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Mineral Content of Honey from Serbia: Analysis of Essential and Toxic Elements

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Abstract: Honey is a natural, sweet, and complex substance produced by honey bees from nectar collected from various plants. After processing, nectar is stored in wax honeycombs, forming honey, which contains several hundred chemical compounds responsible for its diverse biological activities. Traditionally consumed as a nutritious food, honey also exhibits antimicrobial, antioxidant, anti-inflammatory, immunomodulatory, antitumor, and probiotic effects, making it significant for both nutritional and therapeutic purposes. In this study, the mineral composition of honey was analyzed using inductively coupled plasma optical emission spectrometry (ICP-OES) to determine the concentrations of micro- and macroelements, some of which are essential for human health, while others are potentially toxic even at low levels. More than 20 elements were quantified, including Al, As, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Mo, Ni, P, Pb, S, Se, Sr, and Zn. Six honey samples from different geographical regions available on the Serbian market were examined: three domestic samples collected from small family farms and three purchased from retail outlets. Due to the complex honey matrix, sample preparation involved dry digestion with mineral acids to enable efficient metal extraction. Cluster analysis was applied to investigate the relationship between the elemental composition and the geographical origin of honey, providing insights into honey quality, authenticity, and potential health risks associated with toxic element content.

Keywords: microelements, dry digestion, ICP-OES, Cluster analysis

1. Introduction

By processing the nectar of plant secretions, honey bees produce honey, which is a natural chemical substance of complex composition [1]. The agricultural branch that deals with the breeding of bees, the production of honey, and other bee products is called beekeeping. The moderately continental climate of Serbia is extremely favorable and supports this type of production. It is estimated that around 500 tons of honey are produced annually in Serbia. The most well-known types of honey produced in this region are acacia (Robinia pseudoacacia), sunflower (Helianthus annuus), and linden

(Tilia cordata) honey, as representatives of monofloral honey, and meadow honey as a representative of polyfloral honey [2].

According to its chemical composition, honey represents a mixture of various organic and inorganic compounds. It mainly (80–85%) consists of sugars, primarily the monosaccharides glucose and fructose, but also oligosaccharides such as anderose and panose. In addition to sugars, organic compounds such as proteins, amino acids (amylase, peroxidase, catalase, acid phosphatase), polyphenols and phenolic compounds, vitamins (B3, B6, B9, C), and many others can be found [1,2]. Inorganic constituents in honey are present in significantly smaller amounts, and it is estimated that their content does not exceed 0.2% [2]. The amount of present inorganic minerals mostly depends on the type of honey and the geographical area where pollen is collected. Minerals present in honey can be essential – those necessary for human health – but also toxic, which indicates soil contamination within the foraging radius of the bee of 7 km^2 [3].

The botanical origin, geographical location, climatic conditions, and the degree of anthropogenic environmental pollution are just some of the factors that influence the elemental composition of honey [1]. In this paper, the essential and toxic elements present in honey from different geographical locations in Serbia and of different botanical origins will be analyzed. Additionally, by applying cluster analysis, the correlation between the analyzed samples and the examined elements will also be investigated.

2. Methodlogy

In this study, six different types of honey originating from various geographical regions of Serbia were analyzed. Table 1 presents detailed information on the investigated honey samples.

Table 1. Overview of the Analyzed Honey Samples by Type and Geographical Origin

Sample	Honey Type	Geographical Location
1	Acacia honey	Avala
2	Acacia honey	Kruševac
3	Meadow honey	Fruška gora
4	Floral extracted honey	Novi Sad
5	Linden honey	Velika Plana
6	Meadow honey	Velika Plana

The samples for analysis were prepared using the dry digestion method, as follows: Approximately 1.25 g of honey was weighed into a porcelain crucible, followed by the addition of 10 mL of 65% HNO_3 and 2 mL of 35% H_2O_2. The mixture was heated in a furnace at 180 °C for 30 minutes. The dry residue was then rinsed with small portions of concentrated HNO_3, filtered, and quantitatively transferred into a 50.0 mL volumetric flask. The concentrations of the elements present in the samples were determined using inductively coupled plasma–optical emission spectrometry, ICP-OES (Agilent model 5800). The elements Al, As, Ba, Ca, Cd, Co, Cr, Cu, Fe, Hg, K, Mg, Mn, Mo, Ni, P, Pb, S, Sr, and Zn were analyzed at their characteristic wavelengths.

Cluster analysis, one of the methods of multivariate chemometric analysis, was performed for parameter reduction and data grouping based on similarity. The HCA Ward linkage method was used, and the analysis was carried out using Python 3.13.3.

3. Results and Discussion

Table 2 presents the results of the determination of essential and non-essential elements in honey samples collected from the territory of the Republic of Serbia.

