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MINERAL MICRONUTRIENTS COMPOSITION OF BEE'S POLLEN

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Abstract

It were analyzed the total metals contents for some bee's pollen samples collected from Berini, Banat County. Thirteen metals were quantified for each pollen samples. The analysed minerals are: Na, K, Ca, Mg, Fe, Mn, Zn, Cu, Cd, Ni, Co, Pb and Cr. Mineral contents were determinated by flame atomic absorption spectrometry (F-AAS) with high-resolution continuum source ContrAA 300 spectrometer. The results were statistically interpreted using multivariate analyses.
***Keywords**: minerals, pollen, flame atomic absorption spectrometry, multivariate analyses.*

Introduction

Pollen is the male genetic material of flowers and represents the main source of normal non-liquid feed for bees (Mondal, 1998).

Bee pollen is used for its therapeutic properties. Daily ingestion of bee pollen is recommended because it is capable to regulate the function of intestines and has benefits on cardiovascular system, skin and vision. It is used also in chronic prostates treatment for its presumed anti-inflammatory and anti-androgenic effects (Narajo, 2004).

The metals contents of pollen are variable, due to the factors like differences between the plants species, geographical area and conditions of drying process (Kabata, 2001).

In this paper were analyzed the total metals contents for some bee's pollen samples collected from Berini, Banat County, thirteen metals being quantified for each pollen samples.

Experimental

Pollen Samples: Ten bee pollen samples were collected in different months of the year 2006 from Berini, Banat County: **Pollen 1** – harvested time: 20-30 Mars; **Pollen 2** – harvested time: 1-10 April; **Pollen 3** – harvested time: 11-20 April; **Pollen 4** – harvested time: 21-29 April; **Pollen 5** – harvested time: 30 April-5 May; **Pollen 6** – harvested time: 6-15 May; **Pollen 7** – harvested time: 16-30 May; **Pollen 8** – harvested time: 31 May-10 June; **Pollen 9** – harvested time: 11-25 June; **Pollen 10** – harvested time: 26 June-5 July.

Pollen samples preparation: The metals from bee pollen samples were analyzed after dry burning of 10 g in the quartz capsules at 650°C for 4 hours. After complete burning a nitric acid 0.5 N solution was added up to 50 mL. The solutions obtained were used for total metals contents determination by flame atomic absorption spectrometry (F-AAS) with high-resolution continuum source.

Reagents: The standard solutions (1000 mg/L) were analytical grade from Riedel de Haen (Germany). The nitric acid 65% solution used was of ultra pure grade (Merck, Germany). All solutions were prepared using deionized water.

Metals determination from bee pollen: Analysis of metals was made with ContrAA-300, Analytik-Jena device, by flame atomic absorption spectrometry (FASS) in air/acetylene flame. The device working parameters (air, acetylene, optics and electronics) were adjusted for maximum absorption for each element. Acetylene was of 99.99 % purity. Under the optimum established parameters, standard calibration curves for metals were constructed by plotting absorbency against concentration (Gergen, 2006). In a definite range for each metal a good linearity was observed. The correlation coefficient for the calibration curves (r^2) ranged between 0.9745 - 0.9891. All analyses were made in triplicate and the mean values were reported. All the values obtained for metals contents in pollen samples were calculated in mg/kg pollen.

Statistical interpretation of data obtained using multivariate analyses was performed with Statistica-6 software.

Results and Discussion

The results obtained for contents in *Sodium*, *Potassium*, *Calcium* and *Magnesium* for analyzed pollen samples are presented in Table 1.

