



# Macro-element ratios provide improved identification of the botanical origin of mono-floral honeys

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## Abstract

Major macronutrient concentrations (K, Ca, Mg, Na, P, and S) and element ratios were determined in 140 Hungarian mono-floral honey samples (acacia, linden, sunflower, rape, chestnut, forest, silk grass, and facelia) by inductively coupled plasma-optical emission spectrometry (ICP-OES). One-way ANOVA (LSD and Dunnett T3 test) and linear discriminant analysis (LDA) were used to determine the botanical origin based on the element content and element ratio of different honey types. Analysing six element concentrations in the honeys of different botanical origin with LDA allowed the botanical origin of 96% of honeys to be predicted. Reducing the examined elements to K, Mg, and Na increased the accuracy of predictions, but it was still not possible to distinguish acacia and facelia honeys. However, examination of element ratios showed that K/Na and K/Mg ratios were able to separate every honey type from each other with 100% cross-validation. It is concluded that using macro-element ratios rather than macro-element concentrations, it is possible to precisely discriminate the floral origin of honey samples.

**Keywords** Honey · Element · Element ratio · ICP · Botanical origin

## Introduction

Honey is a naturally sweet food produced by *Apis mellifera* bees, with an origin of nectar or honeydew, which has an important effect on its properties and in particular element content. Although the concentration of some elements (e.g., Ca, Mg, B, Cu, Zn, and Fe) is higher in nectar than honey, the Na, K, and P contents are similar [1]. Honey has a low total mineral content, about 1.0% in honeydew, and 0.1–0.2% in nectar honey, which depends on floral type of honey plants, floral density, soil properties, and treatment (e.g., centrifuging, filtration, and storage) [2]. In spite of the low mineral content, the determination of macro-, micro-, and trace elements can provide useful information about the botanical origin of honey, soil properties, and environmental

conditions of the collecting area [3]. In Hungary the annual honey production is about 20,000 tons and the most important honey types are acacia, linden, rape, chestnut, silk grass, and sunflower [4].

Several studies have attempted to investigate honey's botanical origin, based on contents of proline and total phenolic compounds [5, 6], vitamin B2 [7], viscosity [8], sugar composition [9], or combined parameters [10]. Recently, the determination of the element composition of different honey types has been suggested as a possible method to determine the floral origin of honey [11]. According to our previous examinations [12], the micro-element content of soil has significant effects on the micro-element concentration of honeys, especially in the case of Cu, Ba, Sr, Ni, Zn, Mn, Pb, and As. However, the macro-element concentrations of different honey types are influenced mainly by floral origin, and the element content of soil has only a very low effect [12]. In the literature, there are many studies about the element concentration of different honey types and the determination of botanical origin of honeys based on their element concentration. However, none of these studies examined element ratios of different honey types, which is important, because many elements interact and considering them in isolation ignores this. The aims of this study were to: (1) determine

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the macro-element content in mono-floral honey samples with a variety of floral origins; (2) test whether the botanical origin of the honeys can be determined from their macro-element content or the ratio of macro-elements.

## Materials and Methods

### Honey samples

140 mono-floral honey samples of 500 g each were collected from Hungarian beekeepers in sterile glass jars in 2016. The botanical origins of these samples were: acacia (*Robinia pseudoacacia*,  $n = 20$ ), linden (*Tilia* sp.,  $n = 20$ , also known as lime), sunflower (*Helianthus annuus*,  $n = 20$ ), rape (*Brassica* sp.,  $n = 20$ ), chestnut (*Castanea sativa*,  $n = 20$ ), silk grass (*Asclepias* sp.,  $n = 10$ ), facelia (*Phacelia tanacetifolia*,  $n = 10$ ), and forest ( $n = 20$ ) honeys. All honey samples were stored in the dark at room temperature until analysis. Melissopalynological analysis was carried out to verify the botanical origin of honeys using the method described by MSZ 6950-3:1977 (microscopic analysis of honey) [13]. Predominant pollen (> 50% of pollen grains counted) was determined.

