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COMPARATIVE STUDY OF DIFFERENT EXTRACTION METHODS FOR EVALUATION OF BIOAVAILABILITY OF TERRESTRIAL SEDIMENT – BOUND METALLIC ELEMENTS

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The objective of this paper is to compare different extraction methods for evaluation of bioavailability of some metallic elements, such as: Cd, Cu, Ni, Pb and Zn from the lake sediments collected from Roşia Montana aria and also from Săsar river and Racoş stream, situated in Baia Mare aria. The paper contains also information about mobility tests applied to a BCR Certified Reference Material 701, lake sediment. The experimental data proved that NFX31 – 120/1-92 (EDTA and NH_4OAc solution) can be successfully used for estimate the release of metallic elements from terrestrial sediments polluted by mining activities.

INTRODUCTION

Persistent and potentially harmful products have been widely dispersed and concentrated in different spheres of the environment (water, air, soil, sediment, living organism) as the result of human activities. Among the most dangerous of these are toxic metals. Such materials have been accumulated in materials dredged from waterways, harbors, in industrial sites, in landfills, in lake sediment, stream beds and deltas deposited.[1] Some organisms have accumulated high enough level of metals to do harm to themselves or to human that use them as a food source. [1]

Metals are known to concentrate in sediments of natural aquatic systems and this accumulation is postulated to be caused by different mechanisms (ion-exchange, outer and inner-sphere surface complexation, precipitation or co-precipitation) with various geochemical phases such as: organic matter, oxy-hydroxides of iron, aluminum and manganese, phyllosilicate minerals, carbonates and sulfides. [2]

Only a fraction of the metal present in sediment is mobile or bio-available, therefore the determination and monitoring of specific chemical forms of metals in environmental samples are extremely important in order to assess the toxic effects in aquatic systems and geochemical pathways. Among the factors involved in metal availability are the identity of the metal, its chemical form (type of binding, oxidation state), the nature of the suspended material, the type of organism taking the metal and the physical and chemical condition in the water (*e.g.* pH, pE).[1]

The increasing concern to assess the bio-available metal fraction (and thus to estimate the related phyto-toxic effects) and the mobility of trace metals upon disposal of sediment is reflected by a considerable increase in the frequency of analysis over the last 10 years.[3-8] Single and sequential extraction schemes have been designed in order to assess the different retention / release of metals in soil and sediment samples.[9-15]

The objective of this paper is to compare different extraction methods for evaluation of bioavailability of some metallic elements, such as: Cd, Cu, Ni, Pb and Zn from the lake sediments collected from Roşia Montana aria and also from Săsar river and Racoş stream, situated in Baia Mare aria. The sediment samples were collected from two sites, both polluted by mining activities. Also, the paper contains data about mobility tests applied to BCR Certified Reference Material 701, lake sediment.

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The extraction methods tested were two standard methods (SR ISO 14870/99[16], NF X 31 – 120/92[17]), used in environmental study for evaluation and monitoring the mobile metallic species from soil, sediment and sludge [12, 18, 19] and a BCR method for mobile metallic elements extracted from lake sediment, proposed by Standard Measurement and Testing Program of the European Community.

The standard methods use buffer solution with neutral pH and extract the mobile metallic elements from the matrix. In 0.11 M acetic acid solution used by the BCR method for lake sediment are released exchangeable metallic elements, water-soluble and weak acid soluble fraction (carbonates).

Assuming that bioavailability is related to solubility, then metal bioavailability decreases in the order: water-soluble > exchangeable > carbonates.

EXPERIMENTAL

The sediment samples were collected from 8 artificial lakes situated in Roşia Montana aria, in the proximal vicinity of a gold and silver mining site. The samples were taken from 1.5 to 10.5 m within the lakes using a Van Veen Bottom Sampler and were stored in plastic recipients. All samples were kept in cooling boxes at 4^0 C during transportation and the analyses were performed immediately after receiving the samples in the laboratory.

From another site polluted by mining activities, were collected 2 sediment samples, one from Racoş stream, situated in the neighbor of a mining sterile dump and the other one from Săsar river, also situated in the aria polluted with metallic elements. In table 1 there are present the symbol of the samples and location for each of them.

Dry sediment (at room temperature) with particle dimension lower than 60 µm was digested with a mixture of aqua regia[18] in a Microwave Laboratory System Ethos Millestone.

The total and fractionated concentration of metals (Cd, Cu, Cr, Pb, Ni, Zn) was determinated by an UNICAM SOLAAR 929 atomic absorption spectrometer. Air/acetylene type flame was used for all the elements, with exception of Cr. For this element was used a protoxide/acetylene flame. All the chemicals used were of analytical reagent grade (Merck quality).

The working procedure for mobile forms of metallic elements was as follows: for methods no. 1 and 2 (table 2): 4 g of dry sediment (particle size < 60 µm) were mixed with 40 mL of buffer solution. The obtained suspension was shaken at 20ºC for two hours with 40 rpm. In the BCR method, 1 g of dry sediment was mixed with 40 mL of 0.11 M CH_3COOH for 16 hours at 20º C and 30 rpm. The sediment was in complete suspension during the extraction procedure. The solution was centrifuged and then analyzed with AAS methods. For each experiment of extraction a blank sample (i.e. a container with no sediment) was prepared through the entire procedure and were used external standard, prepared from reference standard material - Merck quality in the corresponding extraction solution, for calibration curve.

