Research Article

Phytochemical Characterization, *In Vitro* Antioxidant Activity, and Quantitative Analysis by Micellar Electrokinetic Chromatography of Hawthorn (*Crataegus pubescens*) Fruit

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Received 31 January 2018; Revised 1 June 2018; Accepted 25 June 2018; Published 25 July 2018

Academic Editor: Müberra Koşar

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Due to their antioxidant properties, polyphenolic compounds are considered beneficial for human health. In this work, we investigated the polyphenol profile and antioxidant activity of edible tejocote (*Crataegus pubescens*) fruit extracts by micellar electrokinetic chromatography (MEKC) and HPLC/UV. The major phenolic compounds in the pulp extracts were (+)-catechin (9.17 ± 0.20 mg/100 mg dry fruit), (–)-epicatechin (4.32 ± 0.11 mg/100 mg dry fruit), and chlorogenic acid (5.60 ± 0.24 mg/100 mg dry fruit). The total phenolic content was 168.6 ± 0.9 mg gallic acid equivalent/g dry fruit; the total proanthocyanidin content was 84.6 ± 1.4 mg cyanidin/100 g dry fruit; and the total flavonoid content was 55.89 ± 1.43 mg quercetin/g dry fruit. Interestingly, procyanidins (dimers, trimers, and tetramers of (–)-epicatechin and (+)-catechin) were detected in the extract. This is the first study reporting the presence of polymeric polyphenols in *Crataegus pubescens* fruit. Accordingly, these fruits demonstrate great potential as a natural source of antioxidant phenolic compounds and could therefore be used as a nutraceutical and functional food.

1. Introduction

The *Crataegus* genus has been recognized as a medicinal and ornamental plant tree, comprising approximately 1000 species worldwide [1, 2]. The genus *Crataegus* belongs to the Rosaceae family and is widespread in the Northern Hemisphere, distributed mainly in Asia, Europe, and North America [1]. In Mexico, hawthorn (*C. pubescens*) is known as “tejocote,” which is a native species with reports of its edible use in native populations and in traditional medicine; however, the components involved in its biological activities have not been described, and its use has been limited mainly to making jelly and jam due to the high content of pectin [3, 4], with limited agroindustrial applications [5]. Several studies of *Crataegus* spp. worldwide have demonstrated their potential as a source of valuable phytochemical compounds

[1, 2, 6–8]. Among the antioxidant phytochemicals reported in the extracts from the genus *Crataegus* are procyanidins, flavonoids, flavonols, glycosylated flavanones, and triterpene pentacyclic acids [6, 9–11], all considered as important therapeutic options against hypertension, angina pectoris, heart arrhythmia, and the first stages of congestive heart failure. Typically, procyanidins and triterpene pentacyclic acids are most abundant in fruits [6, 9, 11], and flavonoids are most abundant in leaves [10, 11]. However, for *C. pubescens*, information is limited, and it is mainly focused on the extraction and characterization of pectin [12, 13], leaving aside the importance of phytochemical compounds as phenolic antioxidant compounds that play a significant role in preventing chronic diseases by reducing the oxidative damage caused by highly reactive oxygen species [14–16]. The use of several *Crataegus* species in traditional medicine

has motivated the scientific study of some of these species [4]. The determination of the chemical composition and antioxidant activity of some endemic species of this genus could lead to identifying potentially active compounds [17] or including new species as possible nutraceuticals. The interest in these products increases when they are obtained from unconventional crops, such as tejocote (*C. pubescens*) [7]. Thus, the aim of this work was to investigate the potential of tejocote (*C. pubescens*) fruits as an alternative source to the *Crataegus* originating from Asia and Europe to obtain bioactive compounds and to contribute to the identification of the compounds involved in the pharmacological activities empirically reported for this native species from Mexico. The profile of the polyphenolic compounds in *C. pubescens* was studied using micellar electrokinetic chromatography (MEKC), HPLC/UV, and ESI-MS/MS to identify and quantify the main phenolic compounds. The antioxidant activity was evaluated using the DPPH (2,2-diphenyl-1-picrylhydrazyl) method, and the trapping capability of superoxide radical was evaluated by the xanthine-xanthine oxidase system.

