Ciência Rural, Santa Maria, v.53:7, e20210900, 2023
http://doi.org/10.1590/0103-8478cr20210900
ISSNe 1678-4596
FOOD TECHNOLOGY

Phenolic composition of jussara (*Euterpe edulis* Martius) from Minas Gerais and Espírito Santo, Brazil, determined by high performance liquid chromatography coupled to diode array detection and electrospray ionization tandem mass spectrometry (HPLC–DAD-ESI-MS)

Carolina Tatagiba da Rocha Dadalto[1] Isidro Hermosín-Gutiérrez[2] José Pérez-Navarro[2] Rosa Maria Ojeda-Amador[2] Sergio Gómez-Alonso[2] Paulo César Stringheta[1] Daniele de Almeida Paula[3*] Marcos Roberto Moacir Ribeiro Pinto[4] Afonso Mota Ramos[1]

[1]Departamento de Ciência e Tecnologia de Alimentos, Universidade Federal de Viçosa (UFV), Viçosa, MG, Brasil.
[2]Universidad de Castilla-La Mancha, Ciudad Real, Castilla-La Mancha, ES.
[3]Instituto Federal de Educação, Ciência e Tecnologia do Paraná (IFPR), 85935-000, Assis Chateaubriand, PR, Brasil. E-mail: daniele.paula@ifpr.edu.br.
[*]Corresponding author.
[4]Instituto Federal de Educação, Ciência e Tecnologia do Espírito Santo (IFES), Venda Nova do Imigrante, ES, Brasil.

ABSTRACT: This research assessed the phenolic composition of Jussara pulp from the Brazilian states of Minas Gerais (MG) and Espírito Santo (ES) using HPLC-DAD-MS/MS. Seventeen anthocyanins were detected in fruits, derived from cyanidin, pelargonidin and peonidin. Among the non-anthocyanic phenolic compounds, flavonols (kaempferol, quercetin and isorhamnetin derivatives), flavan-3-ols (catechin, epicatechin, B-type procyanidins and unknown dimers) and resveratrol in its glycosylated form have been identified. Catechin (32.41-60.56 mg $100g^{-1}$) and epicatechin (18.86-40.92 mg $100g^{-1}$) were the main flavan-3-ols present in the fruits. The samples showed small concentrations of resveratrol glycosides (0.02-0.91 mg $100g^{-1}$). The analytical methodology used (HPLC-DAD-ESI-MS/MS) permitted the identification of newly reported compounds in this fruit.
Key words: anthocyanins, flavonols, hydroxycinnamic acid derivatives, flavan-3-ols, jussara fruit, HPLC-DAD-ESI-MS/MS.

Composição fenólica de Jussara (*Euterpe edulis* Martius) de Minas Gerais e Espírito Santo, Brasil, determinada por cromatografia líquida de alta eficiência acoplada à detecção de arranjo de diodos e espectrometria de massa contendo sistema de ionização por eletrospray (HPLC-DAD-ESI MS/MS)

RESUMO: O objetivo desta pesquisa foi avaliar a composição fenólica da polpa de Jussara dos Estados brasileiros de Minas Gerais (MG) e Espírito Santo (ES) usando HPLC-DAD-MS / MS. Dezessete antocianinas foram detectadas, dentre elas, derivadas de cianidina, pelargonidina e peonidina. Dentre os compostos fenólicos não antocianicos, foram identificados flavonóis (derivados de caempferol, quercetina e isorhamnetina), flavan-3-ols (catequina, epicatequina, procianidinas do tipo B e dímeros desconhecidos) e resveratrol em sua forma glicosilada. Catequina (32,41-60,56 mg $100g^{-1}$) e epicatequina (18,86-40,92 mg $100g^{-1}$) foram os principais flavan-3-óis presentes nas frutas. As amostras apresentaram pequenas concentrações de glicosídeos resveratrol (0,02-0,91 mg $100g^{-1}$). A metodologia analítica utilizada (HPLC-DAD-ESI-MS / MS) permitiu a identificação de novos compostos relatados presentes na composição da polpa de Jussara.
Palavras-chave: antocianinas, flavonóis, derivados do ácido hidroxicinâmico, flavan-3-ols, fruta jussara, HPLC-DAD-ESI-MS/MS.

INTRODUCTION

Jussara is the name of the fruit of *Euterpe edulis* Martius, a native palm tree of the Brazilian Atlantic Forest (CARVALHO et al., 2019; BICUDO et al., 2014). The fruit is round and has a dark purple pulp covering a hard seed (SANTANA et al., 2016). After being harvested, the fruit should be macerated with water resulting in a consistent pulp. The consumption of fresh pulp and its use as an ingredient have been encouraged in recent years due to its high nutritional value (GARCIA, et al., 2019; SCHULZ et al., 2015).

This fruit is rich in nutrients such as proteins, carbohydrates, fiber, sugars, and lipids with high proportions of polyunsaturated fatty acids, vitamin C, carotenoids, and minerals (DE SOUZA et al., 2014; PUPIN et al., 2018; BERNARDES et.

Received 12.22.21 Approved 08.03.22 Returned by the author 10.20.22
CR-2021-0900.R1
Editors: Leandro Souza da Silva Juliane Welke

al., 2019). Nevertheless, the interest in Jussara is especially due to its antioxidant compounds, such as phenolic compounds (SEERAM, 2008). The main phenolic compounds of Jussara pulp are anthocyanins. Among them, cyanidin-3-glucoside and cyanidin-3-rutinoside are present at higher concentrations, being identified for the first time in this fruit (VIEIRA et al., 2018; HARBORNE et al., 1994). Among the non-anthocyanin phenolic compounds of Euterpe edulis Martius fruit, phenolic acids, flavonols, flavones and flavan-3-ols have been detected (SILVA et al., 2021; INADA et al., 2015; SCHULZ et al., 2015, 2017). However, flavonols, flavones and flavan-3-ols, specifically, have not been studied in detail up to the present time. Thus, these papers only indicated the group to which the compound belongs without giving information on the structure of the molecules and their substituent.

Most of the published papers evaluated Jussara composition by high-performance liquid chromatography coupled with a diode array detector (HPLC-DAD). This technique compares the UV-Vis spectra and retention times of the compounds present in the extract with those of standards (SEERAM et al., 2006). However, different secondary plant metabolites may have similar UV-Vis spectra or chromatographic retention times, and it is impossible to distinguish them by using only a DAD detector (REBELLO et al., 2013). The mass spectrometry (MS) provides the fragmentation pattern and *m/z* ratio of the compounds, making it possible to define the structure of the molecule by comparing it with data previously depicted in the literature (ARDREY, 2003; LOPES-DA-SILVA et al., 2002).

The main phenolic compounds of Jussara pulp are anthocyanins. Among them, cyanidin-3-glucoside and cyanidin-3-rutinoside are present at higher concentrations, being identified for the first time in this fruit by (HARBORNE et al., 1994). Among the non-anthocyanin phenolic compounds of *Euterpe edulis* fruit, phenolic acids, flavonols, flavones, and flavan-3-ols have been detected (INADA et al., 2015; SCHULZ et al., 2015, 2017). However, flavonols, flavones, and flavan-3-ols, specifically, have not been studied in detail up to the present time. Other than that, the biosynthesis of phenolic compounds via the route of phenylpropanoid can be affected by genetic and agronomic factors, degree of maturation, environmental conditions, and interaction between these factors (TIWARI & CUMMINS, 2013).

Hence, this current study identified and quantified phenolic compounds (anthocyanins, flavonols, flavan-3-ols, stilbenes, and hydrolyzable tannins) of Jussara fruit pulp cultivated in seven regions of Minas Gerais and Espírito Santo, Brazil, using HPLC-DAD -ESI-MS/MS to get specific knowledge of its phenolic composition.

