ASPECTS REGARDING THE LEVEL OF HEAVY METALS IN DIFFERENT HONEY VARIETIES

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Abstract: Heavy metals content in honey represents a decisive factor in estimating its quality. Because their toxicity for human health, EU Standards (European Honey Directive of the European Honey Commission) as well as Romanian Legislation had proclaimed an accepted level of heavy metals in honey. The sources of heavy metals residues were established to be the soil, plant or results from processing and environmental contamination .The heavy metals content in honey is determined by some variables such as weather, season or the flowers botanical origin. The location of the beehives close to roads with heavy car and trucks traffic, industrialized zones or unacceptable materials and equipment of the honey production technology can lead to honey contamination with heavy metals. The purpose of this study is to establish a link between the heavy metal content in honey varieties and the botanical origin of the processing flowers from regions with high or low degrees of pollution in Timis County. The investigated locations are: Timişoara, Timişoara- near a national road, Chevereşu Mare, Pischia, Otvesti and Sacoşu Mare. The goal of this research was to establish the heavy metals concentration such as lead, cadmium, chromium, zinc, manganese and iron in Linden Tree, Mixed Flowers, Rape, Sunflower, Amorpha and Accacia honey samples proceeded straightly from the bee colonies. The heavy metals content in the honey samples was established by using standard methods, as atomic absorption spectrometry (AAS), approved by Romanian STAS 784/2-2009. The obtained results show a variable heavy metal content depending on the floral sources of the honey variety. The highest lead content was established for Linden Tree honey, especially when the hives were located in places with heavy car traffic. The highest chromium content was determined for the Linden Tree honey samples collected from hives placed near a national road. High iron content was measured for the Mixed Flowers honey samples, content which increased for Linden Tree honey samples collected from hives placed near a national road. High cadmium and zinc levels were measured in the Linden Tree honey samples. This research shows that placing the beehives near a polluted area increased the content of cadmium and zinc for all honey varieties.

Keywords: Linden Tree honey, Mixed Flowers honey, rape honey, sunflower honey, amorpha honey, accacia honey, heavy metal content, honey variety.

INTRODUCTION

Honey, obtained by bees from flower nectar or plant secretions like honeydew, which they collect, transform and combine with specific substances, is considered to be a nutritious food [1]. Honey contains water, glucose, fructose, sucrose, dextrin, vitamins of group B, C, pro vitamin A, folic acid, enzymes, polyphenols, flavonoids and mineral salts of Calcium, Sodium, Potassium Phosphorus, Aluminum, Iron, Silica, Magnesium [2].Discovered health effects of honey are among others, immunity boosting, anti-inflammatory, antimicrobial, antiviral and antifungal effects, provided by its complex nature [3]. Therefore, honey is recommended by nutritionists for consumers of all ages. Consumed for its health benefits, the honey composition, must be checked in order to avoid toxic compounds found in honey, such as hydroxymethylfurfural, grayanotoxin, plant alkaloids, heavy metals or other contaminating substances [4, 5, 6].

The presence in honey of heavy metals is a result of honey contamination during the production process, environmental factors or contaminated plants used for processing the honey [7]. When honey is processed from nectar of aromatic plants, well-known to concentrate heavy metals, the heavy metals content in honey is also high. These results allow the use of heavy metals content in honey as an indicator of environmental pollution [8]. The heavy metals level in honey can be linked with beehives location and the entire circuit of honey production beginning with the botanical origin of the flowers. Important heavy metals sources

are the air and soil from locations with intense industry and traffic where beehives are placed, which can contaminate the bee colony and its products [9].

Heavy metal contamination of honey can be the result of various activities such as agricultural practices, several industry branches or waste dump, which pollute the environment. Another heavy metal contamination source for honey is the use of pesticides, OCs, piretrines and piretroids used in plant treatments as agriculture practices [10] and of apicultural procedures, like acaricides and organophosphorus pesticides (OPPs), used for the control of Varroa jacobsonie and Ascosphera apis, pesticides for wax moth and small hive beetle control, antibiotics and other drugs for the treatment of bee diseases and migration from wax to honey [11]. Pesticides used for increase the crop production are linked with residues containing heavy metals, find in plants and other substrates attractive to honey bees, like nectar, pollen or aphid honeydew, reduce the acreages of useful plants for the bee activity[12]. The presence of pesticides residues in nectar plants, on their surface or applied on crops as a pest control measure can affect the hives colonies by killing or altering the physiological functions of bees [13].