2. Materials and Methods

2.1. Plant Material. The tejocote (*C. pubescens*) fruits were from the 2011 harvest (purchased at Central Market, Mexico City). The selected fruits were those with a yellow epidermis, diameter between 2.1 and 3.2 cm and weight between 9 and 16 g, discarding those with obvious mechanical damage. The manually selected fruits were washed and kept at -20°C under a nitrogen atmosphere.

2.2. Phenolic Extract Preparation. Extracts were obtained as reported by Pérez-Jiménez et al. [18], with some modifications. Briefly, fragments of freeze-dried pulp fruit (1 g) were mixed with 50 mL of methanol-water (1 : 1 v/v) for 1 h. The supernatant was separated and reserved, and the solid residue was blended for 1 h with acetone-water (7 : 3 v/v) at room temperature. The mixture was centrifuged at 3000 g for 15 min, the pellet was discarded, and the supernatant was mixed with the first supernatant and concentrated with a rotary evaporator to lyophilize the sample. The lyophilized extract was resuspended in 10 mL of methanol and kept at -20°C .

2.3. Characterization of the Phenolic Extract. The phenolic extract from the freeze-dried fruits of tejocote was phytochemically characterized for its quality, yield, and chemical composition before investigation of its antioxidant activity. Due to the extraction procedure, the carbohydrate content and the phenolic profiles were used as quality and integrity criteria of the extracts, following the suggestion of Dai and Mumper [19].

2.3.1. Total Carbohydrate Content. The total carbohydrate content of the phenolic extract was determined by the phenol-sulfuric acid method [20]. Briefly, 1 mL of sample

was added to 0.6 mL of phenol (5%) and mixed with 3.6 mL of sulfuric acid. The mixture was cooled for 30 min, and the absorbance was measured at 480 nm. A standard curve using glucose (10–100 mg/L) was generated.

2.3.2. Total Phenolic Content. The total phenolic content of the extracts was estimated as reported by Pastrana-Bonilla et al. [21]. Briefly, 100 μL of extract was mixed with 100 μL of deionized water, 1 mL of Folin-Ciocalteu reagent (2 M), and 0.8 mL of sodium carbonate (7.5%). The sample was vortex-mixed for 5 seconds and incubated for 30 minutes in the dark, and the absorbance was measured at 760 nm. The results were expressed as the mean $\pm$ SD ($n = 3$) in mg equivalent to gallic acid/g fruit.

2.3.3. Total Flavonoid Content. The flavonoids content was estimated by a colorimetric method based on aluminum chloride as reported by Vongsak et al. [22] and expressed as the mean $\pm$ SD ($n = 3$) in mg equivalent to quercetin/g fruit.

2.3.4. Proanthocyanidin Content. The total proanthocyanidin content was estimated using a spectrophotometric method based on acid hydrolysis of the sample with HCl/n-butanol [23, 24], with some modifications. Briefly, 5 mg of phenolic extract was resuspended in 1 mL of methanol. Samples of 0.25 mL were added to 3 mL of n-butanol-HCl (95/5, v/v) in screw-cap tubes. Then, 0.1 mL of $\text{NH}_4\text{Fe}(\text{SO}_4)_2 \cdot 12 \text{H}_2\text{O}$ in HCl 2 M (0.2% m/v) was added, and the tubes were incubated for 40 min at 95°C . The mixture was cooled to room temperature and the absorbance measured at 540 nm. A standard curve using cyanidin chloride in methanol (0–10 mg/L) was performed. The results were expressed as the mean $\pm$ SD ($n = 3$) in mg equivalent to cyanidin chloride/100 g fruit.

