

Morphological Characteristics, Nutritional Quality, and Bioactive Constituents in Fruits of Two Avocado (*Persea americana*) Varieties from Hainan Province, China

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Abstract

We studied the morphological characteristics, nutritional quality, and the bioactive compounds in fruits of two avocado accessions, RN-7 and RN-8, produced in Hainan province, China. Edible and non-edible parts of the fruit (pulp and seed) were compared to evaluate their possible contribution to improve the sustainability of the food and pharmaceutical industries. The basic characteristics evaluated were moisture, ash, total lipid, fatty acid composition, soluble sugars, titratable acid, soluble protein, and minerals. We also measured the concentrations of six types of bioactive compounds; total phenolics, flavonoids, tannin, ascorbic acid, tocopheryl acetate, and carotenoids. Our analyses of the nutritional compositions demonstrated that the pulp of the RN-7 and RN-8 proved to be rich in moisture, total lipid, and soluble protein. The seed, in turn, had higher soluble sugar, titratable acidity, sodium, potassium, calcium, iron, copper, and zinc contents. Other nutritional compositions (ash, magnesium, and manganese) had little differences between the pulp and seed of avocado fruit. With regard to the contents of bioactive compounds, the seed was superior to the pulp in the contents of total phenolics, flavonoids, and tannin. Regarding the concentrations of ascorbic acid, tocopheryl acetate, and total carotenoids, the highest values were found in the pulp. The results of fatty acid compositions displayed that the palmitic, palmitoleic, stearic, oleic, and linoleic acid contents of the pulp were higher than those of the seed, while myristic and arachic acid had higher contents in the seed.

Keywords: chemical compositions, pulp, seed

1. Introduction

Avocado (*Persea americana* Mill.) belongs to the botanical family Lauraceae, and originated in Mexico, or possibly Central or South America (Dreher & Davenport, 2013). Avocado fruit is rich in lipids, proteins, minerals, vitamins, and other nutrients and active ingredients (Dreher & Davenport, 2013; Galvão et al., 2014). The lipid content can comprise 15-30% of the fresh weight of the fruit depending on the cultivar, season, and growing conditions (Meyer & Terry, 2008). It is remarkable is that the lipids in avodado fruit contain ~60% monounsaturated fatty acids and ~13% essential fatty acids such as linoleic and linolenic acid, which are beneficial to human cardiovascular health (Villa-Rodríguez et al., 2011; Giraldo & Moreno-Piraján, 2012; Dreher & Davenport, 2013; Donetti & Terry, 2014; Pedreschi et al., 2016). In contrast to lipid content, the sugar content of avocado fruit is relatively low (Meyer & Terry, 2008, 2010). Hence, avocado fruit is generally recommended for people suffering from diabetes because it is a high-energy food. Natural antioxidants, in particular flavonoids and other groups of polyphenols, have potential uses in the pharmaceutical and food

industries because of their many benefits such as reducing the risk of inflammatory diseases and preventing lipid oxidation (Chen et al., 2014). Avocado fruit contains more phenolic compounds than other kinds of tropical and subtropical fruits (Kosinska et al., 2012; Vinha et al., 2013; Chen et al., 2014). Recent studies have also demonstrated that avocado contains other bioactive compounds that are equally beneficial to human health, such as vitamins and phytochemicals (vitamin C, vitamin E, carotene, etc.) and minerals (phosphorus, sodium, potassium, calcium, and magnesium) (Dreher & Davenport, 2013; Vinha et al., 2013).

It is widely recognized that the avocado was initially introduced to China in 1918 (Papademetriou, 2000). Hundreds of avocado varieties were also introduced to China from the USA, Israel, and other countries in the late 1950s (He, 2012; Zhang et al., 2015). Traditional selective breeding and hybridization were widely used and continue to this day at the Chinese Academy of Tropical Agricultural Sciences, resulting in the introduction of more than a dozen high-quality avocado varieties (Zhang et al., 2015). At present, several Chinese superior avocado varieties are widely grown in various regions of Hainan province.

