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Characterization of Total Phenol and Flavonoid Contents, Colour, Functional Properties from Honey Samples with Different Floral Origins

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Abstract

Honey has long been used as a food and has been reported to have potential health benefits. In this work, total phenol content, colour and antioxidant and hepatoprotective activities of honey samples of different floral origins from the State of Hidalgo, Mexico were explored using *in vitro* assays. Hepatoprotective activity was measured by inhibitition of β-glucuronidase; gastroprotective activity was determined by inhibition of urease; antioxidant activity was evaluated by 2,2'-Azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS) and 2, 2-diphenyl-1-picrylhydrazyl (DPPH) methods. All the parameters showed significant differences ($p<0.05$) among honey samples. The total phenolic content ranged from 18.02 to 102.77 mg GAE 100 g^{-1}. The colour ranged from extra light amber to dark amber. Inhibition of β-glucuronidase ranged from 23.70% to 36.00%, while urease inhibition ranged between 7.64% and 63.80%. The antioxidant activity by ABTS was between 44.68 and 441.56 mg AAE 100 g^{-1}, and DPPH showed activities ranging from 35.64 to 573.06 mg AAE 100 g^{-1}. All honey samples contained bioactive compounds and displayed functional properties; therefore, the honeys from this region of Mexico offer attractive characteristics for their potential use in the food industry.

Keywords: Phenols; Flavonoids; Colour; β-glucuronidase; Urease

1 Introduction

Honey is a natural substance produced by honeybees, of the species *Apis mellifera*, from the nectar of flowers and plant secretions. This natural product has several food and clinical applications and contains approximately 200 different chemical components, including fructose, glucose, water, proteins, amino acids, enzymes, vitamins and minerals (Alvarez-Suarez et al., 2010; Rao et al., 2016). Honey is an important food product since it contains bioactive compounds derived from plants and the bees that produce it: honey is rich in phenolic acids and flavonoids, which have a wide range of biological functions and act as natural antioxidants (Kaskoniene et al., 2009). The colour of honey is one of its most distinctive characteristics, and is used as an in-

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dication of the presence of phenolic compounds and their derivatives and their potential antioxidant activities (Estevinho et al., 2008).
There are several reports that suggest that the composition and antioxidant capacity of different honeys depend mainly on the floral origin of the nectar collected by bees, other environmental factors and even processing. These factors demonstrate that there is a correlation between the origin and the identified components of the honey (Baharudin et al., 2017; Marghitas et al., 2009; Pimentel-Gonzalez et al., 2016). Recent research indicates that the main factor that gives diversity of colour, flavour and functional properties to honey is its phenolic composition (Hossen et al., 2017). These compounds demonstrate biological activity that have beneficial effects on health (Bueno-Costa et al., 2016; Estevinho et al., 2008). Phenolic compounds and flavonoids can give insight into the botanical origin of honey; therefore, the phenolic composition and antioxidant capacity of honey depend on the floral sources used to obtain this honey, as well as seasonal and environmental factors (Yao et al., 2003).
Current trends show that there are studies using natural compounds aimed at tumour and cancer inhibition; to reduce the need for chemical therapies and surgical interventions (Khan et al., 2017). Polyphenols in honey can chemically prevent different stages of cancer, including tumour formation and growth, through the inactivation of carcinogenic compounds, cell cycle arrest, apoptosis induction, the reduction of angiogenesis and antioxidation (Fresco et al., 2006).
An important function of the bioactive compounds found in honey is prevention of loss of biological membranes and enzymes functionality by elimination of free radicals that would otherwise induce the oxidation of lipids, proteins and DNA, in living systems (Mouhoubi-Tafinine et al., 2016; Niki, 2011). Various plant species have been studied for their capacity to inhibit certain enzymes to confer health benefits. The hepatoprotective effect is attributed to the inhibition of the β-glucuronidase enzyme (Karak et al., 2017), and gastroprotective activity is related to urease inhibition (Biglar et al., 2012). This study was conducted on honey samples with different floral origins by measuring the total phenol and flavonoid contents, the colour, and the functional properties such as hepatoprotective, gastroprotective and antioxidant activities.

