Food Bioscience 52 (2023) 102501

Contents lists available at ScienceDirect

Food Bioscience

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

Bioaccessibility of phenolic compounds from Brazilian grape juices using a digestion model with intestinal barrier passage

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ARTICLE INFO

Keywords:
Grape cultivars
Functional beverages
Phytochemicals
Digestion simulation
Bioactivity

ABSTRACT

Grape juices are rich in bioactive compounds; however, for these compounds to exert their functionality, they must be bioaccessible. Thus, the present study evaluated a simulated digestion process on the main bioactive compounds of monovarietal grape juices of five Brazilian hybrid cultivars (*V. vinifera x V. labrusca*). Characterization of the chemical profiles in liquid chromatography (HPLC-DAD-RID), behaviour of phenolics in the stages of digestion and bioaccessibility through the INFOGEST protocol plus intestinal barrier passage were carried out. Of the 24 polyphenols identified in the grape juice samples, 11 were bioaccessible, with emphasis on the class of flavanols. Procyanidin B2 (101–426%), (+)-catechin (169–370%) and gallic acid (61–230%) stood out in all juices, showing that these compounds are key to the functionality of these drinks. Particularities were observed to differ between juices, demonstrating that factors such as the cultivar should be explored more extensively in studies on functional foods. The study also suggests that quality components such as sugars and organic acids influence the bioaccessibility of beverages.

1. Introduction

Grape juices are food matrices rich in phenolic compounds associated with consumer health benefits (De Lima Tavares Toscano et al., 2019; Lima et al., 2022; Toaldo et al., 2016). Recently, beneficial effects of polyphenols on the immune system against SARS-CoV-2 infections have been reported (Augusti et al., 2021; Yun Yang et al., 2022). Among other pharmacological effects of phenolic compounds, the prevention of the risk of heart disease, type 2 diabetes mellitus, different types of cancer, obesity, gut-microbiota-modulating, Alzheimer, Parkinson and neurodegenerative diseases stands out (Moradi et al., 2021; Dwibedi et al., 2022; Zhou et al., 2022; Xia et al., 2010). However, for polyphenols to exert their bioactivity, they must be bioaccessible.

The bioaccessibility of polyphenols is often evaluated using *in vitro* digestion systems, due to the difficulty of *in vivo* evaluation caused by significant changes in the structure of these compounds during passage through the gastrointestinal tract (Celep et al., 2015). Models of gastrointestinal digestion have been studied in order to demonstrate its effects on the bioactive compounds of foods and beverages (Barreto et al., 2023; Carneiro et al., 2022; Lingua et al., 2018; Minekus et al., 2014). After simulating the passage of the sample through the stages of the mouth, stomach and intestine, the application of dialysis can simulate the mobility of compounds through the intestinal barrier (Barreto et al., 2023).

Even so, the use of *in vitro* digestion simulation techniques has been applied as a valuable tool to propose an estimate of the bioaccessibility of bioactive compounds, especially those present in grapes and wines (Lingua et al., 2018, 2019; Sun et al., 2020).

Previous studies that established the bioaccessibility of polyphenols present in grape juice are scarce, and it is known that the chemical composition of the grape juice matrix differs from that of wines, mainly due to the high levels of sugars, profile of organic acids and absence of ethanol (Coelho et al., 2018), and these factors may influence the bioaccessibility of phenolic compounds. Some studies have already shown

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https://doi.org/10.1016/j.fbio.2023.102501
Received 23 December 2022; Received in revised form 31 January 2023; Accepted 19 February 2023
Available online 24 February 2023

changes in the structures of phenolic compounds during their passage through the gastrointestinal tract that caused changes in accessibility related to transport through intestinal cells, due to acid-alkaline conditions and enzymatic action (Lingua 2018; Celep et al., 2015; Tagliazucchi et al., 2010; Silva Haas, Marmitt, Toaldo Fedrigo, Goettert, & Bordignon-Luiz, 2020). Additionally, the composition of the food matrix is a determinant of the bioaccessibility of phytochemicals, and thus it is important to verify the effect of interactions between its components (Mohamedshah et al., 2020).

The Sub-middle São Francisco Valley (SSFV) is an important region of Brazil that has invested in the elaboration of high-quality grape juices produced with hybrid cultivars (*V. vinifera x V. labrusca*) adapted to the tropical climate, such as Isabel Precoce, BRS Violeta, BRS Magna, BRS Cora and BRS Carmem (Dutra et al., 2021; Lima et al., 2014, 2022). Previous works with SSFV grape juice were focused on chemical characterization, and no information was found on the bioaccessibility of the phenolic components of these products.

In this context, we evaluated the effect of the digestion process, using a simulated intestinal barrier, on the bioaccessibility of bioactive phenolic compounds present in Brazilian grape juices. Additionally, the association of the grape cultivar used, composition of sugars and organic acids in the matrix and other physicochemical characteristics of the juice on bioaccessibility were also studied.

2. Material and methods

2.1. Raw material and juice processing

Five grape cultivars (Isabel Precoce, BRS Cora, BRS Violeta, BRS Magna and BRS Carmem) were studied. Grapes were harvested in April 2021 and kindly provided by companies localized in Petrolina-PE (latitude 09°21′S and longitude 40°40′W) and Lagoa Grande-PE (latitude 8°59′ S and longitude 40°16′W). Isabel Precoce came from the company Grand Valle Industrial Ltda; the cultivars BRS Violeta, BRS Magna and BRS Carmem from the company Queiroz Galvão Alimentos S/A company, Timbaúba Farm and the cultivar BRS Cora from the company Asa Indústria e Comércio Ltda. For determine the ideal harvest point, during ripeness period and in the day of harvest of the all grape cultivars evaluated, a sampling of 200 grapes berries was randomly collected from the basal, middle and apical regions of grape bunches of the plots groups (4 plots, each plot with 50 berries) and subjected to analysis of the soluble solids and titratable acidity. This procedure guaranteed that were harvested ripe grapes, with soluble solids among 16–18° Brix, and titratable acidity of 0.6–0.7% (expressed as tartaric acid). Monovarietal grape juices were prepared using the hot press method as described by Silva et al. (2019). The juices were elaborated in three repetitions, where each repetition was equivalent to a batch containing 10kg of grapes. The treatments consisted of the monovarietal juices of the five cultivars, totaling 15 process batches. The bottles of each process batch were analysed in triplicate, totaling 45 analysed samples.

2.2. Chemicals

Alpha-amylase, pepsin, pancreatin, bile salts, 12 kDa dialysis bags, TPTZ reagent (2,4,6-Tri(2-pyridyl)-*s*-triazine), Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid), 2,2-azino-bis (3-ethylbenzothiazoline-6 sulfonic acid) (ABTS•+), 2,2-diphenyl-1-picrylhydrazyl (DPPH•) and ferric chloride hexahydrate were obtained from Sigma-Aldrich (St. Louis, MO, USA). Ultrapure water was obtained by purification in a Marte Científica System (São Paulo, SP, Brazil). Ethyl alcohol, potassium persulfate, phosphoric acid, ferrous sulphate, potassium monobasic phosphate, potassium chloride, potassium dihydrogen phosphate, sodium bicarbonate, sodium chloride, magnesium chloride hexahydrate, ammonium chloride, sodium hydroxide, hydrochloric acid, calcium chloride, orthophosphoric acid and sulfuric acid were purchased from Merck (Darmstadt, Germany). Standards of malic, citric, tartaric, succinic and formic acids, glucose and fructose were obtained from Vetec chemistry (Rio de Janeiro, RJ, Brazil). HPLC grade methanol was obtained from J.T. Baker (Phillipsburg, NJ, USA). Standards for the quantification of phenolic compounds, including procyanidin A2, (−)-epicatechin gallate, quercetin-3-*O*-rutinoside (rutin), (−)-epigallocatechin gallate, kaempferol-3-*O*-glucoside, quercetin-3-β-D-glucoside, myricetin, cyanidin-3-*O*-glucoside, malvidin-3-*O*-glucoside, peonidin-3-*O*-glucoside, delphinidin-3-*O*-glucoside, petunidin-3-*O*-glucoside and pelargonidin-3-*O*-glucoside were from Extrasynthese (Genay, France). Caffeic acid, gallic acid, ρ-coumaric acid, chlorogenic acid, caftaric acid, syringic acid, hesperidin, procyanidins B1 and B2, (+)-catechin, (−)-epicatechin, naringenin, cyanidin-3,5-*O*-diglucoside, malvidin-3,5-*O*-diglucoside and pelargonidin-3,5-*O*-diglucoside were from Sigma-Aldrich. The isomers *trans*-resveratrol and *cis*-resveratrol were obtained from Cayman Chemical Company (Michigan, USA).

