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PHD THESIS SUMMARY

Identificarea unor caracteristici ale solurilor, vegetației și apelor prin metode fizico-chimice

Identification of soil, vegetation and water characteristics by physical – chemical methods

Doctorand:

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INTRODUCTION

Heavy metals in water, soil, sediment and vegetation have become a global problem as a result of the increase of industrial activity in the last decades. Due to their toxicity, cumulative and non-biodegradable characteristics, heavy metals are potentially hazardous to terrestrial and aquatic ecosystems and therefore to the lives of humans and animals. Soil degradation as a result of mining for coal extraction, precious metals and due to ballast-type exploitation is the worst form of pollution because soil destruction occurs on large areas [1,2].

Mining operations are areas of potential risk of pollution with toxic metals on the environment, both by accidental discharge of mine water, acidic water with a high content of metals, and through the deposits of mining waste, where there may be seepage into the underground and / or canvas. surface water. Even if the mine has been closed for a long time, metal pollution may continue. Thus, in the case of a closed mining operation, the rainwater or snow continued to wash the abandoned mine galleries and the dumps for mining waste, later pouring into the nearest river. Due to the acidity of these waters and the high content of toxic metals (As, Cd, Cr, Cu, Mn), both soil, surface water and sediments are highly contaminated and devoid of aquatic organisms [3].

The in-depth study of environmental factors in the areas bordering some mining operations may indicate the presence of toxic impurifiers above the maximum limits allowed by the legislation in force, it may alert the population in the area of the danger of use, for example of contaminated groundwater, it may support a starting point for further decontamination of the area. Such a study can also estimate the extent of the polluted area, as well as the affected environmental compartments, surface water, groundwater, soil, sediment, vegetation.

Heavy metals fall into two basic categories: essential and non-essential. Essential metals or micronutrients, such as chromium (Cr), cobalt (Co), copper (Cu), manganese (Mn), molybdenum (Mo), iron (Fe), selenium (Se) and zinc (Zn), are required for optimal functioning of biological and biochemical processes in organisms (including humans) including redox reactions and pigment and enzyme formation. Non-essential metals such as arsenic (As), cadmium (Cd), mercury (Hg) and lead (Pb) have no known biological function. Essential metals, at high concentrations, can have toxic effects on species and ecosystems. Like most organic pollutants, not even heavy metals are usually removed by natural processes [4].

The metals Palladium (Pd), Platinum (Pt), Rhodium (Rh), Ruthenium (Ru), Iridium (Ir) and Osmiu (Os) are known as the platinum element group (PGEs) or the platinum metal group (PGMs). These elements are classified, together with gold and silver, as noble or precious metals, which highlights the high value of these elements.

Platinum metals are found in nature in small quantities, the most abundant is platinum and the least abundant is ruthenium.

Platinum metals, due to their properties (corrosion resistant, high melting point, mechanical strength, ductility) are used mostly as catalytic converters but also in the glass, oil, electricity and electronics industries and in the manufacture of jewelry. Platinum metals are also used in medicine for the treatment of cancer (cisplatin complex PtCl2 (NH3) 2 is a well-known anticancer drug), as well as for the preparation of dental vials.

In the last decade, the concentration of platinum metals from environmental samples such as soil, surface water, sediments and vegetation has increased significantly, the main source of contamination with these platinum metals being the catalytic converters (components of cars fitted to the gas exhaust system). and which have a catalyst whose role is to transform the polluting gases the car produces: it converts carbon monoxide, nitrogen oxides and

hydrocarbons into less dangerous compounds such as water, carbon dioxide and nitrogen). Catalysts containing platinum metals are used mainly in motor vehicles but can also be used in industry as stationary catalysts used for ammonia oxidation. The main sources of platinum metal emission in Europe: 50.4% come from catalysts, 24.7% from jewelry, 6% from the electrical industry, 4.8% from the chemical industry, 4.6% from the glass industry, 2.6% from the oil industry and 6.8% from other sources [5].

Human activity resulted in an increase in the concentration of PGE in the soil, especially in areas with high traffic, as evidenced by a number of international studies [6]. The results of the determination of PGE in the samples taken from areas near the roads / highways indicate a decrease in the concentration of platinum metals as the distance from the highways / highways increases.

Increased concentrations of platinum metals were found in different water samples (rainwater, groundwater, surface water, marine sediments), these concentrations being caused by the emission of exhaust gases. Large concentrations of platinum have been found near the mines, especially those extracting nickel ores.

Due to the fact that PGEs are mainly emitted in metal form, they exhibit low toxicity. Some of them are transformed into a soluble form, becoming bioavailable and dangerous for both flora and fauna.

The doctoral thesis brings original contributions by evaluating polluted areas following the activities of the mining and transport industries and the potential of transfer of metallic pollutants into ecosystems through an in-depth study of the environmental compartments (surface water, sediment, soil and vegetation) from a river sector and its tributaries, in the vicinity of a mining area as well as through original research on the content of platinum metals from soil and vegetation samples taken from the vicinity of high-traffic roads. Also, a study was carried out to determine the metal content of medicinal plants (*Salvia Officinalis and Ocimum Basilicum*) grown on artificially polluted soils to observe the accumulation of metals in different parts of plants over time. In this doctoral thesis, 5 original methods for determining platinum metals and metals that can generate hydrides from environmental samples based on ICP-OES and HG-ICP-OES technique have been elaborated, and these methods have been optimized.

The thesis contains 9 chapters and is structured in two parts: **BIBLIOGRAPHICAL RESEARCH** and **ORIGINAL CONTRIBUTIONS.**

BIBLIOGRAPHICAL RESEARCH comprises 3 chapters:

Chapter 1. Risk elements due to the presence of metals in the environment (water, soil, sediment and vegetation)

Chapter 2. Main characteristics of optical emission spectrometry with inductively coupled plasma (ICP-OES) and the generation of hydride

Chapter 3. Performance parameters used in the validation of analytical methods **ORIGINAL CONTRIBUTIONS** include studies related to the characterization of the mining area, the development of new methods for determining platinum metals and the determination of these metals from environmental samples (soil and vegetation), the elaboration and optimization of the methods for determining As, Se, Sb and Sn by technique of inductive hydride generation coupled plasma optical emission spectrometry (HG-ICP-OES)

Chapter 4. Experimental details regarding the studies carried out

Chapter 5. Development of new methods for simultaneous determination of platinum metals by ICP-OES technique with applications in the evaluation of soil and vegetation samples in the proximity of national roads**.**

Chapter 6. Determination of the variation in time of the metal content by the ICP-OES technique from medicinal plants grown on soils enriched with metals

Chapter 7. Research on the assessment of the concentration of metal pollutants in an area affected by mining activities using the ICP-OES method

Chapter 8. Original experiments for mercury determination by HG-ICP-OES and cold vapor atomic absorption spectrometry (AAS-CV)

Chapter 9. Elaboration of methods for determining As, Se, Sb and Sn by the HG-ICP-OES method.

The doctoral thesis ends with the general conclusions, the original contributions and the prospects for further development.

The articles published during the doctoral thesis, as well as the scientific communications, bibliographic references and the works published during the doctoral period (in extenso) are attached.

ORIGINAL CONTRIBUTIONS

CHAPTER 4

EXPERIMENTAL DETAILS REGARDING THE STUDIES

4.1. EXPERIMENTAL DETAILS FOR SIMULTANEOUS DETERMINATIONS OF PLATINIC METALS FROM ENVIRONMENTAL PROBES BY THE M1 METHOD BASED ON ICP-OES

4.1.1. The equipment used to validate the M1 method based on ICP-OES, for determining platinum metals

The equipment used are:

• Inductive coupled plasma optical emission spectrometer, Optima 5300 DV, manufacturer Perkin Elmer, with simultaneous metal detection

• Ultrasonic nebulizer USN U600 AT +, manufacturer Cetac Tedelyne

• Millipore Sinplicity ultrapure water system, Merk

• Milestone Ethos Up type microwave oven with 15 Teflon dishes and control of both temperature and internal pressure

4.1.3. Experimental tests performed to validate the methods for determining platinum metals

The experimental tests applied in the validation of the determination methods Ir, Pd, Pt, Rh and Ru consisted in determining the limit of detection, limit of quantification, repeatability, intermediate precision, accuracy, of the tests of recovery and linearity, as well as estimation of the uncertainty of measurement.

4.2. EXPERIMENTAL DETAILS FOR DETERMINING THE CONTENT OF METALS FROM MEDICINAL PLANTS CULTIVATED ON SOILS enriched with METALS

4.2.1. Preparation of vegetation samples

The seeds of the two plants (*Salvia Officinalis* and *Ocimum Basilicum*) were grown in the laboratory, each in 2 different pots (one control sample and one sample contaminated with metals). Prior to planting, the concentration of metals in the seeds of the two species of medicinal plants was determined.

The soil on which the seeds were planted was obtained by mixing 75% garden soil with 25% substrate, soil that was analyzed before and after the homogenization with the substrate

took place. It was separated into two pots for each type of plant (sage, basil), constituting the control sample.

In two other pots the sage and basil seeds were cultivated on the same soil mixture, which was enriched with the following metals: cobalt, chromium, cadmium, copper, nickel, lead and zinc in concentrations of 5mg / kg Cd and Ni , 10 mg / kg Co and Cr, 15 mg / kg Cu, 100 mg / kg Zn and 200 mg / kg Pb, respectively. The addition concentrations were established so that the conditions of the polluted soil in the Certej area, contaminated soil of mining activity, were replicated in the laboratory.

All parts of the plants (root, leaves, stem and flowers) were allowed to dry at room temperature for 2-3 weeks, then finely ground and weighed (0.5g-1g) to determine the metal content. Each sample was mineralized with 10 ml nitric acid and 2 ml oxygenated water, ultrapure quality reagents. Berzelius glasses were covered with clock glass, initially digestion at room temperature for 24 hours to destroy organic matter (cold mineralization). After cold mineralization, microwave digestion was performed until the remaining liquid became clear. The samples were filtered and brought to a 25 mL graduated flask, the residue was washed with ultralight water and the resulting water was collected in the volumetric flask.

4.3. EXPERIMENTAL DETAILS IN DETERMINATION OF METAL ELEMENTS IN AN AREA AFFECTED BY MINING ACTIVITIES

The water samples were collected in polyethylene containers, both for the determination of the total metal content and for the washing of the sediment, collected from the same points with the water samples, in order to retain the granulation of 63 µm.

