

Physicochemical characteristics and total phenols content for some kinds of Yemeni honey: A comparative study

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Abstract

Most of Yemeni honey have a locally and internationally Fame. Six of Yemeni honeys were selected according to geographical and floral sources and their fame such as (Osyimi sidir, Dawani sidir, Jardai sidir, Salam Tohamah, Somara Shabwah and SSAI Tawr Albahah). Physic- chemical properties were conducted on tri replicate, according to chemical standards methods. The results showed that all honeys were dark color; the mean results were as following: the highest value of pH was 3.71 of SSal Honey, while the least mean value was 5.3 of Sidir honeys; total acidity was on limit for all, the highest value of moisture was found on SSal, whereas Sidir honeys showed the least mean value 13.62; Hydroxylmethaylfurfural (HMF) of all studied honeys were on limit that is less than permitted value 40 mg/100g; Daistase number of all were on permitted limit except Somra honey that showed 6 shade units; values of sugars analysis were found on permitted limit except Somra was 16 g/100g; electrical conductivity of Jardani sidir honey was the highest mean value 1220, while Somra honey had the least value 544; as well as in comparison of total phenolic compound results, Jardani Sidir had the highest mean value 274.3 mg/100g of tannic acid, while Somra had the least mean value 82.01 g/100g of tannic acid. All studied honeys were in conformity to local, regional and international standards except Somra honey that has non-conformity to them.

Keywords: Six famous Yemeni honey, Physic-chemical properties, TPC, Honey standards.

Introduction

Honey from various floral sources emphasizes the importance of integrating monofloral honey as a daily consumption product due to its antioxidant activity altogether with the in vitro activities of honey [23]. Physicochemical parameters, such as pH, reducing sugar, sucrose, moisture, total solids, ash, free acid and electrical conductivity, were used to determine the quality of honey, together with sensorial, and microbiological distinctions [4]. The physico-chemical properties and antioxidant capacity of honey depend largely on the floral source of the nectar as well as seasonal and environmental conditions [1, 10, 19] Moreover, the diversity of the physical and chemical properties and quality of honey types depend on the plant sources, that also depend on several environmental factors during production such as weather and humidity inside the hive, production methods, processing and storage conditions, honey maturity, and the ability practices of the beekeeper in removing and extracting honey. [26, 6, 22, 25, 8, 5, 10, 19]. Although Yemeni

honey has fame, specially (Sidir Hadramout, Sidir Shabwa, and Soctra honey) and its high price not in the local markets, but also in International markets, and it's classified as the most expensive honey in the world [16] , unless, there was not standard characters for The Yemeni honey that define its general composition and physicochemical characteristics. Also, the research about it is generally rare.

Daniela P. et.al[10], who studied the physicochemical parameters of raspberry, mint, sunflower, thyme, rape and polyfloral honey samples from different regions in Romania were analyzed in order to examine their usefulness in the classification of honey, according to botanical origin. All honey samples have pH and free acidity values in the limits permitted by quality standards, which confirm the freshness of all honey samples, with the exception of three samples (two samples of mint honey and one sample of thyme honey) that have HMF content above the allowed limit. All the samples that were analyzed in that study met the quality requirements for honey. For most of the physicochemical parameters (color parameters, electrical conductivity, HMF content, DPPH, and (free acidity and pH) the differences between the measured levels were determined by the botanical origin of honey and the sugar composition, individual phenolic compounds and organic acids composition of honey varied to some extent between samples.

Eyobel and Miresa [14] studied the physicochemical properties and pesticide residue of honey samples collected from six Woredas of West Shewa Zone of Oromia Regional State Ethiopia and the analyzed physicochemical properties data have shown that almost all the samples of honey analyzed were within the acceptable range of Ethiopian Standard, European Standard and Codex Alimentarius, except sucrose content from Ejere and Toke Kutaye which the results of moisture, electrical conductivity, pH, free acidity, reducing sugar, sucrose, ash, and total solids were found to be 16.61–18.64%, 0.384–0.646 mS/cm, 3.77–4.22, 7.42–13.87 meq/kg, 61.38–72.87%, 6.84–15.94%, 0.030–0.095%, and 81.36–83.28%, respectively.

