Research Article

Bioactive Compounds, Antioxidant Capacity, and Fatty Acids in Different Parts of Four Unexplored Fruits

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Total phenolic content (TPC), total flavonoids (TF), total anthocyanins (TA), and antioxidant capacity of different parts of four unexplored fruits from Brazil (*Syzygium cumini* Lam, *Solanum nigrum* Linn, *Inga edulis* Mart, and *Hovenia dulcis* Thunb) were determined; the bioactive compounds and fatty acids were quantified and identified by high-performance liquid chromatography and gas chromatography, respectively. *S. cumini* peels contained the most TA (63.31 mg/100 g), whilst *H. dulcis* pulp and peels had the highest TPC (518.18 mg GAE/100 g) and TF (76.54 mg EQ/g). Phenolic compounds responsible for antioxidant capacity of fruits were gallic acid, ellagic acid, kaempferol, and epicatechin. *H. dulcis* seed showed the highest level of the essential fatty acid omega-3 (3985.95 mg/100 g). PCA showed that PC1 and PC2 explained 90.43% of the total variability of the antioxidant data. Most of the seeds showed omega-3, omega-6, and omega-9 fatty acids at significant concentrations, with two PCs explaining 93.80% of the total variance of the fatty acid contents.

1. Introduction

Oxidative stress is caused by reactive oxygen species (ROS), which include free radicals (oxygen and nitrogen species), and this condition occurs when there is an overproduction of ROS and/or insufficient antioxidant mechanisms [1]. Among the main consequences of oxidative stress is damage to the biomolecules, particularly DNA, lipids, and proteins, which can severely compromise cell function and activate a cascade of free radical reactions. Oxidative stress may occur as a result of degenerative diseases or may also be a causative factor in the development of diseases. Arthritis, atherosclerosis, cancer, diabetes, and Parkinson's, for example, involve ROS [2].

The interest in studying unexplored species of fruits and their parts, such as peels and seeds, has increased in recent years because they are rich in phenolic compounds, flavonoids, anthocyanins, vitamins, minerals, and other bioactive compounds that are capable of neutralizing free radicals [3–5].

Moreover, fruits have essential polyunsaturated fatty acids (PUFAs), such as linoleic acid (LA, 18:2n-6) and alpha-linolenic acid (LNA, 18:3n-3) that are considered essential because they cannot be synthesized by the human body and must be supplied through the diet [6]. These fatty acids (FAs) are able to protect against oxidative stress by exerting an antioxidant role [7]. Fruits also contain oleic acid (OA, 18:1n-9), which is widely recognized for its action in lowering the risk of ROS-induced diseases and enhancing the immune system [8]. Therefore, increased fruit consumption is associated not only with personal preference but also with the maintenance of health, through the consumption of foods rich in vital micronutrients [7, 9].

Several methods have been developed to measure the antioxidant capacity of fruits. In particular, the ferric reducing antioxidant power (FRAP) and 1,1-diphenyl-2-picrylhydrazyl (DPPH[•]) assays have been widely adopted [7]. Association studies between fruit and vegetable consumption and

TABLE 1: List of the four fruits included in this study.

Common name	Scientific name	Family	Origin
Jambolan	<i>Syzygium cumini</i> Lam	Myrtaceae	India
Black nightshade	<i>Solanum nigrum</i> Linn	Solanaceae	South Africa
Inga edulis	<i>Inga edulis</i> Mart	Leguminosae	Brazil, South and Central America
Japanese grape	<i>Hovenia dulcis</i> Thunb	Rhamnaceae	Japan, China, and Korea

indicators of oxidative stress using antioxidant assays have received the attention of researchers [10]. It is recommended that at least two methods with different mechanisms are combined to provide a reliable picture of the total antioxidant capacity of a food because the results can be affected using a single assay to evaluate the entire sample [11, 12].

Jambolan (*Syzygium cumini* Lam), black nightshade (*Solanum nigrum* Linn), inga edulis (*Inga edulis* Mart), and Japanese grape (*Hovenia dulcis* Thunb) are unexplored fruits and few studies have been published on characterizing the bioactive compounds and antioxidant capacity of the pulps, peels, and seeds of these fruits. Therefore, this study aimed to evaluate the DPPH[•] and FRAP antioxidant capacities, the total phenolic content (TPC), total flavonoids (TF), and total anthocyanins (TA) and to identify and quantify the phenolic compounds and FAs by high-performance liquid chromatography (HPLC) and gas chromatography (GC), respectively.

