

Physicochemical Properties Evaluation for Some Yemeni Honeys

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Abstract: In this study the quality properties of Yemeni honeys were investigated. Two types of honey, monofloral (Sidr) and multi-flora honey originated from three different regions in Yemen (Hadramout, Dhamar and Amran governorates), were included in this study. Honey samples were collected and analyzed to determine their physicochemical properties such as (moisture content, total soluble solid (TSS), pH, free acidity, reducing sugars, sucrose, vitamin (C), electrical conductivity (EC), refractive index, water insoluble matter (WIM) and density) and to determine effect of geographical area and botanical source on the physicochemical properties of honey. The results showed that moisture content ($18.63\% \pm 1.39$), total soluble solid (TSS) content ($79.58 \pm 1.42^\circ\text{Brix}$), pH (4.26 ± 1.17), free acidity (17.16 ± 5.56 meq/kg honey), reducing sugars ($63.86\% \pm 3.50$), sucrose content was ($4.4\% \pm 1.12$), total sugar ($68.27\% \pm 3.55$), vitamin C (0.65 ± 0.226 mg/100g), the electrical conductivity (EC) ($731.08 \pm 273.87 \mu\text{S/cm}$), refractive index (1.4958 ± 0.0038), while water insoluble matter (WIM) was ($0.092\% \pm 0.033$) and the density was (1.3871 ± 0.035 g/cm³). Results obtained from this study showed that honey samples coincide with those specified by Yemeni standard organization (YSMO) and Arabian Gulf standard organization (GSO). Botanical source affected on the pH, sucrose content and the electrical conductivity (EC) values of honey, which was higher in Sidr honey than multi floral honey samples. The geographical area caused a variation in the moisture, free acidity and vitamin C content of honey, while other physicochemical properties for honey had a negligible variation between honey from the different regions and botanical sources.

Keywords: Honey Quality, Yemeni Honey, Physicochemical Properties, Quality Parameters

1. Introduction

Honey is a natural food produced by bees from nectar, secretion of flowers or from excretion of plant sucking insects when living on plants. The chemical composition of honey contain water (15-17%), carbohydrates (80-85%), proteins, (0.3%), ashes (0.2%), amino-acids and vitamins in low concentration [1, 2].

Fresh honey have density value higher than the water density and its color in basically is yellow amber and have low heat conductivity and surface tension [3]. The moisture contents and acidity of honey influencing in all characteristics and quality of honey [4]. Electrical

conductivity (EC) is main parameter for determining the physical properties of honey and it's used as indicator for the authentication of unifloral honeys [5, 6].

Floral source and environmental conditions affects on the quality parameters related to processing and storage of honey. Some quality parameters of honey like moisture content, surface tension, ash and pH differ significantly between honey types. Moisture content, pH and density in *Apis cerana* honey were slightly higher than *Apis mellifera* honey while viscosity, surface tension, optical density and refractive index of *Apis mellifera* were higher than *Apis cerana* honey [7].

AL-Zoreky *et al.*, [8] studied the Yemeni honey properties and conclude that the botanical source of honey influenced on ash and total acidity values, and also conclude that

Yemeni honey has higher concentrations of dry substances (% moisture < 17), total acidity (54.1 ± 10.6 meq/kg) and WIM (pollen grains) which may enhance its stability and nutritional value.

The aims of our study are determining of some physicochemical characteristics for two types of honey originated from three Yemeni governorates and to study the effects of geographical origin and botanical source on the physicochemical properties of honey.

2. Materials & Methods

2.1. Samples Collection

Two types of honey samples, mono-flora (*Sidr*) and multi-flora honey, originated from three different regions (Hadramout, Amran and Dharmar governorates) Yemen, were collected and transported to the laboratory of Biotechnology and Food Technology Department, Faculty of Agriculture, Thamar University where the analysis was carried out.

2.2. Physicochemical Analysis

2.2.1. Moisture Content and Total Soluble Solid (TSS)

Moisture content and total soluble solid was determined by using hand refractometer, Atago (Japan). All measurements were performed at 25°C.

2.2.2. Refractive Index

Refractive index of honey was measured by Abbe refractometer.

2.2.3. Density

Density of honey samples was measured by specific gravity bottle 25ml. All measurements were performed at 25°C.