2.3. Classical quality parameters

Classical quality parameters of grape juices were analysed, following the methodologies described by the International Organization of Grapes and Wine - OIV (OIV–International Organisation of Vine and Wine, 2022). Briefly, the pH was measured using a PHS-3B digital potentiometer (Tecnal, Brazil). SS (in °Brix) were analysed using a HI 96801 digital refractometer (Hanna, USA), titratable acidity (TA) by titrimetry with 0.1 M NaOH solution, colour intensity from the sums of absorbances at 420, 520 and 620 nm and hue by the ratio of absorbances at 420 and 520 nm, measured in a UV 2000A UV–Visible spectrophotometer (Instrutherm, Brazil).

2.4. Antioxidant capacity

The antioxidant capacity (AOX) of the samples was evaluated through *in vitro* assays by the DPPH• (Kim et al., 2002), ABTS•+ (Re et al., 1999) and FRAP (Rufino et al., 2006) methods. For the FRAP method, ferrous sulphate was used for the calibration curve and responses expressed in mmol Fe^{2+} per litre of grape juice (mmol Fe^{2+} L^{-1}). For the DPPH• and ABTS•+ free-radical-scavenging methods, the Trolox analytical standard was used to construct the calibration curves and the results expressed in Trolox equivalents per litre of juice (mmol TE L^{-1}). Briefly, the DPPH• assay was performed with a solution of DPPH• (1 mmol L^{-1}) in absolute ethanol and diluted to an absorbance of 0.900 ± 0.050 (100 μmol L^{-1}). The absorption of free radicals in the solution was measured by the absorbance decay rate at 517 nm, determined at 30 min of incubation. The $ABTS^{\bullet+}$ radical-scavenging assay was performed using a solution of 7 mmol ABTS•+ with 140 mmol of potassium persulfate incubated at 27 °C without incident light for 16 h. The radical was diluted in ethanol to an absorbance of 0.700 ± 0.050 at 734 nm. For the AOX analysis, an aliquot of 30 μL of the juice was combined with 3000 μL of the $ABTS^{\bullet+}$ radical, and the readings were performed 6 min after addition of the sample in a dark environment. In the test with FRAP reagent, a solution of acetate (300 mmol; pH 3.6), 10 mmol TPTZ (2,4,6-Tri(2-pyridyl)-*s*-triazine) in 40 mmol HCl and 20 mmol of $FeCl_3$ solution. A mixture of 90 μL of juice, 270 μL of deionized water and 2.7 mL of FRAP reagent was prepared and incubated at 37 °C in a thermoreactor (AAKER model IT2002, Brazil) for 30 min. Absorbance was measured at 595 nm in a spectrophotometer. All absorbance readings were performed with a spectrophotometer UV–Vis 2000A model (Instrutherm, Brazil).

2.5. Bioactive phenolic compounds, sugars and organic acids

Phenolic compounds, sugars and organic acids were determined by high-performance liquid chromatography (HPLC), using an Agilent 1260 Infinity LC System liquid chromatograph (Agilent Technologies, Santa Clara, USA) coupled to an index and refraction detector (RID) and

a diode array detector (DAD). Data were processed using OpenLAB CDS ChemStation Edition software (Agilent Technologies, Santa Clara, USA).

The determination of phenolics was performed on an HPLC-DAD, following the methodology described by Padilha et al. (2017) and Dutra et al. (2018). The column used was the Eclipse Plus RP-C18 (100 × 4.6 mm, 3.5 μm) Zorbax, with a C18 precolumn (12.6 × 4.6 mm, 5 μm) (Zorbax, USA). The gradient elution were 0–5 min: 5% phase B and 95% phase A; 5–14 min: 23% B; 14–30 min: 50% B; 30–33 min 80% B. Phase A consisted of phosphoric acid in aqueous solution at 0.52% (pH = 2.0), and phase B was methanol acidified with 0.52% orthophosphoric acid (H_3PO_4). The juices were previously diluted (500 μL of juice + 1000 μL of phase A) and filtered through a 0.45-μm membrane and 20 μL was injected. The quantification of phenolic compounds was performed using calibration curves of external standards ($R^2 > 0.998$).

Sugars and organic acids were determined by HPLC-DAD/RID (Coelho et al., 2018). An ion-exchange column (300 × 7.7 mm) with 8.0 μm internal particles (Hi Plex H, Zorbax) and the precolumn (5 × 3 mm) (Agilent Technologies, Santa Clara, CA, USA) were used. Column oven temperature maintained at 70 °C and solvent flow of 0.7 mL min^{-1}. The mobile phase consisted of a 4 mmol L^{-1} solution of sulfuric acid (H2SO4). Grape juice was previously diluted in ultrapure water (500 μL of juice + 1000 μL of ultrapure water), filtered through a 0.45-μm nylon membrane (Millex Millipore, Barueri, SP, Brazil) and injected in a volume of 10 μL. Organic acids were detected on a DAD (210 nm) and sugars using an RID.

2.6. Gastrointestinal digestion simulation and bioaccessibility of phenolic compounds

The evaluation of *in vitro* gastrointestinal digestion (GI) of grape juice was performed according to the INFOGEST protocol (Minekus et al., 2014), adding the simulation of passage through the intestinal barrier as described by Barreto et al. (2023). Briefly, a solution containing 5 mL of grape juice, 3.5 mL of simulated salivary fluid (SSF), 25 μL of 0.3 mol L^{-1} $CaCl_2$, 0.5 mL of α-amylase (1500 U mL^{-1}), and 975 μL of ultrapure water was prepared to simulate the oral phase. The pH of the oral phase solution was adjusted to 7.0, and the mixture was incubated for 2 min (37 ± 1 °C at 90 rpm). Soon after, to mimic the gastric phase, 7.5 mL of simulated gastric fluid (SGF) was added, containing 5 μL of $CaCl_2$ 0.3 mol L^{-1}, 1.6 mL of pepsin (25,000 U mL^{-1}), 200 μL of HCl to 1 mol L^{-1} and 0.695 μL of ultrapure water; the pH was adjusted to 3.0, and the material was incubated (37 ± 1 °C for 2 h at 90 rpm). Then, simulation of the intestinal phase was performed by mixing 20 mL of gastric chyme with 11 mL of simulated intestinal fluid (SIF), containing 1 mL of bile salts (25 mg of bile/mL of sample), 5 mL of pancreatin (800 U mL^{-1}), 40 μL of 0.3 mol L^{-1} $CaCl_2$, 150 μL of 1 mol L^{-1} NaOH and 1.31 mL of ultrapure water; the pH was adjusted to 7.0 (1 mol L^{-1} of NaOH) and the solution maintained with shaking (90 rpm for 2 h at 37 ± 1 °C). This mixture (placed inside a dialysis bag 12 kDa) simulated the passive absorption of phenolics through the membrane of the small intestine. Filled bags were immersed in 0.1 M $NaHCO_3$ and incubated in the dark (90 rpm for 2 h at 37 °C). The solution contained inside the bag (nondialyzable fraction) was separated and stored, representing the material that remained in the gastrointestinal tract. The fraction passing through the dialysis membrane (dialysate), was separated, representing the fraction available for absorption into the circulatory system (potentially bioavailable). The bioaccessibility of phenolic compounds was calculated by Eq. (1):

$$Bioaccessibility\ (\%) = \frac{Dialyzed\ fraction}{non-dialyzed\ fraction\ (whole\ grape\ juice)} \times 100$$

2.7. Statistical analysis

Data were subjected to one-way analysis of variance (ANOVA) and compared using the Tukey test at a 5% probability of error. Multivariate analyses were performed using Principal Component Analysis (PCA) and Hierarchical Cluster Analysis (HCA) techniques and the software Past (Paleontological Statistics) version 4.03 (Hammer et al., 2001) and XLStat version 2015 (Addinsoft, Paris, France).

3. Results and discussion

3.1. Physical-chemical parameters and antioxidant capacity of juices

The results obtained for the characterization of monovarietal whole grape juices are shown in Supplementary Table S1. The values obtained for the basic quality parameters were in accordance with identity and quality standards for Brazilian grape juices (Brasil, 2018). Colour intensity varied between cultivars, with higher values for BRS Violeta (30.52) and BRS Cora (26.62) juice.

