Original Research

Concentration of Chosen Trace Elements of Toxic Properties in Bee Pollen Loads

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Abstract

The aim of this study was to determine the rate of bioaccumulation of chosen heavy metals (As, Pb, Cd, Hg) in fresh pollen obtained in the form of pollen loads. The research material were samples of fresh pollen obtained in stationary apiaries located in two regions: agricultural woodland and a former military airfield. Samples were collected in a period of July – August 2005 and 2006. Samples were mineralized by microwave method. Quantitative analysis of examined metals (arsenic, lead, cadmium and mercury) was done using plasma spectrometry (ICP).

Research demonstrated that mean content of elements of toxic properties in pollen from an agricultural woodland region in 2005 and 2006 were (in $mg\cdot kg^+$ d.m.) as follows: Pb-0.804 and 0.491; Cd-0.234 and 0.272; As-0.060 and 0.036, and Hg-0.0038 and 0.0036, respectively. Lead concentration in 20 samples (n=36) in 2005, and in 11 samples (n=36) in 2006 exceeded acceptable standards (0.50 $mg\cdot kg^+$ d.m.). However, in pollen from the area of the former military airfield the content of particular elements was higher for Pb-0.835 and 0.704, Cd-0.356 and 0.363, As-0.093 and 0.099, and for Hg-0.0066 and 0.0059, respectively. Lead accumulation exceeded permissible standards in 33 samples (n=36) from 2005 and in 21 (n=36) from 2006, and cadmium in 31 and 36 samples (n=36), respectively.

Mercury and arsenic appeared to be metals of toxic properties that do not cause any toxicological problems in pollen from the agricultural woodland and airfield regions since their concentration was very low. High concentration of cadmium in pollen from the agricultural woodland deserves attention. Differences in concentrations of analyzed elements between regions may be used as bioindicators of environmental contamination with elements of toxic properties.

Keywords: pollen, pollen load, heavy metals, lead, cadmium, arsenic, mercury

Introduction

Pollen is a natural plant product that is collected by honey bees mainly as a protein food. A bee colony uses for its own needs from 12 to over 35 kg of pollen per year. Its influx and presence in a bee nest indicates proper colony development, suitable queen oviposition and proper brood growth and development. Pollen contains a rich amino acid composition (32 amino acids) of proteins, including all egzogenous ones that also are essential for humans. It also contains many other valuable components like fats, carbohydrates, enzymes, vitamins, volatile oils and mineral compounds [1-5]. For this reason it is more and more often used as a diet supplement for humans.

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Pollen is exposed to contamination with substances and compounds taken by plants from soil, through root systems with water, and by direct sedimentation of pollution of different origins, including anthropogenic ones. A serious risk for bees' organisms (and also for humans) is caused by elements of toxic properties, including heavy metals, i.e. elements of a mass density above 4.5 g/cm³ that accumulate in a polluted environment. Numerous scientific publications give information concerning the level of heavy metals concentration in bee products, also in pollen obtained in the form of pollen loads [6-9]. Pollen obtained from bees in a form of pollen loads, taken back on the entrance of the hive, does not undergo any technological processes in a beehive. Thus, the level of chemical element content is related to the environment it comes from (soil, water and plants). That is why many authors claim that bees and pollen may be a bioindicator of the environment, including the atmosphere and pollution [10-16]. There is a strict relationship between the level of accumulation of heavy metals in soil and their content in bee products [9, 17].

The aim of this study was to determine the level of bioaccumulation of chosen metals of toxic properties (As, Pb, Cd, Hg) in fresh pollen obtained from bees in the form of pollen loads in two regions: agricultural woodland and a former military airfield.

Material and Methods

The research material were samples of fresh pollen obtained in the form of pollen loads with bee's pollen basket using entrance pollen trap. Pollen was derived from stationary apiaries located in midwestern Opole Province in two regions: agricultural woodland (Lubsza Commune) and a former military airfield (Skarbimierz – southwest from Brzeg). Material was collected June-August in 2005 and 2006 (June 8th, July 12th and August 11th). Thirty-six bee colonies were under examination in each of the analyzed regions. Three pollen collection were conducted in each bee colony, and one cumulative sample representative for each colony, of a mass of about 100 g, was formed. This way 36 samples of pollen were obtained annually in each region (72 samples a year total).

Collected pollen samples were dried (at 42°C), unified by grinding and carefully mixing. The amount of 1000 mg (with accuracy to 0.1 mg) from each sample was weighed for mineralization. Weighed amounts were diluted with 20 ml of concentrated, spectrally pure nitric acid solution (Merck Company), and then mineralized using the microwave technique with increased pressure in an MD-2000 microprocessor station produced by CEM-USA.

