







Article

Physico-Chemical and Melissopalynological Characterization of Czech Honey

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Abstract: Geographical and botanical origin of honeys can be characterized on the basis of physico-chemical composition, sensory properties and on the basis of melissopalynological analysis. No comprehensive description of the characteristics of Czech honey has been published so far. This study provides insights that are important for correct classification. The study analysed 317 samples of authentic honey from randomly selected localities. Due to the diversity of the landscape, the typical honey of the region is blend honey with a predominance of blossom honey. According to the pollen profile and electric conductivity, the honeys were sorted into the following: Brassica honey (BH), Floral honey (FH), Fruit tree honey (PH), Honeydew (HD), Lime tree honey (LH), Robinia pseudoacacia honey (RH), and Trifolium honey (TH). Physico-chemical properties, including higher carbohydrates, were determined for the honeys and their pollen profiles were examined. The physico-chemical properties and pollen profile are partially in compliance with the description of European monofloral honeys, except for RH and TH. Although they had the highest proportion of acacia pollen, amounting to >10% of all the Czech honeys, these RH honeys differ from the European standard, so they cannot be considered acacia honey. Further, PH honeys and FH polyfloral honeys were described. Most honeys contained a significant proportion of rapeseed pollen, which is one of the common agricultural crops grown in the Czech Republic. All the analysed honeys met the parameters defined by the legislation. Due to direct on-site sampling, honeys were characterized by a low 5-(hydroxymethyl)furfural (HMF) content (3.0 mg/kg) and high diastase activity (24.4 DN). Honeydew honeys had the highest proportion of higher carbohydrates, primarily of Melezitose (4.8 g/100 g) and Trehalose (1.3 g/100 g). The presence of higher carbohydrates was also confirmed in LH for Maltose (4.6 g/100 g) and Turanose (2.4 g/100 g).

Keywords: carbohydrates; colour of honey; pollen profile; trisaccharide; disaccharide

1. Introduction

Proclamation of the authenticity of honey is a key step in meeting consumer demands. Honey is a sweetener from which the consumer expects not only its sweet taste, but also other values. Many studies demonstrate that honey is a source of natural antioxidants, an antibiotic, and shows wound healing properties [1]. The honey also enhances the growth of lactic acid bacteria that positively affect liver, cardiovascular and gastrointestinal

problems [2]. Such an additional value can be specific sensory properties, chemical composition, or specific content of bioactive substances, as well as geographical origin. These characteristics are so important for consumers that they are willing to pay a higher price compared to honeys whose characteristic properties are not specific or do not have a clear geographical origin. Specific honeys and honeys with a certain geographical origin can be characterized on the basis of their physico-chemical composition, sensory properties and on the basis of melissopalynological analysis. These parameters can also be used to detect honey adulteration. Mainly, in the case of honey, it is adulterated commonly by starch syrup, inverted syrup, starch or inverted syrup fed to bees and low-quality honey added to high-priced honey [3]. On the other hand, honey has several sources of microbiological and parasitological contamination. Primary sources include pollen, the digestive tracts of honeybees, dust, air, soil, and nectar; secondary sources, such as honey handlers and processing, are easier to control by the application of good manufacturing practices. The major microbiological and parasitological contaminants of honey include molds and yeasts, as well as the spores of *Bacillus* spp., *Clostridium* spp., *Escherichia coli*, *Staphylococcus aureus*, *Ascosphaera apis*, *Nosema* spp., *Aspergillus flavus*, and *Aspergillus fumigatus* [4].

Honey from various countries can differ from each other, depending on the distance in which they are located, which can also be reflected in the occurrence of completely different botanical taxa that contribute to the sources of nectars, as well as the nature of agricultural activity. The presence of specific taxa in individual regions is used to determine the geographical origin as well as monofloral honeys. In both cases, melissopalynological analysis of specific botanical taxa is particularly important [5,6]. The characterization of important monofloral honeys within the European region is described in extensive studies [7–9]. Further characterizations of special or geographically defined honeys are described for some European countries, such as Bulgaria [10], Estonia [11], Canary Islands [12], Poland [13], Portugal [14], Greece [15–17], Romania [18], Spain [19,20], and Ireland [21]. However, there are also studies describing honeys from national park sites [22]. For the Czech Republic, the characterization of the composition of honeys has not yet been described.

The aim of the study is to characterize the physico-chemical properties of honeys and the melissopalynological profile of Czech honeys and their comparison with the published data on European honeys.

2. Materials and Methods

Honey samples were collected in 2019 and 2020 from hobby beekeepers in the Czech Republic. Honey collection was performed directly in the honeycombs and it was extracted separately for each sample. The honey was extracted on a manual four-frame LTR4S honey extractor (Logar trade d. o. o., PLN). The selection of sites was random, to cover the regional units of the Czech Republic including border sites (Figure 1). In total 316 honey samples from various sites were collected. 106 sites were sampled during two honey harvests (spring and summer). In total, 184 sites were selected for sampling and at least two samples were collected at 79 sites. Bee-handling data by the producers were obtained for each sample. Each sample was analysed according to the standard processing in duplicate.

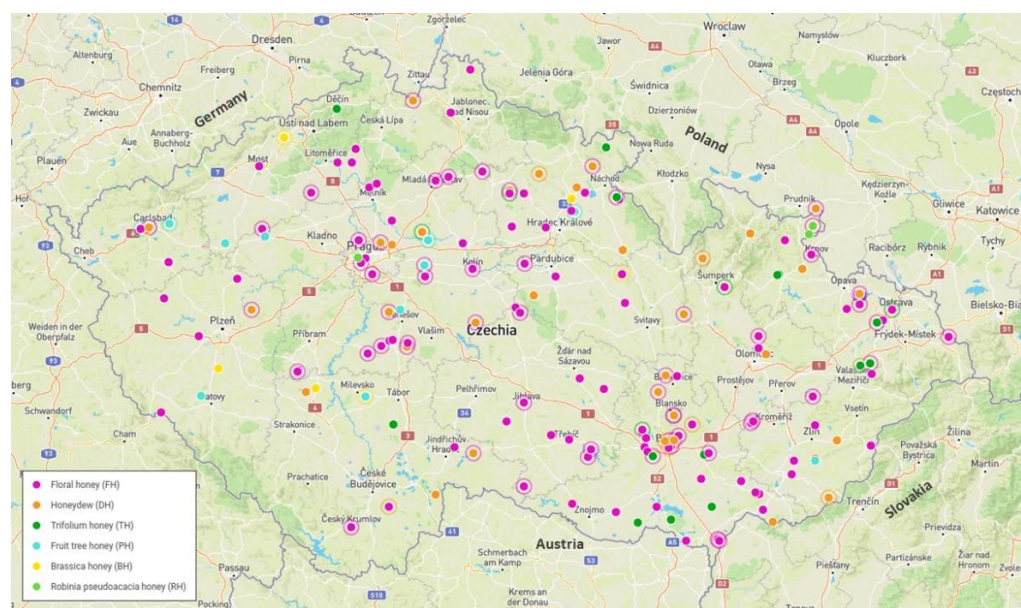


Figure 1. Sampling sites according to honey classification. Note: a double point on the map means multiple sampling in the given location.

The honey collection was classified according to melissopalynological analysis and electric conductivity (E. Cond.) into seven groups (Table 1).

Table 1. Groups of classified honeys.

