

Research Article

Organic Acids, Antioxidants, and Dietary Fiber of Mexican Blackberry (*Rubus fruticosus*) Residues cv. Tupy

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Blackberry fruit processing generates residues comprised of peel, seeds, and pulp that are abundant in flavonoids, colorants, and organic acids. The objective of this study was to determine the organic acids, antioxidants, and dietary fiber content of blackberry residues and compare antioxidants and dietary fiber content of a prune-based commercial product. The ABTS, DPPH, and FRAP methodologies were used for antioxidant capacity. The blackberry residues exhibited a high amount of malic acid (5706.37 mg/100 g db), phenols (4016.43 mg GAE/100 g db), and anthocyanins content (364.53 mg/100 g db) compared with the commercial product. These compounds contributed to the antioxidant capacity (by ABTS) of both products but were 20 times higher in blackberry residues. The fruit residues were able to reduce iron (by FRAP) 4.4 times compared to the prune-based product. Total dietary fiber (44.26%) and functional properties as water retention capacity (2.94 g/g), swelling capacity (5.00 mL/g), and fat absorption capacity (1.98 mL/g) of blackberry residues were significantly higher than those of the commercial sample. The results demonstrated that, due to its antioxidant compounds and functional properties, the blackberry residue can be considered a source of components with potential benefit to human health.

1. Introduction

Berries are mainly consumed fresh but are also used to manufacture products such as juices and concentrates [1]. The residues generated after separation of peels, seeds (4.4% to 12.2%), and pulp are commonly discarded by the industry but still have flavonoids, colorants, pectins, and organic acids [2–4] that can be used or recovered.

Organic acids and sugars are soluble constituents in berries that are responsible for taste and serve as an index of fruit ripeness; both factors affect consumer acceptability [5].

Previous research has reported that different species of whole berries contain organic acids such as citric, malic, tartaric, fumaric, and shikimic acids in total content ranges of 21.5 to 235 mmol/kg, specifically of 45.1 mmol/kg for the *fruticosus* species [6]. Published literature about the content of these individual compounds in blackberry (*Rubus fruticosus*) residues was not found.

Blackberry (*Rubus fruticosus*) fruits also contain phenolic compounds as anthocyanins, flavonols, chlorogenic acid, and procyanidins, which can have beneficial effects on human health [7]. Phenols are considered compounds with potent

antioxidant and free-radical scavenging properties that protect important biomolecules against oxidative damage [8]. Several studies have evaluated these compounds in blackberries (*Rubus*) of different species and varieties (*R. sp. hyb* Marion, *R. laciniatus* Evergreen, *R. spp.* Tupy, and *R. fruticosus*) as a whole fruit and in pulp or seeds, including the use of several extraction technologies (supercritical carbon dioxide extraction, ultrasound assisted extraction, pressurized liquid extraction, etc.) to recover phenols, anthocyanins, fatty acids, phytosterols, and tocopherols, compounds responsible for antioxidant activity [9–14]. Fewer studies have been performed on the evaluation of antioxidants in the bagasse or residues generated by the blackberry fruit processing [11, 13, 15]. Phenolic compounds and dietary fiber are generally studied separately, probably because of the differences in their chemical structures, physicochemical and biological properties, and metabolic pathways [16]. However, these are plant food constituents that are associated with many health benefits and have been demonstrated to reduce the risk of developing cancer and some chronic diseases [17, 18]. Dietary fiber has an essential role in intestinal health and appears to be significantly associated with a reduction of cholesterolemia and modification of the glycemic response [19].

Considering that most studies evaluate the whole fruit but not the residues that may have a great potential as a source of bioactive compounds (dietary fiber, antioxidants, fatty acids, etc.), the objective of this study was to determine the organic acids, antioxidants, and dietary fiber content of blackberry (*Rubus fruticosus*) cv. Tupy residues (comprised of seeds, peel, and pulp) and compare antioxidants and dietary fiber values with a commercial product.

