Usage of physical methods in the characterization of the quality of honey
Fizikālo metožu pielietošana medus kvalitātes raksturošanā

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Abstract. Physical and chemical parameters of 14 samples of honey from different regions of Latvia were analyzed by methods of potentiometry, conductometry, refractometry, photometry, turbidimetry and gravimetry. The obtained results proved that investigated samples of honey were of high quality, natural and mature. Correlation between different parameters of honey was investigated. The results were analyzed by statistical methods (regression and correlation). Certain coherence between the ash content, pH and conductivity of honey was observed.
Key words: honey, content, quality, conductivity, pH.

Introduction

Quality control of food products has become a top issue lately. Honey is practically a concentrated aqueous solution of invert sugar, and it also contains a very complex mixture of other carbohydrates, several enzymes, amino and organic acids, minerals, aroma substances, pigments, waxes, pollen grains, etc. Honey is one of the most widely used food products. The quality of honey may be characterized by various chemical and physical parameters. In this research, the pH, conductivity, absorbency and index of refraction of honey were measured. The content of ash, moisture, reducing sugars, lysozyme and activity of amylases were determined. The obtained data characterize the quality of honey, its nature, maturity, conditions of preservation and treatment. Honey is one of the most complicated natural products, it consists of more than four hundred different components. Its chemical content can vary due to variety of nectar plants, geographical and weather conditions, time of centrifugal extraction and storage. Nevertheless, there are basic groups of substances, which always are present in honey – carbohydrates, ferments, acids and mineral substances (Table 1).

Table 1

Composition of honey, % (Belitz, Grosch, 1987)

Constituents	Average value	Variation range
Moisture (water)	17.2	13.4-22.9
Fructose	38.2	27.3-44.3
Glucose	31.3	22.0-40.8
Saccharose	1.3	0.3-7.6
Maltose	7.3	2.7-16.0
Higher sugars	1.5	0.1-8.5
Other sugars	3.1	0-13.2
Nitrogen	0.04	0-0.13
Minerals (ash)	0.17	0.02-1.03
Free acids*	22.0	6.8-47.2
Lactones*	7.1	0-18.8
Total acids*	29.1	8.7-59.5
pH	3.9	3.4-6.1
Diastase – value	20.8	2.1-61.2

*mequivalents·kg^{-1}

One of the main characteristics of the quality of honey is its water content (moisture). It depends on the grade of honey maturity. In immature honey, water content is higher than in mature honey (for mature honey – 18-20%, for immature – >20%) (Билаш, Бурмистров, Гребцова, 1991). The water content in honey should be less than 20%. Honey having a higher water content is readily susceptible to fermentation by osmophilic yeasts. Yeast fermentation is negligible if the water content is less than 17.1%, while between 17.1 and 20%, fermentation depends on the count of osmophilic yeast buds (Belitz, Grosch, 1987).

The quantity of reducing sugars (fructose, glucose, maltose) in honey is one of parameters characterizing the quality of honey. The content of reducing sugars usually varies between 76-86% (Буренин, Котова, 1994).

Nowadays, amylases (diastase) are the main indicator characterizing the naturalness of honey. Activity of amylases decreases in the result of heat treatment as well as during storage.

Presence of lysozyme determines honey's curing power and efficiency in prevention of diseases. The activity of lysozyme depends on the age of honey.

The amount of mineral substances in honey is very low but of great variety, including approximately 40 macro-, micro- and ultramicroelements (Table 2).

In this research, we analyzed honey samples from different regions of Latvia and investigated mutual coherence between various parameters of honey.

Table 2

Content of prevalent elements in honey
(Билаш, Бурмистров, Гребцова et al., 1991; Скурихин, Волгарев, 1987)

Macro-elements	Content, mg·kg^{-1}		Micro-elements	Content, mg·kg^{-1}	
K	832	360	Fe	9.7	8.0
P	217	180	Mn	4.2	0.34
Ca	190	140	Cu	0.8	0.59
Cl	80	190	Co	0.15	0.003
S	80	10	Zn		0.94
Na	50	100	J		0.02
Mg	50	30	F		1.0

Table 3

The content of moisture, reducing sugars and ash,
the pH and conductivity of honey (average values)