Sample	Frequency	Threshold Criteria
Brassica honey (BH)	10	<0.8 mS/cm E. Cond. >70% specific pollen
Floral honey (FH) *	207	<0.8 mS/cm E. Cond.
Fruit tree honey (PH) *	15	<0.8 mS/cm E. Cond. >20% specific pollen or >15% when is in majority
Honeydew (HD) *	57	>0.8 mS/cm E. Cond. without specific pollen
Lime tree honey (LH)	5	>10% specific pollen
Robinia pseudoacacia honey (RH) **	4	<0.8 mS/cm E. Cond. >10% specific pollen
Trifolium honey (TH) **	18	>20% specific pollen

* not included in new trade specifications in Germany, ** non-compliance with new trade specifications in Germany [23]. Note: Except for FH, HD, RH and TH the honeys with higher content of pollen of the same origin are hereinafter referred to as monofloral honeys.

2.1. Physico-Chemical Analysis

Physico-chemical parameters of honey were determined according to the harmonized IHC (International Honey Commission) methods [24].

2.1.1. Water Content

The water content was analysed using a digital refractometer RM 40 (Mettler-Toledo, Greifensee, Switzerland) with an integrated thermostat (at 20 °C). The determined refractive index (RI) was converted to a percentage of water content (%).

2.1.2. Electrical Conductivity

Electrical conductivity was measured conductively at 20 °C (Multi 9310 IDS conductivity meter, WTW GmbH, Weilheim, Germany; IDS Tetra Con 925 electrode, WTW GmbH, Weilheim, Germany). Results were expressed in milliSiemens per centimeter (mS/cm).

2.1.3. HMF Analysis

HMF content was determined by the HPLC-UV method. A sample of honey was weighed (5 g) into a 100 mL volumetric flask and supplemented with a mobile phase (water/methanol 90:10 *v/v*). Subsequently, the sample was filtered using a 0.45 µm membrane filter (Teknokroma, Barcelona, Spain) into a 1.5 mL vial. The analysis was performed using an HPLC system (Agilent Technology 1290 Series, Santa Clara, CA, USA), a column (Hibar 125-4 Purospher STAR RP-18, 5 µm, Merck, Darmstadt, Germany), and a DAD detector (1290 Infinity Diode Array Detector, Santa Clara, CA, USA). The analysis conditions were as follows: isocratic elution, flow rate 1 mL/min, column temperature 20 °C, and detector wavelength 285 nm.

2.1.4. Diastase Activity

Diastase activity (expressed as diastase number, DN) was determined spectrophotometrically using the Phadebas method (Phadebas Honey Diastase Test, Phadebas AB, Kristianstad, Sweden). The analysis was performed using the manufacturer's instructions [25]. The absorbance of the solution was measured (620 nm) using a Spekol 1300 spectrophotometer (Analytik Jena GmbH, Jena, Germany).

2.1.5. Free Acidity

The free acidity (F. Ac.) of the honey was determined by alkalimetric titration (0.1 M NaOH, Penta, Prague, Czech Republic) to a final pH of 8.3. An automatic titrator T5 (Mettler-Toledo, Greifensee, Switzerland) and an electrode DGi115-SC (Mettler-Toledo, Greifensee, Switzerland) were used for the analysis.

2.1.6. Carbohydrate Content

Carbohydrate analysis was performed using an HPLC system ECP2000 (Ecom, Chrastany, Czech Republic), Separon SGX NH₂ column, 5 µm, 3 × 150 mm (Tessek Ltd., Prague, Czech Republic), tempering at 35 °C, refractometric detector RI2012 (Ecom, Chrastany, Czech Republic). The analysis was performed with external mixture of carbohydrate standards: glucose, fructose (Fluka Chemicals, Gillingham, UK), sucrose, maltose, melibiose, turanose, trehalose, maltotriose, melezitose (Sigma-Aldrich, Burlington, VT, USA). The standard mixture was used before each sample series.

2.1.7. Colour Measurement

Colour was measured photometrically using Hanna HI 96785 (Hanna Instruments, Woonsocket, RI, USA). The glycerol solution (Hanna Instruments, Woonsocket, RI, USA) was used as a standard. The result is expressed in mm Pfund, where the colour is in the range between 1 to 140 mm [26]. Before the colour measurement, the honey samples were tempered to 40 °C.

2.2. *Melissopalynological Analysis*

Honeys were processed by a modified melissopalynological method according to [5], where a 10.0 g sample of honey was mixed with 40 mL of distilled water at a temperature of 40 °C. After complete dissolution, the sample was filtered using a vacuum filtration kit (ThermoFisher, Waltham, MA, USA) through a 25 mm filter with 3 µm pore size (MF-Millipore, Germany). After the filtration, the sample was dried (24 h). Subsequently, the filter with pollen grains was placed on a drop of solacryl (Merci, Czech Republic) on a microscope slide, dripped with another drop of solacryl, and covered with a cover glass.

After drying to the required extent, the edges of the cover glass were covered with a layer of varnish to prevent the slides from drying out.

The samples were analysed under the Eclipse Ci-L microscope (Nikon, Tokyo, Japan) with motorized table of Proscan III (Prior, Rockland, MA, USA). Images were captured by the DFK 23U274 camera (Imaging Source, Bremen, Germany). The photosystem was calibrated using a USB4000-UV-VIS-ES spectrophotometer (Ocean Optics Inc., Orlando, FL, USA) to ensure the same lighting conditions. Pollen grains were captured in five different focal planes at distances of 8 μm from each other, and a depth of focus image was created, which was used subsequently to determine the botanical species of pollen grains. The microscope slides were scanned in random positions according to the counting matrix [5], so that at least 300 pollen grains were captured and subsequently analysed.

2.3. Statistical Analysis

The data were processed statistically using the 2021 XLSTAT software (Addinsoft, Paris, France). The data had no normal distribution (Shapiro-Wilk test). k means Kruskal–Wallis test was used to compare the physico-chemical and melissopalynological characteristics of the honey groups at the 0.05 significance level.

Data for uniform display of groups of Czech classified honeys and monofloral EU honeys are processed as a transformed value according to Persano Oddo and Piro [9]. The box-plot was created from data by Persano Oddo and Piro [9] and the results of our study. Values are expressed as mean value and \pm standard deviation.

3. Results and Discussion

3.1. Physico-Chemical Characterization of Czech Honey

The results of the physico-chemical analyses of the honeys described in Table 1 are summarized in Table 2. Physico-chemical parameters are important indicators for the classification of honey. The most widely accepted criterion is E. Cond., according to which honeys are divided into honeydew and blossom honeys. The limit value is 80 mS/m (0.8 mS/cm), provided that the F + G sum (sum of glucose and fructose) condition for honeydew is not less than 45 g/100 g and not less than 60 g/100 g for blossom and blend honey [27]. For pharmaceutical honey the maximum limit is 0.8 mS/cm [28,29]. Based on physico-chemical parameters, the characterization of monofloral honeys is not established legislatively, but is specified in literature sources [7–9,14,19]. Physico-chemical properties mainly reflect the botanical origin, although small differences are also described for different localities. For example, a difference is described in E. Cond. in Greek Thyme honey (0.40 mS/cm) [30] compared to Italian Thyme honey (0.38 mS/cm). For Lime tree, minor differences are reported in E. Cond. as well, namely 0.67 mS/cm in Italy and 0.69 mS/cm in Bulgaria [8,10]. Physico-chemical parameters can also characterize blend honeys from individual countries, but it is necessary to take into account the great variability. For example, in Portuguese artisanal honey, the average conductivity was 0.6 mS/cm (min./max.; 0.5–0.9 mS/cm) compared to Irish honeys, where the average conductivity was 0.3 mS/cm (min./max.; 0.1–0.5 mS/cm) [21]. For comparison, in Czech FH the average conductivity was 0.4 mS/cm (min./max.; 0.2–0.8 mS/cm) and in HD it reached 1.1 mS/cm (min./max.; 0.8–1.6 mS/cm).