The average of total sugars quantified by HPLC-RID ranged from 122.76 to 206.54 g L^{-1} in BRS Magna and Isabel Precoce juices, respectively. The sum of the total organic acids varied among the five cultivars, and Isabel Precoce (6.65 g L^{-1}) and BRS Violeta (8.22 g L^{-1}) represented the lower and upper limits, respectively of the range of values. The main organic acids quantified in the juices were tartaric and malic acids. In the present study, the results found were similar to those reported in other studies with Brazilian grape juices (Coelho et al., 2018; Dutra et al., 2021; Lima et al., 2014).

A total of 24 phenolics were quantified in the juices by HPLC-DAD. The juices that presented the highest quantified total amount of phenolic compounds, in descending order, were BRS Magna > BRS Violeta > BRS Cora > Isabel Precoce > BRS Carmem. In the juices from Isabel Precoce, BRS Cora, BRS Magna and BRS Carmem, the main flavanol quantified was procyanidin B2, with average values ranging from 13.26 mg L^{-1} (BRS Carmem) to 16.72 mg L^{-1} (BRS Magna). In the juice of cultivar BRS Violeta, flavanols (−)-epigallocatechin gallate and (+)-catechin stood out, with values of 11.80 and 9.20 mg L^{-1}, respectively. From the group of flavonols, myricetin was predominant in all evaluated juices, with values ranging from 69.20 mg L^{-1} (BRS Carmem) to 178.06 mg L^{-1} (BRS Violeta).

Seven anthocyanins were quantified, the major ones in BRS Cora, BRS Violeta and BRS Magna juices being petunidin-3-*O*-glucoside, with mean values of 115.50, 254.90 and 128.74 mg L^{-1}, respectively. For Isabel Precoce and BRS Carmem juice, the predominant anthocyanin was malvidin-3-5-*O*-diglucoside. Of the stilbene group, *trans*-resveratrol was the most prevalent compound in grapes (Lorenzo et al., 2019), being identified in all juices evaluated at levels from 0.33 to 4.70 mg L^{-1} in BRS Violeta and Isabel Precoce juices, respectively.

From the group of flavanones, the presence of naringenin was identified, and from the class of phenolic acids, gallic acid (7.5–11.1 mg L^{-1}), syringic acid (4.1–7.7 mg L^{-1}), caffeic acid (1.1–2.3 mg L^{-1}) and caftaric acid (272–625 mg L^{-1}). This last phytochemical was also highlighted in other works, for example, as the predominant phenolic acid in SSFV juices and grapes (Dutra et al., 2018, 2021; Padilha et al., 2017).

In the present study, AOX was evaluated by the methods of DPPH• and ABTS•+ free-radical scavenging and by the reducing power of iron (FRAP). In general, all evaluated samples showed high AOX (Table S1), with emphasis on the juice of BRS Cora and BRS Violeta cultivars. Values for DPPH• ranged from 7.25 mM TE L^{-1} (BRS Carmem) to 22.97 mM TE L^{-1} (BRS Cora). For the ABTS•+ method, values ranged from 15.27 mM TE L^{-1} (BRS Magna) to 38.35 mM TE L^{-1} (BRS Cora). In the FRAP method, the values ranged from 26.58 mM Fe^{2+} L^{-1} (BRS Magna) to 78.95 mM Fe^{2+} L^{-1} (BRS Cora). The results found corroborate those of other studies that evaluated the antioxidant capacity of juices from viticulture under tropical climate conditions (Lima et al., 2022; Padilha et al., 2017).

3.2. Bioaccessibility of phenolic compounds

Grape juices are matrices rich in phenolic compounds; however, to exert their physiological effects on consumers, polyphenols need to be bioaccessible. In the present study, the behaviour, release into the matrix and stability of phenolic compounds during digestion simulation and their bioaccessibility in monovarietal grape juices were evaluated. In decreasing order, the juices that presented the highest values of bioaccessibility were BRS Carmem > BRS Cora > BRS Magna > Isabel Precoce > BRS Violeta. Table 1 presents a summary of the bioaccessibility of phenolic compounds in all evaluated juices, where 11 compounds were bioaccessible: 3 flavanols, 4 flavonols, 1 stilbene and 3 phenolic acids. The results found suggest that bioaccessibility is not always directly related to the amount of phenolic compounds initially present in the juice (Table S1). The release and absorption of phytochemicals by the human body is influenced by several factors, including the composition of the food matrix (Ribas-Agustí et al., 2018). According to Alqurashi et al. (2017), both potentially bioaccessible compounds and those available in the colon exert beneficial effects, influencing the health of the digestive and systemic systems.

Regarding the classes of phenolic compounds, after simulation of gastrointestinal digestion of monovarietal juices, the order of bioaccessibility was flavanols > flavonols > phenolic acids > anthocyanins (Fig. 1). The most accessible flavanols for absorption via the intestinal barrier were represented by (+)-catechin, (−)-epigallocatechin gallate and procyanidin B2. The second most bioaccessible class comprised the flavonols, represented by rutin, myricetin, quercetin-3-β-D-glucoside and kaempferol-3-*O*-glucoside. Among the phenolic acids, the most bioaccessible compounds were gallic acid and syringic acid. Generally, the bioaccessibility of the phenolics depending on their composition, glycosylation, molecular weight, and esterification, being associated with the interaction of chemical structures of free hydroxyl groups (Naeem et al., 2022; Xia et al., 2010).

According to Lucas-Gonzalez et al. (2016), the bioaccessibility of flavanols and flavonols can be influenced by the ability of these compounds to bind to proteins or fibres in the matrix, through covalent bonds, hydrophobic interactions or hydrogen bonds, which can increase or decrease their solubility. Studies carried out by Augusti et al. (2021) reported greater bioavailability of flavanols when compared to flavonols, in addition to the authors reporting multiple beneficial effects of these phytochemicals, such as antiviral, antioxidant, immunomodulatory and anti-inflammatory actions, also associated with their systemic effects against SARS-CoV-2 infections.

In general, in the monovarietal grape juices subjected to digestion simulation, the bioaccessible compounds present at higher levels were (+)-catechin (169.1%–265.99%), procyanidin B2 (101.42%–423.13%) and gallic acid (61.59%–230.51%), in all cultivars studied. In all juices, quercetin-3-β-D-glucoside (12%–283.11%), rutin (10.53%–430.73%) and syringic acid (15.29%–111.57%) were also accessible after digestion, in reasonable quantities. Several works that evaluated juices from the cultivars in the present study have highlighted procyanidin B2 as one of the main phenolics found, and its presence is strongly associated with a high antioxidant capacity (Dutra et al., 2018; Lima et al., 2014, 2022; Padilha et al., 2017), highlighting this phenolic as an important bioaccessible compound in the SSFV grape juices.

Regarding (+)-catechin according to Augusti et al. (2021), during the digestion stages, proanthocyanidins could be broken down into (+)-catechin monomers. This mechanism of broken may explain by the high bioaccessibility of this flavanol in the grape-derived beverages, corroborating with the data obtained in this study to the different grape cultivars used to produce the juices. In fact occurred a reduction in the procyanidin A2 and B1 contents during the digestion simulations, possibly associated with an increase in the (+)-catechins concentration. The high bioaccessibility of the (+)-catechin was also highlighted in Syrah grapes in the research carried out by Lingua et al. (2018).

Tables 2 and 3 show the influence of the digestion stages in BRS Magna and BRS Carmem juices, which were the juices that presented the highest initial amounts of phenolic compounds in the matrix and bioaccessibility, respectively. Supplementary Table S2, S3 and S4 present the effects of the digestion simulation on the juices of the other evaluated cultivars.

For BRS Magna juice (Table 2), the main bioaccessible compounds present in all stages of digestion were (+)-catechin (180%), procyanidin B2 (174%) and gallic acid (124.6%). The bioaccessibilities of kaempferol-3-*O*-glucoside (62.6%), syringic acid (40.7%), (−)-epigallocatechin gallate (32.9%) and quercetin-3-β-D-glucoside (25.9%) also stand out. In BRS Carmem juice (Table 3), the main bioaccessible compounds were (+)-catechin (214%), procyanidin B2 (315%), rutin (431%), gallic acid (230.5%) and syringic acid (111.6%), present in all stages of digestion. (−)-Epigallocatechin gallate (67.6%), myricetin, quercetin-3-β-D-glucoside and *trans*-resveratrol, all with 35% availability, were also present in all stages of digestion. In BRS Magna and BRS Carmem juices, anthocyanins were only accessible until the digestion phase in the stomach, as was the case for the other cultivars (Table S2, S3 and S4).

