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Original Research

Metal Content in Honey, Propolis, Wax, and Bee Pollen and Implications for Metal Pollution Monitoring

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Abstract

We studied the concentrations of Cd, Ni, Pb, Fe, Mg, and Zn in multi-floral honey, propolis, bee pollen, and wax coming from apiaries situated in different locations in Małopolska Voivodeship in southern Poland. Honey contained the lowest concentrations of all the tested metals, with Cd and Pb concentrations well below allowable levels. Other products showed much higher contamination by Cd and Pb. Propolis and pollen from certain areas were significantly contaminated with Pb. High metal contents occurred in beeswax. Moreover, positive correlations between metals occurred in wax: i.e. Cd vs. Ni, Cd vs. Fe, Ni vs. Fe, Ni vs. Mg, Pb vs. Fe, and Fe vs. Mg. Interestingly, metal contents in honey were not correlated with metal contents in other tested products. Correlations between Fe concentrations in honey and wax and between Mg concentrations in bee pollen and propolis were exceptional. This suggests that honeybee products may be useful in monitoring environmental contamination by metals, although complex studies of all bee products give sufficient information.

Keywords: honeybee, bee products, heavy metals, Małopolska

Introduction

Honey is used as food and as an important ingredient in different kinds of manufactured foods. It may be a significant source of vitamins and micro- and macro-elements essential for human health [1]. Other bee products, such as bee pollen and propolis, often are used as medicines in prevention of cancer, diabetes, and heart diseases. Propolis and bee pollen have anti-inflammatory and antioxidant properties due to high concentrations of flavonoids and other phenolic compounds [2, 3]. Moreover, propolis has been proposed as a germicide for food packaging or as a chemical preservative in some food products [4].

Considering nourishing and healing properties of bee products, it is important to know their contamination. Some apiaries are located in areas exposed to different kinds and levels of pollution. Honeybees may forage over a distance of several kilometers from the hives and they effectively sample the environment for contaminants in plants, soil, and the atmosphere [5]. Large concentrations of heavy metals were found in bee products from hives located in areas of high industrial or agricultural activity. This is why honey and other honeybee products are considered good material for monitoring environmental contamination [6, 7].

Apiculture is very popular in Poland. The number of hives in Poland exceeds 800,000. Yearly, the country produces about 22,000 tons of honey [8]. Numerous apiaries are located among houses or close to crops, which may result in higher contamination of bee products. One of the most industrialized parts of Poland is Małopolska in the south. This is also an area of high agricultural activity. Environmental monitoring here indicates industry and agri-

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Table 1. Cadmium content ($\mu g \cdot g^{-1} \times 10^{-3}$) in tested bee products.

	Bukowno	Proszowice	Wieliczka	Raciechowice	Myślenice	Lubień	S. Beskidzka	Zakliczyn
Honey	5.0 ± 3.0^{PRS}	1.0 ± 0.4^{BLWZ}	4.9 ± 1.6^{PR}	1.6 ± 1.2^{BLWZ}	3.0 ± 0.6^{LZ}	6.3 ± 1.0^{MPRS}	2.0 ± 0.5^{BLZ}	6.5 ± 2.0^{MPRS}
Wax	57.1 ± 35.7^{MPW}	4.7 ± 2.1^{BRSZ}	6.4 ± 2.3^{BRSZ}	70.8 ± 37.8^{MPW}	6.9 ± 1.7^{BRSZ}	47.9 ± 13.0^{Z}	56.6 ± 23.9^{MPW}	98.7 ± 14.2^{LMPW}
Pollen	45.6 ± 10.7^{P}	92.0 ± 50.9^{BLSZ}	48.7 ± 14.4	33.5 ± 8.8^{P}	55.5 ± 15.5	35.9 ± 1.0^{P}	26.1 ± 15.2^{P}	69.3 ± 20.4
Propolis	49.6 ± 21.7^{MW}	18.3 ± 3.6^{L}	15.5 ± 6.0^{BL}	31.2 ± 20.2	12.5 ± 4.7^{BL}	54.4 ± 22.2^{MPWZ}	25.7 ± 24.8	18.7 ± 3.4^{L}

B – significant differences compared to bee product from Bukowno at $p<0.05$
P – significant differences compared to bee product from Proszowice at $p<0.05$
W – significant differences compared to bee product from Wieliczka at $p<0.05$
R – significant differences compared to bee product from Raciechowice at $p<0.05$
M – significant differences compared to bee product from Myślenice at $p<0.05$
L – significant differences compared to bee product from Lubień at $p<0.05$
S – significant differences compared to bee product from Sucha Beskidzka at $p<0.05$
Z – significant differences compared to bee product from Zakliczyn at $p<0.05$

culture as the most important sources of contamination, among which heavy metals belong to the most common and toxic contaminants [9].

