Honey Bees and Their Products as Indicators of Environmental Element Deposition

Jelena Ćirić [1] · Danka Spirić [1] · Tatjana Baltić [1] · Ivana Branković Lazić [1] · Dejana Trbović [1] · Nenad Parunović [1] · Radivoj Petronijević [1] · Vesna Đorđević [1]

Abstract

The aim of this study was to evaluate element (sodium, magnesium, potassium, calcium, chromium, manganese, iron, cobalt, nickel, copper, zinc, arsenic, cadmium, and lead) deposition in honey bees (*Apis mellifera* L.) (worker bees, drone bees, and bee broods) and their products (wax and multifloral honey) in the central and north parts of Serbia using inductively coupled plasma mass spectrometry (ICP-MS). The study was carried out during the spring and summer seasons when the honeybees were active (2019). Fifty-four colonies of honey bees from different apiaries (located in Rudnik, Lazarevac, and Ležimir) were used in this study. Significant differences in element concentrations were found among locations ($P < 0.05$). The highest deposition of elements (sodium, calcium, manganese, iron, cobalt, nickel, zinc, arsenic, and cadmium) was found in worker bees. The most commonly detected elements in wax were chromium and lead. Mg had a significant ($P < 0.01$) correlation with K, Fe, Cu, and As. This study shows that honey bees (worker bees, drone bees, and bee broods) could be more useful as bioindicators of environmental element deposition (toxic and non-toxic) than multifloral honey. Also, this study shows that Serbian multifloral honey meets safety criteria concerning the concentrations of toxic elements.

Keywords Honey · Bee products · Beeswax · Environment pollution

Introduction

Element (toxic and non-toxic) deposition in soil and plants is one of the most pressing concerns in the science of food safety and food quality in Europe [1]. Many studies [2–6] show the impact of different metals/elements on human health as well as the mechanisms of uptake and bioaccumulation of heavy metals by plants and other foods for human nutrition. Some of these elements (lead, cadmium, mercury, and arsenic) are considered potential carcinogens and are associated with the etiology of a number of internal diseases [7]. On the other hand, some elements, such as Zn and Cu, are essential elements for human health [8].

Honey bees and their products can be contaminated with elements from different sources [9–11]. The origin of contamination can be the environment (air, water, plants, and soil), beekeeping practices [12], and then can be the bees themselves, which introduce contaminants into the beehive [13]. Also, the determination of elements in environmental samples including honey bees and their products is an important part of environmental pollution studies [13–16]. One of the most commonly examined honey bee products, due to its potential risk to human health, is honey (which can be floral-specific, e.g., acacia, linden, blossom) [17–21]. On the other hand, previous [10, 22] showed that honey bees can be better indicators of environmental element deposition than honey. This can be explained due to elements from the atmosphere, which firstly can deposit on the honey bees' bodies, and then can be introduced into the beehive.

Thus, information about element concentrations in honeys and their potential dietary intake via this food is very important for assessing the risk to human health. During harvest time, bees are exposed to different sources of contaminants through their pollen and nectar that contains heavy metals of natural and/or anthropogenic origin [9]. Some toxic elements are bioaccumulative, and these can be measured in honey bee and different bee products (honey, wax, propolis, etc.). In this study, a detailed analysis of environmental element deposition in honey bees and their products is reported. The analysis

✉ Jelena Ćirić
 1310jecko@gmail.com

1 Institute of Meat Hygiene and Technology,
 11000 Belgrade, Republic of Serbia

covers the different toxic and non-toxic elements (As, Cd, Cr, Cu, Pb, Zn, Co, Ni, Fe, Zn, Mn, K, Na, Ca, and Mg). The aim of this study was to determine potential correlations between element concentrations in honey bees and their products.

Materials and Methods

Sample Collection

This study was performed in 2019, in three different locations in Serbia: central–Rudnik mountain (coordinates: 44° 07′ 53″ N 20° 32′ 25″ E), Lazarevac (coordinates: 44° 22′ N 20° 15′ E), and Ležimir in northern Serbia (coordinates: 45° 07′ N 19° 34′ E). Site I (Rudnik mountain) is mostly composed of wild areas, while site II (Lazarevac) is surrounded by industrial environments and site III (Ležimir) is near agricultural environments (Fig. 1). The study was conducted from April to July for each sampling site; live honeybees (worker bees, drone bees, and bee broods) were sampled five times. Honey bee samples were prepared according to Nuorteva [23]. Honeys (multifloral, 500 g) from each location were stored in glass containers at 4–8 °C until analysis. Sub-samples of wax (~ 4 g) were taken directly from freshly formed combs using sterile beekeeping knives and put in sterile plastic boxes.