2 Materials and Methods

2.1 Honey samples

A total of 24 samples of honey from different regions of the state of Hidalgo, Mexico were obtained directly from beekeepers and evaluated. The samples were collected in the period March-April 2018 from 24 different sites (Table 1). The collection was carried out at room temperature (20-25 ^{o}C) and the samples were later taken to the Institute of Agricultural Sciences of the Autonomous University of Hidalgo State in closed, dark containers. The honey samples were coded and stored in the dark at room temperature until they were analysed. The floral origin of the honey samples was determined according to the methodology developed by Sodre et al. (2007). For a quantitative analysis of pollen, 300 pollen grains were used for each sample in triplicate. Subsequently, visual comparisons of pollen were made using a BX 45 light microscope (Olympus, Japan).

2.2 Total phenolic content

The phenolic content in honey samples was determined using the Folin-Ciocalteu method (Singleton et al., 1999). Honey was diluted 1:10 with distilled water, and the resulting solution was filtered using Whatman No. 1 paper filter. The filtered solution (5 mL) was mixed with 2.5 mL of 0.2 N Folin-Ciocalteu phenol reagent for 5 minutes, and then 2 mL of Na_2CO_3 solution (75 g L^{-1}) was added. Samples were left to stand for 2 h at room temperature in the dark. After this time, the absorbance was read at 760 nm in a Hitachi U-2000 UV/VIS spectrophotometer (Tokyo, Japan), using water as a blank. A standard curve was prepared with gallic acid in solutions of different concentrations (0-100 mg of gallic acid 100g^{-1}); therefore, results of the phenolic content in honey samples were expressed as mg of gallic acid equivalents (GAE) 100 g^{-1} of honey.

Table 1: Main characteristics of Mexican honey samples

Honey	Collection site	Floral origin
1SA	San Andrés	Cactus
2SF	San Felipe Orizatlán	Cítrus
3AC	Acaxochitlán	Gramineae
4ER	El Real	Conífers
5AT	Atotonilco	Juglans
6HU	Huejutla	Cítrus
7TE	Tehuetlán	Multifloral
8HU	Huautla	Multifloral
9AM	Tenango de Doria	Multifloral
10NR	Tepeji del Rio	Multifloral
11WR	Acatlán	Multifloral
12VZ	Zimapán	Multifloral
13ML	San Felipe Orizatlán	Cítrus
14AB	San Bartolo Tutotepec	Multifloral
15OS	Huejutla	Cítrus
16FA	Zimapán	Multifloral
17JG	Almoloya	Multifloral
18HA	Texcoco	Multifloral
19JJ	Almoloya	Multifloral
20ME	Tepeapulco	Multifloral
21MA	Apan	Multifloral
22VM	Apan	Multifloral
23OR	Zimapán	Multifloral
24MJ	Tlanalapa	Multifloral

2.3 Flavonoid content

The total flavonoid content was measured using the Dowd method as reported by Meda et al. (2005). Approximately 1 g of each honey sample was mixed with 10 mL of pure methanol, and the solution was stirred and then centrifuged at 15,000 rpm for 15 minutes. Then, 2 mL of the suspended liquid was mixed with 2 mL of 2% aluminium trichloride ($AlCl_3$) and left to stand in the dark for 20 minutes. The absorbance was measured at 415 nm in a UV/VIS spectrophotometer (Hitachi U-2000, Tokyo, Japan). A standard curve was prepared of quercetin in different concentrations (0-100 mg of quercetin $100g^{-1}$); therefore, the flavonoid content was expressed in mg of quercetin equivalents (EQ) 100 g^{-1} of honey.