Table 1

The location of the collected sediment samples

Symbol	Description	Location
L 1	Sediment collected from Mare Lake, depth 10.5 m	Roşia Montana
L 2	Sediment collected from Ţapului Lake, depth 2 m	Roşia Montana
L 3	Sediment collected from Corna Lake, depth 3.2 m	Roşia Montana
L 4	Sediment collected from Cartuş Lake, depth 2.1 m	Roşia Montana
L 5	Sediment collected from Brazilor Lake, depth 5.8 m	Roşia Montana
L 6	Sediment collected from Ţarina Lake, depth 3.5 m	Roşia Montana
L 7	Sediment collected from Anghel Lake, depth 6 m	Roşia Montana
L 8	Sediment collected from Găuri Lake, depth 1.5 m	Roşia Montana
R 1	Sediment collected from Săsar River, in the vicinity of a mining site	Baia Mare
R 2	Sediment collected from Racoş Stream, in the vicinity of a mining sterile dump	Baia Mare

Table 2

The extraction methods used for mobile metallic elements

No.	Method	Solution of extraction	Condition
1	SR ISO 14870/99	0.005 M DTPA, 0.1 M TEA, 0.01 M $CaCl_2$, pH = 7.3	Report sediment : solution = 1:10, 2 hour shake at 40 rpm
2	NF X 31 – 120/1-92	0.01 M EDTA, 1 M NH_4OAc, pH = 7.2	Report sediment : solution = 1:10, 2 hour shake at 40 rpm
3	BCR method for lake sediment	0.11 M CH_3COOH, pH = 2.8	Report sediment : solution = 1:40, 16 hour shake at 30 rpm

The sediment dry weight was determined on a separate sub-sample, approximately 1 g, by drying at 105º ± 2º C until constant weight. For each analytical results were applied a correction to "dry matter" (d.m., quantity of metal per g dry sediment).

For the availability comparison tests, each method was applied to the sediment samples (L1 ÷ L8 -lake sediments, R 1 and R 2 – river sediment, BCR CRM 701) using single chemical extraction procedure.

In the study was applied a sequential extraction procedure to BCR CRM 701, lake sediment. In table 3 are present the steps and solutions used in chemical sequential extraction procedure.

Table 3

The steps of the sequential extraction procedure

Step	Solution of extraction	Condition
first	0.005 M DTPA, 0.1 M TEA, 0.01 M $CaCl_2$, pH = 7.3 **or** 0.01 M EDTA, 1 M NH_4OAc, pH = 7	Report sediment : solution = 1:10, 2 hour shake at 40 rpm
second	Distillated water, pH~5	Report sediment : solution = 1:10, 2 hour shake at 40 rpm
third	0.11 M CH_3COOH, pH = 2.8	Report sediment : solution = 1:40, 2 hour shake at 40 rpm

Table 4

The "total concentration" of metallic elements from L1 to L 5 - lake sediments

Element	M.U.	L 1	L 2	L 3	L 4	L 5	Reference values according to Roumanian Order 161/2006
Cd	mg/Kgd.m.	< 0,5	< 0,5	< 0,5	< 0,5	< 0,5	0.8
Cu	mg/Kg d.m	**244±5.2**	**42.1±0.9**	**63.8±1.4**	**131±2.8**	**181±3.9**	40
Cr total	mg/Kg d.m	33.2±1.3	33.2±1.3	15.9±0.6	11.1±0.4	10.8±0.4	100
Ni	mg/Kg d.m	35±1.1	15±0.5	12.1±0.4	**37±1.2**	30.3±0.9	35
Pb	mg/Kg d.m	47.6±2.3	23.4±1.1	25.2±1.2	29.2±1.4	41.2±2.0	85
Zn	mg/Kg d.m	**154±7**	92.5±4.3	106±4.9	**423±20**	**453±21**	150

In order to estimate the values of uncertainty for each concentration of mobile metallic elements, an uncertainty budget was developed. The different uncertainty sources and their influence on the expanded uncertainty are present in the following formula (GUM method):

$$U_{expanded} = k * u_{cs} = k * \sqrt{}\ (u_c^2 + u_v^2 + u_{rep}^2 + 1/u_{mass}^2 + 1/u_{rec}^2)$$

where:
k = coverage factor, value 2 for 95% level of confidence;
u_{cs} = combined standard uncertainty;
u_c =concentration uncertainty (instrument calibration, flasks, pipettes, reference standard material);
u_v = 50 mL volumetric flask (calibration, temperature);
u_{rep} = repeatability uncertainty (mass, volume, absorbance, extraction recovery);
u_{mass} = weight uncertainty (balance calibration, linearity);
u_{rec} = extraction recovery uncertainty.

