THE ANALYSIS OF PLANT-BASED RAW MATERIALS OF UNKNOWN ORIGIN

MAREK WASEK[1,2]*, PIOTR WROCZYŃSKI[1], SYLWIA SOŁOBODOWSKA[1] and NATALIA LAL[1]

[1]Department of Drug Bioanalysis and Analysis, Medical University of Warsaw,
61 Żwirki i Wigury St., 02-091 Warszawa, Poland
[2]Laboratory of Individual and Environmental Doses, Central Laboratory for Radiological Protection,
7 Konwaliowa St., 03-194 Warszawa, Poland

Abstract: Chosen aspects of the safety of use of several herbs received from National Medicines Institute, which came from smuggling, have been examined. The analysis has been conducted in three different aspects:

(1) Possibilities of contamination of plant-based raw materials by metals of heavy elements (As, Cd, Cu, Cr, Pb).

(2) Conscious smuggling of intoxicating preparation or narcotics in plant-based raw materials.

(3) Radioactive contamination originating mostly from ^{137}Cs isotope.

To solve the problem, analytical methods of GFAAS and ICP-MS, X-ray diffraction and high-distributive spectrometry of γ-radiation have been applied. Determined concentration of arsenic in all analyzed samples and the concentration of lead in one sample exceeded allowable concentration recommended by WHO. In analyzed materials, no presence of narcotics or radioactive contamination of ^{137}Cs isotope has been detected.

Keywords: heavy elements, radioactive contamination, analysis of plant

All over the world an increasing trend towards the use of plant origin preparations can been seen recently. These preparations are taken to supplement the diet, improve health and fitness, prevent and cure diseases. Every year Europe, including Poland, is flooded with counterfeit drugs, among which plant origin preparations are found. Smuggling of substances of plant origin and herbal preparations across Polish borders is frequent (1).

Materials used in this study consisted of six samples of plant material of unknown origin, which were seized by the Customs Service of Warsaw airport "Okęcie".

It was expected that seized material could be dangerous because of the presence of the following contaminants: 1. heavy metals; 2. radioactive isotopes; 3. intoxicants and narcotics.

The source of threats mentioned in point 1 and 2 is inappropriate method of collecting plant material. There are several anthropogenic sources of environmental contamination with heavy metals. Various branches of industry, transport, public utilities, power industry, fertilizers and waste used for fertilization, waste dumps to name a few. From these sources heavy metals are dispersed into the environment and cause contamination of soil, water and air. Finally, they enter human and animal organisms, directly or indirectly, by plant consumption. Significant contamination of soil and plant with heavy metals occurs along communication routes. It is mainly caused by lead (which comes from exhaust gases), cadmium and chromium (from tyre abrasion). This source can be responsible for the contamination of herbs and herbal medicines with heavy metals (cadmium, lead, chromium, copper, mercury, arsenic and other) (2, 3).

Another problem is the possibility of plant material contamination with radioactive isotopes, which occurs when plant material is collected from radioactively polluted sites. The main radioactive contaminant is isotope ^{137}Cs. Because of very long half-life (around 30 years) this isotope can be deposited in soil and plants for a very long time. Trial nuclear explosions in the atmosphere caused wild spread of fission and activation products into the environment. Since 1960s, intensive measurements of fallouts have been conducted and it was estimated that around $9.6 \cdot 10^{17}$ Bq of ^{137}Cs was intro-

* Corresponding author: e-mail: marek.wasek@wum.edu.p

duced into the atmosphere, 76% of which was deposited in Northern Hemisphere and 24% in the Southern Hemisphere. Second very important source of radioactive pollutants are nuclear reactor failures. Two, in which the greatest amounts of ^{137}Cs (radioactivity of more than $7 \cdot 10^{16}$ Bq) were emitted, are Windscale and Chernobyl disasters (4).

Completely different health threats arise from consumption of drugs or preparations of plant ori-gin, which can be mixed with intoxicating and nar-cotic substances, what is done intentionally with the aim of smuggling these psychoactive compounds.

Without sophisticated apparatus it is not possi-ble to state whether given material contains dis-cussed types of contaminants.

The aim of this work was to check some safe-ty aspects of six herbal preparation obtained from the National Medicines Institute , including:

1. Determination of the selected heavy metals con-tent (As, Cd, Cu, Cr, Pb) in powdered herbal samples.
2. Identification of crystalline substances and organic compounds with narcotic activity.
3. Radiometric assessment of analyzed materials.

