

THE PHYSICO-CHEMICAL PROPERTIES AND COMPOSITION OF HONEYS RESULTED FROM DIFFERENT BOTANICAL ORIGIN

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ABSTRACT

A total of 24 honey samples from eight different botanical sources were analysed. Results are presented for : specific gravity, viscosity, colour, electrical conductivity (Ec) % and total solids %. Moisture %, pH, free acidity mq/kg, Lactone (mq/kg), total acidity (mq/kg), Proline mg/100gm and hydroxy methyl forfural (H.M.F.).

The results of physical properties showed that the specific gravity was parallel and ranged from (1.39 - 1.42). The viscosity ranged from (13.6 - 48.1 poise), the color ranged from (0.02 - 0.38 ppm), the electrical conductivity (Ec) ranged from (110 - 520 ppm), whereas the total soluble parallel and ranged from (79 - 87.75%).

The results of chemical composition showed that the moisture ranged from (17 - 21%), the pH value ranged from (3.7 - 4.7). The total acidity content is parallel and ranged from (18.5 - 86%) mq/kg., the proline content ranged from (31.6 - 56.6 mg/100 gm). The HMF content ranged from (2 - 23.04 mg/kg) in the different types of tested honeys. From data obtained it is clear that the biochemical variation can be useful separating at least some different honeys within different types.

Discriminant analysis was used to distinguish honeys, using eight variables for 24 samples (eight plant sources). From data obtained it was found that the most useful characteristics were electrical conductivity, HMF, free acidity and Proline content.

INTRODUCTION

Honey, a viscous and aromatic produced appreciated since ancient Grecian times, is prepared by bees mainly from nectar of flowers or honeydew. The chemical composition of honey is complex and the contents of individual constituents vary considerably (Zander and Koch, 1975; White, 1978). Surveys of floral honey composition have established that the three major components are fructose, glucose and water (Doner, 1977). In addition di- and trisaccharides and some higher have also been identified (Nikolov *et al.*, 1984; Mauch *et al.*, 1987; Swallow & Low, 1990; Foldhzi, 1994). Some authors such as Kirkwood, *et al.* (1960). Characterised honeydew honeys by a discriminant function based on the pH value and the percentages of ash and reducing sugars. Other authors, have characterised spanish unifloral honey by discriminant analysis using the following Parameters; pH, water content, sugars, colors and electrical conductivity (Mateo & Boch-Reig (1998), Vorwehl (1964) concluded that, measurement of the electrical conductivity (EL) does give information, on the botanical origin of honeys. EC, is a characteristics of the plant species from which the honey is derived. Honeys of the same floral origin have approximately the same conductivity, even if they are of different harvest years, come from different geographic regions and different climates.

Hydroxy methyl furfural (HMF), is the compound which may be formed by decomposition of fructose in the presence of acid, during unsuitable condition of storage, Crane (1979). The accumulation of a decomposition product of the sugars (Hydroxy methyl furfural or HMF) was measured as an index of heat-induced chemical change in the honey. Honey as a natural product of limited supply and relatively high price traditionally has been a target for adulteration. The adulteration of honey with various sweet syrups without fear of detection is a great threat to the integrity of the honey market and to the fair treatment of consumers. Several research works were done to develop methods for the detection of honey adulteration (Lucchesi, 1979; Deifel *et al.*, 1985; White *et al.*, 1986; Lipp *et al.*, 1988).

The honey has been suggested as an environmental indicator (Bogdanov *et al.*, 1986; Jones (1987) and Fodor & Molnar, 1993). concluded that the low concentration of heavy metals in honey and their inherent variability detract from the reliable use of honey for monitoring purposes.

In recent years research efforts were done in order to find more characteristic components for classifying honeys (Blank, 1990, Castro *et al.*, 1992; Delgado *et al.*, 1994). The aim of this work to search for significant parameters in distinguishing honeys of different botanical origin.

MATERIAL AND METHODS

24 samples of Egyptian honeys (multiflora from El-Minya, camphor, Banana, Bardkoush, multiflora from Beni-Suef, Cedar, Cotton north and Cotton upper) were collected and measured in (2004). All the samples were analyzed for the following properties :

1. **Physical properties**, which includes, specific gravity, viscosity, colour, electrical conductivity (EC) % and total solids %.
2. **Chemical properties**, which includes, Moisture %, pH, free acidity mg/kg, Lactone (mq/kg), total acidity (mq/kg), Proline mg/kg and hydroxy methyl furfural (H.M.F.).