Sample of honey	Region	pH (20°C)	Conductivity*, μS·cm^{-1} (20°C)	Moisture, %	Ash, %	Reducing sugars, %
Various flowers	Cesis	3.66	85.7	17.20	0.094	78.11
Various flowers	Saldus	4.08	122.0	14.40	0.158	77.90
Various flowers	Riga	3.95	144.4	18.40	0.212	75.81
Various flowers	Daugavpils	4.24	127.2	18.85	0.181	80.60
Various flowers	Jekabpils	3.91	94.3	18.09	0.084	80.60
Lime – blossom	Saldus	4.13	144.4	17.20	0.219	82.69
Lime – blossom	Dobele	3.87	78.7	16.98	0.070	75.70
Lime – blossom	Saldus	3.87	109.7	16.80	0.120	78.32
Heather – blossom	Saldus	4.48	185.5	20.49	0.330	76.30
Heather – blossom	Saldus	4.42	188.6	15.20	0.201	77.20
Spring flowers	Rezekne	4.26	107.9	16.00	0.150	80.40
Spring flowers	Aluksne	4.44	122.1	16.09	0.173	75.30
Selected	Cesis	3.85	104.9	16.09	0.069	81.90
Wild flowers	Saldus	4.15	109.8	15.60	0.144	81.55

* For samples of honey with the concentration of 30 g·l^{-1}.

Materials and methods

14 samples of honey from different regions of Latvia were investigated.

The following parameters were determined:

- pH of honey – by method of potentiometry;
- the conductivity of honey solutions – by method of conductometry;
- the content of water in honey – by method of refractometry (Буренин, Котова, 1994);
- the content of reducing sugars – by method of photometry (Аганин, 1985);
- the content (activity) of enzyme amylases – by method of photometry (Matissek, Schnepel, Steiner, 1992);
- the content (activity) of enzyme lysozyme – by method of turbidimetry (Грант, Яворковский, Блумберга, 1972);
- the ash content – by method of gravimetry (James, 1996).

The equipment used:

- pH meter pH 538 (WTW) with electrode Sen Tix Sp;
- conductivity meter LF 92 (WTW) with conductivity cell Tetra Con 96;
- refractometer ИРФ-22;
- spectrophotometer СФ-46;
- analytical balance BP 110 S-DCE (Sartorius);
- heating oven Memmert 400;
- muffle furnace.

Conductivity (Table 3) was measured using honey solutions with the concentration of honey – 30 g·l^{-1}. The use of this concentration was based on the change of conductivity depending on the concentration of solutions. As seen in Fig. 1, with the increase of the concentration of honey solutions, the increase of conductivity diminishes. With the concentration of honey solutions still growing up, the conductivity starts to decrease. Conductivity measurements in the sample of honey showed that there it is very insignificant. Thus, the chosen concentration of honey solutions is optimal for all the honey samples.

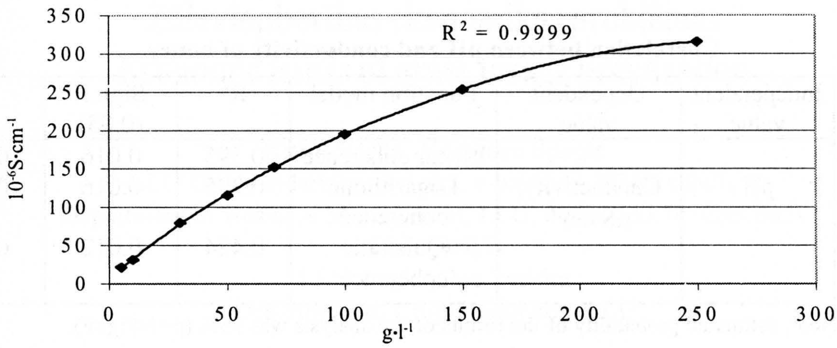

Fig. 1. Change of conductivity depending on the concentration of honey solutions.

Determination of reducing sugars in honey was based on the absorbency after the reaction of reducing sugars with potassium ferricyanide:

$$\underset{\text{glucose}}{C_5H_6(OH)_5CHO} + 2[Fe(CN)_6]^{3-} + H_2O \rightarrow$$
$$\rightarrow \underset{\text{gluconic acid}}{C_5H_6(OH)_5COOH} + 2[Fe(CN)_6]^{4-} + 2H^+. \quad (1)$$

The higher the content of reducing sugars is in honey, the lower is the optical density of honey solution (λ=440 nm).

