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The Physico-chemical Properties of Bee Honey Produced at Matrouh Governorate (Siwa Oasis, El-Alamein and Matrouh) as new reclaimed areas, Egypt

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# ABSTRACT

The honey composition is mainly affected by botanical origin, the geographical factors, as well as beekeeping practice and storage conditions can also influence its quality. Four honey types were collected from different apiaries located in Matrouh Governorate, Egypt during seasons of the year 2020, depending on floral sources, Clover (*Trifolium alexandrinum*) from Matrouh, camphor (*Cinnamomum camphora*) and Alfalfa (*Medicago sativa*) from Siwa Oasis and multiflora from El Alamein City. The results of physical analysis of different kinds of honey showed that the specific gravity, in all honey samples were ranged between 1.3 - 1.44 and viscosity 48.1 - 69 (Pois). The Electrical conductivity (EC) values of samples ranged from 0.008 to 0.039%, the total soluble solids (TSS) percentage ranged between 81.5 to 82.5% and moisture contents ranged from (17.5 to 18.50%). The chemical characteristics of all honey samples tested were found to be acidic in nature, pH (3.01–4.35). The glucose content of all analyzed samples of four varieties ranged from 31.8% to 33.17%, fructose values ranged between 39.19% and 41.23%, the percentage of sucrose varied from 3.36% to 4.57%, the concentration of maltose ranged between 2.37% and 6.0%. The present study concluded that; the quality and physico-chemical properties of honey were varied based on the geographical and botanical origins and fall within the limits of international standards.

Keywords: Bee honey, physicochemical, geographical, and botanical origins.

# 1. Introduction

Botanical origin has the greatest impact on honey composition, although geographical location, beekeeping techniques, and storage conditions can also have a big influence on quality.

The type of nectar flow, on the other hand, has a significant impact on the chemical composition of honey. The biochemical composition of nectar is influenced in both qualitative and quantitative ways by the genetics and physiology of the source plant, as well as by environmental factors (climate conditions) and soil characteristics. Other factors that affect honey quality outside its botanical and geographic origin are the health and vitality of the bee colony, the beekeepers' procedures for gathering and confectioning honey, and how the consumer stores their honey (Soares *et al.*, 2017).

The composition of essential oils is shown to be spatially dependent, even for the same plant species, suggesting that honey from the same floral origin but from various places may have a varying composition (Kaškonienė *et al.*, 2010).

Plant nectar (floral nectar), extrafloral nectar (extrafloral nectar), and honeydew are used by honeybees and stingless bees to create the naturally sweet substance known as honey (Codex

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Alimentarius 2001). The primary component of honey is sugar, but it also contains enzymes, amino acids, organic acids, carotenoids, vitamins, minerals, and aromatic compounds (Alqarni *et al.*, 2012; da Silva *et al.*, 2016 and Ouchemoukh *et al.*, 2007).

For bee honey to avoid fermenting and granulating, its moisture content is essential. Because honey has a low moisture content, it is resistant to microbial growth and can be stored for longer periods of time (Bogdanov, 2009; Buba *et al.*, 2013; Akhtar *et al.*, 2014 and El-Metwally, 2015).

Honey's Rheological properties are influenced not only by moisture content; but also by temperature and chemical makeup (Gomez-Diaz *et al.*, 2009). Moisture content and its association with the fructose to glucose (F/G) ratio are two factors that affect the crystallization rate (Lazaridou *et al.*, 2004; Witczak *et al.*, 2011).

Honey carbohydrates are a complex blend of 70 percent monosaccharides (primarily glucose and fructose) and disaccharides (10–15 percent) (De La Fuente *et al.*, 2006).

The amount of sugar in honey affects physicochemical properties including hygroscopicity, viscosity, and crystallization (Kang and Yoo, 2008).

Numerous scientists have written about the sugar profile of distinct honey types (Ouchemoukh *et al.*, 2010; De La Fuente *et al.*, 2011), and the profile of a particular honey is greatly dependent on the type of flowers visited by bees, as well as regional and climatic factors (Gomez-Barez *et al.*, 2000).