No anthocyanin was detected after any of the digestive process simulations, and the behaviour of these compounds during *in vitro* gastrointestinal digestion can be observed in Tables 2 and 3 and supplementary Table S2, S3 and S4. According to Han et al. (2019), the bioaccessibility of anthocyanins can be considered low, as these compounds are hydrolysed in the oral cavity due to enzymatic and pH conditions, leading to the formation of their corresponding aglycone forms. In the stomach, they are relatively stable, due to the pH of the medium, with only a small portion being hydrolysed. In the intestine, alkaline conditions can also break these compounds down into smaller molecules. *Trans*-resveratrol was present in all stages of the digestive

Table 1
Bioaccessibility of phenolic compounds of grape juice from five new Brazilian cultivars.

Bioaccessibility (%)	Isabel Precoce grape juice	BRS Cora grape juice	BRS Violeta grape juice	BRS Magna grape juice	BRS Carmem grape juice
Flavanols					
(+)-Catechin	295.99 ± 3.31^{ab}	370.9 ± 40.2 ^{a}	169.1 ± 10.2 ^{c}	179.97 ± 7.75^{c}	214.25 ± 46.78^{bc}
(−)-Epicatechin gallate	7.75 ± 0.20^{c}	5.24 ± 0.89^{c}	5.21 ± 0.09^{c}	32.88 ± 1.58^{b}	67.59 ± 5.09^{a}
Procyanidin B2	113.38 ± 15.79^{bc}	101.42 ± 18.95^{c}	426.13 ± 133.90^{a}	173.91 ± 7.85^{bc}	314.82 ± 0.87^{ab}
Flavonols					
Quercetin-3-β-D-glucoside	12.00 ± 1.91^{c}	80.94 ± 14.73^{b}	283.11 ± 23.23^{a}	25.88 ± 2.31^{c}	34.97 ± 1.80^{c}
Rutin	35.75 ± 5.99^{b}	50.47 ± 7.36^{b}	57.56 ± 4.39^{b}	10.53 ± 2.27^{b}	430.73 ± 55.69^{a}
Kaempferol-3-*O*-glucoside	6.83 ± 2.22^{c}	26.74 ± 1.84^{b}	3.00 ± 0.16^{c}	62.65 ± 11.19^{a}	6.66 ± 0.32^{c}
Myricetin	–	–	0.67 ± 0.08^{b}	1.61 ± 0.04^{b}	35.21 ± 0.72^{a}
Stilbenes					
trans-resveratrol	–	85.99 ± 14.44	–	–	35.30 ± 2.21
Phenolic acids					
Gallic acid	79.49 ± 14.65^{bc}	61.59 ± 0.95^{c}	107.09 ± 23.36^{bc}	124.56 ± 3.24^{b}	230.51 ± 18.22^{a}
Syringic acid	20.67 ± 3.61^{b}	15.29 ± 1.62^{b}	16.80 ± 2.56^{b}	40.74 ± 9.57^{b}	111.57 ± 18.35^{a}
Caftaric acid	2.17 ± 0.74^{c}	3.46 ± 1.66^{bc}	5.18 ± 0.67^{b}	1.44 ± 0.08^{c}	10.12 ± 2.04^{a}

The results are expressed as mean ± standard deviation (n = 3). Means followed by the same letters, in lines, do not differ by the Tukey test at a 5% probability of error. Phenolic compounds quantified in the juice and absent from this table were not bioaccessible in the dialyzed fraction.

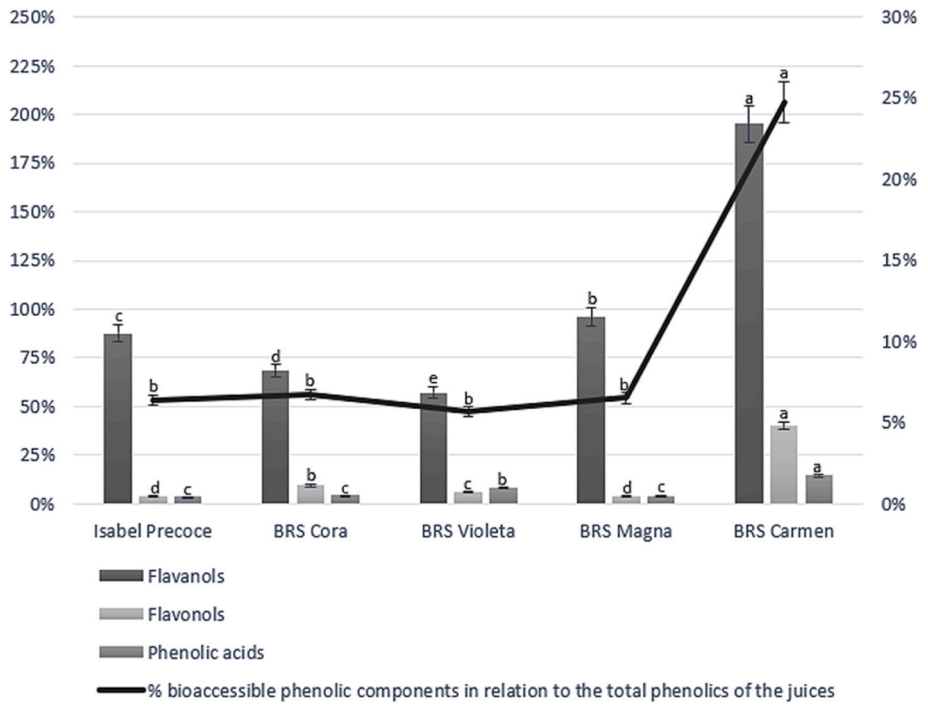

Fig. 1. Mean percentage values of bioaccessibility of phenolic compounds, grouped by family, of grape juices of new Brazilian cultivars. Average bars followed by the same letters do not differ from each other by the Tukey test at a 5% probability of error.

Table 2
Phenolic composition of BRS Magna grape juice, behavior in different fractions of *in vitro* gastrointestinal digestion and bioaccessibility.