Table 2. Physico-chemical parameters of Czech honeys.

	BH	FH	PH	HD	LH	RH	TH
E. Cond. (mS/cm)	0.24 ± 7 ^a	0.4 ± 17.1 ^a	0.5 ± 20.9 ^{ab}	1.1 ± 19.7 ^c	0.7 ± 13.5 ^{bc}	0.6 ± 11.8 ^{ab}	0.6 ± 31.8 ^b
Colour (Pfund)	73.3 ± 27.8 ^{ab}	49.4 ± 20 ^a	59.1 ± 11.5 ^{ab}	76.3 ± 16.8 ^b	45.6 ± 21.2 ^a	47.4 ± 12 ^a	50 ± 17.1 ^a
HMF (mg/kg)	2.5 ± 1.9	3.1 ± 3.9	4 ± 3.4	2.5 ± 3.5	2.5 ± 2.8	1.7 ± 1.3	3.5 ± 6.2
Diastase (DN)	16 ± 1.8 ^a	25.1 ± 7.8 ^b	24.4 ± 6.4 ^b	24.1 ± 7.8 ^b	18.6 ± 7.2 ^{ab}	24.4 ± 1.5 ^b	23.5 ± 6.8 ^{ab}
Water (g/100 g)	17 ± 1.5	17.8 ± 1.6	17.1 ± 1.1	16.6 ± 1.4	16.2 ± 2.4	16.4 ± 0.9	17.2 ± 1.5
F. Ac. (meq/kg)	14.7 ± 6.1 ^a	21.2 ± 8.3 ^{ab}	22 ± 10.8 ^{ab}	34.5 ± 5.6 ^c	20.5 ± 6.6 ^{ab}	23.7 ± 6.4 ^{ab}	25.8 ± 10.8 ^b
Fructose (g/100 g)	37 ± 2.2 ^b	37.3 ± 2.6 ^b	36.7 ± 3 ^b	33.1 ± 2.8 ^a	34.5 ± 3.4 ^{ab}	37 ± 1.1 ^b	36.3 ± 2.5 ^b
Glucose (g/100 g)	36.5 ± 2.9 ^c	34.3 ± 3.2 ^{bc}	31.7 ± 3.9 ^b	28.1 ± 2.4 ^a	30 ± 1.6 ^{ab}	31.8 ± 1.8 ^{bc}	30.7 ± 1.8 ^{ab}
F + G (g/100 g)	73.5 ± 4.8 ^b	71.6 ± 5 ^b	68.4 ± 6.5 ^b	61.2 ± 4.9 ^a	64.6 ± 4.6 ^{ab}	68.8 ± 1.3 ^{ab}	67.1 ± 4 ^{ab}
F/G (g/100 g)	1 ± 0.1 ^a	1.1 ± 0.1 ^{ab}	1.2 ± 0.1 ^{bc}	1.2 ± 0.1 ^c	1.1 ± 0.1 ^{abc}	1.2 ± 0.1 ^{bc}	1.2 ± 0.1 ^c
Sucrose (g/100 g)	0.1 ± 0.2 ^a	0.1 ± 0.3 ^a	0.2 ± 0.7 ^a	0.3 ± 0.4 ^a	0.5 ± 0.6 ^{ab}	0.3 ± 0.4 ^{ab}	0.8 ± 1.3 ^b
Maltose (g/100 g)	2.9 ± 1	3.2 ± 1	4 ± 1.3	3.5 ± 0.7	4.6 ± 1.9	4.5 ± 1.1	3.7 ± 1
Melibiose (g/100 g)	<0.2	<0.2	<0.2	0 ± 0.1 ^b	<0.2	<0.2	0 ± 0.1 ^b
Turanose (g/100 g)	1.3 ± 0.4 ^a	1.7 ± 0.5 ^{ab}	1.9 ± 0.5 ^{bc}	2 ± 0.4 ^c	2.4 ± 1 ^c	2.2 ± 0.4 ^c	2.1 ± 0.6 ^c
Trehalose (g/100 g)	0.4 ± 0.4 ^a	0.6 ± 0.4 ^a	0.4 ± 0.4 ^a	1.3 ± 0.6 ^b	0.5 ± 0.4 ^a	0.6 ± 0.4 ^{ab}	0.8 ± 0.3 ^{ab}
Maltotriose (g/100 g)	0 ± 0	0 ± 0.1	0.1 ± 0.2	0.3 ± 0.4	0.1 ± 0.1	<0.1	0.1 ± 0.3
Melezitose (g/100 g)	0.6 ± 0.8 ^a	0.6 ± 1 ^a	0.7 ± 0.6 ^a	4.9 ± 3.3 ^b	0.8 ± 0.8 ^{ab}	0.8 ± 0.8 ^a	1.3 ± 2.2 ^a

Note: Mean values denoted by different letters at individual rows are significantly different ($p < 0.05$). Data are expressed as mean value ± standard deviation.

Within the Czech Republic, statistically significant differences for E. Cond. were demonstrated. BH and FH were statistically significantly different from TH and also from HD ($p < 0.05$). This finding confirms that honeydew honeys are characterized by higher E. Cond., which also applies to lime honey, which is not a typical blossom honey because honeydew from plant-sucking insects (Hemiptera), in particular *Eucallipterus tiliac* and *Eulecanium coryli* (L.) [31], also play a significant role in its composition. A suitable parameter for the classification of honeys is also its colour, especially for monofloral honeys [32]. Our results confirmed the difference between HD and FH, LH, RH, and TH honeys. This finding is consistent with [9] who also confirmed that there was no difference between lime, rapeseed, and acacia honeys (Figure 2).

HMF, Diastase, % Water, and F. Ac. are physico-chemical parameters that are greatly affected by the handling of honey [33]. These measured values in our study (Table 2) are influenced by the method of sampling, where the honey was taken directly in the honeycombs and then uniformly processed and stored. HMF content in all Czech honeys was <5 mg/kg. This is also consistent with a study [9] where it was found that over 70% of the analysed European honeys are below this limit. Differences in the HMF content between Czech honeys have not been demonstrated, as the HMF content is not the nature of a type of honey, but an indicator of processing and storage conditions. HMF is also used as quality indicator for pharmaceutical honey where maximum limit of 80 mg/kg is allowed [28,29]. The diastase activity in Czech honeys was higher in comparison with one study [32], except for rapeseed honey, for which a lower average value (16 DN) was measured than the value reported in the study (27 DN). It can be assumed that the higher measured values are caused by gentle handling of the honey after extraction, which took place under constant conditions, which is not easy to observe in processing and business practices. A statistically significant difference was demonstrated between RH and FH, PH, HD, and RH honeys ($p < 0.05$). The different water content in honeys is mainly due to climatic conditions and the time of honeycomb harvesting. In Czech honeys, the differences between honeys have not been demonstrated. The last parameter is F. Ac. This parameter is influenced by the content of organic acids from botanical sources and, in case of higher concentrations of organic acids, and it indicates mainly the fermentation of honey. Therefore, it can also be used to classify honeys [32]. This statement was also confirmed by our results, where a statistically significant difference was demonstrated between HD and other honeys and also between BH and TH ($p < 0.05$) (Table 2). In comparison with European honeys, however, the average content of F. Ac. in Czech honeys is lower in RH (15 meq/kg) and higher in HD (34 meq/kg) (Figure 2).

