Research Article

Determination of heavy metal pollution in honey samples collected from Ardahan and counties

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Abstract

In this study the concentrations of some elements in 180 honey samples were investigated. Samples were obtained from beekeepers of all the counties of Ardahan province (Center, Hanak, Çıldır, Göle, Damal) in 2015. The levels of Al, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Na, Ni, Pb, Sr and Zn elements in honey samples were determined by ICP-OES instrument. The samples were digested in microwave oven using nitric acid and hydrogen peroxide. The mean concentration of elements and the lowest and highest values were determined. Of the samples analysed; Cd, Co, Cr and Pb values were found below the detection limit. As a result of analysis, the concentration of elements in honey samples were detected as 124,863 ± 313,44 ppb, 1227,56 ± 892,22 ppb, 67,352 ± 34,636 ppb, 6484,904 ± 2078,892 ppb, 302,551 ± 323,329 ppb, 4,636 ± 3,943 ppb, 3118,69 ± 835,149 ppb and 10,535 ± 14,73 ppb, for Al, Ca, Fe, K, Mg, Mn, Na and Sr respectively. To conclude, the concentrations of detected heavy metals in collected honey samples were below the maximum residue limits of some international residue limits. Therefore it would not pose a risk to human health.

Key Words: ICP-OES, honey, element, pollution, Ardahan

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Introduction

Beekeeping is one of the most ancient and widespread production activities in Turkey. Turkey has rich vegetation and different climatic zones and an important potential for bee colony existence, so beekeeping is an important activity for Turkey (Boluktepe and Yilmaz, 2008; Parlakay et al., 2008).

Honey is among the most ancient nutrients. It has been used as an important source of food supplying energy and used instead of drugs in many diseases for thousands of years (Samarghandian et al., 2017). The mineral content of honey is stated to be low (Mendes et al., 2006). The average mineral content of nectar originated honey have been determined as % 0,169, on the other hand the secretion honey kinds are rich in mineral content (Samarghandian et al., 2017). It is possible to detect elements like calcium, phosphor, chlorine, sulphur, magnesium, manganese, silica, sodium and most particularly potassium in honey. Besides, trace elements such as copper, zinc, iron, iodine, which are important for living beings, can be found in small amounts in honey (Mendes et al., 2006; Samarghandian et al., 2017).

The concentration of metals in honey are affected from some factors. Primarily, the mineral rates vary in flower honey kinds just according to the herbal source (Herrero et al., 2017). The mineral rate in any plant shows dependency on the soil that plant grows up, material inside the soil, kind of the plant and to the environmental factors during the manuring and development period (Kaya and Pirincci, 2002). In addition, the acidity of the soil affect the mineral rate in plants. Infact the plants growing in acidic soils cause Pb, Mn, Fe, and Zn poisoning, and the calcic and the high pH rates of the soil reduce solubility of these elements (Vicil et al., 2012).

Techniques using bioindicators have gained importance in recent years in the identification and observation of environmental pollution. Up to the acquired scientific datas, choosing a relevant organism a bioindicator constitutes an important step of the observation activity. Because bioindicator living beings are beings responding differently against various polluting agents, they can hold the polluting agents for a long time by storing them in their bodies (Conti and Botre, 2001). Honey bees are good biological indicators. Bees show the chemical deterioration in environment by the high death rates in their population or storing the polluters in their bodies. With a proper laboratory analysis the polluters which cause pollution on bees and bee products can be identified (Porrini et al., 2003).

It has been able to produce pretty good quality of honey in different parts of Turkey. Furthermore, it has been stated that %75 percent of the required plant flora for honey is available in Turkey (Sancak et al., 2013). Beekeeping has been stated to be among the occupational sectors which are important and have economical dimension for Ardahan Province. The number of the people dealing with beekeeping is % 0,2 percent of Turkey in Ardahan province (Aygun and Akbulak, 2017).