2. Materials and Methods

2.1. Plant Material. The botanical identification and geographical origin of the four fruits are provided in Table 1. Fresh fruits (ca. 2 kg each) were harvested at Monte Alegre, São Paulo, Brazil (23°35'31"S and 48°38'38"W). The fruits were washed with tap water and the pulps, peels, and seeds were removed manually, chopped, and then homogenized in a multiprocessor to uniformity before analysis. *H. dulcis* pulp and peels were evaluated together because it was not possible to separate them. *I. edulis* peels were discarded.

2.2. Chemical and Standards. Fatty acid methyl esters (FAME) (standard mixture 189-19), methyl tricosanoate (23:0), Folin-Ciocalteu phenol reagent, gallic acid, kaempferol, ellagic acid, epicatechin, quercetin, DPPH[•], 6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (Trolox), 2,4,6-tris(2-pyridyl)-s-triazine (TPTZ), and methanol and acetonitrile (HPLC grade) were purchased from Sigma Chemical Co. (St. Louis, USA). Ultrapure water (Milli-Q system, Millipore Corp, Bedford, MA) was also used. All solvents and chemicals were of analytical grade.

2.3. Extraction of Antioxidants. The extracts were prepared using 10 g (per part of the fruit) in 100 mL of methanol under magnetic stirring for 4 h according to the method developed by Santos et al. [13]. After filtration, the extracts were concentrated under reduced pressure at 40°C. The absorbance values obtained were within the calibration range of the respective methods (Genesys 10-UV scanning, Thermo Scientific). The results were expressed as fresh weight (FW).

2.4. Analysis of DPPH[•] Antioxidant Capacity (Free Radical Scavenging Activity). The DPPH[•] scavenging capacity was measured using the method described by Brand-Williams et al. [14]. Briefly, an aliquot of the methanolic fruit extract (25 μ L) was added to 2 mL of 6.25×10^{-5} mol/L DPPH[•] methanolic solution. After gently mixing and then resting the reaction solutions at room temperature for 30 min, the absorbance was measured at 517 nm. Methanolic solutions of known Trolox concentrations between 0 and 2000 μ mol/L were used for calibration. The results were expressed as μ mol Trolox equivalents (TE)/g FW using the calibration curve ($y = 0.686 - 2.90 \times 10^{-4}x$, $r^2 = 0.997$).

2.5. FRAP Assay. The FRAP assay was determined as previously described by Benzie and Strain [15]. FRAP reagent was prepared by mixing acetate buffer (0.3 mol/L, pH 3.6), TPTZ (10 mmol/L), and FeCl₃ (20 mmol/L) solutions at 10 : 1 : 1, respectively. An aliquot of the methanolic fruit extract (100 μ L) and 300 μ L of distilled water were added to 3 mL of the FRAP reagent and then placed in the dark for 30 min at 37°C. The absorbance was measured against a blank at 593 nm. Aqueous solutions of known Fe²⁺ concentrations between 0 and 1500 μ mol/L (FeSO₄·7H₂O) were used for the calibration curve ($y = 0.006 + 6.55 \cdot 10^{-4}x$, $r^2 = 0.999$) and the results were expressed as μ mol Fe²⁺/g FW.

2.6. TPC. The TPC were determined according to the procedure described by Shahidi and Naczk [16]. An aliquot of the methanolic fruit extract (250 μ L) was mixed with 250 μ L of the Folin-Ciocalteu reagent (prediluted in distilled water at 1 : 1 v/v), 500 μ L of a saturated sodium carbonate solution, and 4 mL of distilled water. The reaction mixture was left to rest for 25 min and then centrifuged at 3000 rpm (1638 g) for 10 min. Then, the absorbance was measured at 725 nm. Methanolic solutions of known gallic acid concentrations between 0 and 250 mg/L were used for calibration. The results were expressed as mg gallic acid equivalents (GAE) 100/g FW using the calibration curve ($y = -0.0273 + 0.00517x$, $r^2 = 0.999$).

2.7. TF. TF were determined according to Woisky and Salatino [17]. An aliquot of the methanolic fruit extract (500 μ L, 2.5 mg/ml) was added to 250 μ L of AlCl₃ (5 g/100 mL methanol) and 4.25 mL of methanol. After 30 min, the absorbance was measured at 425 nm. The results were expressed as mg quercetin equivalents (mg QE/g FW) using the calibration curve ($y = -0.0273 + 0.00517x$, $r^2 = 0.999$).

2.8. TA. TA were extracted according to Lees and Francis [18]. An aliquot (50 g) was then homogenized with 50 mL of

solvent (70% ethanol acidified to pH 2.0 with 0.1% HCl) for 2 min. The volume was adjusted to 200 mL in a volumetric flask using the solvent, covered with parafilm, and stored at 4°C for 12 h. Then, the material was filtered using a Büchner funnel and 125 mL of the filtrate mixed with another 250 mL of the solvent. Triplicate aliquots (2 mL) were adjusted to a final volume of 100 mL in a volumetric flask using the solvent. The reaction mixture was placed in the dark for 2 h. Then, the absorbance was measured at 535 nm. The results were expressed as mg 100/g FW.