Taking into consideration the large application of honeybee products in human nutrition and their possible contamination with toxic metals, we decided to study contents of Cd, Ni, Pb, Fe, Mg, and Zn in multi-floral honey, propolis, bee pollen, and wax coming from apiaries situated in different locations in Małopolska. Another aim of our work was to indicate the most useful bee products for environmental monitoring, taking into consideration the relationships in metal contents between honey, wax, propolis, and pollen.

Material and Methods

The study material was collected in 8 apiaries located in Bukowno, Proszowice, Wieliczka, Raciechowice, Myślenice, Lubień, Sucha Beskidzka, and Zakliczyn in Małopolska Voivodeship (Fig. 1). In each apiary, n=10 hives were chosen randomly. From each selected hive single samples of honey, bee pollen, propolis, and wax were taken. The material was collected in June or August 2009. Honey was taken directly from the combs. Samples of wax were taken from freshly formed combs. Balls of bee pollen were collected from a pollen trap. Each product was represented by 80 samples, and the total number of samples was 320. Each sample weighed 2 g.

In the laboratory all the samples were weighed and next mineralized in 2 ml of concentrated nitric acid at 90ºC until complete dissolution. The acid solutions of the samples were filled with deionized water to a volume of 5 cm. Mineral contents of Cd, Ni, Pb, Fe, Mg, and Zn were estimated with flame atomic absorption spectrometry using a Coleparmer BUCK 200A spectrophotometer using hollow cathode lamps. Reference material was provided by BUCK scientific USA.

Statistical analysis was performed with one-way ANOVA followed by Tukey's test. The distribution of the results was studied with Shapiro-Wilk Test. Homogeneity of variances was checked with Levene's test. In further analysis the data were logarithmically transformed and then correlation coefficient r^2 was calculated. Statistical significance was defined at $p<0.05$.

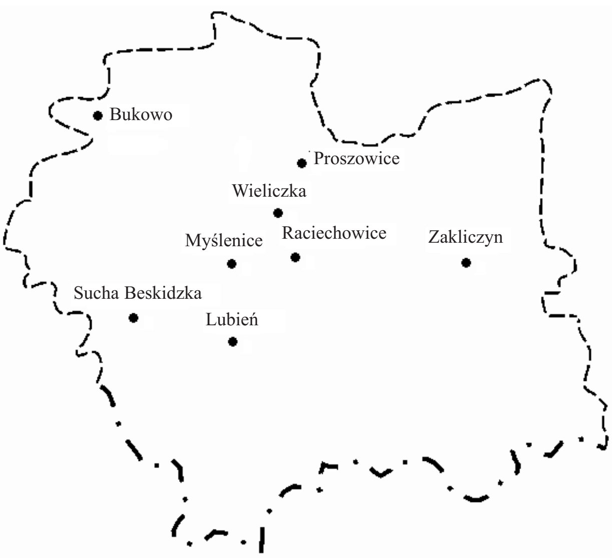

Fig. 1. The distribution of tested apiaries.

Results

Cadmium

One-way analysis of variances indicated highly significant differences in cadmium content between products coming from the studied apiaries. The significance levels were the following: honey (p=0), propolis (p=0.0007), bee pollen (p=0), and wax (p=0). The results of *post hoc* analysis are placed in Table 1. Generally the lowest cadmium content was estimated in honey with the lowest value in honey from Proszowice. Interestingly, propolis and bee pollen from the apiary in Proszowice contained significantly higher cadmium content than propolis and bee pollen

Table 2. Nickel content (μg·g^{-1}) in tested bee products.

	Bukowno	Proszowice	Wieliczka	Raciechowice	Myślenice	Lubień	S. Beskidzka	Zakliczyn
Honey	2.51±0.61	1.24±0.28	1.84±0.47	1.96±0.81	2.45±0.78	4.15±0.62	1.45±0.11	2.73±0.55
Wax	5.12±0.59	2.95±1.11^{Z}	2.29±0.74^{Z}	4.59±1.84	1.89±0.53^{Z}	4.14±0.81	4.89±0.69	7.35±0.61^{MPW}
Pollen	3.61±0.54^{PZW}	8.21±1.13^{BMLRS}	8.41±1.39^{BMLRSZ}	4.89±0.62^{PW}	5.16±0.78^{PW}	4.14±0.63^{PZW}	5.04±0.60^{PW}	6.23±1.68^{BLW}
Propolis	1.99±0.51^{W}	4.55±0.81^{W}	9.81±0.96^{BMPRSZ}	4.96±1.33^{W}	2.63±0.73^{W}	7.63±2.56	4.06±0.93^{W}	3.72±0.56^{W}

See legend under Table 1

from other apiaries. It should also be noted that in most apiaries the highest cadmium content was found in wax, with the highest mean concentration in wax from Zakliczyn (Table 1).