Phenolic compounds(mg L^{-1})	Grape juice	Stomach	Nondialysable	Dialysable	Bioaccessibility (%)
Flavanols					
(+)-Catechin	2.36 ± 0.06^{b}	2.98 ± 0.08^{b}	4.80 ± 0.52^{a}	4.24 ± 0.06^{a}	179.97 ± 7.75
(−)-Epicatechin	0.42 ± 0.01	ND	0.80 ± 0.03	ND	–
(−)-Epicatechin gallate	5.48 ± 0.32^{a}	2.86 ± 0.14^{b}	2.50 ± 0.009^{b}	ND	–
(−)-Epigallocatechin gallate	2.14 ± 0.01^{b}	7.78 ± 0.08^{a}	1.12 ± 0.26^{c}	0.70 ± 0.03^{c}	32.88 ± 1.58
Procyanidin A2	3.18 ± 0.08	4.74 ± 0.21	ND	ND	–
Procyanidin B1	4.22 ± 0.24^{a}	3.36 ± 0.09^{b}	1.05 ± 0.01^{c}	ND	–
Procyanidin B2	16.72 ± 0.52^{c}	22.16 ± 0.56^{bc}	45.40 ± 3.75^{a}	29.12 ± 2.22^{b}	173.91 ± 7.85
Σ Flavanols	34.52 ± 1.24	43.88 ± 1.16	55.67 ± 4.57	34.06 ± 2.31	–
Flavonols					
Quercetin-3-β-D-glucoside	15.08 ± 1.66^{a}	5.88 ± 0.03^{b}	1.32 ± 0.22^{c}	3.21 ± 0.01^{bc}	25.88 ± 2.31
Rutin	7.78 ± 1.48^{b}	12.34 ± 0.04^{a}	0.38 ± 0.005^{c}	0.74 ± 0.006^{c}	10.53 ± 2.27
Kaempferol-3-*O*-glucoside	0.66 ± 0.11^{b}	5.94 ± 0.01^{a}	0.14 ± 0.009^{d}	0.40 ± 0.0006^{c}	62.65 ± 11.19
Myricetin	151.08 ± 13.71^{a}	102.96 ± 0.55^{b}	0.49 ± 0.01^{c}	2.43 ± 0.15^{c}	1.61 ± 0.04
Σ Flavonols	174.60 ± 16.96	127.12 ± 0.63	2.33 ± 0.24	6.78 ± 0.16	–
Anthocyanins					
Cyanidin-3,5-*O*-diglucoside	30.50 ± 1.51	18.52 ± 0.13	ND	ND	–
Delphinidin-3-*O*-glucoside	11.98 ± 1.09	2.40 ± 0.65	ND	ND	–
Malvidin-3,5-*O*-diglucoside	ND	84.66 ± 12.06	ND	ND	–
Cyanidin-3-*O*-glucoside	3.84 ± 0.39	ND	ND	ND	–
Peonidin-3-*O*-glucoside	ND	ND	ND	ND	–
Malvidin-3-*O*-glucoside	ND	ND	ND	ND	–
Petunidin-3-*O*-glucoside	128.74 ± 7.62	297.36 ± 2.18	ND	ND	–
Σ Anthocyanins	175.06 ± 10.61	402.94 ± 15.02	–	–	–
Stilbenes					
trans-resveratrol	2.40 ± 0.26	1.20 ± 0.006	ND	ND	–
Flavanones					
Naringenin	1.02 ± 0.01	ND	ND	ND	–
Phenolic acids					
Gallic acid	11.06 ± 0.50^{a}	11.48 ± 0.09^{a}	13.76 ± 0.2^{a}	15.12 ± 3.49^{a}	124.56 ± 3.24
Syringic acid	6.24 ± 0.65^{a}	7.70 ± 0.47^{a}	2.68 ± 0.55^{b}	2.48 ± 0.32^{b}	40.74 ± 9.57
Caftaric acid	625.40 ± 21.94^{a}	135.00 ± 2.87^{b}	15.56 ± 1.60^{c}	9.04 ± 0.19^{c}	1.44 ± 0.08
Chlorogenic acid	ND	0.74 ± 0.01	ND	ND	–
Caffeic acid	2.26 ± 0.53	ND	ND	ND	–
Σ Phenolic acids	644.96 ± 23.62	154.92 ± 3.44	32.00 ± 2.36	26.64 ± 4.00	–
Total Phenolics quantified	1032.56 ± 52.70	730.06 ± 20.25	90.00 ± 7.17	67.48 ± 6.47	–

The results are expressed as mean ± standard deviation (n = 3). Means followed by the same letters, in lines, do not differ by the Tukey test at a 5% probability of error. ND = Not detected.

Table 3
Phenolic composition of BRS Carmem grape juice, behaviour in different fractions of in gastrointestinal digestion and bioaccessibility.

Phenolic compounds (mg L^{-1})	Grape juice	Stomach	Nondialysable	Dialysable	Bioaccessibility (%)
Flavanols					
(+)-Catechin	2.66 ± 0.21^{c}	3.78 ± 0.31^{bc}	9.52 ± 1.37^{a}	5.60 ± 0.78^{b}	214.25 ± 46.78
(−)-Epicatechin	1.64 ± 0.09	ND	ND	ND	–
(−)-Epicatechin gallate	0.75 ± 0.04^{c}	2.16 ± 0.12^{b}	2.68 ± 0.15^{a}	ND	–
(−)-Epigallocatechin gallate	0.96 ± 0.06^{c}	6.42 ± 0.11^{a}	3.08 ± 0.33^{b}	0.64 ± 0.004^{d}	67.59 ± 5.09
Procyanidin A2	ND	2.52 ± 0.09	ND	ND	–
Procyanidin B1	5.30 ± 0.63^{a}	2.52 ± 0.06^{b}	0.98 ± 0.11^{c}	ND	–
Procyanidin B2	13.26 ± 0.01^{c}	2.46 ± 0.21^{d}	35.79 ± 2.93^{b}	41.68 ± 0.06^{a}	314.82 ± 0.87
Σ Flavanols	24.57 ± 1.04	19.86 ± 0.90	52.05 ± 4.89	47.92 ± 0.84	–
Flavonols					
Quercetin-3-β-D-glucoside	26.84 ± 1.56^{a}	3.78 ± 0.01^{c}	1.31 ± 0.02^{c}	9.36 ± 0.06^{b}	34.97 ± 1.80
Rutin	1.68 ± 0.06^{c}	3.09 ± 0.11^{b}	0.29 ± 0.02^{d}	7.20 ± 0.65^{a}	430.73 ± 55.69
kaempferol-3-*O*-glucoside	4.89 ± 0.27^{a}	3.28 ± 0.06^{b}	3.40 ± 0.16^{b}	0.32 ± 0.002^{c}	6.66 ± 0.32
Myricetin	69.20 ± 2.64^{b}	214.94 ± 6.74^{a}	28.80 ± 1.30^{c}	24.32 ± 0.26^{c}	35.21 ± 0.72
Σ Flavonols	102.61 ± 4.53	225.09 ± 6.92	33.80 ± 1.50	41.20 ± 0.97	–
Anthocyanins					
Cyanidin-3,5-*O*-diglucoside	ND	ND	ND	ND	–
Delphinidin-3-*O*-glucoside	2.44 ± 0.03	ND	ND	ND	–
Malvidin-3,5-*O*-diglucoside	21.24 ± 0.32^{b}	240.04 ± 9.73^{a}	8.48 ± 0.39^{bc}	ND	–
Cyanidin-3-*O*-glucoside	1.92 ± 0.03	ND	ND	ND	–
Peonidin-3-*O*-glucoside	3.02 ± 0.01	ND	ND	ND	–
Malvidin-3-*O*-glucoside	20.29 ± 0.01^{a}	9.32 ± 0.19^{b}	2.52 ± 0.09^{c}	ND	–
Petunidin-3-*O*-glucoside	ND	29.26 ± 1.06	ND	ND	–
Σ Anthocyanins	48.91 ± 0.40	278.62 ± 10.98	2.52 ± 0.09	–	–
Stilbenes					
trans-resveratrol	4.48 ± 0.26^{a}	0.47 ± 0.06^{c}	0.80 ± 0.003^{c}	1.57 ± 0.006^{b}	35.30 ± 2.21
Flavanones					
Naringenin	ND	0.98 ± 0.01	ND	ND	–
Phenolic acids					
Gallic acid	7.54 ± 0.11^{b}	9.92 ± 0.03^{b}	16.20 ± 0.16^{a}	17.36 ± 0.90^{a}	230.51 ± 18.22
Syringic acid	4.07 ± 0.09^{b}	9.50 ± 0.24^{a}	8.32 ± 1.17^{a}	4.56 ± 0.84^{b}	111.57 ± 18.35
Caftaric acid	442.46 ± 0.24^{a}	275.54 ± 8.01^{b}	46.72 ± 0.26^{c}	44.80 ± 9.01^{c}	10.12 ± 2.04
Caffeic acid	0.98 ± 0.27	ND	7.08 ± 0.03	ND	–
Σ Phenolic acids	455.05 ± 0.71	296.06 ± 8.29	93.36 ± 3.44	66.72 ± 10.75	–
Total Phenolics quantified	635.62 ± 6.94	821.08 ± 27.16	182.53 ± 9.92	157.41 ± 12.56	–

The results are expressed as mean ± standard deviation (n = 3). Means followed by the same letters, in lines, do not differ by the Tukey test at a 5% probability of error. ND = Not detected.

process for BRS Cora (Table S3) and BRS Carmem (Table 3) grape juice, with bioaccessibilities of 85.99% and 35.30%, respectively. However, the bioaccessibility of this phytochemical seemed to be influenced by the cultivar, since in the juices of Isabel Precoce grapes (Table S1), BRS Violeta (Table S4) and BRS Magna (Table 2), this compound was not detected in the dialyzed fraction. According to Tagliazucchi et al. (2010), *trans*-resveratrol is significantly degraded by digestion in simulated intestinal fluid.

According to Wojtunik-Kulesza et al. (2020), gallic acid has a high ability to scavenge free radicals. It is suggested that the high bioaccessibility of gallic acid identified in the samples is due to grape juice being a matrix rich in tannins, as the hydrolysis of gallotannin forms gallic acid (Ribéreau-Gayon, Glories, Maujean, & Dubourdieu, 2003).