The sediment samples were collected from 0-5 cm depths using a Van Veen (Wagtech) shallow water sampler; the samples were introduced into polypropylene containers that were previously washed with dilute hydrochloric acid. The samples were transported to the test laboratory in cold boxes and kept at 4ºC until the time of analysis.

In order to evaluate soil pollution, 11 pedological profiles were established, which were representative in all the areas adjacent to the objectives under analysis and from which 22 soil samples were taken on two depths: 0-10 cm and 30-40 cm. Also, soil samples - control (0-10 cm and 30-40 cm) were taken from an open profile in an area considered to be unaffected by the activities carried out on the site under analysis. The sampling was carried out with the Buerkle soil sampling probe.

Vegetation samples were taken from the immediate vicinity of the soil sampling point, with the aim of harvesting various species (leaves; root plants, stem and leaves; tree bark; buds). All the vegetation (except the tree bark and a species of dry grass) was young, freshly sprouted. The collected samples were stored in plastic bags. At the reception of the samples in the laboratory, the vegetation was sorted and sampled, the separation by species being made with the support of 2 specialists in botany, specialists who also established the Latin name of the samples.

4.4. EXPERIMENTAL DETAILS ON MERCURY DETERMINATION HG-ICP-OES AND AAS- CV

4.4.3. Sampling and conditioning of water samples and calibration standards

Because mercury vapors can diffuse through different plastics, only glass containers or FEP (perfluoroethylene-propylene) tubes will be used to collect mercury.

Tap, spring and waste water were collected in borosilicate bottles. All the glassware used to determine the mercury was washed and left overnight in 10% nitric acid and then rinsed with ultrapure water. The samples were preserved with potassium dichromate / nitric acid for method A and for methods B and C 0.5 mL HCl and potassium bromide / potassium bromate at least 24 hours before determination.

CHAPTER 5

DEVELOPMENT OF NEW METHODS OF SIMULTANEOUS DETERMINATION OF SOME PLATINIC METALS BY ICP-OES TECHNIQUE WITH APPLICATIONS IN THE EVALUATION OF SOIL PROBES AND VEGETATION IN THE PROXIMITY OF NATIONAL ROADS

5.1. DEVELOPMENT OF AN ICP-OES-BASED METHOD FOR DETERMINING IR, Pd, Pt, Rh and Ru

The methods for determining the platinum metals Ir, Pd, Pt, Rh and Ru were performed by ICP-OES as follows:

-with ultrasonic nebulizer coupled to inductively coupled plasma atomic emission spectrometer (µg/L)

- directly in plasma (mg/L).

Thus, the experimental tests were performed on the concentration range 10 ÷ 50 µg / L (USN-ICP-OES), integrating the signal obtained both by area and peak height, but also by the concentration range 0.1 ÷ 0, 5 mg / L (ICP-OES). All the wavelengths specific to each element were tested (Table 5.1), selecting only those at which the performance characteristics presented corresponding values for a method of determination by ICP-OES.

Table 5.1. Wavelengths specific to each platinum element

Metal	Wavelengths	Metal	Wavelengths	Metal	Wavelengths
Ir	205,222 nm	Pd	324,270 nm	Pt	193,700 nm
Ir	224,268 nm	Pd	248,892 nm	Rh	343,489 nm
Ir	208,882 nm	Pt	265,945 nm	Rh	233,477 nm
Ir	237,277 nm	Pt	274,423 nm	Ru	240,272 nm
Pd	340,458 nm	Pt	299,797 nm	Ru	349,894 nm
Pd	363,470 nm	Pt	204,937 nm	Ru	279,535 nm

Table 5.7. The values of the performance parameters for platinum metals method 10 ÷ 50 µg / L, integration according to the peak area

Metal Λ, nm	**LOD** µg/L	**LOQ** µg/L	Accuracy µg/L	**RSDr*** %	**RSD_{Ri}*** %	**Uex***** %	Recovery %	Linearity
Ir 224,268	1,36	4,50	0,6 ÷6,8/30 1,8 ÷4,2/50	2,89	4,91	7,21	83,3 ÷ 95,5	R=0,9993 PG1/10=0,09 <5,35
Ir 208,882	1,5	5,00	2,1 ÷1,7/30 2,9 ÷4,4/50	1,92	4,90	12,2	91,3 ÷ 107	R=0,9988 PG1/10=0,04 <5,35
Pd 248,892	1,20	4,00	2,2 ÷3,9/30 1,9 ÷5,9/50	2,56	3,35	13,3	81,2 ÷ 105	R=0,9996 PG10/1=1,41 <5,35
Rh 343,489	0,60	2,00	0,6 ÷0,9/30 0,3 ÷3,6/50	3,48	5,84	7,44	98,8 ÷ 105	R=0,9999 PG1/10=0,06

								<5,35
Ru 349,894	0,72	2,40	1,3 ÷6,4/30 1,7 ÷12/50	2,26	3,26	11,2	75,7 ÷ 96,6	R=0,9999 PG10/1=4,76 <5,35
Ru 240,272	0,31	1,00	3,0 ÷8,0/30 6,3 ÷14/50	2,72	3,92	9,97	71,9 ÷ 88,3	R=0,9999 PG10/1=5,19 <5,35

5.8. APPLICATION OF THE M1 METHOD FOR DETERMINATION OF PLATINIC METALS FROM SOIL AND VEGETATION

The platinum metal determinations from the road and vegetation samples were performed by ICP-OES and ICP-MS (Bruker). It is observed that both the iridium, ruthenium and platinum content are below the limit of quantification of the applied method (50 µg / kg at Ir, 10 µg / kg at Ru and 90 µg / kg at Pt), while rhodium concentrations are low, below 20 µg / kg in both dust and vegetation (Tables 5.11 and 5.12).

Pd concentrations are of the order of hundreds of micrograms per kg dry matter in soil and of the order of tens of micrograms per kg dry matter in vegetation, the statistical processing of the results obtained is presented in Table 5.13 and Table 5.14 respectively.

Table 5.13 PGEs results of the soil samples in comparison with the literature data (mg/kg dm)

Metal	**Ir**	**Pd**	**Pt**	**Rh**	**Ru**
Medium value	<0,05	0,496	<0,09	0,008	<0,01
Minimum	<0,05	0,309	<0,09	0,003 (<0,005)	<0,01
Maximum value	<0,05	0,794	<0,09	0,014	<0,01
Standard deviation	-	0,127	-	0,002 (<0,005)	-
Crust - benchmarks					
Fersman [101]	0,01	0,05	0,2	0,01	0,05
Vinogradov [102]	0,001	0,01	0,005	0,001	0,005
Greenwood [103]	0,001	0,015	0,001 – 0.005	0,0001	0,0001
Linde [104]	0,001	0,015	0,005	0,001	0,001
Dust concentrations - other areas studied					
Dust concentrations - other areas studied	0,0039	0,0708	0,158	0,0179	0,0016
Moscova [106], medie Minimum value Maximum value	0,0039	0,0708	0,158	0,0179	0,0016
	0,0016	0,0077	0,0124	0,0027	0,0007
	0,0062	0,2253	0,3566	0,0545	0,0022
UK [107]	-	0,0093	0,070	-	-
Israel [108]	-	-	1,230	0,381	-
Madrid [109]	-	0,317 (medie) 2,25 (max)	-	0,074 (medie) 0,182 (max)	-
Viena [110]	-	0,20 -1,23	0,21-1,45	-	-

CHAPTER 6

DETERMINATION OF VARIATION IN THE TIME OF METAL CONTENT BY ICP-OES TECHNIQUE FROM MEDICINAL PLANTS GROWED ON SOILS RICH WITH METALS

6.3. DETERMINATION OF THE METAL CONTENT IN *Salvia Officinalis*

6.3.3 Heavy metal accumulation factors in sage

In order to evaluate the plant's ability to accumulate metals from the soil and to move them from the root to the aerial part of the plant, the following coefficients were calculated: the transfer coefficient (TC), the translocation factor (TF) and the enrichment factor (EF).
The transfer coefficient (TC) (or bioaccumulation factor) is defined as the ratio of the concentration of metals in the root and their content in soil (mg / kg) [141]. This provides information about the plant's ability to accumulate metals from the soil.
TC> 1: The plant accumulates metals
TC <1: The plant does not absorb metals [147].
The translocation factor (TF) is calculated as the ratio of the metal concentration in the upper aerial part of the plant to that of the roots [141].
TF> 1: the plant translocates the metals from the root to the surface of the plant.

Enrichment factor (EF) can be used to assess the level of soil pollution (degree of soil contamination) and accumulation of metals in plants growing on contaminated soil relative to soil and plants grown on control soil [148]. It is the ratio between the metal concentration in the contaminated soil / plant and the metal concentration in the soil / uncontaminated plant.
EF> 1 indicates a greater availability and distribution of metals in contaminated soil and, consequently, an increase in the accumulation of metals in plant species grown on contaminated soil relative to their reference values.
The transfer coefficients (TC) calculated as the concentration of metal at the root of the plant inrelation to the concentration of metal in the polluted and control soil are lower than one (Fig. 6.20) for all metals except cadmium (whose value is 1.45 in control sample and 3.54 in the polluted one) and lead (in the polluted sample it reaches 1.67). These values that exceed the value one indicate that the plant is accumulating Cd and Pb.

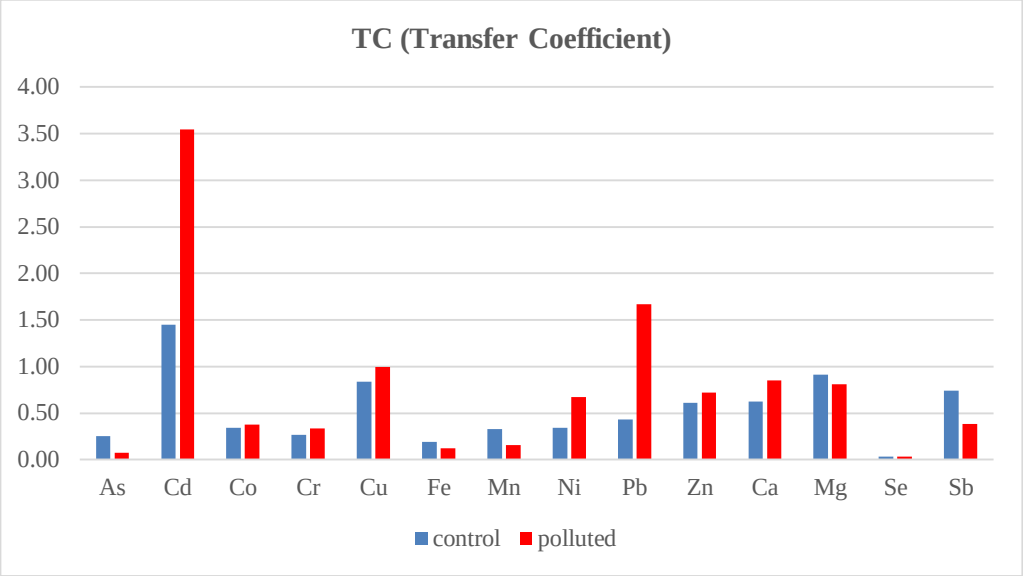


Fig.6.20 TC for sage control and polluted

The enrichment factor (EF) in the polluted soil, the root, stem and leaves of the sage harvested in October are shown in Fig. 6.23. From the calculation of the enrichment factor, a maximum enrichment is observed for Pb in the root (19.02), for Co in the stem (10) and for cadmium in the root (9.66). Although the EF value for lead is very high at the root, the value of this element in the leaves is relatively small (1.46) but above one which indicates an accumulation of lead in the leaves. It should be mentioned that sage leaves are the most used part of this plant.