### Preparation and measurement of samples

Digestion of samples was carried out based on the method of Kovács et al. [14], which has been validated using animal and plant materials. For this method nitric acid (69% v/v; VWR International Ltd., Radnor, USA), hydrogen peroxide (30% v/v; VWR International Ltd., Radnor, USA), and qualitative filter papers (Sartorius Stedim, Biotech S.A., Gottingen, Germany) were used. Ultrapure water produced by a Milli-Q water purification system (Millipore SAS, Molsheim, France) was used for dilution.

Determination of element concentration was carried out using Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES; Thermo Scientific iCAP 6300, Cambridge, UK) in the accredited laboratory (ISO/IEC 17,025:2005) of the Institute of Food Science, Debrecen in 2016. All analytical analyses were carried out in triplicate. The minerals were measured against a standard solution of known mineral concentrations. Operating parameters for the determination were the followings: power rating 1350 W, plasma gas flow rate 16 L/min, auxiliary and nebuliser gas flow rate 1.0 L/min, rinsing time 30 s, rinsing pump speed 75 rpm, and stabilization time 5 s. The emission wavelengths (nm) were as follows: Ca (317.9), K (766.4), Mg (279.5), Na (589.5), P (185.9), and S (182.0). Detection limits (DL) were determined for reagent blank samples ( $n = 12$ ) using the software for ICP-OES (iTEVA) at a confidence level of 99%. DLs were calculated from the following equation:

$LD = 3 \times SD_{\text{reagent blank}} / \text{sensitivity}$ . DLs (mg/kg) were as follows: 2.38 for Ca, 0.53 for K, 0.10 for Mg, 0.49 for Na, 0.49 for P, and 0.11 for S.

### Statistical analysis

Data were described using general terms (mean, standard deviation, minimum, maximum values), and one-way ANOVA with linear discriminant analysis (LDA). The LDA was carried out to confirm differences in element concentration of honeys with different botanical origin. Honey types were designated grouping variables and elements independent variables. Wilks' Lambda, or the ratio of within-groups sums of squares to the total sums of squares, was used to determine the proportion of the total variance in the discriminant scores not explained by differences among groups. Statistical significance was assumed at  $P$  value < 0.05. SPSS 13.0 statistical software program for Windows (SPSS Inc. Chicago, Illinois, USA) was utilised for all calculations.

## Results and discussion

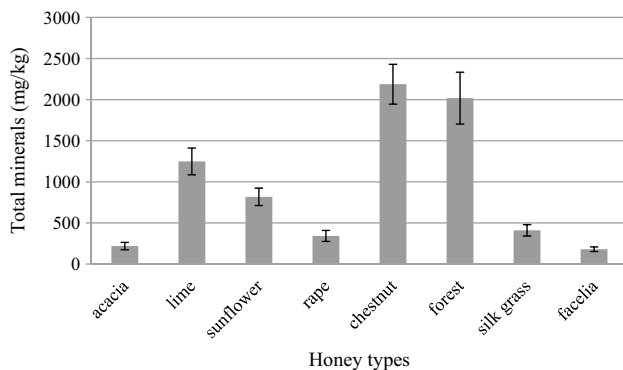
### Determination of botanical origin based on the element content of honey samples

Examining the mean concentration of all samples, the most abundant element in honeys was K, followed by P, Ca, S, Mg, and Na (Table 1). Even though the concentrations of the examined elements had a wide range, K had the highest concentration in all samples from the different botanical origins, and Na content the lowest. Some acacia honeys showed a similar Mg concentration to Na. According to the mean element concentrations, the order of examined elements in linden, sunflower, and chestnut honeys was  $Na < Mg < S < P < Ca < K$ ; in acacia, silk grass, facelia, and forest honeys was  $Na < Mg < Ca < S < P < K$ ; and in rape honey was  $Na < Mg < S < Ca < P < K$ . The highest Ca, K, Mg, Na, P, and S concentrations were determined in chestnut and forest honeys followed by linden and sunflower honeys.