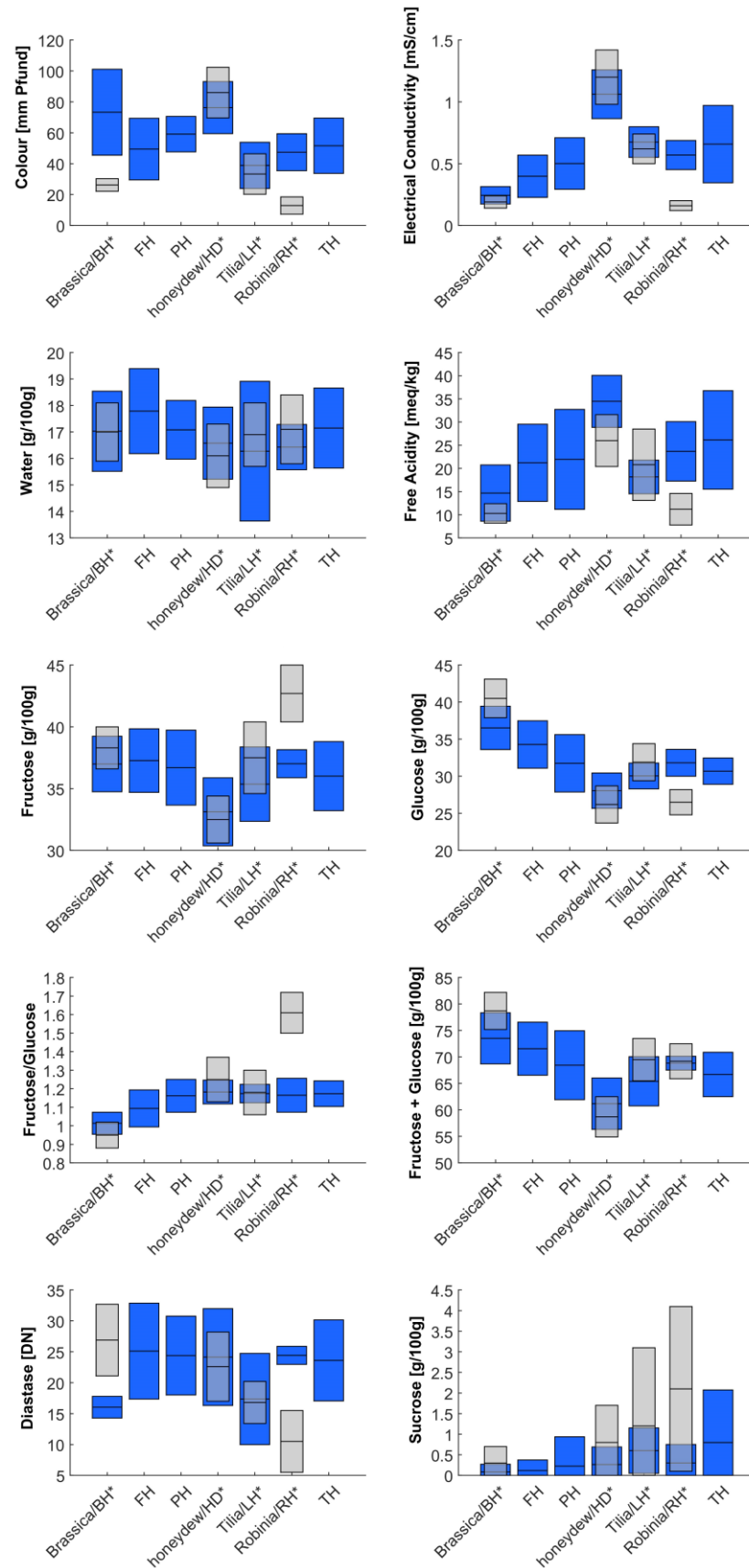


Figure 2. Variability of the various parameters in European and Czech honey. Grey—European honey [9]; blue—Czech honey.

Carbohydrates are an important ingredient in honey. On the one hand, they contribute to the high energy value of honey as a foodstuff, and under certain conditions they can be used for the classification of honeys. The amount and ratio of glucose and fructose can be used to sort monofloral honeys, and other honey carbohydrates can be used to classify honey into blossom honey and honeydew [32]. The representation of carbohydrates in Czech honeys is shown in Table 2. The fructose content of TH and RH was lower compared to European honeys (Figure 2). A statistically significant difference between honeys was confirmed, as the fructose content was lower in HD compared to BH, FH, PH and RH ($p < 0.05$). This finding is in line with [32] and confirms the possibility of using this carbohydrate for honey classification. Another important carbohydrate is glucose (Figure 2). Compared to European honeys, the fructose content was slightly higher for HD, LH, and RH, and slightly lower for BH. The glucose content was highest in BH, which differed statistically significantly from all other Czech honeys ($p < 0.05$). Differences between PH and HD were further confirmed for Czech honeys.

The sum of F + G and F/G ratio (fructose to glucose ratio) are also used for honey characterization purposes. The sum of reducing honey sugars of F + G is important especially as an indicator of the addition or occurrence of other carbohydrates, and must not fall below 60 g/100 g for blossom honey and below 45 g/100 g for honeydew honey and blends of honeydew [27]. The F/G ratio is used as an indicator of the crystallization rate [34]. These parameters can also be used for honey classification. Compared to European honeys, F + G in Czech honeys was slightly higher for HD and slightly lower for BH, RH, and TH (Figure 2). Differences between Czech honeys were also confirmed for this parameter. HD was statistically significantly different from other honeys ($p < 0.05$). Compared to European honeys, the F/G ratio in Czech honeys was higher for BH, RH, and HD, and slightly lower for LH (Figure 2). Within Czech honeys, a statistically significant difference between BH and all other honeys was confirmed, and there were also differences between HD, LH, RH and TH, and BH, FH and PH honeys ($p < 0.05$).

Sucrose is a carbohydrate that occurs naturally in small amounts in honey. For Czech honeys, the content was up to 1 g/100 g (min. 0.1, max. 0.8 g/100 g). EU legislation allows a maximum of 5 g/100 g for honey or 10 g/100 g for *Robinia pseudoacacia*, *Medicago sativa* and other sucrose-rich species that are not produced in the Czech Republic. The highest permissible content is for lavender honeys and borage honeys (*Borago officinalis*), where a maximum of 15 g/100 g is allowed. There was a statistically significant difference in Czech honeys between TH and other honeys. The sucrose content in TH was 0.9 g/100 g, where a higher value is attributed to botanical relationships (*Fabaceae*) in *Trifolium* (*Trifolium* species) and *Medicago sativa* (alfalfa), where a higher sucrose content is described.