Exceeding the heavy metals content in honey established by the European Honey Directive can become dangerous for the consumers health [14]. High levels of heavy metals in honey may appear because their presence in the atmosphere, deposited on the hairy bodies of bees, brought back to the hive with pollen or absorbed together with the nectar of the flowers and through the water or the honeydew [15]. In order to obtain bee or beehive products of high quality without heavy metals content are to be considered: the weather (rain and wind can clean the atmosphere or transfer heavy metals to other environmental sectors), the season (the nectar flow, which is usually greater in spring than in summer and autumn, could dilute the pollutant), the botanical origin of the honey (the nectar of flowers with an open morphology and the honeydew are much more exposed to pollutants)[15]. Therefore since honey composition depends on several factors around the beehives, the presence of heavy metals in honey is related to geographical and botanical origins [16, 17].

The aim of this experiment was to determinate the content of heavy metals in several honey samples collected from bee colonies located in Timis County and to trace the connection between the heavy metal content found in different varieties of honey with the beehives location in areas with different degrees of pollution and the botanical origin of the processing flowers.

MATERIAL AND METHOD

The main objective of this study was to establish the heavy metals content, such as Lead (Pb), Cadmium (Cd), Chromium (Cr), Zinc (Zn), Iron (Fe) and Manganese (Mn) in honey samples collected directly from bee colonies in Timis County and also to establish the dependence between the amount of heavy metals found in different varieties of honey samples from contaminated and pollution free area and the botanical origin of the processing flowers. Therefore the pursued researches tried to point out the relationship between the concentrations of the chosen heavy metals found in several varieties of honey collected from bee colonies placed in urban areas with intense industry and heavy car traffic, rural areas with and without intensive crops plantations and natural forest areas.

The honey samples analyzed in this study were harvested from beehives located in Timiş County, collected from several apiaries and private beekeepers having their beehives located in Timisoara, Cheveresu-Mare, Sacosu Mare, Otveşti and Pişchia. The studied honey varieties are: Acacia, Amorpha, Rape, Linden Tree, Sunflower and Mixed Flowers. The location of the hives mentioned above remained the same during the research. The variety of the honey samples were studied in the same conditions and using the same procedures, having the following location: Acacia is harvested near a natural forest in Sacosu-Mare; Acacia I is harvested also near Sacosu Mare, from another beekeeper; Acacia II is harvest near a forest in Pischia; Amorpha harvested in rural areas of Cheveresu-Mare and Otvesti; Rape and Sunflower harvested in rural areas with intensive crops plantations from Cheveresu-Mare and Otvesti; Linden Tree is harvest near Sacosu-Mare and Linden Tree P1 in Timisoara near a highway; Mixed Flowers is harvested in Cheveresu-Mare and Mixed Flowers P1 is harvested

in Timisoara near a national road. Each honey sample harvested from the locations above was made up as a mixture of ten honey samples collected from each beehive of the same location mentioned above.

The heavy metals content in the honey samples was analyzed by using atomic absorption spectrometry (AAS), as a standard method for detecting heavy metals. To determinate the level of the selected heavy metals: Lead (Pb), Cadmium (Cd), Chromium (Cr), Zinc (Zn), Iron (Fe) and Manganese (Mn) atomic absorption spectrometry (AAS) was used for their detection [8].

The placement of the beehives made possible to study the influence of the mentioned pollution sources on the quality of honey, by analysing the heavy metals content. In order to establish the contamination degree with heavy metals of the studied honey samples, International Standarts (Codex Alimentarius, Standard of F.A.O./ W.H.O. Commision) [14] were used .

RESULTS AND DISCUSSIONS

The results obtained by analyzing the variety of honey samples produced in Timis County showed a variable content of heavy metals in all the studied samples. The influence of the honey variety and location of the beehives on the levels of metals found in the honey samples are showed in Table 1 and Table 2.