2. Materials and Methods

2.1. Sample Preparation. Blackberries (*Rubus fruticosus* cv. Tupy) were collected from Atotonilco, Hidalgo, Mexico, in January 2016. Fruits without external injuries were selected and washed. Blackberries were processed into juice and the resulting residues (seeds, peel, and pulp) were collected as follows: fruit was stirred using an industrial blender (38BL52 LBC10, Waring Commercial, Torrington, CT, USA) and then passed through a conventional strainer; the retained bagasse (seeds and peel) was collected and mixed with the pulp (precipitate) obtained after juice was clarified by centrifugation (Allegra 25™, Beckman Coulter, Palo Alto, CA, USA) at 15,300g for 30 min at 4°C. All blackberry residues (BR) (bagasse plus pulp) were lyophilized (7753020, Labconco, Kansas City, MO, USA), milled, and sieved together to obtain a particle size of 500 µm. The samples were stored in sealed plastic bags at –30°C until further analysis. A prune (*Prunus domestica*)-based commercial product (CP) present on the market as tablets was milled and sieved under the same conditions described for blackberry residues. The CP was included in the present study only for comparison of antioxidants, dietary fiber, and color.

2.2. Chemical Composition. The chemical composition of the BR was determined according to the AOAC [20] for moisture (Official Methods 925.09), ash (Official Methods 923.03),

protein (Official Methods 991.20, conversion factor 6.25), ethereal extract (Official Methods 930.39), and AOAC [21] for total, soluble, and insoluble dietary fiber (TDF, SDF, and IDF, resp.). Total carbohydrates were calculated by difference of moisture, ash, protein, ethereal extract, and total dietary fiber values [22].

2.3. Quantitative Analysis of Mineral Composition. A field emission type scanning electron microscope (FE-SEM) (JEOL SSM 6300, Jeol de Mexico, CDMX, Mexico) with a 300,000 magnification range and a resolution of 30 kv was used. The BR sample was placed on a double-sided graphite tape and was then coated with a thin layer of gold (1 mm) using an ionizing machine (Denton Vacuum LLC, Moorestown, NJ, USA) at a pressure of 20 millitorr and a current of 20 mA for 4 min. Quantitative analysis of mineral composition was obtained by means of a spectrum that forms part of the scanning electron microscopy.

2.4. Quantification of Organic Acids. Individual organic acids (oxalic, malic, citric, fumaric, and ascorbic) were quantified by high-performance liquid chromatography (HPLC). Briefly, 20 mL of metaphosphoric acid at 4.5% (w/v) was added to 500 mg of the sample and the mixture was stirred with magnetic shaking (P-Selecta, Asincro, Spain) for 15 min under dark conditions. After the mixture was centrifuged (Allegra 25, Beckman Coulter, Palo Alto, CA, USA) at 9000 rpm for 15 min, the supernatant was diluted to a final volume of 25 mL with metaphosphoric acid 4.5%. Prior to injection into the HPLC, extracts were filtered through filter paper no. 1242 and then a 0.45 µm Millipore PVDF membrane [23]. The HPLC was equipped with an isocratic pump (model PU-II, Micron Analitica, Madrid, Spain), an AS-1555 automatic injector (Jasco, Tokyo, Japan), a Spherclone ODS(2)250×4.60 mm, 5 µm Phenomenex column (Torrance, CA, USA), and a UV-visible detector (Thermo Separation Spectra Series UV100, San Jose, CA, USA). The mobile phase was 1.8 mM H₂SO₄ at pH 2.6. UV detection at 215 nm and a flow rate of 0.4 mL/min were used for the analysis of organic acids, while a flow rate of 0.9 mL/min and UV detection at 245 nm were set for ascorbic acid. Linear calibration curves were obtained for quantification from stock solutions of all the identified compounds (oxalic, malic, citric, fumaric, and ascorbic acids). Data were analyzed using the Biochrom 2000 (version 3.0) software. Identification was made by comparing retention times of commercial pure standards, and quantification was based on the UV signal response, and the resultant peak areas in the chromatograms were plotted against concentrations obtained from standards (1.34, 1.40, 1.39, 1.45, and 25–100 mg/mL for oxalic, malic, citric, fumaric, and ascorbic acids, resp., in metaphosphoric acid 4.5%). Organic acid contents in blackberry residues were expressed in mg/100 g of dry basis (db).

2.5. Fatty Acid Profile. Blackberry residues were washed according to previous studies by Zafra-Rojas et al. [24] and Siriwoharn et al. [1] with minor modifications to simulate the industrial process. Briefly, 160 mL of deionized water was added to 10 g of sample and centrifuged at 10,000 rpm for

10 min; the supernatant was discarded and centrifugation repeated with 80 mL of deionized water; the resulting precipitate was washed under the same centrifuging conditions with methanol up to six washes until the supernatant was no longer red or was colorless.