Acacia and facelia honey samples showed very similar element concentrations, especially Ca, Na, P, and S concentrations; these two honey types showed the lowest total mineral contents (acacia  $219 \pm 45$  mg/kg and facelia  $181 \pm 28$  mg/kg). Examining the total mineral contents of different honey types (Fig. 1), chestnut ( $2188 \pm 242$  mg/kg), and forest ( $2019 \pm 315$  mg/kg) honeys were highest. Linden honeys also showed high total element content ( $1249 \pm 163$  mg/kg), followed by sunflower ( $818 \pm 106$  mg/kg), silk grass ( $411 \pm 69$  mg/kg), and rape ( $343 \pm 67$  mg/kg) honeys. Blizandik et al. [15] concurred with our results that chestnut honeys showed higher K, Ca and Mg concentration than linden honeys. Bontempo et al. [16] also

**Table 1** Macro-element content of examined mono-floral honeys, together with their standard deviation and range

Element (mg/kg)	Acacia (n=20)	Linden (n=20)	Sunflower (n=20)	Rape (n=20)	Chestnut (n=20)	Forest (n=20)	Silk grass (n=10)	Facelia (n=10)	Total (n=140)
Ca	13.1 ± 1.5 <sup>a</sup>	85.4 ± 7.3	105 ± 13	41.3 ± 5.1	99.6 ± 11.2	64.0 ± 8.5	20.0 ± 2.3	10.3 ± 1.2	60.5 ± 36.5
	10.2–15.5 <sup>b</sup>	71.8–98.3	82.9–124	33.4–50.6	81.9–116	52.4–75.5	17.8–24.1	9.12–12.5	9.12–124
K	137 ± 15	1056 ± 92	580 ± 75	212 ± 46	1918 ± 205	1714 ± 272	295 ± 27	114 ± 10	832 ± 706
	115–176	921–1280	502–735	162–292	1563–2186	1331–2212	262–342	102–130	102–2212
Mg	4.52 ± 0.41	20.1 ± 1.7	27.1 ± 3.7	13.9 ± 1.8	35.3 ± 3.6	50.1 ± 6.1	7.99 ± 0.72	4.49 ± 0.41	22.5 ± 15.6
	3.83–5.30	17.1–24.9	21.8–33.3	11.2–16.9	30.1–41.2	39.7–60.2	6.81–8.88	4.09–5.16	3.83–60.2
Na	3.65 ± 0.37	11.2 ± 0.9	7.82 ± 0.83	7.20 ± 0.98	12.8 ± 1.1	13.7 ± 2.1	5.14 ± 0.44	3.28 ± 0.29	8.64 ± 3.88
	3.13–4.62	10.1–13.9	6.37–9.32	6.13–9.09	10.8–14.6	10.8–17.9	4.51–5.92	3.02–3.81	3.02–17.9
P	38.1 ± 4.1	49.6 ± 4.3	69.3 ± 6.1	43.8 ± 6.5	80.2 ± 6.9	116 ± 10	57.9 ± 6.1	40.4 ± 3.6	63.7 ± 26.4
	32.0–46.2	43.1–58.1	60.3–79.9	33.3–53.9	71.1–92.2	102–138	50.4–67.4	35.4–47.2	32.0–138
S	14.1 ± 1.8	26.7 ± 4.4	28.9 ± 2.5	23.6 ± 3.4	40.4 ± 4.4	64.6 ± 9.0	23.8 ± 2.0	15.9 ± 1.5	31.2 ± 16.3
	12.1–17.2	21.5–34.5	24.8–33.9	20.0–30.9	33.8–48.1	53.5–79.8	20.3–26.4	13.7–18.1	12.1–79.8