1. The Physical properties :

The specific gravity was measured to (Wedmore, 1955).

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

The colour was measured according to (White, 1978).

The electrical conductivity (%) was measured in a 20% solution (regarding the dry substance of the honey) at 20°C using a conductometer (Viorwohl, 1964), the total soluble solids (TSS) (%) was measured according to (The AOAC, 1990).

2. The Chemical Properties :

Moisture determination was performed by measuring the refractive index value (Abbe refractometer at 20°C). Average refractive index value were converted to honey moisture contents using the table developed by Wodmore (A.O.A.C., 1990a).

pH was measured by a pH meter (orion 420A) in a solution containing 10g honey in 75 mL of CO₂ free distilled water (AOAC, 1990). The titrimetric method was used to analyse the acidity (free, lactone and total acid) of the samples (A.O.A.C. 1990b). The proline content was measured by the

German standard method (Din, 1991). The principle of the method is that proline reacts with ninhydrin solution to form colored compounds. The concentration is determined with a spectrophotometer using a wavelength of 520 nm and water blank Hydroxy methyl furfural (H.M.F.) was determined after clarifying samples with carrez reagents (1 and 11) and the addition of sodium bisulfate (A.O.A.C. 1990) absorbance was determined at 284 and 336 nm in a 1 cm quartz cuvette in a spectrophotometer (Milton Roy uv-vis spectronic 3000 Array).

RESULTS AND DISCUSSION

The physical properties :

1.The specific gravity :

Another physical characteristics of practical importance is density. Honey density expressed as specific gravity, it depends on the water content of the honey. Because of the variation in density it is sometimes possible to observe distinct stratification of honey in large storage tanks. The higher water content (less density) honey settles above the denser, drier honey. The densities of tested honeys (Table 1) could be ranged between 1.39 - 1.42, these densities fall within those found by White, (1975); ranging between 1.421 - 1.423.

2.Viscosity :

Viscosity is an important technical parameter during honey processing, because it reduce honey flow during extraction, pumping, setting, filtration, mixing and bottling. Results in Table (1) showed that the tested honeys fall into four groups in relation to viscosity values arranged in descending order as follows :

- 1.Cedr, cotton nourth and cotton upper honeys with 34.9, 36.9, 34.9 Poise.
- 2.Multifloura from El-Minya region and camphare 69.0 Poise.
- 3.Banana and Bardkoush with 20 and 48.1 Poise.
- 4.Multifloura from Beni-Suef with 13.6 Poise.

As pointed out by (White, 1975), the variations in viscosity of honey are du primarily to tempreature and water content where the viscosity values were; 2.6, 10.7, 21.4, 68.4, 189.6 and 600.0 Poise (Crane, 1990) related high viscosity of honey to high sugar and low water content, and (Pierro, 1994) reported that the viscosity is reduced when the temperature raside to 30°C. (Abd El-Bary and Misherf, 1993) found that the viscosity in clover and colton honeys were 24, 34 and 31.52 Poise, respectively, (Mishref *et al.*, 1999) found that the viscosity of clover, cotton and sunflower honeys were 55.56, 63.48, and 116.0 Poise respicively.

3.Color :

Honey varies tremendously in color and flavor, depending largely on its floral source. Its composition also varies widly, depending on its floral sources. Colors of honey form a contrinous range from very pale yellow through ambers to a darkish red amber to nearly black. The variations are almost entirely due to the plant source of the honey although climate may modify the color somewhat through the darkening action of heat, comb color.

Table (1) : Physico properties of some Egyptian honey types.

| Parameters | (Multifloura) from El-Minya | Camphor | Banana | Bardkoush | (Multifloura) from Beni-Suef | Cedr | Cotton Nourth | Cotton Upper |
|-------------------|-----------------------------------|---------|--------|-----------|------------------------------------|-------|------------------|-----------------|
| Specific gravity | 1.42 | 1.42 | 1.4 | 1.4 | 1.39 | 1.41 | 1.41 | 1.41 |
| Viscosity (poise) | 69.0 | 69.0 | 20.0 | 48.1 | 13.6 | 34.9 | 36.9 | 34.9 |
| Colour (ppm) | 0.02 | 0.19 | 0.38 | 0.16 | 0.23 | 0.16 | 0.12 | 0.13 |
| E.C. (ppm) | 170.0 | 200.0 | 520.0 | 170.0 | 380.0 | 470.0 | 110.0 | 260.0 |
| Tss (%) | 87.75 | 83.0 | 80.0 | 81.5 | 79.0 | 80.5 | 81.0 | 80.5 |