The data presented in table 1 show that the lead content in the analysed honey samples lies between 0,21-0,86 mg/kg. The highest concentration (0,86 mg/kg) was found in honey samples of Linden Tree harvested in Timisoara and the lowest value (0,21mg/kg) in Amorpha honey from beehives located in Cheveresu-Mare. The values for the cadmium concentration alters from 0,23-0,72 mg/kg having the highest concentration again in the Linden Tree honey harvested in Timisoara and the lowest one for the Acacia honey with beehives located in Sacosu-Mare. The chromium content registered the largest scale of concentration values, namely 0,14-1,21 mg/kg. Again the highest value of 1,21 mg/kg cadmium was found in honey samples of Linden Tree harvested in Timisoara and the lowest content 0,14 mg/kg in rape honey harvested in Otvesti.

Analysing the heavy metals content of the honey samples a link between the honey variety and the location of the beehives in areas with different degrees of pollution was established. Namely, the highest lead concentration was found in the Linden Tree honey samples (0,86 mg/kg Pb) followed by the Accacia honey variety (0,62 mg/kg Pb). The highest cadmium and chromium content was established again for the Linden Tree honey variety (0,72 mg/kg Cd, 1,21 mg/kg Cr) and lower but close values were found in the honey samples of Mixed Flowers (0,69 mg/kg Cd, 0,85 mg/kg Cr).

Table 1

Impact of beehives location and honey variety on the content of lead, cadmium and chromium in honey

Beehives location	Honey variety	Lead content	Cadmium	Chromium
		(mg/kg)	content (mg/kg)	content
				(mg/kg)
Timisoara	Mixed Flowers P1	$0,45\pm0,3$	$0,69\pm0,4$	0.85 ± 0.5
Timisoara	Linden Tree P1	0.86 ± 0.3	$0,72\pm0,5$	1,21±0,6
Cheveresu-Mare	Amorpha	$0,21\pm0,1$	0.39 ± 0.2	$0,27\pm0,3$
Cheveresu-Mare	Mixed Flowers	0.31 ± 0.1	$0,27\pm0,2$	$0,44 \pm 0,5$
Cheveresu-Mare	Rape	0.35 ± 0.3	0.30 ± 0.2	$0,10\pm0,1$
Cheveresu-Mare	Sunflower	0.38 ± 0.3	0.31 ± 0.2	0.19 ± 0.1
Otvesti	Amorpha	0.39 ± 0.3	$0,41\pm0,3$	0.30 ± 0.3
Otvesti	Rape	$0,47\pm0,3$	0.32 ± 0.3	0.14 ± 0.2
Otvesti	Sunflower	$0,42\pm0,3$	0,35±0,2	$0,22\pm0,1$
Sacosu-Mare	Acacia	$0,58\pm0,2$	$0,23\pm0,1$	0.51 ± 0.3
Sacosu-Mare	Acacia I	$0,44 \pm 0,2$	$0,45\pm0,3$	$0,92\pm0,6$
Sacosu-Mare	Linden Tree	$0,62\pm0,4$	0.32 ± 0.2	0.33 ± 0.3
Pischia	Acacia II	$0,30\pm0,1$	0.36 ± 0.2	0.81 ± 0.5

⁻ Data are presented as mean \pm standard deviation

As seen in table 2 the values of the zinc content registered in honey are between 2,15-7,69 mg/kg,having the highest concentration established for Mixed Flower honey obtained in Timisoara and only 2,15 mg/kg for the rape honey produced in beehives from Cheveresu-Mare.

Table 2
Influence of beehives location and honey variety on the content of zinc, iron and manganese in honey

