



# Properties of Some Commercial honeys Available in Mexican Market: Effect of Overheating on Quality of the Packaged Honey

P.M. Mondragón-Cortez<sup>1</sup>, G.M. Guatemala-Morales<sup>1</sup>, E. Arriola-Guevara<sup>2\*</sup>✉

1. Centro de Investigación y Asistencia en Tecnología y Diseño del Estado de Jalisco A. C. (CIATEJ). Av. Normalistas 800, Col. Colinas de la Normal, P.C. 44270, Guadalajara Jalisco, México

2. Universidad de Guadalajara, Centro Universitario de Ciencias Exactas e Ingenierías, Departamento de Ingeniería Química. Boulevard Marcelino García Barragán 1412, P.C. 44430, Guadalajara, Jalisco, México

## HIGHLIGHTS

- Most of all quality parameters of Mexican honeys were found within international regulations except for hydroxymethylfurfural (HMF).
- Eight out of 15 honeys had HMF levels above the value limit recommended (<40 mg/kg).
- The producers of packaged honey should be encouraged to establish more moderate thermal treatments to avoid adverse changes.

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### Acronyms and abbreviations

HMF=hydroxymethylfurfural

## ABSTRACT

**Background:** Honey is a natural product, but it can be adulterated or heat-treated, both of which damage the properties of the original product. This research was focused on the evaluation of quality parameters of some commercial honeys produced in México.

**Methods:** Fifteen samples of commercial honeys available in Mexican market were collected. Some physicochemical parameters were determined, including pH, moisture, water activity, electric conductivity, color, sugar content (fructose and glucose), and hydroxymethylfurfural (HMF) contents.

**Results:** The results showed that the physicochemical parameters were found within acceptable ranges according to international regulations, with exception of the HMF content in 8 out of 15 honey samples which presented an unacceptable value (>40 mg/kg). The HMF content of the samples ranged from 14.56 to 224.08 mg/kg. Also, all samples of commercial honeys were classified as dark honey according to the L\* values determined less than 50 with range from 14.35 to 35.45.

**Conclusion:** Some commercial honeys from Guadalajara, Mexico had HMF levels above the acceptable limit because of overheating during the packaging process. All evaluated commercial honeys were classified as dark according to the L\* values which could be due to formation of browning pigments, in particular HMF, during the overheating. The producers of packaged honey should be encouraged to establish more moderate thermal treatments in order to avoid adverse changes, which affect the quality of the product.

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## Introduction

Honey, produced by bees (*Apis mellifera*), is a substance from flower nectar with sensory and nutritional

properties that are preferred by consumers around the world. The main constituents of honey are water and

\* Corresponding author. ✉ arriole@hotmail.com  
ORCID ID: <https://orcid.org/0000-0003-1452-7738>

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various sugars, principally fructose, glucose, maltose, and sucrose (Lazaridou et al., 2004; Wang et al., 2010), but it also contains minerals, pigments, volatile compounds, pollen grains, vitamins, amino acids, complex sugars, etc. (Anupama et al., 2003; Haouam et al., 2016; Schramm et al., 2003).

Honey is produced in different regions of the world and it is understandable that each type possesses a unique combination of compounds and physicochemical properties, due to the specific variety of flora localized in each ecosystem of production. Usually, several physicochemical parameters have been commonly used as a measure of honey quality in different countries. Some examples of these include moisture, electrical conductivity, hydroxymethylfurfural (HMF), water activity, pollen profile, and sugars composition (Anupama et al., 2003; Gleiter et al., 2006; Ouchemoukh et al., 2010; Pasiás et al., 2017).

Honey is a natural product, but it can be adulterated or heat-treated, both of which damage the properties of the original product (Zhao et al., 2018). It has been proposed the designation “fresh”, “raw” or “virgin” to show the purity of honey without any heat-treated (European Union, 2002). Similarly, “artisanal” honey is another designation proposed to indicate that honey was produced in a particular geographical region by a small beekeeping apiary.