2.4 Colour determination using the Pfund scale

Honey colour was determined by reading the absorbance of a 50% honey solution in water (w/v) read at 635 nm in a 1 mL cell, in a UV/VIS spectrophotometer (Hitachi U-2000) following the method previously reported by Ferreira et al. (2009). The honey samples were classified, after obtaining the readings, using the Pfund scale by converting the absorbance values, applying equation 1 and comparing the results with the honey colour chart.

$$mmPfund = -38.70 + (371.39 \times absorbance) \quad (1)$$

2.5 Antioxidant Activity

2,2'-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS)

The antioxidant activity was determined by the discolouration of the ABTS radical (Sigma-Aldrich, Canada), as described by Re et al. (1999): 7 μM ABTS radical was reacted with 2.45 μM potassium persulfate ($K_2S_2O_8$) (Mallinckrodt Chemicals, USA) in a 1:1 ratio and stirred for 16 hours in the dark. When the radical was formed, it was diluted with ethanol (20%) until reaching an absorbance of 0.7 $\pm$ 0.01 at 734 nm. Once the radical was stabilized, 3 mL of this solution was placed in a test tube, and 100 μL of honey extract was added. The mixture was stirred rapidly, and the absorbance was measured after 10 minutes of reaction. The absorbance of the ethanol (20%)-diluted solution was used as the blank. A standard curve was prepared with ascorbic acid at different concentrations (0-100 mg of ascorbic acid $100g^{-1}$); therefore, the results were expressed in milligrams of ascorbic acid equivalents (AAE) 100 g^{-1} of honey.

2,2-diphenyl-1-picrylhydrazyl (DPPH)

To prepare the 0.2 mM DPPH solution, 7.8 mg of DPPH radical (Sigma-Aldrich, USA) was weighed and dissolved in 100 mL of methanol (80%), and the mixture was stirred in the dark for two hours for complete dissolution. Then, 2.5 mL of methanolic DPPH solution was added to a test tube, reacted with 0.5 mL of honey solution, and the mixture was left in the dark for 30 minutes and read at an absorbance of 515 nm. The antioxidant activity was determined using a standard curve with ascorbic acid. The results were expressed in milligrams AAE 100 g-1 of honey (Brand-williams et al., 1995).

2.6 β-Glucuronidase inhibition

This analysis was carried out following the methodology described by Karak et al. (2017) with a few modifications. A dilution of the honey with water was made at 10%, from which 340 μL was taken and mixed with 100 μL of a β-glucuronidase solution (986.4 units mL^{-1} in pH 7 phosphate buffer) (Sigma-Aldrich, USA), and the mixture was pre-incubated at 37 ^{o}C for 15 minutes. Subsequently, 60 μL of p-nitrophenyl-β-D-glucuronide (3.15 mg mL^{-1} in pH 7 phosphate buffer) (Sigma-Aldrich, USA) was added and incubated for 50 minutes at 37^{o}C. The absorbance was measured at 405 nm in a spectrophotometer. A control test was carried out with D-saccharic acid 1,4-lactone following the same steps as analysis of honey samples, and the percentage inhibition was obtained using equation 2:

$$\%Inhibition = \left[\frac{absorbance\ of\ Control - absorbance\ of\ Sample}{absorbance\ of\ Control}\right] \times 100 \quad (2)$$

2.7 Urease inhibition

The urease inhibition test was performed as described by Biglar et al. (2012) with a few modifications. For each honey sample, 100 μL of urease solution (Jack bean urease 5 U mL^{-1}) and 100 μL of inhibitor (a 20% dilution of honey in a 50% ethanol-water solution) were mixed and incubated at 37^{o}C for 10 minutes. Then, a solution of urea prepared in phosphate buffer (pH 6.8) was added and incubated at 37^{o}C for 30 minutes. The urease activity was determined by measuring the production of ammonia using the indophenol method. The reaction was stopped with 600 μL of 4% sulfuric acid. Subsequently, 100 μL of solution A (5.0 g phenol and 25 mg of sodium nitroprusside) and 100 μL of solution B (2.5 g sodium hydroxide and 4.2 mL of sodium hypochlorite in 500 mL of distilled water) were added, the samples were left in darkness for 30 minutes at 37^{o}C, and the absorbance was measured at 625 nm.

2.8 Statistical analysis

For the statistical analysis, a completely randomized design was used. The results were interpreted with analysis of variance, and when significant differences were observed ($p<0.05$) between the different honeys, the mean comparison was performed by the Tukey method using the NCSS 2007 software (USA).

