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Abstract
In the context of monitoring environmental factors, metals are one of the major analytical components. Applying appropriate determination methods and obtaining accurate results is a requirement imposed on environmental laboratories that perform quality control of water, soil, waste or vegetation. This study presents some examples of certified reference materials for quality control of the results of toxic metal determination from solid environmental and vegetation samples. The analyzed and verified metals were As, Cd, Cr, Cu, Ni, Pb and Zn. The pre-treatment of the samples, the determination methods of metals and the obtained results are also presented. Inductively Coupled Plasma Optical Emission Spectrometry (ICP-EOS) and Inductively Coupled Plasma Mass Spectrometry (ICP-MS) techniques are suitable for low metal concentrations, while ICP-EOS and Flame Atomic Absorption Spectrometry (FAAS) methods can be used at high concentrations.

Keywords: toxic metals, ICP-EOS, Flame AAS, CRMs, environmental matrices

INTRODUCTION
Due to the advanced industrialization during the last decades of the past century and the first of this century, many areas became contaminated with toxic metals, considerable quantities being found both in soil, sediments, groundwater and surface water. The monitoring and rehabilitation studies have as a starting point the investigation of the quality of environmental factors affected by anthropogenic activities, such as: mining, metallurgical industry, machine building industry, intensive agriculture, etc. Even if some mines have been closed or abandoned, the surrounding areas remain a potential danger to fauna, flora, the aquatic environment and human health [1-4].

Different types of waste with varied metal content are stored either in specially designed landfills, which over time can produce discharges of toxic substances into the environment, or directly on the ground as long as the content of toxic substances is below certain limits imposed by laws [5, 6]. The high metal content in contaminated soils, sediments, waste, biological sludge must be accurately determined using appropriate analytical techniques, so the composition of the complex matrices must not interfere with the determination [7].

In this context, in several international projects, matrix-type Certified Reference Materials (CRMs) have been developed, containing certified values of toxic metal content and the associated uncertainty. Several institutions provide such complex matrices for soils (organic rich soil, sandy soil, road dust, industrial soil), sediments (estuarine, coastal, lake or river sediment), waste (fly ash, sewage sludge amended soil), plant tissue (hay powder, clover, lichen, tomato leaves, apple leaves, plankton, rice, etc.) or animal organs (tuna muscle, cod liver, etc.). Among these, we can mention the best known, namely: European Commission Joint Research Center - JRC (800 CRMs of the BCR- and IRMM-brands as well as the ERM-branded materials that were produced by the JRC) or US Department of Commerce -
National Institute of Standards and Technology NIST. Obtaining such materials implies the application of well-established rules, which ensure the homogeneity and stability of the samples, as well as the assigned value and the uncertainty for each element or compound [8]. The international requirements imposed by the EN ISO 17025/2018 standard enforce accredited laboratories around the world to verify their analytical results for the determined pollutants [9]. Thus, the use of matrix-type certified reference materials has become a common practice in such laboratories (Table 1).