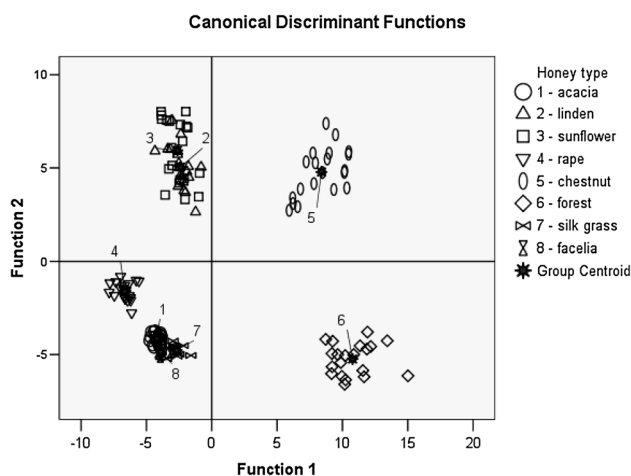
<sup>a</sup>Mean ± standard deviation<sup>b</sup>Minimum–maximum**Fig. 1** Total mineral content of the examined Hungarian honey types, with standard deviations

measured a wide range of elements in honeys from a range of botanical species in Italy (acacia, chestnut, citrus, eucalyptus, honeydew, rhododendron, and polyfloral). Chestnut honey had the highest concentration of K, Ba, Mn, and Rb and high Ca, Mg, and Sr values, and honeydew had the highest Mg, Cu, and Fe concentrations.

Chestnut honeys had higher total element concentrations than linden honeys, particularly in K, P, Mg, and S. Oroian et al. [11] found that K, Mg, and Ca were the predominant elements which were strongly associated with the principal components of acacia, linden, sunflower, and honeydew honeys, and were a suitable tool in predicting their botanical origin. However, Di Bella et al. [17] were unable to determine the botanical origin of honey from their analysis of 12 elements when they examined seven mono-floral honey types (including chestnut and acacia).

In our study, the examined honey types showed important differences in their element concentrations. Because most differences were significant, the non-significant results are presented by the Dunett T3 test (because the homogeneity of variance was  $<0.001$  in all examined elements). There were no statistically verified differences as follows: Ca concentration: sunflower and chestnut ( $P$  value = 0.991); K concentration: chestnut and forest ( $P$  value = 0.245); Mg concentration: acacia and facelia ( $P$  value = 1.000); Na concentration: acacia and facelia ( $P$  value = 0.133), sunflower and rape ( $P$  value = 0.587), chestnut and forest ( $P$  value = 0.904); P concentration: acacia and facelia ( $P$  value = 0.932), linden and rape ( $P$  value = 0.052), rape and facelia ( $P$  value = 0.804); S concentration: acacia and facelia ( $P$  value = 0.261), linden and sunflower ( $P$  value = 0.716), linden and rape ( $P$  value = 0.357), linden and silk grass ( $P$  value = 0.386), and rape and silk grass ( $P$  value = 1.000). In summary, statistically verified differences were not determined between acacia and facelia honeys in case of four examined elements (Na, P, S, and Mg). In addition, there was no difference between chestnut and forest honeys in K and Na contents, between linden and rape honeys in P and S concentrations. Based on these results, we concluded that the analyses of the examined element concentrations were not able to differentiate all of the examined honey types. Nevertheless, as more statistically verified differences than not verified differences were found among the different honey types in the examined element contents, to verify this, LDA analysis was carried out.