Table 1. Sodium, Potassium, Calcium and Magnesium contents for pollen from Berini

Harvesting time	Na (ppm)	K (ppm)	Ca (ppm)	Mg (ppm)
Pollen1	111	4086	2166	702
Pollen2	341	5220	2244	965
Pollen3	322	4914	1323	762
Pollen4	251	5020	1165	952
Pollen5	209	4291	1357	749
Pollen6	287	4994	1476	764
Pollen7	152	3991	2026	749
Pollen8	24	5175	2336	943
Pollen9	91	4463	2312	722
Pollen10	7	4222	2162	744

Sodium and *Potassium* are both alkaline metals and have very important roles in many physiological processes. *Sodium* is present in extra cellular fluids in animals and humans. It is responsible for depolarization of cellular membrane and for the water equilibrium in intra- and extra cellular medium. Potassium is involving in muscles contraction, in lipids metabolism, in proteins synthesis, maintaining the fluid and electrolyte balance in the body and is responsible in the nerve impulses sending. The necessary daily intake of *K* is between 2-4 g/day (Mogos, 1997). The highest *Na* and *K* contents were determinated for Polen 2 (341 ppm for Na, respectively 5220 ppm for K). The smallest quantity for *Na* was obtained in Pollen 10 (7 ppm) and for *K* in Pollen 7 (3991 ppm).

Magnesium and *Calcium* are both alkaline-earth metals and are essential for animals and humans, particularly in cell physiology. Magnesium is present in many enzymes involved in proteins, lipids and carbohydrates metabolisms. *Magnesium* deficiency in humans caused muscle spasms, and has been associated with a high blood

pressure, many cardiovascular diseases, diabetes and osteoporosis. The necessary daily intake is 350 mg/day for men and 300 mg/day for women. A deficit in *Calcium* can affect the formation of bone and tooth, but the excess retention can cause kidney stones. *Calcium* deficiency for a long time is responsible for osteoporosis, when the bones are deteriorated and increased the risk of fractures. The necessary daily intake is between 350 and 1100 mg/day (Mogos, 1997). In pollen samples from Berini the highest content for *Ca* was obtained in Pollen 8 (2336 ppm) and for *Mg* in Pollen 2 (965 ppm). The smallest content in *Ca* was determinated for Pollen 4 (1165 ppm) and in *Mg* for Pollen 1 (702 ppm).

The results obtained for contents in *Cooper*, *Zinc*, *Manganese, Iron, Nickel, Cadmium, Cobalt, Chromium* and *Lead* contents for analyzed pollen samples are presented in Table 2.

Table2. *Cooper*, *Zinc*, *Manganese, Iron, Nickel, Cadmium, Cobalt, Chromium* and *Lead* contents for pollen from Berini

Harvesting time	Cu (ppm)	Zn (ppm)	Mn (ppm)	Fe (ppm)	Ni (ppm)	Cd (ppm)	Co (ppm)	Pb (ppm)
Pollen1	9.4	37.4	17.0	42.9	1.9	0.0	0.0	0.11
Pollen2	6.0	52.5	120.7	65.8	0.3	0.1	0.01	0.40
Pollen3	8.5	42.1	27.2	65.9	1.0	0.0	0.0	0.14
Pollen4	11.1	38.9	41.3	57.4	1.9	0.0	0.0	0.09
Pollen5	6.7	33.5	18.3	35.4	0.4	0.0	0.0	0.01
Pollen6	10.4	42.2	28.1	74.3	1.8	0.0	0.0	0.22
Pollen7	7.9	36.1	17.1	55.9	1.0	0.0	0.0	0.04
Pollen8	10.4	43.1	25.7	94.4	1.6	0.0	0.0	0.16
Pollen9	10.0	40.9	21.7	61.2	1.3	0.0	0.0	0.14
Pollen10	13.8	47.8	21.3	64.0	1.3	0.0	0.0	0.26
Nat. limit in similar products, ppm	*20.0*	*60.0*	-	-	-	-	-	*1.0*

Cooper is one of several trace heavy metals that are essential to life. The national accepted limit for *Cooper* in similar products is 20.0 mg/Kg (Ordinance 975/1998). For analyzed pollen samples Pollen10 has the highest content in *Cooper* (13.8 ppm), followed by Pollen4 (11.1 ppm). Pollen2 has the smallest *Cooper* concentration (6.0 ppm).

Zinc is a constituent of about 300 enzymes and proteins that participate in all major metabolic processes. The national accepted limit for *Zinc* in similar products is 60.0 mg/Kg (Ordinance 975/1998). The analyzed samples contend *Zinc* in range 33 – 53 ppm. The smallest quantity was determinated for Pollen5 (33.5 ppm) and the higher for Pollen2 (52.5 ppm). All the values obtained for *Zinc* contents are under than national accepted limit for this metal in similar products.

