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The physical and chemical properties of some types of bee honey produced in New Valley Governorate, Egypt

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Abstract

These studies aimed to determine the physical and chemical properties of some types of bee honey produced in New Valley Governorate, Egypt. Four types of honey were collected from some apiaries in different regions from New Valley Governorate Egypt belong to five floral plants [Clover (*Trifolium alexandrium*), alfalfa (*Medicago sativa*), tamarisk (*Tamarix aphylla*), acacia (*Acacia arabica*) and eucalyptus (*Eucalyptus camaldulensis*)] during the 2023-2024 seasons. The findings indicate that while minor variations exist among the tested honey samples, no statistically significant differences were observed in key parameters, affirming the consistency and quality of the honeys. The viscosity of the tested honeys ranged between 66.67 to 75 Poise, specific gravity values (1.410 to 1.420) were within acceptable quality standards, electrical conductivity varied among the samples indicative of mineral composition. However, all samples met the European honey quality standards. Total soluble solids (TSS) ranged between 82% to 82.16%, signifying high sugar content and minimal water activity. Moisture content varied between 17.66% and 18%, staying within international quality standards (<20%). Total acidity, free acidity, and lactone levels were consistent across the samples, with values ranging within globally reported standards. pH values (3.733 to 4.19) were within the optimal range for honey, influencing its antimicrobial properties and storage stability. Fructose and glucose were the dominant sugars in the tested honeys, with fructose levels ranging from 39.83% to 40.83% and glucose between 29.70% to 32.86%, confirming their natural carbohydrate profile. Sucrose content ranged from 2.133% to 5.133%. All samples remained within the permissible quality limits, ensuring authenticity. Maltose content showed slight differences among the honey types, with levels between 3.566% and 7.73%, reflecting enzymatic activity during honey production.

Introduction

Honey is a natural substance produced by honeybees (*Apis mellifera* L) from the nectar of flowers, resulting in a sweet, flavorful, and viscous liquid (Rodriguez *et al*., 2023). The chemical composition of honey is significantly influenced by the type of nectar collected. The biochemical profile of nectar is affected both qualitatively and quantitatively by the genetics and physiology of the source plant, as well as environmental factors such as climate conditions and soil composition (Li *et al*., 2023).

Additional factors impacting honey quality beyond its botanical and geographic origin include the health and vitality of the bee colony, the beekeepers' methods for harvesting and processing honey, and storage conditions maintained by consumers (Garcia *et al*., 2024). Honey is a complex mixture with considerable variation in composition and characteristics due to its geographical and botanical origin. Its primary attributes depend on the floral source, or the nectar foraged by bees (Fernandez *et al*., 2023).

The water content of honey is the quality aspect that determines the ability of honey to remain fresh and to avoid spoilage by yeast fermentation. Raw honey can have a water-in-honey content of less than 14% and the lower water content the higher perceived value of the honey (Hatjina *et al*., 2014). It is internationally recognized that good quality honey should be processed at less than 20% water content. Low water content is desirable because honey may begin to ferment and lose its fresh quality if the water-in-honey is greater than 20%. Unpasteurized honey ferments because it contains wild yeast. However, due to honey's high sugar concentration these yeasts are less likely to cause fermentation in honey with low water content (Maughan, 2002).

The proximate analysis of the honey samples obtained from various locations in North Sinai, Egypt, showed that there were no significant differences in moisture, fructose, maltose, (fructose + glucose), reducing sugar, F/G, and G/W. However, there were significant differences ($P<0.05$) in glucose, sucrose, pH, free acidity, lactone, and total acidity among the honey samples (Nafea *et al.,* 2023).