RESULTS

The "total concentrations" of metallic elements dissolved in aqua regia (mixture of nitric acid and hydrochloric acid in the report 1:3) were compared with the reference values for the sediment quality according to the Roumanian Order 161/2006[19] (tables 4 and 5). For the lake sediments, only Cu, Zn and Ni (one sample) contents were higher than the reference value. In the river sediments were recorded very high concentration of Cd (in Săsar river), Cu, Pb and Zn. These data collected from Baia Mare aria conduct to conclusion that mining activities has a harmful action to the surrounding environment.

In the figures 1–10 are present the comparative concentration of mobile metallic elements, extracted from the sediments with three different single chemical extraction procedures (see table 2).

In all test experiments, Cr metallic element was not founded in mobile state inside the sediment structure, either lake or river sediment. Ni element was founded in mobile fraction only in four lake sediments, there are: L2, L4, L5 and L8.

In the figures 11-12 there are data about the sequential extraction procedure (table 3) versus single chemical extraction with the BCR method applied to BCR CRM 701 sediment.

In order to compare the values obtained with three single chemical extraction applied to BCR CRM 701- lake sediment with the certified values, in table 6 are present the comparative results.

Table 5

The “total concentration” of metallic elements from L6 to L 8 - lake sediments, R1 and R2 – river sediments

Element	**M.U.**	**L 6**	**L 7**	**L 8**	**R 1**	**R 2**	Reference values according to Roumanian Order 161/2006
Cd	mg/Kgd.m.	< 0,5	< 0,5	< 0,5	**10.8**	**1.2**	0.8
Cu	mg/Kg d.m	**61.4±1.3**	**69.1±1.5**	**48.2±1**	**432±9**	**391±8**	40
Cr total	mg/Kg d.m	25.7±1	13.5±0.5	13.9±0.5	19.1±0.7	28.6±1.1	100
Ni	mg/Kg d.m	18.7±0.6	6.2±0.2	19.8±0.6	< 2	< 2	35
Pb	mg/Kg d.m	35.1±1.7	50.7±2.5	47±2.3	**708±35**	**643±31**	85
Zn	mg/Kg d.m	131±6.5	120±5.6	**217±10**	**1972±92**	**607±28**	150

Table 6

Comparative results from BCR CRM 701 – lake sediment

Element	**M.U.**	**Experimental data**			**Certified value BCR method**
		SR ISO 14870/99	NFX 31-120/92	BCR method	
Cd	mg/Kg d.m.	4.85 ± 0.24	5.16 ± 0.25	6.88 ± 0.34	7.34 ± 0.35
Cu	mg/Kg d.m	100 ± 3.1	145 ± 4.5	43.4 ± 1.5	49.3 ± 1.7
Ni	mg/Kg d.m	6.96 ± 0.30	9.47 ± 0.40	13.7 ± 0.6	15.4 ± 0.9
Pb	mg/Kg d.m	35.7 ± 2.0	55 ± 3	3.46 ± 0.19	3.18 ± 0.21
Zn	mg/Kg d.m	84 ± 4.5	171 ± 9	196 ± 10	205 ± 6

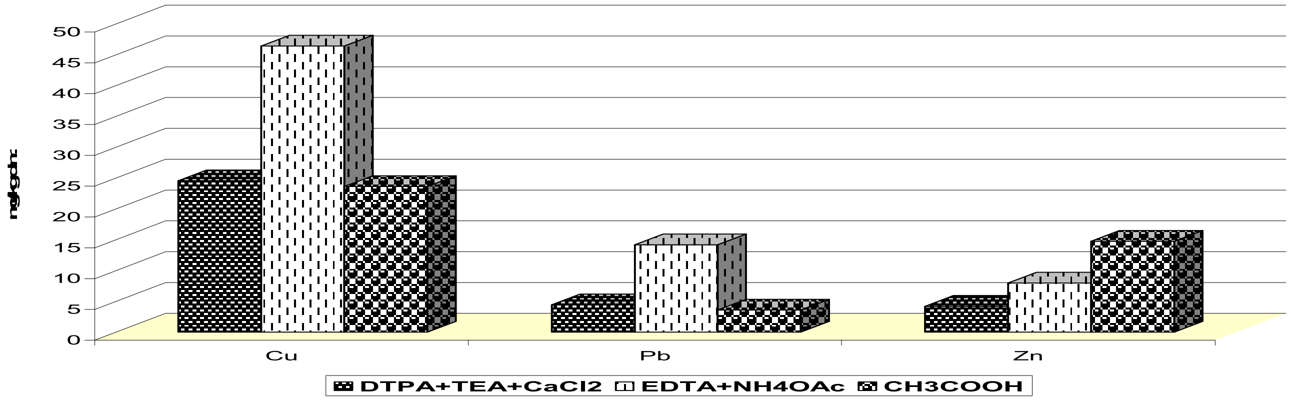

Fig. 1 – The mobile metallic elements from L1 sediment.