Cadmium content was positively correlated with contents of tested metals in honey, wax, and propolis (Table 7, Figs. 2 and 3). Negative correlations occurred between cadmium and magnesium in bee pollen (r^2=0.62, p=0.21) (Table 7). Interestingly, we did not observe any interdependences between cadmium contents in the studied products.

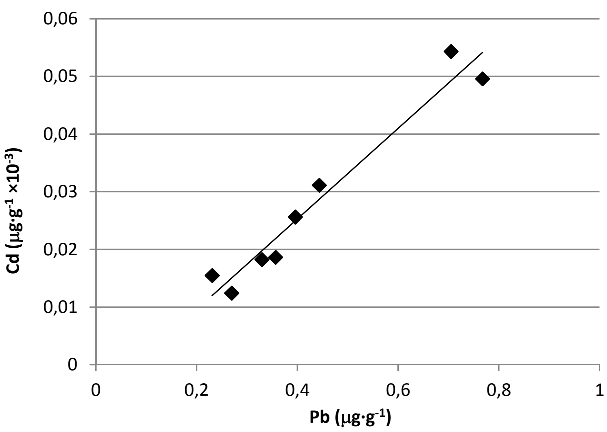

Fig. 2. Strong interdependence (r^2=0.95, p=0) between Cd and Pb contents in propolis, suggesting a common source of both metals.

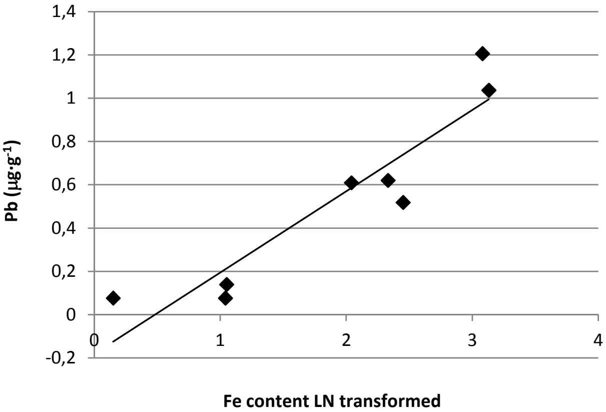

Fig. 3. Positive correlation (r^2=0.84, p=0.001) between Cd and Fe content in wax.

Nickel

Generally, the lowest nickel content was noted in honey. On the other hand some products such as wax from Proszowice, Wieliczka, and Myślenice, or propolis from Bukowno and Myślenice had nickel content in the range of Ni concentrations in honey. The lowest concentration of nickel was found in honey from Proszowice, and the highest in propolis from Wieliczka. High concentrations of nickel were also found in wax collected in the apiary from Zakliczyn. ANOVA indicated significant differences between samples of propolis (p=0), bee pollen (p=0), and wax (p=0.0006) taken from consecutive apiaries. The detailed results together with *post hoc* analysis are presented in Table 2.

Some positive correlations occurred between nickel content and contents of other tested metals in honey, wax, and bee pollen (Table 7). Exceptional was nickel content in propolis, which was not correlated with any of the other tested metals. No correlations were found between nickel contents in the tested products.

Lead

Lead was found in all the tested products. Its content was the lowest in honey. High Pb concentrations were estimated in wax, especially from Bukowno and Zakliczyn. Interestingly, lead contents in honey from these two apiaries were also high compared to other honey samples. High lead contents also were found in pollen from Wieliczka and propolis collected in Bukowno (Table 3, Fig. 4). Analysis of variances ANOVA indicated significant differences between samples of wax (p=0.0001), propolis (p=0), and bee pollen (p=0.002) from consecutive apiaries. Detailed results and *post hoc* analysis are presented in Table 3.

Correlations between lead content and contents of other tested metals are presented in Table 7 and Fig. 2. There were no interdependencies between concentrations of lead in the tested products.

Iron

Among the tested products, iron content was the lowest in honey (Table 4). In most apiaries high iron concentrations occurred in wax and bee pollen. The highest Fe contents were estimated in wax from Bukowno and Zakliczyn.

Table 3. Lead content (μg·g^{-1}) in tested bee products.