The avocado cultivars ‘Fuerte’ and ‘Hass’ are the most commercially valuable varieties and account for up to two-thirds of the avocado production around the world. Hence, most studies of avocado quality characteristics use these two cultivars (Ashton et al., 2006; Meyer & Terry, 2008, 2010; Hurtado-Fernandez et al., 2011, 2014, 2015; Rodríguez-Carpena et al., 2011; Villa-Rodríguez et al., 2011; Reddy et al., 2012; Donetti & Terry, 2014; Ferreyra et al., 2016; Pedreschi et al., 2016; Rohman et al., 2016). However, no similar studies on Chinese native avocado varieties have been published to date. Thus, the objectives of this study were to determine the morphological characteristics, nutritional quality, and the compositions of bioactive compounds in the fruits of two avocado varieties, RN-7 and RN-8. These two varieties have been recommended by the Chinese Academy of Tropical Agricultural Sciences, located in the province of Hainan, China. The non-edible seeds were also investigated in order to evaluate their potential use as cheap waste production for the food, pharmaceutical, and dermocosmetic industries.

2. Materials and Methods

2.1 Plant Material, Reagents, and Sample Preparation

Fruits of the two avocado accessions, RN-7 and RN-8, used in the present study were obtained from the garden of the avocado germplasm resource affiliated with the Chinese Academy of Tropical Agricultural Sciences in Danzhou city, Hainan province, China (North latitude: 19°11', East longitude: 108°50'). Eighteen mature fruits of each accession were collected and selected for their firmness and absence of mechanical damage and visible decay. The fruits were immediately transported in standard polystyrene foam boxes that are used for export packaging and held at 5-6 °C until they reached the laboratory. The pulp and seeds were separated from the fruits, homogenized using a kitchen blender, and stored at 4 °C until analysis, which was conducted within one week.

2.2 Morphological Characteristics and Physicochemical Assays

Fruit skin color was determined from eight points on the equatorial area of each fruit per accession using a SPAD-502 Plus colorimeter. Values were obtained in CIELAB scale (L^* , a^* , b^*), and Hue angle and chroma values were calculated. Length, width, and weight were measured for each fruit and its seed. The measurements were performed on nine fruits of each accession.

2.3 Quantification of Nutritional Compositions

2.3.1 Moisture Assay

Fresh avocado pulp and seed samples (5 g) were homogenized separately using a high-speed homogenizer and placed in an air dry oven (GZX-9146 MBE, Shanghai, China) at 105 °C for 6 h. Dry weights of avocado pulp and seed were measured, and moisture contents (g) were calculated from the differences between fresh and dry weight. The results are expressed as g/100 g on a fresh weight basis. The measurements were performed in three replications per accession.

2.3.2 Ash Assay

A quartz crucible was placed in a muffle furnace at 550 °C for 0.5 h, removed when the temperature had dropped below 200 °C, and weighed after reaching room temperature, followed by repeated burning to attain a constant weight. The quartz crucible containing fresh avocado pulp or seed (5 g) was weighed, the samples were fully carbonized until smoke-free, placed in the muffle furnace at 550 °C for 4 h, and removed when the temperature was < 200 °C. The quartz crucible containing the ash was weighed again after cooling to room temperature. The results are displayed as g/100 g on a fresh weight basis. The experiments were performed in triplicate for each accession.

2.3.3 Oil Content Assay

Oil content was evaluated by the method of Villa-Rodríguez et al. (2011) with slight modifications. The avocado pulp and seed were dried and ground to a powder, and the dry powders (5 g) were transferred to a filter paper cylinder after addition of absolute ether at 50 °C. The ratio of material to ether was 1:20. The filtered solutions were extracted until no more oil was present using a Soxhlet extractor. The extracts were then evaporated on a rotary evaporator and weighed. The results are expressed as g/100 g on a fresh weight basis. The experiments were performed in triplicate for each avocado accession.