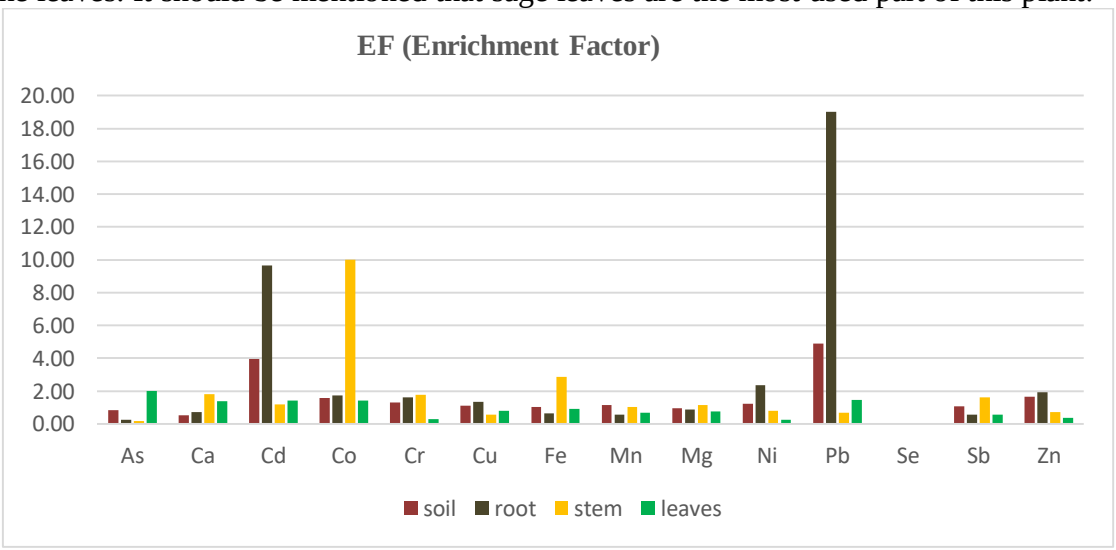


Fig.6.23 EF for sage

6.4. DETERMINATION OF METAL CONTENT FROM *Ocimum Basilicum*

The purpose of this study was to evaluate the effects of heavy metals ecotoxicity on plant growth and accumulation of metals in basil grown on unpolluted and polluted soils. The aromatic basil plant was evaluated in a laboratory experiment using soil contaminated with Cd, Co, Cr, Cu, Ni, Pb and Zn, similar to the one in the mining area. The accumulation of these metals in soil and in different parts of the plants was studied, being made different correlations. Transfer coefficients / bioaccumulation factors, translocation factors, enrichment factors and accumulation indices were also calculated to assess the ability of plants to accumulate metals for phytoremediation purposes.

The basil grown on the control soil had a slower development than the basil from the polluted soil. Thus, despite similar environmental conditions, the height of the plant grown in the polluted soil (42 ± 5 cm) was double that of the basil in the control soil (20 ± 4 cm), being also more vigorous. The length of the basil leaves grown on the control soil ranged from 2 to 5 cm, while the basil leaves in the soil contaminated with metals were between 4 and 9 cm. Moreover, the color of the basil leaves planted in the polluted soil was intensified to darker green than the leaves of the control sample, indicating an active process of photosynthesis. Also, it should be noted that only the basil from the polluted soil bloomed in September and after flowering the plant was harvested in October.

6.4.1 Metal content in soil

The metal content of the control soil and polluted soil samples was determined before basil cultivation (March) and after harvesting (in October) (Table 6.7). The total concentrations of Cd, Co, Cu, Pb and Zn were not very different in the control soil samples from March and October. In contrast, the concentrations of Cr and Ni were about three, respectively, twice lower at the beginning of the study than in October.

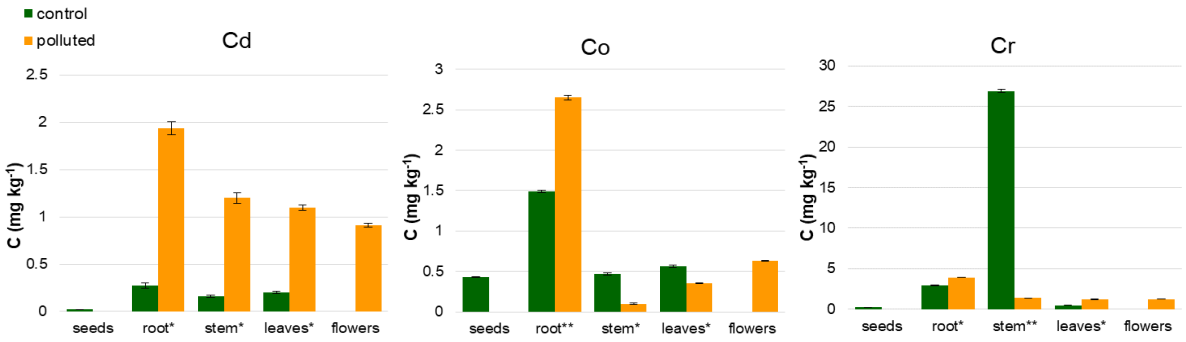
Table 6.7 Metal concentration (mean ± standard deviation) in control soil and polluted soil

Metal	Control soil (mg/kg)		Polluted soil (mg/kg)		t test		Added metal (mg/kg)
	March	October	March	October	March	October	
Cd	0.43 ± 0.03	0.34 ± 0.01	8.20 ± 0.57	4.97 ± 0.21	*	*	5
Co	7.85 ± 0.06	8.22 ± 0.03	25.90 ± 0.11	19.30 ± 0.11	**	**	10
Cr	54.30 ± 0.28	17.50 ± 0.06	73.00 ± 0.23	27.70 ± 0.20	**	**	10
Cu	18.50 ± 0.14	18.80 ± 0.28	48.40 ± 071	29.10 ± 0.28	**	**	15
Ni	33.20 ± 0.42	17.50 ± 0.42	51.00 ± 1.13	27.00 ± 0.85	*	*	5
Pb	10.20 ± 0.28	13.70 ± 0.28	407.00 ± 4.24	280.00 ± 5.66	**	**	350
Zn	74.30 ± 0.71	78.10 ± 0.71	207.00 ± 2.83	175.00 ± 2.83	**	**	100

* P < 0.05, ** P < 0.01

6.4.2 Concentration of metals in different parts of the plant

The accumulation of metals studied in the root, stem, leaves and sweet basil flowers was evaluated in October. The seeds were analyzed before planting in March. The roots of the polluted basil accumulated more of each metal than the corresponding plant grown in the control soil (Fig. 6.24). The Cd, Pb and Zn content of the strain and the concentrations of Cd, Cr, Ni and Zn in the leaves of basil grown on polluted soil were higher than those obtained for the control samples. Moreover, as can be seen from Fig. 6.24, the statistical analysis showed these significant differences.



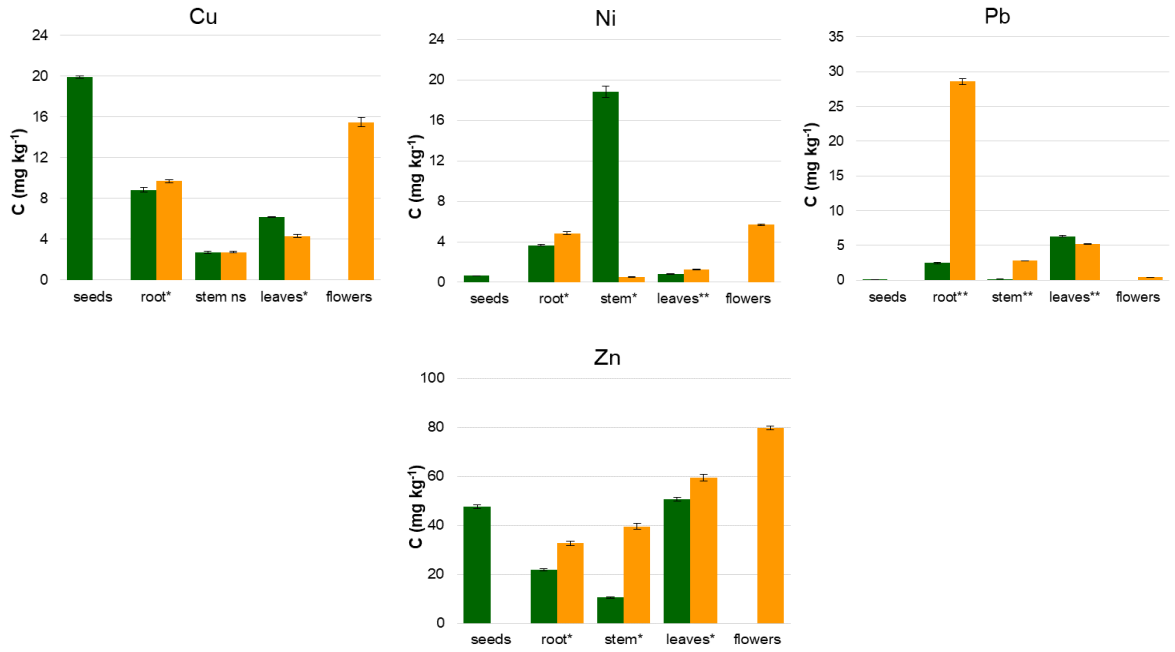


Fig. 6.24. Concentration of metals in different parts of basil in control and polluted samples

There are statistically significant differences between the different parts of the plant in the polluted basil, with a few exceptions, especially for cadmium, as can be seen in Table 6.8.

