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Original Article

Ceratonia siliqua honeys from Morocco: Physicochemical properties, mineral contents, and antioxidant activities

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ABSTRACT

Physicochemical properties, main mineral content, and antioxidant activity were determined for eight floral carob honeys collected from different geographical regions of Morocco. Moroccan honeys showed good chemical and nutritional qualities, fulfilling the criteria described in the standard codex for honey. The percentages obtained for ashes were (0.13–0.69%), electrical conductivity (0.36–1.35 mS/cm), water content (17.30–22.80%), pH (4.17–5.05), free acidity (11.0–42.50 meq/kg), lactone acidity (4.0–16.50 meq/kg), and for total acidity (16.50–59.50 meq/kg). In addition, minerals such as K, Na, Mg, Cu, Zn, and Ca of honey samples were determined and potassium was the major mineral in all samples. The antioxidant activities based on the free radical scavenging, reducing power, and total antioxidant activity were investigated, and the antioxidant capacity of the honey samples was correlated with their biochemical constituents such as total phenol and flavonoids content, and the best antioxidant capacity was confirmed by the honey from Taounate.

1. Introduction

Honey is the natural sweet substance produced by honey bees from the nectar of plants or from secretions of living parts of plants or excretions of plant-sucking insects on the living parts of plants, which the bees collect, transform by combining with specific substances of their own, deposit, dehydrate, store, and leave in the honey comb to ripen and mature [1,2].

The use of honey can be traced back to the Stone Age. Evidence can be found for its nutritional and medicinal use

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beginning with prehistoric and ancient civilizations. Currently, there is a resurgence of scientific interest in natural medicinal products, such as honey, by researchers, the medical community, and even the general public. The medicinal and therapeutic potential of honey on human health, such as cardioprotective, anticancer, antimicrobial, and anti-inflammatory effects, can be attributed to the presence of antioxidant compounds [3–6].

It is well known that both botanical and geographical origin of the honey as well as climatic and environmental conditions influence its composition, antioxidant activity, and health-protective characteristics.

Honey is produced and largely used as an important energy food source and medicinal source [7,8]. This complex mixture has been reported to contain ~200 components, of which, fructose and glucose are the main components that make it a good source of energy for humans. Other constituents such as proteins, free amino acids, organics acids, phenol compounds, vitamins, and traces minerals are also present in honey [6,9], and are responsible for several therapeutic properties including antioxidant, anti-inflammatory, anticancer, mutagenic, antibacterial, antifungal, and antiviral activities [5,10–14].

Moroccan honey is legendary, and some think it is rated among the best in the world. In Morocco, honey is widely used in traditional medicine; unfortunately, there are not enough investigations regarding its quality and characterization, and 80% of the productivity is due to traditional beekeeping [15,16].

The main goals of this work were to determine the physicochemical composition, mineral content, and antioxidant capacity of eight samples of honey from nectar of the famous carob tree in different regions of Morocco in order to understand the influence of geographical ecology on the composition and quality of Moroccan honey.

2. Materials and methods

2.1. Honey samples

The present study was carried out using eight reputed commercial Moroccan carob honeys. The samples from (1) Marrakech, (2) Berkane, (3) Rabat, (4) Sefrou, (5) Meknes, and (8) Benimellal, were obtained from the Beekeepers Associations from six different regions in Morocco during the Ninth International Exhibition of Agriculture in Morocco (2014). Samples (6) and (7) were purchased from the beekeepers of two different places in Taounate Region because of its abundance of the carob tree.

2.2. Physical analysis

2.2.1. Determination of moisture content

The moisture content was determined based on the refractometric method according to the International Honey Commission (IHC). The moisture content was determined from the refractive index of the honey by reference to a standard table [17]. Tests were done in triplicate and the results are given as the mean average.

2.2.2. Determination of total soluble solids and total solid content

The total soluble solids (TSSs) of the honey samples were measured using a refractometer, and the results were expressed in Bx. Total solid (TS) content was calculated using the formula: [9]

$$\text{TS (\%)} = 100 - \text{water content.}$$

Tests were done in triplicate and the results are given as the mean average.