2.3.4 Soluble Sugar Assay

The total sugar content was determined using the colorimetric anthrone method described by Meyer and Terry (2008) with some modifications. Fresh avocado pulp and seed samples (1 g) were homogenized using a high-speed homogenizer with 5 ml of ethanol (80%) and transferred to 10 mL test tubes. The sample solutions were placed in a boiling water-bath for 10 min after decolorization with activated carbon. After cooling, the solutions were filtered and transferred to 25 mL triangular flasks, and the volumes were adjusted to 25 mL with ethanol (80%). Filtrate samples (1 mL) were mixed with 5 mL anthrone reagent, shaken gently, and then placed in a boiling water-bath for 10 min. After cooling, the absorbance was measured at 620 nm using a spectrophotometer (1 mL distilled water plus 5 mL anthrone reagent was used as the blank control). The total sugar content was calculated based on a calibration curve for glucose ($R^2 = 0.997$). The soluble sugar content was expressed as g/100 g fresh weight of sample. All measurements were performed in triplicate for each accession.

2.3.5 Titratable Acidity Assay

Fresh avocado pulp and seed (5 g) samples were homogenized using a high-speed homogenizer (Heidolph, Dixa 900, Germany). The samples were transferred to 50 mL triangular flasks, mixed with 30 mL of distilled water, and incubated in a water-bath at 80 °C for 90 min with constant stirring. After cooling, the solutions were transferred to 50 mL centrifuge tubes, centrifuged at 8000 rpm for 10 minutes. The supernatants were transferred to 50 mL calibrated flasks and the volumes were adjusted to 50 mL with distilled water. Titratable acidity was determined by titrating 15 mL of avocado aqueous extracts with 0.01 M NaOH, using phenolphthalein (1%) as indicator. Distilled water was the blank control. Results were expressed as grams of tartaric acid per 100 g of sample, according to the methodology described by Vinha et al. (2013). The experiments were carried out in triplicate for each accession.

2.3.6 Soluble Protein Assay

Analyses of soluble protein contents were carried out using the Coomassie Blue staining method of Bradford (1976) with some modifications. Fresh avocado pulp and seed samples (1 g) were homogenized using a high-speed homogenizer in 5 mL of distilled water and transferred to 25 mL triangular flasks. The volume was then adjusted to 25 mL with distilled water. After filtration, 0.1 mL samples of the filtrates were transferred to test tubes, 5 mL of the Coomassie Brilliant Blue G-250 reagent was added to each, and they were mixed completely. After incubation for 2 min at room temperature, the absorbance was measured at 595 nm using a Shimadzu UV-1800 spectrophotometer. Distilled water was the blank control. The soluble protein was calculated using 0-100 µg/mL and 1000 µg/mL calibration curves based on bovine serum albumin ($R^2 = 0.997$). The soluble protein content was expressed as g/100 g fresh weight of sample. All measurements were performed in triplicate for each accession.

2.3.7 Fatty Acid Composition Assay

Analyses of the fatty acid profiles were performed using the method of Villa-Rodríguez et al. (2011) with some modifications. The oils extracted from avocado pulp and seed (40 µL) were saponified at 80 °C for 30 min after addition of 5 mL NaOH-MeOH (0.2 mol/L). After cooling, the solutions were mixed with 2.5 mL BF₃-MeOH (14%) and incubated at 80 °C for 30 min to produce methyl esters of the fatty acids. Following this, 2 mL of saturated NaCl and 4 mL *n*-hexane were added, and the resulting solutions were refluxed for 15 min. The upper layers were then removed, filtered through 0.22 µm membranes, and used for fatty acid GC-MS analyses.

The analyses were performed on an Agilent7890B-7000B GC-MS equipped with a DB-5MS (60 m × 0.25 mm i.d., 0.25 µm film thickness) column using helium (1.2 mL/min) as the carrier gas. The oven temperature was programmed as follows: initial temperature of 100 °C held for 3 min, increased to 180 °C at 3 °C/min, held for 1 min, increased to 220 °C at 1 °C/min, held for 1 min, and finally increased to 280 °C at 5 °C/min and held for 5 min. Injector and detector temperatures were 250 °C and 230 °C, respectively. The mass spectrometer was operated in the electron impact mode at 70 eV in the scan range of 35-400 m/z. The fatty acid methyl esters

(FAMES) were identified by comparing peaks retention times to those of commercial standards and by comparing the respective ion chromatograms with those reported in the NIST 2011 library. The FAMES were quantified against methyl nonadecanoate that was added as an internal standard. Quantifications were evaluated from calibration curves of the respective FAMES ($R^2 \geq 0.995$). FAMES were expressed as mg/100 g on a fresh weight basis. The experiments were carried out in triplicate for both avocado accessions.