Element Concentration Analyses

For analyses, approximately 0.5 g of homogenized sample was transferred into a Teflon vessel with 5 ml nitric acid (67% Trace Metal Grade, Fisher Scientific, Loughborough, UK) and 1.5 ml hydrogen peroxide (30% analytical grade, Sigma-Aldrich, St. Louis, MO, USA) for microwave digestion. The microwave (Start D, Milestone, Sorisole, Italy) program consisted of three steps: 5 min from RT to 180 °C, 10 min hold at 180 °C, and 20 min cooling. After cooling, the digested samples were quantitatively transferred into disposable flasks and diluted up to 100 ml with deionized water produced by a water purification system (Purelab DV35, ELGA, High Wycombe, Buckinghamshire, UK). Analysis of the elements (sodium, magnesium, potassium, calcium, chromium, manganese, iron, cobalt, nickel, copper, zinc, arsenic, cadmium, and lead) was performed by inductively coupled plasma mass spectrometry (ICP-MS) (iCap Q mass spectrometer, Thermo Scientific, Bremen, Germany).

Adjustment of physical and electronic parameters was performed before determining the elements, using calibration solution (Thermo Scientific Tune B). The calibration curve consisted of five points in two ranges (including zero). Multielemental internal standard was introduced into the ICP-MS during the measurements. Data analysis software automatically made corrections comparing internal standards.

Fig. 1 Map of sampling

Quality control was performed using certified reference material (CRM) NIST SRM 1577c (Table 1).

Statistical Analysis

Statistical analysis was performed using the GraphPad Prism version 7.00 software. The concentrations of elements in honey bees and their products were expressed as mean and standard deviation (SD) and were subjected to analysis of variance (one-way ANOVA). The parameters were analyzed using the Student's t test at the probability of 0.05. Pearson's correlation analysis was applied to examine the relationship between element concentrations ($P < 0.01$ and $P < 0.05$).

Principal Component Analysis

The results of principal component analysis were used for presentation of the differences/similarities in the metal content of the bees, drones, broods, and products. The varimax method was performed for data rotation. For principal component analysis (PCA) analysis, determination statistic software JMP 10 (SAS Institute Inc., Cary, NC, USA) was employed.

Results and Discussion

Apiculture is very popular in Serbia [24]. According to the Statistical Yearbook of the Republic of Serbia [25], the number of beehives exceeds 840,000, with total honey production of 2263 tons and 15 kg of honey per beehive (in 2015). The mean concentrations of fourteen elements measured in honey from the three different regions are presented in Fig. 2. Concentrations of elements were significantly different between locations ($P < 0.05$). The honey from Ležimir (III) had higher Na, Mg, K, and Ca (6.15 ± 0.34 mg kg^{-1}, 38.45 ± 2.02 mg kg^{-1}, 2824 ± 44.96 mg kg^{-1}, and 1189.70 ± 5.44, respectively) concentrations compared with honey from the two other locations. Potassium was the most abundant element in honey, in agreement with previous studies [9, 17, 26]. According to Alqarni et al. [26], these elements promote fundamental functions in biological systems. The element concentrations in honey are related to the floral source and can vary widely among honeys of different origins and locations [27, 28]. Co was not detected in any of our honeys. Similar, Vukašinović-Pešić et al. [29] examined honey samples from eight different micro-regions of Montenegro, and the results confirm that, with the exception of cadmium in samples from two regions exposed to industrial pollution, none of the 12 elements (Pb, Cd, Cu, Zn, Fe, Cr, Sr, Ba, Ca, Na, K, Mg) analyzed exceeded the maximum allowable level. In Croatia, among the multifloral honeys, the following was determined (Fe, Na, Cu: mg kg^{-1}; As, Pb: μg kg^{-1}): orchard honey—highest of Fe 5.17 and As 276.1, lowest Pb 301; meadow honey—highest Na 36.1, lowest Cu 4.38. The average Ca, Cu, and Pb levels found in multifloral honey types were much higher than those reported in other European countries [30].

The toxic metal concentrations were compared with those established by the European Union [31] and Serbian legislation [32]. The As concentration ranged between $1.13 \pm$

Table 1 Quality control of element analysis

Elements	Limit of detection (μg kg^{-1})	Limit of quantification (μg kg^{-1})	Method repeatability/precision as RSD (%)	Certified value (μg kg^{-1})[a]	Analyzed value (μg kg^{-1})[b]
As	1.2	4	3.57	19.3 ± 1.4	20.5 ± 1.1
Cd	0.4	1	8.99	97 ± 1.4	97.9 ± 2.6
Pb	2	3.8	3.65	62.8 ± 1.0	63.3 ± 2.6
	(mg kg^{-1})	(mg kg^{-1})		(mg kg^{-1})	(mg kg^{-1})
Cu	0.022	0.066	6.26	275.2 ± 4.6	271.9 ± 5.7
Fe	0.08	0.23	4.71	197.94 ± 0.65	197.43 ± 5.21
Zn	0.124	0.372	10.52	181.1 ± 1.0	180.9 ± 1.8
Mn	0.004	0.011	4.47	10.46 ± 0.47	10.55 ± 0.25
Cr	0.002	0.007	4.47	53 ± 14	51 ± 2.8
Co	0.004	0.013	4.24	0.3 ± 0.018	0.31 ± 0.016
Ni	0.050	0.145	9.19	44.5 ± 9.2	52.7 ± 4.3
K	2.21	6.63	2.85	1023 ± 640	1054 ± 300
Na	5.58	16.76	7.02	2033 ± 64	2011 ± 140
Ca	3.08	9.24	3.64	131 ± 10	125 ± 4
Mg	0.13	0.40	3.03	620 ± 42	631 ± 19