Liagia, et.al, [17] who studied the physico-chemical characteristics of honey from *Apis mellifera* in a comparative study; 24 samples of *Apis mellifera* honey had been collected from beehives located in the city of Jandaria. The samples were homogenized and kept in glass flasks of 250 ml, then delivered to the laboratory where they were stored in a dark place at room temperature of 20°C, the samples were subjected to physico-chemical parameters that were analyzed using the official methods of analysis of association of official analytical chemist [3], the harmonized methods of the European honey commission [7] and the codex Alimentarius [9] were considered as well. Samples were analyzed in triplicate and, during the same time in order to ensure uniform conditions and comparability, that were analyzed for moisture, HMF, free acidity, insoluble solids in water, diastase activity, ashes, electrical conductivity, sugars. However, *Apis mellifera* honey samples

demonstrated parameters in accordance with Brazilian legislation. The results of *Apis mellifera* honey samples had presented the values of electrical conductivity $284.00 \mu\text{Scm}^{-1}$, as well as showed darker color 26.67mmpfund ; the mean value of diastase activity was 42.87 with standard deviation of 2.85, the maximum value obtained was 41.45 that was higher than Goth's scale (8). Ash content was $0.18 \pm 0.01\%$. The amount of glucose was $23.50 \pm 0.73\%$, fructose was 38.78 ± 0.69 and sucrose was 5.72 ± 0.23 , moisture content presented a mean of 18.27 ± 0.40 , and its color was 26.67 ± 0.58 .

[18], there was a study conducted by **Vincent Nyan** about physical-chemical qualities of honey harvested from different beehive types that had one of his aim was investigating parameters including ash, moisture, pH, total soluble solids and soluble sugars. After collecting the samples according to certain criteria of study, there were five samples which were collected and stored in clean glass bottles and sealed, then the tightly sealed bottles containing the samples were delivered to the food chemistry and nutrition laboratory at the University of Zambia for analyzing of the previous parameters conducted according to the AOAC. The results showed that the ash content ranged from 0.198 and 0.271%, pH 4.26 and 4.44, moisture 14.9 and 16.4%, total soluble solids 83.6% and 85.7% and soluble sugars 81.6% and 83.4%. Comparisons of the findings on honey quality characteristics to the guidelines stipulated by the codex Alimentarius and European Union standards showed conformity to those standards.

Wajdi [25] who performed a study of physical and chemical properties of some Yemeni honey and collected honey samples: 3 samples for each of the following: Sidir honey, Somrah honey, Kasas honey, Sisban honey, collected directly from beekeeper located in lowder area in Abyan government in order to evaluate some physical and chemical characteristics for honey samples using standard methods for analyzing sugars, electrical conductivity, pH, moisture content and ash. The results revealed that, in Sidir honey , the mean of fructose g/100g was 36.68, Glucose g/100g was 29.00, sucrose g/100g was 0.9122, moisture g/100g was 18.7, pH was 4.08 , ash was 0.47% and the E.C. μScm^{-1} was 1122. Moreover, the results in Somrah honey samples demonstrated that the mean of fructose g/100g was 38.21, Glucose g/100g was 31.17, sucrose g/100g was 0.4178, moisture g/100g was 19.6, pH was 3.81, ash was 0.19 % and the E.C. μScm^{-1} was 692, as well as in kasas honey results showed that the mean of fructose g/100g was 37.07, Glucose g/100g was 33.25, sucrose g/100g was 0.7833, moisture g/100g was 17.0, pH was 4.06, ash was 0.17% and the E.C. μScm^{-1} was 610. Furthermore, in Sisban honey samples, the results showed that a the mean of fructose g/100g was 34.56, Glucose g/100g was 38.06, sucrose g/100g was 3.978, moisture g/100g was 19.7, pH was 3.70, ash was 0.23% and the E.C. μScm^{-1} was 756. Also, results in Maraee honey samples demonstrated that the mean of fructose g/100g was 36.37, Glucose g/100g was 34.26,

sucrose g/100g was 2.473, moisture g/100g was 19.5, pH was 3.811, ash was 0.43% and the E.C. μScm^{-1} was 791. Another study has been conducted by Wajdi about sugars and its effect by temperature and heating.