Table 2: Bioactive compounds and honey color

Honey	Total phenols	Flavonoids	Pfund Scale (mm)	Color
1SA	$25.65\pm0.0.49^{c}$	3.95 ± 0.20^{ijk}	105.3	Amber
2SF	18.02 ± 0.49^{a}	4.97 ± 0.52^{l}	71.8	Light amber
3AC	92.32 ± 1.29^{o}	7.63 ± 0.51^{n}	93.6	Amber
4ER	93.16 ± 0.98^{o}	7.01 ± 0.52^{mn}	135.8	Dark amber
5AT	102.49 ± 0.98^{p}	1.28 ± 0.26^{a}	88.7	Amber
6HU	93.73 ± 0.85^{o}	2.02 ± 0.34^{abc}	69.2	Light amber
7TE	102.77 ± 1.29^{p}	3.49 ± 0.52^{ghij}	138.1	Dark amber
8HU	51.10 ± 0.49^{k}	1.79 ± 0.20^{ab}	65.9	Light amber
9AM	43.10 ± 0.97^{ij}	2.25 ± 0.10^{bcd}	42.9	Extra light amber
10NR	49.44 ± 0.33^{k}	6.33 ± 0.07^{m}	110.7	Amber
11WR	38.46 ± 0.33^{fg}	2.37 ± 0.14^{bcde}	59.6	Light amber
12VZ	72.95 ± 0.72^{n}	5.42 ± 0.16^{l}	125.6	Dark amber
13ML	57.30 ± 0.41^{l}	2.77 ± 0.07^{cdefg}	26.2	White
14AB	44.26 ± 0.23^{j}	3.24 ± 0.23^{fghi}	81.8	Light amber
15OS	64.08 ± 0.23^{m}	5.37 ± 0.09^{l}	64.7	Light amber
16FA	71.58 ± 0.22^{n}	3.56 ± 0.21^{hij}	242.3	Dark amber
17JG	35.65 ± 0.38^{e}	3.19 ± 0.07^{efgh}	140.7	Dark amber
18HA	22.34 ± 0.54^{b}	2.11 ± 0.05^{abc}	40.3	Extra light amber
19JJ	33.02 ± 0.62^{d}	3.04 ± 0.09^{defgh}	233.2	Dark amber
20ME	41.10 ± 0.17^{hi}	4.51 ± 0.05^{kl}	122.6	Dark amber
21MA	36.68 ± 0.711^{ef}	2.94 ± 0.07^{cdefgh}	111.5	Amber
22VM	24.33 ± 0.17^{bc}	2.30 ± 0.04^{bcd}	86.1	Amber
23OR	39.15 ± 0.35^{gh}	4.35 ± 0.07^{jk}	175.1	Dark amber
24MJ	41.50 ± 0.49^{i}	2.44 ± 0.29^{bcdef}	100.2	Amber

The results are expressed in means ± standard deviation. Different letters indicate statistically significant differences between the analyzed samples ($p<0.05$). Total phenols are expressed in mg GAE 100 g^{-1}. Flavonoids are expressed in mg QE 100 g^{-1}.

3 Results and Discussion

3.1 Total phenolic content

Significant differences ($p<0.05$) were found between the different honey samples. The samples with the highest phenolic contents were 5AT and 7TE (of Juglan and multifloral origin, respectively), of 102.49±0.98 and 102.77±1.29 mg EAG 100 g^{-1}, respectively (Table 2). Various analyses of the total phenolic content in different honey samples have been carried out worldwide. Our results are similar to those reported by Ruiz-Navajas et al. (2011) who found values ranging between 51.32 and 134.02 mg EAG 100 g^{-1} in honey from south eastern Mexico with warm and tropical climates. The difference in the content of total phenols among the samples analysed is due to the diverse geographical origin, quality and the floral source from which they were extracted (Al-Farsi et al., 2018).