Beehives location	Honey variety	Zinc content (mg/kg)	Iron content (mg/kg)	Manganese content (mg/kg)
Timisoara	Mixed Flowers P1	$7,69\pm0,9$	8,58± 0,9	$1,65\pm0,3$
Timisoara	Linden Tree P1	$6,93\pm0,8$	15,61± 0,9	$2,87\pm0,3$
Cheveresu-Mare	Amorpha	$3,58\pm0,5$	$7,59\pm0,8$	0.81 ± 0.5
Cheveresu-Mare	Mixed Flowers	$4,59\pm0,5$	9,16± 0,9	$1,53\pm0,5$
Cheveresu-Mare	Rape	2,15±0,8	5,87± 0,9	$0,62\pm0,5$
Cheveresu-Mare	Sunflower	$3,95\pm0,8$	6,25± 0,9	$0,98\pm0,7$
Otvesti	Amorpha	3,92±0,4	$7,84\pm0,9$	$1,02\pm0,5$
Otvesti	Rape	2,60± 0,2	6,21± 0,7	$0,76\pm0,5$
Otvesti	Sunflower	4,02± 0,3	6,71±0,8	$1,13\pm0,5$
Sacosu-Mare	Acacia	2,26± 0,2	4,18± 0,7	$0,38\pm0,2$
Sacosu-Mare	Acacia I	3,15±0,4	4,60± 0,8	$0,69\pm0,4$
Sacosu-Mare	Linden Tree	$5,66\pm0,5$	8,47± 0,9	2,36± 0,4
Pischia	Acacia II	6,86± 0,9	10,18± 0,9	$0,82\pm0,5$

- Data are presented as mean \pm standard deviation

The highest iron concentration of 15,61 mg/kg was detected in Linden Tree honey harvested in Timisoara, unlike 4,18 mg/kg the lowest level for Acacia honey with beehives located in Sacosu-Mare. The manganese content of the studied honey samples lies between 0,38-2,87 mg/kg, having the highest value again for the honey samples of Linden Tree harvested in Timisoara and the lowest concentration of 0,38 mg/kg for Acacia honey with beehives located in Sacosu-Mare.

The results in table 2 spotlight again the influence of the honey variety on the heavy metal content in honey. The highest zinc content was determinated for Mixed Flowers honey $(7,69 \, \text{mg/kg Zn})$ followed by Linden Tree honey $(0,93 \, \text{mg/kg Zn})$ and close by Acacia honey $(6,86 \, \text{mg/kg Zn})$. The presence of iron in the analysed honey samples has the following decreasing order: Linden Tree $(15,61 \, \text{mg/kg Fe})$, Acacia $(10,18 \, \text{mg/kg Fe})$ and Mixed Flowers $(9,16 \, \text{mg/kg Fe})$. The manganese content in the honey samples is much lower, meaning $2,87 \, \text{mg/kg}$ Mn for Linden Tree and $1,65 \, \text{mg/kg}$ Mn for Mixed Flowers.

CONCLUSIONS

In order to show the impact of pollution on the selected heavy metals content of honey, three areas with different pollution degrees were choosen to locate the beehives. An urban area with high air pollution generated by several industry branches, heavy car traffic and other anthropogenic activities represented by Timisoara, capital of Timis County, a rural area with a lower pollution because of intensive agricultural practices and crops plantations represented by Cheveresu-Mare and Otvesti and a forest area with considerable natural forests and plantation crops in Sacosu-Mare and Pischia.

The present investigation of heavy metals content in honey samples revealed the importance of the honey variety and beehives location in polluted area on the contamination with heavy metals of the honey. The results show that the highest level of lead (0,86 mg/kg) was detected in the Linden Tree honey samples with beehives located in Timisoara. The same honey samples registered the highest content of cadmium (0,72mg/kg) and chromium (1,21mg/kg). All three levels of heavy metals exceeds by far the permissible values of heavy

metal (0,20 mg/kg) admited by the International Standarts [14]. Analysing the highest values of the zinc content (7,69mg/kg) for Mixed Flowers honey, iron content (15,61 mg/kg) for Linden Tree honey and manganese content (2,87mg/kg) for Linden Tree honey, it can be observed that once again they where detected in the honey samples produced by the beehives located in Timisoara, an area with high pollution degree and exceed the values admitted by the International Standarts [14]. Another important aspect revealed by this study is that the lowest levels of the selected heavy metals in honey were detected in beehives located in the forest areas of Sacosu-Mare and Pischia, areas considered to have very low air pollution. Based on the assessment of heavy metals contamination, the levels of the selected heavy metals in honey samples are comparatively the highest for Linden Tree honey and Mixed Flowers honey followed by Acacia honey from urban areas (Timisoara), lower in rural areas (Cheveresu-Mare, Otvesti) and the lowest in forest areas (Sacosu-Mare, Pischia).

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