Honey is mostly made up of sugars, mostly fructose (40–50%) and glucose (32–37%), with a small quantity of sucrose (<2%), and mineral components (ash less than 0.1 percent). Water (13–20%) is also present in honey (Alvarez-Suarez *et al.*, 2013).

After harvest, the characteristics of honey are affected by weather exposure, processing, manipulation, packing, beekeeper care, and storage time (da Silva *et al.*, 2016; Chanchao 2013; da Costa Leite *et al.*, 2000; Escuredo *et al.*, 2014 and Tornuk *et al.*, 2013).

The aim of the study was to determine the influence of geographical origin on honey physicochemical quality based on the comparison of different apiaries located in Matrouh Governorate; Siwa Oasis, El-Alamein and Matrouh, as new reclaimed areas, Egypt.

# 2. Materials and Methods

### 2.1. Sample collection and preparation:

The study employed four honey samples harvested from various sites in Egypt's Matrouh Governorate; Siwa Oasis, El-Alamein and Matrouh, as new reclaimed areas. All of the fresh samples were collected in sterile conditions and stored at room temperature in a container labeled with numbers, location, and date of collection.

### 2.2. Physical characteristics:

I. The viscosity of honey was measured according to Munro, (1943).

**II.** Specific gravity was measured according to Wedmore (1955).

**III.** Determination of Electrical Conductivity (EC): EC was measured using an HI 98311 conductivity meter (Hanna Instruments, Mauritius) and a 20% (w/v) solution of honey was suspended in milli-Q water (Bogdanov *et al.*, 1999).

# 2.3. Chemical properties:

**I.** Determination of moisture content: The moisture content of honey was determined by calculating the refractive index value (Abbe refractometer at 20°C) (A.O.A.C, 1995).

# II. pH

The pH meter (Eutech Instruments Pvt Ltd., Singapore) was used for a 10% (w/v) honey solution prepared in distilled water (A.O.A.C., 2012).

**III.** Determination of total soluble solids (TSS) of honey by (Association of Official Analytical Chemists (A.O.A.C., 1980). Equipment: Abbe refractometer was used and expressing the TSS in honey in percentage.

V. Determination of free acids, lactone content, and total acidity according to the method of White *et al.*, (1962).

**IV.** Determination of the quantity of sugars by High performance Liquid Chromatography (HPLC). The concentration of fructose, glucose, sucrose, and maltose in honey samples was determined by HPLC according to the method of Bogdanov and Baumann (1988).

IIV. Determination of pollen sediment content, according to the method of Louveaux et al., (1978).

# 2.4. Statistical analysis

The results were expressed as the mean  $\pm$  standard deviation of the mean, and the significant difference between means was evaluated using a one-way analysis of variance followed by a post hoc test for the comparison of significance using the Statistical Package program SPSS version 23.0 (IBM SPSS Statistics, USA). Values of p<0.05 were considered statistically significant.

# 3. Results and Discussion

Honey's composition was largely determined by the floral source. Also seasonal, environmental and processing variables also played an important role.

# **3.1. Physical properties:**

# 3. 1.1. The specific gravity:

All samples of Matrouh honeys in Table (1) ranged from  $1.4 \pm 0.01$  to  $1.433 \pm 0.005$ , and this value was confirmed to meet European legislation, the European Commission (2001).

All honey types had virtually the same specific gravity of 1.40, ranging from  $1.390 \pm 05$  to  $1.42\pm0.36$  with no significant variances (Abdel-Hameed, 2020). This result is also consistent with the findings of El-Sharawi *et al.* (2009) they reported that the range of the specific gravity was 1.39 to 1.42.

According to Rebiai and Lanez (2014), the specific gravity ranged from 1.29 to 1.41, and Salih (2019) found that the specific gravity ranged from 1.40 - 1.47, with a mean of 1.43 in the Kurdistan area. Nafea *et al.* (2009) studied the specific gravity of different Libyan honey, and he found that, it is ranging from 1.39 - 1.43.

