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Physicochemical properties of Euphorbia honey and determination of antioxidant enzyme activity

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ABSTRACT: In this study, it was aimed to investigate the compliance of the physicochemical properties of 8 honey samples produced in regions where the dominant flora is euphorbia and obtained directly from the producer, to international standards, as well as to determine the total phenolic and flavonoid contents and antioxidant activities of the extracts of these honeys prepared with water and methanol. Determined physical properties such as humidity, pH, electrical conductivity, hydroxy methyl furfural content, acidity, brix, diastase number, proline amount and distribution of sugar ratios are within the appropriate values in terms of the honey notification accepted in our country. Phenolic and flavonoid contents were found to be higher in water extracts, in correlation with antioxidant activity values. It can be expected that the honey that can be consumed safely by consuming this class of flower honey, which has been found to be suitable in terms of physicochemical parameters and has antioxidant activity, will also be beneficial for health.

Keyword: Honey; Euphorbia honey; Physicochemical property; Enzyme activity

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INTRODUCTION

Honey, a sweet and delicious product made by honey bees from plant nectar and honeydew, is considered the main product of honey bees and is defined as monofloral if its pollen, physicochemical and sensory properties are entirely or partially from a botanical origin (Moujanni et al., 2018; Bouhlali et al., 2019; Özkırım et al., 2021). Monofloral honey, which is considered to be more valuable than polyfloral honey, is obtained from the nectar of certain source plants. For this reason, it is important to determine the type of honey before labeling and marketing (Mărgăoan et al., 2021). Moreover, today the international market attaches importance to the suitability of palynological and physicochemical parameters regarding quality control. The chemical and physical properties of monofloral honey are affected by various factors such as plant source, season, packaging conditions, geographical origin, and storage period (Moujanni et al., 2018). The study conducted by Terrab et al., (2022) on euphorbia honey, a unifloral honey, also makes an important contribution to the limited knowledge regarding euphorbia (Euphorbia resinifera) honey, which is considered a unique resin.

Although euphorbia (Euphorbia macroclada Bois.) is a herbaceous plant species that grows widely in Turkey, not all euphorbia plants produce honey nectar. Flower honey, consisting of flower nectar, can be monofloral and heterofloral. Euphorbia or spurge honey is one of the rare unifloral honey produced and it is mostly produced in in the south-eastern Anatolia region, especially in Diyarbakır and Adıyaman province in August and September. Boutoub et al., (2022) also emphasized that although Euphorbia honey is considered the most valuable honey by consumers in Morocco, it has rarely been the subject of research. The taste of euphorbia honey, which is quite viscous, does not crystallize easily, and has a slightly sour taste and aroma, varies depending on the density of the nectar. In this context, the color of euphorbia honey varies from amber to quite dark tones (Kara et al., 2020).

Although Euphorbia honey is valued by consumers in some Mediterranean countries such as Lebanon, Turkey and Morocco, there are very few studies on this type of honey. Although melissopalynological studies on honey containing Euphorbia pollen are found in the literature only in the case of honey obtained from the Canary Islands and Turkey, there is not enough data on their physicochemical characterization (Bettar et al., 2019).

The composition of honey is quite variable, although it depends primarily on the flower source. However, factors such as seasonal, environmental and processing differences also affect the composition. Sugar and water are the basic components of natural honey. Additionally, Honey contains a number of proteins and a large number of free amino acids (Afroz et al., 2023). In order to benefit from the commercial return of honey, consumers must trust the honey they purchase. For this reason, it is important to reveal sensory, chemical, physical and microbiological properties. Physicochemical analysis of honey is also important for the honey industry. Because these factors are closely related to storage quality, granulation, texture, flavor and nutritional and medicinal qualities of honey (Ansari et al., 2023). On the other hand, it is also possible to verify the authenticity of honey and to reveal possible artificial additive components (Liu et al., 2023). Studies have shown that honey types contain a wide variety of components with antioxidant activity, such as flavonoids, phenolic acids, vitamins and enzymes, which have a healing effect on some chronic diseases, in varying sizes depending on these variables (Liu et al., 2023).

In this paper, the antioxidant activity and total phenolic and flavonoid amounts underlying the antioxidant activity, as well as physicochemical properties such as prolin, sugar and 5-hydroxymethyl-2-furaldehyde HMF contents, moisture, pH, electrical conductivity, diastase number, free acidity of honey samples confirmed to be obtained from the euphorbia plant by the local producer were investigated.

MATERIALS AND METHODS

Honey samples

Eight mature honey samples were obtained from local producers and botanical origins were confirmed by palynology analysis. Both water and methanol extracts were prepared from each honey sample at a concentration of 50% and were filtered through Whatman 1 to make them ready for analysis (Serem & Bester, 2012). The samples were stored in the dark at −20°C until analysis.