Quantitative analysis of studied elements (arsenic, lead, cadmium and mercury) were conducted using a Varian ICP-AES plasma spectrometer with mass detection controlled by a P-3202 computer cooperating with Philips Scientific analytical combine (PU-7000 model), and CETAC-5000 AT ultrasonic nebulizer [18]. The level of detection (LOD) in this method is 1 ppb, but the level of quantification (LOQ) was different for individual elements

(Tables 1 and 2). These analysis were conducted in the certified Chemical Laboratory of Multielement Analysis in the Institute of Inorganic Technology and Mineral Fertilizers, Wrocław University of Technology.

Results of the laboratory analysis were worked out statistically. Before calculating average, standard deviations and statistical study, the highest and most abnormal contents were discarded. Mean concentrations of elements, standard deviations and correlations between elements were calculated. The results concerning the contents of elements within the groups were processed statistically by ANOVA. Student's t-test at a significance level of =0.05 and 0.01 was used to examine the significance of differences between years and regions in the treatments under comparison.

Results

Acceptable level of toxic elements in pollen loads is regulated by Polish Norm [19] and Branch Norm [20] concerning pollen loads designed for consumption for human. In both norms the admissible values of studied elements are the same (HAC).

The study revealed that various amounts of elements of toxic properties, including heavy metals, were present in pollen loads.

Arsenic has strong toxic properties, which is why its presence in food products is undesirable. Permissible concentrations of that element in pollen loads was determined on a level that does not exceed 0.20 mg·kg⁻¹ (i.e. 0.22 mg·kg⁻¹ d.m.). The present study revealed that mean values obtained were considerably below acceptable. In the agricultural woodland region they were <0.025 mg·kg⁻¹ d.m. in 2005 and 0.036 mg·kg⁻¹ d.m. in 2006 (Table 3). In pollen loads from the airfield, the level of arsenic accumulation was higher and reached 0.093 in 2005 and 0.062 mg·kg⁻¹ d.m. in 2006 (Table 3). No statistically significant differences in arsenic levels between years and regions of the study were observed. Minimal content of arsenic in 2005 and 2006 was below 0.025 mg·kg⁻¹ d.m. The level of arsenic in 2005 was 1.300 mg·kg⁻¹ d.m., and 0.220 mg·kg⁻¹ d.m in 2006 in a single sample of pollen load from an agricultural woodland region without particular reason. In contrast, in pollen from the airfield a level of 1.517 mg·kg⁻¹ d.m. was determined in a sample from 2006 (Table 3). The rejection of the highest values of As content in pollen from the woodland did not cause the change in qualification of differences between years. However, differences in arsenic content in pollen from the airfield region between years appeared to be statistically significant at a level of p≤0.01. Also, statistically significant differences were shown between regions (Table 3).

Mercury, similar to lead, arsenic and cadmium, is an element frequently met in the environment with strong toxic properties. Results of the present study demonstrated that the level of mercury concentration in pollen, regardless of its origin, was low (Tables 1 and 2). Mean content of that metal in pollen loads from the agricultural woodland region was 0.0038 in 2005 and 0.0036 mg·kg⁻¹ d.m. in 2006, respectively (Table 3). A higher concentration of that metal,

Table 1. Concentration of chosen elements in pollen loads from agricultural-woodland region.

