

Levels of Copper, Selenium, Lead, and Cadmium in Forager Bees

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Abstract

The aim of this study was to determine levels of trace metals (Cu, Se, Pb and Cd) in bodies of forager bees. The study was conducted in 14 stationary apiaries situated in southeastern Opole Province (Poland) in autumn 2007 and in spring, summer and autumn 2008. Samples were mineralized using microwave method. Quantitative analysis of studied metals in bees' bodies and multiflower honey were done using the plasma spectrometry method (ICP).

It was found that copper was an element present in bodies of worker bees in the highest concentration. Its mean content was 22.6 mg·kg⁻¹ d.m. Next was selenium – 7.03, lead – 1.95 and cadmium – 0.65 mg·kg⁻¹ d.m. The highest level of Cu, Pb and Cd accumulation (means on a level of 24.5, 2.59 and 0.83 mg·kg⁻¹ d.m., respectively) was observed in samples of bees obtained during spring 2008, and of selenium in autumn 2008. The lowest concentration of Pb and Cd (means on a level of 0.95 and 0.46 mg·kg⁻¹ d.m., respectively) were in turn observed in autumn 2008, Cu (20.6 mg·kg⁻¹ d.m.) in autumn 2007, and Se (5.81 mg·kg⁻¹ d.m.) in spring 2008. In most cases, the differences in contents of particular metals between periods of sample collection were significant ($p \leq 0.05$) or highly significant ($p \leq 0.01$) statistically.

Higher concentrations of copper (23.3 in comparison to 22.0 mg·kg⁻¹ d.m.), selenium (7.58 in comparison to 6.48 mg·kg⁻¹ d.m.), and lead (1.98 in comparison to 1.91 mg·kg⁻¹ d.m.) were demonstrated in bees from urbanized regions, while cadmium (0.70 in comparison to 0.60 mg·kg⁻¹ d.m.) in those from agricultural-woodland.

The sequence of the range of analyzed metals accumulation in honey was the same as in the case of bees, but concentrations were manifold lower: Cu – 0.82, Se – 0.38, Pb – 0.06 and Cd – 0.06 mg·kg⁻¹ d.m). Any significant relationships between the content of particular elements in honey and bees were demonstrated.

Keywords: honeybee, honey, trace elements, copper, selenium, lead, cadmium

Introduction

The development of anthropogenic sources of pollution emission (industry, motorization, urban development, intense agriculture) negatively influences the environment. Emitted dust, smoke, and industrial or exhaust gases that poisoning the atmosphere, soil, and water also contaminate plants [1, 2].

Increased chemical plant protection and fertilization also contributes to this contamination [3-5]. A prime result of such activity in an increase in concentrations of heavy metals in soil, water, plants, and other elements of an environment [2, 8-11].

Some part of that contamination is accumulated in organisms of honey bees as a result of living processes [16, 17], due mostly to raw material (nectar, pollen, resin, water) transformation into honey [18-21]. There is evidence that

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excessive levels of lead ($LD_{50} = 12$ to $50 \mu\text{g}$ per head), cadmium ($LD_{50} = 6$ to $30 \mu\text{g}$ per head), copper ($LD_{50} = 50$ to $250 \mu\text{g}$ per head), and selenium (LD_{50} to $670 \mu\text{g}$ per head) in bee bodies are important reasons for the regression, and in some regions even of the decay of this species. The level of dose that is toxic for bees is different for particular elements, and depends on the kind and form of chemical compounds the elements occur in [22]. Bees that live in a clean environment perish in lower concentrations of heavy metals, while bees that live in a contaminated environment, adapted for the presence of high concentrations of those metals, perish at considerably their levels [23].

The district of Nysa, where the experimental apiaries were located, occupies an area of about $1,224 \text{ km}^2$. Arable land is about 73% of the district area, and 12.4% is covered with forests. The plants of metal, machine, food, and building-assembly industry are located in the city of Nysa (27.5 km^2 , about 46,500 inhabitants). A galvanizing plant, paint plants, a producer of steel construction materials, semitrailers, plastic systems of soil stabilization, a non-ferrous foundry, and heat engineering are among the biggest contaminators.