Determination of physicochemical properties of honey samples

Physicochemical analyzes that must be determined to determine the quality of honey must be tested as required by the International Honey Commission (IHC) (Bogdanov, 2009; Liu et al., 2023).

Moisture was measured using Abbe refractometer. To determine acidity, sample to be tested was dissolved in CO2 with distilled water and titrated with 0.1 N NaOH. pH value and electrical conductivity (EC) was measured by pH meter and conductometer, respectively. The soluble solids (°Brix) of samples were quantified using a refractometer. HMF was determined calorimetrically after dilution with distilled water and addition of p-toludine solution. Absorbance of solution was determined at 550 nm using 1 cm cells. HMF has been analyzed by the measure of the absorbance at 284 and 336 nm. HPLC system combined with a refractive index detector was used to determine the amount of sugar in honey samples. Diastase activity and proline analysis were determined by a spectrophotometer.

Determination of biological activities of the honey samples

Total phenolic content amounts of extracts prepared from honey samples were determined in terms of gallic acid (mg GAE/g honey) tested under the same conditions as a standard, according to the modified method developed by Singleton and Rossi (1965) using the Folin-Ciocalteu reagent, which is sensitive to reducing compounds, including polyphenols.

In order to reveal the total flavonoid amounts of honey samples, the absorbances occurring 40 minutes after the addition of aluminum nitrate (10%) and ammonium acetate (1 M) onto the extract were measured and recorded at 415 nm (Fukumoto and Mazza, 2000). Using the absorbances obtained at the end of the experiment carried out under the same conditions for different concentrations of quercetin, the amount of flavonoids in the samples was calculated as quercetin equivalent (mg QTE/g honey).

To investigate the antioxidant activity of honey extracts, their scavenging activity on commercially available DPPH radical was firstly investigated. For this purpose, 25 mg/mL portions of honey samples were mixed with the appropriate amount of freshly prepared DPPH solution in methanol. The absorbance of the mixture, which was left at room temperature for 30 minutes in the dark, was measured and the % scavenging activity was calculated using the following equation, using the absorbance of the DPPH solution that did not contain the sample to be tested.

The antioxidant activity of honey extracts was secondly determined as the standard antioxidant trolox equivalent, based on the method known as ferric-reducing antioxidant power (FRAP), which is based on measuring the intensity of the blue color that develops at 595 nm due to the conversion of Fe^{3+} in complex with TPTZ to Fe^{2+} by the components acting as antioxidants in the extract content (Oyaizu, 1986).

Finally, the superiority of the measurable antioxidant contents of the extracts over each other was evaluated according to their ability to scavenge superoxide radicals created in the experimental environment consisting of .1 M EDTA, 2μM riboflavin, 13 mM L-methionine, and 75μM NBT 50 mM in phosphate buffer (pH 7.8). Each reaction mixture containing extracts to be tested (Asample) and extract solvent (Ablank) only was incubated for 10 min in the presence of fluorescent light and absorbances were measured at 560 nm (Beauchamp and Fridovich, 1971).

RESULTS AND DISCUSSION

Due to different purposes and the diversity of results obtained, different methods such as ABTS (2,2-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid radical), DPPH (2,2-diphenyl-picrylhydrazyl radical), FRAP (ferric reducing antioxidant power) and ORAC (oxygen radical absorbance capacity) are used in studies to determine the antioxidant activities of honey (Seraglio et al., 2019).

Solvent selection is critical as it may cause differences in the dissolution of antioxidant compounds with different chemical structures and polarities. It is widely known that for the recovery of polyphenols, polar solvents are preferred, mostly in the form of aqueous mixtures containing methanol, ethanol, acetone or ethyl acetate (Monteiro et al., 2020). In all but one of the eight samples, DPPH radical scavenging activity was higher in water extracts than in methanol extracts. The percentage of DPPH radical scavenging for all extracts varies between 10-30 % for 25 mg/ml concentrations **(Table 1**).

Although a high correlation (R^2= 0.32) was not observed between phenolic content values and DPPH radical scavenging rates, the phenolic content values of all samples, calculated as gallic acid equivalent according to the Folin Ciocalteu method, were also found to be high in the case of extracts prepared with water. The same holds true when we compare total flavonoid content results with DPPH radical scavenging activities. It has been reported that 0.027 mg/ml of euphorbia honey purchased from a locally known beekeeping company in the steppe region of

