

ESTIMATION OF SOME QUALITY PARAMETERS OF HONEY

Aida Albu^{1*}, Gabriela Frunză¹, Roxana Zaharia¹, I.M. Pop¹

¹Faculty of Animal Sciences, University of Agricultural Sciences and Veterinary Medicine „Ion Ionescu de la Brad” Iași, Romania

Abstract

Twenty-four samples of honey, from eight Romanian producers from three types (acacia, linden and polyflora-for each producer) were physico-chemical analyzed. The results obtained were variable: 0.2-72.1 mm Pfund for color, 0.038-0.050% for water insoluble matter, 1.4876-1.4980 for refractive index, 15.48-19.52% for moisture, 80.48-84.52% for solids substances, 79.00-83.00 °Brix for total soluble substances and 1.420-1.448 g/cm³ for specific gravity. The values for pH and free acidity were in the range of 3.77-5.32 pH units, respectively 6.13-26.63 meq kg⁻¹; the ash content varied between 0.027% and 0.344% and the electrical conductivity recorded values between 129 μS cm⁻¹ and 775 μS cm⁻¹. The analysis were performed in accordance with Romanian/EU standards. For all honey samples, the Pearson coefficient results indicated strong positive correlation between specific gravity and total soluble substances, between ash and electrical conductivity, respectively a strong negative correlation between moisture and refractive index, between moisture and total soluble substances and between moisture and specific gravity. As a conclusion, the studied honey bee samples correspond qualitatively to the legislation requirements.

Key words: honey, acacia, linden, polyfloral, quality

INTRODUCTION

Honey produced by bees from floral nectar is a natural sweet substance [6]. Romania have various relief forms and a rich melliferous flora, this advantage, in European Union, give it an important place as producer of many types of bee honey: monofloral, polyfloral and honeydrew [19].

Honey is composed of sugar, water and other minor constituents like: proteins, enzymes, aminoacids, organic acids, phenolic compounds, vitamins and minerals. The composition of this natural food, its nutritional and therapeutical properties depends on the geographical origin, floral source and other external factors like: season, environmental components, honey processing, storage condition and time [2], [14].

The aim of this study was to estimate some quality parameters of commercial honey from different producers: color, water insoluble matter, refractive index, moisture, solids substances, total soluble substances,

specific gravity, pH, free acidity, electrical conductivity and their correlation.

MATERIAL AND METHODS

Twenty four samples of commercial honey (acacia-A, linden-L and polyfloral-L), purchased from Iasi market, from eight producers were analyzed. Samples were kept in original jars, in the laboratory at room temperature. Qualitative parameters were evaluated according to the standards or to literature.

For the determination of Pfund value, 50% honey aqueous solutions were read spectrophotometrically at wave length $\lambda=635$ nm (Shimadzu UV-mini-1240) and conversion the absorbances in mm Pfund. Color of honey samples was established according to the Pfund scale [7], [23]. Water insoluble solids (WIS) were determined according to the Harmonized Methods of the International Honey Commission [3]. Refractive indices (RI) of the honey samples, were read on the ABBÉ Kruss AR 2008 refractometer. The temperature of the honey samples was checked using Heidolph EKT Hei-Con temperature sensor and corrected

*Corresponding author: aida.albu@gmail.com

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refractive index with ± 0.00023 for every 1°C . The values of the moisture content (M) were taken from the table of relationship of water content of honey to refractive index [3], [24], [28]. Moisture content is expressed as a percentage. The content of solids substances (SS) was found by the difference between 100 and moisture and expressed as a percentage. For determination of total soluble solids (TSS) content were used tables of correlating refractive index with total soluble solids content (expressed as Sucrose) and expressed in degrees Brix [16].

The specific gravity (SG) of honey was determined by dividing the pycnometer weight filled with honey by the pycnometer weight filled with water. Specific gravity values are expressed in g/cm^3 [20]. pH was determined on an aqueous honey solution using a specific electrode of WTW apparatus MULTI 3320 [3], [25]. A 10% (w/v) honey solution, with 2-3 drops of phenolphthalein, was titrated with 0.1 N NaOH. Free acidity (FA) was measured in meq kg^{-1} [20], [28]. Ash content was determined by calcination of

samples in a muffle furnace (SUPER THERM) at 550°C [4], [20], [28].