Following the procedure suggested by Añorve-Morga et al. [25], 110 mL of chloroform-methanol (CHCl_3 :MeOH, 2:1 vol/vol) was added to the washed samples and vortexed for 5 minutes and stored at 4°C for 48 h. Subsequently, 500 μL of the aqueous phase containing CHCl_3 :MeOH was transferred to a tube and 1 mL of boron trifluoride-methanol (BF_3 :MeOH) (12.5:100, vol/vol) was added prior to heating the sample at 95°C in a water bath for 6 min [26]. After the mixture was allowed to cool and 1 mL of hexane plus 1 mL of hexane-saturated water was added, it was stirred and the aqueous phase was discarded. Two milliliters of hexane-saturated water was added, stirred, and reserved in a freezer until use. Prior to analysis, the organic phase was evaporated and the fatty acids were taken up in hexane. FAME (methyl esters of fatty acids) were analyzed and identified by comparing their retention times to standards of known amounts (FAME Mix C4-C24 Supelco® by Sigma-Aldrich, St. Louis, MO, USA).

For analysis of the fatty acids, a gas chromatograph (PerkinElmer, Australia) was used: AutoSystem XL equipped with a flame ionization detector (FID) and a Supelco® SP TM-2560 capillary column (75 m \times 0.18 mm \times 0.14 μm) using the splitless injection mode (1 μL). The initial column temperature was set at 150°C, increased at a rate of 4°C/min to 214°C, held for 2 min, and then increased to 244°C at 2.5°C/min and held for 5 min. The injector and detector temperatures were 230°C and 250°C, respectively. The ramp split was approximately 1 μL and nitrogen was used as a carrier gas at a constant flow of 1 mL/min.

2.6. Antioxidants Extraction. Two hundred milligrams of sample (BR and CP) was placed in a capped centrifuge tube; 10 mL of acidic methanol/water (50:50, v/v) was added and the tube was thoroughly shaken at room temperature for 1 h. The mixture was centrifuged at 3400 rpm for 20 min and the supernatant was recovered. Ten milliliters of acetone/water (70:30, v/v) was added to the residue and the shaking and centrifugation were repeated. Methanolic and acetone extracts were combined and used to determine the antioxidant capacity associated with extractable antioxidants [27].

2.6.1. Determination of Total Phenolic Content (TPC). Total phenolic content was determined by the Folin-Ciocalteu procedure [28]. Briefly, 100 μL of the sample was mixed with 500 μL of 1:10 diluted Folin-Ciocalteu reagent. Then, 400 μL (7.5%) of sodium carbonate was added and the mixture was incubated for 30 min at room temperature. The absorbance of the mixture was measured at 765 nm in the microplate reader (Power Wave XS UV-Biotek, software Gen5 2.09, Winooski, VT, USA). The standard curve was developed with concentrations of 0, 100, 200, and 300 mg of gallic acid/L. Gallic acid was used as reference standard and the results were expressed as mg of gallic acid equivalents per 100 grams

of dry basis (mg GAE/100 g db) of blackberry residues and commercial product.

2.6.2. Determination of Anthocyanins. The anthocyanin content was determined through the differential pH method [29]. Two buffer solutions were prepared: 0.025 M potassium chloride at pH 1.0 and 0.4 M sodium acetate at pH 4.5. The pH was adjusted by adding concentrated HCl. The extracts of blackberry residues and prune (commercial product) were diluted in these two buffer solutions. After 15 min under dark conditions at room temperature, absorbance was measured at 510 and 700 nm using a microplate reader (Power Wave XS UV-Biotek, software Gen5 2.09, Winooski, VT, USA). The absorbance of the anthocyanins was calculated according to

$$\text{Abs} = (\text{Abs}_{510} - \text{Abs}_{700}) \text{pH}_{1.0} - (\text{Abs}_{510} - \text{Abs}_{700}) \text{pH}_{4.5} \quad (1)$$

The concentration of anthocyanins in the extracts was calculated according to

$$\text{Anthocyanins (mg/L)} = \frac{(\text{Abs} \times \text{MW} \times \text{DF} \times 1000)}{\epsilon \times 0.52}, \quad (2)$$

where Abs is the absorbance, MW is the molecular weight, DF is the dilution factor, and ϵ is the molar absorptivity, considering the molar absorptivity (ϵ) of 26,900 $\text{L mol}^{-1} \text{cm}^{-1}$, the molecular weight of 449.2 g/mol for cyanidin-3-glucoside, and the path length (0.52 cm) of the well. The results were expressed as mg of cyanidin-3-glucoside equivalent/100 grams on a dry basis (mg cy-3-gl/100 g db).