2.4. Phenolic Profile Determination. Due to the complexity of the phenolic extracts, three methods were used for the assay and quantification of their chemical constituents.

2.4.1. HPLC/UV Analysis. The HPLC/UV analysis of the phenolic profile was performed with acetonitrile and trifluoroacetic acid (TFA) as the mobile phase as reported by Martínez-Juarez et al. [25], using an HPLC system with a 20 μL loop and a Spectra System UV-visible optical scanning detector (Thermo Separation Products Inc.) that allows the UV/VIS detection of all solutes eluting from the HPLC column. The detector signals were captured and processed by the PC1000 system software (Thermo Separation Products Inc.).

2.4.2. ESI-MS/MS Analysis. MS/MS analysis was performed using a 3200 QTRAP system (Applied Biosystems/MDS, USA). The ionization was performed by electrospray (ESI-MS) with a collision energy of 50 V. The ions were detected in positive mode.

2.4.3. Micellar Electrokinetic Chromatography (MEKC) Analysis. The analysis was performed as reported by Lee et al. [26] using a P/ACE™ MDQ Capillary Electrophoresis system (Beckman Coulter™) with a photodiode array detector and a fused silica capillary (50 cm effective length and 60 cm total length) with 75 μm ID. The assay conditions were adjusted as required by the standards ((-)-epicatechin, (+)-catechin, and chlorogenic acid) in ethanol. The final conditions for good resolution were as follows: hydrodynamic injection (4 seconds, 0.5 psi), 24 kV for 15 minutes, capillary temperature at 22°C, buffer (28 mM $\text{Na}_2\text{B}_4\text{O}_7$, 3.2 mM KH_2PO_4 , and 24 mM SDS, pH 8.8), and a resultant current of ~80 amperes. The spectral data were evaluated from 190 to 360 nm, and the wavelength for integration and quantification was 217 nm.

2.5. Antioxidant Activity

2.5.1. DPPH Activity. The antioxidant activity of the extracts was evaluated through the DPPH $\cdot$ (1,1-diphenyl-2-picrylhydrazyl) radical scavenging assay as described by Sánchez-Moreno et al. [27]. The reaction was performed using 3.9 mL of DPPH $\cdot$ in methanol (0.025 g/L) and 0.1 mL of the phenolic extracts diluted at several concentrations. The absorbance was read at 515 nm. The remaining quantity of DPPH $\cdot$ (% remaining DPPH $\cdot$) was defined as follows: $100 \times (\text{absorbance of the sample} / \text{absorbance of the control})$.

2.5.2. Antiradical Efficiency (AE). The antiradical efficiency (AE) or the trapping capability of free radicals by the phenolic extract was evaluated as suggested by Sánchez-Moreno et al. [27]. The AE was calculated by the mathematical expression $1/(\text{EC}_{50} \text{ TEC}_{50})$. EC_{50} is the amount of phenolic extract necessary to decrease by 50% the initial DPPH $\cdot$ concentration, and it was extrapolated from the curve of the remaining DPPH $\cdot$ against the concentrations for each phenolic extract. On the other hand, TEC_{50} is the reaction time to reach a steady state at the concentration of the phenolic extract corresponding to the EC_{50} value, and it was determined graphically by plotting the times at the steady state against the concentration for each phenolic extract.