[a] Certified value as given by the manufacturer

[b] Data are mean ± standard deviation

Fig. 2 Element concentrations in honeys from three different regions (mean); concentrations of elements were significantly different between locations (P < 0.05)


Honey
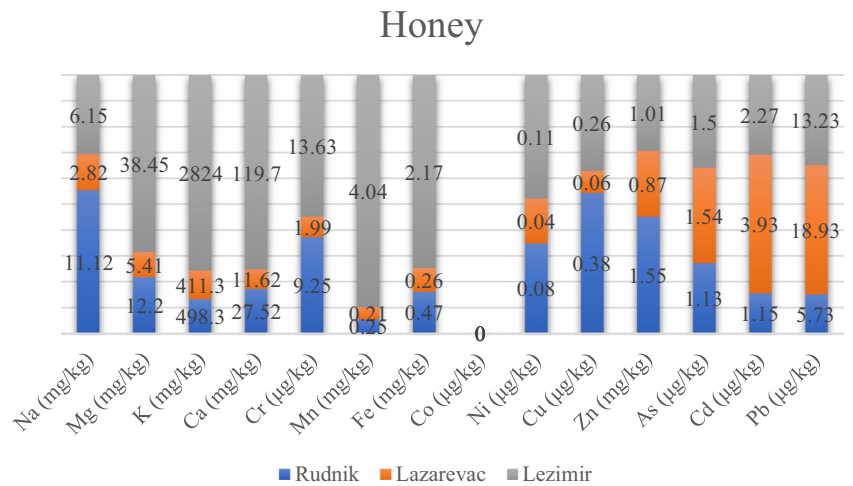
0.12 μg/kg (location I) and 1.54 ± 0.27 μg/kg (location II). This difference was significant (P < 0.05) and can be explained by the nature of the different locations; in fact, location I is mostly wilder areas, while location II (Lazarevac) is surrounded by industrial environments. The mean As concentration in honeys was different in previous studies in Serbia [17] and Italy [19] (5.20 μg/kg, 6.96 ± 2.71 μg/kg, respectively). According to Serbian legislation, the maximum residue limit for As is 0.500 mg kg^{-1}, and all tested samples of honey met this legislation. The highest Pb and Cd concentrations were in honey from location II (18.93 ± 1.24 μg/kg and 3.93 ± 0.63 μg/kg, respectively). Previous studies show that Pb contamination of honey in polluted and non-polluted areas is not significantly different [13, 33]. However, the highest Pb values were often found in honey from polluted areas. Similar results were presented in the study by Spirić et al. [17]. The contamination levels of the toxic elements Pb and Cd measured in our study were low, and the honeys were safe, according to EU Regulation (The MRL—maximum residue limit; proposal for EU is at 0.1 mg kg^{-1} for cadmium and 0.10 mg kg^{-1} for lead).

Element concentrations in waxes from the three different locations are presented in Fig. 3. The concentrations of elements were significantly different between locations (P < 0.05). The wax collected from all three locations (I, II, and III) contained, from the most abundant down, the highest level of K (238.70 ± 46.24 mg kg^{-1}, 693.40 ± 28.41 mg kg^{-1}, and 1572 ± 178.40 mg kg^{-1}, respectively), Pb (29.25 ± 3.82 μg/kg, 111.80 ± 6.73 μg/kg, and 401.10 ± 16.02 μg/kg, respectively), Ca (36.96 ± 5.69 mg kg^{-1}, 212.70 ± 22.71 mg kg^{-1}, and 396.70 ± 21.18 mg kg^{-1}, respectively), and Cr (131.20 ± 39.34 μg/kg, 85.77 ± 4.02 μg/kg, and 247.60 ± 33.59 μg/kg, respectively). Wax, as important honey bee product, was in the scope of large scientific studies as a potential source of heavy metal deposition [34–37].