2.3.8 Mineral Elements Assay

Avocado pulp and seeds were dried and ground to a powder. The dry powders (0.5 g) were transferred to 50 mL beakers containing 5 mL concentrated nitric acid and digested at a temperature of 140 °C for 1 h on an electric hot plate. Determination of Na, Mg, K, Ca, Mn, Fe, Cu, and Zn in the previously mineralised samples was conducted in an AAnalyst 400 atomic absorption spectrometer (Perkin Elmer Ltd., Shanghai, China). Concentrated nitric acid was used as the blank control. Quantification was obtained from a calibration curve of certified reference materials ($R^2 \geq 0.995$). Mineral elements were expressed as mg/100 g on a fresh weight basis. The experiments were performed in triplicate for both accessions.

2.4 Quantification of Bioactive Compounds

2.4.1 Total Polyphenolic Assay

Total phenolic content was determined based on the method of Villa-Rodríguez et al. (2011) with slight modifications. Dry powder residues of avocado pulp and seed (0.2 g, after lipid removal) were dissolved in 5 mL ethanol (50%), subjected to ultrasonic extraction for 15 min, and centrifuged at 8000 rpm for 10 minutes. The 0.1 mL extracts were transferred to centrifuge tubes, mixed with 3 mL distilled water, 0.25 mL Folin and Ciocalteu phenol reagent (1 N), and 0.75 mL Na_2CO_3 (20%), and then diluted with distilled water to a volume of 5 mL. After incubation for 30 min in the dark at room temperature, the absorbance was read at 760 nm using a Shimadzu UV-1800 spectrophotometer. Distilled water was used as the blank control. Total phenolic compounds were calculated using a calibration curve of gallic acid ($R^2 = 0.997$) and displayed as gallic acid equivalents (mg GAE/100 g on a fresh weight basis). The experiments were performed in triplicate for the two accessions.

2.4.2 Tannin Assay

Fresh avocado pulp and seed samples (5 g) were homogenized separately in 80 mL distilled water using a high-speed homogenizer (Heidolph, Diap 900, Germany), placed in a boiling water-bath for 30 min, and then adjusted to a volume of 100 mL with distilled water. Subsamples (5 mL) of the solutions were transferred to centrifuge tubes and centrifuged at 8000 rpm for 5 minutes. For the assays, 1 mL aliquots of the extracts were combined with 1 mL $\text{Na}_2\text{WO}_4 \cdot 2\text{H}_2\text{O}$ and Na_2MoO_4 mixture solution, 3 mL Na_2CO_3 , and 5 mL distilled water, and incubated for 2 hour in the dark at room temperature to allow for color development. The 0 mg/L sample was used as the blank control. Tannin content was measured spectrophotometrically on a Shimadzu UV-1800 spectrophotometer at 765 nm. The values on the calibration curve were referred to total polyphenolic assay and displayed as gallic acid equivalents. The results are displayed as mg GAE/100 g on a fresh weight basis. The experiments were performed in triplicate.

2.4.3 Total Flavonoid Assay

Total flavonoid concentrations were determined by the method of Villa-Rodríguez et al. (2011) with some modifications. As in the polyphenol assay, dry powder residues of avocado pulp and seed (0.2 g, after lipid removal) were dissolved in 5 mL ethanol (50%), extracted ultrasonically for 15 min, and centrifuged at 8000 rpm for 10 minutes. Aliquots of the extracts (0.1 mL) were transferred to centrifuge tubes, mixed with 2 mL distilled water and 0.15 mL NaNO_2 (5%), and incubated for 6 min. After 0.15 mL of $\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$ (10%) was added, the extracts were allowed to stand for 1 min, and 2 mL of NaOH (1 M) were then added. The volume was adjusted to 10 mL with distilled water. For the seed samples, 0.5 mL aliquots of the extracts were diluted with distilled water to 6 mL, and the absorbance was determined at 510 nm with a Shimadzu UV-1800 spectrophotometer. Distilled water was used as the blank control. Total flavonoid concentrations were calculated using a calibration curve of rutin ($R^2 = 0.995$) and expressed as rutin equivalents (mg RE/100 g on a fresh weight basis). The experiments were performed in triplicate.