2.9. Chromatographic Analysis. Total lipids (TL) were extracted by Bligh and Dyer [19] method. FAME were prepared by methylation of the TL as described by Hartman and Lago [20] and the methyl esters separated by GC in a Trace Ultra model 3300 (Thermo Scientific) equipped with a flame ionization detector (FID) and a cyanopropyl capillary column (100 m × 0.25 i.d., 0.25 µm film thickness, CP7420). The gas flow rates were 1.2 mL/min carrier gas (H₂), 30 mL/min make-up gas (N₂), and 35 and 300 mL/min flame gasses (H₂ and synthetic air, respectively). The split ratio was 1:80 and 2 µL of the samples was injected in triplicate. The detector and injection port were set at 240°C and the column was set at 185°C for 7.5 min. The peak areas were determined by ChromQuest 5.0 software. The FAs were identified by comparing their retention times with those of standard methyl esters.

Quantification (as mg FA/g of TL) was determined against tricosanoic acid methyl ester (23:0) as an internal standard (IS), as described by Joseph and Ackman [21]. Theoretical FID correction factor [22] values were used to obtain the concentrations. The FA contents were calculated as mg/g TL using

$$\text{FA (mg/g TL)} = \frac{A_X \cdot W_{IS} \cdot CF_X}{A_{IS} \cdot W_X \cdot CF_{AE}} \times 100, \quad (1)$$

where FA is expressed as mg/g of TL, A_X is the peak area (FA), A_{IS} is the peak area of the IS (the methyl ester of tricosanoic acid, 23:0), W_{IS} is the IS mass (mg) of IS added to the sample (mg), W_X is the sample mass (mg), CF_X is the theoretical correction factor, and CF_{AE} is the conversion factor necessary to express results as mg of FA rather than as methyl esters.

2.10. Quantification of Phenolic Compounds by HPLC. The phenolic compounds were quantified according to Fratianni et al. [23] using a Thermo Scientific chromatograph, model Finnigan Surveyor PDA, with a manual injection valve, a photodiode array detection (DAD), connected to ChromQuest 5.0 software and equipped with a EC 250/4.6 Nucleodur 100-5 C18ec column (Macherey-Nagel) (250 mm × 4.6 mm). The mobile phases consisted of Milli-Q water (containing 1% acetic acid, phase A) and acetonitrile (containing 1% acetic acid, phase B). The gradient started at 90% A and decreased to 60% A in 25 min. After 26 min, phase A returned to 90%, where it remained until 35 min (total run time). The flow rate was 0.7 mL/min, the injection volume was 50 µL, and the detection wavelengths were set at 250 and 270 nm. The phenolic compounds of the methanolic fruit extracts

were identified by comparison of their retention times with standards (Sigma) and UV-visible light spectra. The bioactive compounds were quantified by comparing the sample with the standard calibration curves.

2.11. Statistical Analysis. The data were analyzed using analysis of variance (ANOVA) and the means were compared by Tukey's test. The results were analyzed by principal component analysis (PCA) using Statistica 7.0 software. Data before treatment was not necessary.

3. Results and Discussion

3.1. TPC, TF, TA, and Antioxidant Capacity. The TPC, TF, and TA amounts and the antioxidant capacity (DPPH• and FRAP) of the different parts of the fruits are shown in Table 2. The TPC ranged from 19.27 mg GAE/100 g in *S. nigrum* seeds to 518.18 mg GAE/100 g in *H. dulcis* pulp and peels. The TPC in *H. dulcis* seed (85.56 mg GAE/100 g) was similar to that found by Santos et al. [13] in Benitaka and Brazil grape seed varieties (89.83 and 91.53 mg GAE/100 g, respectively). The TPC of *S. nigrum* pulps was 412.71 mg GAE/100 g. This value can be compared to those of jaboticaba (440 mg GAE/100 g), açai (454 mg GAE/100 g), and gurguri (549 mg GAE/100 g), in the study by Rufino et al. [9] because of the scarcity of studies on this particular fruit.

TF among the parts of the fruits varied from 2.59 mg EQ/g in *I. edulis* seeds to 76.54 mg EQ/g in *H. dulcis* pulps and peels. According to Volp et al. [24], flavonoids are polyphenolic structures found naturally in plants and many of these flavonoids are responsible for the color of fruits. In the current study, *H. dulcis* pulp and peels revealed a higher TF value compared to the other pulps evaluated, which could be associated with the heavy pigmentation of these fruits.