2.2.3. Determination of pH

The determination of pH value was performed according to the IHC [17], with a pH meter from a solution of 10 g of honey samples dissolved in 75-mL ultrapure water. Tests were done in triplicate and the results are given as the mean average.

2.2.4. Determination of free, lactone, and total acidity

Free, lactone, and total acidity was determined using the titrimetric method according to the IHC [17]. The free acidity was determined in a solution of 10-g honey samples dissolved in 75-mL ultrapure water by titration with 0.05M NaOH to pH 8.3 (free acidity). After, 10 mL of the same solution (0.05M NaOH) was added and immediately back-titrated by 0.05M HCl to pH 8.3 (lactone acidity). Total acidity was obtained adding lactone to free acidity [18]. Tests were done in triplicate and the results are given as the mean average, and expressed as milliequivalents/kg (meq/kg).

2.2.5. Determination of electrical conductivity

Electrical conductivity was determined using a conductivity meter at 20°C in a 20% (dry matter basis) solution of honey samples prepared with ultrapure water, according to the IHC [17]. Tests were done in triplicate and the results are given as the mean average, and expressed as (ms/cm).

2.2.6. Determination of color and melanoidin content

Honey color was determined by measuring the absorbance of 50% honey solutions [weight/volume (w/v)] at 635 nm using a UV–visible spectrophotometer. The mm Pfund values were obtained using the following algorithm [19]:

$$\text{mm Pfund} = -38.7 + 371.39 \times \text{absorbance}$$

Melanoidin content was estimated based on the Browning index by measuring the net absorbance of the honey samples at 450 nm and 720 nm [19].

2.2.7. Determination of ash content

The ash content was determined by placing 5-g honey samples in a crucible in a muffle furnace and heating at 550°C for 5 hours. The residue was weighed in an analytical balance. Measurements of ash were done in triplicate and the mean was expressed as a percentage [17]. Tests were done in triplicate and the results are given as the mean average.

2.2.8. Determination of mineral elements

Ash values were obtained by the calcinations method as described above. After that, 5 mL of nitric acid 0.1M was added and the mixture was stirred on a heating plate to almost

complete dryness. Then, 10 mL of the same acid was added and the mixture was made up to 25 mL with ultrapure water [6]. The mineral components were determined by atomic absorption spectrometry (Thermo Scientific ICE 3000 Series AA Spectrometer, ISBS Tunisia) after calibrating the instrument, using K (0.1–5 mg/L), Na (0.1–5 mg/L), Ca (0.1–5 mg/L), Mg (0.1–5 mg/L), Fe (0.1–5 mg/L), Cu (0.5–4 mg/L), Zn (0.1–2 mg/L), Pb (0.1–4 mg/L), Co (0.1–5 mg/L), Cd (0.1–2 mg/L), and Ni (0.1–5 mg/L).

2.3. Bioactive compounds and antioxidant activities

2.3.1. Determination of total phenolic content

Total phenol in all samples was determined with the Folin–Ciocalteau method [21], with slight modifications. The honey samples were dissolved in distilled water (50% w/v). Then, 10-μL dissolved honey was mixed with 180-μL distilled water in a well of a 96-well plate. This solution was mixed with 10-μL Folin–Ciocalteau reagent solution for 6 minutes, and then 30-μL 20% sodium carbonate solution was added to the mixture. After incubation at room temperature for 2 hours, the absorbance was measured at 760 nm against a water blank solution. Gallic acid was used as a standard, and the total phenolic content was expressed as mg gallic acid equivalents per 100 g honey (mg GAE/100 g). Tests were done in triplicate and the results are given as the mean average.

2.3.2. Flavone and flavonol content

The flavone and flavonol content was quantified according to the method described by Miguel et al [22] with minor modifications. Briefly, to 100 μL of sample or standard, we added 100 μL 2% $AlCl_3$–ethanol solution in a well of a 96-well plate. After 1 hour at room temperature, the absorbance was measured at 420 nm. Quercetin (10–25 μg/mL) was used as a standard, and total content was expressed as mg quercetin equivalents per 100 g honey (mg QE/100 g). Tests were done in triplicate and the results are given as the mean average.