Table 1. Examples of CRMs in environmental analysis

| CRMs            | Matrix type               | Aqua regia extractable content                  |
|-----------------|---------------------------|-------------------------------------------------|
| ERM-CC 141      | Loam Soil                 | As, Cd, Co, Cr, Cu, Hg, Mn, Ni, Pb, Zn          |
| ERM-CC018       | Sandy Soil                | As, Cd, Cr, Co, Cu, Pb, Hg, Ni, V, Zn           |
| RTC SPE-001     | Soil                      | Al, As, Ba, B, Cd, Ca, Cr, Co, Cu, Fe, Li, Pb, Mg, Mn, Hg, Mo, Ni, K, Se, Ag, Sb, Na, Sr, Sn, Ti, V, Zn |
| WQB-1           | Sediment                  | Al, As, Ba, B, Cd, Ca, Cr, Co, Cu, Fe, Li, Pb, Mg, Mn, Hg, Mo, Ni, K, Se, Ag, Sb, Na, Sr, Sn, Ti, V, Zn |
| RTC CRM016      | Sediment                  | Al, As, Ba, B, Cd, Ca, Cr, Co, Cu, Fe, Li, Pb, Mg, Mn, Hg, Mo, Ni, K, Se, Ag, Sb, Na, Sr, Sn, Ti, V, Zn |
| BCR 146R        | Sewage sludge from industrial origin | Cd, Co, Cr, Cu, Hg, Mn, Ni, Pb, Zn                        |
| NIST 2782       | Industrial sludge         | As, Cd, Cr, Cu, Pb, Hg, Mo, Ni, Se, Zn          |
| CRM 029         | Sewage sludge             | Al, As, Ba, B, Cd, Ca, Cr, Co, Cu, Fe, Pb, Mg, Mn, Hg, Mo, Ni, K, Se, Ag, Sb, Na, Sr, Sn, Ti, V, Zn |
| BCR 483         | Sewage sludge amended soil | Cd, Cr, Cu, Ni, Pb, Zn                          |
| BCR 176R        | Fly ash                   | As, Cd, Co, Cr, Cu, Fe, Ni, Pb, Sb, Se, Tl, Zn  |
| ERM-CJ464       | Tuna fish                 | Hg total, CH\textsubscript{3}Hg\textsuperscript{+} |
| BCR 482         | Lichen                    | Al, As, Cd, Cr, Cu, Hg, Ni, Pb, Zn              |
| NIST 1515       | Apple leaves              | Al, B, Ba, Cd, Ca, Cu, Fe, Pb, Mg, Mn, Hg, Mo, Ni, K, Na, V, Zn |
| NIST 1573a      | Tomato leaves             | Al, Sb, As, Ba, B, Cd, Ca, Cr, Cu, Co, Fe, Mn, Hg, Mo, Ni, K, Se, Ag, Na, Sr, V, Zn |

Another way of testing the obtained results is by participating in proficiency testing (PT) schemes, where the entire applied procedure is tested (pretreatment, determination, reporting results). There are specialized international organizations that organize annually such schemes, these being diversified according to the pollutants and the matrix type (water, soil, waste, sediment, plant tissue). A database for such PTs in which the topics, the type of matrix and the accreditation of the organization that organizes the scheme are presented is EPTIS database [10].

The aim of this study is to present the results obtained by using various CRMs to check the metal content (As, Cd, Cu, Cr, Ni, Pb, Zn) from different environmental matrices (soil, sediment, fly ash, sewage sludge and vegetable tissues) as well as the applied analytical techniques for metal determination.

EXPERIMENTAL PART

Methods

In order to determine the metal content in solid environmental samples (soil, sediment, fly ash, sewage sludge), FAAS and ICP-EOS techniques were applied.

Table 2 presents the operational parameters for the method that uses ICP-EOS technique [11], while the performance parameters (quantification limit, accuracy and measurement uncertainty) are listed in table 3. The working conditions [12] and performance parameters for FAAS method are presented in table 4.

In the case of vegetation samples, due to their low metal contents, ICP-MS and ICP-EOS methods were applied. The operational parameters, respectively the performance
parameters of the applied ICP-MS method are presented in tables 5 and 6.

**Table 2. Operating parameters for ICP-EOS AVIO 500 equipment**

| Spectrometer parameters | Plasma parameters |
|-------------------------|-------------------|
| Purge gas flow rate: normal | Argon flow rate 15 L/min |
| Delay time: 40s | RF power 1400 W |
| Replicates: 3 times | Auxiliary agent flow rate: 0.2 L/min |
|                        | Plasma view: Axial |
|                        | Nebulizer flow rate: 0.7 L/min |
|                        | View distance: 15.0 mm |

**Processing spectral peaks**

- Peak Algorithm: peak area
- Points per peak: 10 points
- Spectral corrections: background correction