The aim of the study was to determine levels of Cu, Se, Pb, and Cd in bodies of worker bees in two regions (urban – pollution region and agricultural-woodland – clean region) and subsequent periods of a year – autumn 2007, and spring, summer, and autumn 2008, and also to establish if there is a relationship between the content of analyzed elements in honey and bees.

Material and Methods

The research materials were samples of forager bees obtained from bee colonies maintained in stationary apiaries located in the southeastern part of Opole Province (Nysa administrative district). Their bodies were collected in autumn 2007 (from September 25 to October 5), spring 2008 (from March 20 to April 2), summer 2008 (from June 25 to July 10), and autumn 2008 (as in the previous year). The study covered 100 bee colonies in 14 apiaries (7 apiaries in an urban region and 7 in an agricultural-woodland region). In 8 smaller apiaries (up to 15 bee colonies), these bodies were collected from 5 hives. In 6 bigger apiaries (above 15 bee colonies), they were collected from 10 hives. Bees coming back from a field were captured while alighting boards of hives. About 100 bees from each hive were collected in sterile, plastic containers. 100 samples of bodies of forager bees were obtained in total (8 apiaries \times 5 hives + 6 apiaries \times 10 hives = 100 samples). Additionally, one pooled sample of multiflower honey of a mass of about 100 g each, was collected in each apiary – 14 samples in total.

Collected bees were slain in a laboratory by freezing at about -18°C , and then dried at 45°C , unified by grinding and carefully mixed. A test portion for mineralization of a mass of about 1,000 mg (with an accuracy of 0.10 mg) was prepared from each sample. Test portions were then diluted with 20 ml of solution of concentrated, spectrally pure nitric

acid (Ridel-de Haën 30702 Company), and then mineralized using microwave technique under increased pressure in microprocessor microwave furnace of MARS 5 Company CEM (14-stands). Particular samples of honey were carefully mixed and a test portion of a mass of 1,000 mg (with an accuracy of 0.10 mg) was prepared from each of them. Mineralization was conducted in the same manner as in the case of samples of bees.

The quantitative analysis of studied elements (Cu, Se, Pb and Cd) was done using Varian ICP-AES plasma spectrometer with mass detection, controlled by P-3202 computer, cooperating with an analytical combine Philips Scientific- model PU-7000 and ultrasonic nebulizer CETAC-5000 AT. Calibration curves for particular elements were determined using standards of IPC class of a special purity [24]. The level of detection (LOD) in this method was $1 \mu\text{g}\cdot\text{kg}^{-1} \text{ d.m.}$, but the level of quantification (LOQ) was different for individual elements (Table 2). The above analysis was performed in the Analytical Laboratory of Wrocław University of Environmental and Life Science.

Results of laboratory analysis obtained were worked out statistically. Mean concentrations of elements, standard deviations and correlations between elements were calculated. The results concerning the content of elements within the groups were processed statistically by ANOVA. The Student's t-test (normal distribution) at a significance level of 0.05 and 0.01 was used to examine the significance of differences between period of sample collections and also between regions.

Results

The level of copper in organisms of worker bees was the highest among all analyzed elements. An average concentration of this metal was $22.6 \text{ mg}\cdot\text{kg}^{-1} \text{ d.m.}$ (Table 1). It was demonstrated that content of copper in bees was dependent on a region of apiary localization and period of sample collection for the study. The lowest concentration of copper was found in samples collected in autumn 2007 – $20.6 \text{ mg}\cdot\text{kg}^{-1} \text{ d.m.}$, while the highest in spring 2008 – $24.5 \text{ mg}\cdot\text{kg}^{-1} \text{ d.m.}$ Highly significant differences ($p \leq 0.01$) were demonstrated between the content of copper in bees samples from those two periods. However, differences between the concentration of copper in bee samples collected in autumn 2007 and summer 2008 were significant statistically on a level of $p \leq 0.05$. It turned out that the level of copper accumulation in bees was the most stable. In subsequent periods of sample collections, the lowest values of coefficient of variation (from 12.7 to 22.1%) were determined for that element. Copper content in bees coming from apiaries within Nysa was slightly higher than from apiaries situated in an agricultural-woodland region: mean values were 23.3 and $22.0 \text{ mg}\cdot\text{kg}^{-1} \text{ d.m.}$, respectively (Table 2).