Faria et al. [25] detected 91.2 mg EQ/g TF in *S. cumini*. These values were higher than those found in this study (59 mg and 65.51 mg EQ/g in pulps and peels, respectively). In contrast, Benherlal and Arumughan [26] studied the same fruit and only found 7 mg EQ/g. These variations may occur due to the influence of various factors, such as climate conditions, geographic location, fruit maturation stage, and method used [25, 27].

The highest TA concentration was in *S. cumini* peels (63.31 mg/100 g). This value was lower than that found by Rufino et al. [9], when evaluating the combined peel and pulp of the fruit (93.3 mg/100 g) but higher when compared with jaboticaba (58.1 mg/100 g) and camu-camu (42.2 mg/100 g), in the same study. Relatively low TA concentrations were found in *I. edulis* pulps (0.06 mg/100 g) and none were found in the seeds of this fruit. The DPPH• antioxidant capacity was lower in the pulp of this fruit, compared to the other pulps evaluated in this study, in agreement with its lower content of bioactive compounds.

The DPPH• antioxidant capacity of the fruit extracts varied from 68.66 µmol TE/g in *S. nigrum* peels to 13240.60 µmol TE/g in *H. dulcis* peel and pulp. The FRAP antioxidant activity varied from 22.93 µmol Fe²⁺/g in *S. nigrum* seed to 690.68 µmol Fe²⁺/g in *H. dulcis* pulp and peel. Both methods indicated that *H. dulcis* pulp and peel had the

TABLE 2: Total phenolic content (TPC, mg GAE/100 g), total flavonoids (TF, mg/EQ g), total anthocyanins (TA, mg/100 g), and antioxidant capacity by DPPH* ($\mu\text{mol}/\text{TE g}$) and FRAP ($\mu\text{mol}/\text{Fe}^{2+} \text{ g}$) in different parts of fruits based on fresh weight (FW).

Parts of fruits	Analysis				
	TPC	TF	TA	DPPH*	FRAP
Pulp					
<i>Syzygium cumini</i> Lam	313.66 ^b $\pm$ 8.93	59.00 ^b $\pm$ 2.36	9.26 ^a $\pm$ 0.41	2810.90 ^b $\pm$ 114.56	323.50 ^b $\pm$ 10.30
<i>Solanum nigrum</i> Linn	411.01 ^a $\pm$ 9.46	55.42 ^b $\pm$ 5.95	5.41 ^b $\pm$ 1.09	1726.60 ^{bc} $\pm$ 105.83	300.32 ^b $\pm$ 12.54
<i>Inga edulis</i> Mart	72.06 ^c $\pm$ 7.36	5.88 ^c $\pm$ 1.01	0.06 ^c $\pm$ 0.05	447.08 ^c $\pm$ 9.54	41.27 ^c $\pm$ 3.61
<i>Hovenia dulcis</i> Thunb	518.18 ^a $\pm$ 30.89	76.54 ^a $\pm$ 5.47	10.76 ^a $\pm$ 1.14	13240.60 ^a $\pm$ 309.61	690.68 ^a $\pm$ 9.43
Peel					
<i>Syzygium cumini</i> Lam	331.83 ^a $\pm$ 7.84	65.51 ^a $\pm$ 7.34	63.31 ^a $\pm$ 1.64	2010.90 ^b $\pm$ 32.49	341.95 ^b $\pm$ 6.08
<i>Solanum nigrum</i> Linn	21.89 ^b $\pm$ 1.38	43.39 ^b $\pm$ 5.44	4.63 ^c $\pm$ 0.12	68.66 ^c $\pm$ 1.27	48.36 ^c $\pm$ 3.38
<i>Inga edulis</i> Mart	NE	NE	NE	NE	NE
<i>Hovenia dulcis</i> Thunb	518.18 ^a $\pm$ 30.89	76.54 ^a $\pm$ 5.47	10.76 ^b $\pm$ 1.14	13240.60 ^a $\pm$ 309.61	690.68 ^a $\pm$ 9.43
Seed					
<i>Syzygium cumini</i> Lam	411.01 ^a $\pm$ 9.46	38.97 ^a $\pm$ 3.37	0.70 ^b $\pm$ 0.18	10396.40 ^a $\pm$ 639.45	209.30 ^a $\pm$ 3.77
<i>Solanum nigrum</i> Linn	19.27 ^c $\pm$ 1.06	10.24 ^c $\pm$ 1.42	1.40 ^a $\pm$ 0.28	95.12 ^c $\pm$ 1.15	22.93 ^c $\pm$ 1.11
<i>Inga edulis</i> Mart	66.32 ^b $\pm$ 8.68	2.59 ^d $\pm$ 0.69	ND	173.20 ^c $\pm$ 12.43	30.13 ^c $\pm$ 3.46
<i>Hovenia dulcis</i> Thunb	85.56 ^b $\pm$ 7.28	18.60 ^b $\pm$ 4.36	1.51 ^a $\pm$ 0.25	4877.50 ^b $\pm$ 195.27	75.22 ^b $\pm$ 1.22

Results expressed as mean $\pm$ standard deviation of three replicates. Equivalent parts of the different fruits were compared. Means followed by different letters in the same column are significantly different by Tukey's test ($p < 0.05$). *H. dulcis* pulp and peel were evaluated together. ND = not detected. NE = not evaluated.