3.2 Flavonoid content

The results for flavonoid revealed that the honey samples with the highest content were 3AC and 4ER both with more than 7 mg EQ 100 g^{-1} (of Gramineae and Conifers origin, respectively). It should be mentioned that none of the samples had values less than 1 mg EQ 100 g^{-1} or more

than 10 mg EQ 100 g^{-1}. The results obtained in this study are comparable to those presented by Meda et al. (2005) in Burkina Faso honey samples with an average flavonoid content of 2.57 EQ 100 g^{-1}, similar to the samples with the lowest content in this study. A study conducted in Brazil by Bueno-Costa et al. (2016) in which they also determined flavonoid content obtained a range of results between 2.98 and 10.46 mg EQ 100 g^{-1}, very similar to the results presented here; however, the upper concentration limit in this Brazilian honey was slightly higher than that obtained in our samples. Unlike the studies mentioned above, Boussaid et al. (2018) reported very low content compared to the other results. Flavonoids are phenolic compounds and are present as various types of structures in honey; among them, the one with the highest content according to Alvarez-Suarez et al. (2012) is quercetin, while Quintero-Lira et al. (2017) reported the major flavonoid was rutin. The concentrations of these compounds are determined by the botanical and geographical origins of the samples, as well as the climate and environmental conditions such as humidity, temperature and type of soil where the plants grew (Al-Farsi et al., 2018). Flavonoid content and pigments derived from these compounds are also directly related to the colour of honey and its different shades (Alvarez-Suarez et al., 2010). Also, phenolic compounds and flavonoids can be used as an indicator of antioxidant activity since they have sequestering and free radical reduction capacity (da Silva et al., 2016).

3.3 Colour evaluation

The results showed that the darkest honey samples classified as "dark amber" were 4ER, 7TE, 12VZ, 17JE, 19JJ, 20ME, 23OR and 16FA (mainly of Conifers and Multifloral origin), with the highest level on the scale reaching 242.3 mm. Most samples had dark tones and fell under the "amber" classification or higher (Table 2). Compared to the results of honey colour in other studies, the honey colour in the samples analysed here showed large variations, with very high to low values on the Pfund scale that depended on when the colour analysis was performed. An analysis performed in Brazil by Pontis et al. (2014) identified honey samples classified as very clear "extra light amber"; however, the study also had samples within the "dark amber" range but with values lower than those found in our samples. Many authors agree that honey colour is directly related to the presence of phenolic compounds and their antioxidant capacity (Bertoncelj et al., 2007; Ferreira et al., 2009) due to the high correlation values in the tests performed. Therefore, the colour, smell and taste of honey are related to the phenolic content and are indicators and characteristics of floral origin (Bertoncelj et al., 2007; Kaskoniene et al., 2009).

3.4 Antioxidant activity

The antioxidant activity in the honey samples was determined by the ABTS and DPPH methods, obtaining results in equivalent milligrams of ascorbic acid (Figure 1). In both cases, significant differences ($p < 0.05$) were found between the analysed samples. In the ABTS test, the highest content were found in the following honey sample: 13ML (405.10±30.83 mg EAA 100 g^{-1}) of a citrus floral origin, and 24MJ (410.31±3.12 mg EAA 100 g^{-1}) and 17JG (441.56±6.25 mg EAA 100 g^{-1}), both of multifloral origin.
In DPPH test the highest content were found in the 12VZ honey sample, of multifloral origin (573.06±10.20 mg EAA 100 g^{-1}), showing significant differences with respect to the rest of the honey samples. Several studies have presented information on the antioxidant activity of honey. El-haskoury et al. (2018) performed tests on honey from Morocco and found average results of 60.94 mg EAA 100 g^{-1}, which is lower than the results obtained in this study; however, they obtained superior antioxidant activity in the ABTS test in relation to DPPH. A Portuguese honey was analysed by Ferreira et al. (2009) using DPPH test, and reported values between 106.67 and 168.94 mg EAA 100 g^{-1} with few samples being characterised as light honey and more as dark honey; a similar trend was also found in a study reported by Estevinho et al. (2008) with similar samples from Portugal. The results of antioxidant activity are directly related to the total phenol content in honey, as determ-