2.3.3. Determination of antiradical activity: 2,2-diphenyl-1-picrylhydrazyl assay

The radical scavenging activity of the honey samples against 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical was measured using the method of Clarke with minor modifications [23]. Twenty microliters of the honey solutions (50% w/v) in distilled water or the ascorbic acid standard were mixed with 180 μL of 100μM solution of DPPH radical prepared in ethanol (96%) in the wells of a 96-well plate. The absorbance of the solution was measured at 540 nm after 15 minutes of incubation in the dark at room temperature, against a blank solution containing water instead of honey. Ascorbic acid was used as positive control. The percentage of inhibition was calculated using the formula:

$$\% \text{ Inhibition} = [(\text{absorbance blank} - \text{absorbance sample})/\text{absorbance blank}) \times 100]$$

The concentration of sample required to scavenge 50% of DPPH was also determined. Tests were done in triplicate and the results are given as the mean average.

2.3.4. Determination of reducing power

The reducing power or ferric reducing antioxidant power (FRAP) of honey samples was determined using the method of Saxena et al [9] with slight modifications. One hundred microliters of honey solutions (50% w/v) in distilled water were mixed with 250 μL 0.2M phosphate buffer (pH 6.6) and 250-μL 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 minutes and 250 μL = 10% trichloroacetic acid was added and the mixture was centrifuged at 2200g for 10 minutes. Five hundred microliters of the above solution from each reaction was diluted with 500 μL distilled water. One hundred microliters of 0.1% $FeCl_3$ was added. The absorbance against a blank was measured at 700 nm using a UV–visible spectrophotometer, and ascorbic acid was used as reference standard. Tests were done in triplicate and the results are given as the mean average.

2.3.5. Determination of total antioxidant activity

The total antioxidant capacity of the honey samples was evaluated by the phosphomolybdenum method as described by Prieto et al [24] with minor modifications. Briefly, 0.2-mL honey samples were mixed with 2-mL reagent solution (0.6M sulfuric acid, 28mM sodium phosphate, and 4mM ammonium molybdate). All tubes were capped and incubated in a boiling water bath at 95°C for 90 minutes. The absorbance of the cooled mixture was measured at 695 nm against a blank sample using a UV–visible spectrophotometer and the results are expressed as equivalents ascorbic acid/g honey. Tests were done in triplicate and the results are given as the mean average.

2.4. Statistical analysis

All analyses were run in triplicate and the results were expressed as means ± standard deviation. Pearson correlation was performed using IBM SPSS statistics version 20.

3. Results and discussion

3.1. Physicochemical analysis

The moisture content is an important criterion to determine the quality of honey; it depends on environmental conditions, harvest season, storage ability, and source of nectar used by the bee. The moisture content (%) of analyzed carob honey samples ranged from 17.30% to 22.0% (Table 1). The moisture values for all samples were in accordance with values recommended by the IHC in 2009 and Codex Standard for Honey 2001, and were similarly compared with the values obtained for carob honey from Italy [25] and Portugal [20]. The honey from Meknes presented the highest value for moisture, which facilitates the growth of molds on the honey surface and fermentation during storage.

The ash content is an indicator of the botanical origin and influences the mineral content of honey [9]. In our study, the ash content in honey samples varied between 0.13% and 0.69%, and the largest value was detected in carob honey from Taounate Region (0.69 ± 0.01%; Table 1). Similar levels of ash content were obtained for honeys from Italy [25] and Turkey [8].