Determination of amylase activity was based on its capacity to hydrolyse starch (λ=565 nm):

$$\underset{\text{starch}}{(C_6H_{10}O_5)n} + H_2O + \text{amylases} \rightarrow \underset{\substack{\text{dextrines}\\ n>x}}{(C_6H_{10}O_5)x} + H_2O +$$
$$+ \text{amylases} \rightarrow \underset{\text{maltose}}{C_{12}H_{22}O_{11}}. \quad (2)$$

The basis for turbidimetric determination of lysozyme was the ability of lysozyme to split cell tegument of *Micrococcus lysodeikticus*. In the result, the optical density of *Micrococcus lysodeikticus* suspension diminishes (λ=540 nm).

The following formula was used for refractometric determination of water content:

$$\omega\%_{H_2O} = (1.538 - n_D^{20}) \cdot 400, \quad (3)$$

where $n_D^{20} = n_D^t + 0.00023(t - 20)$. (4)

Results

The results of honey analysis are shown in Table 3. The content (activity) of enzyme amylases was 19.46-35.70 g starch degraded by 100 g honey at 40 °C. The investigated samples of honey had high activity of amylases.

The content (activity) of lysozyme varied between 0.5-2.0 μg·g⁻¹.

The average values of different indices correspond with the data in the literature (Belitz, Grosch, 1987).

Discussion

The SPSS 8.0 program was used to analyze the obtained results and to establish correlation and regression. With the help of this program coherence between the determined values was analyzed, using several models of functions (Tables 4-6).

Table 4

Correlation between ash content and conductivity of honey

Independent value	Dependent value	Function model	R^2	Sign F (0.05)	r_{xy}
Content of ash, %	Conductivity, μS·cm⁻¹	Linear coherence	0.776	0.0000	0.881
		Logarithmic coherence	0.787	0.0000	0.887
		Quadratic coherence	0.793	0.0002	0.891

Table 5

Correlation between ash content and pH of honey

Independent value	Dependent value	Function model	R^2	Sign F (0.05)	r_{xy}
Content of ash, %	pH	Linear coherence	0.528	0.0032	0.726
		Logarithmic coherence	0.545	0.0025	0.738
		Quadratic coherence	0.541	0.0137	0.736

Table 6

Correlation between pH and conductivity of honey

Independent value	Dependent value	Function model	R^2	Sign F (0.05)	r_{xy}
pH	Conductivity, $\mu S \cdot cm^{-1}$	Linear coherence	0.395	0.016	0.628
		Logarithmic coherence	0.395	0.016	0.628
		Quadratic coherence	0.424	0.012	0.628

Note: estimated probability of the results of the analysis was 95% (p=1-SignF).

The results of the analysis (Tables 4, 5) show that, regardless of the low determination quotient R^2, the assessment of probability as well as the correlation quotient indicate to a certain correlation between conductivity and pH and ash content in honey. It is clearly demonstrated in linear and logarithmic models of functions. This can be easily explained, as mineral substances constitute the main part of the ash content.

Analysis of correlation between conductivity and pH (Table 6) suggests that in this case conductivity and pH of honey are interdependent even though the probability is not very high, as seen from the level of significance and correlation quotients.

Conclusions

The obtained results show that the investigated samples of honey were of high quality, they were natural and mature.

Certain coherence between the ash content, pH and conductivity of honey was determined.

All determined parameters correspond to the standards of the Republic of Latvia and the EU.

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Anotācija

Darbā analizēti no dažādiem Latvijas rajoniem iegūtu 14 medus paraugu fizikāli-ķīmiskie parametri. Medus paraugi analizēti ar potenciometrijas, konduktometrijas, refraktometrijas, spektrofotometrijas, turbidimetrijas un gravimetrijas metodēm. Iegūtie medus paraugu analīžu rezultāti liecina par medus augsto kvalitāti un dabīgumu. Pētīta medus parametru savstarpējā sakarība. Rezultātu izvērtēšanai izmantotas regresijas un korelācijas analīzes metodes. Novērota sakarība starp medus pelnu saturu, pH un elektrovadītspēju.