Mexico is one of the largest producers of the honey in the world, with an annual production of about 55000 tons, of which approximately 40% is exported principally to Germany, England, Switzerland, and the United States (Mondragón-Cortez et al., 2013). In Mexico, the fresh and artisanal honey has a good reputation among consumers, but commercial honey (labeled as 100% pure) is also sold on a larger-scale according to Mexican regulations (Mondragón Cortez et al., 2013). Commercial honey is made when artisanal, fresh or raw honey is subjected to heating and filtering processes in order to keep the product in a liquid state. It serves with the purpose of facilitating its packaging and preserving the brightness and fluidity of the samples during its shelf life, or in other words, delaying the crystallization phenomenon. A crystallized honey is unattractive to some consumers. In the honey industry, the filtration process also helps to prevent crystallization by removing particles in the honey (dust, pollen, bits of wax, propolis or crystals of glucose hydrate), which can act as nucleation centers of glucose crystal. Yeast is destroyed and glucose crystals are dissolved during a heat treatment process; however, an excessive heat treatment can vary the physicochemical properties, principally the HMF content (Khalil et al., 2010; Turhan et al., 2008).

Physicochemical properties of Mexican artisanal honey have been reported (Moguel-Ordoñez et al., 2005; Mondragón Cortez et al., 2013; Viuda-Martos et al., 2010); however, there is no complete information in relation to commercial honeys, which are usually subjected to a packaging process. For this reason, the aim of this study was to evaluate the quality of different samples of commercial honeys (with the legend of 100% pure) available in the Mexican market in Guadalajara, Jalisco, through the determination of various physicochemical parameters.

## Materials and methods

### *Honey samples*

Fifteen samples of commercial honeys were purchased in glass and plastic jars (identified as H1-H15) during May to July 2018 in different stores, including supermarkets and health food stores, located in Guadalajara as the second largest city in commercial importance of Mexico. The honey samples were stored at laboratory temperature (20 °C) and then taken for the next analysis of physicochemical parameters. The honey samples with visible crystallization were heated before being analyzed to dissolve the crystals to 45 °C for 1 h.

### *Water content*

Water content was measured by an Abbe refractometer (Leica, Buffalo, N. Y., USA) at ~20 °C, based on relation between honey water content as well as refractive index (AOAC, 1999).

### *Water activity*

Water activity of the honey samples were evaluated at 25±0.2 °C using an electronic dew-point water activity meter Aqua-Lab model 4-TEV (Decagon Device, Inc., USA) equipped with a temperature control system.

### *The pH level*

The assessment of pH was done in a solution of 10 g of honey in 75 ml of ultrapure water with a pH meter (Orion 3 Star; Thermo scientific, USA).

### *Electrical conductivity*

Electrical conductivity was measured in a solution of honey (dry matter basis) at 20% (w/v) in deionized water using an Orion conductivity meter 2100 (Thermo scientific, USA), according to the procedure performed by Bogdanov et al. (2004).

### Hydroxymethylfurfural (HMF)

HMF content was determined according to the spectrophotometric method recommended by Zappala et al. (2005) using a Cintra 6 UV-Vis spectrophotometer (GBS Scientific Equipment, Victoria, Australia). Results were expressed in HMF mg/kg of honey.

### Color

Color parameters L\* (Lightness; 0=black, 100=white), a\* (-a\*=greenness, +a\*=redness), and b\* (-b\*=blueness, +b\*=yellowness) were measured using a Minolta CR-300 Chroma-meter (Minolta Ltd, Tokyo, Japan) with a standard illuminant D<sub>65</sub> under an observed angle of 10°. About 40 g honey sample was weighed into glass petri dish (30 mm in diameter) for color determination. Hence, the instrument was calibrated with a white tile standard.

### Sugars content

The fructose, glucose, and sucrose content were assessed using the High-Performance Liquid Chromatography (HPLC) method according to the AOAC (1999). The chromatographic mobile phase consisted of a mixture of water-acetonitrile (25-75); the flow was kept constant at 1 ml/min. The HPLC equipment comprised a binary pump, an auto-sampler, and a refractive index detector, all from Varian Prostar (Varian Inc., Palo Alto, CA, USA). Separation was performed on a 5 µm LC-NH<sub>2</sub> column of 250 mm x 4.6 mm (Supelco, Bellefonte, PA, USA).