**Table 3. Performance parameters of the methods applied with the ICP-EOS AVIO 500 equipment**

| Element | Wavelength, nm / matrix | LOQ, mg/kg | Precision, % | Uncertainty, % |
|---------|-------------------------|------------|--------------|----------------|
| As      | 188.979 (vegetation)    | 0.75       | 4.7          | 14.0           |
|         | 197.197 (solid samples) |            |              |                |
| Cd      | 228.802 (vegetation)    | 0.08       | 3.8          | 0.9            |
|         | 214.440 (solid sample)  |            |              |                |
| Cr      | 267.716                 | 0.04       | 2.4          | 13.3           |
| Cu      | 324.754 (vegetation)    | 0.05       | 5.3          | 11.7           |
|         | 327.393 (solid samples) |            |              |                |
| Ni      | 231.604 (vegetation)    | 0.11       | 4.8          | 11.7           |
|         | 341.476 (solid sample)  |            |              |                |
| Pb      | 220.353 (vegetation)    | 0.33       | 2.7          | 9.4            |
|         | 217.000 (solid sample)  |            |              |                |
| Zn      | 206.200                 | 0.11       | 3.2          | 10.0           |

**Table 4. FAAS - Performance parameters and working conditions**

| Element | Wavelength, nm | Flame type | Background correction | LOQ, mg/kg | Precision, % | Uncertainty, % |
|---------|----------------|------------|-----------------------|------------|--------------|----------------|
| Cd      | 228.8          | air / acetilene | deuterium            | 2.1        | 5.56         | 9.8            |
| Cu      | 324.8          | air / acetilene | deuterium            | 6.0        | 3.82         | 15.0           |
| Ni      | 232.0          | air / acetilene | deuterium            | 14.0       | 4.61         | 9.4            |
| Pb      | 217.0          | air / acetilene | deuterium            | 16.0       | 5.21         | 9.7            |
| Zn      | 213.9          | air / acetilene | deuterium            | 3.0        | 2.83         | 12.8           |

**Table 5. Operating parameters for ICP-MS spectrometer**

| Parameter                               | Value or Condition |
|-----------------------------------------|--------------------|
| Delay time: 60s                         | Purge gas flow: normal |
| Replicates: 3 times                     | Peristaltic pump: 1.5mL/min |
| **Tune parameters**                     |                    |
| Plasma flow rate: 15L/min               |                   |
| Auxiliary flow rate: 0.90 L/min         |                   |
| Nebulizer Pump: 0.10rps                 |                   |
| **Plasma parameters**                   |                   |
| RF Power: 1550W                         |                   |
| Plasma view: axial                      |                   |
| RF matching: 1.30V                      |                   |
| **Plasma mode**                         |                   |
| Plasma Mode: General Purpose            | Sample Depth: 10 mm|
| **Cell parameters**                     |                   |
| He Flow: 4.3mL/min                      | Octp Bias: -8.0 V  |
| **Spectral peak processing**            |                   |
| Peak algorithm: Peak area               | Peak pattern: 3 points |
| Replicates: 3 times                     | Integration time: 0.2001 sec. |
All experimental tests were performed in an accredited laboratory according to EN ISO 17025/2018 [9], respecting the requirements for traceability and quality assurance of the analytical determinations results.

**Materials and equipment**

Multielements Certified Reference Material ME 21, 100 mg/L (Sigma-Aldrich)
Reference Material (RM) Quality Control Standard 21, 100 mg/L (LGC)
Hydrochloric acid ≥ 30% TraceSelect for trace analysis (Fluka)
Nitric acid ultra trace grade 69% (Scharlau)
Hydrogen peroxide solution, trace select ultra, ≥ 30% for trace analysis (Sigma Aldrich)
Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES) Perkin Elmer AVIO 500
Flame Atomic Absorption Spectrometer (FAAS)
Thermo Scientific M6 Dual
Inductively Coupled Plasma Mass Spectrometer (ICP-MS) 7900 Agilent Technologies
Microwave Oven Ethos Up Milestone

**Samples preparation**

The content of metals extracted in aqua regia solution from the soil, sediment and sewage sludge samples, was determined. To 1 g of solid sample, 9 mL HCl and 3 mL HNO₃ were added. For the pretreatment of these solid samples (soil, sediment, and sewage sludge), a digestion program was applied (table 7). After cooling, the solutions were filtered and brought quantitatively to a 50 mL volumetric flask with ultrapure water.