2.7. Antioxidant Capacity

2.7.1. ABTS Method. The radical cation (ABTS^{*+}) was produced by reacting 7 mM ABTS stock solution with 2.45 mM potassium persulfate under dark conditions at room temperature for 16 h before use. The ABTS solution was diluted with deionized water until an absorbance of 0.70 ± 0.10 at 754 nm. An aliquot of 20 μL of the extract was added to 980 μL of diluted ABTS solution, and absorbance readings were taken after incubation for 7 min at room temperature. The absorbance of the mixture was measured at 754 nm in a microplate reader (Power Wave XS UV-Biotek, software Gen5 2.09, Winooski, VT, USA). The standard curve was linear with concentrations of 0, 10, 20, 30, 40, and 50 mg of ascorbic acid/L [30]. The antioxidant capacity of blackberry residues and commercial product (prune) was expressed as milligrams of ascorbic acid equivalents per 100 grams of dry basis (mg AAE/100 g db).

2.7.2. DPPH Method. Antiradical activity was measured with 1,1-diphenyl-2-picrylhydrazyl radical (DPPH) [31]. An ethanolic solution (7.4 mg/100 mL) of the stable DPPH radical was prepared. Then, 100 μL of the extract was taken into vials and 500 μL of the DPPH solution was added, and the mixture was left to stand for 1 h at room temperature. Finally, absorbance was measured at 520 nm using a microplate reader (Power Wave XS UV-Biotek, software Gen5 2.09,

Winooski, VT, USA). The standard curve was developed with concentrations of 0, 50, 100, 200, and 300 μmol of Trolox. Free-radical scavenging activity was expressed as μmol of Trolox equivalents per 100 g of dry basis ($\mu\text{mol TE}/100 \text{ g db}$) of blackberry residues and commercial product.

2.7.3. Ferric Reducing Antioxidant Power (FRAP). The FRAP was assayed according to Benzie and Strain [32] with some modifications. The stock solutions included 300 mM acetate buffer (3.1 g $\text{C}_2\text{H}_3\text{NaO}_2 \cdot 3\text{H}_2\text{O}$ and 16 mL $\text{C}_2\text{H}_4\text{O}_2$) (pH 3.6), 10 mM TPTZ (2,4,6-tripyridyl-s-triazine) solution in 40 mM HCl, and 20 mM $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ solution. The fresh solution prepared by mixing 25 mL acetate buffer, 2.5 mL TPTZ solution, and 2.5 mL $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ solution was warmed at 37°C before use. The sample (30 μL) was mixed with 90 μL of distilled water and 900 μL of the FRAP solution for 10 min in the dark. Absorbance was measured at 593 nm using a microplate reader (Power Wave XS UV-Biotek, software Gen5 2.09, Winooski, VT, USA). The standard curve was developed with concentrations of 0, 200, 400, 600, 800, and 1000 μmol ferrous sulfate (FeSO_4) 5 M, and the results were expressed as $\mu\text{mol Fe(II)}$ per 100 g dry basis ($\mu\text{mol Fe(II)}/100 \text{ g db}$) of blackberry residues and commercial product.

2.8. Properties of Dietary Fiber

2.8.1. Water Retention Capacity (WRC). An aliquot (0.5 g) of BR and CP was placed in 15 mL centrifuge tubes and 10 mL of distilled water was added. The tubes were shaken manually for 10 minutes and then allowed to stand for 24 h at room temperature prior to centrifugation (Hamilton Bell VanGuard V6500, New Jersey, USA) at 3400 rpm for 20 minutes. Immediately, the supernatant was removed and the pellet weighed. The water retention capacity was measured as the amount of water retained by the sample (g/g dry basis) [33, 34] and was calculated by

$$\text{WRC (g/g)} = \frac{(\text{WFS} - \text{WDS})}{\text{WDS}}, \quad (3)$$

where WFS is the weight of the fresh sample (g) and WDS is the weight of the dry sample (g).