TABLE 3: Quantification of phenolic compounds in different parts of fruits (mg/100 g FW).

Parts of fruits	Phenolic compounds			
	Gallic acid	Kaempferol	Epicatechin	Ellagic acid
Pulp				
<i>Syzygium cumini</i> Lam	5.59 ^a $\pm$ 0.09	14.29 ^b $\pm$ 5.67	ND	ND
<i>Solanum nigrum</i> Linn	0.82 ^b $\pm$ 0.08	16.62 ^b $\pm$ 5.49	ND	ND
<i>Inga edulis</i> Mart	ND	11.45 ^c $\pm$ 6.34	ND	ND
<i>Hovenia dulcis</i> Thunb	0.56 ^b $\pm$ 0.21	22.49 ^a $\pm$ 7.84	ND	ND
Peel				
<i>Syzygium cumini</i> Lam	9.63 ^a $\pm$ 0.04	18.32 ^b $\pm$ 7.98	ND	ND
<i>Solanum nigrum</i> Linn	0.53 ^b $\pm$ 0.02	57.38 ^a $\pm$ 12.46	ND	ND
<i>Inga edulis</i> Mart	NE	NE	NE	NE
<i>Hovenia dulcis</i> Thunb	0.56 ^b $\pm$ 0.21	22.49 ^b $\pm$ 7.84	ND	ND
Seed				
<i>Syzygium cumini</i> Lam	57.10 ^a $\pm$ 5.67	23.69 ^b $\pm$ 5.63	4.81 $\pm$ 0.57	3.63 $\pm$ 4.13
<i>Solanum nigrum</i> Linn	0.40 ^c $\pm$ 0.04	21.87 ^b $\pm$ 6.92	ND	ND
<i>Inga edulis</i> Mart	0.81 ^c $\pm$ 0.46	12.24 ^c $\pm$ 4.13	ND	ND
<i>Hovenia dulcis</i> Thunb	34.61 ^b $\pm$ 9.84	28.77 ^a $\pm$ 5.25	ND	ND

Results expressed as mean $\pm$ standard deviation of three replicates. Equivalent parts of the different fruits were compared. Means followed by different letters in the same column are significantly different by Tukey's test ($p < 0.05$). *H. dulcis* pulp and peel were evaluated together. ND = not detected. NE = not evaluated.

highest antioxidant capacity, probably due to having the highest TPC. Rufino et al. [9] evaluated the FRAP antioxidant capacity of methanolic extracts (methanol-water, 50 : 50 v/v) from the combined pulps and peels of *S. cumini* and found 35.5 $\mu\text{mol Fe}^{2+}/\text{g}$. This value was lower than that found in the current study, in pulp and peel of the same fruit (341.95 and 323.50 $\mu\text{mol Fe}^{2+}/\text{g}$, respectively).

3.2. Identification of Phenolic Compounds by HPLC. The phenolic compounds responsible for the antioxidant capacity

of the methanolic fruit extracts were gallic acid, kaempferol, epicatechin, and ellagic acid (with peak retention times of 3.6, 5.4, 14.7, and 19.1 min, respectively) (Table 3). The flavonoid, kaempferol, showed λ_{max} at 250 nm and epicatechin at 270 nm. The gallic and ellagic acids also showed λ_{max} at 270 nm, characteristic of phenolic acids derived from hydroxybenzoic acid [25].

Gallic acid was the main contributor to the antioxidant capacity of *S. cumini* seeds (57.10 mg/100 g) and *H. dulcis* seeds (34.61 mg/100 g). *S. cumini* pulps presented 5.59 mg/100

g of gallic acid, which was comparable to that found in walnut (3.39 mg/100 g) by Donno et al. [28]. As stated previously, phenolic compounds, such as gallic acid, exhibit potent antioxidant, antidiabetic, and antihyperlipidemic activities [29].

Although the *H. dulcis* pulp and peel showed the highest TPC content and the highest DPPH[•] and FRAP antioxidant capacities, it did not have the highest content of gallic acid. This was probably due to the limited interaction between the extraction solvent and gallic acid due to the high reactivity of this phenolic compound with oxidative materials and macromolecules. These factors make the identification process complex [30].