In addition to monosaccharides and sucrose, honey also contains other disaccharides and trisaccharides [35]. With regard to the connection of these carbohydrates to the origin of honey, disaccharides (maltose, melibiose, turanose, trehalose) and trisaccharides (maltotriose, melezitose) were analysed in our study (Table 2). These carbohydrates were not detected in all samples. Therefore, we also provide the number of occurrences for individual carbohydrates. Separate groups of honeys did not differ significantly in the content of maltose ($n = 317$), and the total content was below 8 g/100 g. Melibiose was present in honey in small amounts ($n = 4$), and in most honey samples it was below the detection limit of the method used. Melibiose has not been confirmed at all in BH, FH, PH, LH, or RH. It was confirmed at low concentrations in HD (0.02 g/100 g, $n = 3$) and TH (0.01 g/100 g, $n = 1$). Another disaccharide confirmed in Czech honeys was turanose ($n = 315$), where the total content was below 5 g/100 g. Higher amounts were confirmed in HD, LH, RH, and TH honeys compared to BH and FH. Among these groups, there was no statistically significant difference confirmed ($p < 0.05$). Trehalose was also confirmed in Czech honeys ($n = 266$), but the total content was below 3 g/100 g. A significant difference was confirmed between BH, FH, PH, LH, and HD.

Of the confirmed trisaccharides, the content of Melezitose was higher (max. 14 g/100 g, $n = 194$) compared to Maltotriose (max. 1 g/100 g, $n = 64$). The content of Melezitose

was significantly different in HD compared to BH, FH, PH, RH ($p < 0.05$). No statistically significant difference was confirmed for Maltotriose compared to Melezitose. Disaccharides and trisaccharides are mainly present in honeydew honeys. In a New Zealand study, Melezitose, Turanose, Maltose, Maltotriose, Trehalose were confirmed in honeydew honeys [36]. Melezitose was also confirmed in honeydew honeys in a Bulgarian study, with Maltose and Trehalose also occurring in blossom honey [37]. The presence of these carbohydrates was also confirmed in a Spanish monofloral honey [20]. The reason for the occurrence of di- and trisaccharides in honeydew honeys is their high content in honeydew, which is produced by insects (Hemipteran species). The proportion of di- and trisaccharides differs according to the Hemipteran species but also from the botanical species on which the hemipteran species feed on [31].

3.2. Pollen Profile of Czech Honey

Pollen is also an integral part of honey. Pollen enters honey together with nectar from nectariferous plants and also through social contact of bees in the hive. Thus, in honey we can find pollen of both nectariferous as well as non-nectariferous plants, which are sought by bees for the creation of pollen reserves. Melissopalynological analysis is a suitable tool to determine the botanical origin of honey. For this purpose, the relative frequencies of pollen types of nectariferous species are calculated [38]. Melissopalynological analysis can also be used to determine the geographical origin of honey, especially if the honey comes from other climatic regions. It is possible to use relative frequencies of pollen types for this purpose, but qualitative melissopalynology analysis is the most common [3,5,39]. The results of the melissopalynological analysis are summarized in Table 3.

Table 3. Relative pollen frequency of Czech honeys.

	BH	FH	PH	HD	LH	RH	TH
Aceraceae							
<i>Acer</i> sp. ($n = 106$)	0.3 ± 0.4	1.2 ± 2.8	2.9 ± 8.7	0.3 ± 0.8	<0.1	<0.1	1 ± 1.8
Asteraceae							
<i>Achillea</i> sp. ($n = 20$)	<0.1	0.7 ± 4.6	0.1 ± 0.3	0.1 ± 0.2	<0.1	<0.1	0.1 ± 0.5
<i>Artemisia</i> sp. * ($n = 163$)	0.8 ± 1.3	1.8 ± 3.1	4.6 ± 7.7	2.8 ± 4	1.2 ± 1.9	<0.1	0.4 ± 1
<i>Bellis</i> sp. * ($n = 47$)	0 ± 0	0.1 ± 0.5	0.3 ± 0.6	0.4 ± 1.4	0.6 ± 0.9	0.3 ± 0.5	0.0 ± 0.1
<i>Helianthus</i> sp. ($n = 36$)	0.1 ± 0.2	0.2 ± 1	0.1 ± 0.4	0.1 ± 0.4	0.1 ± 0.2	0.5 ± 1	0.3 ± 1
<i>Taraxacum</i> sp. ($n = 89$)	0.1 ± 0.2	0.4 ± 1.6	0.2 ± 0.5	0.5 ± 1.4	<0.1	0.1 ± 0.2	0.6 ± 0.6
Betulaceae							
<i>Alnus</i> sp. * ($n = 23$)	<0.1	0.1 ± 0.3	0.0 ± 0.1	0.1 ± 0.3	<0.1	<0.1	0.1 ± 0.2
<i>Corylus</i> sp. * ($n = 28$)	0.2 ± 0.8	0.2 ± 0.8	0.8 ± 2.1	0.8 ± 5.2	<0.1	<0.1	<0.1
Boraginaceae							
<i>Myosotis</i> sp. ($n = 274$)	1.9 ± 1.9 ^a	6.9 ± 10.4 ^a	1.9 ± 1.5 ^a	16.1 ± 17.9 ^b	9.4 ± 5.3 ^b	14.7 ± 15.6 ^b	10.7 ± 12.2 ^{ab}
Brassicaceae							
<i>Brassica</i> sp. ($n = 291$)	79.5 ± 7.1 ^c	41.6 ± 22.5 ^b	24.1 ± 23.6 ^b	21.7 ± 23.2 ^a	6.8 ± 7.3 ^a	15 ± 14.4 ^a	10.84 ± 9.5 ^a
Hydrophyllaceae							
<i>Phacelia</i> sp. ($n = 184$)	1.1 ± 1.5	4.3 ± 8.1	7.8 ± 10.7	5.6 ± 7.8	12.3 ± 19.2	0.3 ± 0.6	2.7 ± 3.4
Fagaceae							
<i>Quercus</i> sp. * ($n = 59$)	0.9 ± 2.8	0.4 ± 1.8	1 ± 1.9	0.3 ± 0.7	<0.1	<0.1	0.3 ± 0.9
Rhamnaceae							
<i>Rhamnus</i> sp. ($n = 35$)	<0.1	0.19 ± 1.4 ^a	0.48 ± 0.88 ^b	0.22 ± 0.86 ^{ab}	<0.1	<0.1	0.12 ± 0.49 ^a
Rosaceae							
<i>Pyrus/Prunus</i> sp. ($n = 253$)	1 ± 1 ^a	3.9 ± 4.4 ^a	31.7 ± 13.9 ^b	6.4 ± 8.2 ^a	1.6 ± 2.5 ^a	1.5 ± 3 ^a	3.7 ± 4.9 ^a
<i>Rubus</i> sp. ($n = 81$)	<0.1	0.5 ± 1.9	0.8 ± 1.8	0.4 ± 1.2	1.8 ± 3.8	0.5 ± 1	1 ± 2.9
Salicaceae							
<i>Salix</i> sp. ($n = 164$)	3.2 ± 5.1	4.7 ± 7.3	0.5 ± 1.3	0.7 ± 1.6	0.7 ± 1.1	2.9 ± 1.9	1.4 ± 2.3
Fabaceae							
<i>Robinia pseudoacacia</i> ($n = 117$)	0.0 ± 0.1 ^a	0.8 ± 1.6 ^a	1.2 ± 1.8 ^{ab}	1.6 ± 3.3 ^a	0.3 ± 0.7 ^a	12.7 ± 0.9 ^b	1.2 ± 2.6 ^a
<i>Trifolium</i> sp. ($n = 238$)	0.6 ± 0.7 ^a	3.7 ± 4.4 ^{ab}	1.9 ± 2.8 ^{ab}	3.6 ± 4.7 ^{ab}	18.6 ± 21.1 ^{bc}	8.6 ± 7 ^{bc}	33 ± 12 ^c
<i>Vicia</i> sp. ($n = 223$)	2.5 ± 4.4	2.3 ± 4.4	4.3 ± 4.7	3.3 ± 4.6	2 ± 3.8	9.7 ± 6.9	5.1 ± 7.1
Tiliaceae							
<i>Tilia</i> sp. ($n = 183$)	0.1 ± 0.1 ^a	1.1 ± 1.8 ^a	2.4 ± 4.2 ^{ab}	1.9 ± 3.2 ^{ab}	22.8 ± 14.4 ^b	2.3 ± 2.1 ^{ab}	3.8 ± 4 ^b
Apiaceae (Umbelliferae)							
<i>Apiales</i> *# ($n = 282$)	2.8 ± 2.9	6.4 ± 8.4	3.4 ± 4.8	6.9 ± 8	4.2 ± 3.8	8.5 ± 5.5	5.8 ± 3.7