2.2.4. Reducing Sugars and Sucrose Contents

The estimation of reducing sugars was carried out using the Layne-Enyon method. About 2.6 g of honey was weighed and transferred to a 500 ml volumetric flask. 5 ml of standardized Fehling's solutions A and B were transferred to a 250 ml Erlenmeyer flask containing 7.0 ml of water and 15.0 ml of honey solution. The Erlenmeyer flask was heated and 1.0 ml of methylene blue (0.2%) was added. Titration was carried out by adding the diluted honey solution until the indicator decolorizes.

Sucrose content was determined by inversion, adding 10 ml of dilute HCl, 50 ml of diluted honey solution and water in a 100 ml volumetric flask. The solution was then heated in a water bath, cooled and diluted to the mark. Finally, the Layne-Enyon method was applied and the sucrose content was obtained by difference.

2.2.5. Free Acidity and pH

10 g of sample was dissolved in 75 ml of CO₂-free water in a 250 ml beaker. The Electrode of calibrated pH meter (Hanna Instruments, Switzerland) was immersed into the solution and

the pH value was recorded. To determine the free acidity, the solution was then titrated with 0.10M NaOH to the end point of titration with phenolphthalein indicator and the amount of 0.10M NaOH solution consumed was recorded. Free acidity was calculated from the following equation.

$$\text{Free acidity (meq/kg)} = \text{Volume of 0.10M NaOH (ml)} \times 10.$$

2.2.6. Electrical Conductivity (EC)

20 g of dry matter of honey was weighed and dissolved in 100 ml distilled water. Electrical conductance of the specified solution was measured using previously calibrated digital conductivity meter (HANNA).

2.2.7. Water Insoluble Matter (WIM)

WIM was determined as the method of (IHC) [9]. 20 g of honey was weighed and dissolved in 200ml of water at about 80°C and mixed well. Sample solution was filtered through a dry crucible then the crucible was washed carefully and extensively with warm water until free from sugars, then the crucible was dried by microwave oven and cooled in the desiccators and weighted.

$$\% \text{ Insoluble matter in g/100 g} = \frac{m}{m_1} \times 100$$

Where: m=mass of dried insoluble matter.
m₁=mass of honey.

2.2.8. Vitamin C

Vitamin C content in honey samples was determined by titration method with iodine and starch as indicator [10].

2.3. Statistical Analysis

Statistical analysis was carried out by using (SPSS) program version 16 to establish the difference between the honey samples by means of their physicochemical parameters. The results are expressed as mean values, range of values and standard deviation (SD).

3. Results & Discussions

3.1. Quality Properties of Yemeni Honey

The results in table 1 showed that the physicochemical properties of honey samples studied were coinciding with those specified by YSMO [37] and GSO [38] standards for honey. All samples of honey included in the present study had good quality parameters.

These results are similar to the results obtained in honey from Mexico [11], Bosnia and Herzegovina [12] and the Northeast of Brazil respectively [13] which the physicochemical properties were within the ranges of international regulations. It also coincides with the results found in Nigerian honey [14], but it's not coincide with the results that found in some honey samples collected from local markets of apiary sites which had higher values in certain parameters than recommended as a result of a minor adulteration [15].

Table 1. Physicochemical properties of Yemeni honey.

Parameters	Range	Mean & SD
Moisture content (%)	16–19.5	18.63±1.39
Total soluble solid (TSS) (°Brix)	79–82.5	79.58±1.42
Reducing sugars (%)	60.90–70.44	63.86±3.50
Sucrose (%)	2.8–6.10	4.4±1.12
Total sugar (%)	64.05–74.29	68.27±3.55
pH	3.3–6.3	4.26±1.17
Free acidity (meq/kg)	12–25	17.16±5.56
Vitamin C (mg/100 g)	0.44–0.88	0.65±0.226
WIM (%)	0.06–0.145	0.092±0.033
Density	1.3234–1.4166	1.3871±0.035
EC (µS/cm)	306–980	731.08±273.87
Refractive index	1.4918–1.5029	1.4958±0.0038

3.1.1. Moisture Content

Honey quality depends on moisture content which can effect on its stability and resistance to microbial spoilage during storage [4]. The organoleptic properties and biological activity of honey is depends on its moisture content [16]. The results in table 1 shows that moisture content was 18.63±1.39% and ranged between 16–19.5% in Yemeni honey samples included in this study. These results are fallen in the range of Ethiopian honey (17.56–22.57%) [15], Yemeni honey (13.5–19.5%) [17] and the eastern Moroccan honey (15.39–19.37%) [18] but the moisture content was higher than the moisture contents of Nigerian honey which had average value (16.00±2.19 g/100 g) [14]. The moisture contents in monoflora honeys from Andean Region of Ecuador was 16.42±2.53, 18.62±1.84 and 14.63±2.74 in Avocado, Eucalyptus and Rapeseed honeys respectively [19]. The mean values of moisture found of honey from this study are similar to the values found in honey from Ibadan which was (18.30%) [20], but it was higher than the results previously obtained in Yemeni honey which was less than (17%) [8].