Generally, the relatively large amounts of trace elements (K, Ca, Pb, and Cr) in wax could be due to atmospheric pollutant deposition on plants and flowers, which might be

Fig. 3 Element concentrations in wax from three different regions (mean); concentrations of elements were significantly different between locations (P < 0.05)

Wax
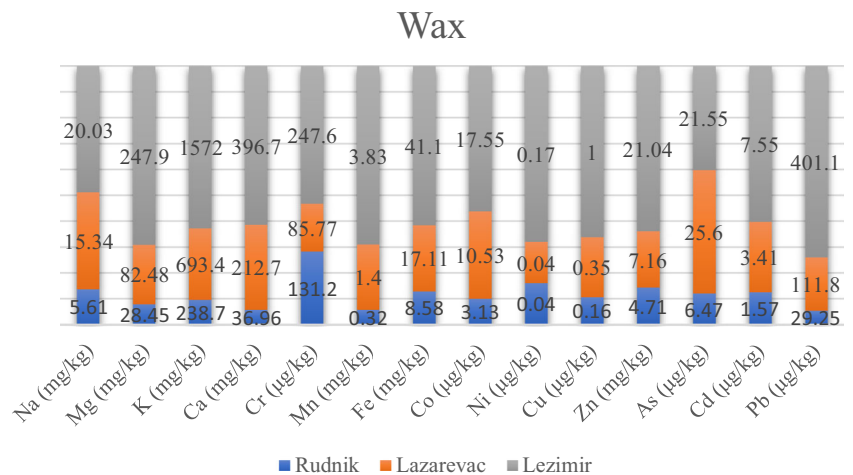
Fig. 4 Element concentrations in worker bees from different regions (mean); concentrations of elements were significantly different between locations ($P < 0.05$)
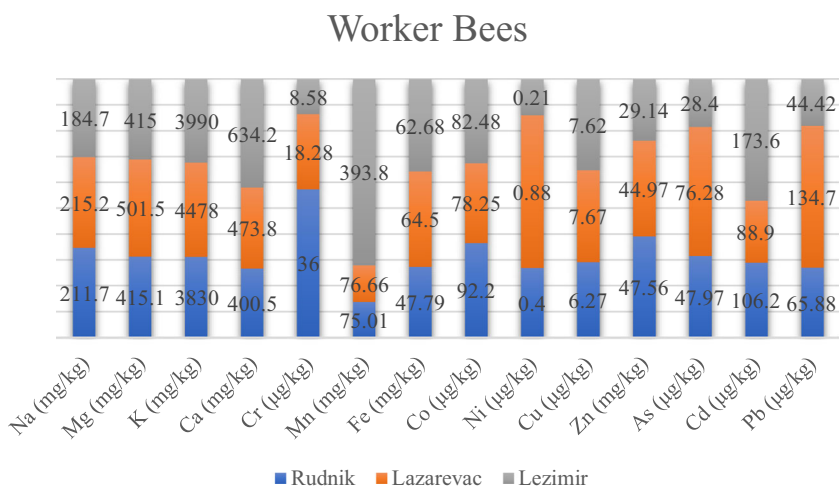
absorbed together with flower nectar [34, 38]. It should also be noted that in most of our studied apiaries, higher Cd, Pb, Mg, K, and Ni concentrations were found in wax than in honey, with the general highest mean concentration in wax from Ležimir (location III). Similar results were present in a study by Formicki et al. [35].

One-way ANOVA indicated significant differences ($P < 0.05$) in element concentrations in the honey bees (worker bees, drone bees, and bee broods) (Figs. 4, 5, and 6).

Honey bees fly intensively in a radius of up to 7 km [13, 17]. For this reason, they are suitable to be used as bioindicators for environmental element deposition [33, 36, 38, 39]. In the present study, element concentrations in honey bees were affected by location. The high concentrations of Na, Mg, K, Fe, Ni, Cu, As, and Pb (215.20 ± 1.13 mg kg^{-1}, 501.50 ± 1.38 mg kg^{-1}, 4478 ± 103.10 mg kg^{-1}, 64.50 ± 0.16 mg kg^{-1}, 0.88 ± 0.01 μg/kg, 7.67 ± 0.12 μg/kg, 76.28 ± 1.61 μg/kg, 134.70 ± 0.52 μg/kg, respectively) were found in worker bees collected in the apiary from Lazarevac (location
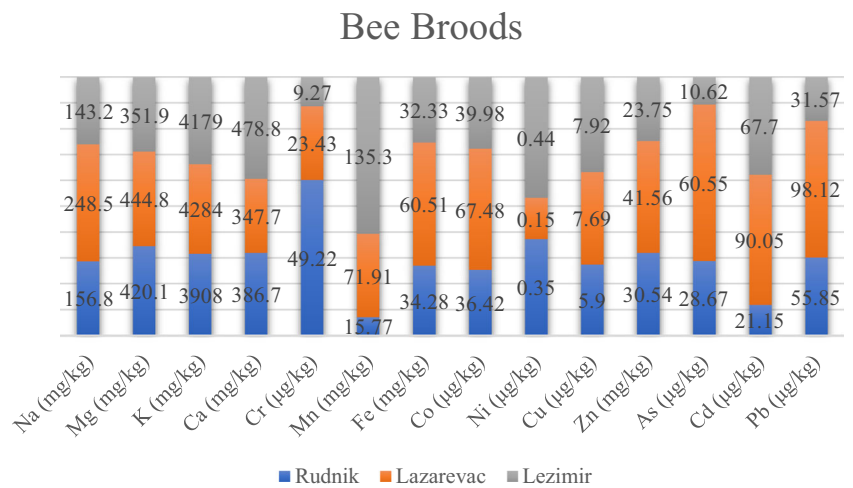
II). The results of the present study reinforce the idea that high concentrations of different elements (toxic) are reflected in the worker bees due to local environmental exposure (as for our Lazarevac bees, surrounded by industrial environments).