A honey solution 20% (in dry matter honey) was prepared with ultrapure water and with the electrode specific of WTW apparatus MULTI 3320 was determined electrical conductivity (EC) expressed in $\mu\text{S cm}^{-1}$ [3], [20], [25].

All analyses were made in triplicate. For descriptive statistics it used one-way ANOVA ($p=0.05$), correlations between studied parameters were obtained by Pearson's correlation coefficient (r) and for hierarchical cluster analysis (HCA) it was used SPSS Statistical version 14.0.

RESULTS AND DISCUSSIONS

The results from physicochemical analysis for the twenty four honey samples are presented in Table 1 and Table 2. All acacia honey samples have water-white color. Color is the first physical property observed by consumers and is directly influenced by the botanical origin, climate and soil [5]. The color of all acacia samples presented water-white color.

Table 1 Descriptive statistics of mm Pfund, water insoluble matter (WIS), refractive index (RI), moisture (M) and solids substances (SS) of honey samples

Type	Descriptive statistics	mm Pfund	WIS %	RI	Moisture %	SS %
A	Min-Max	0.2-4.7	0.035-0.048	1.4900-1.4980	15.48-18.57	81.43-84.52
	Mean \pm SD	1.5 \pm 1.64	0.042 \pm 0.00	1.4934 \pm 0.00	17.23 \pm 1.08	82.77 \pm 1.08
	CV	106.42	11.56	0.19	6.28	1.31
L	Min-Max	12.3-53.2	0.035-0.049	1.4876-1.4954	16.50-19.52	80.48-83.50
	Mean \pm SD	36.2 \pm 12.96	0.042 \pm 0.01	1.4926 \pm 0.00	17.56 \pm 0.96	82.44 \pm 0.96
	CV	35.79	14.24	0.17	5.47	1.17
P	Min-Max	21.0-72.1	0.038-0.050	1.4900-1.4964	16.08-18.56	81.44-83.92
	Mean \pm SD	44.5 \pm 19.15	0.043 \pm 0.00	1.4933 \pm 0.00	17.29 \pm 0.72	82.72 \pm 0.73
	CV	43.05	10.40	0.13	4.19	0.88

A-acacia, L-linden, P-polyfloral

The highest of mm Pfund value was at A3 (0.2 mm Pfund) sample and the lowest value at A4 (4.7 mm Pfund) sample, values with statistically significant difference for $p < 0.05$. Different color has linden honey samples from extra white at L5 (12.3 mm Pfund) to light amber at L2 (53,2 mm Pfund), with values statistically significant difference ($p < 0.05$).

The highest color difference was observed at polyfloral honey samples from 21 mm Pfund at P8 to 72,1 mm Pfund at P6 statistically with significant difference results ($p < 0.05$). Fig. 1 shows significant differences of color between honey samples of the same type and between all the three types of studied honey. Water-insoluble

solids (WIS) like: wax caps, particles of honeycombs, fragments of the body of bees, bee larvae, plant wood, soil, affect the quality of the honey [20]. The lowest WIS value of 0.035% was found at one acacia sample (A1) and at one linden sample (L2). The highest value was obtained at one polyfloral sample (P8) of 0.050%. Statistics there are no difference ($p < 0.05$) between the three types of studied honey. All values are under the maximum acceptable content of water-insoluble solids at marketed bee honey, having a value of 0.1% [6], [27].

The lowest and highest average value of refractive index was found at linden honey of 1.4926 and of 1.4934 at acacia honey, respectively. There are no statistically differences between honey types for $p < 0.05$.