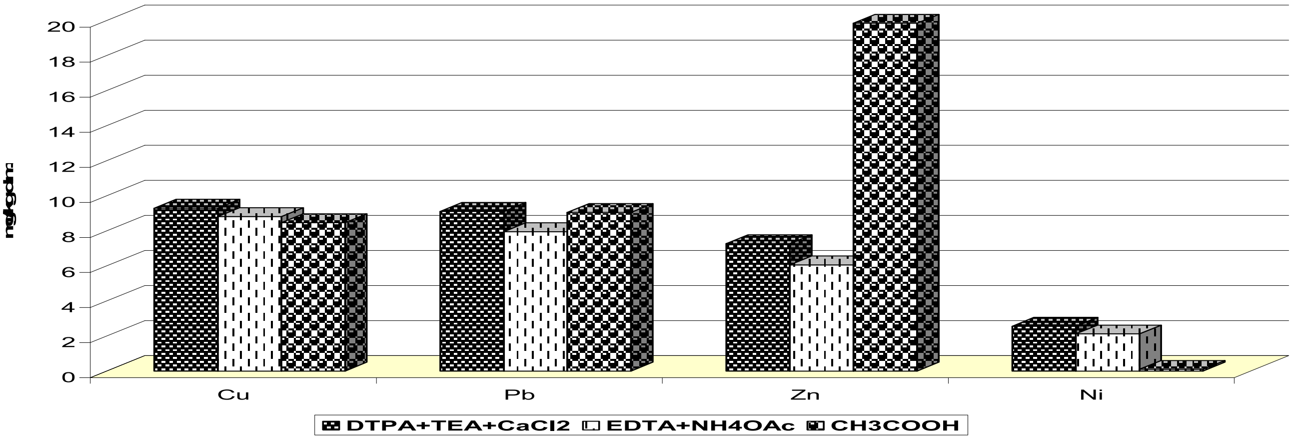

Fig. 2 – The mobile metallic elements from L2 sediment.

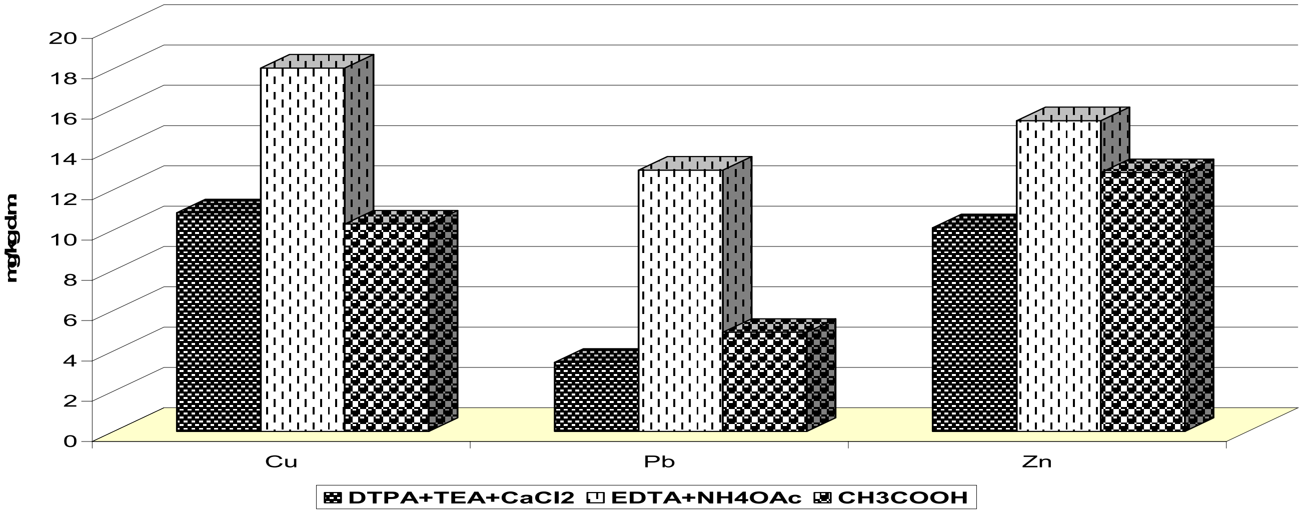

Fig. 3 – The mobile metallic elements from L3 sediment.