Kaempferol ranged from 11.45 mg/100 g in *I. edulis* pulps to 57.38 mg/100 g in *S. nigrum* peels. Using HPLC-DAD/mass spectrometry, Donno et al. [5] found 0.55 mg/100 g of kaempferol in strawberries, being a value lower than that found in all parts of fruits in this study. Ellagic acid and epicatechin were only identified in *S. cumini* seeds (3.63 and 4.81 mg/100 g, respectively). Epicatechin has been shown to exert several physiological effects, such as antioxidant activity and antitumor effects [31]. More recently, Rodríguez-Ramiro et al. [32] have shown that epicatechin was able to activate the detoxifying enzyme glutathione S-transferase pi 1 (GSTP1) related to the neutralization of reactive oxygen species and nitrogen species, in epithelial colon cells. Flavonols, like kaempferol and ellagic acid, possess health-promoting properties and may also contribute to the neutralization of free radicals and the maintenance of heart health [28, 33].

3.3. Fatty Acids (FAs). A total of eight FAs were detected in pulps, peels, and seeds of fruits (Table 4). *S. nigrum* peels presented 1212.20 mg/100 g in the sum of saturated fatty acid (Σ SFA), representing 87% of TL. Among the seeds, *I. edulis* presented the highest values of SFA, followed by *H. dulcis* (613.47 mg/100 g and 526.66 mg/100 g, respectively), showing a significant difference between them.

S. nigrum seeds presented a value of 1895.10 mg/100 g in the sum of monounsaturated fatty acid (Σ MUFA), representing 62.45% of TL. The major MUFA found in these seeds was OA (18:1n-9), showing a value of 1891.36 mg/100 g. This value of OA found was higher than those found by Santos et al. [13], in grape seed variety Isabel (1690.76 mg OA/100 g). The positive effect of dietary monounsaturated fats in preventing cancer has extensively been reported. In addition, the ingestion of this FA is associated with the improvement of the immune system due to its anti-inflammatory activity [8, 34].

H. dulcis seeds presented a value of 5921.07 mg/100 g in the sum of polyunsaturated fatty acid (Σ PUFA) (78.32% TL). The main PUFA was LNA (18:3n-3), which was found at the highest concentration in *H. dulcis* seeds (3985.95 mg/100 g). These seeds contained 8.73% TL, with 53% represented by LNA. Various n-3 PUFAs have been reported to exert beneficial effects, such as cholesterol regulation, blood pressure reduction, and cardiovascular disease prevention [35, 36].

According to Simopoulos [37], the relationship between PUFA/SFA should be greater than 0.4 to be adequate to prevent heart disease. The ratio of Σ SFA / Σ PUFA from all parts of the fruits has been higher than the recommended

value, except in *S. nigrum* peels. This result was low because the peel of this fruit is rich in eicosanoic acid (20:0), presenting statistical difference among the other peels. This result also reflected the sum of saturated fatty acid (Σ SFA) of the *S. nigrum* peels (1212.20 mg/100 g), which was the largest among all the peels.

The high n-6 to n-3 ratio in the diets of Western countries which occurred after the introduction of modern agriculture and animal farming techniques has, at least partly, contributed to the rise in cardiovascular disease. Studies indicate that the human diet can decrease the chances of developing several diseases, so the nutrition societies recommend a healthy diet containing n-6 to n-3 ratio of 5:1 [38, 39]. The n-6 to n-3 ratio of the fruits ranged within 2.38–51.01 in pulps, 0.24–19.54 in peels, and 0.06–78.30 in seeds. All parts of the *H. dulcis* and the *S. nigrum* peels, *S. cumini* seeds, and *I. edulis* seeds presented an ideal ratio of n-6 to n-3.

3.4. Principal Component Analysis (PCA). PC1 (68.70%) and PC2 (21.73%) explained 90.43% of the total variance (Figure 1(a)). Loadings (variables)/score (samples) of antioxidant analyses of the fruits showed the formation of four groups (Figure 1(b)). One group consisted of DPPH[•], FRAP, TPC, TF, and sample 9, due to higher values in this sample, in all of these variables. In PC1, DPPH[•], FRAP, TPC, and TF contributed positively to the separation of this group, whilst, in PC2, DPPH[•], FRAP, and TPC contributed negatively and TF positively. Sample 2 was grouped with TA and also has highlighted the contents of this variable. Sample 3 showed high DPPH[•] and TPC values; for this reason, its position was closer to these variables and separated from the other samples. A fourth group was formed by the remaining samples, which showed lower values in all analyses.