In general, the evaluated juices showed high bioaccessibility of their phenolic components, which were identified in the dialyzed fraction 11 of the 24 phytochemicals initially quantified in the matrices. It is noteworthy that studies on the bioaccessibility of phenolic compounds present in grape juice are scarce, which may suggest possible

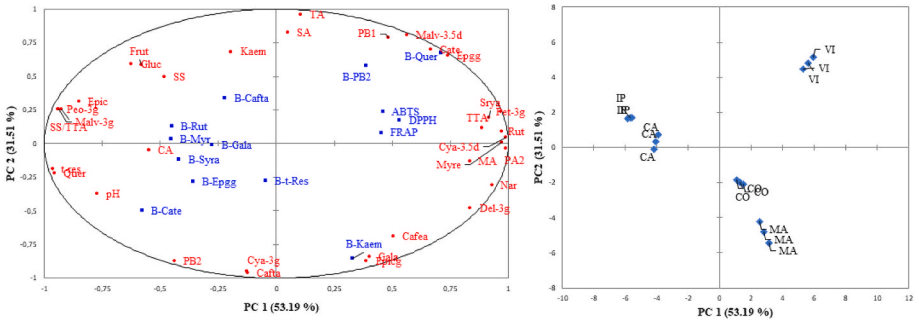

Fig. 2. Principal component analysis of grape juice of new Brazilian cultivars. Legends: IP = Isabel Precoce grape juice; CO = BRS Cora grape juice; VI = BRS Violeta grape juice; MA = BRS Magna grape juice; CA = BRS Carmem grape juice. Gala = gallic acid; Srya = syringic acid; Nar = naringenin; PB1 = procyanidin B1; Cate = (+)-catechin; PB2 = procyanidin B2; Epgg = (−)-epigallocatechin gallate; Epic = (−)-epicatechin; Epicg = (−)-epicatechin gallate; PA2 = procyanidin A2; Cafta = caftaric acid; Cafea = caffeic acid; t-res = trans resveratrol; Myre = myricetin; Quer = quercetin-3-β-D-glucoside; Rut = rutin; Kaem = kaempferol-3-*O*-glucoside; Cya-3.5d = cyanidin-3.5-*O*-diglucoside; Del-3g = delphinidin-3-*O*-glucoside; Malv-3.5d = malvidin-3.5-*O*-diglucoside; Cya-3g = cyanidin-3-*O*-glucoside; Peo-3g = peonidin-3-*O*-glucoside; Malv-3g = malvidin-3-*O*-glucoside; Pet-3g = petunidin-3-*O*-glucoside; B-Gala = bioaccessible gallic acid; B-Srya = bioaccessible syringic acid; B-Cate = bioaccessible (+)-catechin; B-PB2 = bioaccessible procyanidin B2; B-Epgg = bioaccessible (−)-epigallocatechin gallate; B-Cafta = bioaccessible caftaric acid; B-t-Res = bioaccessible trans resveratrol; B-Myr = bioaccessible myricetin; B-Quer = bioaccessible quercetin-3-β-D-glucoside; B-Rut = bioaccessible rutin; B-Kaem = bioaccessible kaempferol-3-*O*-glucoside; DPPH$^{\bullet}$ = antioxidant capacity by DPPH$^{\bullet}$; ABTS$^{\bullet+}$ = antioxidant capacity by ABTS$^{\bullet+}$; FRAP = antioxidant capacity by FRAP; TTA = titratable total acidity; °Brix = soluble solids in °Brix; °Brix/TTA = ratio °Brix/TTA; pH = Potential Hydrogen; CA = citric acid; TA = tartaric acid; MA = malic acid; SA = succinic acid; Glu = glucose; Frut = fructose.

compounds associated with *in vivo* benefits previously reported for this product (Leal et al., 2019).

3.3. Principal component analysis and hierarchical cluster analysis

Chemometric analyses using principal component analysis (PCA) and hierarchical cluster analysis (HCA) were performed to assess, in an integrated manner, similarities and differences between juices of the five cultivars with respect to quality parameters, sugar and organic acid contents, antioxidant capacity, phenolic profiles and the phenolic compounds that showed bioaccessibility. The PCA analysis can be seen in Fig. 2. Principal components 1 and 2, PC1 and PC2, respectively, explained 84.70% of the variability between samples.

The PC1 >0 grouped the juices of the BRS Magna and BRS Cora cultivars, associating them with the presence of higher concentrations of the phytochemicals delphinidin-3-*O*-glucoside, gallic acid, caffeic acid and (+)-epicatechin gallate. Regarding the 11 bioaccessible polyphenols, the two juices stood out for the percentage of bioaccessible kaempferol-3-*O*-glucoside, and the BRS Cora juice also had a higher percentage of bioaccessible *trans*-resveratrol. BRS Violeta juice contained higher levels of organic acids (tartaric and succinic acid) and the phenolic compounds malvidin-3,5-*O*-diglucoside, (+)-catechin, (−)-epigallocatechin gallate and procyanidin B1 and had a higher bioaccessibility of procyanidin B2 and quercetin-3-β-D-glucoside. Additionally, the BRS Violeta juice (PC2 >0) was closer to the BRS Cora juice than to the other juices, because both possessed outstanding AOX according to the ABTS•+, FRAP and DPPH• tests. In turn, the juices of the Isabel Precoce and BRS Carmem cultivars were in the opposite position with regard to PC1 >0, which may be related to the higher ratio value and content of the phenolic compounds malvidin-3-*O*-glucoside, peonidin-3-*O*-glucoside, (+)-epicatechin, quercetin-3-β-D-glucoside and *trans*-resveratrol. The cultivar BRS Carmem also featured the largest bioaccessible fraction of most phenolic compounds identified as bioaccessible, including rutin, gallic acid, syringic acid, (−)-epicatechin gallate, myricetin and caftaric acid. The highest amount of bioaccessible compounds, associated with the BRS Carmem cultivar, showed a positive correlation with the highest levels of fructose, glucose, Brix degree, lower acidity and organic acids (PC1 <0).

Thus, the results show that each grape cultivar evaluated is a complex and unique matrix, corroborating with other works (Lima et al., 2014; Padilha et al., 2017). The release and absorption of phenolic compounds could had be influenced by different chemical composition of these matrixes, suggested a direct relationship between the bioaccessibility of phytochemicals compounds with a higher content of sugars and lower of organic acids. So, possibly, a more advanced stage of grape maturation at harvest can be associated with a greater bioaccessibility of phenolics of the juice. The results found in the research also corroborated with the findings of Seraglio et al. (2018) that studied the bioaccessibility of the "juçara" fruit.

In addition to PCA, hierarchical cluster analysis (HCA) was performed, and the results obtained are shown in Fig. 3. The grape juices of the five grape cultivars were grouped into two groups, with high similarities between the samples. Cluster 1 consisted of the juice of cultivar BRS Carmem and Isabel Precoce. The juices of these cultivars differed in terms of the total percentage of phenolic compounds that were bioaccessible and were similar to the amount of sugars (Fig. 1). Cluster 2 consisted of BRS Violeta juices and a subgroup with BRS Cora and BRS Magna cultivar juices; these juices were the ones with the highest amounts of phenolic compounds (Supplementary Table S1). In short, the clustering analysis strengthens the results presented in the PCA regarding the clustering of samples and the correlation of matrix composition with bioaccessibility.

4. Conclusions

Grape juices made with Brazilian hybrid grapes showed a high content of bioaccessible phenolic compounds. Of the 24 phenolic compounds initially identified and quantified in the juices, 11 were bioaccessibles. BRS Carmem grape juice stood out, with the highest content of bioaccessible phenolics, demonstrating that factors such as the cultivar should be better explored in studies with functional foods. The most bioaccessible phenolic compounds were (+)-catechin, procyanidin B2, and gallic acid, all with accessibility greater than 100%, highlighting the class of flavanols. This research also suggests that the bioaccessibility of phenolic compounds is related with the contents of sugars and organic acids of the matrix. Evidencing that factors such as the choice of the cultivar and degree of ripeness of the grape must be considered to the juice processing industry. Finally, Brazilian grape juice produced from all cultivars evaluated, can be characterized as a functional beverage with appreciable content of bioaccessible phytochemicals, especially flavanols.

Declaration of competing interest

None.