Table 6.8. Differences in different parts of plants for heavy metals

		Cd	Co	Cr	Cu	Ni	Pb	Zn
Root - stem	**Control**	ns	**	**	**	*	*	**
	Polluted	**	**	**	**	*	**	ns
Root - leaves	**Control**	ns	**	**	*	**	**	*
	Polluted	ns	**	**	**	*	**	*
Stem - leaves	**Control**	ns	**	**	*	*	**	*
	Polluted	ns	**	*	*	**	**	**

ns: P > 0.05, *: P < 0.05, **: P < 0.01

The analysis of the different parts of the plant is important when estimating the degree of absorption of the metal and the mobility of the elements in the plant. The absorption and accumulation of metals in different parts of the plant are significantly influenced by the concentration of metal in the soil and the distribution of metals in the plant tissues, being an indicator for evaluating the potential of plants in remedying contaminated sites.

6.4.4 Factors accumulation of heavy metals in the basil

Concentrations of metal detected in soil and different parts of basil were used to calculate the transfer coefficient, the translocation factor, the enrichment factor and the geo-accumulation index.

All the transfer coefficients calculated as the concentration of metal at the root of the plant divided by the polluted concentration and control of the metallic soil are lower than one (Fig. 6.26), varying between 0.10 for Pb in the polluted samples and 0.79 for Cd in the control samples.

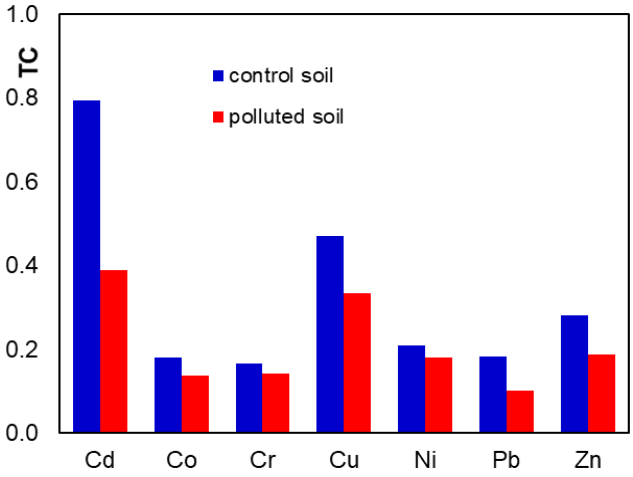


Fig. 6.26. Coefficient of transfer of metals from the soil to the root of the basil

The translocation factor was calculated as the ratio of the total metal concentration in the stem, leaf or flower to the root metal content (Fig. 6.27).

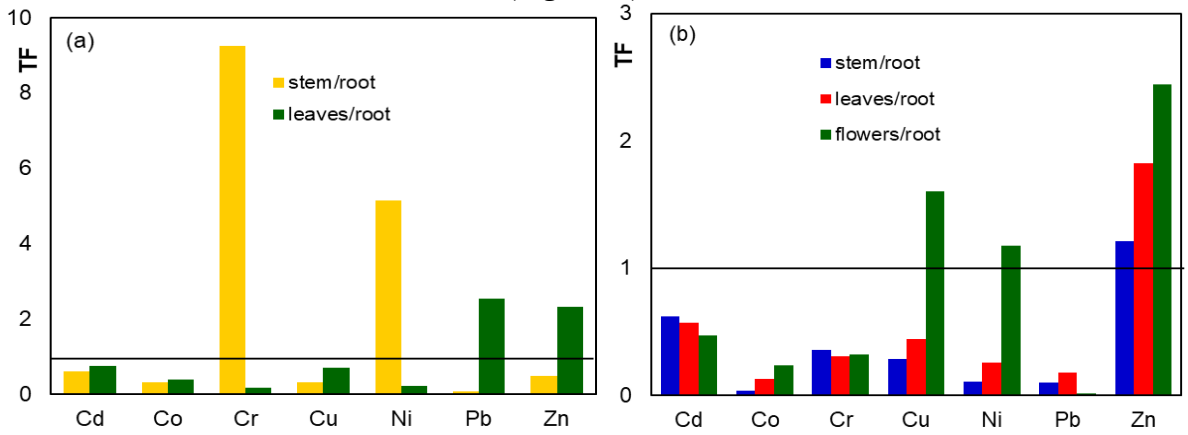


Fig. 6.27. The translocation factor from the root to the aerial parts of the plant (a) control sample, (b) polluted sampl

The translocation from the roots to the stem is significant for Cr and Ni in the case of uncontaminated soil, whereas a TF value greater than one was obtained only for Zn for the soil enriched with heavy metals. Regarding the translocation from the root to the leaves, TF values higher than 1 were found for Pb and Zn for the control soil and for Zn in the case of the contaminated soil. The transfer factor from flower to root can only be calculated for contaminated soil: Zn> Cu> Ni> Cd> Cr> Co> Pb.

Plants are good indicators of soil quality, monitoring metal concentrations in plant tissues, indicating the presence and degree of environmental pollution.

The coefficient of transfer from soil to plant is influenced by different factors, such as the restrictive absorption of the roots, the translocation from the root to the aerial parts of the plant or the bioavailability of metals [140]. The transfer coefficients were lower than one, indicating that basil did not accumulate metals at the root. Root bioaccumulation factors in the control

soil indicate higher values compared to the enriched soil which needles could be one reason why the control plants were less developed.
As already mentioned above, TF values higher than 1 were obtained for the control samples for Cr and Ni (stem / root) and for Pb and Zn (leaves / roots), and in the case of polluted samples for Zn (stem / root and leaf / root) and for Zn, Cu and Ni (flowers / root).

6.4.5 Conclusions regarding the accumulation of metals in basil

The plant biomonitoring is an affordable and valuable tool for the study of the effect of different pollutants like heavy metals. Thus, the simulation, in a laboratory experiment, of the sweet basil growth on a contaminated soil similar to one polluted by mining activities has shown that the accumulation of metals in plants organs was different: Cd, Co, Cr and Pb were accumulated in roots; Cu, Ni and Zn in flowers. Cr and Pb exceed the toxic levels in roots. The intake of heavy metals depends, also, on the plants development stage, thus Cd, Cr and Pb were accumulated more in the mature plants leaves. Taking into consideration that the Cd and Pb contents were higher than the WHO and EC permissible limits in herbal medicines or food supplements it is not safe to consume the plant grown on this type of contaminated soil.

CAPITOLUL 7

RESEARCH ON EVALUATION OF METAL POLLUTANT CONCENTRATIONS IN A AREA IMPACTED BY MINING ACTIVITIES USING THE ICP-OES

7.1. DETERMINATION OF METAL CONTENT IN WATER AND SEDIMENTS

The objective of this research was an in-depth study of the environmental compartments (surface water, sediments, soil and vegetation) from a river sector and its tributaries, in the vicinity of the mining area and the tailings dumps in Certejului Valley, Deva, Hunedoara County, in order to highlight the high pollution points.

Thus, the content of metals (As, Ag, Cr, Co, Hg, Ni, Sb, Mo, V, Ti, Te and W) was monitored in sediment, surface water and vegetation samples from Baiaga stream and its tributaries from Certejului Valley, as well as the content of heavy metals from soils polluted by mining. Also, within the study was carried out a model of distribution of metals on various environmental compartments (surface water, sediments, soil, vegetation), following the transfer of metals between these compartments.

In order to assess the degree of pollution with heavy metal sediments have been used five parameters: the contaminant (CF), the index of geo-accumulation (IgeoE), the index of ecological risk and load index of pollution (PLI) Index of Bioavailability (Ibio).

To achieve goals, it has conducted a sampling campaign in which they collected 10 sediment samples and 10 samples of surface water in the area under study - Basin Certej (jud. Hunedoara).

In the following, the sediments are classified using the geo-accumulation indices calculated and presented in Table 7.17, a classification in which the Igeo values for the 7 metals taken in the study were taken into account (Table 7.18).

Table 7.18. Quality class according to geo-accumulation indices

Sediment	$\mathbf{I_{geo}}$	Sediment	$\mathbf{I_{geo}}$	Sediment	$\mathbf{I_{geo}}$	Sediment	$\mathbf{I_{geo}}$	Sediment	$\mathbf{I_{geo}}$
Sd_1	4	**Sd_3**	6	**Sd_5**	4	**Sd_7**	5	**Sd_9**	6
Sd_2	4	**Sd_3**	5	**Sd_6**	5	**Sd_8**	5	**Sd_{10}**	4

Table 7.19 presents the ecological risk factors calculated for each sediment and the 7 metals taken in the study, factors used to estimate 3 pollution indicators, namely: CH pollution coefficient, RI ecological risk index and pollution load index PLI (Table 7.20).

Table 7.19. Ecological risk factors on metal / sediment

Nr.crt	Metal	**Ecological risk factor EF**									
		Sd_1	**Sd_2**	**Sd_3**	**Sd_4**	**Sd_5**	**Sd_6**	**Sd_7**	**Sd_8**	**Sd_9**	**Sd_{10}**
1	As	188	108	1210	413	204	404	323	238	531	124
2	Cr	0.62	1.1	3.24	1.08	2.64	0.66	5.5	1.3	3.1	2.8
3	Hg	29.2	10.8	42.8	6.8	14.8	65.2	93.2	41.2	161	99
4	Ni	9.36	3.9	11.58	3.72	53.4	4.02	66	6.7	40.3	28.4
5	Pb	2.25	1.5	15.2	2	3.2	25.1	26	11	27.5	10.8
6	Cd	690	393	3990	1305	672	1392	1260	828	1848	507
7	Cu	4.45	8.5	19	4.8	4.75	4.5	37.3	9.5	47.5	20

7.1.3 The distribution of metals in the surface water system - sediment

Although in sediments, the total As content is in very high concentration (314 ÷ 3497 mg / kg su), concentrations higher than the maximum value allowed in sediment from 11 to 121 times, in the mobile fraction the arsenic is found only in 3 sediments (Sd3, Sd4, Sd7) the concentration being <LOD in the other 7 sediment samples. In surface water, arsenic concentrations were determined in the range <LOD ÷ 1070 µg / L, the highest being recorded in sample P3. All samples (except for sample P3) fall into the third grade class, having concentrations less than 50 µg / L. Arsenic concentrations in surface water relatively low compared to large amounts of arsenic in sediment correlate with calculated bioavailability indices. For example, in Sd3 sediment, the bioavailability index of 0.03% applied to the concentration of 3497 mg / kg su indicates a concentration of 1090µg / L, concentration in mobile fraction and concentration found in surface water.