Manganese is both a constituent and an activator of several enzymes and proteins in plant, animal and humans, and has around 20 identified functions. The recommended ESADDI values for adults range from 2 to 5 mg *Mn*/day (Schäfer, 2004). The contents in *Mn* for analyzed samples were in range 17 – 121 ppm.

Iron is an essential element for humans, ranging to approx 4200 mg/body. Approximately 60% of it is bound in hemoglobin and 10% in *Fe*-dependent tissue enzymes. The remaining 20% and 10% are stored as ferritin and respectively hemosiderin (Schümann, 2004). The highest content in *Fe* was obtained for Pollen2 (120.7 ppm).

Excessive soluble *Nickel* compounds are hepatotoxic and nephrotoxic but as aerosols or dusts, insoluble *Ni* compounds or elemental *Ni* are very toxic (carcinogenic), justifying a lot of country imposed restricted limits, 0.05-1 mg/m^3 (Sunderman, 2004). In analyzed samples *Ni* content was in range 0.3-1.9 ppm.

Lead is not an essential element for life and it is very toxic for the nervous system and the kidneys. The national accepted limit for *Lead* in similar products is 1.0 mg/Kg (Ordinance 975/1998). All the values obtained for *Pb* contents are under than national accepted limit for this metal in similar products.

Cadmium inhibits or activates a great enzymes number, like those rich in accessible sulfhydryl groups (Vollenweider, 2006). Only for Pollen2 were determinated *Cd* and *Co* contents (0.01 ppm). Chromium was not detected in the analyzed samples.

For cluster analysis it was used Statistica-6 software. The coefficients of matrix correlation of variables are presented in Table 3.

From the coefficients of matrix correlation of variables result that the values obtained for *Cd* and *Co* contents in analyzed pollen are in very good correlation with those determinate for *Mn* (0.97) and *Zn*

content with *Pb* (0.97). That's way *Cd, Co* and *Pb* were eliminated between the variables.

Table 3. The coefficients of matrix correlation of variables

	Na	K	Ca	Mg	Cu	Zn	Mn	Fe	Ni	Cd	Co	Pb
Na	1.00	0.48	-0.61	0.26	-0.59	0.11	0.55	-0.14	-0.34	0.47	0.47	0.20
K	0.48	1.00	-0.19	0.79	-0.07	0.53	0.58	0.67	0.03	0.43	0.43	0.51
Ca	-0.61	-0.19	1.00	-0.01	0.08	0.41	0.16	0.29	-0.04	0.29	0.29	0.40
Mg	0.26	0.79	-0.01	1.00	-0.11	0.46	0.67	0.49	-0.06	0.54	0.54	0.40
Cu	-0.59	-0.07	0.08	-0.11	1.00	0.15	-0.45	0.33	0.71	-0.53	-0.53	0.07
Zn	0.11	0.53	0.41	0.46	0.15	1.00	0.71	0.56	-0.19	0.69	0.69	0.97
Mn	0.55	0.58	0.16	0.67	-0.45	0.71	1.00	0.17	-0.47	0.97	0.97	0.76
Fe	-0.14	0.67	0.29	0.49	0.33	0.56	0.17	1.00	0.26	0.09	0.09	0.47
Ni	-0.34	0.03	-0.04	-0.06	0.71	-0.19	-0.47	0.26	1.00	-0.58	-0.58	-0.18
Cd	0.47	0.43	0.29	0.54	-0.53	0.69	0.97	0.09	-0.58	1.00	1.00	0.75
Co	0.47	0.43	0.29	0.54	-0.53	0.69	0.97	0.09	-0.58	1.00	1.00	0.75
Pb	0.20	0.51	0.40	0.40	0.07	0.97	0.76	0.47	-0.18	0.75	0.75	1.00

In all analyzed pollen samples the highest values were obtained for *K* content, followed by *Ca, Na* and *Mg*. The distribution of these metals in analyzed pollen is presented in Figure 1.