2.8.2. Swelling Capacity (SC). The lyophilized samples (200 mg) were placed in a graduated cylinder (0.1 mL) and the volume occupied (V_0) (mL) was measured. Then, 10 mL of distilled water was added and the mixture was stirred manually for 5 min and allowed to stand for 24 hours at room temperature. The final volume of the samples (V_1) (mL) was measured [33, 35] and the swelling capacity of BR and CP (mL/g) was calculated using

$$\text{SC (mL/g)} = \frac{(V_1 - V_0)}{\text{sw}}, \quad (4)$$

where V_1 is the final volume (mL), V_0 is the volume occupied by the lyophilized sample (mL), and sw is the sample weight (g).

2.8.3. Fat Absorption Capacity (FAC). Five hundred milligrams (WDS) of BR and CP was placed in separated 15 mL centrifuge tubes and 10 mL of sunflower seed oil was added. The mixtures were shaken manually for 10 minutes and then allowed to stand for 24 h at room temperature. Samples were centrifuged at 3400 rpm (Hamilton Bell VanGuard V6500, New Jersey, USA) for 20 minutes and immediately the supernatant was removed and the pellet weighed (WFS). The fat absorption capacity was measured as the amount of fat retained by the sample (mL/g dry basis) [33, 34] and was calculated using

$$\text{FAC (mL/g)} = \frac{(\text{WFS} - \text{WDS})}{\text{WDS}}, \quad (5)$$

where WFS is the weight of the fresh sample (g) and WDS is the weight of the dry sample (g).

2.9. Color Measurement. Color of BR and CP was measured using a colorimeter (Konica Minolta CM-608d, Sensing, Inc., Japan) set with the D65 illuminant and 10° observer angle. The CIE-Lab values were recorded, where L^* indicates lightness ($L = 0$ or 100 indicates black or white, resp.); a^* is the axis of chromaticity between green (–) and red (+), and b^* is the axis between blue (–) and yellow (+). Numerical values of L^* , a^* , and b^* were used to obtain chroma ($C = [a^2 + b^2]^{1/2}$) and Hue angle (h°) ($h^\circ = \text{tg}^{-1}(b/a)$) [36].

2.10. Statistical Analysis. All experiments were carried out in triplicate and expressed as mean \pm standard deviation (SD). The variables used to compare the blackberry residues with commercial product were analyzed by a *t*-test and a confidence level of 95% using the SPSS® System for WIN™ version 15.0.

3. Results and Discussion

3.1. Chemical Composition, Minerals, and Organic Acids. Table 1 shows the chemical composition, mineral content, and organic acids of blackberry residues. The BR had a moisture content of $74.83 \pm 0.76\%$ and could be described as a semisolid fibrous product from which the liquid has been extracted during juice production. Other fruit residues have similar moisture percentages, such as blackberry (*Rubus* sp.), mango (*Mangifera indica*), and guava (*Psidium guajava*) residues (values between 70 and 90%) [11, 37].

Protein ($2.60 \pm 0.11\%$) and lipid ($1.84 \pm 0.03\%$) content in BR was higher than values reported for orange, guava, soursop, and mango residues, which ranged from 0.6 to 1.9% and from 0.25 to 0.48%, respectively [37]. Differences could be attributed to the high seed content in BR and the fact that these are mainly composed of oils [38]. Carbohydrates ($9.07 \pm 0.80\%$) and total dietary fiber ($11.12 \pm 0.22\%$) were the major components after moisture, while ashes were minor constituents ($0.54 \pm 0.02\%$), although they were higher than ash percentages reported for blackberries bagasse by Pasquel Reátegui et al. [11].

The minerals found in BR were sodium, magnesium, potassium, calcium, iron, sulfur, silicon, and chlorine (Table 1).