MATERIALS AND METHODS

Collecting and processing of fruits

Jussara fruits were collected in seven regions during the 2015 harvest (Table 1). The plants were spontaneously occurring. Samples from each region were processed independently and were collected 3 lots of approximately 5 kg. The selected fruits (~2.3 kg) were washed and sanitized with chlorinated water (200 mg/L) for 10 min and rinsed with chlorinated water (20 mg/L) for 5 min. Then, the fruits were immersed in water at 40 °C for 15 min. The pulp was extracted with the addition of water (0.6 L/kg of fruit) (BICUDO et al., 2014) and was lyophilized and used to prepare the extracts.

Table 1 - Geographic data of the regions where the jussara samples were collected.

Região	UF	Month of harvest	Latitude*	Longitude*	Altitude*
Rio Novo do Sul	ES	May	S20º48`34.529”	W40º56`05.826”	452
Viçosa	MG	June	S20º47`28.082”	W42º52`45.120”	685
Rio Pomba	MG	June	S21º9`6.062”	W43º8`19.397”	839
Canaã	MG	July	S20º41`16.881”	W42º36`15.417”	759
Araponga	MG	July	S20º41`06.609”	W42º32`12.001”	959
Rosário de Limeira	MG	September	S20º53`58.275”	W42º32`41.884”	1092
Vargem Alta	ES	November	S20°31'47.900"	W40°58'33.500"	1058

*Data obtained by GPS. Altitude (m). UF: Federative Unit. MG: Minas Gerais. ES: Espírito Santo.

Phenolic extract preparation

The extraction procedure was adapted by REBELLO et al. (2013). Approximately 0.1 g of lyophilized pulp was extracted with 10 mL of a methanol/water/formic acid (50:48.5:1.5) mixture using a Q.Sonic ultrasonic nozzle at 4 °C for 3 min (every 30 s, a pause of 5 s). The extract was centrifuged (4200 *g* for 10 min; Digicel, Spain), and the supernatant was filtered. The pellet was further extracted with 10 mL of methanol/water/formic acid, centrifuged, and filtered. The two filtered extracts were combined and mixed with hexane (10 mL) to remove the lipids. The hexane was separated from the methanolic extract by decantation. The methanolic extract was dried in a rotary evaporator (35 °C) and its volume was made up to 25 mL, in a volumetric flask, with Milli-Q water.

Determination of anthocyanins by HPLC-DAD-ESI-MS-MS

The methodology was adapted by REBELLO et al. (2013). For the analysis of anthocyanins, the fresh extract (section 2.2) was filtered with polyester membranes (0.20 µm; Chromafil PET 20/25), and 10 µL was injected into an Ascentis Express C18 column (2.1 × 150 mm, 2.7 µm particle; Sigma-Aldrich). The flow rate was 0.22 mL/min and the solvents were A (water/formic acid/acetonitrile, 88.5:8.5:3, v/v/v) and B (water/formic acid/acetonitrile, 41.5:8.5:50, v/v/v). The solvents were eluted using the following gradients: 0 min, 98% A and 2% B; 15 min, 85% A and 15% B; 20 min, 70% A and 30% B; 25 min, 100% B; 29 min, 100% B; 35 min, 98% A and 2% B. The ion trap detector was configured in positive ionization mode. The mass and UV-VIS spectra obtained by the injection were compared to the mass and UV spectra with data from standards and literature. For quantification, the peak area of the chromatograms at 520 nm was used. For compounds with similar retention time were utilized ion the chromatograms of the m/z for each compound to estimate their percent area. The calculated factor was applied to the total peak area. Results were expressed as a molar percentage of each compound and the total content as cyanidin-3- rhamnosyl-glucoside equivalents.

Determination of flavonols by HPLC-DAD-ESI-MS-MS

The non anthocyanin phenolic compounds were isolated from fresh extract using SPE cartridges (40 µm, 500 mg, 6 mL; Agilent, Bond Elut Plexi PCX). Firstly, the cartridges were conditioned with 5 mL of methanol and 5 mL of water. After that, 10 mL of Jussara extract, diluted with 10 mL of 0.1 N HCl, was passed through the SPE cartridges. One cartridge was composed of cationic PCX and the other of C18. The cartridges were washed with 5 mL of 0.1 N HCl and 5 mL of water. Then, the purified fraction (without anthocyanin) was recovered with 2 × 3 mL ethanol. This fraction was concentrated until completely dry in a rotary evaporator at 35 °C, re-dissolved with 1 mL of 20% methanol solution, and 20 µL was injected into the HPLC equipment (REBELLO et al., 2013).

The column used was a ZORBAX Eclipse XDB-C18 (2.1 × 150 mm, 3.5 µm). The flow rate was 0.19 mL/min. The solvents A (water/formic acid/acetonitrile, 88.5:8.5:3, v/v/v), B (water/formic acid/acetonitrile, 41.5:8.5:50, v/v/v) and C (water/formic acid/methanol, 1.5:8.5:90, v/v/v) were eluted in the following gradients: 0 min, 96% A and 4% B; 10 min, 96% A and 4% B; 60 min, 70% A, 15% B and 15% C; 61 min, 50% A, 25 % B and 25% C; 65 min, 50% B and 50% C; 68 min, 50% B and 50% C; 74 min, 96% A and 4% B. For identification, the ion-trap ESI-MS/MS detector was used in positive ion mode for flavonols, using standard solutions of quercetin 3-galactoside for setting the optimal ionization and fragmentation conditions (CASTILLO-MUÑOZ et al., 2009; REBELLO et al., 2013). For the quantification of flavonols, DAD chromatograms at 360 nm were extracted and standard curves of kaempferol-3-glucoside (0,0053x + 0,0006; 0 – 100 mg/L, R2 0,9988) , kaempferol-3-rutinoside (0,0070x + 0,0005); 0 – 100 mg/L, R2 0,9999) , quercetin-3-glucoside (y = 0,0053x + 0,0009; 0 – 100 mg/L, R2 0,9978), quercetin-3-rutinoside (0,0088x + 0,0007; 0 – 100 mg/L, R2 0,9988) and isorhamnetin 3-glucoside (0,0052x + 0,0008; 0 – 100 mg/L, R2 0,9999) were used. Results were expressed as molar percentage of each compound and the total concentration as kaempferol-3-glucoside.

Determination of flavan-3-ols and stilbenes by HPLC-DAD-ESI-MS/MS

SPE C18 cartridges (Waters Sep-Pak C18 cartridges) were conditioned with 10 mL of ethyl acetate, 10 mL of methanol, and 10 mL of Milli-Q water. Then, 10 ml of the fresh extract was passed through the cartridge. The cartridges were then washed with 3 × 5 mL of methanol, 5 mL of ethyl acetate, fully dried by passing air, and the eluate dried in a rotary evaporator at 35 °C. The dried residue was re-dissolved in 2 mL of methanol (REBELLO et al., 2013).

The chromatographic conditions were set according to Rebello et al. (2013) and are described below. The analysis was performed using

an Agilent 1200 series system equipped with a DAD (Agilent, Germany), coupled to an AB Sciex 3200 Q TRAP (Applied Biosystems) ESI-MS/MS system. The column used was a reversed-phase Agilent Eclipse XDB-C18 (2.1 × 150 mm; 3.5 µm particle; Agilent, Germany), thermostatted at 16 °C. The flow rate was 0.1 mL/min, and the injection volume was 10 µL. The solvents A (water/methanol/formic acid, 97:2:1, v/v/v) and B (methanol) were eluted in the following gradients: 0 min, 5% B; 2 min, 5% B; 25 min, 30% B; 40 min, 55% B; 50 min, 65% B; 55 min, 95% B; 65 min, 95% B; 70 min, 5% B; 80 min, 5% B. The following standards were used: flavan-3-ol monomers and dimers of (+)-catechin, (−)-epicatechin, (−)-gallocatechin, (−)-epigallocatechin, (−)-epicatechin-3-gallate and procyanidins (B1, B2, and B4); and resveratrol *trans*/*cis* isomers and their corresponding 3-glucosides. Results were expressed as the molar percentage of each compound. The total amount of flavan-3-ols was expressed as (+)-catechin equivalents and stilbenes were quantified as resveratrol glycoside.