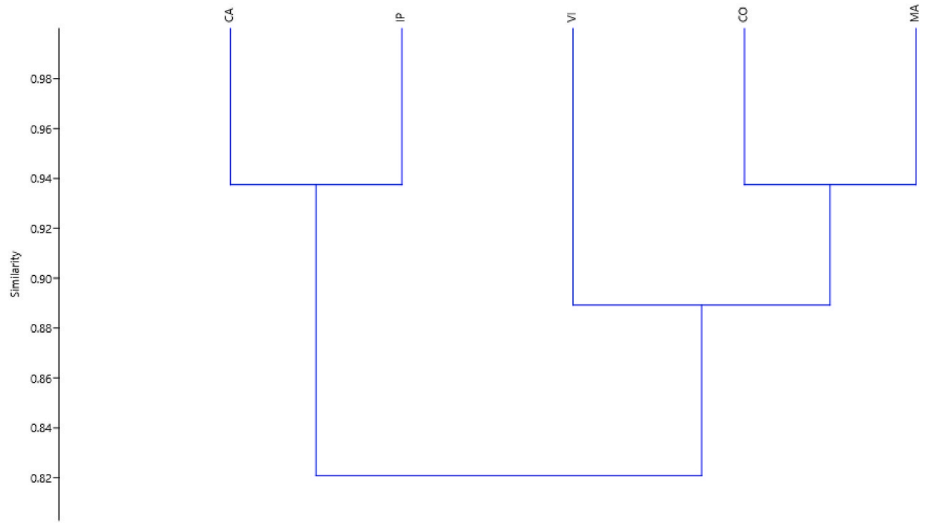

Fig. 3. Dendrogram showing clustering of grape juices based on bioaccessible phenolic compounds, chemical profile and antioxidant capacity. Legends: IP = Isabel Precoce grape juice; CO = BRS Cora grape juice; VI = BRS Violeta grape juice; MA = BRS Magna grape juice; CA = BRS Carmem grape juice.

Data availability

Data will be made available on request.

Acknowledgments

The authors would like to acknowledge Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for granting a scholarship. They also thank the companies Queiroz Galvão Alimentos S/A, EBFT/ASA Ind. Com. Ltda and Grand Valle for the donation of grapes analysed in this study. The AEB Group for the donation of enzymes and to the Federal Institute of Sertão Pernambucano, Embrapa Semiarid and the Federal University of Bahia for use of the laboratory and the donation of reagents.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fbio.2023.102501.

References

Alqurashi, R. M., Alarifi, S. N., Walton, G. E., Costabile, A. F., Rowland, I. R., & Commane, D. M. (2017). In vitro approaches to assess the effects of açai (Euterpe oleracea) digestion on polyphenol availability and the subsequent impact on the faecal microbiota. *Food Chemistry, 234*, 190–198. https://doi.org/10.1016/j.foodchem.2017.04.164

Augusti, P. R., Conterato, G. M. M., Denardin, C. C., Prazeres, I. D., Serra, A. T., Bronze, M. R., & Emanuelli, T. (2021). Bioactivity, bioavailability, and gut microbiota transformations of dietary phenolic compounds: Implications for COVID-19. *Journal of Nutritional Biochemistry, 97*, Article 108787. https://doi.org/10.1016/j.jnutbio.2021.108787

Barreto, S. M. A., da Silva, A. B. M., Dutra, M. C. P., Bastos, D. C., Carvalho, A. J. B. A., Viana, A. C., Narain, N., & Lima, M. S. (2023). Effect of commercial yeasts (Saccharomyces cerevisiae) on fermentation metabolites, phenolic compounds, and bioaccessibility of Brazilian fermented oranges. *Food Chemistry, 408*, 132–151. https://doi.org/10.1016/j.foodchem.2022.135121

Brasil. (2018). Instrução Normativa nº 14, de 08 de fevereiro de 2018. Estabelece a complementação dos padrões de identidade e qualidade do vinho e derivados da uva e do vinho. *Diário Oficial da República Federativa do Brasil*, 4–6. Ed. 47, Seção 1.

Carneiro, T. S., Dutra, M. C. P., Andrade, D. L., Araújo, A. J. B., Constante, P. L. C., & Lima, M. S. L. (2022). Phenolic compounds in peel, seed and cold pressed pink pepper (Schinus terebinthifolia R.) oil and bioaccessibility of peel using a digestion model with intestinal barrier simulation. *Food Bioscience, 49*, Article 101930. https://doi.org/10.1016/j.fbio.2022.101930

Celep, E., Charehsaz, M., Akyüz, S., Acar, E. T., & Yesilada, E. (2015). Effect of in vitro gastrointestinal digestion on the bioavailability of phenolic components and the antioxidant potentials of some Turkish fruit wines. *Food Research International, 78*, 209–215. https://doi.org/10.1016/j.foodres.2015.10.009

Coelho, E. M., da Silva Padilha, C. V., Miskinis, G. A., de Sá, A. G. B., Pereira, G. E., de Azevêdo, L. C., & dos Santos Lima, M. (2018). Simultaneous analysis of sugars and organic acids in wine and grape juices by HPLC: Method validation and characterization of products from northeast Brazil. *Journal of Food Composition and Analysis, 66*, 160–167. https://doi.org/10.1016/j.jfca.2017.12.017

De Lima Tavares Toscano, L., Silva, A. S., De França, A. C. L., De Sousa, B. R. V., De Almeida Filho, E. J. B., Da Silveira Costa, M., Marques, A. T. B., Da silva, D. F., De Farias Sena, K., Cerqueira, G. S., & Da Conceição Rodrigues Gonçalves, M. (2019). A single dose of purple grape juice improves physical performance and antioxidant activity in runners: A randomized, crossover, double-blind, placebo study. *European Journal of Nutrition, 59*, 2997–3007. https://doi.org/10.1007/s00394-019-02139-6

Dutra, M. C. P., Rodrigues, L. L., Viana, A. C., De Oliveira, D., Pereira, G. E., & Dos Santos Lima, M. (2018). Integrated analyses of phenolic compounds and minerals of Brazilian organic and conventional grape juices and wines: Validation of a method for determination of Cu, Fe and Mn. *Food Chemistry, 269*, 157–165. https://doi.org/10.1016/j.foodchem.2018.07.014

Dutra, M. C. P., Viana, A. C., Pereira, G. E., Resende Nassur, R. C. M., & Lima, M. S. (2021). Whole, concentrated and reconstituted grape juice: Impact of processes on phenolic composition, "foxy" aromas, organic acids, sugars and antioxidant capacity. *Food Chemistry, 343*, 128–399. https://doi.org/10.1016/j.foodchem.2020.128399

Dwibedi, V., Jain, S., Singhal, D., Mittal, A., Rath, S. K., & Saxena, S. (2022). Inhibitory activities of grape bioactive compounds against enzymes linked with human diseases. Review. *Microbial Biotechnology, 106*(4), 1399–1417. https://doi10.1007/s00253-022-11801-9.

Hammer, Ø., Harper, D. A. T., & Ryan, P. D. (2001). Past: Paleontological statistics software package for education and data analysis. *Palaeontologia Electronica, 4*(1). art. 4: 9pp http://palaeo-electronica.org/2001_1/past/issue1_01.htm.

Han, F., Yang, P., Wang, H., Fernandes, I., Mateus, N., & Liu, Y. (2019). Digestion and absorption of red grape and wine anthocyanins through the gastrointestinal tract. *Trends in Food Science & Technology, 83*(November 2018), 211–224. https://doi.org/10.1016/j.tifs.2018.11.025

Leal, J. B., Portela Leal, J. B., Borges, Y. L., Moura, M. I. L., Teixeira-Araujo, A. A., Serafim da Costa, V., & Carvalho, F. O. (2019). Grape juice and aerobic exercise on blood pressure. *Nutrition & Food Science*. https://doi.org/10.1108/NFS-08-2019-0256

Lima, M. S., Ferreira, E. T. J., Olinda de Souza, M. E. A., Elias Pereira, G., & Toaldo Fedrigo, I. M. (2022). Artifcial neural network: A powerful tool in associating phenolic compounds with antioxidant activity of grape juices. *Food Analytical Methods, 15*, 527–540. https://doi.org/10.1007/s12161-021-02144-8

Lima, M. D. S., Silani, I. D. S. V., Toaldo, I. M., Corrêa, L. C., Biasoto, A. C. T., Pereira, G. E., Bordignon-Luiz, M. T., & Ninow, J. L. (2014). Phenolic compounds, organic acids and antioxidant activity of grape juices produced from new Brazilian varieties planted in the Northeast Region of Brazil. *Food Chemistry, 161*, 94–103. https://doi.org/10.1016/j.foodchem.2014.03.109