It is worth noting that an average level of selenium content in organisms of worker bees was $7.03 \text{ mg}\cdot\text{kg}^{-1} \text{ d.m.}$ The lowest concentration of that element in bees was in spring 2008 – 5.81, and the highest in autumn 2008 – $7.95 \text{ mg}\cdot\text{kg}^{-1} \text{ d.m.}$ (Table 1). Differences between selenium content in samples of bees collected in spring and summer 2008 were

Table 1. Concentration of trace elements in bees in subsequent periods of the study [$\text{mg}\cdot\text{kg}^{-1}$ d.m.].

Element	Specification	Subsequent periods of the study				In total N=400
		Autumn 2007 N=100	Spring 2008 N=100	Summer 2008 N=100	Autumn 2008 N=100	
Cadmium	min.	0.29	0.33	0.38	0.09	0.09
	max.	0.93	1.82	1.58	1.24	1.82
	\bar{x}	0.53 A	0.83 B	0.77 B	0.46 A	0.65
	SD	0.17	0.36	0.30	0.26	0.32
	V [%]	31.1	43.6	38.5	56.0	49.0
Copper	min.	15.3	15.7	13.9	12.1	12.1
	max.	24.9	35.2	32.1	31.2	35.2
	\bar{x}	20.6 Aa	24.5 B	23.2 b	22.2	22.6
	SD	2.6	5.4	4.7	4.6	4.6
	V [%]	12.7	22.1	20.3	20.5	20.4
Lead	min.	0.81	0.94	0.003	0.003	0.003
	max.	4.94	6.73	3.82	3.53	6.73
	\bar{x}	1.73 aA	2.59 bAB	2.55 B	0.95 C	1.95
	SD	0.78	1.56	0.71	1.09	1.27
	V [%]	45.2	60.3	27.7	115.3	64.8
Selenium	min.	4.70	2.55	1.84	1.40	1.40
	max.	9.27	10.76	11.90	15.81	15.81
	\bar{x}	6.69	5.81 Aa	7.69 B	7.95 b	7.03
	SD	1.28	2.32	2.26	3.85	2.70
	V [%]	19.1	39.9	29.3	48.4	38.1

A, B, C – significant statistical differences on a level of $p \leq 0.01$ between periods;

a, b – significant statistical differences on a level of $p \leq 0.05$ between periods;

V – coefficient of variation.

highly significant statistically on a level of $p \leq 0.01$, while those between samples collected in spring and autumn 2008 were significant on a level of $p \leq 0.05$. Mean selenium concentration in bees coming from apiaries within Nysa was $7.58 \text{ mg}\cdot\text{kg}^{-1}$ d.m., and was higher than the mean value from apiaries from neighborhood agricultural-woodland region – $6.48 \text{ mg}\cdot\text{kg}^{-1}$ d.m. (Table 2).