	Content in mg·kg-1 d.m.									
Sample No.	As		Cd		Hg		Pb			
	2005	2006	2005	2006	2005	2006	2005	2006		
1.	≤0.025	≤0.025	0.380	0.170	0.004	0.002	0.520	0.200		
2.	≤0.025	≤0.025	0.090	0.120	0.002	0.002	0.045	0.340		
3.	1.300	0.220	0.280	0.160	0.001	0.002	0.550	0.360		
4.	≤0.025	≤0.025	0.130	0.410	0.004	0.003	0.580	0.390		
5.	≤0.025	≤0.025	0.280	0.410	0.005	0.003	0.760	0.980		
6.	≤0.025	≤0.025	0.330	0.340	0.007	0.003	0.810	0.560		
7.	≤0.025	≤0.025	0.210	0.093	0.003	0.002	0.360	0.620		
8.	≤0.025	≤0.025	0.260	0.660	0.003	0.003	1.400	0.440		
9.	≤0.025	≤0.025	0.240	0.480	0.002	0.003	0.280	0.450		
10.	≤0.025	≤0.025	0.180	0.320	0.002	0.005	0.210	0.460		
11.	≤0.025	≤0.025	0.220	0.170	0.003	0.005	0.330	0.800		
12.	≤0.025	≤0.025	0.049	0.300	0.002	0.003	1.300	0.270		
13.	≤0.025	≤0.025	0.029	0.530	0.002	0.003	0.310	0.440		
14.	≤0.025	≤0.025	0.290	0.068	0.004	0.002	0.540	0.950		
15.	≤0.025	≤0.025	0.280	0.024	0.002	0.005	0.490	0.600		
16.	≤0.025	≤0.025	0.068	0.130	0.002	0.002	0.190	0.380		
17.	≤0.025	≤0.025	0.150	0.220	0.005	0.002	0.520	0.440		
18.	≤0.025	≤0.025	0.022	0.190	0.003	0.004	0.480	0.810		
19.	≤0.025	≤0.025	0.270	0.092	0.003	0.006	0.830	0.550		
20.	≤0.025	≤0.025	0.260	0.150	0.010	0.006	2.600	0.880		
21.	≤0.025	≤0.025	0.340	0.220	0.002	0.004	0.300	0.180		
22.	≤0.025	≤0.025	0.240	0.260	0.003	0.005	0.630	0.380		
23.	≤0.025	≤0.025	0.250	0.480	0.006	0.004	0.330	0.360		
24.	≤0.025	≤0.025	0.058	0.380	0.006	0.008	1.300	0.370		
25.	≤0.025	≤0.025	0.280	0.100	0.006	0.004	3.900	0.420		
26.	≤0.025	≤0.025	0.420	0.300	0.004	0.004	0.770	0.300		
27.	≤0.025	≤0.025	0.460	0.280	0.004	0.004	1.200	0.370		
28.	≤0.025	≤0.025	0.260	0.460	0.004	0.005	0.670	0.770		
29.	≤0.025	≤0.025	0.160	0.170	0.002	0.004	0.280	0.140		
30.	≤0.025	≤0.025	0.310	0.380	0.006	0.004	1.500	0.600		
31.	≤0.025	0.220	0.440	0.110	0.009	0.006	3.000	0.340		
32.	≤0.025	0.043	0.200	0.290	0.002	0.003	0.300	0.250		
33.	≤0.025	≤0.025	0.290	0.530	0.003	0.003	0.290	0.280		
34.	≤0.025	≤0.025	0.140	0.052	0.004	0.004	0.720	0.500		
35.	≤0.025	≤0.025	0.090	0.440	0.004	0.003	0.260	0.810		
36.	≤0.025	≤0.025	0.470	0.290	0.002	0.002	0.340	0.470		
LOQ	≤0.025	≤0.025	≤0.001	≤0.001	≤0.0001	≤0.0001	≤0.01	≤0.01		

 $LOQ-level\ of\ quantification.$

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Table. 2. Concentration of chosen elements in pollen loads from the region of a former military airfield.

Sample No.	Content in mg·kg ⁻¹ d.m.									
	As		Cd		Hg		Pb			
	2005	2006	2005	2006	2005	2006	2005	2006		
1.	0.099	0.108	0.318	0.303	0.0055	0.0097	0.587	0.630		
2.	0.085	0.112	0.095	0.566	0.0041	0.0069	0.550	0.189		
3.	0.087	1.417	1.798	0.448	0.0069	0.0083	0.607	3.559		
4.	0.082	0.108	0.277	0.395	0.0055	0.0069	0.967	0.548		
5.	0.099	0.027	0.166	0.751	0.0124	0.0014	0.717	0.329		
6.	0.103	0.025	0.360	0.514	0.0124	0.0028	0.530	0.712		
7.	0.109	0.030	0.636	0.448	0.0083	0.0055	0.580	1.259		
8.	0.092	0.033	0.166	0.632	0.0124	0.0014	0.542	0.329		
9.	0.095	0.055	0.180	0.514	0.0083	0.0014	0.597	0.356		
10.	0.082	0.045	0.387	0.408	0.0041	0.0041	0.520	0.767		
11.	0.085	0.065	0.622	0.303	0.0041	0.0069	0.516	1.232		
12.	0.087	≤0.025	0.113	0.290	0.0041	0.0069	0.797	0.225		
13.	0.096	0.028	0.484	0.171	0.0041	0.0069	0.499	0.958		
14.	0.079	0.065	0.318	0.263	0.0028	0.0041	0.540	0.630		
15.	0.087	0.105	0.041	0.158	0.0041	0.0069	0.522	0.082		
16.	0.109	0.110	0.498	0.105	0.0041	0.0097	0.550	0.986		
17.	0.106	0.025	0.470	0.435	0.0055	0.0028	0.526	0.931		
18.	0.085	≤0.025	0.360	0.253	0.0083	0.0041	0.657	0.712		
19.	0.092	0.050	0.066	0.473	0.0041	0.0028	0.549	0.131		
20.	0.098	≤0.025	0.055	0.374	0.0083	0.0069	0.587	0.110		
21.	0.079	≤0.025	0.539	0.330	0.0083	0.0055	0.665	1.068		
22.	0.084	0.056	0.152	0.628	0.0055	0.0055	0.361	0.301		
23.	0.084	0.165	0.401	0.429	0.0028	0.0069	0.649	0.794		
24.	0.082	≤0.025	0.024	0.374	0.0069	0.0097	1.078	0.047		
25.	0.084	0.103	0.401	0.529	0.0069	0.0055	1.037	0.794		
26.	0.082	0.070	0.526	0.429	0.0028	0.0138	0.647	1.040		
27.	0.104	0.094	0.913	0.341	0.0041	0.0069	2.137	1.807		
28.	0.085	0.049	0.024	0.253	0.0055	0.0069	0.885	0.047		
29.	0.104	0.069	0.360	0.242	0.0069	0.0069	0.470	0.712		
30.	0.077	0.115	0.003	0.143	0.0014	0.0124	2.997	0.007		
31.	0.077	≤0.025	0.055	0.220	0.0014	0.0014	0.700	0.110		
32.	0.179	≤0.025	1.024	0.132	0.0427	0.0028	1.151	2.026		
33.	0.084	< 0.025	0.318	0.088	0.0041	0.0069	0.530	0.630		
34.	0.087	0.104	0.360	0.363	0.0028	0.0097	1.338	0.712		
35.	0.079	0.093	0.037	0.518	0.0028	0.0028	0.602	0.074		
36.	0.107	0.038	0.249	0.242	0.0028	0.0041	2.867	0.493		
LOQ	≤0.025	≤0.025	≤0.001	≤0.001	≤0.0001	≤0.0001	≤0.01	≤0.01		