Table 1. Physicochemical properties of Euphorbia honey

TEST PARAMETER	I	II	III	IV	V	VI	VII	VIII
Prolin (mg/kg)	647.8	622	872.7	754.4	794.3	561.7	867.7	580.9
Moisture (g/100 g honey)	15.96	16.01	15.4	15.45	16.71	16.05	15.53	15.12
Brix	82.29	82.23	82.82	82.77	81.56	82.2	82.7	83.09
pH	4.01	3.99	4.1	4.12	4.02	3.95	4.06	3.9
Electrical Conductivity (µS/cm)	329	328	371	376	351	276	347	249
Free acidity (meq/kg)	17	26	26	24	26	24	24	19
Diastase number	19.6	21	24.9	20.1	22.7	16.5	24.9	14.5
HMF (mg/kg)	4.4	4.3	4	4.4	4.6	ND	4.6	9.9
Fructose (g/100 g)	38.7	39.4	38.8	38.9	39.2	37.5	37.7	39.5
Glucose (g/100 g)	29.4	30.6	29.7	29.8	30.3	28.9	29.2	29
Glucose/Moisture	1.84	1.91	1.93	1.93	1.81	1.80	1.88	1.92
Fructose+Glucose (g/100 g)	68.1	70	68.5	68.7	69.5	66.4	66.9	68.5
Fructose/Glucose	1.32	1.29	1.31	1.31	1.29	1.30	1.29	1.3
Sucrose(g/100 g)	ND	ND	ND	ND	ND	ND	ND	ND
Maltose (g/100 g)	1.1	0.8	0.5	0.6	0.5	0.8	0.9	0.9

Tiaret scavenges 50% of DPPH radicals in the environment (Abderrahim et al., 2019). Although antioxidant activity is mainly related to the phenolic content, it is not the only contributing component and is also highly affected by the number and position of hydrogen-donating hydroxyl groups. Possible contributions from other compound families such as anthocyanins, carotenoids, ascorbic acids, terpenoids, tannins and tocopherols should also be kept in mind (Muflihah et al., 2021). However, there is a higher correlation between the antioxidant activity values revealed by the other two methods, namely FRAP and SOD methods, and total phenolic content values, 0.82 and 0.85, respectively. The total phenolic content of the honey in question was reported as 0.54 mg GAE/g extract, and the total flavonoid content was reported as 0.77 mgQE/g extract, and the fact that the total flavonoid content was considerably higher than the samples in the current study suggested that its effectiveness in scavenging DPPH radicals may be due to its flavonoid content Abderrahim et al., 2019. Although it is especially higher in the case of water extracts, the high correlation between total phenolic content and total flavonoid content values in the case of both water and methanol extracts indicates that the contribution of both phenolics and flavonoids to antioxidant activity is significant (Muflihah et al., 2021). Total phenolic content values per 100 g of euphorbia honey studied from Diyarbakır region were determined as 72 on average in terms of gallic acid equivalent. The maximum amount of flavonoids that can be detected for the same samples is equivalent to 1.55 mg quercetin for 100 g of honey. The average amount of the samples in question, sufficient to scavenge half of the DPPH radicals in the environment, is 30.20 mg/ml (Kara et al., 2020). Total phenolic and flavonoid content values of euphorbia honey obtained from Diyarbakır region were reported as 136.92 GAE and 40.68 QE per kilogram of honey, respectively (Özkök et al., 2019). In the study conducted by Gül and Pehlivan on monofloral honey varieties, both DPPH and FRAP values of euphorbia honey obtained from the Mardin region were lower than the samples in the current study.

Superoxide dismutase activities of natural sources are particularly important because the superoxide radical plays an important role in free radical chain oxidations, oxygen toxicity, inflammation, reperfusion damage and even aging (Fridovich, 1995). Although superoxide is actually a weak reagent, it is a fact that it plays a role in cellular damage by causing the reduction of Fe^{3+} to Fe^{2+} in the Haber_weiss reaction, leading to the formation of the highly reactive hydroxyl radical (Benov, 2001).

In a study comparing the antioxidant activities of commercial and natural honey, it was shown that the superoxide radical scavenging abilities of natural honey are much better than commercial honey (Mir et al., 2022).

Since it is known that strenuous exercise triggers oxidative stress leading to the formation of free radicals, the effects of Euphorbia cheriradenia Honey Supplementation on the Superoxide dismutase level of plasma after extensive exercise in rats were investigated. It has been revealed that honey supplementation may be useful to prevent acute extensive exercise-induced oxidative stress in erythrocytes by upregulating antioxidant enzyme activities (Goodarzi and Khosravi, 2013). As in the DPPH radical scavenging and iron reduction antioxidant power trials, the effectiveness of the tested honey samples in scavenging superoxide radicals formed in the environment is significantly higher in the case of water extracts. Although there are reports in the literature about various honey and honey products scavenging superoxide radicals, there are not many studies on euphorbia honey's scavenging of superoxide radicals (Nagai et al., 2001).