Alzoreky, N. et.al, [2] who performed a quality spectrum of Yemeni honey study, who collected 20 representative honey samples from beekeeper and local markets, ten of each *Z. Spina Christi L.* and *Acacia spp* that were evaluated for their physico-chemical characteristics or properties (moisture, total acidity, water insoluble matter (WIM), ash, HMF, Diastase Number, Reducing sugar and sucrose) in which they were analysed according to standard methods (AOAC). The standard methods of the Association of Official Analytical Chemists, [3] were used for determining of the acidity method # 927,19), moisture (method # 969,38A), ash (method # 920,181A), HMF (method # 980,23), Diastase number (method # 958,09), (reducing sugar (method # 920,183b) and apparent sucrose (method # 920,184). The properties results demonstrated the mean value of each property as following: Moisture g/100g was 16.4, WIM ml/100g was 0.39, Diastases number shade units was 6.9, HMF mg/100g was 1.5, total acidity mcq/kg was 54.1 ± 10.6 , ash was 0.33, reducing sugar g/100g was 71.6, and sucrose g/100g was 2.1.

Composition Criteria for Honey:

Honey consists essentially of different sugars, predominantly fructose and glucose as well as other substances such as organic acids, enzymes and solid particles derived from honey collection. When placed on the market as honey or used in any product intended for human consumption, honey must meet the following composition criteria [12, 13, 15, 26, 27].

Aims of the Study:

- Determination of physical properties of collected famous Yemeni honeys,
- chemical composition analysis of investigated samples, and
- assessing properties of the studied famous Yemeni honey with regional and international standards (GSO, EU, CAC).

Materials and Methods:

1. Collection and Preservation of Honey Samples

Six of different kinds of famous Yemeni honey were collected during August 2010 to April-May 2012 direct from the beekeepers. They are H1= Somar Shabwa, H2= Osymi Sidir, H3= Salam Tuhama, H4= Dawani Sidir, H5= Ssal tawer Al-Bahah, H6= Jardani Sidir. They kept in sterile dark steel containers at room temperature till analyzed. [11, 12, 20, 21].

2. Physical and Chemical Properties of the Selected Six Honey Types Studied

Chemical and physical analysis of honey samples was performed in the Chemical Analysis Lab. of Honey Products, at YSMO Yemen standardizations and metrology organization and other medical labs. These famous Yemeni honey studied were examined for Physicochemical parameters and were analysed, using The Official Methods of Analysis of Association of Official Analytical Chemists [3]. The Harmonised Methods of the European Honey Commission [22] and the Codex Alimentarius Codex Alimentarius [12]. Every sample was analyzed in three parallel determinations for each.

3. Total Phenolic Compound (TPC)

Estimation of Total Phenolic Compounds:

Total phenolic compounds of honey were determined, using spectrophotometric method (Singleton and Rossi, 1965 [24]); Tannic acid was prepared according to Galicia et al., 2008. The colorimetric assay is based on the reaction of Folin-Ciocalteu reagent (FCR) which is a method widely used for the determination of total phenols in honey. The total phenolic content was expressed in mg of tannic acid equivalents/100 g of honey.

Results:

Six samples of famous Yemeni honey have been collected from beekeepers located in different region of Yemeni appraise which were chosen according to some criteria, such as floral source, geographical region, locally and internationally fame, beside importing some of them to outside of Yemen which were (Osymi sidir, Dawani sidir, Jardai sidir, Salam Tohamah, Somara Shabwah and SSAI Tawr Albahah).