2.4.4 Ascorbic Acid Assay

Ascorbic acid content was determined using the modified 2,6-dichlorophenolindophenol method (Franck et al., 2003). Samples of fresh avocado pulp and seed (5 g) were homogenized separately using a high-speed homogenizer (Heidolph, Diap 900, Germany) in 5 mL of oxalic acid (2%). The volume was adjusted to 100 mL with distilled water. The sample solutions were transferred to centrifuge tubes, centrifuged at 8000 rpm for 10 minutes, and 10 mL aliquots of the extracts were titrated with 2,6-dichlorophenolindophenol. Oxalic acid (2%)

was used as the blank control. Ascorbic acid was expressed as mg/100 g on a fresh weight basis. The experiments were performed in triplicate.

2.4.5 Tocopheryl Acetate Assay

Samples of fresh avocado pulp and seed (5 g) were homogenized using a high-speed homogenizer as described above for the moisture, acidity, and soluble protein assays. After addition of 50 mL ethanol (95%), 10 mL ethyl ether, and 20 mL NaOH solution (50%), the solutions were transferred to 250 mL saponification flasks and saponified by refluxing in a boiling water bath for 30 min. The saponification reactions were transferred to 250 mL separatory funnels and mixed with 40 mL distilled water and 50 mL ethyl ether. After shaking vigorously for 1 min, the supernatants were transferred to 250 mL volumetric flasks, and the volumes were adjusted to 250 mL with ethyl ether. Samples (5 mL) of the extracts were heated at 90 °C in a water-bath until the flask contents were reduced almost to dryness. The end-products were dissolved in methanol to a final volume of 10 mL. The absorbance at 284 nm was measured using a Shimadzu UV-1800 spectrophotometer. Methanol was used as the blank control. Tocopheryl acetate was expressed as mg/100 g on a fresh weight basis. The experiments were performed in triplicate.

2.4.6 Total Carotenoid Assay

Fresh avocado pulp and seed samples (5 g) were homogenized using a high-speed homogenizer as described in the previous section. The volume was adjusted to 100 mL with petroleum ether. The sample solutions were filtered through sodium sulphate, transferred to 100 mL volumetric flasks, and then diluted to 100 mL with petroleum ether. After incubation for 24 h at room temperature, total carotenoid content was measured spectrophotometrically at 445 nm using a Shimadzu UV-1800 spectrophotometer. Petroleum ether was the blank control. The results are presented as β -carotene equivalents (mg/100 g on a fresh weight basis). The experiments were carried out in triplicate.

2.5 Statistical Analyses

The data were analyzed using SPSS version 20.0 software (SPSS Inc., Chicago, IL, USA). The results were presented as the mean \pm standard deviation of three or nine measurements.

3. Results and Discussion

3.1 Morphological Characteristics and Physicochemical Analyses

Table 1 shows the morphological parameters of the fruits of two avocado accessions. The fruits of RN-7 are ovate, large in size (505.56 ± 39.09 g), and are larger than those of many of the widely cultivated avocado varieties such as 'Hass', 'Fuerte', 'Gwen', and 'Lamb Hass', etc. (Gómez-López et al., 1999, 2002; Schaffer et al., 2012). While the fruits of RN-8 were pyriform and of medium size (336.67 ± 26.46 g). The seeds of RN-7 and RN-8 were all of medium size (50.90 ± 9.08 and 69.73 ± 4.26 g) and nearly spherical in shape. Others have reported the seed weight of more than thirty avocado cultivars from around the world from 26.54 g for Duke to 115.82 g for Ceniap 2 (Gómez-López et al., 1999, 2002; Rodríguez-Carpena et al., 2011; Galvão et al., 2014). This indicates that these two Chinese native avocado accessions had the medium-sized seeds in comparison with foreign avocado varieties. Lightness and chroma of RN-7 avocado fruit were greater than those of RN-8; however, the hue angle of RN-7 avocado fruit was the same as RN-8, which suggest that the degree of saturation and color intensity of RN-7 avocado fruit are higher than those of RN-8 avocado fruit, but peel color of RN-7 and RN-8 were very similar.