An average level of lead accumulation in organisms of worker bees was $1.95 \text{ mg}\cdot\text{kg}^{-1}$ d.m. (Table 1). The highest mean concentration of lead was observed in samples of bees collected in spring 2008 – $2.59 \text{ mg}\cdot\text{kg}^{-1}$ d.m., and the lowest in autumn 2008 – $0.95 \text{ mg}\cdot\text{kg}^{-1}$ d.m. Dispersion of results obtained in these periods was significant. Coefficient of variation was 60.3 and 115.3%, respectively. These were one of the highest values of coefficient of variation observed among all analyzed elements. Significant differences (on a level of $p \leq 0.01$) were demonstrated between lead content in samples of bees obtained in autumn 2007, and summer and autumn 2008, and also between

spring and autumn 2008, and summer and autumn 2008. Mean content of lead in organisms of bees coming from apiaries in urban and agricultural-woodland areas were convergent: 1.98 and $1.91 \text{ mg}\cdot\text{kg}^{-1}$ d.m., respectively (Table 2).

Interestingly, the mean content of cadmium in organisms of bees was $0.65 \text{ mg}\cdot\text{kg}^{-1}$ d.m. (Table 1), so it was the lowest among all analyzed elements. In samples of bees obtained in spring 2008, an average concentration of cadmium was the highest – $0.83 \text{ mg}\cdot\text{kg}^{-1}$ d.m., while the lowest in autumn 2008 – $0.46 \text{ mg}\cdot\text{kg}^{-1}$ d.m. Cadmium accumulation in bees from apiaries in Nysa was $0.60 \text{ mg}\cdot\text{kg}^{-1}$ d.m., and was lower than the mean from apiaries from an agricultural-woodland region – $0.70 \text{ mg}\cdot\text{kg}^{-1}$ d.m. (Table 2).

The second part of our study concerning the content of above elements in multiflower honey demonstrated that their level was multi-fold lower than in bees. An average content of copper was 0.82 , selenium – 0.38 , lead – 0.06 , and cadmium $0.06 \text{ mg}\cdot\text{kg}^{-1}$ d.m. (Table 2). Any statistically

Table 2. Average content of analyzed elements in honey and bees coming from subsequent apiaries, N=14 [in mg·kg⁻¹ d. m.].

Region	No. of apiary	Cd		Cu		Pb		Se	
		honey	bees	honey	bees	honey	bees	honey	bees
Urban region	1.	0.01	0.69	0.01	20.2	0.11	1.46	0.50	7.11
	2.	0.11	0.81	0.05	23.5	0.04	2.14	0.35	8.23
	3.	0.10	0.69	0.01	22.9	0.08	1.61	0.32	6.68
	4.	0.03	0.58	1.16	23.9	0.04	2.18	0.29	6.12
	5.	0.04	0.44	0.20	24.2	0.11	1.93	0.22	11.04
	6.	0.09	0.61	0.23	23.0	0.14	2.32	0.17	6.72
	7.	0.10	0.39	1.14	25.5	0.02	2.22	0.47	7.13
	Mean	0.07 *	0.60	0.40	23.3 *	0.08	1.98	0.33	7.58
	SD	0.04	0.15	0.52	1.63	0.04	0.33	0.12	1.66
Agricultural-woodland region	8.	0.03	0.42	0.01	19.5	0.04	2.31	0.20	4.99
	9.	0.04	1.04	0.07	23.9	0.01	1.25	0.50	7.89
	10.	0.01	0.57	0.13	20.5	0.02	2.69	0.83	5.28
	11.	0.03	0.92	6.02	21.4	0.06	3.11	0.23	8.12
	12.	0.04	0.53	1.70	24.1	0.13	1.13	0.11	6.32
	13.	0.07	0.75	0.25	21.7	0.01	1.08	0.77	5.82
	14.	0.09	0.64	0.44	22.7	0.07	1.82	0.31	6.97
	Mean	0.04 *	0.70	1.23	22.0 *	0.05	1.91	0.42	6.48
	SD	0.03	0.22	2.19	1.71	0.05	0.81	0.29	1.23
Total	LOQ	≤ 0.0003	≤ 0.009	≤ 0.0006	≤ 0.01	≤ 0.003	≤ 0.003	≤ 0.095	≤ 0.175
	Mean	0.06 A	0.65 B	0.82 A	22.6 B	0.06 A	1.95 B	0.38 A	7.03 B
	SD	0.04	0.19	1.59	1.7	0.05	0.60	0.22	1.51
	V [%]	65.8	28.9	194.4	7.7	74.8	30.6	57.4	21.5
Correlation coefficient (r)		0,281		0.119		0.164		0.394	

A, B – highly significant statistical differences on a level of $p \leq 0.01$ between honey and bees;

* – significant statistical differences on a level of $p \leq 0.05$ between regions;

LOQ - the level of quantification.

significant correlations in particular element contents between honey and bees were demonstrated (Table 2).