	Bukowno	Proszowice	Wieliczka	Raciechowice	Myślenice	Lubień	S. Beskidzka	Zakliczyn
Honey	0.12±0.06	0.06±0.01	0.08±0.03	0.09±0.05	0.09±0.03	0.10±0.04	0.08±0.01	0.13±0.03
Wax	3.13±1.98^{MPW}	1.09±0.17^{BZ}	0.15±0.02^{BRSZ}	2.33±0.21^{W}	1.04±0.07^{BZ}	2.04±0.11	2.45±0.13^{W}	3.08±0.15^{MPW}
Pollen	1.56±0.09^{W}	1.24±0.07^{W}	2.49±0.16^{BMPZ}	1.68±0.09	1.31±0.06^{W}	1.76±0.13	1.91±0.11	1.43±0.14^{W}
Propolis	2.94±0.27^{MSPWZ}	1.26±0.04^{BL}	0.89±0.12^{BL}	1.70±0.06	1.03±0.07^{BL}	2.70±0.30^{MPZW}	1.52±0.13^{B}	1.37±0.11^{BL}

See legend under Table 1

Table 4. Iron content (μg·g^{-1}×10^{2}) in tested bee products.

	Bukowno	Proszowice	Wieliczka	Raciechowice	Myślenice	Lubień	S. Beskidzka	Zakliczyn
Honey	0.16±0.12	0.08±0.02	0.15±0.03	0.16±0.05	0.12±0.05	0.21±0.13	0.10±0.03	0.24±0.11
Wax	2.82±1.13^{MPW}	1.15±0.12^{BZ}	1.08±0.13^{BZ}	1.86±0.64^{Z}	1.08±0.40^{BZ}	1.84±0.78^{Z}	1.68±0.38^{Z}	3.34±0.97^{LMPRSW}
Pollen	1.06±0.08^{W}	1.39±0.26	1.69±0.36^{BMLRSZ}	1.07±0.13^{W}	1.10±0.14^{W}	1.15±3.75^{W}	1.09±0.09^{W}	1.25±0.11^{W}
Propolis	0.65±0.21	1.01±0.27^{RSZ}	0.72±0.46	0.30±0.06^{P}	0.72±0.41	0.43±0.14	0.28±0.07^{P}	0.31±0.02^{P}

See legend under Table 1

Special attention should be also paid to high iron content in bee pollen from the apiary in Wieliczka and propolis from the apiary in Proszowice. One-way ANOVA indicated statistically significant differences between samples of wax (p=0), propolis (p=0.004), and bee pollen (p=0) collected in the studied apiaries. *Post hoc* analysis together with the detailed results are shown in Table 4.

Correlations between iron contents and contents of other tested metals are presented in Table 7 and Fig. 3. Interestingly, iron content in honey was correlated with iron content in wax (r^2=0.54, p=0.039).

Magnesium

Bee pollen was the richest in magnesium, and the highest Mg concentration was estimated in pollen from Sucha Beskidzka. High magnesium concentrations also occurred in wax and propolis, especially in wax from Zakliczyn and propolis from Proszowice. Honey contained low concentrations of magnesium compared to other tested bee products. Comparison of products coming from different apiaries by one-way analysis of variances showed significant differences between samples of bee pollen (p=0), propolis (p=0.07), and wax (p=0.006). Mean magnesium contents and *post hoc* analysis are placed in Table 5.

Concentration of magnesium in honey was not correlated with any of the tested metals in this product. Some interdependences between magnesium and other metals occurred in wax, pollen, and propolis (Table 7). Positive correlations were also noted between magnesium contents in bee pollen and propolis (r^2=0.64, p=0.018).

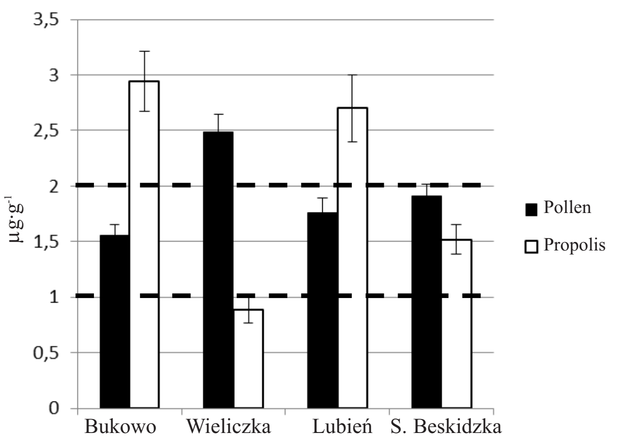

Fig. 4. Lead content in pollen and propolis from chosen apiaries. Bold lines indicate the range of allowable Pb concentrations in dried vegetables and fruits.