The chemical and physical properties of monofloral honey vary depending on various factors such as herbal source, seasonal period, packaging conditions, geographical origin, storage conditions and period. According to today's international marketing strategies, quality control should be carried out in accordance with palynological and physicochemical rules.

Although alternative methods have been tried to be identified in recent years for botanical origin verification and characterization of honey, the most effective and reliable method is still melissopalynological analysis. These analyzes are based on taxon identification by examining the morphological characteristics of the pollen contained in honey using a microscope. After this process, the botanical origins of the identified taxa are determined according to their presence and frequency in honey. Generally, honey containing 45% or more of plant pollen is considered monofloral honey and the honey is named after that plant (Ricciardelli D'Albore, 1997). However, there are exceptions to this situation. Some plants may naturally produce a small amount of pollen or their nectar production rates may be higher than pollen. Since the pollen of these types of plants is less added to honey, their presence in honey is much lower and they are called species represented with less pollen. It is thought that this exceptional situation may also be valid for the honeys examined.

Honey or pollen whose physicochemical and sensory properties come entirely or partially from a single botanical origin is defined as monofloral (EU Council, 2002).

As consumers begin to know more about the honey they consume, the honey industry is increasingly demanding information on honey characterization. In this context, the newly emerging market for monofloral honey in many countries demands the characterization of honey and its differentiation from other honey varieties (Moujanni et al., 2018). Common physicochemical parameters (moisture, pH, acidity, EC, HMF and ash) determined for the euphorbia honeys investigated in the study are listed in **(Table 2**).

In addition, it is inevitable that osmophilic yeast and other fungi will be included in honey during the foraging activities of bees, which must be processed to extend its shelf life. Therefore, heat application may be a useful alternative for moisture protection as appropriate treatment to prevent fermentation by controlling moisture content to inhibit the growth of fungi. However, heat treatment causes undesirable changes such as decrease in enzymes, increase in hydroxymethylfurfural (HMF) level, rapid loss of antioxidant activity, darkening of the color of honey and deterioration of its taste, resulting in a decrease in the quality of honey. Therefore, it is important to provide the optimum heating condition that can keep the moisture content below 20 g/100 g of honey and extend the shelf life of the honey without any negative effects on its properties (Wongsa et al., 2023). Although it depends on many factors, water content, which ranks second among the components of honey, is closely related to the maturity of honey (Seraglio et al., 2019).

The moisture contents of the samples examined vary within a small range. By looking at these values, it is possible to say that the honey samples are sufficiently mature. Because all values are below the maximum value allowed for moisture, which is 20 g per 100 g of moisture (Seraglio et al., 2019). The moisture content of 5 euphorbia honeys from the Algerian region is between 14-16.50%. The pH values of the mentioned honeys selected from the literature are in the range of 3.7-4.21, which is compatible with the current study (Dahmani et al., 2020).

Like humidity, Brix degree is an indicator of honey maturity (Dahmani et al., 2020). While the average Brix degree values measured for 5 euphorbia honeys from Algeria were reported as 83.30 on average (Dahmani et al., 2020), the values calculated for eight honeys in the current study are in the range of 81.56-83.09, giving an average of 82.46. Most of the total soluble solids measured at 20 °C

Table 2. Antioxidant enzyme activities of water (W) and methanol (M) extracts of euphorbia honeys

	FRAP (mg TXE/g honey)	TPC (mg GAE/g honey)	TFC (mg QTE/g honey)	DPPH (% inhibition)	SOD (% inhibition)
IW	0.283	0.333	0.056	22.2	31.48
IM	0.016	0.178	0.019	14.09	ND
IIW	0.329	0.366	0.057	18.3	37.31
IIM	0.022	0.211	0.047	10.91	0.27
IIIW	0.290	0.386	0.063	31.7	29.80
IIIM	0.027	0.240	0.045	22.84	6.11
IVW	0.332	0.387	0.059	33.8	34.22
IVM	0.023	0.216	0.027	24.87	11.03
VW	0.241	0.321	0.049	26.2	37.58
IVM	0.024	0.223	0.049	32.99	7.17
VIW	0.260	0.268	0.038	21.9	16.62
VIM	0.014	0.166	0.019	11.55	TE
VIIW	0.288	0.333	0.056	32.4	28.29
VIIM	0.017	0.181	0.017	26.40	TE
VIIIW	0.149	0.186	0.027	20.9	4.42
VIIIM	0.005	0.117	0.009	17.51	TE

and represented by the Brix degree correspond to total soluble sugars. According to the definition of the United States Department of Agriculture, a Brix degree above 80 means that the water content is below 20% and indicates that the honey is of high quality. Since the moisture content decreases with the increase in the total soluble solids level, it can be predicted that it will have better stability during storage (Albu et al., 2021). As a matter of fact, there is a highly correlated inverse relationship between the Brix degrees and moisture content of the tested honeys. In another study, The Brix degrees of two commercial honeys, whose botanical source was Euphorbiaceae, obtained from Morocco in 2021, with moisture values of 17.60 and 17.00, respectively, were determined as 80.67 and 81.13, respectively (Mehdi et al., 2023).