Determination of hydrolyzable tannins by HPLC-DAD-ESI-MS/MS

The level of gallotannins and ellagitannins was determined following modification of the method of PENG et al. (1991). The anthocyanin-free extract (the same extract used for flavonol determination; 300 µL) was mixed with methanol (2100 µL) and HCl at 37% (600 µL) in a sealed test tube. The mixture was heated in boiling water for a total of 2 h for hydrolysis of the sample. Finally, the tube was cooled in cold water, and 3 mL of Milli-Q water was added to the sample tube. Polyester membrane (0.20 µm; PET Chromafil 20/25, Germany) was used to filter the homogenized mixture before its injection into the HPLC system.

The hydrolyzed sample (50 µL) was injected into a ZORBAX Eclipse XDB-C18 column (2.1 × 150 mm, 3.5 µm). The flow rate was 0.1 mL/min. The mobile phases were: A (97% water/2% methanol/1% formic acid) and B (100% methanol). The gradient of solvent B used was: 0 min, 5%; 2 min, 5%; 25 min, 30%; 40 min, 55%; 50 min, 65%; 55 min, 95%; 70 min, 5%; 80 min, 5%. Identification was based on spectroscopic data (UV-Vis and MS/MS) obtained from authentic standards (gallic acid and ellagic acid).

Statistical analysis

The statistical analysis was performed using Minitab 17 (Demo version). An analysis of variance (one-way ANOVA) was conducted to identify significant differences at the 5% level. Differences between means were compared by the Tukey HSD test.

RESULTS AND DISCUSSION

Antho*cyanins from Jussara pulp*

Figure 1 shows the HPLC-DAD anthocyanin chromatographic profile (detection at 520 nm). Mono-substituted anthocyanins in the 3-position of the oxygenated heterocyclic (ring C) correspond to 14 of the compounds found, being predominant in the Jussara pulp. Besides that, one 3,5-disubstituted anthocyanin (peak 1) and two compounds that could not be identified with current data (peaks 13 and 14) were detected. Table 2 shows the retention time and mass spectra (MS and MS/MS) of the detected compounds and their tentative assignment.

Qualitative analysis of the anthocyanins, including those co-eluting under the chromatographic conditions used, was possible by comparing data from MS and MS/MS spectra with those obtained from standards or previously reported. Cyanidin (cy), pelargonidin (pg), and peonidin (pn) structures were detected by MS/MS spectra (*m/z* 287, 271, and 301, respectively) (BRITO et al., 2007; REBELLO et al., 2013).

For cyanidin, the 3-glucoside (3-glc), 3-galactoside (3-gal), 3-sophoroside (3-soph), 3-rhaminosylglucoside (3-rhmglc), 3-pentosylgalactoside (3-pentgal), 3-rhamnosylgalactoside (3-rhmgal), 3-pentosylglucoside (3-pentglc), 3-*cis*-rhamnoside (3-*cis*-rhm), 3-pentidoside (3-pent), 3-*trans*-rhamnoside (3-*trans*-rhm) and 3,5-diglucoside (3,5-diglc) were found. The 3-glc and 3-rhmglc were also detected for pelargonidin (pg) and peonidin (pn), minor aglycones in Jussara fruit.

Differentiation between cy-3-gal (peak 2) and cy-3-glc (peak 4), which have almost identical MS/MS spectra, was performed through the injection of standard compounds of both compounds and checking their retention times. It was verified that cy-3-gal eluted before cy-3-glc. Based on this, it was determined that galactosylated derivatives elute before glucosylated ones, so 3-pentgal and 3-rhmgal were differentiated from 3-pentglc and 3-rhmglc, respectively.

The MS/MS spectra for compounds A and C showed neutral losses of pentose (132 Da) together with additional galactosyl and glucosyl (162 Da) residues, respectively (Truchado et al., 2009). Compounds 5, 7, and 11 had neutral losses of residues of rhamnose (146 Da) and glucose (162 Da),

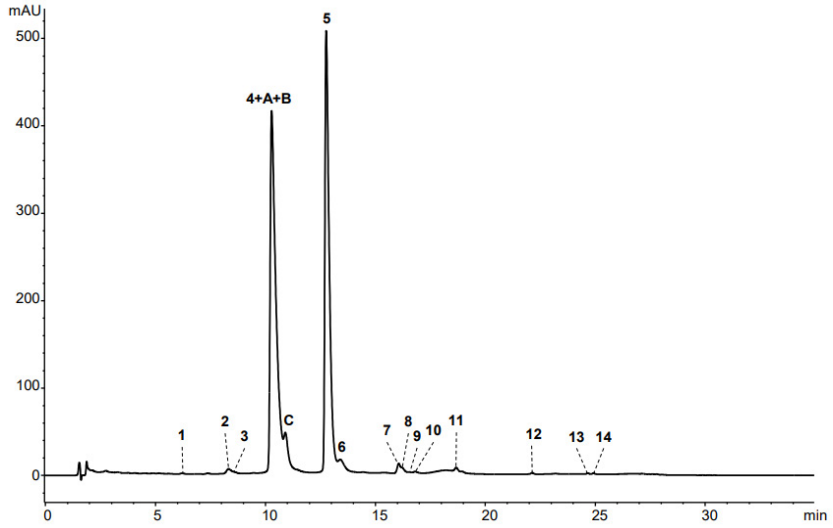

Figure 1 - HPLC-DAD chromatogram (detection at 520 nm) for the anthocyanins from Jussara fruit pulp (sample from Araponga, MG, Brazil). Peaks numbering indicated in the table 2.

suggesting the presence of the 3-rhmglc of cyanidin, pelargonidin, and peonidin, respectively (RUIZ et al., 2013). The retention time of suspected cy-3-rhmglc (peak 5) matched that of a commercial standard.

Peak B had the same fragmentation pattern as cy-3-rhmglc, both exhibiting the molecular ion at m/z 595 and a neutral loss of rhamnose (146 Da) followed by a second neutral loss of 162 Da, but it eluted before cy-3-rhmglc and thus was identified as cy-3-rhmgal.

Peaks 8 and 12 had molecular ions at m/z 433, and MS/MS fragmentation with a lone signal at m/z 287; based on this, they were identified as two isomers of cy-3-rhm (SILVA et al.,2014), maybe positional isomers. The MS and MS/MS data suggest that compound 10 is cy-3-pent, since the molecular ion and products of fragmentation had m/z signals at 419 and 287, respectively, being compatible with those found in the literature (DALL`ASTA et al., 2012). Cy-3,5-diglc (peak 1) and cy-3-soph (peak 3) showed mass spectra (MS and MS/MS) and retention times matching those of commercial standards.

Compounds 1, 4, 5, 6, 7, 8, 9, and 11 had been already reported in previous research dealing with Jussara pulp from São Paulo and Paraná (BICUDO et al., 2014; SILVA et al., 2014). However, as far as we know, compounds 2, 3, A, B, C, 10, 12, 13, and 14 have not yet been reported to occur in Jussara pulp. In this research, spectroscopic evidence was obtained for these compounds in Jussara from Minas Gerais and Espírito Santo.The anthocyanin profiles of the samples were very similar, but there was a concentration difference of each compound according to the culture region. According to table 2 the fruits of Canaã contain a higher concentration of total anthocyanins (737.00 mg/ 100 grams of fruit) than those of the other regions.