TABLE 1: Chemical composition, analysis of minerals content, and organic acids of blackberry residues.

| Blackberry residues ^a | |
|----------------------------------|------------------|
| Composition (%) | |
| Moisture | 74.83 ± 0.76 |
| Protein | 2.60 ± 0.11 |
| Lipids | 1.84 ± 0.03 |
| Total carbohydrates | 9.07 ± 0.80 |
| Total dietary fiber | 11.12 ± 0.22 |
| Ash | 0.54 ± 0.02 |
| Element (%) | |
| C | 48.02 ± 4.69 |
| N | 7.54 ± 0.43 |
| O | 43.04 ± 4.16 |
| P | 0.19 ± 0.08 |
| Na | 0.01 ± 0.01 |
| Mg | 0.11 ± 0.05 |
| K | 0.81 ± 0.17 |
| Ca | 0.08 ± 0.04 |
| Fe | 0.03 ± 0.05 |
| Cu | - |
| Zn | - |
| Mn | - |
| S | 0.05 ± 0.01 |
| Si | 0.02 ± 0.04 |
| Cl | 0.06 ± 0.05 |
| Organic acids (mg/100 g db) | |
| Oxalic acid | 59.51 ± 8.18 |
| Malic acid | 5706.37 ± 123.38 |
| Citric acid | 125.54 ± 0.61 |
| Fumaric acid | 230.25 ± 2.47 |
| Ascorbic acid | 6.00 ± 1.00 |

^aMean ± standard deviation ($n = 3$).

In other blackberry studies, in addition to the mentioned minerals, copper, zinc, manganese, and selenium were also found [39–41]. These differences may be mainly attributed to the type of soil in which these fruits were cultivated; for instance, the soil of Atotonilco, Hidalgo, Mexico (where the blackberry was harvested) consists of layers of volcanic ash and pumice and a conglomerate of gravel and sand characterized by the presence of potassium and other minerals in low concentrations [42].

Many fruits accumulate organic acids in their pulp and peel at certain stages of their development [43, 44]. In the BR, ascorbic acid content was low (6.00 ± 1.00 mg/100 g db) while malic acid was in higher concentrations (5706.37 ± 123.38 mg/100 g db) (Table 1) as has been reported for several fruits such as strawberry tree (*Arbutus unedo* L., Ericaceae), blackberry (*Rubus ulmifolius* Schott), and red apples (*Malus domestica* Borkh.) [45–47]. Evidence suggests that organic acids such as malic or citric acids may have a positive health benefit as antioxidants thanks to their ability to chelate metals [48]. On the other hand, environmental factors and

TABLE 2: Fatty acids profile of blackberry residues (mg/100 g).

| | |
|---------------------------|-------------|
| Palmitic acid (C16:0) | 1.68 ± 0.40 |
| Linolenic acid (C18:3n-3) | 0.43 ± 0.07 |
| Arachidic acid (C20:0) | 7.68 ± 1.88 |
| Eicosenoic acid (C20:1) | 0.20 ± 0.00 |

Mean ± standard deviation ($n = 2$).

cultivation practices (e.g., temperature, cultivar, minerals, and water availability) could affect the content of these organic acids in fruits [43].

3.2. Fatty Acids Profile. Table 2 shows the fatty acids profile of BR. Four fatty acids were found and corresponded to 9.99 mg/100 g of blackberry residues. The saturated palmitic (C16:0) and arachidic (C20:0) acids were found in higher quantities as compared to the polyunsaturated and monounsaturated linolenic (C18:3n-3) and eicosenoic (C20:1) acids. These same fatty acids have been found in oil seeds in different species of blackberries (*Rubus ursinus* var. Marion and *Rubus laciniatus* Willd. var. Evergreen, *Rubus ulmifolius* var. Schott, and *Rubus fruticosus*) [38, 49, 50]. However, the amount found in the BR was lower than the values reported in these studies probably due to the presence of peel and pulp in addition to seeds so that the final concentration was affected.

3.3. Total Phenolic Content and Anthocyanins. The whole blackberry fruit is a good source of antioxidant compounds and has considerable levels of phenolic compounds, including anthocyanins. In blackberry (*Rubus fruticosus* L. var. Caingangue) residues such as peel and seeds, high concentrations (366 to 736 mg GAE/100 g fresh residues) have also been reported [15]. The BR exhibited significantly higher total phenolic content ($p < 0.05$) and anthocyanins in comparison with the commercial product in which anthocyanins were absent (Table 3), probably due to complete degradation during the transformation of fresh plums of prunes [51]. The phenolic content of the BR was higher than that reported by Huang et al. [52] for whole rabbiteye blueberry fruits (*Vaccinium ashei* cv. Brightwell) including peel, seeds, and pulp, for thornless blackberry (*Rubus laciniatus* cv. Hull), and for strawberry (*Fragaria × ananassa* cv. Toyonoka). The values for anthocyanins in the present study were similar to those described by Dai et al. [53] for Hull blackberry puree (534 mg/100 g db).