The concentrations of elements in drone bees were similar to those in worker bees. The high concentrations of Ca, Mn, Fe, Co, Ni, Cu, Zn, As, Cd, and Pb in drone bees ranged between 168.50 ± 1.38 and 377.20 ± 7.19 mg kg^{-1}, 4.30 ± 0.22 and 52.35 ± 0.82 mg kg^{-1}, 18.61 ± 1.57 and 54.26 ± 0.95 mg kg^{-1}, 6.50 ± 0.09 and 70.23 ± 3.11 μg/kg, 0.04 ± 0.01 and 0.52 ± 0.14 μg/kg, 5.93 ± 0.09 and 7.41 ± 0.16 μg/kg, 24.15 ± 2.38 and 42.46 mg kg^{-1}, 5.65 ± 0.24 and 73.63 ± 0.50 μg/kg, 5.12 ± 0.38 and 106.30 ± 1.48 μg/kg, and 9.12 ± 0.26 and 68.07 ± 1.35 μg/kg, respectively. Similarly, Conti and Botre [36] found the concentrations of Cd and Pb in honey worker bees ranged between 2.87 and 4.23 ppm for Cd and 0.61 and 1.25 ppm for Pb. In our study, these differences may be attributed to the varying degrees of heavy metal contaminations at each location.

Fig. 5 Element concentrations in drone bees from different regions (mean); concentrations of elements were significantly different between locations ($P < 0.05$)
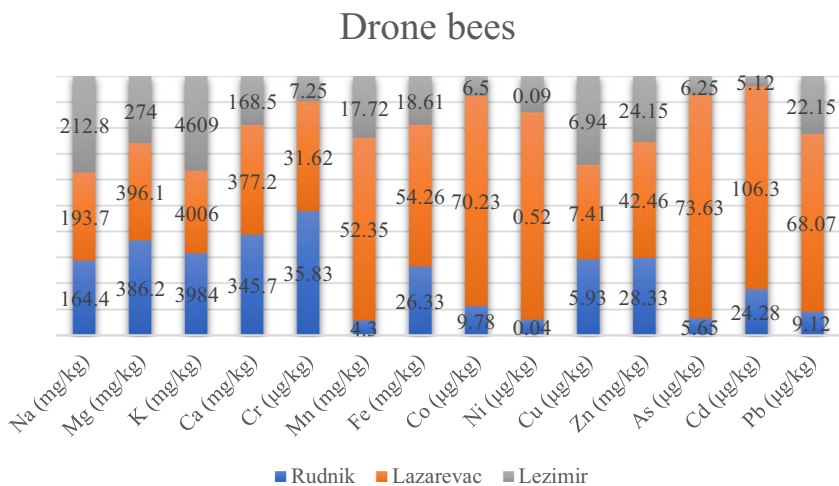
Fig. 6 Element concentrations in tested bee broods from different regions (mean); concentrations of elements were significantly different between locations (*P* < 0.05)


According to the results obtained in the present study (Fig. 6), some of the Cd, Pb, As, Zn, Co, and Fe concentrations in bee broods collected from apiaries were similar to concentrations of these elements in worker bees and drone bees.

PCA components 1 and 2 explain 80.5% variability of data, cumulatively (Fig. 7). Principal component 1 (PC1) explained 64.4% of variability with corresponding eigenvalue of 9.01, and PC2 explained 16.1% with eigenvalue of 2.25. As it can be seen, data were separated in two distinctive groups: products on the one side and bees, drones and broods on the other.

Grouping was mainly established by component 1 based on the differences in content of As, Fe, Ca, Ni, Zn, Co, Mg, Cd, Na, Cu, K, and Mn. The content of these metals was found higher in bees and broods. Higher amounts of Cr and Pb were found only in wax samples originated from Ležimir.