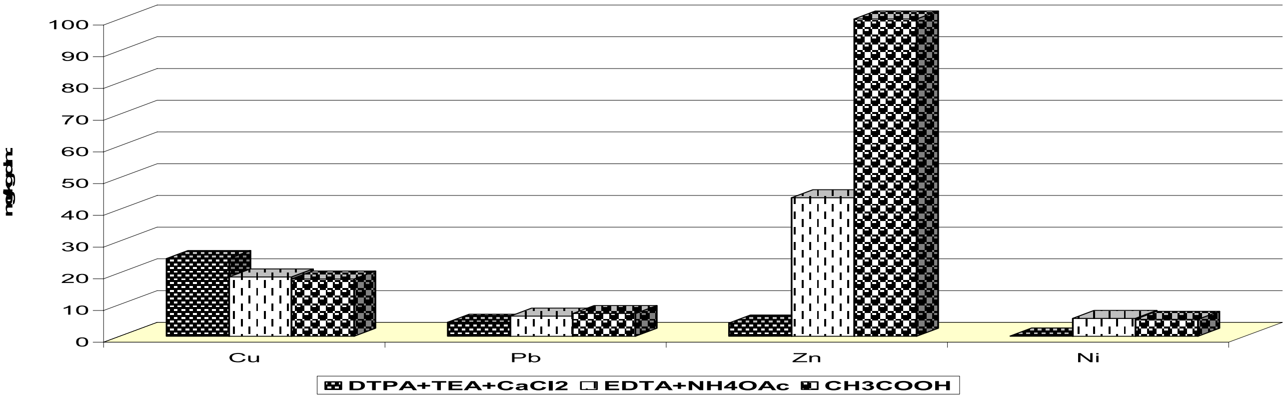

Fig. 4 – The mobile metallic elements from L4 sediment.

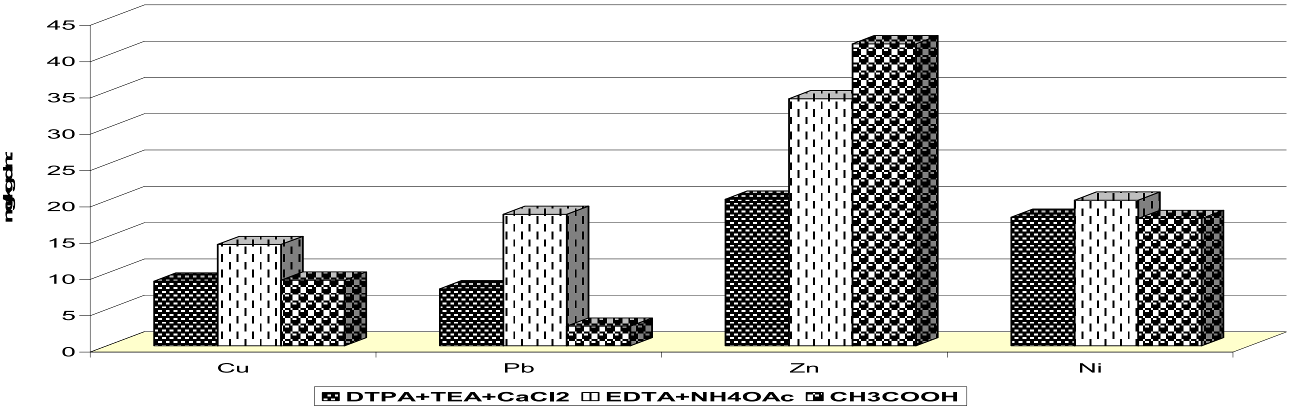

Fig. 5 – The mobile metallic elements from L5 sediment.

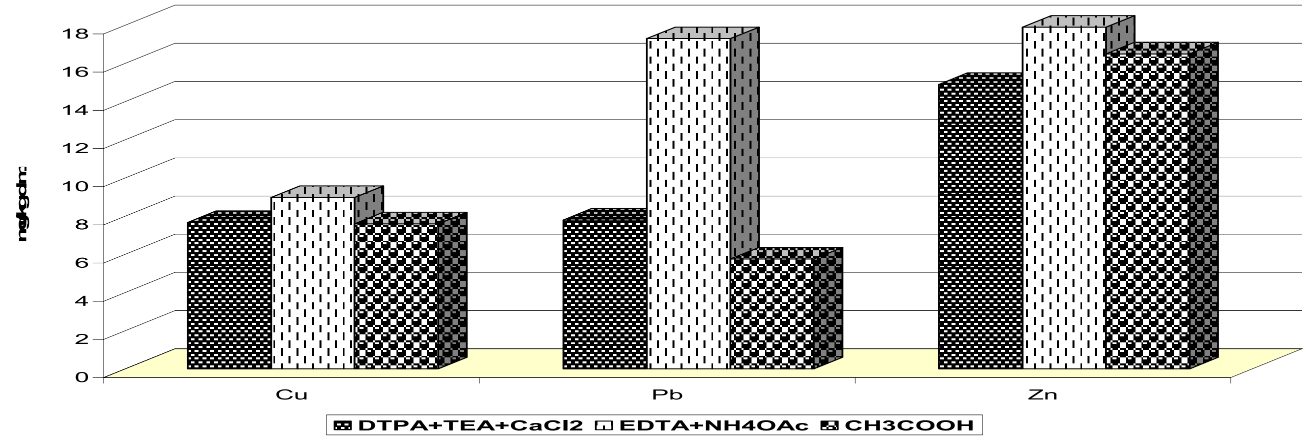

Fig. 6 – The mobile metallic elements from L6 sediment.