	Sd_1/P_1	Sd_2/P_2	Sd_3/P_3	Sd_4/P_4	Sd_5/P_5	Sd_6/P_6	Sd_7/P_7	Sd_8/P_8	Sd_9/P_9	Sd_{10}/P_{10}
Sed										
Apa										
I_{bio}										

Fig. 7.3. Distribution of As in the aquatic compartment

Legend:

Sediment	< 1mg/kg su	1÷10 mg/kg su	11÷100 mg/kg su	101÷1000 mg/kg su	>1000 mg/kg su
Surface water	Class 1	Class 2	Class 3	Class 4	Class 5
	> 1 µg/L	1 ÷10 µg/L	>10÷50 µg/L	>50÷100 µg/L	>100 µg/L
Bioavailability index	0 ÷ <1%	> 1 ÷ 20%	> 20÷ 35%	>35 ÷ 65%	>65 ÷100%
Color					

Regarding the Cd content, the determined values are located in all the sediments analyzed over VMA (0.8 mg / kg su), the concentration range being between 17.9 and 106 mg / kg su, exceeding 22, 3 up to 132.5 times higher. The bioavailability indices of Cd in sediment

fall in the range 2.8 ÷ 13.9%, which is reflected in the quality of the surface water in which Cd is found in high concentrations (155 ÷ 1206µg / L), classifying the waters in V class for quality.

	Sd_1/P_1	Sd_2/P_2	Sd_3/P_3	Sd_4/P_4	Sd_5/P_5	Sd_6/P_6	Sd_7/P_7	Sd_8/P_8	Sd_9/P_9	Sd_{10}/P_{10}
Sed										
Water										
I_{bio}										

Fig. 7.8. Cd distribution in the aquatic compartment

The Sb content is around 25 mg / kg su for Sd1, Sd2, Sd4, Sd5, Sd10, 60 mg / kg su sediments (Sd7, Sd8), the highest concentration being recorded at Sd3 (174 mg / kg su). The percentage of bioavailability is low, below 1%, reflected in the quality of surface water, where the Sb content is below 10 µg / L in most samples, except for samples P3 and P6, where Sb concentrations of 61 µg / were recorded L, respectively 20.4 µg / L. In the surface water, the Sb content is abnormal. The concentrations correlate with high Sb values in the sediment, over 100 mg / kg su and bioavailability indices greater than 0.1%.

	Sd_1/P_1	Sd_2/P_2	Sd_3/P_3	Sd_4/P_4	Sd_5/P_5	Sd_6/P_6	Sd_7/P_7	Sd_8/P_8	Sd_9/P_9	Sd_{10}/P_{10}
Sed										
Water										
I_{bio}										

Fig. 7.12. Sb distribution in the aquatic compartment

7.2. DETERMINATION OF METAL CONTENT IN SOIL AND VEGETATION

Soil and vegetation samples were taken from the Certej River Basin (Hunedoara County). 10 soil samples were taken, each of them on 2 depths (0-10 cm, respectively 30-40 cm), as well as 43 vegetation samples.

7.2.1 Determination of the metal content in soil samples

Table 7.22 presents the results of the physico-chemical analyzes obtained for the soil samples taken from the reference profile on two depths (0-10 cm and 30-40 cm) - sampling point located at about 5 km on the SV direction of Coranda Quarry.

The analysis of the values determined for the control samples reflects:

- the existence of an acidic pH on both levels of sampling;
- on the first sampling level: the classification of the quality indicators Cr and Co in the normal reference value, according to the MAPPM Order no. 756/1997 and exceeding the alert threshold for the Pb quality indicator for soils with sensitive uses;
- on the depth 30-40 cm: the quality indicators Co, Mn and Zn are below the normal reference value. All other quality indicators investigated are below the alert threshold for soils with sensitive uses.

The values obtained on the two sampling levels for the pH of soils, sulphates and for the quality indicator Fe were compared with the values obtained in the reference profile (control), and the values obtained for the rest of the quality indicators were compared on both harvesting levels with the values normed by MAPPM Order no. 756/1997, for soils with "sensitive uses".

Table 7.22 - Physico-chemical analysis results for the control sample

Indicator	MARTOR			Reference values for soils with sensitive uses (The order MAPPM 756/1997)		
	UM	SM/1	SM/2	Normal value (VN) (mg/kgsu)	Alert threshold (PA) (mg/kgsu)	Intervention threshold (PI) (mg/kgsu)
pH	unit. pH	4,81	4,68	-	-	-
umiditate	%	2,71	2,46	-	-	-
Cd	mg/kg su	1,27	1,19	1	3	5
Cr tot	mg/kg su	29,07	32,16	30	100	300
Co	mg/kg su	8,74	8,87	15	30	50
Cu	mg/kg su	70,48	68,74	20	100	200
Fe	mg/kg su	11584	11046	-	-	-
Mn	mg/kg su	1044	871	900	1500	2500
Ni	mg/kg su	26,23	29,75	20	75	150
Pb	mg/kg su	**52,65**	48,80	20	50	100
Zn	mg/kg su	105,8	98,12	100	300	600
As	mg/kg su	7,69	6,59	5	15	25
Sulfati	mg/kg su	803	766	-	2000	10000
Humus	%	0,71	0,68	-	-	-
Hg	mg/kg su	0,59	0,23	0,1	1	2

7.2.3 Metal content in the vegetation bordering the Certej mining area

In the vegetation samples were determined 14 metallic elements, namely: Al, As, Ca, Cd, Co, Cr, Cu, Fe, Mg, Mn, Ni, Pb, Se and Zn, metals for which were found in literature indications on normal content in plants. Vegetation was collected from 6 points bordering the area polluted by the mining activity and a blank sample, located 5 km along the career direction SV. From each location several samples were selected so that, in total, 40 vegetation samples were analyzed, including different types of leaves, plants, buds and tree bark. Harvesting was done in early spring, when the vegetation was in the budding period. Correlations will be made between soil and plant samples for the following metals Cd, Cr, Cu, Ni, Pb and Zn.

7.2.4. Bioavailability indices for soil metals

Soil quality was evaluated using pollution and bioavailability indices in order to highlight the possible pollution generated by a decommissioned mining area in Certej, Hunedoara County. The metals Cd, Cr, Cu, Ni, Pb and Zn were studied in pseudo-total and mobile form and correlated with the content found in the vegetation harvested from the same points. High mobility was observed for Ni, Pb and Zn, respectively moderate mobility for cadmium.

The contamination factor (C_d), which summates the individual contamination factors, is 24.4 if the normal value of the soil is used as the reference value, respectively 31.9 when the normal value of the terrestrial crust is used as the reference value.

7.2.4.1.Contamination factors

Table 7.47. Contamination factor calculated by reference to the earth's crust

	Cd	**Cr**	**Cu**	**Ni**	**Pb**	**Zn**
Ci	1,25	27,2	61,9	84,6	224	369
Cn crust*	0,15	102	60,0	84,0	14,0	70.0
C_f c	**8,33**	0,27	1,03	1,01	**16,0**	5,27
$C_d = \Sigma C_f$ c	**31,9**		$mC_d = C_d/6$		**5,32**	

Cn crustă* [186]

7.2.4.2 The enrichment factor

In the calculation of the enrichment factor were used the reference values from the terrestrial crust, normalizing to the iron content. The factors were calculated for all the collected samples and the average value for each element and the standard deviation were calculated. In Table 7.49 are presented the enrichment factors (minimum, maximum, average, standard deviation).

Table 7.49. The enrichment factor (EF)

		Cd	**Cr**	**Cu**	**Ni**	**Pb**	**Zn**
	Min	1.37	0.16	0.65	0.18	3.93	3.37
EF	Max	**182**	1.86	**15.1**	**19.0**	**163**	**75.6**
	Mean	47.8	6.22	2.98	2.90	55.9	15.6
	SD	45.6	0.57	3.86	5.51	58.3	19.4

The highest EF values were recorded at Cd and Pb. Regarding the Cd content, 45% of the samples are in the extremely high enrichment class (EF> 40), 30% being included in the very high enrichment domain (20 <EF <40), the average value of all factors being located above the maximum value of 40. EF indices with very high values are found in soils S2, S3, S4, S5 and S10. The highest value was recorded in sample S10 (first level), where EF is 182.

For Pb, 40% of the samples are in the extremely high enrichment class, 20% each being in the moderate enrichment classes or significant to very high enrichment classes. The samples with high EF factor values are S2, S3, S5 and S10. The average EF value for all soil samples is in the case of Pb above the maximum value of 40, corresponding to an extremely high enrichment (55.9). The highest values were determined in soil samples S3, respectively S5 (values greater than 150).

7.2.4.4. Bioavailability index

In S1 samples it is observed that the Ibio bioavailability index is very high for the metals Cd, Cu, Ni and Zn (> 70%), even though the pH of the soil is in the neutral domain, which implies the smoothing of these metals towards vegetation and water. underground (table 7.51). It should be mentioned that at 45% Ibio in the case of lead, due to its high metal content in a pseudo-total form, the mobile fraction exceeds the value of the alert threshold for land with less sensitive use. The other contents of metals in mobile form, except lead, are around the normal value of the soil.

In the case of S2 samples with acid pH (pH = 4) it is observed that, although the Pb values exceed the intervention threshold, the mobile content (Ibio = 5 ÷ 15%) is low, being around the normal value. The other metals in mobile form do not exceed the concentrations of normal values in the soil.