Many previous studies found correlations between element concentrations in honey and/or in their products [34, 35]. Table 2 shows the correlations between element concentrations in the studied bees and their products. Mg had a significant (*P* < 0.01) positive correlation with K (*r* = 0.909). Also, Mg

Fig. 7 Principal component analysis
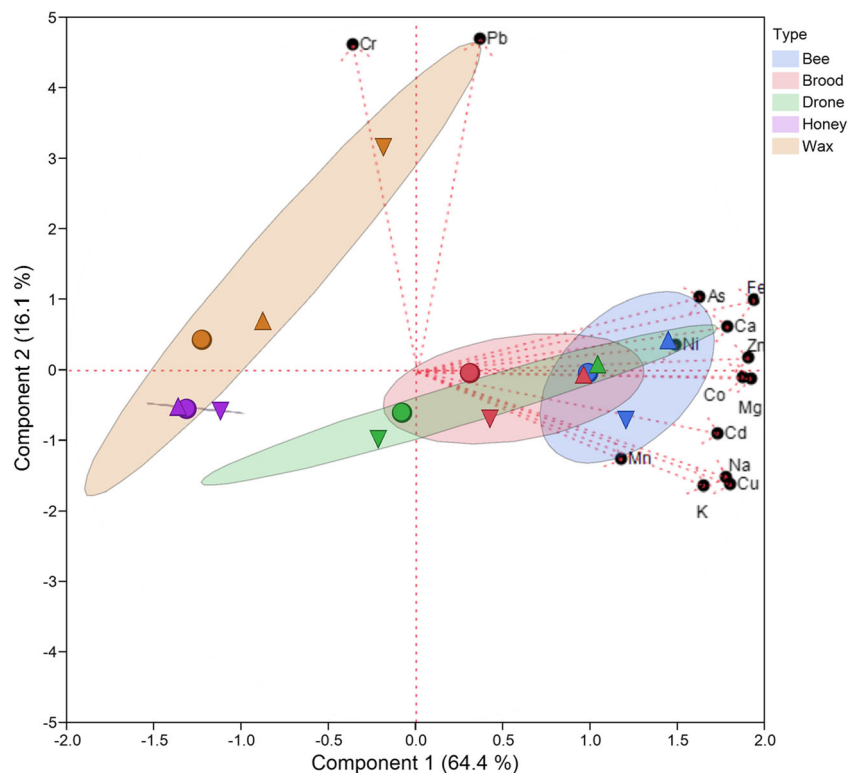
Table 2 Pearson correlation coefficients for element concentrations in different honey bees and honey bee products

	Na	Mg	K	Ca	Cr	Mn	Fe	Co	Ni	Cu	Zn	As	Cd	Pb
Na	1													
Mg	0.921	1												
K	0.919	0.909**	1											
Ca	0.724	0.891	0.779	1										
Cr	−0.390*	−0.181**	−0.393*	−0.048**	1									
Mn	0.447	0.493	0.439	0.737	−0.276*	1								
Fe	0.811	0.901	0.744	0.895	−0.012**	0.627**	1							
Co	0.663	0.711	0.569	0.764	−0.507*	0.584	0.922*	1						
Ni	0.565	0.655	0.563	0.636	−0.205**	0.259	0.706**	0.669	1					
Cu	0.959	0.941	0.935	0.801	−0.398**	0.543	0.806	0.628	0.600	1				
Zn	0.800	0.898	0.765	0.834	0.017**	0.337	0.822	0.649	0.676**	0.791	1			
As	0.583	0.634	0.513	0.565	0.062**	0.153	0.605	0.340	0.580	0.517	0.794*	1		
Cd	0.740	0.738	0.673	0.793	−0.253**	0.741	0.732	0.619	0.367	0.713	0.682	0.617	1	
Pb	−0.074*	−0.046**	−0.163*	0.063**	0.261	−0.093	0.033**	−0.202	0.055*	−0.142*	0.230**	0.238**	0.024*	1

*Correlation is significant at the 0.05 level (2-tailed)

**Correlation is significant at the 0.01 level (2-tailed)

positively correlated (but not significant) with Fe ($r = 0.901$), Cu ($r = 0.941$), and As ($r = 0.634$). However, K had a significant ($P < 0.05$) negative correlation with Cr ($r = −0.393$) and Pb ($r = −0.163$). Cr had a significant ($P < 0.05$; $P < 0.01$) negative correlation with Mn ($r = −0.276$), Fe ($r = −0.012$), Co ($r = −0.507$), Ni ($r = −0.205$), Cu ($r = −0.398$), and Cd ($r = −0.253$).

Our results provide evidence that honey bees can be used as bioindicator for element deposition (including toxic elements). Also, higher element deposition was found in worker bees, drone bees, and bee broods rather than in honey and wax. Finally, honey bees (worker bees, drone bees, and bee broods) could be better bioindicators of the deposition of different elements than honey.

Compliance with Ethical Standards

The manuscript does not contain clinical studies or patient data.

Conflict of Interest The authors declare that they have no conflict of interest.