Honey Characterizations:

1. Physical Characterizations

The results showed that all honey samples were of dark color. In contrast, the highest value of moisture was found in Ssal, while Sidir samples showed the lowest that ranged from 13.2-19.2%. Water insoluble matters were ranged from 80.8-86.7 g. In addition, Ssal honey had the lowest value of pH 3.71, whereas Sidir sample was the highest 5.3. Acidity of all were at limited value of less 50 milieq/Kg. Ssal honey exhibited the highest electrical conductivity, while Jardani Sidir had the lowest as presented in Tables. (1,2,4)

Table 1. Physical and chemical properties of samples

Honey type/ property	colour			pH			Acidity			Moisture		
	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N
H1	134.5167	1.574685	3	4.716667	0.0152753	3	35.66667	0.9451646	3	15.4	0.2000003	3
H2	234.77	2.009055	3	5.023334	0.0513159	3	21.7	0.5	3	13.26667	0.1154699	3
H3	311.2867	1.708703	3	4.003333	0.1464012	3	32.2	0.2000008	3	15.2	0.1999998	3
H4	198.2833	0.6650072	3	5.663333	0.0152751	3	13.4	3.4	3	13.2	0.1999998	3
H5	251.07	1.383504	3	3.716667	0.0251661	3	23.5	0.1000004	3	19.2	0.1999998	3
H6	329.9167	0.8357847	3	5.196667	0.0416333	3	26.9	1.3	3	14.2	0.1999998	3

2. Chemical Characterizations

The activity of diastase and the total Sugars analysis for all samples was normal range except Somar honey which showed 6 units and 16 g/100g. Hydroxymethylfurfural (HMF) of all studied samples was at permitted value as presented in Table (2). Sugers, sucrose and reduced sugers were found in permitted values of standards except Somrah honey (H1) as shown in Table (3). Total phenolic compounds as the total phenolic compound was highly elevated in Jardani, but the higher reduction was recorded in Somar. Therefore, when comparison TPC content, the highest TPC content was found on Jardani Sidir that was obtained with amount of 274.3±3.491mg/100g of tannic acid and Somar Shabwa with amount 82.14±0.367 mg/100g of tannic acid showing the least honey content of TPC amongst of all studied honey. Despite their content of TPC, there was no relation between color and their TPC in which Salam Tuhamah was visually as presented in Table(4).

Table 2. Chemical properties of samples

Honey type/ property	TSS/WIS			Ash			HMF			Daistase		
	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD	N
H1	84.53333	0.2516614	3	0.3633	0.4994838	3	11.81107	0.0216907	3	6.806667	0.6361079	3
H2	86.56667	0.1154683	3	0.3592333	0.2350867	3	0.1251667	0.0077365	3	21.25	3.307189	3
H3	84.7	0.2000008	3	0.1703333	0.0477249	3	4.579	0.0528002	3	17.55	2.5015	3
H4	86.7	0.2000008	3	0.5858666	0.1263865	3	1.732333	0.0086217	3	35.4	6.672901	3
H5	80.8	0.1732068	3	1.0285	0.6636617	3	0.4906	0.0200651	3	50.95333	8.609676	3
H6	85.73334	0.2081677	3	0.4806667	0.0908397	3	0.0271667	0.0127064	3	26.66667	2.886751	3

Table 3. Chemical properties of samples

Honey type/ property	Glucose			Fructose			Sucrose		
	Mean	SD	N	Mean	SD	N	Mean	SD	N
H1	23.63	0.0099993	3	29.01333	0.0757194	3	16.61333	0.0152746	3
H2	25.27	0.0655749	3	35.74333	0.0378587	3	9.613333	0.0351187	3
H3	27.92667	0.0577342	3	34.49	0.0100002	3	6.04	0.07	3
H4	33.89	0.0100002	3	36.26	0.0100002	3	10.25	0.0500002	3
H5	30.78	0.401497	3	42.33	0.0953933	3	1.45	0.01	3
H6	25.86333	0.0404146	3	46.24667	0.1006647	3	10.12333	0.0208166	3