Table 1 – Physicochemical parameters for Moroccan carob honey.

	pH	Moisture (%)	Free acidity (mEq/kg)	Lactone acidity (mEq/kg)	Total acidity (mEq/kg)	Ash content (%)
H1	4.17 ± 0.05	19.3 ± 0.14	42.50 ± 2.29	16.50 ± 1.73	59.0 ± 3.24	0.36 ± 0.0
H2	4.90 ± 0.01	18.6 ± 0.28	40.0 ± 1.50	15.50 ± 1.50	57.50 ± 2.45	0.58 ± 0.01
H3	4.88 ± 0.13	18.7 ± 1.83	13.50 ± 1.50	4.00 ± 0.87	17.50 ± 0.71	0.23 ± 0.00
H4	4.86 ± 0.16	17.3 ± 0.70	11.0 ± 2.29	5.50 ± 1.73	16.50 ± 2.55	0.24 ± 0.01
H5	4.73 ± 0.04	22.0 ± 1.41	26.00 ± 0.87	16.0 ± 2.29	42.0 ± 1.87	0.39 ± 0.0
H6	5.05 ± 0.13	19.8 ± 0.28	27.0 ± 3.01	12.50 ± 3.0	39.50 ± 4.24	0.69 ± 0.01
H7	4.79 ± 0.03	19.5 ± 0.42	31.50 ± 0.87	14.00 ± 1.50	46.0 ± 1.87	0.65 ± 0.01
H8	4.17 ± 0.01	20.8 ± 0.84	17.0 ± 0.87	7.25 ± 1.06	24.5 ± 1.50	0.13 ± 0.01
	Electrical conductivity (mS/cm)	TSS (Bx)	TS (%)	Pfund scale (mm)	Honey color	Melanoidin
H1	0.55 ± 0.01	79.05 ± 0.07	80.7 ± 0.14	148.85 ± 2.59	Dark amber	1.45 ± 0.01
H2	1.17 ± 0.02	79.60 ± 0.28	81.4 ± 0.28	141.42 ± 0.37	Dark amber	1.49 ± 0.04
H3	0.77 ± 0.01	80.50 ± 0.28	81.3 ± 1.83	47.71 ± 0.77	Extra light amber	0.70 ± 0.01
H4	0.56 ± 0.02	84.15 ± 0.63	82.7 ± 0.70	129.16 ± 2.22	Dark amber	0.86 ± 0.02
H5	0.82 ± 0.01	76.55 ± 1.62	78.0 ± 1.41	150.83 ± 4.39	Dark amber	1.36 ± 0.04
H6	1.35 ± 0.03	78.60 ± 0.28	80.2 ± 0.28	104.40 ± 1.86	Amber	1.21 ± 0.04
H7	1.27 ± 0.02	78.80 ± 0.42	80.5 ± 0.42	137.82 ± 3.24	Dark amber	1.26 ± 0.02
H8	0.36 ± 0.02	77.50 ± 0.84	79.2 ± 0.84	305.82 ± 4.22	Dark amber	1.45 ± 0.02

All values are expressed as means of triplicate determinations ± standard deviation.
TS = total solid; TSS = total soluble solid.

The electrical conductivity depends on the mineral content of the honey. In the present work, the electrical conductivity varied in the range of 0.36 mS/cm to 1.35 mS/cm (Table 1). High values were also found in carob honey from Taounate Region (1.35 ± 0.03 mS/cm in Sample 6 and 1.27 ± 0.02 mS/cm in Sample 7). These values agreed with the results obtained in carob honey from Italy [25].

The pH for all studied honey is acidic in character. In this work, the pH ranged from 4.17 to 5.05 (Table 1). These were acceptable values and comparable with those obtained in other studies [19,25,26]. The values of free acidity, lactone acidity and total acidity ranged from 11.0 to 42.5, 4.0 to 16.50 and 17.50 to 59.0, respectively (Table 1), which agree with data reported in the literature from other geographical locations [6,27,28]. The determined acidity of honey is due to the presence of organic acids and inorganic ions. Free acidity values of all honeys were within the limits (< 50 meq/kg).

The obtained data for TSSs and TS content ranged from 76.55 to 84.15°Brix and 78.0 to 82.7%, respectively. The mean values of TSS and TS agreed with those for honeys obtained from India [9] and Portugal [6]. In our study, the TSS and TS values did not show a wide variation among different honeys, and the highest value for the two tests found in honey from Sefrou were 84.15 ± 0.63 and 82.7 ± 0.70, respectively.