2.5.3. Neutralizer Activity Superoxide Radical. The analysis of the trapping capability of the superoxide radical of the phenolic extracts was performed as described by Aruoma et al. [28]. The superoxide radical was generated by using a xanthine-xanthine oxidase as described by McCord and Fridovich [29]. The reaction mixture was as follows: 2 mL of buffer KH_2PO_4 -KOH (100 mM, pH 7.4), 25 μL of xanthine (20 mM), 50 μL of EDTA (0.3 mM), 25 μL of Nitro blue tetrazolium (NBT) (3 mM), 100 μL of sample (changeable phenolic concentration), and finally, 75 μL of xanthine oxidase diluted in the above phosphate buffer (1 unit/mL). The absorbance was measured continuously at 560 nm in a closed 1 cm quartz cell at 25°C for 20 minutes. A control sample was prepared using deionized water instead of the phenolic extracts. The trapping activity of the superoxide

radical of each phenolic extract was expressed as EC_{50} and as the inhibition percentage of the NBT reduction (% inhibition) and calculated using the relation: $100 \times (\text{absorbance control} - \text{absorbance sample}) / (\text{absorbance control})$.

2.5.4. Statistical Analyses. For the quantification of polyphenols by MEKC, the data obtained from the calibration curves were adjusted by linear regression with coefficients of variation less than 3% and a value of R^2 greater than 0.9900. Data were plotted using SigmaPlot (version 12.3 Systat Software, Inc., San Jose, California). All experiments were done in triplicate, and the results are reported as the mean $\pm$ S.D.

3. Results and Discussion

3.1. Characterization of the Phenolic Extract. Since quality, yield, and chemical composition of plant extracts depends on the plant material and the extraction processes, the phenolic extracts from the freeze-dried fruits of tejocote (*C. pubescens*) were phytochemically characterized for their purity and phenolic profile before investigation of their antioxidant activity (Table 1). Due to the polarity of the solvents used, the carbohydrate content and the phenolic profiles were used as quality and integrity criteria of the extract as suggested by Dai and Mumper [19]. The content of total carbohydrates in the phenolic extracts was $69.8\% \pm 1.61\%$, which agrees with reports from the literature showing that this method can simultaneously extract carbohydrates, terpene, and polyphenolic compounds from freeze-dried plant biomass [18, 19]. However, when the extracts are from air-dried biomass, the yield of phenolics is usually lower, and further, the natural phenolic profile may change by side reactions during the drying and extraction processes [19, 30, 31]. On the other hand, the total phenolic content in the extract was 168.6 ± 0.9 mg gallic acid equivalent/g dry fruit, which is higher than the value reported for callus tissue of *Crataegus monogyna* (58.9 mg gallic acid equivalent/g dry fruit) by Baborun et al. [32] and for fruits of *Crataegus pinnatifida* (42.89 mg gallic acid equivalent/g dry fruit) by Cai et al. [33]. However, it is also important to note that the Folin-Ciocalteu method is not an absolute measurement of the amount of phenolic compounds since other substances such as organic acids, sugars, amino acids, proteins, and other compounds present in the extracts may interfere with this assay [34]. Of phenolic compounds, flavonoids have been reported as the most important and principal compounds responsible for the major pharmacological activities of the *Crataegus* species [35]. For example, Swaminathan et al. [36] reported a cardioprotective and antiradical effect of phenolic extracts from *Crataegus oxyacantha* fruits. In the present study, the total content of flavonoids in the phenolic extract from tejocote (*C. pubescens*) fruits was 55.89 ± 0.11 mg equivalents quercetin/g dry fruit (Table 1). This value is within the reported range for other *Crataegus* species, such as *Crataegus scabrifolia* (84 mg/g) and *C. pinnatifida* (31 mg/g) by Gao et al. [37], and higher than that reported by Froehlicher et al. [38] for *C. monogyna* (1.47 mg/g).

TABLE 1: Phytochemical characterization and content of free polyphenolic compounds in *Crataegus pubescens* fruits.

Group/compound	Concentration
Total carbohydrates (g/100 g extract)	69.8 ± 1.61
Total phenolics (mg gallic acid equivalent/g fruit dry)	168.6 ± 0.9
Total flavonoids (mg equivalents quercetin/g dry fruit)	55.89 ± 0.11
Proanthocyanidins (mg cyanidin/100 g dry fruit)	84.57 ± 1.43
(-)-Epicatechin (mg/100 mg dry fruit)	4.32 ± 0.11
(+)-Catechin (mg/100 mg dry fruit)	9.17 ± 0.20
Chlorogenic acid (mg/100 mg dry fruit)	5.60 ± 0.24

The response factor for estimation of the phenolic content was from the calibration curve using MEKC. The results are shown as the mean ± SD ($n = 3$).