The cyanidines group was the most abundant in all samples analyzed, independent of the region. Except for the Araponga region, cy-3-rhmglc was the most dominant anthocyanin in the fruits of each region, ranging from 38.09% in samples from Araponga to 59.17% in those of Canaã. The cy-3-glc was predominant in Jussara from Araponga and was the second most concentrated anthocyanin in Jussara samples of the other regions, ranging from 34.57% (Canaã) to 52.43% (Araponga).

Fruits from Araponga have a higher proportion of total pelargonidin derivatives (total pg) than those of Rio Novo do Sul, Viçosa, Canaã and Vargem Alta. The pg-3-glc content is higher in samples from Araponga (2.44% total anthocyan content) than in the other regions. Rio Pomba presented a higher proportion of pg-3-rhmglc (1.02%) than Canaã (0.23%), Vargem Alta (0.41%), Rosário de Limeira (0.46%) and Rio Novo do Sul (0.45%).

The total content of peonidin derivatives (total pn) did not significantly vary between regions.

Table 2 - Retention time, mass spectra and molar percentage of anthocyanins identified in the Jussara fruit pulp.

						% Molar						
eak[1]	Assignment*	λ_{max} (nm)	t_r(min)	M]$^+$(*m/z*)	S/MS (*m/z*)	ARA	CAN	RL	RN	RP	VA	VIC
	cy-3,5-diglc	94, 508	.21	11	49, 287	0.09±0.01	nq	nq	nq	nq	nq	nq
	cy-3-gal	79, 515	.32	49	87	0.57±0.08^{a}	nq	nq	0.19±0.06^{b}	nq	nq	nq
	cy-3-soph	279, 515	.50	11	87	0.17±0.01^{a}	0.05± 0.00^{b}	0.12±0.04^{a}	0.13±0.01^{a}	nq	0.12±0.02^{a}	nq
	cy-3-glc	80, 514	0.31	49	87	52.43±0.56^{a}	34.57±2.75^{c}	42.93±6.54^{bc}	41.68±2.16^{bc}	43.81±1.23^{ab}	37.77±1.16^{bc}	39.44±4.32^{bc}
	cy-3-pentgal	80, 514	0.60	81	49, 287	0.45±0.05^{b}	0.58±0.09^{ab}	0.76± 0.11^{a}	0.55±0.02^{ab}	0.66±0.21^{ab}	0.54±0.07^{ab}	0.47±0.07^{ab}
	cy-3-rhmgal	80, 514	1.00	95	49, 287	1.01±0.10^{a}	0.91±0.04^{ab}	1.02±0.11^{a}	0.95±0.24^{a}	0.57±0.15^{b}	1.01±0.10^{a}	0.85±0.07^{ab}
	cy-3-pentglc	80, 514	1.10	81	49, 287	2.92±0.05^{bc}	3.45±0.23^{ab}	4.14±0.76^{a}	2.89±0.46^{bc}	2.14± 0.32^{c}	2.48± 0.13^{bc}	2.39±0.35^{bc}
	cy-3-rhmglc	81, 516	2.75	95	49/287	38.09±0.29^{c}	59.17±3.12^{a}	48.34±7.29^{b}	51.14±2.48^{ab}	51.11±1.11^{ab}	56.72±1.07^{ab}	54.47±4.54^{ab}
	pg-3-glc	81, 516	3.39	33	71	2.44±0.17^{a}	0.26±0.19^{b}	1.14±0.98^{b}	0.66±0.34^{b}	0.69±0.09^{b}	0.39± 0.07^{b}	0.66±0.25^{b}
	pg-3-rhmglc		6.06	79	33, 271	0.65±0.08^{ab}	0.23±0.12^{b}	0.46± 0.43^{b}	0.45±0.06^{b}	1.02±0.15^{a}	0.41±0.03^{b}	0.54±0.17^{ab}
	cy-3-rhm		6.16	33	87	0.47±0.06^{abc}	0.19±0.03^{d}	0.47±0.10^{bc}	0.52±0.12^{ab}	nq	0.28±0.08^{cd}	0.68±0.02^{a}
	pn-3-glc		6.50	63	01	0.12±0.00^{ab}	0.08±0.01^{b}	0.13±0.03^{ab}	0.15±0.04^{a}	nq	nq	nq
0	cy-3-pent		6.81	19	87	0.17±0.02^{a}	0.10±0.01^{b}	0.17±0.02^{a}	0.14±0.03^{ab}	nq	nq	nq
1	pn-3-rhmglc	72, 517	8.67	09	463, 301	0.22±0.02^{b}	0.29±0.06^{ab}	0.20±0.06^{b}	0.34±0.07^{ab}	nq	0.28±0.02^{ab}	0.50±0.20^{a}
2	cy-3-rhm-iso	80, 519	2.13	33	87	0.10±0.02^{ab}	0.05±0.01^{b}	0.12±0.04^{ab}	0.13±0.03^{a}	nq	nq	nq
13	385-glc	87, 527	4.66	47	85	0.05±0.00^{a}	0.02±0.00^{b}	nq	0.03±0.01^{ab}	nq	nq	nq
4	385-rhmglc		4.92	93	47, 385	0.04±0.00^{a}	0.04±0.01^{a}	nq	0.05±0.02^{a}	nq	nq	nq
	Total cy					96.48±0.21^{b}	99.07±0.31^{a}	98.08±1.46^{ab}	98.32±0.27^{ab}	98.29±0.08^{ab}	98.92±0.08^{a}	97.85±0.92^{ab}
	Total pg					3.09±0.20^{a}	0.50±0.30^{b}	1.60±1.40^{ab}	1.10±0.40^{b}	1.71±0.08^{ab}	0.80±0.09^{b}	1.20±0.42^{b}
	Total pn					0.34±0.01^{a}	0.37± 0.07^{a}	0.33±0.08^{a}	0.49±0.10^{a}	nq	0.28±0.02^{a}	0.50±0.20^{a}
	Total[2]					96.37±11.00^{b}	737.00±341.00^{a}	98.10±30.90^{b}	318.60±120.50^{b}	11.97±6.08^{b}	96.36±2.91^{b}	61.30±22.70^{b}

*cy, cyanidin; pg, pelargonidin; pn, peonidin; diglc, diglucoside; gal, galactoside; soph, sophoroside; glc, glucoside; pent, pentoside; rhm, rhamnoside; ARA, Araponga; CAN, Canaã; RL, Rosário de Limeira; RN, Rio Novo do Sul; RP, Rio Pomba; VA, Vargem Alta; VIC, Viçosa; Underlined *m/z* value in the MS/MS spectra indicated the most intense signal. [1]Peak numbers used in Figure 1. [2]expressed in mg of cy 3-rhmglc/100 g of fruit. [3]NQ, non quantified.. Different superscript letters between regions denote significant differences.

In the fruits of all regions except Araponga, the total peonidin derivatives are lower than the total pelargonidin derivatives. This suggested that Araponga cultivation conditions may stimulate B-ring methylation. The proportion of pn-3-rhmglc is higher than that of pn-3-glc in fruits of all regions except Rio Pomba, which presents non-quantifiable levels of both compounds.

Flavonols from Jussara pulp

Twelve compounds derived from kaempferol (K), quercetin (Q) and isorhamnetin (I) were identified by MS/MS spectra (m/z values 303, 287 and 317, respectively). The retention time and mass spectra data (MS and MS/MS) are summarized in table 3. Figure 2 shows the HPLC-DAD chromatogram, obtained at 360 nm, of the flavonol profile of one of the purified Jussara extracts.