Honey color is closely related to the storage conditions, phenols and flavonoids content, and mineral and pollen content [19]. The degree of color measured by the absorbance of samples at 635 nm is presented in Table 1. The major samples of Moroccan carob honey analyzed were dark amber; these results are in accordance with those reported in Portuguese honeys [20].

Correlation analysis was performed on the obtained findings by measuring the values of physicochemical characteristics in eight honey samples collected from different regions of Morocco (Table 2). The Pearson's correlation coefficient

Table 2 – Pearson correlation coefficients among the analyzed parameters.

Parameter	pH	AC	EC	FA	TA	WC	TSS	PS	M
pH	1	0.549	0.732*	−0.180	−0.148	−0.320	0.320	−0.714*	−0.489
AC		1	0.949**	0.610	0.637	0.016	−0.215	−0.369	0.297
EC			1	0.400	0.426	−0.024	−0.157	−0.493	0.075
FA				1	0.992**	0.134	−0.399	−0.010	0.733*
TA					1	0.207	−0.447	0.009	0.764*
WC						1	−0.927**	0.446	0.533
TSS							1	−0.401	−0.674
PS								1	0.629
M									1

AC = ash content; EC = electrical conductivity; FA = free acidity; M = melanoidin; PS = Pfund scale; TA = total acidity; TSS = total soluble solid; WC = water content.
* Correlation between analyzed physicochemical parameters were statistically significant ($p < 0.05$).
** Correlation between analyzed physicochemical parameters were statistically significant ($p < 0.01$).

Table 3 – Concentrations of major and minor metals in carob honeys from Morocco.

Sample	Major metals				
	Na	K	Ca	Mg	Fe
H1	367.52 ± 4.13	644.02 ± 1.98	180.34 ± 0.28	18.42 ± 0.15	2.81 ± 0.18
H2	497.54 ± 2.75	1809.32 ± 4.12	458.11 ± 2.19	86.33 ± 0.22	1.13 ± 0.11
H3	700.27 ± 2.75	897.16 ± 9.52	286.01 ± 5.79	43.09 ± 0.11	0.77 ± 0.11
H4	672.01 ± 1.37	811.42 ± 4.76	129.35 ± 0.21	29.08 ± 0.15	2.09 ± 0.18
H5	655.44 ± 5.51	1030.53 ± 6.30	396.1 ± 2.19	53.74 ± 0.15	4.68 ± 0.11
H6	855.24 ± 4.13	1634.67 ± 6.87	584.66 ± 6.57	131.21 ± 0.28	1.19 ± 0.18
H7	656.41 ± 1.37	1883.15 ± 23.45	688.43 ± 8.76	100.67 ± 0.51	0.71 ± 0.11
H8	683.12 ± 1.73	777.07 ± 0.95	422.67 ± 5.79	64.44 ± 0.95	0.71 ± 0.10
	Minor metals				
	Cu	Zn	Ni	Cd	Pb	Co
H1	1.82 ± 0.0	4.26 ± 0.03	0.15 ± 0.03	ND	ND	ND
H2	0.88 ± 0.0	1.95 ± 0.02	0.11 ± 0.0	ND	ND	ND
H3	0.32 ± 0.06	2.48 ± 0.03	0.02 ± 0.01	ND	ND	ND
H4	1.05 ± 0.0	3.36 ± 0.02	0.07 ± 0.01	ND	ND	ND
H5	0.36 ± 0.0	1.41 ± 0.03	0.11 ± 0.02	ND	ND	ND
H6	0.11 ± 0.02	2.24 ± 0.03	0.07 ± 0.01	ND	ND	ND
H7	0.36 ± 0.12	3.51 ± 0.02	0.03 ± 0.01	ND	ND	ND
H8	0.32 ± 0.06	2.27 ± 0.03	0.07 ± 0.0	ND	ND	ND