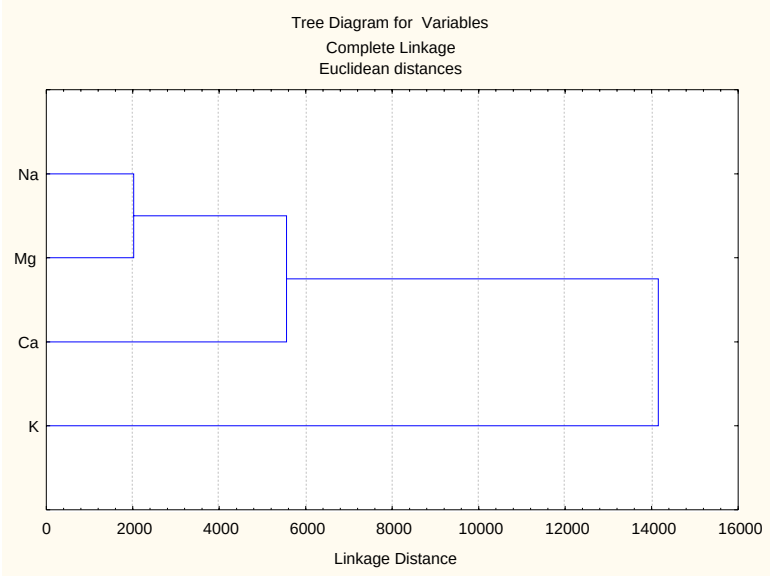

Figure 1. Variables Na, K, Ca and Mg - dendrogram

The highest contents in all analyzed pollen samples were obtained for *Mn* content, followed by *Zn* and *Fe*, *Cu* and *Ni*. The distribution of *Cu, Ni, Zn, Fe* and *Mn* in all analyzed pollen is presented in Figure 2.

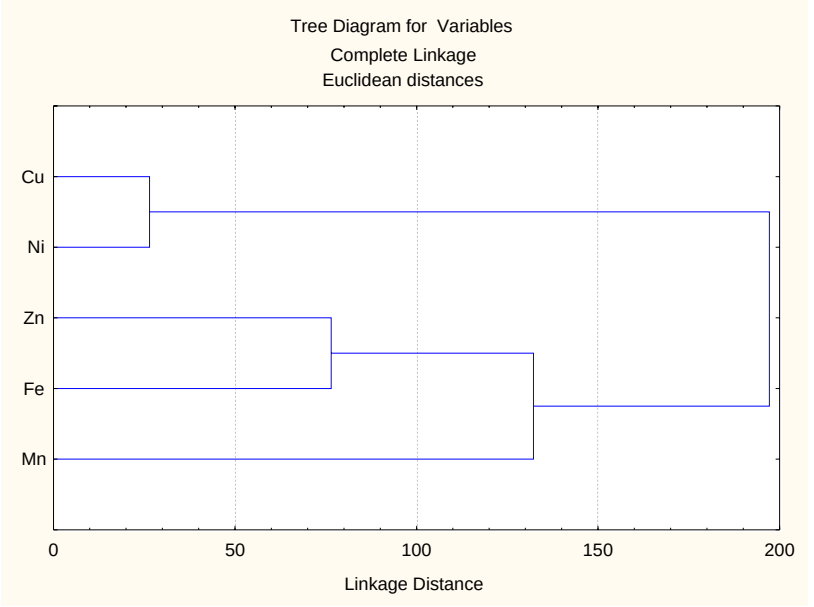

Figure 2. Variables Cu, Ni, Zn, Fe and Mn - dendrogram

Conclusions

In all pollen samples from Berini the highest content was determinated for *Potassium*, followed by *Calcium, Sodium* and *Magnesium*. Between trace metals the highest contents were identified for *Mn* content, followed by *Zn* and *Fe, Cu* and *Ni*.

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*** Excerpt from Romanian Ministry of Public Health Ordinance *no 975/1998*. *Maximum limits accepted for heavy metal in foods*