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CONTENT OF SOME TRACE ELEMENTS IN FRESH HONEYBEE POLLEN

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Key words: honeybee, pollen, pollen load, lead, cadmium, arsenic, mercury

The aim of the study was to determine the content of arsenic, lead, cadmium and mercury in fresh honeybee pollen. Samples of fresh pollen were collected in a stationary apiary in July and August 2005 and 2006. The samples were to mineralized by microwave method and plasma spectrometry was applied for determination.

An average content of the elements was as follows: lead – 0.910 and 0.490; cadmium – 0.239 and 0.265; arsenic – 0.069 and 0.025, and mercury – 0.004 and 0.0036, respectively (in mg/kg dry mass). Lead concentration exceeded permissible standards (0.50 mg/kg dry mass) in 19 samples (n=29) in 2005 and in 10 samples (n=29) in 2006. Concentration of cadmium was high and only in three samples did not exceed the permissible standard (0.05 mg/kg). Concentrations of mercury and arsenic were very low.

INTRODUCTION

Pollen, as a natural plant product, is exposed to different contaminations absorbed by plants from the soil through root system or with water intake, as well as to numerous pollutants of different origin, including anthropogenic ones, deposited directly on pollen. Heavy metals concentration in pollen reflects the safety state of the environment it comes from. Therefore, numerous authors consider pollen as a bioindicator of environment, including the atmosphere, pollution [Wolters & Martens, 1987; Comtois & Schemenauer, 1991; Roman, 1997; Iannotti *et al*., 2000; Onorari *et al*., 2000; Gottardini *et al*., 2004].

There is strict relationship between the level of hazardous elements accumulation in soil and their content in bee products [Free *et al*., 1983; Roman, 1997]. There are numerous reports concerning the level of heavy metals concentration in bee products, including pollen obtained in the form of pollen load [Crane, 1984; Ćermaková, 1997].

The aim of the work was to determine the content of lead, cadmium, arsenic and mercury in fresh honeybee pollen collected in agricultural and forestry areas.

MATERIALS AND METHODS

Samples of fresh pollen were collected in a stationary apiary in middle-west part of Opole region with the use of pollen trap with a 5-mm mesh screen, in July and August 2005 and 2006. Pollen samples were collected 10 times in each bee colony, and one cumulative sample, representative for a particular colony, weighing about 100 g, was created. This way, 29 cumulative samples of pollen load were collected each year (58 collective samples in total).

Samples of pollen load were dried at 40°C, unified and thoroughly mixed. Portions of 1000±0.10 mg were dissolved in 20 mL of concentrated, spectrally pure nitric acid (Merck) and then mineralized using microwave method involving high pressure (microprocessor station MD-2000 CEM-USA). Arsenic, lead, cadmium and mercury concentration was determined [Górecka, 1995] using plasma spectrometer Varian ICP-AES with mass detection, P-3202 computer controlled, cooperating with analytic device Philips Scientific PU-7000 model and ultrasonic nebulizer CETAC-5000 AT.

Statistical analysis was performed using Statgraphics 5.1. software, and Duncan test was applied to evaluate differences.

RESULTS AND DISCUSSION

Permissible content of chemical elements featuring toxic properties in pollen loads is set by the Polish Norm [PN-R-78893:1997] and Trade Norm concerning the pollen loads [BN-89/9161-06:1989].

Permissible content of arsenic in pollen loads was specified on a level below 0.20 mg/kg (*i.e.* 0.22 mg/kg dry mass). The arsenic content (Table 1) amounted to 0.069 mg/kg dry mass in 2005 and 0.025 mg/kg dry mass in 2006 and was lower than the standard. In the both analysed years, minimum arsenic content in pollen loads was on a level of 0.015 and 0.013 mg/kg dry mass. Only in a single sample from 2005 the content of arsenic was 0.934 mg/kg dry mass. Also other authors determined low pollen contamination with arsenic. Falco *et al.* [2005] proved that average content of this metal in pollen from Poland was 0.04 mg/kg (max. 0.19 mg/kg). Bees and bee products contamination with arsenic can be a significant

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problem in the region of chemical industry and non-ferrous metals processing, which especially affects honey and propolis [Jędruszuk, 1987]. Negative effect of this chemical element was also noticed by Konopacka *et al*. [1993] who observed that the presence of chemical contaminants can disqualify pollen as trade turnover commodity. Kirkor [1953] described the case of chronic intoxication with arsenic compounds in bee colonies in the region of Cresson. Similar phenomenon of elevated arsenic concentration in bee bodies and poisoning caused by this metal in the vicinity of zinc works was reported by Nikodemska & Patryn [1972].