### Results

Table 1 shows the identification, visual color, consistency, purity, geographical origin, and floral classification stated in label of honey samples purchased in the Mexican market (Guadalajara). All the samples indicated on the label that they are authentic or 100% pure. It was declared that only one honey sample was heated during the packaging process. Theoretically, the rest of commercial honeys purchased for this study were subjected to different packaging process, which had a stage where the honey was presumably heated. However, in five samples, the presence of total or partial crystallization was evident. The rest of the samples were perfectly clear and fluid.

The results of HMF and color are shown in Table 2. The HMF content was from 14.56 to 224.08 mg/kg. The L\* parameter showed values in the range from 14.35 to 35.45. The a\* values of samples varied from 2.76 to 11.24; while the b\* values of samples ranged from 6.50 to 21.57. The relationship between the HMF content and the L\* (lightness) values in the samples of commercial honeys is shown in Figure 1.

Table 3 shows the physicochemical properties of the fifteen honey samples evaluated in this work. The water content in the honey samples varied from 15.13 to 18.74%. The electrical conductivity in the honey samples analyzed varied from 0.290 to 0.913 mS/cm. The honey samples showed a pH in the range of 3.71 to 4.21.

Table 4 shows the sugars content, fructose/glucose ratio and glucose/water ratio in the honey samples. The fructose content varied from 39.91 to 31.76%, while glucose content was from 34.71 to 28.44%.

**Table 1:** Visual characteristics and labeling information on commercial honeys in Mexico

Honey sample	Visual color	Consistency	Purity declared on the label	Geographical origin declared on the label (Mexico)	Floral classification on the label
H1	Dark amber	Fluid	100%	U.I	U.I
H2	Dark amber	Fluid	100%	U.I	U.I
H3	Amber	Fluid	100%	U.I	U.I
H4	Amber	Crystallized	100%	Jalisco	multifloral
H5	Amber	Fluid	100%	U.I	U.I
H6	Dark amber	Fluid	Authentic	U.I	U.I
H7	Amber	Crystallized	100%	Jalisco	multifloral
H8	Amber	Crystallized	100%	Michoacán	multifloral
H9	Light amber	Crystallized	100%	Jalisco	multifloral
H10	Dark amber	Fluid	100%	Jalisco	multifloral
H11	Dark amber	Fluid	100%	Michoacán	U.I
H12	Dark amber	Fluid	Authentic	Michoacán	U.I
H13	Light amber	Fluid	100%	Jalisco	U.I
H14	Dark amber	Fluid	Authentic	Jalisco	U.I
H15	Dark amber	Crystallized	100%	U.I	U.I

U.I: Undeclared Information

**Table 2:** HMF and color (L\*, a\*, and b\* parameters) in some commercial honey samples in Mexico (mean values±SD)

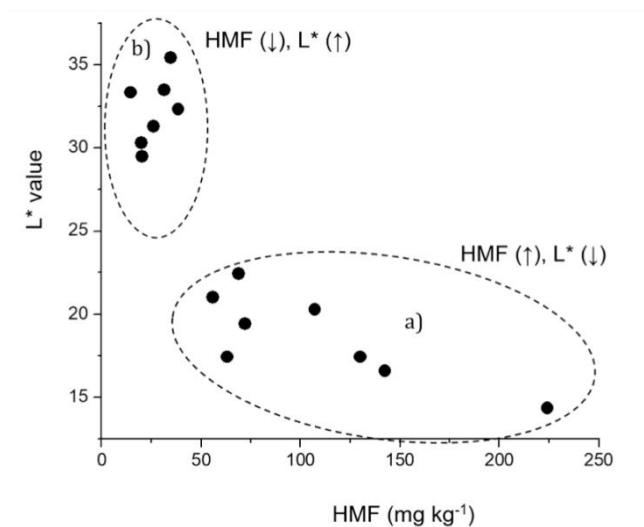
Honey sample	HMF (mg/kg)	L*	a*	b*
H1	107.19±3.53	20.29±0.71	4.47±0.68	9.22±0.54
H2	130.08±4.98	17.41±0.95	6.11±0.22	6.50±0.62
H3	19.92±2.61	30.32±1.46	7.43±0.41	19.28±0.45
H4	38.52±3.32	32.35±1.26	4.86±1.22	14.97±0.72
H5	26.07±2.41	31.29±0.89	4.18±0.45	6.75±0.55
H6	224.08±5.87	14.35±0.83	3.46±0.26	5.75±0.47
H7	14.56±2.54	33.35±0.45	11.24±0.81	16.01±0.78
H8	20.23±0.62	29.49±0.42	3.96±0.85	18.84±0.64
H9	31.50±1.75	33.48±0.53	2.76±1.35	21.57±0.67
H10	72.25±1.72	19.41±0.51	9.25±0.51	9.22±0.36
H11	142.45±3.46	16.58±0.73	3.47±0.45	7.42±0.47
H12	68.87±4.55	22.95±0.32	5.79±0.42	10.89±0.83
H13	34.78±2.13	35.45±0.58	2.78±0.32	9.54±0.54
H14	55.94±1.83	21.02±0.89	6.54±0.52	9.13±0.27
H15	63.12±3.65	17.14±0.81	4.23±0.94	11.35±0.53