The aim of this study is to detect and make comparison of some element concentrations in honey samples collected in Ardahan province which is known to let beekeeping carried out intensively in Turkey. The data that obtained from the study will help to show the level of enviromental pollution, and evaluate the importance of this subject in terms of public health.

Material and Method

The honey samples were collected in 2015 from Ardahan Province and its counties (Centre, Hanak, Posof, Cıldır, Gole and Damal) A total of 180 samples, 30 from each locations, were obtained. 100 grams of honey samples were taken into sample containers, the contact between air and honey were prevented by securing the covers of the sample containers tightly. The samples were stored in dark, under room temperature until the analysis were carried out.

The process of extraction was carried out according to the method advised by Yucel and Sultanoglu (2013), and measurements were actualized in inductively coupled plasma optical emission spectrometry (ICP-OES, Spectroblue, Germany). According to this, 20 honey samples were taken from each sampling field and transferred into the sterilized tubes and in order to prevent crystallization. The samples were taken into the water tank (NÜVE ST 30, Turkey) which is 70 °C average for a while to hemogenize the honey samples. 0,5 g was taken from these samples, then 9 ml (HNO_3) %65 nitric acid and 1 ml %30 hydrogen peroxide were added. The samples were burned in a microwave device (CEM MARS 6 System 240/50, USA). The working conditions of the microwave device were given in Table 1. The burned samples were subtilized with 5 ml of pure water. The prepared blind (Blank) samples were applied with the same process.

Table 1. The working conditions of Microwave Device

Temperature and power	Time
At 70°C 400W	5 min.
At 100°C 800W	5 min.
At 150°C 800W	10 min.
At 200°C 800W	10 min.
Ventilation	10min.

The main stock solution (1000 mg/L) was prepared from ICP Multi-Element standard Solution IV (Merck Millipore, 111355, Darmstadt, Germany) including aluminum (Al), barium (Ba), calcium (Ca), cadmium (Cd), cobalt (Co), chrome (Cr), copper (Cu), iron (Fe), potassium (K), magnesium (Mg), manganese (Mn), sodium (Na), nickel (Ni), lead (Pb) and Strontium (Sr) and Zinc (Zn) which were diluted with citric acid. Calibration standards have been prepared from this main stock solution as 0, 125, 250, 500, 1000, 2500, 5000 µg/L for Ba, Fe, Mg, Na, Ni and for the others as 0, 10, 20, 50, 100, 200, 500, 1000 µg/L. The metal levels in honey were carried out by ICP-OES device in Kırıkkale University Centre for Scientific and Technological Researches and Applications. The device was calibrated with prepared calibration standards. The working conditions of the ICP-OES Device were given on Table 2.

Table 2: The working conditions of ICP-OES device.

Property	Value
Plasma Power:	1430 W
Coolant Flow:	13 L/minute
Nebulizer Flow:	0,75 L/minute
Pump Speed:	30 rpm
Auxiliary Flow:	0,80 L/minute

According to the analyses carried out, the limits of determination for the elements Al, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Na, Ni, Pb, Sr and Zn were detected as 1.77 ppb; 0,112 ppb; 4,44 ppb; 0.0366 ppb; 0.289 ppb; 0.148 ppb; 0.51 ppb; 0.65 ppb; 63,5 ppb; 20,7 ppb; 0.375 ppb; 94 ppb; 0.649 ppb; 1.44 ppb; 1,27 ppb; 0.107 ppb, respectively.

"SPSS 15.0 for Windows" statistic packet programme was used for the statistical calculations. The data were explained by arithmetic mean ± standard deviation. The data were statistically evaluated by one-way analysis of variance (ANOVA). If the *F* values were significant, Duncan's Multiple Range Test was performed to identify the specific differences between the metal accumulation means at a probability level of $P<0.05$.