Initially, every examined element was involved in the statistical analysis (Fig. 2; Table 2). LDA determined six discriminant functions, with eigenvalues from 41.26 for the first to 0.007 for the last. The first function explained 97.6%,



**Fig. 2** LDA of mono-floral honeys examining K, Ca, Mg, Na, P, and S contents

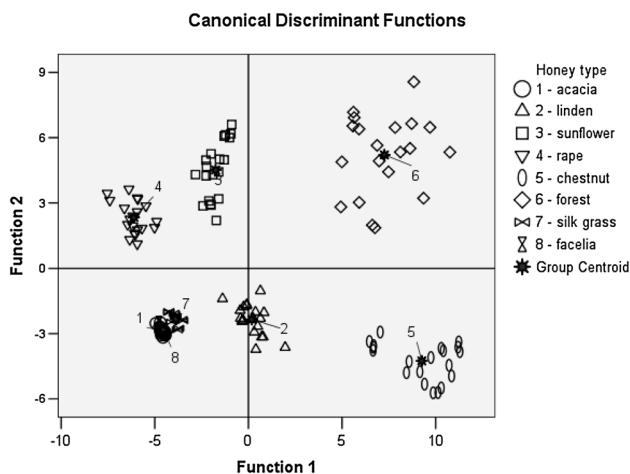
the second 95.8%, the third 94.3%, the fourth 77.3%, the fifth 26.7%, and the last only 0.72% of the variance of dependent variables. The first function contained K and Mg, the second one Ca, the fourth one Na, the fifth one P, and the last one S. In the first dimension, the chestnut (8.39) and forest (11.8) honeys showed the highest centroids; however, in the second dimension, these values were varied (4.79 and  $-5.24$ ). Centroids of linden and sunflower honeys showed the highest values in the second dimension (6.06 and 5.95), which were very similar, and there were no important differences between these two centroids in the first dimension ( $-2.38$  and  $-2.63$ ). Rape honeys showed the lowest centroids in the first dimension ( $-6.67$ ) and in the second dimension, this value was also low ( $-1.59$ ). Acacia, silk grass, and facelia honeys showed similar centroids in the second dimensions ( $-4.13$  for acacia,  $-4.77$  for silk grass, and  $-4.89$

for facelia honeys). In the other dimensions, the differences among these centroids were higher ( $-4.20$  for acacia,  $-2.70$  for silk grass and  $-3.88$  for facelia honeys). According to the cross-validation, in the case of acacia honeys, the number of correctly categorized cases was 17 (85%), with three samples in the group of facelia honeys. In the case of facelia honeys, this number was three (80%), because two samples were assigned to the acacia honey group. Overall 96.4% of cross-validated grouped cases were correctly classified. Based on these results, these six examined elements were not able to separate each honey type exactly, with difficulties in discriminating linden and sunflower, as well as acacia, silk grass, and facelia honey samples.

Then, the element number was reduced and only K, Mg, and Na were examined in the next step (Table 2). The grouping variables were the honey types and the independent variables were K, Mg, and Na. Three discriminant functions were determined and based on Wilks' Lambda, K had the highest effect, followed by Mg and Na. All three variables were significant. The first discriminant function explained 97.0% of the variance of variables, the second one explained 93.3%, and the third one explained 76.2%. The first discriminant function involved K and Mg and the third one involved Na. The scatter plot (Fig. 3) shows six separated groups, so linden, rape, chestnut, forest, and sunflower honeys were separated from each other and from the group of acacia, silk grass, and facelia honeys. In the first dimension, the highest centroids were determined in chestnut (9.26) and forest (7.26) honeys, followed by linden (0.19), sunflower ( $-1.76$ ), silk grass ( $-3.89$ ), facelia ( $-4.55$ ), acacia ( $-4.61$ ), and rape ( $-6.11$ ) honeys. In the second dimension, the highest centroids were determined in forest (5.21) and sunflower (4.52) honeys, followed by rape (2.33), silk grass ( $-2.32$ ), linden ( $-2.35$ ), facelia ( $-2.82$ ), acacia ( $-2.88$ ), and chestnut ( $-4.29$ ) honeys. Acacia, silk grass, and facelia