Lead is one of the most common heavy metal occurring in the environment. Lead level (Table 1) in pollen loads in 2005 (0.910 mg/kg dry mass) considerably exceeded permissible standard [BN-89/9161-06:1989]. However, in 2006 lead concentration decreased to a level of 0.490 mg/kg dry mass, what is close to permissible limit value. Maximum content of this metal in pollen collected in 2005 amounted 3.900 mg/kg dry mass and in 2006 - 0.980 mg/kg dry mass (Table 1). Differences between lead content in particular years proved to be statistically significant ($p<0.01$). Some authors evaluated the degree of environmental pollution on the basis of examination regarding bees and their products contamination, including pollen. Konopacka *et al.* [1993] detected elevated lead concentration (1.20 mg/kg dry mass) in pollen collected in farm experimental apiaries localized near a busy road in the region of Puławy and Gliwice. Free *et al*. [1983] proved that in the region of higher lead content in soil, the concentration

TABLE 1. Content (mg/kg dry mass) of the elements in honeybee pollen.

Number of the sample	Pb		Cd		As		Hg	
	2005	2006	2005	2006	2005	2006	2005	2006
1.	0.520	0.950	0.380	0.068	0.029	0.017	0.0037	0.0015
2.	0.550	0.300	0.280	0.300	0.934	0.194	0.0009	0.0035
3.	0.580	0.770	0.130	0.460	0.025	0.015	0.0043	0.0049
4.	0.760	0.360	0.280	0.160	0.025	0.022	0.0046	0.0024
5.	0.810	0.810	0.330	0.190	0.021	0.015	0.0066	0.0038
6.	1.400	0.340	0.260	0.120	0.019	0.025	0.0029	0.0016
7.	0.280	0.440	0.240	0.660	0.025	0.02	0.0021	0.0030
8.	0.330	0.180	0.220	0.220	0.025	0.013	0.0027	0.0041
9.	1.300	0.420	0.049	0.100	0.025	0.021	0.0018	0.0042
10.	0.310	0.270	0.029	0.300	0.031	0.021	0.0024	0.0029
11.	0.540	0.550	0.290	0.092	0.042	0.013	0.0042	0.0064
12.	0.490	0.880	0.280	0.150	0.025	0.018	0.0023	0.0060
13.	0.190	0.380	0.068	0.130	0.025	0.020	0.0024	0.0023
14.	0.520	0.600	0.150	0.024	0.015	0.022	0.0048	0.0046
15.	0.480	0.450	0.022	0.480	0.022	0.024	0.0028	0.0025
16.	0.830	0.140	0.270	0.170	0.029	0.025	0.0027	0.0035
17.	2.600	0.360	0.260	0.480	0.068	0.033	0.0098	0.0040
18.	0.630	0.560	0.240	0.340	0.039	0.017	0.0032	0.0029
19.	0.300	0.380	0.340	0.260	0.025	0.014	0.0022	0.0050
20.	0.330	0.390	0.250	0.410	0.041	0.020	0.0064	0.0026
21.	1.300	0.980	0.058	0.410	0.025	0.029	0.0058	0.0029
22.	0.770	0.370	0.420	0.280	0.053	0.013	0.0042	0.0038
23.	1.000	0.800	0.460	0.170	0.044	0.018	0.0041	0.0052
24.	0.670	0.370	0.260	0.380	0.062	0.019	0.0039	0.0079
25.	0.280	0.200	0.160	0.170	0.138	0.014	0.0023	0.0023
26.	3.900	0.440	0.280	0.220	0.031	0.020	0.0064	0.0022
27.	0.210	0.460	0.180	0.320	0.028	0.016	0.0022	0.0047
28.	1.500	0.620	0.310	0.093	0.072	0.014	0.0059	0.0024
29.	3.000	0.440	0.440	0.530	0.056	0.025	0.0092	0.0026
$\bar{x}$	**0.910^{A}**	**0.490^{B}**	**0.2392**	**0.2651**	**0.069**	**0.025**	**0.0040**	**0.0036**
SD	0.8764	0.2267	0.1190	0.1589	0.168	0.033	0.0021	0.0015
Standard[1)]	0.50		0.05		0.20		0.02	

A-B – statistically significant differences at $p \leq 0.01$ between years; [1)] – Polish Standard PN-R-78893:1997 and Trade Norm BN-89/9161-06:1989

of this element in pollen was higher (2.50-3.80 mg/kg) and in the regions poorer in this metal – it was lower (1.10 mg/kg). Loper *et al.* [1980] conducted their research near the highway and recorded lead content in pollen on a level of 6-15 mg/kg. Zalewski & Szymaniuk [1985] reported lead content values lower than 1.00 mg/kg dry mass in collected pollen loads. Investigating the contamination with heavy metals of nectar, honey and pollen coming from plants growing close to busy roads Jabłoński *et al.* [1995] proved that lead amount in pollen loads (regardless their place of origin) exceeded permissible standard and its highest level (1.31 mg/kg dry mass on average) was found in pollen coming from bee colonies situated very close to the road. However, on the area assumed as a "clear" an average lead concentration was 0.48 mg/kg dry mass. Conti & Botre [2001] proved relatively low lead concentration in pollen, ranging from 0.020 to 0.332 mg/kg.