**Table 3:** Physicochemical properties of honey samples in Mexico (mean values±SD)

Honey sample	Water activity (crystallized)	Water activity (fluid)	Water content (%)	Conductivity (mS/cm)	pH
H1	-	0.584±0.004	18.74±0.09	0.524±0.005	3.82±0.03
H2	-	0.561±0.003	17.43±0.06	0.463±0.007	3.84±0.02
H3	-	0.538±0.003	16.48±0.11	0.813±0.006	3.93±0.02
H4	0.546	0.525±0.003	15.68±0.10	0.469±0.004	3.94±0.06
H5	-	0.548±0.002	16.81±0.08	0.581±0.004	3.71±0.03
H6	-	0.537±0.003	16.40±0.09	0.647±0.007	4.14±0.03
H7	0.538	0.516±0.002	15.52±0.11	0.394±0.004	3.81±0.04
H8	0.549	0.524±0.003	15.63±0.07	0.341±0.003	3.94±0.05
H9	0.541	0.509±0.002	15.13±0.02	0.388±0.004	3.92±0.04
H10	-	0.557±0.001	17.27±0.08	0.587±0.006	3.72±0.03
H11	-	0.532±0.002	16.01±0.06	0.290±0.005	4.12±0.02
H12	-	0.553±0.002	17.09±0.08	0.913±0.007	3.92±0.02
H13	-	0.532±0.002	17.57±0.06	0.592±0.004	3.63±0.04
H14	-	0.563±0.003	17.27±0.12	0.354±0.003	4.21±0.04
H15	0.597	0.573±0.002	17.11±0.10	0.767±0.007	3.81±0.01

**Table 4:** Fructose (F), glucose (G), and G/water content as well as F/G relation of honey samples in Mexico (mean values±SD)

Honey sample	Fructose (%)	Glucose (%)	G/Water content	F/G
H1	34.87±0.54	30.27±0.38	1.61	1.15
H2	38.40±0.32	32.51±0.35	1.86	1.18
H3	35.34±0.21	31.73±0.15	1.92	1.11
H4	38.35±0.15	31.48±0.31	2.00	1.21
H5	36.55±0.36	32.24±0.24	1.92	1.13
H6	36.51±0.24	31.69±0.22	1.93	1.15
H7	39.64±0.47	32.85±0.20	2.12	1.20
H8	39.91±0.23	34.71±0.18	2.22	1.15
H9	38.58±0.38	32.48±0.29	2.15	1.18
H10	35.54±0.36	31.50±0.12	1.83	1.12
H11	33.77±0.25	28.67±0.30	1.79	1.17
H12	31.76±0.86	28.44±0.34	1.66	1.11
H13	34.65±0.28	31.47±0.31	1.79	1.10
H14	35.28±0.17	32.36±0.29	1.87	1.09
H15	38.43±0.31	34.62±0.41	2.02	1.11



**Figure 1:** Correlation between hydroxymethylfurfural (HMF) content and L\* value of honey samples: a) the first group showed that the honey samples with a high concentration of HMF had the lowest L\* values (light amber), b) the second group showed that the samples with low concentrations of HMF had the highest L\* values (dark-amber).