It is also known that the glucose/moisture (G/M) ratio is effective in the crystallization of honey. If the ratio in question is lower than 1.7, it is considered that crystallization is very slow or even not at all, but if it is higher than 2, it is considered to be quite fast (Seraglio et al., 2019). If we examine the G/M ratios in the table, we see that they are below the critical value. Moderate crystallization is expected to develop.

The pH, acidity and electrical conductivity of honey vary depending on the variability of the organic acids it contains. When acidity is mentioned, free, lactonic and total acidity come to mind. The maximum allowed value for free acidity in European legislation is 50 meq/kg. Acidity values below this value indicate that honey samples have not undergone unwanted fermentation and are still fresh (Silva et al., 2017). The free acidity value measured for Euphorbia resinifera A. Berger honey obtained from the Ben Slimane region was reported as 27.2 meq/kg, which is compatible with the current study (Aazza et al., 2014).

While low molecular weight aliphatic organic acids in balance with lactones, esters and some inorganic ions such as and phosphate are effective on free acidity, botanical source, mineral content and production time also affect the amount of free acidity (Navarrete et al., 2005; Seraglio et al., 2019). Proline value such as electrical conductivity (related to mineral content), free acidity and pH values. In addition to being used to classify honey, proline content is also an indicator of adulteration in honey, such as HMF and diastase number (Aazza et al., 2014).

The main sugars in honey are fructose and glucose. Although the composition of sugars in honey

varies according to plant flora, geographical conditions and climate also contribute significantly to the amount of sugar in honey. The average of fructose+glucose values determined by determining the glucose and fructose amounts of the examined euphorbia honeys was calculated as 68.3%. This calculated average value is higher than the 60% value, which is determined as the lowest value according to the CAC Honey standard, TFC Honey Communiqué and 2001/110/EC EU standards.

Besides fructose and glucose as monosaccharides, In studies reported from Morocco and Algeria, in addition to fructose and glucose monosaccharides, various disaccharides, mostly sucrose, maltose, turanose, trehalose and hybriditose, were also detected in Euphorbia honey (Sahri et al., 2023). According to melissopalynological analysis, euphorbia honey obtained from Diyarbakır region, which was found to contain a minimum of 42% and a maximum of 78% of the relevant pollen, had an average fructose content of 40% and a glucose content of 23.20%; and based on the F/G ratios calculated with these reported values, the authors comment that euphorbia honey is an ideal honey for the use of diabetic patients because honey with a high F/G ratio has low glycemic index values. (Kara et al., 2020). Sucrose was not detected in any of the tested samples, and this is consistent with the literature information that 3% sucrose was detected in only one of 10 euphorbia honeys. When evaluated in terms of maltose, which is a disaccharide, the maltose content of the samples was calculated to be between 0.5-1.1%. In the case of euphorbia honey supplied from Diyarbakır region, maltose content was reported as 1.5% in only one honey (Kara et al., 2020). The F/G ratio of euphorbia honeys, which were collected from the Diyarbakır region and in which 11 pollen types belonging to 10 botanical families were detected, with only 7.5% being euphorbia at the end of Melissopalynological Analysis, was calculated as 1.33, which is quite compatible with the current study (Özkök, 2019).