FIGURE 1: Continued.

FIGURE 1: Electropherogram 217 nm (a) and HPLC 230 nm chromatogram (b) of a phenolic extract of *Crataegus pubescens* fruits. The spectral pattern and identity of the major compounds are shown in (c); 1. (–)-epicatechin; 2. (+)-catechin; 3. procyanidin dimer; 4. procyanidin trimer; 5. procyanidin tetramer; 6. chlorogenic acid. Calibration curves were linear for both methods for concentration between 10 and 160 μM for (–)-epicatechin and (+)-catechin, with correlation coefficients of 0.9901 and 0.9959, respectively, and in a range of 10–240 μM with a correlation coefficient of 0.9976 for chlorogenic acid. The electropherogram with the resolved standards is presented (inset) in (a).

Proanthocyanidins are another group of plant phenolic compounds with antioxidant and pharmaceutical properties. The total content of these compounds in the phenolic extracts of tejocote (*C. pubescens*) fruits was 84.57 ± 1.43 mg cyanidin/100 g dry fruit (Table 1). This content is comparable to that reported by Froehlicher et al. [38] for *C. monogyna* fruits (108.7 ± 9.2 mg/100 g) and higher than that reported for edible fruits from other plant species [39] such as apple (17–50 mg/100 g) and grape (1–160 mg/100 g). Proanthocyanidins belong to a flavonoid subgroup containing a flavan-3-ol unit, epicatechin, and/or catechin [40], which have been reported to have beneficial effects for the endothelium blood vessels by inhibiting endothelin-1 release and improving the blood circulation [41]; consequently, they are considered as the phenolic compounds responsible for *Crataegus* species to be used for treatment of diseases such as heart failure [42].

The characterization of the extract of *C. pubescens* showed the presence of several phytochemicals that in several scientific studies are correlated with diverse biological and pharmacological effects of several species of *Crataegus*. For example, *C. monogyna* has a long history as a medicinal plant used to treat kidney stones, digestive ailments, dyspnea, and cardiovascular disorders and is currently being used for the treatment of cardiovascular diseases [4]. These results demonstrate the potential of tejocote (*C. pubescens*) over other *Crataegus* species with a smaller content of flavonoids already used for therapeutic purposes [35, 36].

3.2. Phenolic Profile. The major free phenolic compounds identified by both HPLC and MEKC in the phenolic extract

from tejocote (*C. pubescens*) fruits were (–)-epicatechin, (+)-catechin, and chlorogenic acid (Figure 1). These results show that MEKC is an alternative technique for analysis of phenolic compounds. These compounds were confirmed by comparing their migration time or retention time with respect to the corresponding standards. The estimation of the quantitative data is performed on the basis of the integration of the areas of the peaks using the software of the instrument. Figure 1(a) shows the electropherogram of the tejocote extract. Table 1 shows the content of (–)-epicatechin, (+)-catechin, and chlorogenic acid in the tejocote (*C. pubescens*) fruits. Likewise, the UV spectra (Figure 1(c)) revealed the presence of compounds that are similar to the absorption spectra of (–)-epicatechin and (+)-catechin. This observation shows that the detected molecules might be oligomers of such compounds, known as procyanidins, which agrees with reports in the literature for polyphenols in extracts from flowers [43] and cell suspensions, fresh fruits, and medicinal dried parts of this plant species [38]. Further, to confirm the presence of these compounds in the phenolic extracts, a mass analysis (MS/MS) was carried out. Six HPLC peaks were collected (Figure 1(b)) and analyzed by MS/MS using a mass spectrometer. Ions for a procyanidin dimer (m/z 579.1), a procyanidin trimer (m/z 867.2), and a procyanidin tetramer (m/z 1153.2) were detected and positively identified by MS/MS fragmentation (Figure 2). The presence of these compounds is congruent with the antioxidant activity shown by extracts of tejocote (*C. pubescens*) in the presence of the superoxide radical and DPPH, since the phenolic compounds, especially procyanidins and flavonoids, are the principal bioactive compounds in *Crataegus* fruits [2]. This result demonstrates the nutraceutical