In the case of kaempferol derivatives, the 3-galactoside (3-gal), 3-glucoside (3-glc), 3-(6”-malonyl)-glucoside, 3-(2”-malonyl)-glucoside and 3-(6”-rhamnosyl)-glucoside (3-rhmglc) were detected. Among the derivatives of quercetin, the 3-gal, 3-glc, 3-rhmglc and 3-(6”-malonyl)-glucoside were found. Three isorhamnetin derivatives were observed: 3-gal, 3-glc and the 3-rhmglc. The identity of the flavonols was tentatively confirmed by comparison of their MS and MS/MS spectrometric data with those reported in the literature or obtained from commercial standards when available.

The presence of K-3-gal (peak 3), K-3-glc (peak 5), and K-3-rhmglc (peak 7) were confirmed by matching their retention times and MS/MS and UV spectra with those obtained from standards analyzed in the same conditions. Besides both compounds K-3-gal and K-3-glc having or presenting the same mass spectrum with a molecular ion at *m/z* 449 and neutral loss of 162 Da, they can be identified since their retention times are different, according to reported data (CASTILLO-MUÑOZ et al., 2009). K-3-rhmglc has a molecular ion with an m/z signal at 595 and fragmentation products with m/z signals at 449 and 287, the latter being the most intense signal. Peaks 9 and 11 present a molecular ion of m/z 535 with a loss of 248 Da, suggesting they correspond to the malonyl-glucoside of kaempferol (CARAZZONE et

Table 3 - Retention time, mass spectra and molar percentage of flavonols identified in the Jussara fruit pulp.

	% Molar[4]											
Peak[1]	Assignment*	λ_{max} (nm)	R_t (min)	$[M+H]^+$ (*m/z*)	MS/MS (*m/z*)	ARA	CAN	RL	RN	RP	VA	VIC
1	q-3-gal	255, 288 sh, 322 sh, 341	23.2	465	303	2.79±0.82^{ab}	1.34±0.52^{bc}	3.68±0.90^{a}	1.45±0.68^{bc}	nq	2.41±0.36^{ab}	0.33±0.09^{c}
2	q-3-glc	360	26.6	465	303	3.20±0.82^{b}	11.05±4.68^{a}	8.76±2.62^{ab}	9.40±0.27^{ab}	7.88±0.78^{ab}	11.98±0.40^{a}	9.28±2.72^{ab}
3	k-3-gal	284, 312 sh, 347	27.1	449	287	nq	nq	nq	nq	nq	nq	nq
4	q-3-rhmglc	255, 289 sh, 346	27.9	611	465, 303	23.41±0.28^{b}	54.91±5.02^{a}	44.32±8.48^{a}	45.91±5.42^{a}	50.64±1.52^{a}	54.64±0.78^{a}	45.78±8.78^{a}
5	k-3-glc	352	29.3	449	287	48.15±1.52^{a}	16.78±6.00^{bc}	19.87±4.09^{b}	20.20±2.79^{b}	7.58±0.71^{cd}	1.79±0.43^{d}	9.86±7.16^{bcd}
6	q-3-(6”-mal)-glc	-	30.9	551	303	1.22±0.18^{bc}	nq	0.89± 0.36^{c}	2.04±0.50^{ab}	1.80±0.51^{abc}	1.60±0.21^{abc}	2.23±0.11^{a}
7	k-3-rhmglc	265, 284 sh, 346	38.1	595	449, 287	15.81±0.98^{abc}	7.73±4.60^{c}	14.39±6.11^{abc}	12.35±3.86^{bc}	22.87±1.31^{a}	16.78±0.58^{abc}	16.99±0.41^{ab}
8	i-3-gal	-	39.7	479	317	nq	nq	nq	nq	nq	nq	nq
9	i-3-glc	-	42.5	479	317	0.49±0.29^{b}	nq	nq	nq	1.01±0.12^{a}	0.56± 0.09^{b}	0.45±0.03^{b}
10	k-3-(6”-mal)-glc	347	43.0	535	287	0.85±0.04^{a}	nq	nq	nq	nq	nq	0.54±0.10^{b}
11	i-3-rhmglc	356	43.8	625	317	2.90±0.04^{c}	6.64±1.74^{b}	5.15±0.93^{bc}	5.97±1.16^{bc}	8.22±0.65^{ab}	10.25±0.94^{a}	6.73±1.77^{b}
12	k-3-(2”-mal)-glc	-	46.5	535	449, 287	1.18±0.19^{b}	1.53±0.35^{b}	2.93±1.00^{b}	2.69±1.36^{b}	nq	nq	7.80±0.71^{a}
	Total kaempferol					65.99±1.23^{a}	26.05±10.17^{bc}	37.19±8.78^{b}	35.24±5.21^{bc}	30.45±0.62^{bc}	18.57±0.17^{c}	35.19±7.96^{bc}
	Total quercetin					30.62±0.95^{b}	67.31±9.12^{a}	57.66±9.64^{a}	58.79±6.35^{a}	60.32±1.20^{a}	70.62±1.13^{a}	57.63±6.20^{a}
	Total isoramnetin					3.39±0.28^{d}	6.64±1.74^{bcd}	5.15±0.934^{cd}	5.97±1.16^{bcd}	9.23±0.65^{ab}	10.81±1.03^{a}	7.18±1.77^{bc}
	Total flavonols[2]					28.52±4.84 ^{b}	89.20±39.00^{a}	15.08±4.19^{b}	42.27±3.48^{b}	7.90±1.25^{b}	15.99±0.88^{b}	18.90±1.99^{b}

*Q, quercetin; K, kaempferol; I, isorhamnetin; gal, galactoside; glc, glucoside; rhmglc, 6”-rhamnosyl-glucoside; (6”-mal)-glc, 6”-malonyl-glucoside; ARA, Araponga; CAN, Canaã; RL, Rosário de Limeira; RN, Rio Novo do Sul; RP, Rio Pomba; VA, Vargem Alta; VIC, Viçosa; Underlined *m/z* value in the MS/MS spectra indicated the most intense signal; [1]Peak numbers used in Figure 2. [2]expressed in mg of kaempferol-3-glucoside/100 g of fruit. [3]NQ, non quantified. [4]Results as mean ± SD from triplicates. Different superscript letters between regions denote significant differences.

al., 2013); the relative abundance of their fragments allowed their assignation.

Peaks 1, 2, 4, and 6 were identified as Q-3-gal, Q-3-glc, Q-3-rhmglc and Q-3-(6”-mal)-glc based on matching their retention times and MS/MS spectra with those of standards. Isorhamnetin derivatives are the minor flavanols found in purified extracts of Jussara. I-3-glc (peak 9) and I-3-rhmglc (peak 11) were identified by matching their retention times and MS/MS spectra with those of commercial standards. I-3-gal (peak 8) had a MS/MS spectrum matching data from the literature.

The flavonols of *Euterpe edulis* fruit had not been deeply studied until now. In this research, the presence of 11 flavonols was identified in Jussara pulp from Minas Gerais, which had not yet been described in the literature for this fruit. Table 3 shows the molar percentage of each flavonol and the total flavonol content reported in the fruits of each region. The flavonol profiles reported in the fruits of each region were very similar to each other. As reported for anthocyanin compounds, only the presence of mono- or di-substituted flavonols in ring B is observed.

Fruits of the Canaã region presented the higher total flavonol content (89.2 mg/ 100 grams of fruit) than fruits of the other regions. The total quercetin glycosides (total quercetin) were predominant in fruits of all regions except Araponga. The main flavonol in all regions was q-3-rhmglc, where the fruits of Araponga have the lowest proportion of this compound (23.41%).