5-hydroxymethyl-2-furaldehyde (HMF), a furan-derived water-soluble heterocyclic organic molecule with both alcohol and aldehyde functional groups, is naturally present in small amounts in fresh honey. However, if honey is subjected to heat treatment and pasteurization, it also occurs as a result of the dehydration of sugars such as glucose and fructose in honey through the Maillard reaction. The color and taste of honey change after the interaction between proteins and sugars. HMF formation occurs as a result of keeping honey in an acidic environment for a long time, so the HMF value is evidence of overheating, spoilage and mixing with other sugars and syrups. Although fresh honey contains less than 15 mg/kg HMF, varying with factors such as pH, temperature and age, the maximum HMF limit is 40 mg/kg according to the Codex alimentarius standard. However, this limit value has been increased to 80 mg/kg for tropical honey. In the study conducted in the Moroccan region, HMF amounts in honey obtained from Euphorbia resinifera were found to be low, ranging from 1.5 to 40 mg/kg, meaning that the honey was subjected to appropriate processing and storage conditions and was fresh (Sahri et al., 2023). The HMF values detected in the range of 4-9.9 mg/kg for the euphorbia honeys examined in the current study indicate that all samples do not exceed the values allowed by international regulations. In the study conducted on honey obtained from euphorbia, strawberry tree and carob plants in the Moroccan market, it was reported that the amount of HMF was detected at low levels in samples other than carob honey (Mehdi et al., 2023). HMF was measured as 4.05 mg/kg in Euphorbia honey obtained from Mardin Midyat region (Pehlivan and Gül, 2016). Chakir et al. (2016) reported HMF values of 12.8 mg/kg and 20.32 mg/kg in Euphorbia echinus and Euphorbia resinifera honey, respectively. Diastase activity is also an indicator of freshness. If it is low, it is an indication that the honey has been subjected to heat treatment and adulteration (Sahri et al., 2023). The number of diastase reported for euphorbia honey obtained from the Mardin region is reported as 38.5, and it is seen that it is above 8, which is the minimum limit determined by the FAO / Food Codex, Turkish Food Codex Honey Communiqué and the European Union standard (Pehlivan and Gül, 2016). Although the same situation is valid for the euphorbia honeys detected in the current study, the average of the diastase values calculated for 8 honeys is 20.525. Some variables, such as botanical and biological sources, may also affect diastase activity. Diastase data in the range 11-103 have been reported for Euphorbia honey samples taken from various geographical regions (Sahri et al., 2023). The average diastase number of 5 euphorbia honeys from the Algerian region was reported as 15.18 (Dahmani et al., 2020).

The amount of proline, which is determined by the standards as a minimum of 180 mg/kg, is a parameter used to distinguish natural and adulterated honey (Da Silva et al., 2016). The fact that the proline contents of the tested euphorbia honeys were determined as 561.7 to 872. 7 mg/kg is proof that

these honeys are natural honey. In a study conducted on 47 samples, including Euphorbia honey samples, proline content values were reported in the range of 0. 35 -302.15 mg/kg (Ouradi et al., 2021).

pH is another important parameter for honey for the growth of microorganisms and fermentation. The organic acids contained in honey, ions such as phosphate and chloride, ionizable minerals and its botanical source are the factors on which pH depends. Although it is an important parameter in the quality control of honey, there is no limiting value in any legislation on this issue. However, since both depend on the amount of ions, a positive correlation between pH and electrical conductivity is inevitable (Seraglio et al., 2019). The correlation value observed between pH values and electrical conductivity measurements of the tested euphorbia honeys was calculated as 0.91, which is compatible with this literature information.

The electrical conductivity of honey varies depending on the amount of minerals, organic acids and proteins it contains. It also varies depending on the floral source of the honey, so it is a distinguishing parameter for classifying honey (Bettar et al., 2019). Electrical conductivity values of the eight samples tested were found to be between 249-376 µS/cm. These values are is compatible with the values of 227-874 µS/cm and 322-690 µS/cm given for E. officinarum subsp. echinus and E. regis-jubae honeys, respectively reported in the literature (Bettar et al., 2019). The average value of ten euphorbia honey samples, whose dominant pollen was determined as 45%, obtained from Diyarbakır region, was reported as 300 µS/cm (Kara et al., 2020).

The relationship between free acidity and electrical conductivity reveals the presence of organic ions and acids in honey (Pereira et al., 2020). However, in the current study, a high correlation that could indicate this relationship could not be calculated. The calculated correlation value is only 0.24. As a result, the biochemical properties and antioxidant activity of euphorbia honey were evaluated. It was found that the biochemical structure and antioxidant capacity of euphorbia honey differed from those of other honeys.

CONCLUSION

The results confirm that Euphorbia honey is a high-quality monofloral honey with measurable antioxidant capacity. It was found that the biochemical properties of the honey samples used in the study were comparable to those of other studies. However, it was found that the extract prepared with water from different solvents was more suitable in terms of antioxidant activity properties. The higher antioxidant activity observed in water extracts may indicate that hydrophilic bioactive compounds contribute substantially to the overall antioxidant potential of Euphorbia honey.

REFERENCES

Aazza S, Lyoussi B, Antunes D, Miguel MG (2014) Physicochemical characterization and antioxidant activity of 17 commercial Moroccan honeys. International Journal of Food Sciences and Nutrition, 65(4), 449-457.

Abderrahim LA, Taïbi K, Abderrahim NA, Boussaid M, Rios-Navarro C, Ruiz-Saurí A (2019) Euphorbia honey and garlic: Biological activity and burn wound recovery. Burns, 45(7), 1695-1706.

Afroz R, Tanvir EM, Hossain MM (2023) Physical Properties of Honey. Honey: Composition and Health Benefits, 12-31.