 $LOQ-level\ of\ quantification.$

	Content in mg·kg ⁻¹ d.m.								
Specification	As		Cd		Hg		Pb		
	agrwood.	airfield	agrwood.	airfield	agrwood.	airfield	agrwood.	airfield	
HAC	0.200		0.050		0.033		0.500		
Year 2005									
Average	0.025 A	0.093 B **	0.234 a	0.356 в	0.0038 a	0.0066 в	0.804 *	0.838	
Sd	0.00	0.018	0.120	0.347	0.002	0.0068	0.824	0.608	
min.	0.025	0.077	0.022	0.003	0.001	0.0014	0.045	0.361	
max.	1.300	0.179	0.470	1.798	0.010	0.0427	3.900	2.997	
% of samples >HAC	2.8%	0	91.7%	86.1%	0	2.8%	55.6%	91.7%	
				Year 2006		,			
Average	0.036 a	0.062 b **	0.272 a	0.363 в	0.0036 A	0.0059 в	0.491 *	0.704	
Sd	0.045	0.038	0.159	0.161	0.001	0.0030	0.219	0.686	
min.	0.025	0.025	0.024	0.088	0.002	0.0014	0.140	0.007	
max.	0.220	1.517	0.660	0.751	0.008	0.0138	0.980	3.559	
% of samples >HAC	5.6%	2.8%	9.27%	100%	0	0	33.3%	58.3%	

Table 3. Concentration of chosen elements in pollen from agricultural-woodland and former military airfield regions.

HAC - Highest Acceptable Concentration.

amounting to 0.0066 and 0.0059 mg·kg¹ d.m., respectively, was observed in pollen from the airfield (Table 3). These values were considerably below the acceptable norm (0.033 mg·kg¹ d.m.). Only in one pollen sample from the airfield was a higher concentration of mercury detected (0.0427 mg·kg¹ d.m.). Statistically significant differences in content of mercury between regions at a level of p≤0.05 in 2005 and p≤0.01 in 2006 were determined (Table. 3).

Lead is presently recognized as the most commonly occurring heavy metal. The analysis of results obtained in the present study point out that the mean level of lead in pollen loads coming from the airfield in both years considerably exceeded permissible concentrations [Branch Norm 1989] and was 0.835 mg·kg⁻¹ d.m. in 2005 and 0.704 mg·kg⁻¹ ¹ d.m. in 2006 (Table 3). In pollen from the agricultural woodland the determined amount of lead in 2005 was 0.804 mg·kg⁻¹ d.m. However, in 2006 its level was 0.491 mg·kg⁻¹ d.m. and was on the threshold of an acceptable norm (Table 3). In 2005 acceptable norms of lead content were exceeded in 55.6% of samples from the agricultural woodland and in 91.7% of samples from the airfield, while in 2006 in 33.3% and 58.3% of samples, respectively. Differences in lead content in pollen from the agricultural woodland region between years appeared to be statistically significant at a level of p \leq 0.05.