References

1. CEC Thematic Strategy for Soil Protection. COM (2006) 231 Final Commission of the European Communities, Brussels (2006) (22.9.2006)

2. Peralta-Videa JR, Lopez ML, Narayan M, Saupe G, Gardea-Torresdey J (2009) The biochemistry of environmental heavy metal uptake by plants: implications for the food chain. Int J Biochem Cell Biol 41(8–9):1665–1677. https://doi.org/10.1016/j.biocel.2009.03.005

3. Saracoglu S, Tuzen M, Mendil D, Soylak M, Elci L, Dogan M (2004) Heavy metal content of hard biscuits produced in Turkey. Bull Environ Contam Toxicol 73:264–269. https://doi.org/10.1007/s00128-004-0422-0

4. Sari A, Mendil D, Tuzen M, Soylak M (2008) Biosorption of cd(II) and Cr(III) from aqueous solution by moss (*Hylocomium splendens*) biomass: equilibrium, kinetic and thermodynamic studies. Chem Eng J 144:1–9. https://doi.org/10.1016/j.cej.2007.12.020

5. Sodre GD, Marchini LC, Zucchi O, Nascimento VF, Otsuk IP, Moreti A (2007) Determination of chemical elements in Africanized *Apis mellifera* (Hymenoptera: Apidae) honey samples from the state of Piaui, Brazil. Quim Nova 30:920–924

6. Uluozlu OD, Tuzen M, Mendil D, Soylak M (2007) Trace metal content in nine species of fish from the black and Aegean seas, Turkey. Food Chem 104:835–840. https://doi.org/10.1016/j.foodchem.2007.01.003

7. Jarup L (2003) Hazards of heavy metal contamination. Br Med Bull 68:167–182. https://doi.org/10.1093/bmb/ldg032

8. Jovanović D, Marković R, Teodorović V, Šefer D, Krstić M, Radulović S, Ivanović J, Janjić J, Baltić ŽM (2017) Determination of heavy metals in muscle tissue of six fish species with different feeding habits from the Danube River, Belgrade-public health and environmental risk assessment. Environ Sci Pollut Res Int 24:11383–11391. https://doi.org/10.1007/s11356-017-8783-1

9. Spiric D, Ciric J, Teodorovic V, Nikolic D, Nikolic A, Radicevic T, Jankovic S (2019) Trace elements and heavy metals in multifloral honeys from Serbia. In IOP Conference Series: Earth and Environmental Science (Vol. 333, No. 1, p. 012104). IOP Publishing

10. Silici S, Uluozlu OD, Tuzen M, Soylak M (2013) Honeybees and honey as monitors for heavy metal contamination near thermal power plants in Mugla, Turkey. Toxicol Ind Health 32(3):507–516. https://doi.org/10.1177/0748233713503393

11. Pohl P, Sergiel I, Prusisz B (2011) Direct analysis of honey for the total content of Zn and its fractionation forms by means of flame atomic absorption spectrometry with solid phase extraction and ultrafiltration approaches. Food Chem 125(4):1504–1509. https://doi.org/10.1016/j.foodchem.2010.10.077

12. Costa-Silva F, Maia M, Matos CC, Calçada E, Barros AI, Nunes FM (2011) Selenium content of Portuguese unifloral honeys. J Food Compos Anal 24:351–355. https://doi.org/10.1016/j.jfca.2010.09.019

13. Bogdanov S (2006) Contaminants of bee products. Apidologie 37(1):1–18. https://doi.org/10.1051/apido:2005043

14. Duran A, Tuzen M, Soylak M (2008) Trace element levels in some dried fruit samples from Turkey. Int J Food Sci Nutr 59:581–589. https://doi.org/10.1080/13561820701507910

15. Enrich C, Boeykens S, Caracciolo N, Custo G, Vazquez C (2007) Honey characterization by total reflection x-ray fluorescence: evaluation of environmental quality and risk for the human health. X-Ray Spectrometry 36(4):215–220. https://doi.org/10.1002/xrs.944

16. Oroian M, Prisacaru A, Hretcanu EC, Stroe SG, Leahu A, Buculei A (2016) Heavy metals profile in honey as a potential indicator of botanical and geographical origin. Int J Food Prop 19(8):1825–1836. https://doi.org/10.1080/10942912.2015.1107578

17. Spirić D, Ćirić J, Đorđević V, Nikolić D, Janković S, Nikolić A, Teodorović V (2019) Toxic and essential element concentrations in different honey types. Int J Environ Anal Chem 99(5):474–485. https://doi.org/10.1080/03067319.2019.1593972

18. Kadri SM, Zaluski R, de Oliveira OR (2017) Nutritional and mineral contents of honey extracted by centrifugation and pressed processes. Food Chem 218:237–241. https://doi.org/10.1016/j.foodchem.2016.09.071

19. Pisani A, Protano G, Riccobono F (2008) Minor and trace elements in different honey types produced in Siena County (Italy). Food Chem 107(4):1553–1560. https://doi.org/10.1016/j.foodchem.2007.09.029