Albu A, Radu-Rusu CG, Pop IM, Frunza G, Nacu G (2021) Quality assessment of raw honey issued from eastern Romania. Agriculture, 11(3), 247.

Ansari H, Agnihotri M, Khan MS, Baweja HS, Rao KL, Nandan H (2023) Report on nutritional, antibiotic and physicochemical properties of honey from Garhwal region of Uttarakhand. The Pharma Innovation Journal 2023; 12(3): 4115-4120.

Beauchamp C, Fridovich I (1971) Superoxide dismutase:improved assays and an assay applicable to acrylamide gels. Analytical Biochemistry, 44, 276–287.

Benov L (2001) How superoxide radical damages the cell. Protoplasma, 217, 33-36.

Bettar I, González-Miret ML, Hernanz D, Marconi A, Heredia FJ, Terrab A (2019) Characterisation of Moroccan Spurge (Euphorbia) honeys by their physicochemical characteristics, mineral contents and colour. Arabian Journal of Chemistry, 12(8), 2052-2060.

Bogdanov S, 2009 Physical properties of honey. In: Book of Honey, Chapter 4. Bee Product Science.

Boutoub O, Ghadraoui LE, Miguel MG (2022) Biological properties of latex, aqueous extracts and bee products of Euphorbia officinarum L.: A short review. *Molecules*, *27*(21), 7200.

Bouhlali EDT, Bammou M, Sellam K, El Midaoui A, Bourkhis B, Ennassir J, ... & Filali-Zegzouti Y (2019) Physicochemical properties of eleven monofloral honey samples produced in Morocco. Arab Journal of Basic and Applied Sciences, 26(1), 476-487.

Chakir A, Romane A, Marcazzan GL, Ferrazzi P (2016) Physicochemical properties of some honeys produced from different plants in Morocco. Arabian Journal of Chemistry, 9, S946-S954.

Council EU (2002) Council Directive 2001/110/EC of 20 December 2001 relating to honey. Official Journal of the European Communities L, 10, 47-52.

Dahmani K, Houdeib JB, Zouambi A, Bendeddouche B, Fernández-Muiño M, Osés SM, Sancho MT (2020) Quality attributes of local and imported honeys commercialized in Algeria. Journal of apicultural science, 64(2), 251-262.

Da Silva PM, Gauche C, Gonzaga LV, Costa ACO, Fett R (2016) Honey: Chemical Composition, Stability and Authenticity. Food Chemistry, 196, 309-323.

Fridovich I (1981) Superoxide radical and superoxide dismutases. Oxygen and Living Processes: An Interdisciplinary Approach, 250-272.

Fridovich I. (1995) Superoxide radical and superoxide dismutases. Annu Rev Biochem 64:97-112.

Fukumoto LR, Mazza G (2000) Assessing antioxidant and prooxidant activities of phenolic compounds. Journal of Agricultural and Food Chemistry, 48(8), 3597–3604.

Goodarzi B, Khosravi A (2013) The Effects of simultaneous 8 weeks Astragalus sp/Euphorbia Cheriradenia Honey supplementation and endurance training on membrane lipid peroxidation of erythrocytes after a bout acute exhaustive treadmill exercise in rats. European Academic Research, 1(2), 114-137.

Gül A, Pehlivan T (2018) Antioxidant activities of some monofloral honey types produced across Turkey. Saudi Journal of Biological Sciences, 25(6), 1056-1065.

Kara Y, Birinci C, Birinci E, Can Z (2020) Characteristic Properties of Spurge (Euphorbia macroclada Boiss.) Honey in Diyarbakır Region. Journal of Apitherapy and Nature, 3(1), 37-43.

Liu M, Yao X, Zhang G, Yi S, Huang X, Ning F, Luo L (2023) Physicochemical properties, chemical constituents, and antioxidant activity of Eurya honey. International Journal of Food Science & Technology, 58(6), 3458-3468.

Mărgăoan R, Topal E, Balkanska R, Yücel B, Oravecz T, Cornea-Cipcigan M, Vodnar DC (2021) Monofloral honeys as a potential source of natural antioxidants, minerals and medicine. Antioxidants, 10(7), 1023.

Mehdi R, Zrira S, Vadalà R, Nava V, Condurso C, Cicero N, Costa R (2023) A preliminary investigation of special types of honey marketed in Morocco. Journal of Experimental and Theoretical Analyses, 1(1), 1-20.

Moujanni A, Partida L, Essamadi AK, Hernanz D, Heredia FJ., & Terrab A (2018) Physicochemical characterization of unique unifloral honey: Euphorbia resinifera. CyTA-Journal of Food, 16(1), 27-35.