Table 1. Morphological characteristics and physicochemical parameters (mean value \pm standard deviation, $n = 9$) of fruits of two Chinese avocado accessions

Morphological characteristic	RN-7	RN-8
Fruit weight (g)	505.56 \pm 39.09	336.67 \pm 26.46
Fruit length (cm)	12.30 \pm 0.50	16.88 \pm 1.30
Fruit diameter (cm)	8.92 \pm 0.38	6.91 \pm 0.31
Seed weight (g)	69.73 \pm 4.26	50.90 \pm 9.08
Seed length (cm)	4.49 \pm 0.24	5.90 \pm 0.60
Seed diameter (cm)	4.71 \pm 0.18	4.23 \pm 0.22
L*	45.27 \pm 2.35	38.35 \pm 2.36
Hue (°)	1.09 \pm 0.02	1.09 \pm 0.04
Chroma (%)	40.37 \pm 3.02	31.01 \pm 4.66

Note. L* = Luminance; Hue (°) = Hue angle.

3.2 Nutritional Compositions Analyses

The avocado pulp has a higher water and total lipid content than does the seed (Table 2), which is in agreement with previous studies (Rodríguez-Carpena et al., 2011; Vinha et al., 2013; Galvão et al., 2014). The total lipid contents in the pulp of RN-7 and RN-8 fruits were all $\leq 8\%$, placing them in the group of varieties with low oil content, which are inferior to the widely grown 'Hass' cultivar (Gómez-López, 1999, 2002; Rodríguez-Carpena et al., 2011; Villa-Rodríguez et al., 2011; Dreher and Davenport, 2013). The pulp of RN-7 and RN-8 contains more soluble protein (0.42 g/100 g), more than twice that measured in the seed in the present study (Table 2), although previous studies found that soluble protein levels in the pulp are lower than levels in the peel and seed (Rodríguez-Carpena et al., 2011; Vinha et al., 2013; Galvão et al., 2014). Comparable levels of ash differed between the pulp and the seed depending on the variety (Rodríguez-Carpena et al., 2011; Vinha et al., 2013; Galvão et al., 2014). Similarly, we found that comparisons of the ash content between the pulp and seed were the exact opposite for RN-7 and RN-8 (Table 2). The soluble sugar content and titratable acidity of the pulp were higher than in the seed, which agreed with the results of a previous study (Vinha et al., 2013). In addition, recent research has shown that soluble sugars may be the precursors of lipid synthesis in avocado fruit (Kilaru et al., 2015). This was supported in the present study, where we found that the higher the lipid content, the lower the soluble sugar levels in the pulp and seed of avocado fruit (Table 2). The contents of six mineral elements (sodium, potassium, calcium, iron, copper, and zinc) were higher in the seed than in the pulp, but two other mineral elements (magnesium and manganese) showed very small differences between the pulp and seed (Table 2).

Table 2. Nutritional compositions (mean value \pm standard deviation, proximates for g/100 g FW and minerals for mg/100 g FW, $n = 3$) of pulp and seed of fruits of two Chinese avocado accessions

Nutritional composition	RN-7		RN-8	
	Pulp	Seed	Pulp	Seed
<i>Proximates</i>				
Moisture	82.85 \pm 0.17	69.61 \pm 0.20	83.59 \pm 0.32	69.71 \pm 0.38
Ash	0.52 \pm 0.00	0.64 \pm 0.01	0.74 \pm 0.01	0.63 \pm 0.01
Total lipid	7.33 \pm 0.15	1.40 \pm 0.04	6.53 \pm 0.14	3.18 \pm 0.17
Soluble sugar	0.56 \pm 0.02	1.78 \pm 0.03	0.72 \pm 0.05	2.43 \pm 0.03
Titrateable acidity	1.78 \pm 0.03	2.57 \pm 0.08	2.63 \pm 0.03	2.87 \pm 0.03
Soluble protein	0.42 \pm 0.03	0.19 \pm 0.02	0.42 \pm 0.02	0.16 \pm 0.01
<i>Minerals</i>				
Sodium	0.52 \pm 0.04	1.54 \pm 0.01	0.47 \pm 0.04	1.11 \pm 0.09
Magnesium	1.40 \pm 0.01	1.41 \pm 0.06	1.73 \pm 0.02	1.83 \pm 0.03
Potassium	247.01 \pm 10.58	336.45 \pm 25.24	240.24 \pm 1.24	310.95 \pm 0.64
Calcium	10.87 \pm 0.52	18.51 \pm 0.76	9.01 \pm 0.20	14.32 \pm 0.47
Manganese	0.03 \pm 0.00	0.05 \pm 0.00	0.03 \pm 0.00	0.03 \pm 0.00
Iron	0.91 \pm 0.03	1.18 \pm 0.09	0.80 \pm 0.11	1.48 \pm 0.05
Copper	0.18 \pm 0.00	0.35 \pm 0.00	0.16 \pm 0.00	0.33 \pm 0.03
Zinc	0.05 \pm 0.00	0.07 \pm 0.00	0.06 \pm 0.00	0.10 \pm 0.00