**Table 2** Statistical analysis of the examined elements

Analysis	Test of equality of group means			Eigenvalues			Wilks' Lambda		
	Elements and ratios	Wilks' Lambda 1	Sig	Functions	Eigenvalues	Canonical correlations	Test of functions	Wilks' Lambda 2	Sig
No.1. (Fig. 1)	K	0.036	0.000	1	41.3	0.988	1 through 7	0.000	0.000
	Na	0.077	0.000	2	23.4	0.979	2 through 7	0.000	0.000
	Ca	0.047	0.000	3	16.3	0.971	3 through 7	0.010	0.000
	P	0.057	0.000	4	3.39	0.879	4 through 7	0.166	0.000
	S	0.073	0.000	5	0.364	0.517	5 through 7	0.728	0.000
	Mg	0.040	0.000	6	0.007	0.085	6 through 7	0.993	0.000
No.2. (Fig. 2)	K	0.036	0.000	1	33.0	0.985	1 through 3	0.000	0.000
	Na	0.077	0.000	2	13.9	0.966	2 through 3	0.016	0.000
	Mg	0.040	0.000	3	3.21	0.873	3	0.238	0.000
No.3. (Fig. 3)	K/Na	0.012	0.000	1	85.6	0.994	1 through 2	0.001	0.000
	K/Mg	0.029	0.000	2	14.9	0.968	2	0.063	0.000



**Fig. 3** LDA of mono-floral honeys examining K, Mg, and Na contents

honeys showed very similar centroids in both dimensions. Examining six elements, the linden and sunflower honeys showed very similar centroids in both dimensions. However, examining only these three elements, these two honey groups were separated from each other, mainly in the second dimension. The cross-validation method was applied to verify the results. Based on the independent variables, 100% of original grouped cases were correctly categorized into linden, sunflower, rape, chestnut, forest, and silk grass honeys. However, in the case of acacia honeys, the number of correctly categorized cases was only 11 (55%), because one sample was assigned to silk grass and eight samples to

facelia honey groups. In case of facelia honeys, the number was 8 (20%), because two samples were moved into the acacia honey group. In summary, the analysis of three elements showed better results than the examination of six elements, because linden and sunflower honeys were clearly separated from each other. However, the examination of these three element concentrations was not satisfactory because of the inability to discriminate acacia and facelia honeys.

### Determination of botanical origin based on element content ratios of honey samples

Examining the element ratios, the values of K/Na were the highest, followed by K/Mg in all examined honey types (Table 3). The third highest ratio value was K/S in linden, sunflower, rape, chestnut, and forest honeys; K/Ca in acacia; and P/Na in facelia honeys. The lowest element ratio value was the P/Ca in linden, sunflower, rape, and chestnut honeys, and the Ca/S in acacia, forest, silk grass and facelia honeys. The highest K/Na ratio was determined in chestnut honeys and the highest K/Ca value was calculated in forest honeys. The highest K/P, K/S, and K/Mg ratio values were determined in linden and chestnut honeys. Linden and sunflower honeys showed the highest Ca/S and Ca/Mg ratio values. The highest P/S, P/Mg, and S/Mg ratio values were calculated in acacia and facelia honeys, the highest Ca/Na ratio values in sunflower honeys, the highest P/Ca ratio values in facelia honeys, the highest P/Na ratio values in silk grass and facelia honeys, and the highest S/Na ratio values in forest, silk grass, and facelia honeys.