Significant (on a level of $p \leq 0.05$) or highly significant ($p \leq 0.01$) positive correlations between concentrations of cadmium and copper, cadmium and lead, and copper and selenium in bodies of worker bees were observed (Table 3). Statistically significant differences (on a level of $p \leq 0.05$)

were demonstrated only in the case of copper level in bees, and cadmium in honey between the urban (Nysa) and agricultural-woodland regions.

It should be emphasized that the level of accumulation of heavy metals in organisms of forager bees is in a significant manner dependent on an origin region and season the bees function in.

Table 3. Relationship between concentration of particular elements in bees (N=100).

Elements	Cu-Cd	Cu-Pb	Cu-Se	Cd-Pb	Cd-Se	Pb-Se
Values of correlation coefficient (r)	0.364*	0.240	0.369*	0.541**	0.181	0.141

* - correlation coefficient significant at a level of $p < 0.05$;

** - correlation coefficient significant at a level of $p < 0.01$.

Discussion

The Author found that bodies of worker bees accumulated trace elements of heavy metals in various concentrations whose range depended on the region of apiary location and period of sample collection. This observation (Tables 1 and 2) is confirmed in the paper by Höffel [16], who demonstrated that the content of lead and cadmium in tissues of bees coming from industrialized regions may be even 8-10 fold higher compared to bees from unindustrialized areas. Bromenshenk [21] and Veleminsky et al. [44] also believe that the level of heavy metals content in bees reflects their concentration in the environment the insects live in. Thus, bees and their products might be used in monitoring an environment with respect to its contamination by heavy metals [1, 6, 7, 13, 17, 44-46]. Jones [47] demonstrated that seasonal changes in heavy metals content (including Cu, Pb and Cd) in honey and soil is obvious. It is connected with their variable supply to the environment and species of blooming nectar giving plants change over the time of the season. For that reason, the content of trace elements also changes during the season in bees fed with the nectar of those plants. However, the literature available does not devote many attention to that issue.

An average concentration of copper was $22.6 \text{ mg}\cdot\text{kg}^{-1}$ d.m. Higher concentration of that metal was noted in bees from an urbanized region ($23.3 \text{ mg}\cdot\text{kg}^{-1}$ d.m.), as compared to agricultural-woodland one ($22.0 \text{ mg}\cdot\text{kg}^{-1}$ d.m.). The results concerning the content of copper in bees from industrialized region comparable to those obtained in the present study were demonstrated by Fakhimzadeh [48] – $19.00 \mu\text{g}\cdot\text{g}^{-1}$ (i.e. $19.00 \text{ mg}\cdot\text{kg}^{-1}$) and Szymanowska-Bielawska [33] – $25.4 \mu\text{g}\cdot\text{g}^{-1}$ d.m. (i.e. $25.4 \text{ mg}\cdot\text{kg}^{-1}$ d.m.). Also Roman [17], in his previous study in the Głogów region, demonstrated a concentration of copper in bees on an average level of $23.40\text{-}26.16 \text{ mg}\cdot\text{kg}^{-1}$ d.m. However, in samples of bees from the Rudna region (dumping site of copper industry flotation wastes) the level of copper was a bit higher – 28.33 to $29.80 \text{ mg}\cdot\text{kg}^{-1}$ d.m. In single samples in turn, the author [17] demonstrated a level of copper up to $47.53 \text{ mg}\cdot\text{kg}^{-1}$ d.m.