PC1 and PC2 accounted for 93.80% of the total variance of the FA data (Figure 2(a)) and 66.21% (PC1) of this variance was attributed to the positive contribution of the sum of PUFAs, n-6 and n-3 (Figure 2(b)). Sample 10 showed the highest values for these variables, forming group 1. The PUFAs and n-3 loadings of this group contributed negatively and only n-6 contributed positively to PC2, which accounted for 27.59% of the total variance. Sample 6 stood out in the MUFA values, contributing positively to PC2 and separating this group. The third group had the lowest values of the analyzed variables.

4. Conclusion

The bioactive compounds gallic acid and kaempferol were largely responsible for the antioxidant capacity of the parts of the fruits analyzed by HPLC, since the ellagic acid and the epicatechin were only found in *S. cumini* seeds. The parts of the fruits that stood out for their high TPC and TA contents showed a positive correlation between the contents of these compounds and the antioxidant capacity by the methods used. The PCA analysis showed that PC1 and PC2 explained 90.43% of the total variability of the antioxidant data. Most of the seeds studied showed omega-3, omega-6, and omega-9 FAs, in significant concentrations, with two PCs explaining

TABLE 4: Fatty acid composition in different parts of fruits (FA, mg/100 g sample).

Parts of fruits	Fatty acids														
	16:0	18:0	18:1n-7	18:1n-9	18:2n-6	18:3n-3	20:0	20:1n-9	Σ SFA	Σ MUFA	Σ PUFA	$\frac{\sum PUFA}{\sum SFA}$	Σ n-6	Σ n-3	$\frac{n-6}{n-3}$
Pulp															
<i>Syzgium cumini</i> Lam	2.19 ^c ± 0.03	6.38 ^c ± 0.05	0.26 ^b ± 0.01	18.19 ^b ± 0.17	5.10 ^d ± 0.03	0.10 ^d ± 0.01	0.04 ^b ± 0.01	0.08 ^c ± 0.01	8.61 ^c ± 0.08	18.52 ^b ± 0.10	5.20 ^d ± 0.03	0.60 ^b ± 0.01	5.10 ^d ± 0.03	0.10 ^d ± 0.01	51.01 ^a ± 0.24
<i>Solanum nigrum</i> Linn	1.05 ^c ± 0.15	7.88 ^c ± 0.27	8.63 ^a ± 0.23	0.20 ^d ± 0.09	48.09 ^b ± 6.02	0.94 ^b ± 0.23	0.22 ^a ± 0.38	0.10 ^c ± 0.03	9.20 ^c ± 0.74	8.93 ^c ± 0.93	49.03 ^b ± 6.25	5.32 ^c ± 0.23	48.09 ^b ± 6.02	0.94 ^b ± 0.23	51.15 ^a ± 1.41
<i>Inga edulis</i> Mart	11.66 ^b ± 4.74	13.93 ^c ± 3.46	0.07 ^c ± 0.01	97.36 ^c ± 33.8	8.45 ^c ± 10.56	0.22 ^c ± 0.01	0.05 ^b ± 0.01	0.47 ^a ± 0.05	25.64 ^b ± 2.92	97.90 ^c ± 12.39	8.67 ^c ± 0.12	0.33 ^c ± 0.05	8.45 ^c ± 10.56	0.22 ^c ± 0.01	38.40 ^b ± 5.68
<i>Hovenia dulcis</i> Thunb	58.28 ^a ± 9.95	8.10 ^b ± 0.24	9.02 ^a ± 0.20	2.14 ^c ± 0.23	98.69 ^a ± 10.47	41.36 ^a ± 8.58	0.36 ^a ± 0.02	0.25 ^b ± 0.01	66.74 ^a ± 2.26	11.41 ^c ± 0.32	140.05 ^a ± 19.30	2.09 ^c ± 0.17	98.69 ^a ± 10.47	41.36 ^a ± 8.58	2.38 ^c ± 0.14
Peel															
<i>Syzgium cumini</i> Lam	29.66 ^c ± 4.92	83.33 ^a ± 5.28	2.64 ^c ± 0.78	257.24 ^a ± 27.03	58.44 ^a ± 3.55	2.99 ^b ± 0.34	0.12 ^b ± 0.07	1.46 ^b ± 0.17	112.11 ^b ± 10.13	261.34 ^a ± 9.33	61.43 ^b ± 7.52	0.54 ^b ± 0.18	58.44 ^a ± 3.55	2.99 ^c ± 0.34	19.54 ^a ± 1.79
<i>Solanum nigrum</i> Linn	29.95 ^a ± 13.92	24.43 ^b ± 0.99	34.41 ^a ± 2.35	1.69 ^b ± 0.02	1.46 ^b ± 0.20	6.04 ^b ± 0.82	888.22 ^a ± 43.50	138.01 ^a ± 32.20	1212.20 ^a ± 56.44	174.11 ^b ± 12.19	75.0 ^c ± 0.23	0.01 ^c ± 0.01	1.46 ^b ± 0.20	6.04 ^b ± 0.82	0.24 ^c ± 0.02
<i>Inga edulis</i> Mart															
<i>Hovenia dulcis</i> Thunb	58.28 ^b ± 9.95	8.10 ^b ± 0.24	9.02 ^b ± 0.20	2.14 ^b ± 0.23	98.69 ^a ± 10.47	41.36 ^a ± 8.58	0.36 ^b ± 0.02	0.25 ^b ± 0.01	66.74 ^c ± 2.26	11.41 ^c ± 0.32	140.05 ^a ± 19.30	2.09 ^c ± 0.17	98.69 ^a ± 10.47	41.36 ^a ± 8.58	2.38 ^b ± 0.14
Seed															
<i>Syzgium cumini</i> Lam	1.56 ^c ± 0.45	0.95 ^d ± 0.10	0.24 ^d ± 0.12	291.01 ^c ± 41.70	0.45 ^d ± 0.05	7.08 ^d ± 0.16	0.40 ^c ± 0.15	1.55 ^c ± 0.43	2.91 ^d ± 0.26	292.79 ^c ± 9.38	7.53 ^d ± 0.21	2.58 ^c ± 0.13	0.45 ^d ± 0.05	7.08 ^d ± 0.16	0.06 ^d ± 0.01
<i>Solanum nigrum</i> Linn	37.90 ^b ± 5.37	15.14 ^c ± 1.06	2.67 ^b ± 0.38	1891.36 ^a ± 123.18	957.72 ^b ± 9.50	12.23 ^c ± 6.40	116.47 ^a ± 2.41	1.07 ^c ± 0.08	169.51 ^c ± 96.71	1895.10 ^a ± 176.61	969.95 ^b ± 14.68	5.72 ^b ± 0.34	957.72 ^b ± 9.50	12.23 ^c ± 6.40	78.30 ^a ± 3.04
<i>Inga edulis</i> Mart	313.70 ^a ± 52.29	2991.7 ^a ± 61.3	1.09 ^c ± 0.04	192.85 ^d ± 9.98	9.84 ^c ± 0.04	60.76 ^b ± 0.42	0.60 ^c ± 0.04	9.41 ^b ± 0.03	613.47 ^a ± 82.29	203.35 ^d ± 3.57	70.60 ^c ± 2.10	0.11 ^d ± 0.01	9.84 ^c ± 0.04	60.76 ^b ± 0.42	0.16 ^c ± 0.01
<i>Hovenia dulcis</i> Thunb	326.76 ^a ± 34.29	161.06 ^b ± 7.89	66.69 ^a ± 7.56	994.30 ^b ± 24.54	1935.12 ^a ± 120.17	3985.95 ^a ± 652.26	38.84 ^b ± 4.92	50.72 ^a ± 2.75	526.66 ^b ± 5.82	1111.71 ^b ± 2.28	5921.07 ^a ± 786.25	11.24 ^a ± 1.22	1935.12 ^a ± 120.17	3985.95 ^a ± 652.26	0.48 ^b ± 0.04