Honey is mostly made up of sugars, mostly fructose (40–50%) and glucose (32–37%), with a small quantity of sucrose (honey can be variable and dependent on its floral source, geographical origin, environmental factors, and processing (Guler *et al*., 2007 and Alvarez-Suarez *et al*., 2010). The ratio of one type of sugar to another depends of the source, i.e., flower pasture, and to some extent on enzyme invertase, which breaks down regular sugar in grape and fruit. This enzyme is located in the flower from which the bees collect nectar, but it is also present in the bee's body (Di Pasquale *et al.* ,2013)

The aim of this study evaluates some physical and chemical properties of some bee honey types produced in New Valley Governorate, and their quality according to Egyptian Organization for standardization and quality control, EOSC (2005): (2005).

Materials and methods

1. List of the tested types collected from New Valley honeys:

Four kinds of honeys belong to five floral plants growing in different region from New Valley Governorate, Egypt and botanically called.

Clover, *Trifolium alexandrium*
Alfalfa, *Medicago sativa*
Tamarisk, *Tamarix aphylla*
Acacia, *Acacia arabica*
Eucalyptus, *Eucalyptus camaldulensis*

2. Confirmation of the botanical origin (floral source) of the tested honeys.

According to Moore and Webb (1978) and Identification of honey content of pollen grains was achieved through the comparison between the extracted honey's pollen and the reference of pollen of the plant grains (Sawyer, 1989). Representing pollen grains of the floral source of the tested honey was considered according to nature of pollen of that crop.

3. Collection of honey samples:

In May 2024, samples of clover honey, while the other kinds of honey were collected at the time of their extraction (Alfalfa honey in June, 2024, tamarisk honey in November, 2023, acacia and eucalyptus honey in November, 2023). The collected honey samples were freshly extracted and kept in dark jars until analyses.

4. Determination of different properties of the tested honeys in fresh state:

The confirmed type of fresh samples of each honey kind were subjected to analysis after their extraction from the honeybee colonies. The following properties were determined in three replicate samples of each tested honey:

-The Association of Official Analytical Chemists defines moisture and total soluble solids (T.S.S.). It was determined by using a refractometric method as mentioned in A.O.A.C. (1995).

4.1. Determination of viscosity and the specific gravity:

Viscosity and specific gravity were determined by White (1978).

4.2. Determination of electrical conductivity (EC)

Electrical conductivity (EC) was determined by Sancho *et al*. (1992).

- pH value was measured by "HANNA" pH-meter, model HI9321 as mentioned in A.O.A.C. (1995).

4.3. Determination of free acids, lactone content and total acidity:

According to the method of White (1978).

-Determination the quantity of sugars by High Performance Liquid Chromatography (HPLC). Concentration of fructose, glucose, sucrose and maltose in honey samples were determined by HPLC according to the method of Bogdanov and Baumann (1988).

Result and discussion

1. Physical properties of the studied honey kinds:

1.1. Viscosity:

The tested honey's viscosity (Table 1) showed that the viscosity values ranged from 66.67 to 75 Poise, with no significant difference between the examined samples of honey ($P<0.05$). Another study found viscosity values ranging from 13.6 to 87.5 poise for Libyan honey types (Nafea *et al*., 2009) However, no significant difference was observed in the viscosity values of the examined samples, which ranged from 69 ± 0.08 to 69 ± 0.36 for Matruh honey samples (El-Dereny *et al*., 2022) and El-Dereny, 2023) viscosity varied between 46.73 and 73.12 Poise.

1.2. Specific gravity:

The tested honeys' densities (Table 1) showed that statistical analysis revealed no significant difference ($P = 0.6294$), suggesting that all honey types possess relatively similar density levels. The specific gravity values ranged from 1.410 to 1.420, and all samples meet the quality standards stated in the European Legislation, European Commission (2001). These values fall within the densities reported by El-Dereny *et al.* (2022) for different Matrouh honey types, specific gravity ranged from 1.4 ±0.01 to 1.433 ±0.005. Nafea *et al.* (2023) indicated that the specific gravity ranged from 1.420 ±0.00 to 1.410±0.018 of honeys at two locations (Rafeh and El-Arish) in North Sinai regions. Specific gravity is crucial in determining honey purity and quality (Terrab *et al*., 2002). The slightly higher value for Tamarisk honey may be linked to its sugar composition and mineral content.