Zinc

Similar to other elements, zinc occurred in the lowest concentration in honey. The lowest zinc concentration was estimated in honey from Sucha Beskidzka and the highest in honey from the apiary in Zakliczyn. Bee pollen contained the highest concentration of zinc, and pollen from Wieliczka was the richest in Zn. High concentrations of zinc also occurred in wax and propolis, particularly in wax from Bukowno and propolis from Proszowice. One-way ANOVA indicated statistically significant differences in Zn content between the samples of honey (p=0.01), bee pollen (p=0.04), and wax (p=0.04) from the consecutive apiaries. Detailed results and *post hoc* analysis are presented in Table 6.

There were not any correlations between zinc content and contents of other metals in propolis and wax. Correlations between metals in other products are presented in Table 7. No interdependencies occurred between zinc contents in consecutive products.

Table 5. Magnesium ($\mu g \cdot g^{-1} \times 10^2$) in tested bee products.

	Bukowno	Proszowice	Wieliczka	Raciechowice	Myślenice	Lubień	S. Beskidzka	Zakliczyn
Honey	0.77±0.28	0.42±0.11	0.85±0.67	0.65±0.30	0.86±0.34	0.67±0.22	0.43±0.13	0.44±0.12
Wax	2.47±0.69^{Z}	2.67±0.29^{Z}	2.22±0.39^{Z}	2.42±0.88^{Z}	2.25±1.16^{Z}	2.61±0.82^{Z}	2.32±0.85^{Z}	4.50±1.41^{BLMPRSW}
Pollen	22.6±5.29	13.7±4.37	19.3±1.32	22.1±4.43	12.9±1.90	20.6±2.02	25.8±0.94	18.7±2.73
Propolis	2.77±1.46^{P}	8.23±4.22^{BLRS}	4.09±0.59	3.06±0.51^{P}	3.49±1.01	2.59±1.17^{P}	1.37±0.43^{P}	3.37±0.38

See legend under Table 1

Table 6. Zinc content ($\mu g \cdot g^{-1}$) in tested bee products.

	Bukowno	Proszowice	Wieliczka	Raciechowice	Myślenice	Lubień	S. Beskidzka	Zakliczyn
Honey	2.60±1.28	2.56±0.74	3.37±1.12	3.12±1.37	3.30±1.19	5.05±3.34	1.66±0.57^{Z}	5.97±2.36^{S}
Wax	81.2±20.3^{L}	41.9±16.6	34.2±15.4	34.2±15.3	33.4±16.8	19.1±4.1^{B}	43.6±32.9	47.4±20.4
Pollen	107.8±16.4	113.4±15.1	159.3±88.9^{M}	89.8±7.2	75.2±12.2^{W}	120.8±37.9	103.5±17.6	99.5±5.22
Propolis	40.9±8.42	71.5±27.2	44.8±9.4	30.8±7.7	17.7±13.5	62.4±12.5	24.9±16.9	25.7±5.36

See legend under Table 1

Discussion

The mineral content of bee products is mostly related to soil type. This results in high differences in mineral content between bee products coming from different locations. For instance, the differences in zinc concentration in propolis from different regions of the world may be as large as 400%. Similarly, high differences reaching 130% occurred in iron content in propolis from different apiaries [10, 11]. The concentrations of iron, zinc, and magnesium in honey estimated in our studies were within the range of these elements measured in honey coming from apiaries located in different regions of the world, including Poland [12]. Interestingly, our studies indicated much higher magnesium content in honey than other studies conducted in Poland [12, 13]. Less is known about the mineral composition of other bee products. Propolis coming from different regions of Argentina contained zinc in concentrations of 23 µg/g and iron in concentrations of 162 µg/g [10]. Both values were several-fold higher than zinc and iron concentrations in most of our samples of propolis. Exceptional was propolis from Sucha Beskidzka, which had similar zinc concentrations as propolis from the Argentinean studies. Among the products that can serve as diet supplements, bee pollen had the largest content of essential metals tested in our studies i.e. iron, zinc, and magnesium. Magnesium was the most abundant element in bee pollen. Its mean concentration exceeded 1200 µg/g in all the tested samples of pollen. Bee pollen may thus be a beneficial magnesium source in different diet supplements. The concentrations of iron and zinc were also relatively high and similar to the concentrations of these elements measured in bee pollen in other studies [14].

Much attention has been paid to the presence of trace elements in bee products. The pollution of bee products with toxic elements is mostly related to human industrial and agriculture activity. It was established that the concentration of such elements as cadmium, lead or manganese reach relatively high values in bee products coming from polluted areas of Europe [16, 17].