An average concentration of mercury amounted to 0.004 in 2005, and 0.0036 mg/kg dry mass in 2006 (Table 1). Those values can be classified below permissible standard (0.022 mg/kg dry mass). Bratkowski & Wilde [2003] reported mercury content in pollen (bee bread) collected in the north-western part of Poland at a level of 0.00143 mg/kg dry mass. Significantly higher mercury content in Polish pollen, *i.e*. exceeding permissible standards, was recorded by Falco *et al.* [2005] (av. 0.01, max. 0.04 mg/kg).

Due to cadmium presence in plant pesticides and mineral fertilizers, its concentration in rural areas can be considerably high. Therefore, in bee products including pollen coming from agricultural and forestry areas, high concentration of cadmium was observed. The average concentrations of this metal (Table 1) in pollen loads significantly exceeded permissible standard (0.050 mg/kg dry mass). In the samples from 2005, an average cadmium level was 0.239, and 0.265 mg/kg dry mass in 2006. Maximum content of this element in the examined pollen loads ranged from 0.480 to 0.660 mg/kg dry mass. Only in three samples, cadmium concentration was below permissible standard. There were no statistically significant differences in the level of cadmium between the years of examination.

Numerous authors point the fact that cadmium level in most examined apiaries many folds exceeded permissible standards [Migula & Kafel 1992]. Jabłoński *et al.* [1995] reported that cadmium was present in all the examined pollen samples and its highest level (0.181 mg/kg dry mass) was recorded in pollen loads collected in the apiary situated near the road. Konopacka *et al.* [1993] determined the cadmium content in pollen samples in the region of Puławy and Gliwice on a level of 0.26 mg/kg dry mass. Lipińska & Zalewski [1989] also detected considerable cadmium concentration in pollen, ranging from 0.03 to 0.50 mg/kg dry mass, and in as many as 50% of samples cadmium values were higher than permissible standard. However, Szczęsna *et al.* [1993] reported that among detected concentrations of cadmium in pollen from 0.032 to 0.154 mg/kg, only in the case of 9% of samples permissible standard was exceed. Comparable to mentioned cadmium level in pollen was recorded by Falco *et al.* [2005] – 0.12 mg/kg on average, while the maximum value was 0.21 mg/kg. Lower (0.015 to 0.0901 mg/kg) cadmium concentration in pollen originating from the area of Rome was reported by Conti & Botre [2001].

Summing up the research, it was demonstrated that cadmium and lead are the elements that may possess some toxicological problem in pollen. However, arsenic and mercury did not show any tendencies to an excessive accumulation in pollen.

CONCLUSIONS

1. The examined elements (arsenic, lead, cadmium and mercury) were determined in all samples of pollen loads.
2. Higher level of lead, arsenic and mercury was found in 2005 and that of cadmium in 2006.
3. Lead concentration exceeded permissible standards in 65.5% of samples in 2005, and in 34.5% samples in 2006.
4. High contamination with cadmium was observed in pollen from agricultural and forestry area.
5. Contents of mercury and arsenic do not pose any toxicological problem - their concentrations in pollen from agricultural and forestry area were low.

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ZAWARTOŚĆ WYBRANYCH PIERWIASTKÓW ŚLADOWYCH W ŚWIEŻYM PYŁKU KWIATOWYM

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Zakład Higieny Zwierząt i Środowiska, Uniwersytet Przyrodniczy we Wrocławiu

Celem badań było określenie stopnia akumulacji wybranych pierwiastków śladowych (arsen, ołów, kadm i rtęć) w świeżym pyłku kwiatowym. Próby świeżego pyłku kwiatowego, pozyskiwane w pasiece stacjonarnej, w lipcu i sierpniu 2005 i 2006 roku. Próbki zmineralizowano techniką mikrofalową, natomiast analizę ilościową badanych metali wykonano techniką spektrometrii plazmowej.

Średnie zawartości badanych metali w roku 2005 i 2006 kształtowały się odpowiednio (w mg/kg s.m.): ołowiu - 0,910 i 0,490; kadmu - 0,239 i 0,265; arsemu - 0,069 i 0,025 oraz rtęci - 0,004 i 0,0036. Koncentracja ołowiu w 2005 r. w 19 próbkach (n=29), a w 2006 r. w 10 próbkach (n=29) przekroczyła dopuszczalne normy (0,50 mg/kg s.m.). Koncentracja kadmu w pyłku pochodzącym z rejonu rolniczo-leśnego była wysoka i tylko w trzech próbach nie przekroczyła dopuszczalnej normy. Zawartości rtęci i arsenu okazały się bardzo niskie.