The present study demonstrated that mean content of cadmium in pollen considerably exceeded the permissible norm (0.05 mg·kg-1 d.m.) – Tables 1 and 2. In samples from 2005, the mean reached a value of 0.234, and in 2006 - 0.272 mg·kg⁻¹ d.m. (Table 3). Maximal content of that element in analyzed pollen loads was 0.480 and 0.660 mg·kg⁻¹ d.m., respectively. Only in three samples from 2005 and in one from 2006 was the content of cadmium below the acceptable norm. Even higher concentrations of that element were detected in pollen from the airfield. The mean values were at a level of 0.356 and 0.363 mg·kg⁻¹ d.m., respectively (Table 3). In the first year of a study cadmium concentration was below the permissible norm in five samples, and in the next year such samples were not observed. In a range of studied regions no statistically significant differences in a level of accumulation of cadmium between years were observed. However, in given years of study, statistically significant differences between regions at a level of $p \le 0.05$ were determined (Table 3).

Overall, it may be stated that statistically significant correlations were demonstrated between the level of arsenic and lead, cadmium and mercury while between lead and mercury only in the first year of study (Table 4).

A, B – significant differences on a level of p≤0.01 between regions,

a, b – significant differences on a level of p≤0.05 between regions,

^{* –} significant differences on a level of p≤0.05 between years,

^{** –} significant differences on a level of p≤0.01 between years,

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Discussion

Bee products can be contaminated by different sources. Environmental contaminants are the heavy metals like lead, cadmium and mercury [21]. Heavy metals present in the atmosphere can be brought back to the hive through pollen, or flower nectar or honeydew [16, 22]. The degree of contamination of pollen by the different contaminants was studied.

Concentrations of arsenic in pollen should not cause any toxicological problem, as the present study (Tables 1 and 2), and works by other authors point out the low level of that element. Falco et al. [23] demonstrated that mean content of that element in pollen originating from Poland was at a level of 0.04 mg·kg⁻¹, while maximal value was 0.19 mg·kg⁻¹. Similarly the low level of arsenic content was demonstrated by Roman [24] in the early study -0.013-0.194 mg·kg⁻¹ d.m. and only in an individual sample was it 0.934 mg·kg⁻¹ d.m. Contamination by arsenic of bees and bee products may be a problem in regions of the chemical industry and non-ferrous metals industry, and honey and propolis are especially susceptible [25]. The unprofitable influence of that element was observed by Konopacka et al. [26], who revealed that the presence of chemical contaminations may disqualify pollen from trade turnover. The phenomenon of elevated arsenic concentration in bodies of bees and poisoning caused by that metal in the neighborhood of a zinc plant was demonstrated by Nikodemska and Patryn [27].

Similarly, the low level of mercury concentration was demonstrated in the present study (Tables 1 and 2). Analogically, Bratkowski and Wilde [28] have shown that the content of mercury in pollen (bee bread) collected in northwestern Poland at a level of 0.00143 mg·kg¹ d.m. Żarski et al. [29] and Roman [24] detected the content of mercury in pollen on a comparable level – in a range from 0.0009 to 0.0098 mg·kg¹ d.m. Considerably higher concentrations of mercury in pollen, also coming from Poland, was observed by Falco et al. [22] – mean value of 0.01 mg·kg¹, with maximal value of 0.04 mg·kg¹, i.e. exceeding permissible norms. In turn; Čermaková [8] in bees bread originating in different regions of Slovakia observed mercury content at levels from 0.0003 to 0.0110 mg·kg¹.

The content of lead in pollen collected in 2001 was determined by Čermaková [8] on a considerably lower level compared to the present study (Tables 1 and 2). Čermaková [8] obtained from 0.228 to 0.280 mg·kg⁻¹. However, in samples of pollen from 1991, the author [8] determined manyfold higher content of that metal, in a range from 1.510 to 3.240 mg·kg⁻¹. Similarly, Konopacka et al. [26] in their study observed elevated content of lead at a level of 1.20 mg·kg-1 d.m. in pollen obtained from apiaries located close to a motorway. Free et al. [17] proved that in a region of elevated lead level in soil, the content of that element in pollen was higher (2.50-3.80 mg·kg⁻¹), and lower in regions poorer in that metal – 1.10 mg·kg⁻¹. Loper et al. [30] conducted a study close to a motorway, and they obtained the content of lead in pollen in a range of 6-15 mg·kg⁻¹. In turn, Roman [24 and 31] revealed an average level of lead below 1.00 mg·kg⁻¹ d.m. (0.659-0.910 mg·kg⁻¹ d.m.) in collected pollen

Table 4. Values of correlation coefficient determined between particular elements.