# 3.1.2. The viscosity

Different kinds of honey as a function of moisture content and temperature. The viscosity decreases with the increase of temperature and moisture content because average intermolecular forces reduce as the tempraturer rises i.e., kinetic energy increases, and molecules become more mobile (Patil and Muskan, 2009). The most essential aspects that should be taken into consideration to understand the flow mechanism at a structural and molecular level during the rheological characterization of honey are glucose, fructose, and F/G ratio (Dobre *et al.*, 2012).

There was no significant difference between the Matrouh honey samples examined, which ranged from 48.1 to 69 Pois. (Table 1).

According to Nafea *et al.* (2014), the viscosity ranged from 13.6 to 87.8 in some Egyptian honey. Abdel-Hameed (2020) clarified the viscosity of honey types were ranged between 13.60  $\pm$ 05 to 69.00  $\pm$ 11 Poise.

### **3.1.3. The Electrical Conductivity** (EC%):

It is one of the most crucial factors for determining the physical characteristics of honey (Serrano *et. al.*, 2004). The EC, rather than the ash content, is an excellent measure for determining the botanical origin of honey (Adenekan *et al.*, 2010). This criterion was recently added to international standards, replacing the ash content determination (Codex Alimentarius Commission, 2001).

The EC value is based on the amount of ash and acid in honey; the higher the amount, the higher the conductivity (Bogdanov *and* Martin, 2002).

As shown in Table (1) the Electrical conductivity (EC) values of samples ranged from 0.008 to 0.039%. However, the results were similar to the findings previously reported by Saxena *et al.* (2010) and Alvarez-Suarez *et al.* (2010).

Zidan, (2019) discovered that the EC percent values of Egyptian and Algerian honey samples, which were 0.046  $\pm 0.009$  and 0.036  $\pm 0.004$ , respectively, were significantly higher than those of Libyan and Yemeni honey samples, which were 0.011  $\pm 0.002$  and 0.015  $\pm 0.003$ , respectively.

In some Egyptian honey, the EC ranged from 110.0 ppm  $\pm 10$  to 520.0 ppm  $\pm 10$ . (Abdel-Hameed, 2020). Also, in various Saudi Arabia honey, Tharwat and Nafea (2006) found that the EC ranged from 0.01 - 0.09.

#### **3.2.** Chemical properties:

**3.2.1. The moisture content** of all analyzed samples was within the range of 17.5 - 18.5% as shown in Table (1) as recommended by Codex Alimentarius Commission (2001) (<20%) and European Commission (2002). The highest moisture value was detected in alfalfa honey and the lowest value was in camphor honey.

There were no significant differences, between all honey samples, indicating that these honeys had high storage ability. The different plants were grow in the same approximately environmental condition nearly. All of the honey samples evaluated had a moisture content of 17.5 - 19.1 percent (Nayik *et al.*, 2019).

Abdel-Hameed (2020) revealed that the moisture percentages of honey samples ranged between 17.25%  $\pm 0.66$  to 21.0%  $\pm 1.11$ , There were no significant changes in moisture content across Sidr honey samples from Arab countries, which varied from 17.70%  $\pm 0.224$  to 18.00%  $\pm 0.447\%$  (Zidan, 2019).

**3.2.2. Total soluble solids (TSS)** are a measure of the combined content of all inorganic and organic substances in honey in the molecular, ionized, micro-granular suspended forms.

The present data in Table (1) revealed non-significant among all Matrouh honey samples, it ranged between  $81.5\% \pm 0.5$  to  $82.5\% \pm 0.5$ , and these results demonstrated a good correlation between EC and TSS, indicating that both parameters can be used to determine honey purity European Commission (2001).