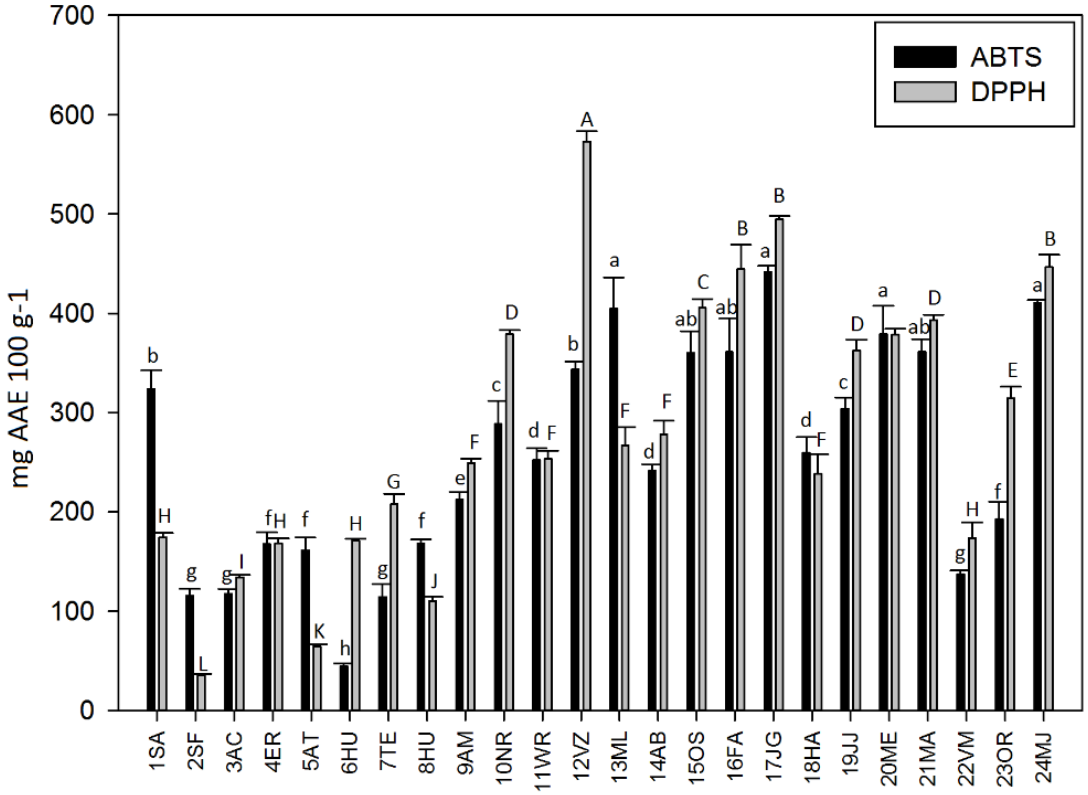

Figure 1: Antioxidant activity of Mexican honeys. The results are expressed in means ± standard deviation. Lowercase letters indicate differences between treatments for the ABTS test ($p<0.05$). Capital letters indicate differences between treatments for the DPPH test ($p<0.05$).

ined by Bertoncelj et al. (2007), who found high correlation factors between both tests, as with the colour intensity.

Based on the determination of the ABTS and DPPH tests performed by Bueno-Costa et al. (2016), it was found that the most appropriate test for antioxidant determination is ABTS since it shows high correlations with the content of phytochemical analytes such as phenols, flavonoids, phenolic acids and carotenoids. The literature mentions that the mechanisms of antioxidant activity in honey include the ability to sequester free radicals, the chelation of metal ions and the oxidizing-reducing capacity of phenolic compounds due to the presence of hydroxyl groups attached to aromatic rings (Bastos & Sampaio, 2013). It is also known that the antioxidant capacity and the colour of honey are affected by the floral source, differences in geographical origin, humidity, soil type and post-harvest conditions (Al-Farsi et al., 2018).

3.5 β-Glucuronidase inhibition

The results shown in figure 2 were obtained from the β-glucuronidase inhibition test. All the honey samples have positive inhibition values higher than 20%. The following samples had the highest percentages: 8HU (33.21±1.96%), 1SA (33.64±1.83%) and 2SF (36.00±0.47%) (mainly