The k-3-rhmglc is the main kaempferol derivative present in fruits from Rio Pomba (22.87%), Vargem Alta (16.78%) and Viçosa (16.99%). Conversely k-3-glc predominates in fruits from Araponga (48.15%), Canaã (16.78%), Rosário de Limeira (19.87%) and Rio Novo do Sul (20.20%). Isorhamnetin derivatives are the flavonols in lowest concentration in Jussara.

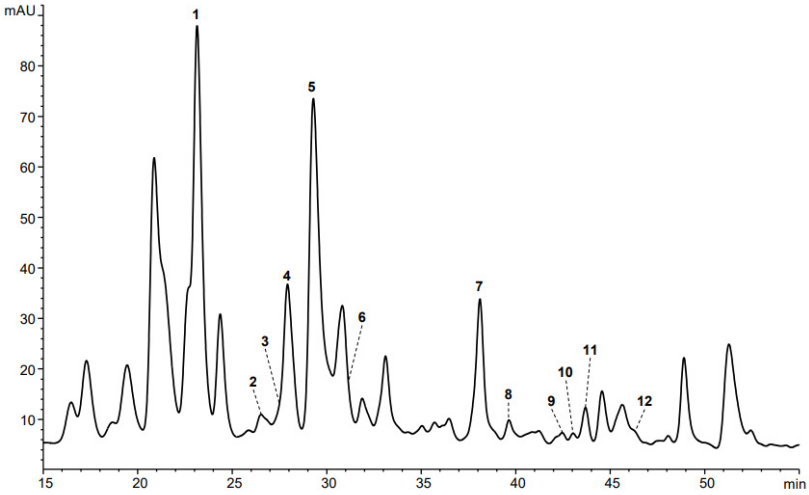

Figure 2 - HPLC–DAD chromatogram (detectionat360 nm) for the flavanols from Jussara fruit pulp (sample from Araponga, MG, Brazil). Peaks numbering indicated in the table 3.

Monomers and dimers of flavan-3-ols and resveratrol from Jussara pulp

Two monomers and eight dimers of flavan-3-ols were found in the Jussara pulp, as well as two resveratrol glucosides (Table 4).

Monomers are represented only by (+)-catechin and (−)-epicatechin ([M-H]$^-$ signal at m/z 289 and MS/MS at m/z 164 and 137). The m/z 289 signal in negative ion mode indicates the presence of flavan-3-ol monomers (MONAGAS et al., 2003); the fragments m/z 164 and 137 may be due to fission in the heterocyclic ring and retro-Diels–Alder fission, respectively (FLAMINI & TRALDI, 2009).

Mass spectra ([M-H]$^-$ signal at m/z 577) confirmed the presence of B-type procyanidin dimers (PB1, PB2, and PB4) and other (epi) catechin dimers for which the structures could not be defined with the current data. These compounds fragmented under MS/MS conditions, releasing ions at m/z 425 and 407. The first fragment results from the loss of ring B by retro-Diel–Alder fission of the upper unit, and the second fragment from a subsequent loss of water (DI LECCE et al., 2014). (+)-Catechin and (−)-epicatechin have been identified in Jussara fruit from Santa Catarina (BORGES et al., 2011). The dimers of flavan-3-ols had not been reported in Jussara fruits until now.

The catechin and epicatechin monomers were the major flavan-3-ols found in Jussara. The proanthocyanidins B1, B2, and B4, dimers formed by catechin and epicatechin units, were also detected at lower concentrations.

The plant growth region influenced the flavan-3-ol content of the fruits. The total content of flavan-3-ols varied from 9.75 mg of catechin/100 g of fruit in Rio Pomba to 49.9 mg of catechin/100 g of fruit in Rio Novo do Sul.

The catechin fraction varies from 32.41% in Rio Novo do Sul to 60.56% in Rio Pomba. The proportion of epicatechin is lower in Rio Pomba (18.86%) than in the other regions studied. The unknown dimer with a retention time of 20.43 min (dimer-3) is the third most abundant flavan-3-ol in fruits of all regions, except for Viçosa, which has a greater fraction of PB1.

Regarding stilbenes, resveratrol 3-glucoside isomers (*cis* and *trans*) were confirmed because of the presence of the deprotonated molecular ion of m/z 389 with fragments of m/z 227 and 185. The first fragment is caused by the loss of one molecule of glucose ([M-162]$^-$), and the second fragment by the subsequent loss of a C_2OH_2 group [M-162-C_2OH_2]$^-$(BUIARELLI et al., 2007). Resveratrol in its free form had already been identified in Jussara by SCHULZ et al. (2015), but its glucosides had not yet been reported.

The proportion of *cis* and *trans*-resveratrol glycoside was affected by the raw material region of origin. The total resveratrol content was derived from the glycosylated isomers of resveratrol, varying from

Table 4 - Retention time, mass spectra and molar percentage of the flavan-3-ol monomers and dimers, and stilbenes identified in samples of the Jussara fruit pulp by means of MRM* experiments.

				%Molar						
Compound	R_t (min)	[M-H]⁻ (*m/z*)	MS/MS (*m/z*)	ARA	CAN	RL	RN	RP	VA	VIC
Flavan-3-ols										
(+)-catechin	25.60	289	164, 137	42.69±5.31^{b}	36.53±11.29^{b}	46.49±4.55^{ab}	32.41±4.95^{b}	60.56±0.56^{a}	35.98±1.83^{b}	32.77±9.06^{b}
(-)-epicatechin	32.64	289	164, 137	36.82±3.54^{a}	39.40±4.90^{a}	32.58±0.81^{a}	40.84±7.49^{a}	18.86±0.59^{b}	39.11±2.15^{a}	40.92±7.32^{a}
PB1	21.80	577	425; 407	7.09±1.12^{ab}	7.50±1.75^{ab}	7.37±2.53^{ab}	7.36±1.12^{ab}	6.08±0.61^{b}	5.84±0.07^{b}	10.24±1.25^{a}
PB2	27.40	577	425; 407	1.14±0.19^{bc}	2.12±0.48^{ab}	0.96±0.07^{c}	2.40±0.85^{a}	1.06±0.13^{bc}	2.15±0.36^{ab}	1.47±0.10^{abc}
PB4	23.10	577	425; 407	0.17± 0.04^{b}	nq	0.18±0.03^{b}	0.34±0.10^{a}	nq	nq	nq
dimer-3	20.43	577	425; 407	8.00±0.15^{a}	10.05±3.13^{a}	8.83±2.45^{a}	10.06±3.94^{a}	10.06±0.91^{a}	11.17± 0.51^{a}	8.17± 0.91^{a}
dimer-4	26.10	577	425; 407	2.80±0.65^{ab}	3.79±1.11^{ab}	2.20±0.72^{ab}	4.99±1.63^{a}	1.88±0.34^{b}	4.45±1.30^{ab}	4.86±1.43^{ab}
dimer-5	30.80	577	425; 407	0.64±0.04^{a}	nq	0.70±0.22^{a}	0.78±0.09^{a}	0.68±0.19^{a}	0.53±0.11^{a}	0.80±0.12^{a}
dimer-6	32.00	577	425; 407	0.50±0.05^{a}	0.58± 0.11^{a}	0.55±0.19^{a}	0.57±0.08^{a}	0.66±0.04^{a}	0.66±0.04^{a}	0.54±0.18^{a}
dimer-7	38.30	577	425; 407	0.14±0.016^{ab}	nq	0.13± 0.01^{b}	0.25±0.05^{a}	0.16±0.02^{ab}	0.20± 0.06^{ab}	0.21±0.06^{ab}
Total flavan-3-ols[1]				39.02±14.80^{ab}	36.42±15.77^{ab}	15.82±4.26^{ab}	49.9±27.7^{a}	9.75±3.86^{b}	14.42±4.87^{ab}	22.59±10.54^{ab}
Stilbenes										
trans-resveratrol-3-O-glucoside	38.25	389	227; 185	52.18±2.59^{ab}	90.79±3.88^{a}	68.76±6.61^{ab}	48.77±9.69^{ab}	42.00±41.70^{ab}	40.93±4.06^{ab}	25.00±43.20^{b}
cis-resveratrol-3-O-glucoside	43.80	389	227; 185	47.82±2.59^{ab}	9.21±3.88^{b}	31.24±6.61^{ab}	51.23±9.69^{ab}	58.00±41.70^{ab}	59.07±4.06^{ab}	75.00±43.20^{a}
Total resveratrol[2]				0.09±0.01^{b}	0.91±0.61^{a}	0.12±0.05^{ab}	0.69±0.45^{ab}	0.01± 0.00^{b}	0.09±0.05^{b}	0.02±0.01^{b}