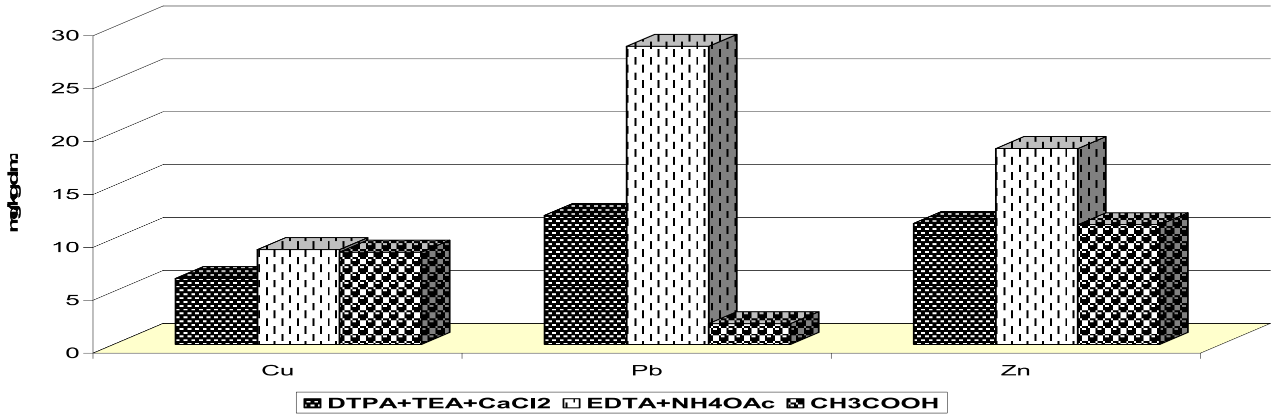

Fig. 7 – The mobile metallic elements from L7 sediment.

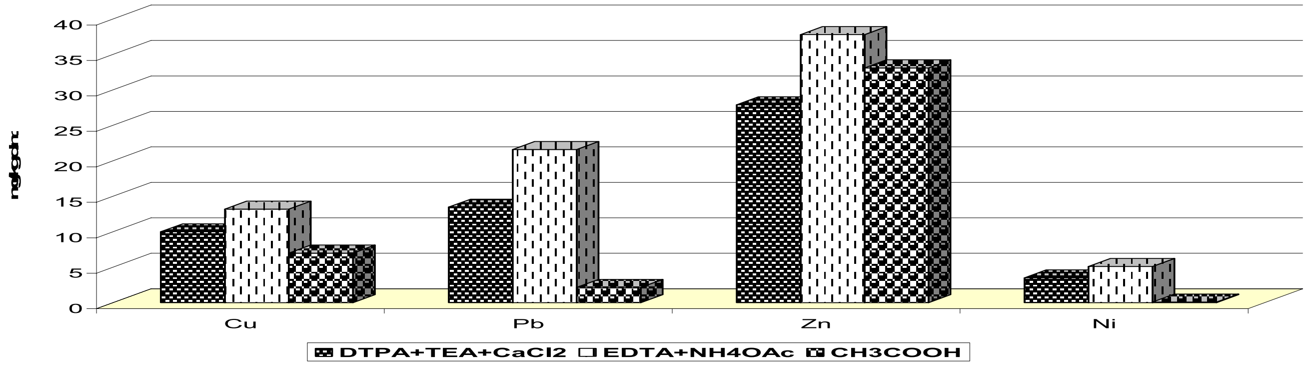

Fig. 8 – The mobile metallic elements from L8 sediment.

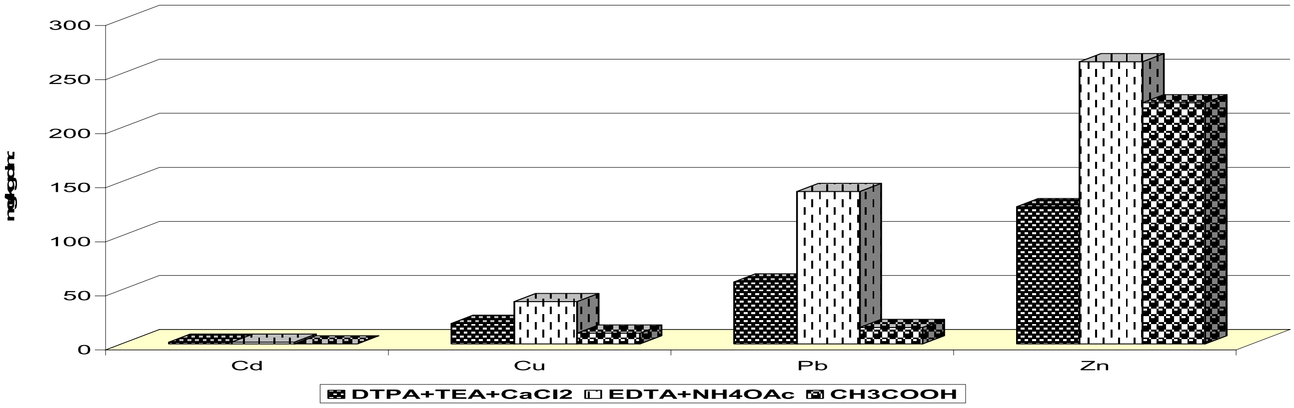

Fig. 9 – The mobile metallic elements from R1 sediment.