	Values of coefficient (r)					
Elements	Season 2005 (n=36)	Season 2006 (n=36)	Total (n=72)			
As - Pb	-0.053	0.516**	0.277*			
As - Cd	0.065	0.076	0.075			
As - Hg	-0.233	0.178	0.101			
Pb - Cd	0.272	0.079	0.035			
Pb - Hg	0.691**	0.079	0.152			
Cd - Hg	0.265	0.224	0.252*			

^{* -} value of correlation coefficient significant on a level of p≤0.05,
** - value of correlation coefficient significant on a level of p≤0.01.

loads. These values were considerably higher than in honey. Bogdanov et al. [32] reports an average of 0.037 mg·kg¹, as well as Przybyłowski and Wilczyńska [33] show the average at a level 0.048 mg·kg¹. Jabłoński et al. [7] analyzed the level of pollution with heavy metals of nectar, honey and pollen coming from plants growing by a busy road demonstrated that the level of lead in pollen loads (irrespective of site of origin) exceeded the acceptable norm, and its highest content (1.31 mg·kg¹ d.m. on average) was in pollen obtained from bee colonies situated just by the road. However, in a region regarded as "clean one," the mean was 0.48 mg·kg¹ d.m. Conti and Botre [34] demonstrated relatively low concentration of lead in pollen, in a range from 0.020 to 0.332 mg·kg¹.

Cadmium possesses the ability to accumulate, especially in soil. Due to the presence of cadmium in pesticides and mineral fertilizers, its concentration in rural regions may be significant [35 and 36]. For that reason, in bee products (including pollen coming from agricultural-woodland regions) the elevated concentration of that element is observed - even up to 0.660 mg·kg⁻¹ d.m. [24, 31]. Numerous authors have pointed out that the level of cadmium in most controlled apiaries exceeded many times the acceptable norms [37]. The level of cadmium in pollen analogous to that obtained in the present study (Tables 1 and 2) was determined by Čermaková [8] – from 0.076 to 0.330 mg·kg⁻¹. Jabłoński et al. [7] demonstrated that cadmium was present in all analyzed samples and they noted its highest level in pollen loads collected in an apiary situated close to a road - to 0.181 mg·kg⁻¹ d.m. Konopacka et al. [26], conducting research in the region of Puławy and Gliwice, observed that cadmium concentrations in samples of pollen was high (0.26 mg·kg⁻¹ d.m.). Lipińska and Zalewski [38] also determined considerable concentrations of cadmium in pollen, in a range of 0.03 to 0.50 mg·kg⁻¹ and in as many as 50% of samples it exceeded permissible norms. However, Szczęsna et al. [39] demonstrated the content of cadmium in pollen in a range of 0.032 to 0.154 mg·kg⁻¹, and only 9% of samples exceeded the acceptable norm. A comparable level of cadmium in pollen was detected by Falco et al. [23] – mean of 0.12, and maximal of 0.21 mg·kg¹. Even lower content of cadmium in pollen coming from a region of Rome, in a range of 0.015-0.0901 mg·kg¹ was demonstrated by Conti and Botre [34]. These values were higher than in honey. Bogdanov et al. [32] observed that cadmium concentration in samples of honey was lower (0.002 mg·kg¹ d.m.). However, Erbilir and Erdoğrul [40] demonstrated the content of cadmium in honey in a range of 0.31 to 0.34 mg·kg¹.

The study demonstrated that the level of chosen elements of toxic property accumulation in pollen was dependent on the region of sample origin. In samples coming from the region of the former airfield higher concentrations of analyzed elements were demonstrated. Especially in the case of cadmium and mercury in comparison to pollen from the agricultural woodland region. A very high level of lead and cadmium accumulation in pollen, independent of the location of sample origin, is of special concern. Arsenic and mercury did not show any tendencies to excessive accumulation levels in pollen. Demonstrated differences between regions in a level of accumulation of chosen elements in pollen loads point out that they may be, as other bee products and bees themselves, applied also as bioindicators of environmental pollution with those elements (Table 3) [8, 29, 33, 34, 41-43].

Conclusions

- The concentration of cadmium, mercury and arsenic (Cd, Hg and As) in pollen loads from the former airfield was indeed statistically higher than from the agricultural woodland region. It was not confirmed in the case of lead statistical differences.
- 2. The highest levels of concentration were observed for cadmium and lead in 91.7 and 97.2% (2005 and 2006 year) of the samples from the agricultural woodland region as well as 86.1 and 100% of samples from the former military airfield region the admissible norms of cadmium were exceeded and suitable lead content was exceeded in 55.6 and 33.3% of samples, as well as 91.7 and 58.3% of samples, respectively.
- 3. Mercury and arsenic appeared to be metals of toxic properties that do not pose any toxicological problem in pollen, irrespective of the region of origin their concentration was at a low level.
- 4. The high content of cadmium in pollen from agricultural woodland deserves special attention.
- 5. Each lot of pollen loads aimed at human consumption needs to be analyzed with respect to toxic contents.