3.1.2. Total Soluble Solid (TSS)

Total soluble solid results were found to be in the range of 79–82.5°Brix and the mean value was 79.58±1.42 (Table 1). These values are greater than those found in Mustard, Litchi and Nigella honey from Tangail, Bangladesh which was 71%, 70% and 73% respectively [21].

3.1.3. Reducing Sugars, Sucrose and Total Sugar

Honey could be adulterating by addition of sucrose, corn syrup, molasses and banana. This practice can cause nutritional and health problems for consumers [22]. Reducing sugars and sucrose analysis results in honey were found (63.86%±3.50) and (4.4%±1.12) and ranged between (60.90–70.44%) and (2.8–6.1%) respectively (table 1). These results of reducing sugars content in honey are agree with those obtained in Ethiopian honey (50.31–79.56%) [15] and in Algerian honey (64.72±0.52 g/100g) [23], but don't agree with the results previously reported for Yemeni honey which was the mean percentage 71.6 g/100g [8].

The results of sucrose content were similar to that obtained in Argentina honey (4.05%) [2] and in Ethiopian honey (2.24–12.21%) [15], but it was more than that obtained in Nigerian honey (1.84±0.79 g/100 g) [14]. The previously results reported that the mean percentage of sucrose in

Yemeni honey was (2.1 g/100g) [8], whereas it was (10.2±0.45 g/100g) in Yemeni *Acacia Shabowah* (ASH) honey [24] which was higher than the standards limit of Codex Alimentations [25]. The mean value of total sugar in investigated honey was (68.27±3.55) and ranged from (64.05–74.29%) (Table 1). These results are similar to those obtained in Algerian honey (67.03±0.68 g/ml) [23], but they are lower than the results obtained in the honey from guinea savannah zones, Nigeria that the total carbohydrate are (98.69%) [26] and in the honey from northeast sub-region of Nigeria (82.30±2.03 g/100 g) [14].

3.1.4. Free Acidity

The free acidity of honey depends on the equilibrium between organic acids and lactones, or internal esters, and some inorganic ions, such as phosphate [27], whereas high acidity can be indicative of the fermentation of sugars into organic acids. In general, honey is acidic due to the presence of organic acids [28]. Results in table 1 show that the free acidity in Yemeni honey samples was (17.16±5.56) and ranged from (12 to 25 meq/kg honey). Its falls in the limit of Codex Alimentarius that free acidity should be less than (50 meq/kg honey) [29]. These results are fall in the range of the results obtained from Ethiopian honey (3.99 to 45.17 meq/kg) [15] and are similar to the results reported for the eastern Moroccan honey (5.77 and 19.47 meq/kg) [18]. They are also similar to the results found in honey from the guinea savannah zones of Nigeria (22.2 to 30.2 meq/kg of honey) [26], but they are lower than the results previously reported for Yemeni honey (54.1±10.6 meq/kg) [8].

3.1.5. pH

pH value of honey during the extraction process affects on the texture, stability and shelf life of honey, [28]. Results in table 1 shows that the mean value of pH in honey samples was 4.26±1.17 and it ranged from 3.3 to 6.3. These results are most similar to the results of the pH value in Nigerian honey (4.2) [26]. The pH values in Yemeni honey obtained in this study are in agreement with those found in Mustard, Litchi and Nigella honey from Tangail, Bangladesh which was 6.97, 5.8 and 4.78 respectively [21]. The range of the pH value in honey sample studied was higher than that obtained in Ethiopian honey (3.40–4.65) [15], in the eastern Moroccan honey (3.53–4.94) [18], in Algerian honey (3.70–4.00) [23] and in honeys available in Mexican market (3.71–4.21) [11]. It's also higher than the results previously in *Sidr Dowany* and *Somer Shabowah* Yemeni honey which was 4.5 and 5.2 respectively [17].

3.1.6. Vitamin C

The results in table 1 show that the mean value of vitamin C in Yemeni honey samples was (0.65±0.226 mg/100 gm honey) and ranged between (0.44 to 0.88 mg/100 g honey). These results are in agreement with those reported in Mustard, Litchi and Nigella honey from Tangail, Bangladesh which was 0.41, 0.63 and 0.7 mg/100ml respectively [21].