In the case of S3 samples, which are highly polluted and present pH in the acidic domain (pH = 3.6), it is observed that the mobile fraction is above the intervention threshold in the case of

Pb and Ni even though Ibio is in the range 15 ÷ 30%. Cd is 60% bioavailable but the obtained values are below the alert threshold.
In S4 samples (pH = 5.7) the total metal contents are either low (Cr, Cu, Ni and Zn) or the percentage of bioavailability is low (Pb, Cd), which causes the values of the mobile fraction to be in the normal range concentration.
For S5 samples (pH = 3.6) low percentages of bioavailability are recorded (2 ÷ 38%), so that all the mobile values (except Pb in the depth layer where the alert threshold exceeds) are lower than the normal values.
S6 samples (neutral pH) have very high bioavailability percentages (30% - 77%), but the reduced metal content in a pseudo-total concentration leads to contents in mobile form below normal values.
A situation similar to S6 is found in samples S7, S8 and S9, but these have pH in the acid domain (S7, pH = 3.7, S8, pH = 5.5 and pH = 5.2 in S9).
In the case of S10 samples, although the pH is neutral, due to the very high metal contents and also due to the bioavailability percentages in the 22 ÷ 77% range, the mobile metals Cd (first depth), Cu, Pb and Zn are above the threshold value. intervention, representing a danger to both vegetation and to the pollution of ground and surface water.
The values of metals in the mobile fraction recorded in the control sample (pH = 4.8) are below or around the normal value.
As mentioned in our previous study [190], in which we investigated the quality of vegetation harvested in early spring from the studied area of the Certej area, significant amounts of Cd, Ni, Pb and Zn were detected in the vegetation samples. Thus, both leaf and root contents accumulated above normal values in the vegetation for Ni (70% of the samples collected), correlated with either high Ibio indices or acid pH. A low percentage of 19% of the vegetation samples showed Pb values above the maximum allowed limit, despite the very high content of lead in the soil, but correlated with the relative low mobility of this metal. The accumulation of Pb content in the bark of trees (*Carpinus betulus*) has been observed over time.
Regarding the Cr and Cu content, in all 36 vegetation samples collected they fall within the normal concentration range, either due to the low pseudo-total content (Cr, Cu) or due to the low mobility (Cu).
A low percentage of 11% of the vegetation samples contains Cd quantities above the maximum allowed value in both leaves and bark (Carpinus betulus). Regarding the zinc content, 25% of the vegetation samples contain Zn over the upper value of the allowed range, content found in the root, bark, leaves. The highest concentrations are found in the vegetation collected from points S3 and S10, correlated with both the high zinc content and the high Ibio percentage.

CHAPTER 8
ORIGINAL EXPERIMENTS FOR HG-ICP-OES MERCURY DETERMINATION AND ATOMIC ABSORPTION SPECTROMETRY WITH COLD STEAM (AAS- CV)

8.1. COMPARISON BETWEEN METHODS FOR DETERMINING MERCURY BY HG-ICP-OES AND AAS- CV

This comparative study shows the determination of mercury in water samples, at very low concentrations (µg/L). Mercury was determined by two different techniques: cold vapor atomic absorption spectrometry (CV-AAS) and inductive plasma optical emission spectrometry coupled with the hydride generator (HG-ICP-OES).

Monovalent or divalent mercury and organo-mercury compounds are converted to divalent mercury by oxidation with bromine-bromide (KBrO3-KBr) or a mixture of potassium permanganate (KMnO4) and potassium peroxodisulfate (K2S2O8).

For the detection of mercury by the HG ICP-OES technique, the sample was pretreated (both for calibration standards and for water samples) by two modes:

- with a mixture of acids and oxidants (sulfuric acid, nitric acid, potassium permanganate, potassium bichromate and potassium peroxodisulfate) and heating on an ultrasonic bath - method A

-with hydrochloric acid and bromine-bromide solution - method B. For mercury reduction

sodium hydroxide solution (NaBH4) 0.3% in sodium hydroxide solution (NaOH) 0.5% was used.

To determine the mercury using the CV-AAS technique, pretreatment of the samples was performed using only hydrochloric acid and bromine-bromide solution. In this case the reducing agent was tin chloride (SnCl2) of 10% concentration - method C.

The coefficient of variation (Hg - 0.84% for method A; 1.45% - for method B and 1.25% - for method C) is less than 3%, including the methods tested in the category of good analytical methods [193,194]. For a concentration level of 10 µg / L the recovery percentage should be in the range of 60 to 115% [195]. For the determination of mercury by the three methods, the experimental results were in the range 92.8% -104.3%. The limits of detection and the limits of quantification for methods A and C allow the determination of mercury from samples of drinking water and mineral water at the concentrations allowed by the legislation in force. The results for the limit of detection, limit of quantification, repeatability, accuracy and recovery for methods A, B and C are presented in Table 8.5.

Table 8.5. Performance parameters for Hg

Performance parameters	**Method A**	**Method B**	**Method C**
LOD and LOQ	LOD = 0.10 µg/L LOQ = 0.38 µg/L	LOD = 0.21 µg/L LOQ = 0.29 µg/L	LOD = 0.04µg/L LOQ = 0.12 µg/L
Repeatability, precision	X=8.821 µg/L s=0.285 µg/L r=0.806 µg/L RSDr= 3.23%	X= 5.36 µg/L s= 0.19 µg/L r= 0.54 µg/L RSDr= 3.57%	X= 6.01 µg/L s= 0.10 µg/L r= 0.27 µg/L RSDr = 1.62%
Recovery $\bar{x}_f \pm s_{xf}$, µg/L η_m	6 µg/L 6.70 ± 0.130 µg/L 92.8 %	5 µg/L 5.326 ± 0.094 µg/L 104.3. %	5 µg/L 4.937 ± 0.218 µg/L 96.7%

CHAPTER 9

DEVELOPMENT OF METHODS (M2-M5) FOR DETERMINATION OF AS, SE, Sb AND SN BY THE HG-ICP-OES METHOD

9.1. DETERMINATION OF ANALYTICAL PERFORMANCE PARAMETERS FOR ARSEN BY HG –ICP-OES (M2)

The determination of arsenic from water samples was performed by optical emission spectrometry coupled with the hydride generator (HG-ICP-OES). In the case of arsenic, As5 + ions are reduced to As3 + ions by adding 5 mL 5% KI and 5% ascorbic acid for 45 minutes at room temperature. Two types of freshly prepared solutions were used to generate arsenic hydride: 10% HCl carrier solution and 0.2% NaBH4 reduction solution in 0.05% NaOH.

Arsenic was determined at 3 wavelengths: 193.696 nm, 197.197 nm and 188.979 nm. Following the calculation of the performance parameters of the arsenic from drinking water, it was found that at the wavelength of Λ = 188,979 nm the lowest detection limit was determined, which is why the performance parameters for this wavelength will be presented widely, while for the other two wavelengths the final values will be presented.

Table 9.2. The performance parameters of the internally validated method for arsenic at the 3 wavelengths

Parametru statistic	**As 193,696**	**As 197,197**	**As 188,979**
Limita de detectie , LOD (µg/L)	0,12	0,33	0,09
Limita de determinare LOQ (µg/L)	0,40	1,08	0,29
Coeficientul de corelaţie, R	0,9995	0,999	0,9995
Abaterea standard a metodei, s_{x0}	0,19	0,30	0,15
Recuperare	100,98÷103,74	100,48÷102,39	98,43÷102,04
Abaterea standard procentuală a repetabilităţii, RSDr %	1,87	2,03	1,02
Coeficientul de variatie	1,77	2,77	1,39
Panta dreptei de regresie (sensibilitatea) µg/L	764	461,17	587,96

From the data summarized in Table 9.2 it is observed that the best performance characteristics (limit of detection, limit of quantification, standard deviation of the method and coefficient of variation) are achieved at the wavelength 188,979 nm. The validated method for quantitative determination of arsenic from drinking water, using inductive coupled plasma optical emission spectrometry - coupled with flow hydride generation, is specific and selective, linear, precise,

accurate, having the detection limit and the quantification limit suitable for the intended purpose [198].

III. FINAL CONCLUSIONS

PhD thesis *IDENTIFICATION OF CHARACTERISTICS OF SOILS, VEGETATION AND WATERS BY PHYSICO-CHEMICAL METHODS* make original contributions related to the characterization of the environmental compartments (surface water, sediments, soil and vegetation) from a river sector and its tributaries, as well as from the vicinity of a mining area. In this PhD thesis, 5 new methods for determining metals from environmental samples were developed: one method (M1) for platinum metals Ir, Pd, Pt, Rh, Ru based on inductive coupled plasma optical emission spectrometry and 4 methods (M2-M5) based on hydride generation inductively coupled plasma optical emission spectrometry (HG-ICP-OES) for elements that can generate hydrides (As, Se, Sb and Sn).
The research directions addressed were:
1. Elaboration of an original method (M1) based on ICP-OES for simultaneous determinations of platinum metals from environmental samples and application of this method on real soil and vegetation samples.
2. Determination of the time variation of the metal content by ICP-OES technique from medicinal plants grown on artificially polluted soils.
3. Researches regarding the presence of metallic elements in an area affected by mining activities through the ICP-OES technique.
4. Experiments for the determination of mercury using the HG-ICP-OES technique and cold vapor atomic absorption spectrometry (AAS-CV).
5. Elaboration and optimization of 4 new methods (M2-M5) for the determination of As, Se, Sb and Sn, metals that can generate hydrides through HG-ICP-OES.

The elaboration of the methods of determining platinum metals from water and vegetation samples was performed by the inductive plasma optical emission spectrometry technique coupled with ultrasonic nebulizer (USN-ICP-OES), over a range of 10 ÷ 50 µg / L for Ir, Pd , Rh and Ru and in the range 100 ÷ 500 µg / L for Pt. For each wavelength specific to each element, the performance parameters were determined and calculated: limit of detection, limit of quantification, linearity, homogeneity of dispersions test, repeatability, intermediate precision, accuracy and recovery. The wavelengths for the determination of metals in water and vegetation samples were selected according to the values of the performance parameters as follows: recoveries in the range of 60 ÷ 115% for concentrations of 30 and 50 µg / L; intermediate precision expressed in RSD between 15 ÷ 21% for concentrations in the range 10 ÷ 100 µg / L.

The most suitable methods for determining platinum metals from water and vegetation samples using USN-ICP-EOS are those in which the signal is integrated according to the peak area, in the concentration range 10 ÷ 50 µg / L, the specific wavelengths are selected as follows: Ir 224.268 nm; Pd 248.892 nm; Rh 343.489 nm; Ru 349, 894 nm.

Due to the higher quantification limit, platinum was verified in a range of 100 ÷ 500 µg / L when integrating the signal after the area, the best response being obtained in the case of the wavelength of 193,700 nm (Pt 193,700 nm).

The following wavelengths were selected for the determination of platinum metal content from soil samples: Ir 224.268 nm; Pd 248.892 nm; Pt 214,423 nm; Rh 343.489 nm; Ru 240.272 nm.