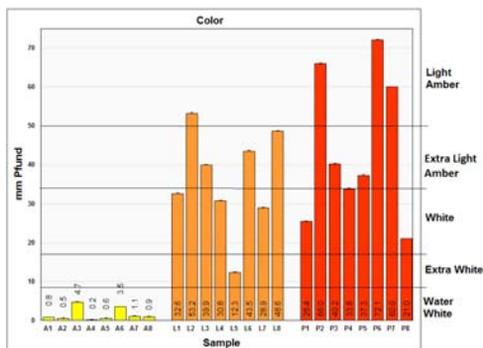


Fig. 1 Average values of mm Pfund of honey samples

The total soluble solids (TSS), of the honey samples analyzed, had ranged from 79 °Brix to 83 °Brix (Table 2) and the values has no statistically difference for $p < 0.05$. Specific gravity (SG) is related to water content, temperature and the solid substances content; this parameter has a practical significance to calculate the weight of honey [20]. The values of SG ranging between 1.420 g/cm³ and 1.448 g/cm³ with no statistically difference for $p < 0.05$. In general, the pH values of bee honey varies between 3.5 and 5.5. There is no pH reference value, but, as much as pH value is lower, the more the growth of microorganisms is inhibited and gives stability and duration product preservation [17], [20]. The pH values of

The moisture content is the second largest constituent of honey and an important parameter because low value give honey stability against fermentation and increase the storage time [24]. The Council of the European Union and Romanian standard, established maximum values of moisture of 20% [6], [27]. All values of moisture content of honey samples are less than 20%, the maximum value recorded was of 19.52% at one linden sample-L8. Results of one-way ANOVA with $p < 0.05$ confidence level were considered statistically no significant between the types of honey.

In studied literature, the moisture content has values from 3.9% [21] to 22.8% [29] at Romanian acacia honey (Table 3).

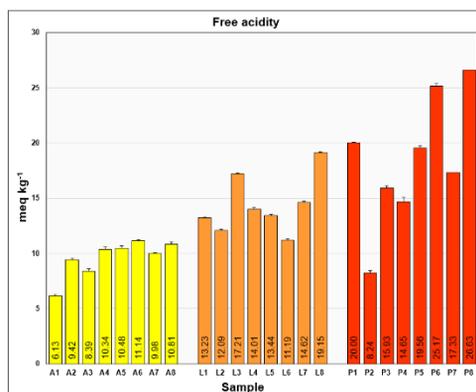


Fig. 2 Average values of free acidity of honey samples

samples are within the range 3.77-5.32 pH units. The results of one-way ANOVA can be considered with significant difference between linden and polyfloral honey and between acacia and linden honey. Between acacia and polyfloral honey type there are no statistically difference for $p < 0.05$. Many research registered similar values (Table 3). The lowest average value of free acidity (FA) was found on acacia honey samples of 9.59 meq kg⁻¹, followed by linden honey samples of 14.37 meq kg⁻¹ and polyfloral honey samples of 18.44 meq kg⁻¹. Free acidity value is a measure of freshness honey, high values of this parameter indicates the presence of fermentation process. The highest value of free acidity was found at polyfloral honey

sample (P8) of 26.63 meq kg⁻¹ (Fig. 2), value under the 50 milliequivalents acid per 1000 g, maximum allowed value request in

legislation [6]. Differences between free acidity values, of all types of studied honey, are statistically significant.

Table 2 Descriptive statistics of total soluble substances (TSS), specific gravity (SG), pH, free acidity (FA), ash and electrical conductivity (EC)

Type	Descriptive statistics	TSS (°Brix)	Specific gravity (g/cm ³)	pH	Free Acidity (meq kg ⁻¹)	Ash%	EC (µS cm ⁻¹)
A	Min-Max	79.95-83.00	1.427-1.448	4.15-4.52	6.13-11.14	0.027-0.074	129-219
	Mean±SD	81.26±1.07	1.436±0.01	4.31±0.11	9.59±1.64	0.049±0.02	157±28.09
	CV	1.32	0.51	2.57	17.11	35.60	17.92
L	Min-Max	79.00-81.99	1.420-1.441	4.29-5.32	11.19-19.15	0.149-0.344	365-775
	Mean±SD	80.93±0.95	1.433±0.01	4.78±0.33	14.37±2.63	0.228±0.06	551±125.21
	CV	1.17	0.46	6.84	18.34	25.99	22.72
P	Min-Max	79.96-82.38	1.427-1.444	3.77-4.69	8.24-26.63	0.137-0.228	241-511
	Mean±SD	81.20±0.71	1.435±0.00	4.22±0.32	18.44±5.88	0.183±0.03	349±86.82
	CV	0.88	0.35	7.57	31.89	14.46	24.91