Results

The means and standard deviation of the ash contents of 180 honey samples for Al, Ca, Fe, K, Mg, Mn, Na and Sr were founds as 124.86±313.44 ppb, 1227.56 ± 892.22 ppb, 67.35 ± 34.63 ppb, 6484.90 ± 2078.89 ppb, 302.55 ± 323.32 ppb, 4,63± 3.94 ppb, 3118.69 ± 835.14 ppb, 10.53 ± 14.73 ppm, respectively. The Cd, Co, Cr and Pb levels of the analyzed samples were all below the detection limit. Ba were found only in samples taken from Çıldır County, Ni were found under determination limit in counties of Cıldır and Göle.

The mean concentrations of minerals in Centre, Cıldır and Damal were as follows: Mn<Sr<Fe<Al<Mg<Ca<Na<K; the mean concentrations of minerals in Göle and Horat and Posof were as follows: Mn<Sr<Al<Fe<Mg<Ca<Na<K. There were statistically significant differences ($p <0,05$) in Al, Ca, Fe, K, Mg, Mn and Na concentrations in terms of mean element levels; but no statistical difference ($p <0.05$) was found for Sr. Statistical comparisons were calculated for regional differences (Table 3).

Discussion

The increase in the industrial activities and the human population cause environmental pollution. The problems due to environmental and food pollution pose a threat to the public health. There can be elemental residue problem at a potentially toxic level in nutrients which are essential for human and living beings. Just because of the mining and industrialization, poisonous materials can join into the environment thereby into the food chain of human and animal. Toxic elements are intensively used in various industrial sectors and used as an agricultural fertilizer (Leita et al., 1996; Vicil et al., 2012).

Just like the animals, the plants need some minerals throughout their progress. Taking these elements in high levels may cause poisonings both in plants and living beings. Plants may store the elements in their structures which are not necessary for their progress and for their growth. The animals feeding with these plants may accumulate these elements in their tissues. This may cause residue problem in animal products. (Kaya and Pirincci, 2002). Throughout their life, the bees are constantly in relation with environment. Thereby bees can be affected from these harmful materials. Consequently the pollutants can be found in honey and other bee products (Carmen and Cristina, 2001).

As the results obtained from this study were surpassed maximum Pb (1 mg/kg) and Cd (0.1 mg/kg) residue limits accepted by European Union and as any kind of residue limit was not determined for other metals, such a comparison was not done.

Different kinds of honey are produced in different regions of Turkey just peculiar to their plant flora. The element content of honey depends on the soil which shelters the plant with the nectar, the period the plant was grown, the climate, season and environmental pollution (Tuzen et al., 2007; Sultanoglu, 2011). The importance of determining the element content of honey have been gaining importance. Thereby, it has been thought that bees and bee products can be important indicators for observation of environmental pollution (Przybylowski and Wilczyńska, 2001; Yucel and Sultanoglu, 2013). In a study carried out to determine the metal concentrations of honey samples taken from different parts of Italy, it was determined that the regional differences did not cause any problem in terms of Pb, Ni and Cr (Porrini et al., 2003).

The results of our study showed that the concentrations of Al was found pretty lower than the results obtained from a study carried out by Van Der Steen et al. (2012). When considered in terms of Cu, the concentration of Cu was lower than the results obtained from a study carried out by Tuzen (2002), Demirezen and Aksoy (2005), Erbilir and Erdogrul (2005), Tuzen and Soylak (2005), Silici et al. (2008) and Van Der Steen et al. (2012). In terms of Fe, the concentrations were lower than the results obtained from a study carried out by Tuzen (2002), Erbilir and Erdogrul (2005), Tuzen and Soylak (2005), Silici et al. (2008) and Saghaei et al. (2012). The concentrations of Mn found in our study was lower than the results obtained from a study carried out by Tuzen (2002), Erbilir and Erdogrul (2005), Tuzen and Soylak (2005), Silici et al. (2008) and Van Der Steen et al. (2012); higher than a study carried out by Saghaei et al. (2012). Ni concentrations was found to be lower than the results of a study carried out by Porrini et al. (2003) and Van Der Steen et al. (2012).