Honey contains phenolic compounds and vitamin C which have antioxidant properties. The mean value of vitamin C in

honey from the northeast sub-region of Nigeria was 21.15 ± 3.99 mg/100 g [14].

3.1.7. Water Insoluble Matter (WIM)

The results in table 1 show that the mean value of water insoluble matter (WIM) content in Yemeni honey samples was (0.092 ± 0.033) and ranged from (0.06 to 0.145%). These results are similar to the results founded in Bosnia and Herzegovina [12], Brazilian honey [30] and in Algerian honey [31]. They are also similar to the results in honey from Northeast of Brazil $(0.003-0.067$ g/100 g) [13].

3.1.8. Density

The density of honey is related to its moisture content; which the honey density is decreasing if its moisture content increased [7].

The results in table 1 show that the density of honey samples ranged from (1.3234) to $(1.4166$ g/cm³) and the mean value was $(1.3871 \pm 0.035$ g/cm³). These results are agree with the results reported in honey from the guinea savannah zones of Nigeria $(1.31-1.65)$ [26]. The mean values of density reported here are similar to those reported for Algerian honey (1.329 ± 0.048) [31], while they are higher than the density of *A.cerana* and *Apismellifera* honeys which was (1.056) and (1.060) respectively [7], but they are lower than those reported in Sidr, Manuka and Nigella stiva honeys $(1.47, 1.433$ and 1.47 g/ml) respectively [32].

3.1.9. Refractive Index

The results in table 1 show that the mean value of refractive index in Yemeni honey samples was (1.4958 ± 0.0038) and ranged from $(1.4918$ to $1.5029)$. These results reported here for Yemeni honey are most similar to the results of Libyan honey $(1.4935$ and $1.5065)$ [33], but they are lower than the refractive index values of *A. mellifera* and *A. cerana* honey from Jammu and Kashmir which was (2.235 ± 0.9) and (2.2305 ± 0.3) respectively [7].

3.1.10. Electrical Conductivity (EC)

The electrical conductivity (EC) of the honey is closely related to the concentration of mineral salts, organic acids and proteins; it is variable depends on the floral origin and it uses as a parameter for differentiating between honeys with different floral origins [34]. The mean value of electrical conductivity (EC) in honey samples included in this study was $(731.08 \pm 273.87$ μ S/cm) and ranged from $(306$ to 980 μ S/cm). The upper limit of the EC range in Yemeni honey samples reported here are above the Codex Alimentations limit [25]. These results are similar to that previously reported in *Sidr Dowany* and *Acacia Shabowah* Yemeni honey which was (1.70 ± 1.4) and $(2.95 \pm 1.0$ mS/cm) respectively [24]. These results are also similar to the results found in commercial honeys available in Mexican market $(0.290-0.913$ mS/cm) [11] and in honey sample obtained from Ibadan $(0.64$ mS/cm) [20], but these results are higher than the results obtained in Ethiopian honey $(0.13-0.56$ mS/cm) [15] and in the eastern Moroccan honey $(108$ and 626 μ S/cm) [18].

3.2. Effect of the Botanical Source on Honey Physicochemical Properties

Honey composition influence by many factors, like the floral origin and environmental conditions [35]. These factors have a role in honey quality parameters especially during the processing and storage [7].

The results in table 2 shows that the botanical source had an effect in some honey properties. The sucrose content, pH and electrical conductivity (EC) were higher in *Sidr* honey than multi-floral honey samples, which were 4.93%, 5.16 and 918.16 (μ S/cm) in *Sidr* honey and 3.87%, 3.36 and 544 (μ S/cm) in multi floral honey samples respectively, while the moisture content, total soluble solid, free acidity and vitamin C content of both types of honey samples included in this study have a little variation. It means that the botanical source had a negligible effect on these properties of honey.

Table 2. Effect of the botanical source on honey physicochemical properties.