Honey tested in our studies contained the lowest concentrations of metals compared to other bee products. Mean concentrations of cadmium and lead in honey did not exceed the highest allowable concentrations established by Polish legislation [18]. Cadmium content was also low in all the other tested products. There is high variability concerning cadmium content in honey coming from different areas of Poland, and numerous reports have informed us about its excess in honey. Spodniewska and Romaniuk [19] reported exceeding cadmium content in honey from 3 per 15 tested apiaries in the Warmia and Masuria province, one of the clearest areas in Poland. The above studies indicated that cadmium concentration in honey from an apiary in Orneta contained 0.121 $\mu g \cdot g^{-1}$, which is a significantly higher concentration than the Cd concentration estimated in our studies. On the other hand, our results indicated relatively high concentrations of lead in some products. There are no established allowable concentrations for lead in propolis and bee pollen. On the other hand, our results may be compared to allowable lead concentrations in some dried vegetables or fruits. Such a comparison points to serious contamination of propolis from Bukowno and Lubień and bee pollen from Wieliczka. Moreover, some samples of bee pollen collected in Sucha Beskidzka were also significantly contaminated with lead, taking into consideration ranges of standard deviation (Fig. 4). Fortunately, propolis and pollen are used in very low amounts as supplements of human diet and thus do not pose a serious source of toxic metals. Studies conducted in other areas of Poland revealed that lead concentrations in honey may exceed allowable concentrations. This was indicated by Spodniewska and Romaniuk [19] during their studies in Warmia and Masuria.

Table 7. Correlations between metal contents in tested products.

	Cd	Ni	Pb	Fe	Mg	Zn
Honey						
Cd		r^2=0.58 (+) p=0.0028	r^2=0.55 (+) p=0.036	r^2=0.71 (+) p=0.009		r^2=0.59 (+) p=0.025
Ni	r^2=0.58 (+) p=0.0028		r^2=0.60 (+) p=0.024	r^2=0.58 (+) p=0.028		r^2=0.52 (+) p=0.043
Pb	r^2=0.55 (+) p=0.036	r^2=0.60 (+) p=0.024		r^2=0.61 (+) p=0.023		
Fe	r^2=0.71 (+) p=0.009	r^2=0.58 (+) p=0.028	r^2=0.61 (+) p=0.023			r^2=0.78 (-) p=0.004
Zn	r^2=0.59 (+) p=0.025	r^2=0.52 (+) p=0.043		r^2=0.78 (-) p=0.004		
Wax						
Cd		r^2=0.91 (+) p=0	r^2=0.86 (+) p=0.001	r^2=0.84 (+) p=0.001		
Ni	r^2=0.91 (+) p=0		r^2=0.87 (+) p=0.001	r^2=0.89 (+) p=0	r^2=0.59 (+) p=0.027	
Pb	r^2=0.86 (+) p=0.001	r^2=0.87 (+) p=0.001		r^2=0.86 (+) p=0.001		
Fe	r^2=0.84 (+) p=0.001	r^2=0.89 (+) p=0	r^2=0.86 (+) p=0.001		r^2=0.56 (-) p=0.033	
Mg		r^2=0.59 (+) p=0.027		r^2=0.56 (-) p=0.033		
Pollen						
Cd					r^2=0.62 (-) p=0.021	
Ni				r^2=0.83 (+) p=0.002		
Pb						r^2=0.59 (+) p=0.027
Fe		r^2=0.83 (+) p=0.002				r^2=0.66 (-) p=0.014
Mg	r^2=0.62 (-) p=0.021					
Zn			r^2=0.59 (+) p=0.027	r^2=0.66 (-) p=0.014		
Propolis						
Cd			r^2=0.95 (+) p=0			
Pb	r^2=0.95 (+) p=0					
Fe					r^2=0.65 (-) p=0.015	
Mg				r^2=0.65 (-) p=0.015		

Interestingly, lead concentrations in honey coming from Lower Silesia were estimated in the range of 0.59 to 2.10 $\mu g \cdot g^{-1}$, which is significantly higher than values estimated in our studies [20]. On the other hand, high lead concentrations in propolis and bee pollen from Bukowno and Wieliczka seem to reflect the influence of Silesia and Kraków agglomerations. Indeed, increased lead concentrations were estimated in dust fallout in some areas of Silesia between 2008 and 2010 [21]. In Małopolska, lead concentrations in dust fallout did not exceed allowable concentrations (taking into consideration the years 2008-10). On the other hand, monitoring data from areas adjacent to Silesia

(Bukowno) indicate the highest concentrations of lead in dust fallout compared to other areas of Małopolska [9, 22, 23]. It should be also noted that other sources of contamination, such as coal or detritus combustion, may influence lead content in bee products, which is a probable explanation for the high lead content in propolis and bee pollen from such locations as Sucha Beskidzka or Lubień.