All values are expressed in mg/kg as means of triplicate determinations ± standard deviation.
ND = not determined.

between electrical conductivity and ash content indicated a strong positive correlation ($r = 0.94$), which established the dependence of electrical conductivity of honey on ash content. This correlation has also been reported in a previous study [26]. There was a significant positive correlation found between melanoidin, free acidity, and total acidity ($r = 0.733$, $r = 0.766$, respectively), which indicate that acidity contributed to melanoidin formation. In addition, a strong negative correlation was found between TSSs and water content ($r = -0.927$). This significant negative correlation was an expected result given that the water content of honey decreases with increasing TSSs.

3.2. *Mineral content*

The mineral content is one of the parameters used for the evaluation of nutritional values of honey. It can be considered as a potential indicator of geographical origin of honey as well as an important biomarker for environmental pollution of honey with heavy metals. This analysis is important to beekeepers and consumers because it helps them to avoid possible contamination during honey processing and assures them that the product is of good quality [29,30]. Also, the mineral content helps to estimate the environmental quality in different regions. In this work, chemical analysis of major minerals and heavy metals present in Moroccan carob honey was performed (Table 3).

According to the obtained results, K was the main mineral present in Moroccan carob honey with a wide variation ranging from 644.02 mg/kg in honey from Marrakech Region to 1883.15 mg/kg in that from Taounate Region. Na was the second most prevalent mineral in honey samples and the concentration varied in the range of 367.52–855.24 mg/kg. Ca and Mg were also important quantitatively in carob honey, with values ranging from 129.35 mg/kg to 688.43 mg/kg and 18.42 mg/kg to 131.21 mg/kg, respectively. The highest concentration of these two minerals was found in honey from Taounate Region (Table 4). In addition, the trace minerals of Fe, Zn, Cu, and Ni were detected in all honey samples at low concentrations ranging from 0.71 mg/kg to 4.68 mg/kg, 1.41 mg/kg to 4.26 mg/kg, 0.11 mg/kg to 1.82 mg/kg, and 0.02 mg/kg to 0.15 mg/kg, respectively. The analyzed heavy metals Pb, Cd, and Co were not detected in all carob honey samples, which indicated honey of good quality and purity. In comparison with other studies, the values found for K, Na, Ca, and Mg were similar to those reported in honey from the United Arab Emirates [31], Portugal [30], Morocco [19], and Spain [32].

3.3. *Biochemical analysis and antioxidant activity*

Phenol and flavonoid contents of carob honey from Morocco and the capacity for scavenging free radicals (DPPH), reducing power, and total antioxidant activity are presented in Table 4. The determination of phenols, flavones, and flavonols contents is an important criterion for determining the nutritional quality of honey; this composition depends on the botanical origin of honey [31,33]. Our results showed the richness of the carob honey for these bioactive molecules. The total phenols and flavonoids (flavones and flavonols) contents varied from 75.52 mg GAE/100 g to 245.22 mg GAE/100 g and 2.26 mg QE/100 g to 4.79 mg QE/100 g, respectively. The phenolic content of our honey samples was similar to that reported in Yemen honey (75.13–246.21 mg/100 g) [35], and was higher than that obtained in honey from Brazil (61.16–111.37 mg GAE/100 g) [34], India (49–98 mg GAE/100 g) [9], Serbia (27.44–61.42 mg GAE/100 g) [7], and Algeria (15.84–61.63 mg/100 g) [36].

The antioxidant activity cannot be evaluated by a single method, but at least two test systems have been

Table 4 – Phenols, flavone, and flavonol contents and antioxidant activities of Moroccan carob honeys.