Table (2) : Chemical properties and composition of some Egyptian honey types.

| Parameters | (Multifloura) from El-Minya | Camphor | Banana | Bardkoush | (Multifloura) from Beni-Suef | Cedr | Cotton Nourth | Cotton Upper |
|--------------------|-----------------------------------|---------|--------|-----------|------------------------------------|------|------------------|-----------------|
| Moisture (%) | 17.25 | 17.0 | 20.0 | 18.5 | 21.0 | 19.5 | 19.0 | 19.5 |
| PH | 4.1 | 4.1 | 3.7 | 3.8 | 4.1 | 4.7 | 3.9 | 4.0 |
| Free acidity | 19.0 | 11.0 | 68.3 | 21.0 | 33.5 | 16.0 | 13.5 | 25.0 |
| Lacton (mq/kg) | 12.5 | 10.0 | 17.5 | 12.5 | 15.0 | 7.5 | 10.0 | 12.5 |
| Total acidity | 31.5 | 21.5 | 86.0 | 33.5 | 18.5 | 23.5 | 23.5 | 37.5 |
| Proline (mg/100gm) | 36.6 | 31.6 | 45.0 | 56.6 | 55.0 | 56.6 | 35.0 | 38.3 |
| HMF (mg/kg) | 7.68 | 7.6 | 13.4 | 7.7 | 17.3 | 2.0 | 23.04 | 5.7 |

Color (as optical density) varies with botanical origin, age and storage condition, but transparency or clarity depends on the amount of suspended particles such as pollen. The color of the tested honeys falls into four groups in descending order as follows :

1. Banana and multiflora from Beni-Suef region honeys with 0.38 and 0.23 ppm respectively.
2. Camphor, Bardkoush and cedar honeys with 0.19, 0.16 and 0.16 p.p.m. respectively.
3. Cotton upper and cotton nouth honeys with 0.13 and 0.12 p.p.m. respectively.
4. Multiflora honeys from El Minya region with 0.02 ppm.

According to the color standard table (given by White, 1978) and Thawley (1969) explained that the dark color is partially attributed to condensation between amino acid and sugars in honey of the coloring materials are plant pigments (Hassan *et al.*, 1985) described the color of citrus, clover, and cotton honeys to fall between water white and extra white-Nour (1988) the range of color for clover honey was between 0.12 to 0.24 and for citrus honey between 0.12 + 0.24. The color by Abd-Elbarry and Mishref (1993) for clover and cotton honeys from Fayom (0.15 and 0.25) Nour (1991) showed lower values of color of Egyptian honeys (0.125, 0.202 and 0.18, respectively). Mishref *et al.* (1999) reported that the color ranged (0.12 - 0.24).

4. Electrical Conductivity (EC) ppm. :

The electrical conductivity first of all depends on the mineral content of the honey. Nectar honey have lower mineral content than honeydew honeys, so measuring their (E.C.) they can be distinguished. Considerable differences in the electrical conductivity (E.c.) values of the tested honeys fall into three groups in descending order as follows :

1. Multiflora honey from El-Minya region, Bardkoush, and cotton Nourth honeys with 170 ppm, 170 ppm and 110 ppm, respectively.
2. Camphor, cotton upper honeys with 200 ppm and 260 ppm respectively.
3. The mean of (E.C.) were 520 ppm, 470 ppm and 520 ppm for Banana, cedar and multiflora honeys respectively.

The statistical analysis showed that there was a significant difference in EC values of different honeys produced by different botanical origin. From data obtained it could be noticed that the (EC) value of Banana honey had the highest average 520 ppm. The lowest value was found in both multiflora from El-Minya, and Bardkoush honeys, 170 ppm and 170 ppm respectively. The high (EC) values are attributed to high mineral content (Nour, 1988; Accorti *et al.*, 1986) suggested that the measurement of (EC) could replace that of ash content in official analysis methods. Laurina and Gelli (2002) found that EC of citrus honeys was 0.185%.

Vorwohl (1964) suggests using the method for detecting adulteration with sugar feeding honey. It is also that the biochemical variation can be useful in separating at least some honeys within different types.