## Discussion

Fresh honey does not contain HMF, and therefore a high value of HMF can be an indication of excessive heating honey or unsuitable storage conditions (Escriche et al., 2008; Tosi et al., 2002). According to international regulations, HMF content should be under 40 mg/kg in honey samples (Codex Alimentarius, 2001; European Union, 2002). It has been found that the formation of HMF increases rapidly when honey is heated at high temperature (>100 °C) in short periods of time that might occur in the industrial packaging of honey. In this work, HMF content of eight honey samples exceed the acceptable limit of HMF most probably because of overheating during the packaging process. Similar results were found in honeys from Pakistan market which showed high HMF values between 220.5 and 509.8 mg/kg (Khan et al., 2006). Also, for an Orange (*Citrus aurantium* L.) honey from Sicily, Italy heated at 70 °C during 48 h, the HMF content varied from 6.0 to 149 mg/kg (Fallico et al., 2004). However, as seen in Table 2, seven honey samples showed HMF content inferior to the limit recommended by the international regulation (<40 mg/kg), which were in the range of 14.56 to 38.52 mg/kg. Presumably, these low HMF values may be due to a moderate heat treatment during the packaging process of these honeys.

The color of the honey is one of the main attributes that determine its acceptability by consumers. In several investigations, CIE LAB (L\*, a\*, b\*) scale has been used to measure color of artisanal honey produced in various countries (Ahmed et al., 2007; Bertoncelj et al., 2007). In fact, the L\* parameter, which indicates lightness has been used to classify honey samples as light if L\* > 50 or dark if L\* < 50 (González-Miret et al., 2005). In fresh honey, color is the result of mineral content, and presence of antioxidants compounds such as carotenoids and flavonoids (Viuda-Martos et al., 2010). However, fresh honey can be darkened if it overheats during the packaging process, due principally to the Maillard reactions, which involves the formation of melanoidin and HMF (Turkmen et al., 2006). Therefore, the L\* value will tend to decrease in a honey sample subjected to overheating. In the present study, all samples showed values L\* < 50, in the range of 14.35 to 35.45, therefore these types of honey can be classified as dark, according to González-Miret et al. (2005). The a\* values of samples varied from 2.76 to 11.24, while the b\* values of samples ranged from 6.50 to 21.57. These values (a\* and b\*) could be indicating have a transformation of yellow and reds components such as carotenoids and flavonoids in the samples during its packaging process.

On the other hand, the HMF content in honey samples subjected to different overheating has been proportional to the darkening ( $L^*$  value) which is observed (Onur et al., 2018; Turkmen et al., 2006). This is due to that HMF is formed by decomposition of sugars through the Maillard reaction, in addition to the caramelization of sugars, which promotes the formation of dark-brown color in a sample honey. As illustrated in Figure 1, the first group showed that the honeys with a high concentration of HMF had the lowest  $L^*$  values. On the contrary, the second group showed that the samples with low concentrations of HMF had the highest  $L^*$  values. These tendencies to darken of the honeys as function of the HMF content could be presumably due to the different levels of heating intensity (temperature and time) at which the samples were subjected during its packaging process. However, it must be taken into consideration the fact that some honeys, prior to heating, could have relatively certain levels of HMF due to an inadequate or long storage period. Hence, these values of residual HMF influenced relatively on the values found in the honeys studied. However, Tosi et al. (2002) stated that the HMF content in the initial honey samples has no influence on the rate of HMF formation in the honeys heated during the packaging process.

The water activities measured in samples of packaged honey with original crystallization (samples H4, H7, H8, H9, and H15) showed values higher in comparison with these same samples when it was measured in fluid state (Table 3). Zamora and Chirife (2006) observed that honey samples with crystallization from Argentina, showed higher values of water activity in comparison with the same samples but without crystals (fluid form). Water activity is a parameter highly affected by the presence of crystallization in a honey sample. Crystallization of honey occurs naturally, and is due to glucose content, since glucose has lower solubility than fructose. Honey samples with high values of water activity ( $>0.61$ ) increased the possibility of damage by fermentation (Gleiter et al., 2006). From this point of view, the analyzed honey samples with and without crystals, can be considered without risk for suffering fermentation.