Beeswax is not used in the human diet, thus its mineral content is generally out of scope of scientific studies. The only exceptions are the studies of some isotopes whose presence and concentrations may serve in recognition of honey and honeycomb origin [15]. Nonetheless, our studies revealed some interesting data that in our opinion may be useful in understanding the deposition of essential and trace elements in different bee products. Beeswax collected in our studies contained the highest levels of iron of all the tested products and relatively high concentrations of zinc. Our studies showed high contamination of beeswax with lead and cadmium. Wax from Bukowno, Raciechowice, Sucha Beskidzka, and Zakliczyn contained the highest concentrations of lead and cadmium compared to other products from the same apiaries. What is interesting is strong positive correlations that occurred between concentrations of Pb and Ni, Pb and Cd, and Pb and Fe in beeswax. Similarly, strong positive correlations occurred between Ni and Cd, Ni and Fe, and Ni and Mg in the studied samples of wax. Positive correlations also occurred between Cd and Fe as well as between Cd and Zn contents in beeswax (Fig. 3). Such interdependences may suggest that metals generally occur in the same kind of pollution (Fig. 2). For instance, dust fallout may contain significant concentrations of cadmium, lead, or iron [21]. In honeybee, beeswax is secreted by wax glands. Perhaps the excess of some elements as well as toxic metals may be transported to the glands and next deposited in wax. Detoxification combined with deposition of toxic elements in wax may be beneficial for the functioning of bee families, although there is no information about such a mechanism in scientific literature. Similar but somewhat weaker correlations between tested metals occurred in honey, a partially transformed product in the gut of bees. There is no information whether bees can actively regulate mineral content of honey. Most authors indicate that chemical composition of honey is mainly related to the chemical composition of the floral source [24]. Thus, low concentration of toxic metals in honey, indicated in our studies, should be related to low metal content in floral nectar collected by bees.

Interestingly enough, there were no correlations concerning concentrations of tested metals between consecutive products. Bee pollen and propolis are far less transformed by bees than wax and honey [25, 26]. Thus, pollen and propolis more precisely reflect environmental contamination. On the other hand, one can expect that high contamination of pollen or propolis with toxic metals should be reflected in increased contamination of honey and wax. Surprisingly, this relation was rare. Correlations between iron contents in honey and wax and between magnesium contents in bee pollen and propolis were exceptional. Bees use different plants, plants organs, and/or exudates during production of wax, honey, propolis, and pollen, which may be one of the reasons for the lack of interdependences between mineral composition of consecutive bee products [12]. Moreover, numerous ecotoxicological research indicate that exposure to metals of different animals is often accidental [27, 28]. Bees from the same apiary may sample quite different sites showing different kinds and levels of contamination [29]. This is probably reflected in relatively large values of standard deviation ranges of mean concentrations of metals in products tested in our studies.

Conclusions

Our results indicate that honey produced in apiaries from Southern Poland contain toxic metals in concentrations well below allowable levels. On the other hand, contamination of wax, propolis, and bee pollen was quite serious in some of the apiaries. Correlations occurring between concentrations of some metals in wax may suggest the important role of wax in metal deposition. The lack of interdependences in metal accumulation between the consecutive products probably reflects accidental exposure of bees and plants to contamination. Moreover, different plants and plant organs may contain different concentrations of metals. Thus, exclusively complex studies of all bee products may be useful in monitoring environmental contamination by metals.

References

1. NANDA V., SARKAR B.V., SHARMA H.K., BAWA A.S. Physico-chemical properties and estimation of mineral content in honey produced from different plants in Northern India. J. Food Comp. Anal., **16**, 613, **2003**.
2. PAULINO N., ABREU S.R., UTO Y., KOYAMA D., NAGASAWA H., HORI H., DIRSCH V.M., VOLLMAR A.M., SCREMIN A., BRETZ W.A. Anti-inflammatory effects of a bio available compound, Artepilin C, in Brasilian propolis. Eur. J. Pharmacol. **587**, 296, **2008**.
3. NEWAIRY A.A., SALAMA A.F., HUSSIEN H.M., YOUSEF M.I. Propolis alleviates aluminium-induced lipid peroxidation and biochemical parameters in male rats. Food Chem. Toxicol., **47**, 1093, **2009**.
4. TOSI E.A., RE E., ORTEGA M.E., CAZZOLI A.F. Food preservative based on propolis: bacteriostatic activity of propolis polyphanols and flavonoids upon Escherichia. Food Chem., **104**, 1025, **2007**.
5. PRZYBYŁOWSKI P., WILCZYŃSKA A. Honey as an environmental marker. Food Chem., **74**, 289, **2001**.
6. GILBERT M. D., LISK D.J. Honey as environmental indicator of radionuclide contamination. Bul. Environ. Contam. Tocicol., **19**, 32, **1978**.
7. ÜREN A., ŞERIFOĞLU A., SARIKAHYA Y. Distribution of elements in honey and effect of a thermoelectric power plant on the element contents. Food Chem., **61**, 185, **1998**.
8. GUS. Concise statistical yearbook of Poland. ZWS Warszawa, **2009**
9. WIOŚ KRAKÓW. Report on the state of the environment in Lesser Poland in 2008. Environmental Monitoring Library, Kraków **2009**.