3.3 Fatty Acid Profiles Analyses

The fatty acid compositions of the pulp and seed oils are presented in Table 3. Eight fatty acids were detected in the pulp and seeds of fruits of avocado accessions RN-7 and RN-8. The palmitic (C16:0), palmitoleic (C16:1), stearic (18:0), oleic (C18:1), and linoleic (18:2) acid contents of the pulp were higher than those in the seed. Myristic (C14:0) and arachic (C20:0) acid levels were higher in the seed, while the linolenic acid (C18:3) levels differed considerably between the two accessions. These results differed slightly from those reported previously, since Galvão et al. (2014) suggested that levels of oleic acid (C18:1) in the pulp and linoleic (18:2) in the seed were higher among the cultivars 'Fortuna', 'Collinson', and 'Barker'. Nevertheless, other fatty acids such as myristic (C14:0), palmitic (C16:0), palmitoleic (C16:1), stearic (18:0), and arachic (C20:0) acids showed different comparable levels between the pulp and seed among these three cultivars. The contents of palmitic (C16:0), oleic acid (C18:1), and linoleic (18:2) acids in the pulp of RN-7 and RN-8 avocado fruits all exceeded 1000 mg/100 g fresh weight, and represented the majority of the total fatty acids quantified. These results agree with those reported previously by other authors for avocado cultivars such as 'Hass' and 'Fuerte', etc. (Ozdemir & Topuz, 2004; Meyer & Terry, 2008, 2010; Villa-Rodríguez et al., 2011; Dreher & Davenport, 2013; Donetti & Terry, 2014; Galvão et al., 2014; Ferreyra et al., 2016; Pedreschi et al., 2016; Rohman et al., 2016). In all of them, more than 63% of total fatty acids (TFA) of the pulp and seed of avocado were unsaturated, the remaining were saturated (37%) (Table 3). Total unsaturated fatty acids (Σ UFA), total saturated fatty acids (Σ SFA), and TFA were all much higher in the pulp of two accessions than in the seed (Table 3). The ratios of Σ UFA/ Σ SFA were larger than 1.0 for the pulp and seed of two accessions, especially in the pulp of RN-8, which indicated that avocado could serve as a food supplement in the diet to decrease the level of cholesterol and fats, preventing the risk of cardiovascular disease (Richard et al., 2008).

Table 3. Fatty acid compositions (mean value \pm standard deviation, mg/100 g FW, $n = 3$) of the pulp and seed oils obtained from fruits of two Chinese avocado accessions