20. González Paramás AM, Gómez Bárez JA, Garcia-Villanova RJ, Rivas Palá T, Ardanuy Albajar R, Sánchez Sánchez J (2000) Geographical discrimination of honeys by using mineral composition and common chemical quality parameters. J Sci Food Agric 80(1):157–165. https://doi.org/10.1002/(SICI)1097-0010(20000101)80:1<157::AID-JSFA506>3.0.CO;2-B

21. Golob T, Doberšek U, Kump P, Necemer M (2005) Determination of trace and minor elements in Slovenian honey by total reflection X-ray fluorescence spectroscopy. Food Chem 91:593–600. https://doi.org/10.1016/j.foodchem.2004.04.043

22. Perugini M, Manera M, Grotta L, Abete MC, Tarasco R, Amorena M (2011) Heavy metal (Hg, Cr, Cd and Pb) contamination in urban reserves: honeybees as bioindicators. Biol Trace Elem Res 140:170–176. https://doi.org/10.1007/s12011-010-8688-z

23. Nuorteva P (1990) Metal distribution patterns and forest decline: seeking Achilles Heel for metals in Finnish forest biocoenoses. In Publications of Department Environmental Conservation 11. University of Helsinki New York

24. Ivanović J, Baltić MŽ, Jelić D, Janjić J, Bošković M, Marković R, Starčević-Dokmanović M (2015) Research of production volume and market turnover of honey from 2004 to 2014. Vet Glas 69(5–6):467–478

25. Statistical Yearbook of the Republic of Serbia 2018. https://publikacije.stat.gov.rs/G2018/Pdf/G20182051.pdf. Accessed Oct 2018

26. Alqarni AS, Owayss AA, Mahmoud AA, Hannan MA (2014) Mineral content and physical properties of local and imported honeys in Saudi Arabia. Int J Environ Anal Chem 18(5):618–625. https://doi.org/10.1016/j.jscs.2012.11.009

27. Chua LS, Abdul-Rahaman NL, Sarmidi MR, Aziz R (2012) Multi-elemental composition and physical properties of honey samples from Malaysia. Food Chem 135(3):880–887. https://doi.org/10.1016/j.foodchem.2012.05.106

28. Karabagias IK, Badeka AV, Kontakos S, Karabournioti S, Kontominas MG (2014) Botanical discrimination of Greek unifloral honeys with physico-chemical and chemometric analyses. Food Chem 165:181–190. https://doi.org/10.1016/j.foodchem.2014.05.033

29. Vukašinović-Pešić V, Blagojević N, Brašanac-Vukanović S, Savić A, Pešić V (2020) Using chemometric analyses for tracing the regional origin of multifloral honeys of Montenegro. Foods 9(2):210

30. Bilandžić N, Gačić M, Đokić M, Sedak M, Šipušić ĐI, Končurat A, Gajger IT (2014) Major and trace elements levels in multifloral and unifloral honeys in Croatia. J Food Compos Anal 33(2):132–138. https://doi.org/10.1016/j.jfca.2013.12.002

31. European Union (2006) Commission Regulation No 1881/2006. EU Legislation Which Sets Maximum Levels for Chemical Contaminants in Foodstuffs

32. Official Gazette (2018) Regulation on maximum residue limits of plant protection products in food and feed and on food and feed for which maximum residue limits of plant protection products are set (Official Gazette of the Republic of Serbia No. 22/18, 90/18 and 76/19)

33. Yazgan S, Horn H, Isengard HD (2002) Honig, ein Bioindikator aufgrund seiner Schwermetallgehalte? Lebensmittelchemie 56:97–116

34. Taha EKA, Manosur HM, Shawer MB (2010) The relationship between comb age and the amounts of mineral elements in honey and wax. J Apic Res 49(2):202–207. https://doi.org/10.3896/IBRA.1.49.2.10

35. Formicki G, Greń A, Stawarz R, Zyśk B, Gał A (2013) Metal content in honey, propolis, wax, and bee pollen and implications for metal pollution monitoring. Pol J Environ Stud 22:99–106

36. Conti ME, Botrè F (2001) Honeybees and their products as potential bioindicators of heavy metals contamination. Environ Monit Assess 69(3):267–282. https://doi.org/10.1023/A:1010719107006

37. Madras-Majewska B, Jasinski Z (2003) Lead content of bees, brood and bee products from different regions of Poland. J Apic Sci 47(2):47–55

38. Porrini C, Sabatini AG, Girotti S, Ghini S, Medrzycki P, Grillenzoni F, Celli G (2003) Honey bees and bee products as monitors of the environmental contamination. Apiacta 38(1):63–70

39. Lebedev VI, Murashova E (2004) Ekologicheskaja cistota produktov pcelovodstvoch. Pcelovodstvo 82:42–44