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Improvement of X-ray fluorescence sensitivity by dry ashing method for elemental analysis of bee honey

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ABSTRACT

Elements, K, Ca, Ti, Cr, Mn, Fe, Ni, Cu, Zn, Rb, and Sr in bee honey samples were determined using an improved dry ashing (DA) method for XRF with Mo-secondary target (Mo-XRF). The sensitivity of the DA method was significantly improved in comparison to the wet ashing (WA) and the direct (D) methods. The limits of detection (LODs) obtained by DA (3.4–0.007 μg/g) method were better by an order of magnitude than those obtained by WA (34.0–0.120 μg/g) and D (61.2–0.270 μg/g) methods. Further improvements in the sensitivity of the DA-XRF were achieved by using a Cu-secondary target for the excitation of the elements of K, Ca, Ti, Cr, and Mn. In this instance, the LODs were in the range of 0.220–0.024 μg/g. The results of DA-XRF analysis revealed a very good accuracy with errors less than 7.1% and a precision with a relative standard deviation (RSD) better than ±8.8%.

The improved DA-XRF analysis was applied for the determination of the above mentioned elements in several Syrian bee honey samples. The results were comparable to those obtained by the atomic spectrometry method with correlation coefficients better than 0.9927.

1. Introduction

During the last decades the importance of trace elements in such different fields as biology [1], medicine [2,3], environmental science [4], and foods [5–7] has been well established. Different analytical methods, including neutron activation analysis (NAA) [7,8], atomic absorption spectrometry (AAS) [9,10], inductively coupled plasma atomic emission or mass spectrometry (ICP-AES, ICP-MS) [11–14], particle-induced X-ray emission (PIXE) and total reflection X-ray fluorescence (TXRF) [2,15–17] were used to determine the trace elements in the previously mentioned matrices. Energy dispersive X-ray fluorescence (ED-XRF) with a combination of different tube and radionuclide sources showed also a reliable determination of a limited number of elements in the biological matrices, including blood [18], plant [19] and honey [20,21] samples.

For determination of the trace elements in the different biological samples, often it is necessary to use preconcentration methods for increasing the sensitivity and usability of the analytical methods. This may be achieved by different preconcentration methods, including dry ashing (DA) of the biological materials [22]. The gain in DA method is mainly dependent on the mass reduction factor and the composition of the residual ash, e.g. the enhancement factor for the pine needles was 7 and the limit of detection (LOD) for PIXE analysis was below 0.2 μg/g for Zn, Cu, Rb, and Sr. When ashing biological materials with low ash contents such as wood of pine or spruce (0.3% of dry weight) and honey (0.1% of wet weight), the gain was far greater and the LOD was 0.030 μg/g for wood and below 0.010 μg/g for honey.

The analyte losses observed by subjecting the samples for the DA procedure are still problematic. However, the concentrations of some elements determined by a DA method were lower than those observed by some other methods, including the digestion in PTFE bomb and microwave oven [12]. Yun et al. [23] developed the DA procedure for ICP-MS analysis of lichen samples using oven drying at low temperature of 90 °C for 20 h; then, drying at 110 °C for an additional 20 h to reduce the moisture. The results revealed to errors of 10% for concentrations greater than 10 μg/g and errors less than 20% for concentrations near 1 μg/g.

The aim of this study was to (i) improve the procedure of DA method for XRF determinations of different elements, namely K, Ca, Ti, Cr, Mn, Fe, Ni, Cu, Zn, Rb, Sr, and Pb in bee honey samples; (ii) characterize the DA method for XRF analysis with respect to the detection limits, precision, and accuracy; and (iii) apply the DA method for XRF analysis of some Syrian honey samples.

2. Materials and methods

2.1. Sampling

Six bee honey samples (500 g each) were collected in the period spring–summer (2007). Two multiflora honey samples were collected

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from beehives, which were located in Dobaya and Swedah locations in the southern parts of Syria (pollution free places); while, one *Quercus* spp. (Oak) and three *Citrrus* spp. (Citrus) honey samples were collected from beehives, which were located in Jableh and Tartous regions in the western part of the country. All necessary precautions have been taken regarding sampling and storage of the honey materials. The honey samples were taken directly from the honeycombs and stored in the clean glass jars without contacting a metallic machine known as a honey extractor. Table 1 shows the sample code, the botanical and the geographical origins of the bee honey samples.

2.2. Apparatus

The XRF measurements were performed using an energy-dispersive X-ray fluorescence instrument, which was equipped with 2 kW Mo tube and a Si(Li) semiconductor detector (PGT Co.) with an energy resolution of 140 eV at 5.9 keV. The operating conditions were differed, depending on the mode of the X-ray excitation. However, these were: (7 mA and 17 kV) and (5 mA and 45 kV) by using Cu- and Mo-secondary targets, respectively. The live time was 1000 s for both of the X-ray excitation modes. A cylindrical collimation unit made from Teflon material was used to collimate the primary and the secondary X-ray beams. The unit was with dimensions of 2.3 cm, 2.3 cm, and 2.0 cm, corresponding to the distance between the tube's window and the secondary target, the secondary target and the sample, and the sample and the detector, respectively.