Fatty acids	RN-7		RN-8	
	Pulp	Seed	Pulp	Seed
<i>Saturated fatty acids (SFA)</i>				
Myristic acid (C14:0)	15.46 \pm 0.44	28.01 \pm 1.00	12.90 \pm 0.16	25.92 \pm 1.17
Palmitic acid (C16:0)	2431.81 \pm 97.27	288.07 \pm 29.22	1727.92 \pm 6.73	359.00 \pm 24.52
Stearic acid (18:0)	83.71 \pm 4.23	44.92 \pm 1.81	73.20 \pm 2.77	46.31 \pm 2.00
Arachic acid (C20:0)	25.38 \pm 0.37	40.59 \pm 0.11	24.02 \pm 0.24	40.62 \pm 0.15
<i>Mono-unsaturated fatty acids (MUFA)</i>				
Palmitoleic acid (C16:1)	526.63 \pm 26.46	53.99 \pm 4.30	393.11 \pm 6.36	48.32 \pm 5.52
Oleic acid (C18:1)	2090.11 \pm 169.83	266.08 \pm 14.39	1999.57 \pm 181.68	174.51 \pm 10.41
<i>Poly-unsaturated fatty acids (PUFA)</i>				
Linoleic acid (18:2)	1615.41 \pm 98.16	420.23 \pm 6.78	1481.21 \pm 15.94	626.85 \pm 82.75
Linolenic acid (C18:3)	83.20 \pm 5.49	74.37 \pm 4.70	44.46 \pm 3.54	61.30 \pm 5.49
Σ SFA	2556.36 \pm 38.73	401.59 \pm 12.19	1838.04 \pm 9.9	471.85 \pm 12.27
Σ UFA	4315.35 \pm 84.65	814.67 \pm 27.32	3918.35 \pm 48.61	910.98 \pm 26.05
TFA	6871.71 \pm 123.38	1216.26 \pm 39.51	5756.39 \pm 58.51	1382.83 \pm 38.32
Σ UFA/ Σ SFA	1.69 \pm 0.02	2.03 \pm 0.01	2.13 \pm 0.02	1.93 \pm 0.01

Note. Σ SFA = total saturated fatty acids; Σ UFA = total unsaturated fatty acids; TFA = total fatty acids.

3.4 Bioactive Compounds Analyses

Remarkably, the total phenolic, flavonoid, and tannin contents of the seed were 10- to 40-fold greater than in the pulp (Table 4). Previous studies also found that total phenolic and flavonoid contents of the seed far exceeded those of the pulp, and these compounds possess strong *in vitro* antioxidant activity and antimicrobial potential (Hidalgo et al., 2010; Rodríguez-Carpena et al., 2011; Kosinska et al., 2012; Vinha et al., 2013). Therefore, the avocado seed, as a byproduct, could be an interesting and inexpensive raw material for a functional food ingredient or an antioxidant additive (Rodríguez-Carpena et al., 2011; Kosinska et al., 2012). Vinha et al. (2013) suggested that the ascorbic acid and total carotenoid contents of the seed are superior to those in the pulp. However, considering the contents of ascorbic acid and total carotenoids, we found that the highest values were in the pulp (Table 4). Tocopheryl acetate was not detected in the seed of either of the two accessions; however, tocopheryl acetate was present in almost the same amounts in the pulp and seed (Vinha et al., 2013).

Table 4. Bioactive constituents (mean value \pm standard deviation, mg/100 g FW, $n = 3$) present in the pulp and seed of fruits of two Chinese avocado accessions

Bioactive constituents	RN-7		RN-8	
	Pulp	Seed	Pulp	Seed
Total phenolics	44.39 \pm 2.81	685.58 \pm 13.45	109.39 \pm 5.29	798.52 \pm 54.04
Flavonoids	43.85 \pm 6.37	1636.25 \pm 50.88	-	936.60 \pm 56.91
Tannin	0.09 \pm 0.01	2.02 \pm 0.04	0.05 \pm 0.01	2.45 \pm 0.09
Ascorbic acid	18.57 \pm 0.31	10.03 \pm 0.00	17.15 \pm 1.28	12.02 \pm 0.20
Tocopheryl acetate	1.28 \pm 0.03	-	2.32 \pm 0.11	-
Total carotenoids	2.14 \pm 0.18	2.02 \pm 0.04	2.02 \pm 0.02	1.98 \pm 0.05

4. Conclusions

This study is the first to characterize and evaluate the morphological characteristics and the contents of nutrients and bioactive constituents in fruits of two avocado cultivars grown in Hainan province, China. The pulp and seeds were thoroughly compared in fruits from cultivars RN-7 and RN-8. For the main nutrients, the highest lipid and soluble protein contents were found in the pulp, but total phenolics, flavonoids, and tannin concentrations in the seed were 10- to 40-fold higher than in the pulp. The pulp and seed of both avocado accessions were found to contain a variety of fatty acids with carbon chain lengths of C14, C16, C18, and C20

and varying degrees of unsaturation. Relatively high levels of palmitic, oleic, and linoleic acids were present in the pulp.

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