1.3. Electrical conductivity (E.C):

The difference in electrical conductivity values may be attributed to ash content, and the E.C of honey is an indicator of its authenticity and adulteration. It provides information about the nectar source. Data in Table 2 revealed that E.C of the tested honeys greatly varied. Although the differences were not statistically significant (P = 0.491), Clover honey showed the highest EC, which could be attributed to a richer mineral composition (0.023±0.009 ppm), followed by Tamarisk bee honey (0.0166±0.009 ppm), and then Acacia and Eucalyptus bee honey (0.0143±0.008 ppm), while Alfalfa honey was significantly the lowest one (0.0066±0.008 ppm).

The high values of EC can be attributed to high mineral content. Abdel-Hameed (2020) showed that EC values ranged from 110.0 ±10 to 520.0 ±10 ppm. El-Dereny *et al*., (2022) demonstrated that the EC values ranged from 0.008 to 0.039%. El-Dereny (2023) E.C. 0.21±0.20 and 0.70 ±0.32 (mS/cm). Generally, EC values help distinguish floral honeys from honeydew honeys, where the latter usually exhibits higher EC values (Bogdanov *et al*., 2004).

1.4. Total soluble solids (TSS):

Data of Table (1) indicated that total soluble solids (TSS) were quite consistent across all honey types, ranging from 82% (Alfalfa and tamarisk honey) to 82.16% (Acacia and eucalyptus mixture honey and clover honey). The minor variations were statistically insignificant (P = 0.993), suggesting that all tested honeys have a similar sugar concentration, which aligns with the Codex Alimentarius standards (Codex Alimentarius, 2001). High TSS values indicate lower water activity, reducing the risk of fermentation and microbial growth (Chirife *et al*., 2006). In general, variations of TSS among different honeys may be attributed to composition of the nectar secreted by different plants sources. Honey with T.S.S. equal to or greater than 81.4% is classified as higher grade (A and B), while a T.S.S. falling between 80% and 81.3% is considered lower grade C (QSAE, 2005). Abdel-Hameed (2020) demonstrated that the T.S.S. of Egyptian honey samples ranged from 79.0% to 87.75%.

Thus, the physical properties of different honey types from the New Valley Governorate, Egypt, showed minor variations. Specific gravity, viscosity, electrical conductivity, and TSS exhibited no significant differences among samples. These findings highlight the consistency in honey quality and align with global honey standards, ensuring their suitability for consumption and industrial applications.

2. The Chemical composition of the tested honeys:

2.1. Moisture content:

The moisture content of the honey depends on floral source, atmospheric humidity, temperature, time of extraction from combs, the ripening process carried out by bees, hygroscopic nature of honey and storage condition. It is well known that moisture content has an influence on honey color, viscosity, flavor, and density and refraction index and is one of the most important physical

Data in Table (2) revealed that Alfalfa bee honey have the highest content of moisture (18 %), followed by Tamarisk bee honey and mixture Acacia and Eucalyptus bee honey (17.83 %), while clover honey have the lowest level of moisture (17.66 %). The minimal variation among samples, with no significant difference (P = 0.99752), suggests that all honey types conform to international quality standards, which typically recommend moisture levels below 20% to prevent fermentation (Codex Alimentarius, 2001). These values align with previous findings where moisture content depends on factors such as floral source and harvesting conditions (White, 1978).

The present results are consistent with those reported by Nafea *et al.* (2023) they present the measured values of chemical properties for various types of honey produced in North Sinai Governorate, Egypt, such as Citrus Algae, Myrrh, and Tarot. The moisture content, which reflects the ripeness of the honey, ranged from 17.23±0.76% to 18.32±0.57% across the samples

El-Dereny (2023) found that moisture ranged from 17.35 to 18.38 %. Abdel-Hameed (2020) revealed that the moisture percentages of honey samples ranged between 17.25% ±0.66 to 21.0% ±1.11, There were no significant changes in moisture content across Sidr honey samples from Arab countries, which varied from 17.70% ±0.224 to 18.00% ±0.447% (Zidan, 2019).