(a)

(b)

FIGURE 2: Continued.

FIGURE 2: Fragmentation ions MS/MS in positive mode of procyanidins detected in *Crataegus pubescens* extracts. (a) Procyanidin dimer (m/z 579.1); (b) procyanidin trimer (m/z 867.2); (c) procyanidin tetramer (m/z 1153.2).

potential of *C. pubescens* fruits. Some authors [44] reported a hypoglycemic effect of *C. pubescens* root extracts in the presence of flavonoids (gallocatechin gallate and quercetin) and phenolic acids (ferulic acid and coumaric acid). Although the compounds reported in the root are not the same as those in the fruit (present study), it is important to mention that flavan-3-ol (catechin and epicatechin) has a strong antioxidant potential [45] and has antimutagenic, antidiabetic, anti-inflammatory, antibacterial, and antiviral properties. Therefore, due to the presence of (–)-epicatechin, (+)-catechin, and chlorogenic acid identified in the phenolic extract of tejocote (*C. pubescens*) by both HPLC and MEKC, tejocote fruits or its pulp extracts could be used as a new ingredient for the elaboration of functional foods or as a nutraceutical.

3.3. Antioxidant Activity DPPH. Trolox™ and quercetin were used for the determination of antioxidant activity of the phenolic extracts (Figure 3 and Table 2) as a reference, since Trolox is the reference antioxidant used in most studies of antioxidant activity, mainly when it is expressed in EC_{50} [46, 47], owing to its structural analogy with vitamin E. However, Trolox is a synthetic compound, whereas quercetin is a natural flavonoid extensively found in plants and is recognized as a potent antioxidant.

The EC_{50} value for the phenolic extract ($1472.27 \mu\text{g/mL}$) was higher than that for Trolox or quercetin (Table 2), indicating that more of the extract is required than that with Trolox or quercetin. This result might be explained due to the purity of the extract, which was used without any purification and has a high content of carbohydrates (69.8%). Similarly, a DPPH capability of inhibition dependent on the concentration was found (Figure 3). When the sample concentration ranged between 300 and $1800 \mu\text{g/mL}$, the inhibition percentage of the DPPH radical varied from 20.5% to 51.5%, and the EC_{50} of 1.47 mg/mL reduced the DPPH radical concentration by 50%. It is known that antioxidant activity is the result of the synergistic and antagonistic effects of the interactions of different compounds [47]. It has been suggested that antioxidants could prevent many chronic diseases such as cancer, diabetes, and cardiovascular diseases [10]. In the present study, the presence of biologically active compounds such as flavonoids in the extract of *C. pubescens* suggests that they can capture free radicals and confer antioxidant activity, demonstrating the nutraceutical potential of this fruit. Likewise, it was observed that antioxidant efficiency (AE) is a discriminating parameter between among samples. Regarding the phenolic extracts, the order of AE was Trolox > quercetin > tejocote extract (*C. pubescens*).

FIGURE 3: Percentage of remnant DPPH against the standard concentration was plotted in an exponential regression to obtain the amount of antioxidant necessary to decrease the initial DPPH concentration by 50% (EC₅₀) and the time needed to reach the steady state for EC₅₀ (TEC₅₀). The results are shown as the mean $\pm$ SD ($n = 3$).