These results are consistent with those of Abdel-Hameed (2020), who showed that the (TSS) of some Egyptian honey samples ranged from 79.0% to 87.75%, and Nafea *et al.*, (2014), who reported that the (TSS) percentage of some Egyptian honey samples ranged from 77.0% to 83.2%. Furthermore, Zidan (2019) revealed that non-significant among all Sidr honey samples, ranging from 82.0% to 82.3%. Our results obtained disagreed with that reported by Salih (2019) who reported that the total soluble solids of the honey ranged from 82.5 to 86.1% and Nyau *et al.* (2013) who found total soluble solids was 83.6 and 85.7% of different honey samples in Zambia.

#### 2.3. Acidity and pH:

In general, honey is acidic in nature, regardless of its varied geographical origin. All of the honey samples evaluated were acidic  $(3.5 \pm 0.1 - 4.033 \pm 0.153)$ , and statistical analysis revealed no significant differences among all of the tested varieties as shown in Table (1). The average free acidity values in Matrouh honey samples ranged from 12.33 ±2.565 and 37.23 ±9.41 mq/kg, with camphor honey having the highest value and clover honey having the lowest. Lactone acidity ranged from 3.51 to 16.87 mq/kg, with highest value in multiflora honey and the lowest in alfalfa honey. The total acidity of Egyptian honey samples ranged from 27.17 ±1.26 to 54.0 ±4.76 mq./kg, with camphor honey having the highest value and clover honey having the lowest.

The pH values matched those found in Brazilian, Algerian, Spanish, and Turkish honeys (Azeredo *et al.*, 2003; Ozcan and Olmez, 2014). Honey's acidity is measured in terms of free acidity, lactonic acidity, and total acidity. Specifics a free acidity of not more than 50 mq/1000 g (mq/kg) (European Commission, 2002).

The findings of this investigation agree with Ouchemoukh *et. al.* (2007). As for the values of pH of several Egyptian honey recorded ranged from 3.7 to 4.7 (Abdel-Hameed, 2020).

According to Kayacier and Karaman, (2008), high acidity can be a sign of sugar fermentation into organic acids. Free acidity, lactones, and total acidity are all within the acceptable ranges (11

mq/kg to 78 mq/kg), (1 mq/kg to 34 mq/kg), and (12 mq/kg to 93.5 mq/kg), respectively (Nafea *et al.*, 2014).

Abdel-Hameed, (2020) showed that free acidity ( $11.0 \pm 1.32$  to  $68.3 \pm 0.85$ ), Lactone ( $7.5 \pm 0.70$  to  $17.5 \pm 0.70$ ), total acidity ( $18.5 \pm 1.05$  to  $86.0 \pm 0.70$ ). Zidan (2019) noticed that all Sidr honey samples in Arab countries were acidic, with pH ranging from  $3.5 \pm 0.2$  to  $5.3 \pm 0.332$ . The average values for free acidity in samples ranged from  $34.0 \pm 2.288$  to  $49.9 \pm 1.159$  mq/kg, lactonic acidity from  $2.0 \pm 0.356$  to  $0.0 \pm 1.034$  mq/kg, and total acidity detected from  $44.03 \pm 5.02$  to  $51.93 \pm 1.59$  mq/kg. Honey's high acidity is attributed to the fermentation of sugars in the honey into organic acid, which is responsible for two main features of honey: flavour and stability against microbial spoilage (Bogdanov *et al.*, 2008).