EXPERIMENTAL

Chemical analysis

Two analytical techniques were used: Graphite Furnace Atomic Absorption Spectrometry (GFAAS) and Inductively Coupled Plasma Mass Spectrometry (ICP-MS). Analyzed material consist-ed of 6 herbal preparations of unknown origin (4 in the form of powder, 2 as solids). No distinct/obvious

Table 1. Analysis results of heavy metals in blank determined by GFAAS and ICP-MS methods.

Element	Concentration of elements in blank (µg/kg)	
	GFAAS[a]	ICP-MS[b]
As	–	20
Cd	< LD	< LD
Cr	< LD	50
Cu	30	50
Pb	< LD	40

[a] relative uncertainty – 35% (coverage factor k = 2); [b] relative uncertainty – 25% (coverage factor k = 2).
LD = limit of detection

Table 2. Analysis results of heavy metals in certified material Mixed Polish Herbs (INCT-MPH-2) determined by GFAAS and ICP-MS methods.

Element	Certified material (mg/kg)[a]	Measured value (mg/kg)[b] GFAAS	Measured value (mg/kg)[c] ICP-MS
Cd	0.199 ± 0.015	0.209 (10)	0.21
Cu	7.77 ± 0.53	7.31 (84)	6.75
Cr	1.69 ± 0.13	1.40(45)	1.6
Pb	2.16 ± 0.23	2.48 (35)	2.5
As	0.191 ± 0.023	...	0.17

[a] uncertainty of result given as expended uncertainty (coverage factor k = 2); [b] values of the combined standard uncertainty u_c given in brackets refer to the corresponding last digits of the quoted result; [c] the result is given as arithmetic mean of 4 measurements; the relative expanded uncertainty – 15% (coverage factor k = 2).

Table 3. Analysis results of heavy metals in certified material Oriental Tobacco Leaves (CTA-OTL-1) by GFAAS and ICP-MS methods.

Element	Certified material (mg/kg)[c]	Measured value (mg/kg)[a] GFAAS	Measured value (mg/kg)[b] ICP-MS
Cd	1.12 ± 0.12	0.98 (18)	1.11
Cu	14.1 ± 0.5	15.6 (9)	13.18
Cr	2.59 ± 0.32	2.92 (16)	2.95
Pb	4.91 ± 0.80	4.31 (71)	4.32
As	0.539 ± 0.060	...	0.427

[a, b, c] – please, refer to description of Table 2

Table 4. Results of analysis of heavy metals in samples by GFAAS and ICP-MS methods.

Element	Analytical method used	Sample number Measured value [mg/kg]					
		1	2	3	4	5	6
As	ICP-MS[a]	2.15	3.75	8.22	3.73	5.22	1.25
Cd	GFAAS[b]	0.292 (76)	0.257 (39)	0.134 (39)	0.383 (18)	0.078 (8)	0.188 (42)
	ICP-MS	0.205	0.201	0.175	0.38	0.09	0.16
Cr	GFAAS	1.14 (13)	4.28 (37)	10.1 (8)	6.21 (35)	1.38 (11)	1.65 (90)
	ICP-MS	1.21	4.5	8.88	6.39	1.09	1.2
Cu	GFAAS	9.59 (13)	10.81 (18)	7.87 (48)	15.52 (22)	7.82 (19)	7.22 (17)
	ICP-MS	10.41	13.91	7.68	15.97	7.18	8.52
Pb	GFAAS	2.52 (27)	2.85 (55)	30.8 (1.8)	3.58 (52)	2.56 (48)	2.43 (47)
	ICP-MS	2.1	2.9	27.98	3.84	2.78	2.13

[a] results given as arithmetic mean of 4 measurements; relative expanded uncertainty – 15% (coverage factor k = 2) 15%; [b] the combined standard uncertainty u_c given is parentheses refers to the corresponding last digits of the quoted result.

contamination of these preparations was observed. The only sensory observation was sharp odour of Asian spices.

In order to check the validity of obtained results two certified reference materials (CRM) were used: Oriental Tobacco Leaves (CTA-OTL-1) and Mixed Polish Herbs (INCT-MPH-2) (5, 6).