The determination of platinum metals from real samples was performed on 17 soil samples and 17 vegetation samples. The samples were collected from several European and national roads located in the eastern part of Romania, high traffic areas.
The concentrations of Ir, Pt, Ru were below the limit of determination of the method applied in both soil and vegetation samples; Comparing the values obtained in the dust samples on the road for Pd with the values obtained in other geographical areas, the maximum value is below that reported in Madrid in 2001; The concentrations recorded in the sections studied for Rh are close to the values recorded in Moscow in 2018; About 50% of vegetation samples have a translocation factor greater than 0.5, which indicates the presence of bioavailable palladium compounds in the dust on the road.
From the study on the variation in time of the metal content of medicinal plants (Salvia Officinalis and Ocimum Basilicum) grown on artificially polluted soils the following conclusions were obtained:

- The sage, grown on the control soil (without additional addition of metals) had a slower development than the sage raised on the soil enriched with metals. Thus, at maturity the leaves of sage grown on unpolished soil with metals had dimensions between 0.5 ÷ 2 cm while the leaves of sage grown on soil polluted with metals had dimensions between 2 ÷ 7 cm, and the stem of the control sage had a height 10-12 cm from the stalk of the unpolluted sage that was 30 cm high.
- The metals accumulate differently in different parts of the plant so that, in the polluted sage sample collected in October, an accumulation of metals (As, Cd, Co, Cr, Ni, Mn, Pb, Zn and Fe) is observed in the order: root> stem> leaves. The concentration for Cu and Sb is distributed in the sage as follows: root> leaves> stem, calcium concentration is in the order: leaves> root> stem, and for magnesium the distribution is: leaves> stem> root.
- The transfer coefficient (TC) indicates an accumulation only for Cd and Pb. The transfer factor (TF)> 1 from root to stem was obtained for Pb (uncontaminated soil) and Co (contaminated soil). The translocation from the root to the leaves is greater than 1 for Mg and Zn (uncontaminated soil) and for Ca and Mg (uncontaminated soil).
- In basil, the accumulation of metals in different parts of the plant was different compared to sage: Cd, Co, Cr and Pb were accumulated in roots; With, Ni and Zn in flowers. Cr and Pb exceed the toxic levels in the roots. Also, the contribution of heavy metals depends on the stage of plant development, so that Cd, Cr and Pb were more accumulated in the leaves of mature plants. Given that the Cd and Pb content was higher than the allowable limits in plants or food supplements of the World Health Organization and the European Commission, it is not safe to consume the cultivated plant on this type of contaminated soil.
- Although plants have accumulated most of the metals in roots and flowers, the transfer coefficients calculated for the roots suggest that the accumulation of metals is insignificant. This capacity could indicate stabilization of contaminated soil, restricting the bioavailability of metals and translocation into air tissues.
- There was a significant development of the plants tested on the soil enriched with metals compared to the control ones. The presence of other metals in the polluted soil had a stimulating effect on the growth of plants, quantified by the amount of plant biomass, by the fact that the polluted plants have larger leaves and dark green color compared to the basil leaves from the control samples that are colored in pale green. It is assumed that this behavior of the tested plants is the result of a high incidence of metal competition in the absorption processes and also of the mechanisms of regulation of the metal.

- The values obtained for the bioaccumulation factor, the translocation factor, the enrichment factor and the geo-accumulation indices showed that the basil plants, under metallic stress conditions had a great phytostabilization potential..

In the experimental study conducted in the polluted mining area in Certej, Deva area, 10 sediment samples were analyzed in correlation with 10 surface water samples, 22 soil samples and 40 vegetation samples in correlation with soil samples. The obtained values were compared with the maximum limits allowed by the legislation in force for the quality of surface water, sediments and soil.

In the soil environment component, of the 10 analyzed profiles, the highest concentrations of metals were found in samples S3 (sample taken in the immediate vicinity of the Coranda Quarry) and S10 (collection point located on the Băiaga river, downstream of water confluence career and upstream of the confluence with the Ciongani stream). In the case of the S3 profile, a significant pollution induced by the metals Ni, Pb, As and Hg is observed, whose concentrations exceed the intervention threshold for soils with sensitive uses. On both levels of sampling of the S3 profile, it is observed that the quality indicators Cd and Mn exceed the alert threshold for soils with sensitive uses inducing a potentially significant pollution.

The most polluted profile, in which the pollution is reflected on both levels of sampling (0-5 cm and 30-40 cm), is the S10 profile. Most of the metals analyzed in this profile (Cd, Co, Cu, Mn, Pb, Zn, As and Hg) induce a significant pollution, because they are in quantities that exceed the intervention threshold for soils with sensitive uses. The high concentrations for most of the indicators analyzed in this profile are due to the cumulative effect generated by the existence of the Coranda quarry, the tailings dumps and the Nicodim gallery, through the contribution of surface waters and percolation waters loaded with metals infiltrating the soil.

Studies have been carried out regarding the distribution of metals in the sediment and surface water environmental compartments. Thus, the concentrations of metals in sediments (total content and mobile fraction) were correlated with the values determined in the surface water. In this respect, the mobile fraction content was evaluated by applying at the first extraction the sequential extraction method of the Community Bureau of Reference Materials (BCR), determining the bioavailability index.

Another correlation was related to the distribution of metals in plants, referring to the contents of total and mobile metals in the soils bordering the sampling area. Thus, to determine the optimal method for determining the mobile metal fraction, fraction used to evaluate the bioavailability of metals, three simple chemical extraction methods were tested, the best results being obtained by using as an extraction agent the 1M ammonium acetate mixture. and 0.05M EDTA, at pH 7.

For the characterization of the sediment quality, several pollution indices were estimated, indices calculated for the elements As, Cd, Cr, Cu, Hg, Ni, Pb (contamination factor, ecological risk factor, geo-accumulation index). These indices are used to evaluate the ecological risk of the sediment as a whole, by estimating pollution coefficients (CH), ecological risk factors (EF) and pollution load indices (PLI).

The conclusions of the correlations made for the surface water - sediment system are as follows:

- In the aquatic component, the bioavailability index correlates very well with the values obtained in surface water; thus, in the case of extremely high concentrations of metals in the sediment, but with low or zero mobility, in the surface water there is a low content of metals (As, Ti);
- In the case of the metallic elements Cd, Ni and Cu, the surface water concentrations are totally classified in the 5th grade of quality, due either to the high bioavailability index or to the very high total metal content, combined with a higher bioavailability index. low;

- The elements Hg, Mo and W detected in the sediment are not found in the surface water, the bioavailability indices being zero;
- Ag and Te metals were not detected in any of the analyzed samples (water, sediment);

- Another category of metals (Co, Cr, V, Sb) has either a relatively high content of metals correlated with reduced mobility or a low content relative to the maximum permissible value (VMA) in the sediment, but correlated with a high mobility, which places surface waters in quality classes 1 ÷ 4;

Unlike the geo-accumulation index, which only takes into account the total content and VMA in the sediment, the bioavailability index is correlated with the ability of the metal to pass from the sediment into surface water and implicitly in vegetation. In view of the results obtained, the bioavailability index is more suitable for characterizing the surface water - sediment interaction in the studied system (zone with acid pH, low flow rates, variable according to precipitation, very high content of toxic metals).

The pollution coefficients, calculated for As, Cd, Cr, Cu, Hg, Ni and Pb depending on the total concentration and the maximum allowed value in the sediment indicate several risk categories for sediment samples (Sd), as follows:

- Sd2 - low ecological risk;
- Sd1, Sd5, Sd8, Sd10 - moderate ecological risk
- Sd4, Sd6, Sd7, Sd9 - strong ecological risk;
- Sd3 - very strong ecological risk.

The ecological risk indices were calculated, based on the total concentration, the maximum allowed concentration and a toxic response factor, which decreases in the order: Hg> Cd> As> Ni> Cu> Pb> Cr. The obtained values allowed the classification of sediments, as follows:

- Sd2 - strong ecological risk;
- Sd1, Sd3 ÷ Sd10 - very strong ecological risk.

Pollution load indices (PLI) were determined, based on the total sediment concentration and the maximum permissible value. Their values were above the value of 1, being in the range 1.3 ÷ 9.5, which indicates a deterioration of the sediment quality.

The plants analyzed in the thesis are mostly plant leaves, tree leaves, grass, and in a smaller proportion tree bark. Harvesting was done in early spring, when the vegetation was in the bud period. Each soil sample was collected from 2 depths, in the interpretation of the results taking into account the values of the metal concentrations obtained in both samples. The research carried out allowed the following conclusions to be drawn for the correlations between the vegetation-soil system:

- In proportion of 90% the vegetation samples presented Ca and Mg over the normal concentration ranges both in the samples related to the polluted area and in the control sample collected from 5 km on the SV direction of the area;
- For the most part, the soils showed acid pH, which was reflected in the bioavailability index, which recorded high percentages for several metals (Cd, Cu, Ni, Pb and Zn); thus, concentrations were recorded above the normal limits of Cd, Cu, Ni, Pb and Zn in various types of plants analyzed: maple, hornbeam, nettle, tree bark, grass, forest leaf, clover and sage, thyme, etc.
- In some plants and concentrations above the normal limits of As, Se, Mn, Fe were determined.

In order to characterize a contaminated area and establish its degree of pollution, it is necessary to correlate the results obtained on several environmental components. Thus, the bioavailability studies of metals in solid environmental samples provided additional information compared to studies based only on total metal content, which proved useful in reaching the correct conclusions regarding the contamination of the contaminated areas.

The pollution indices calculated for the soil samples mCd, EF, Igeo that were evaluated taking into account the total metal content provided partial information on the environmental pollution potential, and a bioavailability study of the respective metal in the soil is required. Thus, three simple methods of chemical extraction were tested, selecting as the method of assessing mobility, the method using EDTA and CH3COONH4. The study highlighted the transfer of metals from soil to vegetation, the metals being found in all types of vegetation analyzed (root, leaf, bark, stem).

The methods for the quantitative determination of arsenic, selenium, stibium and tin from water samples were optimized using inductively coupled plasma hydride generation optical emission spectrometry. The optimized methods are specific, selective, linear, precise, accurate, with much lower detection limits and quantification limits compared to determining these metals directly in the plasma. The disadvantage of these methods would be the high consumption of reagents and consequently the higher price of one analysis compared to the other analyzes of metals determined directly in the plasma, without the generation of hydrides.

BIBLIOGRAFIE SELECTIVĂ

1.L.C. Soare, E. Visioiu, C. Bejan, C.M. Dobrescu, I. Fierascu, I. Iosub, A. Paunescu "Research on the in vitro bioaccumulation capacity of lead in some pteridophyte species of the Romanian flora", Revista de Chimie (Bucharest), vol.66, no.12, 2015, p.2017-2020.