Patras et al. [54] evaluated TPC in strawberry (*Fragaria × ananassa* cv. EI Santa) and blackberry (*Rubus fruticosus* cv. Loch Ness) puree, and the amount of phenols (855 and 1694 mg GAE/100 g db, resp.) was lower than the value obtained for our BR. Differences in the total concentration of phenols in vegetables could be attributed to variations in the composition of the food matrix, geographical origin, cultivar/genotype, maturity, time and type of drying, and storage time [55]. Other factors that affect the recovery of phenolic compounds are extraction methods and conditions such as extraction temperature, time, particle size, and type of solvent [11, 56, 57].

TABLE 3: Total phenolic content, anthocyanins, and antioxidant capacity of blackberry residues^a and commercial product.

| | BR ^a | Commercial product |
|-----------------------------|--------------------|--------------------|
| Parameters | | |
| TPC (mg GAE/100 g db) | 4016.43 ± 13.44* | 1362.98 ± 52.84 |
| Anthocyanins (mg/100 g db) | 364.53 ± 7.36* | 0.00 ± 0.00 |
| Antioxidant capacity | | |
| ABTS (mg AAE/100 g db) | 5422.38 ± 71.50* | 267.06 ± 13.86 |
| DPPH (μmol TE/100 g db) | 13656.27 ± 532.66* | 16655.88 ± 272.02 |
| FRAP (μmol Fe(II)/100 g db) | 12511.44 ± 147.39* | 2842.23 ± 109.41 |

Mean ± standard deviation ($n = 3$). * indicates significant difference ($p < 0.05$) between samples.

TABLE 4: Dietary fiber properties of blackberry residues and commercial product.

| | BR ^a | Commercial product ^b |
|--------------------------------|-----------------|---------------------------------|
| Parameters | | |
| Total dietary fiber (%) | 44.26 ± 0.09* | 8.06 ± 0.12 |
| Soluble dietary fiber (%) | 5.90 ± 0.52* | 1.96 ± 0.08 |
| Insoluble dietary fiber (%) | 38.35 ± 0.39* | 6.09 ± 0.29 |
| Properties | | |
| Water retention capacity (g/g) | 2.94 ± 0.10* | 0.00 ± 0.00 |
| Swelling capacity (mL/g) | 5.00 ± 0.00* | 0.00 ± 0.00 |
| Fat absorption capacity (mL/g) | 1.98 ± 0.03* | 1.49 ± 0.02 |

^aBR: blackberry residues. ^bPrune. * indicates significant difference ($p < 0.05$) between samples.

3.4. Antioxidant Capacity. BR presented high antioxidant capacity by ABTS and FRAP, but low by DPPH ($p < 0.05$) compared with the CP (Table 3). The antioxidant capacity by ABTS and the iron reduction ability of the BR were 20 and 4 times, respectively, those of the CP which in contrast had a DPPH value 1.2 times that of the BR. The reported antioxidant capacity of fruit residues such as pulp, peel, and seeds from strawberry (*Fragaria vesca* L.), carrot (*Daucus carota* L. spp. *sativus* var. *atrurubens* Alef.), and blueberry (202 mg VCEAC/100 g, 10190 μmol TE/100 g db, and 10400 μmol Fe(II)/100 g db, for ABTS, DPPH, and FRAP, resp.) [30, 58, 59] was lower than the values found for BR in the present study. The responsible compounds for the antioxidant activity are mainly phenols [60], but some pigments such as anthocyanins also contribute to the amount of phenolic compounds. The basic structural orientation of these compounds determines their antioxidant activity since the hydroxyl group has a higher ability to donate hydrogen atoms to free radicals [61]. Moreover, the position of the hydroxyl groups appears to be more important than their number to act as antioxidants [62]. Phenolic compounds also contribute by transferring an electron to reduce the ferric ion (III) to its ferrous (II) form [60]. But the presence of lipids as tocopherols that are found in seeds [38] may impact the overall antioxidant activity of the evaluated BR that does contain seeds.