The element concentrations, which are possible to determine in honey, can be affected from climate and seasonal conditions and flora and the physical conditions of the soil. In addition to this it is necessary to take the polluting factors, which can be exposed to in storage conditions and in production conditions, into consideration. The existence of elements in studies carried out upon honey samples in different levels can be attributed to these reasons above.

Conclusions

In this study, the honey samples were evaluated in terms of Al, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Na, Ni, Pb, Sr and Zn. The results show that Cd, Co, Cr and Pb were under the determination limits of ICP-OES device. The results obtained from this study were under the maximum residue limits when compared with some other international limits. The samples analyzed does not pose any kind of danger against human health. Although the levels detected were below the permissible residue limits, beekeepers dealing with beekeeping will be able to have safer and more qualified products by practicing beekeeping activities in fields that can be affected from pollution factors in minimum levels.

In addition to the studies being carried out across the country and the other studies which will be carried out intended to measure metal levels in bees and bee products, this study is qualified enough to help in determination of polluting sources and their distribution into the environment. This study can contribute other studies, which will be carried out in other regions and provinces, in the determination of possible element concentrations in honey.

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Table 3: The concentrations (ppb) of analyzed elements up the counties (Ardahan Merkez, Cıldır, Damal, Göle, Horat, Posof).*

	Al	Ca	Fe	K	Mg	Mn	Na	Sr
Centre	81.18 ±58.55[a]	1327.81±979.31[bc]	67.42±19.17[b]	5948.46 ±1574.51[ab]	248.93 ±92.28[ab]	3.47 ±2.61[ab]	3135.09 ±450.47[b]	8.84±5.14
	34.65-366.71	688.15-5271.15	30.86-105.47	3701.31-10874.1	162.46-699.72	1.22-13.19	2099.3-4433.79	5.04-30.42
Cıldır	60.23 ±22.21[a]	798.55±216.27[a]	45.73±17.84[a]	5081.74 ±1439.31[a]	206.30 ±37.83[a]	2.80 ±2.59[a]	2644.11 ±140.30[a]	6.98±4.44
	35.36-132.94	80.29-1302.32	22.78-78.17	3219.93-8908.79	132.63-261.11	0.98-11.80	2332.12-2909.52	4.28-30.05
Damal	137.69 ±253.79[c]	1185.28±1199.99[ab]	60.52±18.15[ab]	8097.80 ±2340.05[d]	410.11 ±665.63[b]	7.35 ±4.75[d]	3263.5 ±1226.24[b]	14.99±30.87
	43.63-1290.61	218.41-7454.01	31.21-93.04	5191.69-11749	182.03-3915.36	2.44-14.15	2722.4-9675.72	4.57-172.71
Göle	63.20 ±12.41[a]	1148.13±617.49[ab]	72.83±21.62[b]	5493.47 ±653.98[a]	260.24 ± 132.96[ab]	3.13 ±2.35[a]	3182.29 ±819.86[b]	9.10±6.41
	47.28-97.86	740.82-4042.06	37.56-127.64	3631.02-6971.72	153.02-916.34	1.93-15.40	2349.38-7085.13	4.68-36.09
Horat	61.48 ±9.85[a]	1204.12±815.59[ab]	62.49±21.40[ab]	6748.23 ±2402.79[bc]	311.49 ±229.77[ab]	5.27 ±4.63[bc]	3122.34 ±686.69[b]	10.60±10.10
	46.83-88.81	695.28-4261.2	25.58-122.23	3441.21-11596.3	157.82-1217.02	1.07-15.34	2667.41-5442.31	5.22-47.69
Posof	345.37 ±687.74[b]	1699.10±983.92c	95.10±63.87[c]	7506.64 ± 1775.8[cd]	378.22 ±299[ab]	5.78 ±4.02[cd]	3364.72 ±1055.12[b]	12.68±12.43
	54.68-3715.37	897.06-5503.96	32.71-384.94	4520.94-12414.8	193.11-1875.51	1.63-19.35	2676.24-8684.44	6.29-76.60

*mean±Standard deviation and min-max