Note: * non-nectariferous taxa (n = number of pollen occurrences in analysed honeys, grey—specific pollen taxon; # e.g., *Daucus carota*). Data are expressed as mean value ± standard deviation. Mean values denoted by different letters at individual rows are significantly different ($p < 0.05$).

The results of melissopalynological analysis confirmed statistically significant differences of some pollen taxa between the analysed honey species. Especially in monofloral honeys (BH, PH, and LH), the dominant taxon was pollen from these plants (specific pollen). It is generally not possible to say how much pollen is necessary to classify it as a monofloral honey. There are several reasons: an important role is played by the diverse pollen-producing capacity of plants [40], and geographic aspects (climate, soil acidity or other pedologic conditions) have an influence on the nectar and honey composition [8]. The dominant taxon in BH was *Brassica* sp. pollen, the average amount was 79% (min–max, 70–87%). The amount of *Brassica* sp. pollen in BH was statistically significantly different from other honeys (FH, PH, HD, LH, RH, and TH). Due to the high pollen-producing capacity of the *Brassica* genus, it is typical that monofloral honeys have a content of this taxon higher than 60%, and some authors even report over 80% [5,41]. For rapeseed honeys from Bulgaria, the rapeseed content was 87–93% [10]. In the European survey study the content of rapeseed pollen in rapeseed honeys was 83% (61–99%) [9]. A high amount of *Brassica* sp. pollen was, however, also confirmed in FH and PH (Table 3), where these honeys differed significantly from LH, RH, TH, and BH ($p < 0.05$). This is caused by the high occurrence of rapeseed pollen in hives from near agricultural areas with *Brassica napus* or *Brassica alba* [42].

Polyfloral honeys are characterized by the presence of pollen from various botanical taxa. In the Czech FH, a significant taxon was *Brassica* sp. (42%) pollen. This finding is also reported by other authors, where the content of rapeseed pollen in polyfloral honeys ranges from 1%–96% [11,13,14,21,43]. The *Brassica* sp. pollen content was significant for FH in contrast to BH, HD, LH, RH, and TH ($p < 0.05$). Another important pollen (3–15%) with a higher incidence was *Myosotis* sp. pollen (7%), which differed significantly from HD, LH, and RH. Other important pollens included *Salix* sp. (5%), *Phacelia* sp. (4%), and *Trifolium* sp. (4%). A non-nectariferous taxon high in content was *Umbellifera* (6%). Taxa with low content, but forming statistically different groups, were *Rhamnus*, *Pyrus/Prunus* (differences in PH), *Robinia pseudoacacia* (differences in RH), *Trifolium* sp. (differences in TH), and *Tilia* sp. (differences in LH).

Fruit trees are an important source of nectar and pollen for bee development in the spring. Floral honeys with a dominant occurrence of these taxa are not often described in the literature. For the Czech Republic, we have included this group in order to describe honey, which is dominated by *Pyrus/Prunus* pollen. For these honeys, the average pollen content of *Pyrus/Prunus* taxa was 32% (min–max, 18–65%) and the minimum discriminatory limit was 15%. A secondary pollen (15–45%) in PH was rapeseed pollen (24%), and other important pollens included *Artemisa* sp. 5%, *Vicia* sp. (4%); a non-nectariferous taxon high in content was *Umbellifera* (3%). Pollen with low content, forming statistically significant differences, were *Myosotis* sp. (differences in HD, LH, RH), *Rhamnus* sp. (differences in BH, FH, LH, RH), and *Trifolium* sp. (differences in TH).

A source of carbohydrates for HD is honeydew produced by the *Hemipteran* species. These honeys do not have a uniform composition because they are produced by different representatives of insects and also differ according to the botanical taxa on which the insect sucks honeydew [31]. The most common sources of honeydew include Coniferae of *Abies* sp., *Picea excelsa* (Lam) Link., *Pinus* sp., and Latifoliae of *Quercus* sp., *Tilia* sp. These honeys are microscopically characterized by the presence of honeydew elements. [9]. The pollen profile of Czech HD was characterized by a high content of pollen of nectariferous taxa. The highest amount was of *Brassica* sp. 22% (min–max, 0–63%). In pollen content of *Brassica* sp.; however, HD differed from BH, FH, and PH. The secondary pollen in HD was *Myosotis* sp. which represented 16%. In the amount of this pollen, HD differed significantly from BH, FH, and PH. In HD, important pollens included *Pyrus/Prunus* 6% (differences with PH), *Phacelia* sp. 6%, and *Trifolium* sp. 4% (differences with TH). A non-nectariferous taxon high in content was *Umbellifera* (7%). A pollen with a low content forming a statistically significant difference was *Rhobinia pseudoacacia* (differences in RH). Opinions on the content of pollen in honeydew honeys differ, as reported in a European study, and the pollen

profile of honeydew honey is composed mainly of non-nectariferous taxa [9]; in Spanish honeydew honeys the average pollen content was 26%, most of which was nectariferous taxa (*Castanea* 42% and *Rubus* 32%) [44]. Another Spanish study confirmed the presence of *Castanea* and *Rubus* as the dominant pollens (>45%) in honeydew honeys, and other frequent taxa included, for example, *Salix* and *Trifolium* in Evergreen Oak Honeydew Honeys, among other taxa, such as *Brassica* sp., *Trifolium* sp. and *Taraxacum*. The *Quercus* pollen was present in low levels [45]. Similar results were confirmed by our study. Pollen representation in honeydew honeys was also confirmed in Greek honeydew honey, where the dominant taxon was *Castanea* (23–92%) and the accompanying taxa included *Trifolium* sp. and *Quercus* sp. [46] which were also demonstrated in our study.

Lime trees are a very good source of nectar and honeydew. Due to the low pollen-producing capacity of this botanical taxon, the percentage of *Tilia* pollen is low, with an average of 23% [9,47]. In French lime honeys, the lime pollen content ranged from 5–23% [48]. In the Czech LH, the lime pollen content averaged 23% (min–max, 13–39%). The secondary pollen was *Trifolium* sp. (19%) where there were differences comparing to BH. The important pollens in TH were *Phacelia* sp. 12% and *Myosotis* sp. 9% (differences comparing to BH, FH, and PH). Non-nectariferous important pollen was *Umbellifera* (4%). The presence of other pollen taxa in lime honeys is in accordance with literature. Pollen from dandelions, buckwheat, *Brassica* sp., and other pollen taxa not demonstrated in our study, were reported in lime honey [48]. The presence of *Brassica* sp., *Robinia pseudoacacia*, and *Amorpha fruticosa* pollen were confirmed in Serbian lime honeys [49].