The portions of herbal preparations and certificated materials, around 0.5 g each, were carefully weighed into teflon crucibles and subjected to initial mineralization by adding 2.5 ml of 65% nitric acid to each sample. After 24 h, the samples were mineralized using microwaves in a closed system. The conditions were as follows: time of heating – 8 min, microwave power – 20%, 140 W, number of repetitions – 4. CEM MDS – 2000 microwave oven with quartz crucibles was used . After mineralization, the samples were quantitatively transferred into 100 mL volumetric flasks and filled with water to the mark. They were analyzed by GFAAS directly or after dilution. Atomic Absorption Spectrometer type AVANTA ULTRA Z with GBC PAL auto sampler was used. The content of the following metals was determined: Cd, Cr, Cu, Pb. Their amount, as well as that of As, was additionally analyzed by ICP-MS (PerkinElmer Elan DRC II inductively coupled mass spectrometer with laser ablation/sampling).

The conditions of ICP-MS analysis were: RF power: 100 W; plasma gas flow rate: 15.0 L/min; auxiliary gas flow rate: 1.2 L/min; nebulizer gas flow rate: 0.8 L/min; focus lens voltage/potential: 6.5 V; detector mode: dual Scott spray chamber; cone: NI.

The samples were collected from the precipitate and diluted 200 times with 0.7% HNO_3. The following ions were analyzed: ^{75}As, ^{111}Cd, ^{52}Cr, ^{63}Cu, ^{208}Pb. In case of As and Cr DCR mode was used with ammonia as reaction gas.

Identification of crystalline substances and organic compounds

For qualitative analysis performed in order to identify crystalline substances and organic compounds present in analyzed samples BRUKER D8 ADVANCE X-ray diffractometer was used. No special preparation of samples was required. Two solid herbal preparations were pulverized in agate mortar. Diffrac + software was used to identify the compounds.

γ-Ray spectrometry

For radiometric measurements of samples Canberra γ-ray spectrometer with HPGe detector and analogue-to-digital converter line was used. Spectra were recorded by Tukan 8k Multichannel Pulse Amplitude Analyzer (produced by Institute of Nuclear Problems in Świerk, Poland). As measurements were only qualitative, no initial preparation of samples was required.

The conditions of γ-ray spectrometry analysis were: the energy range: 40–2000 keV; resolution (FWHM): 1.5 keV; measuring time: 10 h.

RESULTS AND DISCUSSION

Chemical analysis

Solution of reagents needed for mineralization procedure was used as a blank. The results of blank analysis by GFAAS and ICP-MS are presented in Table 1. It can be seen that the content of analyzed
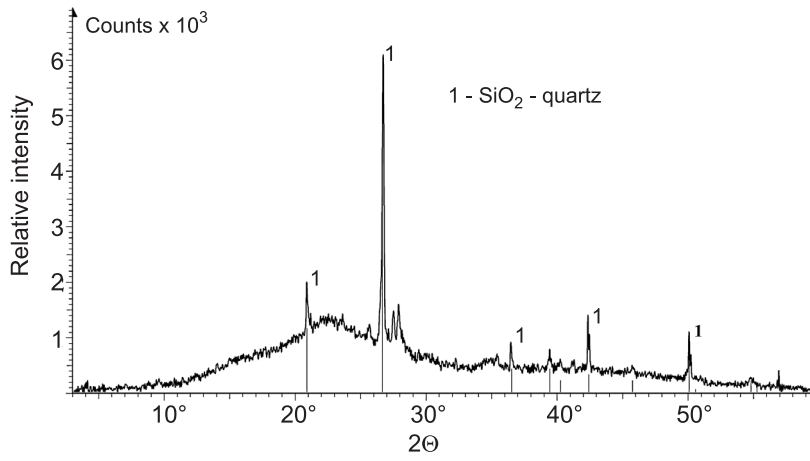
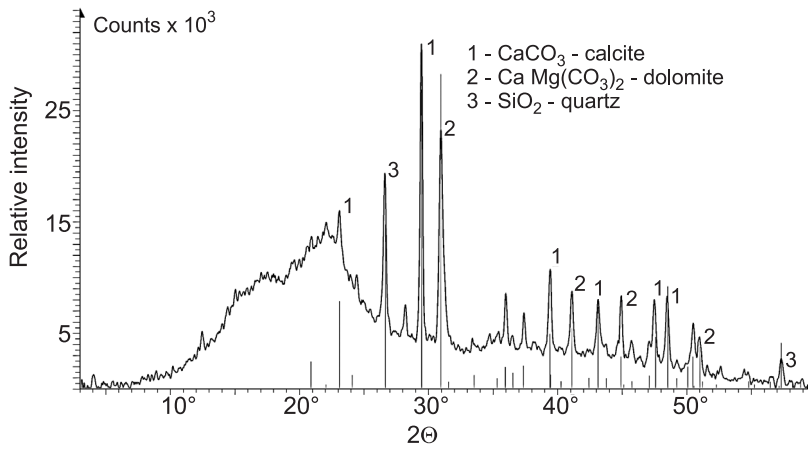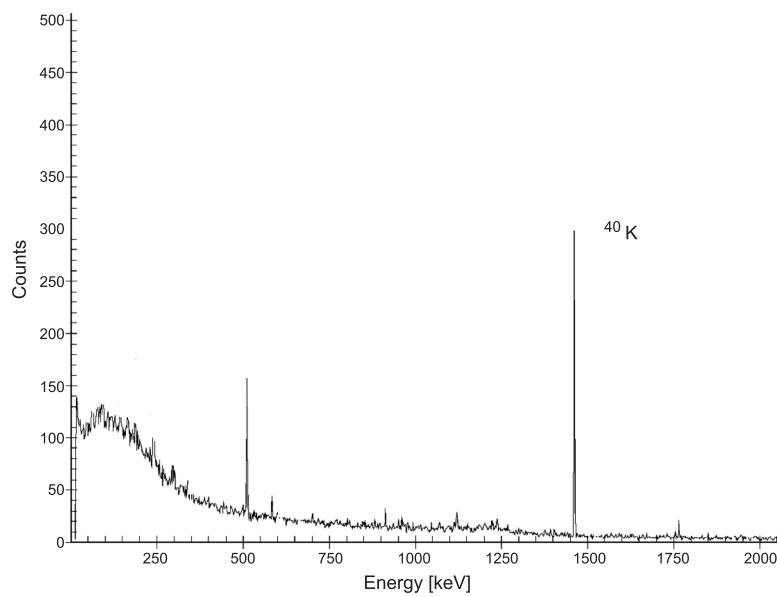Figure 1. X-ray diffractogram of sample number 4