The chemical composition of different honey types from the New Valley Governorate, Egypt, showed minor variations, with no significant differences in moisture content, acidity, free acids, lactone content, or pH. These findings confirm that the tested honeys meet quality standards, with acidity and pH levels contributing to their stability and antimicrobial properties. The variations observed are likely due to differences in floral origin and environmental conditions.

2.2. Total acidity:

Total acidity variations are due to the presence of organic acids in equilibrium with their corresponding lactones, or internal esters, and some inorganic ions such as phosphates or sulfates. Lactonic acidity is the acidity reserve when honey becomes alkaline and total acidity is the sum of free and lactonic acidity. Honey acidity is calculated as free, lactonic and aesthetic acidity. It is an important quality criterion. The total acidity values of Egyptian honey samples ranged from 25 to 43.93 meq/kg. Data in Table (2) indicated that there were no significant differences in the total acidity value between the tested honey types, as these values ranged from 35.6 meq/kg for mixture Acacia and Eucalyptus honey to 49.03 meq/kg for clover honey, and reached 48.2 meq/km and 39.16 meq/km for Alfalfa bee honey and Tamarisk bee honey, respectively, the present results are consistent with those reported by Abdel-Aleem (2002); Downey *et al*. (2005); Sanz *et al*. (2005) and Moussa (2005) found that the total acidity value of honey ranged from 11.20 to 53.50 meq/kg.

2.3. Free acidity and lactone:

The free acidity of honey depends on the organic acids naturally present, as well as the floral source and bee species (De Sousa *et al.*, 2016).

Data in Table (3) revealed that the highest free acidity was observed in clover honey (31.36 meq/kg), while the lowest was found in Mixture Acacia and Eucalyptus honey (19.5 meq/kg).). Although differences were observed, they were statistically insignificant (P = 0.1862). The results also showed that the lactone range was 15.93-21.63 meq/kg (table 3). Abdel-Hameed (2020) showed that free acidity (11.0 ± 1.32 to 68.3 ± 0.85), lactone (7.5 ± 0.70 to 17.5 ± 0.70), and total acidity (18.5 ± 1.05 to 86.0 ± 0.70). El-Dereny (2023) found that free acids were 27.00 ± 3.08 and 36.17 ± 1.26 (meq/kg), lactone 1.13 ± 0.48 and 4.94 ±6.40 (meq/kg) and total acidity 28.13 ±2.95 and 37.83 ±2.02 (meq/kg).

2.4. pH value:

Data tabulated in, Table (2) Indicated that the pH values of the honey samples ranged from 3.733 (Alfalfa honey) to 4.19 (Tamarisk honey), with no significant differences (P = 0.264). Alfalfa bee honey had the lowest value of pH (3.73), followed by honey Mixture Acacia and Eucalyptus bee honey (3.966), Clover bee honey

(4.000), and Tamarisk bee honey had the highest pH value (4.190).

These results are in accordance with those reported by Nafea *et al*. (2023) they recorded that, the pH value of honey was 3.3 -3.93 for various types of honey produced in North Sinai Governorate, Egypt. Honey's pH plays a crucial role in its stability and antimicrobial properties, typically ranging between 3.2 and 4.5 (Bogdanov *et al*., 2008).

In general pH value is affected somewhat by the amounts of the various acids present, but mostly by the mineral content likely calcium, sodium, potassium and other ash constituents, as example honey rich in ash generally show high pH value (White, 1975).

3. The sugars content of the tested honeys:

Honey is super saturated suspension of carbohydrate materials, hence 95 to 99.9 % of the solids being sugars. Dextrose (Glucose) and laevulose (Fructose) the main sugars in honey. They represent about 85 % of the solids in honey. Sugars make up the largest portion of the dry matter in honey, which contributes to its characteristic physical properties such as high viscosity, high density, and resistance to spoilage. Monosaccharides make up approximately 75% of the sugars in honey, while disaccharides account for 10-15%, and small amounts of other sugars are also present (Da Silva *et al*., 2016).