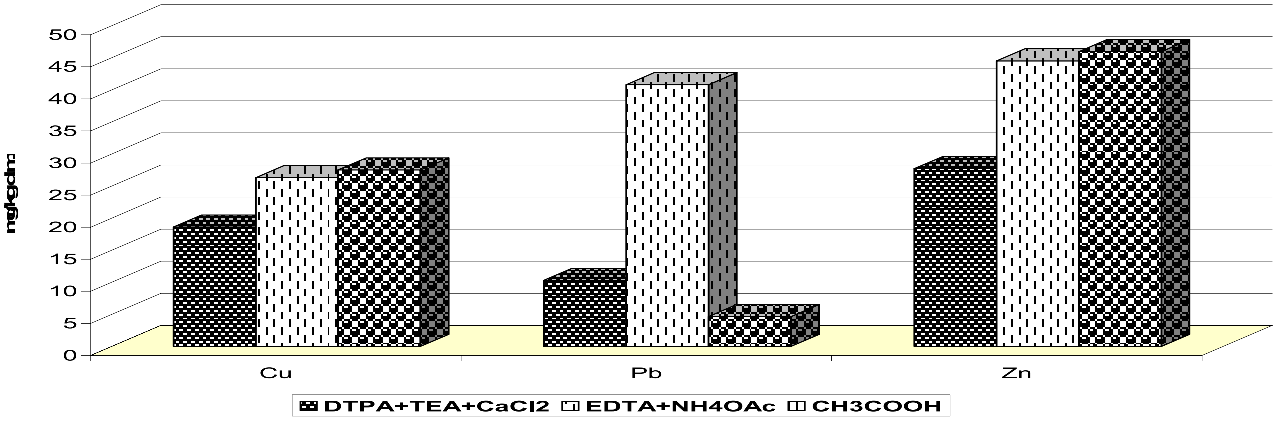

Fig. 10 – The mobile metallic elements from R2 sediment.

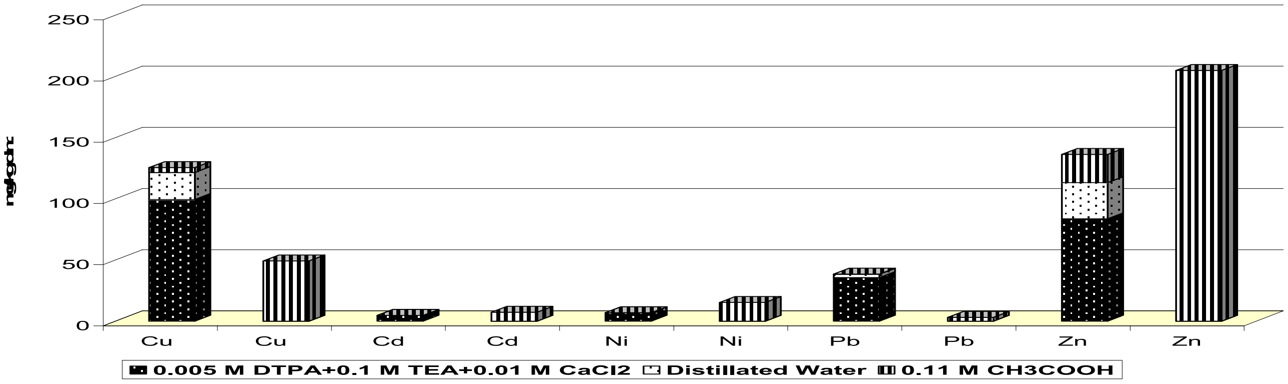

Fig. 11 – Comparative concentrations for sequential extraction (DTPA solution, distilled water and CH3COOH) and single chemical extraction with 0.11 M CH3CCOH from CRM BCR 701, lake sediment.

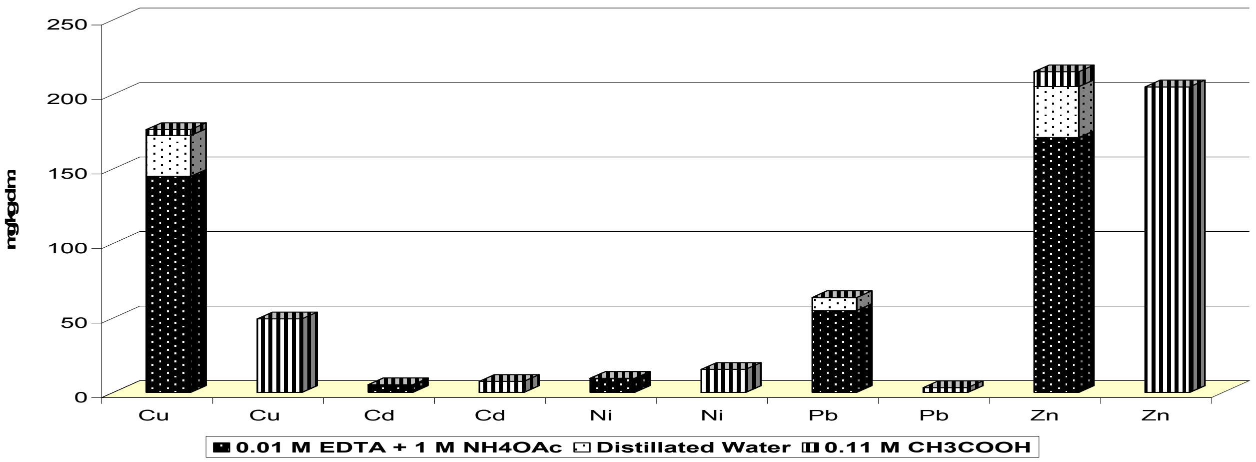

Fig. 12 – Comparative concentrations for sequential extraction (EDTA solution, distillated water and CH3COOH) and single chemical extraction with 0.11 M CH3CCOH from CRM BCR 701, lake sediment.