It is worth noting that an average level of selenium content in organisms of worker bees was $7.03 \text{ mg}\cdot\text{kg}^{-1}$ d.m. The concentration of selenium in bees bodies from urbanized region (more polluted) reached $7.58 \text{ mg}\cdot\text{kg}^{-1}$ d.m., while in bees from agricultural-woodland region – $6.48 \text{ mg}\cdot\text{kg}^{-1}$ d.m. De Jong et al. [49] claim that selenium content in live tissues (of bees) depends on its availability in the environment. The authors demonstrated that in pollen collected by bees from plants growing on a waste dump, selenium concentration was high, while in pollen from plants of the same species growing on arable land selenium was not observed at all. Similar conclusions were drawn by Gutenmann et al. [50]. Roman [51] demonstrated in his earlier study a comparable content of selenium in drones of honeybee – from 3.74 to $7.30 \text{ mg}\cdot\text{kg}^{-1}$ d.m. on average. In worker bees in turn, that concentration was lower – from 2.57 to $5.57 \text{ mg}\cdot\text{kg}^{-1}$ d.m. on average. The author [51] proved also a relationship between the level of selenium concentration in

bees and the region of sample origin – lower in the agricultural-woodland region, and higher in the industrialized one.

An average level of lead accumulation in organisms of worker bees was $1.95 \text{ mg}\cdot\text{kg}^{-1}$ d.m. Lead content in bees from urban and agricultural-woodland regions was comparable and amounted to 1.98 and $1.91 \text{ mg}\cdot\text{kg}^{-1}$ d.m., respectively. While confronting results obtained with those published in literature, it may be noticed that Conti and Botré [15] showed very low content of lead in bees – $0.52\text{-}1.25 \mu\text{g}\cdot\text{kg}^{-1}$ ($0.00052\text{-}0.00125 \text{ mg}\cdot\text{kg}^{-1}$). In turn, Roman [51] obtained lower concentrations of lead – 0.36 on average in worker bees from the agricultural-woodland region, and higher – $0.82 \text{ mg}\cdot\text{kg}^{-1}$ d.m. in the Wrocław region. Similarly, Spodniewska [52], depending on the region of sample origin, demonstrated lead content in bees from 0.390 to $0.531 \mu\text{g}\cdot\text{g}^{-1}$ (i.e. $0.390\text{-}0.531 \text{ mg}\cdot\text{kg}^{-1}$). Comparable or higher amounts of lead in bees were presented by Szymanowska-Bielawska [33] – $2.18 \mu\text{g}\cdot\text{g}^{-1}$ d.m. (i.e. $2.18 \text{ mg}\cdot\text{kg}^{-1}$ d.m.) and Madras-Majewska and Jasiński [53] up to $4.76 \text{ mg}\cdot\text{kg}^{-1}$. In turn, other authors [54, 55] give considerably higher concentrations of lead in bee organisms that are within a very wide range from 12.00 to $607 \text{ mg}\cdot\text{kg}^{-1}$ d.m. (in a region of lead and zinc foundries).