*ARA, Araponga; CAN, Canaã; RL, Rosário de Limeira; RN, Rio Novo do Sul; RP, Rio Pomba; VA, Vargem Alta; VIC, Viçosa; The m/z value in the [M-H]⁻ column corresponds to the deprotonated molecule generated in the MS spectra, whereas the couple of *m/z* values in the MS/MS column are the selected fragment ions used for the identification of each compound. [1]mg of catechin/100 g of fruit. [2]mg of resveratrol glycoside/100 g of fruit.. Different superscript letters between regions denote significant differences.

0.01 mg/ 100 grams of fruit in Rio Pomba to 0.91 mg/ 100 grams of fruit in Canaã.

Hydrolysable tannins by HPLC-DAD-ESI-MS/MS

No hydrolyzable tannins were reported in the Jussara samples. Under the analysis conditions, gallic acid (in its esterified form) elutes at 26.805 min and ellagic acid elutes at 51.632 min. However, no peaks were observed at these times for the hydrolysed sample; in other words, these substances were not present in the sample. This fact was proven with co-injections of the sample mixed with the standards. Besides the UV-Vis spectra, the MS/MS spectra were analysed, in which no m/z 183 or 301 ions were detected, which would indicate the presence of methyl gallate and ellagic acid, respectively.

CONCLUSION

The analytical methodology used (HPLC-DAD-ESI-MS/MS) allowed the identification of compounds that had not yet been reported in this fruit. To date, there has been no research published on the determination of hydrolyzable tannins in *Euterpe edulis*; therefore, this is the first research that analyzed the possible occurrence of this group of compounds.

The anthocyanins identified in the Jussara were derivatives of cyanidin, peonidin, and pelargonidin. Pentosides, galactosides, and sophorosides of cyanidin, cyanidin-3-pentosylgalactoside, and cyanidin-3-pentosylglucoside were first described in the literature for this fruit. The flavonols of Jussara had not been well described in the literature until now. In this study, the presence of kaempferol, quercetin, and isorhamnetin derivatives was tentatively suggested. (+)-Catechin, (−)-epicatechin, procyanidins B1, B2 and B3, and four unknown (epi)catechin dimers were reported among the detected flavan-3-ols. Resveratrol was found only in its glucosylated form.

ACKNOWLEDGMENTS

The authors are thankful to the financial support provided by FAPEMIG [grant numbers 1943) and thankful Isidro Hermosín-Gutiérrez and Universidad Castilla-La Mancha for allowing the doctoral internship abroad.

DECLARATION OF CONFLICT OF INTEREST

There is no conflict of interest regarding the development and publication of the study.

AUTHORS' CONTRIBUTIONS

All authors contributed equally for the conception and writing of the manuscript. All authors critically revised the manuscript and approved of the final version.

REFERENCES

ARDREY, R. E. **Liquid Chromatography-Mass Spectrometry**: an Introduction, Analytical Techniques in the Sciences (AnTS*)*. John Wiley & Sons, Ltd, Chichester, UK (2003).

BICUDO, M. O. P., et al. Anthocyanins, Phenolic Acids and Antioxidant Properties of Juçara Fruits (*Euterpe edulis* M.) Along with the On-tree Ripening Process. **Plant Foods Hum. Nutr**. v.69, p.142–147, 2014. Available from:<https://pubmed.ncbi.nlm.nih.gov/24570272/>. Accessed: Nov. 12, 2019. doi: 10.1007/s11130-014-0406-0.

BERNARDES, A. L; et al. *In vitro* bioaccessibility of microencapsulated phenolic compounds of jussara *(Euterpe edulis* Martius) fruit and application in gelatine model-system. **LWT - Food Science and Technology**, v.102, p.173-180, 2019. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0023643818310673>. Accessed: Nov. 12, 2019. doi: 10.1016/j.lwt.2018.12.009.

BORGES, G. D. S. C., et al. Chemical characterization, bioactive compounds, and antioxidant capacity of jussara (*Euterpe edulis*) fruit from the Atlantic Forest in southern **Brazilian Journal of Food Research**. v.44, p.2128–2133, 2011 Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0963996910004746>. Accessed: Nov. 12, 2019. doi: 10.1016/j.foodres.2010.12.006.

BRITO, E. S. de, et al. Anthocyanins Present in Selected Tropical Fruits: Acerola, Jambolão, Jussara, and Guajiru. **Journal of Agricultural and Food Chemistry**. v.55, p.9389–9394, 2007. Available from: <https://pubs.acs.org/doi/abs/10.1021/jf0715020> Accessed: Dec. 11, 2019. doi: 10.1021/jf0715020.

BUIARELLI, F., et al. Analysis of some stilbenes in Italian wines by liquid chromatography / tandem mass spectrometry. **Rapid Communication in Mass Spectrometry**, v.21, p.2955–2964, 2007. Available from: <https://analyticalsciencejournals.onlinelibrary.wiley.com/doi/10.1002/rcm.3174>. Accessed: Dec. 12, 2008. doi: 10.1002/rcm.3174.

CARVALHO, A. G. S.; et al. Anthocyanins from jussara (*Euterpe edulis* Martius) extract carried by calcium alginate beads pre-prepared using ionic gelation. **Powder Technology**, v.345, p.283-291, 2019 Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0032591019300166>. Accessed: Jan. 08, 2020. doi: 10.1016/j.powtec.2019.01.016.

CARAZZONE, C., et al. Identification of phenolic constituents in red chicory salads (*Cichorium intybus*) by high-performance liquid chromatography with diode array detection and electrospray ionisation tandem mass spectrometry. **Food Chemistry**. v.138, p.1062–1071, 2013. Available from: <https://pubmed.ncbi.nlm.nih.gov/23411215/>. Accessed: Nov. 12, 2019. doi: 10.1016/j.foodchem.2012.11.060.

CASTILLO-MUÑOZ, N., et al.Flavonol 3- O -Glycosides Series of Vitis vinifera Cv. Petit Verdot Red Wine Grapes. **Journal of Agricultural Food Chemistry**. v.57, p.209–219, 2009. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0889157510001638>. Accessed: Oct. 12, 2019. doi: 10.1016/j.jfca.2010.03.017.

DALL`ASTA, M., et al. Identification of microbial metabolites derived from *in vitro* fecal fermentation of different polyphenolic food sources. **Nutrition**, v.28, p.197–203, 2012. Available from: <https://pubmed.ncbi.nlm.nih.gov/22208556/>. Accessed: Nov. 12, 2019. doi: 10.1016/j.nut.2011.06.005.