Table 4. Follow chemical properties of samples

Honey type/ property	G+F			E.C			TPC		
	Mean	SD	N	Mean	SD	N	Mean	SD	N
H1	52.64333	0.0750562	3	545	1	3	82.13667	0.3667898	3
H2	61.01333	0.1001669	3	1097.667	1.527525	3	125.2867	0.7179354	3
H3	62.41667	0.0493284	3	1097.667	1.527525	3	150.8677	0.296799	3
H4	70.15	0.0173198	3	1028	1	3	141.512	0.8902743	3
H5	73.11	0.4850774	3	1181.667	1.527525	3	164.4047	1.430594	3
H6	72.11	0.1410667	3	498	2	3	274.3217	3.491297	3

4. Discussion:

The above results have many similarities and non-similarity with many pervious researches for instance: there are Yemeni studies in 2007 done by Wajdi, M. [25] in which he studied Sidir, Somrah and sisaban honeys that was found to be non-similar in current with selected honeys because there were no geographical regions are mentioned in his study. However, it has the same method of collecting samples in comparison to results that were approximately agreed in limits with the same exception such as mean results of pH of this study that showed around 5 that is higher of 4 because of some previous mentioned factors and inconsistent content of moisture that is 13 for each of three Sidir honeys in a current study which is highly lower than 18.7g/100g and both were in limit, this might be because of the water content of botanical resources where exposed highly number in 2007 which is higher than that in 2010 of the collected date beside other factors as well as Somrah that showed moisture and pH 15.6g/100g, 4.7 in 2012 which is lower than 19.0 g/100g and pH higher than 3.8, this might be because of the same last reasons and shortage of water in that region that is inconsistently affected on water in respectively for each things. However, it is not a good point of Yemen; beside other factors regarding to differences of samples. wgen comparing TPC content, the highest TPC content was found on Jardani Sidir that has been obtained with an amount of 274.3 mg/100g of tannic acid, followed by Ssal Tawer Albahah with an amount of 164.4 mg/100g of tannic acid, then Salam Tuhamah with an amount of 150.87 mg/100g

of tannic acid, as well as Dawani Sidir with an amount of 141.5 mg/100g of tannic acid; Somar Shabwah 82.137mg/100g as well that showed the least honey content of TPC amongst of all studied honey. Despite their content of TPC, there was no relation between color and their TPC in which Salam Tuhamah was visually the darkest one; however it hadn't the highest TPC content. As we notice, the above results had various results from one honey to another depending on honey types; floral sources, geographical region and season of harvest play that an important role in varied results, as well as the similarity of some properties of Sidir honey types such as moisture, pH, TSS, HMF, Diastase activity, some sugars content and various TPC and E. C that might be because of their similarity of floral sources, harvesting on some seasons of different region of Yemen add to that species of bee (*Apis mellifera yementica*). Furthermore, various results of TPC among them rely on their content of chemical composition and physicochemical content of floral sources that might be different from place to another; climate of region from year to year and from place to another as well. Moreover, the results showed that no correlation because of the change of honeys, content from season to another, year to year that might be because of the phytochemical composition and other factors, as illustrated above. We can say that Sidir of this year is better than Somar and vice versa, according to internal and external factors which play almost role in these changes. The above results have many similarity and non-similarity with many previous researches for, instance, Daniela P. et.al, [10] found consistent to current study in the criteria limits of Moisture, pH and free acidity as well as it found inconsistent to other criteria HMF that the current study showed values in below which they are all in range criteria of standards, also, in E.C and TPC which found the current study in higher than this study of Eyobel M. and Miresa T. [14] in the following parameters Ash, moisture, electrical conductivity, total soluble solid, free acidity with similarity in H2, H4, H6 which was found 5.0, 5.6 and 5.1 respectively. While, it found similar to sucrose and reducing suger except H1 that was 52.64 % which emphasize that there are many external and internal factors affected these similarity and non similarity such as floral resources and environmental varieties around the world. Ligia et al, [17] who found consistent to current study in its physic-chemical characteristics with used materials and methods according to AOAC, 1990. On other hand it is found inconsistent to the whole idea of this study. In comparison of results, they were all in agreement of permitted limit of an international standards and approximately regarding similar results in some tests to the best is Yemeni honeys properties. In Zambia as well in 2013, the idea of study physicochemical characteristics is also agreed to Vincent Nyan study with differences of ways of gathering honeys in which results found inconsistent where current findings of study showed pH reach around 5 of Sidir honeys that is higher than Zambia's study findings which was