**Table 3** Range of element ratios of examined mono-floral honeys

Element ratio	Acacia (n=20)	Linden (n=20)	Sunflower (n=20)	Rape (n=20)	Chestnut (n=20)	Forest (n=20)	Silk grass (n=10)	Facelia (n=10)
K/Na	36.3–38.7 <sup>a</sup>	82.2–109	63.8–79.4	24.0–38.9	140–160	121–134	51.9–63.3	33.3–37.5
K/Mg	28.0–33.9	49.0–59.1	20.0–23.5	12.0–17.7	50.3–58.8	28.8–39.9	34.4–38.9	23.4–26.7
K/S	9.00–11.2	32.5–46.1	17.6–22.8	7.89–10.9	42.4–55.3	23.0–31.9	10.9–13.0	6.71–7.66
K/Ca	9.01–12.8	10.3–14.0	4.35–7.42	4.04–6.51	18.2–22.4	23.1–31.6	13.1–16.5	10.0–12.6
K/P	3.13–4.49	18.8–23.0	7.34–9.54	4.00–5.93	20.5–29.2	11.7–19.2	4.68–5.74	2.48–3.02
Ca/Na	3.02–4.13	6.45–9.73	10.5–18.1	3.95–7.85	6.97–8.70	3.94–5.45	3.24–4.45	2.81–3.61
Ca/Mg	2.40–3.57	3.52–5.75	3.09–5.04	2.30–3.42	2.61–3.07	1.09–1.55	2.20–2.88	1.98–2.62
Ca/S	0.72–1.12	2.35–4.37	2.74–4.56	1.42–2.07	2.23–2.95	0.85–1.15	0.70–0.97	0.55–0.74
P/Na	8.48–11.9	3.94–4.97	7.65–10.5	4.74–7.10	5.22–7.50	6.48–10.6	9.81–13.3	11.3–15.1
P/S	2.21–3.14	1.49–2.28	2.12–2.72	1.43–2.12	1.65–2.21	1.40–2.45	2.07–2.73	2.36–3.03
P/Ca	2.42–3.48	0.45–0.68	0.55–0.86	0.77–1.28	0.64–0.94	1.48–2.36	2.51–3.36	3.45–4.47
P/Mg	7.29–10.3	2.26–2.75	2.13–3.16	2.59–3.89	1.88–2.57	1.86–3.08	6.47–8.03	8.35–10.8
S/Na	3.25–4.25	1.97–3.09	3.23–4.51	2.23–4.43	2.78–3.68	3.91–5.63	4.13–4.94	4.39–5.11
S/Mg	2.69–3.62	1.10–1.72	0.89–1.30	1.40–2.04	1.00–1.25	1.09–1.49	2.81–3.26	3.33–3.67
Mg/Na	1.12–1.36	1.58–2.06	3.08–3.88	1.56–2.45	2.46–3.12	3.12–4.38	1.46–1.70	1.31–1.44

<sup>a</sup>Minimum–maximum

In the one-way ANOVA, a LSD test was carried out in case of S/Na and P/S ratio, because the variances were homogenous, and in the case of other element ratios, the Dunnett T3 Test was applied because of the heterogeneity of variances. The K/Na ratio showed a significant difference for every examined honey types. The K/P ratio was also statistically verified for all examined honey types, except between rape and silk grass honeys. Significant differences were determined in case of K/S ratio (except between acacia and rape honeys), P/Ca ratio (except between acacia and silk grass honeys), K/Mg ratio (except between linden and chestnut; between linden and silkgrass), Ca/Na ratio (except between acacia and silk grass and between linden and chestnut), and Mg/Na ratio (except between linden and rape and between sunflower and forest). There were no significant differences in the following element ratios: K/Ca (acacia and facelia; linden and facelia; sunflower and facelia), P/Na (acacia and silk grass; sunflower and forest; rape and forest; silk grass and facelia), Ca/S (acacia and forest; acacia and silk grass; linden and sunflower), Ca/Mg (acacia and rape; acacia and chestnut; linden and sunflower; silk grass and facelia), P/Mg (acacia and facelia; linden and sunflower; linden and forest; sunflower and forest; chestnut and forest), S/Mg (acacia and silk grass; linden and forest; sunflower and chestnut), and S/Na (acacia and sunflower; rape and chestnut; forest and silk grass; forest and facelia; silk grass and facelia). Based on these results, element ratios were selected which showed the most statistically verified differences among the examined honey types: K/Na, K/P, K/S, K/Mg, Ca/Na, and P/Ca. Using these element ratios, LDAs were carried out and we determined that K/Na and K/Mg element ratio could be the suitable for the separation of the examined honey types. The result of this analysis is shown in Fig. 4 and Table 2.