5. Total Soluble Solid (T.S.S.) :

The total soluble solids (T.S.S.) value of different honeys produced from different botanical origin were tested. Data recorded in Table (1)

indicated that the mean of T.S.S. of honeys resulted from different botanical origin were 87.75%, 83%, 80%, 81.5% for multiflora honey produced from El-Minya region, camphor, Banana and Bardkoush respectively. While these mean were 79%, 80.5%, 81.0% and 80.5% for multiflora honey and resulted from Beni-Suef region, cedr, cotton nourth and cotton upper honeys, respectively. According to data obtained, it is obvious that the T.S.S. ranged between 79% - 87.5%. The dry matter, which should be 78% or more, is responsible for protecting honey from fermentation.

In this respect, Hussein (1989) mentioned 76.83% T.S.S. in honey from Oman.

The chemical properties:

1-Water content (Moisture) :

Results presented in Table (2) showed that water content of different types and samples were in the normal value of fresh honey. The values were 17.35%, 17%, 20%, 18.5%, 21%, 19.5%, 19% and 19.5% for different types of honeys produced by different botanical origin srespectively. According to data obtained, it is showed the greatest average of moisture of different types of honeys was 21% for these produced by multiflora from Beni-Suef region. The lowest average was for that produced by camphore honey 17%.

The natural moisture of honey in the comb is that remaining from hte nectar after ripening. The amount of moisture is a function of the factors involved in ripening, including ewather condition and original moisture of the nectar. After esxtraction of the honey, its moisture content may change depending on conditions of storage. It is one of the most important characteristics of honey, influencing keeping quality, groniulation, Ghazali and Sin (1986) found that honeys showed a slight decrease in water content during storage at different temperature, while Vorwhole *et al.* (1988) mentioned that, honey usually loses water content when stored in a country with a relatively low air humidity such as Egypt.

2.Acids :

The flavor of honey results from the he lending of many "Motes" not the least being a slight tartness or acidity. The acids of honey account for less than 0.5 percent of the solids, but this level contributes not only to the flavor, but is in part responsible for the excellent stability of honey against microorganisms. Parameters for acidity and HMF (Table 2) were used to determine, the degree of deterioration of the honey. They were all found to be within acceptable limit of all samples. Owing to the presence of organic acids, in equilibrium with their corresponding lactons, or internalesters, and some inorganic ions such as phosphate and sulphate, the values for the free acidity for eight types of different honeys resulted from different botanical origin ranged from 13.5 to 68.0 mq/kg. According to data obtained, it is obvious that the greatest value of free acidity was found in honey resulted from Banana origin 68 mg/kg while the lowest value of free acidity was found in cotton mourth, 13.5 mq/kg (Table 2). The lactone acidity (considered as the acidity reserve when the honey become alkaline) rquences between 12.5 to 17.5 mq/kg. According to data obtained it is obvious that the greatest value of lacton was found in honey resulted from Banan origin 12.5 mq/kg. The lowest

value of lacton was found in cotton upper and multifloura from El-Minya 12.5 mq/kg respectively. While the total acidity ranges between 18.5 to 86.0 mq/kg.

The statistical analysis showed that there were significant difference among the total acidity of different types of honeys resulted from different botanical origin.

From data obtained, it was found that the greatest value of total acidity was found in honey resulted from Banana origin 86 mq/kg. The lowest value of total acidity was found in multifloura honeys from El-Mynia region 18.5 mq/kg.

Comparable results are reported by other authors Terrab *et al.* (2002) reported that free acidity ranged from 10.3 - 102, lactic acid ranges 0.01 and 21.4, while total acidity ranges between 12.6 - 115 mg/kg. Gomez, *et al.* (1993) found that free acidity 25.34, lactones, 20.94 and total acidity 28.28. Foldhazi, (1994) showed the free acidity ranges between 4.98 - 25.7, lacton 3.39 - 10.9 and total acidity between 8.38 - 31.3 mq/kg. Bogdanov's (1997) reported that, the Bacterial inhibition correlated with acidity but not the pH of the honey.