Robinia pseudoacacia are a good source of nectar and a poor source of pollen. Therefore, their pollen content in RH is low. The average pollen content in one European study was 28% [9]. In the Czech RH, the *Robinia pseudoacacia* pollen content averaged at 13% (min–max, 12–14%). The secondary pollens in RH were *Brassica* sp. (15%) and *Myosotis* sp. (15%), where there were differences comparing to BH, FH, and PH, and *Trifolium* sp. (9%) with differences comparing to BH, *Vicia* sp. (10%). Non-nectariferous important pollen was *Umbellifera* (8%). *Robinia* honey (Black locust) is more typical for eastern region. In Croatia, other pollen taxa were also confirmed in *Robinia* honey; the secondary pollen was *Rosaceae*, and other important pollens included *Fabaceae*, *Amorphafrutcosae*, and *Oleaceae* [50]. These results are not in agreement with our study, but the differences are given by dissimilar climatic conditions. In comparison with Romania, *Robinia* honey contained *Brassica napus* as the secondary pollen [51], which is in agreement with our results.

Trifolium honey does not belong among common monofloral honeys. Even though the *Trifolium* genus is preferred for *Apis mellifera*, these plants have a long flowering time period and they provide a source of nectar and pollen during the season [52]. The total pollen contents varies and can reach up to 78% in some honeys [21,53]. In Czech TH, the mean content was 33% (min–max, 22–59%). The important pollens in TH included *Brassica* sp. 11% (differences comparing to BH, FH, and PH) and *Myosotis* sp. 11%, *Vicia* 5%, *Pyrus/Prunus* 4% (differences comparing to TH), and *Tilia* 4% (differences comparing to BH, FH). Non-nectariferous important pollen was *Umbellifera* (6%). Honeys high in *Trifolium* pollen content were also confirmed in an Irish study. The accompanying pollens included *Brassica* sp., *Rubus* sp., and other taxa.

3.3. Comparison with European Honeys

Honey and its composition has a variable character. It cannot be assumed that its properties will stay unchanged both in place and in time. There are many influences on the physico-chemical properties but also on the pollen profile. An important factor is the location from which the honey comes. To give an impression of the conditions in the Czech Republic, we present a brief description of it. The climate in the Czech Republic is temperate oceanic, with the coldest month averaging above 0 °C (Köppen classification—Cfb) in the lowlands and warm-summer humid continental climate, and the coldest month averaging below 0 °C in the highlands (Dfb). Agricultural land, meadows and forests prevail in the Czech territory. In the Czech Republic, the agricultural land is farmed mainly

by large-scale farms. Farming is concentrated in more fertile areas. The agriculturally marginal areas include grasslands in the areas of higher altitudes, with steeper gradient and worse soil quality [54]. Four major forest types, corresponding to phytosociological associations, can be distinguished in the Czech Republic: Hercynian, widespread in the Bohemian Massif (*Melampyro nemorosi-Carpinetum*); Pannonian, confined to southern Moravia (*Primulo veris-Carpinetum*); Carpathian, common in eastern and central Moravia (*Tilio cordatae-Carpinetum*); and wet habitat type, occurring mainly on soft bedrocks in eastern Bohemia, northern Moravia and Silesia (*Stellario holostea-Carpinetum*) [55].

Neighbouring countries also have a similar climatic and botanical character, but differ in their agricultural activity. Agricultural structure in Poland, Austria, and Germany is more fragmented [54]. This has increased biodiversity in these regions. However, differences can also be expected in comparison with other countries, where especially in the southern and northern regions of Europe, there are different climatic conditions, but also different botanical representation of nectariferous and non-nectariferous plants. For a better description of the achieved results, we compared the results for the Czech Republic with the most comprehensive European survey study [9] characterizing European honeys (Figure 2). This study includes honeys from various European countries, primarily Italy, France, and Germany. For better comparison of the sampled honeys with Czech and European honeys we provide comparison plots (Supplementary I, Figure S1) based on transformed value as in [9].

The dominant agricultural crop grown in the Czech Republic is *Brassica napus*. Intensive growing of this taxon enables the collection of monofloral rapeseed honeys in the Czech Republic, but it also has an effect on the pollen profile of polyfloral honeys from the agricultural areas of the Czech Republic. The effect of agricultural crops on the pollen profile in honey was also confirmed in Finland, when the change in agricultural activity increased the *Brassica* pollen content up to 60%. The authors also confirmed a simultaneous decrease in the *Trifolium* species [56,57]. Both the above taxa were confirmed in the Czech Republic and in some cases they were classified as monofloral honeys. For BH, data overlapped for all physico-chemical parameters except for the colour and diastase activity (Figures 2 and 3, BH). Czech rapeseed honey was darker with greater variability. Differences in the colour of rapeseed honeys were also confirmed in the study by [58] where the colour of *Brassica* honey ranged from light amber to dark amber. However, the colour did not depend on the amount of rapeseed pollen. We assume that the different coloration of BH is due to the proportion of other botanical taxa in the honey, which caused a change in the colour of honey. Typical physico-chemical parameters for European TH are not commonly described. In our study, evaluated TH was non-compliant with Germany trade specifications in pollen profile, E. Cond. and colour [23]. Their summary is given in Figure 2 (TH). We attribute the large variability within TH to the different representation of this taxon in honey, but also to the different representation of species of this family. The most common monofloral honeys of the *Trifolium* family in Europe include *Trifolium alexandrinum*, *incarnatum*, *pratense*, *repens*. In addition, in British and Croatian studies the authors confirmed the high frequency of this family in polyfloral honeys [21,59]. For *Trifolium* it is typical that many *Trifolium* species are represented in the same habitat [52].

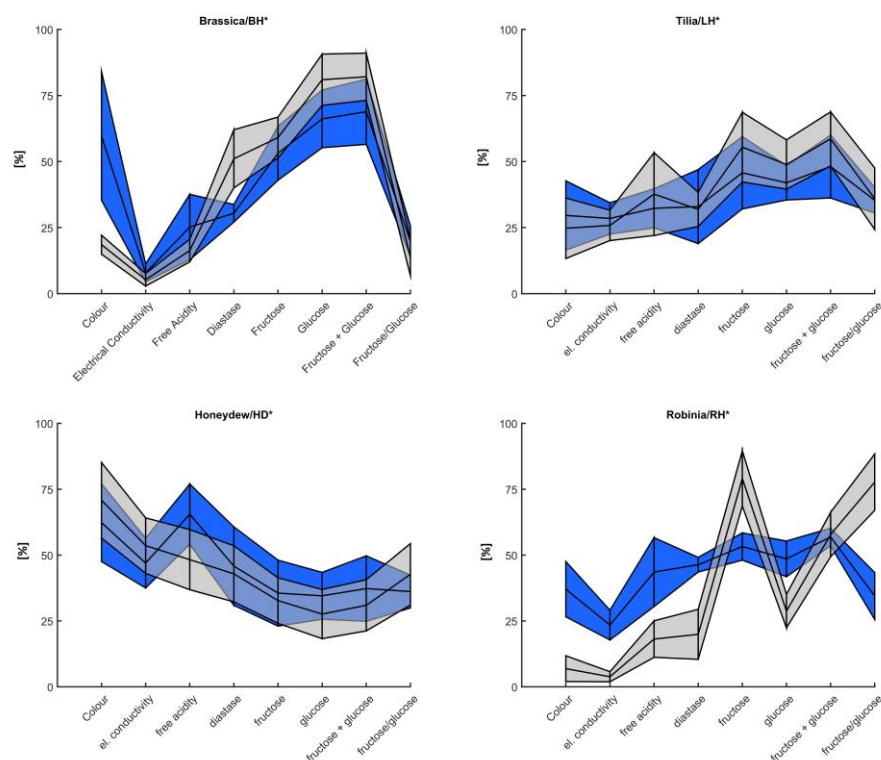


Figure 3. Physico-chemical profiles of the European and Czech honeys. Grey—European honey [9]; blue—Czech honey.