The grouping variables were the honey types and the independent variables were the K/Na and K/Mg element ratios. Two discriminant functions were determined. The eigenvalue of the first function was very high (85.6) and it explained 98.8% of the variance. The second function showed much lower eigenvalue (14.8); however, it explained 93.7% of the variance. According to Wilks' Lambda, the effect of the first function was higher (0.001) than the second one (0.063), but both functions were significant ( $P$  value < 0.001). The first function involved K/Na and the second one involved K/Mg element ratio. Overall, the first function had a more important role based on its eigenvalue and variance. The scatter plot (Fig. 4) shows seven absolutely separated groups from each other. In the first dimension, the highest centroids were determined in case of chestnut honeys (15.1), followed by forest (8.16), linden (5.00), sunflower (−2.57), silk grass (−3.60), acacia (−8.00), facelia (−9.07), and rape (−11.3) honeys. In the second dimension, the highest centroid values were calculated in case of linden honeys (5.79) followed by silk grass (3.43), acacia (2.92), facelia

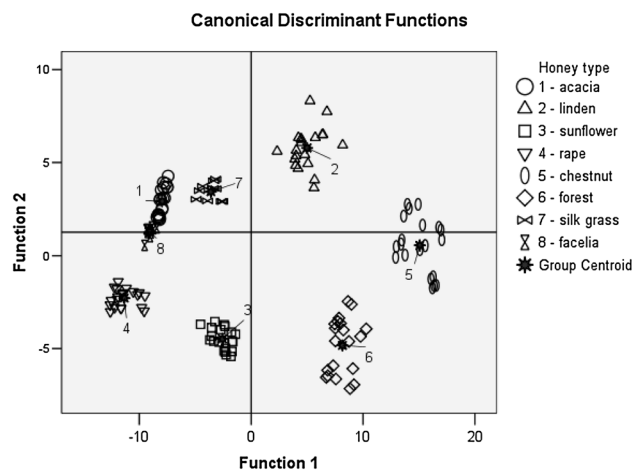


Fig. 4 LDA of mono-floral honeys examining K/Mg and K/Na ratio

(1.26), chestnut (0.544), rape (−2.28), sunflower (−4.48), and forest (−4.83) honeys. In the first dimension, sunflower and silk grass honeys showed very similar centroids; however, in the second dimension, the result was very different. Acacia and facelia honeys also showed very similar centroids in the first dimension; however, in the second, the distance between these centroids was higher. In this second dimension, similar centroids were determined in sunflower and forest honeys, as well as acacia and silk grass honeys; however, in the first dimension, these groups were clearly separated from each other. According to the cross-validation, 100% of original and cross-validated grouped cases were correctly classified. Therefore, our hypothesis was verified, because the separation of acacia, silk grass, and facelia honeys from each other was more successful than in case of analysing individual macro-elements.

Thus, the examination of the element concentration of honeys with different botanical origin can determine most honey types; however, the analysis of element ratios can improve the results.

## Conclusions

In our study, six macro-elements (Ca, K, Mg, Na, P, and S) in 140 Hungarian honey samples with different floral origin (acacia, linden, sunflower, rape, chestnut forest, silk grass, and facelia) were examined. K was the most abundant element in these honey types followed by P, Ca, S, Mg, and Na. Differences in the element concentrations in most cases were significant between floral types. When all six elements were compared between floral types, it was not possible to separate all honey types. Reducing the element number to three (K, Na, and Mg) improved the separation; however, acacia and facelia honeys could not be effectively separated.

When ratios between elements were examined by the LSD test, which differed significantly according to floral origin of the honey, K/Na and K/Mg ratios were able to distinguish the honey type with 100% accuracy. Analysing these two element ratios, therefore, achieved complete separation of the honey types according to their floral origin.

### Compliance with ethical standards

**Conflict of interest** There is no conflict of interest.

**Compliance with ethics requirements** There has not been any research involving human or animal participants performed.

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