References

DOULL K.M. The relative attractiveness to pollen collecting honeybees of some different pollens. J. Apic. Res., 5, 9, 1976.

- 2. KIM J.H., LEE K.H., KIM E.S., PARK S.B. A study on components of pollen load. Journal of the Korean Society of Food and Nutrition. 21 (5), 566. 1992.
- SZCZĘSNA T. Long-chain fatty acids composition of honeybee-collected pollen. Journal of Apicultural Science. 50, (2), 65, 2006.
- SZCZĘSNA T. Protein content and amino acid composition of bee-collected pollen from selected botanical origins. Journal of Apicultural Science. 50, (2), 81, 2006.
- SZCZĘSNA T. Study on the sugar composition of honeybee-collected pollen. Journal of Apicultural Science. 51, (1), 15, 2007.
- CRANE E. Bees, honey and pollen as indicators of metals in the environment. Bee World. 65 (1), 47, 1984.
- JABŁOŃSKI B., KOŁTOWSKI Z., MARCINKOWSKI J., RYBAK-CHMIELEWSKA H., SZCZĘSNA T., WARAKOMSKA Z. Contamination of nectar, honey and pollen collected from roadside plants. Apicultural Scientific Exercise Books. 2, 129, 1995 [In Polish].
- ČERMAKOVÁ T. Bee Bread a bioindicator of the condition of environment. XL-th Scientific Apicultural Conference, Puławy, 11-12 March, pp. 112, 2003.
- ROMAN A. Bees and their products as pollution bioindicator in the copper (LGOM) and lime-cement (Opole) industry areas. Scientific Exercise Books of Agricultural University in Wrocław. 323, 175, 1997 [In Polish].
- MUSZYŃSKA J., WARAKOMSKA Z. Honeybees in monitoring of the environment's contamination: I. The numerousness of monitoring apiary giving samples of pollen loads, which are representative for a neighborhood rich in pollen flow. Apicultural Scientific Exercise Books. 1, 197, 1999 [In Polish].
- 11. WOLTERS M., MARTENS M.J.M. Effects of air pollutants on pollen. Bot. Rev. **53**, 372, **1987**.
- COMTOIS P., SCHEMENAUER R.S. Tree pollen viability in areas subject to high pollutant deposition. Aerobiologia. 7, 144 1991
- IANNOTTI O., MINCIGRUCCI G., BRICCHI E., FRENGUELLI G. Pollen viability as a bio-indicator of air quality. Aerobiologia. 16, 361, 2000.
- ONORARI M., DOMENEGHETTI M.P., FILIPPELLI N., CIUTI P., MAGRINI S. Pollini a inquinamento atmosferico, w Proceedings of the Congresso Nazionale dell' Associazione Italiana di Aerobiologia, Cagliari, Italy, October 5-8, A.I.A., pp. 159, 2000.
- 15. PORRINI C., MEDRZYCKI P., BORTOLOTTI L., SABATINI A.G., GIROTTI S., GHINI S., GRILLENZONI F., GATTAVCCHIA E., CELLI G. Honeybees as bio-indicators of the environmental pollution. XXXVIII-th Apimondia International Apicultural Congress, Ljubljana, Slovenia, Final Programme and Book of Abstracts, pp. 424, 2003.
- GOTTARDINI E., CRISTOFOLINI F., PAOLETTI E., LAZZERI P., PEPPONI G. Pollen Viability for Air Pollution Bio-Monitoring. Journal of Atmospheric Chemistry. 49, 149, 2004.
- FREE J.B., WILLIAMS I.H., PINSENT R.J.F.H., TOWNSEND A., BASI M.S., GRAHAM C.L. Using foraging honeybees to sample an area for trace metals. Environment International. 9, 9, 1983.
- GÓRECKA H., GÓRECKI H., DOBRZAŃSKI Z. An application of plasma spectrometry ICP-OES and ICP-MS for metal Content analysis in biological and environmental samples. Chemistry for Agriculture. 2, 359, 2001.
- Polska Norma PN-R-78893: "Pollen loads"; The Journal of Norms and the Measures, nr 3/1997, poz. 6, 1997 [In Polish].

20. Norma Branżowa BN – 89/9161-06: "Pollen loads"; The journal of Norms and the Measures, nr 11/1989, poz. 28, 1989 [In Polish].