Other types of honey in the Czech Republic, primarily FH and PH, are also affected by agricultural activities. A comparison of the physico-chemical properties of these honeys with European as well as Czech generic honeys is demonstrated in Figure 2. Both types of honey represent a heterogeneous group, which depends on the botanical taxa of the areas. A comprehensive European survey study for FH is not available and, given botanical dependence, is rather a matter of national characterization. For comparison, we present the average values of physico-chemical parameters of some European honeys. For Portuguese polyfloral honeys: E. Cond. 0.6 mS/cm, F. Ac. 28.9 meq/kg, diastase 16, F + G 72.8 g/100 g and sucrose 3.6 g/100 g [14]. For Italian polyfloral honeys: E. Cond. 0.4 mS/cm, and F Ac. 25.1 meq/kg [60]. These results are comparable to Czech FH. For Croatian polyfloral honeys: E. Cond. 0.5 mS/cm, F. Ac. 28.9 meq/kg, and diastase 18 DN, a higher content of HMF than in honeys from the Czech Republic was recorded [61]. For comparison, Turkish polyfloral honeys have higher HMF and lower diastase, which corresponds to a warmer climate (E. Cond. 0.8 mS/cm, HMF 14.7 mg/kg, diastase 9 DN, F + G 57.4 g/100 g and sucrose 0.9 g/100 g [62]. However, even for FH, some differences can be expected depending on the botanical origin, as stated by [63].

There is no comparison for PH with European honeys. The occurrence of monofloral honeys of fruit trees is described rather exceptionally in the southern regions of the EU (Italy, Spain) or in apple trees in England. These are most often honeys of *Malus domestica*, *Prunus avium*, *Prunus dulcis*, or *Prunus mahaleb* taxa, or unspecified origin of fruit trees [7]. The exact botanical origin was not distinguished in our category and it is a group of *Pyrus/Prunus/Malus*. Based on physico-chemical parameters, these honeys resemble polyfloral honeys. This is also consistent with the pollen profile when the secondary pollen is *Brassica*.

Lime honey is a widespread monofloral honey. They occur mainly in Eastern Europe, less often in the western and northern parts of the EU [5]. Comparison with European honeys (Figures 2 and 3, LH) confirmed the similarity of the Czech LH with

European honeys. Glucose, fructose, and sucrose in the Czech LH were at the upper limit of EU Lime honey [9]. On the other hand, in a Serbian study, the average content of glucose, fructose, sucrose, and turanose reached 26.6 g/100 g, 40.6 g/100 g, 6.1 g/100 g, and 2.2 g/100 g respectively [49]. The glucose value in LH is therefore closer in Serbian lime honey. However, the differences in LH do not have to be caused only by the nectar of Lime tree, and we assume that the differences are mainly due to the different representation of honeydew in Lime honey, which is indicated by the presence of higher carbohydrates. In the case of Czech LH, it was mainly turanose (2.4 g/100 g), which is comparable to [49].

Honeydew honeys are characterized by different chemical and melissopalynological compositions, compared to blossom honey [45]. In Europe, honeydew honeys are from plant-sucking insects in different habitats. In particular, it includes *Abies alba*, *Abies cephalonica*, *Picea abies*, *Pinus* sp., *Quercus*, sp. and *Metcalfa pruinosa*, which is however insignificant for the Czech Republic [7]. Although honeydew honeys are a diverse group, a comparison of Czech and EU honeys showed a high relation (Figures 2 and 3, HD). The difference compared to EU honeys was in the parameter of F. Ac. On the other hand, F. Ac. comparable with the Czech HD is reported in Portuguese and Italian honeydew honey, namely 36.4 and 38.0 meq/kg [38,60].

The most different honey compared to EU honeys was RH. There was no overlap of data for RH, except for water content and F + G (Figures 2 and 3, RH). However, the water content does not characterize the honey: it rather describes maturity and beekeeping practices. Robinia honey is the honey of the eastern and south-eastern EU [7]. They are rather exceptional for the Czech Republic, both due to climatic conditions and their low incidence. Robinia honey is also characterized by the low content of pollen grains of this taxon, on average 28.1% [9]. In our study, the average pollen content of RH was 12.7%. We chose 10% of pollen as the threshold value for *Robinia pseudoacacia*. As a comparison with EU honeys shows, this extreme limit for classification was insufficient and RH honey has the character of rather a blossom honey. The presence of *Robinia pseudoacacia* pollen, however, was manifested in honeys, and these honeys were lighter than other FH and had lower HMF, lower diastase, higher E. cond., lower glucose, a higher F/G ratio, and higher sucrose values (Table 2). Based on this comparison, it cannot be said that RH honeys are indeed monofloral honeys. This result points to the importance of physico-chemical parameters for the species classification of honey, because in cases with a limited content of a specific pollen, the pollen analysis is not very specific.

4. Conclusions

Due to its botanical variability and climatic conditions, the Czech Republic is typical especially for FH and HD honeys. In certain localities and while maintaining suitable beekeeping practices, monofloral honeys can also be obtained. BH, LH, and HD honeys are identical to European honeys in most physico-chemical parameters. Czech BH was darker (73.3 Phund) than EU rapeseed honeys, and the average content of rapeseed pollen was 79.5%. RH honeys, which were selected as acacia honeys, based on having the highest proportion of *Rhobinia pseudoacacia* pollen (12.7%), did not correspond to the characteristic physico-chemical parameters and cannot be considered as acacia honeys. HD had the highest proportion of higher carbohydrates, primarily of Melezitose (4.8 g/100 g) and trehalose (1.3 g/100 g). The presence of higher carbohydrates was also confirmed in LH for Maltose (4.6 g/100 g) and TH for Turanose (2.4 g/100 g) and Melezitose (1.3 g/100 g). The average content of specific pollens was 22.8% for LH and 33% for TH. PH had physico-chemical parameters similar to polyfloral honeys, the content of specific pollen was 31.7% and the secondary pollen was Brassica pollen (24.1%). Polyfloral FH honeys had diverse physico-chemical parameters and pollen profile, such as Brassica 41.6% as the secondary pollen. A significant proportion of Brassica pollen was confirmed in majority of evaluated honeys, which is one of the common agricultural crops grown in the Czech Republic, and this pollen has been confirmed in most localities of the Czech Republic. All analysed honeys were characterized by low HMF content (3.0 mg/kg), sucrose (0.2 g/100 g),

and high diastase activity (24.4 DN). The physico-chemical properties and pollen profile of honeys from the Czech Republic are identical to those of EU honeys, although minor differences were demonstrated, which are due to the nature of the landscape and especially agricultural activities.

Supplementary Materials: The following are available online at <https://www.mdpi.com/article/10.3390/app11114989/s1>, Figure S1: Czech honey descriptors.

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