A-acacia, L-linden, P-polyfloral

The values of ash content were between 0.027% at A3-acacia sample and at L5-linden sample of 0.344%. The highest average ash content was at linden honey (L5) of 0.344% and the lowest value was at acacia honey (A3) of 0.027%. Five linden samples had the highest average values of ash content (L3, L4, L5, L6 and L7), over 0.236% (Fig. 3). Between the three types of honey samples, the

values of the ash content showed statistically significant differences. Similar results of ash content were found by Purcarea et al, 2016, of 0.186% at romanian linden honey, and by Aazza et al., 2013, of 0.17% to 0.33% at polyfloral honey from Portugal. Average values of electrical conductivity ranged from 129 µS cm⁻¹ at acacia honey sample (A2) to 775 µS cm⁻¹ at linden honey (L5).

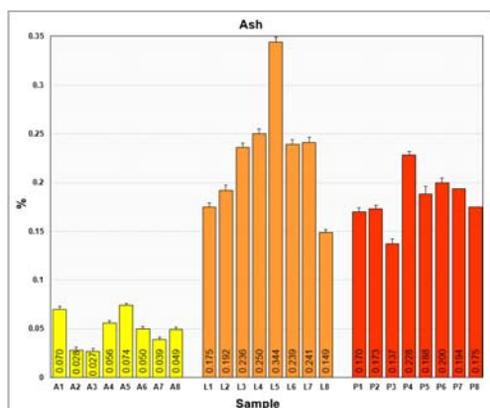


Fig. 3 Average values of ash of honey samples

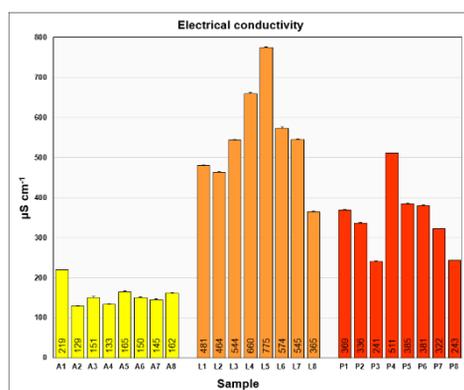


Fig. 4 Average values of electrical conductivity of honey samples

Five linden samples (L3, L4, L5, L6 and L7), recorded highest values of electrical conductivity, over 544 µS cm⁻¹ but under of 800 µS cm⁻¹ (Fig. 4). Electrical conductivity is an indicator of the differentiation of bee

honey from nectar and honeydew. Directive 2001/110/EC of the European Commission, as amended in 2007, provides for a maximum permitted value for honey of nectar of 800 µS cm⁻¹.

Table 3 Some parameters of acacia, linden, and polyfloral honey in literature

Country	Moisture (%)	pH	Free acidity (meq kg ⁻¹)	EC (mS cm ⁻¹)	References
Acacia					
Serbia	13.90-20.57	3.49-5.85	7.8-29.6	0.1-0.68	[12]
România	3.9-22.8	3.65-4.63	1.84-10.87	0.097-0.35	[15],[21], [26],[29]
Poland	17.73	3.79	25.6	0.42	[30]
Slovakia	17.86	3.71	16.1	0.20	[30]
Linden					
Polonia	19.5	3.81-4.13	14.5-34.2	0.53-0.579	[9],[30]
Serbia	13,41-22,48	3,98-5,40	8,2-26,2	0,3-0,76	[12]
România	5.4-18.8	3.6-4.7	-	0.20-0.73	[21], [29]
Slovakia	18.35	3.90	21.6	0.23	[30]
Polyfloral					
Italy	16.3-17.3	3.85-4.51	9-29	0.15-0.78	[8]
Poland	18,4-18.65	3.57-4.73	4.5-37.0	0.35-0.371	[9], [30]
Algeria	11,59-14,13	3,7-4	-	0,417 -0,806	[11]
România	4.8-19.6	3.2-4.6	23.9	0.232-0.831	[18], [21], [26]
Slovakia	18.53	3.68	23.3	0.21	[30]