DE SOUZA, V.R, et al. Determination of the bioactive compounds, antioxidant activity and chemical composition of Brazilian blackberry, red raspberry, strawberry, blueberry and sweet cherry fruits. **Food Chemistry**, v.156, p.362-368, 2014. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0308814614001770>. doi: 10.1016/j.foodchem.2014.01.125. Accessed: Oct. 07, 2019.

DI LECCE, G., et al. Phenolic profiling of the skin, pulp and seeds of Albari?o grapes using hybrid quadrupole time-of-flight and triple-quadrupole mass spectrometry. **Food Chemistry**. v.145,p.874–882,2014.Available from: <https://www.sciencedirect.com/science/article/abs/pii/S030881461301217X>. Accessed: Oct. 08, 2019. doi: 10.1016/j.foodchem.2013.08.115.

FLAMINI, R., TRALDI, P. **Mass Spectrometry in Grape and Wine Chemistry, Mass Spectrometry in Grape and Wine Chemistry.** John Wiley & Sons, Inc., Hoboken, NJ, USA. 2009.

HARBORNE, J.B., et al. Anthocyanins of Cephaelis, Cynomorium, Euterpe, Lavatera and Pinanga. **Biochemical Systematics and Ecology.** v.22, p.835–836, 1994. Available from: <https://www.sciencedirect.com/science/article/pii/0305197894900884>. Accessed: Nov. 12, 2019. doi: 10.1016/0305-1978(94)90088-4.

GARCIA, J. A. A., et al. Chemical composition and biological activities of Juçara (Euterpe edulis Martius) fruit by-products, a promising underexploited source of high-added value compounds. **Journal Functional Foods**, v.55, p.325-332, 2019. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S1756464619300982>. Accessed: Nov. 12, 2019. doi: 10.1016/j.jff.2019.02.037.

INADA, K. O. P., et al. Screening of the chemical composition and occurring antioxidants in jabuticaba *(Myrciaria jaboticaba*) and jussara (Euterpe edulis) fruits and their fractions. **Journal Functional Foods**, v.17, p.422–433, 2015. Available from: <https://www.sciencedirect.com/science/article/pii/S1756464615002868> Accessed: Dec. 06, 2019. doi: 10.1016/j.jff.2015.06.002.

LOPES-DA-SILVA, F., et al. Identification of anthocyanin pigments in strawberry (*cv Camarosa*) by LC using DAD and ESI-MS detection. **European Food Research and Technology**, v.214, p.248–253,2002. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0308814605002682>. Accessed: Nov. 12, 2019. doi: 10.1016/j.foodchem.2005.02.047.

MONAGAS, M., et al. Monomeric , Oligomeric , and Polymeric Flavan-3-ol Composition of Wines and Grapes from Vitis. **Journal**

Agric. Food Chem. v.51, p.6475−6481, 2013. Available from: <https://pubmed.ncbi.nlm.nih.gov/14558765/>. Accessed: Nov. 12, 2019. doi: 10.1021/jf030325.

PENG, S., et al.. Insoluble ellagitannins in Castanea sativa and Quercus petraea woods. **Phytochemistry**. v.30, p.775–778, 1991. Available from: < https://www.sciencedirect.com/science/article/abs/pii/0031942291852504 > Accessed: Nov. 22, 2019. doi. org/10.1016/0031-9422(91)85250-4.

PUPIN, L.; et al. Is the bioaccessibility of minerals affected by the processing steps of juçara fruit (*Euterpe edulis* Mart.) **LWT - Food Science and Technology**, v.91, v.14-25, 2018. Available from: <https://www.sciencedirect.com/science/article/pii/S0023643818300240> Accessed: Nov. 22, 2019. doi: 10.1016/j.lwt.2018.01.024.

REBELLO, L. P. G., et al. Phenolic composition of the berry parts of hybrid grape cultivar BRS Violeta (BRS Rubea×IAC 1398-21) using HPLC–DAD–ESI-MS/MS. **Food Research International**. v.54, p.354–366, 2013. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0963996913003906> Accessed: Nov. 20, 2019. doi: 10.1016/j.foodres.2013.07.024.

RUIZ, A., et al. Anthocyanin profiles in south Patagonian wild berries by HPLC-DAD-ESI-MS/MS. **Food Research International.** v.51, p.706–713, 2013. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0963996913000707>. Accessed: Nov. 25, 2019. doi: 10.1016/j.foodres.2013.01.043.

SANTANA, A. A., et al. Influence of different combinations of wall materials on the microencapsulation of jussara pulp (*Euterpe edulis*) by spray drying. **Food Chemistry**, v.212, p.1-9, 2016. Available from: <https://www.sciencedirect.com/science/article/pii/S0308814616308378>. Accessed: Dec. 10, 2019. doi: 10.1016/j.foodchem.2016.05.148.

SILVA, F. P., et al. Low dose of Juçara pulp (*Euterpe edulis* Mart.) minimizes the colon inflammatory milieu promoted by hypercaloric and hyperlipidic diet in mice. **Journal of Functional Foods**, 2021, v.77, p.104343. Available from: <https://www.sciencedirect.com/science/article/pii/S1756464620305673>. Accessed: Dec. 02, 2022. doi: 10.1016/j.jff.2020.104343.

SCHULZ, M., et al. Bioaccessibility of bioactive compounds and antioxidant potential of juçara fruits (*Euterpe edulis* Martius) subjected to in vitro gastrointestinal digestion. **Food Chemistry**. v.228, p.447–454, 2017. Available from: <https://pubmed.ncbi.nlm.nih.gov/28317748/>. Accessed: Nov. 18, 2019. doi: 10.1016/j.foodchem.2017.02.038.

SCHULZ, M., et al. Chemical composition , bioactive compounds and antioxidant capacity of juçara fruit (*Euterpe edulis* Martius) during ripening. **Food Research International**. v.77, p.125–131, 2015. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0963996915301423>. Accessed: Nov. 24, 2019. doi: 10.1016/j.foodres.2015.08.006.

SEERAM, N. P., et al. Identification of phenolic compounds in strawberries by liquid chromatography electrospray ionization mass spectroscopy. **Food Chemistry.** v.97, p.1–11, 2006 Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0308814605002682>. Accessed: Dec. 10, 2019. doi: 10.1016/j.foodchem.2005.02.047.

SEERAM, N. P. Berry fruits: Compositional elements, biochemical activities, and the impact of their intake on human health, performance, and disease. **Journal of Agricultural and Food Chemistry**, v.56, p.627-629, 2008. Available from: <https://pubmed.ncbi.nlm.nih.gov/18211023/>. Accessed: Dec. 02, 2019. doi: 10.1021/jf071988k.

SILVA, N.A., et al. Phenolic Compounds and Carotenoids from Four Fruits Native from the Brazilian Atlantic Forest. **Journal Agricultural in Food Chemistry**. 62, 5072–5084, 2014. Available from: <https://pubs.acs.org/doi/10.1021/jf501211p>. Accessed: Oct. 10, 2019. doi: 10.1021/jf501211p.

TIWARI, U.; CUMMINS, E. Factors influencing levels of phytochemicals in selected fruit and vegetables during pre- and post-harvest food processing operations. **Food Research International,** v.*50*(2), p.497–506, 2013. Available from: <https://www.sciencedirect.com/science/article/abs/pii/S0963996911005370>. Accessed: Oct. 12, 2019. doi: 10.1016/j.foodres.2011.09.007.

VIEIRA, G. S.; et al. Influence of nanofiltration membrane features on enrichment of jussara ethanolic extract (*Euterpe edulis)* in anthocyanins. **Journal of Food Engineering**, v.226, p.31-41, 2018. Available from: <https://www.sciencedirect.com/science/article/pii/S0260877418300256>. Accessed: Oct. 05, 2019. doi: 10.1016/j.jfoodeng.2018.01.013.