3.The amino acids :

The amino acids are simple compounds obtained when proteins are broken down by chemical or digestive process. They are the "building blocks of the proteins". Several of them are essential to life and must be obtained in the diet. The quantity of free amino acids in honey is small and of no nutritional significance. Break throughs in the separation and analysis of minute quantities of material (Chromatography) have revealed that various honeys contain 11 to 21 free amino acids proline, glutamic acid, alanine, phenylalanine tyrosine e.c. Among free amino acids proline predominates representing 50 - 85% of the total (Whitte, 1978). In this experiment the values for proline content of different types of some different of honeys resulted from different botanical origin ranged between 31.5 mg/100g, 56.6 mg/100 gm. Table (2) According to data obtained, it is obvious that the lowest value of proline was found in honey resulted from camphor origin 31.6 mg/100g, while the greatest value of proline was found in both Bardkoush and cedr honeys, 56.6 and 56.6 mg/100gm respectively. The statistical analysis showed that there was significant difference among the proline content of different types of honeys resulted from different botanical origin. Davies (1976) using data for 98 honey samples has suggested that certain ratios between contents of various amino acids could be used for determining the geographic source of honey.

He found that while there are variations in the ratios between samples in the same area, the variation between sources is much greater. Foldhazi, (1994) who found that the values of proline ranges between 199-426 mg/100g, Gomez *et al.* (1993) found that the mean value of proline is 46.4 µg/100g and Terrab *et al.* (2002) reported that the values of proline ranges 1.5 - 2.3 mg/100mg. In my opinion, by using proline content is a best indicator of honey purity, but the wide range of values found precluded the usability of it. From data obtained it could be said that when the different type

of honey contained different amount of proline it due to biochemical variation can be useful in separating at least some honeys within different types.

4. Hydroxymethyl furfural (HMF) :

HMF, is a breakdown product of certain sugar solution particularly, fructose, stored. Stored at high temperature or for a long time or honey adulteration with invert sugar prepared by acid hydrolysis at high temperature. HMF is the good indicator index of heat-induced chemical change in the honey.

In this study the value of HMF of different types of honey resulted from different botanical origin ranged between 2 to 23.04 mg/kg. From data obtained it is clear that the greatest value of HMF was found in honey resulted from cotton Nourth 23.04 mg/kg, while the lowest value of HMF was found in honey resulted from cedar origin. The statistical analysis showed that there was a significant difference among the HMF parameter of different types of honeys resulted from different botanical origin. comparable results are reported by other authors. Laurino and Gelli (2002) were found that the values for HMF ranged between 2.0 - 26.0 mg/kg. Nour *et al.* (1991) found that the HMF values ranged between 1.27 - 19.13 mg/kg in fresh nesses honeys. Gomez *et al.* (1993) reported that the a mean value of 3.6 mg/kg for HMF. From data obtained it is clear that the biochemical variation can be useful separating at least some different honeys within different types.

In my opinion it was suggested that different factors affected the composition of different types of honeys resulted from different botanical origin and different regions :

- a. Effect of crop year.
- b. Effect of storage.
- c. Effect of area of production.
- d. Granulation.
- e. Color,.
- f. Variation in bee keeping treatment.

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الصفات الطبيعية والكيمائية والتركيب الكيماوى لبعض الأعسال الناتجة من مصادر نباتية مختلفة

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فى هذه الدراسة تم استخدام ٢٤ عينة من أعسال مختلفة من مصادر نباتية مختلفة ومن مناطق مختلفة وهى :-

- ١- أعسال ناتجة من نباتات متعددة من محافظة المنيا.
- ٢- عسل الكافور.
- ٣- عسل الموز.
- ٤- عسل اليردقوش.
- ٥- أعسال من مصادر نباتية متعددة من محافظة بنى سويف.
- ٦- عسل ناتج من نبات السدر.
- ٧- عسل القطن من شمال الدلتا.
- ٨- عسل القطن من جنوب الدلتا.

وقد تم تحليل عدة صفات طبيعية وكيمائية لهذه الأعسال الناتجة من نباتات مختلفة ومناطق مختلفة وذلك للصفات الطبيعية والكيمائية الآتية : نسبة الرطوبة، اللزوجة، اللون، خاصية التوصيل الكهربى، نسبة المواد الصلبة، درجة pH، وكذلك الأحماض الحرة والأمينية مثل البرولين وكذلك HMF. والفرض من هذه الأدراسة هو محاولة تحديد وتحليل الصفات المختلفة لهذه الأعسال حتى يمكن استخدامها فى التفريق والتمييز بين أنواع الأعسال المختلفة. وأوضحت النتائج أن هناك فروق معنوية وأنه يمكن استخدام الصفات الآتية فى تمييز أنواع الأعسال المختلفة حيث يكون ذلك مفيدا فى التعرف على جودة ونقاء الأعسال المختلفة وهى درجة التوصيل الكهربى EC ، HMF ، والأحماض الحرة والأحماض الأمينية والمتمثلة فى حمض البرولين.