methods).

Acknowledgements

The authors would like to thank CAPES, CNPq, EMBRAPA, UFERSA and the European Union (INCO-CT-2005-015279) for financial support.

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