Mean content of cadmium in organisms of bees was $0.65 \text{ mg}\cdot\text{kg}^{-1}$ d.m. That is the only element where concentration was higher in bees from the agricultural-woodland region, as compared to the urbanized one (0.70 and $0.60 \text{ mg}\cdot\text{kg}^{-1}$ d.m., respectively). The high concentration of cadmium in bees may be due to its too large supply to an environment with mineral fertilizers and pesticides [5, 41]. These components are then accumulated in soil and plants [52], and are collected by bees with nectar and pollen [20, 56]. Very low content of cadmium in bees was demonstrated by Conti and Botré [15] – from 2.87 to $4.23 \mu\text{g}\cdot\text{kg}^{-1}$ ($0.00287\text{-}0.00423 \text{ mg}\cdot\text{kg}^{-1}$). Spodniewska [52] demonstrated considerably lower content of cadmium in samples of bees – from 0.081 to $0.178 \mu\text{g}/\text{g}$ (i.e. $0.081\text{-}0.178 \text{ mg}\cdot\text{kg}^{-1}$) depending on the region of sample origin. Roman [51] in his previous study also demonstrated low concentration of cadmium in bees from agricultural-woodland region ($0.22 \text{ mg}\cdot\text{kg}^{-1}$ d.m.), while from the region of Wrocław – $0.12 \text{ mg}\cdot\text{kg}^{-1}$ d.m. Similarly, a low concentration of cadmium in worker bees was observed by Szymanowska-Bielawska [33] – $0.26 \mu\text{g}\cdot\text{g}^{-1}$ d.m. (i.e. $0.26 \text{ mg}\cdot\text{kg}^{-1}$ d.m.). In turn, Muller and Agthe [57] demonstrated the content of cadmium in bees from an unindustrialized region of Germany at a level of $0.45 \text{ mg}\cdot\text{kg}^{-1}$ d.m., and Fakhimzadeh [48] – $0.423 \mu\text{g}\cdot\text{g}^{-1}$ (i.e. $0.423 \text{ mg}\cdot\text{kg}^{-1}$), so on a level comparable to results obtained in the present study. In bees from an industrialized region in turn, the authors [57] determined the content of that element as $1.51 \text{ mg}\cdot\text{kg}^{-1}$ d.m., i.e. on a maximal level obtained in the present study.

In turn, Pratt and Sikorski [58] demonstrated the relationship between the content of heavy metals in organisms of worker bees and honey. The authors demonstrated such a relationship analyzing the content of lead in nectar of golden rod and worker bees. They observed that with the content of lead in nectar on a level of $0.2 \text{ mg}\cdot\text{kg}^{-1}$ d.m., it was $1.4 \text{ mg}\cdot\text{kg}^{-1}$ d.m. in worker bees fed with honey from

that nectar, while with lead level in nectar 13.6 mg·kg⁻¹ d.m., it was 28.1 mg·kg⁻¹ d.m. in bees. Caroli et al. [46] demonstrated copper content in honey on a level from 57.6 to 141 ng·g⁻¹ (0.0576 to 0.141 mg·kg⁻¹). Similarly, Sodr  et al. [59] obtained a low concentration of copper in honey that was on a level of 0.179 µg·g⁻¹ (i.e. 0.179 mg·kg⁻¹). Considerably higher content of copper in honey from the southeastern part of Antolia was obtained by Yilmaz and Yavuz [60] – 1.8 mg·kg⁻¹. Sodr  et al. [59] did not demonstrate selenium presence in bee honey. In turn, lead content determined by other authors was on a level from 23–138 ng·g⁻¹ (0.023–0.138 mg·kg⁻¹) – [46] to 4.2 µg·g⁻¹ (4.2 mg·kg⁻¹) – [23]. Similarly, like in the present study, other authors noted low cadmium content - Caroli et al. [46] on a level of 0.59–0.70 ng·g⁻¹ (0.00059–0.0007 mg·kg⁻¹), while Rashed and Soltan [23] – 0.01 mg·kg⁻¹.

Conclusions

1. The range of accumulation of studied metals was dependent on a period of samples of bee collections – the highest levels of copper, lead and cadmium were in samples from spring 2008, while selenium from autumn 2008.
2. The sequence, as regards the range of analyzed metal concentration in bees, was as follows: Cu >> Se >> Pb > Cd.
3. The most stable level of concentration was demonstrated for copper (coefficient of variation from 12.7 to 22.1%), while the most changeable was the level of lead (coefficient of variation from 27.7 to 115.3%).
4. Higher concentrations of copper, selenium and lead was demonstrated in bees from an urbanized region, while cadmium in those from agricultural-woodland one.
5. The relationship between the content of particular elements in honey and bodies of forager bees was not demonstrated.

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