Figure 2. X-ray diffractogram of sample number 3

Figure 3. γ-Ray spectrum of sample number 3

metals in blank is several times smaller than that expected in analyzed samples. All final results were corrected for the blank values.

The analysis of heavy metals in plant material is particularly difficult because analyzed elements are present only in trace amounts. In order to ensure the required quality of results two certified reference materials CRM: CTA-OTL-1 and INCT-MPH-2 were also analyzed. The results of their analyses by GFAAS and ICP-MS are presented in Tables 2 and 3, respectively. Determined amounts of elements in certified materials are in accordance with those declared, what indicates that the method used was suitable and valid.

In order to compare the results obtained from analyses of 6 samples of unknown origin using two analytical methods, Student's t-test was used. The null hypothesis tested was that the means of results obtained by GFAAS and by ICP-MS are equal. For all analyzed samples the null hypothesis was not rejected. That is why any discrepancies between results obtained by these two methods are purely random. This emphasizes that in the two analytical methods used in this work there are no systematic errors.

Identification of crystalline and organic substances

Figures 1 and 2 present two typical diffractograms obtained for two samples (number 4 and number 3, respectively.). The dependence of intensity of diffraction lines on the angular position of the line is shown. On both diffractograms two distinct peaks for quartz (sample 4) and additionally for dolomite and calcite 9 (sample 3) were identified. Diffraction lines at low scattering angles were absent, which means that the samples are free of narcotic organic compounds. This result refers to all analyzed materials.

Radiometric analysis

Figure 3 shows typical γ-ray spectrum of one analyzed sample (sample number 3). No line indicating the presence of ^{137}Cs was found. Environmental background radiation due to ^{40}K was observed at 1460 keV.

CONCLUSIONS

1. The arsenic concentration in all analyzed plant materials is greater than the WHO norm (7). Frequent use of such preparations can increase the risk of arsenic accumulation and have negative consequences on health.

2. The lead content in sample 3 greatly exceeds the levels permitted by WHO (7) and new monograph of European Pharmacopoeia (8). Safety of such materials is questionable.

3. The comparison of CFAAS and ICP-MS analyses of CRM materials and the results of Student's t-test show great reliability of the results obtained by two analytical methods used in experiments. That is why cheaper GFAAS method can be used in future analyses.

4. The analysis of diffractograms showed that no crystalline organic compounds or pharmacologically active additives are present in the analyzed materials. Relatively high silicon content could indicate the contamination of the samples with silica.

5. The analyzed materials do not show any, not even trace contamination with radioactive elements.

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