Monteiro M, Santos RA, Iglesias P, Couto A, Serra CR, Gouvinhas I, Barros A, Oliva-Teles A, Enes P, Díaz-Rosales P (2020) Effect of extraction method and solvent system on the phenolic content and antioxidant activity of selected macro-and microalgae extracts. Journal of Applied Phycology, 32, 349-362.

Muflihah YM, Gollavelli G, Ling YC (2021) Correlation study of antioxidant activity with phenolic and flavonoid compounds in 12 Indonesian indigenous herbs. Antioxidants, 10(10), 1530.

Mir B, Mushtaq A, Rizwan S, Bashir F, Behlil F, Afzal S (2022) Comparison of Antioxidant Enzymes in Natural and Commercial Honey. Pak-Euro Journal of Medical and Life Sciences, 5(1), 31-38.

Nagai T, Sakai M, Inoue R, Inoue H, Suzuki N (2001) Antioxidative activities of some commercially honeys, royal jelly, and propolis. Food chemistry, 75(2), 237-240.

Navarrete M, Casado S, Minelli M, Segura A, Bonetti A, Dinelli G, Fernández A (2005) Direct determination of aliphatic acids in honey by coelectroosmotic capillary zone electrophoresis. Journal of Apicultural Research, 44(2), 65-70.

Ouradi H, Hanine H, Fauconnier ML, Kenne T, Rizki H, Ennahli S, Hssaini L (2021) Determination of physico-biochemical proprieties and composition in volatile constituents by solid phase micro-extraction of honey samples from different botanical and geographical origins in Morocco. Journal of Apicultural Research, 60(1), 84-98.

Oyaizu M (1986) Studies on product of browning reaction prepared from glucose amine. Japanese Journal of Nutrition and Dietetics, 44(6), 307–315.

Özkok A (2019) Determination of antioxidant capacities, chemical composition and melissopalynological characterization of Verbascum spp. and Euphorbia spp. honeys. Fresenius Environmental Bulletin, 28(4), 2644-2649.

Özkırım A, Schiesser A, Küçüközmen B, İpek A, & Sorkun K. (2021) Antimicrobial activitiy of oak honey (Quercus spp.) on the biofilm microorganisms. Ege Üniversitesi Ziraat Fakültesi Dergisi, 58(2), 203-210.

Pereira JR, Campos ANDR, de Oliveira FC, Silva VR, David GF, Da Silva JG, Nascimentoa WWG, Silvab MHL, Denadai ÂML (2020) Physical-chemical characterization of commercial honeys from Minas Gerais, Brazil. Food Bioscience, 36, 100644.

Pehlivan T, Gül A (2016) Türkiye'de üretilen keçiboynuzu, kekik ve sütleğen ballarının kimyasal özellikleri. Mustafa Kemal Üniversitesi Ziraat Fakültesi Dergisi, 21(1).

Ricciardelli d'Albore G (1997) Textbook of melissopalynology. Apimondia, Bucharest.

Sahri N, Alaoui A, Elherradi E, Mahmoud MF, Louaradi D, Sobeh M (2023) Euphorbia honey: a comprehensive compile of its traditional use, quality parameters, authenticity, adulteration, and therapeutic merits. Phytochemistry Reviews, 1-28.

Sá, F. V. d. S., Geovani, S. d. L., João, B. d. S., Hans, R. G., Lauriane, A. d. A. S., Lourival, F. C., Leandro, d. P. S. (2016). Growth and physiological aspects of bell pepper (Capsicum annuum) under saline stress and exogenous application of proline. African Journal of Biotechnology, 15(36), 1970-1976. https://doi.org/10.5897/ajb2016.15441

Seraglio SKT, Silva B, Bergamo G, Brugnerotto P, Gonzaga LV, Fett R, Costa, ACO (2019) An overview of physicochemical characteristics and health-promoting properties of honeydew honey. Food Research International, 119, 44-66.

Serem JC, Bester MJ (2012) Physicochemical properties, antioxidant activity and cellular protective effects of honeys from southern Africa. Food Chemistry, 133(4), 1544–1550.

Silva LR, Sousa A, Taveira M (2017) Characterization of Portuguese honey from Castelo Branco region according to their pollen spectrum, physicochemical characteristics and mineral contents. Journal of Food Science and Technology, 54, 2551-2561.

Terrab A, Moujanni A, Essamadi AK, Hernanz D, Díez MJ, Berjano R (2022) A palynological and geographical characterization of labeled resin spurge honey: Euphorbia resinifera. Palynology, 46(1), 1-10.

Wongsa K, Meemongkolkiat T, Duangphakdee O, Prasongsuk S, Rattanawannee A (2023) Physicochemical Properties, Phenolic, Flavonoid Contents and Antioxidant Potential of Stingless Bee (Heterotrigona Itama) Honey From Thailand. Current Research in Nutrition & Food Science, 11(1).