3.1. Fructose content %:

Results in Table 3 showed that there was no significant difference between the studied honeys in fructose content.

Clover bee honey had the highest content of fructose 40.83 % followed by Tamarisk bee honey 40.7%, Mixture Acacia and Eucalyptus, 40.16% and Alfalfa bee honey 39.83%.

These results are in general agreement with El-Dereny *et al*, (2022), they found 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%. These values are consistent with previous studies indicating that fructose typically constitutes 38–43% of honey composition (Bogdanov *et al*., 2008 and White, 1978).

3.2. Glucose content (%):

The present results in Table (3). revealed that there are no significant differences between the clover honey, mixture (Acacia and eucalyptus) bee honey, alfalfa bee honey and Tamarisk bee honey in glucose content. Where the samples showed that the glucose content ranged from 29.70 to 32.86 %, clover honey had the highest glucose value, while Alfalfa bee honey had the lowest glucose value. Higher glucose concentrations, as seen in clover honey (32.86%), may promote a faster crystallization rate, which aligns with reports that honeys with higher glucose-to-fructose ratios crystallize more rapidly. These results were in the same line with those obtained by Manikis and Thrasivoulou (2001), indicated that the glucose content of honey was 25.2-41.3%.

3.3. Sucrose content (%):

The sucrose content in honey is a crucial parameter in determining its quality and authenticity. Several recent studies have examined the sucrose levels in honey from different geographical regions and floral sources, contributing to a more comprehensive understanding of its variability.

The analysis of variance carried out for the results of sucrose content for the different studied honey kinds under investigation. Sucrose levels showed significant variation among the honey samples ($p = 0.022$) (Table 3), recorded the sucrose content of the studied honeys varied from 2.133 %for Tamarisk bee honey to 5.133 % for clover honey, and it reached 2.300 % and 4.733 for Alfalfa bee honey and Mixture (Acacia and Eucalyptus) honey, respectively.

Recent research by Berhanu *et al.* (2023) analyzed 320 monofloral honey samples from Ethiopia and found that the sucrose content ranged between 1.11 and 3.40 g/100 g. These values align with international honey quality standards, ensuring that the honey is not adulterated with added sugar. Similarly, Ahmed *et al.* (2024) studied honey samples from different regions in Saudi Arabia and reported that the sucrose concentration in Acacia honey from the Asir region was 4.68%, which was significantly higher than that from the Jazan region at 1.82%. This highlights the influence of geographical and floral sources on the sugar composition of honey.

In another study, Johnson *et al.* (2022) conducted an in-depth analysis of sugar composition in various honey samples, finding that sucrose levels ranged from 3.20% to 3.90%. These values remain below the 5% mass ratio limit, which is a common threshold for unadulterated honey. The study underscores the importance of maintaining strict quality controls to prevent honey adulteration.

3.4. Maltose content (%):

Maltose is a disaccharide formed during the enzymatic conversion of starch and is typically present in honey at varying levels depending on nectar composition and enzymatic activity (Da Costa Leite *et al.,* 2000).

The data presented in Table (4) showed that the maltose content of the tested honeys were varied from 3.566±4.02 % for Clover bee honey to 7.73±1.078 % for Alfalfa bee honey, and it reached 4.00±2.5%, 6.80±4.42 % for Mixture (Acacia and Eucalyptus) bee honey and Tamarisk bee honey, respectively. The present results are consistent with those reported by Nafea *et al.* (2023)) theypresent the measured values of chemical properties for various types of honey produced in North Sinai Governorate, Egypt, such as Citrus Algae, Myrrhan, and Tarot. The sugar values ranged from 39.21% to 40.33% for fructose, 26.53% to 29.6% for glucose,3.733% to 5.966% for sucrose, and 6.66% to 8.36% for maltose.