| Regions           | Matrouh                      | <b>El-Alamein</b>             | Siwa Oasis                       |                                    |
|-------------------|------------------------------|-------------------------------|----------------------------------|------------------------------------|
| Parameters        | Clover                       | Multiflora                    | Alfalfa                          | Camphor                            |
| Moisture %        | 18.33 <sup>a</sup> ±18.33    | $18.00 \ ^{a} \pm 18.0$       | 18.5 <sup>a</sup> ±18.5          | 17.5 <sup>a</sup> ±17.5            |
| TSS %             | $81.67^{a} \pm 0.577$        | $82.00^{a} \pm 0.5$           | 81.5 <sup>a</sup> ±0.5           | 82.5 <sup>a</sup> ±0.5             |
| Specific gravity  | $1.413 \ ^{b} \ \pm 0.011$   | $1.4^{\text{b}}\pm 0.01$      | $1.417 \ ^{\mathrm{b}}\pm 0.006$ | $1.433 \ ^{a} \pm 0.005$           |
| Viscosity (Poise) | $48.1^{\ b} \pm 0.05$        | 69 <sup>a</sup> ±0.011        | $48.1 \ ^{b} \pm 0.05$           | 69 <sup>a</sup> ±0.05              |
| EC %              | $0.008^{\circ}\ \pm 0.001$   | $0.016 \ ^{\rm b} \pm 0.005$  | $0.018^{\ b} \pm 0.075$          | $0.039  {}^{\mathrm{a}} \pm 0.003$ |
| рН                | $3.5$ <sup>a</sup> $\pm$ 0.1 | 3.7 <sup>a</sup> ±0.36        | 3.565 <sup>a</sup> ±0.472        | 4.033 <sup>a</sup> ±0.153          |
| Free acidity      | 12.33 <sup>b</sup> ±2.565    | 18.3 <sup>b</sup> ±4.91       | 22.0 <sup>b</sup> ±6.68          | 37.23 <sup>a</sup> ±9.41           |
| Lactone           | 14.83 <sup>a</sup> ±2.08     | $16.87 ^{\text{a}} \pm 10.43$ | 3.51 <sup>a</sup> ±15.33         | 16.77 <sup>a</sup> ±5.89           |
| Total acidity     | $27.17^{b} \pm 1.26$         | $35.17^{b} \pm 6.91$          | $37.33^{b} \pm 3.27$             | 54.0 <sup>a</sup> ±4.76            |
| Fructose          | 41.23 <sup>a</sup> ±1.33     | 39.19 <sup>a</sup> ±1.306     | 39.96 <sup>a</sup> ±1.66         | 40.67 <sup>a</sup> ±1.22           |
| Glucose           | 33.23 <sup>a</sup> ±1.67     | 33.07 <sup>a</sup> ±3.81      | 33.17 <sup>a</sup> ±3.95         | $31.8^{a}\pm1.42$                  |
| Sucrose           | 3.36 <sup>a</sup> ±1.11      | $4.57 ^{\text{a}} \pm 083$    | $4.37 ^{\mathrm{a}} \pm 0.74$    | 3.93 <sup>a</sup> ±0.15            |
| Maltose           | 2.34 ° ±1.54                 | 6.0 <sup>a</sup> ±2.95        | 4.83 <sup>a</sup> ±1.62          | 4.76 <sup>a</sup> ±0.513           |
| F/G               | 1.24 <sup>a</sup>            | 1.18 ª                        | 1.2 ª                            | 1.27 ª                             |
| G/W               | 1.81 ª                       | 1.84 ª                        | 1.79 ª                           | 1.82 ª                             |

Table 1: The physico-chemical composition of honey produced at Matrouh Governorate, Egypt.

Different letters a, b, c within the same row indicates significant differences p<0.05.

TSS= total soluble solids, EC= electrical conductivity, F/G= fructose/ glucose ratio, G/W= glucose/water.

#### 2.4. Sugars

Dobre *et al.* (2012) and Ouchemoukh *et al.* (2010) found that the major components of honey are sugars, which are influenced mostly by floral and geographical origins and less by seasonal, processing, and storage circumstances.

As demonstrated in Table (1), there were no significant differences in fructose value, glucose, sucrose, maltose, fructose/ glucose ratio (G/F), and glucose/water (G/W) ratio among all Matrouh honey samples evaluated.

The fructose value of all honey samples ranged from 39.19 to 41.23%, with clover having the greatest proportion and multiflora having the lowest.

The glucose content of all analyzed samples from four varieties ranged from 31.8 to 33.32%, with clover having the highest percentage and camphor having the lowest.