4.26-4.44 that might be because differences of honeys types, geographical and climates as well as all found to be consistent to codex and European standard [18].

Conclusion:

This research concluded that all the studied samples of honey have the conformity of local, regional and international standards according to standard criteria, except Somra honey. Moreover, the concentration of different compounds in honey relies mainly on various factors, such as honey type and floral source, and environmental and processing factors. These factors affect the biological activities of each type of honey in the world. Briefly, honeys present a variety of dietary phytochemical compounds with functional properties such as phenolic acids, minerals, phenolics and flavonoids.

Recommendation

- More awareness workshop about Yemeni honey standard locally, regionally and internationally should be held
- Surveillance of honey marketing in Yemen.
- Establishment of government honey research center with all modern equipment system in order to facilitate doing scientific honey research as especially GC-MS/MS and LC-MS/MS.
- Performing more studies of different Yemeni honey in various fields as well as identifying their antimicrobial agents and their chemical compositions, using modern techniques.

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الخصائص الفيزيائية والكيميائية والفينولات الكلية لبعض أنواع العسل اليمني:

دراسة مقارنة

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الملخص

أغلب أنواع العسل اليمني تمتلك شهرة محلية ودولية. تم اختيار ستة منها وفقا للموقع الجغرافي والمصدر الزهري لدراسة الصفات الفيزيائية والكيميائية وتقدير الفينولات الكلية لثلاث مكررات لكل منها. حيث تم اختيار (عسل سدر عصيمي، سدر دوعني، سدر جرداني، سمر شبو، سلام تهامه، صال طور الباحة). تم فحصها وفقا للاختبارات القياسية المعتمدة. وكانت النتائج كالتالي: جميع الأعسال تعتبر ذو لون داكن وأعلى قيمة pH 3,71 لعسل الصال، بينما أقل قيمة كانت في أعسال السدر الثلاثة pH 5.3، أما الحموضة الكلية فكانت في الحدود، وأقل رطوبة ظهرت في أعسال السدر الثلاثة 13جم\100جم، وأعلىها في عسل الصال 19جم\100جم، وأما بالنسبة لتقدير نسبة الهيدروكسيل ميثيل فارفورال فجميع أنواع العسل المدروسة كانت في الحدود المسموح بها، في حين أظهرت نتائج رقم الدياستيز لجميع الأعسال أنها في الحدود المسموح بها، ماعدا عسل السمر أظهر 6وحدات شاد، كما أظهرت نتائج تحليل السكريات لجميع الأنواع أنها في الحدود المسموح بها ماعدا عسل السمر الذي أظهر ارتفاع في نسبة السكروز 16جم\100جم. وأما أعلى قيمة للتوصيلة الكهربائية فقد ظهرت في عسل السدر الجرداني وأقلها في عسل السمر بقيمة 544, 1220 μcm^{-1} على التوالي، وبمقارنة نتائج الفينولات الكلية فإن عسل السدر الجرداني احتوى على أعلى قيمة 274مجم\100جم حمض التانيك، وأقلها في عسل السمر 82مجم\100جم حمض التانيك. جميع أنواع العسل المدروسة كانت مطابقة للمواصفات المحلية والإقليمية والدولية ماعدا عسل السمر الذي أظهر بعض الصفات الغير مطابقة كرقم الدياستيز ونسبة السكروز.

الكلمات المفتاحية: العسل اليمني، خصائص العسل، الفينولات الكلية، مواصفات العسل.