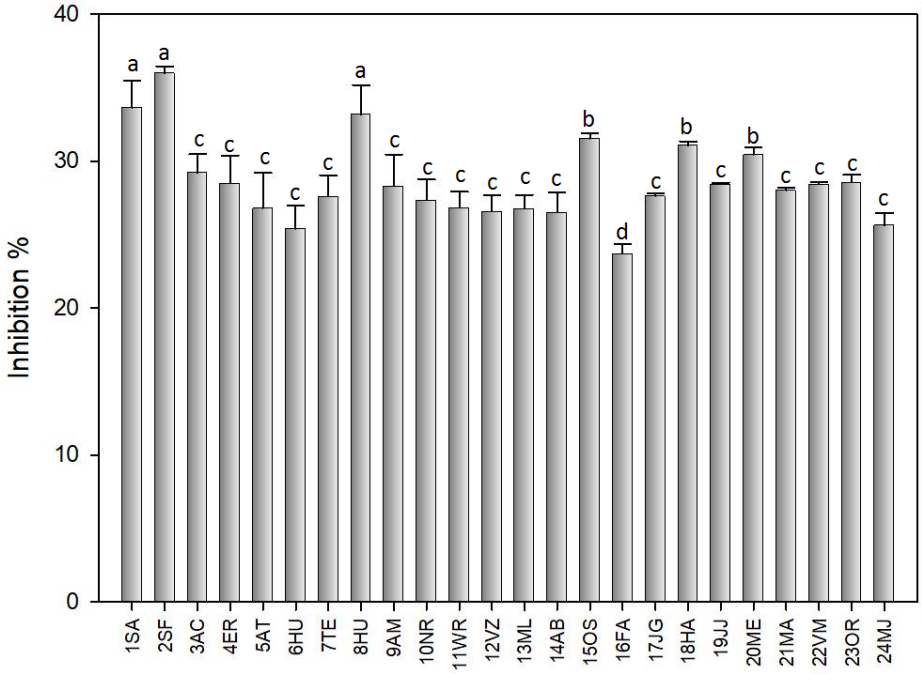

Figure 2: Inhibition percent of β-glucuronidase in Mexican honeys. The results are expressed in means ± standard deviation. Different letters indicate differences between treatments ($p<0.05$).

of cactus, citrus and multifloral origin). The results are similar between different samples that range between 25% and 30%, with some honey samples exhibiting higher peaks. The analyses that have been performed on the inhibition of β-glucuronidase by bioactive compounds are limited. Han et al. (2005) evaluated the hepatoprotective effect of lactic acid bacteria with an inhibition range of 17% to 66% in an in vivo study with rats. *Swertia sp.* plant extracts have been studied as a β-glucuronidase inhibitor; Karak et al. (2017) found an average inhibition percentage of 30.89% from a concentration of 500 μg mL^{-1} of the *S. bimaculate*.

β-Glucuronidase is used as a marker of various diseases because it is a member of the lysosomal glucosidase family, which catalyses the degradation of glycosaminoglycans of normal and cancerous tissue cell membranes (Baharudin et al., 2017). Glucuronic acid conjugates with toxins and diminishes their potency by eliminating them from the body later; this process is limited by high levels of β-glucuronidase (Karak et al., 2017). Liver damage causes an increase in the level of β-glucuronidase in the blood, and liver cancer may be related to this enzyme. Inhibitors of this indicator reduce the carcinogenic potential of toxic compounds. Therefore, inhibitors of β-glucuronidase are suggested as potential hepatoprotective agents (Shim et al., 2000). Some extracts and compounds from plants with hepatoprotective activities are known for their ability to inhibit this type of enzyme, among which are flavonoids (Joshi & Priya, 2007; Kim et al., 1994).

3.6 Urease inhibition

All honey samples exhibited urease inhibition, exceeding 20% in most samples (Figure 3). A clear variation was seen among the samples analysed, showing significant differences ($p<0.05$). Sample 10NR (63.80±0.11%) had the greatest inhibition (of multifloral origin), followed by 15OS (58.73±0.62%) and 4ER (59.24±3.64%). Cur-