- BOGDANOV S. Contaminants of bee products. Apidologie, 37, 1, 2006.
- ILLARIONOV A.I., DERKACZ A.A. Xenobiotics in bees and bee's products. Agrochimia. 3, 85, 2008 [In Russian].
- FALCO G., LIOBET J.M., ZARĘBA S., KRZYSIAK K., DOMINGO J.L. Risk assessment of trace elements intake through natural remedies in Poland. Trace elements and Electrolytes. 22 (3), 222, 2005.
- ROMAN A. content of some trace elements in fresh honeybee pollen. Polish Journal of Food and Nutrition Sciences. 57, 4 (C), 475, 2007.
- JĘDRUSZUK A. Bees and bee's products as indicators of natural environment pollution. Veterinary Medicine. 6 (43), 352, 1987 [In Polish].
- KONOPACKA Z., POHORECKA K., SYROCKA K., CHABER J. The Content of cadmium, lead, nitrates and nitrites in pollen loads collected from different sites in vicinity of Puławy. Apicultural Scientific Exercise Books. 37 (1), 181, 1993 [In Polish].
- NIKODEMSKA E., PATRYN O. The bees' poisoning the industrial wastes on terrain Katowice province. Veterinary Medicine. 28, (6), 358, 1972 [In Polish].
- 28. BRATKOWSKI J., WILDE J. Chemical and biological pollen contamination. Apiary. 3, 4, 2003 [In Polish].
- ŻARSKI T.P., VALKA J., DĘBSKI B., ŻARSKA H., MAJDECKA T., ARKUSZEWSKA E. Comparison of mercury pollution of bees products in selected regions of Poland and Slovakia in aspect of threats for Consumer. Chemistry and Ecological Engineering. 12, (8), 871, 2005.
- LOPER G.M., STANDIFER L.N., THOMPSON M.J., GILLIAM M. Biochemistry and microbiology of bee-collected almond (*Prunus dulcis*) pollen and bee bread. I. Fatty acids, sterols, vitamins and minerals. Apidologie. 11 (1), 63, 1980
- ROMAN A. Bioaccumulation of selected metals featuring toxic properties in fresh pollen obtained from honey bees in agricultural and forest area. Chemicals in Agriculture and Environment. 8, 196, 2007.
- 32. BOGDANOV S., HALDIMANN M., LUGINBUHL W.,

- GALLMANN P. Minerals in honey: environmental, geographical and botanical aspects. Journal of Apicultural Research and Bee World. **46** (4), 269, **2007**.
- PRZYBYŁOWSKI P., WILCZYŃSKA A. Honey as an environmental marker. Food Chemistry. 74, 289, 2001.
- CONTI M.E., BOTRE F. Honeybees and their products as potential bioindicators of heavy metals contamination. Environmental Monitoring and Assessment. 69, 267, 2001.
- OKRUSZKO H. The ecological results of hitherto exists development of Polish village, "The Eko-development of Country Areas" - the conference materials, "Problematic Exercise Book of Agricultural Sciences Progress PAN", PWN, Warszawa, pp. 410, 1992 [In Polish].
- 36. WINIARSKI A. The artificial fertilizers as potential threat for the man's health and the environment the. "Chemical Danger in Agriculture", The Monograph of Village Medicine Institute, Lublin, 1997 [In Polish].
- MIGULA P., KAFEL A. Change of the bee's products quality from contaminated regions. Polish beekeeping. 3, 15, 1992 [In Polish].
- LIPIŃSKA J., ZALEWSKI W. The Content of microelements and harmful elements in bee products from Poland. Apicultural Scientific Exercise Books. 33, 113, 1989 [In Polish].
- SZCZĘSNA T., RYBAK-CHMIELEWSKA H., ARCIUCH H. Application of anodic stripping voltammetry to the determination of Cd, Pb and Cu in pollen loads. Apicultural Scientific Exercise Books. 37, 171, 1993 [In Polish].
- ERBILIR F., ERDOĞRUL Ö. Determination of heavy metals in honey in Kahramanmaras City, Turkey. Environmental Monitoring and Assessment. 109, 181, 2005.
- CELLI G., MACCAGNANI B. Honey bees as bioindicators of environmental pollution. Bulletin of Insectology. 56 (1), 137, 2003.
- 42. FREDES C., MONTENEGRO G. Heavy metals and other trace elements contents in Chilean honey. Ciencia e Investigacion Agraria. 33 (1), 50, 2006.
- PORRINI C., SABATINI A.G., GIROTTI S., GHINI S., MEDRZYCKI P., GRILLENZONI F., BORTOLOTTI L., GATTAVECCHIA E., CELLI G. Honey bees and bee products as monitors of the environmental contamination. Apiacta, 38, 63, 2003.