	Phenols (mg GAE/100 g)	Flavone and flavonol (mg QE/100 g)	TAA (mg AAE/g)	DPPH (IC50 = mg/mL	FRAP (IC50 = mg/mL)
H1	95.31 ± 8.80	4.71 ± 0.35	35.03 ± 0.61	15.94 ± 0.7	2.93 ± 0.06
H2	163.55 ± 8.88	4.51 ± 0.26	60.63 ± 1.70	15.82 ± 0.44	1.98 ± 0.0
H3	75.52 ± 7.45	2.26 ± 0.28	53.75 ± 1.16	23.10 ± 0.54	4.40 ± 0.02
H4	96.31 ± 1.01	2.82 ± 0.25	57.37 ± 0.10	15.05 ± 0.68	3.98 ± 0.02
H5	119.95 ± 1.17	4.02 ± 0.22	45.29 ± 2.29	18.99 ± 0.73	1.93 ± 0.03
H6	199.27 ± 8.71	4.35 ± 0.28	54.25 ± 2.10	14.24 ± 0.24	1.96 ± 0.01
H7	245.22 ± 11.40	4.79 ± 0.32	60.94 ± 1.88	12.54 ± 0.52	1.87 ± 0.04
H8	136.89 ± 5.07	3.30 ± 0.13	46.59 ± 1.50	23.52 ± 0.73	4.72 ± 0.08
Ascorbic acid (μg/mL)	—	—	—	8.66 ± 0.66	2.12 ± 0.06

All values are expressed as means of triplicate determinations ± standard deviation.

recommended for the determination of antioxidant power [37]. In this study, three different antioxidant tests (reducing power, DPPH, and antioxidant total activity) were used to assess the antioxidant ability of carob honey. The total antioxidant power of honey samples was estimated using the phosphomolybdenum method. This assay is based on the reduction of Mo(VI) to Mo(V) by the reducing compounds present in honey and the formation of a green phosphate/Mo(V) complex with a maximal absorption at 695 nm [24]. The total antioxidant power of carob honey varied from 35.03 mg AAE/g to 60.94 mg AAE/g. The highest value was obtained in carob honey from Taounate Region, and the lowest value was found in samples from Marrakech Region.

DPPH is a stable nitrogen-centered radical and has been widely used to estimate the free radical scavenging activity of various samples. The concentration of honey required to inhibit 50% of DPPH varied between 12.54 mg/mL and 23.10 mg/mL. The highest value was observed in the honey sample from Benimellal (H8), whereas the lowest value was observed in honey from Taounate (H6 and H7).

Fe(III) reduction is often used as an indicator of electron-donating activity. The presence of reducing agents in the honey reduced the ferric ions to ferrous ions. This reduction is quantified by an absorbance measurement at 700 nm against a blank. An increase in absorbance indicates a high reducing power [36]. The FRAP assay (Table 4) showed that the ability of carob honey to reduce ferric ions ranged from 1.87 mg/mL to 4.40 mg/mL.

The antioxidant activity of our carob honey samples was higher than that reported in previous studies on Serbian honey (11.16–48.48 mg/mL for DPPH and 39.06–120.0 mg/mL for FRAP) [7] and Turkish honey (12.56–152.40 mg/mL for DPPH) [39], but its lowest was when compared to ascorbic acid as a standard compound (8.66 ± 0.66 μg/mL for DPPH and 2.12 ± 0.06 μg/mL for FRAP). Previous studies on honey indicate that this antioxidant activity is mainly due to the presence of bioactive compounds such as polyphenols and flavonoids. Also, the antioxidant activity of honey varies widely, depending on floral source, geographical origin, climatic conditions, and processing and storage conditions [9,38].

The carob honey from Taounate Region presented the highest capacity for these activities, which give Moroccan carob honeys an important potential in combating oxidant damage and in preventing pathogenesis.

4. Conclusion

This work supplied new information about the physicochemical characterization, polyphenolic content, and antioxidant activity of Moroccan carob honeys obtained from various regions. All results showed that this honey was rich in active biomolecules that attribute good quality. This aspect of Moroccan carob honeys correlates with important antioxidant and antiradical activities observed in all honey samples obtained from different regions of Morocco with different geographical conditions, climate, and environmental factors. Moroccan carob honeys can be considered to have important potential in combating oxidant damage and preventing pathogenesis of many diseases.

Conflicts of interest

The authors declare that they have no conflict of interest.

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