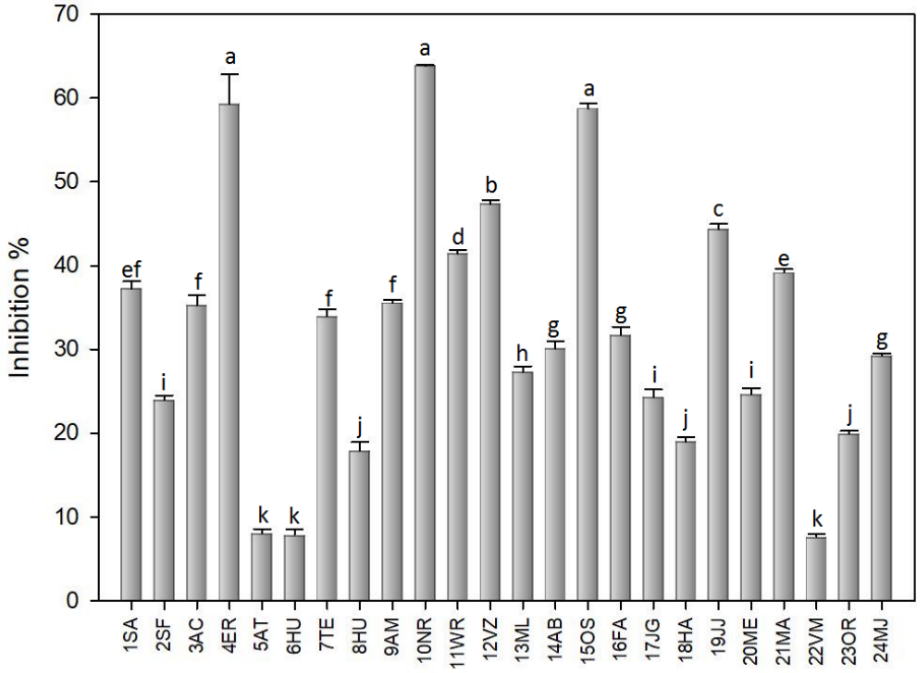

Figure 3: Inhibition percent of urease in Mexican honeys. The results are expressed in means ± standard deviation. Different letters indicate differences between treatments ($p<0.05$).

rent research on urease inhibition is focused on gastric protection and ulcer reduction; however, most studies are based on plant extracts and isolates. On the one hand, Lateef et al. (2012) analysed *Glycyrrhiza glabra* roots and found an approximate average urease inhibition value of 61% with a methanolic extraction and 40% in an extraction with chloroform. These results are within the range of those found in honey. On the other hand, Lin et al. (2005) evaluated the enzymatic inhibition of urease in oregano, and found an approximate value of 40%, which also coincides with results observed here. In the same study, the activity of cranberry was also evaluated, resulting in 9% inhibition.

The study on the inhibition of urease is aimed at determining the presence of *Helicobacter pylori* since this gram-positive bacterium causes chronic gastritis and intervenes directly in the formation of peptic ulcers, carcinoma and gastric lymphoma (Dunn et al., 1997). The gastric mucosa is affected by *H. pylori* since it resists the acidic environment producing urease, which hydrolyses urea-forming bicarbonate and ammonium (Hu & Mobley, 1990; Kuhns et al., 2013). The methanolic extracts of vegetable sources can be considered a natural source for the inhibition of urease, as mentioned by Lateef et al. (2012), so it is possible that these compounds can be transferred to honey, generating a similar effect. The difference in the enzymatic inhibitory activity is mainly due to the type of phenolic compounds in the inhibitor, as verified by Lin et al. (2005), who demonstrated that hydrophobic phenolic compounds such as those present in oregano are superior in urease inhibition than those that are soluble in water, such as those found in blueberries. In honey, phenolic derivatives such as flavonoids behave both ways since they are liposoluble and water soluble (Martínez-Flórez et al., 2002), this is reflected in the varied behaviour of urease inhibition.

4 Conclusion

The honey samples analysed in this study were a good source of phenolic compounds, especially the honey of walnut and multifloral origin, and had good antioxidant activity, as in the case of honeys 17JG and 12VZ (multifloral). β-glucuronidase inhibition values indicated the presence of possible hepatoprotective activity, and some honey samples showed high values of possible gastric protective activity as exhibited by the urease inhibition test *in vitro*. Differences between the honey samples in each test demonstrated that the characteristics of each honey sample were unique, probably due to the compounds they contain derived from different plant origins. This study provides an insight to potential area of opportunity for future research related to the possible beneficial effects of honeys in the region studied, which the native population uses even in traditional medicine.

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