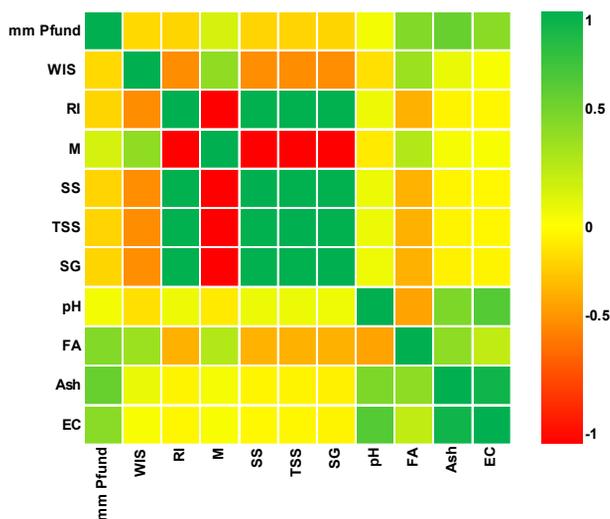


Fig. 5 Heat map of Pearson correlation between studied honey parameters(WIS- water insoluble matter, RI- refractive index, M- moisture, SS- solids substances, TSS-total soluble substances, SG-specific gravity, EC- electrical conductivity)

Correlation Pearson between the investigated parameters of twenty four honey samples is shown in Fig. 5. The results of Pearson coefficient shows strong positive linear correlations between ash content and electrical conductivity ($r=0.94$). A strong positive linear correlation ($r=1$) was found between refractive index and solids substances, between refractive index and total soluble substances and between refractive index and specific gravity. Pearson

Correlation value of 0.67 indicate a medium positive correlation like between mm Pfund and ash ($r=0.60$) and pH and ash ($r=0.51$). Between variables: moisture and refractive index, moisture and solids substances, moisture and total soluble substances and moisture and specific gravity, Pearson coefficient ($r=-1$) show strong negative correlation. Low positive correlation were obtained by Pearson's correlation between free acidity and electrical conductivity

($r=0.24$) and low negative correlation between pH and free acidity with an $r=0.36$ coefficient). The same correlations were also reported by Khalafi et al., 2016 and by

Majewska et al., 2019. Cluster analysis was made to find similarities between studied parameters.

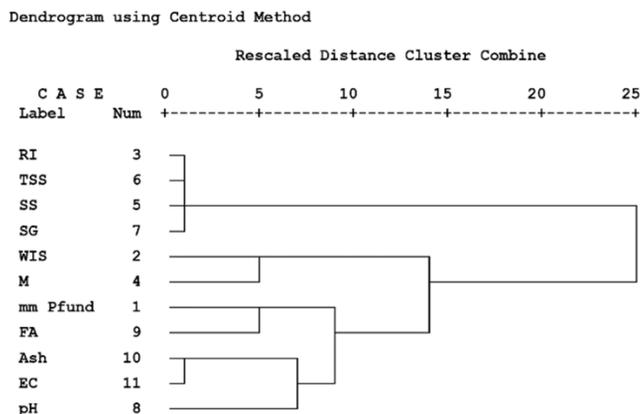


Fig. 6 Hierarchical cluster analysis(HCA) based studied honey parameters

In the obtained dendrogram (Fig. 6) are distinguished clusters of parameters: the first one included refractive index, total soluble substances, solids substances and specific gravity, the second cluster included water-insoluble solids and moisture, the third cluster is made from mm Pfund and free acidity; ash and electrical conductivity forms another cluster and last in pH. The distance cluster represent how different they are, for example the first cluster compared to the first or the second cluster.

CONCLUSIONS

The results obtained in this study skown that the same type of honey can have different quality parameters, these being influenced by different factors (environment elements, time and condition of storage etc.). It is necessary to know the area where the honey was harvested, because this product is an quality environmental bioindicator. The Pearson coefficient values show many correlations between physicochemical properties. All values of results were in the limits permitted by the legislation, which confirm the quality of all honey samples.

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