10. CANTARELLI M.A., CAMINA J.M., PETTENATI E.M., MARCHEVSKY E.J., PELLERANO R.G. Trace mineral content of Argentinean raw propolis by neutron activation analysis (NAA): Assessment of geographical provenance by chemometrics. LWT – Food Sci. Technol., **44**, 256, **2011**.
11. ANKLAM E. A review of the analytical methods to determine the geographical and botanical origin of honey. Food Chem., **63**, 549, **1998**.
12. POHL P. Determination of metal content in honey by atomic absorption and emission spectrometries. Trends Aanalyt. Chem., **28**, 117, **2009**.
13. MADEJCZYK M., BARALKIEWICZ D. Characterization of Polish rape and honeydew honey according to their mineral contents using ICP-MS and F-AAS/AES. Analyt. Chim. Acta, **617**, 11, **2008**.
14. KUMP P., NEČEMER M., ŠNAJDER J. Determination of trace elements in bee honey, pollen and tissue by total reflection and radioisotope X-ray fluorescence spectrometry. Spectrochim. Acta, B, **51**, 499, **1996**.
15. CHESSON L.A., TIPPLE B.J., ERKKILA B.R., CERLING T.E., EHLERINGER J.R. B-HIVE: Beeswax hydrogen isotopes as validation of environment. Part I: Bulk honey and honeycomb stable isotope analysis. Food Chem., **125**, 576, **2011**.
16. FODOR P., MOLNAR E. Honey as an environmental indicator: effect of sample preparation on trace element determination by ICP-AES. Microchim. Acta **112**, 113, **1993**.
17. PIETRA R., FORTANER S., SABBIONI E. Use of Chelex 100 resin in preconcentration and radiochemical separation neutron activation analysis applied to environmental toxicology and biomedical research. J. Trace Micro. Tech. **11**, 235, **1993**.
18. REGULATION OF THE MINISTER OF HEALTH FROM 13.01.2003 concerning the levels of chemical and biological pollutants present in food components, permissible additional substances, substances aiding the processing, or on food surface. No. 37, annex 1, **2003**.
19. SPODNIEWSKA A., ROMANIUK K. Lead and cadmium contents in chosen apiaries of Warmian-Masurian Voivodeship. Med. Wet., **63**, 602, **2007**.
20. ROMAN A. Comparative research over Cd, Pb and Zn contents in honey, propolis and wax from the area of Wałbrzych and Głogów. Apiarist Science Notebooks, Suppl. **I**, 76, **1997**.
21. WIOŚ KATOWICE. Report on the state of the environment in Silesia Voivodeship in 2010. Environmental Monitoring Library, Katowice **2011**.
22. WIOŚ KRAKÓW. Report on the state of the environment in Lesser Poland in 2009. Environmental Monitoring Library, Kraków **2010**.
23. WIOŚ KRAKÓW. Report on the state of the environment in Lesser Poland in 2010. Environmental Monitoring Library, Kraków **2011**.
24. BASUALDO C., SGROY V., FINOLA M.S., MARIOLI J.M. Comparison of antibacterial activity of honey from different provenance against bacteria usually isolated from skin wounds. Vet. Microbiol., **124**, 375, **2007**.
25. MELLO B. C. B. S., PETRUS J. C. C., HUBINGER M. D. Concentration of flavonoids and phenolic compounds in aqueous and ethanolic propolis extracts through nanofiltration. J. Food Engin., **96**, 533, **2010**.
26. DEGRANDI-HOFFMAN G., CHEN Y., HUANG E., HUANG M. H. The effect of diet on protein concentration, hypopharyngeal gland development and virus load in worker honey bees (*Apis mellifera* L.). J. Insects Physiol., **56**, 1184, **2010**.
27. FORMICKI G. Heavy metals in aquatic environment, toxicity, biological properties, availability and accumulation in animal tissues. Scientific Publisher of Pedagogical University of Cracow, Kraków **2010**.
28. MARTINIAKOVA M., OMELKA R., STAWARZ R., FORMICKI G. Accumulation of lead, cadmium, nickel, iron, copper, and zinc in boned of small mammals from polluted areas in Slovakia. Pol. J. Environ. Stud., **21**, 153, **2012**.
29. RASHED M. N., SOLTAN M. E. Major and trace elements in different types of Egyptian mono-floral and non-floral bee honeys. J. Food Comp. Anal., **17**, 725, **2004**.