The sucrose value of all tested honey was between 3.36 and 4.93%, with camphor having the highest percentage and clover having the lowest.

The high glucose level of honey samples resulted in faster crystallization, with a glucose content of more than 30%.

Maltose values range from 2.34 to 6.0%, with multiflora having the greatest amount and clover having the lowest. F/g was between 1.2 and 1.27, and G/W was between 1.79 and 1.84.

All the honeys presented a value of glucose plus fructose higher than 60 g/100g, which is the value, required for all kinds of honey in the European and Codex standards. The sucrose content of all the honey samples was less than the European Community Directive's (5 g/100 g) limit, indicating that these honeys were at an advanced stage of ripening. These results supported the previous several studies on different honey types (Buba *et al.*, 2013 and EL-Metwally, 2015).

The range of quantities measured in our study is comparable to that found in reports for numerous types of Egyptian honey, where fructose values were varied from (38.2  $\pm$ 0.66 to 41.2  $\pm$ 0.30%), glucose values were ranged from (28.0  $\pm$ 1.23 to 32.0  $\pm$ 1.61%), sucrose was ranged from (1.1 to 5.1%), maltose was ranged from (4.5 to 10.0%), and the (F/G) ratio of honey samples was ranged from 1.45 to 1.9 (Abdel-Hameed, 2020).

Also, according to Zidan (2019), the sugar contents of Sidr honey in several Arab countries were, fructose value ranged from 38.0 to 41.9%, glucose ranged from 69.60% to 32.0%, F/G ranged from 1.29 to 1.44 and G/W ranged from 1.644 to 1.78.

Nafea *et al.* (2014) cited that the sugar values of some Egyptian honey were, fructose ranged between 35.1- 38.9%, glucose ranged from 27.7 to 32.0%, sucrose ranged between 0.75 - 2.5% and maltose ranged between 2.0 - 5.0%.

The sucrose contents reported in this realisation are within the range of values recorded for Argentine and Turkish sucrose (Cantarelli *et. al.*, 2008), and Venezuelan sucrose (Vit *et al.*, 2009).

# **3.2.5.** Crystallization ratios

The tendency of honey to granulate is explained by the (F/G) ratio because glucose is less water soluble than fructose, making it an important parameter to predict honey's crystallization tendency (Laos *et al.*, 2011). Our results were consistent with a mean F/G ratio of approximately 1.2, which is prevalent in most honey varieties around the world (Dobre *et al.*, 2012)

While (Amir *et al.*, 2010) stated that honey crystallization is slower when the F/G ratio is greater than 1.3 and faster when the ratio is less than 1.0, also when the (G/W) ratio is less than 1.3, honey crystallization is very slow or even non-existent, and when the ratio is greater than 2.0, honey crystallization is complete and rapid. Crystallization period is mostly influenced by the F/G and glucose/water (G/W) ratios, according to the National Honey Board (2010). According to certain publications, the G/ W ratio can be a better signal for honey crystallization prediction (Dobre *et al.*, 2012; Manikis and Thrasivoulou, 2001).

The lower the moisture content and higher the glucose content of honey, the faster the crystallization will take place. According to the review, honey crystallization is slow or null when the G/W ratio is less than 1.7 and is complete and rapid when the ratio is greater than 2 (Dobre *et al.*, 2012).

The G/W ratio was greater than 1.7 in all four honey samples tested. Our findings correspond with those of (Abdel-Hameed, 2020), who discovered that the G/W ratio of honey samples ranged from 1.45 to 1.9, while the F/G ratio ranged from 1.25 to 1.4. F/G ranged from 1.29 to 1.44 and G/W from 1.644 to 1.78, according to Zidan (2019).

#### 4. Conclusion

The results of physico-chemical analysis of all the honey varieties of Matrouh Governorate, Egypt were within the limits recommended by European Commission and the Codex Alimentarius. The carbohydrates profile of studied honey revealed that all the unique honey varieties possessed reducing sugars, fructose and glucose in the portion and small quantities of disaccharides.

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