3.5. Dietary Fiber and Functional Properties. It is well known that agroindustrial waste is rich in dietary fiber. Dietary fiber contains significant amounts of antioxidant compounds and other substances with positive effects on health [63].

Total, soluble, and insoluble dietary fiber and functional properties of BR and CP are described in Table 4. In general, the percentage of total dietary fiber and their properties was higher in BR compared with CP. The analyzed functional properties such as WRC, SC, and FAC were significantly higher ($p < 0.05$) in BR (2.94 ± 0.10 g/g, 5.00 ± 0.00 mL/g, and 1.98 ± 0.03 mL/g, resp.). The total fiber content of the BR was similar to the one described for grapefruit fiber (44 g/100 g) but SC was higher than that reported for oat and apples fibers (2.3 and 3.4 mL/g, resp.) [64, 65]. In terms of WRC (9.7 g/g), our values were lower than those reported by Basanta et al. [66] who evaluated cherry fibers. The content of soluble dietary fiber affects SC, and insoluble dietary fiber determines the WRC and FAC [67]; hence, the properties of dietary fiber depend on the proportion of SDF and IDF. Since the BR contains IDF, it may help to ease constipation, increase excretion, and bind bile salts, cholesterol, and carcinogenic compounds allowing their excretion through the feces [68, 69]. Intake of IDF may also prevent conditions such as colon cancer, hypercholesterolemia, and intoxication. The BR also exhibited a high capacity to increase its volume in excess of water, which may contribute to enhancing satiety thanks to its content of SDF [64, 70].

3.6. Color Parameters. Color is an important quality attribute because it can condition consumer acceptability. Table 5 shows the color parameters for BR and CP. The luminosity (L^*) for both samples was significantly different ($p < 0.05$); BR presented low values, and according to the a^* and b^* axes, the BR and CP were found in the reddish quadrant. The Hue angle can be between 0° and 90°, representing the red (0°)

TABLE 5: Color parameters of blackberry residues and commercial product.

| Color parameters | BR ^a | Commercial product ^b |
|-----------------------|-----------------|---------------------------------|
| <i>L</i> [*] | 16.73 ± 0.00* | 37.97 ± 0.68 |
| <i>a</i> [*] | 16.77 ± 0.03* | 2.49 ± 0.08 |
| <i>b</i> [*] | 10.88 ± 0.00 | 11.38 ± 0.35 |
| <i>h</i> [°] | 32.98 ± 0.06* | 77.64 ± 0.63 |
| Chroma | 20.00 ± 0.02* | 11.65 ± 0.33 |

^aBR: blackberry residues. ^bPrune. * indicates significant difference ($p < 0.05$) between samples.

and orange-yellow (90°) colors; based on this value, the BR is within the red-orange tone, and the CP exhibited a yellow tone.

Color saturation (chroma) was between 0 and 60 and results showed that the CP had lower saturation than the BR [71, 72]. Malien-Aubert et al. [73] evaluated the color of some commercial extracts from grapes (*Vitis vinifera*), elderberry (*Sambucus nigra*), purple carrot (*Daucus carota*), red radish (*Raphanus sativus*), black currant (*Ribes nigrum*), red cabbage (*Brassica oleracea*), and chokeberry (*Aronia melanocarpa*); the chroma values from these two last sources were close to the values obtained for the BR (around 20). Red, blue, and purple colors in fruits depend on the anthocyanins and also the number and orientation of the hydroxyl and methoxyl groups of the molecule; for example, an increase in hydroxylation produces a shift to blue tones whereas an increase in methoxylation generates red coloration [74]. This suggests that the BR may contain a high amount of methoxyl groups. According to the European Union, all dyes derived from anthocyanins are recognized as natural dyes under the E163 classification, ranging from E163a (cyanidin: red food coloring) to E165f (petunidin: dark red food coloring) [72]. Therefore, based on our results, the BR could be a good source of natural dyes thanks to the presence of anthocyanins.

4. Conclusions

The results demonstrated that blackberry residues are a good source of organic acids and contain elevated amounts of phenolic compounds and anthocyanins and antioxidant capacity compared to the prune-based commercial product. These residues also exhibited better dietary fiber functional properties and a rich reddish color which make them a potential source of components with a beneficial role in human health, particularly for patients with chronic noncommunicable diseases.

Conflicts of Interest

The authors declare that there are no conflicts of interest.

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