Results expressed as mean $\pm$ standard deviation of three replicates. Equivalent parts of the different fruits in the same column are significantly different by Tukey's test ($p < 0.05$). *H. dulcis* pulp and peel were evaluated together. SFA: saturated fatty acid, MUFA: monounsaturated fatty acid, PUFA: polyunsaturated fatty acid, n-6: omega-6 fatty acid, n-3: omega-3 fatty acid.

FIGURE 1: PC: principal component; (a) eigenvalue number; (b) loadings (variables)/scores (samples) biplot antioxidant analyses for the first and second PC of different parts of fruits: 1: *S. cumini* pulp; 2: *S. cumini* peel; 3: *S. cumini* seed; 4: *S. nigrum* pulp; 5: *S. nigrum* peel; 6: *S. nigrum* seed; 7: *I. edulis* pulp; 8: *I. edulis* seed; 9: *H. dulcis* pulp and peel; 10: *H. dulcis* seed.

FIGURE 2: PC: principal component; (a) eigenvalue number; (b) loadings (variables)/scores (samples) biplot of fatty acids for the first and second PC of different parts of fruits: 1: *S. cumini* pulp; 2: *S. cumini* peel; 3: *S. cumini* seed; 4: *S. nigrum* pulp; 5: *S. nigrum* peel; 6: *S. nigrum* seed; 7: *I. edulis* pulp; 8: *I. edulis* seed; 9: *H. dulcis* pulp and peel; 10: *H. dulcis* seed.

93.80% of the contribution of these FA contents and the sum of the MUFAs and PUFAs to these data.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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