Lingua, M. S., Theumer, M. G., Kruzynski, P., Wunderlin, D. A., & Baroni, M. V. (2019). Bioaccessibility of polyphenols and antioxidant properties of the white grape by simulated digestion and Caco-2 cell assays: Comparative study with its winemaking product. *Food Research International, 122*(May), 496–505. https://doi.org/10.1016/j.foodres.2019.05.022

Lingua, M. S., Wunderlin, D. A., & Baroni, M. V. (2018). Effect of simulated digestion on the phenolic components of red grapes and their corresponding wines. *Journal of Functional Foods, 44*(February), 86–94. https://doi.org/10.1016/j.jff.2018.02.034

Lorenzo, J. M., Estévez, M., Barba, F., Thirumdas, R., Franco, D., & Sichetti Munekata, P. E. (2019). Polyphenols: Bioaccessibility and bioavailability of bioactive components. *Innovative Thermal and Non-Thermal Processing, 11*, 309–332. https://doi.org/10.1016/B978-0-12-814174-8.00011-1

Lucas-Gonzalez, R., Navarro-Coves, S., Pérez-Álvarez, J. A., Fernández-López, J., Muñoz, L. A., & Viuda-Martos, M. (2016). Assessment of polyphenolic profile stability and changes in the antioxidant potential of maqui berry (Aristotelia chilensis (Molina) Stuntz) during in vitro gastrointestinal digestion. *Industrial Crops and Products, 94*, 774–782. https://doi.org/10.1016/j.indcrop.2016.09.057

Minekus, M., Alminger, M., Alvito, P., Ballance, S., Bohn, T., Bourlieu, C., Carrière, F., Boutrou, R., Corredig, M., Dupont, D., Dufour, C., Egger, L., Golding, M., Karakaya, S., Kirkhus, B., Le Feunteun, S., Lesmes, U., MacIerzanka, A., MacKie, A., … Brodkorb, A. (2014). A standardised static in vitro digestion method suitable for food-an international consensus. *Food & Function, 5*(6), 1113–1124. https://doi.org/10.1039/c3fo60702j

Mohamedshah, Z., Chadwick-Corbin, S., Wightmanc, J. D., & Ferruzz, M. G. (2020). Comparative assessment of phenolic bioaccessibility from 100% grape juice and whole grapes. *Food & Function, 11*, 6433–6445. https://doi.org/10.1039/d0fo00792g

Naeem, A., Ming, Y., Pengyi, H., Jie, K. Y., Yali, L., Haiyan, Z., Shuai, X., Wenjing, L., Ling, W., Xia, Z. M., Shan, L. S., & Qin, Z. (2022). The fate of flavonoids after oral administration: Acomprehensive overview of its bioavailability. Critical reviews in. *Food Sciences and Nutrition, 62*, 2022. https://doi.org/10.1080/10408398.2021.1898333

OIV – International Organisation of Vine and Wine. (2022). *Compendium of international methods of wine and must analysis, edition 2022. 19th General Assembly, 21 July 2021* (Paris).

Padilha, C. V. da S., Miskinis, G. A., de Souza, M. E. A. O., Pereira, G. E., de Oliveira, D., Bordignon-Luiz, M. T., Lima, M., & dos, S. (2017). Rapid determination of flavonoids and phenolic acids in grape juices and wines by RP-HPLC/DAD: Method validation and characterization of commercial products of the new Brazilian varieties of grape. *Food Chemistry, 228*, 106–115. https://doi.org/10.1016/j.foodchem.2017.01.137

Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., & Rice-Evans, C. (1999). *Antioxidant activity applying an improved ABTS radical cationdecolorization assay* (Vol. 26, pp. 1231–1237). New York: Free Radical Biologyand Medicine.

Ribas-Agustí, A., Martín-Belloso, O., Soliva-Fortuny, R., & Elez-Martínez, P. (2018). Food processing strategies to enhance phenolic compounds bioaccessibility and bioavailability in plant-based foods. *Critical Reviews in Food Science and Nutrition, 58*(15), 2531–2548. https://doi.org/10.1080/10408398.2017.1331200

Ribéreau-Gayon, P., Glories, Y., Maujean, A., & Dubourdieu, D. (2003). *Tratado de Enología: Química del vino estabilización y tratamentos. 1ªed* (Vol. 2, p. 537). Buenos Aires: Hemisferio Sur, 26X18.

Rufino, M. D. S. M., Alves, R. E., de Brito, E. S., de Morais, S. M., Sampaio, C. D. G., Pérez-Jiménez, J., & Saura-Colixto, F. D. (2006). Metodologia científica: Determinação da atividade antioxidante total em frutas pelo método de redução do ferro (FRAP). *Comunicado Técnico Embrapa, 125*, 1–4.

Seraglio, S. K. T., Gonzaga, L. V., Vitali, L., Micke, G. A., Oliveira Costa, A. C., Fett, R., & Campelo Borges, C. S. (2018). Effects of gastrointestinal digestion models in vitro on phenolic compounds and antioxidant activity of juçara (*Euterpe edulis*). *International Journal of Food Science and Technology, 53*, 1824–1831. https://doi:10.1111/ijfs.13816.

Silva Haas, I. C., Marmitt, D. J., Toaldo Fedrigo, I. M., Goettert, M. I., & Bordignon-Luiz, M. T. (2020). Evaluation of antiproliferative and anti-inflammatory effects of non-pomace sediment of red grape juices (Vitis labrusca L.) in healthy and cancer cells after in vitro gastrointestinal simulation. *PharmaNutrition, 13*, 100–204. https://doi.org/10.1016/j.phanu.2020.100204

Silva, G. G., Dutra, M., da, C. P., de Oliveira, J. B., Rybka, A. C. P., Pereira, G. E., & dos Santos Lima, M. (2019). Processing methods with heat increases bioactive phenolic compounds and antioxidant activity in grape juices. *Journal of Food Biochemistry, 43*(3), 1–10. https://doi.org/10.1111/jfbc.12732

Sun, X., Cheng, X., Zhang, J., Ju, Y., Que, Z., Liao, X., Lao, F., Fang, Y., & Ma, T. (2020). Letting wine polyphenols functional: Estimation of wine polyphenols bioaccessibility

under different drinking amount and drinking patterns. *Food Research International, 127*. https://doi.org/10.1016/j.foodres.2019.108704/, 1087-04.

Tagliazucchi, D., Verzelloni, E., Bertolini, D., & Conte, A. (2010). In vitro bio-accessibility and antioxidant activity of grape polyphenols. *Food Chemistry, 120*(2), 599–606. https://doi.org/10.1016/j.foodchem.2009.10.030

Toaldo, I. M., Cruz, F. A., Silva, E. L., & Bordignon-Luiz, M. T. (2016). Acute consumption of organic and conventional tropical grape juices (V. labrusca L.) increases antioxidants in plasma and erythrocytes, but not glucose and uric acid levels, in healthy individuals. *Nutrition Research, 36*, 808–817. https://doi.org/10.1016/j.nutres.2016.04.010

Wojtunik-Kulesza, K., Oniszczuk, A., Oniszczuk, T., Combrzyński, M., Nowakowska, D., & Matwijczuk, A. (2020). Digestion on composition , bioaccessibility and antioxidant activity of food polyphenols — a non-systematic review. *Nutrients, 12*, 1401. https://doi.org/10.3390/nu12051401

Xia, E. Q., Deng, G. F., Guo, J. Y., & Li, H. B. (2010). Biological activities of polyphenols from grapes. *International Journal of Molecular Sciences, 11*, 622–646. https://doi.org/10.3390/ijms11020622

Yun Yang, C., Chen, Y. H., Liu, P. J., Hu, W. J., Lu, K. C., & Tsai, K. W. (2022). The emerging role of miRNAs in the pathogenesis of COVID-19: Protective effects of nutraceutical polyphenolic compounds against SARS-CoV-2 infection. Review. *International Journal of Medical Sciences, 19*(8), 1340–1356. https://doi: 10.7150/ijms.76168.

Zhou, D. D., Li, J., Xiong, R. G., Saimaiti, A., Huang, S. Y., Wu, S. X., Yang, Z. J., Shang, A., Zhao, C. N., Gan, R. Y., & Li, H. B. (2022). Bioactive compounds, health benefits and food applications of grape, 2022 *Foods, 11*, 2755. https://doi.org/10.3390/foods11182755.