In order to compare the results, the bee honey samples were also analyzed by the atomic absorption spectrometer (AAS), Vario 6, Analytica Jena (Germany).

The moisture content was determined using Abbe refractometer (Vip-Carl Zeiss, Germany). In addition, the moisture in the bee honey samples was removed using the oven from Memmert GmbH + Co.KG (Germany). Electrical furnace from Nabertherm Com.(Germany) was used for ashing of the bee honey samples.

2.3. Reagents

A pure cellulose powder (AG) for analysis from Seelze (Hannover/Germany) was used as a binder for preparation of the XRF targets. The solutions of 14 N HNO_3 'Analar' (BDH) were used for the ash dissolutions. The stock standard solutions of K, Ca, Ti, Cr, Mn, Fe, Ni, Cu, Zn, Rb, and Sr with the concentrations of 1000 μg mL^{-1} each were used for the preparation of the multi-element reference targets for XRF calibration of Kα lines; while, the stock standard solutions of Au, Cd, La, Nd, Gd, and Pb with the concentrations of 1000 μg mL^{-1} each were used for the preparation of the multi-element reference targets for XRF calibration of Lα lines.

The solutions of $HClO_4$ 'Analar' and HCl 'Analar' from BDH were used for the bee honey analysis by AAS method.

All aqueous solutions and dilutions were prepared with ultrapure water, which was obtained from a water purification system (New Human power II, South Korea) with 18.3 MΩ cm specific resistivity.

Table 1
Sample code, botanical and geographical origin of the Syrian bee honey samples.

Sample code	Botanical origin	Geographical origin	
		Place	Area
IS2	Multiflora	Dobaya	S[a]
IS4	Multiflora	Sweida	S
IIIS1	*Citrrus* spp. (Citrus)	Jableh	W[b]
IIIS2	*Quercus* spp. (Oak)	Jableh	W
IIIS5	*Citrrus* spp. (Citrus)	Tartous	W
IIIS9	*Citrrus* spp. (Citrus)	Jableh	W

[a] and [b] are the southern and the western part of Syria.

2.4. Moisture content

The moisture content of the bee honey samples was determined by the refractive-index method. The refractometer sampler (compartment and window) was cleaned with acetone prior to each use and the measurements were performed at 22 °C.

2.5. Dry ashing

The procedure of DA method was experimented in a manner consisting of two steps. First, the bee honey samples with 10 g each were put in the crucibles with volumes of 50 mL and dried in an oven at 105 °C for 72 h, covered, cooled in the desiccators and weighed. The samples were re-dried for 1 h in the oven, cooled, and reweighed. The process was repeated at 1 h drying intervals until the differences in the variations in the released water were less than 0.1%. Second, the dried samples in the crucibles were subjected to ashing in an electrical furnace. The temperature was increased in three steps: 200 °C, 300 °C, and 550 °C; where the first and the second steps were lasted for 20 min each, while the third step was lasted for 16 h. The ashes were weighed and kept in the desiccators for further use.

2.6. Effect of binder

The effect of the binder made of the cellulose material on the precision of the XRF analysis was studied. For this purpose, a 1-mL volume of a multi-element standard solution, containing elements of K, Ca, Ti, Cr, Mn, Fe, Ni, Cu, Zn, Rb, and Sr with a concentration of 10 μg/mL each, was put in a spectrocup with a surface area of 4.91 cm^2. A 100 μL volume of a suspended cellulose solution (0.120 g/mL) was added to the multi-element standard solution, thoroughly mixed, and finally evaporated by IR lamp. The above mentioned procedure, except the addition of cellulose, was repeated to prepare a multi-element target without the binder.

2.7. Preparation of bee honey targets

In order to compare between the analytical results, three kinds of bee honey targets were prepared as follows:

(i) A mass of 10 g of the IIIS5 honey sample was dried at 105 °C for 72 h. Then, the sample was subjected to the previously mentioned DA procedure (Section 2.5). The obtained ash was dissolved in 1-mL volume of 6 N HNO_3. The ash solution in the crucible was removed to a small vial with a 5-mL volume. Then, a 100 μL volume of the suspended cellulose solution (0.120 g/mL) was added to the dissolved ash. The obtained mixture was thoroughly shaken by an electrical shaker (KS 125 basic, IKALABORTECHNIK Co., Japan) for 5 min, removed to a spectrocup with a surface area of 4.91 cm^2, and dried under IR lamp. Finally, the obtained target with mass of 0.0525 g was kept in a desiccator for further XRF analysis.

(ii) A mass of 10 g of the IIIS5 honey sample was put in a 100 mL volume glass beaker. The sample was dried at 105 °C for 72 h. A 25 mL volume of concentrated HNO_3 was added to the honey sample, then, the temperature of the mixture was adjusted at 130 °C using oil bath. After drying of the sample, the obtained residue (the mass of the residue was about 2.5 g; a black charcoal powder was left in the sample) was cooled, ground into a fine powder using an agate hand mortar (Retsch, Germany) and sieved with a mesh width of 75 μm. A portion with a mass of 1.0 g of the obtained residue was put in a spectrocup with a surface area of 4.91 cm^2; the sample was gently pressed with a glass rod. Finally, the obtained target was kept in a desiccator for further XRF analysis.