The water content in honey is a parameter that could change during its packaging process, because honey can lose water by evaporation. However, the water content in our honey samples varied from 15.13 to 18.74%. These values are within of the limits of the water content ( $<20\%$ ) recommended by international regulations (Codex Alimentarius, 2001). Therefore, it is possible to infer that the moisture loss during the thermal processing was relatively low.

The electrical conductivity in the analyzed samples varied from 0.290 to 0.913 mS/cm, these values are similar to those found in artisanal honeys such as Hungarian

honeys with electrical conductivity range of 0.101 and 1.036 mS/cm (Czipa and Kovacs, 2014), and Tunisia honeys with ranged between 0.39 and 0.89 mS/cm (Boussaid et al., 2018). The electrical conductivity is related mainly to the presence of salts, organic acids, minerals, and proteins. This parameter may have high variability because it greatly depends on the floral origin of honey.

Our analyzed honeys showed a pH in the range of 3.71 to 4.21. The acid characteristic in honey is due principally to the presence of organic acids (Viuda-Martos et al., 2010). The pH values found in the honey samples of the current survey are similar to those obtained in raw honey produced in India ranged from 3.62 to 5.46 (Anupama et al., 2003) and Spain with range of 3.73 to 4.71 (Bentabol-Manzanares et al., 2014).

According to the results of this research, the fructose content varied from 39.91 to 31.76%, while glucose content was from 34.71 to 28.44%. These findings can be considered as normal compared to the values found in fresh honeys coming from different countries. In some previous reports, the range of fructose contents in honey samples were 33.12 and 39.12% from USA (Wang et al., 2010); 37.2 and 35.9% from Argentina (Baroni et al., 2009); and 41.4 and 37.75% from Spain (Bentabol-Manzanares et al., 2014). Also, for these same honeys, the glucose contents have been found to vary between 34.3 to 25.3%, 30.9 to 29.4%, and 37.3 to 28.9%, respectively. In all honey samples the fructose content was higher than the glucose content. In authentic honey, the ratio of fructose/glucose is between 1 and 1.2 (White Jr, 1978), which was observed in our results. Thus, it is possible to interpret that the honey samples of this study were not adulterated during the packaging process.

On the other hand, some researchers have explained that glucose/water content ratio can be a good indicator for prediction of honey crystallization (Dobre et al., 2012; Manikis and Thrasivoulou, 2001). Honey crystallization is entire when the glucose/water content ratio is greater than 2 (Dobre et al., 2012). We found that 5 out of the 15 analyzed samples showed glucose/water content ratio equal or higher than 2 (Table 3), which is congruent with the original consistency (crystallized) of the samples H4, H7, H8, H9, and H15 shown in Table 1. Furthermore, it could be assumed that although these honeys were subjected to a potential overheating during the packaging process, they did not lose the ability to crystallize rapidly during shelf life.

## Conclusion

Some commercial honeys from Guadalajara, Mexico had HMF levels above the value limit recommended by international regulations because of overheating during

the industrial packaging process. All samples of commercial honeys evaluated can be classified as dark according to the L\* values which could be due to the formation of browning pigments, in particular HMF, during the overheating. The producers of packaged honey should be encouraged to establish more moderate thermal treatments in order to avoid adverse changes, which affect the quality of the product.

This study only covered honey samples purchased in a particular region of Mexico and only determined few physicochemical properties. Thus, more investigations are needed in future to assess the quality of honeys produced in the other parts of the county.

### Author contributions

P.M.M-C., G.M.G-M., and E.A-G designed the methodology, did the experimental work and wrote the manuscript; P.M.M-C. and E.A-G. conceptualized and supervised of the research; P.M.M-C and G.M.G-M. analyzed the data. All authors revised and approved the final manuscript.

### Conflicts of interest

There are no conflicts of interest.

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