3.4. Superoxide Trapping Activity. The inhibition capability of the superoxide radical is important because in addition to the hydroxyl radical, they are the main reactive oxygen species (ROS) continuously generated in normal body metabolism, particularly through the mitochondrial energy production pathway. Control of ROS production is fundamental to avoid oxidative stress, which is implicated in the incidence and progression of several disease conditions [48]. Figure 4 shows the inhibitory relationship of the superoxide radical dependent on the phenolic extract. When the extract

concentrations ranged between 200 and 1000 $\mu\text{g/mL}$, the inhibition percentage of the superoxide radical ranged from 19.5% to 61.7%. These results are comparable with those of other *Crataegus* species, such as *Crataegus aronia* [49], that are used for medicinal purposes, whose aqueous extracts inhibit the formation of the superoxide radical (82%) with concentrations between 500 and 1000 $\mu\text{g/mL}$. It has also been reported that the concentration of the superoxide radical decreases by more than 50% in a 500 $\mu\text{g/mL}$ concentration of ethanolic extract from *Crataegus oxyacantha*

TABLE 2: Antioxidant activity through the DPPH radical.

Sample	Concentration ($\mu\text{g/mL}$)	% inhibition DPPH	EC ₅₀ ($\mu\text{g/mL}$)	TEC ₅₀	AE
Extract tejocote fruits	300	20.5	1472.27	36	1.88×10^{-5}
	600	32.4			
	900	41.6			
	1200	49.1			
	1500	50.3			
	1800	51.5			
Trolox	120	41.4	154.19	1.33	4.87×10^{-3}
	140	46.2			
	160	51.1			
	180	56.4			
	200	62.5			
Quercetin	20	14.8	55.48	14.03	1.28×10^{-3}
	40	38.7			
	60	56.3			
	80	74.8			
	100	85.4			

EC₅₀: amount of antioxidant necessary to decrease the initial DPPH concentration by 50%; TEC₅₀: time required to reach the steady state for EC₅₀; EA: antiradical efficiency, where $AE = 1/EC_{50} \cdot TEC_{50}$. The results are shown as the mean ($n = 3$).

FIGURE 4: Effect of different concentrations of extracts from tejocote on the inhibition of superoxide radical generated by the xanthine-xanthine oxidase system. The results are shown as the mean $\pm$ SD ($n = 3$).

[36]. Thus, the results from the present work suggest that the phenolic extract from tejocote (*C. pubescens*) is able to trap superoxide radicals.

4. Conclusions

This is the first study to report the presence of (–)-epicatechin, (+)-catechin, chlorogenic acid, and procyanidins represented in dimers, trimers, and tetramers in the tejocote (*Crataegus pubescens*) fruits in important concentrations in order to consider such fruit as a potential source of extraction of such polyphenols to be used in the food or nutraceutical industries. Tejocote is of interest because of these antioxidant activities, and the trapping capability of free radicals is comparable to that of other *Crataegus* species that are used for therapeutic

purposes. Likewise, micellar electrokinetic chromatography (MEKC) proved to be an alternative to the use of HPLC for the qualitative and quantitative analysis of phenolic compounds in tejocote (*C. pubescens*). The time and use of reagents were reduced, and the resolution of the analyzed compounds was not affected. This result demonstrates that tejocote (*C. pubescens*) could be considered as a potential new ingredient for the elaboration of functional foods or nutraceuticals due to the presence of antioxidants.

Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Conflicts of Interest

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

Acknowledgments

The authors would like to thank CONACYT for the economic support provided in order to carry out this project using scholarship 268626/219101 granted to Francisco Erik González-Jiménez. Moreover, they would like to thank the biologist María del Pilar Méndez-Castrejón, chemist Octavio Gómez-Guzmán, and M.Sc. Emmanuel Ríos-Castro for their valuable support in the mass spectrometry analysis carried out in the National Laboratory of Experimental Services (LaNSE-Cinvestav).

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