**Table 7. Microwave program for digestion of soil, sediment and sewage sludge samples**

| Stage | Power  | Time    | Temperature T1 (inside vessels) | Temperature T2 (outside vessels) |
|-------|--------|---------|---------------------------------|----------------------------------|
| 1     | 1800 W | 15 minutes | up to 180°C                      | 120°C                            |
| 2     | 1800 W | 20 minutes | 180°C                           | 120°C                            |
| 3     |        | 30 minutes | cooling                         | cooling                          |

Vegetation samples (0.5 g) were pretreated with a mixture of 8 mL HNO₃ and 2 mL H₂O₂ and then subjected to the digestion program presented in table 8. The resulting solutions were filtered on low porosity filter paper and brought to the mark with ultrapure water in 25 mL volumetric flask.

**Table 8. Microwave program for digestion of apple leave and lichen samples**

| Stage | Power  | Time    | Temperature |
|-------|--------|---------|-------------|
| 1     | 1800 W | 15 minutes | up to 200°C |
| 2     | 1800 W | 15 minutes | 200°C       |
| 3     |        | 30 minutes | cooling     |

For each matrix type and each applied digestion program, blank samples were used in order to verify the interference of reagents used during sample preparation.

The metals from the following CRMs: loam soil (ERM-CC 141), sandy soil (ERM-CC 018), sediment (WQB-1), fly ash (176 R), industrial
sludge (NIST 2782) were extracted in aqua regia mixture. The analyzed vegetation CRMs were: apple leaves (NIST 1515), lichen (BCR 482).

RESULTS AND DISCUSSION

The results obtained for the analyzed metals (As, Cd, Cr, Cu, Ni, Pb, Zn) in the CRMs are presented in this section. Thus, the obtained data for solid environmental samples (ICP-EOS, respectively FAAS results) are compared to the certified values from the quality certificates of the used CRMs (tables 9 ÷ 13).

| Table 9. Loam soil ERM-CC 141(JRC, IRMM brand), mg/kg dry matter |
|-----------------|-----------------|-----------------|
| Element         | Certified value | Determined values |
|                 |                 | ICP-EOS | FAAS |
| As              | 7.5 ± 1.4       | 8.8 ± 1.3 | -     |
| Cr              | 31 ± 4          | 30.4 ± 4.0 | -     |
| Cu              | 12.4 ± 0.9      | 11.6 ± 1.4 | 11.5 ± 1.7 |
| Ni              | 21.9 ± 1.6      | 20.3 ± 2.4 | 22.1 ± 2.1 |
| Pb              | 32.2 ± 1.4      | 30.9 ± 2.9 | -     |

| Table 10. Sandy soil ERM-CC 018, mg/kg dry matter |
|-----------------|-----------------|-----------------|
| Element         | Certified value | Determined values |
|                 |                 | ICP-EOS | FAAS |
| As              | 22.9 ± 1.3      | 21.7 ± 3.1 | -      |
| Cd              | 5.4 ± 0.5       | 5.9 ± 0.5 | -      |
| Ni              | 25.8 ± 1.8      | 27.3 ± 3.2 | 24.4 ± 2.3 |
| Pb              | 289 ± 10        | 298 ± 28 | 281 ± 27 |

| Table 11. Sediment WQB-1, mg/kg dry matter |
|-----------------|-----------------|-----------------|
| Element         | Certified value | Determined values |
|                 |                 | ICP-EOS | FAAS |
| As              | 23 ± 1.8        | 24.7 ± 3.5 | -      |
| Cu              | 79.6 ± 16.1     | 66.4 ± 7.8 | 63.5 ± 9.5 |
| Ni              | 61.5 ± 17.6     | 57.1 ± 6.7 | 59.7 ± 5.6 |

| Table 12. Fly Ash 176 R, mg/kg dry matter |
|-----------------|-----------------|-----------------|
| Element         | Certified value | Determined values |
|                 |                 | ICP-EOS | FAAS |
| Cu              | 10000 ± 70      | 1013 ± 119 | 989 ± 148 |
| Pb              | 50000 ± 500     | 4572 ± 430 | 4563 ± 443 |
| Zn              | 16800 ± 400     | 17039 ± 1700 | 16670 ± 2130 |

| Table 13. Industrial sludge NIST 2782, mg/kg dry matter |
|-----------------|-----------------|-----------------|
| Element         | Certified value | Determined values |
|                 |                 | ICP-EOS | FAAS |
| Cu              | 2594 ± 52       | 2626 ± 307 | 2555 ± 383 |
| Pb              | 574 ± 11        | 569 ± 53 | 573 ± 56 |
| Zn              | 1254 ± 196      | 1062 ± 106 | 1170 ± 150 |