Table 2. Results of the mineral composition analysis of six different types of honey

Results	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6
Al	n.d.	48.4	4.3	n.d.	n.d.	1.7
As	n.d.	1.0	1.2	0.7	1.2	n.d.
Ba	17.6	1235.0	17.4	2.4	5.6	1.0
Ca	113.8	97.1	108.5	219.5	186.4	159.0
Cd	0.1	0.2	0.1	0.3	0.1	0.2
Co	0.4	1.7	0.8	0.4	0.4	0.4
Cr	0.9	3.5	0.2	4.8	0.3	n.d.
Cu	6.6	20.6	5.3	2.8	2.5	2.2
Fe	75.5	95.4	58.4	60.4	15.0	27.3
Hg	0.8	1.6	0.5	2.3	1.0	1.5
K	294.8	235.2	264.4	385.5	1096.5	666.4
Mg	19.2	18.8	23.5	35.7	38.8	40.4
Mn	0.7	0.6	0.6	11.6	0.9	0.6
Mo	0.9	1.0	0.8	1.0	1.2	1.3
Ni	36.2	88.6	18.0	17.2	3.6	3.2
P	28.2	35.9	27.8	46.6	55.0	65.5
Pb	5.5	1.8	0.3	7.2	1.4	0.8
S	28.6	192.5	55.2	35.7	55.5	58.0
Sr	0.7	0.9	0.6	0.8	0.8	0.7
Zn	631.3	84.7	685.0	8.5	7.1	5.9

*n.d. – not detect

Elemental analysis showed significant variations in certain honey samples. The greatest variations that can be identified are for Ba in sample 2, for K in sample 5, for Zn in samples 1 and 3, and for S in sample 2. Other elements are present in approximately similar concentrations. The highest amounts were detected for the macroelements K > Ca > S > P > Mg, while the others were detected in significantly lower amounts. Elements detected in amounts less than 2 mg/kg are: Cd, As, Co, Sr, Mo, Hg, Cr, and Mn.

Figure 1 shows a dendrogram representing the clustering of the obtained results based on the similarity in metal content among the honey samples. In subfigure a), the clustering of samples is presented, while subfigure b) displays the clustering of elements.

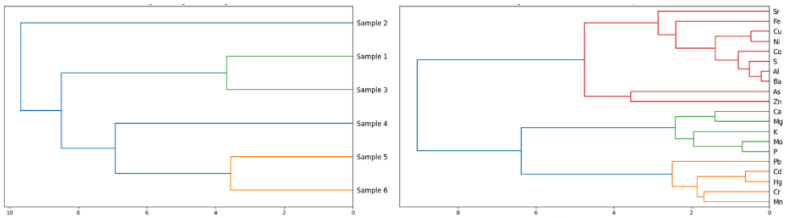

Figure 1. Dendrogram of a) samples and b) elements, using HCA and Ward linkage method

Based on the cluster analysis, the honey samples were grouped into three clusters. Samples 4, 5, and 6 belong to the first cluster, samples 1 and 3 belong to the second cluster, while sample 2 belongs to the third cluster. The first cluster consists of samples with moderate content of essential and low content of toxic elements. Samples 5 and 6, although belonging to different types of honey, showed the most similar elemental composition, which can be associated with their geographical location. The second cluster is characterized by its high nutritional value and high Zn content, while the third cluster is characterized by a high content of S and Ba. This sample is also characterized by an increased presence of other toxic elements, which may indicate the presence of industrial pollution in the vicinity.

Clustering of elements based on their concentrations in the honey samples also resulted in three groups. The first cluster consisted of toxic trace elements: Pb, Cd, Hg, Cr, and Mn. The second cluster mainly included macroelements: Ca, Mg, K, Mo, and P. The third cluster included: Sr, Fe, Cu, Ni, Co, S, Al, Ba, As, and Zn. Ovi elementi This distribution indicates that certain elements tend to co-occur depending on their which can be linked to both natural and anthropogenic factors affecting the samples.

4. Conclusions

This study analyzed micro- and macroelements in six different honey samples. The obtained results show that the following elements were detected in higher concentrations in most samples: K > Ca > S > P > Mg, while toxic elements such as Cd, As, Co, Hg, Pb, and Cr were mostly detected at concentrations below 2 mg/kg. Cluster analysis successfully identified the similarity and grouping of the samples into three clusters, as well as the grouping of the elements also into three clusters. It was also shown that the content of heavy metals in honey samples is more influenced by geographical origin than by the type of honey.

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References

[1] R. Ullah, F.A. Jan, H. Gulab, S. Saleem, N. Ullah, Wajidullah, Metals contents in honey, beeswax and bees and human health risk assessment due to consumption of honey: A case study from selected districts in Khyber Pakhtunkhwa, Pakistan, Arch. Environ. Contam. Toxicol., 82 (2022) 341–354.

[2] M. Jovetić, J. Trifković, D. Stanković, D. Manojlović, D. Milojković-Opsenica, Mineral content as a tool for the assessment of honey authenticity, J. AOAC Int., 100 (2017) 862–870.

[3] H. Bayir, A. Aygun, Heavy metal in honey bees, honey, and pollen produced in rural and urban areas of Konya Province in Turkey, Environ. Sci. Pollut. Res., 29 (2022) 74569–74578