2.R. Chandra, S. Yadav, " Phytoextraction potential of heavy metals by native wetland plants growing on chloroling containing sludge of pulp and paper industry", Ecological Engineering, vol.98, 2017, p.134-135.

3.G.G. Vasile, L. Kim, S. Gheorghe, B.Stanescu, "Ecological Assessment Of Mobile Cadmium In Sediments From Certej Mining Site, Hunedoara County, Romania" SGEM Conference Proceedings, 2013, vol. 1, p.299 – 306.

4.J. Pérez-Esteban, et al., "Bioavailability and extraction of heavy metals from contaminated soil by Atriplex halimus". Environmental and Experimental Botany, vol. 88, 2013, p. 53-59.

5.G. Bonanno, J.A. Borg, V. Di Martino, "Levels of heavy metals in wetland and marine vascular plants and their biomonitoring potential: A comparative assessment", Science of the Total Environment 576, 2017, p.796–806.

101. A.E. Fersman, Geochemistry, Academy of Science of Soviet Union Publishing House, Moscow, vol. 1-4, p.1955-1959.

102.A.P. Vinogradov, „Regularities of the distribution of chemical elements in the Earth's crust", Geokhimiya, 1, 1962, p.6-52.

103.N.N. Greenwood, A. Earnshaw, Chemistry of the Environment, Pergamon, Oxford, 1989, p.1242-1363.

104.„Abundence of Elements in the Earth's Crust and in the Sea" in D.R. Lide, ed., 2005, CRC Handbook of Chemistry and Physics, Section 14: Geophysics, Astromony and

Acoustics, pag. 2373, Internet Version 2005, <http://www.hbcpnetbase.com>, CRC Press, Boca Raton, FL.

105.K. Ravindra, L. Bencs, R. Van Grieken, „Platinum group elements in the elements and their health risk", Science of the Total Environment, 318, 2004, p.1-43.

106.D.V. Ladonin, „Platinum-Group Elements in Soils and Street Dust of the Southeastern Administrative District of Moscow", Eurasian Soil Science, 51, 3, 2018, p. 268-276.

107.M.E. Farago, E.J. Hutchinson, N. Chandran, P.R. Simpson, „Increases in platinum metals in the UK and possible health effects, in Metal Ions in Biology and Medicine", John Libbey Eurotext, Paris, vol. 6. 2000.

108.N. Teutsch, Y. Harlavan, L. Halicz, „Level and distribution of Platinum Group Metals (PGM) in Israel, Ministry of National infrastructures", Geological Survey of Israel, report GSI/23/2013.

109.B. Gomez, M. Gomez, J. Sanchez, R. Fernandez, M.A. Palacois, „Platinum and rhodium determination in airborne particulate matter and road dust", Science of the Total Environment, 2001, p.131-144.

110.C.L.S. Wiseman, J. Niu, C. Levesque, M. Chenier, „An assessment of the inhalation bioaccessibility of platinum group elements in road dust using a simulated lung fluid", Environmental Pollution, 241, 2018, p.1009-1017.

140.C. L. Boechat, F.S. Carlos, C. Gianello de Oliveira, F.A. Camargo, „Heavy metals and nutrients uptake by medicinal plants cultivated on multi-metal contaminated soil samples from an abandoned gold ore processing site", Water Air Soil Pollut, 2016, p.227:392.

141.D. Adamczyk-Szabela, Z. Romanowska-Duda, K. Lisowska, W.M. Wolf, „ Heavy metal uptake by herbs. V. Metal accumulation and physiological effects induced by thiuram in Ocimum basilicum L", Water Air Soil Poll., 2017, p. 228:334.

147.J.O. Olowoyo, E. van Heerden, J.L. Fischer, C. Baker, „Trace metals in soil and leaves of Jacaranda mimosifolia in Tshwane area, South Africa", Atmos Environ 44, 2010, p.1826–1830.

148.G.C. Kisku, S.C. Barman, S.K. Bhargava, „Contamination of soil and plants with potentially toxic elements irrigated with mixed industrial effluent and its impact on the environment", Water Air Soil Pollut 120, 2000, p.121–137.

186.D.R. LIDE, CRC Handbook of Chemistry and Physics, „Abundance of elements in the earth's crust and in the sea".

190.C.Dinu, E.M.Ungureanu, G.G.Vasile, L.Kim, I.Ionescu, C.Ene, M.Simion, "Soil and vegetation from an abandoned mining area situated in Hunedoara county, Romania", Revista de Chimie(Bucharest), 69, No.1, 2018.

193.D. B. Hibbert, "Quality Assurance for the Analytical Chemistry Laboratory", Oxford, University Press, 2007.

194.I. Taverniers, M. de Loose, E. V., Bockstaele, Trends in Analytical Chemistry, 23 (8), 535, 2004.

195.I. Gh Tanase, G.L Radu, A.Pana., M..Buleandra, "Analytical method validation", Printech Press, Bucharest, 2007.

198.C. Dinu, G. Vasile, L. Cruceru, J. Petre, "Flow injection hydride analysis using ICP-EOS technique", International Symposium „Environmental and Industry", 28-30 0ctober 2009, Bucuresti.

A.1. ARTICLES PUBLISHED ON THE THESIS

1.**C.Dinu,** G. Vasile, L. Cruceru, *Advanced analytical methods for mercury determination in slightly contamined water samples,* Journal of Environmental Protection and Ecology, 2013, 14 (4), p.1515-1524; FI = 0.338; SRI=0.016

2.L.R. Mandoc (Popescu), **C.Dinu**, M. Iordache, G.-O. Buica, E.-M. Ungureanu, L. F. Pascu *Application of flow injection hydride generation coupled with ICP-EOS for mercury detection in waters*, U.P.B. Sci. Bull., Series B, ISSN 1454-2331, Vol. 77, Iss. 3, 2015,p. 33 – 38

3.L. Kim, G.G. Vasile, B. Stanescu, **C.Dinu**, C.Ene, *Distribution of trace metals in surface waters and streambed sediments in the vicinity of an abandoned gold mine from Hunedoara county, Romania*, Revista de Chimie, vol.67, nr.8, 2016, p.1441-1446. FI = 1.232; SRI=0.057

4.**C.Dinu**, G.-G. Vasile, E.-M. Ungureanu, L. Kim, L.V. Cruceru, *Metal Accumulation in Salvia Officinalis Grown in Contaminated Soil from a Waste Mining Dump*, Ecology and Environmental Pollution, 17th International Multidisciplinary Scientific GeoConference SGEM 2017, conference proceedings contents isi, section ecology and environmental protection, 29 June - 5 July, 2017, Albena, Bulgaria, DOI: 10.5593/sgem2017/51, ISSN 1314-2704, ISBN 978-619-7408-08-9, Volume 17, ISSUE 51, 643), p.

5.**C.Dinu**, E.M.Ungureanu, G.G.Vasile, L.Kim, I.Ionescu, C.Ene, M.Simion, *Soil and vegetation from an abandoned mining area situated in Hunedoara county*, Romania, Revista de Chimie(Bucharest), 69, No.1, 2018, p.14-20. FI = 1.605; SRI=0.135

6.**C.Dinu**, E.M.Ungureanu, G.G.Vasile, L.Kim, L.F. Pascu, M.Simion, *Evaluation of the bioavailability and pollution indexes of some toxic metals in polluted soils from an abandoned mining area*, Revista de Chimie(Bucharest), 69, No.11, 2018, p.4141-4147. FI = 1.605; SRI=0.135

7.A.Tenea, **C.Dinu**[*], E.M.Ungureanu, G.G.Vasile, M.Simion, *Determination of Rhodium from wastewater sample using ICP-OES technique*, International Symposium "The Environment and the Industry" SIMI 2018, p.366-372

8.G.G.Vasile, **C.Dinu*,** L.Kim, A.Tenea, M.Simion, C.Ene, C.Spanu, E.M.Ungureanu, D.Manolache, *Platinum group elements in road dust and vegetation from some European and National Roads with intensive car traffic in Romania*, Romania, Revista de Chimie, vol 70, nr. 1., 2019, p.286-292. FI = 1.605; SRI=0.135

9. C.Dinu, G.G.Vasile, M.Buleandra, D.E.Popa, St.Gheorghe, E.M. Ungureanu, *Translocation and accumulation of heavy metals in Ocimum basilicum plants grown in a mining contaminated soil* , Journal of Soils and Sediments,-in press FI=2.669, SRI=1.101

Total FI 9,054 Total SRI 1.579

A.2. SCIENTIFIC COMMUNICATIONS ON THE DOCTORAL THESIS

1. L.-R. Popescu (Mandoc), E.-M. Ungureanu, M.Iordache, G.-O. Buica, G.-A. Inel, P. Pavel, **C. Dinu**, *Assesment of mercury polution on ecosystems water/sediment from Olt river basin and soil near industrial sources of pollution*, Conference on Chemistry and Chemical Engineering, RICCCE 18, 4-7 september 2013, Sinaia, Romania, oral presentation, S5-33.

2. **C. Dinu**, G.-G. Vasile, E.-M. Ungureanu, L. Kim, L. V. Cruceru, *Metal Accumulation In Salvia Officinalis Grown In Contaminated Soil From A Waste Mining Dump*, 17th International Multidisciplinary Scientific GeoConference *SGEM 2017*, 29 June - 5 July, 2017, Albena, Bulgaria, poster.

3. **C.Dinu,** I.Chilibon, C.Vasiliu, R.Stefan, R.Isopescu, E.-M. Ungureanu, *Characterization of polyazulene complexing films*, Workshop in the 21st Romanian International Conference on Chemistry and Chemical Engineering Constanta- Mamaia, ROMANIA - September 4 – 7, 2019 (Atelier scientifique *Nouveaux Matériaux pour la Reconnaissance Electrochimique des Minéraux et des Espèces Biologiques (NOMARES)*, poster.
4. A.Tenea, **C.Dinu**[*], E.M.Ungureanu, G.G.Vasile, M.Simion, *Determination of Rhodium from wastewater sample using ICP-OES technique*, International Symposium "The Environment and the Industry" SIMI 2018, 21st Bucharest, September 2018 poster.
5. **C. Dinu**, E.-M. Ungureanu, L.Kim, L.F.Pascu, M.Simion, *Pollution Indexes for soil quality assessment*, International Symposium "The Environment and the Industry" SIMI 2018, 21st Bucharest, September 2018 poster.