Parameters	Honey Type.	
	Mono-floral (<i>Sidr</i> honey).	Multi-floral honey.
Moisture (%).	18.23 \pm 1.93	19.03 \pm 0.80
Total soluble solid (TSS) ($^{\circ}$ Brix)	80.16 \pm 2.02	79 \pm 0.0
Reducing sugars (%).	63.51 \pm 1.09	64.21 \pm 5.39
Sucrose (%).	4.93 \pm 1.05	3.87 \pm 1.09
Total sugar (%).	68.44 \pm 1.35	68.09 \pm 5.44
pH	5.16 \pm 1.00	3.36 \pm 0.11
Free acidity (meq/kg).	17.66 \pm 4.93	16.66 \pm 7.23
Vitamin C (mg/100 g).	0.73 \pm 0.25	0.56 \pm 0.23
WIM (%).	0.091 \pm 0.02	0.093 \pm 0.04
Density.	1.3824 \pm 0.05	1.3919 \pm 0.02
EC (μ S/cm).	918.16 \pm 56.25	544 \pm 281.68
Refractive index.	1.4980 \pm 0.004	1.4936 \pm 0.001

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, [11]. The EC values of honey change if the plant pollen content was decreased [23].

High pH value in *Sidr* honey samples are similar to the results obtained in *Sidr* honey which had higher pH (6.0) than Manuka and Nigella stiva honey [32].

The variation in EC value between honey floral types included in this study is similar to the variation found in Avocado, Eucalyptus and Rapeseed honeys which was (1.31 ± 0.47) , (0.39 ± 0.03) and (0.20 ± 0.01) (mS/cm) respectively [19]. Moisture content is varies among the different types of floral honey [18].

3.3. Effect of the Geographical Origin on Honey Physicochemical Properties

The honey composition and moisture contents change by many factors including environmental factors, geographical floral origin, season, treatment of beekeepers and honey storage [19, 36].

Table 3. Effect of the geographical origin on honey physicochemical properties.

Parameters (Mean).	The Region		
	A (Hadramout)	B (Dhamar)	C (Amran)
Moisture content (%).	17.05 a±1.48	19.35 b±0.21	19.5 b±0.0
Total soluble solid (TSS) (°Brix)	80.75±2.47	79±0.0	79±0.0
Reducing sugars (%).	61.92±1.35	66.66±5.34	63.15±2.49
Sucrose (%).	4.50±0.67	4.975±1.59	3.735±1.32
Total sugar (%).	66.425±0.68	71.635±3.75	66.22±3.81
pH	4.8±2.12	3.95±0.77	4.05±0.92
Free acidity (meq/kg).	12 a±0.0	16.5 a±4.94	23 b±2.82
Vitamin C (mg/100 g).	0.835 b±0.063	0.66 ab±0.31	0.44 a±0.0
WIM (%).	0.115±0.042	0.065±0.070	0.0975±0.035
Density.	1.3677±0.0626	1.4046±0.0168	1.3891±0.0255
EC (µS/cm).	687.75±306.53	688±398.80	912.5±88.38
Refractive index.	1.4992±0.052	1.4951±0.023	1.4931±0.019

* Different superscript along a row indicates significantly different (P<0.05).

The results in table 3 show that the geographical origin had an effect on some honey physicochemical properties. These results indicated that moisture, free acidity and vitamin C contents in honey samples were varying significantly (P<0.05), while no significant differences (P<0.05) between other physicochemical properties of honey samples from different regions. The free acidity in honey samples from region (A) was lower than the honey samples from regions (B) and (C) which was 12, 16.5 and 23 (meq/kg honey) respectively.

Honeys contain some organic acids which make it naturally acidic, contribute to its flavor and enhance the resistance of honey to microbial spoilage. The source of nectar and environmental conditions may cause the variation in free acidity of honeys [28].

Moisture content in honey samples obtained from region (A) was lower than honey samples from (B) and (C) region area which had higher moisture content and it was (17.05), (19.35) and (19.5%) respectively. This variation in moisture content of honey samples is due to the difference in climate conditions between the regions. The region (A) climate temperature was higher than the regions (B) and (C).

Moisture content of honey differs depending on the temperature and relative humidity values in the geographical origin during honey producing [17].

Vitamin C content was the highest in honey from region (A) and the lowest in the honey sample from region (C).

4. Conclusions

This evaluation study intended to determine some physicochemical characteristics of two types of honey originated from three geographical areas of Yemen and sold in Dhamar markets. More investigations are needed in future to assess the quality of honeys produced in different regions of the county. The results showed that both honey types from these regions of Yemen, had good quality. The results showed that the botanical source had an effect on the pH, sucrose content and electrical conductivity (EC) values of honey, which was higher in *Sidr* honey than multi floral honey samples. The geographical area caused a variation in the moisture, free acidity and vitamin C content of honey, while

other physicochemical properties for honey had a negligible variation between honey from the different regions and botanical sources.

More investigations are needed in future to assess other quality parameters of honeys and other honey types produced in Yemen.

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