Nafea *et al*. (2014) reported that the sugar values of some Egyptian honey ranged from 35.1% to 38.9% for fructose, 27.7% to 32.0% for glucose, 0.75% to 2.5% for sucrose, and 2.0% to 5.0% for maltose. El-Dereny (2023) found that medicinal and aromatic honeys sugar were fructose ranged 38.50 ±5.16 and 43.11±3.88 (%), glucose 31.55 ±5.54 to 33.46 ±1.29 (%), sucrose 1.24 ±1.00 and 3.00 ±0.78%, maltose (%) 1.90 ±0.72 to 3.75 ±1.45.

Table (1): The Physical Properties of some types of bee honey produced in New Valley Governorate, Egypt.

Physical properties	Alfalfa bee honey	Tamarisk bee honey	Mixture Acacia and Eucalyptus bee honey	Clover bee honey	F	P	L.S. D
V.*	66.67±	72±	75±10.68	69±0.000	0.465	0.714	16.86
S.g*	1.416±0.006(a)	1.420±0.00 (a)	1.413±0.002 (a)	1.410±0.018 (a)	0.6060	0.6294	0.0180
EC*PPm	0.0066±0.008 (a)	0.0166±0.009 (a)	0.0143±0.008 (a)	0.023±0.009 (a)	0.9310	0.491	0.0144
TSS (%)*	821.00 (a)	821.144 (a)	82.16±0.289 (a)	82.16±0.288 (a)	0.0266	0.993	1.921

L.S.D: Least significant differences V.: (Viscosity) S.g : (Specific gravity) EC :(Electrical conductivity) TSS :(Total soluble solids)

Table (2) : The chemical composition of some types of bee honey produced in New Valley Governorate, Egypt.

The Chemical composition	Alfalfa bee honey	Tamarisk bee honey	Mixture Acacia and eucalyptus bee honey	Clover bee honey	F	P	L.S. D
Moisture (%)	18±1.0(a)	17.83±1.44(a)	17.83±0.29(a)	17.66±0.288(a)	0.0683	0.99752	0.697
Total Acidity meq/km	48.2± 10.153(a)	39.16±1.3(a)	35.6±2.869(a)	49.03±10.639(a)	1.997	0.193	15.357
Free acids meq/km	26.66±6.408(a)	22.13±5.536(a)	19.5±0.289(a)	31.36±9.35(a)	2.044	0.1862	11.90
lactone meq/km	21.63±3.87(a)	17.13±9.358(a)	15.93±2.759(a)	17.66±4.53(a)	0.6408	0.586	10.519
pH	3.733(a)	4.190(a)	3.966(a)	4.000(a)	1.600	0.264	0.486

Table (3): The sugars contents of some types of bee honey produced in New Valley Governorate, Egypt.

Physical properties	Alfalfa bee honey	Tamarisk bee honey	Mixture (Acacia and eucalyptus) bee honey	Clover bee honey	F	P	L.S.D
Fructose %	39.83±1.76(a)	40.7±1.756(a)	40.16±1.258(a)	40.83±1.803(a)	0.0249	0.8593	3.041
Glucose %	29.70±0.874(a)	31.23±2.483(a)	32.76±3.042(a)	32.86±2.203(a)	1.225	0.3019	4.318
Sucrose %	2.300±0.77(b)	2.133±1.40(b)	4.733±1.527(ab)	5.133±3.21(a)	5.694	0.0220	2.137
Maltose %	7.73±1.078(a)	6.80±4.42(a)	4.00±2.5(a)	3.566±4.02(a)	1.214	0.3653	6.079

The findings indicate that while minor variations exist among the tested honey samples, no statistically significant differences were observed in key parameters, affirming the consistency and quality of the honeys. Overall, the studied honey samples exhibit high-quality attributes with minimal variation in physical and chemical characteristics. The findings confirm compliance with international honey standards, reinforcing their suitability for human consumption and industrial applications. These results highlight the impact of floral source and environmental conditions on honey properties while ensuring that the tested honeys maintain their purity, stability, and beneficial health attributes.

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