The reported values for all metals in all solid matrices that were analysed are within the confidence intervals according to the quality certificates data.
Tables 14 and 15 show the results obtained for vegetation samples after metal determination by ICP-EOS, respectively ICP-MS techniques.

| Table 14. Apple Leaves NIST 1515, mg/kg dry matter |
|-----------------|-----------------|-----------------|-----------------|
| Element        | Certified value | Determined values |              |
|                |                 | ICP-EOS          | ICP-MS         |
| Cd             | 0.0132 ± 0.0015 | < 0.08           | 0.013 ± 0.0013 |
| Cu             | 5.69 ± 0.13     | 5.58 ± 0.88      | 5.57 ± 0.56    |
| Ni             | 0.936 ± 0.094   | 1.025 ± 0.132    | 1.02 ± 0.10    |
| Pb             | 0.470 ± 0.024   | < 1.5            | 0.49 ± 0.05    |
| Zn             | 12.45 ± 0.43    | 12.75 ± 1.67     | 12.18 ± 1.22   |

| Table 15. Lichen BCR 482, mg/kg dry matter |
|-----------------|-----------------|-----------------|-----------------|
| Element        | Certified value | Determined values |              |
|                |                 | ICP-EOS          | ICP-MS         |
| As             | 0.85 ± 0.07     | -                | 0.91 ± 0.09    |
| Cd             | 0.56 ± 0.02     | -                | 0.57 ± 0.06    |
| Cr             | 4.12 ± 0.15     | 5.58 ± 0.88      | 5.57 ± 0.56    |
| Cu             | 7.03 ± 0.19     | 6.87 ± 0.80      | 7.17 ± 0.72    |
| Ni             | 2.47 ± 0.07     | 2.53 ± 0.30      | 2.44 ± 0.24    |
| Pb             | 40.9 ± 1.4      | 41.3 ± 3.9       | 40.5 ± 4.1     |
| Zn             | 100.6 ± 2.2     | 99.6 ± 10.0      | 98.7 ± 9.9     |

Table 16 contains the results obtained after participating in an inter-laboratory comparison scheme for a soil sample analysis, the obtained values being compared to the assigned values. The table also presents the Z scores obtained for the reported data, all values falling within the \(-2 \div 2\) accepted range of standard deviation.

| Table 16. LGC sample 14/2019, results of PTS, mg/kg dry matter |
|-----------------|-----------------|-----------------|-----------------|
| Element        | Assigned value  | ICP-EOS          | FAAS            |
|                |                 | Value            | Z score | Value | Z score |
| As             | 32.9 ± 4.2      | 27.9 ± 4.2       | -1.44    | -     | -       |
| Cd             | 6.28 ± 0.63     | 5.14 ± 0.51      | -1.72    | -     | -       |
| Cr             | 153 ± 15.3      | 145.4 ± 17.4     | -0.47    | 145.0 ± 18.7 | -0.49 |
| Cu             | 112.8 ± 16.8    | 106.3 ± 16.8     | -0.57    | 118.3 ± 13.6 | -0.47 |
| Ni             | 43.9 ± 4.4      | 37.8 ± 5.3       | -1.39    | 45.7 ± 4.4  | 0.44    |
| Pb             | 226.7 ± 22.7    | 228.3 ± 31.5     | 0.07     | 226.7 ± 22.6 | 0.00    |
| Zn             | 815 ± 81.5      | 898.2 ± 90.2     | 1.02     | 935 ± 87    | 1.47    |

CONCLUSIONS
The results obtained in the verification of some methods applied for the determination of toxic metals both from solid environmental (soil, sediment, ash waste, respectively sewage sludge) and vegetation samples indicated that the applied ICP-EOS, FAAS, respectively ICP-MS techniques led to good results both on CRM matrices and in the case of participation in a proficiency test scheme.

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