DISCUSSION

The aim of this study was to investigate which method is appropriate for these types of sediments and to observe the differences between the obtained results with three single chemical extraction procedures. The experimental data (figures 1 – 10) show that in the lake sediments, the 0.11 M acetic acid solution extract more quantity of Zn than EDTA method(L1, L2, L4, L5) or similarly contents (L3, L4, L7, L8). The EDTA solution release highest quantity of mobile Ni; also, for the mobile Cu and Pb, the EDTA method extract more quantity (L1, L3, L5-L8) than the other two methods.

In Brazilor Lake was recorded high concentration of exchangeable Ni, obtained with all three methods (20 mg/Kg d.m, represent 66%.), even if the total concentration is under the reference value; in Cartuş Lake, where the total Ni was over the reference value, the mobile Ni (5.5 mg/Kg d.m.) represent only 15 % from the "total concentration". The higher value for de mobile Zn content was obtained in Cartuş Lake (100 ppm) and represent approximately 24% from the total concentration (431 ppm). Even if in L5 sediment (Brazilor Lake) was high concentration of total Zn (453 ppm), three times more than the reference value, the mobile Zn was only 10% from the total content, without problem for the environment. The same situation was recorded for the other two sediments with high concentration of total Zn situated over the reference value; L1 and L8, where the mobile Zn represent 9.5%, respectively 15%.

The highest value for mobile Cu (46 ppm) was obtained in Mare Lake, where 19% from the total concentration (244 ppm) was found; for the other sediment samples, the values for mobile Cu were situated in the range 7 – 19 ppm.

In both river sediments (figures 9, 10), for mobile Pb contents the extraction order was EDTA > DTPA > acetic acid. The EDTA solution extract 10 times more Pb than 0.11 M acetic acid; also, for mobile Cu in Săsar river the EDTA method release higher quantity than DTPA and acetic acid solutions; for mobile Zn, EDTA and acid acetic solutions extract similarly contents, higher than DTPA solution.

In Săsar river was founded 1.5 mg/Kg d.m. of mobile Cd (with DTPA and EDTA solutions), that represent a potential source of pollution for the quality of surface water; also high quantity of mobile Pb (140 mg/Kg d.m) and Zn (261 mg/Kg d.m) were recorded, these mobile concentration been higher than the reference values for the total content of metals in the sediment structure according Roumanian Order no. 161/2006.

The aim of the sequential chemical extraction procedure was to clarify the differences between the tested methods. In BCR CRM 701 lake sediment (figures 11, 12) the sequential procedure shows that after first extraction, for the mobile Cu and Pb, the extracted concentrations increase in order EDTA > DTPA > acetic acid. For the other elements (Cd, Ni and Zn), the order was the following: acetic acid > EDTA > DTPA. The efficiency of extraction was 3 times higher in EDTA solution than in 0.11 M acetic acid for Cu and 17 times higher for mobile Pb. The quantity of mobile Zn release by BCR method is only with 1.2 times more than the concentration obtained with EDTA solution. For the mobile Cd and Ni, the values obtained were situated in the same range.

The other two extraction solutions used in sequential procedure release significant concentration of Cu and Zn (distillated water) and low contents for the same metals in 0.11 M acetic acid. This fact conducts to the conclusion that after the first extraction can be used a second step with distillated water and the total content of mobile metallic elements can be a sum of this two fractions.

CONCLUSIONS

The paper proposed a method for extraction of mobile metallic elements[17] from different terrestrial sediment. In the paper were compared the results obtained with three different extraction methods applied to eleven sediments: CRM BCR 701, eight sediments collected from Roşia Montana aria and two surface waters sediments.

The exchangeable fraction corresponds to the form of metals that is most available for plant uptake and can be release by changing the ionic strength of the medium.[2] The method NF X 31 – 120/1-92 use an organic complexant (EDTA) and the solution can modify the ionic strength in the sediment structure using different mechanism (ion-exchange, complexation) and also, can release more quantity of metallic elements than 0.11 M acetic acid (method BCR).

The metal content bound to carbonates is sensitive to pH changes and can became mobile when pH is lowered.[2] For this reason, 0.11 M acetic acid (pH=2.8) can release higher quantity of metallic carbonates then the other two solutions with the neutral pH. This case was recorded for Zn, higher quantity of Zn was obtained in this solution.

The experimental data proved that NFX31 – 120/1-92 (EDTA and NH_4OAc solution) can be successfully used for estimate the release of metallic elements from terrestrial sediments polluted by mining activities.

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